Cellular Energy Metabolism



Energy uptake and utilization in cells is a dynamic process regulated by interacting metabolic networks. Interrogating this complex network would benefit from rapid, simple techniques to specifically measure key metabolites. We have several bioluminescent assays for reproducible and sensitive detection of glucose, lactate, glutamate and glutamine in a plate-based format. These versatile assays are amenable to higher-throughput formats, and compatible with many sample types.

Metabolite Detection Assays:

Glucose-Glo[™] Assay

Promega

- Lactate-Glo[™] Assay
- Glutamate-Glo™ Assay
- Glutamine/Glutamate-Glo™ Assay

Assay Advantages

Rapid, Flexible Sample Preparation

Measure metabolites in a variety of sample types for intracellular and extracellular detection. The assays offer easy in-well preparation steps without the need for centrifugation or spin columns.

Convenient Analysis

Offers broad linearity up to 3 logs for simple sample measurement across a wide range of metabolite concentrations.

Detect Small Changes

Has a wide assay window for better discrimination of small changes in metabolite levels compared to colorimetric and fluorometric assays. Signal-to-background window is $S/B_{max} > 100.$

Gain More Information Per Sample

Can measure multiple metabolites from the same sample. Multiplex with cell viability assays for normalization. Requires only small volumes of medium, enabling repeated measurements over time.

Assay Chemistry





Figure 1. Metabolite Detection Assay Principle. A metabolite-specific dehydrogenase catalyzes the oxidation of the metabolite with concomitant reduction of NAD⁺ to NADH. In the presence of NADH, reductase enzymatically reduces a pro-luciferin reductase substrate to luciferine. Luciferin is detected in a luciferase reaction by adding Ultra-Glo[™] rLuciferase and ATP, and the amount of light produced is proportional to the amount of metabolite in the sample.

Assay Workflow



Figure 2. Sample preparation and protocol overview.

Monitor Nutrient Consumption and Metabolite Secretion in the Medium

Monitoring metabolites in cell culture medium can provide information about changes that occur in cellular metabolic pathways. Glucose consumption and lactate secretion can serve as indicators of glycolysis, while glutamate secretion can provide information about glutaminolysis. Changes can be monitored over time or after treatments, such as exposure to hypoxic conditions, by assaying small amounts of diluted medium.



Figure 3. Hypoxia-induced metabolic changes. Two different breast cancer cell lines responded differently under hypoxic conditions (1% oxygen). The MCF7 cells shifted to a more glycolytic phenotype with increased lactate secretion, whereas no significant change was observed for the already highly glycolytic MDA-MB-231 cell line.

Learn more about the design and use of these assays

Leippe, D. *et al.* (2017) Bioluminescent assays for glucose and glutamine metabolism: High-throughput screening for changes in extracellular and intracellular metabolites. *SLAS Discov. 22*, 366–77.doi: 10.1177/1087057116675612



Glutamate Secretion



Figure 4. Cell-dependent glutamine metabolism. Two ovarian cancer cell lines known to have different glutamine requirements exhibited different patterns of glutamine consumption and glutamate secretion over 2 days. SKOV-3 or OVCAR-3 cells were plated in 96-well plates at 10,000 cells/well. At the indicated time points, 2µl of medium was removed, diluted and assayed. The ratio of glutamate secreted to glutamine consumed was less for OVCAR-3 cells (~0.17) compared to SKOV-3 cells (~0.42).

To sample an assay today, visit: www.promega.com/MetabolismAssays

Ordering Information

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Product	Size	Cat.#
Lactate-Glo™ Assay	5ml	J5021
	50ml	J5022
Glucose-Glo™ Assay	5ml	J6021
	50ml	J6022
Glutamate-Glo™ Assay	5ml	J7021
	50ml	J7022
Glutamine/Glutamate-Glo™ Assay	5ml	J8021
	50ml	J8022

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