



THE POLYCLONAL ANTIBODY SPECIALISTS

ANTIBODIES TO KINETOCHORE/CENTROMERE COMPLEX PROTEINS

In the early 20th century, cytologists and anatomists first used the terms “kinetochore” and “centromere” as a general description of the differentiated regions observed on mitotic chromosomes that associated with spindle microtubules. Today, the term “centromere” is commonly used as a morphological description to describe the constricted region that divides a chromosome into two arms, while the term “kinetochore” has become a term that more specifically describes the proteinaceous structure embedded on the surface of the centromere and the location where spindle microtubules attach. The kinetochore is a button-like trilami-

nar structure that is comprised of an inner plate of compacted chromatin, a dense osmophilic outer plate, and a fibrous corona. During cell division the kinetochore associates with spindle microtubules and functions to partition sister chromatids toward the poles to daughter cells. The kinetochore also performs a critical function in mitotic check-point signaling that ensures chromosomal attachment to spindle microtubules for the prevention of chromosomal loss. Precise chromosome segregation requires the assembly of a single kinetochore on each chromosome. The assembly and layering of the trilaminar kinetochore structure is complex and

relies on specific centromeric DNA sequences and their associated DNA binding proteins. A collection of factors have been found to specifically localize to the outer and inner kinetochore plate as well as to the centromere and contribute to kinetochore function. Although this aggregation of proteins and their relationships in the kinetochore/centromere complex have been extensively studied, the assembly order and dynamics of the complex is not yet completely understood. The availability of antibodies to these components will be valuable to understanding the assembly and function of the kinetochore/centromere complex.

BETHYL LABORATORIES' PORTFOLIO OF ANTIBODIES TO KINETOCHORE/CENTROMERE COMPLEX PROTEINS INCLUDE:

Antibody	Applications						Antibody	Applications						Antibody	Applications								
	WB	ICC	F	IP	ICC-IF	IHC		WB	ICC	F	IP	ICC-IF	IHC		WB	ICC	F	IP	ICC-IF	IHC			
53BP1	✓	✓	✓				CENP-C1 (AbVanced™)										MSH2	✓			✓		✓
Phospho 53BP1 (S25)	✓		✓	✓			CENP-E				✓						NUP96	✓			✓		
APC	✓			✓			CENP-F/Mitosis	✓			✓	✓	✓				NUP107	✓			✓		
APC1	✓			✓			CENP-H (AbVanced™)										NUP160	✓			✓		
APC3/CDC27	✓			✓			CENP-J (AbVanced™)										PLK1	✓			✓	✓	
APC4	✓			✓			CRM1	✓			✓		✓				PP2A	✓			✓		
APC5	✓			✓			ELYS	✓			✓						RanBP2	✓			✓		
APC6/CDC16	✓			✓			HEC1	✓			✓						RCC2	✓			✓		✓
APC8/CDC23	✓			✓			HELLS	✓			✓						ROD/KNTC1	✓			✓		
Astrin/MAP126	✓			✓			HP1-beta	✓									SGO2	✓			✓		
Aurora B	✓			✓	✓	✓	INCENP						✓				Sin3A	✓			✓		✓
Bub1	✓			✓			KIF18A	✓			✓						SMC1	✓			✓	✓	✓
Bub3	✓					✓	KIF22/KID	✓			✓						Phospho SMC1 (S957)	✓			✓		
BubR1	✓			✓			LIS-1	✓			✓						Phospho SMC1 (S966)	✓			✓		✓
CBX3	✓			✓		✓	MAD1	✓			✓		✓				SPINDLY/CCDC99KIF2A	✓			✓		
CBX5	✓			✓		✓	MAD2	✓			✓						TTK	✓			✓		
CDC20	✓			✓			MCAK	✓			✓						WSTF	✓			✓		
CENP-A (AbVanced™)							MIS12	✓			✓						ZWINT-1	✓			✓	✓	✓
CENP-B					✓		MOF/MYST1	✓			✓												

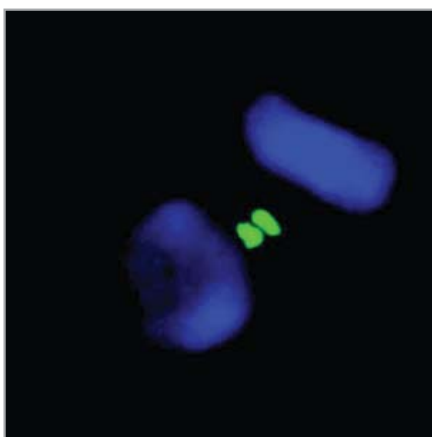
WB Western Blot, ICC Immunocytochemistry, F Flow Cytometry, IP Immunoprecipitation, ICC-IF Immunocytochemistry-Immunofluorescence, IHC Immunohistochemistry.

AbVanced™ Panel of antibodies and peptides to target tested by ELISA only.

