abcam

Product datasheet

Alexa Fluor® 647 Anti-Ki67 antibody [B56] ab283699

Recombinant

4 Images

Overview

Product name Alexa Fluor® 647 Anti-Ki67 antibody [B56]

Description Alexa Fluor® 647 Mouse monoclonal [B56] to Ki67

Host species Mouse

Conjugation Alexa Fluor® 647. Ex: 652nm, Em: 668nm

Tested applications Suitable for: ICC/IF, Flow Cyt (Intra)

Species reactivity Reacts with: Human

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control ICC/IF: HeLa (human cervix adenocarcinoma epithelial cells) Flow Cyt: HeLa (Human cervix

adenocarcinoma epithelial cells).

General notes This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact orders@abcam.com.

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.

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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.

Avoid freeze / thaw cycle. Store In the Dark.

Storage buffer pH: 7.20

Preservative: 0.02% Sodium azide

Constituents: 30% Glycerol (glycerin, glycerine), 1% BSA, 68% PBS

Purity Protein A purified

Monoclonal Clonality

Clone number **B**56 Isotype lgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab283699 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
ICC/IF		1/500.
Flow Cyt (Intra)		1/5000.

Target

Function

Required to maintain individual mitotic chromosomes dispersed in the cytoplasm following nuclear envelope disassembly (PubMed:27362226). Associates with the surface of the mitotic chromosome, the perichromosomal layer, and covers a substantial fraction of the chromosome surface (PubMed:27362226). Prevents chromosomes from collapsing into a single chromatin mass by forming a steric and electrostatic charge barrier: the protein has a high net electrical charge and acts as a surfactant, dispersing chromosomes and enabling independent chromosome motility (PubMed:27362226). Binds DNA, with a preference for supercoiled DNA and AT-rich DNA (PubMed:10878551). Does not contribute to the internal structure of mitotic chromosomes (By similarity). May play a role in chromatin organization (PubMed:24867636). It is however unclear whether it plays a direct role in chromatin organization or whether it is an indirect consequence of its function in maintaining mitotic chromosomes dispersed.

Sequence similarities

Contains 1 FHA domain. Contains 16 K167R repeats. Contains 1 PP1-binding domain.

Developmental stage

Expression occurs preferentially during late G1, S, G2 and M phases of the cell cycle, while in cells in G0 phase the antigen cannot be detected (at protein level) (PubMed:6206131). Present at highest level in G2 phase and during mitosis (at protein level). In interphase, forms fiber-like structures in fibrillarin-deficient regions surrounding nucleoli (PubMed:2674163,

PubMed:8799815).

Post-translational

Phosphorylated. Hyperphosphorylated in mitosis (PubMed:10502411, PubMed:10653604).

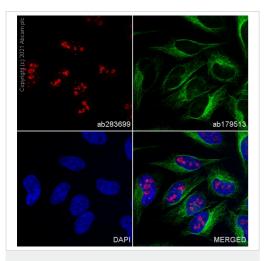
modifications

Cellular localization

Hyperphosphorylated form does not bind DNA.

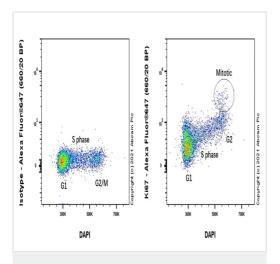
Chromosome. Nucleus. Nucleus, nucleolus. Associates with the surface of the mitotic chromosome, the perichromosomal layer, and covers a substantial fraction of the mitotic chromosome surface (PubMed:27362226). Associates with satellite DNA in G1 phase (PubMed:9510506). Binds tightly to chromatin in interphase, chromatin-binding decreases in mitosis when it associates with the surface of the condensed chromosomes (PubMed:15896774, PubMed:22002106). Predominantly localized in the G1 phase in the perinucleolar region, in the later phases it is also detected throughout the nuclear interior, being predominantly localized in the nuclear matrix (PubMed:22002106).

Images



Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 647 Anti-Ki67 antibody [B56] (ab283699)

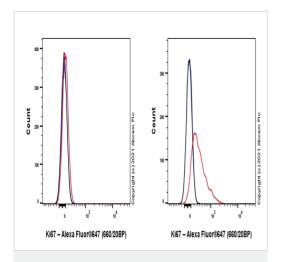
Immunofluorescent analysis of 4% Paraformaldehyde HeLa (human cervix adenocarcinoma epithelial cells) labelling Ki67 with ab283699 at 1/500 dilution, (Red). Confocal image showing nucleolar staining on HeLa cells. ab179513 Anti-beta Tubulin rabbit monoclonal antibody - Microtubule Marker was used to counterstain tubulin at 1/200 dilution. ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) preadsorbed was used as the counterstain secondary antibody at 1/1000 (Green). The nuclear counterstain was DAPI (Blue).



Flow Cytometry (Intracellular) - Alexa Fluor® 647 Anti-Ki67 antibody [B56] (ab283699)

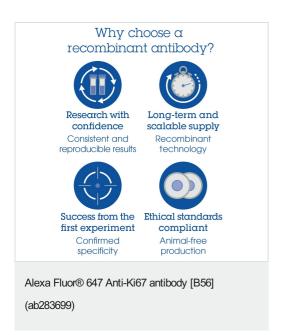
Intracellular Flow cytometric scatterplot showing 4% paraformaldehyde fixed HeLa (Human cervix adenocarcinoma epithelial cells) permeabilized with 90% methanol labelling Ki67 with ab283699 at 1/5000 dilution (Red). Mouse monoclonal IgG (Left)

Cells were co-stained with DAPI to differentiate cell cycle phase.



Flow Cytometry (Intracellular) - Alexa Fluor® 647 Anti-Ki67 antibody [B56] (ab283699) Intracellular Flow cytometric overlay histogram showing 4% paraformaldehyde fixed HeLa (Human cervix adenocarcinoma epithelial cells (Right)) permeabilized with 90% methanol labelling Ki67 with ab283699 at 1/5000 dilution (Red). MKl67 KO HeLa (Left). Isotype control was Mouse monoclonal IgG (Black). Unlabelled control: Cells without incubation with primary antibody and secondary antibody (Blue).

Positive staining on HeLa cells (<u>ab255448</u>), while no staining on Ki67 knockout HeLa cells (<u>ab255407</u>).



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