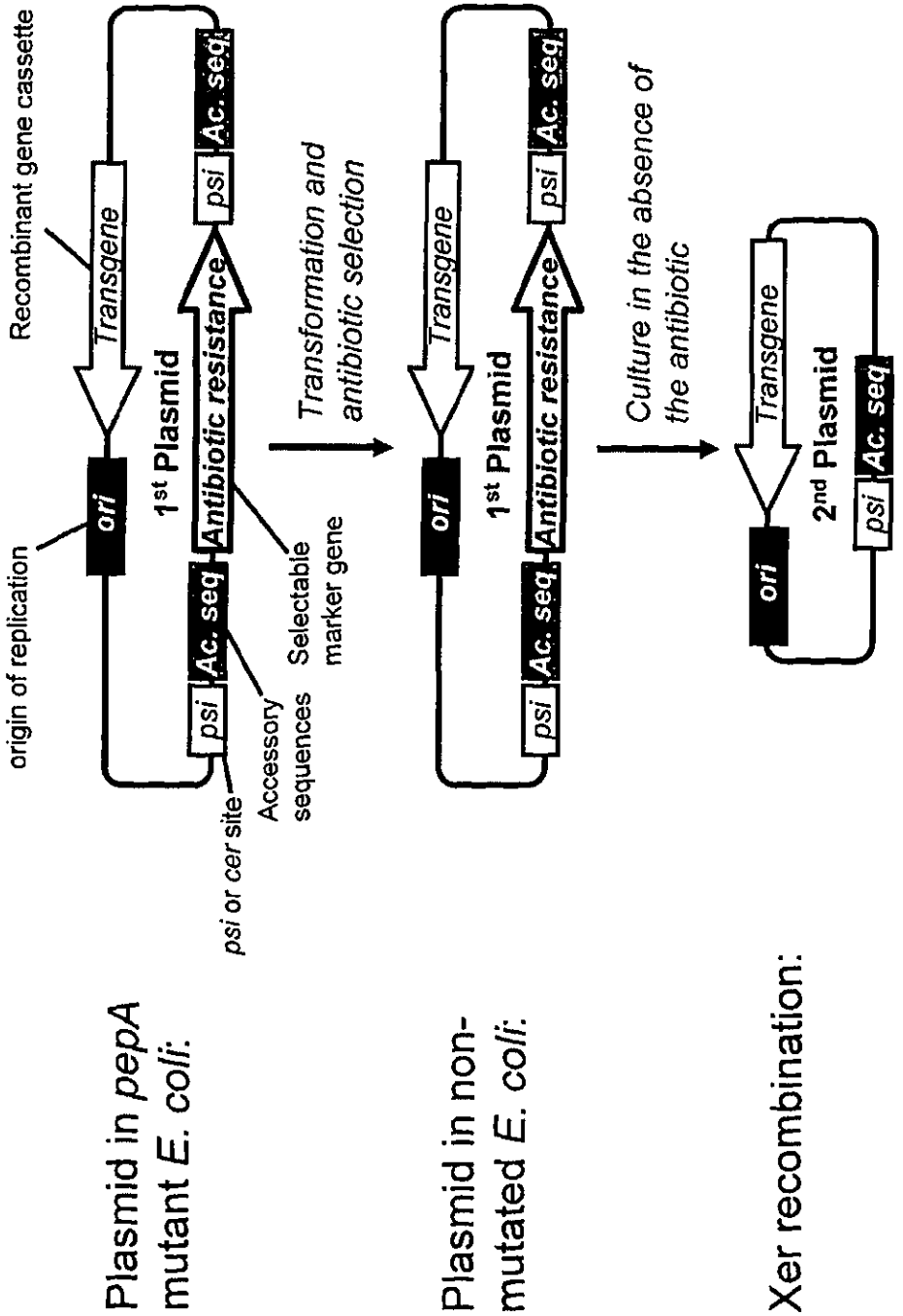

Recombinant gene cassette
origin of replication
Transgene
ori
1st Plasmid
Antibiotic resistance
psi
Ac. seq.
Selectable marker gene
Accessory sequences
psi or cer site
Plasmid in pepA mutant E. coli:
Transformation and antibiotic selection
Plasmid in non-mutated E. coli:
Culture in the absence of the antibiotic
Xer recombination:
2nd Plasmid

