

Genotyping protocol

General information:

Strain name	B6.129S2-Cdkn1a<tm1Tyj>/RvOCnbc
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Primers:

Name	Sequence	Primer type
Primer 1 (p21+1116F)	AAG CCT TGA T TC TGA TGT GGG C	
Primer 2 (p21 -135)	TGA CGA AGT CAA AGT TCC ACC G	
Primer 3 (Neo191)	GCT ATC AGG ACA TAG CGT TGG C	

In case more than two primers are introduced, please indicate how they should be combined:

	Forward primer	Reverse primer
wt	Primer 1 (p21+1116F)	Primer 2 (p21 -135)
mutant	Primer 1 (p21+1116F)	Primer 3 (Neo191)

Reaction mix:

ddH ₂ O	18,5	μl
PCR Buffer 15mM MgCl ₂	2,5	μl
dNTPs 2,5mM	2,5	μl
Primer 100μM	0,125	μl
Primer 100μM	0,125	μl
Taq Polymerase 5U/μl	0,250	μl
DNA	1	μl
Final volume	25	μl

PCR program:

94	°C	4	min	X40
94	°C	60	sec	
64	°C	60	sec	
72	°C	120	sec	
72	°C	10	min	

Expected fragment size:

wt	900	bp
mutant	750	bp

Comments/Additonal information:

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