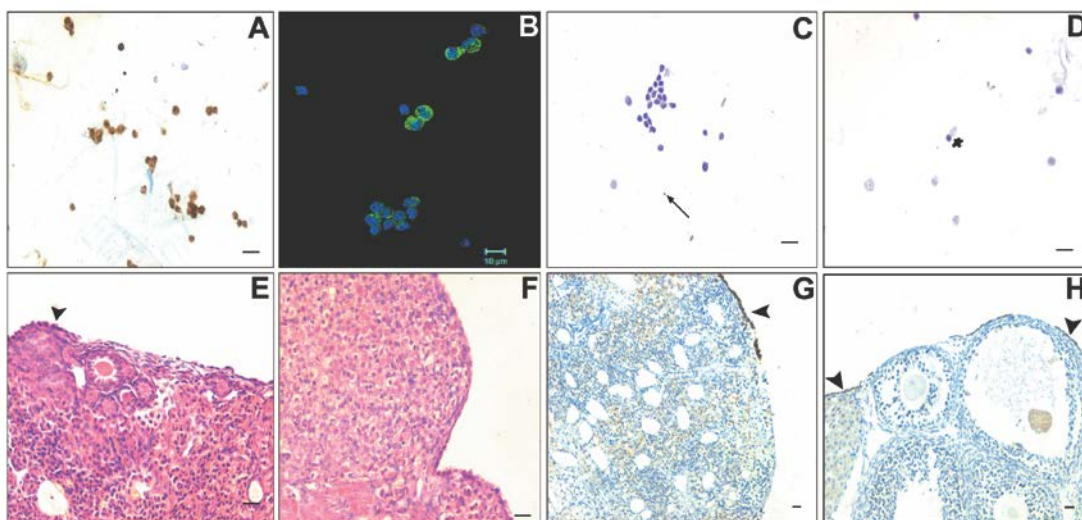
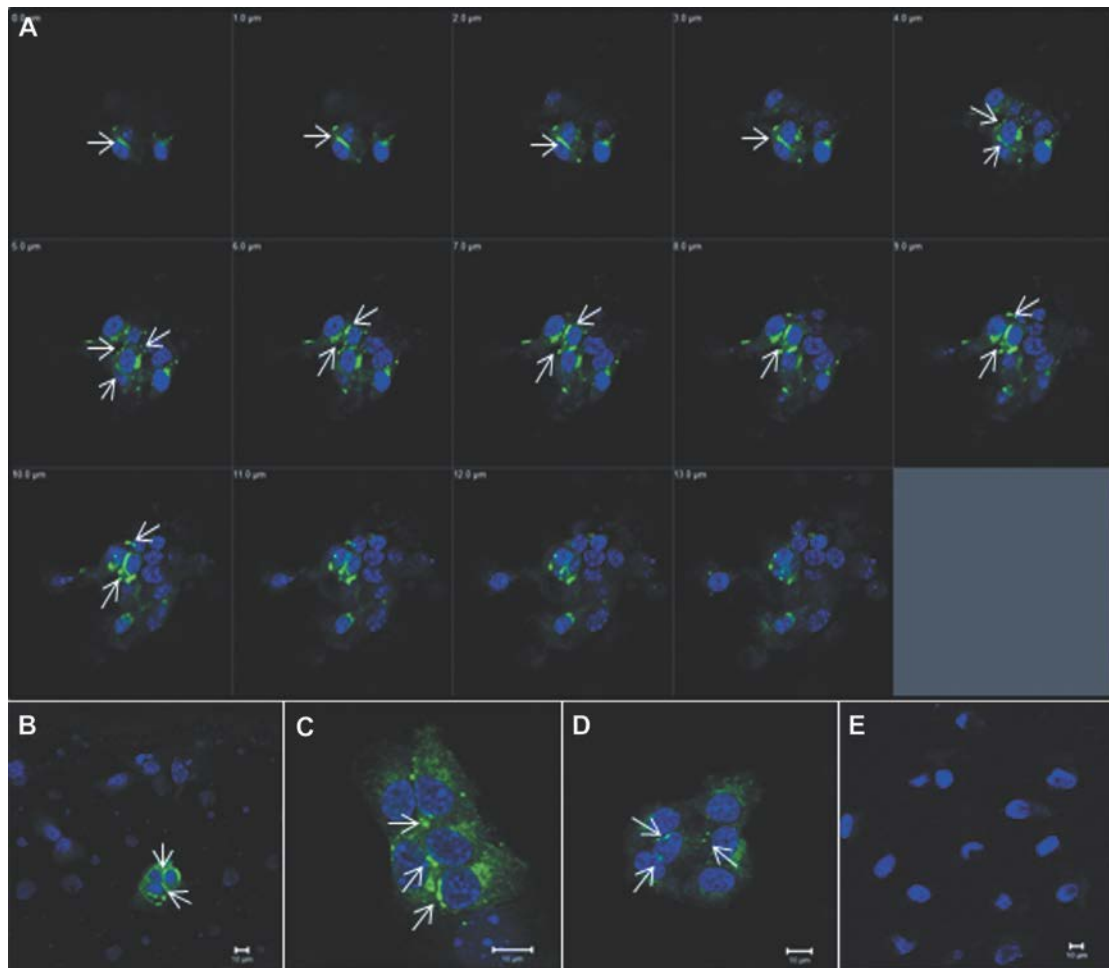


