

Genotyping protocol

EM:09220

B6.129-Grin1^{tm1Rsp}/Kctt

Primers

N1ex18do1 5' CTG GGA CTC AGC TGT GCT GG 3'

N1in18up 5' AGG GGA GGC AAC ACT GTG GAC 3'

PCR profile

95 °C, 5 min

94 °C, 30 s 35 cycles

55 °C, 45s

72 °C, 45 s

72 °C, 10 min

4 °C, ∞

PCR mix

10 x PCR Buffer II (Life technologies)	3.0 µl
MgCl ₂ (25 mM)	1.8 µl
dNTP mix (25 mM of each nucleotide)	0.24 µl
N1ex18do1 (100 µM)	0.15 µl
N1in18up (100 µM)	0.15 µl
AmpliTaq Gold (5 U/µl)	0.15 µl
DNA template (~ 0.5 µg tail DNA)	2.0 µl
H ₂ O	<u>22.51 µl</u>
	30 µl

Post-PCR analysis

3% agarose gel.

Expected pattern is **wt: 455bp**
Tg: 500bp