Figure S1. PTTG1 expression in relation to cell proliferation, cell cycle and apoptosis in YD-10B cell lines. (A) PTTG1 expression was analyzed by reverse transcription-quantitative PCR in oral squamous cell carcinoma cells. (B) Protein expression of PTTG1 and PCNA in YD-10B cell lines revealed by western blotting. The left panel shows the membranes stained with antibodies, with GAPDH as an internal control and the right panel shows the relative quantification of protein expression. (C) Protein expression (left) and the fold change (right) of cell cycle markers, including cyclins D1, E and B1 in YD-10B cell lines. (D) Protein expression (left) and fold change (right) related to apoptosis markers, including Cas-7, c-Cas-7 and c-PARP in YD-10B cell lines. *P<0.05, **P<0.01 and ***P<0.001 vs. VC using Student's t-test. All experiments were performed in triplicate. PTTG1, pituitary tumor-transforming gene 1; PCNA, proliferating cell nuclear antigen; c-, cleaved-; Cas-7, Caspase-7; PARP, cleaved poly (ADP-ribose) polymerase; siR-PTTG1, small interfering RNA-PTTG1; VC, vehicle control.

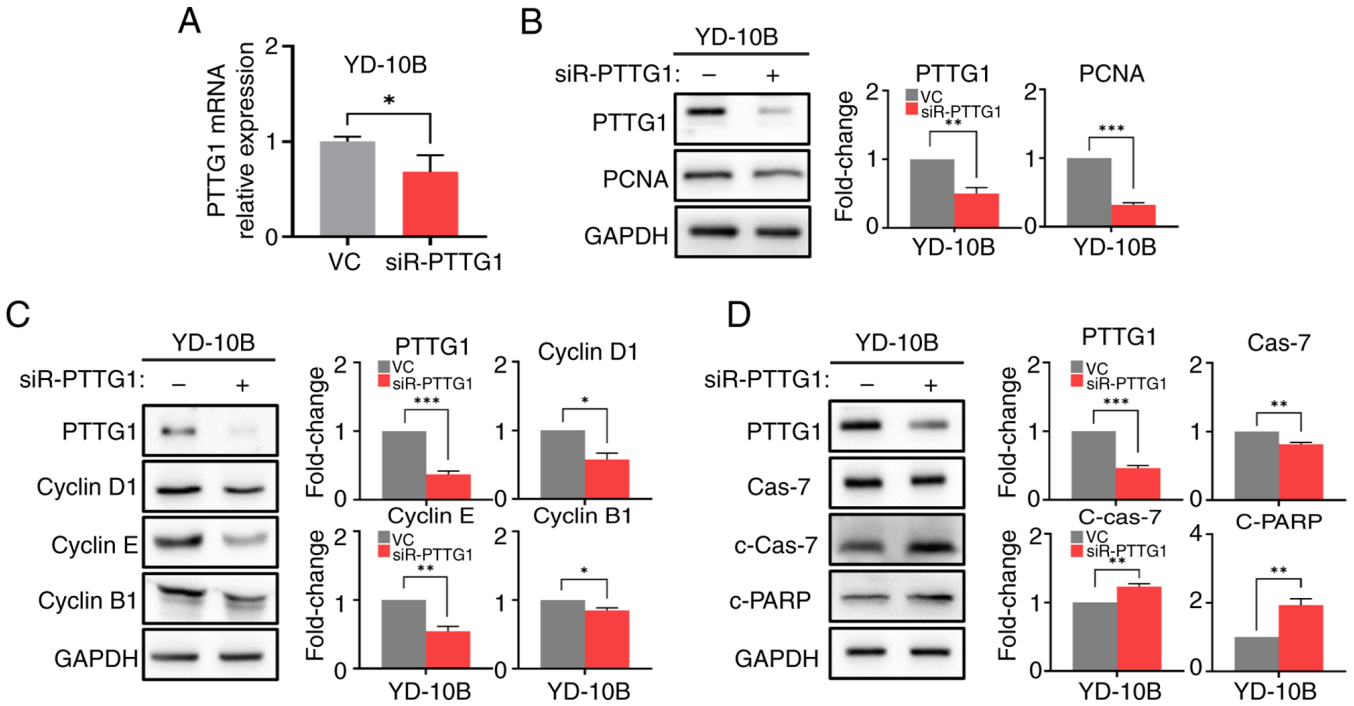

Figure S2. The effect of PTTG1 on cellular senescence and DNA damage in YD-10B cell lines. (A) Protein expression (left) and quantification (right) of γH2AX in YD-10B cell lines. GAPDH was used as an internal control. (B) Representative images (left) and numbers (right) of cellular senescence in YD-10B cell lines detected by senescence-associated β-galactosidase staining (blue). The percentages of YD-10B cell lines determined by β-galactosidase incorporation (scale bars, 50 μm and 200 μm, respectively; original magnification, x40). *P<0.05 and ***P<0.001 vs. VC using Student's t-test. All experiments were performed in triplicate. PTTG1, pituitary tumor-transforming gene 1; siR-PTTG1, small interfering RNA-PTTG1; γH2AX, phosphorylated histone H2AX; VC, vehicle control.

