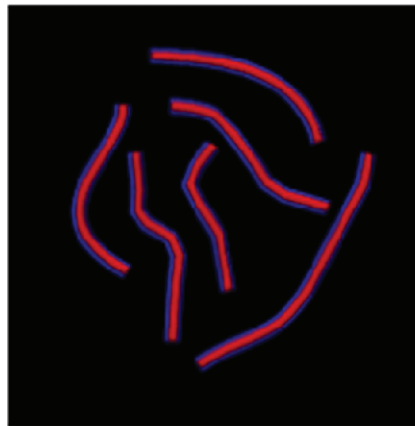
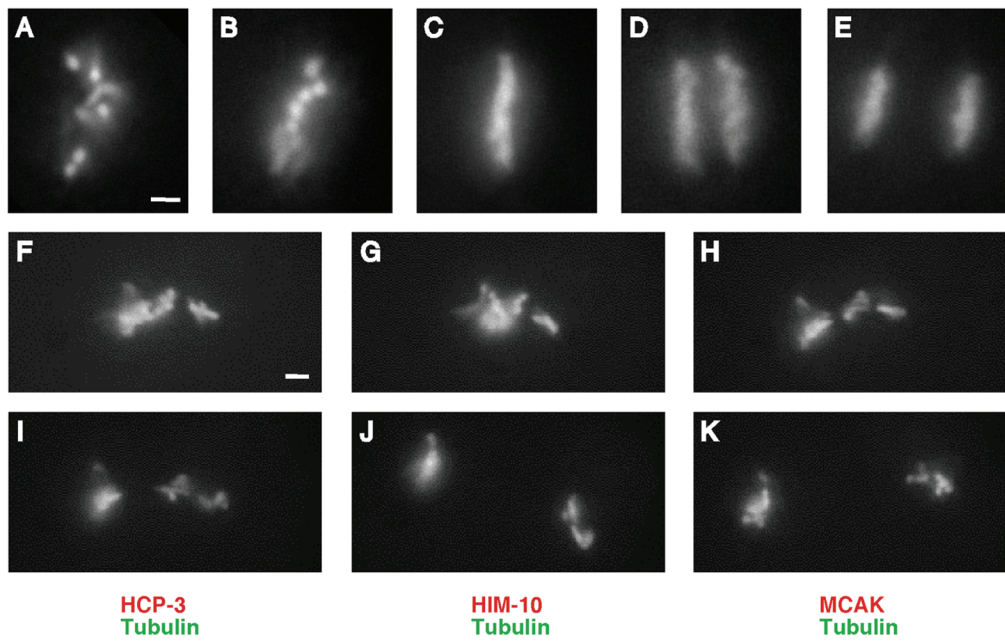


Perpendicular perspective



Centrosomal perspective

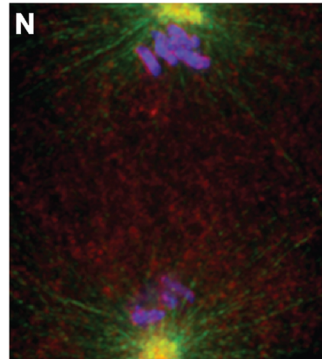
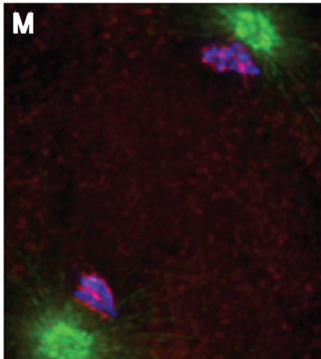
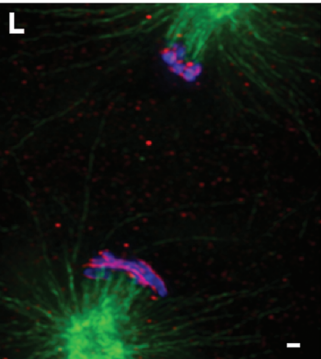
Stear and Roth, Supplemental Figure 2



