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2 Supplementary Material and Methods

3 Human embryonic stem cell culture of the WA09 line

The WA09 line was cultured at the University of California, San Francisco under
normoxic conditions in 76.5% DMEM/F12; 20% Knockout Serum Replacer; 0.5% LGlutamine; 1% MEM non-essential aminoacids; 2% penicillin/Streptomycin and 3.5 μl βmercaptoethanol. Cells were enzymatically passaged using Dispase and then plated onto
irradiated MEFs. Media was changed every other day and new drug was added at that
time.

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14 ELISA for bFGF secretion

15 In order to determine the levels of bFGF secretion by the feeders and hESC, we used the 16 Quantikine human bFGF ELISA kit (R&D Systems) following manufacturer's 17 instructions. This kit has been previously determined to accurately measure bFGF 18 secretion by mouse feeders and hESC [26]. MEF's were plated in CF1 media. Twenty 19 four hours later the cells were washed with PBS, and hESCs were plated in KO media 20 (plus or minus bFGF) as described above. Two days after hESC plating, the KO Media 21 was removed and fresh media (with or without Antimycin A) was added to each plate. 22 500 µl of media was collected from each well at 6 hr and 24hr and bFGF content was 23 determined by ELISA.