Product Information Sheet for NR-49804

Escherichia coli K-12, Strain DC10B

Catalog No. NR-49804

For research use only. Not for use in humans.

Contributor:
Ian Monk, Postdoctoral Researcher, Department of Microbiology and Immunology, Peter Doherty Institute for Infection and Immunity, University of Melbourne, Victoria, Australia, Tim Stinear, Professor, Peter Doherty Institute for Infection and Immunity, University of Melbourne, Victoria, Australia and Timothy Foster, Professor, Department of Microbiology, Trinity College Dublin, University of Dublin, Dublin, Ireland

Manufacturer:
BEI Resources

Product Description:
Bacteria Classification: *Enterobacteriaceae, Escherichia*
Species: *Escherichia coli*
Strain: K-12, DC10B
Original Source: *Escherichia coli (E. coli)* K-12, strain DC10B is a DNA cytosine methyltransferase *(dcm)* deletion mutant that was produced from *E. coli* K-12 derivative strain DH10B via recombination-mediated genetic engineering (recombineering).[1,2,3]
Comments: *E. coli* K-12, strain DC10B is a universal host for constructing plasmids for introduction into staphylococci and was deposited as Δ*dcm* and resistant to streptomycin.[1,2]

E. coli K-12, strain DC10B is a Δ*dcm* mutant of the high-efficiency cloning strain, DH10B, and provides a background for plasmid production in the absence of cytosine methylation. The lack of methylation allows plasmid DNA to bypass a conserved type IV restriction-modification (RM) barrier in staphylococci which has been identified as a major barrier to transformation with foreign DNA.[2,3,4] *E. coli* K-12, strain DC10B is an ideal host for the construction of recombinant plasmids for subsequent direct transformation into *Staphylococcus aureus (S. aureus)* or *Staphylococcus epidermidis*.[1]

Additional transformation efficiency can be gained by the introduction of the *S. aureus* clonal complex (CC) specific methylation profiles observed in type 1 RM. The type 1 RM protein complex is comprised of three protein components: a methylase (HsdM), a specificity protein (HsdS) and a restriction protein (HsdR). The complex recognizes a target recognition motif (TRM) determined by HsdS and detects its methylation status via HsdM. DNA that is correctly hemimethylated will be fully methylated, which will prevent the restriction of the DNA by the RM protein complex.[4] *E. coli* K-12 strains recombineered to contain genes from *S. aureus* CC1, CC8, CC30 and CC93, in a Δ*dcm* background, are available from BEI Resources as NR-49805 through NR-49808, respectively.

Material Provided:
Each vial contains approximately 0.5 mL of bacterial culture in Tryptic Soy broth containing 25 µg/mL streptomycin supplemented with 10% glycerol.

Note: If homogeneity is required for your intended use, please purify prior to initiating work.

Packaging/Storage:
NR-49804 was packaged aseptically in cryovials. The product is provided frozen and should be stored at -60°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

Growth Conditions:
Media:
Tryptic Soy broth or Nutrient broth or equivalent containing 25 µg/mL streptomycin
Tryptic Soy agar or Nutrient agar or Tryptic Soy agar with 5% defibrinated sheep blood or equivalent containing 25 µg/mL streptomycin
Incubation:
Temperature: 37°C
Atmosphere: Aerobic
Propagation:
1. Keep vial frozen until ready for use, then thaw.
2. Transfer the entire thawed aliquot into a single tube of broth.
3. Use several drops of the suspension to inoculate an agar slant and/or plate.
4. Incubate the tube, slant and/or plate at 37°C for 1 day.

Citation:
Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Escherichia coli* K-12, Strain DC10B, NR-49804."

Biosafety Level: 1
Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories (BMBL). 6th ed. Washington, DC: U.S. Government Printing Office, 2020.

Disclaimers:
You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at www.beiresources.org.

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC® nor the U.S. Government makes any warranties or representations as to its accuracy. Citations

BEI Resources
www.beiresources.org
E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898

© 2016/2018/2020/2024 American Type Culture Collection (ATCC). All rights reserved.

from scientific literature and patents are provided for informational purposes only. Neither ATCC® nor the U.S. Government warrants that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, use and disposal. ATCC® and the U.S. Government are not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to ensure authenticity and reliability of materials on deposit, the U.S. Government, ATCC®, their suppliers and contributors to BEI Resources are not liable for damages arising from the misidentification or misrepresentation of products.

Use Restrictions:

This material is distributed for internal research, non-commercial purposes only. This material, its product or its derivatives may not be distributed to third parties. Except as performed under a U.S. Government contract, individuals contemplating commercial use of the material, its products or its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale.

References:

1. Monk, I., Personal Communication.
2. Monk, I., et al. "Transforming the Untransformable: Application of Direct Transformation to Manipulate Genetically *Staphylococcus aureus* and *Staphylococcus epidermidis*." mBio 20 (2012): e00277. PubMed: 22434850.
3. Monk, I. and T. J. Foster. "Genetic Manipulation of Staphylococci-Breaking Through the Barrier." Front. Cell Infect. Microbiol. 12 (2012): e00049. PubMed: 22919640.
4. Monk, I., et al. "Complete Bypass of Restriction Systems for Major *Staphylococcus aureus* Lineages." mBio 26 (2015): e00308-15. PubMed: 26015493.

ATCC® is a trademark of the American Type Culture Collection.

Support Provided by NIAID

BEI Resources
www.beiresources.org

E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898

© 2016/2018/2020/2024 American Type Culture Collection (ATCC). All rights reserved.