

GENOTYPING PROTOCOL: SOX17em3Mgn

Investigator: Mark Magnuson, VICC and DRTC **Genome Edit:** Sox17 CR2 -50 bp deletion

Allele Name: Sox17^{em3Mgn}

Primers:

Sox17CR2Fwd1: CTTGGACTTGTTCTTCAATCTTCC Sox17CR2Rev2: CCAGAGGAACTCGTAAAGCTG

PCR products:

WT = 394 bp

Sox17 -50 bp allele = 344 bp

Components	50 ul reaction	Final concentration	PCR program*
5 x Phusion Reaction Buffer (NEB #M0530S)	10.0 µL	1X	98°C, 30 seconds
10 mM dNTPs	1.0 µL	200 μΜ	98°C, 10 seconds
10 μM Sox17CR2Fwd1	2.5 µL	0.5 μΜ	62°C, 10 seconds
10 μM Sox17CR2Rev2	2.5 µL	0.5 μΜ	72°C, 40 seconds
Phusion HF DNA Polymerase	0.5 μL	0.02 U/μΙ	Go to 2, 35 X
Nuclease-free water	32.5 µL		72°C, 2 minutes
Genomic DNA	1.0 µL	Less than 1 μg, variable	4°C, ∞

