Protocol



TD-P Revision 2.1

Creation Date: 6/29/2015 Revision Date: 11/16/2023

ProBlock™ Gold Yeast/Fungi

Protease Inhibitor Cocktail Specific for Yeast and Fungi

Introduction

ProBlock™ Gold Yeast/Fungi is an easy-to-use protease inhibitor cocktail specifically developed for protein purification from yeast and fungi. *ProBlock™ Gold Yeast/Fungi* contains optimized concentrations of both reversible and irreversible protease inhibitors to inhibit fungal serine, cysteine, aspartic and metalloproteases.

Since some proteins require divalent cations like Ca^{2+} , Mg^{2+} or Mn^{2+} for their biological activity, the presence of EDTA may be detrimental to the protein activity. Furthermore, if the protein of interest is purified using immobilized metal chelate affinity chromatography (IMAC), EDTA must be removed from the buffer before the chromatography. The *ProBlock Gold Yeast/Fungi* is therefore supplied with an optional EDTA solution and which may be added in the extraction buffer or lysate as needed.

ProBlock™ Gold Yeast/Fungi is a concentrated solution that prevents the proteolytic degradation of yeast and fungal proteins from lysed cells in vitro. ProBlock™ Gold Yeast/Fungi inhibits over 95% of protease activities at 1X concentration (pH 7-8) in extraction buffer.

Items Included

- ProBlock™ Gold Yeast/Fungi [100X] (GoldBio Catalog #GB-333)
- 0.5M EDTA

Storage Conditions

Shipped at ambient temperature. Upon arrival, store it refrigerated at 4°C. If stored properly, this product should be stable for at least 2 years from the date of receipt.

Method

- 1. Allow solution to warm to room temperature. The solution is in suspension form, vortex the vial before removing the solution.
- 2. Add *ProBlock™ Gold Yeast/Fungi* 10 µl/ml directly in an appropriate volume of extraction buffer or protein extract to 1X final concentration. For more potent protease inhibition, add *ProBlock™ Gold Yeast/Fungi* 20-30 µl/ml to give 2-3X final concentration.



Gold Biotechnology/ FM-000008 ProBlock™ Gold Yeast/Fungi

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* When ProBlock™ Gold Yeasts/Fungi is added to the buffer or extract, it is stable for 1-2 weeks at 4°C and 4-6 weeks at -20°C.

3. Mix solution thoroughly.

Note: (OPTIONAL). For inhibition of metalloproteases (if the buffer does not contain EDTA), add 0.5M EDTA 10 μ l/ml directly in an appropriate volume of extraction buffer or extract to 1X final concentration.

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