b|**e**|**i** resources

SUPPORTING INFECTIOUS DISEASE RESEARCH

Clostridium difficile, Isolate 20110979

Catalog No. NR-49285

For research use only. Not for human use.

Contributor:

Brandi Limbago, Ph.D., Deputy Chief, Clinical and Environmental Microbiology Branch, Division of Healthcare Quality Promotion, Centers for Disease Control and Prevention, Atlanta, Georgia, USA and Ryan T. Ranallo, Ph.D., Program Officer, Enteric and Hepatic Diseases Branch, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Rockville, Maryland, USA

Manufacturer:

BEI Resources

Product Description:

<u>Bacteria Classification</u>: Clostridiaceae, *Clostridium* (A taxonomy change to *Peptostreptococcaceae, Peptoclostridium* has been proposed.)¹

<u>Species</u>: Clostridium difficile (Peptoclostridium difficile) <u>Isolate</u>: 20110979

- <u>Original Source</u>: *Clostridium difficile* (*C. difficile*), isolate 20110979 was obtained from the stool of an elderly female patient with a community-associated (CA) *C. difficile* infection in midwestern USA in 2011.²
- <u>Comments</u>: *C. difficile,* isolate 20110979 is part of the <u>Emerging Infections Program - *Clostridium difficile* <u>Surveillance Project</u> at the Centers for Disease Control and Prevention.^{2,3} Isolates were selected to represent the diversity of strain types and geographical locations circulating in the U.S. during 2010-2011. Isolate 20110979 was deposited as PCR ribotype 027, North American pulsed-field gel electrophoresis type 1 (NAP1), containing *tcdA, tcdB* and *tcdC* (with 18 base pair deletion) of the PaLoc operon as well as the *C. difficile* binary toxin (CDT).²</u>

C. difficile is a Gram-positive, spore-forming, obligate anaerobe that commonly inhabits the intestinal tract of various mammalian species, reptiles and birds, and may also be found in the environment. C. difficile infection is the leading cause of gastroenteritis-associated death and has become the most common cause of hospital-associated (HA) infections in the USA.³ Epidemic strains of *C. difficile* associated with severe disease are generally positive for CDT, contain an 18 base pair deletion in tcdC, are resistant to fluoroquinolones, have PCR ribotype 027 and pulse-field gel electrophoresis type NAP1, restriction endonuclease analysis (REA) type B1 and toxinotype III (CDT⁺, TcdA⁺ and TcdB⁺).⁴ C. difficile produces a cytotoxin (TcdB) and an enterotoxin (TcdA) whose genes are part of the PaLoc operon. The operon also contains the tcdC gene which is a negative regulator of the tcdA and tcdB genes. The CDT is comprised of two parts encoded by *cdt*A (enzymatic component) and *cdt*B (binding component).⁴ The production of these toxins in the gut ultimately leads to pseudomembranous colitis (PMC) and *C. difficile* associated diarrhea (CDAD), which often occur as a complication of antibiotic therapy in elderly hospitalized patients.⁵

Material Provided:

Each vial contains approximately 0.5 mL of bacterial culture in Modified Reinforced Clostridial medium supplemented with 10% glycerol.

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

Packaging/Storage:

NR-49285 was packaged aseptically, in screw-capped plastic 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:

Modified Reinforced Clostridial medium or equivalent

Tryptic Soy agar with 5% defibrinated sheep blood or equivalent

Incubation:

Temperature: 37°C

Atmosphere: Anaerobic

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 to 2 days.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Clostridium difficile*, Isolate 20110979, NR-49285."

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. <u>Biosafety in</u> <u>Microbiological and Biomedical Laboratories</u>. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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 <u>www.beiresources.org</u>.

E-mail: <u>contact@beiresources.org</u> Tel: 800-359-7370 Fax: 703-365-2898 e i resources

SUPPORTING INFECTIOUS DISEASE RESEARCH

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 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, noncommercial 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:

- Yutin, N. and M. Y. Galperin. "A Genomic Update on Clostridial Phylogeny: Gram-Negative Spore-Formers and Other Misplaced Clostridia." <u>Environ. Microbiol.</u> 15 (2013): 2631-2641. PubMed: 23834245.
- 2. Limbago, B., Personal Communication.
- Lessa, F. C., et al. "Burden of *Clostridium difficile* Infection in the United States." <u>N. Engl. J. Med.</u> 372 (2015): 2369-2370. PubMed: 26061850.
- Persson, S., M. Torpdahl and K. E. P. Olsen. "New Multiplex PCR Method for the Detection of *Clostridium difficile* Toxin A (*tcd*A) and Toxin B (*tcd*B) and the Binary Toxin (*cdt*A/*cdt*B) Genes Applied to a Danish Strain Collection." <u>Clin. Microbiol. Infect.</u> 14 (2008): 1057-1064. PubMed: 19040478.
- Kelly, C. P. and J. T. LaMont. "*Clostridium difficile* More Difficult than Ever." <u>N. Engl. J. Med.</u> 359 (2008): 1932-1940. PubMed: 18971494.

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

