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Kinetochores and disease: keeping microtubule dynamics *in check!*

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Abstract

The essential role of microtubules in cell division has long been known. Yet the mechanism by which microtubule attachment to chromosomes at kinetochores is regulated has only been recently revealed. Here, we review the role of kinetochore-microtubule (kMT) attachment dynamics in the cell cycle as well as emerging evidence linking deregulation of kMT attachments to diseases where chromosome mis-segregation and aneuploidy play a central role. Evidence indicates that the dynamic behavior of kMTs must fall within narrow permissible boundaries, which simultaneously allow a level of stability sufficient to establish and maintain chromosome-microtubule attachments and instability, which permits error correction required for accurate chromosome segregation.

Introduction

Microtubules are a major component of the cytoskeleton [1,2]. Functions of microtubules range from intracellular transport, cellular motility, structural support, chromosome segregation and cell division [1]. Defects in microtubule function or structure leads to a range of cellular dysfunction and disease with the severity depending on the nature of the defect. For instance, in patients with Chédiak-Higashi, cellular microtubules have a subtle defect in microtubule polymerization [3]. This defect impairs phagocytosis in neutrophils leading to recurrent pyogenic infections, it also impairs melanosome transport in melanocytes, leading to partial albinism, and these patients also suffer from peripheral neuropathy as a result of axonal defects [4]. Understanding the dynamic nature of mitotic microtubules in cancer however has been underappreciated, despite the fact that microtubules constitute a longstanding, important, and effective drug target for cancer treatment [5]. Recent work is uncovering an important relationship between microtubule dynamics in mitosis and chromosomal instability (CIN) [6,7]. CIN, which constitutes increased rates of chromosome mis-segregation, is thought to play an important role in cancer progression, adaptation, and drug resistance [8-10].

Microtubule behavior in mitosis

Microtubules are not static polymers of tubulin subunits but they are in a state of constant polymerization and depolymerization through addition and loss of tubulin subunits from the polymer's ends. This growth and shrinkage behavior has been termed “dynamic instability”

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[11,12] and it confers to microtubules the ability to adapt to diverse functions, which require precise spatial and temporal coordination. Strikingly, the dynamic behavior of microtubules is vastly different based on the cell type and the phase of the cell cycle. For instance, in differentiated neurons, axonal microtubules turnover with a half-life of several days [13]. In contrast, microtubules in dividing cells polymerize and depolymerize so rapidly that their half-lives range from seconds to minutes [14]. Recent work suggests that the precise regulation of the dynamic behavior of microtubules is just as important to proper functioning as the structural integrity of microtubules themselves [15]. It was demonstrated that proper cell division and faithful chromosome segregation are meticulously sensitive to minor perturbation in microtubule stability [6,7]. This vulnerability appears to stem from the nature in which chromosomes attach to microtubules at kinetochores. In early mitosis, highly dynamic microtubules search and capture kinetochores and, in human cells, each kinetochore binds 20-25 microtubules [16]. Each kinetochore must bind microtubules emanating from a single spindle pole if the attached chromosome is to properly segregate during anaphase. Yet at kinetochores, microtubules constantly attach and detach. On average, individual microtubules are released from kinetochores every 10-20 seconds and new ones reattach [6]. Such dynamic behavior is error prone as occasionally microtubules emanating from one spindle pole could attach erroneously to a kinetochore that is mostly connected to microtubules from the opposite spindle pole [17-19]. These errors are called merotelic attachments and if left uncorrected prior to anaphase they will impair the proper movement of chromosome segregation and lead to increased probability of mis-segregation [17-19]. The consequence of merotely is lagging chromosomes in anaphase, which were shown to be the primary mechanism of chromosomal instability (CIN) in cancer cell lines [7,20]. Understanding the precise regulation of microtubule dynamics in mitosis has become an emerging focus in recent years due to its importance in the progression as well as fidelity of cell division.

Regulation of microtubule dynamics at kinetochores

Kinetochores are macromolecular organelles formed of ~100 proteins [21,22], many of which have been identified as important regulators of kinetochore-microtubules (kMT) dynamics [23]. Most notably, is the aurora kinase B (aurora B) and protein phosphatase 1 (PP1) [24-26]. Aurora B is localized to the centromeric region between sister kinetochores. From there, it can directly phosphorylate kinetochore components by forming an activity gradient that decays in the direction of kinetochores [27,28]. Inhibition of aurora B dramatically increases kMT stability. kMT half-lives in the presence of the aurora kinase inhibitor, hesperadin, increases from ~2-5 minutes to ~200-700 minutes [24]. Conversely, mislocalizing an active form of aurora B closer to kinetochores appears to perpetually destabilize microtubule attachments [28]. A major mechanism by which aurora B acts on kinetochore-microtubule attachments is through its phosphorylation of the outer kinetochore protein hec1, a component of the conserved Ndc80 complex [29]. In cells transfected with hec1 phosphorylation-resistant mutants, kMT stability mimics these in cells where aurora B is pharmacologically inhibited. On the other hand, PP1 localizes to kinetochores where it potentially antagonizes the actions of aurora B [25,26]. Inhibition of PP1 was shown to increase aurora B activity [30]. This simple relationship however is far from sufficient to explain the precise spatial and temporal regulation of kMTs. Many of the approximately 100 kinetochore proteins play a role in regulating kMT attachments [16]. Furthermore, the attachment stability of microtubules at kinetochores is cell cycle dependent. In early mitosis, while kinetochores are not yet firmly attached to the mitotic spindle, kMT are relatively unstable with a half-life of ~2min [7,29]. As chromosomes align along the metaphase plate and the spindle matures, kMT stability increases 2-3 fold. These subtle changes in microtubule stability are unaccounted for by the all-or-none effects of the aurora B/PP1 axis

indicating that other kinetochore components participate in the subtle temporal control of kMT dynamics.

