

## PCR for Tg ColVI Cre

### PCR REACTION:

10xPCR buffer	2µl
MgCl <sub>2</sub> 25mM	1,2µl
dNTP's 2.5mM	1µl
Primer A (324)	5 pmol / reaction
Primer B (325)	5 pmol / reaction
DNA	1µl
Taq polymerase	0,4µl
milliQ H <sub>2</sub> O	Up to volume (20λ)

**Primer A (sense):** 5'-att-acc-ggt-cga-tgc-aac-gag-t-3'

**Position in relating gene:** (bacteriophage P1 cre recombinase )68-80\*

**Primer B (antisense):** 5'-cag-gta-tct-ctg-acc-aga-gtc-a-3'

**Position in relating gene:**(bacteriophage P1 cre recombinase )852-873\*  
(\*starting point: atg of cre recombinase)

### PCR CONDITIONS:

1. 94<sup>0</sup>C for 5 min.
2. 94<sup>0</sup>C for 1 min.
3. 57<sup>0</sup>C for 1 min.
4. 72<sup>0</sup>C for 1 min.
5. Steps 2-4 for 5 more cycles.
6. 94<sup>0</sup>C for 10 sec.
7. 55<sup>0</sup>C for 40 sec.
8. 72<sup>0</sup>C for 1:30 min.
9. Steps 6-8 for 25 more cycles.
10. 72<sup>0</sup>C for 10 min.
11. 16<sup>0</sup>C for 1 min.
12. End

**PCR PRODUCT:** Transgene band: 813bp.