

## B6J.Cg-Gt(ROSA)26Sor<sup>tm96(CAG-GCaMP6s)Hze</sup>/MwarJ

Stock No: 028866

Protocol 19604: Standard PCR Assay - Gt(ROSA)26Sor<sup>tm96(CAG-GCaMP6s)Hze</sup>

Version 1.0

### 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

Mutant = 356 bp

Heterozygote = 297 bp and 356 bp

Wild type = 297 bp

### JAX Protocol

#### Protocol Primers

PRIMER	5' LABEL	SEQUENCE 5' → 3'	3' LABEL	PRIMER TYPE	REACTION	NOTE
19908		ACG AGT CGG ATC TCC CTT TG		Mutant Forward	A	
oIMR2088		AGA CTG CCT TGG GAA AAG CG		Mutant Reverse	A	
oIMR9020		AAG GGA GCT GCA GTG GAG TA		Wild type Forward	A	
oIMR9021		CCG AAA ATC TGT GGG AAG TC		Wild type Reverse	A	

#### Reaction A

COMPONENT	FINAL CONCENTRATION
ddH <sub>2</sub> O	
Kapa 2G HS buffer	1.30 X
MgCl <sub>2</sub>	2.60 mM
dNTP KAPA	0.26 mM
19908	0.50 uM
oIMR2088	0.50 uM
oIMR9020	0.50 uM
oIMR9021	0.50 uM
Glycerol	6.50 %
Dye	1.00 X
Kapa 2G HS taq polym	0.03 U/ul
DNA	

#### Cycling

STEP	TEMP °C	TIME	NOTE
1	94.0	--	
2	94.0	--	
3	65.0	--	-0.5 C per cycle decrease
4	68.0	--	
5		--	repeat steps 2-4 for 10 cycles (Touchdown)
6	94.0	--	
7	60.0	--	
8	72.0	--	
9		--	repeat steps 6-8 for 28 cycles
10	72.0	--	
11	10.0	--	hold

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

JAX uses a 'touchdown' cycling protocol and therefore has not calculated the optimal annealing temperature for each set of primers.

