Supplementary material

Supplementary movies

Real-time records of stable cyclin expression in embryos also expressing the histone-GFP transgene. Stable cyclin expression was induced with a 30 minute heat shock at 150–195 minutes AED. All QuickTime movies were made using time lapse imaging with the DeltaVision microscopy system. Images were taken with a 20×, 40×, or 100× lens, as indicated. The clock in the lower right corner rotates once per minute of elapsed time.

Movie 1. CYC-A^S metaphase arrest. 100× - Bin 2; 22 min.Three cells are shown arrested in metaphase for the duration of the movie.

Movie 2. Escape from CYC-A^S metaphase arrest. 20×; 26.5 min.
Several cells underwent an aberrant anaphase after a prolonged block in metaphase. Three cells of particular interest are marked. Cell 1: after at least 7.5 minutes in metaphase, slow movement of the chromosomes towards the poles can be seen. Chromosome disjunction occurred over acouple of minutes, instead of a few seconds. During this time, the separating chromosome masses stretched out along the mitotic spindle. Eventually, full chromosome separation was accomplished, chromosome decondensation occurred, and mitosis appeared to be completed. Cell 2: After at least 9 minutes in metaphase, chromosome separation started slowly, and chromosomes were stretched along the spindle, as with cell 1. However, in this case, the chromosomes never fully separated, and the chromosome masses decondensed to give a "dumbbell" configuration. Cell 3: After at least 17 minutes in metaphase, chromosome separation started slowly, as seen in cells 1 and 2. Over the final 9 minutes of the record, the chromosomes stretched out along the spindle and were in this configuration as the movie ended. Six cells were examined carefully, and all showed evidence of prolonged chromosome disjunction. Additional anaphase cells in the movie were not considered because the limited record did not include documentation of a metaphase arrest.
Movie 3. Initial anaphase movements in cells expressing CYC-B^S. 100×; 16 min.
Detail of three cells that pass the metaphase/anaphase transition and initiate chromosome movements toward the poles. After an initial period of separation, the chromosomes stopped their poleward movements and collapsed back toward the midline. The cells then entered into the dynamic terminal arrest state.
Movie 4. CYC-B^S dynamic arrest. 40×; 20 min.
The majority of the cells that are shown displayed the CYC-B^S terminalarrest phenotype, with individual chromosomes moving along the spindle.Three cells can be seen that came into the plane of focus while inmetaphase and then underwent initial anaphase movements before collapsinginto the terminal arrest state. The movements of individual chromosomescan be followed in some of the cells. The slow movementsnot led by the kinetochore are particulary obvious in this record (seeFigure 4).
Movie 5. Detail of CYC-B^S terminal arrest. 40×, enlarged 2×; 20 min.
One cell (lower right-hand corner at the beginning of the movie) from Movie 4 is focused on, and individual chromosome movements along the spindle are readily apparent. Chromosomes can be seen to move rapidly from one side of the spindle midline to the other.
Movie 6. CYC-$B3^S$ arrest. 20×; 25 min.
Cells underwent anaphase normally and arrested with condensed chromosomesat the poles. In some cases, individual chromosome movements along the spindle can be seen as well as rotation of the spindle.Three examples of cells showing this behavior are marked with arrows.
Movie 7. CYC-$B3^S$ does not block cytokinetic furrow formation. 100×; 11 min.
A cell near the center of the screen underwent anaphase and thenarrested with condensed chromosomes at the poles. Background fluorescence from the histone-GFP protein allows visualization of the pinching of the plasma membrane. Pinching can be seen with the chromosomes still condensed. Pixelated spots on the screen are an artifact of data collection.
Movie 8. CYC-$B3^S$ does not block cytokinetic furrow formation. 40×, Bin 2, enlarged 2×; 11 min.
Several cells are shown in which pinching of the plasma membrane canbe clearly seen due to background fluorescence from the histone-GFP protein. Chromosomes can be seen to remain condensed in these cells.