

Supplementary figures

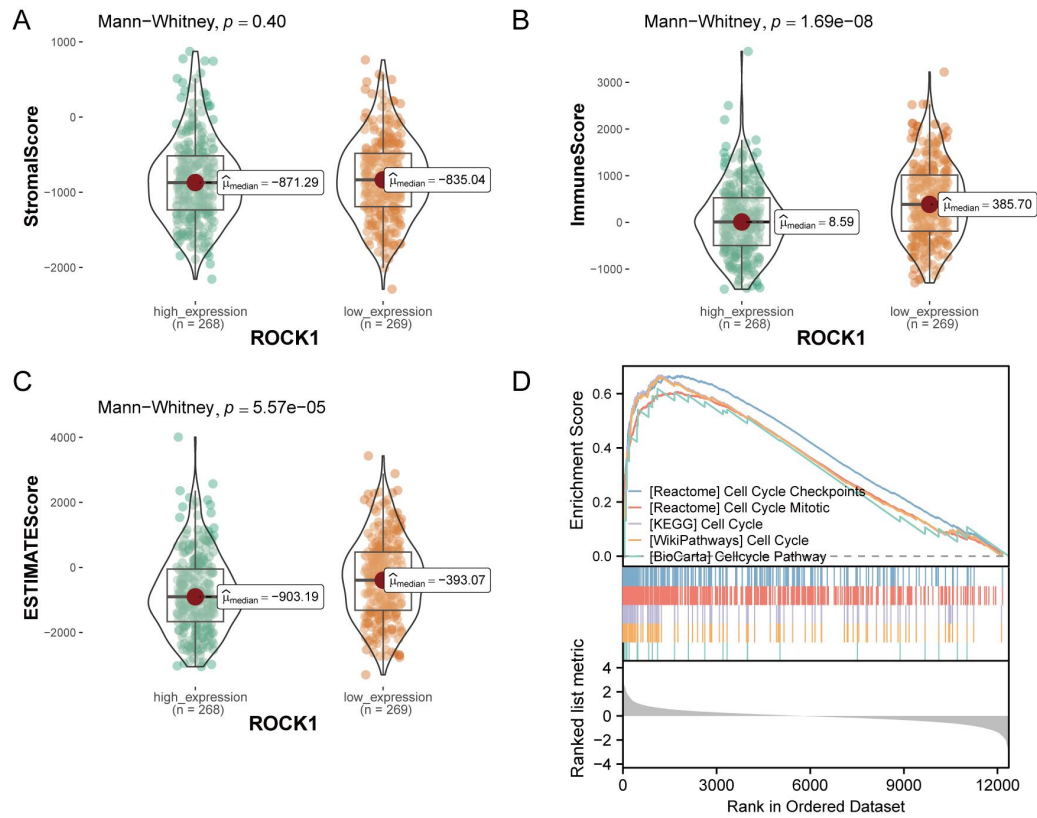


Figure S1 Estimate and GSEA analysis of EC patients according to ROCK1 expression.

- A . Estimate algorithm revealed no significant difference in the stromal score between patients with high ROCK1 expression and those with low ROCK1 expression.
- B . Patients with low ROCK1 expression exhibited higher immune scores.
- C . Patients with low ROCK1 expression had higher Estimate score.
- D . Patients with high ROCK1 expression were enriched in cell cycle related pathways.

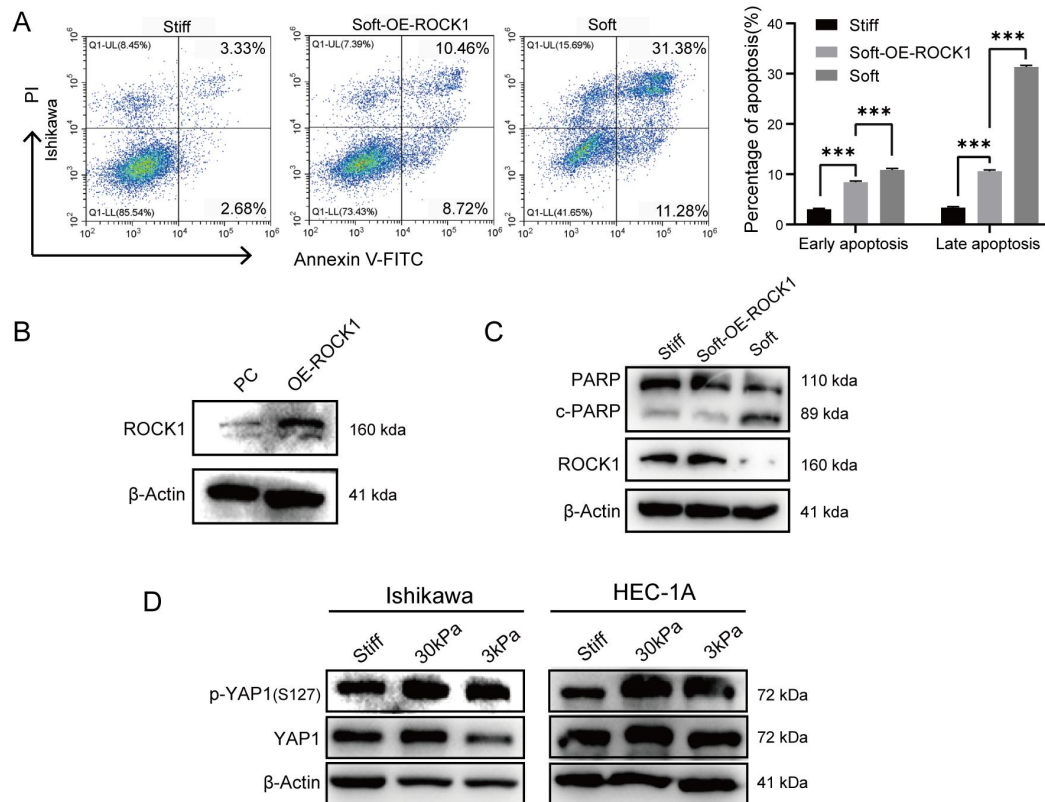


Figure S2 Cell apoptosis and protein expression cells cultured under different stiffness conditions.

A . Cell apoptosis of Ishikawa with different ROCK1 expression cultured on stiff and soft substrate.

