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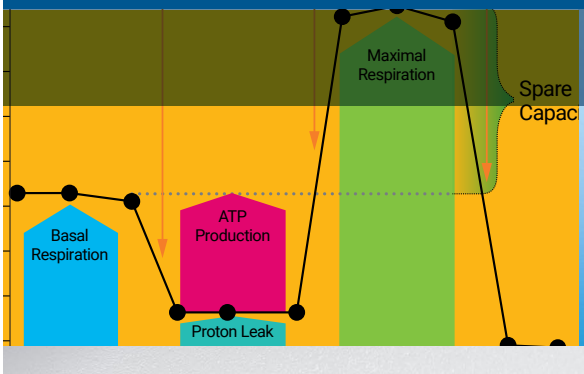
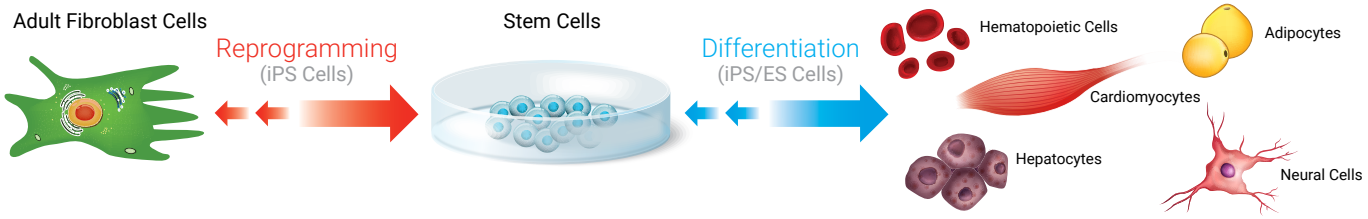
Trusted Answers

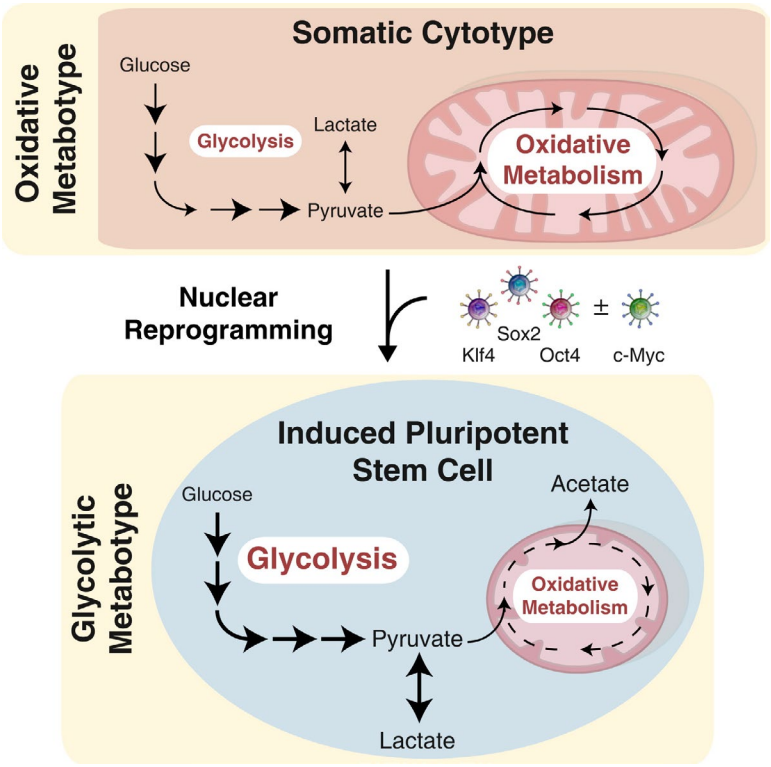
Agilent Seahorse XF Live-Cell Metabolism Solutions for Stem Cell Research



Improve Differentiation and Reprogramming Outcomes



Cellular age and origin, in addition to donor variability, protocol differences, growth rates and media choices all contribute to inconsistent reprogramming and/or differentiation efficiencies. Metabolic energy utilization, characterized before and after cell fate changes occur, identifies the metabolic phenotype and enables researchers to predict and confirm cell function, revealing actionable reprogramming and differentiation potential.



Somatic oxidative bioenergetics transitions into pluripotency-dependent glycolysis to facilitate nuclear reprogramming.
Folmes, C. D., *et al.* Cell Metab. 2011. 14: 264-71.

