SUPPLEMENTARY MATERIALS

Movie S1. Cytokinesis defects in *Lis1* mutant MEFs.
Time-lapse live cell imaging of mitotic cell division from *Lis1* mutant MEFs (*CreER^{TM}; Lis1$^{hc/hc}$* +TM$^{12\ h}$) treated with 4-hydroxy tamoxifen (TM). H2B-GFP and mCherry-α-Tubulin labeled fluorescence signals were acquired with a 1 minute interval by Nikon Ti epifluorescence microscope. During cytokinesis of *Lis1*-deficienct MEFs, severe cytokinesis defects were observed such as vigorous cell shape oscillation and spindle rocking.

Movie S2. Formation of binucleated daughter cells by cytokinesis failure in *Lis1* mutant MEFs.
Time-lapse live cell imaging of mitotic cell division from *Lis1* mutant MEFs (*CreER^{TM}; Lis1$^{hc/hc}$* +TM$^{12\ h}$) treated with 4-hydroxy TM. H2B-GFP and mCherry-α-Tubulin labeled fluorescence signals were acquired with a 30 second interval by Nikon Ti epifluorescence microscope. The *Lis1*-deficienct MEFs underwent abnormal cytokinesis and resulted in formation of binucleated daughter cells.

Movie S3. Myosin II localization during cytokinesis of wild-type (WT) MEFs.
Time-lapse live cell imaging of mitotic cell division from wild-type (WT) MEFs. Myosin regulatory light chain 1 (MRLC1)-GFP and H2B-tdTomato labeled fluorescence signals were acquired with a 30 second interval by Nikon Ti spinning disk confocal microscope. In normal cytokinesis, MRLC1 was recruited to the equatorial cortex and formed the cleavage furrow.

Movie S4. Abnormal Myosin II movements during cytokinesis of *Lis1* mutant MEFs.
Time-lapse live cell imaging of mitotic cell division from *Lis1* mutant MEFs (*CreER^{TM}; Lis1$^{hc/hc}$* +TM$^{24\ h}$). Myosin regulatory light chain 1 (MRLC1)-GFP and H2B-tdTomato labeled fluorescence signals were acquired with a 30 second interval by Nikon Ti spinning disk confocal microscope. In *Lis1* mutant MEFs, MRLC1 was first recruited to the equatorial cortex. However, we found failure to properly restrict the cleavage furrow at the equatorial cortex.

Movie S5. Uncoupling between chromosome segregation and cytokinesis in *Lis1* mutant MEFs.
Time-lapse live cell imaging of mitotic cell division from *Lis1* mutant MEFs (*CreER^{TM}; Lis1$^{hc/hc}$* +TM$^{24\ h}$). Myosin regulatory light chain 1 (MRLC1)-GFP and H2B-tdTomato labeled fluorescence signals were acquired with a 30 second interval by Nikon Ti spinning disk confocal microscope. We observed frequent uncoupling between chromosome segregation and cytokinesis in *Lis1* mutant MEFs.

Movie S6. Septin localization during cytokinesis of wild-type (WT) MEFs.
Time-lapse live cell imaging of mitotic cell division from wild-type (WT) MEFs. Septin 6 (SEPT6)-GFP labeled fluorescence signals were acquired with a 30 second interval by Nikon Ti spinning disk confocal microscope. In normal cytokinesis, Septin-associated contractile ring complex was recruited to the equatorial cortex and formed the cleavage furrow.

Movie S7. Abnormal Septin localization during cytokinesis of *Lis1* mutant MEFs.
Time-lapse live cell imaging of mitotic cell division from *Lis1* mutant MEFs (*CreER^{TM}; Lis1$^{hc/hc}$* +TM$^{24\ h}$). Septin 6 (SEPT6)-GFP labeled fluorescence signals were acquired with a 30 second interval by Nikon Ti spinning disk confocal microscope. We observed SEPT signals at the equatorial cortex initially but then it regressed with vigorous cortical deformation and chromosome oscillation/rocking.