EVALUATION OF AUTOMATIC CLASS III DESIGNATION FOR
BD MAX Vaginal Panel

DECISION SUMMARY

A. DEN Number:

DEN160001

B. Purpose for Submission:

De Novo request for evaluation of automatic class III designation for the BD MAX Vaginal Panel

C. Measurands:

The assay detects and identifies nucleic acids of the following organisms:

- Bacterial vaginosis (BV) markers (Results for individual organisms are not reported. Qualitative BV results are based on detection and quantitation of targeted organisms)
    - *Lactobacillus* spp (*L. crispatus* and *L. jensenii*)
    - *Gardnerella vaginalis*
    - *Atopobium vaginae*
    - Bacterial Vaginosis Associated Bacteria-2 (BVAB-2)
    - *Megasphaera*-1
- *Candida* spp. (Reported as Cgroup: includes *C. albicans*, *C. tropicalis*, *C. parapsilosis*, *C. dubliniensis*)
- *Candida glabrata*
- *Candida krusei*
- *Trichomonas vaginalis*

D. Type of Test:

The BD MAX Vaginal Panel, performed on the BD MAX System, is a nucleic acid-based test for the detection of the above listed bacteria, yeast and parasites in vaginal specimens obtained from symptomatic patients.

E. Applicant:

GeneOhm Sciences Canada, Inc. (BD Diagnostics)

F. Proprietary and Established Names:

BD MAX™ Vaginal Panel

BD MAX™ (Instrument)

G. Regulatory Information:

1. Regulation section:

   21 CFR 866.3975. Device that detects nucleic acid sequences from microorganisms associated with vaginitis and bacterial vaginosis.

2. Classification:

   Class II (Special Controls)

3. Product code(s):
   PQA
   OUY
   OOI
   NSU

4. Panel:

   83 - Microbiology

H. Indications for Use:

1. Indications for Use:

   The BD MAX Vaginal Panel performed on the BD MAX System is an automated qualitative *in vitro* diagnostic test for the direct detection of DNA targets from bacteria associated with bacterial vaginosis (qualitative results reported based on detection and quantitation of targeted organism markers), *Candida* species associated with vulvovaginal candidiasis, and *Trichomonas vaginalis* from vaginal swabs in patients who are symptomatic for vaginitis/vaginosis. The test utilizes real-time polymerase chain reaction (PCR) for the amplification of specific DNA targets and utilizes fluorogenic target-specific hybridization probes to detect and differentiate DNA from:

   - Bacterial vaginosis markers (Individual markers not reported)
     - *Lactobacillus* spp. (*L. crispatus* and *L. jensenii)*
     - *Gardnerella vaginalis*
     - *Atopobium vaginae*
     - Bacterial Vaginosis Associated Bacteria-2 (BVAB-2)
     - *Megasphaera*-1

   - *Candida* spp. (*C. albicans, C. tropicalis, C. parapsilosis, C. dubliniensis*)
   - *Candida glabrata*
   - *Candida krusei*
   - *Trichomonas vaginalis*

The BD MAX Vaginal Panel is intended to aid in the diagnosis of vaginal infections in women with a clinical presentation consistent with bacterial vaginosis, vulvovaginal candidiasis and trichomoniasis.

3. Special conditions for use statement(s):

   For Prescription Use Only

4. Special instrument requirements:

   The BD MAX Vaginal Panel is performed on the BD MAX System.

I. Device Description:

The BD MAX System and the BD MAX Vaginal Panel are comprised of an instrument with associated hardware and accessories, disposable microfluidic cartridges, master mixes, unitized reagent strips, and extraction reagents. The instrument automates sample preparation including target lysis, DNA extraction and concentration, reagent rehydration, target nucleic acid amplification and detection using real-time PCR. The assay includes a Sample Processing Control (SPC) that is present in the Extraction Tube. The SPC monitors DNA extraction steps, thermal cycling steps, reagent integrity and the presence of inhibitory substances. The BD MAX System software automatically interprets test results. For the BD MAX Vaginal Panel, a test result may be called as POS, NEG or UNR (Unresolved) based on the amplification status of the targets and of the Sample Processing Control. IND (Indeterminate) or INC (Incomplete) results are due to BD MAX System failure.

