# ***PCR FOR CRE (common)***

## PCR REACTION:

| | |
|---|---|
| 10X PCR buffer | **2ul** |
| $MgCL_2$ 25 mM | Depends on Taq polymerase **(usually 2ul)** |
| dNTP'S 2,5 mM | 1ul |
| Primer A | 5 pmol per reaction **(1ul from 5pmole/ul stock)** |
| Primer B | 5 pmol per reaction **(1ul from 5pmole/ul stock)** |
| DNA | 1ul |
| Taq | Variable **(0.4ul)** |
| dd.$H_2O$ | Up to volume (20λ) **11.6ul** |

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

PCR PRODUCT: Transgene band: ~800bp.

## PHOTO EXAMPLE: