

Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free ab251504

KO VALIDATED Recombinant RabMAb

13 Images

Overview

Product name	Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free
Description	Rabbit monoclonal [EPR19244] to Galectin 3 - BSA and Azide free
Host species	Rabbit
Tested applications	Suitable for: Flow Cyt (Intra), WB, IP, IHC-P, ICC/IF
Species reactivity	Reacts with: Human
Immunogen	Recombinant full length protein. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa, SW480, MCF7, A431 and Wild-type A549 whole cell lysates; human heart, kidney and stomach lysates. IHC-P: Human liver, stomach, diffuse large B cell lymphoma and colon cancer tissues. ICC/IF: HeLa and A431 cells. Flow Cyt (intra): HeLa cells. IP: HeLa whole cell lysate.
General notes	<p>ab251504 is the carrier-free version of ab209344.</p> <p>Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.</p> <p>This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.</p> <p>Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.</p> <p>This product is compatible with the Maxpar[®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.</p> <p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none">- High batch-to-batch consistency and reproducibility- Improved sensitivity and specificity- Long-term security of supply- Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit</p>

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. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR19244
Isotype	IgG

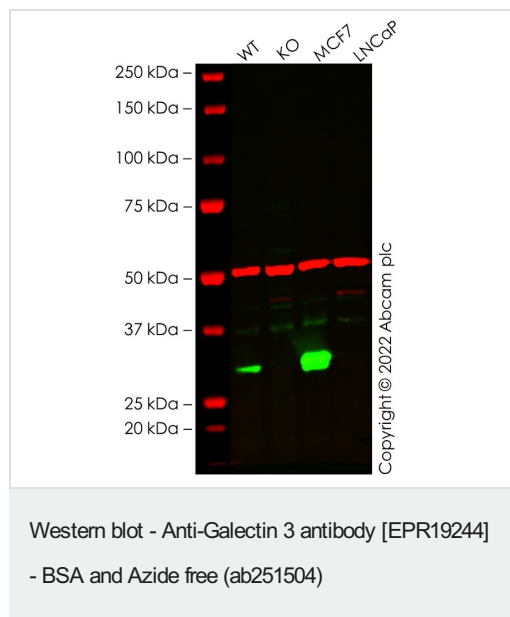
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab251504 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)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 26 kDa (predicted molecular weight: 26 kDa).
IP		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.

Target

Function	Galactose-specific lectin which binds IgE. May mediate with the alpha-3, beta-1 integrin the stimulation by CSPG4 of endothelial cells migration. Together with DMBT1, required for terminal differentiation of columnar epithelial cells during early embryogenesis.
Tissue specificity	A major expression is found in the colonic epithelium. It is also abundant in the activated macrophages.
Sequence similarities	Contains 1 galectin domain.
Cellular localization	Nucleus. Cytoplasmic in adenomas and carcinomas. May be secreted by a non-classical secretory pathway and associate with the cell surface.



All lanes : Anti-Galectin 3 antibody [EPR19244] ([ab209344](#)) at 1/1000 dilution

Lane 1 : Wild-type A549 cell lysate

Lane 2 : LGALS3 knockout A549 cell lysate

Lane 3 : MCF7 cell lysate

Lane 4 : LNCaP cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

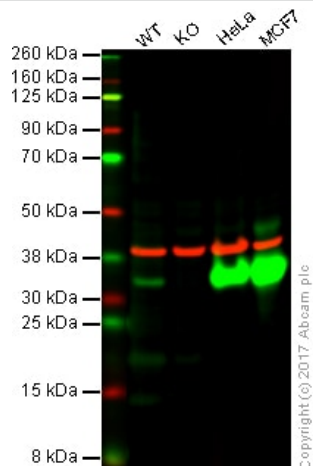
Performed under reducing conditions.

Predicted band size: 26 kDa

Observed band size: 30 kDa

False colour image of Western blot: Anti-Galectin 3 antibody [EPR19244] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] ([ab7291](#)) loading control staining at 1/20000 dilution, shown in red. In Western blot, [ab209344](#) was shown to bind specifically to Galectin 3. A band was observed at 30 kDa in wild-type A549 cell lysates with no signal observed at this size in LGALS3 knockout cell line. To generate this image, wild-type and LGALS3 knockout A549 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.

This data was developed using [ab209344](#), the same antibody clone in a different buffer formulation.



Western blot - Anti-Galectin 3 antibody [EPR19244]
- BSA and Azide free (ab251504)

This data was developed using [ab209344](#), the same antibody clone in a different buffer formulation.