Figure 1

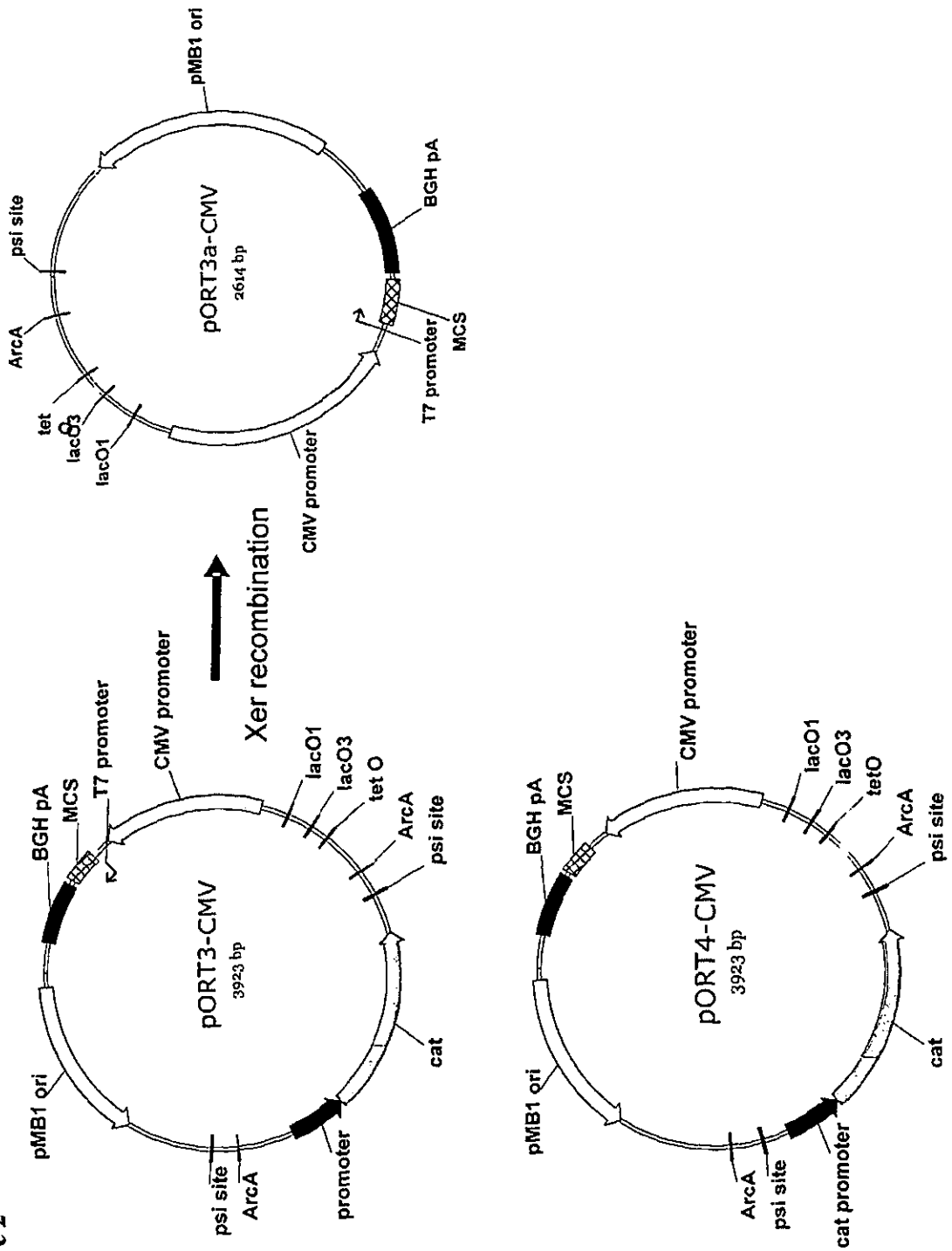

pMB1 ori
BGH pA
psi site
ArcA
tet
lacO3
lacO1
pORT3a-CMV
2614 bp
T7 promoter
MCS
CMV promoter
Xer recombination
BGH pA
MCS
T7 promoter
CMV promoter
lacO1
lacO3
tet O
ArcA
psi site
pMB1 ori
pORT3-CMV
3923 bp
cat
psi site
ArcA
promoter
BGH pA
MCS
CMV promoter
lacO1
lacO3
tetO
ArcA
psi site
pMB1 ori
pORT4-CMV
3923 bp
cat
ArcA
psi site
cat promoter

