ALPHA DIAGNOSTIC INTERNATIONAL

LIFE TECH Life Technologies™ Your Molecular & Cell Technology Partner

Product Specification Sheet

Human Beta-Secretase (BACE/Asp2) Peptide Substrates

Cat. # BACE-SW	Human beta-Secretase Peptide Substrate (**wild**) # 1	**SIZE**: 100 ug
Cat. # BACE-SS	Human beta-Secretase Peptide Substrate (**Swedish**) # 2	**SIZE**: 100 ug

β-amyloid **(Aβ)** deposition in the brain is the hallmark of Alzheimer's Disease (**AD**). To initiate Aβ formation, **β-secretase** cleaves APP at the N-terminus of Aβ to release APPsβ (~100 kDa soluble NT-fragment), and C99, a 12-kDa CT membrane fragment. Alternatively, **α-secretase** cleaves within the Aβ to prevent the formation of Aβ. Cleavage by α-secretase produces a soluble N-terminal fragment, APPsα, and a 10-kDa membrane C-terminal fragment, C83. Both C99 and C83 can be further cleaved by **γ-secretase** releasing Aβ and a nonpathogenic p3 peptide, respectively.

Recently, **BACE (Beta-site APP Cleaving Enzyme)** has been identified as **β-secretase**. BACE belongs to the family of **Aspartyl proteases (Asp)** also known as **Memapsins**. At least four related Asps, located on chromosome IV and X, have been cloned (**Asp1, Asp2, Asp3, and Asp4).** Human **BACE/Asp2/Memapsin2,** located on chromosome 11, is a transmembrane protein of 501 aa (signal peptide 1-21 aa, a proprotein domain 22-45 aa, 1 TM domain near the CT, and a short cytoplasmic CT-tail of 24 aa; mature protein 46-460 aa). The lumenal portion of BACE has two active site motifs at 93 aa and 289 aa with signature sequence of aspartic proteases. Rat and mouse BACE (501 aa) are 96% identical with human BACE. BACE expression was most prominent in most areas of the rat brain and pancreas. It has been localized in the compartments of the secretory pathways.

Source of Peptides

A 14 aa peptide (**designated BACE-SW**) corresponding to the known b-secretase site of human APP590-603 were synthesized and purified by HPLC. The sequence of the wild type and Swedish mutation peptides are as follows:

β-Secretase Substrate (wild type) Human APP590-603

Acetyl-EISEVKM ↓ DA EFR HD-amide

β-Secretase Substrate (Swedish) Human APP590-603

Acetyl-EISEV**NL** ↓ DA EFR HD-amide

↓ denotes the known cleavage site of β-secretase.

These sequences are slightly larger than previously used by Yan and Lin et al (1999). They extend by 2 aa at both ends to facilitate their detection by HPLC.

Form & Storage

Control peptide Solution is provided in water at 1 mg/ml (100 ug/100 ul) (100 ul in solution or Lyophilized). It is also available as 1 mg vial in lyophilized form. **Lyophilized products** should be reconstituted in 100 ul water and gently mixed for 15 min at room temp. All peptide received in solution or reconstituted from lyophilized vials should be stored frozen at -20°C or below in suitable aliquots. It is not recommended to store diluted solutions. Avoid repeated freeze and thaw.

Recommended Usage

Purified BACE/Asp2 cleaves APP and synthetic **APP peptide substrates** at the β-secretase site, and the rate of cleavage is increase 10-fold by a **Swedish type mutation** associated with early onset of Alzheimer's disease. BACE has an acidic optimum (pH 4.0). Typically, purified BACE/Asp2 prepared by membrane solubilization in 25 mM Tris, pH 8.0, containing 50 mM **β**-octylglucoside retained the enzyme activity. Enzymatic reactions are performed in 50 mM 2-(N-morpholino)ethane-sulfomate, pH 5.5, containing 70 uM peptide substrate and the reaction performed at 37oC [see details in Yan et al (1999) and Lin et al (1999)]. Reaction products are then analyzed by reverse phase HPLC.

General References

Yan R et al (1999) Nature 402, 533-537; Lin X et al (2000) PNAS 97, 1456-1460; Sinha S et al (1999) Nature 537-540; Hussain I et al (1999) Mol. Cell Neurosci. 14, 419-427; Vassar R et al (1999) Science 286, 735-741.

*This product is for In vitro research use only.

Related material available from ADI

Ant-Beta amyloid 1-40, 1-42, APP, Parkin, Synucleins (α, β, γ), Presenilins 1, 2

Antibodies to α-secretase (TACE), beta-Secretase (BACE/Asp2 & BACE/Asp1)

ReadyBrain Blot- Study distribution of protein in 12 regions of mouse/rat brain using pre-made protein blots.

BACE-SS-SW 71219A

India Contact:
Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura, Delhi – 110034 (INDIA). Ph: +91-11-42208000, 42208111, 42208222, Mobile: +91-9810521400, Fax: +91-11-42208444
Email: customerservice@lifetechindia.com Website: www.lifetechindia.com