

For life science research only.  
Not for use in diagnostic procedures.



# Leupeptin

## Ac-Leu-Leu-argininal $\times \frac{1}{2} \text{H}_2\text{SO}_4$ , synthetic

 **Version: 14**

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<b>Cat. No. 11 017 101 001</b>	5 mg
<b>Cat. No. 11 017 128 001</b>	25 mg
<b>Cat. No. 11 034 626 001</b>	50 mg
<b>Cat. No. 11 529 048 001</b>	100 mg

**Store the product at +2 to +8°C.**

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# 1. General Information

## 1.1. Contents

Vial / Bottle	Label	Function / Description	Catalog Number	Content
1	Leupeptin	White powder	11 017 101 001	1 vial, 5 mg
			11 017 128 001	1 vial, 25 mg
			11 034 626 001	1 vial, 50 mg
			11 529 048 001	1 vial, 100 mg

## 1.2. Storage and Stability

### Storage Conditions (Product)

When stored at +2 to +8°C, the product is stable through the expiry date printed on the label.

Vial / Bottle	Label	Storage
1	Leupeptin	Store at +2 to +8°C. <b>⚠ Store dry.</b>

### Storage Conditions (Working Solution)

Store an aqueous solution of Leupeptin for 1 week at +2 to +8°C, stored under nitrogen, or in aliquots for at least 6 months at –15 to –25°C.

**⚠ Avoid repeated freezing and thawing.**

### Reconstitution

Leupeptin is readily soluble in water (1 mg/ml), methanol, ethanol, acetic acid, dimethylformamide, and dimethylsulfoxide. It is poorly soluble in acetone, chloroform, ethyl ether, and n-hexane.

## 1.3. Application

Leupeptin inhibits serine and thiol proteases, such as:

- Trypsin
- Plasmin
- Proteinase K
- Kallikrein
- Papain
- Thrombin
- Cathepsin A and B

Leupeptin as well as other protease inhibitors such as Antipain\*, Chymostatin\*, Pepstatin\* and phosphoramidon are useful for protection of proteins during their isolation from tissues or membranes. Not affected are α-, β-, γ- and δ-chymotrypsins, pepsin, cathepsin D, elastase, renin, and thermolysin.

## 2. How to Use this Product

### 2.1. Before you Begin


#### General Considerations

##### Classes of proteases

Proteases can be assigned to various classes on the basis of their characteristic active centers:

Protease Type	Active Center
Serine	Serine and histidine
Cysteine	Cysteine (thiol, SH-)
Metallo	Metal ions, such as Zn <sup>2+</sup> , Ca <sup>2+</sup> , Mn <sup>2+</sup>
Aspartate	Aspartic acid moiety

##### Protease classes and their specific inhibitors

Serine	Cysteine	Metallo	Aspartate
Aprotinin*	E-64*	Bestatin (aminopeptidase)*	Pepstatin*
Pefabloc® SC*		Phosphoramidon	
Pefabloc® SC PLUS*			
Leupeptin*			
 <i>Inhibits serine and cysteine proteases with trypsin-like specificity.</i>			
PMSF*			
cOmplete Protease Inhibitor Cocktail Tablets, EDTA-free*			
cOmplete Protease Inhibitor Cocktail tablets*			
α2-Macroglobulin* (endoproteinases)			

##### Cell permeability

Leupeptin is not cell permeable.

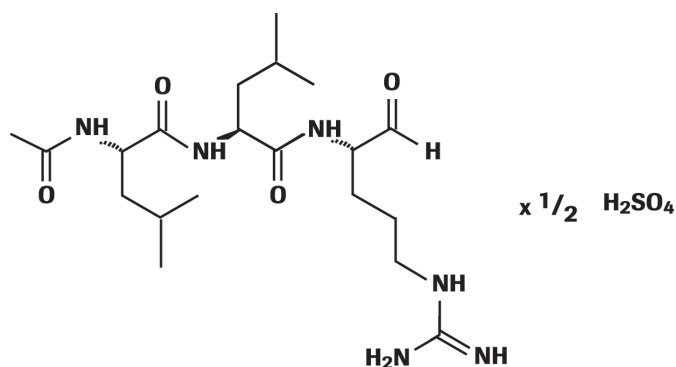
##### Removal of Leupeptin

Leupeptin can be removed from the reaction by dialysis.

## 2.2. Parameters

### Chemical Name

### Structural formula



**Fig. 1:** Chemical structure of Leupeptin.

### Inhibition

#### Mechanism of inhibition

The strong inhibitory effect is explained by the formation of a covalent hemiacetal adduct between the aldehyde group in the inhibitor and the serine hydroxyl function in the active site of the protease.

