

Genotyping protocol

EMMA ID: EM:10720

Gene symbol: miR455

Strain designation: 7C6 miR455 het

Primers:

Primer name	Primer sequence (5'- 3')
miR455F	CAGCAGAGGAGTCTCTAACC
miR455R	GTGCCAAAGGGAGGAGAGAG

Products:

Allele	Primer 1	Primer 2	Product size (bp)
Wild-type	miR455F	miR455R	180
Mutant	miR455F	miR455R	210

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 ₂)	3.0
MgCl ₂ (25 mM)	1.0
dNTPs (25 mM)	0.25
DNA polymerase (5 U/μl)	0.05
H ₂ O	21.7
Total	30.0

PCR program:

Time	Temperature (+ °C)	Cycles
5 min	95	
1 min	95	35
30 sec	57	
1 min	72	
10 min	72	
∞	4	