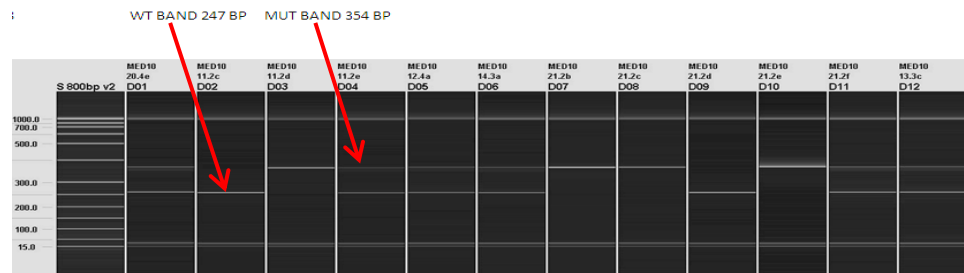




T-Cre Genotyping Strategy

Introduction

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.

T-Cre gel based primers

T-Cre_F GTGGCGAGAGAAGTGAAGGTG
T-Cre_R GACCGCAAACGGACAGAAG

PCR mix

KAPA Taq PCR master mix 5µl
T-Cre_F 0.5µl
T-Cre_R 0.5µl
H₂O 3.0µl
DNA 1

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

Mut band = 225 bp