

# RbBP5 Antibody

Rabbit Polyclonal

Antigen Affinity Purified	RefSeq ID	NP_005048.2
Catalog No. A300-109A	Uniprot ID	Q15291
Lot No. 5	GeneID	5929

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<b>APPLICATIONS</b>	WB, IP, IHC, ICC, ChIP, ChIP-chip, ChIP-Seq
<b>SPECIES REACTIVITY</b>	Human, Mouse
<b>AMOUNT</b>	100 µl
<b>CONCENTRATION</b>	1000 µg/ml
<b>STORAGE/SHELF LIFE</b>	2 – 8°C / 1 year from date of receipt
<b>PHYSICAL STATE</b>	Liquid
<b>BUFFER</b>	Tris-citrate/phosphate buffer, pH 7 to 8 containing 0.09% Sodium Azide
<b>ISOTYPE</b>	IgG
<b>ORIGIN</b>	USA
<b>PRODUCTION PROCEDURES</b>	Antibody was affinity purified using an epitope specific to RbBP5 immobilized on solid support.

The epitope recognized by A300-109A maps to a region between residue 500 and the C-terminus (residue 538) of human retinoblastoma binding protein 5 using the numbering given in entry NP\_005048.2 (GeneID 5929).

Immunoglobulin concentration was determined by extinction coefficient: absorbance at 280 nm of 1.4 equals 1.0 mg of IgG.

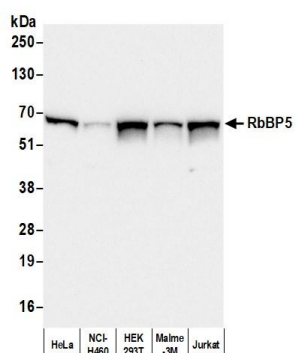
<b>APPLICATIONS</b>	Centrifuge tube to remove product from lid. Optimal working dilutions should be determined experimentally by the investigator. Prepare working dilution immediately before use.
Western Blot	1:10,000 – 1:25,000
Immunoprecipitation	2 – 10 µg/mg lysate
Immunohistochemistry	1:1,000 – 1:5,000. Epitope retrieval with citrate buffer pH6.0 is recommended for FFPE tissue sections.
Immunocytochemistry	1:250 – 1:1,000
ChIP	1 – 3 µg as per Dou et al., Nat Struct Mol Biol 13 (8):713-719, 2006. Previous lots of this antibody have performed in this application.
ChIP-chip	10 µg. Previous lots of this antibody have performed in this application.
ChIP-Seq	2 µg. Previous lots of this antibody have performed in this application.

**IHC HUMAN CONTROLS** Breast Carcinoma, Colon Carcinoma, Ovarian Carcinoma, Prostate Carcinoma

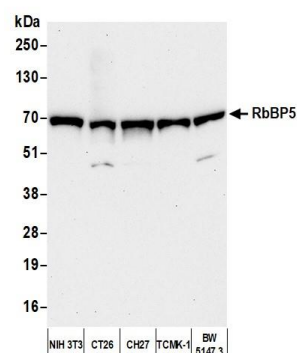
**ADDITIONAL INFO** <https://www.fortislifecom.com/p/A300-109A>

Use the link above to view SDS, a current list of citations, and other product specific information.

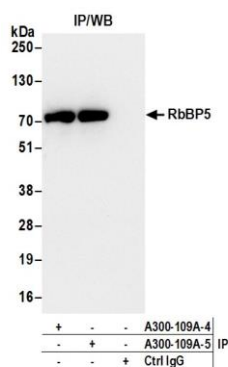
This document certifies that this product has met all of the quality control standards defined by Bethyl Laboratories, Inc.  
Michael Spencer, PhD Date: May 31, 2023



