abcam

Product datasheet

Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free ab182618

重组 RabMAb

11 References 17 图像

概述

产品名称	Anti-EGFR (phospho Y1068)抗体[EP774Y] - BSA and Azide free
描述	兔单克隆抗体[EP774Y] to EGFR (phospho Y1068) - BSA and Azide free
宿主	Rabbit
特异性	Recognises EGFR phosphorylated on Tyrosine 1068 of the mature human isoform 1 (corresponding to Y1092 from the precursor form P00533-1/p170) The mouse recommendation is based on the WB results. We do not guarantee IHC-P for mouse.
经测试应用	**适用于:** Flow Cyt (Intra), WB, IHC-P, Dot blot, ICC/IF
种属反应性	**与反应:** Mouse, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	IHC-P: Human breast adenocarcinoma, papillary carcinoma of thyroid glad, glioma, cervical carcinoma and prostate cancer tissue; Mouse E17 embryo head tissue and mouse pancreas tissue. ICC/IF: A431 cells and WD-PBEC cultures. WB: A431 cells treated with EGF. Dot Blot: EGFR (pY1068) peptide.
常规说明	ab182618 is the carrier-free version of **ab40815**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity

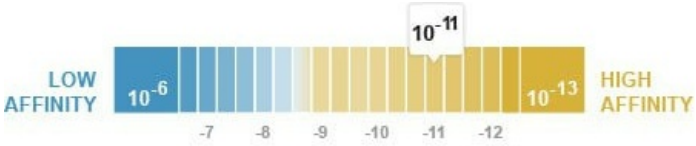
- Long-term security of supply
- Animal-free production

For more information **see here**.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C. Do Not Freeze.
解离常数 (K_D)	K_D = 3.60 x 10^{-11} M



Learn more about K_D

存储溶液	pH: 7.20 Constituent: PBS
无载体	**是**
纯度	Protein A purified
克隆	单克隆
克隆编号	EP774Y
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab182618于以下的经测试应用**

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

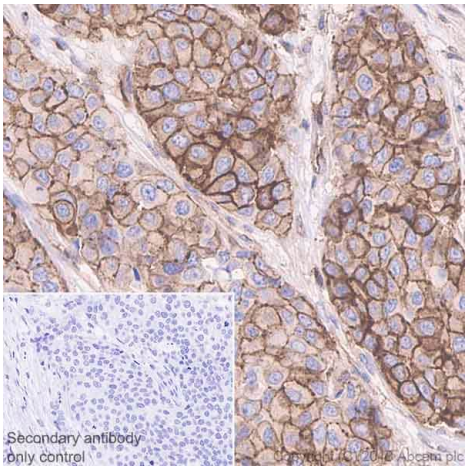
应用	Ab评论	说明
Flow Cyt (Intra)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 134 kDa. Can be blocked with EGFR (phospho Y1092) peptide (**ab190788**).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**. The mouse recommendation is based on the WB results. We do not guarantee IHC-P for mouse.
Dot blot		Use at an assay dependent concentration.

应用	Ab评论	说明
ICC/IF		Use at an assay dependent concentration.

靶标

功能	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. Isoform 2 may act as an antagonist of EGF action.
组织特异性	Ubiquitously expressed. Isoform 2 is also expressed in ovarian cancers.
疾病相关	Lung cancer Inflammatory skin and bowel disease, neonatal, 2
序列相似性	Belongs to the protein kinase superfamily. Tyr protein kinase family. EGF receptor subfamily. Contains 1 protein kinase domain.
翻译后修饰	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.
细胞定位	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).