Many kinetochore proteins have been recently identified as important regulators microtubule attachment. An incomplete list includes Kif2b, MCAK, ska1, CLASP, astrin, nuf2, CenpE, CenpH, CenpF, CenpS, Cep57, HURP, SKAP, and Kif18a [7,15,31-43]. The magnitude of the effect of most of these proteins on kMT dynamics is much smaller than that of aurora B and hec1 and studies on various kinetochore components reveal that the function of kinetochores goes far beyond the simple act of their attachment to microtubules.

Work on kinetochore proteins, astrin, Kif2b and CLASPs recently provided important evidence supporting the precise regulation of kinetochores to their attached microtubules [15,38]. Most important was the temporal nature of this regulation whereby different proteins were recruited to kinetochores at different stages of mitosis to regulate microtubule dynamics. During prometaphase CLASP proteins recruit Kif2b – a kinesin-13 protein that depolymerizes microtubules – where it promotes the release of kMTs. On the other hand, as chromosomes align along the metaphase plate, Kif2b is replaced by astrin, which also binds CLASP proteins and in turn increases kMT stability. Interestingly, the presence of Kif2b and astrin on kinetochores was always mutually exclusive and their localization appears to be a response to aurora B activity and tension between sister-kinetochores. It was also shown that other kinetochore proteins, such as Kif18a, may act through the CLASP/astrin/Kif2b network suggesting the presence of a central, temporally regulated, molecular network at kinetochores that serves to integrate inputs from a plethora of kinetochore proteins to influence kMT dynamics [15,44-47].

Narrow permissible window of kMT dynamics

A striking feature of the CLASP/astrin/Kif2b network is the small magnitude by which it affects kMT stability. Whereas aurora B and Hec1 perturbation can influence stability by 100-200-fold [24,29], perturbation of either Kif2b or astrin only affects kMT turnover by 2-4 folds [6,7]. This relatively minor perturbation does not abrogate the physical attachment of kinetochores to microtubules however it has profound consequences on the progression and fidelity of cell division. Small reductions in kMT stability were sufficient to maintain spindle assembly checkpoint (SAC) signaling and delay or prevent the onset of anaphase. On the other hand, minor increases in kMT stability lead to the persistence of kMT microtubule attachment errors into anaphase, which in turn led to chromosome mis-segregation and chromosomal instability (CIN) [6,7].

The role of microtubule occupancy at kinetochores in silencing the SAC has long been appreciated [48-50]. Studies using quantitative fluorescence microscopy and electron microscopy show that kinetochores require a minimum number of attached microtubules to silence the SAC and allow anaphase onset. Decreasing kMT stability is synonymous to increasing kMT turnover or the rate of detachment of microtubules from kinetochores. At any specific time the number of microtubules attached to kinetochores is the product of the attachment and detachment rates, and subtle increases in the frequency of detachment can lead to a significant decrease in the number attached microtubules. Most perturbations that decrease the half-life of kMT by 2-fold during metaphase lead to anaphase delay [15]. Such a decrease means that individual microtubules are released from kinetochores every ~12 seconds instead of every 18 seconds [6]. It is striking that SAC signaling is maintained in conditions of unstable kMT attachments despite of chromosome alignment along the metaphase plate [15,28]. In experiments where astrin was depleted or a constitutively active aurora B was placed closer to kinetochores cells achieved full chromosome alignment but failed to progress through anaphase despite achieving typical inter-kinetochore tension

[15,28]. This demonstrates that kMT occupancy can serve as the primary regulator of the SAC signaling independent of the tension between sister-kinetochores brought about by alignment during metaphase [49], although this is a matter of continuous debate [49,51-56]. In addition, in experiments where kMT stability was prematurely increased either by inhibiting aurora B, modifying hec1 or adding low doses of drugs that stabilize microtubules, cells prematurely underwent anaphase without achieving proper alignment of inter-kinetochore tension [57-60].

In the same way that subtle reductions in kMT stability influence cell cycle progression, minor increases in the stability of kMT attachments have a profound effect on chromosome segregation fidelity [7]. This effect is due to the stochastic nature of kinetochore microtubule interaction throughout mitosis, particularly in prometaphase [2]. As kinetochore pairs align along the mitotic spindle, they attach to microtubules emanating from opposite spindle poles. The constant and frequent attachment and release of microtubules together with the rigid centromere geometry that links sister kinetochores favor their biorientation, whereby each kinetochore of a pair is attached to only one spindle pole [6,61]. Increasing kMT stability however, means decreasing kMT release rates in such a way that if individual microtubules were improperly attached that they would remain until the onset of anaphase. Individual kinetochores that are merotelically attached are not detected by the SAC [17,18,62]. Consequently, the persistence of merotelic chromosomes into anaphase leads to a tug of war between opposing microtubules and frequently the lagging of these chromosomes in the middle of the anaphase spindles. These lagging chromosomes were shown to have a higher propensity to mis-segregate and are a common feature of cells with chromosomal instability (CIN) [20].

Examples where increasing kMT stability leads to CIN are plenty and a partial list includes depletion, inhibition, or perturbation of the following proteins: Kif2b, MCAK, hec1, aurora B, CLASP, APC [6,7,15,29,63]. The function of some of these proteins has been shown to be perturbed in tumors and the search is underway to find the mechanisms underlying the relationship between regulators of kMT dynamics and CIN. In human-derived cell lines, an increase of kMT by 2 to 4-fold is sufficient to induce chromosome mis-segregation. This change would increase the mean lifetime of a microtubule at kinetochores of a diploid chromosomally stable cell line (RPE1) from ~5 minutes to ~10-20 minutes or even higher [6]. This extension of attachment stability means that if a microtubule erroneously attaches to kinetochores it is likely to remain attached for that period of time before being released and this is likely not to take place prior to anaphase onset.

