

PCR FOR TNF k/I (A86)

PCR REACTION:

10X PCR buffer	2 λ
MgCL2 25mM	Depends on the taq polymerase 2 λ
dNTP'S	1.5 λ
Primer A	5 μ mol per reaction 1 λ
Primer B	5 μ mol per reaction 1 λ
Taq	Variable -> 0,4 λ
DNA	1 λ
H2O	Up to volume -> 11,1 λ

Primer A: 5'-TAA TGG GCA GGG CAA GGT GG-3'

Position in relating gene:4799 4818 in mmTNFAB pc/gene

Primer B: 5'-TTC ACT TCC GGT TCC TGC ACC CT-3'

Position in relating gene:5695 5717 in mmTNFAB pc/gene

PCR CONDITIONS:

1. 95C for 3min
2. 95C for 30sec
3. 60C for 30sec
4. 72C for 1min
5. Step 2 for 29 more cycles.
6. 72C for 5min
7. 12C for 10min
8. End.

PCR PRODUCT: wild type band: 930bp
Transgenic allele band: 800bp

