abcam

Product datasheet

Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade ab150402

Recombinant RabMAb

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Overview

Product name	Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade
Description	Rabbit monoclonal [EPR6171(2)(B)] to Histone H2A.Z - ChIP Grade
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), WB, IHC-P, ICC/IF, ChIP
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Human Histone H2A.Z aa 1-100. The exact sequence is proprietary.
Positive control	WB: Neuro-2a, HeLa, HepG2, RAW 264.7, C6 and PC-12 cell lysates. IHC-P: Human colon and lung carcinoma tissues, human breast tissue, mouse and rat cerebrum tissue. ICC/IF: HepG2 and HeLa cells. Flow Cyt (intra): HeLa cells.
General notes	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 production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	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.
Storage buffer	Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR6171(2)(B)

Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab150402 in the following tested applications.

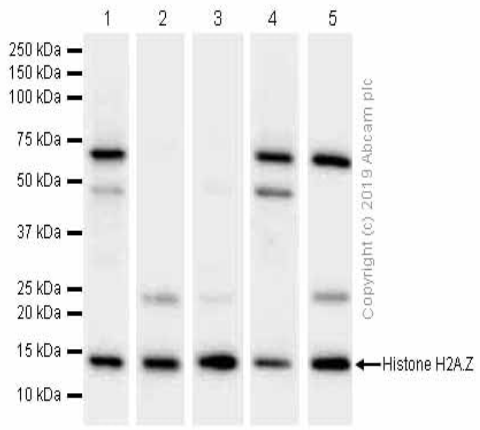
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/2500.
WB	★★★★★ **(5)**	1/1000 - 1/10000. Predicted molecular weight: 13 kDa.
IHC-P		1/2000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**. **For unpurified format use at 1/250 - 1/500 dilution.**
ICC/IF		1/100 - 1/250.
ChIP		Use 2 µg for 25 µg of chromatin.

Target

Function	Variant histone H2A which replaces conventional H2A in a subset of nucleosomes. Nucleosomes wrap and compact DNA into chromatin, limiting DNA accessibility to the cellular machineries which require DNA as a template. Histones thereby play a central role in transcription regulation, DNA repair, DNA replication and chromosomal stability. DNA accessibility is regulated via a complex set of post-translational modifications of histones, also called histone code, and nucleosome remodeling. May be involved in the formation of constitutive heterochromatin. May be required for chromosome segregation during cell division.
Sequence similarities	Belongs to the histone H2A family.
Post-translational modifications	Monoubiquitination of Lys-122 gives a specific tag for epigenetic transcriptional repression. Acetylated on Lys-5, Lys-8 and Lys-12 during interphase. Acetylation disappears at mitosis. Monomethylated on Lys-5 and Lys-8 by SETD6. SETD6 predominantly methylates Lys-8, lys-5 being a possible secondary site. Not phosphorylated.
Cellular localization	Nucleus. Chromosome.

Images



Western blot - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

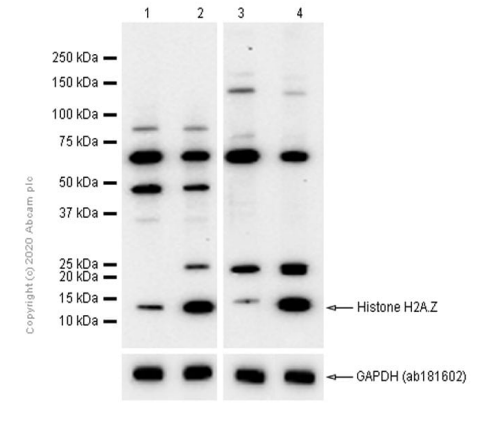
All lanes : Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402) at 1/5000 dilution (Purified)

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates
Lane 2 : PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates
Lane 3 : Neuro-2a (Mouse neuroblastoma neuroblast) whole cell lysates
Lane 4 : RAW 264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysates
Lane 5 : C6 (Rat glial tumor glial cell) whole cell lysates

Lysates/proteins at 15 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 13 kDa
Observed band size: 13 kDa



Western blot - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

All lanes : Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402) at 1/1000 dilution

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates prepared in RIPA lysis method
Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates prepared in 1% SDS Hot lysis method
Lane 3 : PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates prepared in RIPA lysis method
Lane 4 : PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates prepared in 1% SDS Hot lysis method

