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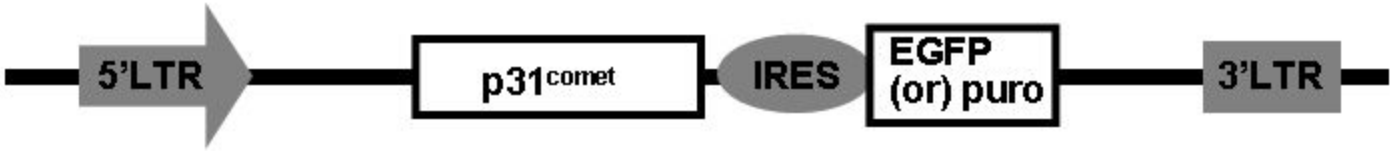

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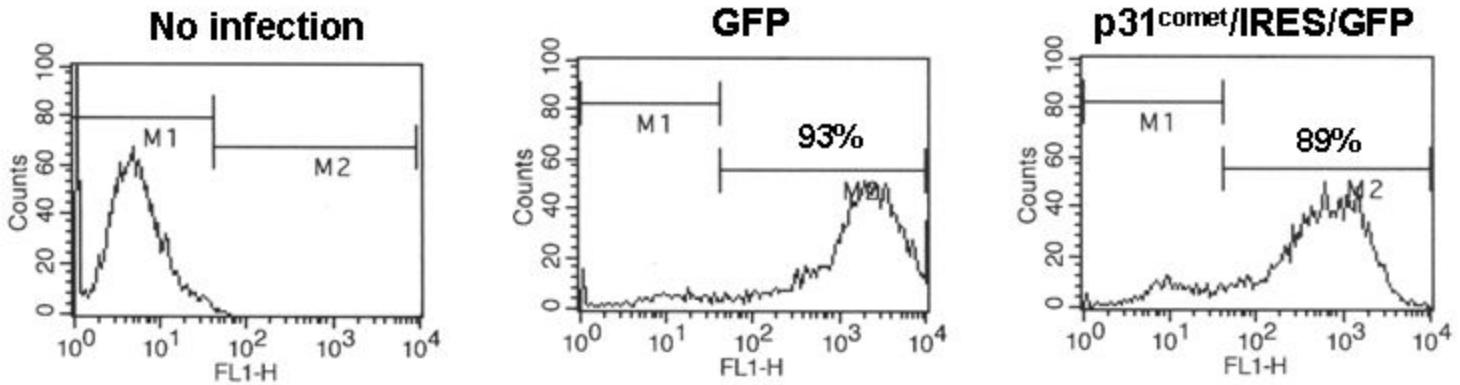
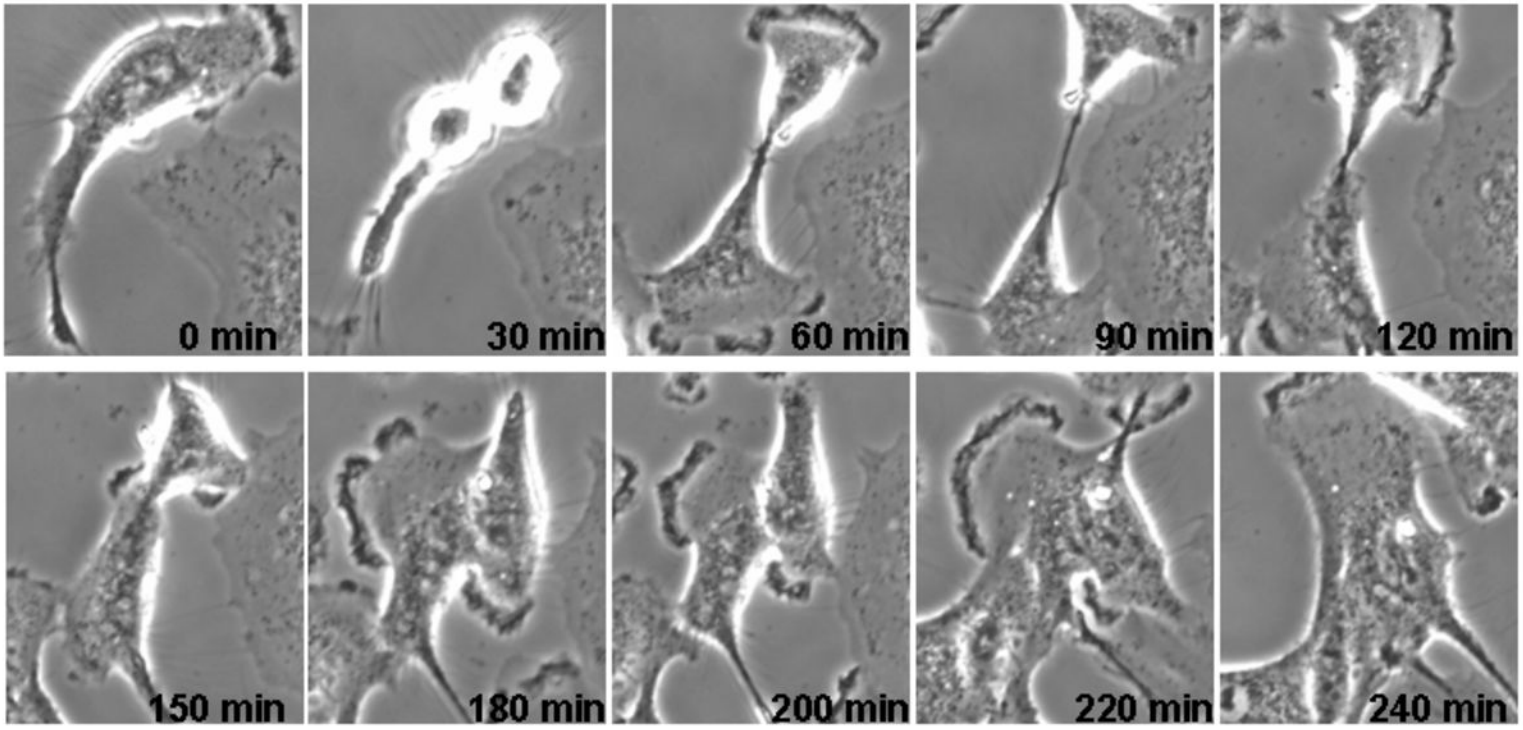


