abcam

ab240647

Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade

Recombinant | RabMAb

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR23430-12
Purification technique	Affinity purification Protein A
Concentration	0.546 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/2000
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Not recommended

Species	Mouse
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Species	Rat
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

IP

Tested

Species	Mouse
Dilution info	1/30
Notes	-

Species	Human
Dilution info	1/30
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

ChIP

Tested

Species	Mouse
Dilution info	5 µg for 25 µg chromatin
Notes	-

Species	Human
Dilution info	5 µg for 25 µg chromatin
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested

Species	Rat
Dilution info	1/1000
Notes	-

Species	Human
Dilution info	1/1000
Notes	-

Species	Mouse
Dilution info	1/1000
Notes	-

ICC/IF

Tested

Species	Mouse
Dilution info	1/50
Notes	-

Species	Human
Dilution info	1/50

Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/500
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information HEXIM1

Function Transcriptional regulator which functions as a general RNA polymerase II transcription inhibitor (PubMed:14580347, PubMed:15201869, PubMed:15713661). Core component of the 7SK RNP complex: in cooperation with 7SK snRNA sequesters P-TEFb in a large inactive 7SK snRNP complex preventing RNA polymerase II phosphorylation and subsequent transcriptional elongation (PubMed:12832472, PubMed:14580347, PubMed:15201869, PubMed:15713661). May also regulate NF-kappa-B, ESR1, NR3C1 and CIITA-dependent transcriptional activity (PubMed:15940264, PubMed:15941832, PubMed:17088550). Plays a role in the regulation of DNA virus-mediated innate immune response by assembling into the HDP-RNP complex, a complex that serves as a platform for IRF3 phosphorylation and subsequent innate immune response activation through the cGAS-STING pathway (PubMed:28712728).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	HEXIM1 also known as hexamethylene bis-acetamide inducible protein 1 is a protein with an approximate molecular mass of 49 kDa. It predominantly resides in the nucleus and is ubiquitously expressed across various tissues. This protein functions primarily in the regulation of transcription by modulating the activity of P-TEFb (Positive Transcription Elongation Factor b). HEXIM1 acts as an inhibitor by binding to the cyclin T1 and CDK9 components of P-TEFb preventing its role in promoting RNA polymerase II activity during transcription elongation.
Biological function summary	HEXIM1 serves a critical regulatory role within the transcription machinery in cells. It acts as an essential component of the HEXIM1/P-TEFb/7SK snRNP complex which helps maintain transcriptional homeostasis. This complex involves 7SK small nuclear RNA which makes the transcription process more efficient by sequestering P-TEFb in an inactive state. By doing this HEXIM1 controls the release and

	activation of P-TEFb ensuring accurate transcription regulation and preventing aberrant gene expression.
Pathways	HEXIM1 is an integral element in managing transcription elongation processes. It is known to be part of significant signaling pathways such as the P-TEFb dependent transcription pathway. HEXIM1 interacts with proteins like CDK9 and cyclin T1 that are fundamental to its regulatory function. By controlling these interactions HEXIM1 influences RNA polymerase II's transition into productive elongation mode essential for proper gene expression.
Associated diseases and disorders	Scientists have associated dysregulation of HEXIM1 activity with several conditions including cancer and cardiac hypertrophy. Aberrant HEXIM1 function or expression can lead to unchecked cellular proliferation in cancer due in part to misregulated P-TEFb activity. Moreover disruptions in this protein's regulatory capability have links to cardiac disorders where related proteins like CDK9 play roles in pathological cardiac growth. Therefore HEXIM1 serves as a potential target for therapeutic interventions concerning transcriptional dysregulation-related diseases.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

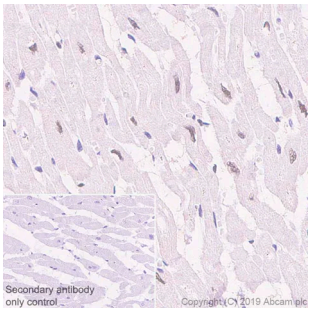
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

10 product images

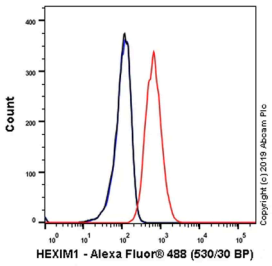


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

HEXIM1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-HEXIM1 antibody

Immunohistochemical analysis of paraffin-embedded Human cardiac muscle tissue labeling HEXIM1 with ab240647 at 1/2000 dilution dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Nuclear staining on human cardiac muscle (PMID: 23300697). The section was incubated with ab240647 for 10 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.
Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

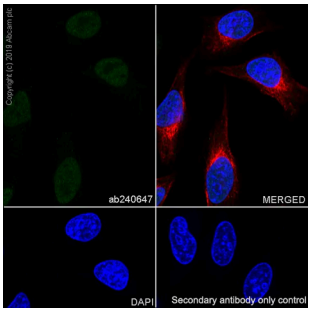
Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



Flow Cytometry (Intracellular) - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

HEXIM1 Flow Cytometry (Intracellular) staining using rabbit Anti-HEXIM1 antibody

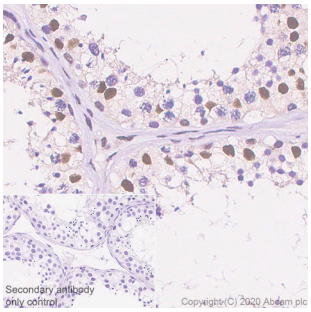
Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol permeabilized HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling HEXIM1 with ab240647 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).
A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

HEXIM1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-HEXIM1 antibody

Immunofluorescent analysis of 100% methanol-fixed HeLa cells labelling HEXIM1 with ab240647 at 1/50 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 (Green). Confocal image showing nuclear staining in HeLa cells. ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).
Secondary antibody only control: Secondary antibody is ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.

