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Datasheet for ABIN129671 anti-Histone H2A Variant (HIS2AV) (Internal Region), (pSer137) antibody

27

Publications



Overview

10

Images

Quantity:	100 µg
Target:	Histone H2A Variant (HIS2AV)
Binding Specificity:	Internal Region, pSer137
Reactivity:	Drosophila melanogaster
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	Un-conjugated
Application:	ELISA, Western Blotting (WB)
Product Details	
Immunogen:	Histone H2AvD pS137 Antibody was prepared from whole rabbit serum produced by repeated
	immunizations with a synthetic peptide corresponding to amino acids 132-141 of Drosophila
	melanogaster (fruit fly) H2AvD protein.
	Immunogen Type: Peptide
Isotype:	lgG
Specificity:	This affinity purified anti-Histone H2AvD pS137 Antibody is directed against the phosphorylated
	form of Drosophila H2AvD protein at the pS137 residue. The product was affinity purified from
	monospecific antiserum by immunoaffinity purification. Antiserum was first purified against the
	phosphorylated form of the immunizing peptide. The resultant affinity purified antibody was
	then cross-adsorbed against the non-phosphorylated form of the immunizing peptide.
	Reactivity occurs against Drosophila H2AvD pS137 protein and the antibody is specific for the
	phosphorylated form of the protein. Reactivity with non-phosphorylated Drosophila H2AvD is

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	minimal by ELISA. A BLAST analysis was used to suggest little to no cross reactivity with
	H2AvD proteins from other sources based on a comparison using the immunizing sequence.
	Reactivity against homologues from other sources is not known.
Characteristics:	Variant histones H2A are synthesized throughout the cell cycle and are very different from
	classical S-phase regulated H2A. H2AvD is vital for viability, but the exact function of variant
	histones H2A is not known. H2A is a core component of the nucleosome, an octamer
	containing two molecules each of H2A, H2B, H3 and H4. The octamer wraps approximately 146
	bp of DNA. HsAvD is expressed both maternally and zygotically and is found in embryos
	through to adults (female only). The human homologue, H2AX, is phosphorylated by ATM
	protein kinase when double strand DNA breaks occur. In mouse, H2AX "knock out" mice have
	an increased incidence of cancer.
Sterility:	Sterile filtered

Target Details

Target:	Histone H2A Variant (HIS2AV)
Alternative Name:	Histone H2AvD
Background:	Variant histones H2A are synthesized throughout the cell cycle and are very different from classical S-phase regulated H2A. H2AvD is vital for viability, but the exact function of variant histones H2A is not known. H2A is a core component of the nucleosome, an octamer containing two molecules each of H2A, H2B, H3 and H4. The octamer wraps approximately 146 bp of DNA. HsAvD is expressed both maternally and zygotically and is found in embryos through to adults (female only). The human homologue, H2AX, is phosphorylated by ATM protein kinase when double strand DNA breaks occur. In mouse, H2AX "knock out" mice have an increased incidence of cancer. Synonyms: H2AvD protein antibody
Gene ID:	43229, 17738227
UniProt:	P08985

Application Details

Application Notes:	Histone H2 AvD pS137 Antibody has been tested for use in ELISA and by western blot. Specific
	conditions for reactivity should be optimized by the end user. Expect a band approximately 14
	kDa in size corresponding to phosphorylated H2 vD protein by western blotting in the
	appropriate Drosophila tissue or cell lysate or extract. Less than 0.2 % reactivity is observed

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Application Details	
	against the non-phosphorylated form of the immunizing peptide. This antibody is phospho specific for pS137 of H2 vD protein.
Comment:	Gene Name: HIS2AV
Restrictions:	For Research Use only
Handling	
Format:	Liquid
Concentration:	1.0 mg/ml
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	Sodium azide
Precaution of Use:	WARNING: Reagents contain sodium azide. Sodium azide is very toxic if ingested or inhaled. Avoid contact with skin, eyes, or clothing. Wear eye or face protection when handling. If skin or eye contact occurs, wash with copious amounts of water. If ingested or inhaled, contact a physician immediately. Sodium azide yields toxic hydrazoic acid under acidic conditions. Dilute azide-containing compounds in running water before discarding to avoid accumulation of potentially explosive deposits in lead or copper plumbing.
Handling Advice:	Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature.
Storage:	4 °C/-20 °C
Storage Comment:	Store vial at 4 ° C prior to restoration. For extended storage aliquot contents and freeze at -24 ° C or below. This product is stable for several weeks at 4 ° C as an undiluted liquid. Dilute only prior to immediate use. Expiration date is one (1) year from date of opening.
Expiry Date:	12 months
Publications	
Product cited in:	Balmus, Karp, Ng, Jackson, Adams, McIntyre: "A high-throughput in vivo micronucleus assay for genome instability screening in mice." in: Nature protocols , Vol. 10, Issue 1, pp. 205-15, (2015) (PubMed). There are more publications referencing this product on: Product page



Immunofluorescence (Paraffin-embedded Sections)

