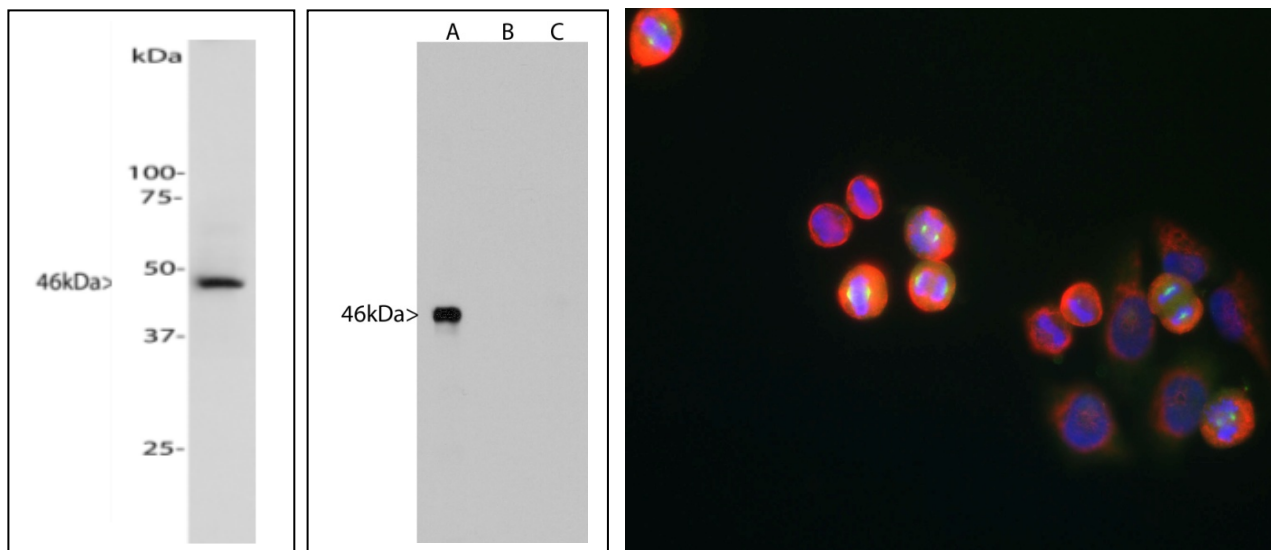


**Catalogue# MCA-1A14: Mouse Monoclonal Antibody to Aurora A Kinase**

**The Immunogen:** Aurora-A is a member of the Aurora/Ipl1-related kinase family, a novel family of serine/threonine kinases crucial for cell cycle control. The first Aurora kinase was discovered in *Drosophila* (1). Because mutations of this kinase cause monopolar spindles, it was given the name "Aurora", reminiscent of the North Pole (1). Mammals contain at least three aurora kinases: Aurora A, Aurora B, and Aurora C. In particular, aurora A is overexpressed in various types of cancer and considered to play critical roles in tumorigenesis (2,3). Overexpression of Aurora A overrides the cell cycle checkpoint, interferes with mitotic exit (4), and induces transformation in mammalian cells (2,3). In contrast, silencing of aurora A interrupts centrosome separation, spindle assembly and arrests G2-M transition in *C. elegans* and human cells (5,6). Moreover, aurora A plays an essential role for neuronal migration by modulation of microtubule organization and is required for normal axon formation (7,8). Aurora A is activated by phosphorylation at Thr-288 and activated aurora A phosphorylates numerous target proteins including p53, TPX2 and BRCA1. Expression of aurora A is cell-cycle regulated, low in G1/S, accumulates during G2/M, and decreases rapidly after. Aurora A localizes next to the centrosome late in the G1/S phase. As the cell cycle progresses, aurora A associates with the mitotic poles and the adjacent spindle microtubules. It remains associated with the spindles through telephase and relocates to the mid-zone of the spindle right before mitotic exit (9). Monoclonal antibody MCA-1A14 was raised against full length recombinant human aurora A expressed in and purified from *E. coli*. The antibody was tested for binding to expressed human aurora A, B, C and shown to react with only Aurora A and no cross-reaction with aurora B and aurora C (see below). The HGNC name for this protein is AURKA .