These findings uncover several important concepts about the process of chromosome segregation fidelity in human cells. First, at all times prior to anaphase, the rate of attachment and detachment of microtubules from kinetochores is balanced in such a way that attachment errors are corrected at sufficiently high probability before anaphase onset [6,24,64]. Second, the machinery involved in allowing for the correction of kMT attachment errors may not be sufficiently robust to withstand minor perturbations. These perturbations can either increase in attachment error rate or a decrease the correction rate [7,29,65,66]. The former was shown in cells containing multiple centrosomes where the transition from multipolar to bipolar spindles increases the formation of attachments errors and consequently lagging chromosomes [67,68]. Furthermore, transient treatment of cells with molecules that perturb spindle geometry such as monastrol or nocodazole leads to increased rates of lagging chromosomes without affecting kMT dynamics prior to anaphase. Concomitant increases in kMT stability dramatically compromises chromosome segregation fidelity whereas slightly promoting kMT turnover can increase the rate of microtubule release from kinetochores and lead to suppression of the drug treatment effects [7]. It was also shown that chromosomally unstable cancer cell lines have an inherent increase in kMT

attachment stability indicating their reduced efficiency to correct attachment errors whenever they arise [6]. Interestingly, promoting the turnover of kMT attachment in these cells was sufficient to significantly reduce the rates of lagging chromosomes and suppress CIN [7]. Thus it is evident that cancer cells exhibit defects in one or both processes of formation and correction of attachment errors [66].

Taken together, the evidence points to a narrow acceptable range for the attachment stability of kMTs (Figure 1). kMTs must be stable enough to reach full occupancy of kinetochores to satisfy the checkpoint, but sufficiently unstable to permit error correction. These narrow boundaries suggest the importance of the precise regulation of kMT attachment dynamics. In a peculiar manner, a kinetochore acts as a stabilostat for kMTs whereby stability must be appropriately maintained during all stages of mitosis (Figure 1). Phylogenetic analysis of kinesin-13 proteins from three species: humans (H*s*), mice (M*m*) and Drosophila Melagnogaster (D*m*) shows that gene duplication occurred independently throughout evolution despite the fact that each has three copies of kinesin-13 gene that perform distinct function that is conserved among the three species [69] (Figure 2). This suggests that individual kinetochore proteins may not necessarily display high sequence conservation among various eukaryotes yet homologous genes have evolved to maintain the appropriate regulation of microtubules based on the requirement of the species and the number of microtubules that attach to its kinetochores.

Mosaic Variegated Aneuploidy (MVA) Syndrome

Given the tight requirements for the regulation of kMT dynamics it is expected that genetic mutations found in disease conditions may influence the kMT interface and perturb either the timely progression of the cell cycle or the fidelity of chromosome segregation process. Such an example is Mosaic Variegated Aneuploidy (MVA), which is a rare autosomal recessive syndrome associated with various trisomies and monosomies involving different chromosomes and tissues [70]. It causes severe intrauterine growth retardation, microcephaly, dysmorphism, and most importantly, increased risk of malignancy such as rhabdomyosarcoma, Wilms tumors, and leukemia [71-73]. MVA can be caused by mutations in BubR1 (encoded by Bub1B) as patients harbor two mutant alleles and are compound heterzygotes. BubR1 is a serine/threonine kinase that phosphorylates important SAC proteins and is thought be necessary to maintain SAC signaling [74,75]. Thus, it is reasonable to suggest that MVA is caused by defects in the SAC [76,77]. It was postulated that sister chromatids lost cohesion prematurely, well before sister kinetochores were able to bi-orient and establish sufficiently stable attachments with microtubules. However, BubR1 is also known to be involved in regulating kMT dynamics and its depletion leads to unstable kMT attachments [78]. This finding together with the observation that in some patients with MVA many sister chromatids do not prematurely separate [71,74,79], suggest that kMT attachment dynamics may play an important role in the pathogenesis of MVA. This hypothesis is further supported by the finding that mutations in Cep57, a kinetochore and centrosomal protein involved in stabilizing kinetochore microtubules [40,41,80], is found in some patients with MVA [81]. It is thus reasonable to postulate that deregulated kMT attachments may have a role in MVA and there may be many molecular etiologies given the complexity of the kMT interface.

Kinetochore microtubule dynamics in cancer

In contrast to the inherited condition of MVA, most tumors arise due to the accumulation of somatic mutations. The effect of individual gene mutations on kMT dynamics is largely an unexplored field [82], however given the complexity of the kMT interface, it is possible that many of these mutations either directly or indirectly influence kMT stability. Many proteins

were shown to be overexpressed in cancer with the putative consequence of increasing kMT stability. Examples include CenpH, Cyclin E, Hec1/Ndc80 complex, and MCT-1 [83-88]. Conversely, other proteins were shown to be mutated, deleted, or underexpressed such as APC, beta catenin, pRB, and BRCA1 [6,89-93]. It is unclear however if these genes are initial triggers of chromosome mis-segregation and CIN or if they simply act by exacerbating mis-segregation rates after the onset of CIN. Furthermore, genetic analysis of CIN may be complicated by the fact that after the initial chromosome mis-segregation event, the levels of hundreds to thousands of proteins encoded by the mis-segregated chromosome would be disturbed [6,82,94]. This would lead to a significant imbalance in the ratio of proteins that either stabilize or destabilize kMT attachments. Such an imbalance may by itself lead to further mis-segregation events given the narrow permissible boundaries of kMT dynamics, in such a way that CIN may be a self-propagating event [6]. This hypothesis is testable and would lead to an important insight into the progression of CIN. Preliminary work on cancer cell lines with CIN demonstrates that each has a unique set of ratios of microtubule stabilizers and destabilizers suggesting that this imbalance could both be the consequence as well as the continuous driver for chromosome mis-segregation.