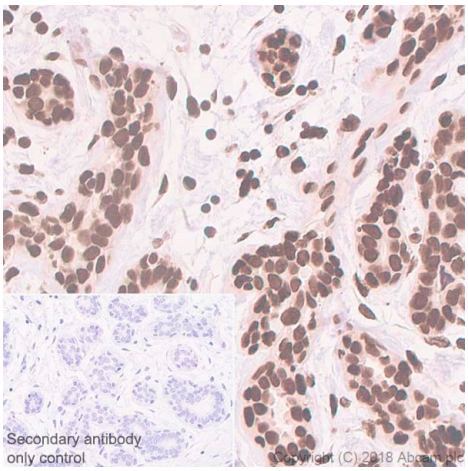
Lysates/proteins at 15 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000

dilution

Predicted band size: 13 kDa
Observed band size: 13 kDa

Blocking buffer: 5% NFDM/TBST.

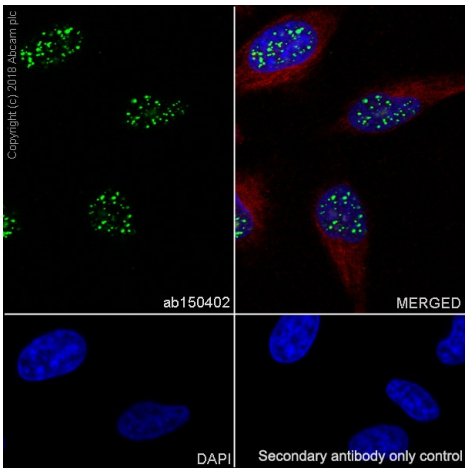


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast tissue sections labeling Histone H2A.Z with purified ab150402 at 1/2000 dilution (1.09 µg/ml). Heat mediated antigen retrieval was performed using heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

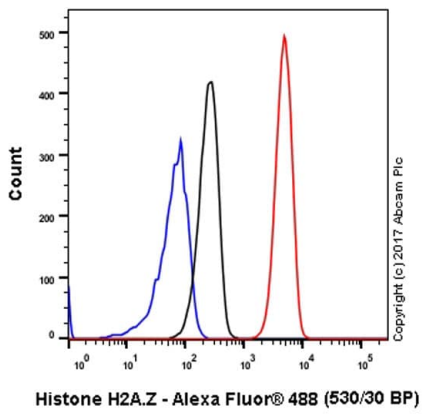
ChIP - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Chromatin was prepared from HeLa cells according to the Abcam X-ChIP protocol*. Cells were fixed with formaldehyde for 10 minutes.
The ChIP was performed with 25 µg of chromatin, 2 µg of ab150402 (red), and 20 µl of Protein A/G sepharose beads. 2 µg of rabbit normal IgG was added to the beads control (gray). 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 located in the first kb of the transcribed region.
*http://www.abcam.com/resources?keywords=X%20ChIP%20protocol



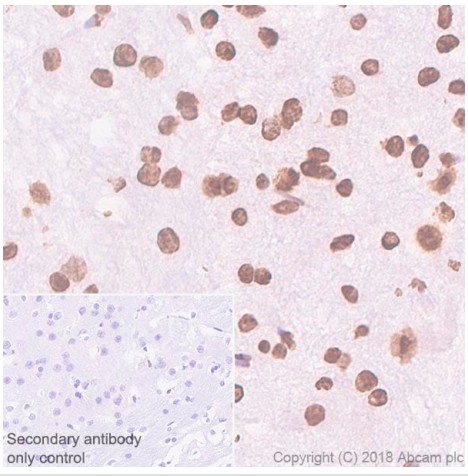
Immunocytochemistry/ Immunofluorescence - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Histone H2A.Z with purified ab150402 at 1/200 dilution (10 µg/ml). Cells were fixed in 100% Methanol. Cells were counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



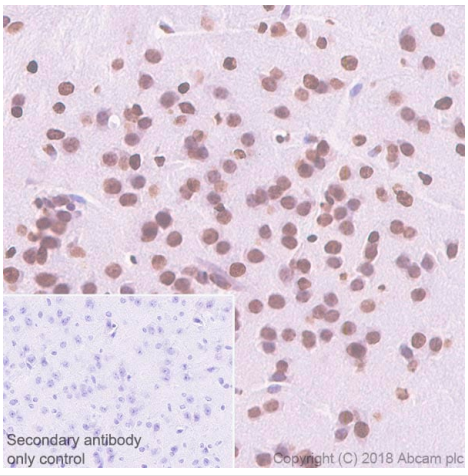
Flow Cytometry (Intracellular) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling Histone H2A.Z (red) with purified ab150402 at a 1/2500 dilution. Cells were fixed with 80% methanol and permeabilized with 0.1% Tween-20. A goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) was used as the secondary antibody at a 1/2000 dilution. Black - Rabbit monoclonal IgG (**ab172730**). Blue (unlabeled control) - Cells without incubation with the primary and secondary antibodies.