J. Standard/Guidance Document Referenced:

- CLSI EP 17-A2, Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures, 2012
- CLSI EP5-A2, Evaluation of Precision Performance of Quantitative Measurement Methods, Approved Guideline, 2004
- CLSI EP12-A2, User Protocol for Evaluation of Qualitative Test Performance, 2008

K. Test Principle:

The BD MAX Vaginal Panel is designed for use with the BD MAX™ UVE Specimen Collection kit. Samples are transported to the testing laboratory in BD MAX UVE Sample Buffer Tubes (SBT). The Sample Buffer Tubes, are vortexed to release cells from the swab into the buffer. The Sample Buffer Tubes, Unitized Reagent Strips and PCR Cartridges are loaded on the BD MAX System. No further operator intervention is necessary and the following automated procedures occur.

A combination of lytic and extraction reagents are used to perform cell lysis and DNA extraction. Nucleic acids released from the target organisms are captured on magnetic affinity beads. The beads, together with the bound nucleic acids, are washed and the nucleic acids are eluted by a combination of heat and pH. Eluted DNA is neutralized and transferred

to the Master Mix Tubes to rehydrate the PCR reagents. After reconstitution, the BD MAX System dispenses a fixed volume of PCR-ready solution containing extracted nucleic acids into the PCR Cartridge. Microvalves in the cartridge are sealed by the system prior to initiating PCR in order to contain the amplification mixture and thus prevent evaporation and contamination.

The amplified DNA targets are detected using hydrolysis probes, labeled at one end with a fluorescent reporter dye (fluorophore), and at the other end, with a quencher moiety. Probes labeled with different fluorophores are used to detect the target analytes in different optical channels of the BD MAX System. When the probes are in their native state, the fluorescence of the fluorophore is quenched due to its proximity to the quencher. However, in the presence of target DNA, the probes hybridize to their complementary sequences and are hydrolyzed by the 5'-3' exonuclease activity of the DNA polymerase as it synthesizes the nascent strand along the DNA template. As a result, the fluorophores are separated from the quencher molecules and fluorescence is emitted. The amount of fluorescence detected in the optical channels used for the BD MAX Vaginal Panel is directly proportional to the quantity of the corresponding probe that is hydrolyzed. The BD MAX System monitors these signals at each cycle of the PCR and interprets the data at the end of the reaction to provide qualitative test results for each vaginitis analyte as well as qualitative results for bacterial vaginosis based on detection and quantitation of targeted bacterial vaginosis markers.

L. Performance Characteristics:

1. Analytical Performance:

a. *Precision/Reproducibility Studies*

Reproducibility/Precision Study Panel Member Composition

For the precision and reproducibility studies, panel members were prepared with targeted organisms (or plasmid DNA for *Megasphaera*-1 and BVAB-2) spiked into simulated vaginal matrix. Table 1 describes organisms that were used to prepare panel members.

Table 1: Organisms for Reproducibility/Precision Study Panel Members

MasterMix	Assay Target	Organism
Vaginosis	BV Markers	*Lactobacillus crispatus*
		Lactobacillus jensenii
		Gardnerella vaginalis
		Atopobium vaginae
		Megasphaera type 1
		BVAB-2
Vaginitis	Cgroup	*Candida albicans*
	Ckru	*Candida krusei*
	Cgla	*Candida glabrata*
	TV	*Trichomonas vaginalis*

For Cgroup (*Candida albicans*), *C. glabrata* and *C. krusei* panel members, samples were spiked at high negative, low positive and moderate positive concentrations based on the assay Limit of Detection (LoD).

For BV panel members, sample compositions were designed to represent the flora of BV positive and negative specimens with specific target organism combinations based on results from clinical specimen testing. Because a variety of targeted BV organism combinations can be present in vaginal specimens, multiple panel members for each level were prepared with different targeted organism compositions at varying loads. Each BV negative panel member was spiked with two target organisms. Each BV low positive and moderate positive panel member was prepared with three or more target organisms. Sample compositions were determined based on assay cutoffs for positive and negative BV results.

