Datasheet for ABIN129664

anti-HMGN1 antibody (pSer20, pSer24)

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Overview

Quantity:	100 µg
Target:	HMGN1
Binding Specificity:	AA 19-28, pSer20, pSer24
Reactivity:	Human
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This HMGN1 antibody is un-conjugated
Application:	Western Blotting (WB), Immunohistochemistry (IHC), ELISA

Product Details

Immunogen:	This affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to amino acids 19-28 of human HMGN protein (see below).
Isotype:	IgG
Characteristics:	Concentration Definition: by UV absorbance at 280 nm

Target Details

Target:	HMGN1
Abstract:	HMGN1 Products
Background:	This antibody is designed, produced, and is suitable for Cancer, Immunology and Nuclear Signaling research. HMGNs are proteins that bind chromatin effectively reducing the

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Target Details

compaction of the chromatin fiber and enhancing access to DNA regulatory sequences. Members of this family have a conserved chromatin binding domain which is phosphorylated during mitosis. The sequence immunized is conserved in several species. As such, this reagent is designed as a "universal" reagent for the detection of all phosphorylated HMGN proteins. The High Mobility Group (HMG) proteins were originally isolated from mammalian cells and were named according to their electrophoretic mobility in polyacrylamide gels. HMGs were arbitrarily classed as a specific type of nonhistone proteins based on the observation that they are ubiquitous to mammalian cells, that they share certain physical properties, and that they are associated with isolated chromatin. HMG proteins and are now subdivided into 3 families: the HMGB (formerly HMG-1/-2) family, the HMGN (formerly HMG-14/-17) family, and the HMGA (formerly HMG-I/Y/C) family. Each HMG family has a characteristic functional sequence motif. The functional motif of the HMGB family is called the "HMG-box;" that of the HMGN family, the "nucleosomal binding domain;" and that of the HMGA family, the "AT-hook." The functional motifs characteristic of these canonical HMGs are widespread among nuclear proteins in a variety of organisms. Proteins containing any of these functional motifs embedded in their sequence are known as "HMG motif proteins."
Synonyms: High mobility group nucleosome binding domain containing protein 1 antibody, High mobility group protein 14 antibody, High-mobility group nucleosome binding domain 1 antibody, HMG14 antibody

Gene ID:	3091, 386779
UniProt:	P05114
Pathways:	Chromatin Binding

Application Details

Application Notes:	This affinity purified antibody has been tested for use in ELISA, immunohistochemistry and by western blot. Specific conditions for reactivity should be optimized by the end user. Expect a band approximately 17 kDa in size corresponding to phosphorylated HMGN proteins by western blotting in the appropriate cell lysate or extract. Less than 0.5% reactivity is observed against the non-phosphorylated form of the immunizing peptide. This antibody is phospho specific for pS20 and pS24 of HMGN proteins. The antibody was tested against HMGN1 by performing a standard phopshorylation in vitro assay using HMGN1 as a substrate and Msk1 as a kinase (known as a main effector kinase of MAPK signaling pathway and in vivo kinase for HMGN1). This kinase specifically phosphorylates S20 and S24 of HMGN1 (S24 and S28 of HMGN2). The reaction was resolved on SDS PAGE and immunoblotted, using antibody diluted

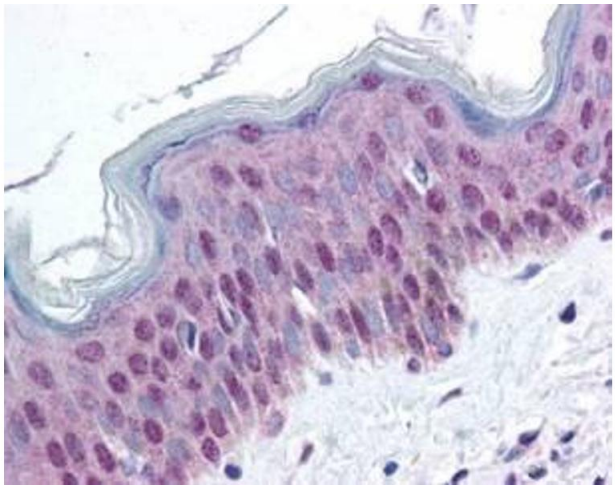
Application Details

	at 1:1,000 and 1:5,000. The antibody shows strong positive signal for Msk-phosphorylated HMGN1 and no cross-reaction for non-treated HMGN1.
Restrictions:	For Research Use only

Handling

Format:	Liquid
Concentration:	1.20 mg/mL
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	Sodium azide
Precaution of Use:	This product contains sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Storage:	-20 °C

Images



Immunohistochemistry

Image 1. affinity purified anti-HMGN pS20/pS24 antibody was used at 20 ug/ml to detect signal in a variety of tissues including multi-human, multi-brain and multi-cancer slides. This image shows moderate nuclear and faint cytoplasmic positive staining of epidermal keratinocytes at 40X. Tissue was formalin-fixed and paraffin embedded. The image shows localization of the antibody as the precipitated red signal, with a hematoxylin purple nuclear counterstain.

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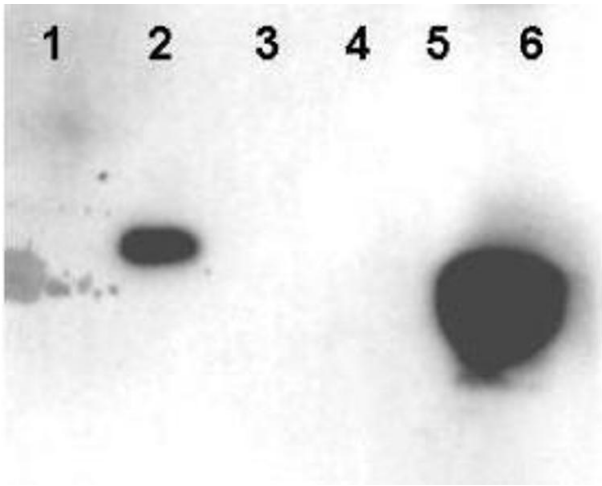

Western Blotting

Image 2. Western blot using Affinity Purified anti-HMNG antibody shows detection of phosphorylated HMGN1 and HMGN2. Recombinant native and mutant HMGN proteins were treated with kinase PKCalpha to specifically phosphorylate S20 and S24 residues. Lanes contain: 1 - HMGN1, non-phosphorylated, 2 - HMGN1, phosphorylated, 3 - HMGN1 delta20E, delta24E, non-phosphorylated, 4 - HMGN1, delta20E, delta24E, phosphorylated, 5 - HMGN2, non-phosphorylated, and 6 - HMGN2, phosphorylated. Molecular weight markers (not shown) confirm the size of each recombinant protein. The primary antibody was diluted 1:1,000 for this experiment. The blot was processed using a 5 sec exposure time. Personal Communication, Yuri Postnikov, NIH, CCR, Bethesda, MD.

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