

## Product datasheet

# Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade ab239364

Recombinant RabMAb

[2 References](#) [15 Images](#)

### Overview

<b>Product name</b>	Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade
<b>Description</b>	Rabbit monoclonal [EPR22956-37] to Cdk9 - ChIP Grade
<b>Host species</b>	Rabbit
<b>Tested applications</b>	<b>Suitable for:</b> Flow Cyt (Intra), IP, WB, IHC-P, ICC/IF, ChIP, ChIP-sequencing, ChIC/CUT&RUN-seq
<b>Species reactivity</b>	<b>Reacts with:</b> Mouse, Rat, Human
<b>Immunogen</b>	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
<b>Positive control</b>	WB: HEK-293T, HeLa, RAW 264.7, PC-12, NIH/3T3 and C6 whole cell lysate; Mouse brain and rat lung tissue lysate; IHC-P: Human pancreas, pancreatic cancer, mouse and rat liver tissue. ICC/IF: HeLa and NIH/3T3 cells. Flow: HeLa and NIH/3T3 cells. IP: NIH/3T3 cell lysate. ChIP: Chromatin from MEF and HeLa cells. ChIC/CUT&RUN-Seq: HeLa cells.
<b>General notes</b>	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"><li>- High batch-to-batch consistency and reproducibility</li><li>- Improved sensitivity and specificity</li><li>- Long-term security of supply</li><li>- Animal-free production</li></ul> <p>For more information <a href="#">see here</a>.</p> <p>Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <a href="#">RabMAb<sup>®</sup> patents</a>.</p>

### Properties

<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
<b>Storage buffer</b>	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
<b>Purity</b>	Protein A purified

<b>Clonality</b>	Monoclonal
<b>Clone number</b>	EPR22956-37
<b>Isotype</b>	IgG

## Applications

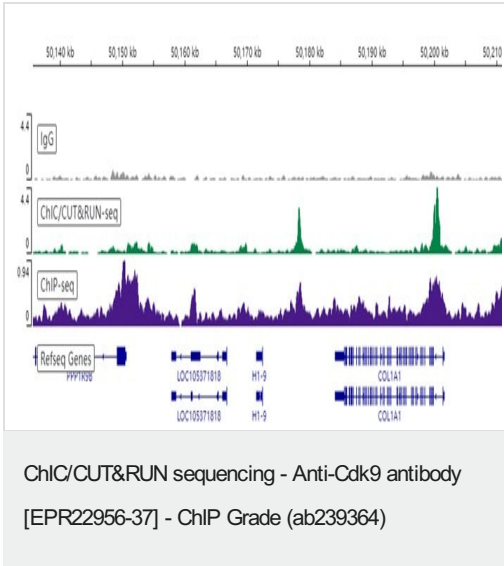
**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab239364 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
<b>Flow Cyt (Intra)</b>		1/600.
<b>IP</b>		1/30.
<b>WB</b>		1/1000. Detects a band of approximately 42, 55 kDa (predicted molecular weight: 43 kDa).
<b>IHC-P</b>		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.
<b>ICC/IF</b>		1/100.
<b>ChIP</b>		Use 5 µg for 25 µg of chromatin.
<b>ChIP-sequencing</b>		Use 8µg for 10 <sup>7</sup> cells.
<b>ChIC/CUT&amp;RUN-seq</b>		Use at an assay dependent concentration. 5 µg

## Target

<b>Function</b>	Member of the cyclin-dependent kinase pair (CDK9/cyclin-T) complex, also called positive transcription elongation factor b (P-TEFb), which facilitates the transition from abortive to production elongation by phosphorylating the CTD (C-terminal domain) of the large subunit of RNA polymerase II (RNAP II), SUPT5H and RDBP. The CDK9/cyclin-K complex has also a kinase activity toward CTD of RNAP II and can substitute for P-TEFb in vitro.
<b>Tissue specificity</b>	Ubiquitous.
<b>Sequence similarities</b>	Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. CDC2/CDKX subfamily. Contains 1 protein kinase domain.
<b>Cellular localization</b>	Nucleus.