Supplementary Figure 1: Immunofluorescence for OCT-4A on whole ovarian smears from control and chemotherapy treated mice: Only small nuclear OCT-4 positive VSELs were observed in both control **(A)** and chemoablated **(B)** mouse whole ovarian smears using Oct-4A specific antibody (MAB4419; Millipore).



Supplementary Figure 2: Characterization of mouse OSE preparation. CK-8 (a specific mouse OSE marker) staining of OSE from control mice **(A)** showing most cells are positive for CK-8. Immunofluorescence analysis of isolated OSE from control ovaries using epithelial cell marker E-Cad **(B)** also showed most cells positive for E-Cad. The image in B is a merge of green (E-Cad) and blue (DAPI) channels. In H and E staining of OSE preparations from control mice **(C-D)**, two sizes of small cells with dark nucleus and high nucleus-cytoplasm ratio were observed in addition to epithelial sheaths, which are characteristic of VSELs (arrow) and OGSCs (asterisks) described earlier.² H and E analysis of control **(E)** and chemoablated **(F)** ovarian sections after removal of OSE confirmed removal of OSE. The inner regions (including follicles in control) were intact and not disturbed by the enzymatic treatment. Arrowhead represents region where OSE was not removed. CK-8 staining of ovarian sections prepared after removal of OSE on ovarian sections from control **(G)** and chemoablated **(H)** mice confirmed removal of OSE after enzymatic digestion. Arrowhead represents region of OSE (positive for CK-8) that is not removed. Bar in A and C-H equals 20µm.



Supplementary Figure 3 : CNX-43 staining on germ cell cyst-like structure using confocal microscopy. (A) shows Z-Stack image of the cluster shown in Figure 5D taken at 1µm intervals. **B-D** represents additional unique clusters showing CNX-43 staining from different culture well/ experiment. **E** represents negative control without the addition of CNX-43 antibody. The images shown are merge of CNX-43 (green) and DAPI (blue) staining. Arrows represent typical CNX-43 staining between the cells. Bar equals 10µm.



Supplementary Figure 4: *In vitro* culture of OSE isolated from chemoablated mouse ovaries: (A) Gene expression analysis of mRNA isolated from OSE of chemoablated ovaries on day 0 showing absence of Msy2 expression (Lane 3) suggesting complete absence of oocytes in initial culture conditions. Lanes: 1- PCR negative for Msy2, 2- Msy2 expression in whole ovary (Positive control), 3-Msy2 expression on day 0 of culture , 4- 100bp ladder, 5- Gapdh (house keeping) expression on day 0 of culture, 6-Gapdh expression in whole ovary (positive control), 6-PCR negative for Gapdh. **(B)** Immunofluorescence analysis shows the cell clusters with ovoid cells observed on day 2 of culture were positive for OCT-4 (green) in cytoplasm and BrdU (red) in nucleus suggesting they are proliferating stem cell clusters. Blue represents the nuclear counterstain DAPI.