abcam

Product datasheet

Anti-CHFR antibody ab4184

2 References 3 Images

Overview

Product name	Anti-CHFR antibody
Description	Rabbit polyclonal to CHFR
Host species	Rabbit
Specificity	The immunogen for this antibody is identical in all 3 isoforms of CHFR listed in SwissProt ID Q96EP1.
Tested applications	**Suitable for:** WB, IHC-P, ICC/IF
Species reactivity	**Reacts with:** Human **Predicted to work with:** Mouse
Immunogen	Synthetic peptide conjugated to KLH derived from within residues 450 - 550 of Human CHFR.Read Abcam's proprietary immunogen policy(Peptide available as ab24681.)
Positive control	This antibody gave a positive signal in Hela, Jurkat and A431 whole cell lysates

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Add glycerol to a final volume of 50% for extra stability and aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.02% Sodium Azide Constituents: PBS, pH 7.4
Purity	Immunogen affinity purified
Clonality	Polyclonal
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab4184** in the following tested applications.

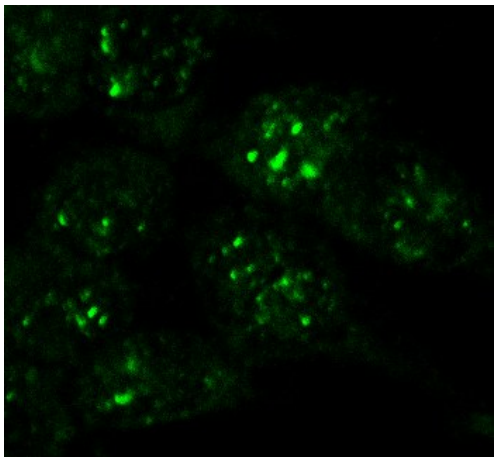
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use a concentration of 1 µg/ml. Detects a band of approximately 76 kDa (predicted molecular weight: 73 kDa).
IHC-P		Use a concentration of 5 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration. We believe this antibody is probably specific for CHFR because in IF with this antibody we see nuclear dots in SAOS2 cells (which have wt CHFR) but not in U2OS cells (that have a mutation in CHFR and hence low levels of it).

Target

Function	E3 ubiquitin-protein ligase that functions in the antephase checkpoint by actively delaying passage into mitosis in response to microtubule poisons. Acts in early prophase before chromosome condensation, when the centrosome move apart from each other along the periphery of the nucleus. Probably involved in signaling the presence of mitotic stress caused by microtubule poisons by mediating the 'Lys-48'-linked ubiquitination of target proteins, leading to their degradation by the proteasome. Promotes the ubiquitination and subsequent degradation of AURKA and PLK1. Probably acts as a tumor suppressor, possibly by mediating the polyubiquitination of HDAC1, leading to its degradation. May also promote the formation of 'Lys-63'-linked polyubiquitin chains and functions with the specific ubiquitin-conjugating UBC13-MMS2 (UBE2N-UBE2V2) heterodimer. Substrates that are polyubiquitinated at 'Lys-63' are usually not targeted for degradation, but are rather involved in signaling cellular stress.
Tissue specificity	Ubiquitous.
Pathway	Protein modification; protein ubiquitination.
Sequence similarities	Belongs to the CHFR family. Contains 1 FHA domain. Contains 1 PBZ-type zinc finger. Contains 1 RING-type zinc finger.
Developmental stage	Weakly expressed in G1 phase, and highly expressed during S phase.
Domain	The PBZ-type zinc finger (also named CYR) mediates non-covalent poly(ADP-ribose)-binding. Poly(ADP-ribose)-binding is dependent on the presence of zinc and is required for its function in antephase checkpoint. The FHA domain plays a key role in the anti-proliferative properties of the protein and is involved in initiating a cell cycle arrest at G2/M. The FHA domain may be required to interact with phosphorylated proteins.
Post-translational modifications	Poly-ADP-ribosylated. In addition to binding non covalently poly(ADP-ribose) via its PBZ-type zinc finger, the protein is also covalently poly-ADP-ribosylated by PARP1. Autoubiquitinated; may regulate its cellular level. Phosphorylated upon DNA damage, probably by ATM or ATR (By similarity). Phosphorylated by PKB. Phosphorylation may affect its E3 ligase activity.
Cellular localization	Nucleus > PML body.

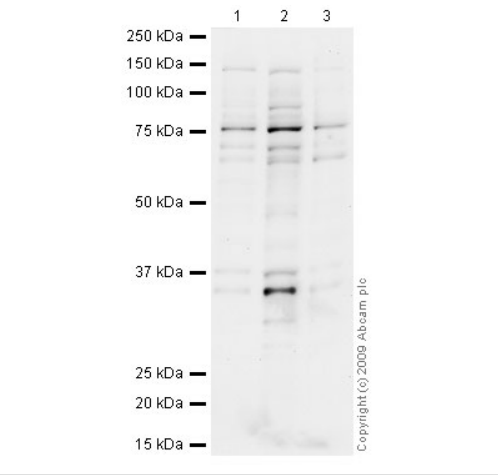
Images



Immunocytochemistry/ Immunofluorescence - Anti-CHFR antibody (ab4184)

Immunofluoresence using ab4184 on SAOS2 cells with a FITC conjugated secondary antibody.

The foci seen are confined to the nuclei of the cells present in the picture.



Western blot - Anti-CHFR antibody (ab4184)

All lanes : Anti-CHFR antibody (ab4184) at 1 µg/ml

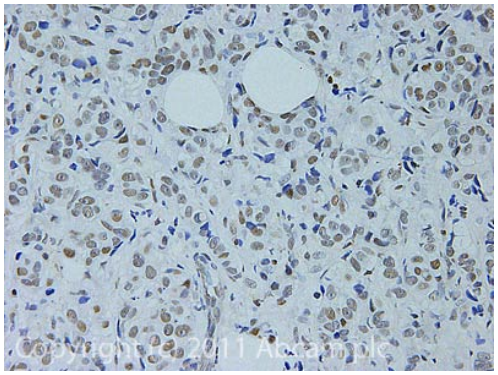
Lane 1 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate
Lane 2 : Jurkat (Human T cell lymphoblast-like cell line) Whole Cell Lysate
Lane 3 : A431 (Human epithelial carcinoma cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Performed under reducing conditions.

Predicted band size: 73 kDa
Observed band size: 76 kDa
Additional bands at: 34 kDa. We are unsure as to the identity of these extra bands.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CHFR antibody (ab4184)

IHC image of ab4184 staining in Human Breast adenocarcinoma 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 ab4184, 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.

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.

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