

PCR for tmTNF(k/i)

PCR REACTION:

10x PCR buffer	2 λ
MgCl ₂ 25mM	depends on the Taq polymerase
dNTP's 2.5mM	1 λ
Primer A (78)	5 pmol / reaction
Primer B (79)	5 pmol / reaction
Taq polymerase	Variable
DNA	1 λ
milliQ H2O	Up to volume

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) 94.0, 2min
 - 2) 95.0, 1 min
 - 3) 65.0, 1 min
 - 4) 72.0, 1 min
 - 5) GOTO 2, for 29 cycles
 - 6) 72.0, 10 min
 - 7) 20.0, 1min
- END

PCR PRODUCT:

wild type band: 2kb

k/i band: 2.1kb