

## Product Information | Certification of Analysis

### I Product Information

CAS: 9002-07-7

Lot No.

#### Trypsin, Mass Spec Grade

| Part No.    | Name                          | Size/pkg |
|-------------|-------------------------------|----------|
| HLS TRY001C | Trypsin, Mass Spec Grade      | 100 µg   |
| HLS HCL001  | 1 mM hydrochloric acid buffer | 0.5 mL   |

**Description:** Trypsin specifically hydrolyzes peptide bonds at the carboxyl side of lysine and arginine residues. Unmodified trypsin is subject to auto-proteolysis, generating fragments that can interfere with protein sequencing or HPLC peptide analysis. In addition, autoproteolysis can result in the generation of pseudo trypsin, which has been shown to exhibit chymotrypsin-like specificity. Mass Spec Grade modified trypsin is porcine trypsin modified by reductive methylation, rendering it resistant to proteolysis digestion. In enzymatic stability tests, modified trypsin was not found to self-hydrolyze but can retain higher the activity of general trypsin.

**Physical Appearance:** Lyophilized powder.

**Molecular Weight:** 23 kDa

**Resuspension Buffer (HLS CHL001C):** Trypsin Resuspension Buffer is composed of 1mM hydrochloric acid.

**Storage Conditions:** Store the lyophilized powder at -20 °C; store reconstituted enzyme at -80 °C.

**Shelf life:** 24 months at -20 °C

**Stability:** Modified trypsin is maximally active in the pH range of 7–9 and reversibly inactivated at pH 4.

#### Usage Notes:

1. For maximum activity, resuspend trypsin in the resuspension buffer provided, and heat at 30 °C for 15 min before use.
2. Thaw the reconstituted trypsin at room temperature, placing on ice immediately after thawing.
3. Add trypsin to a protease : protein ratio to be in range of 1:100 - 1:25 (w/w) for protein identification (recommended). Mix well and incubated for 37 °C for 4 h.

### I Quality Control

**Purity:** > 99.5% trypsin peak area analyzed by HPLC at 280 nm.

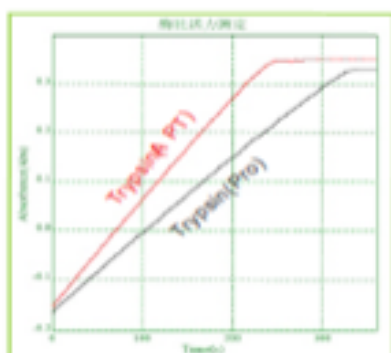
**Specificity:** < 5% nonspecific cleavage with Human Serum Albumin (HSA) sample. Digested products that were incubated at 37 °C for 16 h were compared with those incubated at 37 °C for 1 h, and the nonspecific cleavage was analyzed by LC-MS/MS.

**Activity:** 13,397 units/mg

**Unit Definition:** One unit is the amount of Mass spectrum Grade Modified Trypsin required to produce a  $\Delta A_{253}$  of 0.001 per minute at 37°C with the substrate N $\alpha$ -benzoyl-L-arginine ethyl ester (BAEE).

**MALDI-TOF Analysis:** Trypsin is analyzed by MALDI-TOF, impurity peak is not found.

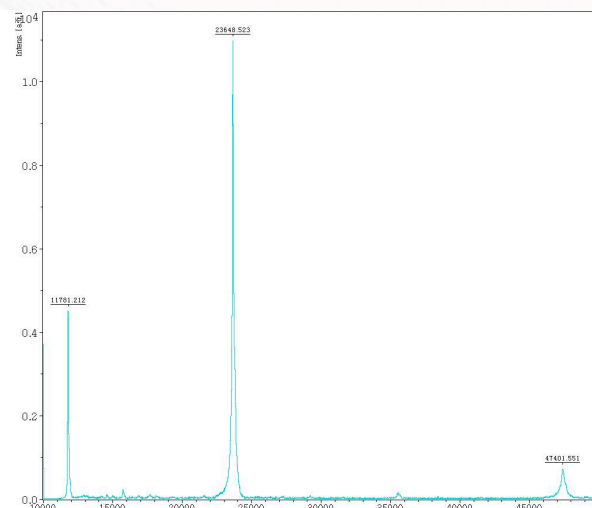
**LC-MS/MS Analysis:** HSA was dissolved and denatured for at 37 °C for 1 h, diluted at pH 8.0, and incubated with trypsin for 4 h. The digests were analyzed by LC-MS/MS, and experimental peptides results matched the peptides generated in a theoretical digestion results of HSA by trypsin.



Enzyme activity =  $\frac{\Delta A_{247nm}}{\Delta t \times \text{protein}} \times 1000 \times 5$   
 540<sup>th</sup> = 100% Trypsin activity (100% activity)  
 100% activity = 100% activity of protein (100% activity) at 100% activity

| 样品               | 斜率<br>( $\Delta A_{247}/\mu$ ) | TAME<br>(units/mg protein) | BAE<br>(units/mg protein) |
|------------------|--------------------------------|----------------------------|---------------------------|
| Trypsin (pro)    | K=0.001548                     | 172                        | 9890.0                    |
| Trypsin (apt-01) | K=0.002098                     | 233                        | 13397.5                   |

方法：分光光度计时间扫描  
 (条件：底物为TAME, 247nm, 25℃, 6min)



1 MEQVTFISLL FLSSAYSRG VFRDAHKSE VAHRFDLGE ENFKALVLI  
 51 FAQYLQCCPF EDHVKLNVET TEFARTCVAD ESAENCDSKL HTLFGDKLCT  
 101 VATLRETYGE MADCCARQEP ERNECFLQHK DDNPMLPRLV RPEVDVMCTA  
 151 FHDNEETFLR KYLYEIARRH FYFYAPELLF FAKRYKAAPT ECCQAADKAA  
 201 CLLPKLDELK DEGRKASSAQ RLKASLQKF GERAFKAMAV ARLSQRFFKA  
 251 EFAEVSKLVT DLTRVHTCC HGDLLCADD RADLAKYICE NQDSISSRLK  
 301 ECCEKPLLEK SHCIAEVEND EMPADLPSLA ADFVESKIDVC KNYAEAKDVF  
 351 LGMFLYFYAR RHPDYSVLL LRLAKTYETT LERCCAAADP HECYARVDE  
 401 FKPLVEEPQN LIKQNCLEFE QLGEYKQNA LLVRYTKVP QVSTPTLVEV  
 451 SRNLGKVGSK CCKHPEAKGM PCAEDYLSVV LNLCLVLEK IFVSDRVTEC  
 501 CTESLVNRRP CFSALEVDET YVFKFNAET FTFHADICTL SEKERQIKKQ  
 551 TALVELVKHK FKATKEQLKA VMDDFAAPVE KCKKADDEET CFAESGKGLV  
 601 AASQAALGL

QA Manager Signature:

Beijing Shengxia Proteins Scientific. Ltd.

I www.coowins.com I E-mail: [yu.feng@coowins.com](mailto:yu.feng@coowins.com) I Tel: +86 139 1067 9882

Add: Room 101, Building 3, No.2 Zhuyuan 2th Street, Tianzhu Free Trade Zone, Shunyi District, Beijing, P.R. China