

***Escherichia coli* – *Staphylococcus aureus*
Shuttle Vector pCN49, Recombinant in
*Escherichia coli***

Catalog No. NR-46145

For research use only. Not for human use.

Contributor:

Richard P. Novick, M.D., Departments of Microbiology, Medicine and Molecular Pathogenesis, New York University School of Medicine, New York, New York, USA

Manufacturer:

BEI Resources

Product Description:

NR-46145 is a culture of *Escherichia coli* (*E. coli*) DH5α (RN9609, NRS609) containing the *E. coli*-staphylococcal shuttle vector pCN49. Vector pCN49 contains the *E. coli* ColE1 replication origin, the thermosensitive *Staphylococcus aureus* (*S. aureus*) pT181 *cop-634ts-rep4C* replicon and the *blaZ* transcriptional terminator. Vector pCN49 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 pCN49 have been determined and are available on the Certificate of Analysis for lot 63025316. The BEI Resources vector sequence was deposited into GenBank as NR-46145 (GenBank: KR781467).

Vector pCN49 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 (pT181-based low-copy number, high-copy-number or 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 φ11 phage fragment, staphylococcal pathogenicity island SaPI1 fragment, an inducible or constitutive promoter, and reporter genes.¹

Material Provided:

Each vial of NR-46145 contains approximately 0.5 mL of bacterial culture in Luria-Bertani (LB) broth containing 100 µg/mL ampicillin supplemented with 10% glycerol.

Packaging/Storage:

NR-46145 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:

LB broth containing 100 µg/mL ampicillin

LB agar containing 100 µg/mL ampicillin

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 18 to 24 hours.

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 pCN49, Recombinant in *Escherichia coli*, NR-46145.”

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

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References:

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

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