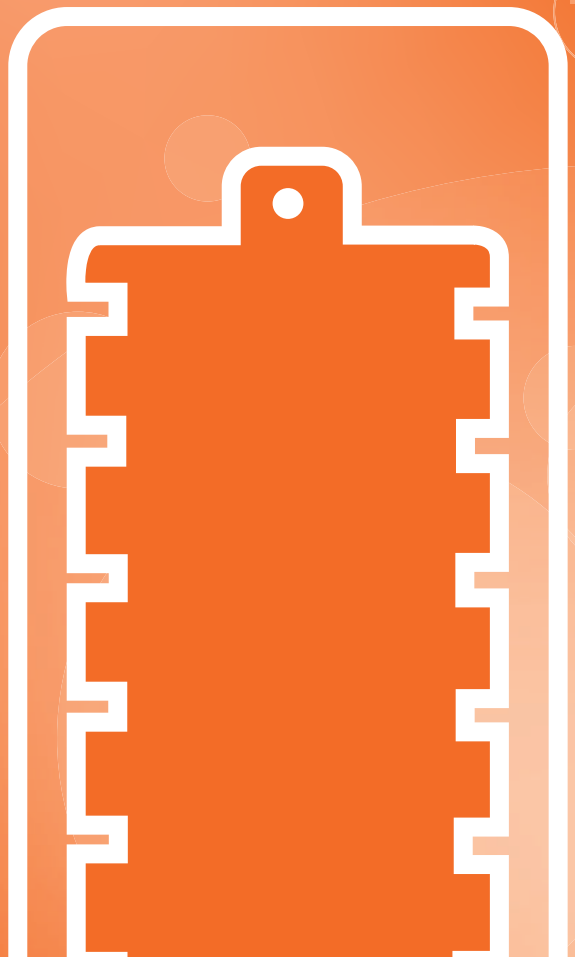
isoplexis

Single-Cell Metabolome

Multi-Omic Energy State Application for Integrated Cancer Biology



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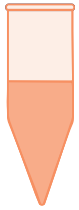
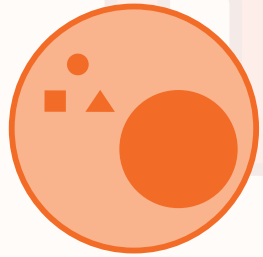


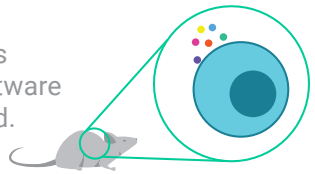


Solving your research challenges one app at a time

IsoPlexis' systems enable you to move your programs forward in a comprehensive fashion – each week – with minimal human resources.

DAY 1 Low Volume High Multiplexed Proteomics

Uniquely highly multiplexed and low sample volume, in an automated format, for precious preclinical samples and in vitro human – software delivers insights day 1; no technician required.

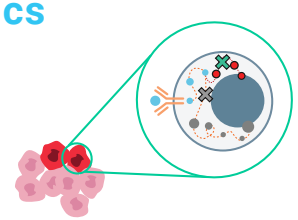


DAY 2 Single-Cell Secreted Proteomics

Build upon bulk multiplexed data, with **unique & gold standard single-cell secreted proteomics insights** which have created 50+ predictive data sets. Reveal optimal complex immune therapy candidates, with higher more durable efficacy – in a predictive fashion.

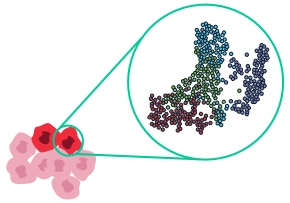
DAY 3 Single-Cell Phosphoproteomics

Reveal how heterogeneous target tumor cells are responding to or resisting therapy by leveraging **uniquely high multiplexed phosphoprotein readouts per cell.**



DAY 4 Multi-Omic Energy States

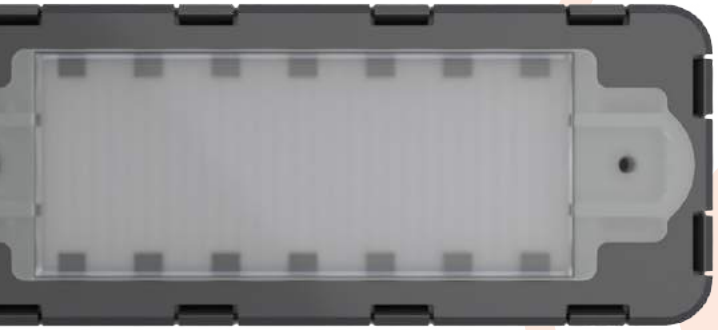
Connect functional proteins and metabolites to energy states for the first time. Identify functional activation pathways and metabolic function of single cells with **highly multiplexed metabolomics.**



