510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number:

K181029

B. Purpose for Submission:

Clearance of the Solana Bordetella Complete Assay on the Solana instrument

C. Measurand:

Bordetella pertussis: IS481 Insertional Element sequence;
Bordetella parapertussis: IS1001 Insertional Element sequence

D. Type of Test:

Helicase-Dependent Amplification (HDA)

E. Applicant:

Quidel Corporation

F. Proprietary and Established Names:

Solana Bordetella Complete Assay

G. Regulatory Information:

1. Regulation section:

   866.3980

   Respiratory viral panel multiplex nucleic acid assay

2. Classification:

   Class II

3. Product code:

   OZZ

4. Panel:

   Microbiology (83)

H. Intended Use:

1. Intended use(s):

   The Solana Bordetella Complete Assay is an *in vitro* diagnostic test for the qualitative detection of *Bordetella pertussis* and *Bordetella parapertussis* nucleic acids isolated from nasopharyngeal swab specimens obtained from patients suspected of having respiratory tract infection attributable to *Bordetella pertussis* and *Bordetella parapertussis*.

   The Solana Bordetella Complete Assay is an HDA-based duplex assay that targets the IS481 and IS1001 sequence of *Bordetella pertussis* (BP) and *Bordetella parapertussis* (BPP) genomes, respectively. The IS481 sequence may also be found in strains of other organisms (i.e., *B. holmesii* and *B. bronchiseptica*). The IS1001 sequence may also be found in strains of other organisms (i.e., *B. bronchiseptica*). *B. holmesii* infection may cause clinical illness similar to *B. pertussis*, and mixed outbreaks involving both *B. pertussis* and *B. holmesii* infection have been reported. Additional testing should be performed if necessary to differentiate *B. holmesii* and *B. pertussis*. *B. bronchiseptica* is a rare cause of infection in humans. When clinical factors suggest that *B. pertussis* or *B. parapertussis* may not be the cause of respiratory infection, other clinically appropriate investigation(s) should be carried out in accordance with published guidelines.

   Negative results for the Solana Bordetella Complete Assay do not preclude *B. pertussis* or *B. parapertussis* infection and positive results do not rule out co-infection with other respiratory pathogens. Results from the Solana Bordetella Complete Assay should be used in conjunction with information obtained during the patient's clinical evaluation as an aid in diagnosis of *B. pertussis* and or *B. parapertussis* infection and should not be used as the sole basis for treatment or other patient management decisions.

2. Indication(s) for use:

   Same as intended use.

3. Special conditions for use statement(s):

   - For *in vitro* diagnostic use only
   - For prescription use only

4. Special instrument requirements:

   Solana Instrument

I. Device Description:

The Solana Bordetella Complete Assay amplifies, detects and differentiates DNA of *Bordetella pertussis* and *Bordetella parapertussis* from nasopharyngeal swabs.

The assay consists of two major steps: 1) specimen preparation, and 2) amplification and detection of target sequences specific to *B. pertussis* and *B. parapertussis* using isothermal Helicase-Dependent Amplification (HDA) in the presence of target-specific fluorescence probes.

A patient nasopharyngeal swab specimen in transport media is transferred to a Process Buffer Tube, subjected to heat treatment at 95°C for 5 minutes and mixed. The processed sample is transferred to a Reaction Tube. The Reaction Tube contains lyophilized HDA reagents, dNTPs, primers and probes. Once rehydrated with the processed sample, the Reaction Tube is placed in Solana for amplification and detection of *B. pertussis* and *B. parapertussis*-specific target sequences. In Solana, the target sequences are amplified by *B. pertussis* and *B. parapertussis* specific primers and detected by *B. pertussis* and *B. parapertussis*-specific fluorescence probes, respectively. A process control (PRC) is included in the Process Buffer Tube to monitor sample processing, inhibitory substances in clinical samples, reagent failure or device failure. The PRC target is amplified by specific primers and detected by a PRC specific fluorescence probe.

The two target probes and PRC probe are labeled with a quencher on one end and a fluorophore on the other end. In addition, the two target probes and PRC probe have one or more bases that are comprised of ribonucleic acid. Upon annealing to *B. pertussis* and *B. parapertussis* or PRC amplicons, the fluorescence probes are cleaved by RNaseH2 and 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 an attached printer.

Materials Provided:

Solana Bordetella Complete Assay Kit: M308, 48 Tests per kit

Kit Components		
Component	**Quantity**	**Storage**
Process Buffer	48 tubes/kit, 1.45 mL	2°C to 8°C
Reaction Tubes	48 tubes/kit	2°C to 8°C

Materials required but not provided:

- External controls for *B. pertussis* and *B. parapertussis* (e.g. Quidel Molecular Bordetella Control Set, Cat. No. M117 which contains positive and negative controls, serves as an external processing control)
- Sterile DNase-free filter-blocked positive displacement micropipettor tips

- Micropipettor
- Stopwatch or timer
- Scissors or a blade
- Workflow tray
- Transfer Rack
- Heat block capable of 95° ± 2°C temperature
- Thermometer
- Solana instrument

