**Product datasheet**

**Anti-Cdk4 antibody ab226474**

**Overview**

**Product name**
Anti-Cdk4 antibody

**Description**
Rabbit polyclonal to Cdk4

**Host species**
Rabbit

**Tested applications**
Suitable for: WB, IP, IHC-P

**Species reactivity**
Reacts with: Human

**Immunogen**
Synthetic peptide within Human Cdk4 aa 253-303. The exact sequence is proprietary. Database link: P11802

**Positive control**
WB: HeLa, HEK-293T and Jurkat cell lysate. IP: HEK-293T cell lysate. IHC: Human lung cancer and breast carcinoma tissue.

**Properties**

**Form**
Liquid

**Storage instructions**

**Storage buffer**
Preservative: 0.09% Sodium azide
Constituent: Tris citrate/phosphate

pH 7 to 8

**Purity**
Immunogen affinity purified

**Clonality**
Polyclonal

**Isotype**
IgG

**Applications**

Our Abpromise guarantee covers the use of ab226474 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

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**Function**

Ser/Thr-kinase component of cyclin D-CDK4 (DC) complexes that phosphorylate and inhibit members of the retinoblastoma (RB) protein family including RB1 and regulate the cell-cycle during G(1)/S transition. Phosphorylation of RB1 allows dissociation of the transcription factor E2F from the RB/E2F complexes and the subsequent transcription of E2F target genes which are responsible for the progression through the G(1) phase. Hypophosphorylates RB1 in early G(1) phase. Cyclin D-CDK4 complexes are major integrators of various mitogenic and antimitogenic signals. Also phosphorylates SMAD3 in a cell-cycle-dependent manner and represses its transcriptional activity. Component of the ternary complex, cyclin D/CDK4/CDKN1B, required for nuclear translocation and activity of the cyclin D-CDK4 complex.

**Involvement in disease**

Defects in CDK4 are a cause of susceptibility to cutaneous malignant melanoma type 3 (CMM3) [MIM:609048]. Malignant melanoma is a malignant neoplasm of melanocytes, arising de novo or from a pre-existing benign nevus, which occurs most often in the skin but also may involve other sites.

**Sequence similarities**

Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. CDC2/CDKX subfamily.

Contains 1 protein kinase domain.

**Post-translational modifications**

Phosphorylation at Thr-172 is required for enzymatic activity. Phosphorylated, in vitro, at this site by CCNH-CDK7, but, in vivo, appears to be phosphorylated by a proline-directed kinase. In the cyclin D-CDK4-CDKN1B complex, this phosphorylation and consequent CDK4 enzyme activity, is dependent on the tyrosine phosphorylation state of CDKN1B. Thus, in proliferating cells, CDK4 within the complex is phosphorylated on Thr-172 in the T-loop. In resting cells, phosphorylation on Thr-172 is prevented by the non-tyrosine-phosphorylated form of CDKN1B.

**Cellular localization**

Cytoplasm. Nucleus. Membrane. Cytoplasmic when non-complexed. Forms a cyclin D-CDK4 complex in the cytoplasm as cells progress through G(1) phase. The complex accumulates on the nuclear membrane and enters the nucleus on transition from G(1) to S phase. Also present in nucleoli and heterochromatin lumps. Colocalizes with RB1 after release into the nucleus.

**Images**

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<tr>
<td>IP</td>
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<td>Use at 2-10 µg/mg of lysate.</td>
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<tr>
<td>IHC-P</td>
<td>1/500 - 1/2000. Epitope retrieval with citrate buffer pH6.0 is recommended for FFPE tissue sections.</td>
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Western blot - Anti-Cdk4 antibody (ab226474)

**All lanes**: Anti-Cdk4 antibody (ab226474) at 0.1 µg/ml

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

**Lane 2**: HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) cell lysate

**Lane 3**: Jurkat (human T cell leukemia cell line from peripheral blood) cell lysate

Lysates/proteins at 50 µg per lane.

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size**: 34 kDa

**Exposure time**: 30 seconds

Cdk4 was immunoprecipitated from HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate (1.0 mg for IP, 20% of IP loaded) with ab226474 at 6 µg/mg lysate. Western blot was performed from the immunoprecipitate using a different rabbit anti-Cdk4 antibody at 1 µg/ml.

**Lane 1**: ab226474 IP in HEK-293T whole cell lysate.

**Lane 2**: Control IgG IP in HEK-293T whole cell lysate.

Detection: Chemiluminescence with exposure time of 10 seconds.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdk4 antibody (ab226474)

Formalin-fixed, paraffin-embedded human lung cancer tissue stained for Cdk4 using ab226474 at 1:1000 dilution in immunohistochemical analysis.

Formalin-fixed, paraffin-embedded human breast carcinoma tissue stained for Cdk4 using ab226474 at 1:1000 dilution in immunohistochemical analysis.

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