This Procedural Bulletin is intended to provide a ready outline reference for performance of the assay. These abbreviated directions for use are not intended to replace the complete Package Insert. It is the obligation of every manufacturer of medical devices labeled FOR *IN VITRO* DIAGNOSTIC USE to provide a complete Package Insert in accordance with FDA labeling regulation (21 CFR 809.10).

Quidel Corporation provides CLSI procedures for your use. The procedures are required to include the same information as listed in the Package Insert. Any modifications to this document are the sole responsibility of the Laboratory.

Solana C. difficile Assay

CLIA Complexity: Moderate

INTENDED USE

The Solana C. difficile Assay is an *in vitro* diagnostic test for the direct, qualitative detection of the *Clostridioides (Clostridium) difficile* Toxin A gene *(tcdA)* in unformed stool specimens of patients suspected of having *Clostridioides (Clostridium) difficile*-infection (CDI). The Solana C. difficile Assay is intended for use as an aid in diagnosis of CDI. The assay utilizes helicase-dependent amplification (HDA) for the amplification of a highly-conserved fragment of the Toxin A gene sequence. The Solana C. difficile Assay is intended for use only with the Solana instrument.

SUMMARY AND EXPLANATION

Clostridioides (Clostridium) difficile is the most frequently identified enteric pathogen in patients with antibiotic-associated diarrhea and colitis. Each year in the United States, *C. difficile* infection results in approximately half a million infections among patients in the United States.[1] These infections account for considerable increases in the length of hospital stays and more than $1.1 billion in health care costs.[2] Recently, the incidence and severity of *C. difficile*-associated disease corresponding to short-term hospital stays has been on the rise.[3,4]

The majority of *C. difficile* infections are acquired nosocomially, and most patients remain asymptomatic following acquisition. It is thought that the exposure to antibiotics disrupts the flora of the intestine, allowing an opportunistic colonization by *C. difficile*. The virulence of *C. difficile* is believed to be mediated by the production of two toxins (Toxin A and Toxin B). However, the presence of both Toxin A and Toxin B proteins is not required for pathogenicity.[6] Both toxin genes (*tcdA* and *tcdB* respectively) are located within a 19.6 Kb pathogenicity locus (PaLoc) found within the genome of all known toxogenic *C. difficile* strains.[7] The Solana C. difficile Assay targets a highly conserved region of the PaLoc, which is intact in all known A+B+ and A-B+ toxinotypes of *C. difficile*.

PRINCIPLE OF THE TEST

The Solana C. difficile Assay combines simple sample processing and Helicase-Dependent Amplification (HDA) performed in Solana for the detection of toxigenic *Clostridioides (Clostridium) difficile* directly from CDI-suspected diarrheal specimens.

A small amount of specimen is transferred to a Lysis Tube using a swab. The Lysis Tube is then subjected to heat-treatment at 95°C for 5 minutes. The heat-treated sample is added to a Dilution Tube, and then transferred to a Reaction Tube. The Reaction Tube contains lyophilized HDA reagents, dNTPs, primers and probes. Once rehydrated with the diluted sample, the Reaction Tube is placed in Solana for amplification and detection of target sequence. In Solana, the target sequence is amplified by specific primers and detected by a specific fluorescence probe included in the Reaction Tube. A competitive process control (PRC) is included in the Lysis Tube to monitor sample processing, inhibitory substances in clinical samples, reagent failure or device failure. The PRC is amplified by the target-specific primers and detected by a PRC specific fluorescence probe.

The target and PRC probes are labeled with a quencher on one end and a fluorophore on the other end. Upon annealing to target or PRC amplicons, the fluorescence signal increases due to physical separation of fluorophore from quencher. Solana measures and interprets the fluorescent signal, using on-board method-specific algorithms. Solana then reports the test results to the user on its display screen, and it can print out the results via a printer.

