Technical Data Sheet

Fixation and Permeabilization Solution

Product Information

Material Number: 554722
Size: 125 mL

Description
BD Cytofix/Cytoperm™ solution is supplied as a 1X solution and can be used for the simultaneous fixation and permeabilization of cells prior to intracellular cytokine staining.

Preparation and Storage
Store undiluted at 4°C.

Application Notes

Application

| Intracellular staining (flow cytometry) | Routinely Tested |

Recommended Assay Procedure:
For the stimulation of cells, various in vitro methods have been reported for stimulating cells to produce cytokines. Polyclonal activators have been particularly useful for inducing cytokine-producing cells. These activators include the following: concanavalin A, lipopolysaccharide, phorbol esters plus calcium ionophore or ionomycin, phytohaemagglutinin, staphylococcus enterotoxin B, and monoclonal antibodies directed against subunits of the TCR/CD3 complex (with or without antibodies directed against costimulatory receptors, such as CD28).

1. Fix and Permeabilize Cells
   a. Thoroughly resuspend cells in 100 µL of BD Cytofix/Cytoperm solution per well for microwell plates (or 250 µL for tubes) and incubate for 20 min. at 4°C. Note: Cell aggregation can be avoided by vortexing prior to the addition of the BD Cytofix/Cytoperm™ solution.
   b. Wash cells two times in a buffer that contains a cell permeabilizing agent such as saponin (BD Perm/Wash™ buffer, Cat. 554723, which can be used as the wash buffer and as the antibody diluent).

2. Stain for Intracellular Cytokines
   a. Thoroughly resuspend fixed/permeabilized cells in 50 µL of a saponin-containing buffer (e.g., BD Perm/Wash™ buffer) containing a predetermined optimal concentration of a fluorochrome-conjugated anti-cytokine antibody or appropriate negative control. Incubate at 4°C for 30 minutes in the dark. Note: Because saponin-mediated cell permeabilization is a reversible process, it is important to keep the cells in the presence of saponin during intracellular cytokine staining.
   b. Wash cells 2 times with saponin-containing buffer (e.g, BD Perm/Wash™ buffer) and resuspend in staining buffer prior to flow cytometric analysis.

Note: Both the BD Cytofix/Cytoperm™ (Cat. No. 554722) and BD Perm/Wash buffer (Cat. No. 554723) are included in the Fixation/Permeabilization Solution Kit (Cat. No. 554714) as well as the Fixation/Permeabilization Solution Kit with BD GolgiStop (containing monensin); Cat. No. 554715) and Fixation/Permeabilization Solution Kit with GolgiPlug (containing brefeldin A); Cat. No. 555028).
Danger

BD Cytofix/Cytoperm™ Buffer (Fixation and Permeabilization Solution) contains 4.2% formaldehyde (w/w).

Hazard statements

Harmful if inhaled.
Causes skin irritation.
Causes serious eye damage.
May cause an allergic skin reaction.
Suspected of causing genetic defects.
May cause cancer. Route of exposure: Inhalative.
May cause respiratory irritation.

Precautionary statements

Wear protective clothing / eye protection.
Wear protective gloves.
Do not breathe mist/vapours/spray.
IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do.
Continue rinsing.
If skin irritation or rash occurs: Get medical advice/attention.

Suggested Companion Products

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<thead>
<tr>
<th>Catalog Number</th>
<th>Name</th>
<th>Size</th>
<th>Clone</th>
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<tbody>
<tr>
<td>554723</td>
<td>Perm/Wash Buffer</td>
<td>100 mL</td>
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<tr>
<td>554714</td>
<td>BD Cytofix/Cytoperm™ Fixation/Permeabilization Kit</td>
<td>250 Tests</td>
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<tr>
<td>554715</td>
<td>BD Cytofix/Cytoperm Plus Kit (with BD GolgiStop)</td>
<td>250 Tests</td>
<td>(none)</td>
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<tr>
<td>555028</td>
<td>BD Cytofix/Cytoperm Plus Kit (with BD GolgiPlug)</td>
<td>250 Tests</td>
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Product Notices


References