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)

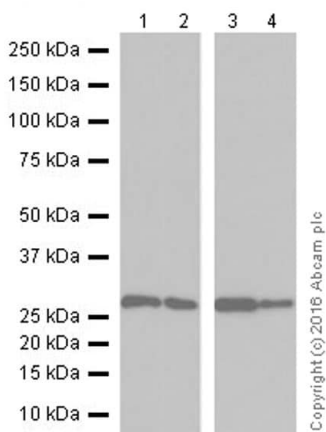
Lane 2: alectin 3 (KO) knockout HAP1 whole cell lysate (20 µg)

Lane 3: HeLa whole cell lysate (20 µg)

Lane 4: MCF7 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - [ab209344](#) observed at 35 kDa. Red - loading control, [ab8245](#), observed at 37 kDa.

[ab209344](#) was shown to specifically react with Galectin 3 (KO) in wild-type HAP1 cells. No band was observed when LGALS3 (KO) knockout samples were examined. Wild-type and Galectin 3 (KO) knockout samples were subjected to SDS-PAGE. [ab209344](#) and [ab8245](#) (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10,000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed [ab216773](#) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed [ab216776](#) secondary antibodies at 1/20,000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Galectin 3 antibody [EPR19244]
- BSA and Azide free (ab251504)

Lanes 1-2 : Anti-Galectin 3 antibody [EPR19244] ([ab209344](#)) at 1/10000 dilution

Lanes 3-4 : Anti-Galectin 3 antibody [EPR19244] ([ab209344](#)) at 1/1000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 2 : SW480 (Human colorectal adenocarcinoma cell line) whole cell lysate

Lane 3 : MCF7 (Human breast adenocarcinoma cell line) whole cell lysate

Lane 4 : A431 (Human epidermoid carcinoma cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

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

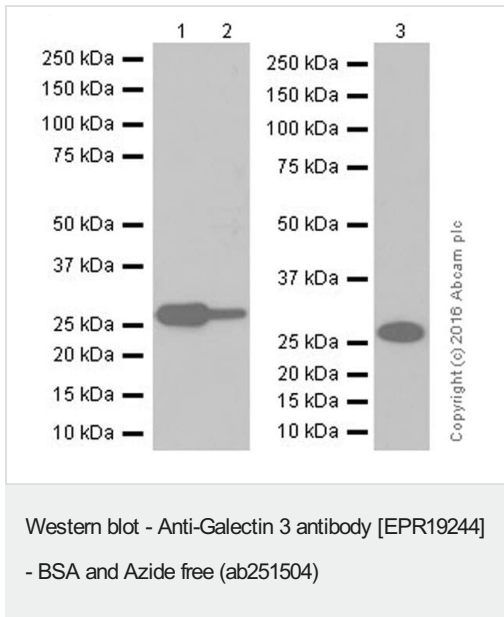
Predicted band size: 26 kDa

Observed band size: 26 kDa

This data was developed using [ab209344](#), the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure times: Lanes 1-2: 30 seconds; Lanes 3-4: 8 seconds.



All lanes : Anti-Galectin 3 antibody [EPR19244] ([ab209344](#)) at 1/1000 dilution

Lane 1 : Human heart tissue

Lane 2 : Human kidney tissue

Lane 3 : Human stomach tissue

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

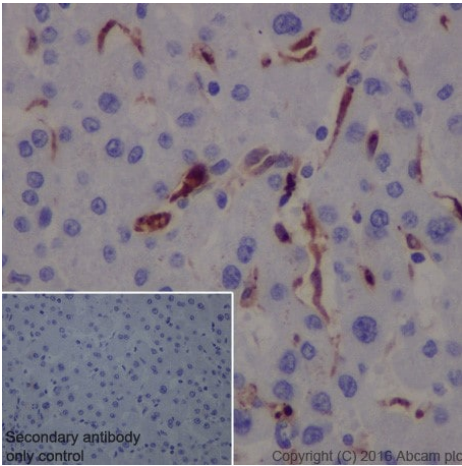
Predicted band size: 26 kDa

Observed band size: 26 kDa

This data was developed using [ab209344](#), the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.

