

Cor Mouse Monoclonal Antibody

Host

Isotype

lgG1

MCA-6B4

Species Cross-Reactivity

Ordering Information Web www.encorbio.com Email admin@encorbio.com Phone 352-372-7022 Fax 352-372-7066

HGNC Name: MKI67 UniProt: P46013 RRID: AB_2637051

Immunogen: Recombinant construct containing the 2nd, 3rd and 4th Ki67 repeats of the human sequence (amino acids 1,111-1,490) expressed in and purified

Format: Purified antibody at 1mg/mL in 50% PBS, 50% glycerol. 5mM azide

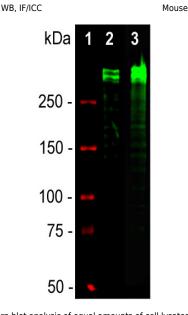
50% glycerol, 5mM azide **Storage:** Stable at 4°C for one year, for longer term

store at -20°C

Recommended dilutions: WB: 1:1,000-5,000. IF 1:2,000-5,000.

References:

- 1. Gerdes J, Schwab U, Lemke H, Stein H. Production of a mouse monoclonal antibody reactive with a human nuclear antigen associated with cell proliferation. Int. J. Cancer 31:13-20 (1983).
- 2. Kill IR, Faragher RGA, Lawrence K. Shall S. The expression of proliferation-dependent antigens during the lifespan of normal and progeroid human fibroblasts in culture. J. Cell Sci. 107:571-9 (1994).
- 3. Yerushalmi R, et al. Ki67 in breast cancer: Prognostic and predictive potential. Lancet Oncol. 11:174-83 (2010).
- 4. Josefsson A, et al. Low endoglin vascular density and Ki67 index in Gleason score 6 tumours may identify prostate cancer patients suitable for surveillance. Scand. J. Urol. Nephrol. 46:247-57 (2012)
- 5. Ishihara M, et al. Retrospective analysis of risk factors for central nervous system metastases in operable breast cancer: effects of biologic subtype and Ki67 overexpression on survival. Oncology, 84:135-140 (2013).
 6. Cheang MC, et al. Ki67 Index, HER2 Status,
- 6. Cheang MC, et al. Ki67 Index, HER2 Status, and Prognosis of Patients With Luminal B Breast Cancer. J. Natl. Cancer Inst. 101:736-50 (2009). 7. Margulis V, et al. Multi-institutional validation of the predictive value of Ki-67 labeling index in patients with urinary bladder cancer. J. Natl. Cancer Inst. 101:114-9 (2009).
- 8. Cuylen S, et al. Ki-67 acts as a biological surfactant to disperse mitotic chromosomes. Nature. 535:308-12 (2016).



Western blot analysis of equal amounts of cell lysates using mouse mAb to Ki67, MCA-6B4, dilution 1:2,000, in green: [1] protein standard (red), [2] rapidly dividing HeLa cell cultures, [3] rapidly dividing HEK293 cell cultures. Strong double bands above 250kDa correspond to the two major Ki67 isoforms of apparent molecular weight of 345kDa and 395kDa. Smaller proteolytic fragments of these isoforms are also invariably detected on the blot.

Molecular Wt.

345kDa, 395kDa

Confcoal immunofluorescence image at high magnification of HeLa cell culture stained with mouse monoclonal antibody to Ki-67, MCA-6B4, in green, and costained with chicken polyclonal antibody to Lamin A/C, CPCA-LaminAC, in red. The antibody against Ki67 stains the nuclei of rapidly dividing cells within their nucleoli, but doesn't stain nearby quiescent cells. The antibody against Lamin A/C stains nuclear lamina. The blue is DAPI staining of nuclear DNA.

Background:

Applications

The Ki67 protein was first discovered when researchers attempted to generate cancer cell specific monoclonal antibodies by injecting mice with nuclear preparations from Hodgkin's lymphoma cells (1). They obtained a monoclonal antibody which recognized two large proteins of apparent molecular weight 345kDa and 395kDa. The clone was named Ki67 after Kiel, Germany where the original work was done and the number of the 96 well plate in which the clone was found. The two proteins were found to be heavily expressed in proliferating cells, but to be absent in quiescent cells, and later work showed that they were the product of a single gene. The presence of the Ki67 protein is frequently used as an indicator of cell proliferation and its level of expression is one of the most reliable biomarkers of proliferative status of cancer cells (2-5). Much research shows a correlation between Ki67 protein level and prognosis in cancer patients, when high Ki67 levels being associated with poorer outcomes (e.g. 6,7). The original Ki67 antibody and several others have become so widely used that a search for "(Ki67 or Ki-67) and antibody" in PubMed in August 2018 produced over 5,600 results. Recent studies show that Ki67 functions as a "biological surfactant", which is essential for the fidelity of separation of condensed chromosomal DNA into the two daughter cells during cell division (8). This presumably explains the highly basic nature of Ki67, allowing a charge-based interaction with nucleic acids, the lack of this protein in non-dividing cells and the relative lack of protein sequence conservation.

The MCA-6B4 antibody was made against a recombinant construct including amino acids 1,111-1,490 of the human sequence P46013, a region corresponding to 2nd, 3rd and 4th Ki67 type repeats. This product is not recommended for use on rodent tissues: The Ki67 protein sequence is rather poorly conserved across species boundaries so antibodies raised against the human form, like this one, are often unreactive with the rodent form.

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Abbreviation Key:

mAb—Monoclonal Antibody pAb—Polyclonal Antibody WB—Western Blot IF—Immunofluorescence ICC—Immunocytochemistry IHC—Immunohistochemistry E—ELISA Hu—Human Mo—Monkey Do—Dog Rt—Rat Ms—Mouse Co—Cow Pi—Pig Ho—Horse Ch—Chicken Dr—D. rerio Dm—D. melanogaster Sm—S. mutans Ce—C. elegans Sc—S. cerevisiae Sa—S. aureus Ec—E. coli.