abcam

ab232571

Anti-PRAME antibody [EPR20330] - BSA and Azide free

Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571) is a rabbit recombinant monoclonal antibody provided in a PBS only buffer for easy conjugation detecting PRAME in **Western Blot, Flow Cytometry (Intra), IP, IHC-P, ICC/IF**. Suitable for **Human**.

- Clone EPR20330 is the most cited clone to PRAME
- BSA, sodium azide, and glycerol-free for easy conjugation
- Biophysical QC for unrivalled batch-batch consistency

Recombinant | RabMAb

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR20330
Purification technique	Affinity purification Protein A
Specificity	PRAME is expressed in malignant cells, including leukaemias, Hodgkin's lymphoma, breast cancer, and primary and metastatic melanomas.
Concentration	1.011 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	Recommend ab219650 incubation at +4°C overnight.

Target data

See full target information PRAME

Function

Substrate-recognition component of a Cul2-RING (CRL2) E3 ubiquitin-protein ligase complex, which mediates ubiquitination of target proteins, leading to their degradation (PubMed:21822215, PubMed:26138980). The CRL2(PRAME) complex mediates ubiquitination and degradation of truncated MSRB1/SEPX1 selenoproteins produced by failed UGA/Sec decoding (PubMed:26138980). In the nucleus, the CRL2(PRAME) complex is recruited to epigenetically and transcriptionally active promoter regions bound by nuclear transcription factor Y (NFY) and probably plays a role in chromstin regulation (PubMed:21822215). Functions as a transcriptional repressor, inhibiting the signaling of retinoic acid through the retinoic acid receptors RARA, RARB and RARG: prevents retinoic acid-induced cell proliferation arrest, differentiation and apoptosis (PubMed:16179254).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

What is this antibody validated in?
Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Flow Cytometry (Flow Cyt), Immunoprecipitation (IP), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF) in Human samples.

What is the molecular weight of PRAME?
Anti-PRAME [EPR20330] - BSA and Azide free (ab232571) specifically detects a band for PRAME (UniProt: P78395) at a molecular weight of 57kDa.

Other related products

We have a range of other formats of antibody clone [EPR20330] also available for your convenience: ab219650, Carrier free - ab232571, Alexa Fluor® 555 - ab307626, Alexa Fluor® 647 - ab307630, Alexa Fluor® 488 - ab307769, Alexa Fluor® 594 - ab312896

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.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. 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.

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.

Compatibility
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.

Supplementary info

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

Activity summary	PRAME also known as Preferentially Expressed Antigen in Melanoma is a protein typically weighing around 50 kDa. It is found mostly in testis tissue but also appears in several types of tumors including melanoma and some leukemias. PRAME does not show significant expression in most normal tissues making it a potential target for cancer therapies. Immunohistochemistry (IHC) techniques such as PRAME IHC and PRAME staining often use monoclonal antibodies like mAb EPR20330 to detect PRAME protein presence in various samples.
Biological function summary	PRAME acts as a repressor of retinoic acid signaling a process important for cell differentiation and growth. It does not directly form part of a complex but interacts with components involved in retinoic acid pathways. By binding to retinoic acid receptor complexes PRAME prevents activation of genes involved in cell cycle arrest and apoptosis contributing to unchecked cellular proliferation seen in certain cancers.
Pathways	PRAME participates in the retinoic acid and various oncogenic signaling processes. It influences the retinoic acid pathway by interfering with the retinoic acid receptor (RAR) signaling. This interference with key proteins like RAR disrupts the normal

regulatory processes that typically inhibit cancer progression. PRAME's modulation of these pathways highlights its role in promoting tumor growth and survival.

Associated diseases and disorders

PRAME has significant relevance to cancer particularly melanoma. It is often used as a biomarker for melanoma identification due to its overexpression in such tumors. Additionally PRAME's connection to acute myeloid leukemia (AML) emerges from its ability to act similarly in diverse malignant cells. In these diseases the overexpression of PRAME interferes with normal differentiation processes and allows for sustained proliferation of malignant cells suggesting its potential role as a therapeutic target or diagnostic marker in oncology.

