



Primary Cilia Restrain PI3K-AKT Signaling to Orchestrate

Human Decidualization

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Supplemental materials

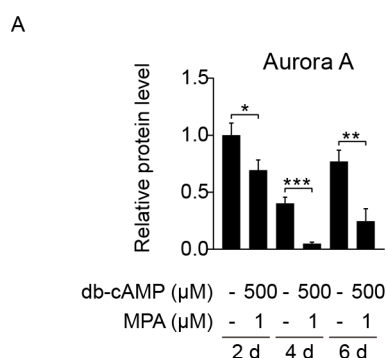


Figure S1. Aurora A-mediated primary cilia disassembly inhibits human decidualization. **(A)** Western blot quantification of Aurora A during stromal cells decidualization for 2, 4, and 6 days. In **(A)**, data are presented as means \pm SD from three independent experiments. * $P < 0.05$, ** $P < 0.01$, and *** $P < 0.001$ by two-tailed Student's t test for comparing two groups or one-way ANOVA test for comparing more than two groups.

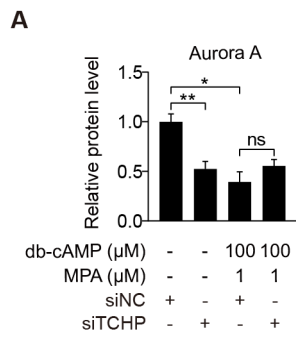


Figure S2. Trichoplein act as the activator of Aurora A. **(A)** Western blot quantification of Aurora A after stromal cells were transfected with siTCHP for 24 h then in vitro decidualization for 48 h. In **(A)**, data are presented as means \pm SD from three independent experiments. ns, ($P>0.05$), * $P<0.05$, and ** $P<0.01$ by two-tailed Student's *t* test for comparing two groups or one-way ANOVA test for comparing more than two groups.