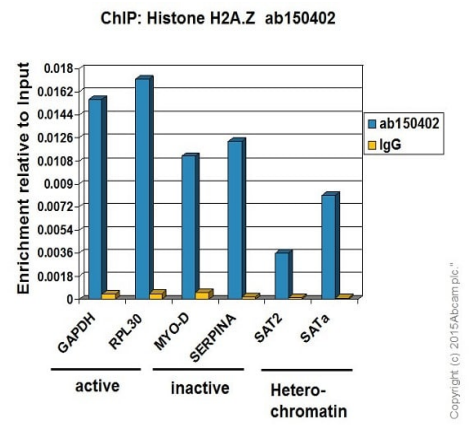
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat cerebrum tissue sections labeling Histone H2A.Z with purified ab150402 at 1:2000 dilution (1.09 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



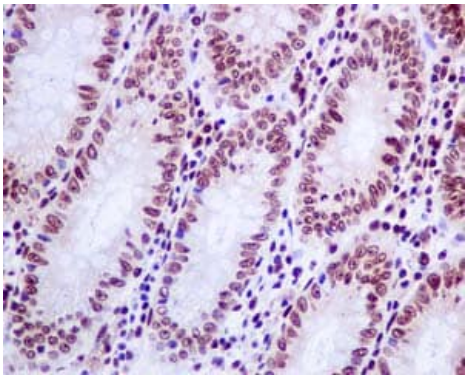
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse cerebrum tissue sections labeling Histone H2A.Z with purified ab150402 at 1/2000 dilution (1.09 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



ChIP - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

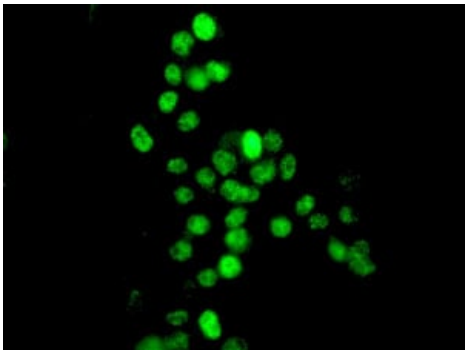
Chromatin was prepared from HeLa (Human epithelial cell line from cervix adenocarcinoma) cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 2µg of ab150402 (unpurified) (blue), and 20µl of Anti rabbit IgG sepharose beads. 2µg of rabbit normal IgG was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach). Primers and probes are located in the first kb of the transcribed region.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

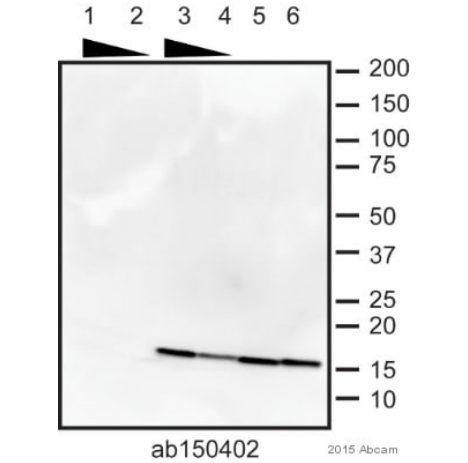
Immunohistochemical analysis of paraffin-embedded human colon tissue labeling Histone H2A.Z with ab150402 (unpurified) at 1/250 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunofluorescent analysis of HepG2 (Human liver hepatocellular carcinoma cell line) cells labeling Histone H2A.Z with ab150402 (unpurified) at 1/100 dilution.



