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) → 30: 5'-att-acc-ggt-cga-tgc-aac-gag-t-3'
Position in relating gene: (bacteriophage P1 cre recombinase)68-80*
Primer B (antisense) → 31: 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. 94^0C for 5 min.
2. 94^0C for 1 min.
3. 57^0C for 1 min.
4. 72^0C for 1 min.
5. Steps 2-4 for 5 more cycles.
6. 94^0C for 10 sec.
7. 55^0C for 40 sec.
8. 72^0C for 1:30 min.
9. Steps 6-8 for 25 more cycles.
10. 72^0C for 10 min.
11. 16^0C for 1 min.
12. End

PCR PRODUCT: Transgene band: ~800bp.
Samples:

PHOTO EXAMPLE: