abcam

Product datasheet

Anti-ErbB 4 antibody [EPR22665-104] ab219208

Recombinant RabMAb

4 Images

Overview

Product name	Anti-ErbB 4 antibody [EPR22665-104]
Description	Rabbit monoclonal [EPR22665-104] to ErbB 4
Host species	Rabbit
Specificity	ab219208 is not recommended for mouse IHC.
Tested applications	**Suitable for:** WB, IHC-P, Flow Cyt **Unsuitable for:** ICC/IF or IP
Species reactivity	**Reacts with:** Mouse, Human
Immunogen	Recombinant fragment within Human ErbB 4 aa 700-1000. The exact sequence is proprietary. Database link: Q15303
Positive control	WB: HEK-293, T-47D, MCF7 and 4T1 lysates. IHC-P: Human breast carcinoma tissue. FC: T-47D and MCF7 cells.
General notes	Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab® patents. This product is a recombinant rabbit monoclonal antibody.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR22665-104
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab219208** 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
WB		1/1000. Predicted molecular weight: 147 kDa.
IHC-P		1/1000.
Flow Cyt		1/60.

Application notes Is unsuitable for ICC/IF or IP.

Target

Function Specifically binds and is activated by neuregulins, NRG-2, NRG-3, heparin-binding EGF-like growth factor, betacellulin and NTAK. Interaction with these factors induces cell differentiation. Not activated by EGF, TGF-A, and amphiregulin. The C-terminal fragment (CTF) of isoform JMA-A CYT-2 (containing E4ICD2) can stimulate transcription in the presence of YAP1. ERBB4 intracellular domain is involved in the regulation of cell growth. Conflicting reports are likely due at least in part to the opposing effects of the isoform-specific and nuclear-translocated ERBB4 intracellular domains (E4ICD1 and E4ICD2). Overexpression studies in epithelium show growth inhibition using E4ICD1 and increased proliferation using E4ICD2. E4ICD2 has greater in vitro kinase activity than E4ICD1. The kinase activity is required for the nuclear translocation of E4ICD2.

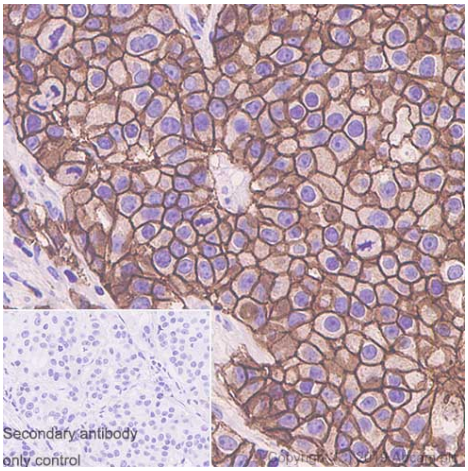
Tissue specificity Expressed at highest levels in brain, heart, kidney, in addition to skeletal muscle, parathyroid, cerebellum, pituitary, spleen, testis and breast. Lower levels in thymus, lung, salivary gland, and pancreas. Isoform JM-A CYT-1 and isoform JM-B CYT-1 are expressed in cerebellum, but only the isoform JM-B is expressed in the heart.

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

Post-translational modifications Isoform JM-A CYT-1 and isoform JM-A CYT-2 but not isoform JM-B CYT-1 and isoform JM-B CYT-2 are processed by ADAM17. Proteolytic processing in response to ligand or 12-O-tetradecanoylphorbol-13-acetate stimulation results in the production of 120 kDa soluble receptor forms and intermediate membrane-anchored 80 kDa fragments (m80HER4), which are further processed by a presenilin-dependent gamma-secretase to release the respective cytoplasmic intracellular domain E4ICD (either E4ICD1/s80Cyt1 or E4ICD2/s80Cyt2). Membrane-anchored 80 kDa fragments of the processed isoform JM-A CYT-1 are more readily degraded by the proteasome than fragments of isoform JM-A CYT-2 suggesting a prevalence of E4ICD2 over E4ICD1.
Ligand-binding increases phosphorylation on tyrosine residues. Isoform JM-A CYT-2 is constitutively phosphorylated on tyrosine residues in a ligand-independent manner. E4ICD2 but not E4ICD1 is phosphorylated on tyrosine residues.
Ubiquitinated. The ERBB4 intracellular domain is ubiquitinated and targeted to proteosomal

