

#3197 Store at -20°C

EGF Receptor (L858R Mutant Specific) (43B2) Rabbit mAb

100 µl
(10 western blots)



Orders ■ 877-616-CELL (2355)
orders@cellsignaling.com
Support ■ 877-678-TECH (8324)
info@cellsignaling.com
Web ■ www.cellsignaling.com

rev. 02/01/12

This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IHC-P, IF-IC, IF-P, F Endogenous	H	175 kDa	Rabbit IgG**

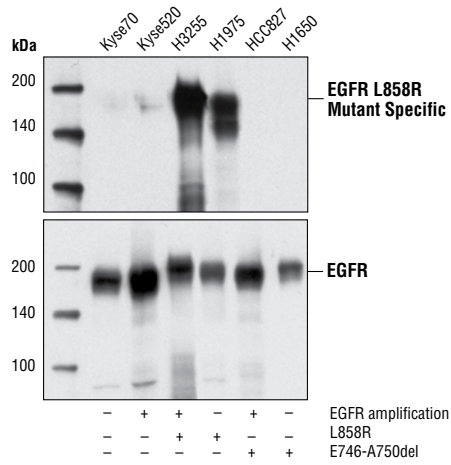
Background: Epidermal growth factor receptor (EGFR) is a 170 kDa transmembrane receptor tyrosine kinase that belongs to the HER/ErbB protein family. Somatic mutations in the tyrosine kinase domain of EGFR are present in a subset of lung adenocarcinomas (1-3). Two types of mutations account for approximately 90% of mutated cases: a specific point mutation L858R which occurs in exon 21 and short in-frame deletions in exon 19 (4,5). A common lesion in exon 19 is the deletion of E746-A750, although other variants occur.

Specificity/Sensitivity: EGFR (L858R Mutant Specific) (43B2) Rabbit mAb detects endogenous levels of EGFR mutant L858R protein. Careful titration of this antibody may be required to obtain optimal specificity.

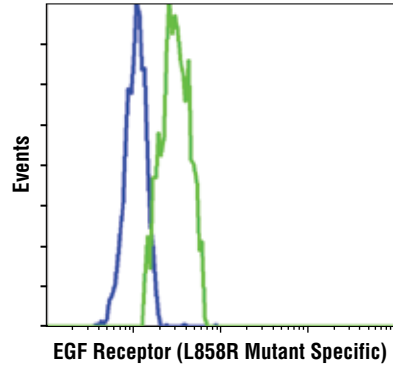
Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to L858R mutant sequence of human EGF receptor.

Background References:

- (1) Lynch, T.J. et al. (2004) *N Engl J Med* 350, 2129-39.
- (2) Pao, W. et al. (2004) *Proc Natl Acad Sci U S A* 101, 13306-11.
- (3) Haber, D.A. et al. (2005) *Cold Spring Harb Symp Quant Biol* 70, 419-26.
- (4) Kosaka, T. et al. (2004) *Cancer Res* 64, 8919-23.
- (5) Riely, G.J. et al. (2006) *Clin Cancer Res* 12, 7232-41.



Western blot analysis of extracts from various EGFR expressing cell lines, showing the specificity of detection of mutant EGFR (L858R) protein using EGF Receptor (L858R Mutant Specific) (43B2) Rabbit mAb (upper), and total EGFR control antibody (lower).



Flow cytometric analysis of two non-small cell lung cancer cell lines using EGF Receptor (L858R Mutant Specific) (43B2) Rabbit mAb. H3255 cells (green) harbor the L858R EGFR mutation while the H1650 cells (blue) have the exon 19 mutation (delE746-A750).