J. Substantial Equivalence Information:

1. Predicate device name(s):

   ARIES Bordetella Assay

2. Predicate 510(k) number(s):

   K163626

3. Comparison with predicate:

Similarities		
Item	**Solana Bordetella Complete Assay** (K181029)	**ARIES Bordetella Assay (K163626)**
Intended Use	An assay for the qualitative detection of *Bordetella pertussis* and *Bordetella parapertussis* nucleic acids isolated from nasopharyngeal swab specimens obtained from individuals suspected of having a respiratory tract infection attributable to *B. pertussis* or *B. parapertussis*. When clinical factors suggest that *B. pertussis* or *B. parapertussis* may not be the cause of respiratory infection, other clinically appropriate investigation(s) should be carried out in accordance with published guidelines. Negative results for the Solana Bordetella Complete Assay do not preclude *B. pertussis* or	An assay for the direct detection and identification of *Bordetella pertussis* (*B. pertussis*) and *Bordetella parapertussis* (*B. parapertussis*) nucleic acid in nasopharyngeal swab (NPS) specimens obtained from individuals suspected of having a respiratory tract infection attributable to *B. pertussis* or *B. parapertussis*. When clinical factors suggest that *B. pertussis* or *B. parapertussis* may not be the cause of respiratory infection, other clinically appropriate investigation(s) should be carried out in accordance with published guidelines. Negative results for the ARIES

	B. parapertussis infection and positive results do not rule out co-infection with other respiratory pathogens. Results from the Solana Bordetella Complete Assay should be used in conjunction with information obtained during the patient's clinical evaluation as an aid in diagnosis of *B. pertussis* and or *B. parapertussis* infection and should not be used as the sole basis for treatment or other patient management decisions.	Bordetella Assay do not preclude *B. pertussis* or *B. parapertussis* infection and positive results do not rule out co-infections with other respiratory pathogens. The direct detection and identification of *B. pertussis* and *B. parapertussis* nucleic acids from symptomatic patients aids in the diagnosis of *B. pertussis* and *B. parapertussis* respiratory infection in conjunction with other clinical findings and epidemiological information.
Sample Type	Nasopharyngeal swab	Same

Differences		
Item	**Solana Bordetella Complete Assay**	**ARIES Bordetella Assay (k163626)**
Target	*Bordetella pertussis*: IS481 Insertional Element sequence; *Bordetella parapertussis*: IS1001 Insertional Element sequence	*Bordetella pertussis*: Toxin promoter (ptxA-pr); *Bordetella parapertussis*: IS1001 sequence
Amplification Technology	Helicase-Dependent Amplification (HDA)	Real Time PCR
Detection Techniques	Different fluorescent reporter dyes for each target. Fluorescence Emissions and Detection	Different fluorescent reporter dyes for each target and melt analysis. Fluorescence Emissions and Detection
Extraction Methods	None	Automated by the ARIES Systems
Instrument	Solana	ARIES Systems

K. Standard/Guidance Document Referenced (if applicable):

Guidance for Industry and FDA Staff - Class II Special Controls Guidance Document: Respiratory Viral Panel Multiplex Nucleic Acid Assay

Guidance for Industry and FDA Staff: Statistical Guidance on Reporting Results from Studies Evaluating Diagnostic Tests (Final, 3/13/2007)

Guidance on Informed Consent for In Vitro Diagnostic Device Studies Leftover Human Specimens that are Not Individually Identifiable (April 2006)

Guidance for Industry and Food and Drug Administration Staff - eCopy Program for Medical Device (December 2012)

L. Test Principle:

The assay consists of two major steps: 1) specimen preparation, and 2) amplification and detection of target sequences specific to *B. pertussis* and *B. parapertussis* using isothermal Helicase-Dependent Amplification (HDA) in the presence of target-specific fluorescence probes.

A patient nasopharyngeal swab specimen in transport media is transferred to a Process Buffer Tube, subjected to heat treatment at 95° ± 2°C for 5 minutes and mixed. The processed sample is transferred to a Reaction Tube. The Reaction Tube contains lyophilized HDA reagents, dNTPs, primers and probes. Once rehydrated with the processed sample, the Reaction Tube is placed in Solana for amplification and detection of *B. pertussis* and *B. parapertussis*-specific target sequences. In Solana, the target sequences are amplified by *B. pertussis* and *B. parapertussis* specific primers and detected by *B. pertussis* and *B. parapertussis*-specific fluorescence probes, respectively. A process control (PRC) is included in the Process Buffer Tube to monitor sample processing, inhibitory substances in clinical samples, reagent failure or device failure. The PRC target is amplified by specific primers and detected by a PRC specific fluorescence probe.

The two target probes and PRC probe are labeled with a quencher on one end and a fluorophore on the other end. In addition, the two target probes and PRC probe have one or more bases that are comprised of ribonucleic acid. Upon annealing to *B. pertussis* and *B. parapertussis* or PRC amplicons, the fluorescence probes are cleaved by RNaseH2 and 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 an attached printer.

M. Performance Characteristics:

1. Analytical Performance:

   a. Precision/Reproducibility

   A repeatability study was conducted using a panel of 30 contrived samples with two (2) subsets of 15 samples, where operator 1 tested samples 1-15 and operator 2 tested samples 16-30. Each subset was manufactured as negative samples (n=3), high negative samples containing BP and BPP (n=3; C_{20} – C_{80} concentration per target), low positive samples containing BP or BPP (n=3; near the assay limit of detection), and moderate positive samples containing BP and BPP (n=3; 2x LoD per target) (see Table below). The positive samples were generated from two (2) separate strains each of BP and BPP in equal proportions. The samples were randomized and blind-coded within each panel, and each operator tested one (1)

panel subset, along with three (3) positive and three (3) negative external controls, in two (2) runs.