Supplementary Fig 1. Yun et al.



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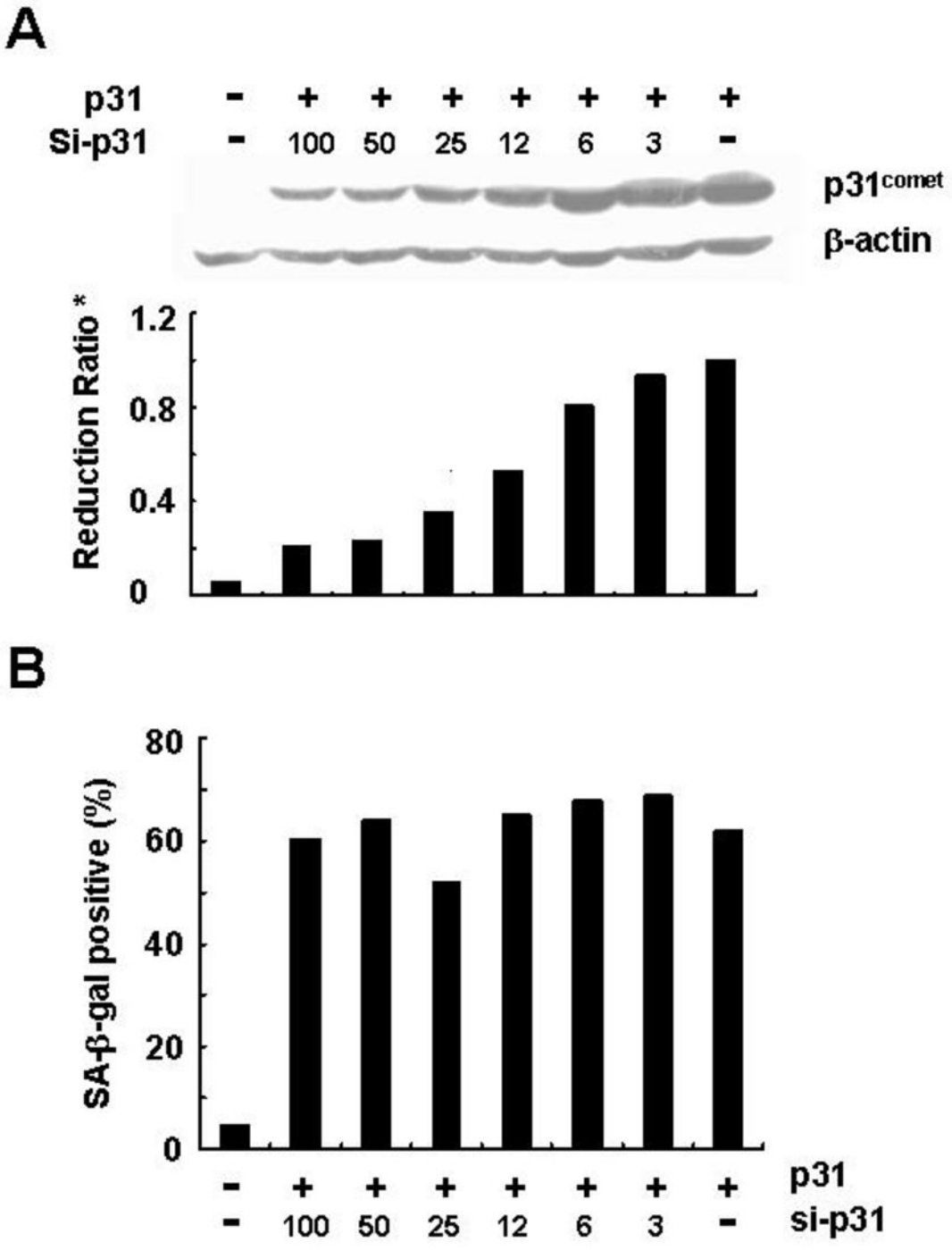
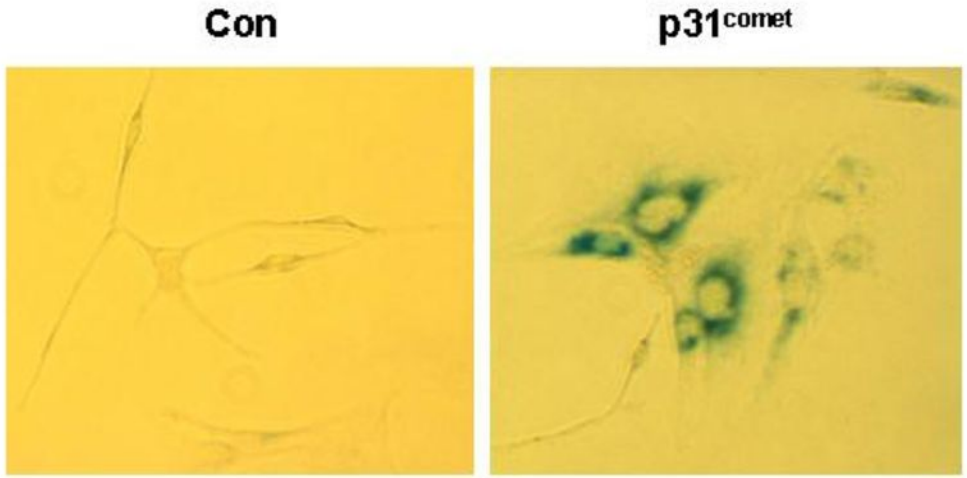


