abcam

Product datasheet

Anti-BubR1 antibody [EPR12259(2)] - BSA and Azide free ab250655



7 Images

Overview

Immunogen

Product name Anti-BubR1 antibody [EPR12259(2)] - BSA and Azide free

Description Rabbit monoclonal [EPR12259(2)] to BubR1 - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), IHC-P, IP, WB

Unsuitable for: ICC/IF

Species reactivity Reacts with: Human

Predicted to work with: Mouse, Rat

Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

General notes ab250655 is the carrier-free version of <u>ab183496</u>.

Our <u>carrier-free</u> 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.

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.

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.

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**.

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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 EPR12259(2)

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab250655 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.
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.
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 120 kDa (predicted molecular weight: 120 kDa).

Application notes

Is unsuitable for ICC/IF.

Target

Function

Essential component of the mitotic checkpoint. Required for normal mitosis progression. The mitotic checkpoint delays anaphase until all chromosomes are properly attached to the mitotic spindle. One of its checkpoint functions is to inhibit the activity of the anaphase-promoting complex/cyclosome (APC/C) by blocking the binding of CDC20 to APC/C, independently of its kinase activity. The other is to monitor kinetochore activities that depend on the kinetochore motor CENPE. Required for kinetochore localization of CENPE. Negatively regulates PLK1 activity in interphase cells and suppresses centrosome amplification. Also implicated in triggering apoptosis in polyploid cells that exit aberrantly from mitotic arrest. May play a role for tumor suppression.

Tissue specificity

Highly expressed in thymus followed by spleen. Preferentially expressed in tissues with a high mitotic index.

Involvement in disease

Note=Defects in BUB1B are associated with tumor formation.

Defects in BUB1B are the cause of premature chromatid separation trait (PCS) [MIM:176430]. PCS consists of separate and splayed chromatids with discernible centromeres and involves all

or most chromosomes of a metaphase. It is found in up to 2% of metaphases in cultured lymphocytes from approximately 40% of normal individuals. When PCS is present in 5% or more of cells, it is known as the heterozygous PCS trait and has no obvious phenotypic effect, although some have reported decreased fertility. Inheritance is autosomal dominant.

Defects in BUB1B are the cause of mosaic variegated aneuploidy syndrome (MVA) [MIM:257300]. MVA is a severe autosomal recessive developmental disorder characterized by mosaic aneuploidies, predominantly trisomies and monosomies, involving multiple different chromosomes and tissues. The proportion of aneuploid cells varies but is usually more than 25% and is substantially greater than in normal individuals. Affected individuals typically present with severe intrauterine growth retardation and microcephaly. Eye anomalies, mild dysmorphism, variable developmental delay, and a broad spectrum of additional congenital abnormalities and medical conditions may also occur. The risk of malignancy is high, with rhabdomyosarcoma, Wilms tumor and leukemia reported in several cases. MVA is caused by biallelic mutations in the BUB1B gene.

Sequence similarities

Belongs to the protein kinase superfamily. Ser/Thr protein kinase family. BUB1 subfamily.

Contains 1 BUB1 N-terminal domain. Contains 1 protein kinase domain.

Domain

The D-box targets the protein for rapid degradation by ubiquitin-dependent proteolysis during the transition from mitosis to interphase.

The BUB1 N-terminal domain directs kinetochore localization and binding to BUB3.

Post-translational modifications

Proteolytically cleaved by caspase-3 in a cell cycle specific manner. The cleavage might be involved in the durability of the cell cycle delay. Caspase-3 cleavage is associated with

abrogation of the mitotic checkpoint. The major site of cleavage is at Asp-610. Acetylation at Lys-250 regulates its degradation and timing in anaphase entry.

Ubiquitinated. Degradated by the proteasome.

Sumoylated by SUMO2 and SUMO3. The sumoylation mediates the association with CENPE at

the kinetochore.

