

## Product Information

### PROTEASE INHIBITOR COCKTAIL

For Plant Cell Extracts

Product Number **P 9599**

Storage Temperature  $-20\text{ }^{\circ}\text{C}$

#### Product Description

Crude cell extracts contain a number of endogenous enzymes, such as proteases and phosphatases, which are capable of degrading the proteins present in the extract. The best way to improve the yield of intact proteins is to add inhibitors of these enzymes known to be present in the source material. This protease inhibitor cocktail has been optimized and tested for plant cell extract use.

This is a mixture of protease inhibitors with a broad specificity for the inhibition of serine, cysteine, aspartic, metalloproteases, and aminopeptidases

#### Preparation Instructions

The cocktail is supplied as a clear, faint pink solution in dimethyl sulfoxide (DMSO).

#### Storage/Stability

Store cocktail at  $-20\text{ }^{\circ}\text{C}$ . The product is stable for two years as supplied.

#### Recommended Usage

One ml of the cocktail solution is recommended for the inhibition of protease activity found in 100 ml of cell lysate from 30 g (wet weight) of various plant tissues. Whole extracts of plant seedlings from pea (*Pisum sativum*), bean (*Phaseolus vulgaris*), wheat (*Triticum aestivum*), tobacco (*Nicotiana tobaccum*), and arabidopsis (*Arabidopsis thaliana*) have been tested. Extracts of leaves or roots from pea, wheat, and tobacco have also been tested. Since not all organisms contain the same level of endogenous proteases, it may sometimes be necessary to increase the concentration of inhibitors.

#### Components

The individual components of this proprietary formulation have specific inhibitory properties. A description of each inhibitor is given below.

AEBSF (Product Code A 8456) inhibits serine proteases, such as trypsin and chymotrypsin.

1,10-Phenanthroline (Product Code P 9375) inhibits metalloproteases.

Pepstatin A (Product Code P 4265) inhibits acid proteases, such as pepsin (human or porcine), renin, cathepsin D, chymosin (bovine rennin), and protease B (*Aspergillus niger*).

Leupeptin (Product Code L 2884) inhibits both serine and cysteine proteases, such as calpain, trypsin, papain, and cathepsin B.

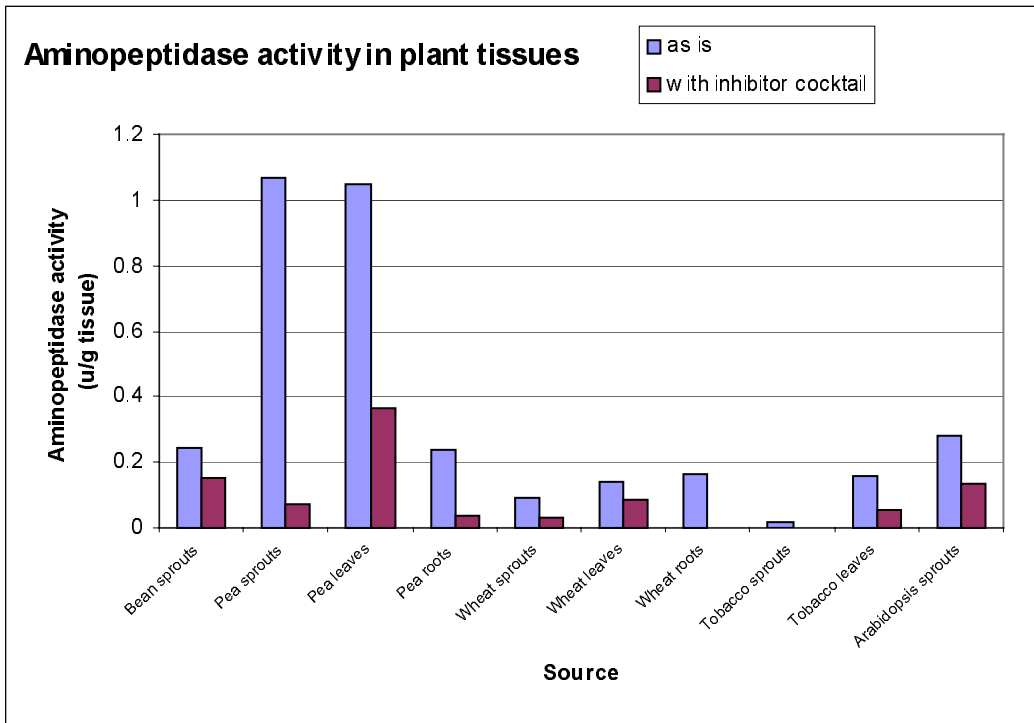
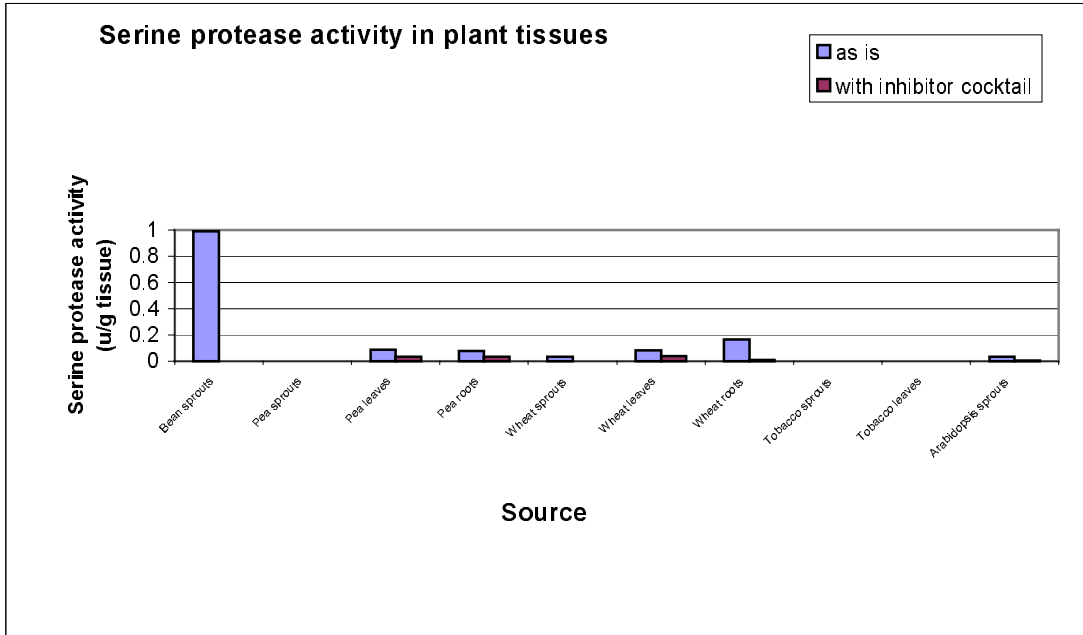
Bestatin (Product Code B 8385) inhibits aminopeptidases, such as leucine aminopeptidase and alanyl aminopeptidase.<sup>1,2,3,4</sup>

