Store at -20°C

#35869

RPA32/RPA2 (E8X5P) XP® Rabbit mAb

Cell Signaling TECHNOLOGY®

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Entrez-Gene ID #6118
UniProt ID #P15927

New 01/19

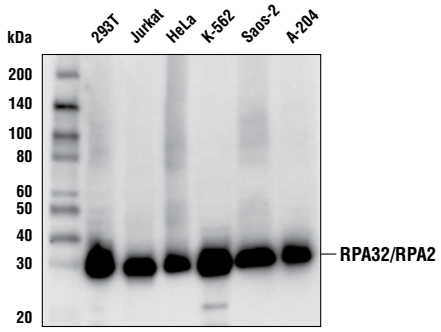
For Research Use Only. Not For Use In Diagnostic Procedures.

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IHC-P, IF-IC	H	32 kDa	Rabbit IgG**
Endogenous			

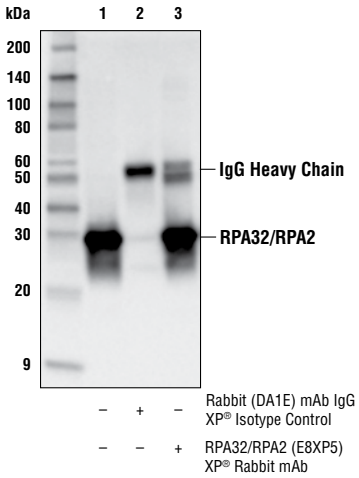
Background: RPA70 (HSSB, REPA1, RF-A, RP-A, p70) is a component of a heterotrimeric complex, composed of 70, 32/30 and 14 kDa subunits, collectively known as RPA. RPA is a single stranded DNA binding protein, whose DNA binding activity is believed to reside entirely in the 70 kDa subunit. The complex is required for almost all aspects of cellular DNA metabolism such as DNA replication (1-3), recombination, cell cycle and DNA damage checkpoints, and all major types of DNA repair including nucleotide excision, base excision, mismatch and double-strand break repairs (4-7). In response to genotoxic stress in eukaryotic cells, RPA has been shown to associate with the Rad9/Rad1/Hus1 (9-1-1) checkpoint complex (8). RPA is hyperphosphorylated upon DNA damage or replication stress by checkpoint kinases including ataxia telangiectasia mutated (ATM), ATM and Rad3-related (ATR), and DNA-dependent protein kinase (DNA-PK) (9-11). Phosphorylation of RPA32 occurs at serines 4, 8 and 33 (11). Hyperphosphorylation may alter RPA-DNA and RPA-protein interactions. In addition to the checkpoint partners, RPA interacts with a wide variety of protein partners, including proteins required for normal replication such as RCF, PCNA and Pol α, and also proteins involved in SV40 replication, such as DNA polymerase I and SV40 large T antigen (10,12).

Specificity/Sensitivity: RPA32/RPA2 (E8X5P) XP® Rabbit mAb recognizes endogenous levels of total RPA32 protein.

Source/Purification: Monoclonal antibody is produced by immunizing animals with recombinant protein specific to the amino terminus of human RPA32 protein.



Western blot analysis of extracts from various cell lines using RPA32/RPA2 (E8X5P) XP® Rabbit mAb.



Immunoprecipitation of RPA32/RPA2 protein from HeLa cell extracts. Lane 1 is 10% input, lane 2 is Rabbit (DA1E) mAb IgG XP® Isotype Control #3900, and lane 3 is RPA32/RPA2 (E8X5P) XP® Rabbit mAb. Western blot analysis was performed using RPA32/RPA2 (E8X5P) 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	1:50
Immunohistochemistry (Paraffin)	1:600
Optimal IHC dilutions determined using SignalStain® Boost IHC Detection Reagent.	
Unmasking buffer: SignalStain® Citrate Unmasking Solution (10X) #14746	
Antibody diluent: SignalStain® Antibody Diluent #8112	
Detection reagent: SignalStain® Boost (HRP, Rabbit) #8114	
Immunofluorescence (IF-IC)	1:800
Fixative:	4% Formaldehyde
Permeabilization:	0.3% Triton X-100

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com.

