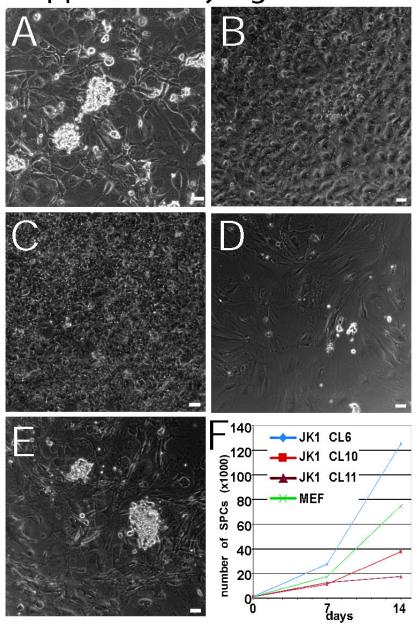
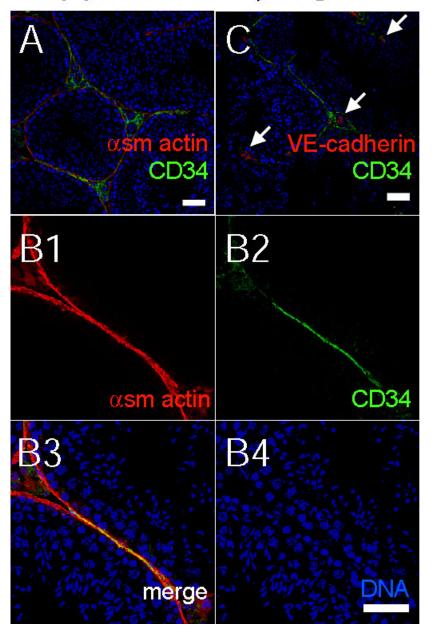
Supplementary Information Supplementary Figure 1



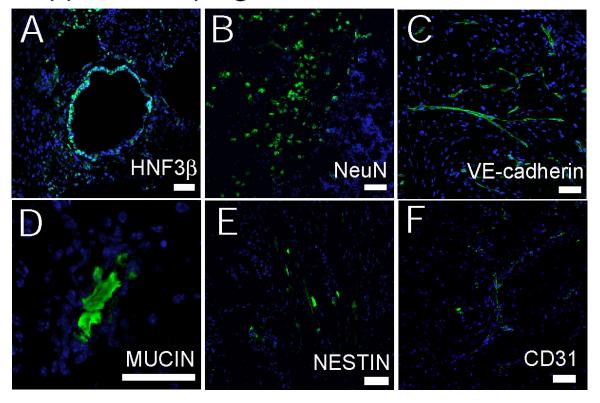
SPC growth. (A-E): Phase contrast images (original magnification 200X). (A) Prominent SPC colonies were visible on JK1 clone 6 after two weeks. (B) SPCs had adhered to JK1 clone 10 after two weeks. (C) Sparse SPCs on JK1 clone 11 were discernable after two weeks. (D) Small colonies of SPCs grew on MTS after two weeks. (E) Larger SPC colonies developed on MEFs after two weeks. (F): An equal number of SPCs were seeded onto mitomycin-C inactivated JK1 clones 6, 10, 11, and MEFs. Colonies were allowed to grow for three weeks. At the end of each week, SPCs were trypsinized, stained with trypan blue, and counted with a hemocytometer. SPC proliferation analysis on the different feeders indicated that JK1 clone 6 promoted the most SPC expansion. Clones 10 and 11 also supported proliferation, but were not as effective as clone 6. All size bars indicate 50 μm.

Supplementary Figure 2



Supplementary Figure 2. Normal mouse testis contains non-germ cells positive for CD34, α-smooth muscle actin (SMA), and VE-cadherin. (A): Cells surrounding the tubules were positive for CD34 (green) and SMA (red). (B 1 - 4): A more magnified view shows that peritubular cells expressed SMA (B1) and CD34 (B2). The merged confocal image seemed to indicate that both are expressed on the same cells (B3). Nuclear counterstaining (blue) is presented in B4. (C): While CD34 (green) is expressed on the peritubular cells lining the testis tubules, VE-cadherin (red) seems to be limited to testis endothelial cells lining blood vessels (arrows) in the interstitium. All size bars indicate 50 μm.

Supplementary Figure 3



Supplementary Figure 3. EGs derived and expanded on JK1 are capable of generating cells of all three germ layers. (A,D): HNF3 β (A) and Mucin (D) staining indicated that the EGs were capable of forming cells of the endoderm. (B,E): The presence of ectoderm is indicated by cells positive for neural markers NeuN (B) and nestin (E). (C,F): The presence of VE-cadherin (C) and CD31 (F) positive cells demonstrated that EGs were capable of forming mesoderm. All markers are shown in green. Nuclear counterstaining is blue. All size bars indicate 50 μ m.