

Certificate of Analysis

Product: Affinity Purified Anti-Histone H2AvD pS137 (Rabbit)

Code: 600-401-914

Lot #: 16986

Size: 100 µg

Physical State: Liquid (sterile filtered)

Antibody Concentration: 1.09 mg/ml (by UV absorbance at 280 nm)

Buffer: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

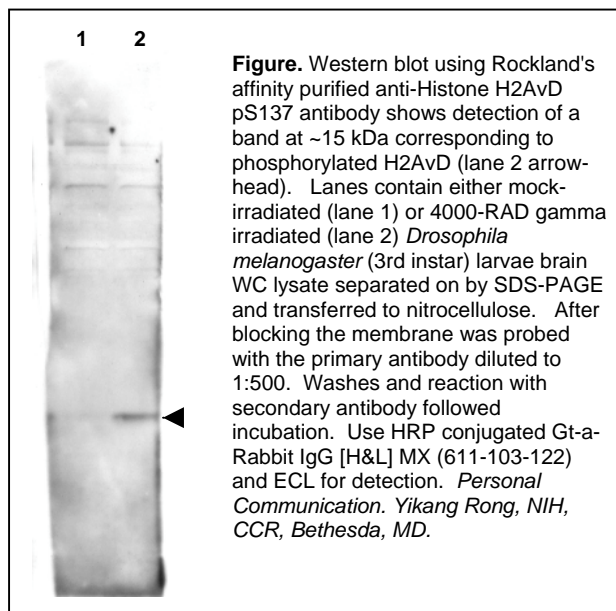
Stabilizer: None

Preservative: 0.01% (w/v) Sodium Azide

Storage Conditions: Store vial at -20° C prior to opening. Dilute only prior to immediate use. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Expiration date is one (1) year from date of opening.

Background Information: 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.

Application Note(s): This affinity purified 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 H2AvD protein by western blotting in the appropriate *Drosophila* tissue or cell lysate or extract. Less than 0.2% reactivity is observed against the non-phosphorylated form of the immunizing peptide. This antibody is phospho specific for pS137 of H2AvD protein.



Recommended Dilutions:

ELISA	1:4,000 - 1:16,000
WESTERN BLOT	1:400 - 1:1,600
IF MICROSCOPY	User Optimized
OTHER APPLICATIONS	User Optimized

Purity and Specificity: This affinity-purified 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 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.

Immunogen: This affinity purified 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.

Relevant Links: NCBI [S08118](#) (see also [NP_524519](#)) Swiss-Prot [P08985](#)

Protein Sequence: *Drosophila melanogaster* H2AvD pS137, 140 aa, predicted MW 14.8 kDa

1	maggkagkds	gkakakavsr	saraglqfpv	grihrhlksr	ttshgrvgat	aavysaile
61	yltaevlela	gnaskdlkvk	ritprhlqla	irgdeeldsl	ikatiagggv	ipihkslig
121	kkeetvqdpq	rkgnvilsqa	y			

General References:

Mason, J.M., Ransom, J. and Konev, A.Y. (2004) A deficiency screen for dominant suppressors of telomeric silencing in *Drosophila*. *Genetics* **168** (3), 1353-1370.

Kimura, K., Kodama, A., Hayasaka, Y. and Ohta, T. (2004) Activation of the cAMP/PKA signaling pathway is required for post-ecdysial cell death in wing epidermal cells of *Drosophila melanogaster*. *Development* **131** (7), 1597-1606.

Madigan, J.P., Chotkowski, H.L. and Glaser, R.L. (2002) DNA double strand breaks-induced phosphorylation of *Drosophila* histone variant H2Av helps prevent radiation-induced apoptosis. *Nucleic Acid Res.* **30**:17; 368-3705.

van Daal, A. and Elgin, S.C. (1992) A histone variant, H2AvD, is essential in *Drosophila melanogaster*. *Mol. Biol. Cell* **3** (6), 593-602.

van Daal, A., White, E.M., Elgin, S.C. and Gorovsky, M.A. (1990) Conservation of intron position indicates separation of major and variant H2As is an early event in the evolution of eukaryotes. *J. Mol. Evol.* **30** (5), 449-455.

van Daal, A., White, E.M., Gorovsky, M.A. and Elgin, S.C. (1988) *Drosophila* has a single copy of the gene encoding a highly conserved histone H2A variant of the H2A.F/Z type. *Nucleic Acids Res.* **16** (15), 7487-7497.

Related Products:

#611-703-127 Peroxidase Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (DONKEY) MX10

#611-132-122 IRDye800 Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (GOAT) MX10

#MB-070 Blocking Buffer for Fluorescent Western Blotting

#KIA-003 **MaxTag**TM Anti-RABBIT IgG Kit for Immunoblotting

#MB-070 Blocking Buffer for Fluorescent Western Blotting

USDA Certification: All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation.

Note: This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information.