Figure 2

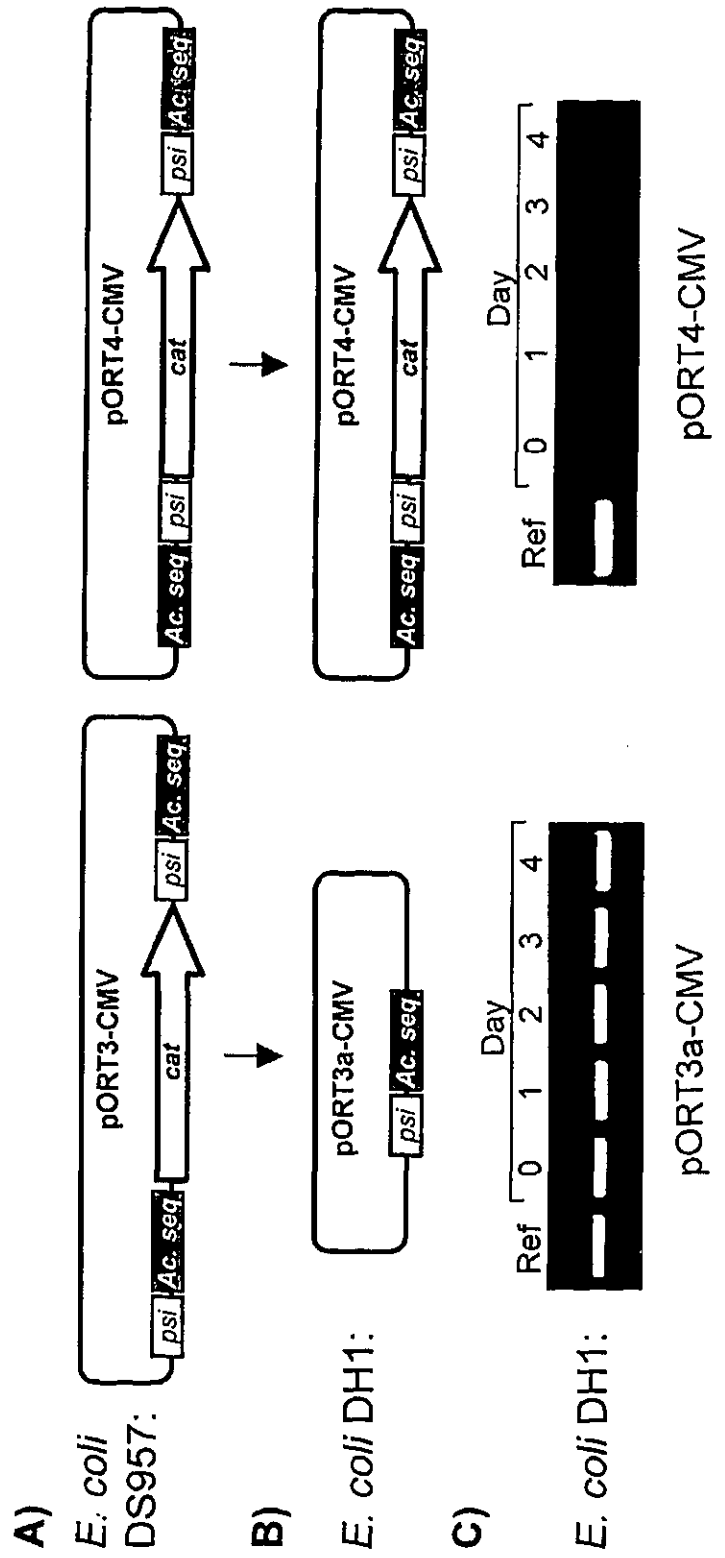

A)
E. coli DS957:
pORT3-CMV
psi
Ac. seq
cat
psi
Ac. seq
pORT4-CMV
psi
Ac. seq
cat
psi
Ac. seq
B)
E. coli DH1:
pORT3a-CMV
psi
Ac. seq
pORT4-CMV
psi
Ac. seq
cat
psi
Ac. seq
C)
E. coli DH1:
Day
Ref
0
1
2
3
4
pORT3a-CMV
