Anti-HER2 / ErbB2 Antibody [PD00-99]

HA721210

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IHC-P, IF-Tissue
Molecular Wt:	Predicted band size: 138 kDa
Clone number:	PD00-99

Description: HER-2 (also called HER-2/neu, c-erbB2, ERBB2 or neu) is a transmembrane receptor tyrosine kinase. HER-2 is a proto-oncogene, i.e. its activation causes malignant transformation and increases the malignant potential (cell proliferation, invasiveness etc.) of the cells. Amplification of HER-2 gene invariably leads to over-expression of its protein product. The magnitude of over- expression is usually 10-100 folds or even more. Over-expressed HER-2 protein disturbs the HER-receptor family signalling networks, i.e. signalling mediated via EGFR receptor, HER-3 and HER-4. In tumours, HER-2 is over-expressed in 15-25% of primary breast cancers. Metastases usually have the same amplification status as the primary tumours. HER-2 amplification and over-expression are typical features of hormone receptor negative, rapidly growing histologic grade 2-3 tumours. Of the histologic types, Paget's disease is almost invariably HER-2 positive, whereas only a small minority of lobular and tubular carcinomas shows HER-2 amplification. HER-2 amplification and over-expression can also be found in intestinal type gastric and gastroesophageal carcinomas, ovarian carcinomas, high grade endometrial carcinomas and some salivary duct tumours. Low-level copy number increases have been found also in rare cases of lung tumours. Because of its central importance in breast cancer therapy selection, standardization of HER-2 IHC assays and slide interpretation are of outmost clinical and economical importance.

Immunogen:	Recombinant fragment within Human ErbB2/ HER2 aa 500-650.
Positive control:	SK-Br-3 cell lysates, human breast carcinoma tissue.
Subcellular location:	Cell membrane, Nucleus, Cytoplasm.
Database links:	SwissProt: P04626 Human
Recommended Dilutions:	
WB	1:1,000
IHC-P	1:4,000
IF-Tissue	1:200
Storage Buffer:	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.
Purity:	Protein A affinity purified.

Images

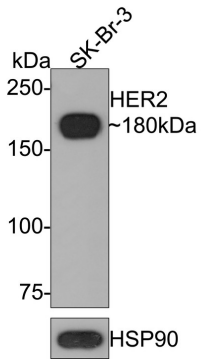

Fig1: Western blot analysis of HER2 / ErbB2 on SK-Br-3 cell lysates with Rabbit anti-HER2 / ErbB2 antibody (HA721210) at 1/1,000 dilution.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 138 kDa
Observed band size: 180 kDa

Exposure time: 2 minutes;

6% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA721210) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

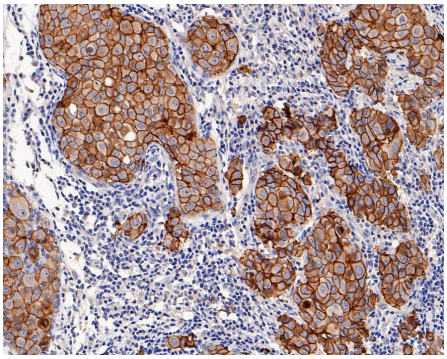

Fig2: Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue with Rabbit anti-HER2 / ErbB2 antibody (HA721210) at 1/4,000 dilution.

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 (HA721210) at 1/4,000 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.

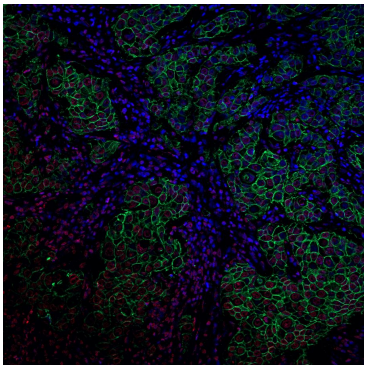

Fig3: Immunofluorescence analysis of paraffin-embedded human breast cancer tissue labeling HER2 / ErbB2 (HA721210, green) and Histone H3 (HA601335, red).

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS. And then probed with the primary antibodies HER2 / ErbB2 (HA721210, green) at 1/200 dilution and Histone H3 (HA601335, red) at 1/200 dilution overnight at 4 ℃, washed with PBS.

iFluor™ 488 conjugate-Goat anti-Rabbit IgG (HA1121) and iFluor™ 594 conjugate-Goat anti-Mouse IgG (HA1126) were used as the secondary antibodies at 1/1,000 dilution. DAPI was used as nuclear counterstain.

Background References

1. Ellis CM, Dyson MJ, Stephenson TJ, Maltby EL. HER2 amplification status in breast cancer: a comparison between immunohistochemical staining and fluorescence in situ hybridisation using manual and automated quantitative image analysis scoring techniques. J Clin Pathol. 2005 Jul;58(7):710-4.
2. Gancberg D, Jarvinen T, di Leo A, Rouas G, Cardoso F, Paesmans M, Verhest A, Piccart MJ, Isola J, Larsimont D. Evaluation of HER-2/NEU protein expression in breast cancer by immunohistochemistry: an interlaboratory study assessing the reproducibility of HER-2/NEU testing. Breast Cancer Res Treat. 2002 Jul;74(2):113-20.