B . Western blot analysis to detect protein efficiency of ROCK1 overexpression in Ishikawa cells.

C . Related protein expression of Ishikawa with different ROCK1 expression cultured on stiff and soft substrate.

D . YAP1 expression of Ishikawa and HEC-1A cells cultured with different stiffness substrate.

Note: The β -actin band for the HEC-1A group in this panel is reused from Figure 2D, as both panels represent the same sample under identical stiffness conditions (Stiff, 30 kPa, and 3 kPa).

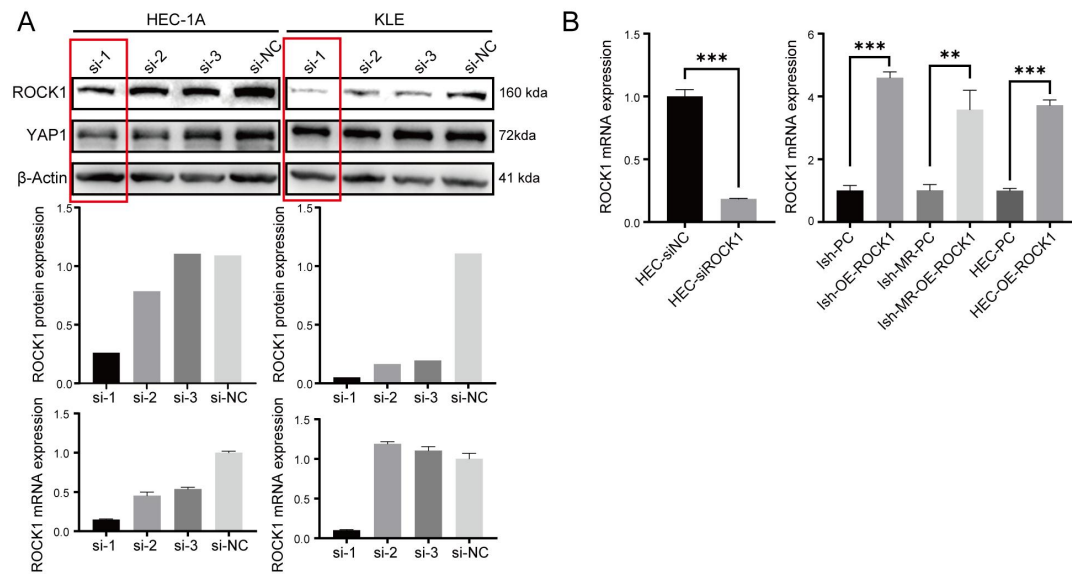


Figure S3 Efficiency of ROCK1 knockdown and overexpression.

- A . Comparison of the knockdown efficiency of 3 ROCK1-targeting siRNA on ROCK1 protein and mRNA expression in HEC-1A and KLE cells (the most efficient siRNA marked in red).
- B . ROCK1 mRNA expression after ROCK1 knockdown in HEC-1A cells and ROCK1 overexpression in Ishikawa, Ish-MR, and HEC-1A cells. **: $P < 0.01$, ***: $P < 0.001$.

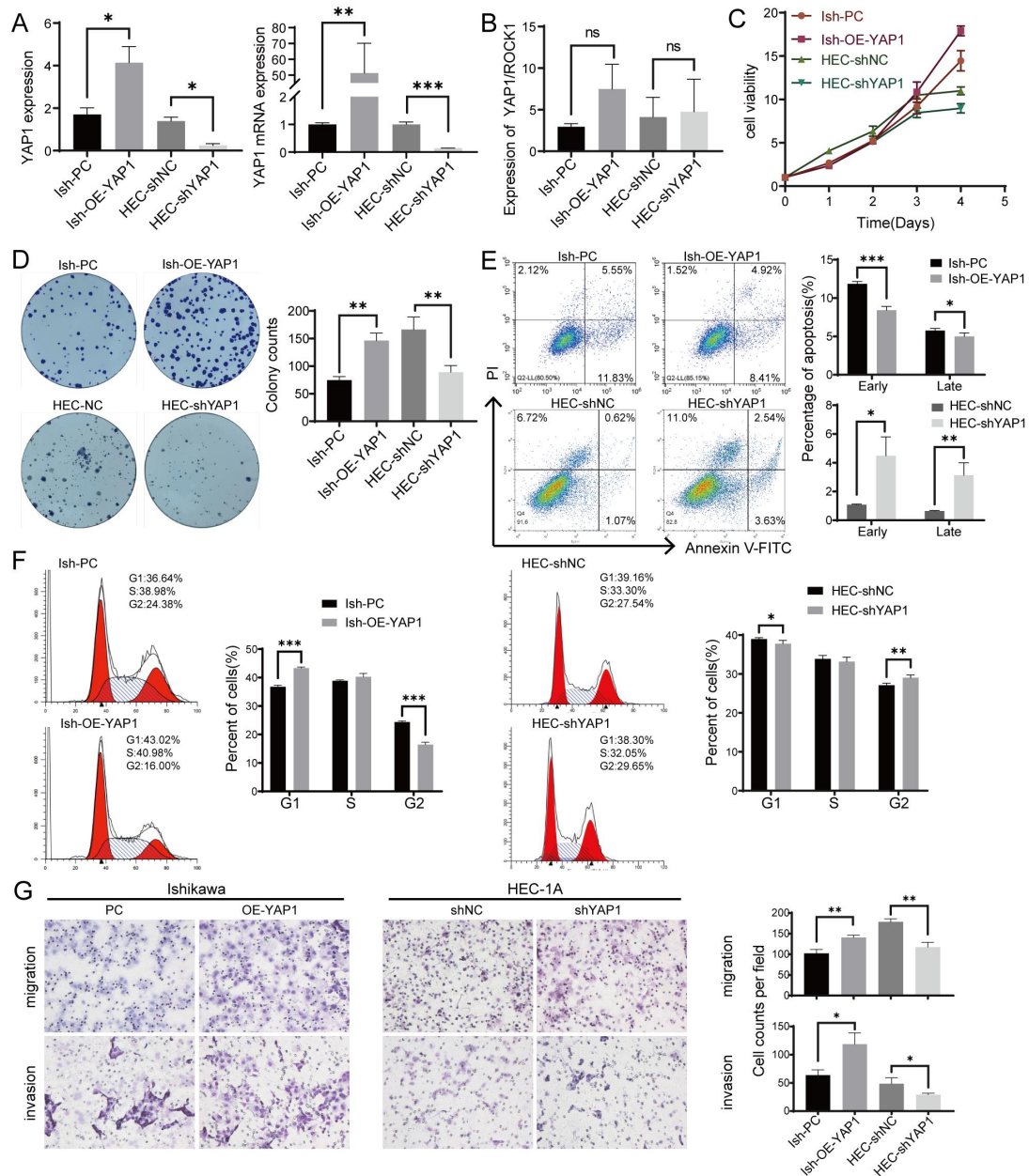


Figure S4 Elevated YAP1 expression contributes to the malignant biological behavior of endometrial cancer cells.