Concluding remarks

When put together, recent data in the field indicate that, during cell division, microtubule dynamics is a finely and temporally regulated process whereby the stability of kMT attachments must be constrained within a defined narrow window that is permissible for faithful mitotic progression. Decreasing kMT stability leads to cell cycle arrest whereas increasing their stability directly leads to chromosome segregation defects. Furthermore, cancer cells are either less efficient in correcting attachment errors or possess defects which lead to increased formation of these errors, necessitating a compensatory increase in the correction machinery. Thus, in theory, it is possible to exploit microtubule dynamics during mitosis to influence the progression of cell division. A targeted approach could either reduce the stability of the attachment for the purpose of halting cell division or reducing the rates of chromosome mis-segregation in an effort to reduce cancer's ability to adapt to the microenvironment or chemotherapeutic drugs. The advantage of such targeted therapy is the exquisite sensitivity of mitotic cells to minor perturbation of microtubules, which might spare novel drugs from the dose-limiting and neurotoxic side-effect exhibited by the widely used taxane class of anti-microtubule drugs.

References

1. Olmsted JB, Borisy GG. Microtubules. AnnuRevBiochem. 1973; 42:507–540.
2. Compton DA. Spindle assembly in animal cells. AnnuRevBiochem. 2000; 69:95–114.
3. White JG. Platelet microtubules and giant granules in chediak-higashi-syndrome. American Journal of Medical Technology. 1978; 44(4):273–278. [PubMed: 206143]
4. Kaplan J, De Domenico I, Ward DM. Chediak-higashi syndrome. Current Opinion in Hematology. 2008; 15(1):22–29. [PubMed: 18043242]
5. Wilson L, Jordan MA. New microtubule/tubulin-targeted anticancer drugs and novel chemotherapeutic strategies. Journal of Chemotherapy. 2004; 16:83–85. [PubMed: 15688618]
6**. Bakhoum SF, Genovese G, Compton DA. Deviant kinetochore microtubule dynamics underlie chromosomal instability. Curr Biol. 2009; 19(22):1937–1942. Paper showing that cancer cells have defective kinetochore-microtubule dynamics. It also provides estimates for kinetochore microtubule attachment/detachment rates. [PubMed: 19879145]
7. Bakhoum SF, Thompson SL, Manning AL, Compton DA. Genome stability is ensured by temporal control of kinetochore-microtubule dynamics. NatCell Biol. 2009; 11(1):27–35.
8. Lengauer C, Kinzler KW, Vogelstein B. Genetic instability in colorectal cancers. Nature. 1997; 386(6625):623–627. [PubMed: 9121588]

9. Lengauer C, Kinzler KW, Vogelstein B. Genetic instabilities in human cancers. Nature. 1998; 396(6712):643–649. [PubMed: 9872311]
10. Swanton C, Lee AJX, Endesfelder D, Rowan AJ, Walther A, Birkbak NJ, Futreal PA, Downward J, Szallasi Z, Tomlinson IPM, Howell M, et al. Chromosomal instability confers intrinsic multidrug resistance. Cancer Res. 2011; 71(5):1858–1870. [PubMed: 21363922]
11. Burbank KS, Mitchison TJ. Microtubule dynamic instability. CurrBiol. 2006; 16(14):R516–517.
12. Desai A, Mitchison TJ. Microtubule polymerization dynamics. AnnuRevCell DevBiol. 1997; 13:83–117.
13. Meininger V, B S. Characteristics of microtubules at the different stages of neuronal differentiation and maturation. International Review of Cytology. 1989; 114:21–79. [PubMed: 2661457]
14**. Zhai Y, Kronebusch PJ, Borisy GG. Kinetochore microtubule dynamics and the metaphase-anaphase transition. JCell Biol. 1995; 131(3):721, 734. first paper to measure kinetochore-microtubule half-life during mitosis. [PubMed: 7593192]
15***. Manning AL, Bakhoum SF, Maffini S, Correia-Melo C, Maiato H, Compton DA. Clasp1, astrin and kif2b form a molecular switch that regulates kinetochore-microtubule dynamics to promote mitotic progression and fidelity. Embo Journal. 2010; 29(20):3531–3543. Paper showing a regulatory network at kinetochores that allows microtubules to remain within the permissible boundaries of dynamics. [PubMed: 20852589]
16. Cheeseman IM, Desai A. Molecular architecture of the kinetochore-microtubule interface. NatRevMolCell Biol. 2008; 9(1):33–46.
17. Cimini D, Howell BJ, Degrassi F, Salmon ED. Merotelic attachment of single kinetochores to microtubules from opposite poles is not detected by the mitotic spindle checkpoint and induces chromosome loss during mitosis. MolBiolCell. 2000; 1143:0a–430a.
18. Cimini D, Howell B, Maddox P, Khodjakov A, Degrassi F, Salmon ED. Merotelic kinetochore orientation is a major mechanism of aneuploidy in mitotic mammalian tissue cells. JCell Biol. 2001; 153(3):517–527. [PubMed: 11331303]
19. Cimini D, Fioravanti D, Salmon ED, Degrassi F. Merotelic kinetochore orientation versus chromosome mono-orientation in the origin of lagging chromosomes in human primary cells. JCellSci. 2002; 115(Pt 3):507–515.
20. Thompson SL, Compton DA. Examining the link between chromosomal instability and aneuploidy in human cells. JCell Biol. 2008; 180(4):665–672. [PubMed: 18283116]
21. Gascoigne KE, Cheeseman IM. Kinetochore assembly: If you build it, they will come. Current Opinion in Cell Biology. 2011; 23(1):102–108. [PubMed: 20702077]
22. Bower J. Mammalian kinetochore structure and function. Genet Res. 1987; 49(3):258–258.
23. Wan XH, O'Quinn RP, Pierce HL, Joglekar AP, Gall WE, DeLuca JG, Carroll CW, Liu ST, Yen TJ, McEwen BF, Stukenberg T, et al. Protein architecture of the human kinetochore microtubule attachment site. Cell. 2009; 137(4):672–684. [PubMed: 19450515]
24. Cimini D, Wan XH, Hirel CB, Salmon ED. Aurora kinase promotes turnover of kinetochore microtubules to reduce chromosome segregation errors. Curr Biol. 2006; 16(17):1711–1718. [PubMed: 16950108]
25. Emanuele MJ, Lan W, Jwa M, Miller SA, Chan CSM, Stukenberg PT. Aurora b kinase and protein phosphatase 1 have opposing roles in modulating kinetochore assembly. J Cell Biol. 2008; 181(2): 241–254. [PubMed: 18426974]
26. Liu D, Vleugel M, Backer CB, Hori T, Fukagawa T, Cheeseman IM, Lampson MA. Regulated targeting of protein phosphatase 1 to the outer kinetochore by knl1 opposes aurora b kinase. Journal of Cell Biology. 2010; 188(6):809–820. [PubMed: 20231380]
27. Fuller BG, Lampson MA, Foley EA, Rosasco-Nitcher S, Le KV, Tobelmann P, Brautigan DL, Stukenberg PT, Kapoor TM. Midzone activation of aurora b in anaphase produces an intracellular phosphorylation gradient. Nature. 2008; 453(7198):1132–U1114. [PubMed: 18463638]
28***. Liu D, Vader G, Vromans MJM, Lampson MA, Lens SMA. Sensing chromosome bi-orientation by spatial separation of aurora b kinase from kinetochore substrates. Science. 2009; 323(5919):1350–1353. Important paper showing the relationship between Aurora B kinase and kinetochore microtubule dynamics. In addition, it shows that perpetually increasing kMT turnover leads to metaphase arrest. [PubMed: 19150808]

