

#12708 Store at -20°C

HER3/ErbB3 (D22C5) XP[®] Rabbit mAb



- Small 100 µl (10 western blots)
- Petite 40 µl (4 western blots)

Orders ■ 877-616-CELL (2355)
orders@cellsignal.com

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rev. 04/06/15

For Research Use Only. Not For Use In Diagnostic Procedures.

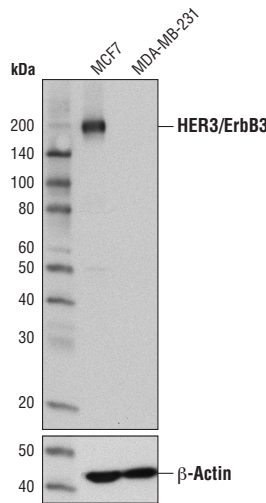
Applications W, IP, IHC-P, IF-IC, F Endogenous	Species Cross-Reactivity* H, (M, R)	Molecular Wt. 185 kDa	Isotype Rabbit IgG**
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Background: HER3/ErbB3 is a member of the ErbB receptor protein tyrosine kinase family, but it lacks tyrosine kinase activity. Tyrosine phosphorylation of ErbB3 depends on its association with other ErbB tyrosine kinases. Upon ligand binding, heterodimers form between ErbB3 and other ErbB proteins, and ErbB3 is phosphorylated on tyrosine residues by the activated ErbB kinase (1,2). There are at least 9 potential tyrosine phosphorylation sites in the carboxy-terminal tail of ErbB3. These sites serve as consensus binding sites for signal transducing proteins, including Src family members, Grb2, and the p85 subunit of PI3 kinase, which mediate ErbB downstream signaling (3). Both Tyr1222 and Tyr1289 of ErbB3 reside within a YXXM motif and participate in signaling to PI3K (4).

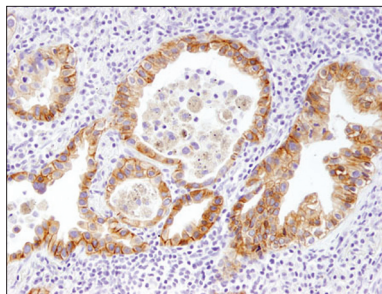
Investigators have found that ErbB3 is highly expressed in many cancer cells (5) and activation of the ErbB3/PI3K pathway is correlated with malignant phenotypes of adenocarcinomas (6). Research studies have demonstrated that in tumor development, ErbB3 may function as an oncogenic unit together with other ErbB members (e.g. ErbB2 requires ErbB3 to drive breast tumor cell proliferation) (7). Thus, investigators view inhibiting interaction between ErbB3 and ErbB tyrosine kinases as a novel strategy for anti-tumor therapy.

Specificity/Sensitivity: HER3/ErbB3 (D22C5) XP[®] Rabbit mAb recognizes endogenous levels of total HER3/ErbB3 protein. This antibody does not cross-react with other HER family proteins.

Source/Purification: Monoclonal antibody is produced by immunizing animals with recombinant protein corresponding to the carboxy terminus of human ErbB3 protein.



Western blot analysis of extracts from MCF7 (HER3+) and MDA-MB-231 (HER3-) cells using HER3/ErbB3 (D22C5) XP[®] Rabbit mAb (upper) or β-Actin (D6A8) Rabbit mAb #8457 (lower).



Immunohistochemical analysis of paraffin-embedded non-small cell lung carcinoma using HER3/ErbB3 (D22C5) XP[®] Rabbit mAb.

Entrez-Gene ID #2065
UniProt ID #P21860

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

***Species cross-reactivity is determined by western blot.**

****Anti-rabbit secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western blotting	1:1000
Immunoprecipitation	1:50
Immunohistochemistry (Paraffin)	1:250†
Unmasking buffer:	EDTA
Antibody diluent:	SignalStain [®] Antibody Diluent #8112
Detection reagent:	SignalStain [®] Boost (HRP, Rabbit) #8114
† Optimal IHC dilutions determined using SignalStain [®] Boost IHC Detection Reagent.	
Immunofluorescence (IF-IC)	1:200
Flow Cytometry	1:100

For product specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

Background References:

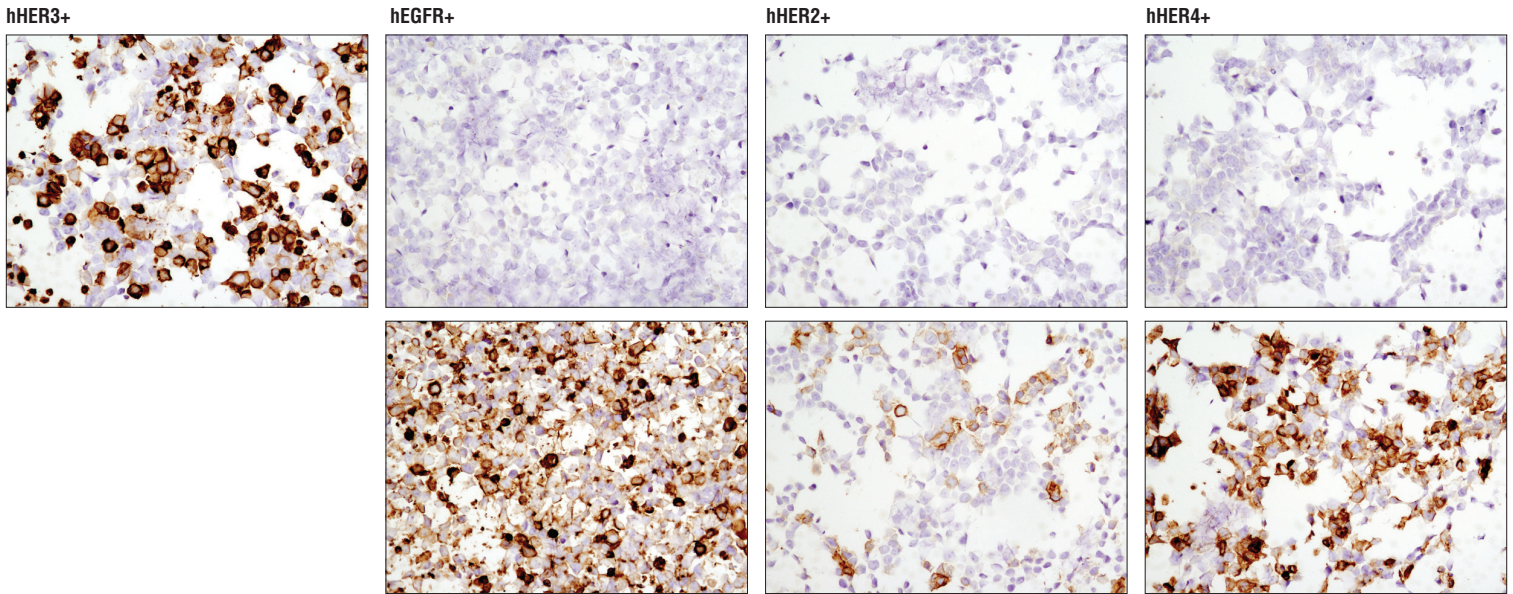
- (1) Yarden, Y. and Sliwkowski, M.X. (2001) *Nature Rev. Mol. Cell. Biol.* 2, 127-137.
- (2) Guy, P.M. et al. (1994) *Proc. Natl. Acad. Sci. USA* 91, 8132-8136.
- (3) Songyang, Z. et al. (1993) *Cell* 72, 767-778.
- (4) Kim, H.H. et al. (1994) *J. Biol. Chem.* 269, 24747-24755.
- (5) Sithanandam, G. et al. (2003) *Carcinogenesis* 24, 1581-1592.
- (6) Kobayashi, M. et al. (2003) *Oncogene* 22, 1294-1301.
- (7) Holbro, T. et al. (2003) *Proc. Natl. Acad. Sci. USA* 100, 8933-8938.

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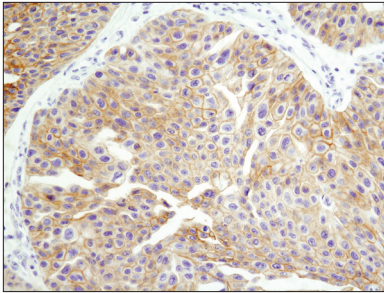
IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween[®] 20 at 4°C with gentle shaking, overnight.

Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide
Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine
 Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—horse AI—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.