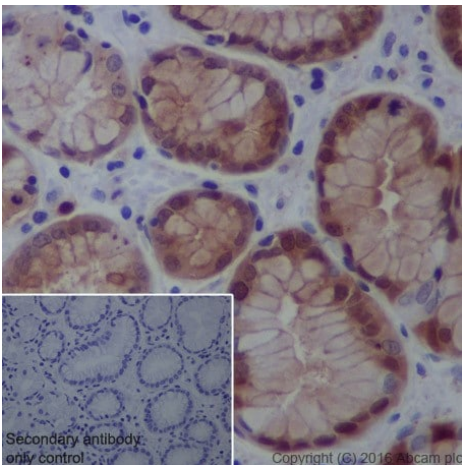
Exposure times: Lanes 1-2: 15 seconds; Lane 3: 30 seconds.



This data was developed using [ab209344](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded human liver tissue labeling Galectin 3 with [ab209344](#) at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Positive staining on Kupffer cells in the liver is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

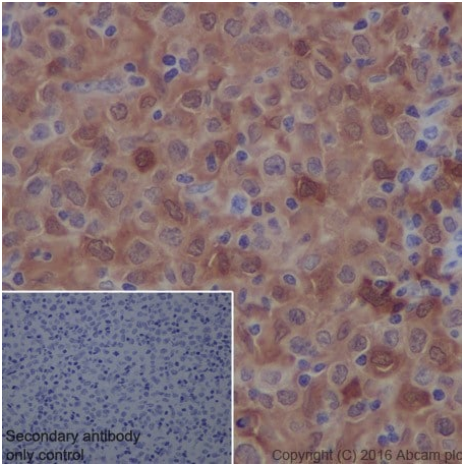
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)



This data was developed using [ab209344](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded human stomach tissue labeling Galectin 3 with [ab209344](#) at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Cytoplasmic and nuclear staining on human stomach is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

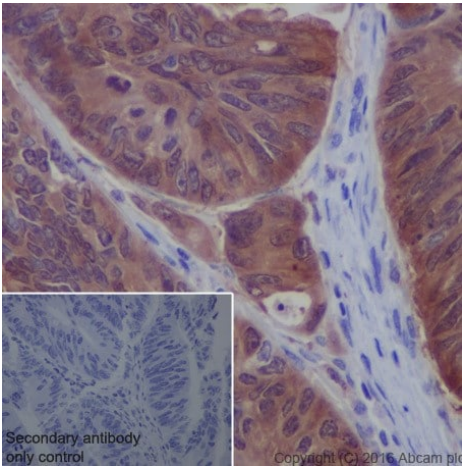
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)

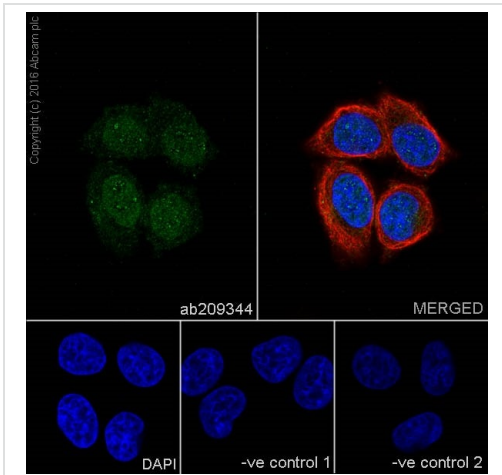
This data was developed using [ab209344](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded human diffuse large B cell lymphoma tissue labeling Galectin 3 with [ab209344](#) at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Cytoplasmic and weak nuclear staining on human diffuse large B cell lymphoma is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)

This data was developed using [ab209344](#), the same antibody clone in a different buffer formulation. Immunohistochemical analysis of paraffin-embedded human colon cancer tissue labeling Galectin 3 with [ab209344](#) at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Cytoplasmic and weak nuclear staining on human colon cancer is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

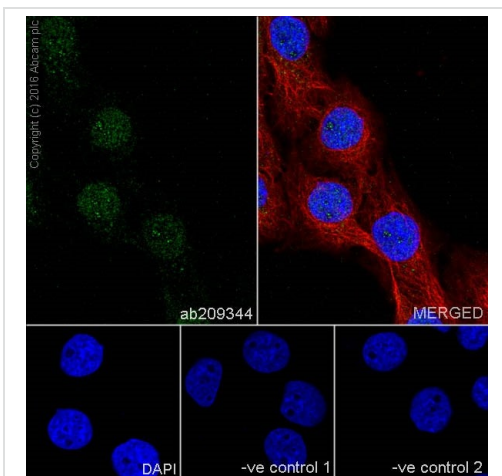


Immunocytochemistry/ Immunofluorescence - Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)

This data was developed using **ab209344**, the same antibody clone in a different buffer formulation. Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling Galectin 3 with **ab209344** at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear and weak cytoplasmic staining on HeLa cell line. The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution and Goat Anti-Mouse IgG H&L (Alexa Fluor[®] 594) (**ab150120**) secondary antibody at 1/1000 dilution (red). The negative controls are as follows:

-ve control 1: **ab209344** at 1/500 dilution followed by Goat Anti-Mouse IgG H&L (Alexa Fluor[®] 594) (**ab150120**) secondary antibody at 1/1000 dilution.

