

Product datasheet

Anti-EGFR antibody [EPR39Y] ab76153

Recombinant RabMAb

[5 References](#) [5 Images](#)

Overview

Product name	Anti-EGFR antibody [EPR39Y]
Description	Rabbit monoclonal [EPR39Y] to EGFR
Host species	Rabbit
Specificity	This antibody detects both phosphorylated and non-phosphorylated EGFR protein. This product yielded a strong signal in western blot using A431 (human squamous carcinoma) lysate which naturally overexpresses the EGFR protein. Western blot conditions may need to be optimised for cell lines and tissues that express lower levels of endogenous EGFR.
Tested applications	Suitable for: Flow Cyt (Intra), ICC/IF, WB Unsuitable for: IHC-P or IP
Species reactivity	Reacts with: Mouse, Human
Immunogen	Synthetic peptide within Human EGFR. The exact sequence is proprietary. A synthetic peptide corresponding to residues surrounding Tyr1068 of mature human EGFR Database link: P00533
Positive control	HeLa whole cell lysate (ab150035); A431 cells; Ms skin tissue lysate.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p> <p>Rat: We have preliminary internal testing data to indicate this antibody may not react with this species. Please contact us for more information.</p>

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer	pH: 7.20

	Preservative: 0.05% Sodium azide
	Constituents: 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR39Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab76153 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/55 - 1/100. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/100 - 1/250.
WB		1/1000 - 1/5000. Detects a band of approximately 170 kDa (predicted molecular weight: 134 kDa). This product yielded a strong signal in western blot using A431 (human squamous carcinoma) lysate which naturally overexpresses the EGFR protein. Western blot conditions may need to be optimised for cell lines and tissues that express lower levels of endogenous EGFR.

Application notes Is unsuitable for IHC-P or IP.

Target

Function	<p>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, amphiregulin, 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.</p> <p>Isoform 2 may act as an antagonist of EGF action.</p>
Tissue specificity	Ubiquitously expressed. Isoform 2 is also expressed in ovarian cancers.
Involvement in disease	<p>Lung cancer</p> <p>Inflammatory skin and bowel disease, neonatal, 2</p>

Sequence similarities

Belongs to the protein kinase superfamily. Tyr protein kinase family. EGF receptor subfamily. Contains 1 protein kinase domain.

Post-translational modifications

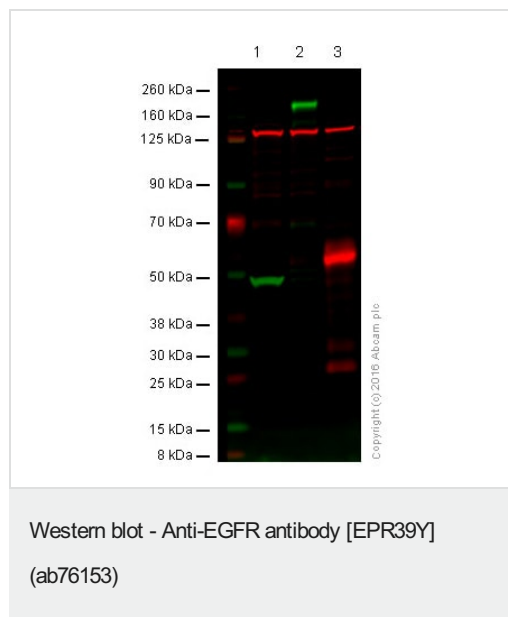
Phosphorylation at Ser-695 is partial and occurs only if Thr-693 is phosphorylated. Phosphorylation at Thr-678 and Thr-693 by PRKD1 inhibits EGF-induced MAPK8/JNK1 activation. Dephosphorylation by PTPRJ prevents endocytosis and stabilizes the receptor at the plasma membrane. Autophosphorylation at Tyr-1197 is stimulated by methylation at Arg-1199 and enhances interaction with PTPN6. Autophosphorylation at Tyr-1092 and/or Tyr-1110 recruits STAT3. Dephosphorylated by PTPN1 and PTPN2.