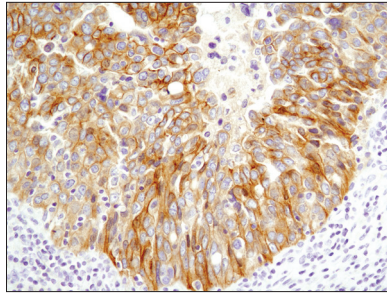
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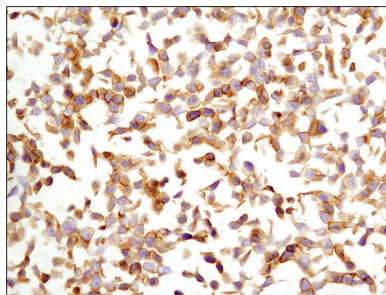
Immunohistochemical analysis of paraffin-embedded 293T cell pellets transfected with human erbB family members HER3, EGFR, HER2 and HER4 (from left to right as indicated) using HER3/ErbB3 (D22C5) XP[®] Rabbit mAb (top panels). Transfections were confirmed using EGF Receptor (D38B1) XP[®] Rabbit mAb #4267 (lower left), HER2/ErbB2 (D8F12) XP[®] Rabbit mAb #4290 (lower middle) and HER4/ErbB4 (111B2) Rabbit mAb #4795 (lower right).



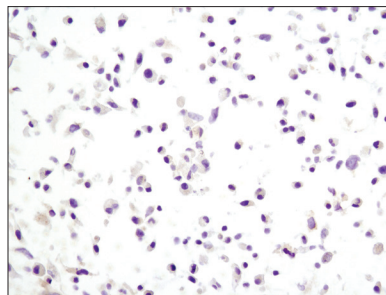
Immunohistochemical analysis of paraffin-embedded breast carcinoma using HER3/ErbB3 (D22C5) XP[®] Rabbit mAb.



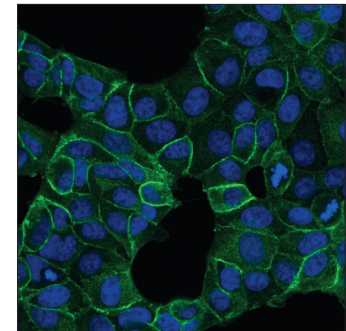
Immunohistochemical analysis of paraffin-embedded ovarian serous adenocarcinoma using HER3/ErbB3 (D22C5) XP[®] Rabbit mAb.



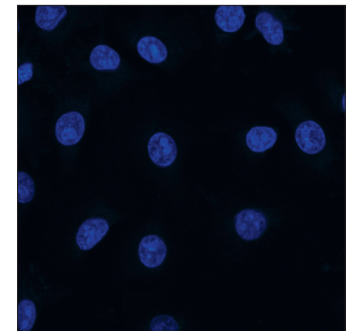
Immunohistochemical analysis using HER3/ErbB3 (D22C5) XP[®] Rabbit mAb on SignalSlide[®] HER3/ErbB3 IHC Controls #12128 (paraffin-embedded MCF7 (HER3+; left) and MDA-MB-231 (HER3-; right) cells).



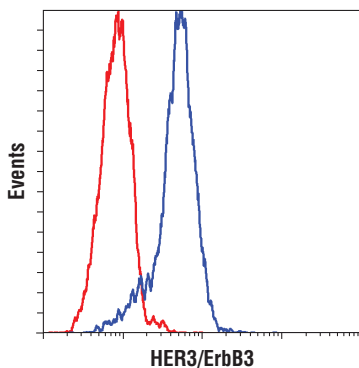
MCF7



MDA-MB-231



Confocal immunofluorescent analysis of MCF7 cells (HER3+, left) and MDA-MB-231 cells (HER3-, right) using HER3/ErbB3 (D22C5) XP[®] Rabbit mAb (green). Blue pseudocolor= DRAQ5[®] #4084 (fluorescent DNA dye).



◀ Flow cytometric analysis of MCF7 cells using HER3/ErbB3 (D22C5) XP[®] Rabbit mAb (blue) compared to Rabbit (DA1E) mAb IgG XP[®] Isotype Control #3900. Anti-rabbit IgG (H+L), F(ab)₂ fragment (Alexa Fluor[®] 488 conjugate) #4412 was used as a secondary antibody.