

Genotyping protocol

EMMA ID: EM:11029

Gene symbol: Col18a1

Strain designation: FVB.129S4-Col18a1^{tm1Hms}/Oulu

Primers:

Primer name	Primer sequence (5'- 3')
Col XVIII-1	CTC TGT AGG GTC CTT ATG GAC G
Col XVIII-3	CCT TTA AGC CAT TCG TTC CAG G
MSRneo	AGG AGT AGA AGG TGG CGC GAA GG

Products:

Allele	Primer 1	Primer 2	Product size (bp)
Wild-type	Col XVIII-1	Col XVIII-3	435
Mutant	Col XVIII-1	MSRneo	600

Reaction mix:

Component	Volume (µl)
Genomic DNA (~50-100 ng)	1.0
Primer 1 (10 µM)	1.0
Primer 2 (10 µM)	1.0
10x Buffer (15 mM MgCl ₂)	3.0
dNTPs (25 mM)	0.25
DNA polymerase (5 U/µl)	0.1
H ₂ O	23.65
Total	30.0

PCR program:

Time	Temperature (+ °C)	Cycles
4 min	94	
1 min	94	30
1 min	60	
1 min	72	
10 min	72	
∞	10	