A . Protein and mRNA expression profiles of YAP1 in YAP1-overexpressed and knocked-down cells (statistical graphs).

B . Expression of YAP1/ROCK1 profiles after overexpression and knockdown of YAP1.

C . Proliferation assay of Ishikawa and HEC-1A cells after YAP1 overexpression and knockdown.

D . Clonogenic assay depicting the colony formation of cells in YAP1 overexpressed and knock-down conditions.

- E . Assessment of apoptosis ratio in cells with YAP1 overexpression and knockdown.
- F . Cell cycle distribution analysis in cells with YAP1 overexpression and knockdown.
- G . Transwell assay to evaluate the invasion and migration capabilities of cells after YAP1 overexpression and knockdown. *: $P<0.05$, **: $P<0.01$, ***: $P<0.001$.

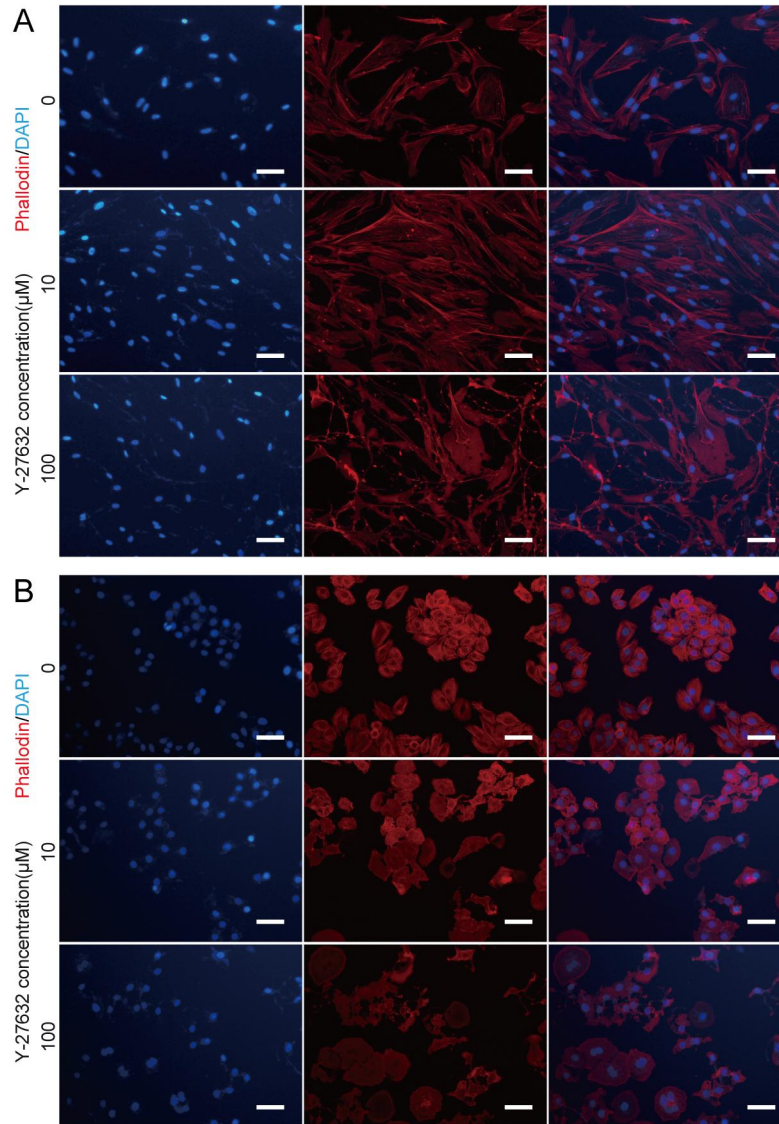


Figure S5 Cytoskeleton staining of hESCs and Ish-MR cells with Y-27632 treatment.

- A . Immunofluorescence staining of the cytoskeleton in hESCs cells (human endometrial stromal cell line) after treatment with 0, 10, and 100 μM Y-27632.
- B . Immunofluorescence staining of the cytoskeleton in Ish-MR cells after treatment with 0, 10, and 100 μM Y-27632. Scale: 50 micrometers.

