PROTOCOL FOR TetO-Cre MOUSE GENOTYPING

Procedure

Genotyping of offspring from tetO-Cre breeding colony is based on PCR.

PCR primers

5' forward primer (Cre2) 5´ **ggt tag cac cgc agg tgt ag** 3´
3' reverse primer (JL23) 5´ **cac atc tag act aat cgc cat ctt cca gca g** 3´

PCR profile – TEDX3

94 ˚C, 5 min

94 ˚C, 45 s — 35 cycles
56 ˚C, 45 s
72 ˚C, 1 min

72 ˚C, 10 min
4 ˚C, ∞

PCR mix

10 x PCR Gold buffer (Perkin Elmer)	3.0 µl
$MgCl_2$ (25 mM)	2.5 µl
dNTPs (10 mM)	0.5 µl
JL23 (20 µM)	0.5 µl
Cre2 (20 µM)	0.5 µl
AmpliTaq Gold (5 U/µl)	0.2 µl
DNA template (~ 0.5 µg tail DNA)	2.0 µl
ddH_2O	20.8 µl
	30 µl

Post-PCR analysis

Load 10 µl of the PCR reaction on a 1 % agarose gel.
Expected results; one band – 430 bp.