**Product datasheet**

**Anti-beta Tubulin antibody [EP1331Y] - Microtubule Marker ab52901**

- **Host species**: Rabbit
- **Tested applications**: Suitable for: WB, IP, IHC-P, IHC-Fr
- **Species reactivity**: Reacts with: Mouse, Rat, Human, Zebrafish
- **Immunogen**: Synthetic peptide within Human beta Tubulin aa 400 to the C-terminus (C terminal). The exact sequence is proprietary. Database link: P07437
- **Positive control**: WB: PC12 and HeLa whole cell lysate (ab150035) and mouse brain, rat brain, mouse spinal cord and rat spinal cord tissue lysates. IHC-P: Human gastric carcinoma. ICC/IF: HeLa cells.
- **General notes**: 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.

**Properties**

- **Form**: Liquid
- **Storage instructions**: Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
- **Storage buffer**: pH: 7.20
  - Preservative: 0.01% Sodium azide
  - Constituents: 49% PBS, 50% Glycerol, 0.05% BSA
- **Purity**: Tissue culture supernatant
- **Clonality**: Monoclonal
Clone number: EP1331Y
Isotype: IgG

Function
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.

Tissue specificity
Ubiquitously expressed with highest levels in spleen, thymus and immature brain.

Involvement in disease
Cortical dysplasia, complex, with other brain malformations 6
Skin creases, congenital symmetric circumferential, 1

Sequence similarities
Belongs to the tubulin family.

Domain
The highly acidic C-terminal region may bind cations such as calcium.

Post-translational modifications
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.

Cellular localization
Cytoplasm, cytoskeleton.

Images

Applications
Our Abpromise guarantee covers the use of ab52901 in the following tested applications.
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
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</thead>
<tbody>
<tr>
<td>WB</td>
<td>🌟🌟🌟🌟🌟</td>
<td>1/20000. Detects a band of approximately 50 kDa (predicted molecular weight: 50 kDa).</td>
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<tr>
<td>IP</td>
<td></td>
<td>1/50.</td>
</tr>
<tr>
<td>IHC-P</td>
<td>🌟🌟🌟🌟🌟</td>
<td>Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.</td>
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<tr>
<td>IHC-Fr</td>
<td>🌟🌟🌟🌟🌟</td>
<td>1/100.</td>
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Target

Images 2
**Western blot - Anti-beta Tubulin antibody [EP1331Y] - Microtubule Marker (ab52901)**

**All lanes**: Anti-beta Tubulin antibody [EP1331Y] - Microtubule Marker (ab52901) at 1/20000 dilution

**Lane 1**: Brain (Mouse) Tissue Lysate

**Lane 2**: Brain (Rat) Tissue Lysate

**Lane 3**: Spinal Cord (Mouse) Tissue Lysate

**Lane 4**: Spinal Cord (Rat) Tissue Lysate

**Lane 5**: PC12 (Rat adrenal pheochromocytoma cell line) Whole Cell Lysate

Lysates/proteins at 20 µg per lane.

**Secondary**

**All lanes**: Goat Anti-Rabbit IgG H&L (Alexa Fluor® 790) (**ab175781**) at 1/10000 dilution

**Predicted band size**: 50 kDa

**Observed band size**: 52 kDa

*why is the actual band size different from the predicted?*

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using Licor blocking buffer before being incubated with ab52901 overnight at 4°C. Antibody binding was detected using ab175781 at a 1:10,000 dilution for 1hr at room temperature and then imaged using the Licor Odyssey CLx.
Immunohistochemistry (Frozen sections) - Anti-beta Tubulin antibody [EP1331Y] - Microtubule Marker (ab52901)
This image is courtesy of an abreview submitted by Dr. Ryan MacDonald (Cambridge University, United Kingdom)

IHC-Fr image of beta Tubulin staining on zebrafish retina sections using ab52901 (1:100). The sections were paraformaldehyde and permeabilized using Triton-X. Antigen retrieval was performed using Sodium Citrate and blocking was performed using 5% BSA for 1 hour at 23°C. ab52901 was diluted 1:100 and incubated with the sections for 16 hours at 4°C. The secondary antibody was goat polyclonal to rabbit IgG conjugated to Alexa Fluor 488 (1:1000).

Western blot - Anti-beta Tubulin antibody [EP1331Y] - Microtubule Marker (ab52901) at 1/20000 dilution + HeLa cell lysate at 10 µg

Secondary
goat anti-rabbit HRP at 1/2000 dilution

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

ab52901 at 1/250 dilution staining beta Tubulin in human gastric carcinoma by Immunohistochemistry, Paraffin embedded tissue.
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
Western blot - Anti-beta Tubulin antibody [EP1331Y] - Microtubule Marker (ab52901) at 1/1000 dilution (in PBS +0.5% Tween20 for 2 hours at 23°C) + 293 human embryonic kidney whole cell lysate at 25 µg

**Secondary**
An HRP-conjugated Goat anti-rabbit IgG polyclonal at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size:** 50 kDa

**Observed band size:** 55 kDa

why is the actual band size different from the predicted?

**Exposure time:** 45 seconds

**Blocking Step:** 5% Milk for 1 hour at 23°C

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