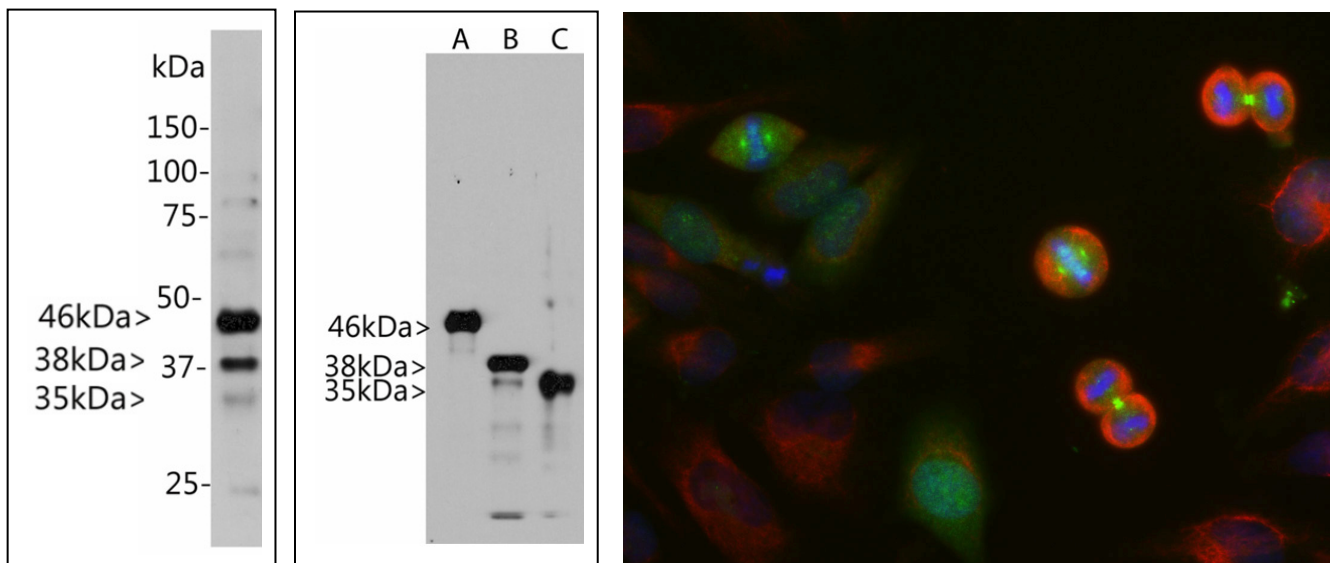


Catalogue# MCA-4A7: Mouse Monoclonal Antibody to Aurora A, B and C Kinases

The Immunogen: Aurora proteins are a family of serine/threonine protein kinases that play a key role in the regulation of cell division. The first Aurora kinase was discovered in *Drosophila* (1). Mutations of this kinase cause monopolar spindles surrounded by kinase, and the appearance of this was reminiscent of the Aurora borealis at the poles of the earth (1). Mammalian genomes encode 3 Aurora kinases named Aurora A, Aurora B, and Aurora C. All 3 contain a regulatory domain at the N terminus which is quite different between the molecules followed by a catalytic serine/threonine kinase domain which is almost identical between them. To download a sequence alignment of the 3 human Aurora proteins go to <http://encorbio.com/Alignments/Aurora%20alignment.pdf>. As a consequence antibodies raised against one Aurora family member frequently cross-react with other family members. There is a short C-terminal peptide which is also variable between the three molecules (2). Aurora A is required for centrosome duplication, entry into mitosis, formation of bipolar spindle and mitotic checkpoint (3). Aurora B is a chromosomal passenger protein and essential for chromosome condensation, kinetochore functions, spindle checkpoint activation and cytokinesis completion (4). Aurora C is heavily expressed in testis and is involved in spermatogenesis, but is also expressed in many cell lines and cancer cells and has been less well studied to date (5). Aurora A is first associated with centrosomes and then with spindle microtubules whereas Aurora B localizes to the spindle midzone and finally accumulates at the midbody. Aurora C is localized to centrosomes and midbodies, like Aurora A and Aurora B (6,7).

Although MCA-4A7 was raised against full length recombinant human aurora C expressed in and purified from *E. coli*, it was shown to react with all three aurora kinase (Blot image). The HGNC names for Aurora A, B and C are AURKA, AURKB and AURKC.



Left: Western blot analysis of MCA-4A7 in HeLa cells. Blot of HeLa cells treated with 100ng/ml nocodazole for 18 hours was probed with MCA-4A7. Nocodazole is a microtubule depolymerizing agent which induces cells to halt at the G2/M phase and also induces Aurora kinase expression. The MCA-4A7 monoclonal binds strongly to bands at about 46 kDa and 38kDa, corresponding to Aurora A and Aurora B. Also, it recognizes a weak band at 35 kDa which is Aurora C. **Middle:** Blot of recombinant full length human Aurora A, B and C was probed with MCA-4A7. This antibody reacts strongly with all three Aurora kinases proteins. **Right:** HeLa cell cultures were stained with MCA-4A7 antibody (green). In this image, strong staining in spindle poles was seen in cells at anaphase. This antibody also stains the midbodies between the two daughter cells. Cells were counterstained with our chicken polyclonal antibody to Vimentin **CPCA-Vim** in red. Blue is a DNA stain.

Antibody characteristics: MCA-4A7 is a mouse IgG1 class antibody with a κ light chain. MCA-4A7 recognizes aurora A, B and C kinases in western blots and in immunocytochemical experiments.

Suggestions for use: The antibody is protein G purified from tissue culture supernatant and is diluted in phosphate buffered saline at 1 mg/mL. The preparation contains 10 mM sodium azide preservative (Link to <http://www.encorbio.com/MSDS/azide.htm> for a material safety data sheet (MSDS)). The antibody solution can be used at dilutions 1:500-1:1,000 for immunofluorescence. For western blots try at 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. Hohegger H, Hegarat N, Pereira-Leal JB. Aurora at the pole and equator: overlapping functions of Aurora kinases in the mitotic spindle. *Open Biol.* Mar 20;3(3):120185 (2013)
3. Barr AR, Gergely F. Aurora-A: the maker and breaker of spindle poles. *J Cell Sci.*120:2987-96 (2007). Review.
4. Andrew PD, Knatko E, Moore WJ, Swedlow JR. Mitotic mechanics: the auroras come into view. *Curr Opin Cell Biol.*15(6):672-83 (2003). Review.
5. Tang CJ, Lin CY, Tang TK. Dynamic localization and functional implications of Aurora-C kinase during male mouse meiosis. *Dev Biol.* Dev Biol. 290(2):398-410 (2006).
6. Dutertre S, Hamard-Péron E, Cremet JY, Thomas Y, Prigent C. The absence of p53 aggravates polyploidy and centrosome number abnormality induced by Aurora-C overexpression. *Cell Cycle.* Dec;4(12):1783-7 (2005).
7. Kimura M, Matsuda Y, Yoshioka T, Okano Y. Cell cycle-dependent expression and centrosome localization of a third human aurora/Ipl1-related protein kinase, AIK3. *J Biol Chem.* 274(11):7334-40 (1999).

©EnCor Biotechnology Inc. December 31, 2015.