倍沃医学

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BW-SCCP Single Step Competent Cell Preparation Kit

Contents

Kit Contents

Catalog#	SCCP-00	SCCP-01	SCCP-02
Preps	5	250	500
SCCP Solution	500 μL	25 mL	50 mL
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Introduction

The SCCP Solution is designed to prepare competent *E. coli* cells in a single step and transform the cells without heat-shock. SCCP method is faster and easier than other methods of producing competent cells, such as the traditional $CaCl_2$ method described by *Sambrook et al*, or other high competency protocols. The competent cells obtained by this method can be kept at -80℃ for long term storage. Transformation efficiencies depend on the strain of *E. coli*, as well as the nature and quality of the transforming DNA. Typical transformation efficiencies are 10^6~10^8 pfu/μg plasmid DNA. For example, the transformation efficiencies observed for *E. coli* strains DH1, DH5α, HB101, JM109, LE392, MM294, SCS-1, XL1-blue and TG1 ranged from 1.5×10^6 to 1.0×10^8 pfu per μg of DNA.

SCCP is supplied as a ready to use format of 1x solution.

Applications

Preparation of *E. coli* competent cells for transformation.

Storage and Stability

All contents of the kit should be kept at 4℃. From the date of production, the kit is stable for 12 months.

Features

Simple: No high speed centrifugation required.

Fast: The entire procedure takes only 5 minutes.

Quality Control

Transformation and storage solution is tested for transformation efficiency with appropriate *E. coli* strains and pUC18 or pUC19 DNA.

Protocol (For single preparation)

1. Pick a colony from an overnight cultured agar plate containing the appropriate strain of *E. coli* using a sterile tooth pick or a sterile pipette tip. Inoculate the colony into 2 mL SOB broth and grow overnight on a shaker at 37℃ with vigorous shaking.
2. On Day 2, inoculate 1 mL of the above culture into 50 mL of SOB broth in a 250 mL flask. Continue to grow the cells on the shaker at 37℃ with vigorous shaking until OD_{600}=0.5~0.7.
3. Transfer 2 mL supernatant into a clean tube. Spin at 2,000 x g (4,000 rpm) for 2~3 minutes and discard the supernatant carefully.
4. Add **100 µL** of precooled **SCCP Solution** and resuspend the cells gently.
5. Add 100 pg to 10 ng of transforming DNA to the cells, mix gently and incubate the mixture on ice for 10 minutes, then 37℃ for 5 minutes, followed by on ice for 5 minutes.
6. Add 1 mL preheated SOB broth and incubate the tube at 37℃ for 45-60 minutes.
7. Plate the cells on the appropriate selective or differential media.

Protocol (For batch preparation)

1. Pick a colony from an overnight cultured agar plate containing the appropriate strain of *E. coli*, using a sterile tooth pick or a sterile pipette tip. Inoculate the colony into 2 mL SOB broth and grow overnight on a shaker at 37℃ with vigorous shaking.
2. On Day 2, inoculate 1 mL of the above culture into 50 mL of SOB broth in a 250 mL flask. Continue to grow the cells on the shaker at 37°C with vigorous shaking until OD600=0.5~0.7.
3. Transfer culture to a 50 mL centrifuge tube. Spin at 2,000 x g (4,000 rpm) for 10 minutes and discard the supernatant carefully.
4. Add **2.5 mL** of precooled **SCCP Solution** and resuspend the cells gently.
5. Aliquot the cell mixture **50~100 µL** per tube. The competent cells are ready to use or can be stored at -80℃.
6. Perform *E. coli* transformation according to standard protocol.

Note:

- For large volume of cell preparation, the volume of **SCCP Solution** should be scaled up correspondingly. For example, 1~2 mL **SCCP Solution** can be used for 50~100 mL cultured cells. After the cells are resuspended, aliquot of 50~100 µL per vial and store at -80℃.
- Step **3** to **6** MUST BE CARRIED OUT ON ICE AT ALL TIMES.
- The solution also works well with *Bacillus subtilis*.

Limited Use and Warranty

This product is warranted to perform as described in its labeling and in BEIWO's literature when used in accordance with instructions. No other warranties of any kind, express or implied, including, without limitation, implied warranties of merchantability or fitness for a particular purpose, are provided by BEIWO. BEIWO's sole obligation and purchaser's exclusive remedy for breach of this warranty shall be, at the option of BEIWO, to replace the products, BEIWO shall have no liability for any direct, indirect, consequential, or incidental damage arising out of the use, the results of use, or the inability to use it product.

For technical support or learn more product information, please contact us at 400-115-2855 or visit our website at www.beiwobiomedical.com.

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