The design for study panel members is described in Table 2.

Table 2: Precision/Reproducibility Study Panel Member Design

Concentration Designation	**Bacterial Vaginosis[1]** **(% of positive results expected at the designated concentration)**	***Candida* spp. and *Trichomonas vaginalis*** **(x LoD)**
Moderate Positive	~100	≥ 2 to ≤ 5
Low Positive	~95	< 2
High BV Negative	~20-80	
BV Negative	< 5	
True Negative	0 (No Target)	No Target

[1]Multiple panel members with different organism compositions used for BV positive and BV negative samples.

Precision Study

Within-laboratory precision was evaluated for the BD MAX Vaginal Panel at one site. Testing of two different panels was performed over 12 days. Two operators performed two runs each per day, for a total of 48 runs per panel. For evaluation of BV, testing included four different BV high negative panel members, six different BV low positive panel members and one BV negative panel member, each spiked with varying compositions of targeted BV organisms. Results from the study are shown in Table 3.

Table 3: Qualitative Precision Study Results Summary- Vaginitis/Vaginosis

Concentration	Percent Agreement with Expected Result [95 % Confidence Interval]				
	Bacterial Vaginosis	***Trichomonas vaginalis***	***Candida albicans***	***Candida glabrata***	***Candida krusei***
True Negative[a,b]	100.0 (288/288) [98.7, 100.0]	100.0 (240/240) [98.4, 100.0]	99.6 (239/240) [97.7, 99.9]	100.0 (240/240) [98.4, 100.0]	100.0 (240/240) [98.4, 100.0]
Low Positive[c]	100.0 (287/287) [98.7, 100.0]	100.0 (48/48) [92.6, 100.0]	100.0 (48/48) [92.6, 100.0]	100.0 (48/48) [92.6, 100.0]	100.0 (48/48) [92.6, 100.0]
Moderate Positive[d]	100.0 (192/192) [98.0, 100.0]	100.0 (48/48) [92.6, 100.0]	100.0 (48/48) [92.6, 100.0]		
High BV Negative[d]	37.5 (72/192) [31.0, 44.5]				
BV Negative[a]	100.0 (48/48) [92.6, 100.0]				

[a]The expected assay results were deemed to be negative.
[b]Samples containing specific targets used for analyses of one Master Mix (vaginitis or vaginosis) were used as a TN for the other Master Mix.
[c] Performance includes combined results from replicates of six panel members containing different organism compositions.
[d]Performance includes combined results from replicates of four panel members containing different organism compositions.

Reproducibility Study

A multi-site reproducibility study was performed using the same sample categories as defined above for the precision study with the exception that the high negative category was not evaluated for BV. For BV panel members, the study included two different sample compositions each for low positive and moderate positive samples. Testing was performed using multiple instruments at three different testing sites over eight days. At each site, two operators performed two runs per day on alternating days, for a total of 48 runs tested. The overall Site-to-Site Reproducibility percent agreement for panel member results ranged from 98.5 % to 100% for true negatives, 99.0% to 100% for low positive samples, and 99.5% to 100% for moderate positive samples. Table 4 includes overall qualitative reproducibility results and Table 5 includes qualitative results stratified by site. In addition, Second Derivative Peak Abscissa (SDPA), an internal criterion used to determine a final assay result, was selected as a means of assessing quantitative assay reproducibility. Mean SDPA values with variance components (SD and % CV) are shown in Table 6.

