Mitotic Motors: Kinesin-5 Takes a Brake

A kinesin-5-dependent ‘sliding filament’ mechanism is commonly used to actively push apart the poles during mitotic spindle assembly and elongation, but a recent study now shows that, in C. elegans, kinesin-5 is deployed as a brake to slow down spindle-pole separation.

Gül Civelekoglu-Scholey
and Jonathan M. Scholey

Faithful chromosome segregation depends upon the formation and function of a bipolar, microtubule (MT)-based mitotic spindle, which uses multiple mitotic motors to assemble itself and to separate sister chromatids [1]. Among these motors, members of the kinesin-5 family [2] are thought to have critical and often essential mitotic functions, by pushing apart the spindle poles, for example during anaphase B spindle elongation [3].

Curiously, however, the single kinesin-5 present in Caenorhabditis elegans, BMK-1, is dispensable for mitosis. Now, new work from the Saxton and Strome laboratories, published recently in Current Biology, shows that, in this system, BMK-1 has novel mitotic functions, serving as a brake that restrains the rate of anaphase spindle-pole separation driven by other cortical force generators [4].

Saunders et al. [4] studied the role of kinesin-5 in early C. elegans embryos, where anaphase B spindle elongation represents the major mechanism for chromosome segregation and where anaphase...
A chromatid-to-pole motion contributes little. The kinesin-5, BMK-1, localizes to the region of overlapping interpolar (ip) MTs of wild-type embryo spindles and, therefore, since such ipMTs slide apart during anaphase B [5], the motor is in an appropriate position to generate forces that contribute to spindle elongation. But if BMK-1 were involved in generating forces that push apart the spindle poles (like kinesin-5 motors in other systems), one would expect that the loss of its function would lead to a decrease in the rate and/or extent of spindle-pole separation. Surprisingly, however, abnormally fast pole–pole separation was observed following loss of kinesin-5 function in bmk-1 deletion mutants, suggesting that, in this system, BMK-1 normally functions as a rate-limiting brake that governs the rate of spindle elongation. A plausible explanation is that anaphase B is normally stalled by astral pulling forces is consistent with early micromanipulation and UV laser ablation experiments (e.g. see [6]). The results of Saunders et al. [4] are also consistent with competitive in vitro motility assays in which kinesin-5 was observed to slow down the sliding of a MT being moved by a ‘fast’ MT plus-end-directed motor [7]. Yet in more ‘conventional’ models, kinesin-5 acts on ipMTs to perform a different function that appears consistent with its known biochemical and ultrastructural properties (e.g. Figure 1B–E). For example, purified kinesin-5 is a ‘slow’ plus-end-directed homotetramer with MT-binding motor domains located at opposite ends of a central rod [8–10] and, in at least one system, its assembly into homotetramers is essential for biological function [11]. In clever motility assays, kinesin-5 has been shown to drive a ‘sliding filament’ mechanism by crosslinking adjacent microtubules and sliding them in relation to one another [12]. Within the mitotic spindle, ensembles of kinesin-5 motors are proposed to exert force between adjacent MTs to drive poleward MT sliding, which is coupled to MT depolymerization at spindle poles to produce poleward flux, the persistent translocation of the MT polymer lattice from the spindle equator towards the pole (Figure 1B) [13]. In one model, based on studies carried out in fly embryos, kinesin-5 motors persistently crosslink and slide apart antiparallel ipMTs. Prior to anaphase B, the minus ends of these sliding ipMTs are depolymerized at the poles, creating a force balance within ipMTs that flux poleward as they maintain pole–pole spacing, but at the onset of anaphase B, depolymerization at the poles stops, allowing the sliding ipMTs to drive pole–pole separation and elongate the spindle [14] (Figure 1D).

It is somewhat surprising to find such distinct, indeed opposite, roles for kinesin-5, acting as a brake on ipMT sliding in the spindles of C. elegans embryos versus actively pushing apart ipMTs in spindles of other systems, such as Drosophila embryos [4,15]. Furthermore, in the latter case, the minus-end-directed kinesin-14 motor appears to be the brake that limits the rate of kinesin-5-driven ipMT sliding, producing a force-balance that is essential to maintain the bipolar prometaphase spindle (Figure 1C), which collapases following loss of kinesin-5 function [15]. The phenotypes resulting from the loss of the braking action of kinesin-5 and kinesin-14 in the two systems are quite similar, being characterized by an increase in the onset, rate and extent of spindle pole separation, but ultimately the spindle seems able to ‘correct’ itself so that no severe defects in mitosis and chromosome segregation result [4,15]. However, the plots of spindle-pole dynamics in C. elegans provide clues about the possible contribution of the kinesin-5 brake to spindle function; in pre-anaphase spindles, the early and fast separation of the spindle poles produces a ‘pseudo-metaphase’ steady-state length, which may result from initially unloaded cortical pulling motors stalling and/or detaching if they are not restrained by BMK-1 [4].

What biochemical and biophysical properties of kinesin-5 might allow it to perform these mitotic functions? Recent studies (reviewed in [2]) suggest that kinesin-5 moves in a modestly processive fashion, taking short runs along the MT polymer lattice but readily detaching from sliding MTs rather than stalling in response to competing forces [16]. This may be an adaptation that allows it to act within ensembles in the spindle, by preventing it from acting as a molecular ‘monkey wrench’ which, if it remained bound to MTs in a stalled state, could non-specifically block and dampen MT sliding driven by other motors [16]. In addition, kinesin-5’s speed of movement along its MT track was observed to be relatively insensitive to ‘assisting’ loads as high as 4 pN directed toward the MT plus ends [16]. In the wild-type C. elegans embryo, this may allow kinesin-5 motors to remain engaged as the ipMTs are slid apart by fast cortical motors, producing steady, linear pole–pole separation at rates twice the unloaded rate of BMK-1 motility alone (see Table 1 in [4], where the initial rate of wild-type pole–pole separation, 0.1 μm/s, is twice the rate of some kinesin-5s [8]).

