Agilent Seahorse XF Assay Kits and Reagents

Standardized, optimized, live-cell metabolic assays for use with Agilent Seahorse Analyzers

Agilent provides a wide range of Seahorse XF assay kits, reagents, and media for studying cellular function, including metabolism and energetics and assessing mitochondrial and glycolytic function in real time. These kits are designed to simplify workflows and improve research productivity, enabling generation of consistent data on your Seahorse XF analyzer.

Together with Agilent's excellent technical support, these kits will greatly enhance your overall experience of performing XF assays.







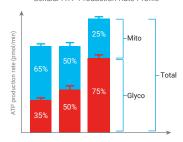


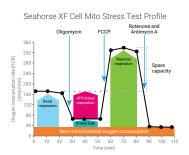
Key features and benefits include:

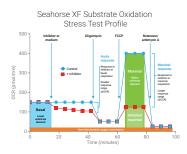
- An optimized open-and-go format for simplified protocol and reduced assay complexity
- Direct reconstitution of compounds in assay medium—eliminating the need for DMSO or other solvents
- A single-use concept to reduce waste and eliminate the need for storage of used compounds
- Rigorous testing and validation for every lot with cells to deliver consistent and robust results eliminating the cost associated with failed experiments

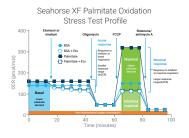


Cellular ATP Production Rate Profile











Agilent Seahorse XF Real-Time ATP Rate Assay Kits

(p/n 103592-100 and 103591-100)

This is the only assay that quantifies the production rates of adenosine triphosphate (ATP) from mitochondrial respiration and glycolysis simultaneously, in real time with live cells. It offers a more informative and quantitative approach to assess the interplay between cellular functions and energy metabolism in response to gene modification, compound exposure, and/or other types of interventions. For more information, visit the Agilent website at http://www.agilent.com/chem/realtimeatp

Agilent Seahorse XF Cell Mito Stress Test Kits

(p/n 103015-100 and 103010-100)

This widely-used assay kit is an optimized solution for assessing mitochondrial function. In this assay, key parameters obtained include ATP-linked respiration, maximal respiration, and spare respiratory capacity. The assay provides insights into the mechanism of mitochondrial dysfunction and enables investigation of functional differences among cell types, drug candidates, as well as genetic or biochemical interventions.

Agilent Seahorse XF Substrate Oxidation Stress Test Kits

(p/n 103672-100, 103673-100, and 103674-100)

A suite of powerful assays enabling interrogation of cellular substrate oxidation under high energy demand conditions in live cells. These kits facilitate the investigation of how cells alter or shift oxidation of mitochondrial substrates (e.g., long-chain fatty acids, glucose/ pyruvate, and/or glutamine) to perform key cellular functions of activation, proliferation and differentiation, survival in microenvironments, as well as responses to genetic or pharmaceutical interventions. For more information, visit the Agilent website at https:// www.agilent.com/en/promotions/seahorse-xf-subox

Agilent Seahorse XF Palmitate Oxidation Stress Test Kit (p/n 103693-100)

This conventional fatty acid oxidation (FAO) assay kit provides ready-to-use reagents, including the XF palmitate-BSA FAO substrate, etomoxir, oligomycin, FCCP, rotenone/ antimycin A, and L-carnitine. It enables determination of the intrinsic rate and capacity of a cell to oxidize long-chain fatty acids (LCFAs) in the absence or limitation of other exogenous substrates, facilitating investigation of how interventions (e.g., genetic manipulations, drug exposures) specifically affect the LCFAs oxidation process.

Agilent Seahorse XF Plasma Membrane Permeabilizer (p/n 102504-100)

Perform isolated mitochondria type assays without isolating mitochondria! This exclusive reagent permeabilizes the plasma membrane of intact cells in culture without damage to mitochondrial membranes, enabling experimental control of substrate provision to the mitochondria and detailed characterization of key components in mitochondrial function, such as transporters, enzymes, and electron transport chain complexes.