MATERIALS PROVIDED

Cat. #M307
48 Tests per Kit

Component	Quantity	Storage
Neonatal flocked swabs	48 tubes/kit	2°C to 30°C
Lysis Buffer	48 tubes/kit 1.0 mL	2°C to 8°C
Dilution Buffer	48 tubes/kit 1.8 mL	2°C to 8°C
Reaction Tubes	48 tubes/kit	2°C to 8°C

MATERIALS REQUIRED BUT NOT PROVIDED

- External controls for toxigenic *C. difficile* (e.g. the laboratory's own internal control materials from isolated and characterized clinical specimen previously submitted for interpretation or Quidel Molecular C. difficile Control Set, Cat. #M108; these controls may serve as external processing and amplification controls and are independent of the PRC).
- Sterile DNase-free filter-blocked or positive displacement micropipettor tips
- Micropipettor
- Stopwatch or timer
- Vortex Mixer
- Scissors (to separate the reaction tubes)
- Solana workflow tray and transfer rack
- Heat block capable of 95°C ± 2°C temperature
- Thermometer
- Solana instrument

WARNINGS AND PRECAUTIONS

- Refer to the Solana User Manual for further information regarding instrument installation and operation.
- Only use the protocol described in this package insert. Deviations from the protocol may give erroneous results.
- All reagents are for *in vitro* diagnostic use only.
- Treat all specimen/samples as potentially infectious. Follow universal precautions when handling samples, this kit and its contents.
- All tubes should be capped tightly prior to vortexing.
- Proper sample collection, storage and transport are essential for correct results.
- Store assay reagents as indicated on their individual labels.
- Reagents are not interchangeable between lots.
- Never pool reagents from different tubes even if they are from the same lot.
- Do not use the reagents after their expiration date.
- Do not use kit components that appear to be broken or damaged.
- Do not interchange caps among reagents as contamination may occur and compromise test results.
- Only open the tubes when adding aliquots into tubes or removing aliquots from tubes. Keep the tubes closed at any other time to avoid contamination.
- To avoid contamination of the environment with amplicons, do not open the reaction tubes post-amplification.
- Avoid microbial and deoxyribonuclease (DNase) contamination of reagents when removing aliquots from tubes. The use of sterile DNase-free disposable filter-blocked or positive displacement pipettor tips is recommended.
- Use a new pipettor tip for each specimen or reagents.
- Performing the assay outside of the recommended time ranges can produce invalid results. Assays not completed within specified time ranges should be repeated.
- To avoid exposure to excessive heat, care should be taken when inserting and removing tubes from the heat block, and when handling the heated tubes.
- Additional controls may be tested according to guidelines or requirements of local, state, provincial and/or federal regulations or accrediting organizations.

- Do not pipette by mouth.
- Do not smoke, drink or eat in areas where specimens or kit reagents are being handled.
- For accurate results, pipette carefully using only calibrated equipment. Use of inaccurate volumes may give erroneous results.
- Thoroughly clean and disinfect all surfaces with a 10% bleach solution followed by molecular grade water.
- Use micropipettes with an aerosol barrier or positive displacement tips for all procedures.
- Testing should be performed in an area with adequate ventilation.
- Dispose of containers and unused contents in accordance with Federal, State and Local regulatory requirements.
- Wear suitable protective clothing, gloves, and eye/face protection when handling the contents of this kit.
- Wash hands thoroughly after handling.
- For additional information on hazard symbols, safety, handling and disposal of the components within this kit, please refer to the Safety Data Sheet (SDS) located at quidel.com.

STORAGE AND HANDLING OF KIT REAGENTS

Store the Assay Kit at 2°C to 8°C until the expiration date listed on the outer kit box.

SPECIMEN COLLECTION, STORAGE AND HANDLING

Specimen Type: unformed stool samples indicative of CDI.

Using a sterile container:

1. Transfer the liquid or soft stool into the sterile container, taking care not to transfer toilet paper, urine, water or soap.
2. Label the container according to hospital standard operating procedures.
3. Transport the labeled specimen to the laboratory.

