

# iFluor™ 647 Conjugated Anti-Ki67 Antibody [SR00-02] HA720163F



<b>Product Type:</b>	Recombinant Rabbit monoclonal IgG, primary antibodies
<b>Species reactivity:</b>	Human, Mouse
<b>Applications:</b>	IF-Cell, IF-Tissue, FC, IHC-Fr
<b>Molecular Wt:</b>	Predicted band size: 359 kDa
<b>Clone number:</b>	SR00-02

**Description:** The Ki-67 protein is a nuclear protein doublet, 345-395 kDa, playing a pivotal role in maintaining cell proliferation. Ki-67 is present in all non-G0 phases of the cell cycle. Beginning in the mid G1, the level increases through S and G2 to reach a peak in M. In the end of M, it is rapidly catabolized. The Ki-67 labelling index (LI), i.e., the percentage of cells in a tissue staining for Ki-67, indicates the growth fraction. For many tumours, the rate of cell proliferation as assessed by Ki-67 immunoreactivity correlates with tumour grade and clinical course. In Non-Hodgkin lymphoma a labelling index of less than 20% is seen in low grade lymphomas, greater than 20% is associated with high grade lymphomas. Low grade lymphomas with a labelling index in excess of 5% have a worse prognosis than those with an index of less than 5%. In Burkitt and Burkitt-like lymphoma, nearly 100% of the nuclei are stained. This can be used as a diagnostic criterion. In gliomas the indices ranges from 0% to 5% for low grade astrocytomas while anaplastic astrocytomas and glioblastomas most frequently show an index above 10%. In soft tissue sarcomas Ki-67 index is positively correlated with mitotic count, cellularity and histological grade. In some benign tumours, like meningioma, a high LI is associated with a high recurrence rate. In dysplasia in Barrett's oesophagus and in granulosa cell tumours and ovarian serous tumours, Ki-67 LI is associated with progression. In the former, reproducibility of dysplasia grading is improved when Ki67 is included. In breast cancer, the proliferative index measured by Ki67 immunoreactivity has both prognostic and predictive value.

<b>Conjugate:</b>	iFluor™ 647
<b>Immunogen:</b>	Synthetic peptide within human Ki67 aa 1,040-1,080.
<b>Positive control:</b>	A431, Jurkat cells, human colon carcinoma tissue, mouse small intestine tissue, MOLT-4.
<b>Subcellular location:</b>	Nucleus, Chromosome.
<b>Database links:</b>	SwissProt: P46013 Human
<b>Recommended Dilutions:</b>	
IF-Cell	1:250
IF-Tissue	1:100
FC	1:1,000
IHC-Fr	1:500
<b>Storage Buffer:</b>	Preservative: 0.02% Sodium azide Constituents: 30% Glycerol, 1% BSA, 68.98% PBS
<b>Storage Instruction:</b>	Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.
<b>Purity:</b>	Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders: 0086-571-88062880

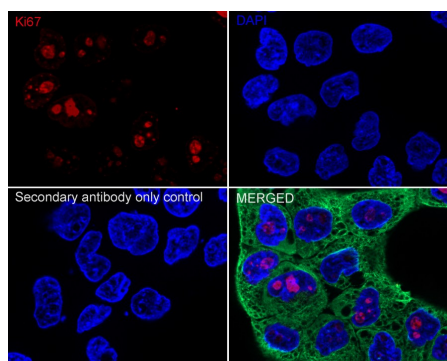
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Applications: WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

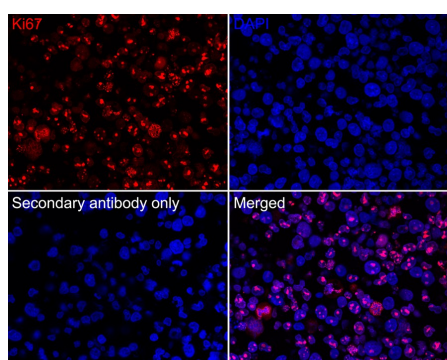
## Images



**Fig1:** Immunocytochemistry analysis of A431 cells labeling Ki67 with Rabbit anti-Ki67 antibody (HA720163F) at 1/250 dilution.

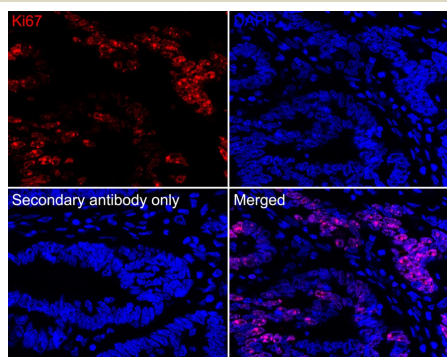
Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Ki67 antibody (HA720163F, red) at 1/250 dilution in 1% BSA in PBST overnight at 4 °C. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, green) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution.



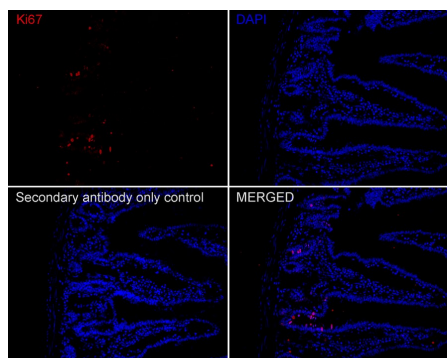
**Fig2:** Immunofluorescence analysis of paraffin-embedded Jurkat cells labeling Ki67 with Rabbit anti-Ki67 antibody (HA720163F) at 1/100 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA720163F, red) at 1/100 dilution overnight at 4 °C, washed with PBS.



**Fig3:** Immunofluorescence analysis of paraffin-embedded human colon carcinoma tissue labeling Ki67 (HA720163F).

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 10% negative goat serum for 1 hour at room temperature, washed with PBS. And then probed with the primary antibody Ki67 (HA720163F, red) at 1/100 dilution overnight at 4 °C, washed with PBS. DAPI was used as nuclear counterstain.



**Fig4:** Immunofluorescence analysis of frozen mouse small intestine tissue with Rabbit anti-Ki67 antibody (HA720163F) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA720163F, red ) at 1/500 dilution overnight at 4 °C, washed with PBS. Nuclei were counterstained with DAPI (blue).

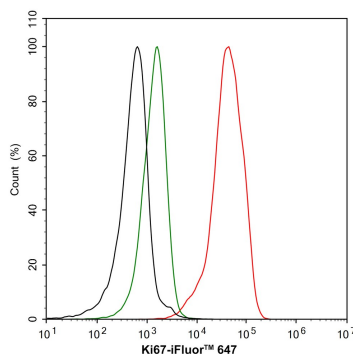
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**Fig5:** Flow cytometric analysis of MOLT-4 cells labeling Ki67.

Cells were washed twice in 1x PBS and fixed in 70% ethanol at 4°C overnight. Then incubated for 1 hour at +4°C with Ki67 (HA720163F, red, 1/1,000) and Rabbit IgG Isotype Control (iFluor™ 647, green, 1/1,000). Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

**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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