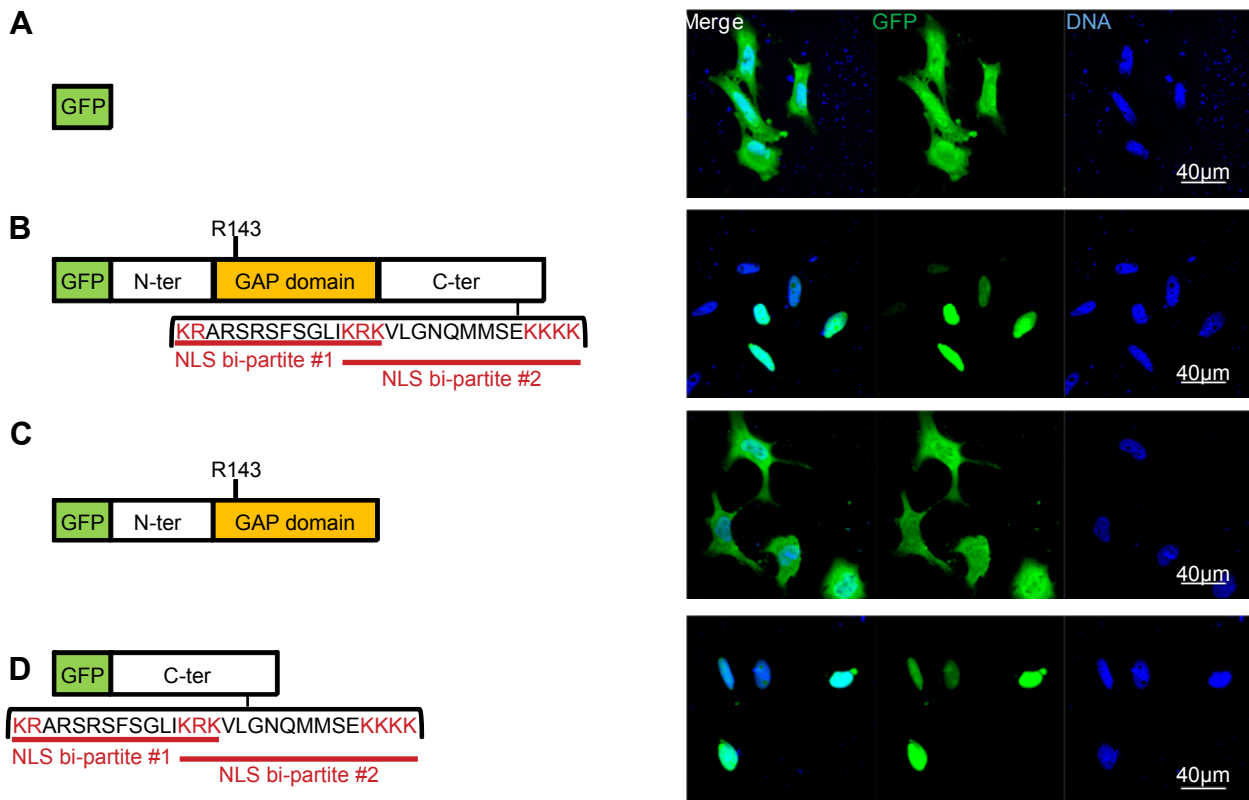
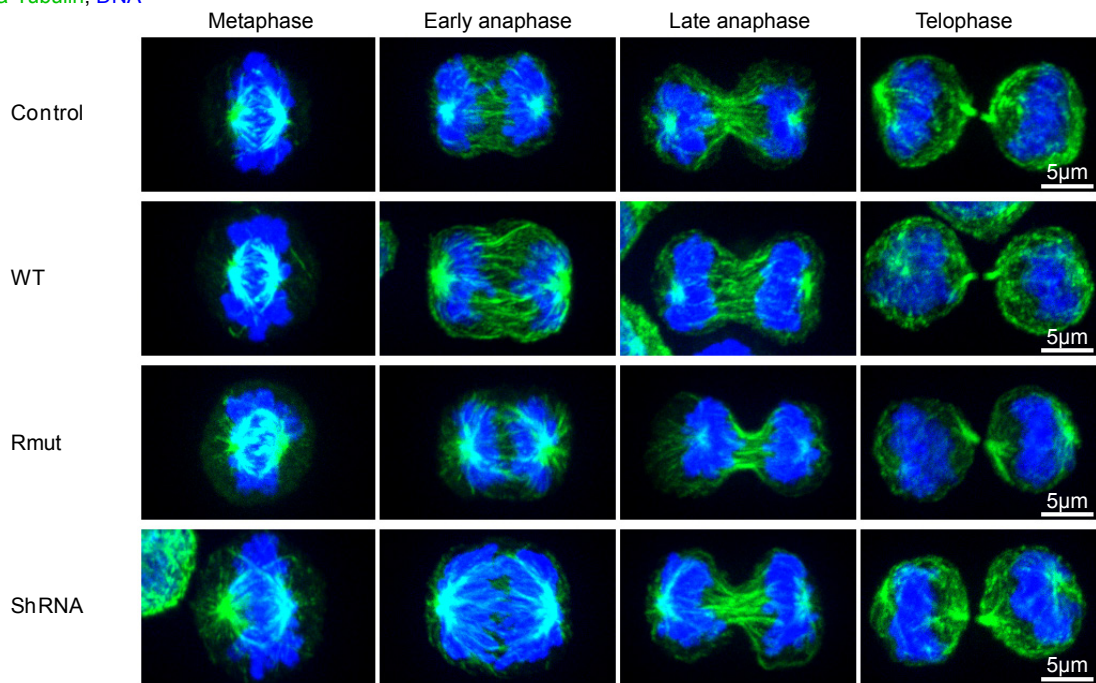


