

Table S1. Primer Sequences and Cycling Conditions for QPCR:

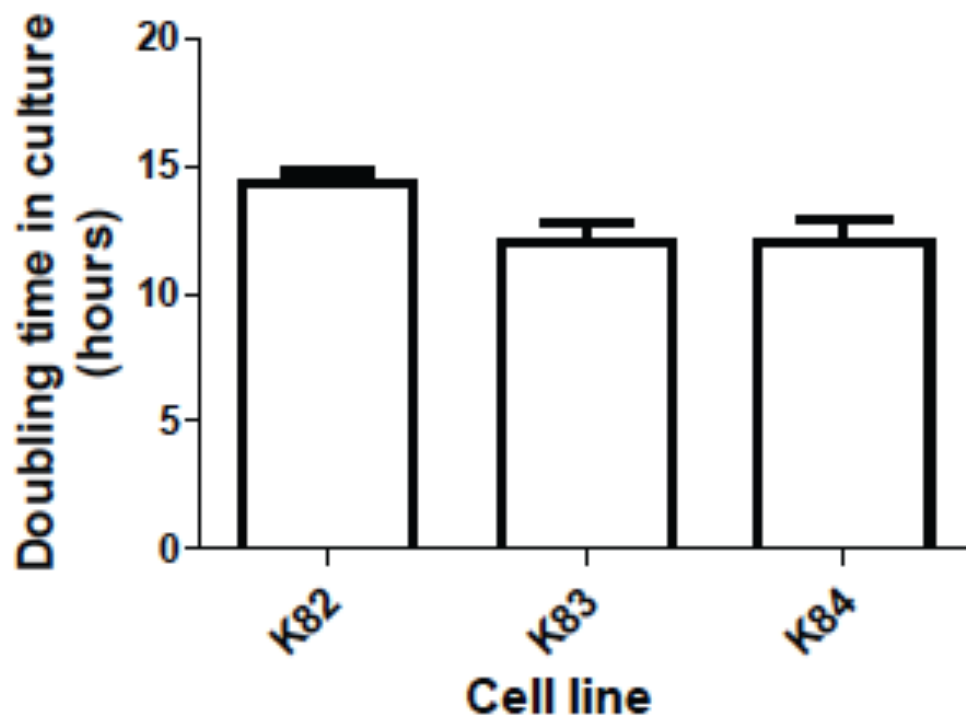
<b>Genes</b>	<b>Forward (5'-3')</b>	<b>Reverse (5'-3')</b>
<i>Gata6</i>	TCCCCTGCCGAAGTCACA	GGCCAGAGCACACCAAGAA
<i>Pou5f1</i>	TTCCCTCTGTTCCCGTCACT	TGGTGCCTCAGTTTGAATGC
<i>Gata4</i>	CCTCCCGCACGATTTCTG	CTCAGGAAAAAGAAAATCCCAAATT
<i>Sox2</i>	CTGGACTGCGAACTGGAGAAG	TTTGCACCCCTCCCAATTC
<i>Sox7</i>	CCAGGGCCCCCTTACTCA	GGCCAAGGGCTAAAGAACCT
<i>Sox17</i>	ATAAGCCCGAGATGGGTCTTC	CCGTGGCTGTCTGAGAGGTT
<i>Sall4</i>	CCCTGGGAACTGCGATGA	GAACTCGGCACAGCATTGT
<i>Lin28a</i>	AGTCTGCCAAGGGTCTGGAA	CGCTCACTCCCAATACAGAACA
<i>Albumin</i>	GAAATATGAAGCCACTCTGGAAAAG	CAAGCACTGTGCCGTAGCA
<i>Nanog</i>	AGGCCTGGACCGCTCAGT	AGTTATGGAGCGGAGCAGCAT

The cycling conditions are :

-95°C for 10 min,

-40 cycles:95°C-15 seconds;60°C-1 min

-Melting curve analysis: 95°C-15 second;60°C-1min;95° C 15 seconds for melt curve.



**Figure S1.** Doubling time of three AFCL. The difference in doubling time between cell lines was not statistically significant.