Storage: Specimens should be transported in tightly sealed, leak proof plastic containers. If specimens can be processed within 3 to 4 hours after collection, transport at room temperature is adequate. Specimens delayed to the laboratory should be promptly cooled and kept at either 2°C to 8°C or –20°C for up to 7 days. Ship samples on ice if transported over long distances.

TEST PROCEDURE

1. Turn on Solana by pressing the power button and wait until it completes self-testing.
   Note: Do not open the lid during the self-testing.
2. 25 minutes prior to the heat lysis step, warm a heating block to 95°C ± 2°C.
3. Place the required number of Lysis Tubes in a rack. Mark the Lysis Tubes on the cap and/or side of the tube.
   Note: One (1) Lysis Tube is required for each specimen or control to be tested.
   Note: A maximum of 12 tests can be performed in a single Solana instrument.
4. Mix stool sample thoroughly.
5. Collect stool sample by using the swabs provided. Dip a swab into the liquid or unformed stool specimen.
   Note: Do not oversample. The swab head should only be lightly coated with stool. Use a new swab for each specimen.
6. Place the swab in a patient identified Lysis Tube and release the specimen by swirling the swab head rapidly within the Lysis Tube for 10 seconds. Remove the swab and discard as appropriate for your laboratory.
   Note: Once the specimen has been added to the Lysis tube proceed to Step 7 without delay.
7. Heat the Lysis Tubes at 95°C ± 2°C for 5 minutes and then vortex the Lysis Tubes for 5 seconds.
   Note: Begin 5-minute lysis procedure when the heat block measures 95° ± 2°C. The timer must be stopped if the temperature falls out of range at any time during the 5-minute period and cannot be restarted until the heat block returns to 95° ± 2°C.
   Note: Samples are stable in Lysis buffer after heating for up to 96 hours at 2° to 8°C and up to 24 hours at 25°C ± 2°C.
8. Place the required number of Dilution Tubes in a rack. Mark the Dilution Tubes on the cap and/or side of the tube.
   Note: One (1) Dilution Tube is required for each specimen or control to be tested.
9. Transfer 50 µL of each specimen to an identified Dilution Tube. Close the cap and mix the solution well by vortexing the tubes for 5 seconds.
   Note: Use a new pipette tip for each specimen.

Note: Samples are stable in Dilution buffer for up to 96 hours at 2° to 8°C and up to 25 hours at 25°C ± 2°C.

10. Remove the required number of Reaction Tubes from the protective pouch, remove the excess air and reseal the bag. Mark the Reaction Tubes on the cap.
11. Transfer 50 µL of the diluted specimen to the labeled Reaction Tube, mix the solution by pipetting up and down a minimum of 3 times and close the cap. The solution should be clear, free of solid material.
    Note: Use a new pipette tip for each diluted sample.
    Note: Proceed immediately to the next step. Do not allow reconstituted reaction mix to sit for longer than 15 minutes.
12. Using the Solana Transfer Rack to hold Reaction Tubes at eye-level, visually inspect each Reaction Tube to ensure pellet rehydration.
13. Open the lid and place the Reaction Tubes in Solana via the Transfer Rack. Close the lid.
    Note: Be sure that all tubes are in tight contact with heat block.
14. Enter User ID and Password and press ↵ (ENTER).
15. Select "NEW TEST". If Solana displays a different screen, go to the home screen.
16. Select the tube positions to use.
17. Scan the assay barcode or select "Cdiff Assay" from the Select Test drop-down menu and manually enter Lot ID/Exp Date, and press "▶".
18. Select sample type (patient or QC) from the drop-down menu and enter Sample IDs (optional; see 2nd Note in next step).
19. Press "Start" to initiate the Solana Cdiff Assay. Solana will display the progress and the count-down to assay completion, and the test results will be displayed on the screen in approximately 30 minutes.
    Note: To avoid laboratory contamination, once the tube has been closed and the amplification reaction started, **DO NOT** open the Reaction Tube.
    Note: While the test is running, sample ID can be entered or edited by pressing the pencil icon.
20. After the run is completed, press the arrow to move to the Test Results screen. The results can be printed by selecting the print button.
    Note: Do not navigate away from this screen before printing results. Once the screen is gone it cannot be revisited. If this occurs the results can be viewed individually by going Home then selecting Review Results.