Image 1. Systemic hormetic responses from musclespecific DNA damage.(A) Detection of DNA damage (double strand breaks) in dissected longitudinal thoracic muscle of young (10 d) Act88FG4>+(w1118) controls, and flies with DNA repair attenuation specifically in thoracic muscle (muspecific, Act88FG4>UAS-Mei-9RNAi or Act88FG4>UAS-Mei-9RNAi, UAS-ERCC1RNAi), assayed by phospho-H2aV immunostaining (red), counterstained with phalloidin (""Phall"", green, actin filaments) and DAPI (blue). Representative images shown. (B) Immunostaining to detect poly-ubiquitin protein (aggregates, ""Poly-Ub."") in dissected longitudinal thoracic muscle from young (10 d) and old (30 d) flies, genotypes described above, anti-polyubiquitin (green), counterstained with phalloidin (red, actin filaments). Representative images shown. (C-D). Survival curves (lifespan, female flies) associated with mu-specific inhibition of Mei-9 using (C) the Act88FGal4 driver (compared to Act88FG4>+[w1118] controls) or (D) a GeneSwitch inducible driver (Act88FGS, +RU486 compared with -RU486 [vehicle alone] sibling controls). (E) Quantification of mitoses per whole dissected midgut (assayed by anti-pH 3 immunostaining) at indicted ages, genotypes described above, bars represent mean \pm SE, n = 25-30. (F) Immunostaining of dissected intestines to assess epithelial integrity of posterior midguts at indicated ages, genotypes described above, pH 3 (green), armadillo (""Arm"", membrane, red), and DAPI (blue). Representative images shown. (G-H) Lineage tracing from ISCs using FRT recombination of a split alpha-tubulin-lacZ transgene (in Act88FG4>+[w1118, controls] or Act88FG4>UAS-Mei-9RNAi genetic background). (G) Changes in clone size (cell per clone form posterior midgut) at indicated ages, represented as box plot (median, red line), n = 25. (H) Representative images of lacZ clones from various genotypes at indicated

ages, immunostaining of dissected midguts (posterior), antilacZ (green), and DAPI (blue). (I) Venn diagrams showing overlap of up-regulated genes (from dissected midguts) between Act88FG4>UAS-Mei-9RNAi and controls Act88FGal4>+(w1118) during aging (transcriptomes at 30 d, compared to Act88FGal4>+[w1118] controls at day 10). The threshold for genes included in the analysis was (i) changes in RPKM values of at least 2-fold up-regulated in intestine compared to young controls and (ii) a minimum RPKM value of 2. (J) Fold change (in intestinal transcriptome RPKM values, Day 30 Act88FG4>UAS-Mei-9RNAi/ Day 10 Act88FGal4>+[w1118] control [black bars] or Day 30 control/Day 10 control [gray bars]) of selected innate immune genes. Underlying data can be found in S1 Data. See also S1 and S2 Figs and S1 and S2 Tables. FRT, flippase recombination target, ISC, intestinal stem cell, pH 3, phospho Histone H3, mu-specific, muscle-specific, RPKM, reads per kbp per million reads, RU486, mifepristone. figure provided by CiteAb. Source: PMID30036358

Western Blotting

Image 2. Western blot using affinity purified anti-histone H2AvD pS137 antibody shows detection of a band at ~15 kDa corresponding to phosphorylated H2AvD (lane 2 arrowhead). Lanes contain either mock-irradiated (lane 1) or 4000-RAD gamma irradiated (lane 2) Drosophila melanogaster (3rd instar) larvae brain WC lysate separated on by SDS-PAGE and transferred to nitrocellulose. After blocking the membrane was probed with the primary antibody diluted to 1:500. Washes and reaction with secondary antibody followed incubation. Use HRP conjugated Gt-a-Rabbit IgG [H&L] MX and ECL for detection. Personal Communication. Yikang Rong, NIH, CCR, Bethesda, MD.





Immunofluorescence (Cultured Cells)

Image 3. dH1 depletion induces DNA damage. a Immunostaining of dH1-depleted (siRNAdH1) and control undepleted cells (siRNAlacZ and untreated) with $\alpha dH1$ and αγH2Av antibodies (both in green). DNA was stained with DAPI (blue). Insets show enlarged images of representative individual cells. Scale bars are 20µm and 2µm in the Insets. On the right, the number of yH2Av foci per cell is presented (n>100 for each condition). Error bars are s.e.m. The p-value of siRNAdH1 respect to siRNAlacZ is indicated (***p<0.005, two-tailed Student's t-test). b WB analyses with adH1, ay H2Av and α H4 of increasing amounts of extracts (lanes 1-3) prepared from siRNAdH1, siRNAlacZ and untreated cells. The positions corresponding to molecular weight markers are indicated. On the right, quantitative analysis of the results (N=3). Error bars are s.e.m. The p-value of siRNAdH1 respect to siRNAlacZ is indicated (***<0.005, two-tailed Student's t-test). c Alkaline and neutral single-cell electrophoresis analyses of siRNAdH1, siRNAlacZ and untreated cells. Scale bar corresponds to 20µm. On the right, relative comet-tail moments are presented (n>100 for each condition). Error bars are s.e.m. The p-values of siRNAdH1 respect to siRNAlacZ are indicated (***<0.005, two-tailed Student's t-test). d On the top, WB analysis with ayH2Av and atubulin at different time points after X-ray irradiation (10Gy) of siRNAdH1 and untreated cells. The positions corresponding to molecular weight markers are indicated. On the bottom, quantitative analysis of the results (N=3) - figure provided by CiteAb. Source: PMID28819201

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