

**STRAIN NAME:****NGF5KO****REAGENTS AND SOLUTIONS:**

- Taq polymerase 10X Buffer (1,5 mM MgCl<sub>2</sub>) (MP QBiogene)
- dNTP (MPQ Biogene)
- Primers (Invitrogen)
- Taq polymerase (MPQBiogene)
- Mineral oil (EUROBIO)

**REACTION MIX:**

- Taq Buffer 1X
- dNTP (0,4 mM total)
- primer NGF5F (0,24 μM)
- primer NGF5R (0,60 μM)
- primer NGF5KO (0,36 μM)
- Taq polymerase (40U/ml)
- DNA

**PROGRAM:**

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

**MIGRATION :**

Migration of the samples on a 1.5% agarose gel.

**PRIMERS SEQUENCES**

- Primer NGF5F 5' CGGTTGGTGGGATTCCTGTGC3'
- Primer NGF5R 5' GCAGCCAGGGGTGATGATAACAGG3'
- Primer NGF5KO 5' CTGGGTGTGGATGGTTTGGGTATACG3'

**AMPLIFICATION**

Transgene : bp  
Mutant allele : 700 bp  
WT allele : 379 bp