Day
Ref
0
1
2
3
4
pORT4-CMV

Figure 3

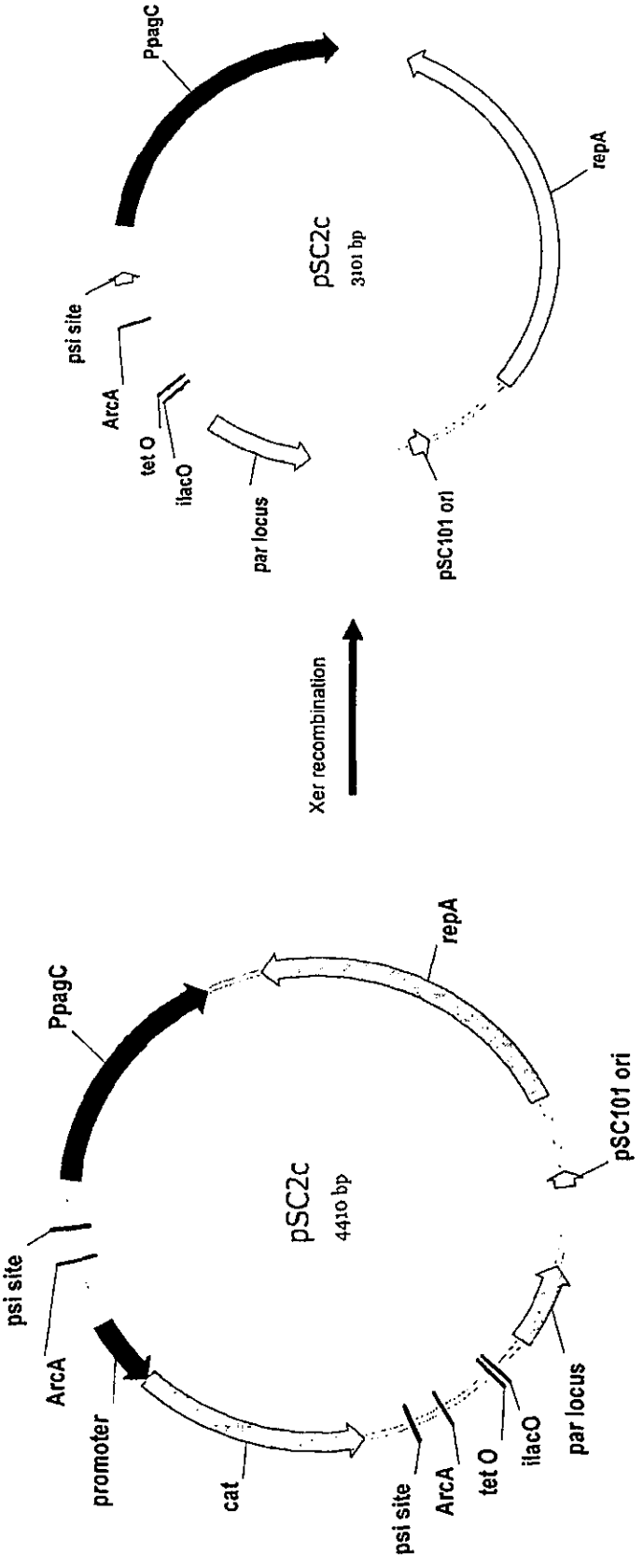

PpagC
repA
psi site
ArcA
tet O
ilacO
par locus
pSC2c
3101 bp
pSC101 ori
Xer recombination
PpagC
repA
psi site
ArcA
promoter
cat
pSC2c
4410 bp
pSC101 ori
psi site
ArcA
tet O
ilacO
par locus

