Technical Data Sheet

Purified Mouse Anti-Ki-67

Product Information

Material Number: 550609
Alternate Name: MKI67; Antigen identified by monoclonal antibody Ki-67; KIA
Size: 1 mL
Concentration: 250 µg/ml
Clone: B56
Immunogen: Human Ki-67
Isotype: Mouse IgG1, κ
QC Testing: Human
Tested in Development: Mouse
Reported Reactivity: Rat, Rhesus

Storage Buffer:
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.

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

<table>
<thead>
<tr>
<th>Application</th>
<th>Routinely Tested</th>
<th>Tested During Development</th>
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</thead>
<tbody>
<tr>
<td>Flow cytometry</td>
<td></td>
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<tr>
<td>Immunohistochemistry-frozen</td>
<td>Tested During Development</td>
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<tr>
<td>Immunohistochemistry-formalin (antigen retrieval required)</td>
<td>Tested During Development</td>
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Recommended Assay Procedure:

**Immunohistochemistry:** The B56 clone reactive against 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 Retrievagen 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 lgs (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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**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.
## Suggested Companion Products

<table>
<thead>
<tr>
<th>Catalog Number</th>
<th>Name</th>
<th>Size</th>
<th>Clone</th>
</tr>
</thead>
<tbody>
<tr>
<td>550946</td>
<td>Streptavidin HRP</td>
<td>50 mL</td>
<td>(none)</td>
</tr>
<tr>
<td>550337</td>
<td>Biotin Goat Anti-Mouse Ig (Multiple Adsorption)</td>
<td>0.25 mg</td>
<td>Polyclonal</td>
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<tr>
<td>550880</td>
<td>DAB Substrate Kit</td>
<td>500 Tests</td>
<td>(none)</td>
</tr>
<tr>
<td>550524</td>
<td>Retrievagen A (pH 6.0)</td>
<td>1000 mL</td>
<td>(none)</td>
</tr>
<tr>
<td>550878</td>
<td>Purified Mouse IgG1 κ Isotype Control</td>
<td>1 mL</td>
<td>MOPC-31C</td>
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<tr>
<td>559148</td>
<td>Antibody Diluent for IHC</td>
<td>125 mL</td>
<td>(none)</td>
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</table>

### Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. 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.
4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
5. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
6. 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.
7. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.

### References


