Glucose Uptake Assay Kit (Colorimetric) ab136955

Overview

Product name: Glucose Uptake Assay Kit (Colorimetric)
Detection method: Colorimetric
Sample type: Adherent cells, Suspension cells
Assay type: Cell-based (quantitative)
Sensitivity: <= 0.01 nmol/well
Assay time: 3h 00m
Species reactivity: Reacts with: Other species, Mammals

Product overview

Glucose Uptake Assay Kit (Colorimetric) (ab136955) is a highly sensitive and easy to use non-radioactive assay kit which can detect glucose uptake as low as 10 pmol/well in a variety of cell types.

2-deoxyglucose (2-DG) is used in glucose uptake assay protocols because of its structural similarity to glucose. 2-DG is taken up by glucose transporters and metabolized to 2-DG-6-phosphate (2-DG6P). 2-DG6P cannot be further metabolized, and thus accumulates within cells. The accumulated 2-DG6P is directly proportional to 2-DG (or glucose) uptake by cells. In this assay, the 2-DG6P is oxidized to generate NADPH, the level of which can be determined by an enzymatic recycling amplification reaction.

Glucose uptake assay protocol summary:
- prepare cells with suitable glucose starvation / uptake stimulation depending on experimental set-up
- add 2-DG to cells and incubate for 20 mins at 37°C
- wash cells with PBS to remove exogenous 2-DG
- lyse cells with extraction buffer and repeated pipetting
- freeze/thaw lysates and heat at 85°C for 40 min
- cool on ice for 5 min
- add neutralizing buffer, spin and transfer supernatant to new tubes
- add supernatants and standards to wells
- add reaction mix A and incubate for 1 hr at 37°C
- add extraction buffer and heat to 90°C for 40 min
- cool on ice for 5 min and add neutralizing buffer
- add reaction mix B
- analyze every 2-3 mins on microplate reader in kinetic mode at 37°C

Notes

Review our Metabolism Assay Guide to learn about assays for metabolites, metabolic enzymes,
mitochondrial function, and oxidative stress, and also about how to assay metabolic function in live cells using your plate reader.

**Platform**

Microplate reader

**Properties**

**Storage instructions**

Store at -20°C. Please refer to protocols.

<table>
<thead>
<tr>
<th>Components</th>
<th>Identifier</th>
<th>100 tests</th>
</tr>
</thead>
<tbody>
<tr>
<td>2-Deoxyglucose</td>
<td>Purple</td>
<td>1 x 1ml</td>
</tr>
<tr>
<td>2-DG6P Standard (Lyophilized)</td>
<td>Yellow</td>
<td>1 vial</td>
</tr>
<tr>
<td>Assay Buffer</td>
<td>WM</td>
<td>1 x 25ml</td>
</tr>
<tr>
<td>Enzyme Mix (Lyophilized)</td>
<td>Orange</td>
<td>1 vial</td>
</tr>
<tr>
<td>Extraction Buffer</td>
<td>NM</td>
<td>1 x 17ml</td>
</tr>
<tr>
<td>Glutathione Reductase (Lyophilized)</td>
<td>Green</td>
<td>2 vials</td>
</tr>
<tr>
<td>Neutralizing Buffer</td>
<td>Clear</td>
<td>1 x 2.5ml</td>
</tr>
<tr>
<td>Recycling Mix (Lyophilized)</td>
<td>Blue</td>
<td>1 vial</td>
</tr>
<tr>
<td>Substrate</td>
<td>Red</td>
<td>2 vials</td>
</tr>
</tbody>
</table>

**Images**

2-DG6P Standard curve (a) and 2-DG uptake in 3T3-L1 cells (b), Human adipocytes (c) and HeLa cells (d) respectively. I=Insulin; P=Phloretin.
Glucose uptake in 3T3-L1 adipocytes stimulated with insulin (I). 3T3-L1 adipocytes were differentiated using:

- Dexamethasone \textbf{ab120743} (1mM, 1:1000)
- IBMX \textbf{ab120840} (11.5 mg/mL, 1:100)
- Insulin \textbf{ab123768} (1 mg/mL, 1:1000)

**Assay Procedure**

Step A: 2-DG oxidation to generate NADPH; Step B: NADPH recycling amplification Reaction.

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