#12290 Store at -20°C

RCAS1 (D2B6N) XP® Rabbit mAb

☐ Small 100 µl
(10 western blots)

☐ Petite 40 µl
(4 western blots)

Cell Signaling TECHNOLOGY®

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rev. 01/05/15

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

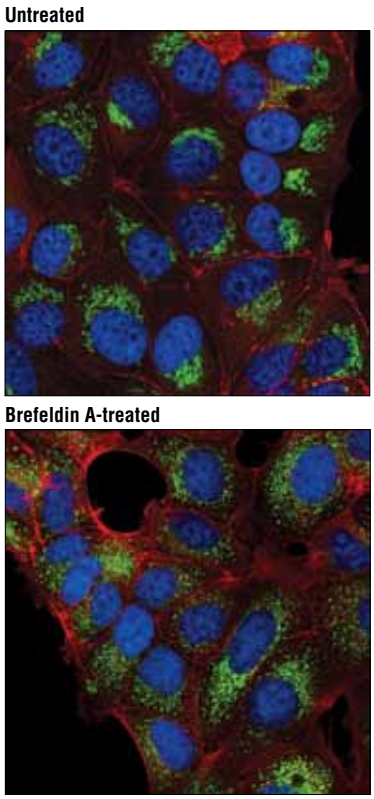
Entrez-Gene ID #9166
UniProt ID #O00559

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IF-IC, F Endogenous	H, M, R, (Mk)	32 kDa	Rabbit IgG**

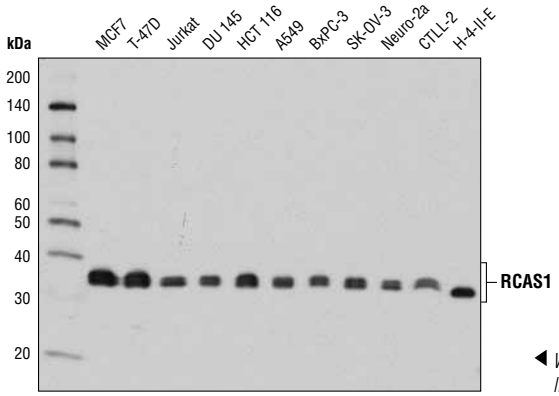
Background: Receptor binding cancer antigen expressed on SiSo cells (RCAS1) is also known as estrogen receptor-binding fragment-associated gene 9 (EBAG9). Originally identified as an estrogen-inducible gene (1), RCAS1 was recently found to play a novel role in the adaptive immune response by negatively regulating the cytolytic activity of cytotoxic T lymphocytes (CTLs) (2). RCAS1 is conserved in phylogeny and is ubiquitously expressed in most human tissues and cells (3,4). There is evidence that tissue expression of RCAS1 is increased in a variety of malignancies, including cancers of the gastrointestinal tract, liver, lung, breast, ovary, endometrium, and cervix. Research studies have shown that levels of RCAS1 tissue expression are negatively correlated with the prognosis of patients harboring the aforementioned malignancies (4). It is also noteworthy that elevated levels of RCAS1 have been detected in the sera of cancer patients (4). Initial studies indicated that RCAS1 was secreted from cancer cells and functioned as a ligand for a putative receptor expressed on NK cells, as well as T and B lymphocytes, inducing their apoptosis, which enabled cancer cells to evade immune surveillance (5,6). Subsequent studies have identified RCAS1 as a type III transmembrane Golgi protein with the ability to regulate vesicle formation, secretion, and protein glycosylation (2,7-9). Indeed, it has been shown that RCAS1 overexpression negatively regulates the cytolytic function of CTLs by negatively regulating protein trafficking from the *trans*-Golgi to secretory lysosomes (2). Furthermore, RCAS1 overexpression delays vesicle transport from the ER to Golgi and causes components of the ER quality control and glycosylation machinery to mislocalize. As a consequence, RCAS1 induces the deposition of tumor-associated glycan antigens on the cell surface, which are thought to contribute to tumor pathogenesis through the mediation of adhesion, invasion, and metastasis (8,9).

