

SUPPORTING INFECTIOUS DISEASE RESEARCH

Product Information Sheet for NR-46131

Escherichia coli – Staphylococcus aureus Shuttle Vector pCN40, Recombinant in Staphylococcus aureus

Catalog No. NR-46131

For research use only. Not for human use.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

NR-46131 is a culture of *Staphylococcus aureus* (*S. aureus*), strain RN4220 (RN9594, NRS594) containing the *Escherichia coli* (*E. coli*)-staphylococcal shuttle vector pCN40 (pNR-46131). Vector pCN40 contains the *E. coli* ColE1 replication origin, the *S. aureus* pT181 *cop-wt-repC* replicon and the P_{blaz} promoter. Vector pCN40 was deposited as resistant to ampicillin and erythromycin in *E. coli* and resistant to erythromycin in *S. aureus*.¹

The complete sequence and vector map of pCN40 have been determined and are available on the Certificate of Analysis for NR-46131 lot 62782861. The BEI Resources vector sequence (pNR-46131) is available (GenBank: KP255996).

Vector pCN40 is a member of a series of novel shuttle vectors that were developed using PCR-designed cassettes to allow for easy exchange of vector components. The base shuttle vectors are comprised of (i) a staphylococcal replicon (pT181based low-copy number, high-copy-number thermosensitive replicons or pl258-based low-copy-number theta replicon), (ii) a staphylococcal selectable marker (erythromycin, tetracycline, chloramphenicol, kanamycin or spectinomycin resistance), (iii) an E. coli ColE1-based replicon (iv) an E. coli selectable marker (ampicillin resistance) and (v) a pUC19-derived expanded multiple cloning site (MCS). Additionally, some of the vectors may contain a staphylococcal \$11 phage fragment, staphylococcal pathogenicity island SaPI1 fragment, an inducible or constitutive promoter, and reporter genes.1

Material Provided:

Each vial of NR-46131 contains approximately 0.5 mL of bacterial culture in Casitone-Yeast (CY) broth containing 0.1 M glycerol phosphate and 10 μ g/mL erythromycin supplemented with 10% glycerol.

Packaging/Storage:

NR-46131 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. Freezethaw cycles should be avoided.

Growth Conditions:

Media:

Casitone-Yeast broth containing 10 µg/mL erythromycin Tryptic Soy agar containing 10 µg/mL erythromycin Incubation:

Temperature: 37°C Atmosphere: Aerobic

Propagation:

- 1. Keep vial frozen until ready for use, then thaw.
- Transfer the entire thawed aliquot into a single tube of broth.
- 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 provided by the Network on Antimicrobial Resistance in *Staphylococcus aureus* (NARSA) for distribution by BEI Resources, NIAID, NIH: *Escherichia coli – Staphylococcus aureus* Shuttle Vector pCN40, Recombinant in *Staphylococcus aureus*, NR-46131."

Biosafety Level: 2

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. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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References:

 Charpentier E., et al. "Novel Cassette-Based Shuttle Vector System for Gram-Positive Bacteria." <u>Appl. Environ. Microbiol.</u> 70 (2004): 6076-6085. PubMed: 15466553.

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