

## Genotyping protocol

EMMA ID: EM:09863

Gene symbol: 2810006K23Rik

Strain designation: C57BL/6N-A<sup>tm1Brd</sup> 2810006K23Rik<sup>tm1(KOMP)Mbp</sup>/Oulu

### Primers:

Primer name	Primer sequence (5'- 3')
23Rik -F2	CAC ATA CCC TAC TCC TCT AAG
23RikR2	CCA TGT CCT TTC ACG AAC TGT T
Construct primer	TCG TGG TAT CGT TAT GCG CC

### Products:

Allele	Primer 1	Primer 2	Product size (bp)
Wild-type	23Rik -F2	23RikR2	555
Mutant	23Rik -F2	Construct primer	335

### Reaction mix:

Component	Volume (µl)
Genomic DNA (~50-100 ng)	2.0
Primer 1 (10 µM)	1.0
Primer 2 (10 µM)	1.0
10x Buffer (15 mM MgCl <sub>2</sub> )	3.0
MgCl <sub>2</sub> (25 mM)	1.0
dNTPs (25 mM)	0.25
DNA polymerase (5 U/µl)	0.05
H <sub>2</sub> O	21.7
<b>Total</b>	<b>30.0</b>

### PCR program:

Time	Temperature (+ °C)	Cycles
5 min	95	
1 min	94	30
1 min	60	
1 min	72	
7 min	72	
∞	4	