Protocol

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ProBlock™ Gold Plant

Protease Inhibitor Cocktail Specific for Plants

Introduction

ProBlock™ Gold Plant is an easy-to-use protease inhibitor cocktail specifically developed for protein purification from plant cells and tissues. *ProBlock™ Gold Plant* contains optimized concentrations of both reversible and irreversible protease inhibitors to inhibit plant serine, cysteine, and metallo- proteases. It also contains specific inhibitors for plant proteases, such as aspartic proteases and aminopeptidases.

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 Plant* is therefore supplied with an optional EDTA solution and which may be added in the extraction buffer or lysate as needed.

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

Items Included

- ProBlock™ Gold Plant [100X] (GoldBio Catalog # GB-332)
- 0.5M EDTA

Storage Conditions

Shipped at ambient temperature. Upon arrival, store it refrigerated at 4°C. If stored properly, it is stable for 1 year*.

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 Plant* 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 Plant* 20-30 µl/ml to give 2-3X final concentration.

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**** When* ProBlock™ Gold Plant *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 µl/ml directly in an appropriate volume of extraction buffer or extract to 1X final concentration.