

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human ErbB2 when phosphorylated at Y1248.
Source	Polyclonal Rabbit IgG
Purification	Antigen Affinity-purified
Immunogen	Phosphopeptide containing human ErbB2 Y1248 site
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.

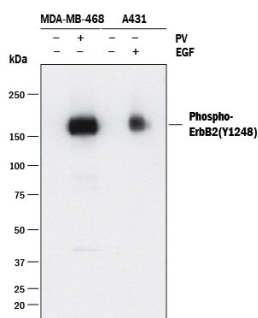
APPLICATIONS

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	0.25 µg/mL	See Below
Immunohistochemistry	5-15 µg/mL	See Below
Simple Western	5 µg/mL	See Below

DATA

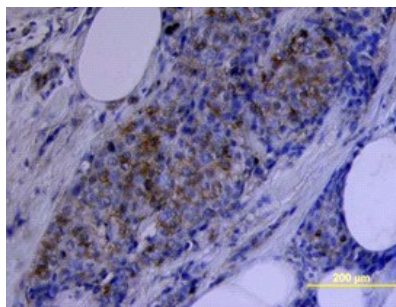
Western Blot



Detection of Human Phospho-ErbB2/Her2 (Y1248) by Western Blot.

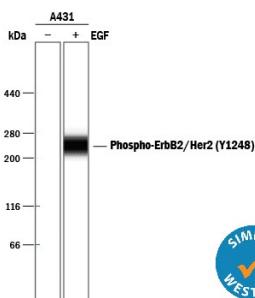
Western blot shows lysates of MDA-MB-468 human breast cancer cell line and A431 human epithelial carcinoma cell line untreated (-) or treated (+) with 1 mM Pervanadate (PV) for 10 minutes or with 10 ng/mL Recombinant Human EGF (Catalog # 236-EG) for 5 minutes. PVDF membrane was probed with 0.25 µg/mL of Rabbit Anti-Human Phospho-ErbB2/Her2 (Y1248) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1768) followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). A specific band was detected for Phospho-ErbB2/Her2 (Y1248) at approximately 170 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry



ErbB2/Her2 in Human Breast Cancer Tissue. ErbB2/Her2 phosphorylated at Y1248 was detected in immersion fixed paraffin-embedded sections of human breast cancer tissue using Rabbit Anti-Human Phospho-ErbB2/Her2 (Y1248) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1768) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Rabbit HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS005) and counterstained with hematoxylin (blue). View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

Simple Western



Detection of Human Phospho-ErbB2/Her2 (Y1248) by Simple Western™.

Simple Western™. Simple Western lane view shows lysates of A431 human epithelial carcinoma cell line untreated (-) or treated (+) with 10 ng/mL Recombinant Human EGF (Catalog # 236-EG) for 5 minutes, loaded at 0.2 mg/mL. A specific band was detected for Phospho-ErbB2/Her2 (Y1248) at approximately 265 kDa (as indicated) using 5 µg/mL of Rabbit Anti-Human Phospho-ErbB2/Her2 (Y1248) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1768). This experiment was conducted under reducing conditions and using the 66-440 kDa separation system.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 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

ErbB2, also called Neu and Her2 (human epidermal growth factor receptor 2), is a type I membrane glycoprotein that is a member of the ErbB family of tyrosine kinase receptors. ErbB family members serve as receptors for the epidermal growth factor (EGF) family of growth factors. ErbB2 is widely expressed in epithelial cells and has also been found to be over-expressed in a large number of breast carcinomas. Among ErbB family members, ErbB2 is unique in that it has no identified ligands. Rather, ErbB2 heterodimerizes with the other members of the ErbB family (ErbB1 (EGFR), ErbB3, ErbB4) to form higher affinity signaling complexes. Because ErbB3 contains a defective kinase domain, the kinase domain of ErbB2 is responsible for initiating the tyrosine phosphorylation signal through the heterodimeric receptor. It has been found that a discrete three amino acid signal in the ErbB3 cytoplasmic domain is critical for transactivation of ErbB2. Interestingly, this same three amino acid signal has also been found in ErbB1 and ErbB4. Phosphoinositide 3-kinase has been shown to play a role in ErbB2 signal transduction. The cytoplasmic domain of ErbB2 has been shown to associate with beta-catenin and plakoglobin. Human ErbB2 consists of 1255 amino acids (aa) with a 21 aa signal sequence, a 631 aa extracellular domain, a 23 aa transmembrane region, and a 580 aa cytoplasmic domain. ErbB2 can be shed from the cell surface by proteolytic cleavage by an unidentified protease. ErbB2 appears to play roles in development, cancer, communication at the neuromuscular junction and regulation of cell growth and differentiation (1-10).

References:

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