Table 4: Qualitative Reproducibility Study Results Summary

Concentration	Percent Agreement with Expected Result [95 % Confidence Interval)]				
	Bacterial vaginosis	***Trichomonas vaginalis***	***Candida albicans***	***Candida glabrata***	***Candida krusei***
True Negative[a]	100.0 (576/576) [99.3, 100.0]	100.0 (480/480) [99.2, 100.0]	98.5 (473/480) [97.0, 99.3]	100.0 (480/480) [99.2, 100.0]	99.6 (478/480) [98.5, 99.9]
Low Positive[b]	99.0 (190/192) [96.3, 99.7]	100.0 (96/96) [96.2, 100.0]	100.0 (96/96) [96.2,100.0]	100.0 (96/96) [96.2, 100.0]	100.0 (96/96) [96.2, 100.0]
Moderate Positive[b]	99.5 (191/192) [97.1, 99.9]	100.0 (96/96) [96.2, 100.0]	100.0 (96/96) [96.2, 100.0]		
BV Negative[a]	100.0 (96/96) [96.2, 100.0]				

[a]The expected assay results were deemed to be negative.
[b]Performance includes combined results from replicates of two panel members containing different organism compositions.

Table 5: Qualitative Site to Site Results

Target	Concentration/ Sample	Percent Agreement with Expected Results			
		Site 1	**Site 2**	**Site 3**	**All**
Bacterial Vaginosis	True Negative	100 192/192	100 192/192	100 192/192	100 576/576
	BV Negative	100 32/32	100 32/32	100 32/32	100 96/96
	Low BV Positive[a]	100 64/64	96.9 62/64	100 64/64	99.0 190/192
	Moderate Positive[a]	100 64/64	98.4 63/64	100 64/64	99.5 191/192
Trichomonas vaginalis	True Negative	100 160/160	100 160/160	100 160/160	100 480/480
	Low Positive	100 32/32	100 32/32	100 32/32	100 96/96
	Moderate Positive	100 32/32	100 32/32	100 32/32	100 96/96
Candida albicans	True Negative	98.8 158/160	98.8 158/160	98.1 157/160	98.5 473/480
	Low Positive	100 32/32	100 32/32	100 32/32	100 96/96
	Moderate Positive	100 32/32	100 32/32	100 32/32	100 96/96
Candida glabrata	True Negative	100 160/160	100 160/160	100 160/160	100 480/480
	Low Positive	100 32/32	100 32/32	100 32/32	100 96/96
Candida krusei	True Negative	99.4 159/160	99.4 159/160	100 160/160	99.6 478/480
	Low Positive	100 32/32	100 32/32	100 32/32	100 96/96

[a] Performance includes combined results from replicates of two panel members containing different organism compositions

Table 6: Quantitative Site to Site Results

Concentration		SDPA		Within Run		Between Run		Between Day		Between Operator		Between Site		Total	
		N	Mean	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Candida albicans	Low Positive	96	29.9	0.71	2.4	0.00	0.0	0.00	0.0	0.04	0.1	0.08	0.3	0.71	2.4
	Moderate Positive	96	28.5	0.48	1.7	0.00	0.0	0.10	0.3	0.00	0.0	0.19	0.7	0.52	1.8
Candida glabrata	Low Positive	96	29.6	0.32	1.1	0.00	0.0	0.00	0.0	0.04	0.1	0.08	0.3	0.33	1.1
Candida krusei	Low Positive	96	30.6	0.25	0.8	0.16	0.5	0.00	0.0	0.11	0.4	0.06	0.2	0.32	1.1
Trichomonas vaginalis	Low Positive	96	32.9	0.33	1.0	0.11	0.3	0.00	0.0	0.05	0.2	0.00	0.0	0.36	1.1
	Moderate Positive	96	31.7	0.31	1.0	0.10	0.3	0.00	0.0	0.01	0.0	0.00	0.0	0.33	1.0

Additional evaluation of lot-to-lot reproducibility of the BD MAX Vaginal Panel was performed at one site with three assay lots over eight days. At the testing site, two operators performed two runs on alternate days, for a total of 48 runs. Lot-to lot reproducibility results are reported below in Tables 7, 8 and 9.