Supplementary Fig 3. Yun et al.

A



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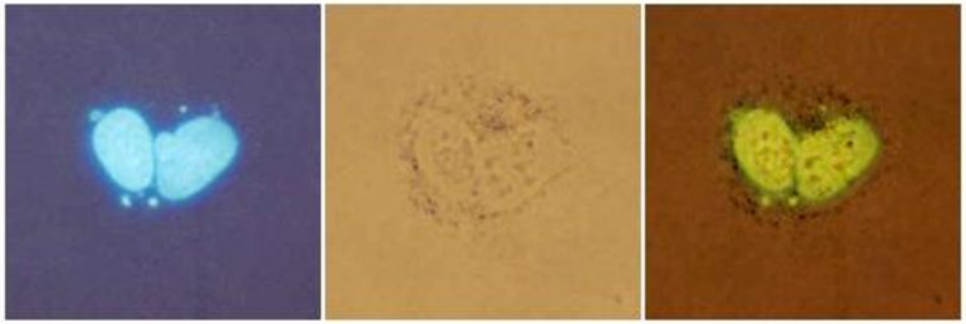
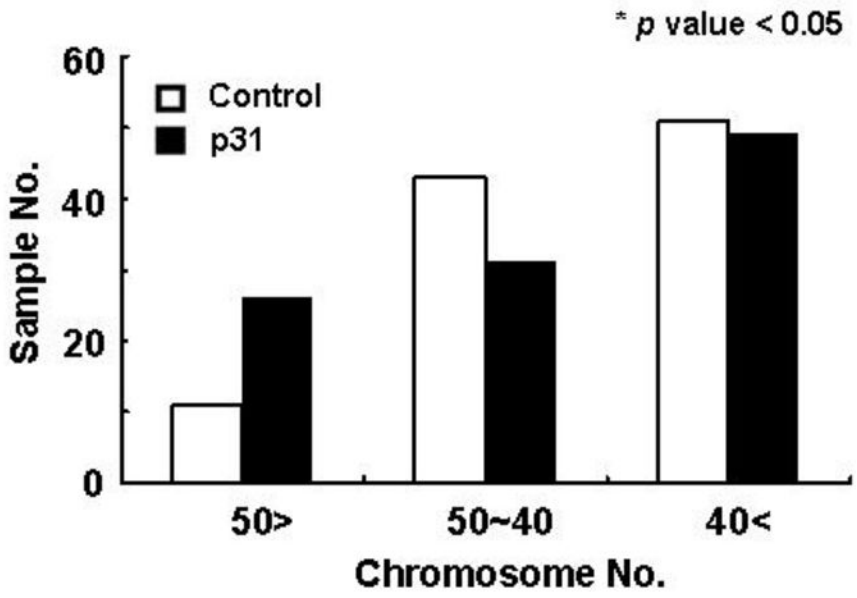