Entrez-Gene ID #1956
Swiss-Prot Acc. #P00533

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	1:200
Immunohistochemistry (Paraffin)	1:100
Unmasking buffer:	EDTA
Antibody diluent:	SignalStain® Antibody Diluent #8112
Immunofluorescence (IF-IC)	1:400
Immunofluorescence (IF-P)	1:800
Flow Cytometry	1:100

For application specific protocols, please see the web page for this product at www.cellsignaling.com

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

The manufacture, use, sale and import of this product is within the scope of one or more intellectual property rights (including patents and patent applications) owned or controlled by Cell Signaling Technology. The purchase of this product conveys to the buyer a non-transferrable right to use the purchased product only in research conducted by the buyer. The sale of the product is expressly conditioned on the buyer not using the products or its components (1) to analyze or reverse engineer the product for its chemical/physical properties and composition (including e.g., identification of the sequence); (2) in manufacturing; (3) to provide a service, information, or data to an unaffiliated third party for payment; (4) for therapeutic, diagnostic or prophylactic purposes; (5) resale, whether or not such product are resold for use in research; or for any other commercial purpose. For information on purchasing a license to this product for purposes other than research, contact Cell Signaling Technology, Inc. Business Development at BUSDEV@CELLSIGNAL.COM.

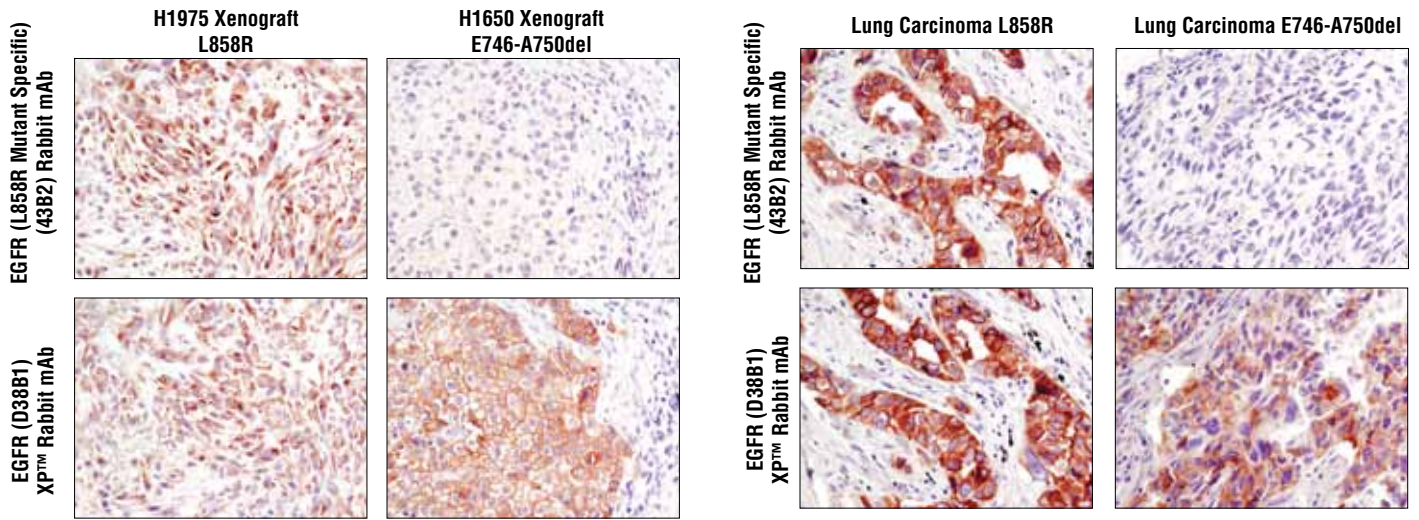
Rabbit monoclonal antibody produced under license (granting certain rights including those under U. S. Patents No. 5,675,063 and 7,429,487) from Epitomics, Inc.