Single-Cell Metabolome

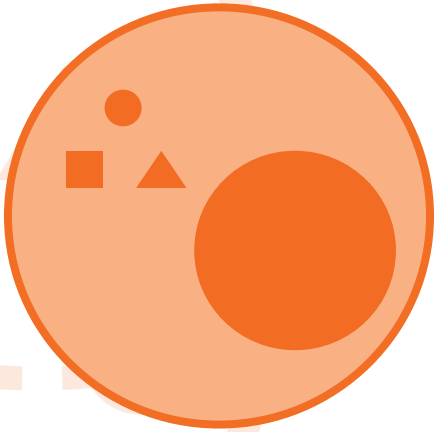
Solution and Cytokine Panel Menu

IsoPlexis' multi-omics (metabolomics + functional proteomics) provides a critical and uniquely capable tool for addressing functional and metabolic changes that accompany adaptive resistance development. Through the use of IsoPlexis' multi-omic energy state application, independent trajectories to drug tolerance can be revealed, enabling researchers to better develop combination therapies to combat this drug-resistant cell state and prevent drug resistance.



Automated Ease-of-Use:

- **Highly Multiplexed:** Targets up to 15+ phosphoproteins and metabolites
- **One System:** Automated imaging, incubation, washing, and ELISA
- **Published:** In a variety of peer-reviewed journals and indication types



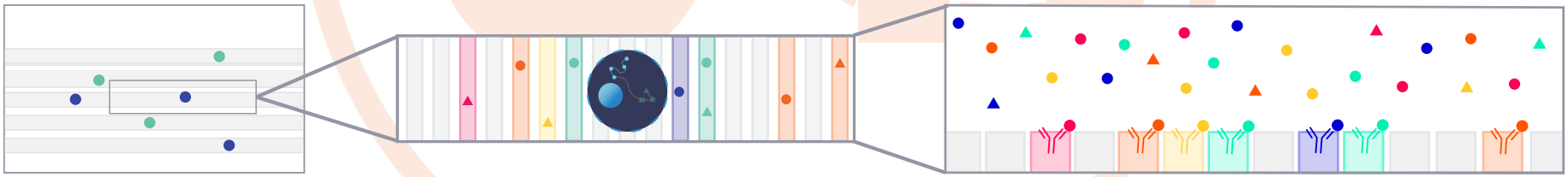
Tumor Metabolome

P-PRAS40, P-IkBα, P-NF-kβ p65, P-Met, P-p44/42 MAPK, P-S6 Ribosomal, P-Rb, P-p90RSK, P-Stat3, P-MEK1/2, P-Stat1, P-Stat5, P-eIF4E, Cleaved PARP, Alpha Tubulin, Glucose-Biotin

Unique Single-Cell Proteomics Technology

Enables Mapping of Multi-Omic Proteomic Pathways and Energy States at the Single-Cell Level

The Single-Cell Metabolome solution measures cellular communication via phosphorylation events and adaptive resistance pathways. Microchambers capture single cells. IsoPlexis' unique proteomic barcode captures the full range of metabolites and functional proteins, enabling predictive intracellular discoveries.



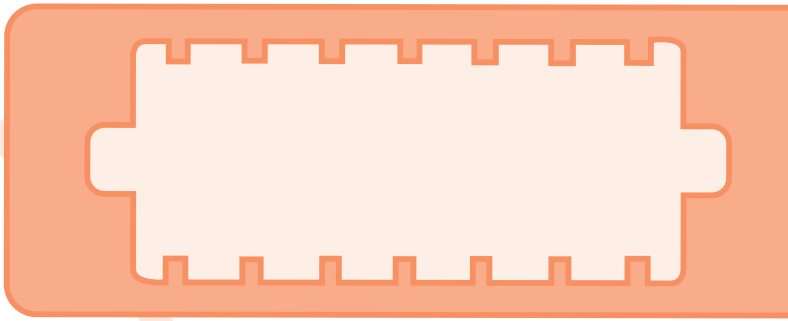
CELLS CAPTURED

CELLS LYSED

SENSITIVE BARCODED ELISAS DETECT MULTIPLEXED PHOSPHOPROTEINS AND METABOLITES

Multi-Omic Energy State:

Connecting Functional Signaling Pathways to Energy States for Integrated Cancer Biology



Multi-Omic Analysis Reveals Paths Toward Drug Resistance in Melanoma with Single-Cell Metabolomics and Single-Cell Phosphoproteomics

IsoPlexis' Single-Cell Metabolomics solution combines the ability to look at phosphoproteins and metabolites, enabling researchers to perform multi-omic analyses on their cells with just one technology. Functional signaling pathways can be identified and connected to energy states for the first time with this solution, leading to critical insights in functional adaptations and resistance.

