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SUPPORTING INFECTIOUS DISEASE RESEARCH

Clostridium difficile, Isolate 20120085

Catalog No. NR-49321

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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>: 20120085

- <u>Original Source</u>: *Clostridium difficile* (*C. difficile*), isolate 20120085 was obtained from the stool of an older male patient with a community-associated (CA) *C. difficile* infection in midwestern USA, in 2011.²
- <u>Comments</u>: *C. difficile,* isolate 20120085 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 20120085 was deposited as PCR ribotype 106, North American pulsed-field gel electrophoresis type 11 (NAP11), containing *tcdA*, *tcdB* and *tcdC* of the PaLoc operon. This isolate is reported to be negative for 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 healthcare-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 *cdtA* (enzymatic component) and *cdtB* (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-49321 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 24 to 72 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Clostridium difficile*, Isolate 20120085, NR-49321."

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.

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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. 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 (*tcdA*) and Toxin B (*tcdB*) and the Binary Toxin (*cdtA*/*cdtB*) 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.

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