Supplementary Fig 4. Yun et al.

A



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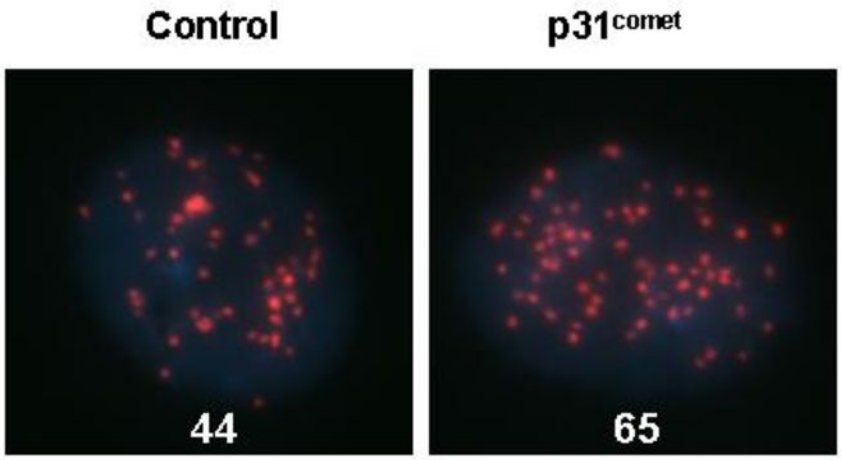
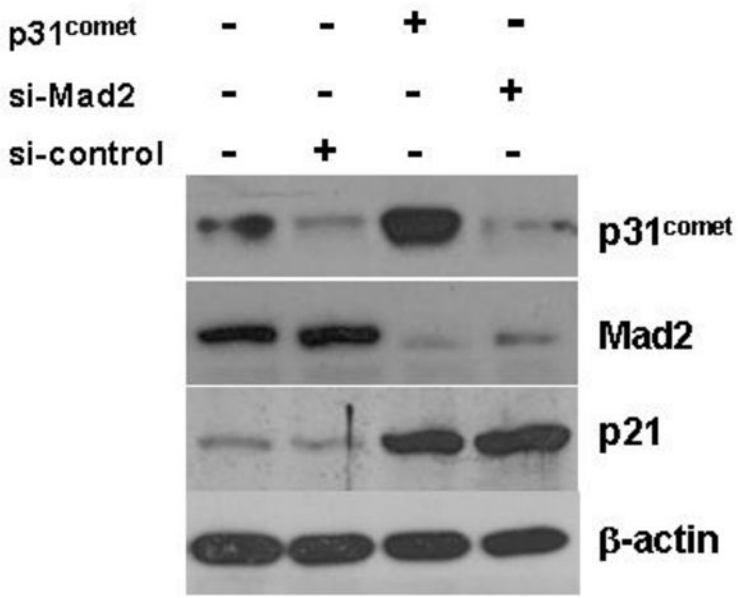


Supplementary Fig 5. Yun et al.