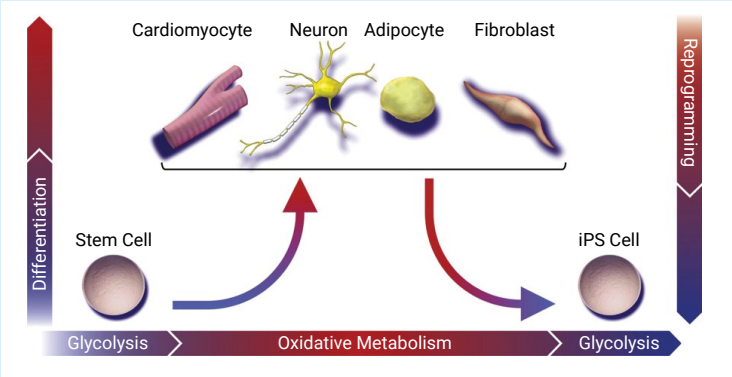
Cellular metabolic phenotyping measures the cell's energy requirement and pathway preference for readying the transition between undifferentiated and differentiated states. Metabolic switching occurs rapidly as cells transition from quiescent to pluripotent and/or from pluripotent to differentiated.

Seahorse XF Technology:

- Live cell
- Real-time
- Label-free
- Dynamic injection ports
- Measures oxygen consumption and glycolytic rates simultaneously

Identify Pluripotency and Differentiation Transitions

Seahorse XF technology enables reliable measurements that predict, monitor, and track cell fate transitions. Discover how these metabolic measurements can be used as indicators to minimize inefficiencies and improve differentiation and reprogramming approaches. Routine assays make identifying cell phenotype and cell transitions easy. What's more, the metabolic phenotyping analysis that Seahorse XF delivers provides the tools and knowledge to customize your approach, and push the conventional boundaries of stem cell research through the development of new assays.



Metabolic plasticity in stem cell homeostasis and differentiation.
Folmes, C. D., *et al.* Cell Stem Cell. 2012. 11: 596-606.

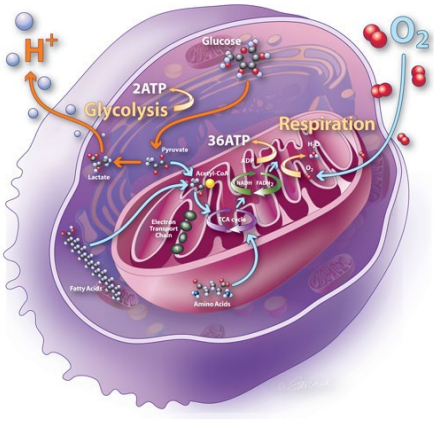
Seahorse XF Technology:

- Measures distinct metabolic signatures
- Characterizes cellular phenotypes at each stage
- Enables routine and reliable stem cell phenotyping
- Facilitates the discovery of new standards and benchmarks

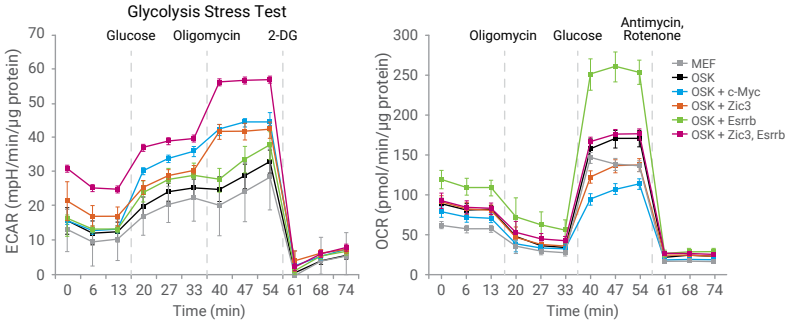
Seahorse XF Technology simultaneously measures rates of oxidative metabolism and glycolysis using label-free methods on live cells, in real-time.

"iPSCs and their differentiated counterparts are metabolically distinct and these metabolic parameters are important for stem cell identity."

-Dr. James Ryall, University of Melbourne, Australia



Reprogramming Efficiency is Improved with Increased Glycolysis and Oxidative Phosphorylation



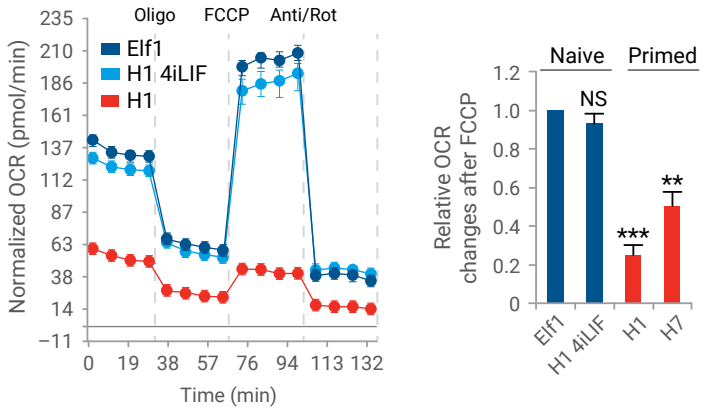
Metabolic phenotype can be a leading indicator for reprogramming efficiency