E-64 (Product Code E 3132) inhibits cysteine proteases, such as calpain, papain, cathepsin B, and cathepsin L.

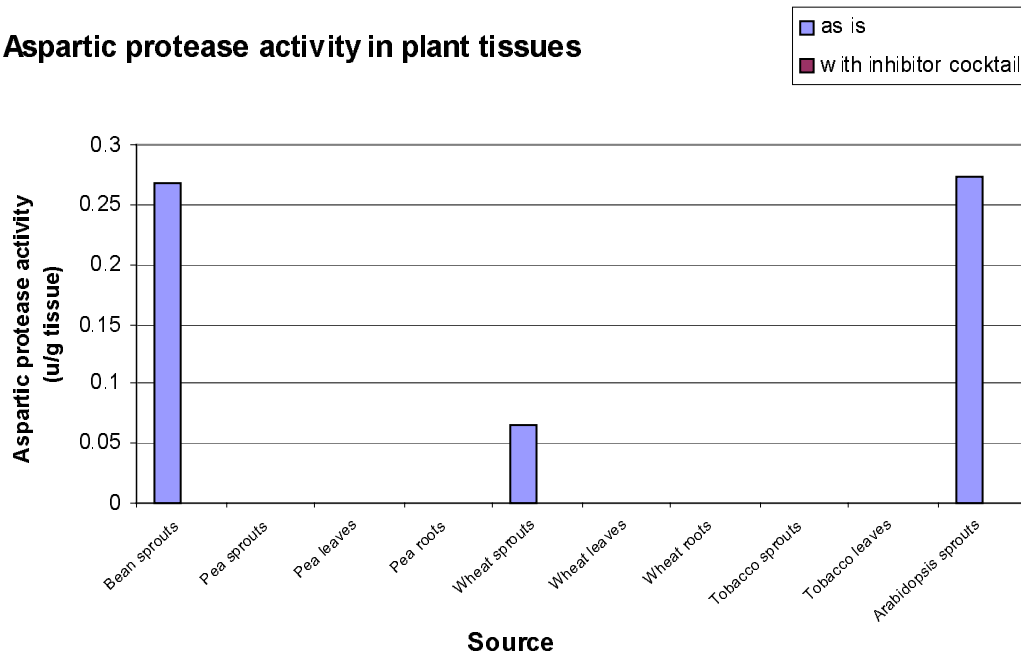
#### References:

1. Umezawa H., Ann. Rev. Microbiol., **36**, 75-99 (1982).
2. Aoyagi, T., et al, Biochem. Int., **9**, 405-411 (1984).
3. Aoyagi, T. & Umezawa, H., Acta Biol. Med. Ger., **40**, 1523-1529 (1981).
4. Mumford, R. A., et al, Biochem. Biophys. Res. Comm., **103**, 565-572 (1981).