INTERPRETATION OF RESULTS

Samples	**Assay Result**	**Interpretation**
Patient specimen	POSITIVE	Toxigenic *C. difficile* DNA detected
	NEGATIVE	No Toxigenic *C. difficile* DNA detected/PRC detected
	INVALID	No Toxigenic *C. difficile* DNA and No PRC detected; for invalid test results, retest the same processed sample first. If the test is invalid upon retesting with the processed sample, re-process another aliquot of the same sample or obtain a new sample and re-test.

QUALITY CONTROL

The Solana C. difficile assay incorporates several controls to monitor assay performance.

- The process control is used to monitor sample processing, to detect HDA inhibitory specimens and to confirm the integrity of assay reagents and cassette detection. The process control is included in the lysis buffer tube.
- External positive controls may be treated as a patient specimen. Dip the provided swab into the external positive control ensuring liquid covers the tip. Identify the Dilution Buffer tube as the positive control and proceed with processing as described above in the Assay Procedure. The external positive control is intended to monitor substantial reagent and instrument failure.
- External negative controls may be treated as a patient specimen. Dip the provided swab into the external negative control ensuring liquid covers the tip. Identify the Dilution Buffer tube as the negative control and proceed with processing as described above in the Assay Procedure. The external negative control is used to detect reagent or environmental contamination (or carry-over) by *C. difficile* DNA or amplicon.

It is recommended that the reactivity of each new lot and each new shipment of the Solana C. difficile Assay be verified on receipt and before use. External control tests should be performed thereafter in accordance with appropriate federal,

state and local guidelines. The Solana C. difficile Assay should not be used in patient testing if the external controls do not produce the correct results.

LIMITATIONS

- A negative *C. difficile* result should not be used as the sole basis for diagnosis, treatment, or patient management decisions. Results should be interpreted in conjunction with other clinical and laboratory findings.
- Although there is no need for reagent preparation, the main laboratory technique required is pipetting; good laboratory technique is essential for the proper performance of this assay. Due to the high analytical sensitivity of this test, extreme care should be taken to preserve the purity of all reagents, especially in cases where multiple aliquots are taken from a tube.
- Mutations or polymorphisms in primer or probe binding regions may affect detection of new or unknown *C. difficile* variants and may result in a false negative result with the Solana C. difficile Assay.
- A positive test result does not necessarily indicate the presence of viable organisms.
- This test detects but does not differentiate hypervirulent strains from other toxigenic *C. difficile* genotypes.
- This test does not indicate the susceptibility of detected *C. difficile* strains to various antimicrobial agents.
- Negative test results may occur from improper specimen collection, handling or storage, presence of inhibitors, technical error, sample mix-up or because the number or organisms in the specimen is below the analytical sensitivity of the test. Careful compliance with the instructions given in this insert is necessary to avoid erroneous results. Use of this assay should be limited to personnel trained on the procedure.
- This test is for use with liquid or soft human stool specimens. Performance characteristics of other specimen types have not been established.

EXPECTED VALUES

The expected values of the Solana C. difficile Assay were established during a prospective study conducted between November 2016 to February 2017. Eight hundred fifty-four (854) specimens used for this study were collected from patients suspected of having *Clostridioides (Clostridium) difficile* infection (CDI) at three distinct geographical sites across the United States. A single specimen was collected per patient. The specimens were processed and tested with the Solana C. difficile Assay on the Solana instrument at the sites.

Patient age and gender for the combined sites are presented below.