Table 7: Qualitative Reproducibility Study Results Summary - Lot to Lot

Category	Percent Agreement with Expected Result [95 % Confidence Interval]				
	Bacterial vaginosis	*Trichomonas vaginalis*	*Candida albicans*	*Candida glabrata*	*Candida krusei*
True Negative[a]	100.0 (576/576) [99.3, 100.0]	100.0 (480/480) [99.2, 100.0]	99.2 (476/480) [97.9, 99.7]	100.0 (480/480) [99.2, 100.0]	99.8 (479/480) [98.8, 100.0]
Low Positive	100.0[b] (192/192) [98.0, 100.0]	100.0 (96/96) [96.2, 100.0]	100.0 (96/96) [96.2, 100.0]	100.0 (96/96) [96.2, 100.0]	100.0 (96/96) [96.2, 100.0]
Moderate Positive	100.0[b] (192/192) [98.0, 100.0]	100.0 (96/96) [96.2, 100.0]	100.0 (96/96) [96.2, 100.0]		
BV Negative	100.0 (96/96) [96.2, 100.0]				

[a] The expected assay results were deemed to be negative.

[b] Performance includes combined results from replicates of two panel members containing different organism compositions

Table 8: Qualitative Lot-to-Lot Results

Target	Concentration/ Sample	Percent Agreement with Expected Results			
		Lot 1	Lot 2	Lot 3	All
Bacterial Vaginosis	True Negative	100 192/192	100 192/192	100 192/192	100 576/576
	BV Negative	100 32/32	100 32/32	100 32/32	100 96/96
	Low BV Positive[a]	100 64/64	100 64/64	100 64/64	100 192/192
	Moderate Positive[a]	100 64/64	100 64/64	100 64/64	100 192/192
Trichomonas vaginalis	True Negative	100 160/160	100 160/160	100 160/160	100 480/480
	Low Positive	100 32/32	100 32/32	100 32/32	100 96/96
	Moderate Positive	100 32/32	100 32/32	100 32/32	100 96/96
Candida albicans	True Negative	98.8 158/160	99.4 159/160	99.4 159/160	99.2 476/480
	Low Positive	100 32/32	100 32/32	100 32/32	100 96/96
	Moderate Positive	100 32/32	100 32/32	100 32/32	100 96/96
Candida glabrata	True Negative	100 160/160	100 160/160	100 160/160	100 480/480
	Low Positive	100 32/32	100 32/32	100 32/32	100 96/96
Candida krusei	True Negative	99.4 159/160	100 160/160	100 160/160	99.8 479/480
	Low Positive	100 32/32	100 32/32	100 32/32	100 96/96

[a] Performance includes combined results from replicates of two panel members containing different organism compositions

Table 9: Quantitative Lot-to-Lot Results

Target	Concentration	SDPA		Within Run		Between Run		Between Day		Between Operator		Between Lot		Total	
		N	Mean	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Candida albicans	Low Positive	96	30.2	0.53	1.7	0.00	0.0	0.00	0.0	0.00	0.0	1.11	3.7	1.23	4.1
	Moderate Positive	96	28.9	0.43	1.5	0.00	0.0	0.00	0.0	0.00	0.0	1.04	3.6	1.13	3.9
Candida glabrata	Low Positive	96	29.6	0.32	1.1	0.00	0.0	0.06	0.2	0.00	0.0	0.21	0.7	0.39	1.3
Candida krusei	Low Positive	96	30.5	0.18	0.6	0.15	0.5	0.05	0.2	0.00	0.0	0.24	0.8	0.33	1.1
Trichomonas vaginalis	LowPositive	96	32.7	0.37	1.1	0.00	0.0	0.00	0.0	0.06	0.2	0.32	1.0	0.50	1.5
	Moderate Positive	96	31.6	0.28	0.9	0.05	0.2	0.00	0.0	0.00	0.0	0.18	0.6	0.34	1.1

b. Linearity/Assay Reportable Range:

Not Applicable

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

Internal Control

Each Extraction Tube contains a Sample Processing Control (SPC) comprised of plasmids containing a synthetic target DNA sequence. The SPC monitors the efficiency of DNA capture, washing and elution during the sample processing steps, as well as the efficiency of DNA amplification and detection during PCR analysis. If the SPC result fails to meet the acceptance criteria, the result of the specimen will be reported as Unresolved for the Master Mix reaction. Each Master Mix contains its own Sample Processing Control; thus Unresolved results are determined independently for each Master Mix. An Unresolved result is indicative of specimen-associated inhibition or reagent failure. The operator is directed to repeat any specimen reported as Unresolved.

