

Broad Spectrum Phosphatase Inhibitor Cocktail

Cat#: orb76033 (User Manual)

Properties

Form Supplied	100X concentrated stock solution
Physical State	Liquid, colorless to light yellow
Pack Size	1 ml
Recommended working concentration	100-fold dilution in tissue lysis buffer; Use 10 μ l of the Phosphatase Inhibitor Cocktail solution to inhibit dephosphorylation of proteins for 1 ml of lysate
Compatibility with reagents	Fully compatible with cell lysis buffers and Broad Spectrum Protease Inhibitor Cocktail
Compatibility with assays	Compatible with IEF/2D studies; MS-compatible: not contain AEBSF; Compatible with immobilized metal chelate affinity chromatography and 2D gel electrophoresis inhibitor components dialysis removal or desalting;
Storage	Store at -20°C for one year, at 4°C for two months
Precautions	FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC AND CLINICAL USE

Description

Broad Spectrum Phosphatase Inhibitor Cocktail is a Western blot related concentrated stock solution reagent containing a mixture of different phosphatase inhibitors that is to be added to cell lysis buffer to protect native phosphoproteins from dephosphorylation during proteins purification and sample preparation used in WB, Co-IP, ChIP, and protein kinase assays.

Application	WB sample preparation, protein purification, Co-IP, protein kinase activity assay experiments
Pack Size	1 mL
Reagent Type	Western Blotting related reagent; Inhibitors
Usage	To preserve phosphorylation state and protein functionality following cell lysis
Content	Proprietary mix of: Sodium fluoride, Sodium orthovanadium, Imidazole, Sodium molybdate, Sodium sulphate, Sodium pyrophosphate, B-phosphoric acid glycerol
Target Specificity	Tyrosine phosphatase, acidic and alkaline phosphatase; Serine/threonine phosphatase, histidine phosphatase, etc.
Target Sample	Cell lysis extracts

BioChemicals Information

Inhibitor	Inhibition Specificity
Sodium fluoride	Ser/Thr and acid phosphatases
Sodium orthovanadium	ATPases
	Tyr and alkaline phosphatases
Imidazole	Alkaline phosphatases
Sodium molybdate	Acid and phosphoprotein phosphatases
Sodium tartrate	Acid phosphatase
Sodium pyrophosphate	Ser/Thr phosphatases
B-phosphoric acid glycerol	Ser/Thr phosphatases

Usage and Handling

The product is supplied as a 100X concentrated stock solution in a liquid format for improved accuracy, solubility, and ease of use in comparison to traditional tablets. Since phosphatase levels may vary among cell and tissue types, it may be necessary to increase the concentration of inhibitors.

Background

Phosphatase inhibitors are used when phosphorylation (activation) states of target proteins need to be studied and the phosphorylated residues of interest must remain intact. They are chemicals that aid in the extraction of intact proteins in their native modification state by inhibiting endogenous phosphatases that would otherwise dephosphorylate the proteins present in cell lysates and tissue extracts. Broad Spectrum Phosphatase Inhibitor Cocktail contains individual components with specific inhibitory properties to provide an all-around protection of the protein phosphorylation state. The six phosphatase inhibitors included in this mixture target a broad spectrum phosphatase categories. Dynamic protein phosphorylation is a key cellular signaling mechanism for cell processes regulation. When tissues are lysed to make whole cell extracts, the loss of natural compartmentalization causes normal regulation of cellular signaling to get distorted, and resident cell phosphatases within the cell extract are free to disorderly dephosphorylate proteins. The usual consequence of this unregulated state is biologically meaningless representation of protein activities (i.e. phosphorylation status) and false negative staining in anti-phosphoprotein immunostaining analyses. The addition of phosphatase inhibitors to the cell lysis buffer aids in the preservation of phosphorylated residues at the time of cell disruption.

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