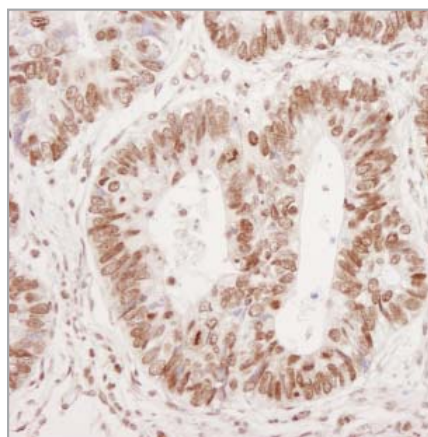
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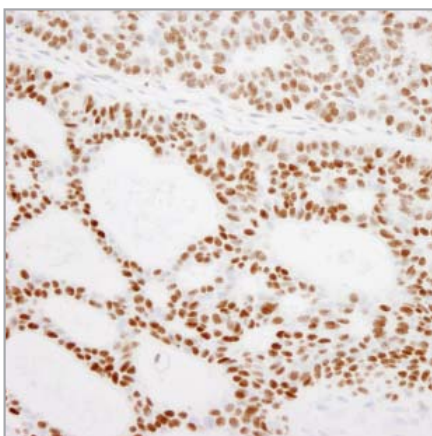
IMMUNOHISTOCHEMISTRY & WESTERN BLOT



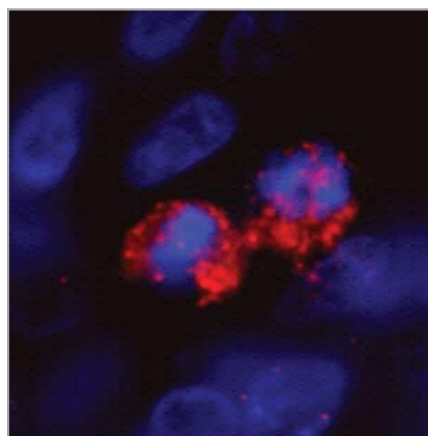
Detection of Human Aurora B by ICC. **Sample:** NBF-fixed HeLa Cells. **Antibody:** Affinity purified rabbit anti-Aurora B (Cat. No. IHC-00003) used at a dilution of 1:100. **Detection:** Goat anti-rabbit IgG (h&l) FITC (Cat. No. A120-101F) used at a dilution of 1:100.



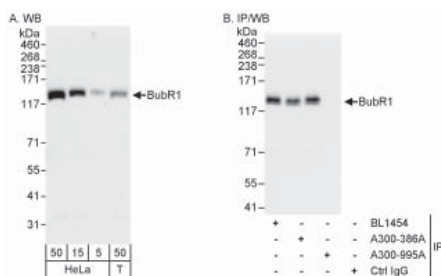
Detection of Human Phospho-53BP1 (Ser 25) by IHC. **Sample:** FFPE section of human colon adenocarcinoma. Mock phosphatase treated section immunostained for Phospho-53BP1. **Antibodies:** Affinity purified rabbit anti-Phospho-53BP1 (Ser 25) (Cat. No. IHC-00053) used at a dilution of 1:250. **Detection:** DAB



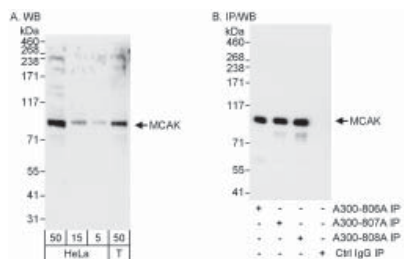
Detection of Human RCC2 by IHC. **Sample:** FFPE section of human basal cell carcinoma. **Antibody:** Affinity purified rabbit anti-RCC2 (Cat. No. IHC-00155) used at a dilution of 1:500. **Detection:** DAB staining using Immunohistochemistry Accessory Kit (Cat. No. IHC-101).



Detection of Human CENP-F/Mitotin by IHC-IF. **Sample:** FFPE section of human ewing sarcoma. **Antibody:** Affinity purified rabbit anti-CENP-F/Mitotin (Cat. No. IHC-00315) used at a dilution of 1:100. **Detection:** Red-fluorescent goat anti-rabbit IgG highly cross-adsorbed Antibody Hilyte Plus™ 555 (A120-501E) used at a dilution of 1:100.



Detection of Human BubR1 by Western Blot and Immunoprecipitation. **Samples:** Whole cell lysate from HeLa (5, 15 and 50 mcg for WB; 1 mg for IP, 20% of IP loaded) and 293T (T; 50 mcg) cells. **Antibodies:** Affinity purified rabbit anti-BubR1 antibody A300-995A used for WB at 0.04 mcg/ml (A) and 1 mcg/ml (B) and used for IP at 3 mcg/mg lysate (B). BubR1 was also immunoprecipitated by rabbit anti-BubR1 antibodies BL1454 and A300-386A, which recognize downstream epitopes. For blotting immunoprecipitated BubR1, the ReliaBLOT® Reagents and Procedures (Cat. No. WB120) were used. **Detection:** Chemiluminescence with exposure times of 30 seconds (A) and 3 seconds (B).



Detection of Human MCAK by Western Blot and Immunoprecipitation. **Samples:** Whole cell lysate from HeLa (5, 15 and 50 mcg for WB; 1 mg for IP, 20% of IP loaded) and 293T (T; 50 mcg) cells. **Antibodies:** Affinity purified rabbit anti-MCAK antibody A300-807A used for WB at 0.4 mcg/ml (A) and 1 mcg/ml (B) and used for IP at 3 mcg/mg lysate (B). MCAK was also immunoprecipitated by rabbit anti-MCAK antibodies A300-806A and A300-808A. For blotting immunoprecipitated MCAK, ReliaBLOT® Reagents and Procedures (Cat. No. WB120) were used. **Detection:** Chemiluminescence with exposure times of 30 seconds (A) and 10 seconds (B).

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