- Enhance reprogramming efficiency by modulating both the glycolytic and mitochondrial pathways.
- Measure gain of pluripotency
- Connect metabolic state with cellular identity

Metabolic reprogramming precedes changes in gene expression, and is required for efficient reprogramming

Hybrid Cellular Metabolism Coordinated by Zic3 and Esrrb Synergistically Enhances Induction of Naive Pluripotency.
Sone M., *et al.* Cell Metabolism 2017. (25)5: 1103-1117.

Determine Differentiation Potential by Distinguishing Naïve and Primed Stem Cells



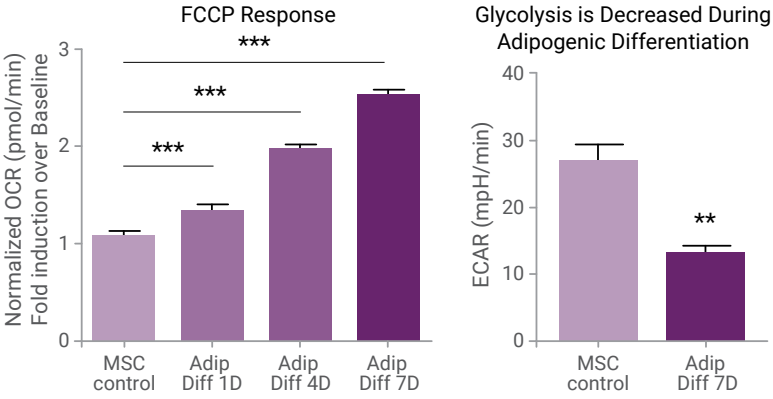
Quality Control Your Cells

- Evaluating pluripotent stem cell metabolic signatures reveals when to bank iPS cells or when to begin differentiation
- Energy pathway prevalence determines a cell's readiness for differentiation
- Calculating the timing and efficiency of the metabolic switch is essential for improving gene targeting effectiveness

Metabolic signature transformations stimulate cell fate changes

The metabolome regulates the epigenetic landscape during naive-to-primed human embryonic stem cell transition.
Sperber, H., *et al.* Nat Cell Biol. 2015. 17: 1523-35.

Monitor Metabolic Switching Events Underlying Differentiation Progression



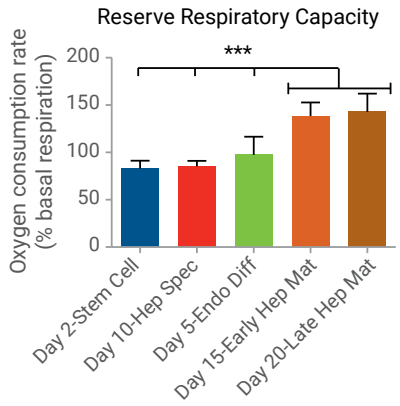
Increasing oxygen consumption rates reflect high energy demand required for lineage commitment, differentiation and maturation

Cell Fate Transitions

- Spare Respiratory Capacity defines cell's propensity to differentiate
- Measure Glycolytic rates to determine proliferation and self-renewal ability
- Determine the commitment stage based on the metabolic switch

Mitochondrial Respiration Regulates Adipogenic Differentiation of Human Mesenchymal Stem Cells.
Zhang, Y., *et al.* PLoS ONE. 2013. 8: e77077.

Confirm Differentiation



Differentiating hepatocytes switch to an oxidative phenotype

Functional Performance

- Measure metabolic switching events to determine lineage commitment at an early stage
- Orient functional potential to actual function during lineage specification
- Confirm disease model efficacy by comparison with the parental phenotypic metabolic profile

Bioenergetic Changes during Differentiation of Human Embryonic Stem Cells along the Hepatic Lineage.
Hopkinson, B. M., *et al.* Oxid Med Cell Longev. 2017. 2017: 5080128.