Combined Sites – Age and Gender Distribution				
Age/Gender	**Female**	**Male**	**Total**	**Total percent positive with the Solana C. difficile Assay in Raw specimens**
≤ 2 years	3	3	6	16.7% (1/6)
3 to 11 years	4	6	10	20.0% (2/10)
12 to 17 years	4	10	14	7.1% (1/14)
18 to 21 years	11	14	25	24.0% (6/25)
22 to 59 years	206	132	338	14.2% (48/337*)
≥ 60 years	268	193	461	12.0% (55/460*)
Total	496	358	854	13.3% (113/852**)

* One (1) specimen was invalid

** Two (2) total specimens were invalid

CLINICAL PERFORMANCE

Performance characteristics of the Solana C. difficile Assay were established during a prospective study conducted November 2016 to February 2017. Eight hundred fifty-four (854) specimens used for this study were collected from patients suspected of having *Clostridioides (Clostridium) difficile* infection (CDI) at three distinct geographical sites across the United States. These specimens were tested raw with the Solana C. difficile Assay at the sites on the day of collection or after storage for up to three days at 2°C to 8°C. The Solana results were compared to a broth enhanced toxigenic bacterial culture (sensitivity/specificity) and a FDA-cleared molecular device (positive/negative percent agreement).

Performance in Comparison to broth Enhanced toxigenic bacterial culture

Eight hundred fifty-four (854) raw specimens were tested by both the Solana C. difficile Assay and enhanced toxigenic culture. For the toxigenic culture method, samples were inoculated into chopped-meat glucose (CMG) broth and after 48-hours sub-cultured onto CCFA-HB plates. Suspicious colonies were further characterized and *C. difficile* identified colonies were sub-cultured in CMG broth for subsequent cytotoxin testing. Two (2) specimens (0.2%) were invalid in the Solana C. difficile Assay when tested according to the Solana C. difficile Assay draft instructions for use. Both specimens remained invalid upon repeat testing. These specimens were removed from further analysis. The data below is for the remaining eight hundred fifty-two (852) specimens.

Combined Sites – Raw Specimen								
	Enhanced Toxigenic Culture							
		POS	NEG	Total			**95% CI**	
Solana C. difficile Assay	POS	107	6*	113	**Sensitivity**	93.0%	86.9%	96.4%
	NEG	8**	731	739	**Specificity**	99.2%	98.2%	99.6%
	Total	115	737	852				

* Three (3) of the six (6) specimens were positive for *C. difficile* toxin gene DNA by an alternative molecular device, three (3) were negative.

** Six (6) of eight (8) specimens were found positive for *C. difficile* toxin gene DNA by an alternative molecular device and two (2) were negative

Performance in Comparison to FDA-cleared molecular device

Eight hundred fifty-four (854) specimens were tested by both the Solana C. difficile Assay and FDA-cleared molecular device. Two (2) specimens (0.2%) were invalid in the Solana C. difficile Assay when tested according to the Solana C. difficile Assay draft instructions for use. Both specimens remained invalid upon repeat testing. These specimens were removed from further analysis. The data below is for the remaining eight hundred fifty-two (852) specimens.

Combined Sites – Raw Specimen								
	FDA-cleared molecular device							
		POS	NEG	Total			**95% CI**	
Solana C. difficile Assay	POS	97	16*	113	**Positive Percent Agreement**	97.0%	91.6%	99.0%
	NEG	3**	736	739	**Negative Percent Agreement**	97.9%	96.6%	98.7%
	Total	100	752	852				

* Twelve (12) of the sixteen (16) specimens were positive for *C. difficile* toxin gene DNA by an alternative molecular device, four (4) were negative.

** Two (2) of three (3) specimens were found positive for *C. difficile* toxin gene DNA by an alternative molecular device, and one (1) was negative

ANALYTICAL PERFORMANCE

Limit of Detection

The LOD of the Solana C. difficile Assay was determined using serial dilutions of two (2) toxigenic C. difficile strains, ATCC® BAA-1805 and CCUG 20309 spiked in negative matrix, and also quantified C. difficile genomic DNA, BAA-1382DQ™ spiked in lysis buffer. Analytical sensitivity (LOD) is defined as the lowest concentration at which 95% of all replicates tested positive.