Figure 4

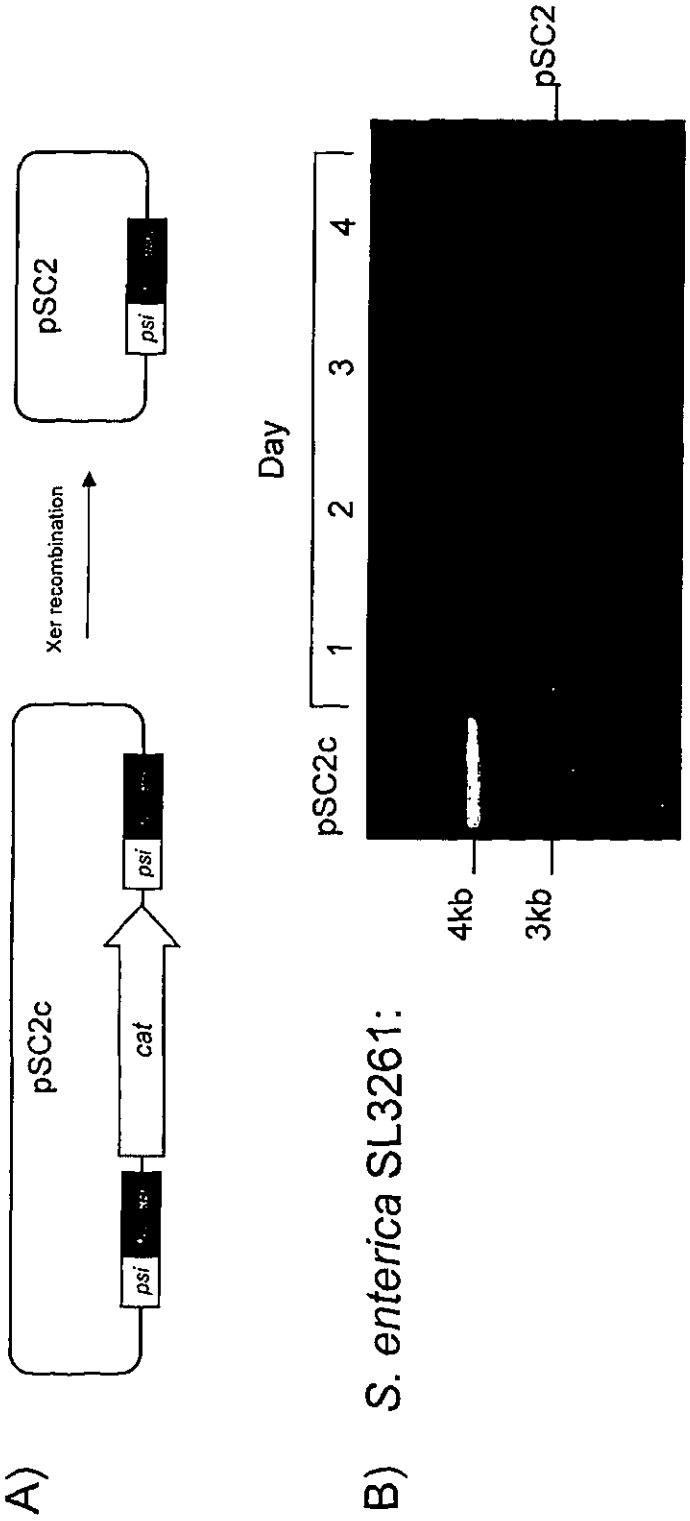

A)
pSC2c
cat
psi
psi
Xer recombination
pSC2
psi
B) S. enterica SL3261:
Day
pSC2c
1
2
3
4
pSC2
4kb
3kb

Figure 5