29. DeLuca JG, Gall WE, Ciferri C, Cimini D, Musacchio A, Salmon ED. Kinetochore microtubule dynamics and attachment stability are regulated by hec1. Cell. 2006; 127(5):969–982. [PubMed: 17129782]
30. Posch M, Khoudoli GA, Swift S, King EM, DeLuca JG, Swedlow JR. Sds22 regulates aurora b activity and microtubule-kinetochore interactions at mitosis. J Cell Biol. 2010; 191(1):61–74. [PubMed: 20921135]
31. Lan WJ, Zhang X, Kline-Smith SL, Rosasco SE, Barrett-Wilt GA, Shabanowitz J, Walczak CE, Stukenberg PT. Aurora b phosphorylates centromeric mcak and regulates its localization and microtubule depolymerization activity. Curr Biol. 2004; 14(4):273–286. [PubMed: 14972678]
32. Maiato H, Fairley EAL, Rieder CL, Swedlow JR, Sunkel CE, Earnshaw WC. Human clasp1 is an outer kinetochore component that regulates spindle microtubule dynamics. Cell. 2003; 113(7): 891–904. [PubMed: 12837247]
33. Welburn JPI, Grishchuk EL, Backer CB, Wilson-Kubalek EM, Yates JR, Cheeseman IM. The human kinetochore ska1 complex facilitates microtubule depolymerization-coupled motility. DevCell. 2009; 16(3):374–385.
34. Cheeseman IM, Hori T, Fukagawa T, Desai A. Knl1 and the cenp-h/i/k complex coordinately direct kinetochore assembly in vertebrates. MolBiolCell. 2008; 19(2):587–594.
35. Cheeseman IM, MacLeod I, Yates JR, Oegema K, Desai A. The cenp-f-like proteins hcp-1 and hcp-2 target clasp to kinetochores to mediate chromosome segregation. Curr Biol. 2005; 15(8): 771–777. [PubMed: 15854912]
36. Amano M, Suzuki A, Hori T, Backer C, Okawa K, Cheeseman IM, Fukagawa T. The cenp-s complex is essential for the stable assembly of outer kinetochore structure. Journal of Cell Biology. 2009; 186(2):173–182. [PubMed: 19620631]
37. Kline SL, Cheeseman IM, Hori T, Fukagawa T, Desai A. The human mis 12 complex is required for kinetochore assembly and proper chromosome segregation. Journal of Cell Biology. 2006; 173(1):9–17. [PubMed: 16585270]
38. Maffini S, Maia ARR, Manning AL, Maliga Z, Pereira AL, Junqueira M, Shevchenko A, Hyman A, Yates JR, Galjart N, Compton DA, et al. Motor-independent targeting of clasps to kinetochores by cenp-e promotes microtubule turnover and poleward flux. Curr Biol. 2009; 19(18):1566–1572. [PubMed: 19733075]
39. DeLuca JG, Moree B, Hickey JM, Kilmartin JV, Salmon ED. Hnuf2 inhibition blocks stable kinetochore-microtubule attachment and induces mitotic cell death in hela cells. J Cell Biol. 2002; 159(4):549–555. [PubMed: 12438418]
40. Momotani K, Khromov AS, Miyake T, Stukenberg RT, Somlyo AV. Cep57, a multidomain protein with unique microtubule and centrosomal localization domains. Biochemical Journal. 2008; 412:265–273. [PubMed: 18294141]
41. Emanuele MJ, Stukenberg PT. Xenopus cep57 is a novel kinetochore component involved in microtubule attachment. Cell. 2007; 130(5):893–905. [PubMed: 17803911]
42. Mattaj IW, Casanova CM, Rybina S, Yokoyama H, Karsenti E. Hepatoma up-regulated protein is required for chromatin-induced microtubule assembly independently of tpx2. MolBiolCell. 2008; 19(11):4900–4908.
43. Gruneberg U, Dunsch AK, Linnane E, Barr FA. The astrin-kinastrin/skap complex localizes to microtubule plus ends and facilitates chromosome alignment. J Cell Biol. 2011; 192(6):959–968. [PubMed: 21402792]
44. Stumpff J, von Dassow G, Wagenbach M, Asbury C, Wordeman L. The kinesin-8 motor kif18a suppresses kinetochore movements to control mitotic chromosome alignment. DevCell. 2008; 14(2):252–262.
45. Mayr MI, Hummer S, Bormann J, Gruner T, Adio S, Woehlke G, Mayer TU. The human kinesin kif18a is a motile microtubule depolymerase essential for chromosome congression. CurrBiol. 2007; 17(6):488–498.
46. Gupta ML Jr, Carvalho P, Roof DM, Pellman D. Plus end-specific depolymerase activity of kip3, a kinesin-8 protein, explains its role in positioning the yeast mitotic spindle. NatCell Biol. 2006; 8(9):913–923.

