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| PCR LTa TNFa |
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1- PCR mix

2,50 µl 10X buffer(Q BIOgene ; 1,5 mM MgCl₂)
 0,50 µl dNTP (20 mM total)
 0,15 µl primer TNF3 (200 ng/µl)
 0,65 µl primer TNF4 (200 ng/µl)
 0,50 µl primer neo2 (200 ng/µl)
 0,20 µl Taq polymerase (Q BIOgene ; 5U/µl)
 19,50 µl distilled water

2- PCR reaction :

24,0 µl mix
 1,0 µl DNA (200ng/µl)

3- PCR program :

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|-------|-------|-----------|
| 5 min | 94°C | |
| 1 min | 94°C | |
| 1 min | 60 °C | 35 cycles |
| 2 min | 72°C | |
| 5 min | 72°C | |

4- Primers:

- Primer TNF3 5' TTCGAGTGACAAGCCTGTAG 3'
 - Primer TNF4 5' TCATACCAGGGTTTGAGCTC 3'
 - Primer neo 2 5' AAGGTGAGATGACAGGAGATC 3'

5- Amplifications:

wild-type allele: 618 bp
 mutant allele: 880 bp