## Images

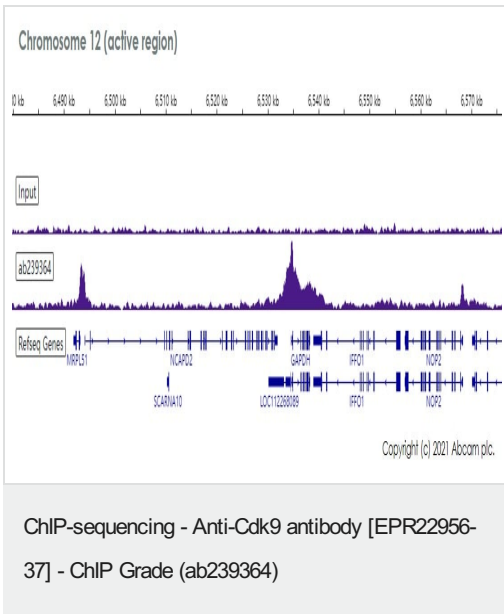


ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL,  $2 \times 10^5$  HeLa (Human cervix adenocarcinoma epithelial cell line) cells and 5  $\mu$ g of ab239364 [EPR22956-37]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control **ab172730** is also shown.

The ChIP data was conducted on chromatin prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with  $10^7$  HeLa cells and 8  $\mu$ g of ab239364. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

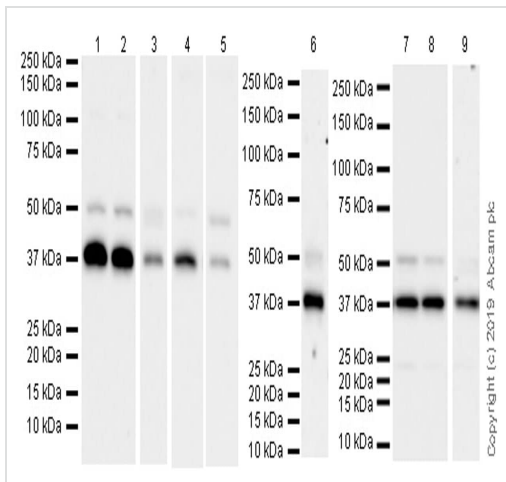
Additional screenshots of mapped reads can be downloaded [here](#).

The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with  $10^7$  HeLa cells and 8  $\mu$ g of ab239364 [EPR22956-37]. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

Additional screenshots of mapped reads can be downloaded [here](#).



Western blot - Anti-Cdk9 antibody [EPR22956-37] -  
ChIP Grade (ab239364)

**All lanes** : Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade  
(ab239364) at 1/1000 dilution

**Lane 1** : 293T (human embryonic kidney epithelial cell), whole cell  
lysate at 10 µg

**Lane 2** : HeLa (human cervix adenocarcinoma epithelial cell),  
whole cell lysate at 10 µg

**Lane 3** : Mouse lung tissue lysate at 20 µg

**Lane 4** : RAW264.7 (mouse Abelson murine leukemia virus-  
induced tumor macrophage), whole cell lysate at 10 µg

**Lane 5** : Mouse brain tissue lysate 20 µg

**Lane 6** : Rat lung tissue lysate at 20 µg

**Lane 7** : PC-12 (rat adrenal gland pheochromocytoma), whole cell  
lysate at 10 µg

**Lane 8** : NIH/3T3 (mouse embryonic fibroblast), whole cell lysate at  
10 µg

**Lane 9** : C6 (rat glial tumor glial cell) whole cell lysate at 10 µg

#### Secondary

**Lanes 1-5 & 7-9** : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at  
1/100000 dilution

**Lane 6** : VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) at  
1/1000 dilution

**Predicted band size:** 43 kDa

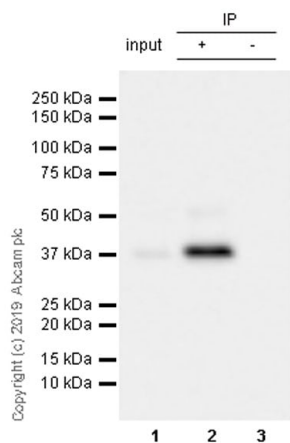
**Observed band size:** 42,55 kDa

Blocking and Dilution Buffer and concentration: 5% NFDM/TBST

Exposure times.