Panels and controls were tested at one (1) site by two (2) operators per instrument for fifteen (15) days, each sample tested in three (3) replicates, for a total of 45 results per level for each organism strain (2 operators x 15 days x 3 replicates). The following table (Table 1) describes the repeatability test panel subset composition, and the corresponding expected results.

Table 1. Repeatability Panel Subset Composition

Panel Members	# of Samples	BP or BPP Concentrations	Expected Results
Negative	3	0	Negative: 100%
BP+BPP High Negative	3	**C20 – C80 per target** BP A639 (103 CFU/mL) BP E431 (86 CFU/mL) BPP A747 (462 CFU/mL) BPP E838 (553 CFU/mL)	BP Positive: 20-80%; BPP Positive: 20-80%;
BP Low Positive	3	**1X LoD** BP A639 (1025 CFU/mL) BP E431 (862 CFU/mL)	BP Positive: ≥ 95%
BPP Low Positive	3	**1X LoD** BPP A747 (4622 CFU/mL) BPP E838 (5533 CFU/mL)	BPP Positive: ≥ 95%
BP+BPP Moderate Positive	3	**2X LoD per target** BP A639 (2050 CFU/mL) BP E431 (1724 CFU/mL) BPP A747 (9244 CFU/mL)BPP E838 (11,066 CFU/mL)	BP Positive: 100%; BPP Positive: 100%

Negative nasal matrix without spiked organism was used for the negative sample. Positive and negative controls were run in triplicate along with the panels. The panels were run by four (4) operators for fifteen (15) non-consecutive days.

A summary of the repeatability testing is as follows:

Table 2. Repeatability Testing Summary Results-*B. pertussis*

Bordetella pertussis	**# Detected**		
	Moderate Positive	Low Positive	High Negative
BP A639	45	45	28
BP E431	45	45	27
% Detected	90/90 (100%)	90/90 (100%)	55/90 (61.1%)

Table 3. Repeatability Testing Summary Results-*B. parapertussis*			
Bordetella parapertussis	**# Detected**		
	Moderate Positive	Low Positive	High Negative
BPP A747	45	45	30
BPP E838	45	45	32
% Detected	90/90 (100%)	90/90 (100%)	62/90 (68.9%)

Table 4. Repeatability Testing Summary Results-Controls		
Samples	**# Detected**	**% Detected**
Negative Specimen	0	0
Negative Control	0	0
BP Positive Control	90	100
BPP Positive Control	90	100

A multi-center reproducibility study was conducted using two different four-sample panel sets each consisting of three levels of combined *B. pertussis* (BP) and *B. parapertussis* (BPP) (two (2) strains of each organism) contrived samples and a negative contrived sample. Set 1 consisted of BP A639 and BPP A747, Set 2 consisted of BP E431 and BPP E838. Samples were diluted in negative nasal matrix (UTM) to 2x LoD for moderate positive, 1x LoD for low positive and diluted to concentrations below the LoD (i.e., C_{20} to C_{80}) for high negative samples. Negative nasal matrix without spiked organism was used for the negative sample. Positive and negative controls were run in triplicate along with the panels.

Panels and controls were tested over five days at three testing sites by two (2) operators per instrument for five (5) days with each sample tested in three (3) replicates, for a total of 45 results per level for each organism strain (2 operators x 5 days x 3 sites x 3 replicates). Results are shown in Table 5 below.

Table 5. Reproducibility Summary								
	SITE						**Overall Percent Agreement**	**95% Confidence Interval**
	Site #1		**Site #2**		**Site #3**			
	# Expected Result/# tested	*% Agreement with Expected Result*	*# Expected Result/# tested*	*% Agreement with Expected Result*	*# Expected Result/# tested*	*% Agreement with Expected Result*		

Table 5. Reproducibility Summary									
	SITE						Overall Percent Agreement		95% Confidence Interval
	Site #1		Site #2		Site #3				
	# Expected Result/# tested	*% Agreement with Expected Result*	*# Expected Result/# tested*	*% Agreement with Expected Result*	*# Expected Result/# tested*	*% Agreement with Expected Result*			
BP strain A639 High Negative[1] (103 CFU/mL)	7/15	46.6	7/15	46.6	6/15	40.0	20/45	44.4	30.9 to 58.8
BP strain A639 Low Positive (1025 CFU/mL)	15/15	100	15/15	100	15/15	100	45/45	100	92.1 to 100
BP strain A639 Moderate Positive (2050 CFU/mL)	15/15	100	15/15	100	15/15	100	45/45	100	92.1 to 100
BP strain E431 High Negative[1] (86 CFU/mL)	4/15	26.7	10/15	66.7	7/15	46.6	21/45	46.7	32.9 to 60.9
BP strain E431 Low Positive (862 CFU/mL)	15/15	100	15/15	100	15/15	100	45/45	100	92.1 to 100
BP strain E431 Moderate Positive (1724 CFU/mL)	15/15	100	15/15	100	15/15	100	45/45	100	92.1 to 100
BPP strain A747 High Negative[1] (462 CFU/mL)	6/15	40.0	7/15	46.6	1/15	6.7	12/45	26.7	16.0 to 41.0
BPP strain A747 Low Positive (4622 CFU/mL)	15/15	100	15/15	100	15/15	100	45/45	100	92.1 to 100
BPP strain A747 Moderate Positive (9244 CFU/mL)	15/15	100	15/15	100	15/15	100	45/45	100	92.1 to 100
BPP strain E838 High Negative[1] (553CFU/mL)	6/15	40.0	8/15	53.3	3/15	20.0	17/45	37.8	25.1 to 52.4
BPP strain E838 Low Positive (5533 CFU/mL)	15/15	100	15/15	100	15/15	100	45/45	100	92.1 to 100
BPP strain E838 Moderate Positive	15/15	100	15/15	100	15/15	100	45/45	100	92.1 to 100