Specificity/Sensitivity: RCAS1 (D2B6N) XP® Rabbit mAb recognizes endogenous levels of total RCAS1 protein.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Gly147 of human RCAS1 protein.



◀ *Confocal immunofluorescent analysis of MCF7 cells, untreated (upper) or treated with Brefeldin A #9972 (5 µg/ml, 1 hr; lower), using RCAS1 (D2B6N) XP® Rabbit mAb (green). Actin filaments were labeled with DyLight™ 554 Phalloidin #13054 (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).*



◀ *Western blot analysis of extracts from various cell lines using RCAS1 (D2B6N) XP® Rabbit mAb.*

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	1:100
Immunofluorescence (IF-IC)	1:100
Flow Cytometry	1:100

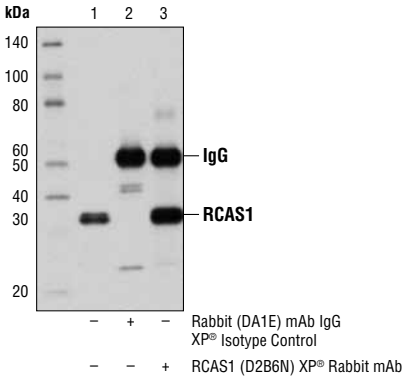
For product specific protocols please see the web page for this product at www.cellsignal.com.

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

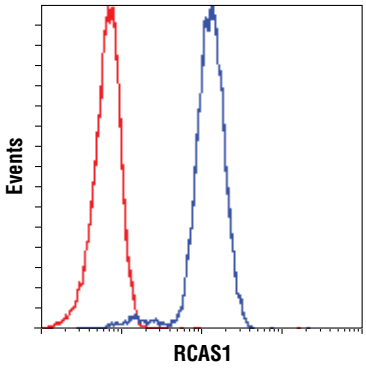
IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween®20 at 4°C with gentle shaking, overnight.

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

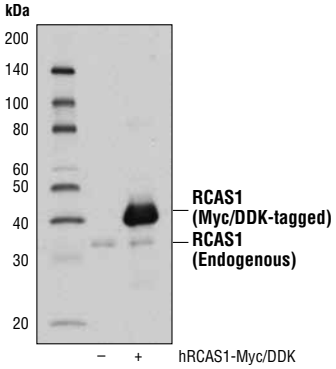
Species Cross-Reactivity Key: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



Immunoprecipitation of RCAS1 from HCT 116 extracts using Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (lane 2) or RCAS1 (D2B6N) XP® Rabbit mAb (lane 3). Lane 1 is 10% input. Western blot analysis was performed using RCAS1 (D2B6N) XP® Rabbit mAb.



Flow cytometric analysis of Jurkat cells using RCAS1 (D2B6N) XP® Rabbit mAb (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (red).



Western blot analysis of extracts from 293T cells, mock transfected (-) or transfected with a construct expressing Myc/DDK-tagged full-length human RCAS1 (hRCAS1-Myc/DDK; +), using RCAS1 (D2B6N) XP® Rabbit mAb.

Background References:

(1) Watanabe, T. et al. (1998) *Mol Cell Biol* 18, 442-9.

(2) Rüder, C. et al. (2009) *J Clin Invest* 119, 2184-203.

(3) Tsuchiya, F. et al. (2001) *Biochem Biophys Res Commun* 284, 2-10.

(4) Giaginis, C. et al. (2009) *Histol Histopathol* 24, 761-76.

(5) Matsushima, T. et al. (2001) *Blood* 98, 313-21.

(6) Nakashima, M. et al. (1999) *Nat Med* 5, 938-42.

(7) Reimer, T.A. et al. (2005) *BMC Cancer* 5, 47.

(8) Wolf, J. et al. (2010) *FASEB J* 24, 4000-19.

(9) Engelsberg, A. et al. (2003) *J Biol Chem* 278, 22998-3007.