GT115 Competent Cells

Cat: C3770
Size: 10×100μL/20×100μL
Storage: Store at -70°C to avoid repeated freezing and thawing.

Product Parameters:
English name: GT115 Competent cells

Genotype: F⁻mcrA Δ(MR-HSDRms-MCRBC) φ80lacZΔM15 ΔlacX74 nupG recA1 araD139 Δ(ara-leu)7697 galE15 galK16 rpsL(StrA) endA1 Δdcm uidA(ΔMluI)::pir-116 ΔsbcC-sbcD

Strain Resistance: Sensitive to ampicillin, kanamycin, spectacular, chloramphenicol, and tetracycline. Resistant to streptomycin.

Introduction:

The genome of strain GT115 contains the pir gene, which is capable of expressing π protein for the recognition of R6K replication initiator. Plasmids carrying R6K replication initiators(such as pCpGL-Basic, pCpGL-CMV/EF1, etc.) can be replicated and amplified in this strain. By deleting sbcC and sbcD genes in the genome, the SbcCD complex of recognizing and cutting hairpin structure could not be formed in GT115 cells, and the stability of the plasmids containing hairpin structure DNA was enhanced. This strain contains the gene rpsL(StrA) and is resistant to streptomycin. This strain has a nuclease(endA) mutation, recombinase(recA) mutation, which enhances the stability of foreign DNA. GT115 competent cells were prepared by special technology and pUC19 detected conversion efficiency >1×10^8cfu/μg DNA.

Protocols:

1. The competent cells are placed in an ice bath to defreeze. After the cells are just defreeze, plasmid DNA or 5-10μL of the connection product was added to the cells, dial the bottom of the tube with your finger, and gently mix;
2. Place in the ice bath for 30min, do not shake;
3. Heat shock at 42℃ for 60s, do not shake;
4. Place in the ice bath for 2min, do not shake;
5. Add 500μL sterile LB medium;
6. The culture was placed in a shaking table at 37℃, 150-200rpm for 60min.
7. Take 50-100μL bacterial solution and apply it on LB plate containing phase resistance. After the liquid was sucked up, the plate was turned upside down and cultured at 37℃ overnight.

(**Plate scribing separation method:** After the recovery culture, centrifuge at 12000rpm for 30s, discard the supernatant, leave about 100μL of liquid, gently blow the bacteria with 200μL suction head, take 10μL of suspended bacterial liquid divided into more drops on the plate, tilt the suction head, and use the side of the suction head to scribing the liquid dripping on the plate. This method can obtain a larger monoclonal colony. This method is mainly suitable for plasmid transformation,

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and the conversion of link products is best by coating.)

(**Rapid plasmid transformation steps:** For ampicillin resistant plasmids, shorten the time of step 2 to 5min. After completing step 4, it can be directly coated or marked on the ampicillin resistant LB plate. Other resistant plasmids still require 60min of resuscitation.)

Notes:

1. If the biochemical reagents produced by our company are not specially marked, they are basically non-sterile packaging. If they are used in cell experiments, please pre-treat them in advance.
2. Once it is prepared into a solution, please pack it separately and store it to avoid product failure caused by repeated freezing and thawing.
3. The product information is for reference only, if you have any questions, please call 400-968-6088 for consultation.
4. This product is for scientific research only. Do not use for medicine, clinical diagnosis or therapy, food or cosmetics. Do not store in ordinary residential areas.
5. For your safety and health, please wear a lab coat and wear disposable gloves and a mask.