Stool Matrix	*C. difficile* Strains	Strain LOD
Unpreserved Stool	ATCC BAA-1805	9.13E+03 CFU/mL
	CCUG 20309	4.90E+03CFU/mL
N/A	Genomic DNA: ATCC® BAA-1382DQ™	15 copy/assay

Analytical Reactivity (Inclusivity)

The reactivity of the Solana C. difficile Assay was evaluated against an additional twenty-three strains of *Clostridioides (Clostridium) difficile* representing multiple toxinotypes. The testing was performed on three replicates of each strain spiked in negative

stool matrix near the level of detection for the assay (1.83E+04 CFU/mL, 2X LOD for ATCC BAA-1805). All twenty-three additional strains were detected in all replicates by the Solana C. difficile Assay in this study.

Strain	Toxinotype
ATCC BAA-1805*	III
CCUG 20309*	X
ATCC BAA-1870	IIIb
CCUG 37770	IV
ATCC BAA-1875	V
ATCC 43598	VIII
ATCC 37774	XXIII
CCUG 9004	Unknown
ATCC BAA-1874	0
ATCC 43600	0
ATCC BAA-1871	0
ATCC BAA-1803	IIIc
ATCC BAA-1872	0
ATCC 700792	0
ATCC 43599	0
CCUG 60276	Unknown
CCUG 60275	Unknown
CCUG 37778	Unknown
CCUG 37777	Unknown
CCUG 37776	Unknown
CCUG 37773	Unknown
ATCC 17857	0
ATCC 43594	0
ATCC 43596	0
ATCC 43255	0

* C. difficile strains, ATCC BAA-1805 and CCUG 20309 were shown to be inclusive in the LOD study.

Reproducibility Study

In order to confirm the reproducibility of the Solana C. difficile Assay a blinded and randomized study panel containing *Clostridioides (Clostridium) difficile* negative and positive samples contrived in negative stool matrix were tested at three (3) test sites (two (2) clinical sites). Each site tested a reproducibility panel and Assay Controls for five (5) days in triplicate. Testing was done by two operators at each site. Each operator ran the panel once a day. A total of 540 specimens were tested (including controls). The Solana C. difficile Assay generated reproducible results in this study.

Sites	**Site #1**		**Site #2**		**Site #3**		**Overall Percent Agreement**		**95% Confidence Interval**
Category	*#expected results/# tested*	*% Agreement*	*#expected results/# tested*	*% Agreement*	*#expected results/# tested*	*% Agreement*			
C. difficile High Negative (4.8×10^2 CFU/mL)	12/30	40%	19/30	63.3%	12/30	40%	43/90	47.8%	37.8% - 58.0%
C. difficile Low Positive (1.7×10^3 CFU/mL)	30/30	100%	30/30	100%	29/30	96.7%	89/90	98.9%	94% - 99.8%
C. difficile Moderate Positive (3.4×10^3 CFU/mL)	30/30	100%	30/30	100%	30/30	100%	90/90	100%	95% - 100%
Negative	30/30	100%	30/30	100%	30/30	100%	90/90	100%	95% - 100%

Sites	Site #1		Site #2		Site #3		Overall Percent Agreement		95% Confidence Interval
Category	#expected results/# tested	% Agreement	#expected results/# tested	% Agreement	#expected results/# tested	% Agreement			
C. difficile Positive Control	30/30	100%	30/30	100%	30/30	100%	90/90	100%	95% - 100%
Assay Negative Control	30/30	100%	30/30	100%	30/30	100%	90/90	100%	95% - 100%

Analytical Specificity – Cross-reactivity and Microbial Interference

The analytical specificity of the Solana C. difficile Assay was evaluated by testing a panel consisting of sixty-eight (68) bacterial, viral and yeast microorganisms and human DNA representing common enteric pathogens, flora or nucleic acid commonly present in the intestine. Microorganisms or nucleic acid was mixed with pooled negative matrix and tested directly or in the presence of 1.83E+04 CFU/mL of *C. difficile* for cross-reactivity and microbial interference, respectively.