Table 5. Reproducibility Summary									
	SITE						Overall Percent Agreement		95% Confidence Interval
	Site #1		Site #2		Site #3				
	# Expected Result/# tested	% Agreement with Expected Result	# Expected Result/# tested	% Agreement with Expected Result	# Expected Result/# tested	% Agreement with Expected Result			
(11,066 CFU/mL)									
Negative Sample	0/30	100	0/30	100	0/30	100	0/90	100	95.9 to 100
BP Positive Control	30/30	100	30/30	100	30/30	100	90/90	100	95.9 to 100
BPP Positive Control	30/30	100	30/30	100	30/30	100	90/90	100	95.9 to 100
Negative Control	0/30	100	0/30	100	0/30	100	0/90	100	95.9 to 100

[1] An expected result for the high negative sample is percent positivity between 20 and 80%.

The results suggest that there are no significant differences in assay performance between different users and different sites on different days.

b. Linearity/assay reportable range:

Not applicable.This assay is qualitative.

c. Traceability, Stability, Expected values (controls, calibrators, or methods):

Traceability

Not applicable. This assay is qualitative.

Specimen Stability

Contrived BP and BPP positive specimens were prepared by diluting BP or BPP cells to the 2x LOD concentration of each target in 3 mL of negative matrix for each of the eight transport media types that are claimed for use with the Solana Bordetella Assay: Molecular Grade Water, Saline (0.9%), Tris-EDTA, Amies Liquid transport media (or Eswab), M4, M4-RT, M5, and UTM were tested with one validation lot of reagents and the Solana Bordetella assay workflow.

The specimens were stored at three different storage conditions—≤ -70°C, 2°C to 8°C, 25°C ± 3°C. Testing indicated that specimens can be stored before processing at 2° to 8°C for up to 97 hours, up to 49 hours at 25°C, or up to 5-months at ≤ -70°C.

No significant difference in assay performance was seen between the four different types of viral transport media, saline (0.85%), Tris EDTA, Molecular Grade Water, or Amies.

Processed Specimen Stability

Contrived samples were prepared in negative nasal matrix with organism concetnrations of 2x LOD for BP (Strain A639, 2050 CFU/mL) and 2x LOD for BPP (Strain E838, 11066 CFU/mL). The stability of samples in Process Buffer at 2°C to 8°C and at 25°C ± 3°C before and after heat lysis was evaluated in this study. Three (3) replicates were processed and tested before heat lysis and after heat lysis conditions according to the instructions for use. The condition, before or after heating, and testing time points for the corresponding samples in Process Buffer kept at the evaluated temperatures are listed in Table 6 below.

Table 6. Process Specimen Stability Conditions

Lysis Buffer Tubes	Time Points Tested
Before Heating	0, 24, 48, 72, 73 hours
After Heating	0, 24, 48, 72, 73 hours

Results from the study demonstrated that BP- and BPP-positive samples in Process Buffer generated the expected positive results after storage at at 2°C to 8°C for 97 hours and at 25°C ± 3°C for 49 hours both before and after undergoing heat lysis.

Controls

Process Control

The process control is used to monitor sample processing, to detect HDA inhibitory specimens, to confirm the integrity of assay reagents and the operation of the Solana instrument. The process control is included in the Process Buffer tube used for each specimen.

External Controls

Controls (Quidel Molecular Bordetella Control Set, which contains positive and negative controls and serves as an external processing control) were run on the Solana Bordetella Complete Assay each day of testing during the clinical study. A total of 117 positive controls (BP-positive and BPP-positive) and 117 negative controls (BP- and BPP-negative) were evaluated across five (5) clinical test sites.

All controls produced the expected results.

These controls are described as follows:

- The Quidel Molecular Bordetella external positive control may be treated as a patient specimen. The control should be sampled and tested as if it were a patient specimen and processed as described above in the Assay Procedure. The external positive control is intended to monitor substantial reagent and instrument failure.
- The Quidel Molecular Bordetella external negative control may be treated as a patient specimen. The control should be sampled and tested as if it were a patient specimen and processed as described above in the Assay Procedure. The external negative control is used to detect reagent or environmental contamination (or carry-over) by *B. pertussis* and *B. parapertussis* DNA or amplicon.

