Datasheet: BUF076A

Description:	CELL STIMULATION REAGENT (WITHOUT BREFELDIN A) (500X)
Name:	CELL STIMULATION REAGENT (WITHOUT BREFELDIN A)
Format:	Reagent
Product Type:	Accessory Reagent
Quantity:	0.1 ml

Product Details

Applications

This product has been reported to work in the following applications. This information is derived from testing within our laboratories, peer-reviewed publications or personal communications from the originators. Please refer to references indicated for further information. For general protocol recommendations, please visit www.bio-rad-antibodies.com/protocols.

	Yes	No	Not Determined	Suggested Dilution
Flow Cytometry	▪			

Where this product has not been tested for use in a particular technique this does not necessarily exclude its use in such procedures. Suggested working dilutions are given as a guide only. It is recommended that the user titrates the product for use in their own system using appropriate negative/positive controls.

Product Form

500x concentrate - liquid

Preparation

500x solution contains: Phorbol-12-myristate-13-acetate (PMA) 40.5 µM and Ionomycin 669.3 µM in DMSO

Product Information

Cell Stimulation Reagent (without Brefeldin A) 500x is a mixture of Phorbol-12-myristate-13-acetate (PMA) and ionomycin. PMA activates protein kinase C and ionomycin acts as a $Ca2^{+}$ ionophore allowing movement of $Ca2^{+}$ across membranes. Activation of cells with PMA and ionomycin leads to an increase in surface expression of certain markers such as CD69 and CD154 and the production of cytokines such as IL-2, IFN-gamma and TNF-alpha.

Instructions For Use

1. Resuspend 1 $x10^{6}$ - 2 $x10^{6}$ cells per ml in cell culture media
2. Thaw the Cell Stimulation Reagent (without Brefeldin A) 500x (BUF076A) in a 37°C water bath
3. To each ml of cell suspension add 2 µl of BUF076A and incubate cells in a CO_2, 37°C incubator for 6 hours
4. Harvest cells and centrifuge at 300-400 g for 5 minutes. Discard supernatant
5. To wash the pellet, add 10-20 ml of Cell Staining Buffer (BUF073), vortex to resuspend the pellet then centrifuge at 300-400 g for 5 minutes. Discard supernatant and repeat step 5 for a second time
6. Stain for surface markers or proteins of interest and analyze cells using flow cytometry

Note: Aliquot product to avoid repeat freeze-thawing. Time and culture conditions may need to be optimized. To detect intracellular staining following activation, the Cell Activation Reagent (without Brefeldin A) 500x (BUF076A) can be used with Monensin Solution (BUF074) or Brefeldin A Solution 1000x (BUF075).

Storage

Store at -70°C.

	Storage in frost-free freezers is not recommended. This product should be stored undiluted. This product is photosensitive and should be protected from light.
Shelf Life	Please see label for expiry date.
Health And Safety Information	Material Safety Datasheet documentation #20387 available at: 20387: https://www.bio-rad-antibodies.com/uploads/MSDS/20387.pdf
Regulatory	For research purposes only

Related Products

Recommended Useful Reagents

CELL STIMULATION REAGENT (WITH BREFELDIN A) (500X) (BUF077A)
MONENSIN SOLUTION (1000X) (BUF074)
BREFELDIN A SOLUTION (1000X) (BUF075)

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