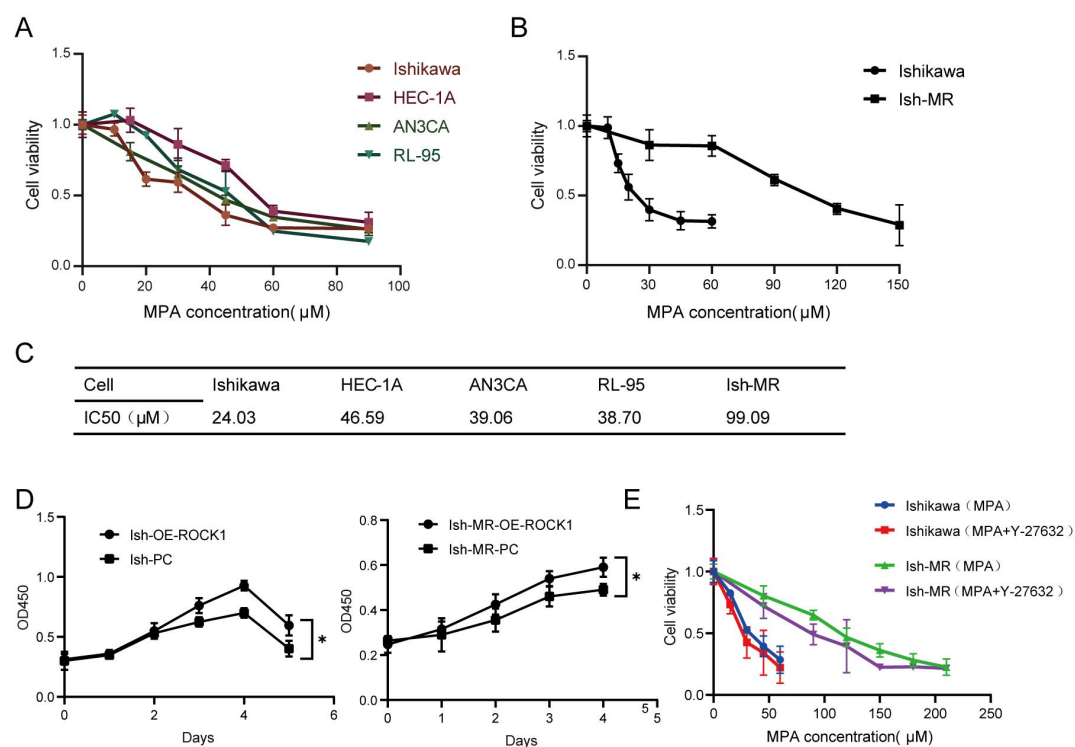


Figure S6 Cell proliferation assay to detect the effect of MPA.

A . IC50 of MPA in 4 endometrial cancer cell lines.

B . IC50 of MPA in Ishikawa and Ish-MR cells.

C . The IC50 value of cell lines.

D . The effect of ROCK1 overexpression on cell proliferation was compared under the treatment of MPA.

E . IC50 of Ishikawa and Ish-MR cells with different ROCK1 expression after treated with combination of Y-27632 and MPA. *: $P < 0.05$.

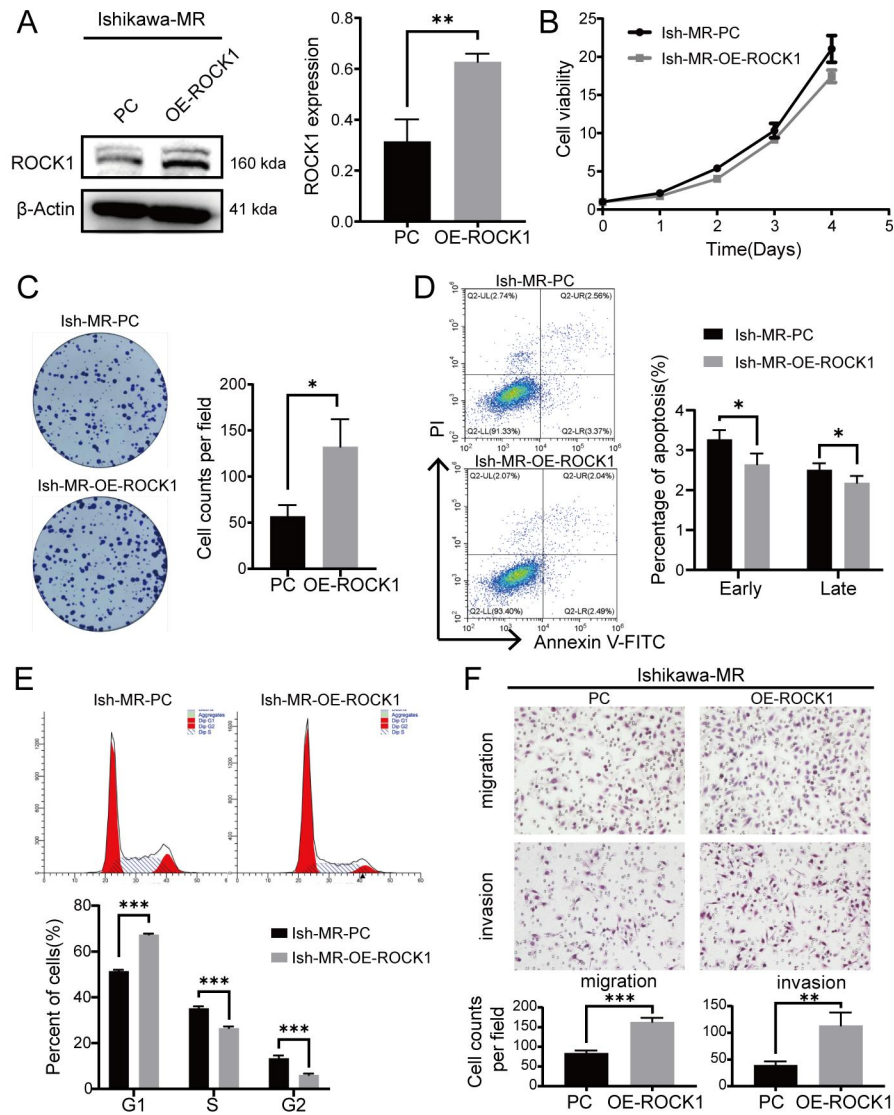


Figure S7 Elevated ROCK1 expression contributes to the malignant biological behavior of progesterone resistance endometrial cancer cells.

- F . Western blot analysis to detect protein efficiency of ROCK1 overexpression in Ish-MR cells.
- G . Proliferation of Ish-MR cells after ROCK1 overexpression.
- H . Clonogenic formation of Ish-MR cells after ROCK1 overexpression.
- I . Apoptosis of Ish-MR cells after ROCK1 overexpression.
- J . Cell cycle distribution of Ish-MR cells after ROCK1 overexpression.
- K . Migration and invasion of Ish-MR cells after ROCK1 overexpression assessed by Transwell assay. *: $P < 0.05$, **: $P < 0.01$, ***: $P < 0.001$.

