Anti-Phospho-EGFR (Y1092) Antibody [SJ0194]

ET1606-44

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IF-Cell, IF-Tissue, IHC-P
Molecular Wt:	170 kDa
Clone number:	SJ0194

Description: Epidermal growth factor mediates its effects on cell growth through its inter-action with a cell surface glycoprotein designated the EGF receptor. Binding of EGF or TGF alpha to the EGF receptor activates tyrosine-specific protein kinase activity intrinsic to the EGF receptor. The carboxy terminal tyrosine residues on EGFR, Tyr 1068 and Tyr 1173, are the major sites of autophosphorylation, which occurs as a result of EGF binding. Once activated, EGFR mediates the binding of the phosphotyrosine binding (PTB) domain of GRB2 through direct interactions with Tyr 1068 and Tyr 1086 and through indirect interactions with Tyr 1173 in the Ras signaling pathway. Tyr 1173 of EGFR also functions as a kinase substrate. Phosphorylation of Tyr 992, Tyr 1068 and Tyr 1086 is required for conformational change in the C-terminal tail of the EGF receptor.

Immunogen: Synthetic phospho-peptide corresponding to residues surrounding Tyr1092 of Human EGFR aa 1,071-1,120 / 1,210.

Positive control: A431 cell lysate treated with EGF, BT-20, human tonsil tissue, mouse skin tissue.

Subcellular location: Cell membrane, Nucleus membrane, Nucleus, Endoplasmic reticulum membrane, Golgi apparatus membrane, Endosome.

Database links: SwissProt: P00533 Human | Q01279 Mouse

Recommended Dilutions:

WB	1:1,000-1:2,000
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:50-1:200

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% SodiumAzide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

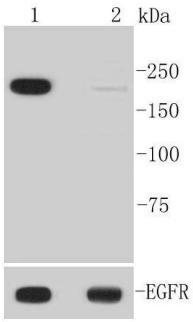

Fig1: Western blot analysis of Phospho-EGFR (Y1092) on different lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET1606-44, 1/1,000) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:5,000 dilution was used for 1 hour at room temperature.

Positive control:
Lane 1: A431 cell lysate treated with EGF
Lane 2: Untreated A431 cell lysate

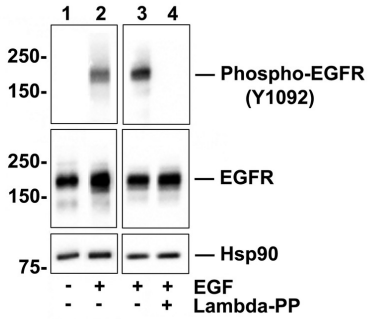

Fig2: Western blot analysis of Phospho-EGFR(Y1092) on A431 cell lysates.

Lane 1: A431 cells, whole cell lysate, 10ug/lane
Lane 2/3: A431 cells treated with 100 ng/ml EGF for 30 minutes, whole cell lysates, 10ug/lane
Lane 4: A431 cells treated with 100 ng/ml EGF for 30 minutes, then treated with 2.8ug/ul lambda-PP for 30 minutes, whole cell lysates, 10ug/lane

All lanes :
Anti-Phospho-EGFR(Y1092) antibody (ET1606-44
) at 1:500 dilution. Anti-EGFR antibody (ET1603-37) at 1:500 dilution. Anti-Hsp90 beta antibody (ET1605-56) at 1:10,000 dilution. Goat Anti-Rabbit IgG H&L (HRP) (HA1001) at 1/200,000 dilution.

Predicted band size: 134 kDa
Observed band size: 170 kDa

Blocking and diluting buffer: 5% BSA.

Exposure time: 15 seconds

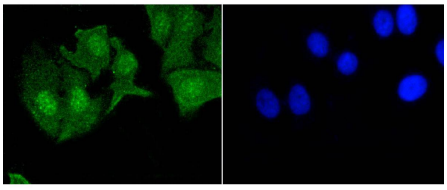

Fig3: ICC staining of Phospho-EGFR (Y1092) in BT-20 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1606-44, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

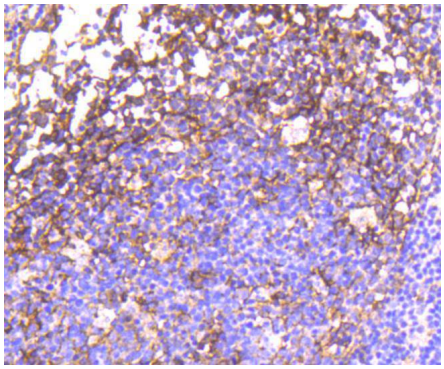

Fig4: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-Phospho-EGFR (Y1092) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1606-44, 1/50) for 30 minutes 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.

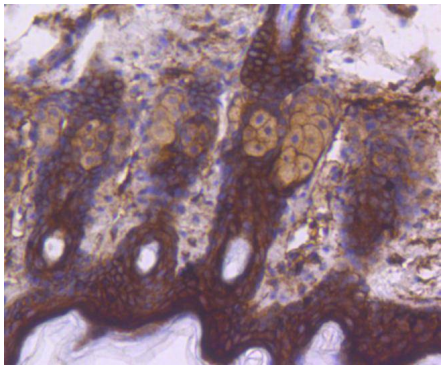

Fig5: Immunohistochemical analysis of paraffin-embedded mouse skin tissue using anti-Phospho-EGFR (Y1092) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1606-44, 1/50) for 30 minutes 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.

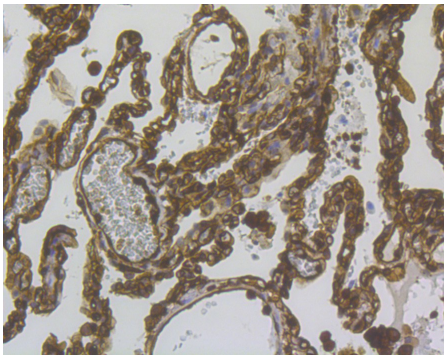

Fig6: Immunohistochemical analysis of paraffin-embedded human lung cancer tissue with Rabbit anti-Phospho-EGFR (Y1092) antibody (ET1606-44) at 1/200 dilution.

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 (ET1606-44) at 1/200 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. Nakamura T et al. LRIG1 inhibits STAT3-dependent inflammation to maintain corneal homeostasis. J Clin Invest 124:385-97 (2014).
2. Furcht CM et al. Multivariate signaling regulation by SHP2 differentially controls proliferation and therapeutic response in glioma cells. J Cell Sci 127:3555-67 (2014).