IRAK1 Recombinant Rabbit Monoclonal Antibody (19H32L12)

Product Details	
Size	100 µg
Species Reactivity	Human, Mouse
Host/Isotype	Rabbit / IgG
Expression system	Expi293
Class	Recombinant Monoclonal
Type	Antibody
Clone	19H32L12
Conjugate	Unconjugated
Immunogen	A peptide corresponding to amino acids 579-591 of human IRAK1
Form	Liquid
Concentration	0.5 mg/mL
Purification	Protein A
Storage buffer	PBS
Contains	0.09% sodium azide
Storage conditions	Store at 4°C short term. For long term storage, store at -20°C, avoiding freeze/thaw cycles.
RRID	AB_2532270

Applications	Tested Dilution	Publications
Western Blot (WB)	1:1,000	-
Immunohistochemistry (Paraffin) (IHC (P))	1:10-1:50	-
ChIP assay (ChIP)	1 µL	-

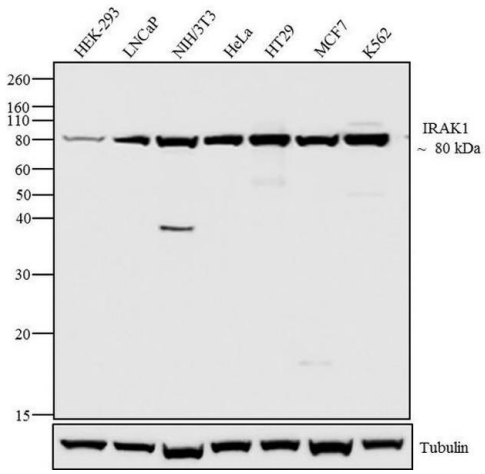
Product Specific Information

This antibody is predicted to react with mouse based on sequence homology.

Intact IgG appears on a non-reducing gel as ~150 kDa band and upon reduction generating a ~25 kDa light chain band and a ~50 kDa heavy chain.

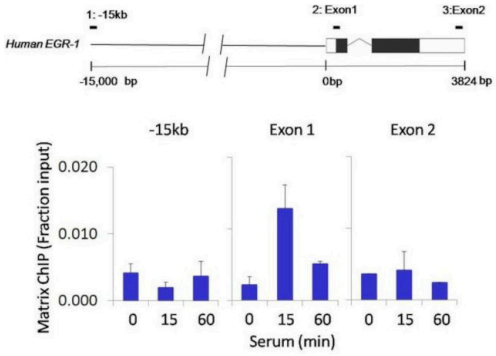
Recombinant rabbit monoclonal antibodies are produced using in vitro expression systems. The expression systems are developed by cloning in the specific antibody DNA sequences from immunoreactive rabbits. Then, individual clones are screened to select the best candidates for production. The advantages of using recombinant rabbit monoclonal antibodies include: better specificity and sensitivity, lot-to-lot consistency, animal origin-free formulations, and broader immunoreactivity to diverse targets due to larger rabbit immune repertoire.

Product Images For IRAK1 Recombinant Rabbit Monoclonal Antibody (19H32L12)



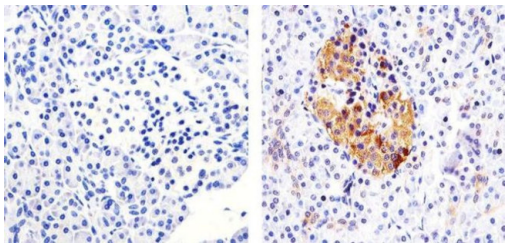
IRAK1 Antibody (700025) in WB

Western blot analysis of IRAK1 was performed by loading 20 µg of HEK-293 (lane1), LNCaP (lane2), NIH/3T3 (lane3), HeLa (lane4), HT-29 (lane5), MCF7 (lane6) and K562 (lane7) cell lysate using Novex®NuPAGE®4-12 % Bis-Tris gel (Product # NP0322BOX), XCell SureLock™ Electrophoresis System (Product # EI0002), Novex® Sharp Pre-Stained Protein Standard (Product # LC5800), and iBlot® Dry Blotting System (Product # IB21001). Proteins were transferred to a nitrocellulose membrane and blocked with 5 % skim milk for 1 hour at room temperature. IRAK1 was detected at ~ 80 kDa using IRAK1 Recombinant Rabbit Monoclonal Antibody (Product # 700025) at 1:1000 dilution in 5 % skim milk at 4° C overnight on a rocking platform. Goat anti-Rabbit IgG - HRP Secondary Antibody (Product # G-21234) at 1:5000 dilution was used and chemiluminescent detection was performed using Pierce™ ECL Western blotting Substrate (Product # 32106).



IRAK1 Antibody (700025) in ChIP

Chromatin immunoprecipitation analysis of IRAK-1 performed using cross-linked chromatin from 1 x 10^6 HCT116 human colon carcinoma cells treated with serum for 0, 15, and 60 minutes. Immunoprecipitation was performed using a multiplex microplate Matrix ChIP assay (see reference for Matrix ChIP protocol: http://www.ncbi.nlm.nih.gov/pubmed/22098709) with 1.0 µL/100 µL well volume of an IRAK-1 rabbit monoclonal antibody (Product # 700025). Chromatin aliquots from ~1 x 10^5 cells were used per ChIP pull-down. Quantitative PCR data were done in quadruplicate using 1 µL of eluted DNA in 2 µL SYBR real-time PCR reactions containing primers to amplify -15kb upstream of the human Egr-1 locus, or exon-1 or exon-2 of Egr-1. PCR calibration curves were generated for each primer pair from a dilution series of sheared total genomic DNA. Quantitation of immunoprecipitated chromatin is presented as signal relative to the total amount of input chromatin. Results represent the mean +/- SEM for three experiments. A schematic representation of the Egr-1 locus is shown above the data where boxes represent exons (black boxes = translated regions, white boxes = untranslated regions), the zigzag line represents an intron, and the straight line represents upstream sequence. Regions amplified by Egr-1 primers are represented by black bars. Data courtesy of the Innovators Program.



IRAK1 Antibody (700025) in IHC (P)

Immunohistochemistry analysis of IRAK1 showing staining in the cytoplasm of paraffin-embedded human pancreas tissue (right) compared to a negative control without primary antibody (left). To expose target proteins, antigen retrieval was performed using 10mM sodium citrate (pH 6.0), microwaved for 8-15 min. Following antigen retrieval, tissues were blocked in 3% H2O2-methanol for 15 min at room temperature, washed with ddH2O and PBS, and then probed with a IRAK1 Recombinant Rabbit Monoclonal Antibody (Product # 700025) diluted in 3% BSA-PBS at a dilution of 1:20 overnight at 4°C in a humidified chamber. Tissues were washed extensively in PBST and detection was performed using an HRP-conjugated secondary antibody followed by colorimetric detection using a DAB kit. Tissues were counterstained with hematoxylin and dehydrated with ethanol and xylene to prep for mounting.