# abcam

#### Product datasheet

## Anti-beta Tubulin antibody - Loading Control ab6046

★★★★★ 61 Abreviews 1124 References 6 图像

概述

免疫原

产品名称 Anti-beta Tubulin抗体- Loading Control

描述 兔多克隆抗体to beta Tubulin - Loading Control

宿主 Rabbit

特异性 This antibody detects a single clean band at 50kD representing beta Tubulin. This band is

significantly reduced by using peptide blocking.

经测试应用 适用于: WB, ICC/IF, IHC-P, IP

预测可用于: Chicken, Pig, Xenopus laevis, Zebrafish 4

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

(Peptide available as ab20775)

阳性对照 WB: HeLa, A431, MCF7, NIH3T3, PC12 CHO/K1, and 293 cell lysates; IP: HeLa whole cell

extract: ICC/IF: HeLa cells: IHC-P: Human liver carcinoma tissue section.

常规说明

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

性能

形式 Liquid

**存放说明** Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

**存储溶液** pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising

agent. If you would like information about the formulation of a specific lot, please contact our

scientific support team who will be happy to help.

纯**度** Immunogen affinity purified

 克隆
 多克隆

 同种型
 lqG

应用

#### The Abpromise guarantee Abpromise™承诺保证使用ab6046于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★★ (38)	1/500. Detects a band of approximately 50 kDa (predicted molecular weight: 50 kDa).
ICC/IF	**** <u>(11)</u>	Use a concentration of 1 µg/ml.
IHC-P	****(1)	Use a concentration of 5 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
IP	<b>★★★★★ (1)</b>	Use at an assay dependent concentration.

靶标

功能 Tubulin is the major constituent of microtubules. It binds two moles of GTP, one at an

exchangeable site on the beta chain and one at a non-exchangeable site on the alpha chain.

组织特异性 Ubiquitously expressed with highest levels in spleen, thymus and immature brain.

**疾病相关** Cortical dysplasia, complex, with other brain malformations 6

Skin creases, congenital symmetric circumferential, 1

序列相似性 Belongs to the tubulin family.

结**构域** The highly acidic C-terminal region may bind cations such as calcium.

翻译后修饰 Some glutamate residues at the C-terminus are polyglutamylated, resulting in polyglutamate

chains on the gamma-carboxyl group (PubMed:26875866). Polyglutamylation plays a key role in microtubule severing by spastin (SPAST). SPAST preferentially recognizes and acts on microtubules decorated with short polyglutamate tails: severing activity by SPAST increases as

the number of glutamates per tubulin rises from one to eight, but decreases beyond this

glutamylation threshold (PubMed:26875866).

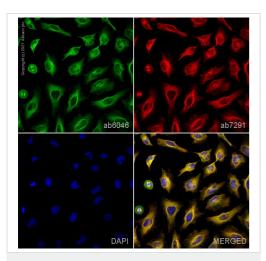
Some glutamate residues at the C-terminus are monoglycylated but not polyglycylated due to the absence of functional TTLL10 in human. Monoglycylation is mainly limited to tubulin incorporated into axonemes (cilia and flagella). Both polyglutamylation and monoglycylation can coexist on the same protein on adjacent residues, and lowering glycylation levels increases polyglutamylation,

and reciprocally. The precise function of monoglycylation is still unclear.

Phosphorylated on Ser-172 by CDK1 during the cell cycle, from metaphase to telophase, but not

in interphase. This phosphorylation inhibits tubulin incorporation into microtubules.

细胞定位 Cytoplasm, cytoskeleton.



Immunocytochemistry/ Immunofluorescence - Antibeta Tubulin antibody - Loading Control (ab6046)

1 2 3 4 5 6 7 8

250 kDa \_\_\_
150 kDa \_\_\_
100 kDa \_\_\_
75 kDa \_\_\_

50 kDa \_\_\_

37 kDa \_\_\_

25 kDa \_\_\_
20 kDa \_\_\_

Western blot - Anti-beta Tubulin antibody - Loading Control (ab6046)

(c) 2005 Abcam plc.

15 kDa 10 kDa ab6046 staining beta Tubulin in HeLa cells. The cells were fixed with 4% paraformaldehyde (10 min), permeabilized with 0.1% PBS-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 ab6046 at 1µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit lgG - H&L (Alexa Fluor® 488), preadsorbed at 1/1000 dilution (shown in green) and ab150120, Goat polyclonal Secondary Antibody to Mouse lgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Also suitable in cells fixed with 100% methanol (5 min).

Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.

**All lanes :** Anti-beta Tubulin antibody - Loading Control (ab6046) at 1/500 dilution

Lane 1: HeLa Cell lysate

Lane 2: A431 Cell lysate

Lane 3: MCF7 Cell lysate

Lane 4: 293 Cell lysate

Lane 5: HeLa Cell lysate with Human beta Tubulin peptide

(<u>ab20775</u>) at 1 µg/ml

Lane 6: A431 Cell lysate with Human beta Tubulin peptide

(ab20775) at 1 µg/ml

Lane 7: MCF7 Cell lysate with Human beta Tubulin peptide

(ab20775) at 1 µg/ml

Lane 8: 293 Cell lysate with Human beta Tubulin peptide

(ab20775) at 1 µg/ml

Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (ab6721) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size:** 50 kDa **Observed band size:** 55 kDa

Exposure time: 10 seconds

**All lanes :** Anti-beta Tubulin antibody - Loading Control (ab6046) at 1  $\mu$ g/ml

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

**Lane 2**: NIH3T3 (Mouse embryo fibroblast cell line) whole cell lysate

**Lane 3**: PC12 (Rat adrenal gland pheochromocytoma cell line) whole cell lysate

Lane 4 : CHO/K1 (Chinese hamster ovary cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

1 2 3 4

250 kDa —
150 kDa —
100 kDa —
75 kDa —
37 kDa —
25 kDa —
20 kDa —
20 kDa —
15 kDa —

Western blot - Anti-beta Tubulin antibody - Loading Control (ab6046)

### Secondary

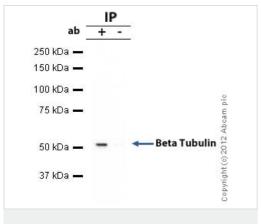
 $\begin{tabular}{ll} \textbf{All lanes:} Goat polyclonal to Rabbit lgG - H\&L - Pre-Adsorbed (HRP) at 1/50000 dilution \end{tabular}$ 

Developed using the ECL technique.

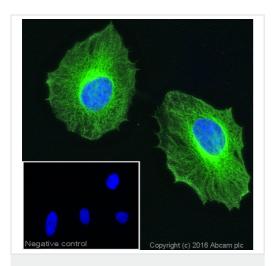
Performed under reducing conditions.

Predicted band size: 50 kDa Observed band size: 51 kDa

Exposure time: 30 seconds



Immunoprecipitation - Anti-beta Tubulin antibody - Loading Control (ab6046)



Immunocytochemistry/ Immunofluorescence - Antibeta Tubulin antibody - Loading Control (ab6046)

Beta Tubulin was immunoprecipitated using 0.5mg Hela whole cell extract,  $5\mu g$  of Rabbit polyclonal to Tubulin and  $50\mu l$  of protein G magnetic beads (+). No antibody was added to the control (-). The antibody was incubated under agitation with Protein G beads for 10min, Hela whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of  $40\mu$ I SDS loading buffer and incubated for 10min at  $70^{o}$ C;  $10\mu$ I of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab6046.

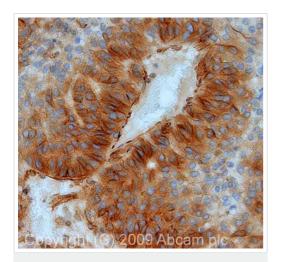
Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit lgG light chain (HRP) (ab99697).

Band: 50kDa: beta Tubulin.

ICC/IF image of ab6046 stained HeLa cells. The cells were 4% formaldehyde fixed (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then incubated in 1% BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to block nonspecific protein-protein interactions. The cells were then incubated with the antibody (ab6046, 1µg/ml) overnight at +4°C. The secondary antibody (green) was  $\underline{ab150081}$  Alexa Fluor  $^{\$}$  488 goat anti-rabbit lgG (H+L) used at a 1/1000 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.

The negative control (inset) is a secondary-only assay to demonstrate low non-specific binding of the secondary antibody.

This product also gave a positive signal under the same testing conditions in HeLa cells fixed with 100% methanol (5 min).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-beta Tubulin antibody - Loading Control (ab6046)

IHC image of beta Tubulin staining in human liver carcinoma FFPE section, performed on a Leica Bond<sup>TM</sup> system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab6046, 5µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

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

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