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

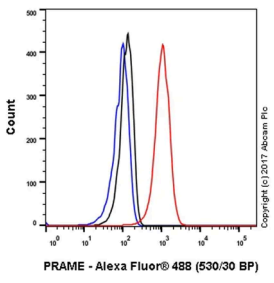
We do not recommend this combination. It is not covered by our product promise.

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.

14 product images

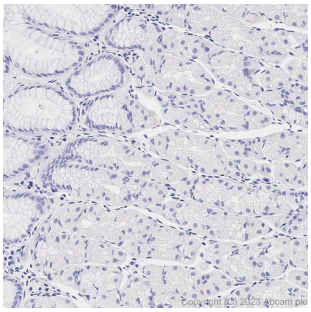


Flow Cytometry (Intracellular) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Flow Cytometry (Intracellular) staining of MeWo (Human malignant melanoma cell line) cells using rabbit Anti-PRAME antibody

PRAME flow cytometry with PRAME antibody ab219650 of 4% paraformaldehyde-fixed and 90% Methanol-permeabilised MeWo (Human malignant melanoma cell line) cells labeling PRAME with ab219650 at 1/500 dilution (red) compared with a rabbit monoclonal IgG isotype control

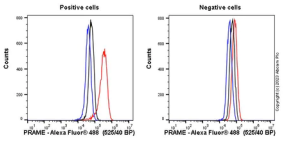
(ab172730; black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (Alexa Fluor® 488) (ab150077) at 1/2000 dilution was used as the secondary antibody.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human stomach using rabbit Anti-PRAME antibody

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650). Immunohistochemical analysis of paraffin-embedded human stomach tissue labeling PRAME with ab219650 at 1/500 dilution, followed by LeicaDS9800 (Bond™ Polymer Refine Detection). Sections were counter stained with Hematoxylin. Antigen retrieval was heat mediated with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Negative control: No staining on human stomach. The section was incubated with ab219650 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Flow Cytometry (Intracellular) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

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

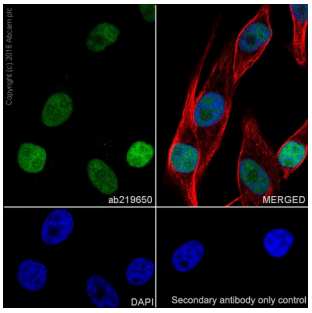
Flow cytometry overlay histogram showing left K562 positive cells and right negative HEK293 stained with ab219650 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilised with 0.1 % PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab219650) (1x 10^6 in 100µl at 0.008µg/ml (1/267500 dilution)) for 30min at 22°C. The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C.

Isotype control antibody (black line) was Recombinant Rabbit IgG monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

This antibody gave a positive signal in K562 Fixed with 80% methanol (5 min) / permeabilised with 0.1 % PBS-Triton X-100 for 15 min under the same conditions.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650).



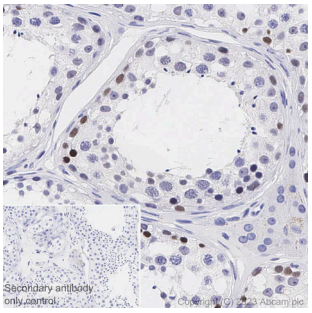
Immunocytochemistry/ Immunofluorescence - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunocytochemistry/ Immunofluorescence staining of MeWo (Human malignant melanoma cell line) cells using rabbit Anti-PRAME antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed 0.1% Triton X-100 permeabilized MeWo (Human malignant melanoma cell line) cells labeling PRAME with ab219650 at 1/500 dilution followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing mostly nuclear staining on MeWo cells. The nuclear counter stain is DAPI (blue). Tubulin is detected with ab195889 (Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594)) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody secondary antibody is Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) at 1/1000 dilution.