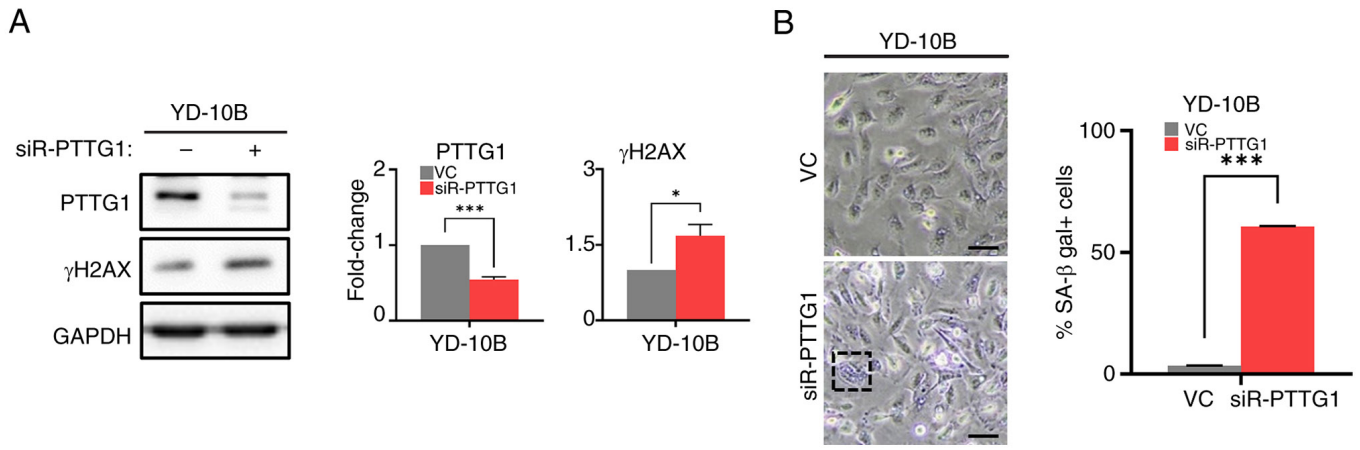

Figure S3. The effect of PTTG1 on p53 dependent pathway in OSCC. Protein expression (left) and quantification (right) of p53, RB and p-RB were analyzed by western blotting in OSCC cells. *P<0.05, **P<0.01 and ***P<0.001 vs. VC using Student's t-test. All experiments were performed in triplicate. PTTG1, pituitary tumor-transforming gene 1; siR-PTTG1, small interfering RNA-PTTG1; OSCC, oral squamous cell carcinoma; Rb, retinoblastoma; pRb, phosphorylated Rb; VC, vehicle control.

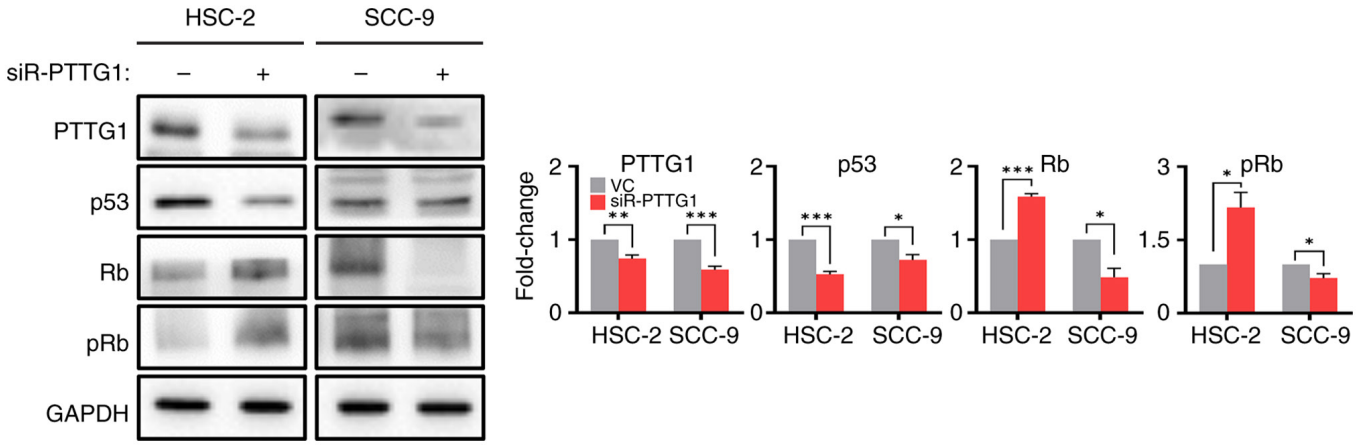

Figure S4. The effect of DOX on PTTG1 and p21 expression in oral squamous cell carcinoma cell lines. (A and B) The cell viability of (A) HSC-2 and (B) SCC-9 cell treated with DOX using MTT assay. *P<0.05, **P<0.01 and ***P<0.001 vs. control using one-way ANOVA followed by Dunnett's test for post-hoc analysis. (C and D) The mRNA expression of (C) PTTG1 and (D) p21 in HSC-2 and SCC-9 cells treated with DOX using reverse transcription-quantitative PCR. (E and F) Representative images (left) and numbers (right) of chromosomal damage detected by γH2AX staining in (E) HSC-2 and (F) SCC-9 cells. *P<0.05 and ***P<0.001 vs. control using Student's t-test. All experiments were performed in triplicate. DOX, doxorubicin; PTTG1, pituitary tumor-transforming gene 1; γH2AX, phosphorylated histone H2AX.