The table below lists the bacterial, viral and yeast microorganisms used in these studies. There was no evidence of cross reactivity or interference with any of the panel members and the Solana C. difficile Assay.

Organisms ID	Identification
Abiotrophia defective	ATCC 49176
Acinetobacter baumannii	ATCC 19606
Aeromonas hydrophila	ATCC 7966
Alcaligenes faecalis subsp. *faecalis*	ATCC 15554
Bacillus cereus	ATCC 13472
Bacteroides fragilis	ATCC 25285
Campylobacter coli	ATCC 43479
Campylobacter jejuni sub sp *.jejuni*	ATCC 33292
Candida albicans	ATCC 10231
Citrobacter freundii	ATCC 8090
Clostridioides (Clostridium) bifermentans	ATCC 638
Clostridioides (Clostridium) botulinum	
Clostridioides (Clostridium) butyricum	CCRI-11128
Clostridioides (Clostridium) haemolyticum	ATCC 19398
Clostridioides (Clostridium) novyi	ATCC 19402
Clostridioides (Clostridium) orbiscindens	ATCC 49531
Clostridioides (Clostridium) perfringens	ATCC 13124
Clostridioides (Clostridium) scindens	ATCC 35704
Clostridioides (Clostridium) septicum	ATCC 12464
Clostridioides (Clostridium) sordellii	ATCC 9714
Clostridioides (Clostridium) sordellii	Z077
Clostridioides (Clostridium) sordellii	CCUG 6329
Clostridioides (Clostridium) sordellii	CCUG 9284
Clostridioides (Clostridium) sordellii	CCUG 33098
Clostridioides (Clostridium) sordellii	CCUG 36938
Clostridioides (Clostridium) sordellii	CCUG 43123
Clostridioides (Clostridium) sordellii	CCUG 47545
Clostridioides (Clostridium) sordellii	CCUG 59819
Clostridioides (Clostridium) difficile	ATCC 43593
Clostridioides (Clostridium) difficile	ATCC 43601
Clostridioides (Clostridium) sporogenes	ATCC 15579
Edwardsiella tarda	ATCC 15947
Enterobacter aerogenes	ATCC 13048
Enterobacter cloacae	ATCC 13047

Organisms ID	Identification
Enterococcus faecalis vanB	ATCC 51299
Escherichia coli	ATCC 23511
Escherichia coli O157:H7	ATCC 700927
Helicobacter pylori	ATCC 43504
Klebsiella oxytoca	ATCC 33497
Lactobacillus acidophilus	ATCC 4356
Listeria monocytogenes	ATCC BAA-389
Peptostreptococcus anaerobius	ATCC 27337
Plesiomonas shigelloides	ATCC 14029
Porphyromonas asaccharolytica	ATCC 25260
Prevotella melaninogenica	ATCC 25845
Proteus mirabilis	ATCC 25933
Providencia alcalifaciens	ATCC 9886
Pseudomonas aeruginosa	ATCC 35554
Salmonella choleraesuis (typhimurium)	ATCC 14028
Salmonella enterica subsp. *arizonae*	ATCC 13314
Salmonella enterica subsp. *enterica*	ATCC 7001
Serratia liquefaciens	ATCC 27592
Serratia marcescens	ATCC 13880
Shigella boydii	ATCC 9207
Shigella dysenteriae	ATCC 11835
Shigella sonnei	ATCC 29930
Staphylococcus aureus	ATCC 43300
Staphylococcus epidermidis	ATCC 14990
Streptococcus agalactiae	ATCC 12973
Vibrio parahaemolyticus	ATCC 17802
Adenovirus	
Rotavirus	
Norovirus	
Enterovirus	
Echovirus	
Coxsackie virus	
Cytomegalovirus	
Human DNA	

Analytical Specificity – Interfering Substances

The performance of Solana C. difficile Assay was evaluated with potentially interfering substances that may be present in stool specimens. The potentially interfering substances were evaluated using two *C. difficile* strains (CCUG#20309 or ATCC BAA#1805) at a concentration of 1.83E+04 CFU/mL. There was no evidence of interference caused by the substances tested.

