

# Anti-Ki67 Antibody [ST50-01]

ET1609-34



<b>Product Type:</b>	Recombinant Rabbit monoclonal IgG, primary antibodies
<b>Species reactivity:</b>	Human
<b>Applications:</b>	WB, IF-Cell, IHC-P, FC
<b>Molecular Wt:</b>	Predicted band size: 359 kDa
<b>Clone number:</b>	ST50-01

**Description:** Ki-67 is a nuclear protein that is expressed in proliferating cells and may be required for maintaining cell proliferation. Ki-67 has been used as a marker for cell proliferation of solid tumors and some hematological malignancies. A correlation has been demonstrated between Ki-67 index and the histopathological grade of neoplasms. Assessment of Ki-67 expression in renal and ureter tumors shows a correlation between tumor proliferation and disease progression, thus making it possible to differentiate high-risk patients. Ki-67 expression may also prove to be important for distinguishing between malignant and benign peripheral nerve sheath tumors.

**Immunogen:** Synthetic peptide within human Ki67 aa 1040-1080.

**Positive control:** HepG2 cell lysates, HepG2, HeLa, human tonsil tissue, human stomach carcinoma tissue, human colon carcinoma tissue.

**Subcellular location:** Nucleus, Chromosome.

**Database links:** SwissProt P46013 Human

**Recommended Dilutions:**

<b>WB</b>	1:1,000-1:2,000
<b>IF-Cell</b>	1:50-1:1,000
<b>IHC-P</b>	1:100-1:500
<b>FC</b>	1:50-1:100

**Storage Buffer:** 1\*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% SodiumAzide.

**Storage Instruction:** Store at +4°C after thawing. Aliquot store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.

**Purity:** Protein A affinity purified.

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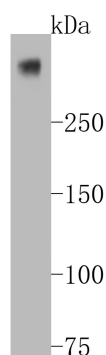
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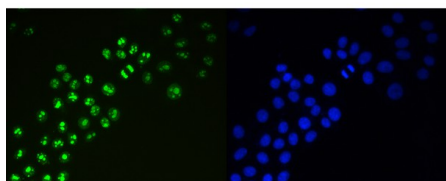
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## Images

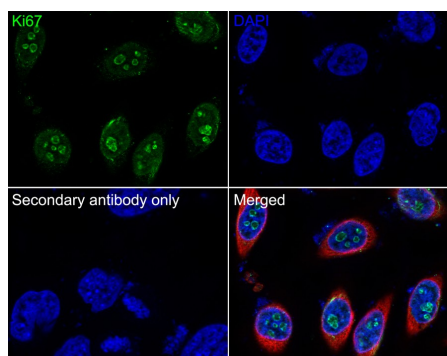


**Fig1:** Western blot analysis of Ki67 on HepG2 cell lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET1609-34, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:5,000 dilution was used for 1 hour at room temperature.



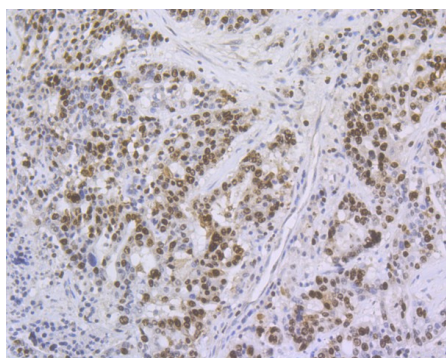
**Fig2:** ICC staining of Ki67 in HepG2 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-34, 1/1,000) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

**Fig3:** Immunocytochemistry analysis of HeLa cells labeling Ki67 with Rabbit anti-Ki67 antibody (ET1609-34) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Ki67 antibody (ET1609-34) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.



**Fig4:** Immunohistochemical analysis of paraffin-embedded human stomach carcinoma tissue using anti-Ki67 antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1609-34, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

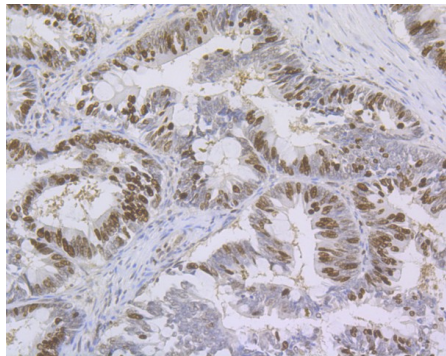
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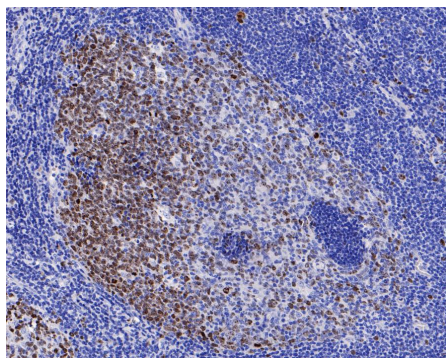
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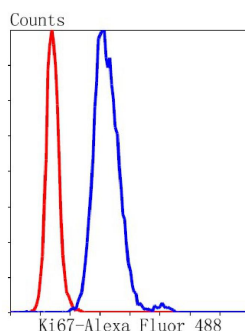


**Fig5:** Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue using anti-Ki67 antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1609-34, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



**Fig6:** Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-Ki67 antibody (ET1609-34) at 1/800 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1609-34) at 1/800 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



**Fig7:** Flow cytometric analysis of Ki67 was done on HeLa cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1609-34, 1/100) (blue). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes. Unlabelled sample was used as a control (cells without incubation with primary antibody; red).

**Note:** All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

### Background References

1. Cuylen S. et al. Ki-67 acts as a biological surfactant to disperse mitotic chromosomes. *Nature* 535:308-312(2016).
2. Booth D.G. et al. Ki-67 is a PP1-interacting protein that organises the mitotic chromosome periphery. *Elife* 3:E01641-E01641(2014).

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