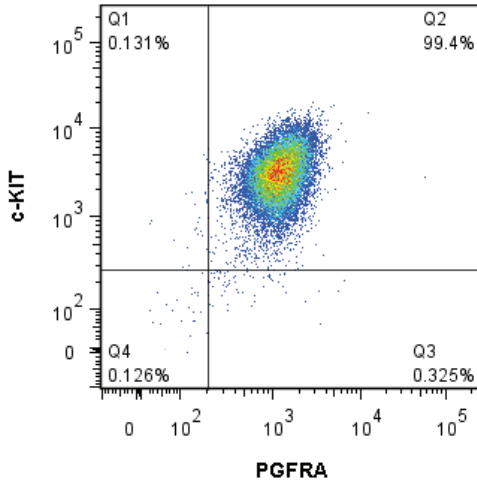
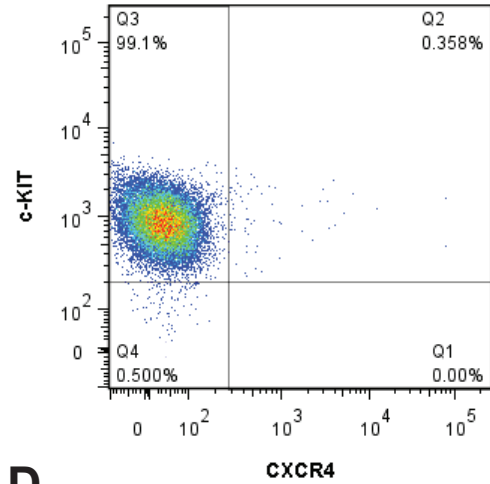
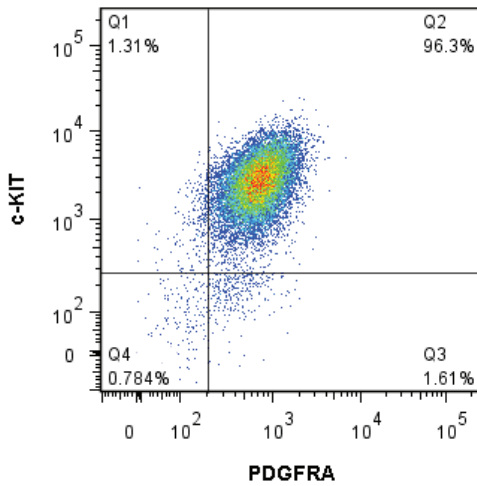
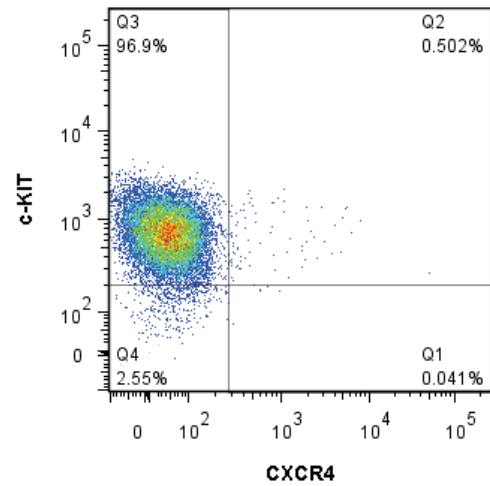
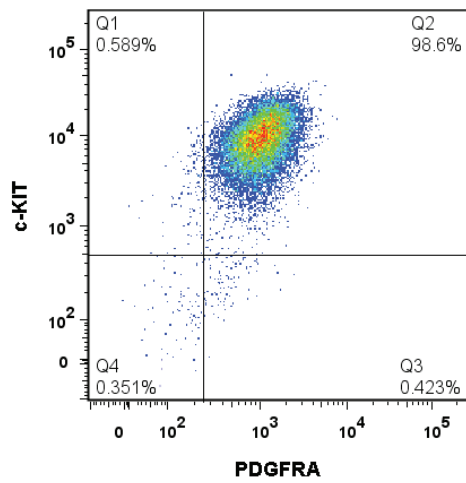
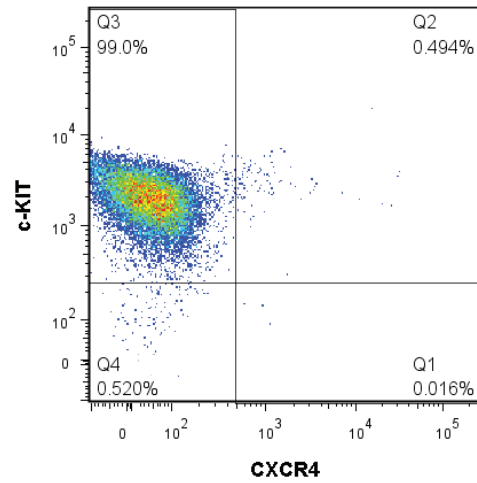
**A****B****C****D****E****F**

Figure S2. Flow cytometric analysis of c-KIT, PDGFRA and CXCR4 expression on low passage AFCL. A and B K82 passage 12; C and D K83 passage 8; E and F K84 passage 10. Approximately 19,000 DAPI negative events are shown on each graph.