Autophosphorylated in vitro. Intramolecular autophosphorylation is stimulated by CENPE. Phosphorylated during mitosis and hyperphosphorylated in mitotically arrested cells. Phosphorylation at Ser-670 and Ser-1043 occurs at kinetochores upon mitotic entry with

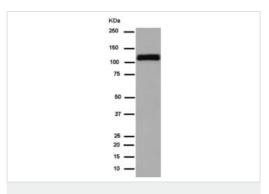
dephosphorylation at the onset of anaphase.

Cellular localization

 $\label{lem:cytoplasm} \begin{tabular}{ll} Cytoplasm. Nucleus. Chromosome > centromere > kinetochore. Cytoplasm > cytoskeleton > centrosome. Cytoplasmic in interphase cells. Associates with the kinetochores in early prophase. \\ \end{tabular}$

Kinetochore localization requires BUB1, PLK1 and CASC5.

Images



Western blot - Anti-BubR1 antibody [EPR12259(2)] -BSA and Azide free (ab250655)

Anti-BubR1 antibody [EPR12259(2)] (ab183496) at 1/20000 dilution + HeLa lysate at 20 μg

Secondary

Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 120 kDa

This data was developed using <u>ab183496</u>, the same antibody clone in a different buffer formulation.

Western blot - Anti-BubR1 antibody [EPR12259(2)] - BSA and Azide free (ab250655)

All lanes : Anti-BubR1 antibody [EPR12259(2)] (<u>ab183496</u>) at 1/100000 dilution

Lane 1 : HepG2 cell lysate

Lane 2 : Jurkat cell lysate

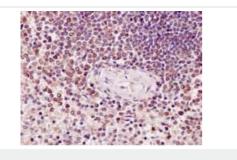
Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 120 kDa

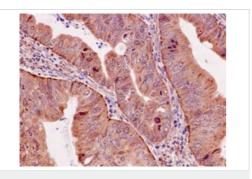
This data was developed using <u>ab183496</u>, the same antibody clone in a different buffer formulation.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-BubR1 antibody

[EPR12259(2)] - BSA and Azide free (ab250655)

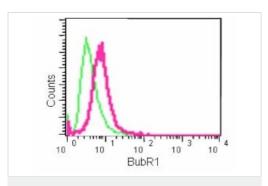
This data was developed using <u>ab183496</u>, the same antibody clone in a different buffer formulation.lmmunohistochemical analysis of paraffin-embedded Human spleen labeling BubR1 with <u>ab183496</u> at 1/100 dilution and HRP conjugated Rabbit lgG. Counterstained with Hematoxylin. Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-BubR1 antibody

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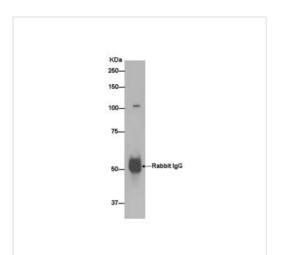
This data was developed using <u>ab183496</u>, the same antibody clone in a different buffer formulation.lmmunohistochemical analysis of paraffin-embedded Human ovarian carcinoma labeling BubR1 with <u>ab183496</u> at 1/100 dilution and HRP conjugated Rabbit lgG. Counterstained with Hematoxylin. Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-BubR1 antibody [EPR12259(2)] - BSA and Azide free (ab250655)

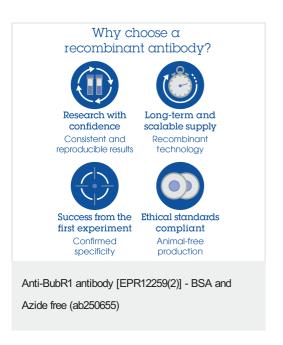
This data was developed using <u>ab183496</u>, the same antibody clone in a different buffer formulation.

Intracellular flow cytometric analysis of HeLa cells fixed in 2% paraformaldehyde labeling BubR1 with <u>ab183496</u> at 1/100 dilution and Goat anti rabbit lgG (FITC). Rabbit monoclonal lgG was used as an isotype control.



Immunoprecipitation - Anti-BubR1 antibody
[EPR12259(2)] - BSA and Azide free (ab250655)

This data was developed using <u>ab183496</u>, the same antibody clone in a different buffer formulation.lmmunoprecipitation of HepG2 cells labeling BubR1 with <u>ab183496</u> at 1/50 dilution and Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000.



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