

## B6(Cg)-Sgta<sup>tm1.1Drmcr</sup>/J

Stock No: 029290

Protocol 22103: Probe Assay - Generic FLP

Version 6.0

### Notes

The genotyping protocol(s) presented here have been optimized for reagents and conditions used by The Jackson Laboratory (JAX). To genotype animals, JAX recommends researchers validate the assay independently upon receipt of animals into their facility. Reaction cycling temperature and times may require additional optimization based on the specific genotyping reagents used.

### Expected Results

### JAX Protocol

#### Protocol Primers

PRIMER	5' LABEL	SEQUENCE 5' → 3'	3' LABEL	PRIMER TYPE	REACTION	NOTE
oIMR1544		CAC GTG GGC TCC AGC ATT		Internal Positive Control Forward	A	
oIMR3580		TCA CCA GTC ATT TCT GCC TTT G		Internal Positive Control Reverse	A	
oIMR8940		AGT AGT GAT CAG GTA TTG CTG TTA TCT G		Forward	A	
oIMR8941		GAC TAA TGT TGT GGG AAA TTG GAG		Reverse	A	
TmoIMR0105	Fluorophore-1	CCA ATG GTC GGG CAC TGC TCA A	Quencher-1	IC Probe	A	
TmoIMR0126	Fluorophore-2	TAT ACG TTG TCC TGG CCA CGG CA	Quencher-2	Tg Probe	A	

#### Reaction A

COMPONENT	FINAL CONCENTRATION
Kapa Probe Fast QPCR	1.00 X
ddH <sub>2</sub> O	
oIMR1544	0.40 uM
oIMR3580	0.40 uM
oIMR8940	0.40 uM
oIMR8941	0.40 uM
Wt Probe	0.15 uM
Mutant Probe	0.15 uM
DNA	

#### Cycling

STEP	TEMP °C	TIME	NOTE
1	95.0	--	
2	95.0	--	
3	60.0	--	
4		--	repeat steps 2-3 for 40 cycles
5	4.0	--	Forever

JAX uses a very high speed Taq (~1000 bp/sec), use cycling times recommended for your reagents.

Endpoint Fluorescence Scatter Plot

