AfCS Solution Protocol

Reagent name: Pervanadate solution, 30 mM

Reagent name abbreviation: 30 mM PVD

Protocol ID: PS00000536

Version: 01

Volume: 1 ml

Components:

<table>
<thead>
<tr>
<th>Reagent</th>
<th>Source</th>
<th>Catalog or Protocol No.</th>
<th>F.W. or Stock Conc.</th>
<th>Quantity</th>
<th>Final Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium orthovanadate, 200 mM</td>
<td>None</td>
<td>PS00000478</td>
<td>200 mM</td>
<td>150 µl</td>
<td>30 mM</td>
</tr>
<tr>
<td>Phosphate buffered saline (PBS)</td>
<td>Invitrogen</td>
<td>20012-027</td>
<td>1X</td>
<td>789 µl</td>
<td>1X</td>
</tr>
<tr>
<td>Hydrogen peroxide in PBS</td>
<td>None</td>
<td>PS00000535</td>
<td>3% (w/w)</td>
<td>61 µl</td>
<td>0.18% (w/w)</td>
</tr>
</tbody>
</table>

Preparation:
1. Dilute sodium orthovanadate in PBS in a 1.5-ml Eppendorf tube.
2. Add hydrogen peroxide in PBS to diluted sodium orthovanadate.
3. Incubate the mixture for 15 min at room temperature, in the dark, before adding to the cells.

Storage:
- Temperature: NA
- Location: __________________________
- Aliquot size: NA
- Special instructions: Prepare solution fresh as needed and add to cells immediately after incubation.

Dilution for treatment of cells at 100 µM:
1. Dilute the solution immediately before use.
2. Dilute 30 mM pervanadate stock thirtyfold into the appropriate cell culture medium to make a 1 mM solution.
3. Dilute 1 mM pervanadate stock tenfold into the existing medium on the cells, resulting in a final concentration of 100 µM pervanadate.

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Date: 07/08/03

Approved: Deirdre Brekken

Comments: None