PCR genotyping Protocol for *Sox17$^{Cre.ERT2}$* mouse line.

PCR reagents:

1. oligo primers at 20 uM

   Sox1.S2 5'- CAGAGGTATGCAGATCTCTGT -3'
   Sox17.3'UTR 5'CATTCTGGTCAACATGTAAGGT 3'

   Sox1.S2+ Sox17.3'UTR amplification yields **660bp TM** (targeted mutant allele) band and a **526bp WT** (wild type allele) band.

2. Perkin Elmer PCR buffer with $MgCl_2$
3. 1.25mM dNTP premix (dNTP premix is made by using 100 mM NEB dNTP's. The premix contains 250 ul of each dNTP - A, C, G, &T - and 19 ml sterile water and is stable at -20°C. I freeze this in 1 ml aliquots and thaw and refreeze as needed.)
4. Perkin Elmer Amplitaq Gold
5. genomic DNA samples diluted to 50 ng/ul with sterile water

PCR reaction mixture:

15.8 ul sterile water
2.5 ul 10X PCR buffer
4 ul dNTP premix
0.75 ul primer #1
0.75 ul primer #2
1 ul dil. DNA template
0.2 ul Amplitaq Gold
25 ul total volume

Cycling conditions:

1 cycle - 94° x 6 min.
40 cycles - 94° x 1 min., 58° x 30 sec., 72° x 2 min.
1 cycle - 72° x 7 min.
hold at 4°C.

Analysis of PCR products:

Load 10 ul aliquots of reactions + 2 ul 6X gel loading buffer in a 1% mini-agarose gel (using a 15-well comb). Run gel approximately half-way down.