Protocol

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ProBlock™ Gold

For Complete Inhibition of Protease Activities

Introduction

ProBlock™ Gold is an easy-to-use, general protease inhibitor cocktail. Due to the optimized concentration of the various inhibitors, the *ProBlock™ Gold* shows excellent, broad-spectrum inhibition of protease activities and is therefore suitable for the protection of proteins extracted from animal cells/tissues, plants, yeast and bacteria etc. *ProBlock™ Gold* contains both irreversible and reversible protease inhibitors and inhibits serine, cysteine 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™* is therefore supplied with an optional EDTA solution and which may be added in the extraction buffer or lysate as needed. An alternative metalloprotease inhibitor is used in the actual *ProBlock™ Gold* cocktail, which allows optimal action of nuclease activity for removing nucleic acids from the samples.

ProBlock™ Gold inhibits over 95% of protease activities at 1X concentration (pH 7-8) in extraction buffer.

Items Included

- ProBlock™ Gold [100X] (GoldBio Catalog #GB-108)
- 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* 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 20-30 µl/ml to give 2-3X final concentration.

**** When* ProBlock™ Gold *is added to the buffer or extract, it is stable for up to week at 4°C and 4-6 weeks at -20°C.***

3. Mix 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.