

Genotyping protocol

General information:

Strain name	Rnf2 ^{tm1Mvi}
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Primers:

Name	Sequence	Primer type
1551	TAGGGATGGAGAGAGGGAGG	gene-specific
1552	TCTGATGGTAACAGAGAGCCAG	gene-specific
1553	GACATAGCGTTGGCTACCCG	selection cassette

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

	Forward primer	Reverse primer
wt	1551	1552
mutant	1553	1552

Reaction mix:

10 x buffer	2	μl
MgCl ₂ 50 mM	0,6	μl
dNTPs 5 mM	0,8	μl
Primer 1551 2.5 uM	1	μl
Primer 1552 10 uM	1	μl
Primer 1553 10 uM	1	μl
Tail DNA 1 :10	1	μl
H ₂ O	12,1	μl
Taq 1u/ul	0,5	μl
Final volume	20	μl

PCR program:

94	°C	4	min	X 34
94	°C	30	sec	
60	°C	20	sec	
72	°C	60	sec	
72	°C	2	min	

Expected fragment size:

wt	384	bp
mutant	784	bp

Comments/Additonal information:

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