

## SUPPLEMENTARY FIGURE LEGENDS

**Supplementary Figure 1.** Analyses of co-localization of MEK1 and tubulin in different regions at various stages of the cell cycle. A, Average values of Pearson's correlation ( $R_p$ ) and Manders' co-localization coefficients (M) around the centrosomes and midbody in MCH603MEK1act cells at various stages of mitosis and cytokinesis. Int.: Interphase, Pro.: Prophase, Meta.: Metaphase, Ana.: Anaphase, Telo.: Telophase, Cyto.: Cytokinesis, CTSM: centrosome, MTOC: microtubule organization center. Bar indicates 5  $\mu$ M. B, Analysis of co-localization in different regions at interphase and prophase. C, Analysis of co-localization in different regions at metaphase. D, Analysis of co-localization in different regions at anaphase. E, Analysis of co-localization in different regions at telophase and cytokinesis.

i. Upper panels; Circled area was analyzed. Bottom panels; Scatter plot displays the co-localization coefficient in the circled area; pixels in channel 1 represent green color (MEK1) signal, pixels in channel 2 represent red color (tubulin) signal, pixels in channel 3 represent overlapping signals from green and red colors.

ii. Summary of co-localization coefficients from each region generated from each scatter plot. \*, Weighted co-localization coefficient from channels 1 or 2.

iii. Left; Signal intensities across different regions of an entire cell. Right; Graph reflecting a pattern of the co-localization of MEK with tubulin along the cross-section, MEK1 (green), tubulin (red), and nucleus (blue).

**Supplementary Figure 2.** MEK1<sup>act</sup> induces microtubule polymerization in vitro. *A*, Tubulin (50 µg, 0.3 mg/ml) was incubated with the indicated reagents in a plate reader spectrophotometer, and the kinetics of microtubule polymerization were monitored by reading at OD<sub>340</sub> for 1 h. *B*, Transmission electron microscopy of in vitro polymerized microtubules. Bars indicate 0.5 µM.

**Supplementary Figure 3.** Co-localization coefficient analyses of MCH603-GFP-MEK1act and MCH603-GFP-MEK1act-D. Less colocalization of MEK1act with tubulin was observed in the MCH603-GFP-MEK1act-D compared to MCH603-GFP-MEK1act. Left panel; Difference in MEK1act and tubulin signal intensities between the two cell lines. Right panel; Average co-localization coefficient values from 10 individually-analyzed cells.