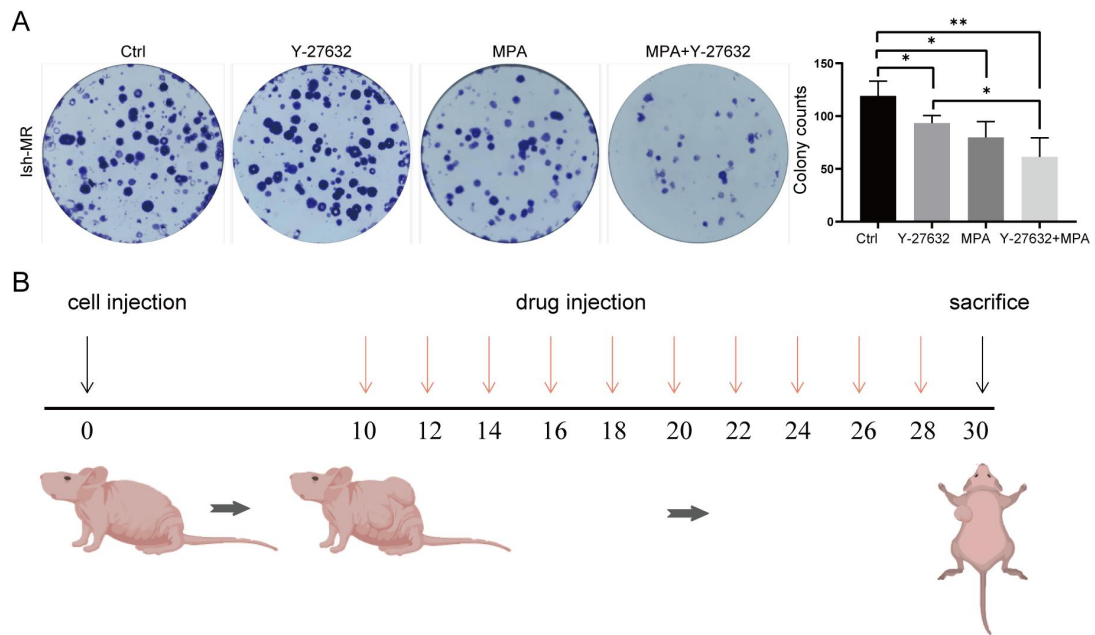


Figure S8 Colony formation assay and in vivo xenograft experiment of endometrial cancer cells.

A . Colony formation assay of Ish-MR cells treated with MPA, Y-27632, or their combination.

B . Schematic diagram of in vivo xenograft formation and drug administration in nude mice.

