



3050 Spruce Street  
Saint Louis, Missouri 63103 USA  
Telephone 800-325-5832 • (314) 771-5765  
Fax (314) 286-7828  
email: techserv@sial.com  
sigma-aldrich.com

## Product Information

### Bromelain from pineapple stem

Product Number **B4882**  
Storage Temperature -20 °C

#### Product Description

Enzyme Commission (EC) Number: 3.4.22.32  
CAS Number: 37189-34-7  
pI: 9.55<sup>1</sup>

Bromelain is a cysteine endopeptidase with broad specificity for cleavage of proteins. Stem source bromelain should not be confused with fruit source bromelain, which is an aspartic endopeptidase.

pGlu-Phe-Leu-p-nitroanilide (Product No. P 3169) is a chromogenic substrate for the assay of thiol proteinases such bromelain (also papain and ficin). Bromelain has a  $K_M$  of 0.30 mM with this substrate. Initial rates of hydrolysis can be measured spectrophotometrically at 410 nm using the molar extinction coefficient of p-nitroaniline ( $E^M = 9,800$ ) at 410 nm.<sup>2</sup>

Bromelain impacts several important biological functions. It inhibits the biosynthesis of proinflammatory prostaglandins.<sup>3</sup> It can reduce clotting efficiency by affecting fibrinogen.<sup>4</sup>

#### Precautions and Disclaimer

For Laboratory Use Only. Not for drug, household or other uses.

#### Preparation Instructions

This product may be suspended in acetate buffer, pH 4.5 (1 mg/ml), yielding a hazy, off-white suspension.

#### Procedure

An assay of bromelain using the general thiol protease chromogenic substrate, pGlu-Phe-Leu-p-Nitroanilide is:

1. Prepare a reaction buffer of 2.5 ml of 0.1 M phosphate buffer, pH 6.5, containing 0.3 M potassium chloride, 0.1 mM EDTA, and 3 mM dithioerythritol.
2. Add 0.3 ml of 1-5 mM substrate in dimethyl sulfoxide.
3. Incubate 5 minutes at 37 °C.
4. Add 0.2 ml of a 133 µg/ml solution of bromelain in water.
5. Incubate at 37 °C until the desired color intensity is achieved.
6. The reaction can be stopped by the addition of 0.2 ml of 3 N hydrochloric acid.
7. Read the result spectrophotometrically at 410 nm.

#### References

1. Biochem., **3**, 48-55 (1964).
2. Filippova, I.Y. et al., L-Pyroglutamyl-L-phenylalanyl-L-leucine-p-nitroanilide--a chromogenic substrate for thiol proteinase assay. Anal. Biochem., **143(2)**, 293-297 (1984).
3. Taussig, S.J., The mechanism of the physiological action of bromelain. Medical Hypotheses, **6(1)**, 99-104 (1980).
4. Livio, M. et al., Drugs, Exptl. Clin. Res., **4(1)**, 49-53 (1978).

MWM/NSB 5/07

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