Monoubiquitinated and polyubiquitinated upon EGF stimulation; which does not affect tyrosine kinase activity or signaling capacity but may play a role in lysosomal targeting. Polyubiquitin linkage is mainly through 'Lys-63', but linkage through 'Lys-48', 'Lys-11' and 'Lys-29' also occurs. Deubiquitination by OTUD7B prevents degradation. Ubiquitinated by RNF115 and RNF126. Methylated. Methylation at Arg-1199 by PRMT5 stimulates phosphorylation at Tyr-1197.

Cellular localization

Secreted and Cell membrane. Endoplasmic reticulum membrane. Golgi apparatus membrane. Nucleus membrane. Endosome. Endosome membrane. Nucleus. In response to EGF, translocated from the cell membrane to the nucleus via Golgi and ER. Endocytosed upon activation by ligand. Colocalized with GPER1 in the nucleus of estrogen agonist-induced cancer-associated fibroblasts (CAF).

Images



All lanes : Anti-EGFR antibody [EPR39Y] (ab76153) at 1/1000 dilution

Lane 1 : Caco-2 cell lysate

Lane 2 : A431 cell lysate

Lane 3 : Mouse skin cell lysate

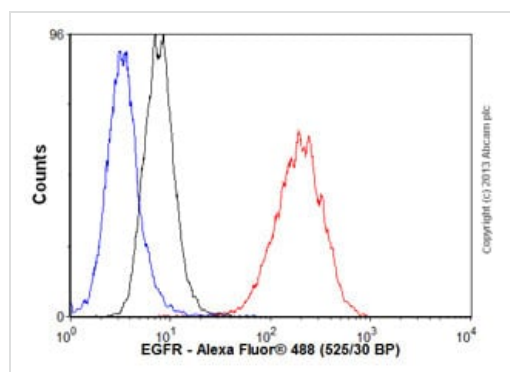
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 134 kDa

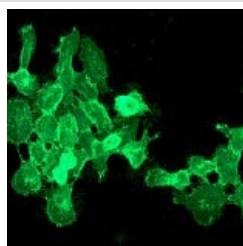
This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour before being incubated with ab76153 overnight at 4°C in the presence of loading control **ab18058** (Mouse monoclonal [SPM227] to Vinculin diluted 1:10000). Antibody binding was detected using IR-labelled goat anti-Rabbit Ab at a 1:10,000 dilution for one hour at room temperature before imaging.

This product yielded a strong signal in western blot using A431 (human squamous carcinoma) lysate which naturally overexpresses the EGFR protein. Western blot conditions may need to be optimised for cell lines and tissues that express lower levels of endogenous EGFR.



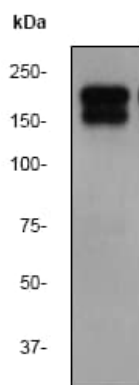
Flow Cytometry (Intracellular) - Anti-EGFR antibody [EPR39Y] (ab76153)

Overlay histogram showing A431 cells stained with ab76153 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab76153, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor[®] 488 goat anti-rabbit IgG (H&L) ([ab150077](#)) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



Immunocytochemistry/ Immunofluorescence - Anti-EGFR antibody [EPR39Y] (ab76153)

ab76153 at 1/100 dilution staining EGFR in A431 cells by Immunofluorescence.



Western blot - Anti-EGFR antibody [EPR39Y] (ab76153)

Anti-EGFR antibody [EPR39Y] (ab76153) at 1/5000 dilution + HeLa cell lysate at 10 µg

Secondary

goat anti-rabbit HRP at 1/1000 dilution

Developed using the ECL technique.

Predicted band size: 134 kDa

Observed band size: 170,180 kDa

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-EGFR antibody [EPR39Y] (ab76153)

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