47. Varga V, Helenius J, Tanaka K, Hyman AA, Tanaka TU, Howard J. Yeast kinesin-8 depolymerizes microtubules in a length-dependent manner. NatCell Biol. 2006; 8(9):957–962.
48. Musacchio A, Salmon ED. The spindle-assembly checkpoint in space and time. NatRevMolCell Biol. 2007; 8(5):379–393.
49. McEwen BF, Dong YM. Releasing the spindle assembly checkpoint without tension. J Cell Biol. 2009; 184(3):355–356. [PubMed: 19193624]
50. Musacchio A, Hardwick KG. The spindle checkpoint: Structural insights into dynamic signalling. NatRevMolCell Biol. 2002; 3(10):731–741.
51. Nicklas RB, Ward SC, Gorbsky GJ. Kinetochore chemistry is sensitive to tension and may link mitotic forces to a cell-cycle checkpoint. J Cell Biol. 1995; 130(4):929–939. [PubMed: 7642708]
52. Uchida KSK, Takagaki K, Kumada K, Hirayama Y, Noda T, Hirota T. Kinetochore stretching inactivates the spindle assembly checkpoint. J Cell Biol. 2009; 184(3):383–390. [PubMed: 19188492]
53. Paliulis LV, Nicklas RB. Release of chromosome cohesion during anaphase is gradual and does not require tension. MolBiolCell. 2004; 1523:9a–240a.
54. King JM, Nicklas RB. Tension on chromosomes increases the number of kinetochore microtubules but only within limits. JCellSci. 2000; 113(Pt 21):3815–3823.
55. Nicklas RB, Campbell MS, Ward SC, Gorbsky GJ. Tension-sensitive kinetochore phosphorylation in vitro. JCellSci. 1998; 111(Pt 21)(Pt 21):3189–3196.
56. King JM, Nicklas RB. Tension, kinetochore microtubule number, and the spindle checkpoint. MolBiolCell. 1999; 1012:7a–127a.
57. Martin-Lluesma S, Stucke VM, Nigg EA. Role of hec1 in spindle checkpoint signaling and kinetochore recruitment of mad1/mad2. Science. 2002; 297(5590):2267–2270. [PubMed: 12351790]
58. Carvalho A, Carmena M, Sambade C, Earnshaw WC, Wheatley SP. Survivin is required for stable checkpoint activation in taxol-treated hela cells. JCellSci. 2003; 116(14):2987–2998.
59. Hauf S, Cole RW, LaTerra S, Zimmer C, Schnapp G, Walter R, Heckel A, van Meel J, Rieder CL, Peters JM. The small molecule hesperadin reveals a role for aurora b in correcting kinetochore-microtubule attachment and in maintaining the spindle assembly checkpoint. J Cell Biol. 2003; 161(2):281–294. [PubMed: 12707311]
60. Rieder CL, Yang ZY, Kenny AE, Brito DA. Cells satisfy the mitotic checkpoint in taxol, and do so faster in concentrations that stabilize syntelic attachments. J Cell Biol. 2009; 186(5):675–684. [PubMed: 19720871]
61. Loncarek J, Kisurina-Evgenieva O, Vinogradova T, Hergert P, La Terra S, Kapoor TM, Khodjakov A. The centromere geometry essential for keeping mitosis error free is controlled by spindle forces. Nature. 2007; 450(7170):745–749. [PubMed: 18046416]
62. Cimini D. Merotelic kinetochore orientation, aneuploidy, and cancer. BiochimBiophysActa. 2008; 1786(1):32–40.
63. Cimini D, Salmon ED. Aurora b contributes to the correction of merotelic kinetochore orientation before anaphase onset. MolBiolCell. 2004; 1526:4a–264a.
64. Cimini D, Moree CB, Maddox PS, Degrassi F, Salmon ED. Prolonging metaphase decreases kinetochore merotelic orientations. MolBiolCell. 2001; 1231:5a–315a.
65. Cimini D, Moree CB, Salmon ED. A delay in bi-polar spindle formation strongly promotes merotelic kinetochore orientation. MolBiolCell. 2002; 1330:9a–309a.
66. Bakhoum SF, Compton DA. Cancer: Cinful centrosomes. Curr Biol. 2009; 19(15):R642–R645. [PubMed: 19674544]
67. Ganem NJ, Godinho SA, Pellman D. A mechanism linking extra centrosomes to chromosomal instability. Nature. 200910.1038/nature08136
68. Silkworth WT, Nardi IK, Scholl LM, Cimini D. Multipolar spindle pole coalescence is a major source of kinetochore mis-attachment and chromosome mis-segregation in cancer cells. Plos One. 2009; 8(4):e6564. [PubMed: 19668340]

69. Manning AL, Ganem NJ, Bakhoum SF, Wagenbach M, Wordeman L, Compton DA. The kinesin-13 proteins kif2a, kif2b, and kif2c/mcak have distinct roles during mitosis in human cells. MolBiolCell. 2007; 18(8):2970–2979.