Seahorse XF T Cell Metabolic Profiling Assays

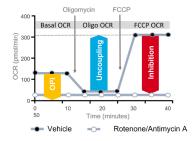
Algorithm A Diligomycin A BAM15 Rotenone/Antimycin A Time (min)

Time (min)

MitoATP GlycoATP

GlycoATP

Agilent Seahorse XF Mito Tox Assay Profile



Seahorse XF Glycolytic Rate Assay Profile

Rev/AA

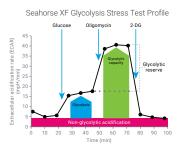
2-DG

Total proton efflux

Glycolytic proton efflux

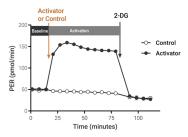
Glycolytic proton efflux

30 40 Time (min)





Seahorse XF Hu T Cell Activation Assay Profile



Agilent Seahorse XF T Cell Metabolic Profiling Kits

(p/n 103771-100 and 103772-100)

This kit is optimized for T cell and NK cell research and therapeutic discovery with improved reagents and assay performance. It provides robust bioenergetic parameters linked to critical attributes for antitumor properties: persistence and metabolic fitness. The kit is suitable for use in evaluating CAR T and CAR NK engineering strategies, starting material selection, and metabolic conditioning during in vitro cell expansion. It is also applicable to assessing the capacity of T cells and NK cells to maintain metabolic fitness in tumor microenvironments.

Agilent Seahorse XF Mito Tox Assay Kit

(p/n 103595-100)

The kit is a part the XF Mito Tox solution that integrates the Agilent Seahorse XF Pro analyzer, a streamlined workflow and enhanced software tools, enabling to easily identify drug-induced mitochondrial toxicity early in drug discovery, thus de-risk drug pipeline. It provides high sensitivity and specificity through direct measurements of mitochondrial dysfunction and allows for screening compound libraries with a throughput of 80 compounds per plate. It also simplifies template design and data analysis with dedicated software tools and delivers actionable conclusions quickly with high confidence.

Agilent Seahorse XF Glycolytic Rate Assay Kit

(p/n 103344-100 and 103346-100)

This assay is an optimized complete solution (kit, media, software) that quantifies the rate of glycolytic activity in live cells by accounting for both glycolytic and mitochondrial (CO_2) acidification. This kit delivers robust, informative, and precise glycolysis data that enable the study of transient responses and metabolic switches in cellular disease models. This low-cost, real-time, functional assay is a superior alternative to the end-point lactate assay.

Agilent Seahorse XF Glycolysis Stress Test Kit

(p/n 103020-100 and 103017-100)

A relative test of glycolytic function in live cells, this assay measures the capacity of the glycolytic pathway by driving cells toward glycolysis and assesses the ability of study models to increase glycolytic activity to meet metabolic and bioenergetic demands.

Agilent Seahorse XF Palmitate-BSA FAO Substrate

(p/n 102720-100)

A bio-available, preconjugated substrate with a quantified free palmitate concentration, used for in vitro studies of LCFA oxidation by intact cells. It is supplied as a part of the Seahorse XF Palmitate Oxidation Stress Test kit and is also available alone for other types of applications.

Agilent Seahorse XF Hu T Cell Activation Assay Kit

(p/n 103759-100 and p/n 103766-100)

The assay measures human T cell activation response in minutes, via monitoring proton efflux rates (PER) associated with glycolysis-dependent energy production, thus providing an early window into the dynamics of immunometabolism that drive many key aspects of immune responses. This metabolism-based T cell activation assay kit, together with standardized protocol and Agilent Seahorse Analytics, offers an optimized turnkey solution and delivers unique insights into the mechanisms underlying T cell activation.

