Store at 4°C

#9415

Phospho-Tyrosine Mouse mAb (P-Tyr-100) (Alexa Fluor® 647 Conjugate)

✓ 100 µl
(50 tests)

New more concentrated formulation

rev.03/17/11

Cell Signaling TECHNOLOGY®

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

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	Source	Isotype
IF-IC, F	All*	Mouse	IgG1

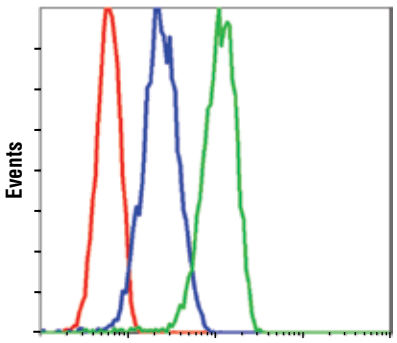
Description: Cell Signaling Technology Antibody conjugated to Alexa Fluor® 647 fluorescent dye and tested in-house for direct flow cytometric analysis of human cells.

*The unconjugated antibody, #9411, reacts with phospho-tyrosine in all species tested. CST expects that Phospho-Tyrosine Mouse mAb (P-Tyr-100) (Alexa Fluor® 647 conjugate) will also recognize phospho-tyrosine in all species.

Background: Tyrosine phosphorylation plays a key role in cellular signaling (1). In cancer, unregulated tyrosine kinase activity can drive malignancy and tumor formation by generating inappropriate proliferation and survival signals (2). Antibodies specific for phospho-tyrosine (3,4) have been invaluable reagents in these studies. The phospho-tyrosine Mouse mAbs developed by CST (P-Tyr-100, #9411 and P-Tyr-102, #9416) provide exceptionally sensitive new tools of increased utility for studying tyrosine phosphoyrlation and monitoring tyrosine kinase activity in high throughput drug discovery.

Specificity/Sensitivity: Phospho-Tyrosine Mouse mAb (P-Tyr-100) is a high affinity antibody. ELISAs against a wide variety of phosphopeptides indicate that (a) P-Tyr-100 binds phospho-Tyr in a manner largely independent of the surrounding amino acid sequence. 2D gel Western blot analysis of pervanadate-treated cell extracts also shows that P-Tyr-100 interacts with a broad range of tyrosine-phosphorylated proteins. P-Tyr-100 does not cross-react with peptides containing phospho-Ser or phospho-Thr. (U.S. Patent No's.: 6,441,140; 6,982,318; 7,259,022; 7,344,714; U.S.S.N. 11,484,485; and all foreign equivalents.)

Source/Purification: Monoclonal antibody is derived from animals immunized with phospho-tyrosine-containing peptides. The antibody was conjugated to Alexa Fluor® 647 under optimal conditions with an F/P ratio of 2–6.



Flow cytometric analysis of K562 cells, untreated (green) or Gleevec®- treated (blue), using Phospho-Tyrosine Mouse mAb (P-Tyr-100) (Alexa Fluor® 647 Conjugate) compared with a nonspecific negative control antibody (red).

Background References:

(1) Schlessinger, J. (2000) *Cell* 103, 211–225.

(2) Blume-Jensen, P. and Hunter, T. (2001) *Nature* 411, 355–365.

(3) Ward, S.G. et al. (1992) *J. Biol. Chem.* 267, 23862–23869.

(4) Glenney, J.R. et al. (1988) *J. Immunol. Methods.* 109, 277–285.

Storage: Supplied in PBS (pH 7.2), less than 0.1% Sodium azide, 2 mg/ml BSA. Store at 4°C. *Protect from light. Do not freeze.*

Recommended Antibody Dilutions:

Immunofluorescence (IF-IC)	1:50
Flow Cytometry	1:50

For application 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.

