

DESCRIPTION

Source	Chinese Hamster Ovary cell line, CHO-derived human EGFR protein			
	Human EGFR Isoform VIII (Leu25-Ser378) Accession # NP_001333870.1	IEGRMD	Human IgG ₁ (Pro100-Lys330)	Avi-tag
	N-terminus		C-terminus	
N-terminal Sequence	Leu25			
Analysis				
Structure / Form	Disulfide-linked homodimer, biotinylated via Avi-tag			
Predicted Molecular Mass	67 kDa			

SPECIFICATIONS

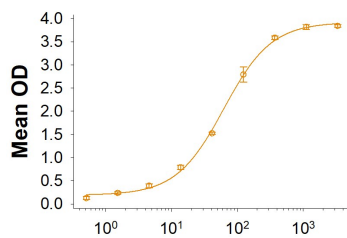
SDS-PAGE	95-115 kDa, under reducing conditions
Activity	Measured by its binding ability in a functional ELISA. When Human EGFR Isoform VIII antibody (Novus Catalog # 50599) is immobilized at 0.1 µg/mL (100 µL/well), Biotinylated Recombinant Human EGFR Isoform VIII Fc Chimera Avi-tag (Catalog # AV110494) binds with an ED ₅₀ of 25-150 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 500 µg/mL in PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	<p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <ul style="list-style-type: none"> • 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

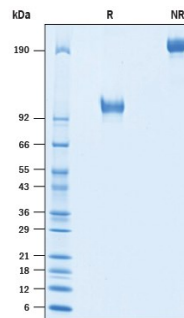
Binding Activity



Biotinylated Recombinant Human EGFR Isoform VIII Fc Avi-tag (ng/mL)

When Human EGFR Isoform VIII antibody (Novus Catalog # BNP-50599) is immobilized at 0.1 µg/mL (100 µL/well), Biotinylated Recombinant Human EGFR Isoform VIII Fc Chimera Avi-tag (Catalog # AV110494) binds with an ED₅₀ of 25-150 ng/mL.

SDS-PAGE



2 µg/lane of Biotinylated Recombinant Human EGFR Isoform VIII Fc Avi-tag Protein (Catalog # AV110494) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 95-115 kDa and 190-230 kDa, respectively.

BACKGROUND

Epidermal growth factor receptor (EGFR), also known as HER-1 and ErbB1, is a member of a subfamily of receptor tyrosine kinases comprised of four members: EGFR, ErbB2 (Neu, HER-2), ErbB3 (HER-3), and ErbB4 (HER-4). All family members are type I transmembrane glycoproteins with an extracellular domain (ECD) containing two cysteine-rich domains separated by a spacer region and a cytoplasmic domain containing a tyrosine kinase domain followed by multiple tyrosine autophosphorylation sites (1, 2). Several soluble isoforms lacking the intracellular domain are generated by alternate splicing (3-4). EGFRVIII is a tumor-specific mutation that results from an in-frame deletion removing 267 amino acids from the ECD and insertion of a glycine residue (5). EGFRVIII has a molecular mass of approximately 145 kDa and has been shown to have weaker activity than full-length EGFR (6). EGFR binds a subset of the EGF family ligands, including EGF, amphiregulin, TGF- α , betacellulin, epiregulin, HB-EGF, and epigen (1, 2). Ligand binding induces EGFR homodimerization as well as heterodimerization with ErbB2, resulting in kinase activation, heterodimerization tyrosine phosphorylation and cell signaling (7-9). EGFR can also be recruited to form heterodimers with the ligand-activated ErbB3 or ErbB4. EGFR signaling regulates multiple biological functions including cell proliferation, differentiation, motility, and apoptosis (7-9). EGFR is overexpressed in a wide variety of tumors, with EGFRVIII overexpressed particularly in glioblastoma multiforme (GMB), and is the target of several anti-cancer therapeutics (5,10,11). Our Avi-tag Biotinylated Recombinant Human EGFRVIII 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:

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6. Batra SK, *et al.* (1995) *Cell Growth Differ* **6**:1251
7. Freed, D. M. *et al.* (2017) *Cell.* **171**:683.
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