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.

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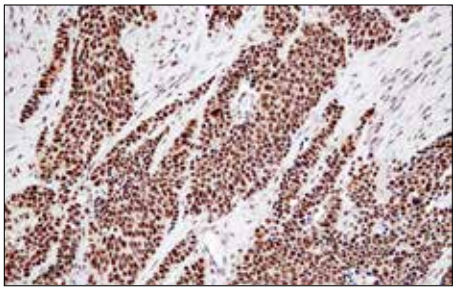
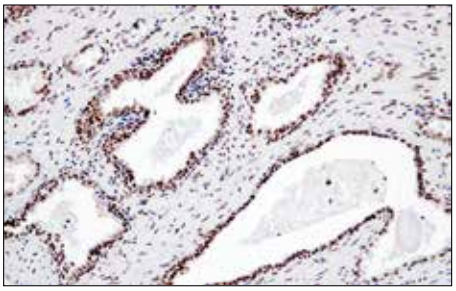
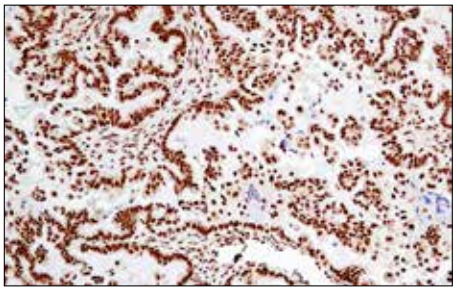
Applications: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide **Species Cross-Reactivity:** **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **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.



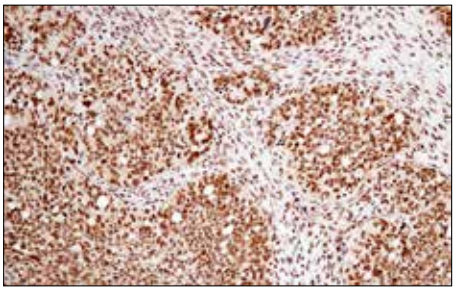
Immunohistochemical analysis of paraffin-embedded human colon carcinoma using RPA32/RPA2 (E8X5P) XP® Rabbit mAb.



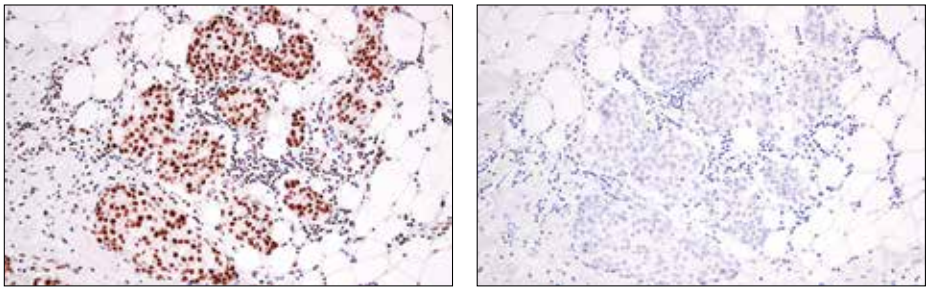
Immunohistochemical analysis of paraffin-embedded human prostate carcinoma using RPA32/RPA2 (E8X5P) XP® Rabbit mAb.



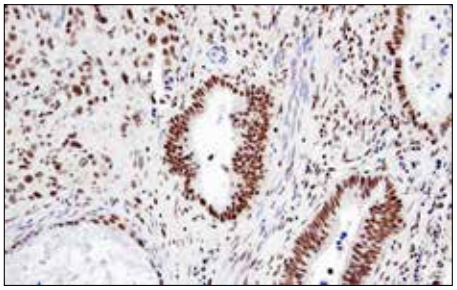
Immunohistochemical analysis of paraffin-embedded human ovarian clear cell carcinoma using RPA32/RPA2 (E8X5P) XP® Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded human serous papillary carcinoma of the ovary using RPA32/RPA2 (E8X5P) XP® Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded human ductal breast carcinoma using RPA32/RPA2 (E8X5P) XP® Rabbit mAb (left) compared to concentration matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (right).