#### Molecular Weight

475.6 Da (Leupeptin  $\times \frac{1}{2} \text{H}_2\text{SO}_4$ )

493.6 Da (Leupeptin  $\times \frac{1}{2} \text{H}_2\text{SO}_4 \times \text{H}_2\text{O}$ )

# Specificity

## Specificity of Leupeptin to proteases

Enzyme	Substrate	ID <sub>50</sub> [µg/ml]
Plasmin	Fibrin	8
	Casein	36
Trypsin	Casein	2
	Hemoglobin	5
Papain	Casein	0.51
	Hemoglobin	0.15
Kallikrein	BAEE <sup>(1)</sup>	70
Cathepsin A	Cb-Glut-Tyr <sup>(2)</sup>	1.7
Cathepsin B	BAA <sup>(3)</sup>	0.44

<sup>(1)</sup> α-N-benzoyl-L-arginine ethyl ester-HCl

<sup>(2)</sup> α-N-carbobenzoxy-L-glutamyl-L-tyrosine

<sup>(3)</sup> α-N-benzoyl-L-arginine amide-HCl

**i** To check other protease inhibitors, try our *Protease Inhibitor Set\** including *Antipain (HCl)<sub>2</sub>*, *Aprotinin*, *Bestatin*, *Chymostatin*, *E-64*, *EDTA-Na<sub>2</sub>*, *Leupeptin*, *Pefabloc® SC*, *Pepstatin*, and *Phosphoramidon*.

See section, **General Considerations** for a table of protease classes and their inhibitors.

## Toxicity

LD<sub>50</sub>: 1.5 g/kg (mouse or rabbit, oral)

## Working Concentration

0.5 to 5 µg/ml

## 3. Additional Information on this Product

### 3.1. Test Principle

Protease inhibitors are very common in animals, plants and microorganisms. These naturally occurring compounds are exclusively oligo- or polypeptides, partly also glycopeptides. Culture filtrates of several streptomyces species are a rich source of peptide-derived protease inhibitors. In contrast to their related compounds from animals and plants, bacterial protease inhibitors are of low molecular weight. Compared with conventional synthetic protease inhibitors, they are more specific, more active at lower concentrations, and show less toxicity. Leupeptin is a tripeptide derivative with an acetylated  $\alpha$ -amino group. The C-terminus carries an aldehyde group instead of a carboxyl function.



### 3.2. Quality Control

Function tested with trypsin.

## 4. Supplementary Information

### 4.1. Conventions

To make information consistent and easier to read, the following text conventions and symbols are used in this document to highlight important information:

Text convention and symbols	
	<i>Information Note: Additional information about the current topic or procedure.</i>
	<b>Important Note: Information critical to the success of the current procedure or use of the product.</b>
① ② ③ etc.	Stages in a process that usually occur in the order listed.
① ② ③ etc.	Steps in a procedure that must be performed in the order listed.
* (Asterisk)	The Asterisk denotes a product available from Roche Diagnostics.

### 4.2. Changes to previous version

Layout changes.  
Editorial changes.

### 4.3. Ordering Information

Product	Pack Size	Cat. No.
Non-finished products		
cOplete, EDTA free	7500 tablets in glass vial	04 574 834 001
Reagents, kits		
E-64, Protease Inhibitor	10 mg	10 874 523 001
	25 mg	11 585 681 001
Pefabloc® SC (AEBSF)	custom fill	11 427 393 103
Chymostatin	10 mg	11 004 638 001
cOplete	20 tablets in a glass vial, for 50 ml each	11 697 498 001
	3 x 20 tablets in glass vials, for 50 ml each	11 836 145 001
	20 tablets, for 50 ml each	04 693 116 001
Antipain dihydrochloride	10 mg	11 004 646 001
cOplete, Mini	25 tablets in a glass vial, for 10 ml each	11 836 153 001
	30 tablets, for 10 ml each	04 693 124 001
cOplete, Mini, EDTA-free	25 tablets in a glass vial, for 10 ml each	11 836 170 001
	30 tablets, for 10 ml each	04 693 159 001
Pepstatin	custom fill	10 253 294 103
Protease Inhibitors Set	1 set, 10 individual protease inhibitors	11 206 893 001
cOplete, EDTA-free	20 tablets, for 50 ml each	04 693 132 001
Aprotinin	custom fill	10 236 632 103
Bestatin	10 mg	10 874 515 001
Pefabloc® SC PLUS	Set I, 100 mg Pefabloc® SC; 5 ml PSC-Protector solution	11 873 601 001
	Set II, 1 g Pefabloc® SC; 2 x 25 ml PSC-Protector solution	11 873 628 001
α <sub>2</sub> -Macroglobulin	25 inhibitor units	10 602 442 001
PMSF	10 g	10 837 091 001
	25 g	11 359 061 001



## 4.4. Trademarks

COMPLETE is a trademark of Roche.

All other product names and trademarks are the property of their respective owners.

## 4.5. License Disclaimer

For patent license limitations for individual products please refer to:

**List of biochemical reagent products.**

## 4.6. Regulatory Disclaimer

For life science research only. Not for use in diagnostic procedures.

## 4.7. Safety Data Sheet

Please follow the instructions in the Safety Data Sheet (SDS).

## 4.8. Contact and Support

To ask questions, solve problems, suggest enhancements or report new applications, please visit our **Online Technical Support Site.**

To call, write, fax, or email us, visit **sigma-aldrich.com**, and select your home country. Country-specific contact information will be displayed.

