

Halt™ Protease and Phosphatase Inhibitor Cocktail

78440 78444 78446

2053.0

Number	Description
78440	Halt Protease and Phosphatase Inhibitor Cocktail , sufficient for 100 ml of sample Contents: Protease and Phosphatase Inhibitor Cocktail (100X) , 1 ml EDTA Solution, 0.5 M (100X) , 1 ml
78444	Halt Protease and Phosphatase Inhibitor Cocktail , sufficient for 500 ml of sample Contents: Protease and Phosphatase Inhibitor Cocktail (100X) , 5 × 1 ml EDTA Solution, 0.5 M (100X) , 5 × 1 ml
78446	Halt Protease and Phosphatase Inhibitor Cocktail , sufficient for 1 L of sample Contents: Protease and Phosphatase Inhibitor Cocktail (100X) , 10 ml EDTA Solution, 0.5 M (100X) , 10 ml

Storage: Upon receipt store at 4°C. Product shipped with an ice pack. Do not freeze product.

Introduction

The Halt Protease and Phosphatase Inhibitor Cocktail protects proteins from degradation by endogenous proteases and phosphatases released during protein extraction and purification. The ready-to-use cocktail contains a mixture of several potent inhibitors (Table 1). The protease inhibitors target aminopeptidases, cysteine and serine proteases. The phosphatase inhibitors target serine/threonine and protein tyrosine phosphatases.

Table 1. Protease and phosphatase inhibitors included in the cocktail.

Inhibitor	Target Protease/Phosphatase
Aprotinin	Serine Proteases
Bestatin	Aminopeptidase Proteases
E-64	Cysteine Proteases
Leupeptin	Serine and Cysteine Proteases
Sodium Fluoride	Serine and Threonine Phosphatases
Sodium Orthovanadate	Tyrosine Phosphatases
Sodium Pyrophosphate	Serine and Threonine Phosphatases
β-glycerophosphate	Serine and Threonine Phosphatases
EDTA*	Metalloproteases

*Provided in a separate vial.

Important Product Information

- This inhibitor cocktail is generally effective when used at a 1X final concentration; however, samples that contain high levels of proteases or phosphatases might require a more concentrated treatment (i.e., 2-3X).
- EDTA inhibits metalloproteases by chelating divalent cations necessary for their activity. By this same mechanism, EDTA might affect the activities of other proteins. Empirically determine if EDTA is beneficial for each experiment.
- This inhibitor cocktail interferes with immobilized metal-chelate affinity chromatography (IMAC) and 2-D gel electrophoresis. Either dialyze or desalt (see Related Products Section) sample to effectively remove inhibitors from sample extracts before performing such procedures.
- Pepstatin A is not included in the cocktail formulation. If you need to inhibit acid proteases, purchase Pepstatin A (see Related Products) and add it to the cocktail.

Procedure

1. Vortex the bottle before use to ensure a homogeneous suspension.
2. Immediately before use, add the cocktail at 10 µl/ml directly to the cell lysis buffer or extract to produce a 1X final concentration.
3. Optional: To inhibit metalloproteases, add EDTA at 10 µl/ml of lysis buffer or extract to achieve a 1X (5 mM) final working concentration.

Troubleshooting

Problem	Possible Cause	Solution
Poor inhibition of protease or phosphatase activity	Sample contains high levels of proteases or phosphatases	Add sufficient cocktail to produce a 2X or 3X final concentration
Cocktail does not provide sufficient protection against a particular protease family	Inhibitor for a specific family is not a component of the cocktail	Add individual inhibitors to the samples for the specific protease
	A specific inhibitor is not at the needed concentration	Use the cocktail at a higher concentration

Related Products

66380 Slide-A-Lyzer® Dialysis Cassette, 3-12 ml, 8 each*

89882 Zeba™ Desalt Spin Columns, 0.5 ml, 25 columns*

78431 AEBSF, 100 mg

78436 Pepstatin A, 25 mg

*Please see the catalog or website for a complete list of Slide-A-Lyzer and Zeba Products.

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Current versions of product instructions are available at www.thermo.com/pierce. For a faxed copy, call 800-874-3723 or contact your local distributor.

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