

STRAIN NAME:**NGF1KO****REAGENTS AND SOLUTIONS:**

- Taq polymerase 10X Buffer (1,5 mM MgCl₂) (MP QBiogene)
- dNTP (MPQ Biogene)
- Primers (Invitrogen)
- Taq polymerase (MPQBiogene)
- Mineral oil (EUROBIO)

REACTION MIX:

- Taq Buffer 1X
- dNTP (0,4 mM total)
- primer NGF1 for (0,60μM)
- primer NGF1 rev (1,20μM)
- primer NGF1 KO (0,60μM)
- Taq polymerase (40U/ml)
- DNA

PROGRAM:

5 min	94°C	35 cycles
30 sec	94°C	
30 sec	60°C	
1 min	72°C	
5 min	72°C	

MIGRATION :

Migration of the samples on a 1.5% agarose gel.

PRIMERS SEQUENCES

- Primer NGF1 for 5' CAGAATAATTTCCGGCAGAGACCGG 3'
- Primer NGF1 rev 5' AGCCAAAGAAGGAAGGTAAGCAACGAA 3'
- Primer NGF1 KO 5' GCGGTCTCTGTGGTCCTTGTAAGAGTCT 3'

AMPLIFICATION

Mutant allele : 1000 bp
WT allele : 444 bp