

Genotyping protocol

General information:

Strain name	ODB
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Primers:

Name	Sequence	Primer type
5'ROSA	tatcagtaaggagctgcagtg	please select one
3'ROSA	acccagatgactacctatctc	please select one
LoxI2-R1	cccactggccttcgtagtagac	please select one
		please select one

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

	Forward primer	Reverse primer
e.g. wt	5'ROSA	3'ROSA
e.g. mut	5'ROSA	LoxI2-R1

Reaction mix:

Enzyme	10	μl
Forward primer (10uM)	1	μl
Reverse primer (10uM)	1	μl
Water	6	μl
DNA	2	μl
		μl
		μl
		μl
Final volume		μl

PCR program:

95	°C	5	min	X30
95	°C	30	sec	
62	°C	30	sec	
72	°C	30	sec	
72	°C	2	min	

Expected fragment size:

wt	300	bp
mutant	750	bp

Comments/Additonal information:

Use different PCR reactions for each allele.
Check Martin et al., 2015 (EMBO J)