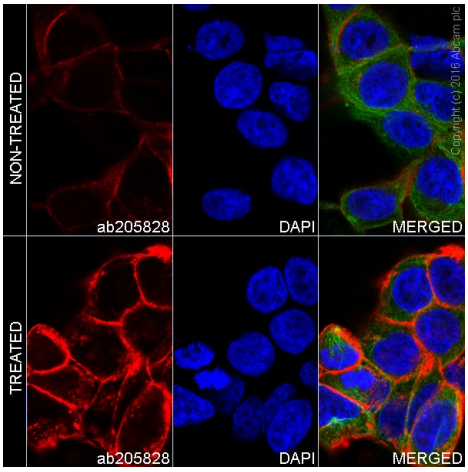
图片



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast cancer tissue sections labeling EGFR with purified **ab40815** at 1/500 dilution (1.75 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



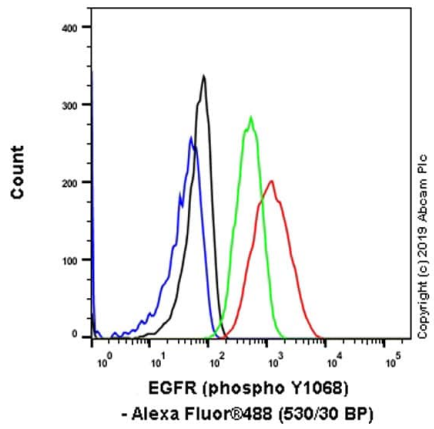
Immunocytochemistry/ Immunofluorescence - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Clone EP774Y (ab182618) has been successfully conjugated by Abcam. This image was generated using Anti-EGFR (phospho Y1068) antibody [EP774Y] (Alexa Fluor® 647). Please refer to **ab205828** for protocol details.

ab205828 staining EGFR (phospho Y1092) in A431 cells +/-EGF (100ng/ml, 5min). The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h.

The cells were then incubated overnight at +4°C with **ab205828** at 1:100 dilution (shown in red) and **ab195887**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 488), at 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

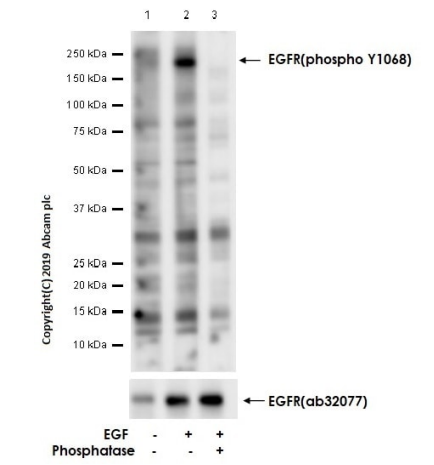
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Flow Cytometry (Intracellular) - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Intracellular Flow Cytometry analysis of A431 (Human epidermoid carcinoma epithelial cell) treated with 200 ng/ml EGF for 15 minutes cells labeling EGFR with purified **ab40815** at 1/800 dilution (1µg/ml) (red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue). Untreated A431 cells (Green).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



Western blot - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

All lanes : Anti-EGFR (phospho Y1068) antibody [EP774Y] (**ab40815**) at 1/1000 dilution

Lane 1 : C2C12 (Mouse myoblasts myoblast) whole cell lysate
Lane 2 : C2C12 (Mouse myoblasts myoblast) treated with 100 ng/ml EGF for 24 hours whole cell lysate
Lane 3 : C2C12 (Mouse myoblasts myoblast) treated with 100 ng/ml EGF for 24 hours whole cell lysate. Then the membrane was incubated with phosphatase.

Lysates/proteins at 20 µg per lane.

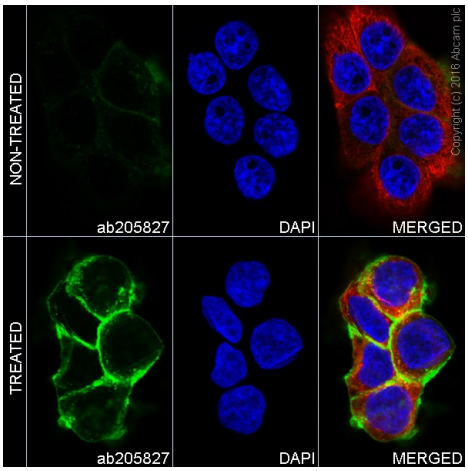
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 134 kDa
Observed band size: 175 kDa

Exposure time: 30 seconds

Blocking and dilution buffer: 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