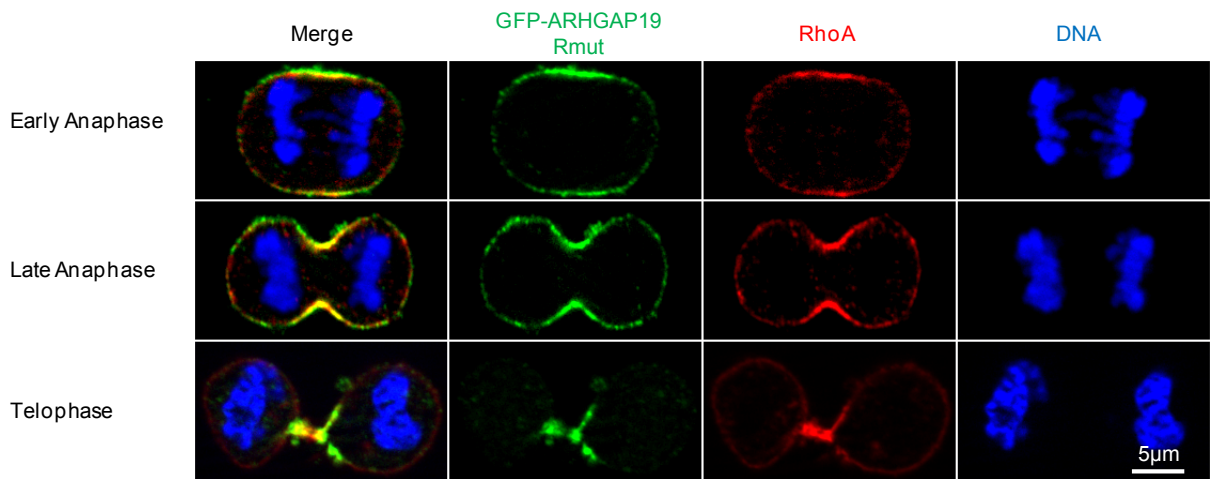
**Fig. S2: Downregulation of ARHGAP19 levels in Jurkat lymphocytes through ShRNA induces excessive cell shape changes in early mitosis as well as chromosome segregation defects in anaphase.** Jurkat T cells were stably transfected with plasmids bearing ShRNA against ARHGAP19 (Sh113 or Sh117). **(A)** Western blot analysis on the cell lysates was performed with our anti-ARHGAP19 antibody. Equal loading was confirmed by reprobing the blot with an anti-Hsc70 antibody. **(B, C)** Mitosis of a control Jurkat T cell **(B)** and of an ARHGAP-19-depleted Jurkat T cell **(C)**, monitored by time-lapse microscopy. The red color corresponds to the fluorescence emitted by the DNA labeling, Syto 59 dye. Time "0 min" was set on the last frame displaying the cell in the metaphase stage. White arrow points out lagging DNA. **(D)** ARHGAP19 silencing induces precocious cell elongation and cleavage furrow ingression in Jurkat cells.



**Fig. S3: Nuclear localization of ARHGAP19 depends on its C-terminal region.** HeLa cells were transiently transfected with plasmids coding for GFP (**A**), GFP-ARHGAP19 full length (**B**), a truncated form of ARHGAP19 devoid of its C-terminal region (GFP-ARHGAP19- Cter) (**C**), or GFP fused to the C-terminal region of ARHGAP19 (GFP+Cter) (**D**). Schemes on the left part of the figure illustrate the primary structure of these proteins and the respective localizations of the GAP domain and of the two potential overlapping bi-partite nuclear localization signals (NLS). ARHGAP19 truncated of its C-terminal region is no longer enriched in the cell nucleus (**C**). Reciprocally, fusion with the C-terminal region of ARHGAP19 is sufficient to target GFP to the cell nucleus (**D**).