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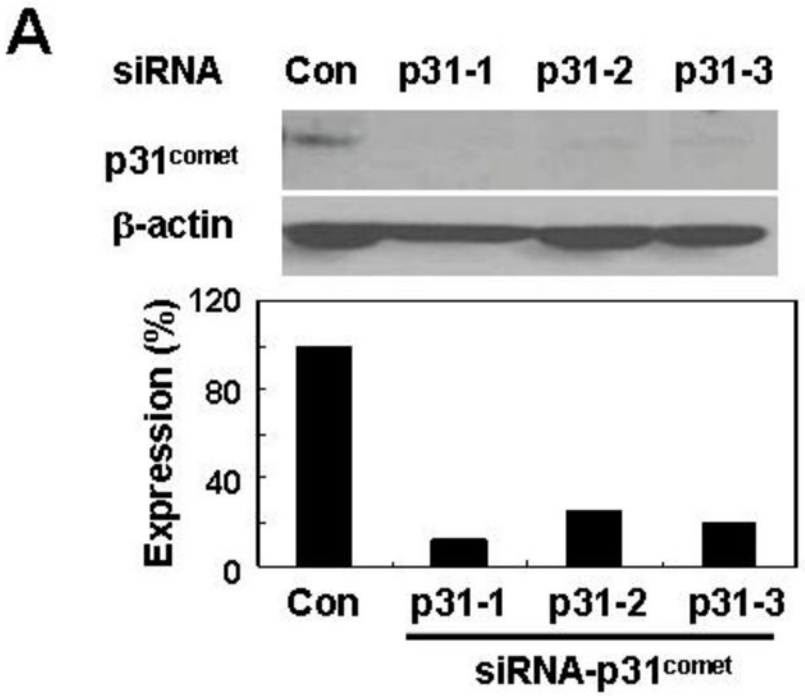

