### **Product Data Sheet**

#### **Purified anti-mouse Ki-67**

**Catalog #** / 3862005 / 25 μg

**Size:**  $3862010 / 100 \mu g$ 

**Clone:** 16A8

**Isotype:** Rat IgG2a, κ

Immunogen: E. coli expressed partial mouse Ki-67

recombinant protein, 1816-2163 aa.

Reactivity: Human, Mouse

**Preparation:** The antibody was purified by affinity

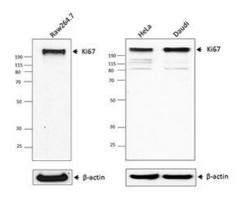
chromatography.

**Formulation:** This antibody is provided in

phosphate-buffered solution, pH 7.2,

containing 0.05% sodium azide.

**Concentration:** 0.5



Total cell lysates (15 microg protein) from Raw264.7, HeLa, and Daudi were resolved by 4-12% Bistris gel electrophoresis, transferred to nitrocellulose, and probed with mouse Ki-67 Antibody

probed with mouse Ki-67 Antibody (clone 16A8) (upper). Proteins were visualized using a goat an

### **Applications:**

**Applications:** Other

Recommended

Usage:

Each lot of this antibody is quality control tested by Western blotting. Western blotting, suggested working dilution(s): Use 5 microL per 5 ml antibody dilution buffer for each mini-gel (0.5 microg/ml). Additionally, each lot of this antibody is quality control tested by our Ki-67 staining protocol below. It is recommended that the reagent be titrated for optimal performance for each application.

Application

Notes:

Additional reported applications (for the relevant formats) include:

immunofluorescence staining.

#### **Ki-67 Staining Protocol:**

- 1. Prepare 70% ethanol and chill at -20°C.
- 2. Prepare target cells of interest and wash 2X with PBS by centrifuge at 350xg for 5 minutes.
- 3. Discard supernatant and loosen the cell pellet by vortexing.
- 4. Add 3 ml cold 70% ethanol drop by drop to the cell pellet while vortexing.
- 5. Continue vortexing for 30 seconds and then incubate at -20  $^{\circ}\text{C}$  for 1 hour.
- 6. Wash 3X with BioLegend Cell Staining Buffer and then resuspend the cells at the concentration of  $0.5-10 \times 10^6$ /ml.
- 7. Mix 100 microL cell suspension with proper fluorochrome-conjugated Ki-67 antibody and incubate at room temperature in the dark for 30 minutes.
- 8. Wash 2X with BioLegend Cell Staining and then resuspend in 0.5 ml cell staining buffer for flow cytometric analysis.

## Application References:

- 1. Medina-Reyes El, et al. 2015. Environ Res. 136:424. PubMed
- 2. Guillaumond F, et al. 2015. PNAS. 112:2473. PubMed
- 3. Sharma SK, et al. 2015. J Immunol. 194:5529. PubMed
- 4. Rodero MP, et al. 2014. J. Invest. Dermatol. 7:1991-7. PubMed

#### **Description:**

The nuclear protein Ki-67 was first identified by the monoclonal antibody Ki-67, which was generated by immunizing mice with nuclei of the L428 Hodgkin lymphoma cell line. Ki-67 protein plays an essential role in ribosomal RNA transcription and cell proliferation. Expression of Ki-67 occurs during G1, S, G2, and M phase, while in G0 phase the Ki-67 protein is not detectable. Ki-67 is strongly expressed in proliferating cells and has been reported as a prognostic marker in various tumors.

# Antigen References:

- 1. Starborg M, et al. 1996. J. Cell. Sci. 109:143.
- 2. Byeon IJ, et al. 2005. Nat. Struct. Mol. Biol. 12:987.
- 3. Yerushalmi R, et al. 2010. Lancet. Oncol. 11:174.
- 4. Beltrami AP, e