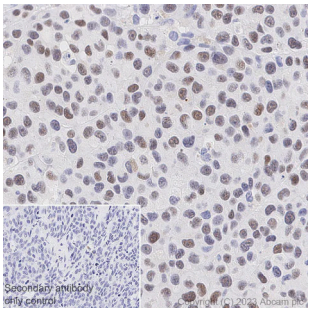
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human testis using rabbit Anti-PRAME antibody

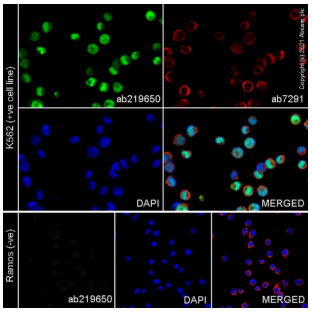
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650). Immunohistochemical analysis of paraffin-embedded human testis tissue labeling PRAME with ab219650 at 1/500 dilution, followed by LeicaDS9800 (Bond™ Polymer Refine Detection). Sections were counter stained with Hematoxylin. Antigen retrieval was heat mediated with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Nuclear staining on human testis. The section was incubated with ab219650 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Secondary antibody only control: Used PBS instead of primary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human melanoma tissue using rabbit Anti-PRAME antibody

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650). Immunohistochemical analysis of paraffin-embedded human melanoma tissue labeling PRAME with ab219650 at 1/500 dilution, followed by LeicaDS9800 (Bond™ Polymer Refine Detection). Sections were counter stained with Hematoxylin. Antigen retrieval was heat mediated with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Nuclear staining on human melanoma. The section was incubated with ab219650 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Secondary antibody only control: Used PBS instead of primary antibody.



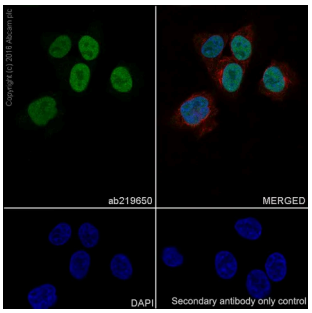
Immunocytochemistry/ Immunofluorescence - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-PRAME antibody

PRAME immunofluorescence with PRAME antibody ab219650 in K562 cells with negative expression in Ramos cells. The cells were fixed with 4% formaldehyde (10 min) permeabilised with 0.1% Triton x-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab219650 at 1 µg/ml and ab7291 Mouse monoclonal [DM1A] to alpha Tubulin at 0.5 µg/ml. Cells were then incubated with ab150081 Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488) pre-adsorbed at 1/1000 dilution (shown in green) and ab150119 Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 647) pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).
Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.

This product also work with 100% methanol (5 min) fixation under the same testing conditions.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650).



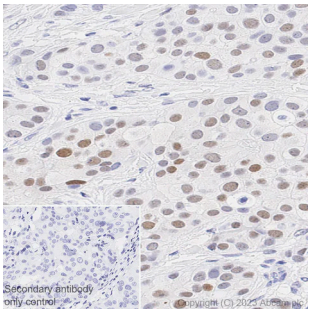
Immunocytochemistry/ Immunofluorescence - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunocytochemistry/ Immunofluorescence staining of A-375 (Human malignant melanoma cell line) cells using rabbit Anti-PRAME antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed 0.1% Triton X-100 permeabilized A-375 (Human malignant melanoma cell line) cells labeling PRAME with ab219650 at 1/500 dilution followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing mostly nuclear staining on A-375 cells.
The nuclear counter stain is DAPI (blue). Tubulin is detected with ab195889 (Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594)) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody secondary antibody is Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) at 1/1000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650).