70. Kawame H, Sugio Y, Fuyama Y, Hagashi Y, Suzuki H, Kurosawa K, Maekawa K. Syndrome of microcephaly, dandy-walker malformation, and wilms tumor caused by mosaic variegated aneuploidy with premature centromere division (pcd): Report of a new case and review of the literature. Journal of Human Genetics. 1999; 44(4):219–224. [PubMed: 10429359]

71. Jacquemont S, Boceno M, Rival JM, Mechinaud F, David A. High risk of malignancy in mosaic variegated aneuploidy syndrome. Am J Med Genet. 2002; 109(1):17–21. [PubMed: 11932988]

72. Kajii T, Ikeuchi T, Yang ZQ, Nakamura Y, Tsuji Y, Yokomori K, Kawamura M, Fukuda S, Horita S, Asamoto A. Cancer-prone syndrome of mosaic variegated aneuploidy and total premature chromatid separation: Report of five infants. American Journal of Medical Genetics. 2001; 104(1): 57–64. [PubMed: 11746029]

73. Kajii T, Kawai T, Takumi T, Misu H, Mabuchi O, Takahashi Y, Tachino M, Nihei F, Ikeuchi T. Mosaic variegated aneuploidy with multiple congenital abnormalities: Homozygosity for total premature chromatid separation trait. American Journal of Medical Genetics. 1998; 78(3):245–249. [PubMed: 9677059]

74. Hanks S, Coleman K, Reid S, Plaja A, Firth H, FitzPatrick D, Kidd A, Mehes K, Nash R, Robin N, Shannon N, et al. Constitutional aneuploidy and cancer predisposition caused by biallelic mutations in bub1b. Nature Genetics. 2004; 36(11):1159–1161. [PubMed: 15475955]

75. Matsuura S, Matsumoto Y, Morishima K, Izumi H, Matsumoto H, Ito E, Tsutsui K, Kobayashi J, Tauchi H, Kajiwara Y, Hama S, et al. Monoallelic bub1b mutations and defective mitotic-spindle checkpoint in seven families with premature chromatid separation (pcs) syndrome. American Journal of Medical Genetics Part A. 2006; 140A(4):358–367. [PubMed: 16411201]

76. Matsuura S, Ito E, Ikeuchi T, Kajii T, Komatsu K. Chromosomal instability syndrome of total pcs with mosaic variegated aneuploidy: Genetic homogeneity of the two japanese infants. American Journal of Human Genetics. 2001; 69(4):311–311.

77. Matsuura S, Ito E, Tauchi H, Komatsu K, Ikeuchi T, Kajii T. Chromosomal instability syndrome of total premature chromatid separation with mosaic variegated aneuploidy is defective in mitotic-spindle checkpoint. American Journal of Human Genetics. 2000; 67(2):483–486. [PubMed: 10877982]

78. Lampson MA, Kapoor TM. The human mitotic checkpoint protein bubr1 regulates chromosome-spindle attachments. NatCell Biol. 2005; 7(1):93–+.

79. Limwongse C, Schwartz S, Bocian M, Robin NH. Child with mosaic variegated aneuploidy and embryonal rhabdomyosarcoma. American Journal of Medical Genetics. 1999; 82(1):20–24. [PubMed: 9916837]

80. Yoshitake Y, Tsuruoka N, Iwai A. A novel centrosomal protein, cep57 stabilizes microtubules in vivo and binds fgf-2 in vitro. Febs Journal. 2005; 272:336–337.

81***. Snape K, Hanks S, Ruark E, Barros-Nunez P, Elliott A, Murray A, Lane AH, Shannon N, Callier P, Chitayat D, Clayton-Smith J, et al. Mutations in cep57 cause mosaic variegated aneuploidy syndrome. Nature Genetics. 2011; 43(6):527–529. Paper showing a potential link between deregulation of kinetochore-microtubule dynamics and Mosaic Variegated aneuploidy. [PubMed: 21552266]

82. Wang Z, Cummins JM, Shen D, Cahill DP, Jallepalli PV, Wang TL, Parsons DW, Traverso G, Awad M, Silliman N, Ptak J, et al. Three classes of genes mutated in colorectal cancers with chromosomal instability. Cancer Res. 2004; 64(9):2998–3001. [PubMed: 15126332]

83. Tomonaga T, Matsushita K, Ishibashi M, Nezu M, Shimada H, Ochiai T, Yoda K, Nomura F. Centromere protein h is up-regulated in primary human colorectal cancer and its overexpression induces aneuploidy. Cancer Research. 2005; 65(11):4683–4689. [PubMed: 15930286]

84. Rajagopalan H, Jallepalli PV, Rago C, Velculescu VE, Kinzler KW, Vogelstein B, Lengauer C. Inactivation of hcdc4 can cause chromosomal instability. Nature. 2004; 428(6978):77–81. [PubMed: 14999283]

85. Spruck CH, Won KA, Reed SI. Deregulated cyclin e induces chromosome instability. Nature. 1999; 401(6750):297–300. [PubMed: 10499591]

86. Tzankov A, Gschwendtner A, Augustin F, Fiegl M, Obermann EC, Dirnhofer S, Went P. Diffuse large b-cell lymphoma with overexpression of cyclin e substantiates poor standard treatment response and inferior outcome. Clinical Cancer Research. 2006; 12(7):2125–2132. [PubMed: 16609025]

87. Benezra R, Diaz-Rodriguez E, Sotillo R, Schvartzman JM. Hec1 overexpression hyperactivates the mitotic checkpoint and induces tumor formation in vivo. ProcNatlAcadSciUSA. 2008; 105(43): 16719–16724.