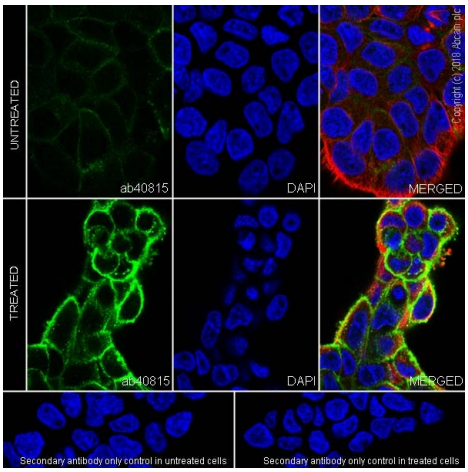
Immunocytochemistry/ Immunofluorescence - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Clone EP774Y (ab182618) has been successfully conjugated by Abcam. This image was generated using Anti-EGFR (phospho Y1068) antibody [EP774Y] (Alexa Fluor® 488). Please refer to **ab205827** for protocol details.

ab205827 staining EGFR (phospho Y1068) in A431 cells +/-EGF (100ng/ml, 5min). The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h.

The cells were then incubated overnight at +4°C with **ab205827** at 1:100 dilution (shown in green) and **ab195889**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 594), at 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

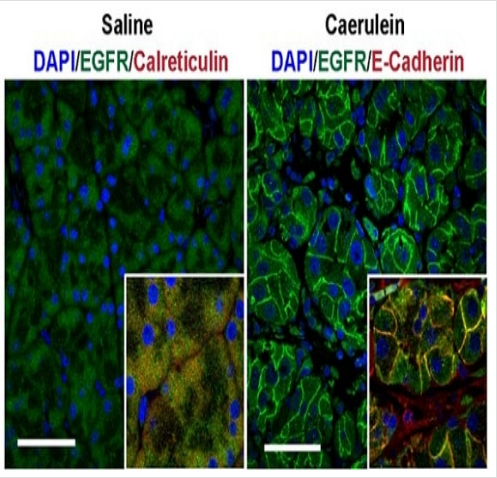
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunocytochemistry/ Immunofluorescence - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Immunocytochemistry/ Immunofluorescence analysis of A431 (Human epidermoid carcinoma epithelial cell) treated with 100 ng/ml EGF for 10 minutes cells labeling EGFR with purified **ab40815** at 1/500 dilution (1.8 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1/200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).

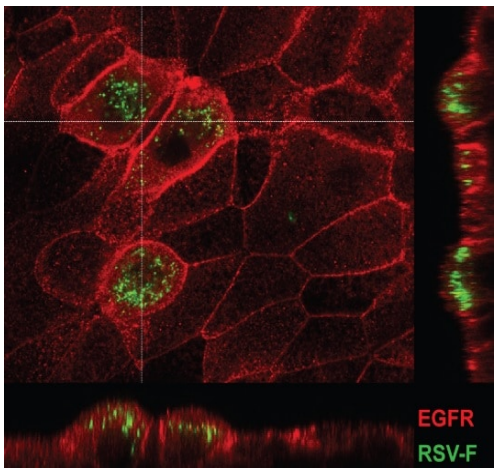


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Total EGFR (green) subcellular localization with and without caerulein-induced pancreatitis as determined by immunohistochemistry and confocal imaging. The nuclei were identified with DAPI stain (blue). Scale bars = 50 µm. Calreticulin and E-cadherin (red) served as markers for the endoplasmic reticulum and plasma membrane, respectively, and were used to quantify EGFR subcellular location. Pancreatitis was induced with the 1-day protocol of 8 hourly caerulein injections in 6–8 week old wild-type (wt) and 3-week old $AGR2^{-/-}$ (ko) mice.