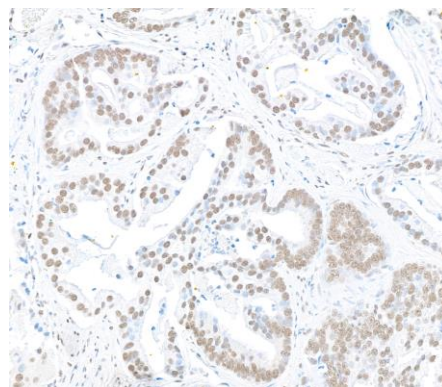
**Detection of human RbBP5 by western blot.** *Samples:* Whole cell lysate (25  $\mu$ g) from HeLa, NCI-H460, HEK293T, Malme-3M, and Jurkat cells prepared using NETN lysis buffer. *Antibody:* Affinity purified rabbit anti-RbBP5 antibody (A300-109A lot 5) used for WB at 0.04  $\mu$ g/ml. *Detection:* Chemiluminescence with an exposure time of 10 seconds.



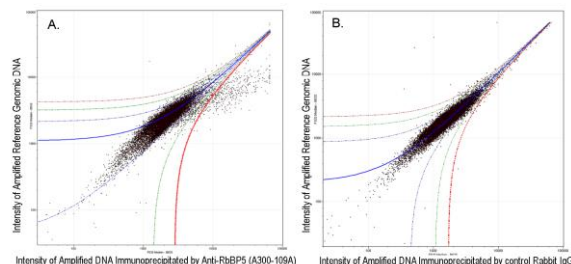
**Detection of mouse RbBP5 by western blot.** *Samples:* Whole cell lysate (25  $\mu$ g) from NIH 3T3, CT26, CH27, TCMK-1, and BW5147.3 cells prepared using NETN lysis buffer. *Antibody:* Affinity purified rabbit anti-RbBP5 antibody (A300-109A lot 5) used for WB at 0.04  $\mu$ g/ml. *Detection:* Chemiluminescence with an exposure time of 75 seconds.



**Detection of human RbBP5 by western blot of immunoprecipitates.** *Samples:* Whole cell lysate (1.0 mg per IP reaction; 20% of IP loaded) from HeLa cells prepared using NETN lysis buffer. *Antibodies:* Affinity purified rabbit anti-RbBP5 antibody (A300-109A lot 5) used for IP at 6  $\mu$ g per reaction. RbBP5 was also immunoprecipitated by a previous lot of this antibody (A300-109A lot 4). For blotting immunoprecipitated RbBP5, A300-109A was used at 0.04  $\mu$ g/ml. *Detection:* Chemiluminescence with an exposure time of 10 seconds.

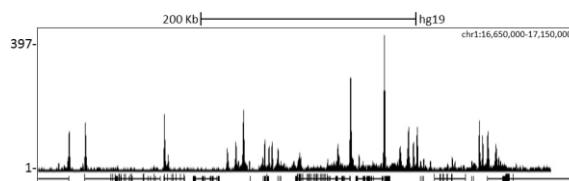


**Detection of human RbBP5 by immunohistochemistry.** *Sample:* FFPE section of human prostate carcinoma. *Antibody:* Affinity purified rabbit anti-RbBP5 (A300-109A lot 4) used at a dilution of 1:5,000 (0.2  $\mu$ g/ml). *Detection:* DAB



### ChIP–chip scatter plot of anti–RbBP5 (A300–109A) enriched DNA binding sites versus input reference DNA.

A. 10 µg of A300–109A was used to immunoprecipitate chromatin from K–562 cells according to Ren et al (Genes Dev. 2002 16: 245–256). immunoprecipitated DNA and reference DNA were amplified via ligation–mediated PCR and the products labeled with fluorescent dNTPs. The labeled ChIP and reference DNA were pooled, hybridized to a DNA microarray, and analyzed. Data points below the +3 SD curve (red line) represent significantly enriched binding sites. B. As a control, a similar experiment was performed using normal rabbit IgG. Compared to the anti–RbBP5 ChIP, normal rabbit IgG showed little enrichment.



### Localization of RbBP5 Binding Sites by ChIP–sequencing.

Chromatin from K562 cells was immunoprecipitated with anti–RbBP5 antibody A300–109A and analyzed by DNA sequencing. The figure illustrates the peak distribution of RbBP5 binding within a 500 Kb region of chromosome 1 as detected using anti–RbBP5 antibody A300–109A. ChIP–seq validation performed by Diogenode, Denville, NJ.