External Controls

External quality control materials are not provided with the BD MAX Vaginal Panel and the BD MAX System software does not require inclusion of external controls for the purpose of sample test results interpretation. However, the instructions for use indicate that one external positive control and one external negative control should be run at least daily until adequate process validation is achieved on the BD MAX System in each laboratory setting. After such validation has been completed, laboratories are directed to perform external quality control testing according to guidelines or requirements of local, state and federal accrediting organizations.

The following are recommended in the package insert for external control testing with the BD MAX Vaginal Panel.

External Negative Controls

- Suspension of commercially available *Lactobacillus iners* strain
- Previously characterized negative clinical specimen

External Positive Controls

- Suspension of available organisms listed in Table 10.
- Previously characterized positive clinical specimen

Table 10: Recommended Organisms for External Controls

	Positive controls	**Negative controls**
Vaginitis	*Trichomonas vaginalis* ATCC 30001	*Lactobacillus iners* ATCC 55195
	Candida albicans ATCC 10231	
	Candida glabrata ATCC 2001	
	Candida krusei ATCC 6258	
Vaginosis	BV Positive External Control[a]	

[a] Mixture of *Gardnerella vaginalis* and *Atopobium vaginae*

In the prospective clinical study, one external positive and one external negative control were evaluated each day of testing. A rotation scheme consisting of five different positive controls was used to cover all assay targets at each testing site. The external control success rate was 97.6% (445/456) with 6/456 (1.3%) controls generating unexpected results and 5/546 (1.1%) controls generating non-reportable results. Results are presented by analyte in Table 11.

Table 11: External Control Results –Clinical Study

Control	External Control Pass Rate
BV Positive	100% (53/53)
C albicans	100% (22/23)
C. glabrata	96.2% (51/53)
C. krusei	98.1% (52/53)
T. vaginalis	93.5% (43/46)
BV Negative	99.2% (119/120)
Negative (No target)	97.2% (105/108)

Specimen Stability

Evaluation of specimen stability was performed to demonstrate that target DNA is stable in vaginal specimens prior to testing with the BD MAX Vaginal Panel. The study combined different storage conditions in a nested design.

The study was conducted using five different combinations of reagent lots (Master Mix, Extraction Tubes, Reagent strips and SBTs). For vaginitis analytes, 25 different strains of targeted vaginitis organisms (*Candida* spp. or *T. vaginalis*) were used to prepare low positive samples at <2x LoD. Each vaginitis sample was prepared in a unique natural negative vaginal matrix. For BV samples, panel members included 13 low-positive BV organism pools and two negative organism pools. A minimum of 24 sample replicates were evaluated for each panel member and storage condition.

To demonstrate stability at each storage condition, a minimum of 95% agreement with the expected result was required. Study results met the study acceptance criteria and therefore substantiate the claimed stability of amplifiable DNA in vaginal specimens containing low positive concentrations of vaginitis DNA targets, low positive compositions of BV analytes, as well as negative specimens for the following claimed storage conditions:

- Dry swabs: Storage of dry swab for up to two hours at 2-30°C after collection and before transfer to BD MAX UVE Sample Buffer Tube (SBT).
- Specimen in capped SBT (transport and pre-testing storage): Storage of specimen in SBT up to eight days at 2-30°C or for a maximum of 14 days at 2-8°C.
- SBT post vortex: Storage of vortexed specimen up to four hours at 2-30°C. After this time period the vortexing step must be repeated before testing.
- SBT post testing: Storage of SBT for up to five hours when stored at 2-30°C after completion of the run.

The study data provided support the specimen handling recommendations described in the BD MAX Vaginal Panel package insert.

d. Limit of Detection:

A study was conducted to determine the LoD for a representative strain of each targeted organism detected by the BD MAX Vaginal Panel. Serial dilutions of targeted strains were inoculated into simulated vaginal matrix in BD MAX UVE Sample Buffer. A total of 24 replicates were evaluated for each dilution to determine the assay LoD for each target (i.e., organism concentration at which >95% of replicates are detected).