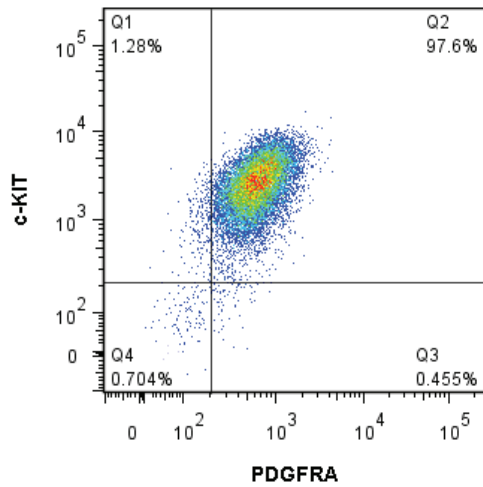
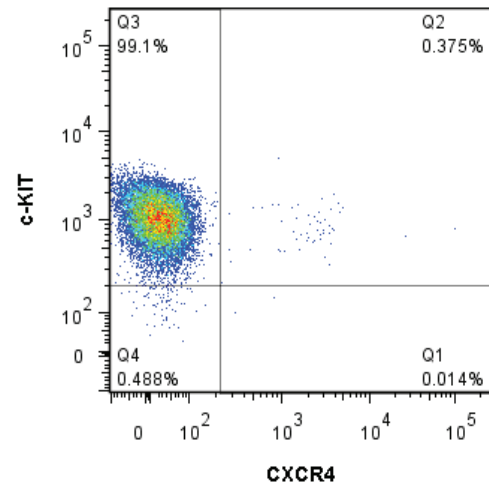
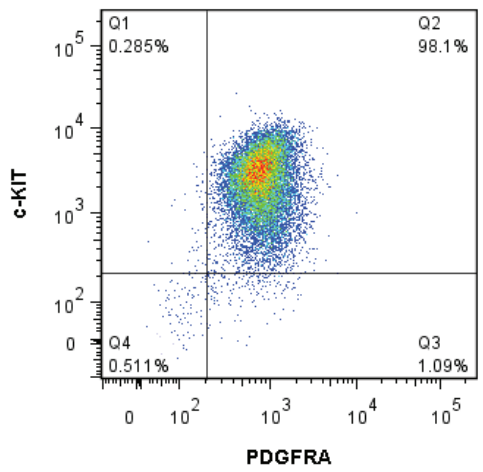
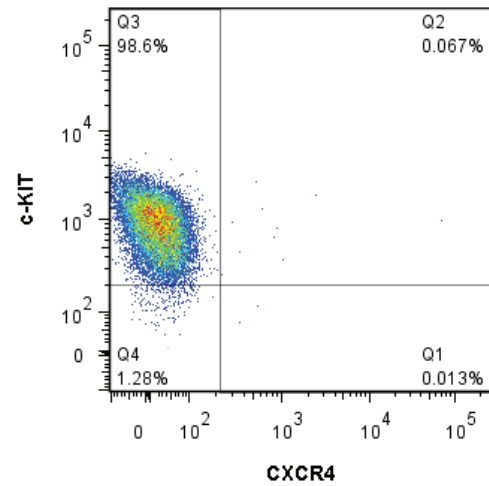
