XL10-Gold Competent Cells

Cat: C1450
Size: 20×100μL
Storage: Store at -70℃ for 6 months to avoid repeated freezing and thawing. Not suitable for storage in liquid nitrogen.

Introduction:

XL10-Gold competent cells produced by our company are competent cells obtained by special processing of Escherichia coli XL10 strain, which can be used for chemical transformation of DNA. Suitable for efficient DNA cloning and plasmid amplification. It can ensure the stable replication of high copy plasmid. Suitable for the transformation of unmethylated DNA. The conversion efficiency reached 10^8 cfu/μg by pUC19 plasmid detection.

Genotype:

Tet^{r}Δ(mcrA)183Δ(mcrCB-hsdSMR-mrr)173endA1supE44thi-1recA1gyrA96relA1lacHte[F'proABlacI^{q}ZΔM15Tn1 0(Tet^r) AmyCam^r]

Strain Resistance: The cells were resistant to tetracycline and chloramphenicol.

Protocols: (The following operations are carried out according to the standard of sterile conditions)

1. The competent cells are placed in an ice bath.
2. The target DNA was added to the receptive cell suspension(100μL receptive cells could be saturated with 1ng superhelix plasmid DNA), the centrifuge tube was gently rotated to mix the contents, and the contents were left to rest in an ice bath for 30min.
3. The centrifuge tube was placed in a water bath at 42°C for 60s, and then quickly transferred to the ice bath to cool the cells for 2min without shaking the centrifuge tube.
4. 500μL sterile SOC or LB medium (without antibiotics) was added to each centrifuge tube, mixed and placed at 37℃, 150rpm, and shaken for 60min. The purpose was to express related resistance marker genes on the plasmid and revive the bacteria.
5. Under sterile conditions, appropriate amount of bacterial solution was added to LB solid medium plate containing corresponding antibiotics, and the cells were evenly coated with sterile bacterial coater or glass beads. After the liquid in the plate was completely absorbed, the plate was inverted and cultured at 37℃ for 12-16h.
6. Keep the remaining bacterial solution in the refrigerator at 4℃, and decide whether to leave or leave according to the growth of the colony on the plate.

Notes:

1. The competent cells should be stored at -70℃, can not be frozen and thawed repeatedly, otherwise its conversion efficiency will be reduced.
2. The experiment should be strictly aseptic operation, to prevent the contamination of other DNA

or miscellaneous bacteria, to avoid the impact on the future screening and identification.

3. During conversion, the conversion efficiency is proportional to the concentration of foreign DNA within a certain range, but when the amount of foreign DNA added is too large or the volume is too large, the conversion efficiency will be reduced. The volume of DNA during transformation should be less than one-tenth of the volume of competent cells.
4. Calculation of conversion rate: Conversion rate = total number of colonies produced/total amount of paving DNA.
5. In order to prevent the conversion experiment from being unsuccessful, part of the connection products can be retained to re-transform and minimize the loss.

Related Products:

I1020 IPTG solution(50mg/mL)
A1170 Ampicillin storage Solution(100mg/mL)
K1030 Kanamycin(100mg/mL)
L1015 LB solid medium(dry powder)
L1020 SOC Liquid medium(dry powder)
X1010 X-gal(20mg/mL)