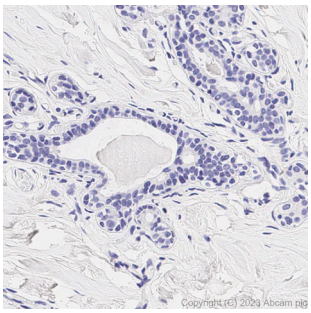


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human breast carcinoma using rabbit Anti-PRAME antibody

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650). Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue labeling PRAME with ab219650 at 1/500 dilution, followed by LeicaDS9800 (Bond™ Polymer Refine Detection). Sections were counter stained with Hematoxylin. Antigen retrieval was heat mediated with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Nuclear staining on human breast carcinoma. The section was incubated with ab219650 for 30 mins at room temperature. The immunostaining was

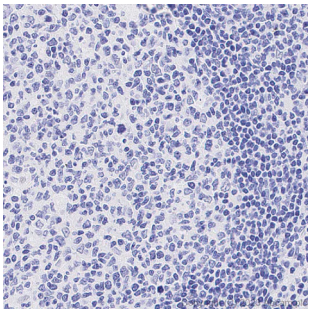
performed on a Leica Biosystems BOND® RX instrument. Secondary antibody only control: Used PBS instead of primary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human breast using rabbit Anti-PRAME antibody

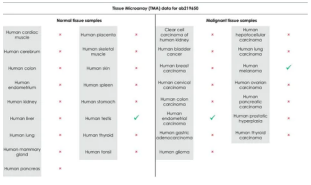
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650). Immunohistochemical analysis of paraffin-embedded human breast tissue labeling PRAME with ab219650 at 1/500 dilution, followed by LeicaDS9800 (Bond™ Polymer Refine Detection). Sections were counter stained with Hematoxylin. Antigen retrieval was heat mediated with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Negative control: No staining on human breast. The section was incubated with ab219650 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human tonsil using rabbit Anti-PRAME antibody

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650). Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling PRAME with ab219650 at 1/500 dilution, followed by LeicaDS9800 (Bond™ Polymer Refine Detection). Sections were counter stained with Hematoxylin. Antigen retrieval was heat mediated with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Negative control: No staining on human tonsil. The section was incubated with ab219650 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

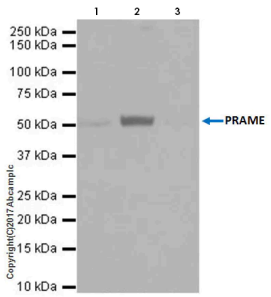


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

Tissue microarray (TMA) analysis using PRAME EPR20330 (ab219650).
Click here to view EPR20330 staining performance on human normal and cancer tissue microarray (TMA).

This table provides a detailed overview of positive PRAME staining (tick mark) and negative PRAME staining (cross mark) per sample type tested. PRAME is expressed in metastatic melanoma (PMID: 30045064). PRAME has low or no expression in normal tissues except in testis, ovary, placenta, adrenals and endometrium (PMID: 30045064).

The sections were pre-treated using Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0). The sections were incubated with ab219650 at +4°C overnight followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer).



Immunoprecipitation - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME was immunoprecipitated from 0.35 mg of MeWo (Human malignant melanoma cell line) whole cell lysate with ab219650 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab219650 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.
Lane 1: MeWo whole cell lysate 10 µg (Input).

Lane 2: ab219650 IP in MeWo whole cell lysate.

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab219650 in MeWo whole cell lysate.

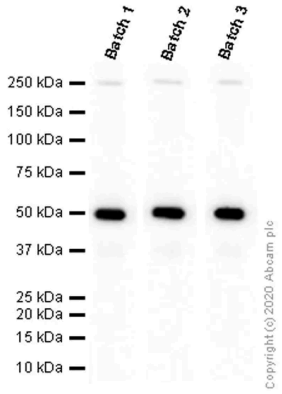
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 1 second.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab219650).

All lanes:
Immunoprecipitation - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

Predicted band size: 57 kDa



Western blot - Anti-PRAME antibody [EPR20330] - BSA and Azide free (ab232571)

PRAME Western blot staining using rabbit Anti-PRAME antibody

This data was developed using ab219650, the same antibody clone in a different buffer formulation.
PRAME western blot with PRAME antibody ab219650.

Different batches of ab219650 were tested on MeWo (Human malignant melanoma fibroblast) whole cell lysate at 0.1 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 50 kDa.

All lanes:
Western blot - Anti-PRAME antibody [EPR20330] (ab219650)

Predicted band size: 57 kDa

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