	degradation during mitosis mediated by the APC/C complex. Isoform JM-A CYT-1 and isoform JM-B CYT-1 are ubiquitinated by WWP1. The ERBB4 intracellular domain (E4ICD1) is ubiquitinated, and this involves NEDD4.
Cellular localization	Membrane and Nucleus. Following proteolytical processing E4ICD (E4ICD1 or E4ICD2 generated from the respective isoforms) is translocated to the nucleus. Significantly more E4ICD2 than E4ICD1 is found in the nucleus. E4ICD2 colocalizes with YAP1 in the nucleus.

Images

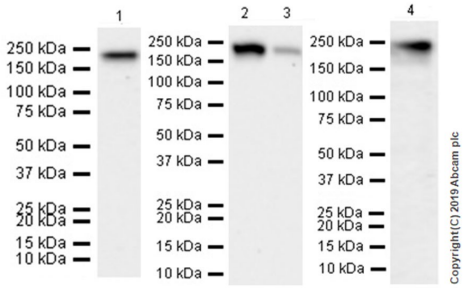


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ErbB 4 antibody [EPR22665-104] (ab219208)

Immunohistochemical analysis of paraffin-embedded Human breast carcinoma tissue labeling ErbB 4 with ab219208 at 1/1000 dilution followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer). Positive staining on the human breast carcinoma (PMID: 19239686) is observed. The section was incubated with ab219208 for 15 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.



Western blot - Anti-ErbB 4 antibody [EPR22665-104] (ab219208)

All lanes : Anti-ErbB 4 antibody [EPR22665-104] (ab219208) at 1/1000 dilution

Lane 1 : HEK-293 (human embryonic kidney epithelial cell) whole cell lysate
Lane 2 : T-47D (human ductal breast epithelial tumor epithelial cell) whole cell lysate
Lane 3 : MCF7 (human breast adenocarcinoma epithelial cell) whole cell lysate
Lane 4 : 4T1 (mouse mammary gland carcinoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 147 kDa

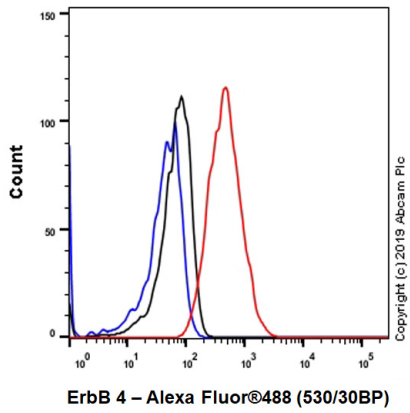
Observed band size: 180 kDa

why is the actual band size different from the predicted?

Blocking and diluting buffer and concentration: 5% NFDM/TBST

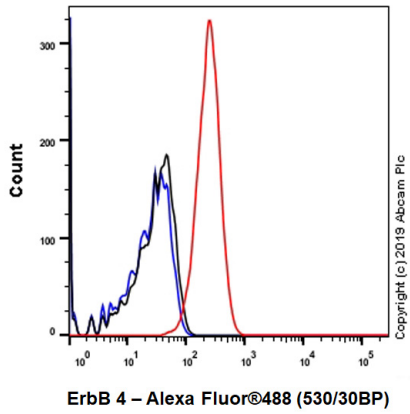
The molecular weight observed is consistent with what has been described in the literature (PMID: 22761786) Lane 4 was developed using a higher sensitivity ECL substrate.

Exposure time: Lane 1:70 seconds; Lanes 2-3:48 seconds; Lane 4: 3 minutes.



Flow Cytometry - Anti-ErbB 4 antibody [EPR22665-104] (ab219208)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized MCF7 (Human breast adenocarcinoma epithelial cell) cells labelling ErbB 4 with ab219208 at 1/60 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730, Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 was used as the secondary antibody.



Flow Cytometry - Anti-ErbB 4 antibody [EPR22665-104] (ab219208)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized T-47D (Human ductal breast epithelial tumor epithelial cell) cells labelling ErbB 4 with ab219208 at 1/60 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730, Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 was used as the secondary antibody.

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