

Allele Name: Rora Twin Strep Tag (TS)

Primer

Forward 5'-3' AGCTTAGGCAGCATCAAGTGT

Reverse 5'-3' TTTCCCCTTTGTGCCTGAACC

PCR product

Wild type: 483 bp

Knock-in mutant: 600 bp

Reagents: KAPA Genotyping Kit

PCR program

Step #	Temp °C	Time	Note
1	94	3 min	
2	94	20sec	
3	65	15sec	-0.5 C per cycle decrease
4	68	20sec	
5			repeat steps 2-4 for 10 cycles (Touchdown)
6	94	20sec	
7	60	15sec	
8	72	20sec	
9			repeat steps 6-8 for 25 cycles
10	72	10min	
11	4		hold