PCR genotyping ADAMTS1 KO mice

Stephanie Lauw

Reaction Mix
3 ul Taq Buffer
1.5 ul MgCl₂
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 H₂O
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 gel image]

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'

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https://labs.feinberg.northwestern.edu/arisp/protocols-reagents/index.html