For immunofluorescence, antigen retrieval was performed in a pressure cooker set to 118°C. The slides were incubated in antigen unmasking solution (DAKO) for 3 min followed by equilibration at room temperature for 1 hr. The slides were then placed in 5% serum blocking solution (goat, horse, or rabbit serum as appropriate) for 30 min to block nonspecific binding of antibody to the tissue. The sections were incubated with primary antibody diluted in 2% serum overnight at 4°C. The respective secondary antibodies were used at predetermined dilutions. Immunofluorescence slides were mounted with media containing DAPI stain (Vectashield, Vector Laboratories).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



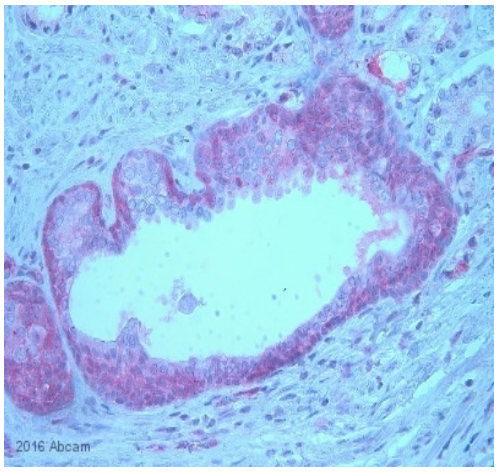
Immunocytochemistry/ Immunofluorescence - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

WD-PBEC cultures were infected with RSV clinical isolate BT2a and stained for EGFR (red) and RSV F (RSV F, green) expression.

For WD-PBECs, pediatric bronchial epithelial cells (PBEC) were obtained, via written parental consent, from bronchial brushings of children undergoing elective surgery at the Royal Belfast Hospital for Sick Children, and the procedures were approved by the Office of Research Ethics Committees Northern Ireland. PBEC were expanded in collagen-coated flasks using airway epithelial cell media and supplements (Lonza), then seeded onto transwell inserts (Corning), and then air-liquid interface (ALI) cultures were initiated and maintained 21 days in order to establish well-differentiated (WD)-PBECs. Paraformaldehyde-fixed and permeabilized WD-PBEC were stained for RSV F protein expression and were stained with anti-phospho-(p)-EGFR (Abcam, **ab40815**). WD-PBEC cultures were infected with RSV subgroup A clinical isolate BT2a. Fluorescent images were obtained with a SP5 confocal DMI 6000

inverted microscope (Leica).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).

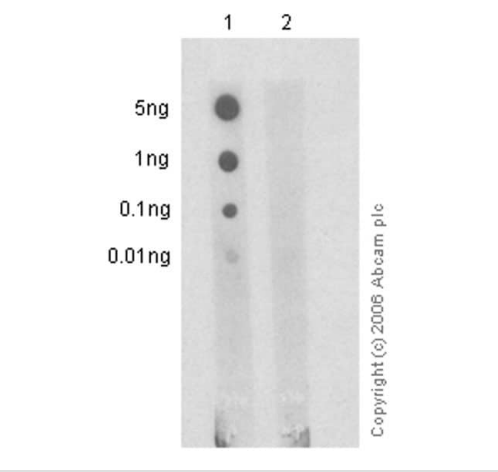


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

This image is courtesy of an anonymous Abreview.

Formaldehyde-fixed, paraffin-embedded human prostate cancer tissue stained for EGFR (phospho Y1068) using **ab40815** (unpurified) at 1/200 dilution in immunohistochemical analysis, followed by Goat anti Rabbit IgG (Biotin).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



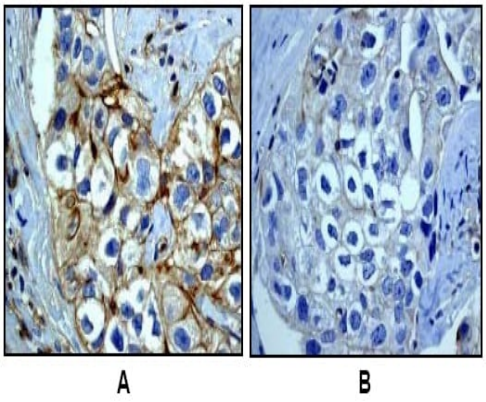
Dot Blot - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Dot blot analysis of EGFR (pY1068) peptide (Lane 1), SMAD5 (unmodified) peptide labelling EGFR (pY1068) with **ab40815** (unpurified) at a dilution of 1/1000. Peroxidase conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody at a dilution of 1/2500.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.

