**Product datasheet**

**Anti-Casein Kinase 1 delta/CSNK1D antibody [AF12G4]**

**Product name**

Anti-Casein Kinase 1 delta/CSNK1D antibody [AF12G4]

**Description**

Mouse monoclonal [AF12G4] to Casein Kinase 1 delta/CSNK1D

**Host species**

Mouse

**Tested applications**

Suitable for: IHC-P, Flow Cyt, WB, IP, ELISA

**Species reactivity**

Reacts with: Mouse, Rat, Human

**Immunogen**

Recombinant full length protein (His-tag) corresponding to Human Casein Kinase 1 delta/CSNK1D aa 1-415. Purified from E.coli.

Database link: P48730

**Positive control**

WB: HeLa, K562, SK-N-MC, 293T and L929 cell lysates. IHC-P: Human Alzheimer brain (hippocampus) tissue section.

**General notes**

This product was changed from ascites to tissue culture supernatant on 18th September 2017. Lot numbers higher than GR155202 will be from tissue culture supernatant. Please note that the dilutions may need to be adjusted accordingly.

Previously labelled as Casein Kinase 1 delta.

**Properties**

**Form**

Liquid

**Storage instructions**

Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

**Storage buffer**

Preservative: 0.03% Sodium azide

Constituents: 0.01% BSA, 50% Glycerol, 0.87% Sodium chloride, HEPES

**Purity**

Protein G purified

**Clonality**

Monoclonal

**Clone number**

AF12G4

**Isotype**

IgG2b

**Light chain type**

kappa
**Applications**

Our [Abpromise guarantee](#) covers the use of [ab85320](#) 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>
</tr>
</thead>
<tbody>
<tr>
<td>IHC-P</td>
<td></td>
<td>Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.</td>
</tr>
<tr>
<td>Flow Cyt</td>
<td></td>
<td>Use at an assay dependent concentration. ab170192 - Mouse monoclonal IgG2b, is suitable for use as an isotype control with this antibody.</td>
</tr>
<tr>
<td>WB</td>
<td>⭐⭐⭐⭐⭐</td>
<td>Use a concentration of 0.05 µg/ml. Detects a band of approximately 47 kDa (predicted molecular weight: 47 kDa).</td>
</tr>
<tr>
<td>IP</td>
<td></td>
<td>Use at an assay dependent concentration. Use at 2µl</td>
</tr>
<tr>
<td>ELISA</td>
<td></td>
<td>Use at an assay dependent concentration.</td>
</tr>
</tbody>
</table>

**Target**

**Function**

Casein kinases are operationally defined by their preferential utilization of acidic proteins such as caseins as substrates. It can phosphorylate a large number of proteins. Participates in Wnt signaling. Central component of the circadian clock. May act as a negative regulator of circadian rhythmicity by phosphorylating PER1 and PER2. Retains PER1 in the cytoplasm.

**Tissue specificity**

Expressed in all tissues examined, including brain, heart, lung, liver, pancreas, kidney, placenta and skeletal muscle. In blood, highly expressed in hemopoietic cells and mature granulocytes. Also found in monocytes and lymphocytes.

**Involvement in disease**

Defects in CSNK1D are a cause of familial advanced sleep-phase syndrome (FASPS) [MIM:604348]. FASPS is characterized by very early sleep onset and offset. Individuals are 'morning larks' with a 4 hours advance of the sleep, temperature and melatonin rhythms.

**Sequence similarities**

Belongs to the protein kinase superfamily. CK1 Ser/Thr protein kinase family. Casein kinase I subfamily.

Contains 1 protein kinase domain.

**Post-translational modifications**

Autophosphorylated on serine and threonine residues.

**Cellular localization**

Cytoplasm. Nucleus.

**Images**
**Western blot - Anti-Casein Kinase 1 delta/CSNK1D antibody [AF12G4] (ab85320)**

- **All lanes:** Anti-Casein Kinase 1 delta/CSNK1D antibody [AF12G4] (ab85320) at 1/500 dilution

- **Lane 1:** Wild-type HeLa cell lysate
- **Lane 2:** CSNK1D knockout HeLa cell lysate
- **Lane 3:** K-562 cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 47 kDa
**Observed band size:** 47 kDa

**Lanes 1-3:** Merged signal (red and green). Green – ab85320 observed at 47 kDa. Red - loading control, ab181602 observed at 37 kDa.

ab85320 was shown to react with CSNK1D in wild-type HeLa cells in Western blot. Loss of signal was observed when knockout sample ab258383 was used. Wild-type and CSNK1D knockout samples were subjected to SDS-PAGE. ab85320 and Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) were incubated overnight at 4°C at 1 in 500 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.
All lanes: Anti-Casein Kinase 1 delta/CSNK1D antibody [AF12G4] (ab85320) at 1/5000 dilution

Lane 1: HeLa cell lysate
Lane 2: K562 cell lysate
Lane 3: SK-N-MC cell lysate
Lane 4: 293T cell lysate
Lane 5: L929 cell lysate

Predicted band size: 47 kDa
Observed band size: 47 kDa

Overlay histogram showing HeLa cells stained with ab85320 (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 followed by the antibody (ab85320, 1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG2b [PLPV219] (ab91366, 2µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min) permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

IHC image of ab85320 staining in human Alzheimer brain (hippocampus) formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ 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 ab85320, 0.2µg/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.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.
Casein Kinase 1 delta/CSNK1D was immunoprecipitated using 0.5mg Hela whole cell extract, 5µg of Mouse monoclonal to Casein Kinase 1 delta / CSNK1D and 50µl 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 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab85320.

Secondary: Goat polyclonal to mouse IgG light chain specific (HRP) at 1/5000 dilution.

Band: 47kDa: Casein Kinase 1 delta/CSNK1D; non specific - 40kDa: We are unsure as to the identity of this extra band.

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

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