Western blot - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

This image is courtesy of an Abreview submitted by Ragnhild Eskeland

All lanes : Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402) at 1/1000 dilution (unpurified)

Lane 1 : Recombinant Human octamers containing H2A at 1 µg
Lane 2 : Recombinant Human octamers containing H2A at 0.5 µg
Lane 3 : Native recombinant octamers K562 cells at 3 µg
Lane 4 : Native recombinant octamers K562 cells at 1.5 µg
Lane 5 : Recombinant Human octamers containing H2A.Z.2.1 at 0.5 µg
Lane 6 : Recombinant Human octamers containing H2A.Z.1 at 0.5 µg

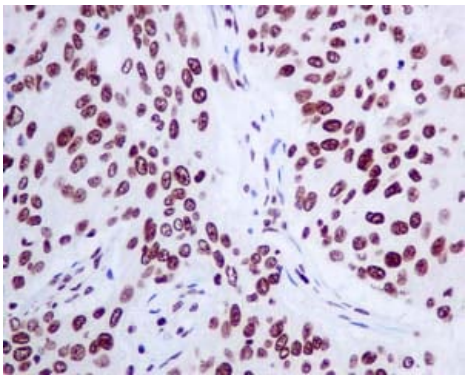
Secondary
All lanes : HRP-conjugated donkey anti-rabbit IgG polyclonal at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 13 kDa
Observed band size: 15 kDa

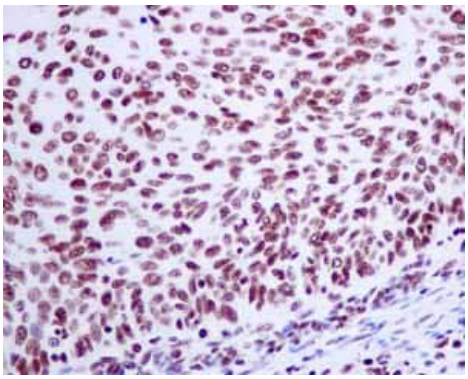
Exposure time: 5 minutes



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunohistochemical analysis of paraffin-embedded human lung carcinoma tissue labeling Histone H2A.Z with ab150402 (unpurified) at 1/250 dilution.

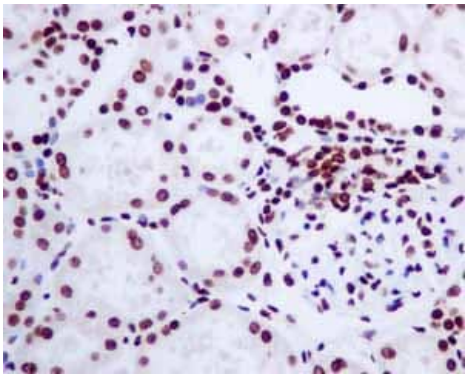
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunohistochemical analysis of paraffin embedded human cervical carcinoma tissue using ab150402 (unpurified) showing positive staining.

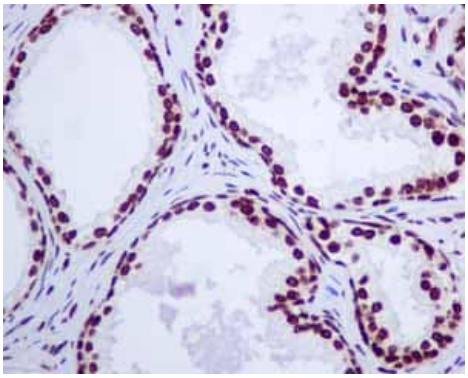
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunohistochemical analysis of paraffin embedded normal human kidney tissue using ab150402 (unpurified) showing positive staining.

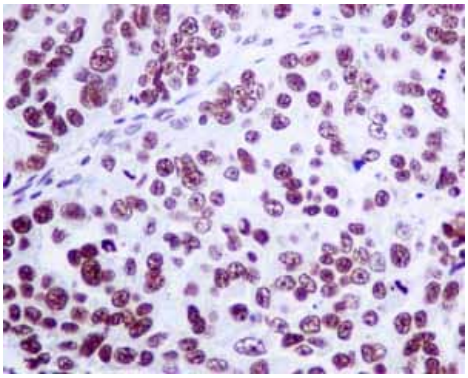
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunohistochemical analysis of paraffin embedded human prostate hyperplasia tissue using ab150402 showing positive staining.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

Immunohistochemical analysis of paraffin embedded human ovarian carcinoma tissue using ab150402 (unpurified) showing positive staining.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Anti-Histone H2A.Z antibody [EPR6171(2)(B)] - ChIP Grade (ab150402)

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