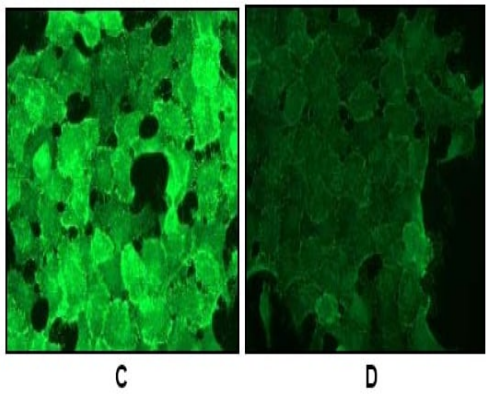
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Immunohistochemical staining of untreated (A) and Phosphatase-treated (B) paraffin-embedded breast adenocarcinoma tissue using **ab40815** (unpurified).

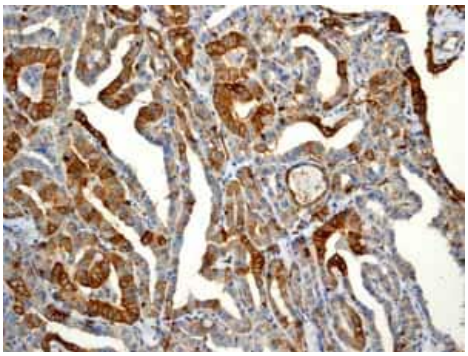
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



Immunocytochemistry/ Immunofluorescence - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Immunofluorescent staining of untreated (C) and Phosphatase-treated (D) A431 cells using **ab40815** (unpurified).

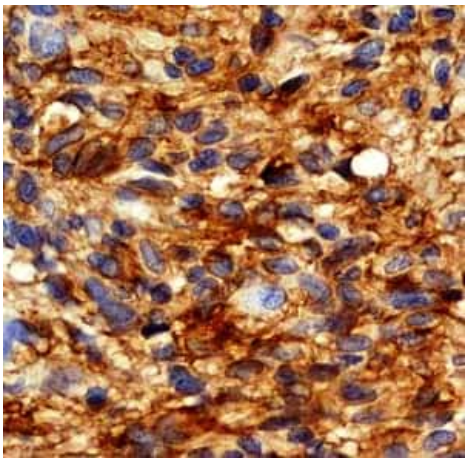
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

ab40815 (unpurified) showing positive staining in Papillary carcinoma of thyroid gland tissue.

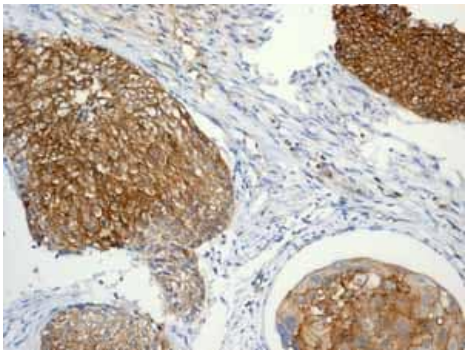
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

ab40815 (unpurified) showing positive staining in Glioma tissue.

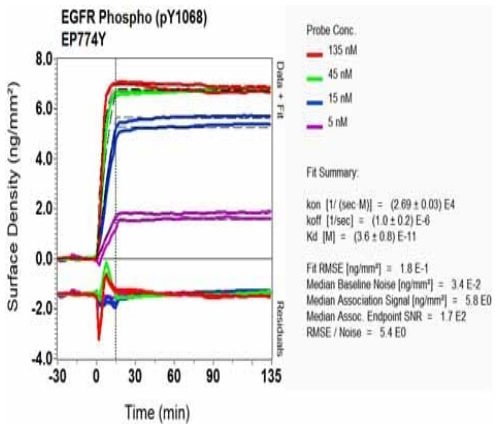
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

ab40815 (unpurified) showing positive staining in Cervical carcinoma tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).



OI-RD Scanning - Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40815**).

Anti-EGFR (phospho Y1068) antibody [EP774Y] - BSA and Azide free (ab182618)

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