**Featured Publication Note**
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# Live cell imaging of *Drosophila* S2 anaphase cells using the Ultra*VIEW* VoX

Dr. Karsenti and colleagues are interested in understanding the fundamental principles involved in microtubule-dependent morphogenetic events in the cell. RanGTP is locally produced inside the nucleus in interphase and in a gradient surrounding the chromosomes during mitosis. The production of RanGTP around chromosomes induces spindle assembly through the activation of nuclear localization signal (NLS)-containing factors. All NLS proteins are potentially involved in spindle assembly or other chromosome-dependent processes. In this study, the role of one of these proteins, ISWI, was investigated.

As part of this research, the authors examined the role of ISWI *in vivo*, by performing RNAi against ISWI in *Drosophila* S2 cells. To directly address whether ISWI is required in anaphase, the researchers used live-cell imaging of S2 cells stably expressing GFP–α-tubulin and mCherry-centromere identifier (CID, a kinetochore marker) at 4 d after treatment with dsRNA (Figure). Live cell imaging was performed using the Ultra*VIEW* VoX and 4D datasets covering the entire cell volume were collected every 1.5 min with 1 µm z steps at 25°C. Using the Ultra*VIEW* VoX, they demonstrated that ISWI is essential for *Drosophila* cells to grow and progress from G2/M phases, and is required for anaphase microtubule stability and chromosome segregation.

The conclusion to the study was that ISWI functions as a RanGTP-dependent microtubule-associated protein which is required for microtubule stability in anaphase and is essential for chromosome segregation. This anaphase function involves a RanGTP-dependent stabilization of spindle microtubules.

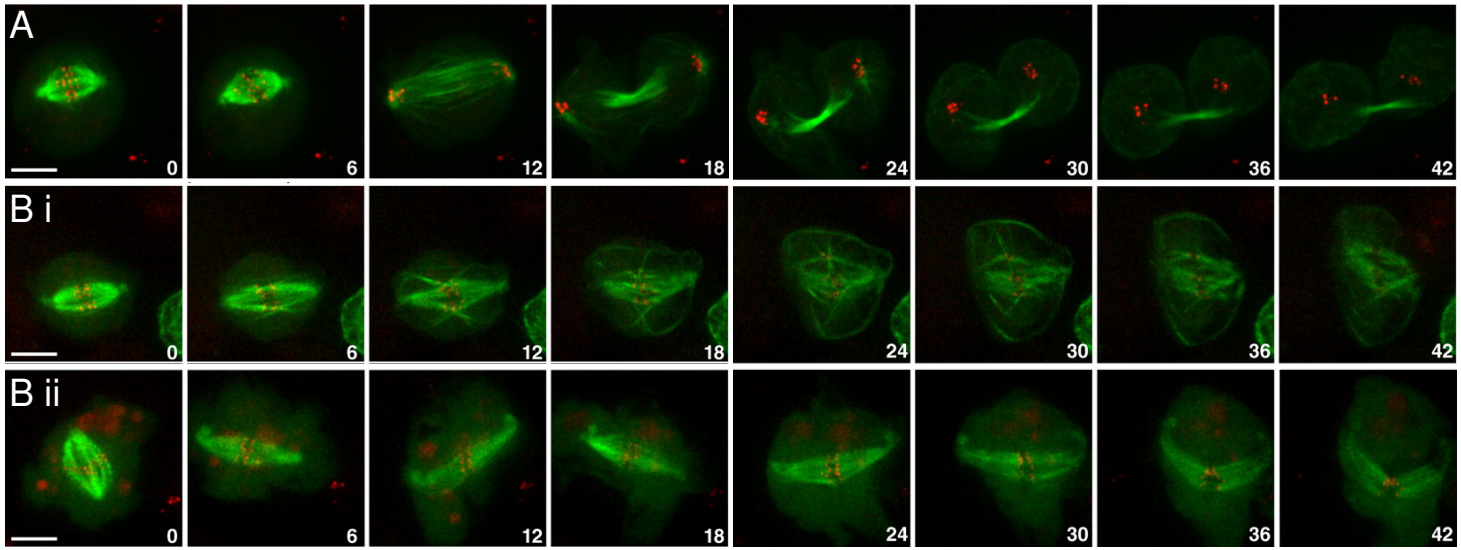


**Figure: Live cell imaging of *Drosophila* S2 cells showing the requirement for ISWI in anaphase.** After 4 d of dsRNA treatment, *Drosophila* Schneider S2 cells stably expressing GFP–α-tubulin (green) and CID-mCherry (red) were analyzed for spindle and chromosome dynamics during anaphase. Z stacks of the acquired images were projected with maximum intensity, and stills of the time-lapse analysis are presented. Times in minutes from the start of the video are indicated in each frame. (A) Control S2 cell treated identically but without dsRNA (B) i) and ii) ISWI RNAi. Bars, 5 µm.

**The videos can be viewed at: http://jcb.rupress.org/cgi/content/full/jcb.200906020/DC1**

**Image adapted from Yokoyama, H, Rybina S, Santarella-Mellwig R, Mattaj I W and Karsenti E (2009). ISWI is a RanGTP-dependent MAP required for chromosome segregation. J. Cell Biol.; 187: 813 - 829. © Yokoyama *et al.*, 2009. Originally published in The Journal of Cell Biology. doi: 10.1083/jcb.200906020.**