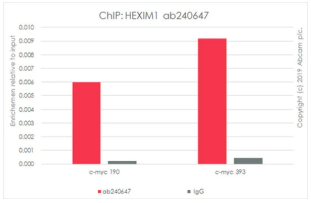


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

HEXIM1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-HEXIM1 antibody

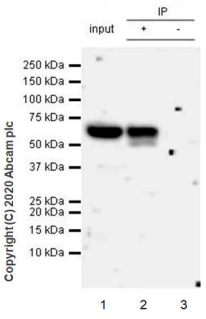
Immunohistochemical analysis of paraffin-embedded Human testis tissue labeling HEXIM1 with ab240647 at 1/2000 dilution dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on human testis (PMID: 23300697). The section was incubated with ab240647 for 10 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



ChIP - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

Chromatin was prepared from LNCaP cells according to the Abcam Dual-X-ChIP protocol. Cells were fixed with 1.5 mM EGS for 30mins and then formaldehyde for 10min.
The ChIP was performed with 25 µg of chromatin, 5 µg of ab240647 (red), or 5 µg of rabbit normal IgG ab172730 (gray) and 20 µl of Protein A/G sepharose beads. The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci).



Immunoprecipitation - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

HEXIM1 was immunoprecipitated from 0.35 mg HeLa (human cervix adenocarcinoma epithelial cell) (treated with 10 µM MG-132 for 24 hours), whole cell lysate with ab240647 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab240647 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP)(ab131366) was used at 1/5000 dilution.
Lane 1: HeLa (human cervix adenocarcinoma epithelial cell) treated with 10 µM MG-132 for 24 hours, whole cell lysate 10 ug

Lane 2: ab240647 IP in HeLa treated with 10 µM MG-132 for 24 hours whole cell lysate

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab240647 in HeLa treated with 10 µM MG-132 for 24 hours whole cell lysate

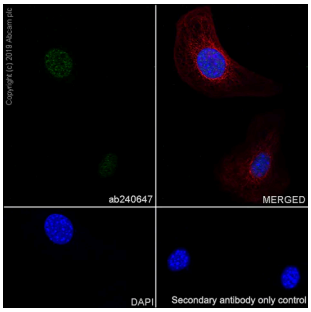
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 3 minutes.

All lanes:
Immunoprecipitation - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

Predicted band size: 41 kDa

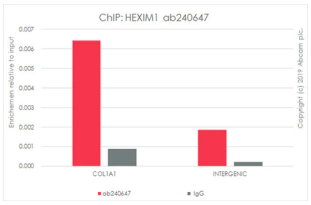
Observed band size: 60 kDa



Immunocytochemistry/ Immunofluorescence - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

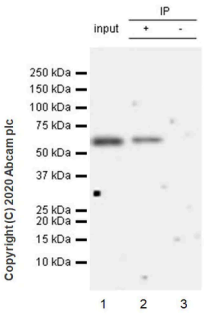
HEXIM1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-HEXIM1 antibody

Immunofluorescent analysis of 100% methanol-fixed MEF cells labelling HEXIM1 with ab240647 at 1/50 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 (Green). Confocal image showing nuclear staining in MEFs. ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 (Red). The Nuclear counterstain was DAPI (Blue).
Secondary antibody only control: Secondary antibody is ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



ChIP - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

Chromatin was prepared from MEF cells according to the Abcam Dual-X-ChIP protocol. Cells were fixed with 1.5 mM EGS for 30mins and then formaldehyde for 10min.
The ChIP was performed with 25 µg of chromatin, 5 µg of ab 240647 (red), or 5 µg of rabbit normal IgG ab172730 (gray) and 20 µl of Protein A/G sepharose beads. The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci).
Primers and probes are from paper PMCID: PMC4103662



Immunoprecipitation - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

HEXIM1 was immunoprecipitated from 0.35 mg MEF (mouse embryonic fibroblast (immortalized)) whole cell lysate with ab240647 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab240647 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP)(ab131366) was used at 1/5000 dilution.
Lane 1: MEF (mouse embryonic fibroblast (immortalized)) whole cell lysate 10 ug

Lane 2: ab240647 IP in MEF whole cell lysate

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab240647 in MEF whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

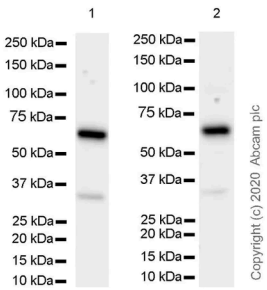
Exposure time: 3 minutes.

Lysate were made freshly and used in IP test immediately to minimize protein degradation. Incubation for immunoprecipitation was carried out overnight at 4°C.

All lanes:
Immunoprecipitation - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

Predicted band size: 41 kDa

Observed band size: 60 kDa



Western blot - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647)

HEXIM1 Western blot staining using rabbit Anti-HEXIM1 antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST.
The molecular weight observed is consistent with what has been described in the literature (PMID: 28254838)

Lysates were made freshly and used in WB test immediately to minimize protein degradation.

Exposure time: 26 seconds.

All lanes:
Western blot - Anti-HEXIM1 antibody [EPR23430-12] - ChIP Grade (ab240647) at 1/1000 dilution

Lane 1:
LNCaP (human prostate carcinoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
PC-12 (rat adrenal gland pheochromocytoma) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 41 kDa

Observed band size: 60 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.