

Genotyping protocol

EM:11843

B6.Cg-Itga11tm1Dgul/Kctt

Primers

Ko-wt/F	5' CCA TCA GAA GAC AGG AGA CGT ATA ACA 3'
Ko-wt/R	5' TGG TCA GTG GAT GGG TTA GGA AG 3'
LacZ1	5' GTG GTG GTT ATG CCG ATC GC 3'
LacZ2	5' TAC CAC AGC GGA TGG TTC GG 3'

PCR profile

98 °C, 30s	
98 °C, 10 s	35 cycles
58 °C, 15 s	
72 °C, 30s	
72 °C, 5 min	
4 °C, ∞	

PCR mix

5 x PCR Buffer	5.0 µl
dNTP mix (10 mM of each nucleotide)	0.5 µl
ko-wt/F or LacZ1 (100 µM)	0.125 µl
ko-wt/R or LacZ2 (100 µM)	0.125 µl
Platinum SuperFipolymerase (2 U/µl)	0.25 µl
DNA template (~ 0.5 µg tail DNA)	2.0 µl
H ₂ O	<u>16 µl</u>
	24 µl

Post-PCR analysis

1.5% agarose gel.
 Expected pattern is wt:688bp, mut:374bp