Product Information

Aprotinin Bovine, Recombinant

Expressed in *Nicotiana* (tobacco), ≥5 TIU/mg protein, ≥ 98% (SDS-PAGE)

A6103

Product Description

CAS Registry Number: 9087-70-1

Synonyms: Antikrein, Antilysin(e), Basic pancreatic trypsin inhibitor (BPTI), Kallikrein-trypsin inactivator, Kunitz protease inhibitor

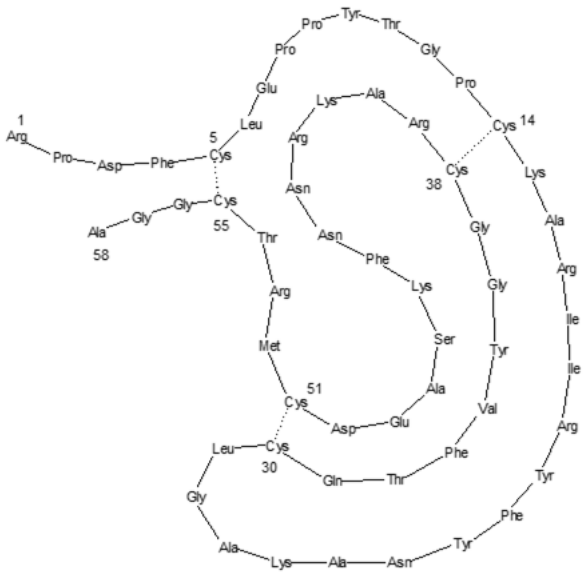
Molecular formula: $C_{284}H_{432}N_{84}O_{79}S_7$

Molecular weight: 6,512 Da

pI: 10.5

Extinction coefficient: $E^{1\%}$ = 8.4 (280 nm, water)

Aprotinin is a protein consisting of 58 amino acids, arranged in a single polypeptide chain that is cross-linked by three disulfide bridges (depicted below). Aprotinin is a competitive serine protease inhibitor that forms stable complexes with and blocks the active sites of various enzymes, such as those listed in Table 1. The binding is reversible. Most aprotinin-protease complexes dissociate at pH >10 or pH < 3.



This product is a recombinant form of the native bovine-sequence aprotinin. Native bovine aprotinin is traditionally isolated primarily from bovine lung by methods involving fractional precipitation, gel filtration, and ion exchange chromatography. Unlike animal-derived aprotinin, this recombinant aprotinin product is isolated and purified from plant tissue (by proprietary methods).

Several theses[1-5] and dissertations[6-12] have cited use of product A6103 in their protocols.

Reagent

Purity: ≥ 98% (SDS-PAGE)

Inhibitory Activity: ≥5 TIU/mg of protein

Unit Definition: One Trypsin Inhibitor Unit (TIU) will decrease the activity of 2 trypsin units by 50%, where 1 trypsin unit will hydrolyze 1.0 µmole of *N*-α-benzoyl-DL-arginine *p*-nitroanilide (BAPNA) per minute at pH 7.8 and 25 °C.

Another commonly used unit of activity is the KIU (Kallikrein Inhibitor Unit). Different conversion factors between aprotinin units have been reported:

- 1 TIU = 1,300 KIU
- 1 TIU = 1,025 KIU[14]

Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions

Aprotinin is freely soluble in water (5 mg/mL) and in aqueous buffers of low ionic strengths.[15]

Storage/Stability

Store the lyophilized powder at 2-8 °C. When stored at 2-8 °C, the product retains activity for at least 2 years.

Dilute solutions of aprotinin are generally less stable than concentrated ones. Solution stability is pH-dependent, although a pH range of 1-12 can be tolerated.[16] Repeated freeze-thaw cycles should be avoided.

The Cys^{14}-Cys^{38} disulfide bridge is readily split by reducing agents like 2-mercaptoethanol.[16] Due to its compact tertiary structure, aprotinin is relatively stable against denaturation due to high temperature, organic solvents, or proteolytic degradation (See Table 2). Only thermolysin has been found capable of degrading aprotinin after heating to 60-80 °C.[16]

The high basicity of aprotinin causes it to adhere to commonly used dialysis tubing and even gel filtration matrices. However, the use of acetylated materials and concentrated salt solutions (such as ≥ 0.1 M NaCl in buffer) minimizes this problem.[16]

Sterilization may be achieved by filtration through a 0.2 µm filter.

References

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Table 1. Inhibition by Aprotinin

Enzyme (Source), Condition	Inhibition (K_i)
Acrosin	Weak inhibition[15]
Chymotrypsin	K_i = 9 nM[17]
CMP-*N*-Acetyl-neuraminate lactosylceramide α-2,3-sialyltransferase	74% Inhibition at 300 nM[17]
Elastase (human leukocyte), pH 8.0	K_i = 3.5 µM[16]
Kallikrein (pancreatic), pH 8.0	K_i = 1.0 nM[16]
Kallikrein (plasma)	K_i = 30 nM; 100 nM[15]
Kallikrein (tissue)	K_i = 1 nM[15]
Kallikrein (urine)	K_i = 1.7 nM[15]
Plasmin (porcine), pH 7.8	K_i = 4.0 nM[16]
Plasminogen activator	K_i = 8 µM; 27 µM[15]
Trypsin (bovine), pH 8.0	K_i = 0.06 pM[16]
Trypsinogen (bovine), pH 8.0	K_i = 1.8 µM[16]
Tryptase TL-2	16% Inhibition at 10 µM[15]
Urokinase (human), pH 8.8	K_i = 8.0 µM[16]

Table 2. Aprotinin solution stability

Solvent	Concentration	Temperature	% Loss/Time
Sterile water with 0.9% NaCl and 0.9% benzyl alcohol, pH 5.7-6.2	10 mg/mL	0-5 °C	< 4.3%/year
2.5% Trichloroacetic acid	N/A	80 °C	No loss[18]
pH < 12.6	N/A	N/A	No loss observed after 24 hours[19]
pH < 12	N/A	N/A	Irreversibly denatured[20]
pH 7-8	0.065-1.95 µg/mL	4 °C	About 1 week[15]
pH 7-8	0.065-1.95 µg/mL	–20 °C	> 6 months[15]

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