**Left:** Western analysis of MCA-1A14. Blot of HeLa cells treated with 100ng/ml nocodazole for 18 hours was probed with MCA-1A11. Nocodazole is microtubule inhibitor which induces cells G2/M phase and induces aurora A expression. The MCA-1A11 monoclonal binds strongly and cleanly to a band at about 46 kDa. **Middle:** Blot of expressed human protein of aurora A, B and C was probed with MCA-1A14. MCA-1A14 recognized only Aurora A protein. **Right:** shows HeLa cell cultures stained with MCA-1A14 antibody (green). Aurora A localizes in centrosome and mitotic spindles at late mitosis. Counterstained is our chicken polyclonal antibody against Vimentin: **CPCA-Vim** (red). Blue is a DNA stain.

**Antibody characteristics:** MCA-1A14 is a mouse IgG2b class antibody. MCA-1A14 recognizes aurora A kinase specifically both in western blots and in immunocytochemical experiments. On blots, MCA-1A14 reveals a prominent 46kDa band, on cells in tissue cultures, MCA-1A14 reveals strong staining in centrosome.

**Suggestions for use:** The antibody is provided in the form of purified antibody at a concentration of 1mg/ml in 50% glycerol/PBS with 10mM sodium azide. Store at 4°C or -20°C. Avoid repeat freezing and thawing. The

antibody solution can be used at dilutions of at least 1:500-1:1,000 in immunofluorescence experiments. In western blotting using chemiluminescence it can be used at dilutions of 1:1,000.

**Storage Instructions:** Shipped on ice. Please store at 4°C for regular uses. For long term storage, please leave frozen at -20°C and avoid freeze/thaw cycles.

**Limitations:** This product is for research use only and is not approved for use in humans or in clinical diagnosis.

#### **References:**

1. Glover DM, Leibowitz MH, McLean DA, Parry H. Mutations in aurora prevent centrosome separation leading to the formation of monopolar spindles. *Cell* 81:95-105 (1995).
2. Bischoff JR, Anderson L, Zhu Y, et al. A homologue of Drosophila aurora kinase is oncogenic and amplified in human colorectal cancers. *EMBO J.* 17:3052-65 (1998).
3. Zhou H, Kuang J, Zhong L, et al. Tumour amplified kinase STK15/BTAK induces centrosome amplification, aneuploidy and transformation. *Nat. Genet.* 20:189-93 (1998).
4. Marumoto T, Hirota T, Morisaki T, Kunitoku N, Zhang D, Ichikawa Y, Sasayama T, Kuninaka S, Mimori T, Tamaki N, Kimura M, Okano, Roles of aurora-A kinase in mitotic entry and G2 checkpoint in mammalian cells. *Genes Cells* 7: 1173-1182 (2002).
5. Hannak E, Kirkham M, Hyman AA, Oegema K. Aurora-A kinase is required for centrosome maturation in *Caenorhabditis elegans*. *J. Cell Biol.* 155:1109-16 (2001).
6. Du J, Hannon GJ. Suppression of p160ROCK bypasses cell cycle arrest after Aurora-A/STK15 depletion. *Proc Natl Acad Sci U S A* 101:8975-80 (2004).
7. Mori D, Yamada M, Mimori-Kiyosue Y, Shirai Y, Suzuki A, Ohno S, Saya H, Wynshaw-Boris A, Hiroutsune S. An essential role of the aPKC-Aurora A-NDEL1 pathway in neurite elongation by modulation of microtubule dynamics. *Nat Cell Biol.* 11(9):1057-68 (2009) *Nat. Cell Biol.*11(9):1057-68(2009).
8. Takitoh T, Kumamoto K, Toba S, Wynshaw-Boris A, Hiroutsune S. Activation of Aurora-A is essential for neuronal migration via modulation of microtubule organization. *J. Neurosci.* 32(32):11050-66 (2012).
9. Marumoto T, Honda S, Hara T, Nitta M, Hirota T, Kohmura E, Saya H. Aurora-A kinase maintains the fidelity of early and late mitotic events in HeLa cells. *J. Biol. Chem.* 278(51): 5178-95 (2003).