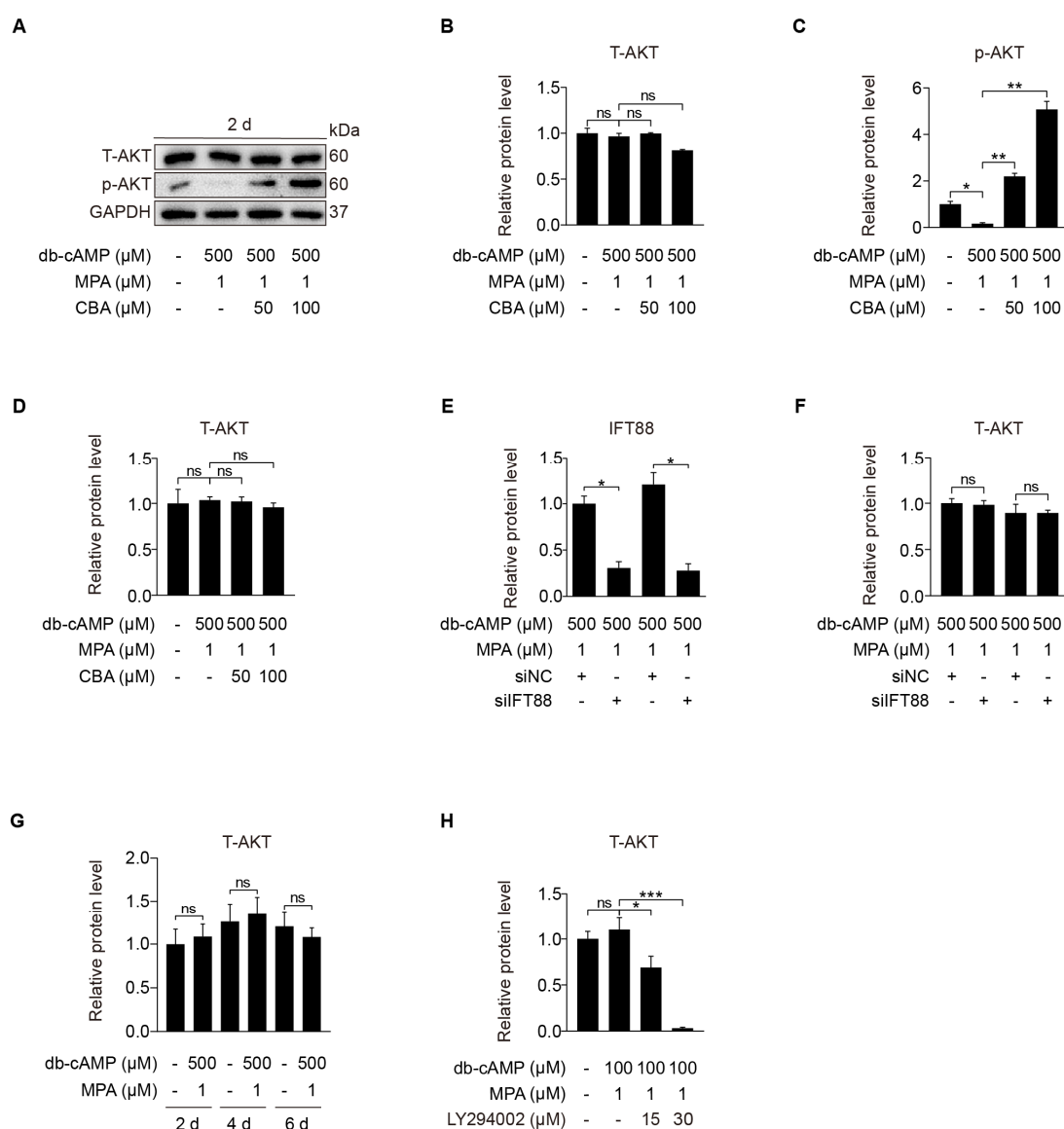


Figure S3. Primary cilia negatively control AKT activation (**A** to **C**) Western blot and quantification of T-AKT and p-AKT (S473) after stromal cells were treated with CBA for 24 h during in vitro decidualization for 2 days. (**D**) Western blot quantification of T-AKT after stromal cells were treated with CBA for 24 h during in vitro decidualization for 4 days. (**E** and **F**) Western blot quantification of IFT88 and T-AKT after stromal cells were treated with siIFT88 for 24 h then in vitro decidualization for 2 and 4 days. (**G**) Western blot quantification of T-AKT in stromal cells during decidualization for 2, 4, and 6 days. (**H**) Western blot quantification of T-AKT in LY294002-treated stromal cells for 48 h. In (**A**), the representative images of three biologically independent experiments are shown. In (**B-H**), data are presented as means \pm SD from three independent experiments. ns, ($P > 0.05$), * $P < 0.05$, ** $P < 0.01$, and *** $P < 0.001$ by two-tailed Student's *t* test for comparing two groups or one-way ANOVA test for comparing more than two groups.

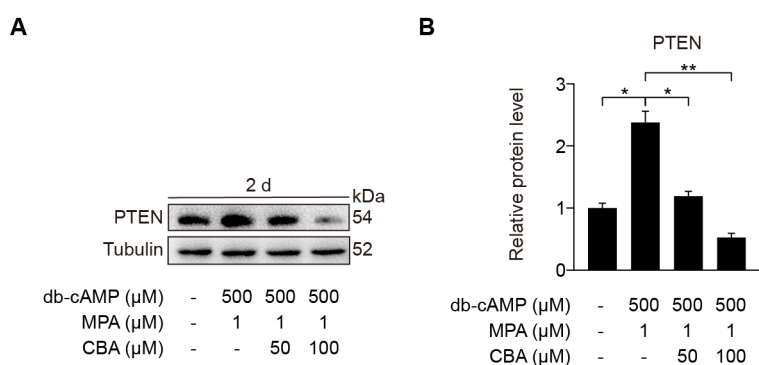


Figure S4. PTEN antagonizes AKT during stromal cells decidualization. **(A and B)** Western blot and quantification of PTEN after stromal cells were treated with CBA for 24 h during in vitro decidualization for 4 days. In **(A)**, the representative images of three biologically independent experiments are shown. In **(B)**, data are presented as means \pm SD from three independent experiments. * $P < 0.05$ and ** $P < 0.01$ by two-tailed Student's *t* test for comparing two groups or one-way ANOVA test for comparing more than two groups.

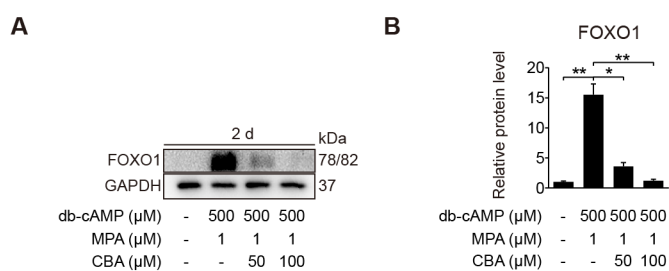


Figure S5. Primary cilia modulate FOXO1 expression by suppressing AKT activity. **(A and B)** Western blot and quantification of FOXO1 after stromal cells were treated with CBA for 24 h during in vitro decidualization for 2 days. In **(A)**, the representative images of three biologically independent experiments are shown. In **(B)**, data are presented as means \pm SD from three independent experiments. * $P < 0.05$ and ** $P < 0.01$ by two-tailed Student's t test for comparing two groups or one-way ANOVA test for comparing more than two groups.

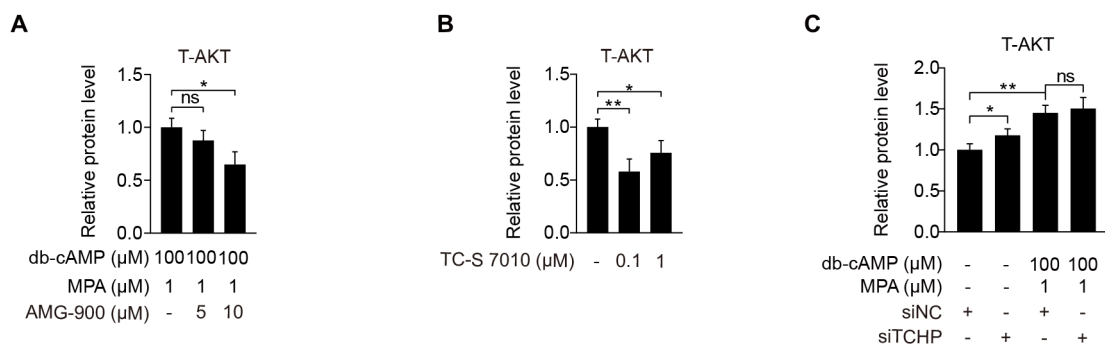


Figure S6. Trichoplein-Aurora A-mediated cilia disassembly contributes to PTEN-AKT-FOXO1 signaling. **(A)**

Western blot quantification of T-AKT in AMG-900-treated stromal cells for 48 h. **(B)** Western blot quantification of T-AKT in TC-S 7010-treated stromal cells for 48 h. **(C)** Western blot quantification of T-AKT after stromal cells were transfected with siTCHP for 24 h then in vitro decidualization for 48 h. In **(A)**, **(B)**, and **(C)**, data are presented as means \pm SD from three independent experiments. ns, ($P > 0.05$), * $P < 0.05$, and ** $P < 0.01$ by two-tailed Student's *t* test for comparing two groups or one-way ANOVA test for comparing more than two groups.