

## C57BL/6J-Mc1r<sup>e</sup>/J

Stock No: 000060

Protocol 22000: End Point Analysis Assay - Mc1r<e>

Version 3.0

### 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 = -/-

Heterozygote = C/-

Wild type = C/C

### JAX Protocol

#### Protocol Primers

PRIMER	5' LABEL	SEQUENCE 5' → 3'	3' LABEL	PRIMER TYPE	REACTION	NOTE
18816		CCA GCA CCC TCT TTA TCA CC		Forward	A	
18817		AGA AAG TGA CGA GGC AGA GC		Reverse	A	
18818	Fluorophore-1	TAC TAC AAG CAC ACA GCC GTT CT	Quencher-1	WT Probe		
18820	Fluorophore-2	CTA CTA CAA GCA ACA GCC GTT CTG	Quencher-2	MUT Probe		

#### Reaction A

COMPONENT	FINAL CONCENTRATION
Kapa Probe Fast QPCR	1.00 X
ddH <sub>2</sub> O	
18816	0.40 uM
18817	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.