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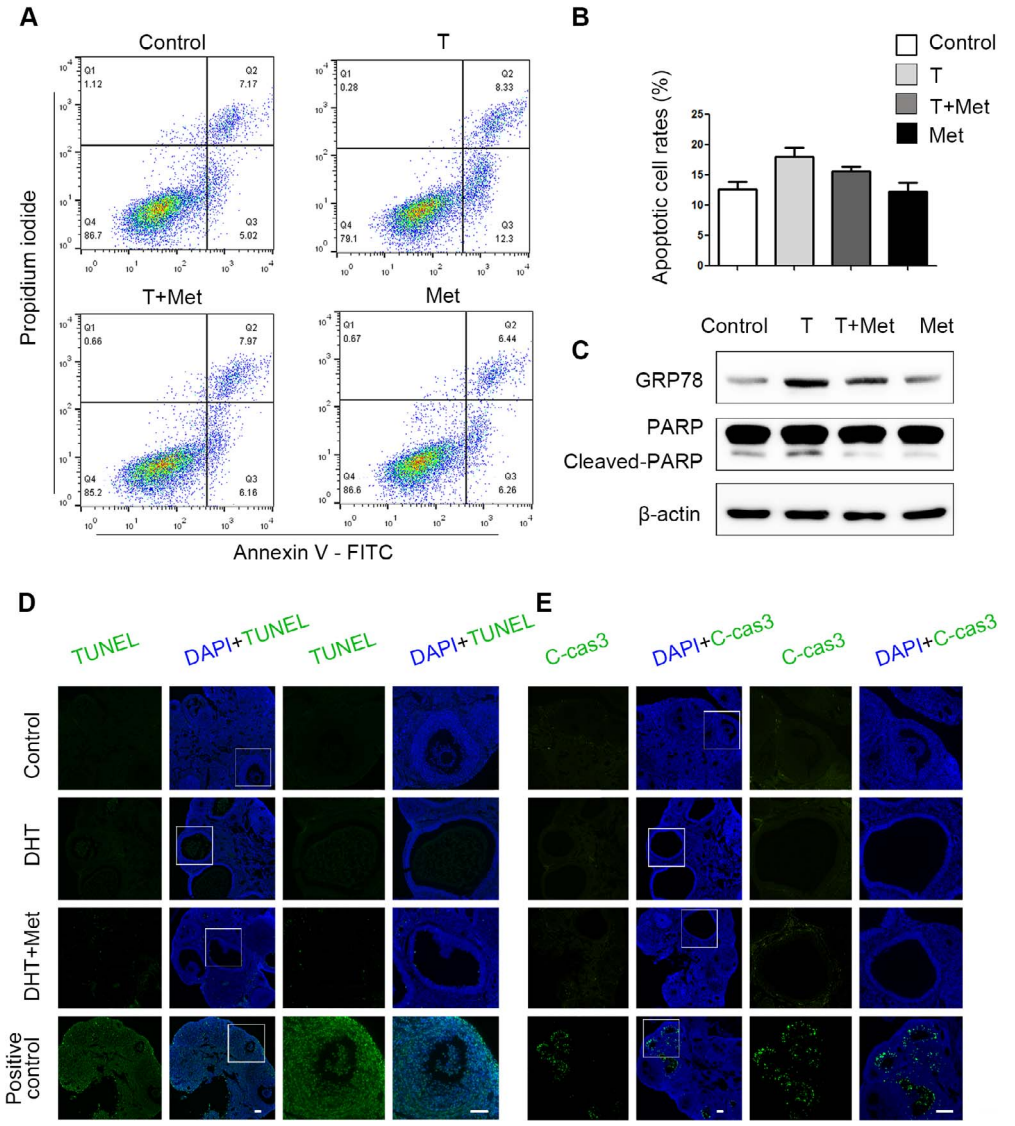
SUPPLEMENTARY DATA



Supplementary Figure S4 Analysis of apoptosis in testosterone-treated granulosa cells (GCs) and dihydrotestosterone (DHT)-induced polycystic ovary syndrome (PCOS) model mouse ovaries. (**A**) FITC-Annexin V and propidium iodide (PI) staining of GCs and flow cytometry analysis of GC apoptotic rates after treatment with 10 μM testosterone (T) for 24 h with or without 1 mM metformin (met). (**B**) Statistical analysis of the apoptosis rates of different groups compared to that of the control group. The data are presented as means ± SEMs. (**C**) Western blot results of total PARP protein and cleaved PARP protein levels in GCs treated with 10 μM testosterone with or without 1 mM metformin for 24 h. (**D**) TUNEL assay for apoptosis in ovarian antral follicles from the control, DHT-treated and DHT- and metformin-treated groups at 16 weeks of age. Slides with ovarian sections treated with DNase I for 30 min served as positive controls. DNA strand breaks were visualized by detecting fluorescein-dUTP signals. Nuclei were stained with DAPI. Scale bar: 100 μm. (**E**) Immunofluorescence (IF) results for cleaved caspase 3 in ovarian antral follicles from the different groups at 16 weeks of age. The positive control samples were from the ovaries of mice injected with 120 mg/kg cyclophosphamide for 72 h. Nuclei were stained with DAPI. Scale bar: 100 μm. C-cas 3, cleaved-caspase 3.