It is recommended that the reactivity of each new lot and each new shipment of the Solana Bordetella Complete Assay be verified on receipt and before use using the Quidel Molecular Bordetella Control Set External control tests should be performed thereafter in accordance with appropriate federal, state and local guidelines. The Solana Bordetella Complete Assay should not be used in patient testing if the external controls do not produce the correct results.

d. Limit of Detection

The analytical sensitivity (limit of detection or LoD) of the Solana Bordetella Complete Assay was determined using quantified (CFU/mL) cultures of two (2) BP strains (A639 and E431) and two (2) BPP strains (A747 and E838) serially diluted in a negative nasal matrix. Results are presented in Table 7.

Table 7. Limit of Detection

Target Type	Target	Validation Lot	Determined LOD	
			CFU/mL	CFU/Assay
Fresh Cells	BP Strain: A639	1	1025	1.71
		2	1025	1.71
		3	1025	1.71
	BP Strain: E431	1	863	1.44
		2	863	1.44
		3	863	1.44
	BPP Strain: A747	1	4622	7.70
		2	4622	7.70
		3	4622	7.70
	BPP Strain: E838	1	5533	9.22
		2	5533	9.22

Table 7. Limit of Detection				
Target Type	**Target**	**Validation Lot**	**Determined LOD**	
			CFU/mL	**CFU/Assay**
		3	5533	9.22
Assay LOD: BP			1025	1.71
Assay LOD: BPP			5533	9.22

e. Inclusivity

The reactivity of the Solana Bordetella Complete Assay was evaluated against an additional eight (8) *Bordetella pertussis* (BP) and eight (8) *Bordetella parapertussis* (BPP) strains that were not used for LOD determination. The testing was performed at 1x LOD (1025 CFU/mL and 5533 CFU/mL, respectively) level of the assay. All additional sixteen (16) strains were detected in the Solana Bordetella Complete Assay (Table 8).

Table 8. Inclusivity Strains	
BP Strains Detected at 1X LOD (1025 CFU/mL)	**BPP Strains Detected at 1X LOD (5533 CFU/mL)**
ATCC 9340	ZeptoMetrix C510
ATCC 9797	ZeptoMetrix E595
ATCC BAA-1335	ATCC 15311
ATCC BAA-589	ATCC 15989
ATCC 51445	ATCC 53892
ATCC 10380	ATCC 53893
ATCC 8478	ATCC BAA-587
ATCC 12743	ATCC 15237

To supplement inclusivity testing for each Solana Bordetella Complete assay target, *in-silico* analysis was performed as an additional evaluation of Solana Bordetella BP and BPP amplicon sequence for the predicted detection of microorganism strains.

f. Analytical Specificity:

Cross-Reactivity

Based on in-silico analysis, *B. holmesii* and *B. bronchiseptica* genomes are expected to share complete or nearly complete sequence identity with the BP amplicon because both contain the IS*481* sequence. Therefore, these organisms are expected to cross-react with the Solana Bordetella Assay. Other microorganisms are not expected to cause cross-reactivity because their sequence alignment with the BP amplicon shows multiple mismatches in the primer and probe binding regions. It is also be noted that

no *Bordetella parapertussis* sequences were retrieved from the blastn search, suggesting that there is little to no sequence identity shared between *B. parapertussis* and the BP amplicon. Therefore, the BP primers are not expected to amplify any BPP sequences.

Furthermore, based on in-silico analysis, the *B. bronchiseptica* genome is expected to share complete or nearly complete sequence identity with the BPP amplicon because it contains the IS*1001* sequence. Therefore, it is expected to cross-react with the Solana Bordetella Assay. Other microorganisms are not expected to cause cross-reactivity because their sequence alignment with the BPP amplicon shows multiple mismatches in the primer and probe binding regions, which are not likely to promote BPP primer or probe binding. It is also be noted that no *Bordetella pertussis* sequences were retrieved from the blastn search, suggesting that there is little to no sequence identity shared between BP and the BPP amplicon. Therefore, the BPP primers are not expected to amplify any BP sequences.

A study was performed to determine if eighty-three (83) microorganisms or viruses likely to be present in nasopharyngeal swabs cross-react with the Solana Bordetella Complete Assay. The microorganisms (Table 9, 10, 11) were tested above clinically relevant levels (bacteria/yeast $\geq 1x10^6$ CFU/mL, viruses $1x10^5$TCID50/mL).

Five (5) non-BP organism strains tested positive for BP for all three (3) replicates. The five organisms include 4 of 4 *Bordetella holmesii* strains (ZeptoMetrix F061, ATCC 51541, ATCC 700053, and ATCC 700052) and 1 of 8 *Bordetella bronchiseptica* strains (ATCC 4617). Cross-reactivity with these organisms is expected due to the presence of the BP target sequence, IS481, in the genomes of these organisms.

None of the potentially cross-reactive organisms tested positive for BPP.

High levels of BP did not generate any BPP-positive results, and high levels of BPP did not generate any BP-positive results demonstrating no cross-reactivity between the two organism targets.