Measure Functional Performance and Model Relevance

Normal Model

ES Cells

Gene Editing

Spontaneous Chromosomal Aberration

Genetic Defect

Somatic Cells

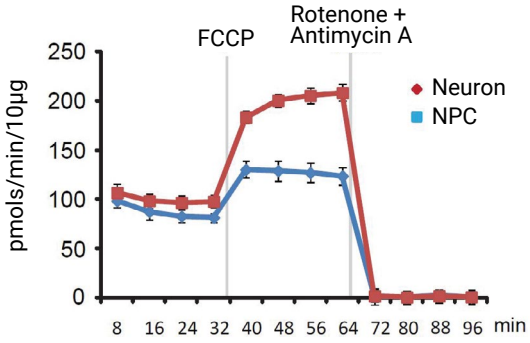
Reprogramming

Optimize Disease Models

- Compare somatic, origin, pluripotent intermediate, and differentiated cells
- Modulate metabolism to improve functional outcome
- Standardize assays for cellular characterization

Pluripotent Stem Cell with a Genetic Defect

Metabolic Characterization



Obesity, Diabetes, and Metabolic Disorder Research

Cancer Research

Hepatic Research

Neurobiology Research

Cardiovascular Research

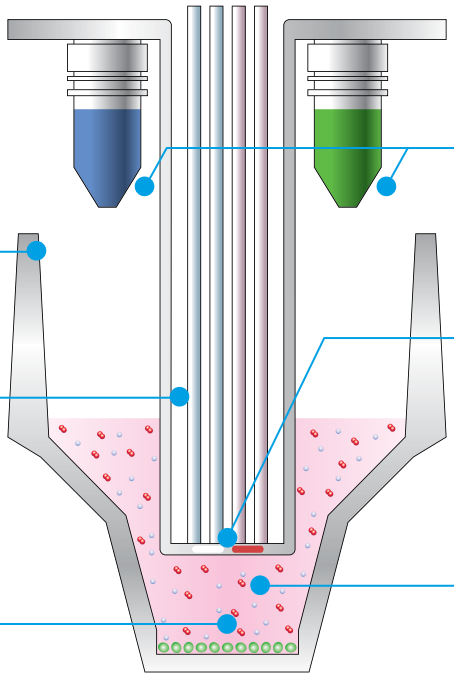
Hematopoietic Research

Metabolic reprogramming during neuronal differentiation from aerobic glycolysis to neuronal oxidative phosphorylation.
Zheng, X., *et al*. Elife. 2016. 5:e13374 (data figure).

Seahorse XF Analyzers

Seahorse XF Analyzers simultaneously measure the two major energy pathways of the cell — mitochondrial respiration and glycolysis — in live cells using label-free, solid-state sensor cartridges in a microplate format. They work with many cell types, including primary cells, cell lines, suspension cells, as well as islets, spheroids, and isolated mitochondria.

The patented design makes it all possible



Integrated injection ports sequentially deliver up to 4 compounds allowing multiple conditions per well.

Seahorse XF Cell Culture Microplates are tissue culture treated and plate reader compatible.

Sensor probes gently lower to create a transient microchamber, allowing rapid, real time measurement of changes in both oxygen and proton concentrations in the extracellular medium.

Inert optical microsensors measure rates of oxygen consumption and extracellular acidification simultaneously, without addition of dyes.

The microplate well requires a small number of cells, 10-20 fold fewer cells compared to conventional respirometers.

Seahorse XF Analyzers utilize patented transient microchambers which enable sensitive, precise, and label-free metabolic measurements of live cells in real time.

Seahorse XF Assays and Kits

Assay	Measure	Significance
XF Cell Mito Stress Test	Mitochondrial function and spare espiratory capacity	Low SRC indicates pluripotency High OXPHOS indicates differentiation
XF Glycolytic Rate Assay	Glycolysis utilization and capacity to compensate for energy demand	High glycolytic capacity indicates pluripotency and proliferation
XF Mito Fuel Flex Test	3 major fuel oxidation pathways: glucose, glutamine, and fatty acids (pathway dependence)	Removal of glutamine prompts cells to differentiate
XF Cell Energy Phenotype Test	Measures glycolysis and OXPHOS simultaneously (pathway preference)	Energy map can easily distinguish differentiated versus stem cell populations Switch is essential for successful differentiation

Measure What's Important to Your Cell

With over 20,000 genes, 200,000 proteins and thousands of pathways, you can't measure everything in a cell at once, but you can measure what provides the energy that drives them—metabolism.

Agilent Seahorse XF technology detects changes in cell bioenergetics in real-time, providing a window into the critical functions driving cell signaling, proliferation, activation, toxicity and biosynthesis.

Move beyond analyzing what your cells are, and reveal a clearer picture of what they do.

Agilent Seahorse Wave Software

Wave, the primary Seahorse software program, enables the transformation of raw kinetic data into powerful results. Wave provides preloaded templates and protocols for each Seahorse XF assay kit, reducing time for assay design, as well as several analysis views and export options that facilitate Seahorse XF data analysis and interpretation.

Learn More

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