G-Biosciences ♦ 1-800-628-7730 ♦ 1-314-991-6034 ♦ technical@GBiosciences.com

A Geno Technology, Inc. (USA) brand name

Protease Screening™

For Testing for Proteases in Protein Samples

(Cat. # 786-137)

INTRODUCTION

Sensitive protease screening is needed to study protease activity present in the sample of interest. Protease screening kit is a simple and quick method for testing the presence of proteases in the protein samples. The screening kit uses dye- labeled protein substrate. The proteases present in the solution will digest the protein substrate and release dye labeled peptides. The absorbance of dye-labeled peptide is measured at 570nm for determination of protease activity. The kit is sufficient for 50 assays in a microtiter format.

ITEM(S) SUPPLIED (Cat. # 786-137)

Description	Size
Incubation Buffer	5ml
Precipitation Agent	5ml
Assay Buffer	6ml
Protease Substrate (lyophilized)	1vial (150µl)

STORAGE CONDITIONS

The kit is shipped at ambient temperature. Store Protease Screening kit at 4°C. When stored and used properly the kit is stable for 1 year.

ADDITIONAL ITEM(S) REQUIRED

96 well titer plates, microfuge tubes, plate reader, spectrophotometer, centrifuge, and micro-plate reader.

PREPARATION BEFORE USE

Substrate

Reconstitute the supplied Protease Substrate (dye-labeled protein) by adding 150µl water into the vial, mix it to dissolve completely. After reconstitution, store the substrate at -20°C and is stable for 3-4 months.

PROTOCOL

NOTE: *The reaction set up is designed for 96 well titer plates. The use of 96 well titer plates requires a centrifuge adapted for 96 well titer plates. Alternatively, the assay may be performed in micro-centrifuge tubes and the final reaction product is transferred to 96 well titer plates for the measurement of optical density.*

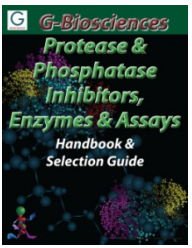
1. Set up the reaction as indicated in the table below. We recommend performing each assay in triplicate:

Blank	**Blank**	**Test Sample**
Test Sample	-	1-45µl
Protease Substrate Solution	2.5µl	2.5µl
Incubation Buffer or buffer of your choice	to 50µl	to 50µl

2. Seal the plate or tube and incubate at 37°C for 2-3 hours
   NOTE: *For slow acting proteases, incubation time may be extended up to 24 hours.*
3. After incubation, add 50µl Precipitation Agent, mix the contents and incubate again at 37°C for 10 minutes.
4. Centrifuge the tubes at 12,000xg for 5 minutes or centrifuge 96 well titer plate at 2,000-4,000xg for 15 minutes.
5. Transfer the supernatant to clean tubes or 96-well plate. Add 120µl Assay Buffer in each tube/well and mix. A pink color will develop instantly.
6. If in tubes, transfer to a 96-well plate or micro-cuvette.
7. Read the reaction color at 570nm against the blank. The intensity of the color developed in each tube is proportional to protease activity.
   NOTE: *The reaction product may be diluted with water to 1ml and read in 1 ml cuvettes, however, the sensitivity will drop by 4-5 fold. Alternatively, the reaction volume may be increased proportionally to give total reaction volume 1ml.*

RELATED PRODUCTS

Download our Protease & Phosphatase Inhibitors, Enzymes & Assays Handbook.



http://info.gbiosciences.com/protease-phosphatase-inhibitors-enzymes-assay-handbook

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