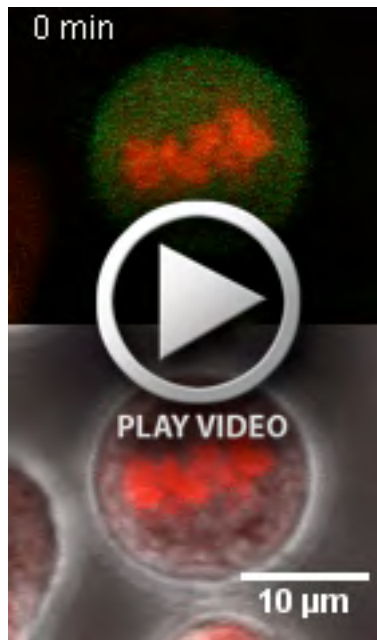
**Fig. S4: Manipulation of ARHGAP19 levels or activity does not induce noticeable defects in microtubule organization in dividing lymphocytes.** Kit225 lymphocytes (control, expressing the WT or Rmut forms of ARHGAP19, or depleted in ARHGAP19) were fixed using PFA. Cells were labeled for DNA (Hoechst, blue) and alpha-Tubulin (green). Representative pictures of cells in metaphase, early anaphase, mid-anaphase or telophase are shown.



**Fig. S5: GFP-ARHGAP19 Rmut and RhoA co-localize in anaphase.** GFP-ARHGAP19 Rmut-expressing Kit225 lymphocytes were fixed using TCA and labeled for DNA (Hoechst, blue) and RhoA (red). GFP signals are displayed in green color. Representative pictures of mitotic cells are shown.



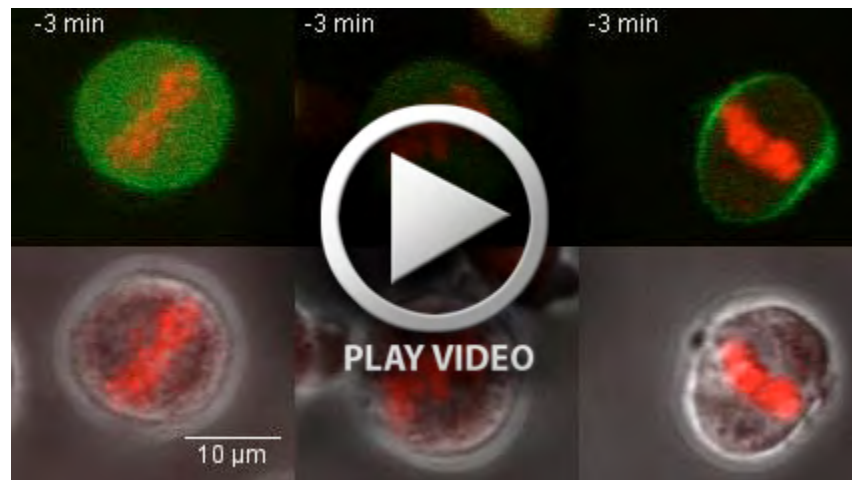
**Movie 1. Control Kit225 lymphocyte undergoing mitosis.** Mitosis progression of control Kit225 lymphocytes was followed by time lapse confocal microscopy. The red color corresponds to fluorescence emitted by the DNA labeling, Syto 59 dye. Frames were taken every minute. Selected video stills are shown in Fig. 3.



**Movie 2. Mitosis of a GFP-ARHGAP19 WT-expressing Kit225 lymphocyte, with delayed cell elongation and cleavage furrow ingression as compared to control cells.** Mitosis progression of Kit225 lymphocytes expressing GFP-ARHGAP19 WT (green) was followed by time lapse confocal microscopy. The red color corresponds to fluorescence emitted by the DNA labeling, Syto 59 dye. Frames were taken every minute. Selected video stills are shown in Fig. 3.



**Movie 3. Examples of dividing ARHGAP19-depleted Kit225 lymphocytes:** Mitosis progression of Kit225 lymphocytes transfected with ShRNA constructs targeting ARHGAP19 was followed by time lapse confocal microscopy. The red color corresponds to fluorescence emitted by the DNA labeling, Syto 59 dye. Frames were taken every minute. Selected video stills are shown in Fig. 3. The left panel shows an ARHGAP19-depleted lymphocyte that divides without major phenotype except for excessive blebbing. The middle panel illustrates the excessive cell elongation and early furrow ingression induced by silencing of *ARHGAP19*. Moderate lagging of DNA is detectable. The right panel shows a dividing ARHGAP19-depleted lymphocyte with moderate cell shape phenotype but major defects in chromosome segregation.



**Movie 4. Examples of dividing lymphocytes expressing the R143A dominant negative form of GFP-ARHGAP19.** Mitosis progression of Kit225 lymphocytes expressing GFP-ARHGAP19 Rmut (green) was followed by time lapse confocal microscopy. The red color corresponds to fluorescence emitted by the DNA labeling, Syto 59 dye. Frames were taken every minute. Selected video stills are shown in Fig. 3. The left panel shows a GFP-ARHGAP19 Rmut-expressing lymphocyte that divides without major phenotype except for excessive blebbing. The middle and right panels represent GFP-ARHGAP19 Rmut-expressing lymphocytes with moderate or severe chromosome segregation defects, respectively. On the right panel, excessive cell elongation long before anaphase onset is also observable.