PROTOCOL FOR WAP-SVT MOUSE GENOTYPING

Procedure

Genotyping of offspring from WAP-SVT breeding colony is based on PCR.

PCR primers

5' forward primer (WAPSVt-1); 5´ **gct ttg caa aga tgg ata aag** 3´
3' reverse primer (WAPSVt-2); 5´ **act aaa cac agc atg act c** 3´

PCR profile – WAP

95 ˚C, 10 min

95 ˚C, 45 s 40 cycles
55 ˚C, 45 s
72 ˚C, 1min

72 ˚C, 10 min
4 ˚C, ∞

PCR mix

10 x PCR Gold buffer (Perkin Elmer)	3.0 µl	
$MgCl_2$ (25 mM)	2.0 µl	
dNTPs (10 mM)	0.5 µl	
WAPSVt-1 (20 µM)	0.5 µl	
WAPSVt-2 (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		21.3 µl
	30 µl	

Post-PCR analysis

Load 10 µl of the PCR reaction on a 1 % agarose gel.
Expected results; one band – 813 bp fragment.