VIASURE MULTIPLEX

Clostridium difficile toxins A+B Real Time PCR Detection Kit

Pathogen and product description

Clostridium difficile is a gram-positive, sporogenic, anaerobic bacillus that belongs to the Clostridiaceae family. Initially, C. difficile was described as a member of the commensal microbiota of neonates. However, later it was identified as a causal agent of antibiotic-associated diarrhea (AAD) and its infection is associated with high morbidity and mortality in the elderly.

The major risk factors for the *C. difficile* infection are broad spectrum antibiotics exposure, hospitalization and advanced age. The severity of its infection ranges from mild diarrhea and pseudomembranous colitis to toxic megacolon, perforations of the colon and occasionally, sepsis and even death. The main routes of transmission are the fecal-oral or aerosols. In fact, infected persons with acute diarrhea can lead to heavy contamination of the environment with spores,

which can persist in dust or on surfaces for months and be transmitted to other hospitalized patients or to healthcare workers once again.

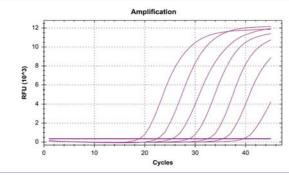
Toxigenic strains of *C. difficile* can colonize the gut, replicate and produce enterotoxin A and cytotoxin B, encoded by *tcdA* and *tcdB* genes. A Real-Time PCR based diagnosis has been described as a sensitive test for detection and identification of *Clostridium difficile* toxin A and/ or B.

VIASURE Clostridium difficile toxins A+B Real Time PCR Detection Kit is designed for the diagnosis of gastroenteritis caused by Clostridium difficile in human stool samples. After DNA isolation, the identification of Toxin A and/or B of Clostridium difficile is performed by the amplification of a conserved region of the tcdA and tcdB genes using specific primers and fluorescent-labeled probes.



Analytical sensitivity

VIASURE Clostridium difficile toxins A+B Real Time PCR Detection Kit has a detection limit of ≥ 10 DNA copies per reaction (figures 1 and 2).



Amplification

8

6

0

10

20

30

40

Cycles

Figure 1. Dilution series of Clostridium difficile Toxin A (10⁷−10¹ copies/rxn) template run on the Bio-Rad CFX96 Touch™ Real-Time PCR Detection System.

Figure 2. Dilution series of Clostridium difficile Toxin A $(10^7-10^1 \text{ copies/rxn})$ template run on the Bio-Rad CFX96 TouchTM Real-Time PCR Detection System.

Components

Reagent/Material	Description	Quantity
Clostridium difficile toxins A+B 8-well strips	A mix of enzymes, primers-probes, buffer, dNTPs, stabilizers and Internal control in stabilized format	6/12 x 8-well strip
Clostridium difficile toxins A+B 96-well plate	A mix of enzymes, primers-probes, buffer, dNTPs, stabilizers and Internal control in stabilized format	1 plate
Rehydration Buffer	Solution to reconstitute the stabilized product	1 vial x 1,8 mL
Clostridium difficile toxins A+B Positive Control	Non-infectious synthetic lyophilized cDNA	1 vial
Negative Control	Non template control	1 vial x 1 mL
Water RNAse/DNAse free	Water RNAse/DNAse free	1 vial x 1 mL
Tear-off 8-cap strips	Optical caps for sealing wells during thermal cycling	6/12 x 8-cap strip
Shell Frame Grid	Shell Frame Grid	1 or 2

Work Flow

One-step rehydration of wells and add your extracted DNA



STEP 1
Add 15 µl of rehydration buffer into each well



STEP 2 Add 5 µl of DNA sample / positive control / negative control



STEP 3 Load the strips into the thermocycler and run the specified protocol



STEP 4 Interpretate results

Kit References

Reference	Description
VS-CDA106L	Viasure <i>Clostridium difficile toxins A+B</i> Real Time PCR Detection Kit 6 x 8-well strips, low profile
VS-CDA106H	Viasure <i>Clostridium difficile toxins A+B</i> Real Time PCR Detection Kit 6 x 8-well strips, high profile
VS-CDA112L	Viasure <i>Clostridium difficile toxins A+B</i> Real Time PCR Detection Kit 12 x 8-well strips, low profile
VS-CDA112H	Viasure <i>Clostridium difficile toxins A+B</i> Real Time PCR Detection Kit 12 x 8-well strips, high profile
VS-CDA113L	Viasure <i>Clostridium difficile toxins A+B</i> Real Time PCR Detection Kit 96-well plate, low profile
VS-CDA113H	Viasure <i>Clostridium difficile toxins A+B</i> Real Time PCR Detection Kit 96-well plate, high profile



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