Substance Name	Active Ingredients	Test Concentration
Nystatin	Nystatin	1% (w/v)
Cortizone 10	Hydrocortisone	1% (w/v)
Fleet Glycerin Suppositories	Glycerin	1% (w/v)
Desitin	Zinc Oxide	1% (w/v)
Anusol Plus	pramoxine hydrochloride and zinc sulfate monohydrate	1% (w/v)
Preparation H	Phenylephrine	1% (w/v)
Nystatin	Nystatin	1% (w/v)
Cortizone 10	Hydrocortisone	1% (w/v)
Fleet Glycerin Suppositories	Glycerin	1% (w/v)

Substance Name	Active Ingredients	Test Concentration
Desitin	Zinc Oxide	1% (w/v)
Anusol Plus	pramoxine hydrochloride and zinc sulfate monohydrate	1% (w/v)
Preparation H	Phenylephrine	1% (w/v)
Tums	Calcium Carbonate	10% (w/v)
Equate Antacid Max Strength	Aluminum hydroxide, Magnesium hydroxide	10% (w/v)
Mesalazine Rectal Suspension Enema	Mesalazine	10% (w/v)
Fleet Mineral Oil Enema	Mineral Oil	10% (w/v)
Gynol II Vaginal Contraceptive	Nonoxynol-9	1% (w/v)
Imodium AD	Loperamide HCl	10% (w/v)
Pepto Bismol	Bismuth subsalicylate	10% (w/v)
Ex-Lax	Sennosides	1% (w/v)
Metronidazole	Metronidazole	12.5 mg/ml
Vancomycin	Vancomycin	12.5 mg/ml
Polysporin	Bacitracin and Polymyxin B	1% (w/v)
Naproxen sodium	Naproxen sodium	12.5 mg/ml
Tucks personal cleaning pads	Witch hazel	10% (v/v)
Benzalkonium Chloride Towelettes	Benzalkonium Chloride	10% (v/v)
Ethanol	Ethanol	10% (v/v)
Mucus	Immunoglobulins, Lysozyme, Polymers, etc.	3.5%
Whole Blood	Glucose, Hormones, Enzymes, Ions, Iron, etc.	10%
Palmitic acid	Palmitic acid	12.5 mg/ml
Steric Acid	Steric Acid	12.5 mg/ml
Triglyceride Mix (C2 – C10)	Triglyceride	10%

None of the thirty-two (32) potential interfering substances that may be present in stool specimens cross-reacted or interfered with the Solana C. difficile Assay.

Carryover – Cross Contamination

A study was conducted to demonstrate that carry-over and cross contamination does not occur when the intended users perform the Solana C. difficile Assay following the Package Insert instructions.

Two (2) samples were prepared: *C. difficile* positive sample and *C. difficile* negative sample. The positive sample was prepared by adding cells of one *C. difficile* strain (CCUG 20309) with a known titer to negative stool matrix at the concentration of 4.9×10^6 CFU/mL (approximately 1000X LOD). The negative stool matrix served as the *C. difficile* negative sample. In each experiment, the positive samples were alternated with the negative samples and tested using Solana C. difficile Assay to assess the risk of cross contamination. In total, two (2) operators tested a total of 50 positive and 50 negative samples over a total of 11 runs.

All positive *C. difficile* samples were positive and all negative samples were negative. No evidence of carry-over/ cross contamination was observed with the Solana C. difficile Assay when performed in accordance with the package insert.

CUSTOMER AND TECHNICAL ASSISTANCE

If you have any questions regarding the use of this product, please contact Quidel Technical Support at 1.800.874.1517 (in the U.S.) or technicalsupport@quidel.com. If outside the U.S., further information can be obtained from your distributor, or directly from Quidel at one of the numbers listed below. Reference **quidel.com** to see more options for Support.

Country	Phone	E-Mail Address
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