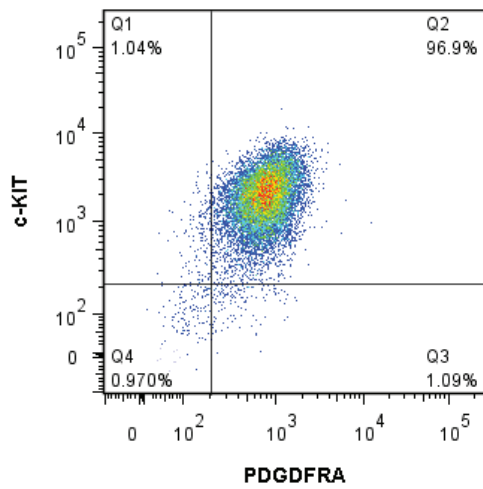
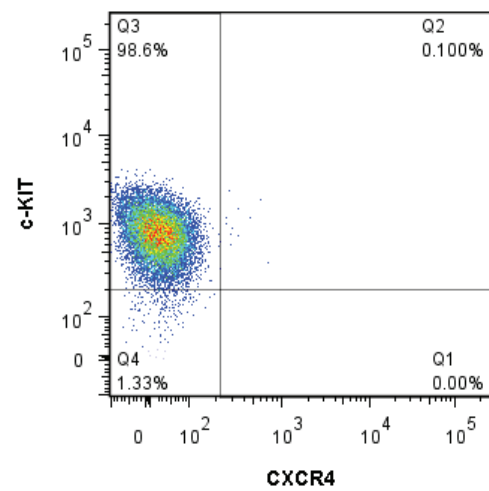
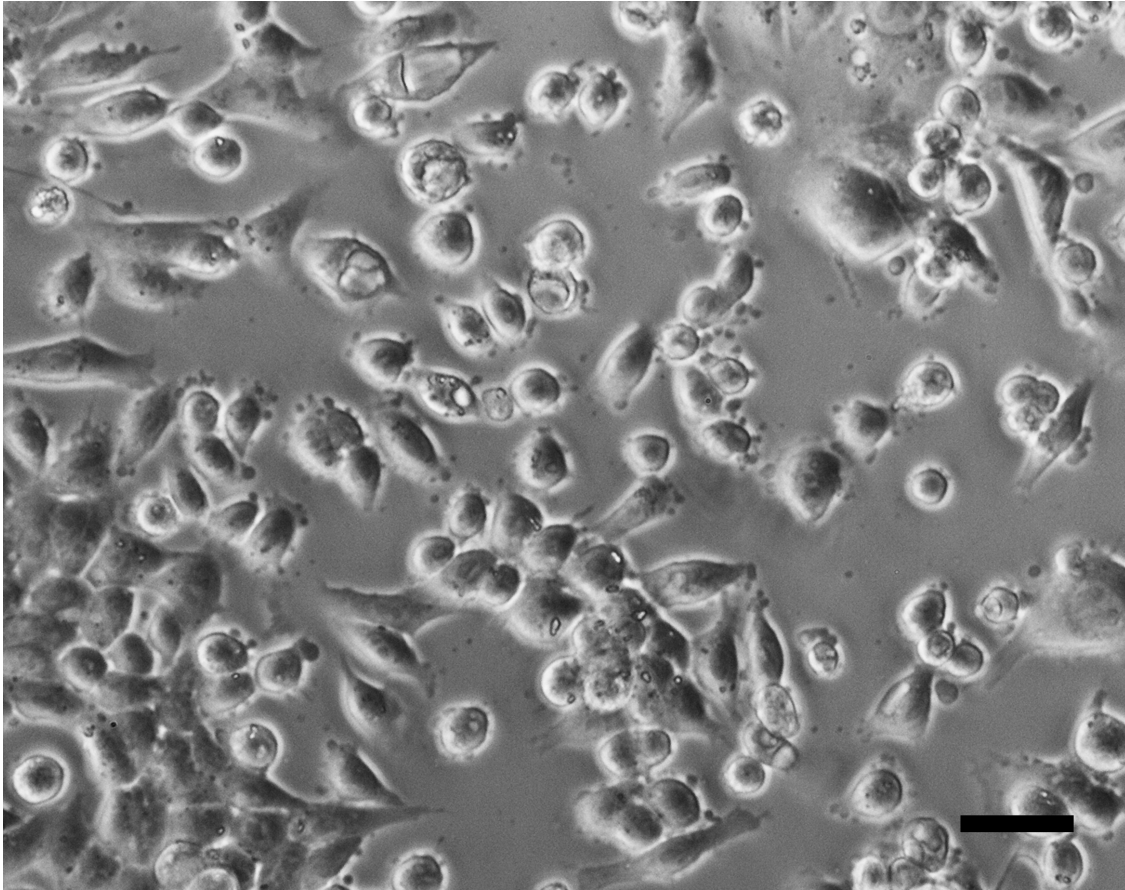
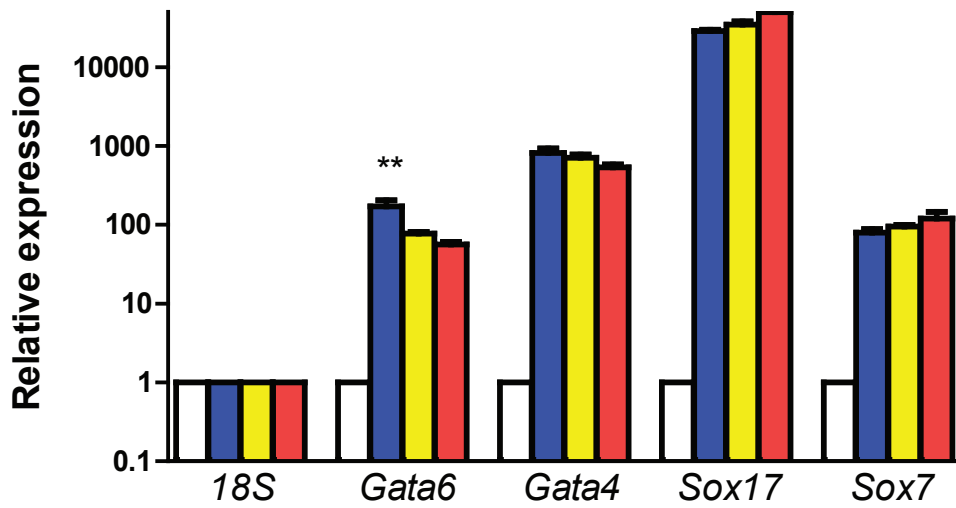
**A****B****C****D****E****F**