88. Hsu HL, Kasiappan R, Shih HJ, Chu KL, Chen WT, Liu HP, Huang SF, Choy CO, Shu CL, Din R, Chu JS. Loss of p53 and mct-1 overexpression synergistically promote chromosome instability and tumorigenicity. Molecular Cancer Research. 2009; 7(4):536–548. [PubMed: 19372582]

89. Dikovskaya D, Schiffmann D, Newton IP, Oakley A, Kroboth K, Sansom O, Jamieson TJ, Meniel V, Clarke A, Nathke IS. Loss of apc induces polyploidy as a result of a combination of defects in mitosis and apoptosis. JCell Biol. 2007; 176(2):183–195. [PubMed: 17227893]

90. Fodde R, Kuipers J, Rosenberg C, Smits R, Kielman M, Gaspar C, van Es JH, Breukel C, Wiegant J, Giles RH, Clevers H. Mutations in the apc tumour suppressor gene cause chromosomal instability. NatCell Biol. 2001; 3(4):433–438.

91. Taketo MM, Aoki K, Aoki M, Sugai M, Harada N, Miyoshi H, Tsukamoto T, Mizoshita T, Tatematsu M, Seno H, Chiba T, et al. Chromosomal instability by beta-catenin/tcf transcription in apc or beta-catenin mutant cells. Oncogene. 2007; 26(24):3511–3520. [PubMed: 17160019]

92. Walter JC, Joukov V, Groen AC, Prokhorova T, Gerson R, White E, Rodriguez A, Livingston DM. The brca1/bard1 heterodimer modulates ran-dependent mitotic spindle assembly. Cell. 2006; 127(3):539–552. [PubMed: 17081976]

93. Dyson NJ, Manning AL, Longworth MS. Loss of prb causes centromere dysfunction and chromosomal instability. Genes & Development. 2010; 24(13):1364–1376. [PubMed: 20551165]

94. Shih IM, Zhou W, Goodman SN, Lengauer C, Kinzler KW, Vogelstein B. Evidence that genetic instability occurs at an early stage of colorectal tumorigenesis. Cancer Res. 2001; 61(3):818–822. [PubMed: 11221861]

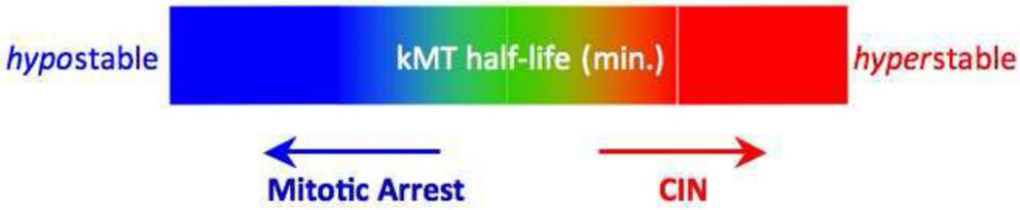

Figure 1.
Kinetochore-microtubule stabilostat model. Kinetochore-microtubules (kMT) attachment stability is regulated in a narrow range for high fidelity chromosome segregation. Hypostable kMT attachments fails to adequately satisfy the spindle assembly checkpoint leading to mitotic delay or arrest. Hyperstable kMT attachments lose efficient error correction with the consequence being the persistence of attachment errors in anaphase and an increase in chromosome mis-segregation leading to chromosomal instability (CIN).

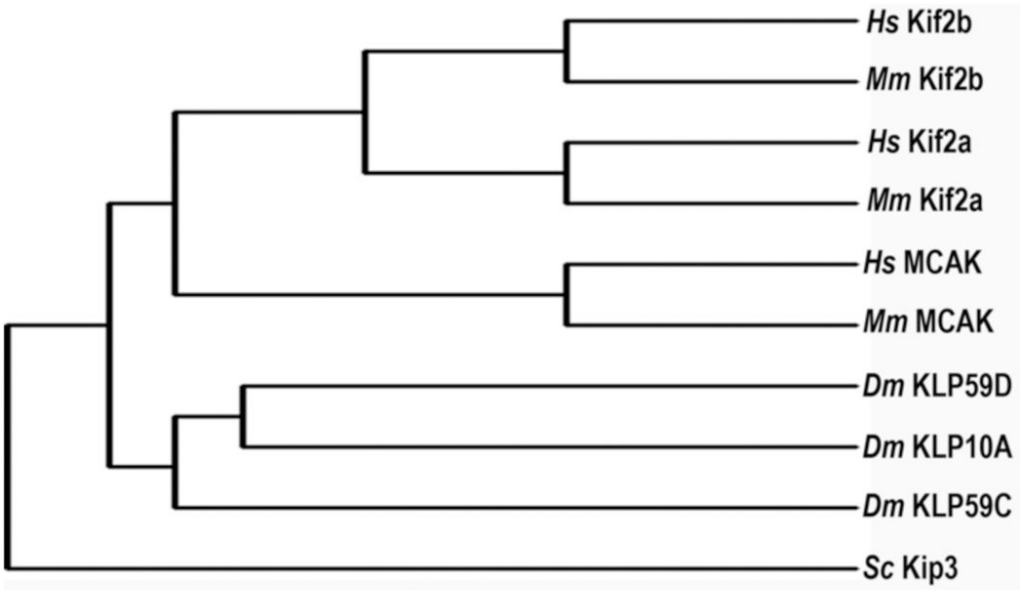

Figure 2.
Phylogenetic comparison of kinesin-13 genes. Amino acid sequences from three kinesin-13 proteins encoded by fruit fly (Dm), mouse (Mm), and human (Hs) are compared using Clustal X. The sequence of yeast Kip3 is used as an outgroup to root the tree. Line engths are not to scale. Reprinted from Manning et *al.* 2007 (http://www.molbiolcell.org/content/18/8/2970)