Original original Western blot images

Fig 2

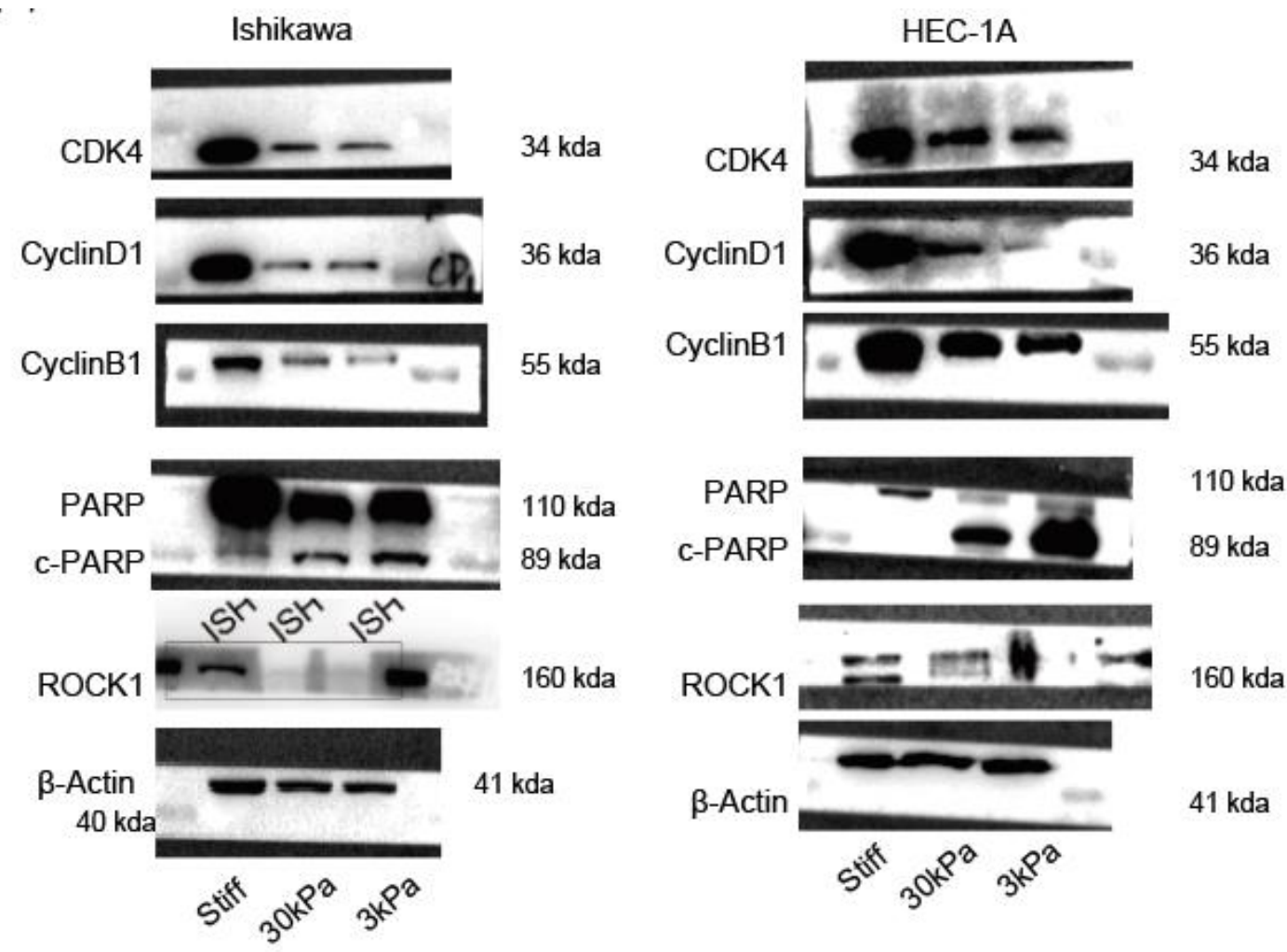


Fig 3

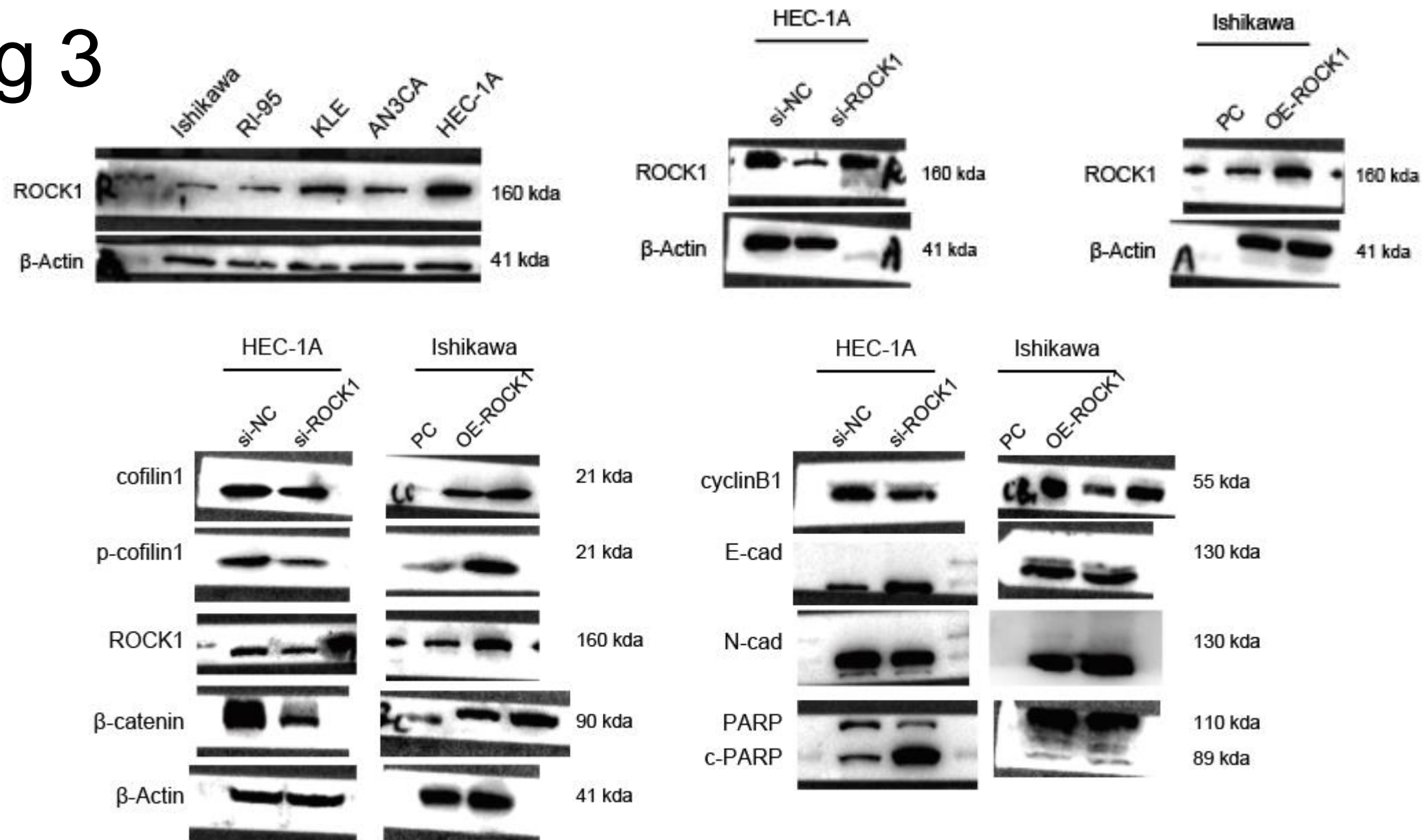