Figure S3. Flow cytometric analysis of c-KIT, PDGFRA and CXCR4 expression on high passage AFCL. A and B K82 passage 23; C and D K83 passage 21; E and F K84 passage 27. Approximately 14,000 DAPI negative events are shown on each graph.



**Figure S4.** AFCL-K82 grown on gelatin coated surface display various morphologies such as rounded cells and epithelial sheets; 20  $\mu\text{m}$  scale bar.

A



B

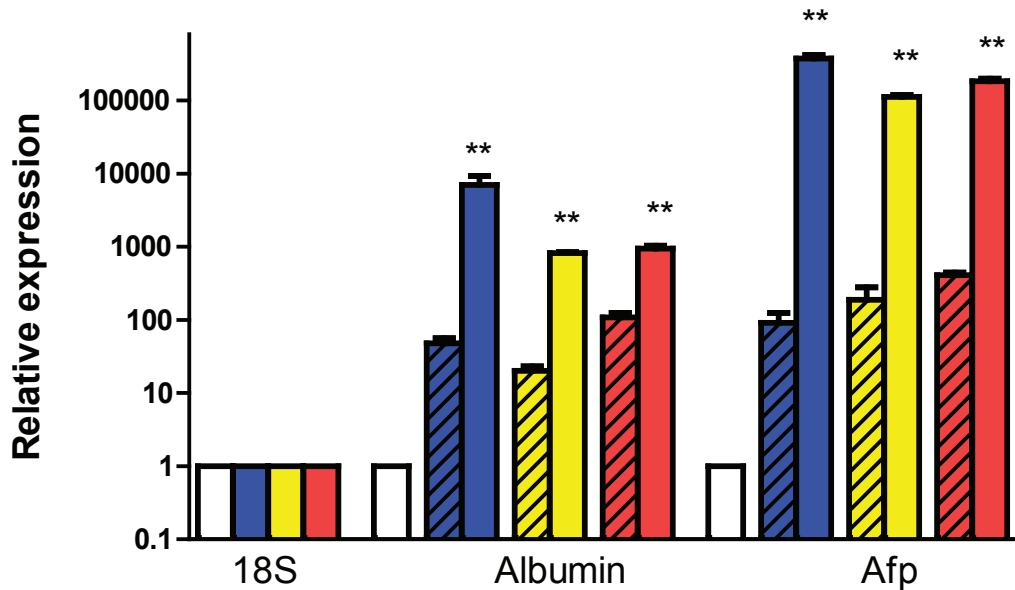


Figure S5. A) QPCR analysis of mRNA expression in AFCL grown in standard conditions. MEF (open bars), K82 (blue bars), K83 (yellow bars) and K84 (red bars). The data is normalized to mRNA expression level of each gene in CF-1 MEF. Error bars denote standard error of the mean. \*\* P<0.01 B) QPCR analysis of mRNA expression in MEF (open bars), cells before differentiation (hatched bars) and post-differentiation for 2 days in Stage I and 7 days in Stage II; MEF (open bars), K82 (open blue bars), K83 (open yellow bars), K84 (open red bars). The data is normalized to mRNA expression level of *Afp* and *Alb* in CF-1 MEFs. Error bars denote standard error of the mean. \*\* P<0.01