

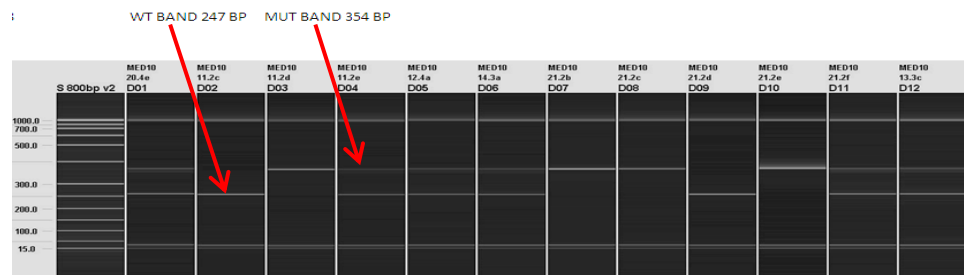


5430427ORik Genotyping Strategy

Introduction

5430427ORik is X-Linked

The gel based assays are normally run on the Qiagen QIAxcel. This is a capillary based system that provides clearer resolution and is quicker than running standard agarose gels. Different size ladders maybe loaded onto runs depending on the fragment sizes being analysed. Typically samples are run with a 50-800bp size ladder.



PCR is performed using KAPA fast Taq polymerase, although alternatives may be used.

5430427ORik gel based primers

54..Rik_RDAI_F4	CTGCAAATCCTCTGGGAAGTTTA
54..Rik_RDAI_R4	CGCCCTGTGGAGAGAAAATAAC

PCR mix

KAPA Taq PCR master mix	5µl
54..Rik_RDAI_F4	0.5µl
54..Rik_RDAI_R4	0.5µl
H ₂ O	3.0µl
DNA	1µl

60TM30FA

1. 95°C 1min.
2. 95°C 10sec.
3. 60°C 10sec.
4. 72°C 1sec.
5. Go to 2 for 29 cycles
6. 72°C 30sec.
7. 16 °C forever
8. end



Example of a 5430427ORik gel based assay (T345593)

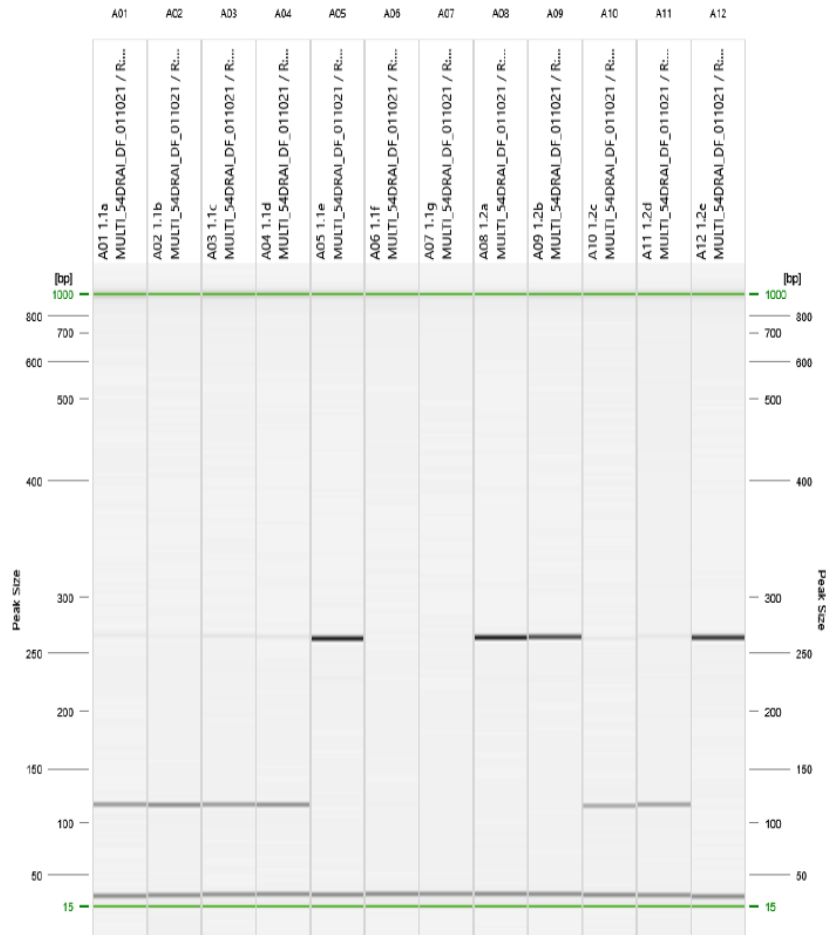


Figure: 1

WT band = No band produced

Flox band = 246 bp

Del band = 99 bp

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