

## Technical Data Sheet

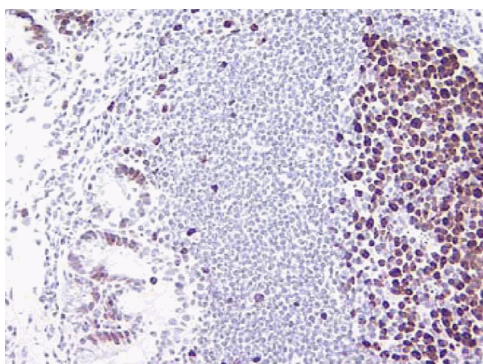
## Purified Mouse Anti-Human Ki-67

## Product Information

<b>Material Number:</b>	<b>550609</b>
<b>Alternate Name:</b>	MKI67; Antigen identified by monoclonal antibody Ki-67; KIA
<b>Size:</b>	1.0 ml
<b>Concentration:</b>	250 µg/ml
<b>Clone:</b>	B56
<b>Immunogen:</b>	Human Ki-67
<b>Isotype:</b>	Mouse IgG1, κ
<b>Reactivity:</b>	QC Testing: Human Tested in Development: Mouse, Pig, Rat
<b>Storage Buffer:</b>	Aqueous buffered solution containing BSA, goat serum, and ≤0.09% sodium azide.

## Description

Recognizes Ki-67, a nuclear cell proliferation-associated antigen expressed in all active stages of the cell cycle. Ki-67 is revealed as a double band (345 and 395 kD) in western blot analysis of proliferating cells. B56 was developed using an immunogen composed of the immunodominant epitope of the Ki-67 protein. Antibodies B56 and MIB 1 react with this immunogen. Flow cytometric analysis reveals that the binding of B56-PE can be blocked by MIB 1 purified antibody. Immunohistochemistry analysis demonstrates a staining pattern similar to MIB 1 on both frozen and paraffin-embedded tissue sections.



**Immunohistochemistry of Ki-67 positive cells.**  
Formalin-fixed paraffin embedded sections of normal human tonsil were reacted with the anti-Ki-67 antibody. Proliferating cells expressing Ki-67 can be identified by the intense brown labeling of their cell nuclei. Magnification 20X.

## Preparation and Storage

Store undiluted at 4°C.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

## Application Notes

## Application

Flow cytometry	Routinely Tested
Immunohistochemistry-frozen	Tested During Development
Immunohistochemistry-formalin (antigen retrieval required)	Tested During Development

## Recommended Assay Procedure:

**Immunohistochemistry:** The B56 clone reactive against human Ki-67 is recommended to test for immunohistochemical staining of formalin-fixed paraffin and acetone-fixed frozen sections. For paraffin sections microwave pretreatment with BD Retrieval A (pH 6.5) (Cat. No. 550524) is required. Tissues tested were human spleen and tonsil. The antibody stains cells in all different stages of proliferation. The isotype control recommended for use with this antibody is purified mouse IgG1 (Cat. No. 550878). For optimal indirect immunohistochemical staining, B56 antibody should be titrated (1:10 to 1:50 dilution) and visualized via a three-step staining procedure in combination with polyclonal, biotin conjugated anti-mouse Igs (multiple adsorbed) (Cat. No. 550337) as the secondary antibody and Streptavidin-HRP (Cat. No. 550946) together with the DAB detection system (Cat. No. 550880).

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## Suggested Companion Products

Catalog Number	Name	Size	Clone
550337	Biotin Goat Anti-Mouse Ig (Multiple Adsorption)	1.0 ml	Polyclonal
550946	Streptavidin HRP	50 ml	(none)
550880	DAB Substrate Kit	500 tests	(none)
550524	Retrievagen A (pH 6.0)	1000 ml	(none)
550878	Purified Mouse IgG1 $\kappa$ Isotype Control	1.0 ml	MOPC-31C
559148	Antibody Diluent for IHC	125 ml	(none)

## Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. This antibody has been developed for the immunohistochemistry application. However, a routine immunohistochemistry test is not performed on every lot. Researchers are encouraged to titrate the reagent for optimal performance.
5. An isotype control should be used at the same concentration as the antibody of interest.
6. Please refer to [www.bdbiosciences.com/pharmlingen/protocols](http://www.bdbiosciences.com/pharmlingen/protocols) for technical protocols.

## References

Kubbutat MH, Key G, Duchrow M, Schluter C, Flad HD, Gerdes J. Epitope analysis of antibodies recognising the cell proliferation associated nuclear antigen previously defined by the antibody Ki-67 (Ki-67 protein). *J Clin Pathol.* 1994; 47(6):524-528. (Biology)

Shi SR, Key ME, Kalra KL. Antigen retrieval in formalin-fixed, paraffin-embedded tissues: an enhancement method for immunohistochemical staining based on microwave oven heating of tissue sections. *J Histochem Cytochem.* 1991; 39(6):741-748. (Biology)

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