

***Clostridium difficile*, Isolate 20120956**

Catalog No. NR-49304

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Contributor:

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Manufacturer:

BEI Resources

Product Description:

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

Species: *Clostridium difficile* (*Peptoclostridium difficile*)

Isolate: 20120956

Original Source: *Clostridium difficile* (*C. difficile*), isolate 20120956 was obtained from the stool of an older male patient with a healthcare-associated (HA) *C. difficile* infection in southern USA in 2011.²

Comments: *C. difficile*, isolate 20120956 is part of the [Emerging Infections Program - Clostridium difficile Surveillance Project](#) 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 20120956 was deposited as PCR ribotype 020, North American pulsed-field gel electrophoresis type 4 (NAP4), containing *tcdA*, *tcdB* and *tcdC* of the PaLoc operon. This isolate is reported to be negative for the *C. difficile* binary toxin (CDT).²

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 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.

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

Packaging/Storage:

NR-49304 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 20120956, NR-49304."

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:

1. Yutin, N. and M. Y. Galperin. "A Genomic Update on Clostridial Phylogeny: Gram-Negative Spore-Formers and Other Misplaced Clostridia." Environ. Microbiol. 15 (2013): 2631-2641. PubMed: 23834245.
2. Limbago, B., Personal Communication.
3. Lessa, F. C., et al. "Burden of *Clostridium difficile* Infection in the United States." N. Engl. J. Med. 372 (2015): 2369-2370. PubMed: 26061850.
4. 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." Clin. Microbiol. Infect. 14 (2008): 1057-1064. PubMed: 19040478.
5. Kelly, C. P. and J. T. LaMont. "*Clostridium difficile* - More Difficult than Ever." N. Engl. J. Med. 359 (2008): 1932-1940. PubMed: 18971494.

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