

## Genotyping protocol

EMMA ID: EM:10723

Gene symbol: Kihl4

Strain designation: STOCK Kihl4<sup>tm1Ali</sup>/Oulu

### Primers:

Primer name	Primer sequence (5'- 3')
EM10723-A	CGGCAGCTTTCTGGCTTCAGCTAG
EM10723-B	AACTCTTTCTTGCCGGACAC

### Products:

Allele	Primer 1	Primer 2	Product size (bp)
Wild-type	EM10723-A	EM10723-B	233
Mutant	EM10723-A	EM10723-B	313

### Reaction mix:

Component	Volume (µl)
Genomic DNA (~50-100 ng)	2.0
Primer 1 (10 µM)	1.5
Primer 2 (10 µM)	1.5
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)	0.1
H <sub>2</sub> O	20.65
<b>Total</b>	<b>30.0</b>

### PCR program:

Time	Temperature (+ °C)	Cycles
5 min	95	
1 min	94	30
1 min	62	
1 min	72	
10 min	72	
∞	10	