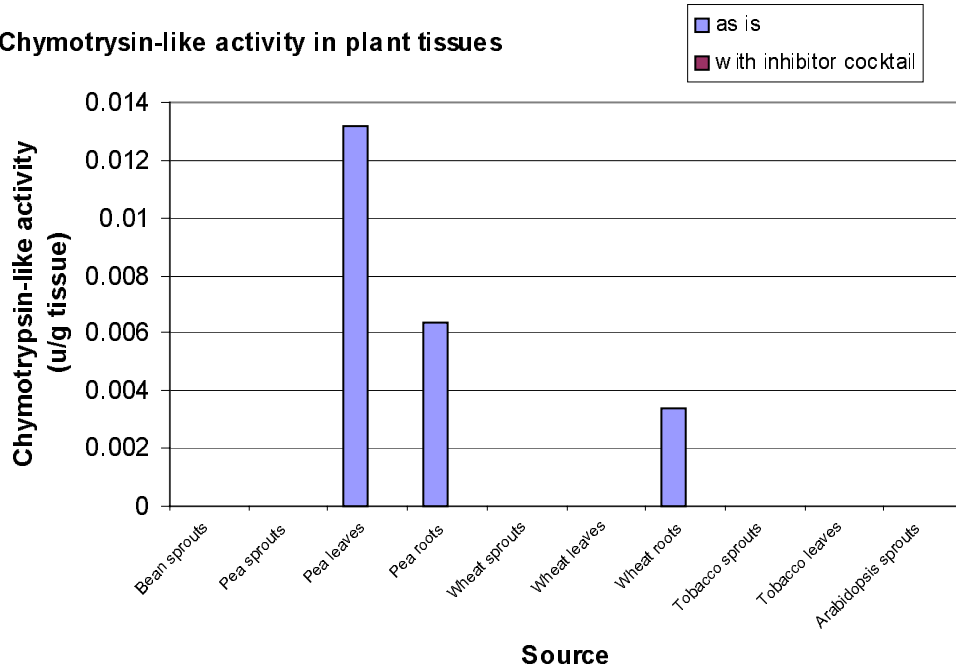
**Endogenous protease activities in plant tissues and their inhibition using Sigma cocktails**

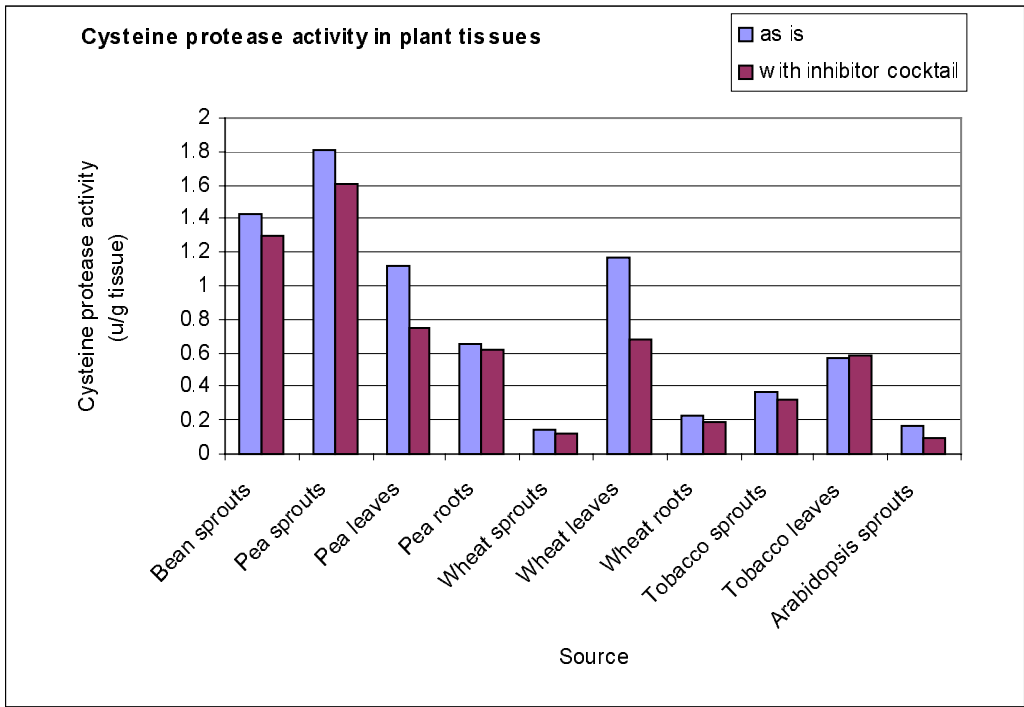


### Aspartic protease activity in plant tissues



### Chymotrypsin-like activity in plant tissues





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