

Datasheet: BUF077A

BATCH NUMBER 172356

Description:	CELL STIMULATION REAGENT (WITH BREFELDIN A) (500X)
Name:	CELL STIMULATION REAGENT (WITH 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: PMA (phorbol-12-myristate 13-acetate) 40.5 µM, Ionomycin 669.3 µM and Brefeldin A 2.5 mg/ml in DMSO

Product Information **Cell Stimulation Reagent (with Brefeldin A) 500x** is a mixture of PMA (phorbol-12-myristate-13-acetate), ionomycin and the protein transport inhibitor Brefeldin A. PMA and ionomycin activate cells and increase cytokine production. Brefeldin A allows for these cytokines to accumulate in the rough endoplasmic reticulum and golgi apparatus allowing for their detection by intracellular fluorescent staining.

- Instructions For Use**
1. Resuspend 1×10^6 - 2×10^6 cells per ml in cell culture media
 2. Thaw the Cell Stimulation Reagent (with Brefeldin A) 500x (BUF077A) in a 37°C water bath
 3. To each ml of cell suspension add 2 µl of BUF077A and incubate cells in a CO₂, 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 by flow cytometry

Note: Aliquot product to avoid repeat freeze-thawing. Time and culture conditions may need to be optimized.

Activation of cells for >12 hours with Brefeldin A may affect cell viability. In these circumstances it is recommended to use Cell Stimulation Reagent (without Brefeldin A) 500x and then add Brefeldin A Solution 1000x ([BUF075](#)) or Monensin Solution ([BUF074](#)).

References

1. Flores-Villalva, S. *et al.* (2025) Polyfunctionality of CD4(+) T lymphocytes in buffaloes and cattle: comparative antigen-specific cytokine responses in bovine tuberculosis infection. [Front Immunol. 16: 1608065.](#)

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.

Guarantee

Guaranteed until date of expiry. Please see product label.

Health And Safety Information

Material Safety Datasheet documentation #20388 available at: <https://www.bio-rad-antibodies.com/SDS/BUF077A>

Regulatory

For research purposes only

Related Products

Recommended Useful Reagents

[BREFELDIN A SOLUTION \(1000X\) \(BUF075\)](#)

[MONENSIN SOLUTION \(1000X\) \(BUF074\)](#)

Product inquiries: www.bio-rad-antibodies.com/technical-support

To find a batch/lot specific datasheet for this product, please use our online search tool at: bio-rad-antibodies.com/datasheets

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