Motility assays might also illuminate how kinesin-5 motors are able to function as active sliding motors that push apart spindle poles in some situations but as brakes that restrain MT–MT sliding and pole–pole separation in others. Recently, the predicted functional antagonism between purified kinesin-5 and kinesin-14 (Figure 1C) was tested in competitive motility assays using varying molar fractions of the two motors, which did indeed behave as mutual ‘brakes’ to slow one another down, producing a stable ‘steady-state’ balance point within a narrow range of molar ratios [10]. Quantitative modeling suggested that the braking effect is not due to the generation of active, opposite polarity ‘power strokes’ typical of...
stalled motors, but instead reflects weaker braking forces due to passive ‘protein friction’ [10]. The resulting drag is plausibly exerted by cycles of MT attachment–detachment by kinesin-5 slowing down active sliding by kinesin-14, and vice versa [10]. Although this requires further testing, perhaps similar protein friction could allow kinesin-5 to slow down the rate of MTs being slid in the same direction as the active kinesin-5 power stroke. However, it is also possible that the slow ATP hydrolysis cycle of kinesin-5 is needed to limit the rate of MT sliding driven by faster, ‘assisting’ motors [4,7]. Perhaps a study of the rate of spindle elongation in C. elegans embryos containing mutant kinesin-5 motors with impaired ATP hydrolysis [17] might be useful for discriminating between these possibilities. Biochemical studies further suggest that sites located outside the kinesin-5 motor domains could exert protein friction, since a headless kinesin-5 motor was observed to crosslink MTs into bundles, and by doing so in the central spindle it might be expected to exert drag and limit the rate of MT sliding driven by other motors [10], but this idea has not been tested yet. In some systems, on the other hand, additional factors may be used to exert protein friction and limit the rate of motor-driven sliding of MTs, including, for example, the microtubule-associated protein, Ase1p, which, like kinesin-5, associates with antiparallel ipMTs in the mitotic spindle midzone [18].

The hypothesis that kinesin-5 serves as a brake on spindle-pole separation in some spindles [4] but drives spindle elongation in others [3,15] predicts that the loss of kinesin-5 function should enhance or restrict the rate and/or extent of centrosome separation and spindle elongation, respectively, as observed. In Drosophila cultured S2 cells, however, spindles depleted of kinesin-5 either collapse to produce monoaasters (plausibly during prometaphase as in embryos) or form bipolar metaphase structures in which no significant length defects were detected, leading to suggestions that here MT sliding motors contribute little to steady-state metaphase spindle length [19] (although effects on rates of attainment of steady state length cannot be ruled out). If this interpretation proves correct, it implies that significant diversity exists in the functions of kinesin-5, even in spindles within the same organism [14,19]. In fact, as noted above, it has long been proposed that kinesin-5 also has functions that go beyond its roles in centrosome separation, including the powering of poleward flux (Figure 1B) [19], as well as forming, stabilizing and ‘sorting’ parallel and antiparallel microtubules and organizing the poles of acentrosomal spindles (Figure 1E) [9,20]. Thus, there is diversity in the functional deployment of this key mitotic motor in different systems, although it is plausible that all of its roles are a consequence of its

---

**Figure 1. Diverse mitotic functions of kinesin-5 motors.**

(A) In C. elegans embryos, kinesin-5 on the interzone (blue) acts as a brake to oppose outward forces generated by cortical dynein (violet red). (B) In Xenopus extracts, kinesin-5 slides MTs poleward, contributing to poleward flux (curly blue arrow). (C,D) In Drosophila embryos and yeast, kinesin-5 maintains spindle bipolarity and drives anaphase B spindle elongation. In (C), a force balance between kinesin-5 and kinesin-14 (red) contributes to the maintenance of prometaphase pole-pole spacing. A model for anaphase B in fly embryos is shown in (D): persistent outward ipMT sliding by kinesin-5 is balanced by depolymerization at spindle poles to produce flux in preanaphase B spindles (upper) and the suppression of depolymerization allows ipMT sliding to drive anaphase B pole–pole separation (lower). (E) In anastral spindles, kinesin-5 crosslinks MTs nucleated from chromosomes into bundles and slides them apart to ‘sort’ them so their minus ends point to the poles for subsequent focusing. In each panel, MTs are shown as black lines with plus ends shaded dark and minus ends shaded light; black dots are polymerizing or depolymerizing tubulin subunits; the cortex is green; chromosomes are violet; centrosomes are black. The direction of movement of motors along MTs is shown in color-coded dashed arrows; direction of force exerted on the pole by the motors is shown in color-coded dashed arrows; curly blue arrows indicate poleward flux.
Kin Recognition: Knowing Who’s Boss in Wasp Colonies

Paper wasps recognise the dominant individual in their colony and surrender reproduction to this alpha individual. Contrary to expectations her dominance status is not signalled by a chemical indicator of fertility.

Duncan E. Jackson

Many behavioural interactions among organisms are determined by genetic relationships and correspond to the prediction of kin selection theory: that close relatives treat each other better[1]. Individuals are more likely to behave altruistically towards close relatives because they share a high proportion of their genes[2]. In nature we find that altruism is widespread in groups and inbreeding is rare, both of which indicate that the ability to recognise kin is ubiquitous. Understanding the proximate mechanisms underlying kin discrimination is an area of intense research. A new study by Dapporto et al.[3], reported recently in Current Biology, shows that the main egg-producer in a paper wasp colony is differentiated by a chemical signal which denotes her dominant status, and that this


References