Product ordering information

P/N	Product Description	Unit	Product Content	Compatible Analyzer
103592-100	Seahorse XF Real-Time ATP Rate Assay Kit	EA	Six (6) pouches ¹	XF Pro, XFe96, XFe24
103015-100	Seahorse XF Cell Mito Stress Test Kit	EA	Six (6) pouches ¹	XF Pro, XFe96, XFe24
103672-100	Seahorse XF Long Chain Fatty Acid Oxidation Stress Test Kit	EA	Three (3) pouches ¹	XF Pro, XFe96, XFe24
103673-100	Seahorse XF Glucose/Pyruvate Oxidation Stress Test Kit	EA	Three (3) pouches ¹	XF Pro, XFe96, XFe24
103674-100	Seahorse XF Glutamine Oxidation Stress Test Kit	EA	Three (3) pouches ¹	XF Pro, XFe96, XFe24
103693-100	Seahorse XF Palmitate Oxidation Stress Test Kit	EA	Contains one (1) each of these products: 103672-100, 102720-100, 103689-100	XF Pro, XFe96, XFe24
103260-100	Seahorse XF Mito Fuel Flex Test Kit	EA	Six (6) pouches ¹	XF Pro, XFe96, XFe24
103344-100	Seahorse XF Glycolytic Rate Assay Kit	EA	Six (6) pouches ¹	XF Pro, XFe96, XFe24
103020-100	Seahorse XF Glycolysis Stress Test Kit	EA	Six (6) pouches ¹	XF Pro, XFe96, XFe24
103772-100	Seahorse XF T Cell Metabolic Profiling Kit	EA	Six (6) pouches ¹	XF Pro, XFe96
103595-100	Seahorse XF Mito Tox Assay Kit	EA	Six (6) pouches ¹	XF Pro Analyzer
103591-100	Seahorse XFp Real-Time ATP Rate Assay Kit	EA	Six (6) pouches ²	XFp, XF HS Mini
103010-100	Seahorse XFp Cell Mito Stress Test Kit	EA	Six (6) pouches ²	XFp, XF HS Mini
103270-100	Seahorse XFp Mito Fuel Flex Test Kit	EA	Six (6) pouches ²	XFp, XF HS Mini
103346-100	Seahorse XFp Glycolytic Rate Assay Kit	EA	Six (6) pouches ²	XFp, XF HS Mini
103017-100	Seahorse XFp Glycolysis Stress Test Kit	EA	Six (6) pouches ²	XFp, XF HS Mini
103771-100	Seahorse XFp T Cell Metabolic Profiling Kit	EA	Six (6) pouches ²	XFp, XF HS Mini
102720-100	Seahorse XF Palmitate-BSA FAO Substrate	EA	3 × 2 mL palmitate-BSA 3 × 2 mL BSA controls	All XF Analyzers
102504-100	Seahorse XF Plasma Membrane Permeabilizer	EA	One (1) vial of 25 µL PMP in 10 µM concentration	All XF Analyzers
103759-100	Seahorse XF Hu T Cell Activation Assay Kit, 200 tests	EA	1 × 2 mL ImmunoCult human CD3/CD28 T cell activator, 2 × 2-deoxy-D-glucose	XF Pro, XFe96, XFp, XF HS Mini Analyzers
103766-100	Seahorse XF Hu T Cell Activation Assay 96-well Pack, 200 tests	Pk	1 × p/n 103759-100 2 × XFe96 PDL FluxPak	XF Pro, XFe96 Analyzers
103335-100	Seahorse XF Base Medium, without phenol red, 500 mL	EA		All XF Analyzers
103575-100	Seahorse XF DMEM Medium, pH 7.4, 500 mL	EA		All XF Analyzers
103680-100	Seahorse XF DMEM Assay Medium Pack	PK	Contains one (1) each of 103575-100, 103577-100, 103578-100, 103579-100	All XF Analyzers
103576-100	Seahorse XF RPMI Medium, pH 7.4, 500 mL	EA		All XF Analyzers
103681-100	Seahorse XF RPMI Assay Medium Pack	PK	Contains one (1) each of 103576-100, 103577-100, 103578-100, 103579-100	All XF Analyzers
103577-100	Seahorse XF 1.0 M Glucose Solution, 50 mL	EA		All XF Analyzers
103578-100	Seahorse XF 100 mM Pyruvate Solution, 50 mL	EA		All XF Analyzers
103579-100	Seahorse XF 200 mM Glutamine Solution, 50 mL	EA		All XF Analyzers

¹ One XF pouch contains sufficient reagents to run one 24-well or 96-well plate

www.agilent.com/chem/discoverXF

DE.5508449074

This information is subject to change without notice.

© Agilent Technologies, Inc. 2020, 2022–2024 Published in the USA, March 19, 2024 5991-8291EN 103582-400 Rev. G

US and Canada

1-800-227-9770 cellanalysis.support@agilent.com

Europe and Asia Pacific

cellanalysis.support@agilent.com



 $^{^{\,2}\,}$ One XFp pouch contains sufficient reagents to run one 8-well mini-plate