

Catalog #	Aliquot Size
T565-31AN-02	0.2 mg
T565-31AN-1	1 mg
T565-31AN-5	5 mg
T565-31AN-25	25 mg

## Immobilized Thrombin, Agarose

Native Human protein derived from human plasma

**Catalog # T565-31AN**

Lot # L2158-5

### Product Description

Native human thrombin was derived from human plasma.

Matrix: highly cross-linked agarose

Average Particle Size: 90  $\mu$ m

Ligand Density: 1 mg protein / ml matrix

Maximum Flow Rate, Pressure: 200 cm/h, 1 bar

### Gene Aliases

F2; PT; RPRGL2; THPH; Coagulation Factor II; Factor IIa

### Storage and Stability

Store product at 2 - 4°C for up to 6 months.

### Digestion Conditions

Catalytic pH Range: 6 ~ 9

Catalytic Temperature Range: 20 ~ 37°C

Catalytic Time Range: 1 ~ 16h

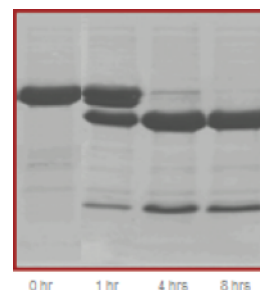
### Scientific Background

Human thrombin derived from human plasma is an endolytic serine protease that has a molecular weight of 36 KDa. Thrombin specifically recognizes the consensus sequence Leu-Val-Pro-Arg- $\blacktriangledown$ -Gly-Ser. Thrombin has high proteolytic specificity and hydrolytic efficiency, making it a widely used tool for cleaving purification tag from recombinant fusion proteins.

Immobilized human thrombin is covalently cross-linked to an agarose matrix circumventing the need for enzyme removal after cleavage. The resin can be reused 10-20 times after regenerating when proper storage conditions are followed.

### Specific Activity

Sample Data



SDS-PAGE image of a DsbA fusion protein cut with 1  $\mu$ g/ $\mu$ l thrombin agarose at 37°C.

## Immobilized Thrombin, Agarose

Native Human protein derived from human plasma

Catalog Number	T565-31AN
Lot #	L2158-5
Format	Stored in 50% glycerol
Storage & Shipping	Store product at 2 - 4°C for up to 6 months

To place your order, please contact us by phone 1-(604)-232-4600, fax 1-604-232-4601 or by email: [orders@signalchem.com](mailto:orders@signalchem.com)  
[www.signalchem.com](http://www.signalchem.com)

**FOR IN VITRO RESEARCH PURPOSES ONLY. NOT INTENDED FOR USE IN HUMAN OR ANIMALS.**

# Assay Protocol

## Reaction Components

### Active Protease (Catalog #: T565-31AN)

Immobilized human thrombin, Agarose.

### Digestion Buffer (User Prepared)

50 mM Tris-HCl pH 8.0

## Reaction Protocol

The following conditions may be different for different proteins. Optimize the protocol for each specific protein. Protein of interest must be purified prior to digestion and dialyzed with digestion buffer.

- Step 1.** Prepare protein sample in digestion buffer at the concentration range of 1-10 mg/ml
- Step 2.** Gently resuspend resin and aliquot 15  $\mu$ l of the uniformed resin to a microfuge tube
- Step 3.** Spin tube at 500 x g and remove supernatant
- Step 4.** Add 500  $\mu$ l of digestion buffer and resuspend
- Step 5.** Spin tube at 500 x g and remove supernatant
- Step 6.** Repeat Steps 4 and 5 twice
- Step 7.** Resuspend the resin with 200  $\mu$ l of digestion buffer
- Step 8.** Add 800  $\mu$ l of protein sample to the resin and mix well
- Step 9.** Incubate reactions in a platform shaker at room temperature for 1-16 hours
- Step 10.** Monitor the cleavage products by SDS-PAGE or HPLC
- Step 11.** Elute protein via one of two methods when desired reaction is achieved
  - a. Centrifugation at 500 x g for 3 minutes and collect supernatant; or
  - b. Transfer the contents of the tube to a column and collect eluate
- Step 12.** In both methods above, a second elution can be performed with 200  $\mu$ l of digestion buffer
- Step 13.** To regenerate resin:
  - a. Wash the resin with 1 ml of deionized water
  - b. Repeat step a twice
  - c. Resuspend the resin in 50% glycerol with 50 mM Tris-HCl, pH 8
  - d. Store at 2-4°C.

To place your order, please contact us by phone 1-(604)-232-4600, fax 1-604-232-4601 or by email: [orders@signalchem.com](mailto:orders@signalchem.com)  
[www.signalchem.com](http://www.signalchem.com)

**FOR IN VITRO RESEARCH PURPOSES ONLY. NOT INTENDED FOR USE IN HUMAN OR ANIMALS.**