

PCR genotyping ADAMTS1 KO mice

Stephanie Lauw

Reaction Mix

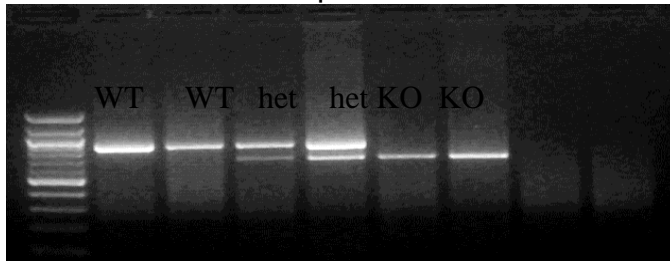
3 ul	Taq Buffer
1.5 ul	MgCl ₂
0.5 ul	20 mM dNTPs
0.5 ul	Left3 Primer 10uM
0.5 ul	PGKNeoleftPrimer 10uM
1 ul	Right1 Primer 10uM
0.5 ul	Invitrogen Taq
0.5 ul	DMSO
20 ul	H ₂ O
2 ul	DNA sample

Total: 30 ul

Make a master mix of the PCR reaction reagents from above minus the DNA sample. Example: if you have 10 DNA samples to be genotyped, multiply everything by 11 so that you have room for pipette error. Then add your DNA.

The primer set must be mixed in order to the PCR to work.

The KO band is 730 bp in size and the WT band is 844 bp in size



PCR reaction conditions:

94C 7min

94C 20sec

56C 20sec

72C 1min

(37 cycles)

72C 5min

4C ∞

1.5% agarose gel, 150V, 30 minutes

Primer Sequences:

Left3 = 5' GGC TAT TAG AGC CGC TGA TG - 3'

Right1 = 5' ATA GTG CTT TGG GGC TCC TT - 3'

NeoLeft2= 5' ATG GGC TGA CCG CTT CCT CGT - 3'