License/Use Restrictions: *Use of CST Motif Antibodies within certain methods (e.g., U.S. Patent No.'s 7,198,896 & 7,300,753) may require a license from CST. For information regarding academic licensing terms please have your technology transfer office contact CST Legal Department at CST_ip@cellsignal.com. For information regarding commercial licensing terms please contact CST Pharma Services Department at ptmscan@cellsignal.com.*

The Alexa Fluor® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc., for research use only, except for use in combination with DNA microarrays. The Alexa Fluor® dyes (except for Alexa Fluor® 430 dye) are covered by pending and issued patents.

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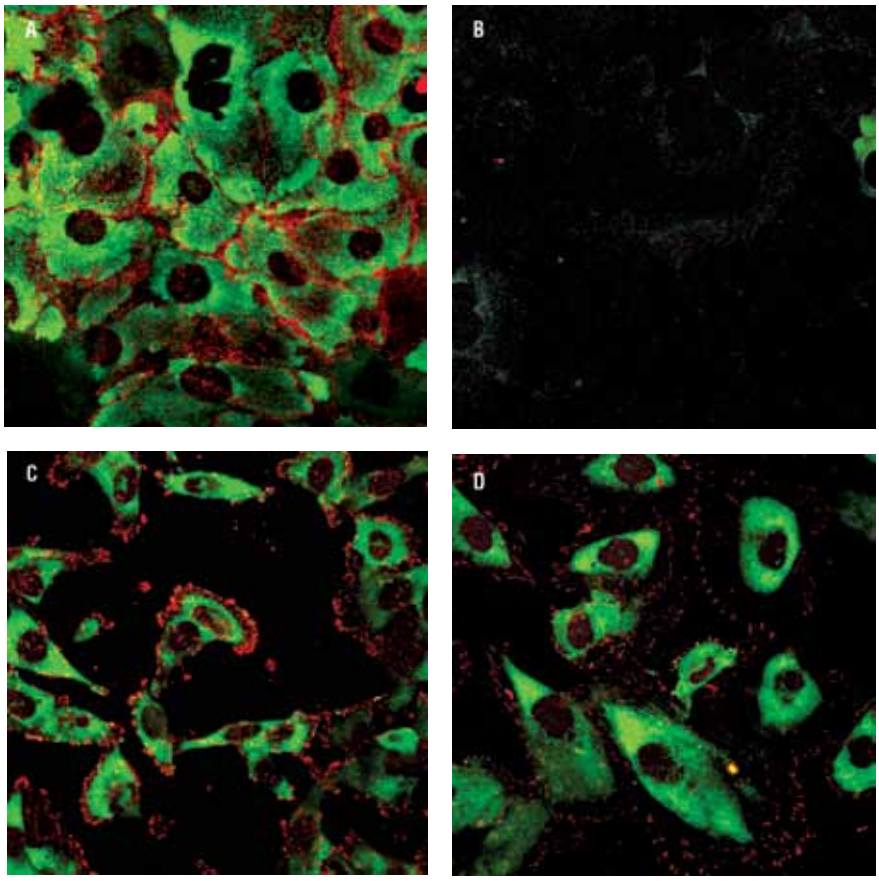
Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide

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



Confocal immunofluorescent image of two Gefinitib (Iressa) treated non-small cell lung cancer cell lines. HCC827 cells have the E746_A750 deletion in exon 19 of the EGFR gene, and are highly sensitive to Gefitinib. H1975 cells have the (T790M) mutation that confers Gefitinib-resistance. Both cell lines were treated and then double-labeled with Phospho-S6 Ribosomal Protein Rabbit mAb (Alexa Fluor® 488 Conjugate) #4854 and Phospho-Tyrosine Mouse mAb (Alexa Fluor® 647 Conjugate) #9415. Untreated HCC827 (A) and H1975 (C) cells show bright phospho-S6 (green) and phospho-tyrosine (red pseudocolor) label. Phospho-S6 and phospho-tyrosine signals dramatically decrease following Gefitinib treatment in HCC827 cells (B), with little or no change in the Gefitinib-resistant H1975 cells (D).

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