Immunohistochemical analysis of paraffin-embedded human esophageal carcinoma using RPA32/RPA2 (E8X5P) XP® Rabbit mAb.

Background References:

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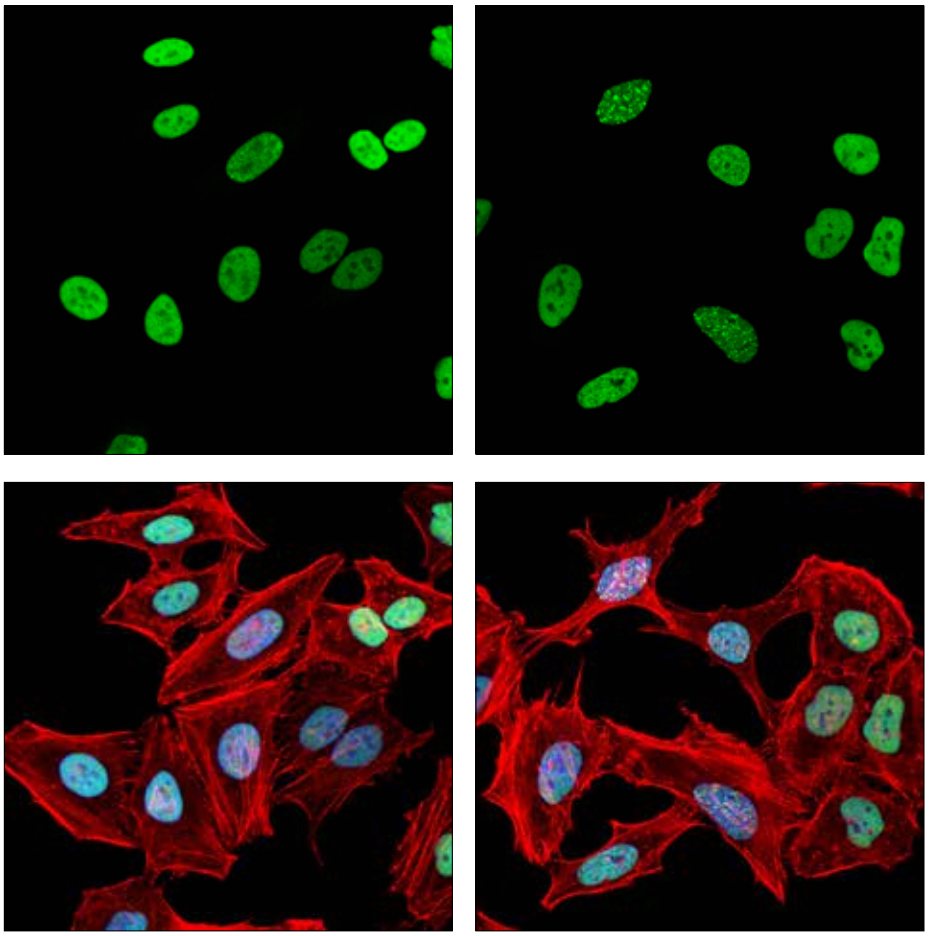

Confocal immunofluorescent analysis of HeLa cells, untreated (left) or UV-treated (right), using RPA32/RPA2 (E8X5P) XP® Rabbit mAb (green) showing translocation to distinct nuclear foci after UV-induced damage. Actin filaments were labeled with DyLight™ 554 Phalloidin #13054 (red). Samples were mounted in ProLong® Gold Antifade Reagent with DAPI #8961 (blue).