

**DB3.1 Chemically Competent Cell**

Catalogue No.: abx098865

DB3.1 Chemically Competent Cell is designed for chemical transformation of DNA. This cell contains the *gyrA462* gene which provides resistance to the toxic effects from the *ccdB* gene. This product can be used for transformation and propagation of plasmids containing the *ccdB* gene, with a transformation efficiency of over  $10^8$  cfu/ $\mu$ g DNA (tested by pUC19 plasmid DNA).

The genotype is: F<sup>-</sup> *gyrA462 endA1 Δ(sr1-recA) mcrB mrr hsdS20(r<sub>B</sub><sup>-</sup>, m<sub>B</sub><sup>-</sup>) supE44ara-14 galK2 lacY1 proA2 rpsL20(Sm<sup>R</sup>) xyl-5 λ- leu mtl1.*

**Target:** DB3.1 Chemically Competent Cell

**Storage:** Store at -70°C for up to 6 months. Do not store in liquid nitrogen. Avoid repeated freeze/thaw cycles.

**Buffer:** Contains 10% glycerol.

**Directions for use: Recommended Protocol:**

1. Equilibrate a water bath to 42 °C. Bring a vial of SOC medium or LB medium to room temperature. Warm selective plates to 37 °C for 30 minutes.
2. Thaw 100  $\mu$ l of DB3.1 Chemically Competent Cell on ice. Aliquot 50  $\mu$ l of cells into a pre-chilled 1.5 ml tube, then add target DNA (1-5  $\mu$ l). Do not mix by pipetting up and down. Leave on ice for 30 minutes.
3. Heat-shock the cells for 45 seconds at 42 °C, without shaking. Immediately transfer to the tube to ice. Leave on ice for 2 minutes without shaking.
4. Add 500  $\mu$ l of prewarmed SOC medium or LB medium (without antibiotics) into the tube. Mix well and shake at 37 °C for 1 hour at 200 RPM for cell recovery and expression of antibiotic resistance.
5. Spread 20-200  $\mu$ l from each transformation vial onto a pre-warmed selective plate. Cells that have not been plated can be stored at 4 °C and plated the next day if required.
6. Invert the plates and incubate at 37 °C overnight.
7. Select colonies and analyse by restriction enzyme digestion, PCR or sequencing.

**Note:**

Higher efficiency transformation can be achieved by transforming cells immediately following thawing. All samples and reagents require gentle handling throughout the entire procedure.

**Note:** This product is for research use only. This product is shipped with dry ice.