-ve control 2: Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (**ab150077**) secondary antibody at 1/1000 dilution.



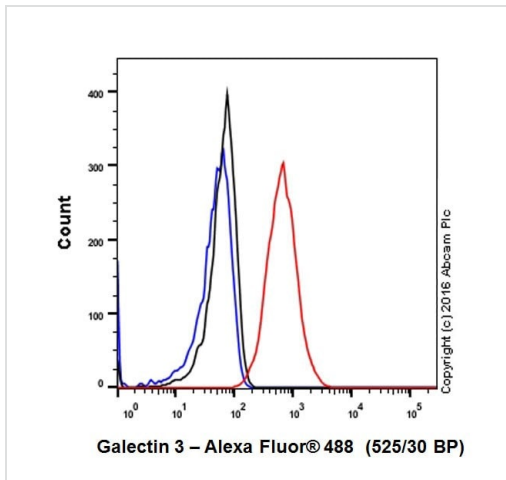
Immunocytochemistry/ Immunofluorescence - Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)

This data was developed using **ab209344**, the same antibody clone in a different buffer formulation. Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized A431 (Human epidermoid carcinoma cell line) cells labeling Galectin 3 with **ab209344** at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear and weakly cytoplasmic staining on A431 cell line. The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution and Goat Anti-Mouse IgG H&L (Alexa Fluor[®] 594) (**ab150120**) secondary antibody at 1/1000 dilution (red). The negative controls are as follows:

-ve control 1: **ab209344** at 1/500 dilution followed by **ab150120** Goat Anti-Mouse IgG H&L (Alexa Fluor[®] 594) (**ab150120**)

secondary antibody at 1/1000 dilution.

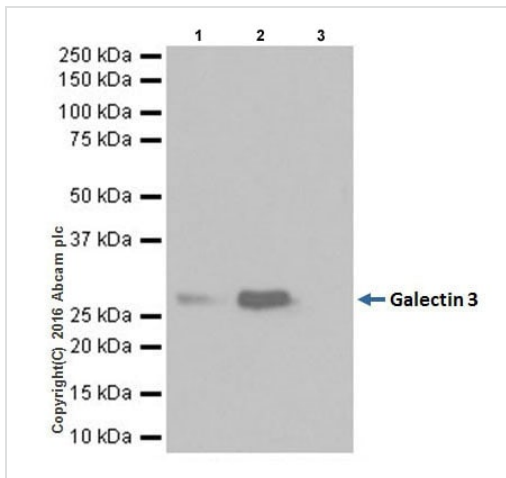
-ve control 2: Anti-alpha Tubulin mouse MAAb (**ab7291**) at 1/1000 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution.



Flow Cytometry (Intracellular) - Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)

This data was developed using **ab209344**, the same antibody clone in a different buffer formulation.





Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling Galectin 3 with **ab209344** at 1/50 dilution (red) compared with a rabbit monoclonal IgG isotype control (**ab172730**; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat Anti-Rabbit IgG (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)

This data was developed using **ab209344**, the same antibody clone in a different buffer formulation. Galectin 3 was immunoprecipitated from 0.35 mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate with **ab209344** at 1/30 dilution. Western blot was performed from the immunoprecipitate using **ab209344** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution. Lane 1: HeLa whole cell lysate 10µg (Input). Lane 2: **ab209344** IP in HeLa whole cell lysate. Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab209344** in HeLa whole cell lysate. Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 8 seconds.

Why choose a recombinant antibody?

 <p>Research with confidence Consistent and reproducible results</p>	 <p>Long-term and scalable supply Recombinant technology</p>
 <p>Success from the first experiment Confirmed specificity</p>	 <p>Ethical standards compliant Animal-free production</p>

Anti-Galectin 3 antibody [EPR19244] - BSA and Azide free (ab251504)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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