Fig 4

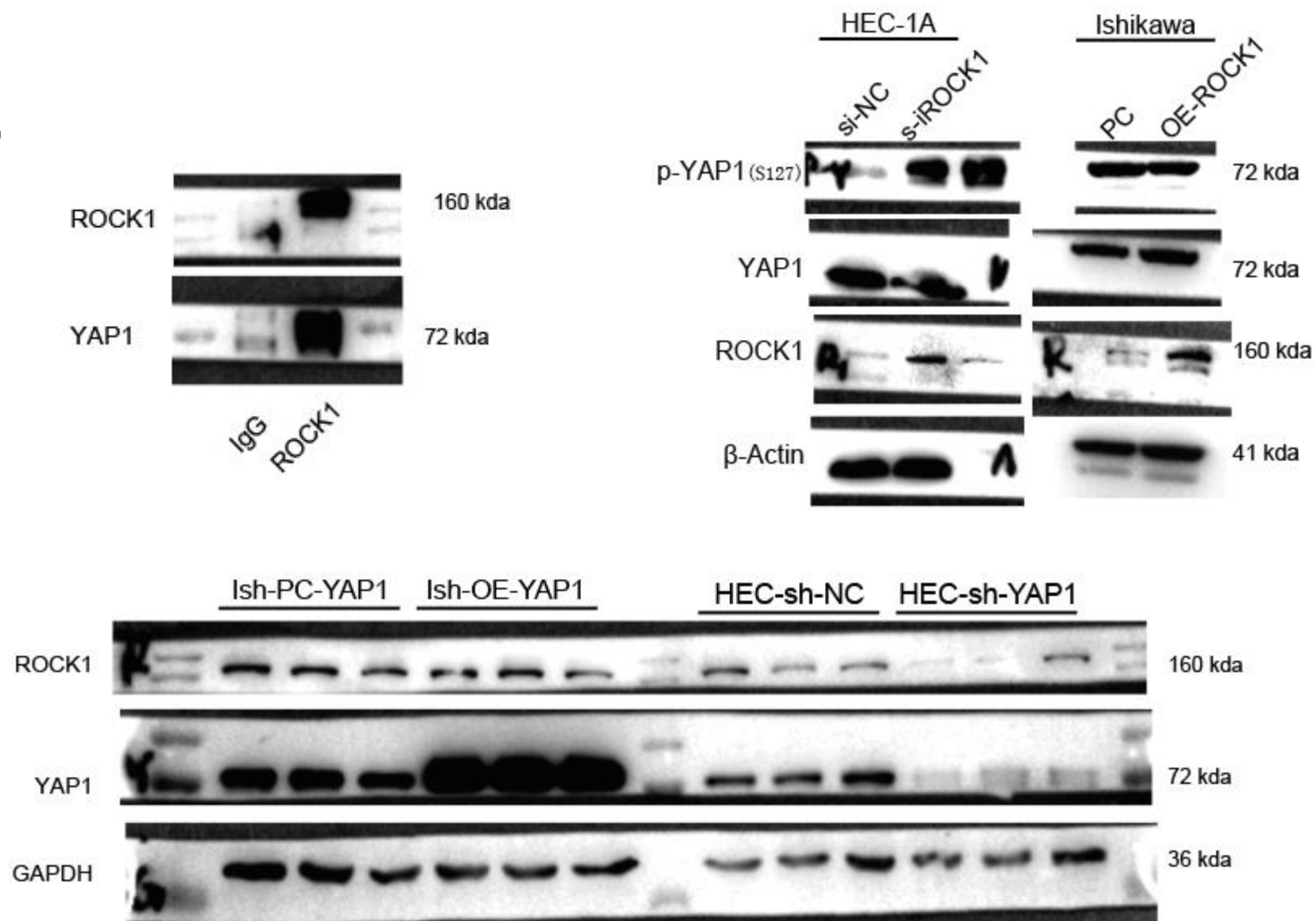


Fig 5

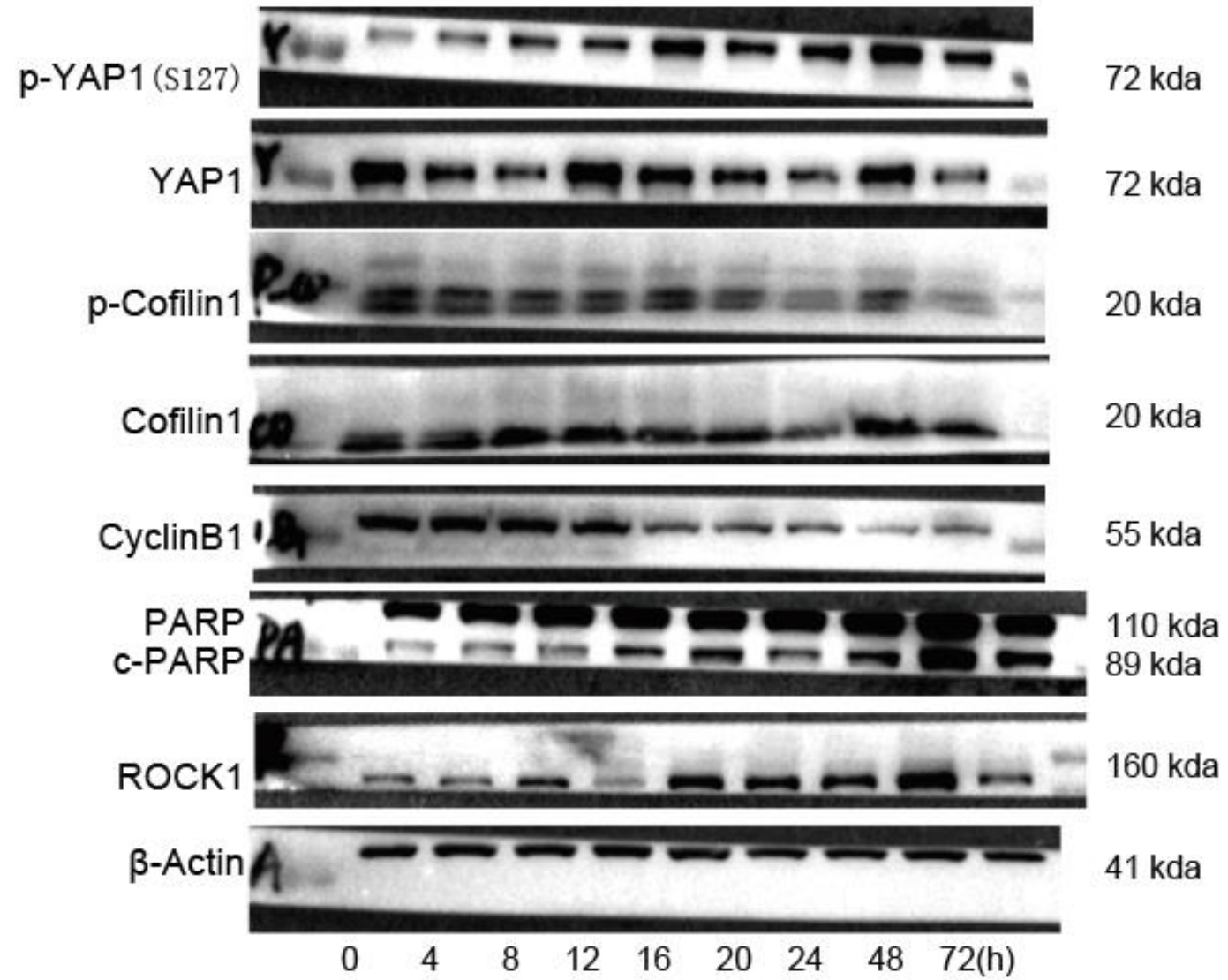


Fig 6