Table 9. Organism - Bacteria		
Acinetobacter baumanii	*Bordetella pertussis A639*	*Legionella pneumophilia*
Arcanobacterium haemolyticum	*Bordetella petrii*	*Moraxella catarrhalis*
Bacteroides fragilis	*Bordetella trematum*	*Morganella morganii*
Bordetella avium	*Burkholderia cenocepacia*	*Mycobacterium avium*
Bordetella bronchiseptica (ATCC 780)	*Burkholderia cepacia*	*Mycobacterium tuberculosis (avirulent)*
Bordetella bronchiseptica (ZeptoMetrix 801649)	*Burkholderia multivorans*	*Mycoplasma pneumoniae*

Table 9. Organism - Bacteria		
Bordetella bronchiseptica (ATCC 4617)	*Burkholderia thailandensis*	*Neisseria gonorrhoeae*
Bordetella bronchiseptica (ATCC 10580)	*Chlamydia trachomatis*	*Neisseria meningitidis*
Bordetella bronchiseptica (ATCC BAA-588)	*Chlamydophila pneumoniae*	*Neisseria mucosa*
Bordetella bronchiseptica (ATCC 785)	*Corynebacterium diptheriae*	*Parvimonas micra*
Bordetella bronchiseptica (ATCC 786)	*Enterobacter aerogenes*	*Proteus mirabilis*
Bordetella bronchiseptica (ATCC 14064)	*Enterococcus faecalis*	*Proteus vulgaris*
Bordetella hinzii	*Escherichia coli*	*Pseudomonas aeruginosa*
Bordetella holmesii (ZeptoMetrix F061)	*Fusobacterium necrophorum*	*Staphylococcus aureus (MRSA)*
Bordetella holmesii (ATCC 51541)	*Haemophilus influenzae*	*Staphylococcus epidermidis*
Bordetella holmesii (ATCC 700053)	*Klebsiella pneumoniae*	*Stenotrophomonas maltophilia*
Bordetella holmesii (ATCC 700052)	*Lactobacillus acidophilus*	*Streptococcus pneumoniae*
Bordetella parapertussis E838	*Lactobacillus plantarum*	*Streptococcus pyogenes*
		Streptococcus salivarius

Table 10. Yeast
Candida albicans

Table 11. Viruses	
Adenovirus 31	HSV Type 2 G strain
Coronavirus 229E	Influenza A/Mexico/4108/2009
Coronavirus NL63	Influenza B/Florida/04/2006
Coronavirus OC43	Measles virus
Coxsackievirus B4	Metapneumovirus A1
Coxsackievirus B5/10/2006	Mumps virus
Echovirus 6	Parainfluenza Type 1 (#2)
Echovirus 7	Parainfluenza Type 2 (Greer)
Echovirus 9	Parainfluenza Type 3 (C234)
Echovirus 11	Parainfluenza Type 4 (VR-1377)
Enterovirus 70	Respiratory Syncytial Virus A
Enterovirus 71	Rhinovirus 1A
Epstein-Barr Virus	Varicella Zoster Virus
HSV Type 1 Maclnytre strain	

Microbial Interference

A study was performed to determine if eighty-three (83) microorganisms or viruses likely to be present in nasopharyngeal specimens interfere with detection of the *Bordetella pertussis* (BP) or *Bordetella parapertussis* (BPP) with the Solana Bordetella Complete Assay. Samples were prepared with one BP strain (A639) and one BPP strain (E838) at 2x LoD (2050 CFU/mL and 11066 CFU/mL, respectively) combined with each potentially interfering microorganism. The potentially interfering microorganisms (Table 9, 10, 11) were tested above clinically relevant levels (bacteria/yeast $\geq 1x10^6$ CFU/mL, viruses $1x10^5$ TCID50/mL).

None (0) of the organisms or viruses tested above interfered with the performance of the Solana Bordetella Complete Assay for detection of BP or BPP.

High levels of BP A639 did not cause interference with BPP E838 detection, and high levels of BPP E838 did not cause interference with BP A639 detection.

Interfering Substances

The performance of Solana Bordetella Complete Assay was evaluated with sixteen (16) potentially interfering substances that may be present in nasopharygeal specimens collected to test for *Bordetella pertussis* (BP) and *Bordetella parapertussis* (BPP) infection. The substances listed in Table 12 were diluted in negative nasal matrix and tested in the absence or presence of 2x LOD BP (Strain A639 2050 CFU/mL) and 2x LOD BPP (Strain E838 11066 CFU/mL) in the Solana Bordetella Complete Assay.

Table 12. Interfering Substances			
Substance	**Concentration Tested**	**Substance**	**Concentration Tested**
Cepacol Sore Throat Lozenges	5% w/v	Neo-Synephrine	15% v/v
Halls Cherry Menthol-Lyptus Cough Drops	15% w/v	Afrin Nasal Spray Original	15% v/v
Children's Dimetapp	15% v/v	Zicam Non-Drowsy Allergy Relief Nasal Gel	5% v/v
Chloraseptic Sore Throat Lozenges	10% w/v	Rite Aid Brand Saline Nasal Spray	15% v/v
Ricola Original Swiss Sugar-Free Herb Cough Suppressant Throat Drops	15% w/v	Zanamivir (Relenza)	5 mg/mL
Sucrets Complete Lozenges-Vapor Cherry	5% w/v	Tobramycin	4 μg/mL
Mucin (Bovine Submaxillary Gland, Type I-S)	5 mg/mL	Mupirocin	10 mg/mL
Human Blood, EDTA anticoagulated	5% v/v	Oseltamivir Phosphate (Tamiflu)	10 mg/mL