Lanes 1-3: 36 seconds Lanes 4-5: 5.5 seconds Lane 6: 3 minutes

Lanes 7-8: 15 seconds Lane 9: 48 seconds



Immunoprecipitation - Anti-Cdk9 antibody  
[EPR22956-37] - ChIP Grade (ab239364)

CDK9 was immunoprecipitated from 0.35 mg NIH/3T3 (mouse embryonic fibroblast), whole cell lysate 10 ug with ab239364 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab239364 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

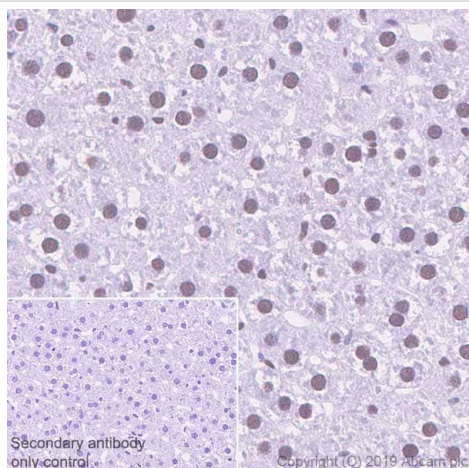
Lane 1: NIH/3T3 (mouse embryonic fibroblast), whole cell lysate 10 ug

Lane 2: ab239364 IP in NIH/3T3 whole cell lysate

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab239364 in NIH/3T3 whole cell lysate

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

Exposure time: 6 seconds

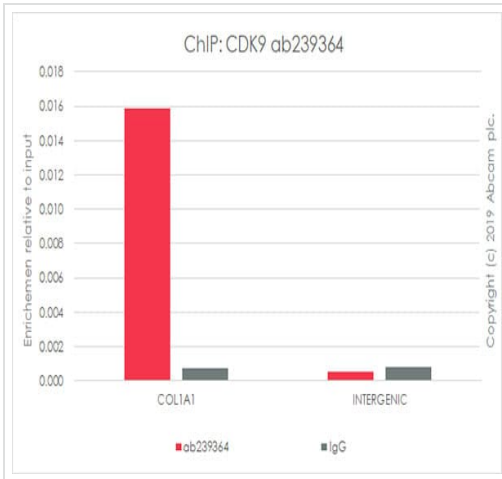


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdk9 antibody  
[EPR22956-37] - ChIP Grade (ab239364)

Immunohistochemical analysis of paraffin-embedded Rat liver tissue labeling CDK9 with ab239364 at 1/2000 dilution (0.30 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Nuclear staining on rat liver (PMID: 9766517, 11282025). The section was incubated with ab239364 for 10 mins at RT. The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> 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 Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

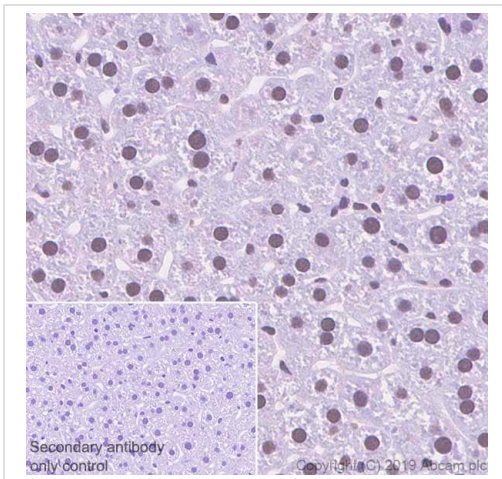


ChIP - Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade (ab239364)

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 ab239364 (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 PMC4103662 (PMID: 23663783)

