

# Genotyping protocol

## General information:

Strain name	Cnbc:NMRI-conls
-------------	-----------------

## Primers:

Name	Sequence	Primer type
Fw	CATCGAGACCAAGTTTTCTG	
Rev	ACCATGTCGTAGGCACTGAG	

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

	Forward primer	Reverse primer
Mutant	Fw	Rev

## Reaction mix:

ddH <sub>2</sub> O	18,5	μl
PCR Buffer 15mM MgCl <sub>2</sub>	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	X36
94	°C	15	sec	
51	°C	60	sec	
72	°C	60	sec	
72	°C	10	min	

## Expected fragment size:

wt	bp
mutant	bp

## Comments/Additonal information:

PCR followed by an MseI restriction digest. Digesting PCR products with MseI generates a single band of 362 bp.