

Supplemental Figure 1. Ribonuclease protection assay of mRNAs associated with granulosa cell differentiation in cells stimulated by FSH or PKA-CQR in the absence or presence of testosterone. Granulosa cells were infected with PKA-CQR lentiviral vector (5 MOI) as described in Figure 1. The next morning unattached cells and free lentivirus were removed and replaced with M199. Twenty-four h later medium was replaced with fresh medium without stimulatory agents or containing testosterone (30 ng/ml), FSH (100 ng/ml) or both as indicated in the Figure. After 48 h, total RNA was extracted from monolayers and analyzed for mRNA by ribonuclease protection assay. The housekeeping gene cyclophilin was included as a loading control. Results shown are representative of 3 separate groups of granulosa cells (top). Densitometry analysis of the ribonuclease protection assay products was performed using the NIH Image software (bottom). The signal of each mRNA was normalized to the housekeeping gene cyclophilin (relative intensity). Results show the mean \pm 1 SEM of 3 groups of granulosa cells. Significant differences were considered when $P < 0.05$ and are indicated by different letters in the figure.

