# abcam

## Product datasheet

## Anti-Bubl antibody [2F9] ab54893

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#### Overview

Product name Anti-Bub1 antibody [2F9]

**Description** Mouse monoclonal [2F9] to Bub1

Host species Mouse

Tested applications
Suitable for: Flow Cyt
Species reactivity
Reacts with: Human

**Immunogen** Recombinant fragment corresponding to Human Bub1 aa 1-130.

Database link: **O43683** 

**General notes**This product was changed from ascites to tissue culture supernatant on 25/02/19. Please note

that the dilutions may need to be adjusted accordingly. If you have any questions, please do not

hesitate to contact our scientific support team.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

## **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw

cycles.

Storage buffer pH: 7.40

**Purity** Tissue culture supernatant

**Clonality** Monoclonal

Clone number 2F9

Isotype IgG1

Light chain type kappa

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The Abpromise guarantee

Our Abpromise guarantee covers the use of ab54893 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		Use at an assay dependent concentration. <u>ab170190</u> - Mouse monoclonal lgG1, is suitable for use as an isotype control with this antibody.

### **Target**

#### **Function**

Serine/threonine-protein kinase that performs 2 crucial functions during mitosis: it is essential for spindle-assembly checkpoint signaling and for correct chromosome alignment. Has a key role in the assembly of checkpoint proteins at the kinetochore, being required for the subsequent localization of CENPF, BUB1B, CENPE and MAD2L1. Required for the kinetochore localization of PLK1. Plays an important role in defining SGOL1 localization and thereby affects sister chromatid cohesion. Acts as a substrate for anaphase-promoting complex or cyclosome (APC/C) in complex with its activator CDH1 (APC/C-Cdh1). Necessary for ensuring proper chromosome segregation and binding to BUB3 is essential for this function. Can regulate chromosome segregation in a kinetochore-independent manner. Can phosphorylate BUB3. The BUB1-BUB3 complex plays a role in the inhibition of APC/C when spindle-assembly checkpoint is activated and inhibits the ubiquitin ligase activity of APC/C by phosphorylating its activator CDC20. This complex can also phosphorylate MAD1L1. Kinase activity is essential for inhibition of APC/CCDC20 and for chromosome alignment but does not play a major role in the spindle-assembly checkpoint activity. Mediates cell death in response to chromosome missegregation and acts to suppress spontaneous tumorigenesis.

## Tissue specificity

High expression in testis and thymus, less in colon, spleen, lung and small intestine. Expressed in fetal thymus, bone marrow, heart, liver, spleen and thymus. Expression is associated with cells/tissues with a high mitotic index.

## 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 KEN box is required for its ubiquitination and degradation.

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

## Post-translational modifications

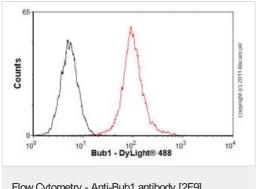
Phosphorylated upon DNA damage, probably by ATM or ATR. Upon spindle-assembly checkpoint activation it is hyperphosphorylated and its kinase activity toward CDC20 is stimulated. Phosphorylation at Thr-609 is required for interaction with PLK1, phosphorylation at this site probably creates a binding site for the POLO-box domain of PLK1, thus enhancing the PLK1-BUB1 interaction.

Ubiquitinated and degraded during mitotic exit by APC/C-Cdh1.

#### **Cellular localization**

Nucleus. Chromosome > centromere > kinetochore. Nuclear in interphase cells. Accumulates gradually during G1 and S phase of the cell cycle, peaks at G2/M, and drops dramatically after mitosis. Localizes to the outer kinetochore. Kinetochore localization is required for normal mitotic timing and checkpoint response to spindle damage and occurs very early in prophase. AURKB, CASC5 and INCENP are required for kinetochore localization.

### **Images**



Flow Cytometry - Anti-Bub1 antibody [2F9] (ab54893)

Overlay histogram showing HeLa cells stained with ab54893 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab54893,  $1\mu g/1x10^6$  cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse lgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse lgG1 [ICIGG1] (ab91353,  $2\mu g/1x10^6$  cells ) used under the same conditions. Acquisition of >5,000 events was performed.

This image was generated using the ascites version of the product.

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

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