abcam

Product datasheet

Anti-FANCD2 antibody [EPR2279(3)] ab178705

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-FANCD2 antibody [EPR2279(3)]
Description	Rabbit monoclonal [EPR2279(3)] to FANCD2
Host species	Rabbit
Specificity	This antibody only detects FANCD2 when dephosphorylated at Serine 1418.
Tested applications	**Suitable for:** Flow Cyt (Intra), WB **Unsuitable for:** ICC/IF,IHC-P or IP
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HAP1 and HeLa whole cell lysate (**ab150035**). Flow Cyt (intra): 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**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture supernatant

Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR2279(3)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab178705 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
Flow Cyt (Intra)		1/10 - 1/100. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
WB	★★★★★ (1)	1/1000 - 1/10000. Predicted molecular weight: 166 kDa.

Application notes Is unsuitable for ICC/IF,IHC-P or IP.

Target

Function Required for maintenance of chromosomal stability. Promotes accurate and efficient pairing of homologs during meiosis. Involved in the repair of DNA double-strand breaks, both by homologous recombination and single-strand annealing. May participate in S phase and G2 phase checkpoint activation upon DNA damage. Promotes BRCA2/FANCD1 loading onto damaged chromatin. May also be involved in B-cell immunoglobulin isotype switching.

Tissue specificity Highly expressed in germinal center cells of the spleen, tonsil, and reactive lymph nodes, and in the proliferating basal layer of squamous epithelium of tonsil, esophagus, oropharynx, larynx and cervix. Expressed in cytotrophoblastic cells of the placenta and exocrine cells of the pancreas (at protein level). Highly expressed in testis, where expression is restricted to maturing spermatocytes.

Involvement in disease Defects in FANCD2 are a cause of Fanconi anemia complementation group D type 2 (FANCD2) [MIM:227646]. It is a disorder affecting all bone marrow elements and resulting in anemia, leukopenia and thrombopenia. It is associated with cardiac, renal and limb malformations, dermal pigmentary changes, and a predisposition to the development of malignancies. At the cellular level it is associated with hypersensitivity to DNA-damaging agents, chromosomal instability (increased chromosome breakage) and defective DNA repair.

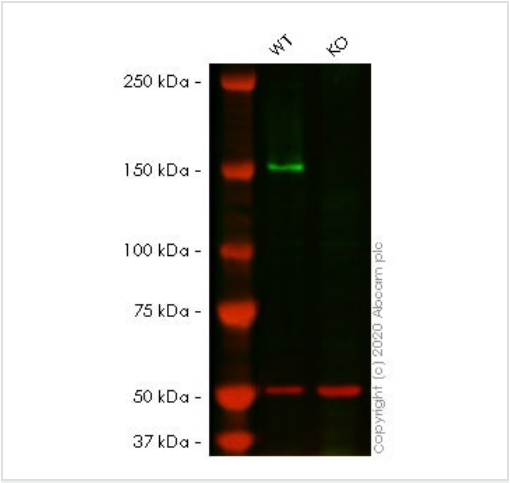
Developmental stage Highly expressed in fetal oocytes, and in hematopoietic cells of the fetal liver and bone marrow (at protein level).

Domain The C-terminal 24 residues of isoform 2 are required for its function.

Post-translational modifications Monoubiquitinated on Lys-561 during S phase and upon genotoxic stress (isoform 1 and isoform 2). Deubiquitinated by USP1 as cells enter G2/M, or once DNA repair is completed. Monoubiquitination requires the FANCA-FANCB-FANCC-FANCE-FANCF-FANCG-FANCM complex, RPA1 and ATR, and is mediated by FANCL/PHF9. Ubiquitination is required for binding to chromatin, interaction with BRCA1, BRCA2 and MTMR15/FAN1, DNA repair, and normal cell cycle progression, but not for phosphorylation on Ser-222 or interaction with MEN1.

	Phosphorylated in response to various genotoxic stresses by ATM and/or ATR. Upon ionizing radiation, phosphorylated by ATM on Ser-222 and Ser-1404. Phosphorylation on Ser-222 is required for S-phase checkpoint activation, but not for ubiquitination, foci formation, or DNA repair. In contrast, phosphorylation by ATR on other sites may be required for ubiquitination and foci formation.
Cellular localization	Nucleus. Concentrates in nuclear foci during S phase and upon genotoxic stress.

