Anti-EGFR Antibody [A8E12]

HA601073

Product Type:	Mouse monoclonal IgG2a, primary antibodies
Species reactivity:	Human, Monkey
Applications:	WB, IHC-P
Molecular Wt:	Predicted band size: 134 kDa
Clone number:	A8E12

Description: Receptor tyrosine kinase binding ligands of the EGF family and activating several signaling cascades to convert extracellular cues into appropriate cellular responses. Known ligands include EGF, TGFA/TGF-alpha, AREG, epigen/EPGN, BTC/betacellulin, epiregulin/EREG and HBEGF/heparin-binding EGF. Ligand binding triggers receptor homo- and/or heterodimerization and autophosphorylation on key cytoplasmic residues. The phosphorylated receptor recruits adapter proteins like GRB2 which in turn activates complex downstream signaling cascades. Activates at least 4 major downstream signaling cascades including the RAS-RAF-MEK-ERK, PI3 kinase-AKT, PLCgamma-PKC and STATs modules. May also activate the NF-kappa-B signaling cascade. Also directly phosphorylates other proteins like RGS16, activating its GTPase activity and probably coupling the EGF receptor signaling to the G protein-coupled receptor signaling. Also phosphorylates MUC1 and increases its interaction with SRC and CTNNB1/beta-catenin. Positively regulates cell migration via interaction with CCDC88A/GIV which retains EGFR at the cell membrane following ligand stimulation, promoting EGFR signaling which triggers cell migration. Plays a role in enhancing learning and memory performance. Ubiquitously expressed. Isoform 2 is also expressed in ovarian cancers.

Immunogen:	Recombinant protein within human EGFR aa 25-645 (Extracellular)
Positive control:	A431 cell lysate, MDA-MB-468 cell lysate, Human placenta tissue.
Subcellular location:	Endoplasmic reticulum, Cell membrane, Endosome, Nucleus, Golgi apparatus.
Database links:	SwissProt: P00533 Human
Recommended Dilutions:	
WB	1:2,000
IHC-P	1:800
Storage Buffer:	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.
Purity:	Protein A affinity purified.

Images

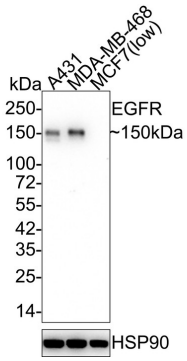

Fig1: Western blot analysis of EGFR on different lysates with Mouse anti-EGFR antibody (HA601073) at 1/2,000 dilution.

Lane 1: A431 cell lysate
Lane 2: MDA-MB-468 cell lysate
Lane 3: MCF7 cell lysate (low expression)

Lysates/proteins at 15 µg/Lane.

Predicted band size: 134 kDa
Observed band size: 150 kDa

Exposure time: 21 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA601073) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

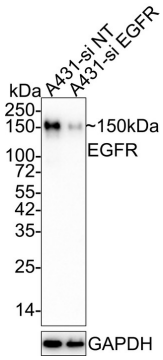

Fig2: Western blot analysis of EGFR on different lysates with Mouse anti-EGFR antibody (HA601073) at 1/2,000 dilution.

Lane 1: A431-si NT cell lysate
Lane 2: A431-si EGFR cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 134 kDa
Observed band size: 150 kDa

Exposure time: 1 minutes 2 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA601073) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

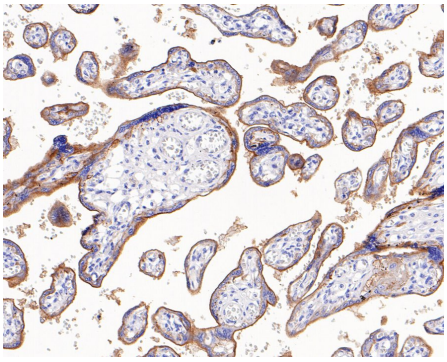

Fig3: Immunohistochemical analysis of paraffin-embedded Human placenta tissue with Mouse anti-EGFR antibody (HA601073) at 1/800 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601073) at 1/800 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Background References

1. Lu C., Mi L.Z., Grey M.J., Zhu J., Graef E., Yokoyama S., Springer T.A. Structural evidence for loose linkage between ligand binding and kinase activation in the epidermal growth factor receptor. Mol. Cell. Biol. 30:5432-5443(2010)
2. Red Brewer M., Choi S.H., Alvarado D., Moravcevic K., Pozzi A., Lemmon M.A., Carpenter G. The juxtamembrane region of the EGF receptor functions as an activation domain. Mol. Cell 34:641-651(2009)