DRAQ5® is a registered trademark of Biostatus Limited.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

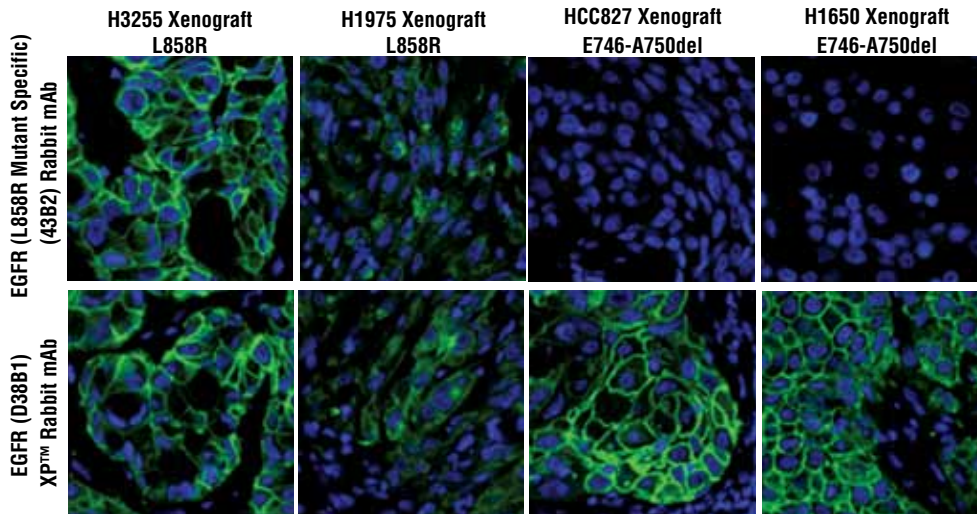
Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide
Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine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

© 2012 Cell Signaling Technology, Inc. Patent pending. XP™ and SignalStain® are trademarks of Cell Signaling Technology, Inc.

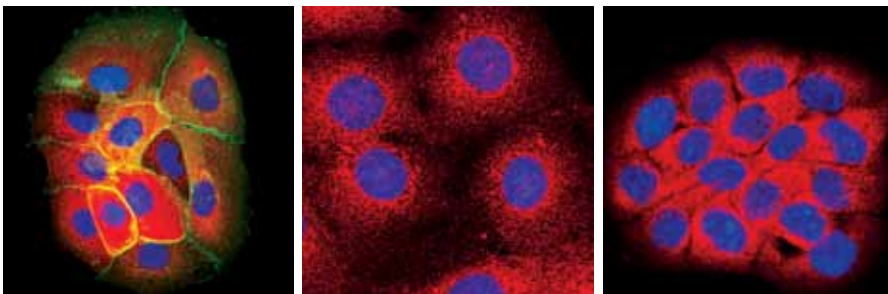


Immunohistochemical analysis of paraffin-embedded H1975 (left, EGFR L858R positive) and H1650 xenograft (right, EGFR deletion positive) using EGF Receptor (L858R Mutant Specific) (43B2) Rabbit mAb (upper) and total EGFR (D38B1) XP™ Rabbit mAb #4267 (lower).

Immunohistochemical analysis of paraffin-embedded human lung carcinoma of known mutational status using EGF Receptor (L858R Mutant Specific) (43B2) Rabbit mAb (upper) and total EGFR (D38B1) XP™ Rabbit mAb #4267 (lower). EGFR L858R positive lung (left), EGFR deletion positive lung sections (right).



Confocal immunofluorescent analysis of paraffin-embedded H3255, H1975, HCC827 and H1650 xenografts using EGF Receptor (L858R Mutant Specific) (43B2) Rabbit mAb (upper, green) and EGFR (D38B1) XP™ Rabbit mAb #4267 (lower, green). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).



Confocal immunofluorescent analysis of H3255 (left), HCC827 (middle) and A431 cells (right) using EGF Receptor (L858R Mutant Specific) (43B2) Rabbit mAb (green) and MEK1/2 (L38C12) Mouse mAb #4694 (red). H3255 cells have the L858R EGFR mutation, HCC827 cells have an exon 19 EGFR deletion, and A431 cells overexpress wild type EGFR. Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).