

# Genotyping protocol

EM:12708

**C57BL/6-Tg(Gpr101-cre)BKldr/Kctt**

## Primers

Chrna2\_fwr            5' GAC AGC CAT TTT CTC GCT TC 3'  
Chrna2\_rev            5' AGG CAA ATT TTG GTG TAC GG 3'

## PCR profile

95 °C, 5 min  
  
95 °C, 30 s  
55 °C, 45 s                    35 cycles  
72 °C, 1 min  
  
72 °C, 10 min  
4 °C, ∞

## PCR mix

10 x PCR Buffer II (Life technologies)	2.0 µl
MgCl <sub>2</sub> (25 mM)	1.2 µl
dNTP mix (25 mM of each nucleotide)	0.16 µl
Chrna2_fwr (100 µM)	0.1 µl
Chrna2_rev (100 µM)	0.1 µl
AmpliTaq Gold (5 U/µl)	0.13 µl
DNA template (~ 0.5 µg tail DNA)	1.0 µl
H <sub>2</sub> O	<u>15.31 µl</u>
	20 µl

## Post-PCR analysis

1.5% agarose gel.  
Expected pattern is Tg:210bp