Supplemental Figure 1: Validation of PROK1 via PROKR1 induction of COX2. (A) Further characterisation of the parental TCam-2 cells demonstrates they express significantly less PROKR2 transcript that human fetal ovarian tissue (15-18 weeks). (B) Further, treatment of parental TCam-2 cells with 40 nM PROK1 (+PROK) did not induce COX2 expression compared to vehicle control (VEH), demonstrating that mRNA induction in PROKR1-TCam-2 cells is the result of PROK1 via PROKR1 downstream signalling. Data shown is relative to housekeeping gene RPL32 and is mean ± SEM, *p≤0.05.
Supplemental Figure 2: COX2 induction over time. COX2 induction by PROK1 via PROKR1 was examined over a timecourse at 2, 4, 8, 12 and 24 hours (H). Enhanced COX2 mRNA was seen as early as 4 hours post 40 nM PROK1 treatment (+PROK) compared to vehicle control (VEH).
Supplemental Figure 3: Investigation of PROK1 functionality in PROKR1-TCam-2 cells. (A) As COX2, a prostaglandin synthesis enzyme, was induced by 40 nM PROK1 treatment (+PROK) compared to vehicle treated PROKR1-TCam-2 cells (VEH), the prostaglandin E$_2$ receptors (EP1-4) were also investigated for changes in mRNA expression via qRT-PCR after 12 hours of treatment, as they form a positive feedback loop in other tissues. No change in expression was determined. (B) Changes in proliferation and survival were investigated downstream of PROK1 treatment of PROKR1-TCam2 cells treated with PROK or vehicle over 24, 48, and 96 hours. No significant difference was determined via colorimetric SRB assay suggesting PROK1 does not function to alter cell turnover in these cells.