

Anti-EGFR Antibody Picoband™

Catalog Number: A00023-4

About EGFR

The epidermal growth factor receptor (EGFR; ErbB-1; HER1 in humans) is the cell-surface receptor for members of the epidermal growth factor family (EGF-family) of extracellular protein ligands. It is a member of the ErbB family of receptors, a subfamily of four closely related receptor tyrosine kinases: EGFR (ErbB-1), HER2/c-neu (ErbB-2), Her 3 (ErbB-3) and Her 4 (ErbB-4). EGFR exists on the cell surface and is activated by binding of its specific ligands, including epidermal growth factor and transforming growth factor alpha (TGFalpha). EGFR and its ligands are cell signaling molecules involved in diverse cellular functions, including cell proliferation, differentiation, motility, and survival, and in tissue development. Mutations that lead to EGFR overexpression (known as upregulation) or overactivity have been associated with a number of cancers, including lung cancer and glioblastoma multiforme. In this latter case a more or less specific mutation of EGFR, called EGFRvIII is often observed.

Overview

Product Name	Anti-EGFR Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-EGFR Antibody Picoband™ catalog # A00023-4. Tested in ELISA, Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	ELISA, Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.01mg NaN3.
Storage Instructions	Store at -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	P00533

Technical Details

Immunogen	E.coli-derived human EGFR recombinant protein (Position: N36-N497).
Predicted Reactive Species	Chicken
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG
Form	Lyophilized

Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	<p>Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.</p> <p>If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.</p> <p>Some PubMed article(s) citing the expression level of this target are as follows:</p> <p>Boster Bio's internal QC testing used:</p> <p>Western blot, 0.1-0.25ug/ml, Human, Mouse, Rat</p> <p>Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human, Mouse, Rat</p> <p>Immunocytochemistry/Immunofluorescence, 4ug/ml, Human</p> <p>Immunofluorescence, 4ug/ml, Human</p> <p>Flow Cytometry, 1-3ug/1x10⁶ cells, Human</p> <p>Direct ELISA, 0.1-0.5ug/ml, Human</p>

Anti-EGFR Antibody Picoband™ (A00023-4) Images

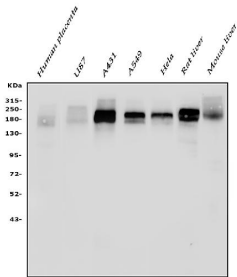


Figure 1. Western blot analysis of EGFR using anti-EGFR antibody (A00023-4).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: human placenta tissue lysates,

Lane 2: human U87 whole cell lysates,

Lane 3: human A431 whole cell lysates,

Lane 4: human A549 whole cell lysates,

Lane 5: human Hela whole cell lysates,

Lane 6: rat liver tissue lysates,

Lane 7: mouse liver tissue lysates.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes.

Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-EGFR antigen affinity purified polyclonal antibody (Catalog # A00023-4) at 0.25 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for EGFR at approximately 180KD. The expected band size for EGFR is at 180KD.

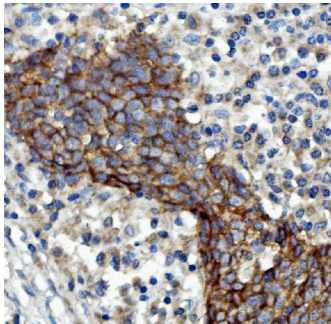


Figure 2. IHC analysis of EGFR using anti-EGFR antibody (A00023-4).

EGFR was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-EGFR Antibody (A00023-4) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

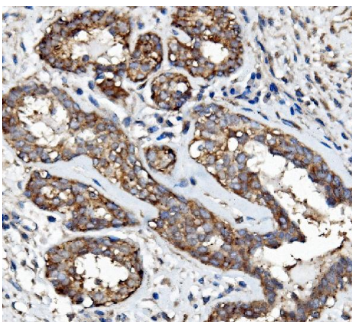


Figure 3. IHC analysis of EGFR using anti-EGFR antibody (A00023-4).

EGFR was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-EGFR Antibody (A00023-4) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

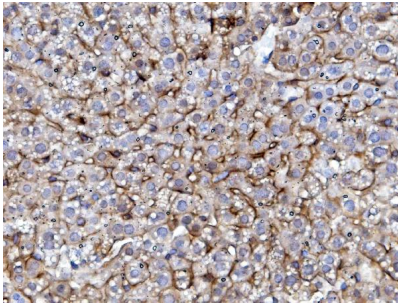


Figure 4. IHC analysis of EGFR using anti-EGFR antibody (A00023-4).
EGFR was detected in paraffin-embedded section of mouse liver tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-EGFR Antibody (A00023-4) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

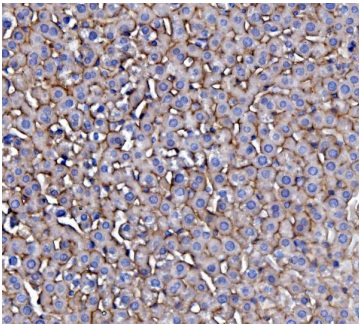


Figure 5. IHC analysis of EGFR using anti-EGFR antibody (A00023-4).
EGFR was detected in paraffin-embedded section of rat liver tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-EGFR Antibody (A00023-4) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

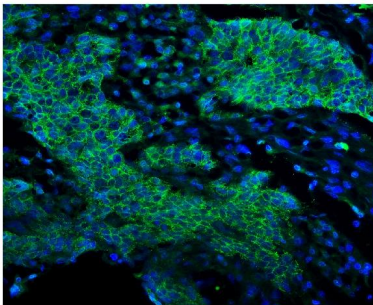


Figure 6. IF analysis of EGFR using anti-EGFR antibody (A00023-4).
EGFR was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 4ug/mL rabbit anti-EGFR Antibody (A00023-4) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

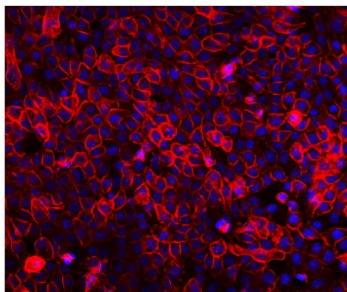


Figure 7. IF analysis of EGFR using anti-EGFR antibody (A00023-4).
EGFR was detected in immunocytochemical section of A431 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 4ug/mL rabbit anti-EGFR Antibody (A00023-4) overnight at 4°C. DyLight®550 Conjugated Goat Anti-Rabbit IgG (BA1135) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

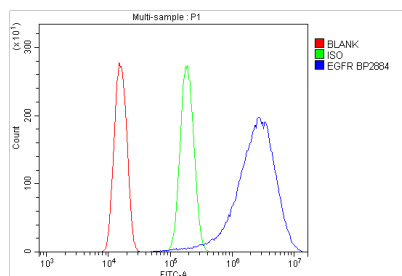


Figure 8. Flow Cytometry analysis of A431 cells using anti-EGFR antibody (A00023-4). Overlay histogram showing A431 cells stained with A00023-4 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-EGFR Antibody (A00023-4, 1ug/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

1 Publications Citing This Product

1. PubMed ID: -, Study of the correlation between the expression of nuclear factor kappa B and proliferation regulatory proteins and chronic superficial gastritis, Hu Hui, Ma Zhijian, Ren Shouzhong, Xie Yiqiang, Vojnosanitetski pregled 2020 OnLine-First, 00, SP - 135 EP-135

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