

**DESCRIPTION**

<b>Source</b>	Human embryonic kidney cell, HEK293-derived human CD3 epsilon protein			
	Human CD3 epsilon (Asp23-Asp126) Accession # P07766.2	IEGRMD	Human IgG <sub>1</sub> (Pro100-Lys330)	Avi-tag
	N-terminus		C-terminus	

**N-terminal Sequence Analysis** Gln22 inferred from enzymatic pyroglutamate treatment revealing Asp23

**Structure / Form** Disulfide-linked homodimer  
Biotinylated via Avi-tag

**Predicted Molecular Mass** 40 kDa

**SPECIFICATIONS**

**SDS-PAGE** 45-60 kDa, under reducing conditions.

**Activity** Measured by its binding ability in a functional ELISA.  
Biotinylated Recombinant Human CD3 epsilon Fc Chimera Avi-tag binds to Human CD3 epsilon Antibody (Catalog # MAB10670) with an ED<sub>50</sub> of 0.600-6.00 ng/mL.

**Endotoxin Level** <0.10 EU per 1 µg of the protein by the LAL method.

**Purity** >95%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.

**Formulation** Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.

**PREPARATION AND STORAGE**

**Reconstitution** Reconstitute at 250 µg/mL in water.

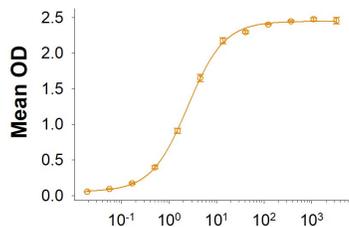
**Shipping** The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

**Stability & Storage** Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 3 months, -20 to -70 °C under sterile conditions after reconstitution.

**DATA**

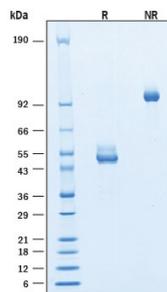
**Binding Activity**



**Biotinylated Recombinant Human CD3ε Avi-tag (ng/ml)**

**Biotinylated Recombinant Human CD3 epsilon Fc Chimera Avi-tag Protein Binding Activity.** Measured by its binding ability in a functional ELISA. Biotinylated Recombinant Human CD3 epsilon Fc Chimera Avi-tag Protein (Catalog # AVI9850) binds to Human CD3 epsilon Antibody (Catalog # MAB1067) with an ED<sub>50</sub> of 0.600-6.00 ng/mL.

**SDS-PAGE**



**Biotinylated Recombinant Human CD3 epsilon Fc Chimera Avi-tag Protein SDS-PAGE.** 2 µg/lane of Biotinylated Recombinant Human CD3 epsilon Fc Chimera Avi-tag Protein (Catalog # AVI9850) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 45-60 kDa and 90-120 kDa, respectively.

**BACKGROUND**

CD3 $\epsilon$  (T-Cell Surface Glycoprotein CD3 Epsilon) is a type I transmembrane protein that belongs to the Ig (Immunoglobulin) superfamily. Human CD3 $\epsilon$  consists of 104 amino acid (aa) extracellular domain, 26 aa transmembrane domain and 55 aa cytoplasmic domain (1). With the extracellular domain, human CD3 $\epsilon$  shares 44.2% and 49% homology with mouse and rat CD3 $\epsilon$  respectively. There are four CD3 proteins: CD3 $\delta$ , CD3 $\epsilon$ , CD3 $\gamma$ , and CD3 $\zeta$ . All CD3 proteins contain ITAM (Immunoreceptor Tyrosine-based Activation Motifs) in the cytoplasmic tail, which becomes phosphorylated by Src family protein tyrosine kinases LCK and FYN upon TCR engagement (2). CD3 proteins form heterodimers of CD3 $\delta$ /CD3 $\epsilon$  and CD3 $\gamma$ /CD3 $\epsilon$ , which bind to TCR and form trimeric TCR $\alpha$ /CD3 $\epsilon$ /CD3 $\gamma$  and TCR $\beta$ /CD3 $\gamma$ /CD3 $\epsilon$ . The resulting heterohexamer further associates with CD3 $\zeta$  homodimer and forms TCR/CD3 signaling complex (3, 4). Similar complex is also formed with TCR $\gamma$  and TCR $\delta$  (5). CD3 $\epsilon$  plays essential role in adaptive immune response. Mutations in CD3 $\epsilon$  lead to SCID (severe combined immunodeficiency) (6). Our Avi-tag Biotinylated CD3 $\epsilon$  features biotinylation at a single site contained within the Avi-tag, a unique 15 amino acid peptide. Protein orientation will be uniform when bound to streptavidin-coated surface due to the precise control of biotinylation and the rest of the protein is unchanged so there is no interference in the protein's bioactivity.

**References:**

1. Gold, D.P. *et al.* (1986) *Nature* **321**:431.
2. Barber, E.K. *et al.* (1989) *Proc. Nat. Acad. Sci. U.S.A.* **86**:3277.
3. Manolios, N. *et al.* (1991) *EMBO J* **10**:1643.
4. Dietrich, J. *et al.* (1999) *Eur J Immunol* **29**:1719.
5. Pan, Q. *et al.* (2006) *Mol Immunol* **43**:1741.
6. Soudais, C. *et al.* (1993) *Nat. Genet.* **3**:77.