

Supplementary Legends

Supplementary Figure S1. Comparison of SOX2 and p53 expression in endometrial cancer.

(a) Distribution of SOX2 expression in early (FIGO stages I and II; n = 210) and advanced (stages III and IV; n = 31) stages of endometrial cancer (n = 241). (b, c) Kaplan-Meier analyses of progression-free survival (b) and overall survival (c) in patients suffering from stage I endometrial cancer. The patients were stratified into SOX2-positive (red lines, n = 33) and SOX2-negative (blue lines, n = 168) groups. (d) Representative immunostaining images for p53-negative (left) and p53-positive (right) cases. Scale bars: 100 μ m. (e, f) Distribution of p53 expression in SOX2-positive and SOX2-negative endometrial cancer of all cases (e, n=258), and pathologically high-grade cases (f, n=25).

Supplementary Figure S2. SOX2 expression in human endometrial cancer cell lines.

(a) Quantitative real-time PCR analysis of SOX2 mRNA expression levels in endometrial cancer cell lines. PA-1 cells were used as a SOX2-positive control. (b) Images of immunostaining for SOX2, DAPI, and phalloidin. SOX2 expression

was localized mainly in the nuclei of endometrial cancer cells. PA-1 and HEC1A cells were used as positive and negative controls, respectively. Scale bars: 50 μm . (c) Flow cytometric analysis of SOX2 expression. (d) Bright-field images of EN cells (left, day 0) and immunofluorescence images of cells after staining with anti-SOX2 antibodies following 7 days of culture (right, day 7). (e) Cell cycle distribution of EN cells, as analyzed using a FACS Aria II instrument after staining of the cells with Hoechst 33342 dye. Cells were separated into G₁ and G₂/M phase subpopulations. (f) Western blot analysis and (g) quantitative real-time PCR analysis of EN cells after separation into G₁ and G₂/M phases. Cyclin B1 and cyclin E1 were also used to confirm the cell cycle phases.

Supplementary Figure S3. SOX2 is required to modulate the proliferation of endometrial cancer cells.

(a) Western blot analyses in EN, and JHUEM7 cells transfected with SOX2 siRNA or control siRNA. (b, c) Time course of cell proliferation for EN (b) and JHUEM7 (c) cells transfected with SOX2 siRNA or control siRNA. (d) SA- β -Gal activity in EN cells transfected with SOX2 siRNA or control siRNA. (e) *CDKN1A* mRNA expression levels in EN cells transfected with SOX2 siRNA, as analyzed

by quantitative real-time PCR. (f) Chromatin immunoprecipitation (ChIP) was used to detect the SOX2-*CDKN1A* interaction in EN cells.

Supplementary Figure S4. Stratification of SOX2/p21 immunostaining based on p53 expression.

p53 expression in four subtypes of SOX2 and p21 staining pattern in advanced endometrial cancer (n=31).

Supplementary Table S1. Clinicopathological factors of all 258 patients with uterine corpus endometrioid adenocarcinoma presented in Figure 1b.

Clinicopathological factors were analyzed by Fisher's exact test.

Abbreviations: ER, estrogen receptor; PR, progesterone receptor.

Supplementary Table S2. Clinicopathological factors of 241 patients with uterine corpus endometrioid adenocarcinoma presented in Figure 1c, 1d, and Supplementary Figure S1.

Supplementary Table S3. Univariate (a) and multivariate analyses (b) of prognostic factors for overall survival (OS) and progression-free survival (PFS) in 31 cases of advanced-stage cancer presented in Figure 1c and 1d.

Cox proportional hazards regression models were used to calculate hazard ratios (HRs) and 95% confidence intervals (95% CIs). Baseline variables with *p* values of less than 0.05 in univariate analysis were included in the multivariate analysis. Statistically significant differences are indicated in bold letters.

Supplementary Table S4. Univariate (a) and multivariate analyses (b) of prognostic factors for overall survival (OS) and progression-free survival (PFS) in 201 cases of stage endometrial cancer presented in

Supplementary Figure S1b and S1c.

Statistically significant differences are indicated in bold letters.

Supplementary Table S5. List of antibodies used in this study.

Abbreviations: WB, western blot; IHC, immunohistochemistry; ICC, immunocytochemistry; ChIP, chromatin immunoprecipitation.