Negative sample testing in the presence of each of the sixteen (16) substances produced negative results for 3 of 3 replicates. BP+BPP sample testing at 2x LoD levels in the presence of each of the 16 substances produced BP and BPP positive results for 3 of 3 replicates. Based on these results, the 16 substances tested in this study are considered to not interfere with the Solana Bordetella Complete Assay.

g. Carry-over/Cross-contamination

A study was conducted to evaluated the potential for carry-over or cross-contamination for the Solana Bordetella Complete Assay. For the study two (2) samples were prepared: a *Bordetella pertussis* (BP) positive sample and a BP negative sample. The positive sample was prepared by adding cells of one BP strain with a known titer to negative nasal matrix at the concentration of 1.0×10^6 CFU/mL. The negative nasal matrix served as the BP negative sample. In each experiment, the positive samples were alternated with the negative samples and tested using Solana Bordetella Complete Assay to assess the risk of cross contamination. In total, two (2) operators tested a total of 30 positive and 30

negative samples over a total of five (5) runs. All positive BP samples tested positive and all negative BP samples tested negative. No evidence of carry-over/cross contamination was observed with the Solana Bordetella Complete Assay when performed in accordance with the package insert.

h. Fresh versus frozen study

A study was performed to demonstrate equivalency between fresh and frozen specimens. Two (2) BP strains (A639 394 and E431) and two (2) BPP strains (A747 and E838) were serially diluted in a negative nasal matrix at varying concentrations above and below the assay LOD. The dilutions were tested fresh and then after freezing at -70°C. The dilutions demonstrating at least 95% detection (≥19 of 20) were thawed and re-tested. For all four (4) organism strains tested, the fresh and frozen results matched at LOD concentrations indicating equivalence between fresh and frozen samples when tested with the Solana Bordetella Complete Assay.

i. Assay cut-off:

The preliminary baseline, cut-off, and threshold settings were determined for the Solana Bordetella Complete Assay on the Solana instrument during development. The settings were based on the analysis of negative clinical specimens (these specimens were not part of the clinical study), as well as analysis of contrived specimens for the Limit of Detection Study. The baseline, threshold, and cut-off settings were established and used during the clinical trial.

2. Comparison studies:

a. Method comparison with predicate device:

Not applicable

b. Matrix comparison:

Not applicable

3. Clinical studies:

A multi-center study was conducted to evaluate the Solana Bordetella Complete Assay using nasopharyngeal swab specimens obtained from patients suspected of having respiratory tract infection attributable to *Bordetella pertussis* or *Bordetella parapertussis*. The study was performed from October, 2017 to January, 2018 at one (1) internal and four (4) external sites in the United States. Specimens were collected using either Copan NP flocked swab or rayon swab at the various test sites, and both swab types were used for each site's test of record *Bordetella* testing. The study collected seven hundred forty-

one (741) fresh specimens at four (4) sites, and two hundred thirty-three (233) frozen archival specimens at four (4) sites.

Specimen collection followed the FDA guidance entitled "Guidance on Informed Consent for In Vitro Diagnostic Device Studies Using Leftover Human Specimens that are Not Individually Identifiable". The gender and age demographics for each category are listed below.

Table 13. Combined Fresh Specimen Study–Age and Gender Distribution		
Gender	Female	Male
Total	384	357
Age		
< 2 years	106	137
3 to 12 years	99	113
13 to 21 years	54	48
> 22 years	125	59

Testing with contiguous fresh nasopharyngeal swabs on the Solana device was performed at four (4) laboratories. Specimens were eluted into the following sample buffers during the clinical studies: Remel M6 (n=236 or 31.8%), UTM (n=226 or 30.5%), Tris EDTA (n=175 or 23.6%), and Molecular Grade Water (n=104 or 14.0%). An aliquot from each buffer was frozen and shipped on dry ice to the central laboratory for testing with comparator reference assays.

Clinical performance was based on comparison of the Solana Bordetella Complete Assay results to those obtained by a composite reference method that included two (2) manufacturer validated, IS481-targeted real-time PCR assays and bi-directional sequencing. The PCR assay protocols included 37 amplification cycles.

a. Prospective clinical study

Seven hundred forty-one (741) fresh nasopharyngeal swab specimens, obtained from female and male patients suspected of having respiratory tract infection attributable to *Bordetella pertussis* or *Bordetella parapertussis,* were prospectively collected and transported to each laboratory for testing with the Solana Bordetella Complete Assay. Four (4) specimens (0.5%) were invalid (in both the initial and repeat test) and have been removed from further analysis. The tables below present the data for the remaining seven hundred thirty-seven (737) specimens.

