Flow Cytometry Fixation/Permeabilization Kit

B1511174

Storage 2-8℃. Do not freeze.

Shipping Ship at low temperature. Upon receipt, store immediately at the recommended storage conditions.

Product Introduction

Flow Cytometry Fixation/Permeabilization Kit contains optimally formulated buffers for fixation and permeabilization of cells for immunofluorescence staining of intracellular antigens for analysis by flow cytometry.

Product Components

B1511174	Components	50 T	100 T	500 T	Storage
B1511174A	Flow Cytometry Fixation Buffer	5 mL	10 mL	50 mL	2-8℃
B1511174B	Flow Cytometry Permeabilization Buffer	5 mL	10 mL	50 mL	2-8℃

Usage Protocol

1. Pellet cells by centrifuging at 400g for 5 minutes and resuspend in PBS at a density of 10^7 cells/mL.
2. Aliquot 100 µL of cell suspension (10^6 cells) per tube into 96 well plate.
3. Optional: Staining for surface antigens.

a. Dilute the appropriate amount of surface antigen antibody(s) in PBS.
b. Add 100 µL of the antibody solution to each well.
c. Incubate for 15 minutes at room temperature.
d. Wash cells twice with 200 µL PBS as described in step 1.
e. Resuspend cells in 100 µL PBS.

4. Add 100 µL of Flow Cytometry Fixation Buffer to each sample and vortex gently to mix.
5. Incubate for 20 minutes at room temperature.
6. Centrifuge for 5 minutes at 400g. Wash cells twice by resuspending in 200 µL PBS, then centrifuge for 5 minutes at 400g, and gently pour off supernatant.
7. Add 100 µL of Flow Cytometry Permeabilization Buffer to each sample and vortex gently to mix.
8. Incubate for 10 minutes at room temperature.
9. Wash cells twice with PBS as described in step 6.

10. Dilute the appropriate amount of primary antibody in PBS and vortex gently to mix.
11. Add 100 µL of the primary antibody solution to each tube.
12. Incubate samples at room temperature for 30 minutes.
13. Wash cells twice with PBS as described in step 6. Note: If staining with fluorescently-labeled primary antibodies, add 200 µL PBS and analyze by flow cytometry. If staining with unconjugated primary antibodies and fluorescently-labeled secondary antibodies, proceed to step 14.
14. Dilute the appropriate amount of fluorescent secondary antibody in PBS and vortex gently to mix.
15. Add 100 µL of the secondary antibody solution to each tube.
16. Incubate for 30 minutes at room temperature in the dark.
17. Wash cells twice with PBS as described in step 6.
18. Resuspend cell pellet in 200 µL PBS and analyze by flow cytometry.

Precautions

1. This product contains formaldehyde. Personal protective measures should be taken during use, such as wearing gloves and a lab coat, to avoid contact with eyes or skin.
2. During incubation, ensure that the liquid covers the cells and that the cells do not adhere to the tube wall, to prevent insufficient cell processing that may lead to a negative signal population.

Results Presentation

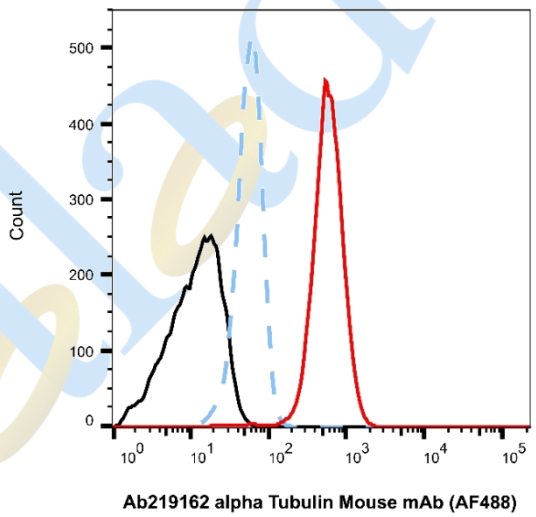

Figure1 Jurkat cells were fixed and permeabilized with Flow Cytometry Fixation/Permeabilization Kit (B1511174) and then stained with alpha Tubulin Mouse mAb

(AF488) (Ab219162) (red) at for 30 minutes at 4°C in the dark. Blue - Mouse IgG (AF488) (Ab190245). Black - Unlabelled control, cells without incubation with primary antibody.