\*<http://www.abcam.com/resources?keywords=X%20ChIP%20protocol>



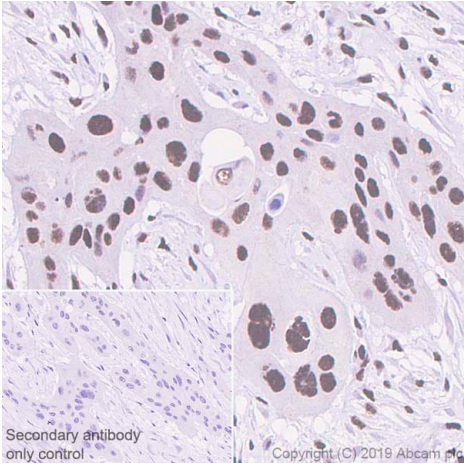
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade (ab239364)

Immunohistochemical analysis of paraffin-embedded Mouse liver tissue labeling CDK9 with ab239364 at 1/2000 dilution (0.30 µg/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Nuclear staining on mouse liver (PMID: 9766517, 11282025) The section was incubated with ab239364 for 10 mins at RT. 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 Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.



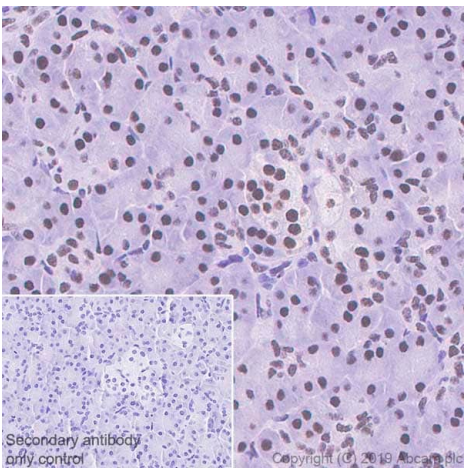


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade (ab239364)

Immunohistochemical analysis of paraffin-embedded Human pancreatic cancer tissue labeling CDK9 with ab239364 at 1/2000 dilution (0.30 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)). Nuclear staining on human pancreatic cancer (PMID: 28231737) The section was incubated with ab239364 for 10 mins at RT. 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 Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

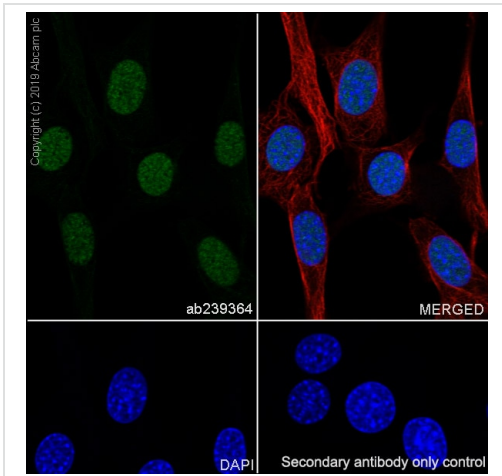


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade (ab239364)

Immunohistochemical analysis of paraffin-embedded Human pancreas tissue labeling CDK9 with ab239364 at 1/2000 dilution (0.30 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)). Nuclear staining on human pancreas (PMID: 28231737) The section was incubated with ab239364 for 10 mins at RT. 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](#)).

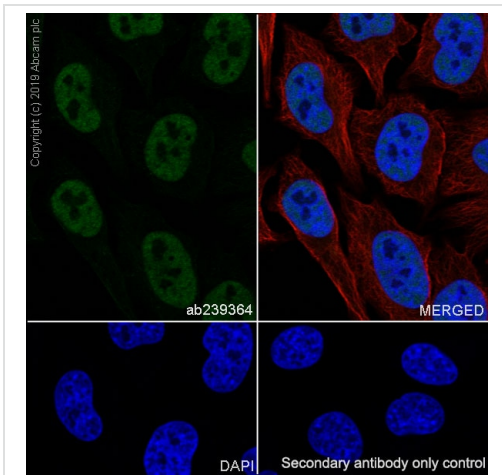
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.