Table 14. Prospective Study: Combined Site Fresh Specimens - Composite Reference Method versus Solana Bordetella Complete Assay for *B. pertussis*			
	Composite Reference Method		
Solana Bordetella Complete Assay	Positive	Negative	Total
Positive	11	3	14
Negative	0	723	723
Total	11	726	737
			95% CI
Positive Percent Agreement	11/11	100%	74.1% to 100%
Negative Percent Agreement	723/726	99.6%	98.8% to 99.9%

Table 15. Prospective Study: Combined Site Fresh Specimens - Composite Reference Method versus Solana Bordetella Complete Assay for *B. parapertussis*			
	Composite Reference Method		
Solana Bordetella Complete Assay	Positive	Negative	Total
Positive	10	0	10
Negative	0	727	727
Total	10	727	737
			95% CI
Positive Percent Agreement	10/10	100%	72.2% to 100%
Negative Percent Agreement	727/727	100%	99.5% to 100%

b. Retrospective Clinical Study, Archived Specimens

Two hundred thirty-three (233) selected frozen archival specimens were collected at four (4) sites. The frozen archival nasopharyngeal swab specimens were obtained from female and male patients previously tested for the presence of *Bordetella pertussis* or *Bordetella parapertussis*. The specimens were tested with the Solana Bordetella Complete Assay and the Composite Reference Method. Archival specimens were eluted into the following sample buffers: Amies (eSwab) (n=17 or 7.3%), UTM (n=102 or 43.8%), Remel M6 (n=48 or 20.6%), 0.85% Saline (n=10 or 4.3%) and Molecular Grade Water (n=56 or 24.0%). An aliquot from each specimen was re-frozen and shipped on dry ice to the central laboratory for testing with comparator assays. The testing with the comparator assays was performed concurrently and in a blinded fashion. The tables below present the data for the specimens.

Table 16. Combined Site Archival Specimens - Composite Reference Method versus Solana Bordetella Complete Assay for *B. pertussis*			
	Composite Reference Method		
Solana Bordetella Complete Assay	Positive	Negative	Total
Positive	155	6	161
Negative	3	69	72
Total	158	75	233
			95% CI
Positive Percent Agreement	155/158	98.1%	94.6% to 99.4%
Negative Percent Agreement	69/75	92.0%	83.6% to 96.3%

Table 17. Combined Site Archival Specimens - Composite Reference Method versus Solana Bordetella Complete Assay for *B. parapertussis*			
	Composite Reference Method		
Solana Bordetella Complete Assay	Positive	Negative	Total
Positive	12	3	15
Negative	0	218	218
Total	12	221	233
			95% CI
Positive Percent Agreement	12/12	100%	75.7% to 100%
Negative Percent Agreement	218/221	98.6%	96.1% to 99.5%

An additional study was performed to demonstrate the sensitivity of Solana Bordetella Complete Assay using three (3) levels of *Bordetella parapertussis* (BPP) (two (2) strains) in negative specimen matrix at two (2) testing locations. Each individual contrived specimen was prepared using a unique negative NP specimen matrix.

Table 18. Contrived *Bordetella parapertussis* Panel Result Summary			
Panel Members	**# of Samples**	**BPP Concentrations**	**Results**
Negative	20	0	Negative: 100% (20/20)
BPP Low Positive	 10 10	**2.5X LOD** BPP A747 (1.2 x10^4 CFU/mL) BPP E838 (1.4 x10^4 CFU/mL)	 100% (10/10) 100% (10/10)
BPP Moderate Positive	 6 6	**10X LOD** BPP A747 (4.6 x10^4 CFU/mL) BPP E838 (5.5 x10^4 CFU/mL)	 100% (6/6) 100% (6/6)
BPP High Positive	 4 4	**100X LOD** BPP A747 (4.6 x10^5 CFU/mL) BPP E838 (5.5 x10^5 CFU/mL)	 100% (4/4) 100% (4/4)
Negative Control	6	**N/A**	0% (0/6)
BPP Positive Control	6	**N/A**	100% (6/6)

4. Clinical cut-off:

   Not applicable

5. Expected values/Reference range:

   The prevalence of *Bordetella pertussis* and *Bordetella parapertussis* detected with the Solana Bordetella Complete Assay has been calculated for the combined sites based on the age of the patient. Four (4) specimens (0.5%) were invalid (in both the initial and repeat test) and have been removed from the Expected Values table. The table below presents the data for the remaining seven hundred thirty-seven (737) specimens.

Table 19. Combined Fresh Specimen Study Expected Values (N=737)					
		Bordetella pertussis		***Bordetella parapertussis***	
Age	Total #	Total Positive	Prevalence	Total Positive	Prevalence
≤2 years	241*	3	1.2%	4	1.7%
3 to 12 years	210**	4	1.9%	6	2.9%
13 to 21 years	102	6	5.9%	0	0.0%
≥22 years	184	1	0.5%	0	0.0%

* Two (2) specimens were invalid
** Two (2) specimens were invalid

N. Instrument Name:

Solana

O. System Descriptions:

1. Modes of Operation:

   Does the applicant's device contain the ability to transmit data to a computer, webserver, or mobile device?

   Yes ____X____ or No _____

   Does the applicant's device transmit data to a computer, webserver, or mobile device using wireless transmission?

   Yes _____ or No ____X___

2. Software:

   FDA has reviewed the applicant's Hazard Analysis and software development processes for this line of product types: Yes _____X_____ or No _____

3. Specimen Identification:

   Nasopharyngeal swab (NPS) specimens collected from the human nasopharynx region.

4. Specimen Sampling and Handling:

   N/A

5. Calibration:

   N/A

6. Quality Control:

   See Section M.1.c. (*Traceability, Stability, Expected values (controls, calibrators, or methods)*) for further details.

P. Other Supportive Instrument Performance Characteristics Data Not Covered in the "Performance Characteristics" Section above:

N/A

Q. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

R. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.