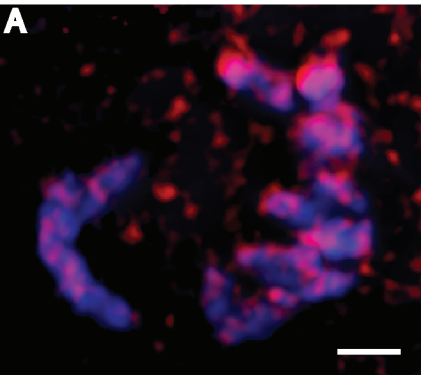
HCP-3
Tubulin

HIM-10
Tubulin

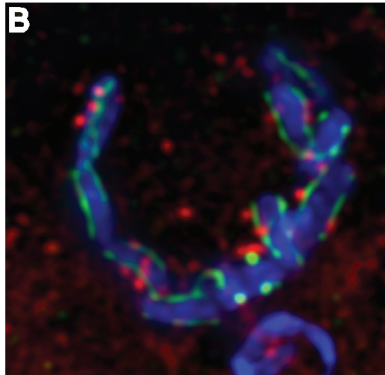
MCAK
Tubulin



MCAK



HCP-3
HCP-1



Supplemental Figure 1: Schematic representations of the metaphase plate in *C. elegans*

(Left panel) This image displays the metaphase plate viewed from a perspective perpendicular to the spindle axis. The centrosomes lie to the left and right of the metaphase plate. The individual chromosomes cannot be resolved and appear as a solid bar of DNA (blue). The centromeres and kinetochores (red), which lie on the poleward faces of the metaphase plate, sandwich the DNA between them. (Right panel) This image displays the metaphase plate viewed parallel to the spindle axis. From this centrosomal perspective, the poles lie above and below the plane of the screen. It is possible to visualize individual metaphase chromosomes (blue), and detect a single line of centromere staining (red) coincident with each chromosome. Although only a single sister centromere on each chromosome is visible from this perspective, the other sister centromere lies below the plane of the screen.

Supplemental Figure 2: *hcp1/hcp2(RNAi)* embryos display defects in congression and spindle attachment.

In the histone H2B::GFP background, wild-type (**a-e**) and *hcp1/hcp2(RNAi)* (**f-k**) chromosomes were visualized in one cell embryos shortly after pronuclear fusion. We filmed the progression of these embryos from prometaphase to anaphase. The interval between each frame is 25 sec. Wild-type chromosomes congress to the center of the cell (**a-b**), assemble into a metaphase plate (**c**), and execute a clean separation of sister chromatids at anaphase (**e-f**). In contrast, *hcp1/hcp2(RNAi)* chromosomes fail to align into a metaphase plate (**f-h**), and ultimately segregate in a highly disorganized fashion into the daughter cells (**i-k**). *hcp1/hcp2(RNAi)* embryos were fixed and stained with DAPI (blue), α -tubulin (green), and either α -HCP-3 (**l**), α -HIM-10 (**m**), or α -MCAK (**n**) (red). In these images, collected from an one cell embryo, we

show that the chromosomes in a *hcp1/hcp2(RNAi)* background represent pairs of sister chromatids that are attached to microtubules from only one pole (**l**). Furthermore, these chromosomes are able to direct assembly of other late kinetochore proteins such as HIM-10/CeNUF2 (**m**) and MCAK (**n**). Bar equals 1 μm .

Supplemental Figure 3: *him-10(RNAi)* chromosomes are able recruit to recruit a subset of kinetochore components.

him-10(RNAi) embryos were stained with DAPI (blue) and antibodies against MCAK (**a**) or HCP-1 (red) and HCP-3/CeCENP-A (green) (**b**). Examining metaphase plates in four cell embryo demonstrates that the kinetochore protein MCAK (**a**, red) is recruited normally onto chromosomes in this background. However, the levels of HCP-1 (**b**, red) are dramatically reduced on *him-10(RNAi)* chromosomes. Bar equals 1 μm .