Product datasheet

Anti-p27 KIP 1 (phospho T198) antibody ab64949

Overview

Product name
Anti-p27 KIP 1 (phospho T198) antibody

Description
Rabbit polyclonal to p27 KIP 1 (phospho T198)

Host species
Rabbit

Specificity
Detects endogenous levels of p27 Kip 1 only when phosphorylated at threonine 198.

Tested applications
Suitable for: WB, ELISA

Species reactivity
Reacts with: Mouse, Human

Immunogen
Synthetic phosphopeptide (Human) from around the phosphorylation site of threonine 198 (RRRQT<sup>3</sup>)

Positive control
WB: HeLa, HepG2 and COLO205 cell extracts.

Properties

Form
Liquid

Storage instructions
Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.

Storage buffer
Preservative: 0.02% Sodium Azide
Constituents: 50% Glycerol, PBS (without Mg<sup>2+</sup> and Ca<sup>2+</sup>), 150mM Sodium chloride, pH 7.4

Purity
Immunogen affinity purified

Purification notes
The antibody was affinity purified from rabbit antiserum by affinity chromatography using epitope specific phosphopeptide. The antibody against non phosphopeptide was removed by chromatography using non phosphopeptide corresponding to the phosphorylation site.

Clonality
Polyclonal

Isotype
IgG

Applications

Our Abpromise guarantee covers the use of ab64949 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.
Function
Important regulator of cell cycle progression. Involved in G1 arrest. Potent inhibitor of cyclin E- and cyclin A-CDK2 complexes. Forms a complex with cyclin type D-CDK4 complexes and is involved in the assembly, stability, and modulation of CCND1-CDK4 complex activation. Acts either as an inhibitor or an activator of cyclin type D-CDK4 complexes depending on its phosphorylation state and/or stoichiometry.

Tissue specificity
Expressed in all tissues tested. Highest levels in skeletal muscle, lowest in liver and kidney.

Involvement in disease
Defects in CDKN1B are the cause of multiple endocrine neoplasia type 4 (MEN4) [MIM:610755]. Multiple endocrine neoplasia (MEN) syndromes are inherited cancer syndromes of the thyroid. MEN4 is a MEN-like syndrome with a phenotypic overlap of both MEN1 and MEN2.

Sequence similarities
Belongs to the CDI family.

Domain
A peptide sequence containing only AA 28-79 retains substantial Kip1 cyclin A/CDK2 inhibitory activity.

Post-translational modifications
Phosphorylated; phosphorylation occurs on serine, threonine and tyrosine residues. Phosphorylation on Ser-10 is the major site of phosphorylation in resting cells, takes place at the G(0)-G(1) phase and leads to protein stability. Phosphorylation on other sites is greatly enhanced by mitogens, growth factors, cMYC and in certain cancer cell lines. The phosphorylated form found in the cytoplasm is inactivate. Phosphorylation on Thr-198 is required for interaction with 14-3-3 proteins. Phosphorylation on Thr-187 by CDK2 leads to protein ubiquitination and proteasomal degradation. Tyrosine phosphorylation promotes this process. Phosphorylation by PKB/AKT1 can be suppressed by LY294002, an inhibitor of the catalytic subunit of PI3K. Phosphorylation on Tyr-88 and Tyr-89 has no effect on binding CDK2, but is required for binding CDK4. Dephosphorylated on tyrosine residues by G-CSF. Ubiquitinated; in the cytoplasm by the KPC complex (composed of RNF123/KPC1 and UBAC1/KPC2) and, in the nucleus, by SCF(SKP2). The latter requires prior phosphorylation on Thr-187. Ubiquitinated; by a TRIM21-containing SCF(SKP2)-like complex; leads to its degradation. Subject to degradation in the lysosome. Interaction with SNX6 promotes lysosomal degradation.

Cellular localization
Nucleus. Cytoplasm. Endosome. Nuclear and cytoplasmic in quiescent cells. AKT-or RSK-mediated phosphorylation on Thr-198, binds 14-3-3, translocates to the cytoplasm and promotes cell cycle progression. Mitogen-activated UHMK1 phosphorylation on Ser-10 also results in translocation to the cytoplasm and cell cycle progression. Phosphorylation on Ser-10 facilitates nuclear export. Translocates to the nucleus on phosphorylation of Tyr-88 and Tyr-89. Colocalizes at the endosome with SNX6 and this leads to lysosomal degradation.

Application | Abreviews | Notes
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WB | 1/500 - 1/1000. Detects a band of approximately 28 kDa (predicted molecular weight: 22 kDa). |
ELISA | 1/20000. |
Western blot - Anti-p27 KIP 1 (phospho T198) antibody (ab64949)

All lanes: Anti-p27 KIP 1 (phospho T198) antibody (ab64949) at 1/500 dilution

Lane 1: HeLa cell extract
Lane 2: HepG2 cell extract
Lane 3: COLO205 cell extract
Lane 4: COLO205 cell extract with immunising peptide at 10 µg

Lysates/proteins at 30 µg per lane.

Predicted band size: 22 kDa
Observed band size: 28 kDa

Western blot - Anti-p27 KIP 1 (phospho T198) antibody (ab64949) at 1/1000 dilution + Mouse liver tissue lysate at 40 µg

Secondary
HRP-conjugated goat anti-rabbit IgG polyclonal at 1/10000 dilution

Developed using the ECL technique.
Performed under reducing conditions.

Predicted band size: 22 kDa
Observed band size: 34 kDa

Exposure time: 1 second

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