To further confirm the LoD for vaginitis analytes, a total of 24 sample replicates were each tested at the LoD in both simulated and natural vaginal matrix. Because natural vaginal matrix contains BV analytes as part of the normal vaginal flora, confirmation of the LoD for BV analytes was performed only in simulated matrix.

Table 12 lists the confirmed LoD for organisms strains evaluated in the study.

Table 12: Limit of Detection Results

Assay Target	Organism	Strain ATCC#	LoD	
			Concentration	Units
Vaginitis	*Candida albicans*	18804	17787	CFU/mL
	Candida glabrata	2001	202	
	Candida krusei	6258	1035	
	Candida dubliniensis	MYA-646	4002	
	Candida tropicalis	750	313	
	Candida parapsilosis	22019	30660	
	Trichomonas vaginalis	30001	22	Cells/mL
Bacterial Vaginosis Markers	*Atopobium vaginae*	BAA-55	127	CFU/mL
	Gardnerella vaginalis	14018	962	
	Lactobacillus crispatus	33820	55	
	Lactobacillus jensenii	25258	510	
	Megasphaera-1	NA[a]	2265	Copies/mL
	BVAB 2		464	

[a] LoD determined with plasmid DNA.

e. Analytical Inclusivity:

An analytical inclusivity study was conducted to evaluate the BD MAX Vaginal Panel for detection of a variety of organism strains, taking into account phylogenetic diversity, geographic origin and temporal diversity. The microbial strains evaluated were from public collections or well-characterized clinical isolates. Testing included five strains each for targeted *Candida* species (*C. albicans*, *C. dubliniensis, C. parapsilosis, C. tropicalis, C. glabrata,* and *C. krusei)* and nine strains of *Trichomonas vaginalis* (including one metronidazole resistant strain). In addition, ten strains of *Gardnerella vaginalis* and five strains each of *Atopobium vaginae, Lactobacillus crispatus,* and *Lactobacillus jensenii* were evaluated. Samples were inoculated at < 3x LoD of the corresponding reference strain evaluated in the LoD study. The BD MAX Vaginal Panel correctly identified 60 of the strains tested upon initial testing. Results from four strains

of *Gardnerella vaginalis* and one strain of *Lactobacillus crispatus* did not meet acceptance criteria and were further evaluated to determine the minimum concentration sufficient for detection. Upon repeat, one *G. vaginalis* strain was detected at < 3x LoD and three strains were detected at ~9x LoD. The *L. crispatus* strain was detected at ~5x LoD.

f. Mixed Infection/Competitive Interference Study:

A mixed infection/competitive interference study was designed to evaluate the BD MAX Vaginal Panel for detection of targeted analytes at low positive concentrations in the presence of other targets at high concentrations. The following organisms and concentrations were evaluated:

- Assay targets at high concentrations: *L. crispatus* (8.7×10^4 CFU/mL), *G. vaginalis* (5.0×10^6 CFU/mL), *A. vaginae* (8.0×10^5 CFU/mL), *Megasphaera*-1 (2.4×10^7 cp/mL) and *T. vaginalis* (3.3×10^5 to 1.0×10^6 cells/mL), *C. albicans* (1×10^6 CFU/mL), *C. glabrata* (1×10^6 CFU/mL)

- High concentrations of non-targeted vaginal flora organisms: *Dialister microaerophilus, Prevotella melaninogenica, Streptococcus mitis, Bifidobacterium breve and Mobiluncus curtisii* (each at 1.0×10^6 CFU/mL).

- Low positive loads of vaginitis targets were evaluated at < 2x LoD. Low positive BV samples were prepared with BV organism compositions sufficient to obtain 95% positive BV results.