Researchers Su, et al. used the Single-Cell Metabolome technology to analyze mutant melanoma cancer cells ($BRAF^{V600E}$M397). The researchers sought to gain a deeper understanding of the transition from drug responsive to drug tolerant in $BRAF^{V600E}$M397 cells, as these cancer cells have demonstrated the ability to quickly become resistant to targeted inhibitors.

The M397 cell cultures were treated with BRAF inhibitor (BRAFi) for five days and analyzed each day with IsoPlexis' multi-omic energy state technology using a panel to simultaneously capture phosphoproteins and metabolites from single cells to get a snapshot of how these cells were functioning day to day. To resolve "the complex cell-state space traversed by the cells during the first few days of BRAFi treatment"[1] Su, et al. conducted single-cell analysis consisting of phenotypic markers and markers of oncogenic signaling, cell proliferation, and metabolic activity, which are all transformed in the preliminary drug response.

After the first day of BRAFi treatment, inhibited glucose intake and the suppression of most metabolic regulators and signaling phosphoproteins as well as Ki67 indicated that the treatment was effectively blocking a key oncogenic signaling pathway. There was, however, a small subset of M397 cells that were still Ki67-High at this time, demonstrating a slower drug response.[1] Many of the analytes showed a sharp and temporary rise in variance on day 3 which was reduced by day 5, including metabolic enzymes, resistant state markers, and signaling phosphoproteins. This rise in variation at day 3 suggests that one or more cell state changes may occur at this point in time.

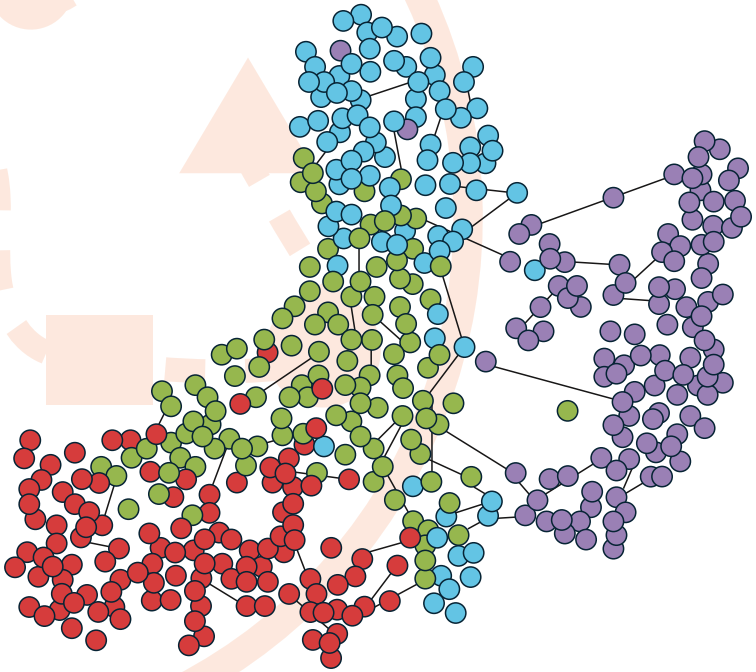
On day 5, most of the cells went into a state of senescence, but there was no increase in apoptosis. Additionally, researchers noted an elevation of factors generally associated with drug resistance on day 5, such as AXL, N-cadherin, NGFR, and TNFR.

Overall, single-cell metabolomics and phosphoproteomics provided evidence of initial drug response at day 1, a drug-induced cell-state change at day 3, and emerging drug tolerance at day 5, followed by an increase in cell proliferation (full drug resistance) a few weeks later.[1]

Multi-Omic Energy State Application Enables Development of Better Combination Therapies Combatting Drug Resistance

Based on these findings, Su, et al. determined a small subpopulation of the M397 cells did not respond well to BRAF inhibition. Single-cell analysis with IsoPlexis' technology showed that untreated cells "contain both MITF-Low and MITF-High cell populations, which tend to take different paths to develop drug tolerance."[1] The $BRAF^{V600E}$M397 cells took multiple pathways to drug resistance, making targeting the resistance mechanisms of these cells more difficult. The researchers were able to identify drug susceptibilities for both pathways which significantly inhibited tumor growth.