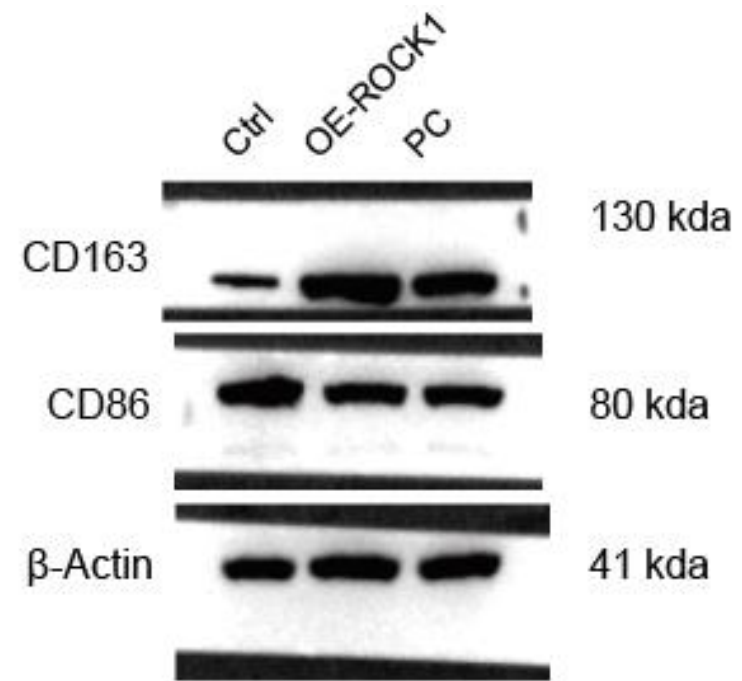


Fig S2

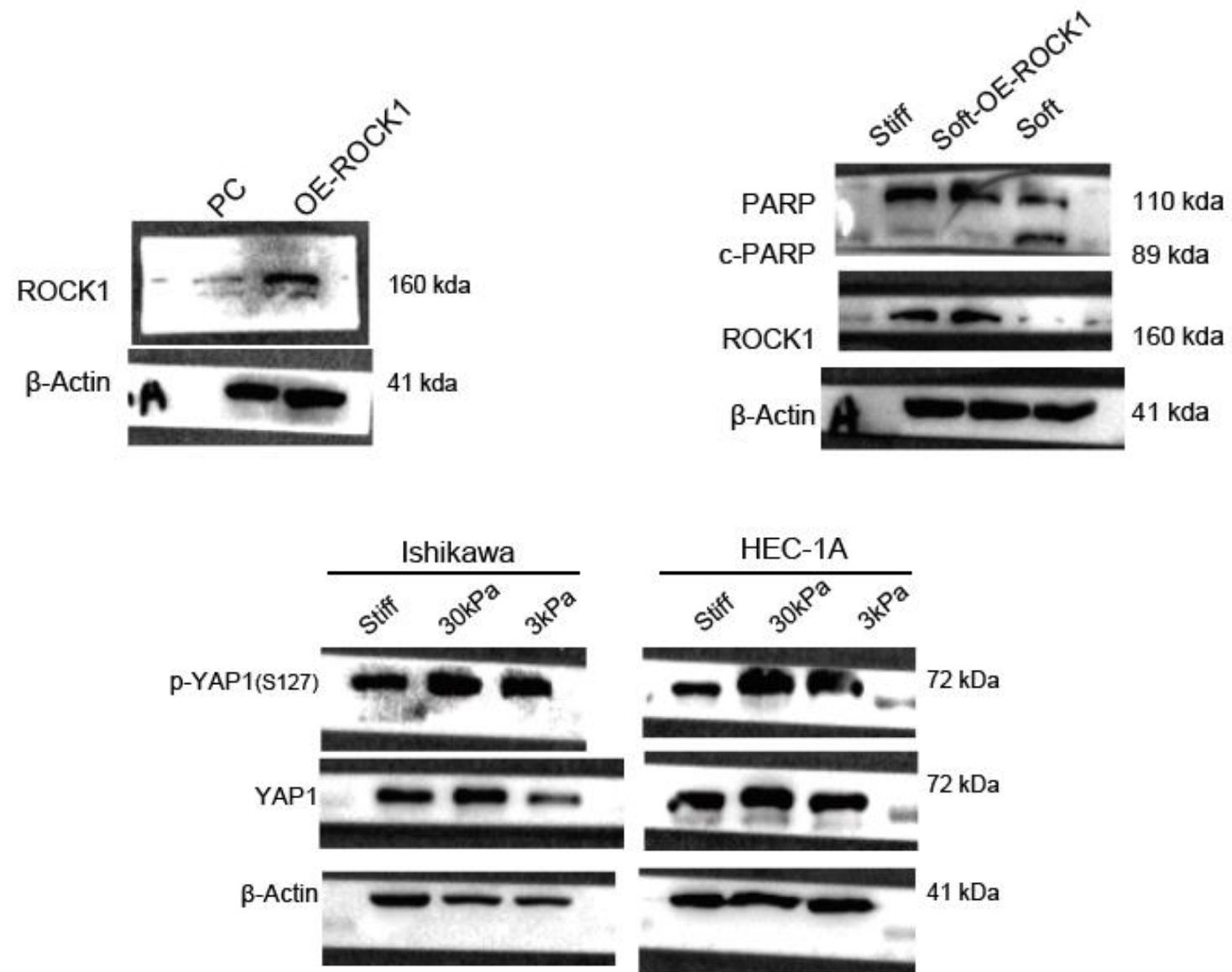


Fig S3

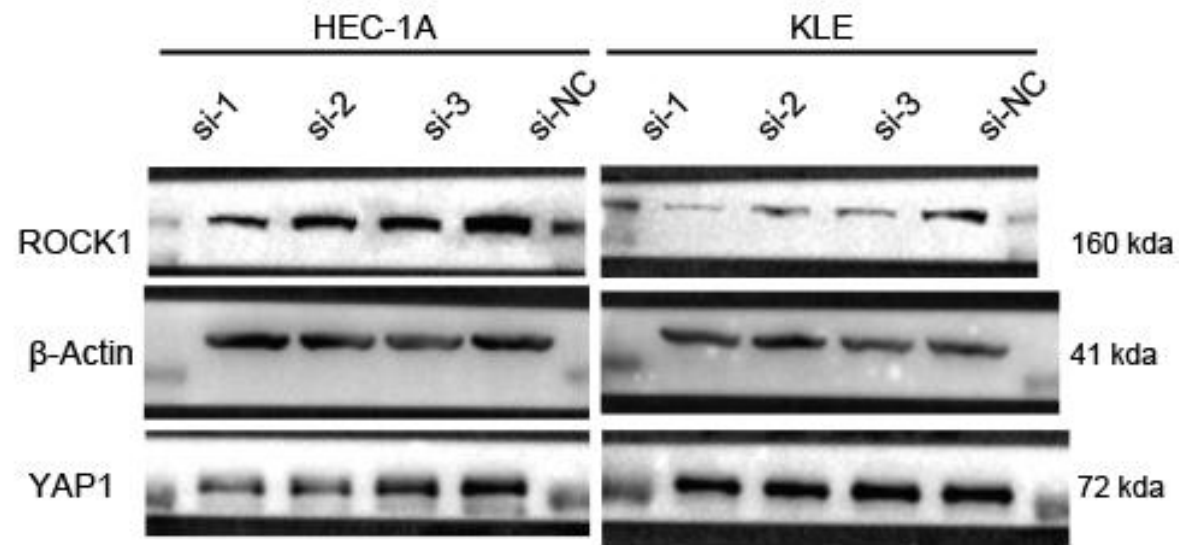


Fig S7