Immunocytochemistry/ Immunofluorescence - Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade (ab239364)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized NIH/3T3 (mouse embryonic fibroblast) cells labelling CDK9 with ab239364 at 1/100 (6 ug/ml) dilution, followed by [ab150077](#) AlexaFluor<sup>®</sup>488 Goat anti-Rabbit secondary antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing nuclear staining in NIH/3T3 cell line is observed. [ab195889](#) Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab239364 anti-CDK9 [ab150077](#) AlexaFluor<sup>®</sup>488 Goat anti-Rabbit secondary at 1/1000 (2 ug/ml) dilution.

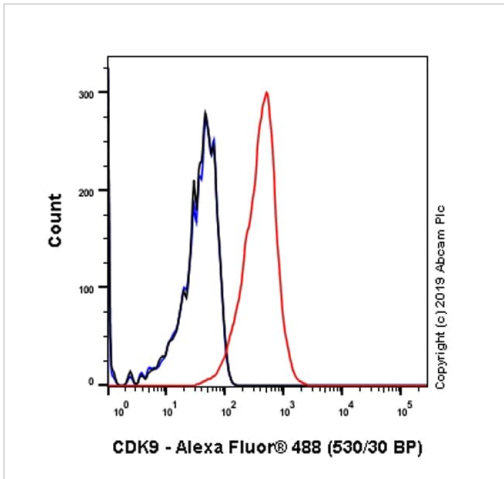


Immunocytochemistry/ Immunofluorescence - Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade (ab239364)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized HeLa (human cervix adenocarcinoma epithelial cell) cells labelling CDK9 with ab239364 at 1/100 (6 ug/ml) dilution, followed by [ab150077](#) AlexaFluor<sup>®</sup>488 Goat anti-Rabbit secondary antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing nuclear staining in HeLa cell line is observed. [ab195889](#) Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

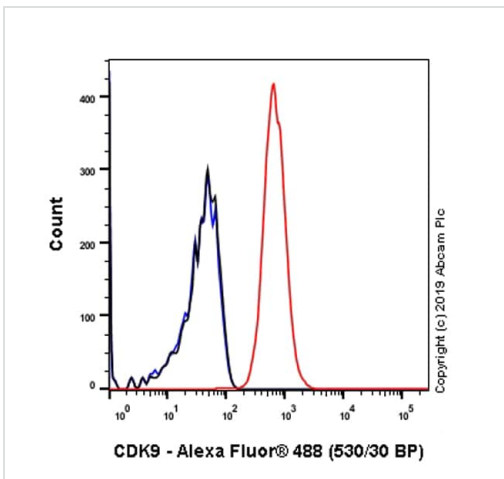
Secondary antibody only control: Secondary antibody is ab239364 anti-CDK9 [ab150077](#) AlexaFluor<sup>®</sup>488 Goat anti-Rabbit secondary at 1/1000 (2 ug/ml) dilution.





Flow Cytometry (Intracellular) - Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade (ab239364)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized NIH/3T3 (Mouse embryo) cells labelling CDK9 with ab239364 at 1/600 (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.



Flow Cytometry (Intracellular) - Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade (ab239364)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized HeLa (Human cervix adenocarcinoma) cells labelling CDK9 with ab239364 at 1/600 (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.



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Chromatin was prepared from HeLa 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 ab239364 (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 PMID: PMC2756882.

\*[https://www.abcam.com/resources?](https://www.abcam.com/resources?keywords=X%20ChIP%20protocol)

**keywords=X%20ChIP%20protocol**

### Why choose a recombinant antibody?



**Research with confidence**  
Consistent and reproducible results



**Long-term and scalable supply**  
Recombinant technology



**Success from the first experiment**  
Confirmed specificity



**Ethical standards compliant**  
Animal-free production

Anti-Cdk9 antibody [EPR22956-37] - ChIP Grade  
(ab239364)

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