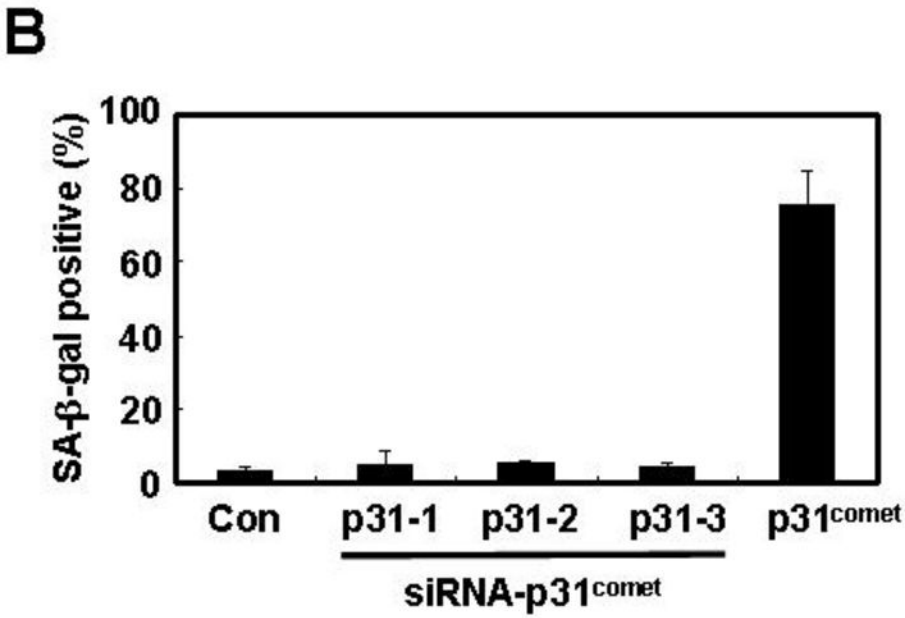

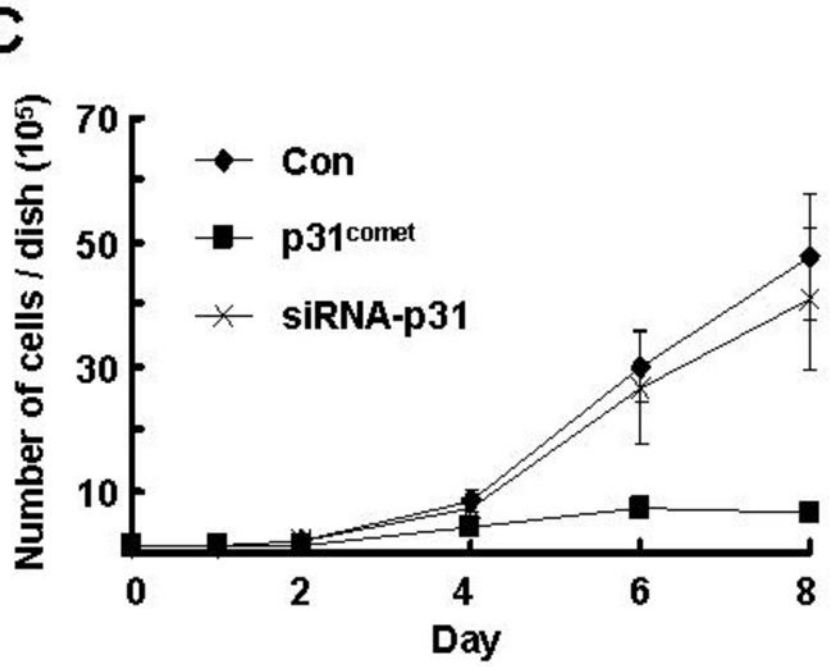

Supplementary Fig 7. Yun et al.

Supplementary figure legends

Supplementary Fig. 1. Overexpression of p31^{comet} in A549 cells. (A) The structure of bicistronic retroviral vector for ectopic expression of p31^{comet}. The open reading frame of human p31^{comet} is linked through an internal ribosome entry sequence (IRES) to the enhanced green fluorescence protein (EGFP) as a reporter or puromycin resistance gene (puro) as a selectable marker. (B) Analysis of virus infectivity for A549 cells using flow cytometry. Ten thousand cells were analyzed per experiment. The percentage of EGFP-positive cells is shown on top of the M2 picks. (M1, EGFP-negative region; M2, EGFP-positive region)

Supplementary Fig. 2. Time-lapse imaging of A549 cells overexpressing p31^{comet}. Frames from a low-magnification video microscopy sequence show a field of A549 cells progressing into mitosis over a period of 4 hours (time stamp in min: minute) after p31^{comet} infection. Note the prominent inhibition of cytokinesis and the eventual reforming of cell shape.

Supplementary Fig. 3. Senescence induction even in the reduced amounts of p31^{comet}. Four hours after retroviral infection of p31^{comet}, p31-1 siRNA for p31^{comet} (Supplementary Fig. 7) was added in a concentration-dependent manner. (A) The level of p31^{comet} was determined by Western blotting and (B) the percentage of senescent cells was determined by counting the SA-β-gal-positive cells. Si-p31 indicates the addition of p31^{comet} siRNA at the indicated concentration (pmol). In this experiment, the

duration of retroviral infection was minimized for 2 h. Similar results were obtained in cells receiving the siRNA, 12 h after retroviral infection with p31^{comet}. * The reduction ratio was calculated by the following method: (each p31 intensity/β-actin intensity)/(intensity of p31^{comet} without si-p31^{comet} treatment/its β-actin intensity).

Supplementary Fig. 4. p31^{comet} overexpression in Calu-1 is associated with cellular senescence. (A) Alteration of cell morphology and induction of SA-β-gal activity by p31^{comet}. Calu-1 cells were infected with p31^{comet} or control retrovirus. Images of the infected cells were obtained after 8 days. (B) Abnormal nuclear formation by p31^{comet}. At 8 days after infection with p31^{comet} retrovirus, cell nuclei were stained with DAPI. Bright field images for cell morphology and fluorescent images for DAPI staining were acquired using an inverted fluorescence microscope from the same field.

Supplementary Fig. 5. Centrosome imbalance in A549 cells overexpressing p31^{comet}. (A) Representative images of centrosomes at the interphase nucleus. The centrosome images of A549 cells were captured at interphase 3 days after infection with p31^{comet} or the control retrovirus (x400). (B) Chromosome numbers were determined by counting the fluorescent signals with centromere probe. FISH analysis for centromere detection was performed with biotin labeled pan-centromere probes according to manufacture's instructions (Chembio, England). * The difference in the distribution of number of chromosomes between two groups was calculated by the chi-square test.

Supplementary Fig. 6. Depletion of Mad2 induces accumulation of p21. Mad2 siRNA was transfected as described in Fig. 5A and protein samples were harvested at day 5 and a detail immunoblotting method was described in Material and Method.

Supplementary Fig. 7. Depletion of p31^{comet} protein does not affect cell growth or senescence. (A) A549 cells were transfected with three different siRNAs for p31^{comet}, and the reduced protein levels analyzed by Western blotting. β-actin was used as the loading control. (B) SA-β-gal positive cells were scored from 400 cells per experiment, 6 days after siRNA transfection. (C) Viable cell numbers were counted at the indicated days after virus infection or siRNA transfection (p31-1 of present figure A).