With the help of IsoPlexis' multi-omic energy state application, Su, et al. were able to resolve heterogeneous drug-response trajectories that provided a powerful method for treating therapeutic resistance. IsoPlexis' Single-Cell Metabolomics measures functional energy states and adaptive resistance pathways to combat drug resistant cell states. Without this multi-omic technology from IsoPlexis, researchers may not have been able to identify both resistance pathways, which ultimately led to identifying a combination therapy that resolved therapeutic resistance. With this methodology and IsoPlexis' unique multi-omic platform, researchers can develop effective combination therapies and identify highly functional cell subsets which are predictive of patient outcome in complex cancers and beyond.



soSpeak:

Automated Push-Button Analytics

The integrated IsoSpeak data informatics software provides researchers with publication-ready data with a same-day turnaround. The advanced visualizations offer multiple ways to stratify responders and non-responders and identify other meaningful information not previously accessible--all at the touch of a button.

t-SNE
High-Dimensional Single-Cell Mapping

PF OVERVIEW
Reveal the Polyfunctionality of Your Samples

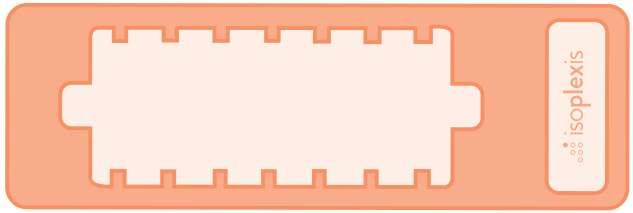
UMAP
Highlight Differences in High Dimensional Datasets

PAT PCA
Stratify Donor/ Patient Response

PF Heatmap
Uncover Critical Cells and Subpopulations

PSI
Reveal the Potency of Different Immune Cell Types

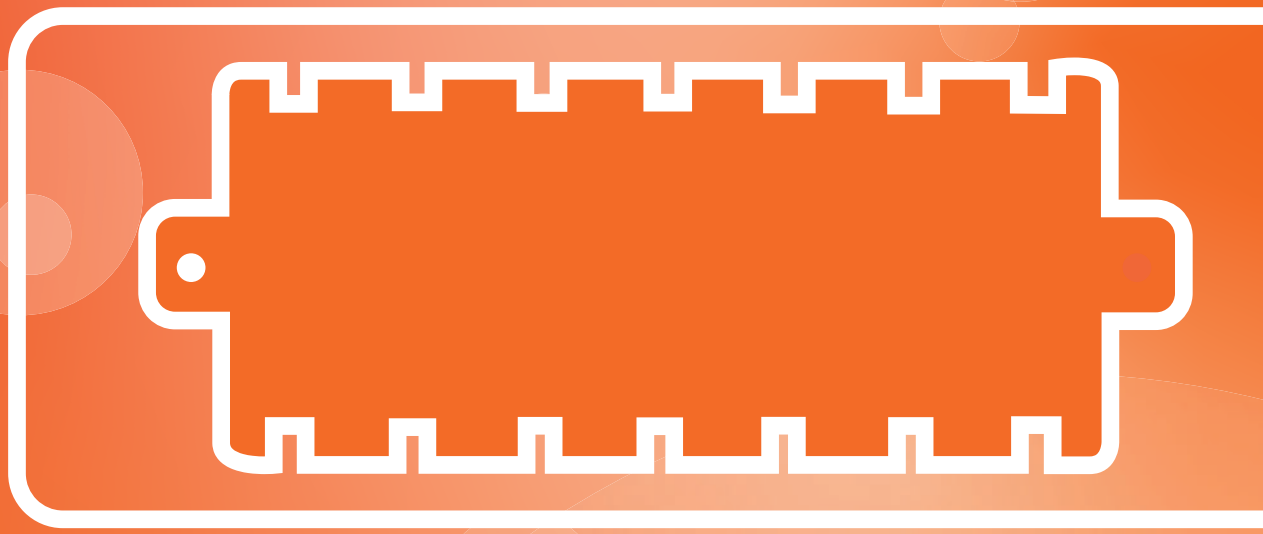
Conclusion



- IsoPlexis' multi-omic energy state application simultaneously identifies functional signaling pathways and connects them to energy states. For the first time, the intracellular proteome and metabolome are connected together in an integrated multi-faceted technology.
- It is well documented that cancer cells use metabolic pathways to rewire their metabolism and energy production so that they can rapidly grow and become resistant to cancer treatments through functional adaptations. In order to gain deeper clarity on these functional adaptations, tumor cells must be analyzed at the single-cell level with multi-omic approaches to detect and predict multiple paths to resistance.
- IsoPlexis' multi-omic energy state application reveals these critical functional adaptations with unique single-cell metabolomics and intracellular signaling proteomics, enabling researchers to better develop combination therapies to combat drug-resistant cell states.

References

1. Su Y et al. Multi-omic single-cell snapshots reveal multiple independent trajectories to drug tolerance in a melanoma cell line. Nature Communications 11: 2345, 2020.



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