

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human EGFR when phosphorylated at Y1086 in Western blots.
Source	Monoclonal Rat IgG _{2A} Clone # 938835
Purification	Protein A or G purified from cell culture supernatant
Immunogen	Phosphopeptide containing the human EGFR Y1086 site Accession # P00533
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

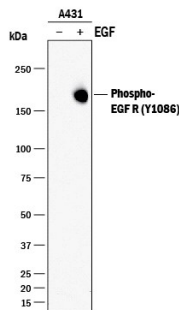
APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Western Blot	1 µg/mL	See Below
Immunocytochemistry	8-25 µg/mL	See Below
Simple Western	10 µg/mL	See Below

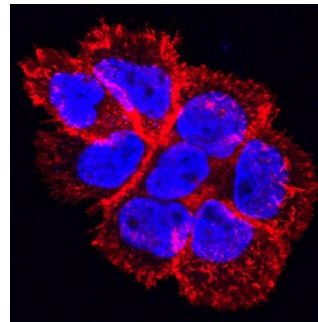
DATA

Western Blot



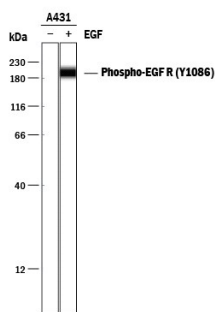
Detection of Phospho-EGFR (Y1086) by Western Blot. Western blot shows lysates of A431 human epithelial carcinoma cell line untreated (-) or treated (+) with 100 ng/mL Recombinant Human EGF (Catalog # [236-EG](#)) for 5 minutes. PVDF membrane was probed with 1 µg/mL of Rat Anti-Human Phospho-EGFR (Y1086) Monoclonal Antibody (Catalog # MAB89671) followed by HRP-conjugated Anti-Rat IgG Secondary Antibody (Catalog # [HAF005](#)). A specific band was detected for Phospho-EGFR (Y1086) at approximately 180 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



EGFR in A431 Human Cell Line. EGFR phosphorylated at Y1086 was detected in immersion fixed A431 human epithelial carcinoma cell line treated with Recombinant Human EGF (Catalog # [236-EG](#)) using Rat Anti-Human EGFR Monoclonal Antibody (Catalog # MAB89671) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rat IgG Secondary Antibody (red; Catalog # [NL013](#)) and counterstained with DAPI (blue). Specific staining was localized to cell membrane. View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

Simple Western



Detection of Human Phospho-EGFR (Y1086) by Simple Western™. Simple Western lane view shows lysates of A431 human epithelial carcinoma cell line untreated (-) or treated (+) with 100 ng/mL Recombinant Human EGF (Catalog # [236-EG](#)) for 5 minutes, loaded at 0.2 mg/mL. A specific band was detected for Phospho-EGFR (Y1086) at approximately 197 kDa (as indicated) using 10 µg/mL of Rat Anti-Human Phospho-EGFR (Y1086) Monoclonal Antibody (Catalog # MAB89671) followed by 1:50 dilution of HRP-conjugated Anti-Rat IgG Secondary Antibody (Catalog # [HAF005](#)). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Epidermal growth factor receptor (EGFR, also known as ErbB1 and HER1) is the founding member of the ErbB family of receptor tyrosine kinases. Ligand binding induces receptor dimerization and autophosphorylation on multiple tyrosine residues. Phosphorylation of Y1086 creates a binding site for the adaptor protein GRB2, leading to attenuation of receptor signaling.