Images



Western blot - Anti-FANCD2 antibody [EPR2279(3)] (ab178705)

All lanes : Anti-FANCD2 antibody [EPR2279(3)] (ab178705) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : FANCD2 knockout HeLa cell lysate

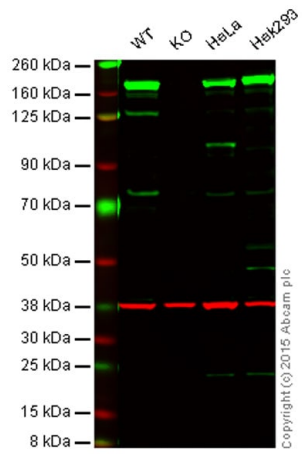
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 166 kDa
Observed band size: 166 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab178705 observed at 166 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) observed at 50 kDa.

ab178705 was shown to react with FANCD2 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab261743** (knockout cell lysate **ab257173**) was used. Wild-type HeLa and FANCD2 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab178705 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-FANCD2 antibody [EPR2279(3)] (ab178705)

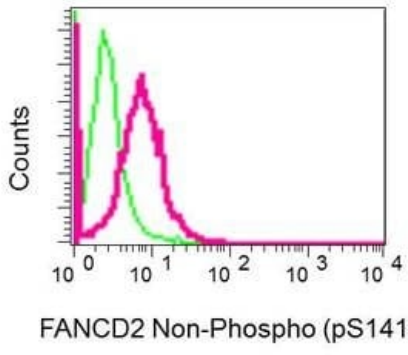
All lanes : Anti-FANCD2 antibody [EPR2279(3)] (ab178705) at 1/1000 dilution

Lane 1 : Wild-type HAP1 cell lysate
Lane 2 : FANCD2 knockout HAP1 cell lysate
Lane 3 : HeLa cell lysate
Lane 4 : HEK293 cell lysate

Lysates/proteins at 20 µg per lane.

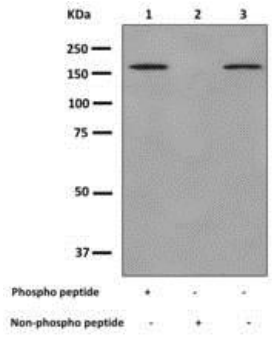
Predicted band size: 166 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab178705 observed at 166 kDa. Red - loading control, **ab8245**, observed at 37 kDa.
ab178705 was shown to specifically react with FANCD2 when FANCD2 knockout samples were used. Wild-type and FANCD2 knockout samples were subjected to SDS-PAGE. ab178705 and **ab8245** (loading control to GAPDH) were diluted 1/1000 and 1/2000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed **ab216776** secondary antibodies at 1/10000 dilution for 1 h at room temperature before imaging.



Flow Cytometry (Intracellular) - Anti-FANCD2 antibody [EPR2279(3)] (ab178705)

Intracellular flow cytometric analysis of permeabilized HeLa cells labeling FANCD2 with ab178705 at 1/10 dilution (red) compared toa rabbit IgG negative control (green).



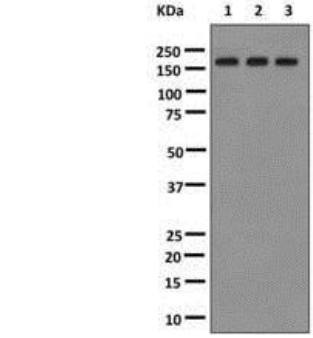
Western blot - Anti-FANCD2 antibody [EPR2279(3)] (ab178705)

All lanes : Anti-FANCD2 antibody [EPR2279(3)] (ab178705) at 1/1000 dilution

Lane 1 : HeLa cell lysate with FANCD2 (pS1418) peptide
Lane 2 : HeLa cell lysate with Non-phospho peptide
Lane 3 : HeLa cell lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 166 kDa



Western blot - Anti-FANCD2 antibody [EPR2279(3)] (ab178705)

All lanes : Anti-FANCD2 antibody [EPR2279(3)] (ab178705) at 1/1000 dilution

Lane 1 : HeLa cell lysate untreated
Lane 2 : HeLa cell lysate treated with Alkaline Phosphatase
Lane 3 : HeLa cell lysate treated with Lambda Phosphatase

Lysates/proteins at 10 µg per lane.

Predicted band size: 166 kDa

Anti-FANCD2 antibody [EPR2279(3)] (ab178705)

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