Study samples were each prepared in simulated vaginal matrix. The following series of organism pools simulating mixed infections were evaluated:

- High positive BV organisms with low positive loads of:
  - *T. vaginalis* and *C. albicans*
  - *C. krusei* and *C. glabrata*

- High positive *C. albicans* with low positive loads of*:*
  - *C. krusei* and *C. glabrata*
  - *T. vaginalis*
  - BV

- High positive *C. glabrata* with low positive loads of:
  - *T. vaginalis*
  - BV

- High positive *C. krusei* with low positive BV

- High positive *T. vaginalis* with low positive loads of:
  - BV
  - *C. albicans*

- *C. krusei*
- *C. glabrata*

- High positive loads of non-targeted vaginal flora organisms with low positive loads of:
  - *T. vaginalis* and BV
  - *C. albicans* and BV
  - *C. glabrata* and BV
  - *C. krusei* and BV

The study demonstrated that samples containing *T. vaginalis* at low concentrations as well as low positive BV samples were successfully detected by the BD MAX Vaginal Panel when tested in combination with high concentrations of other assay targets or high concentrations of other selected organisms of the vaginal flora.

Competitive inhibition was observed for samples containing low positive *Candida* spp. when present in samples containing high concentrations of *T. vaginalis* or BV analytes.

- For low positive samples containing *Candida albicans,* 92% of positive results were obtained in presence of BV analytes at high loads

- For low positive samples containing *Candida albicans*, *C. krusei* or *C. glabrata*, BD MAX Vaginal Panel generated 42%, 61% and 33% of expected results respectively in presence of *Trichomonas vaginalis* at a load of 3.3×10^5 cells/mL.

g. Analytical Specificity/Cross-reactivity:

The BD MAX Vaginal Panel was evaluated for potential cross-reactivity with samples containing phylogenetically related species and other organisms likely to be present in vaginal specimens. Bacteria, yeasts, parasites and viruses were tested in the BD MAX UVE Sample Buffer Tube at $\geq 10^6$ bacteria, cells or genome equivalents/mL, or $\geq 10^5$ PFU/mL or $TCID_{50}$/mL or equivalent amount of RNA/DNA per PCR reaction. In total, 118 organisms were evaluated and those organisms are listed in Table 14 below.

For organisms that generated unexpected positive results, additional testing was performed to evaluate the organism load that no longer cross-reacts with the BD MAX Vaginal Panel. Table 13 describes organisms that demonstrated cross-reactivity and the concentrations at which detection was observed. A limitation is included in the package insert describing all cross-reactive organisms.

Table 13: Cross Reacting Organisms

Cross Reacting Organism	BD MAX Vaginal Panel Target	Additional Testing
Candida guillermondii[1]	Cgroup	Not detected at <6.0 x 10^3 CFU/mL
Candida haemulonii[1]	Cgroup	Detected at all concentrations evaluated
Candida orthopsilosis[12]	Cgroup	Detected at all concentrations evaluated
Pichia fermentans	*C krusei*	Not detected at <6.0 x 10^3 CFU/mL
Trichomonas tenax	*Trichomonas vaginalis*	Detected at all concentrations evaluated
Atopobium rimae	*Atopobium vaginae* (BV)	Not detected at <4.4 x 10^4 CFU/mL
Olsenella uli	*Atopobium vaginae* (BV)	Not detected at <6.6x 10^4 CFU/mL
Lactobacillus delbrueckii subsp. lactis	*L. crispatus/jensenii* (BV)	Not detected at <3.9 x 10^3 CFU/mL
Lactobacillus acidophilus	*L. crispatus/jensenii* (BV)	Detected at all concentrations evaluated

[1]*Candida guilliermondii*, *Candida haemulonii* and *Candida orthopsilosis* have each been reported as occasional causes of vulvovaginal candidiasis

[2]*Candida metapsilosis* and *Candida orthopsilosis* are both subgroups of *C. parapsilosis*, which is a target of the assay. These two targets were predicted to cross-react based on *in silico* analysis.

Table 14: Organisms Evaluated For Specificity/Cross-Reactivity

Organisms tested (Cross-reacting Organisms Bolded)					
BACTERIA		**BACTERIA**		**BACTERIA**	
Genus	**Species**	**Genus**	**Species**	**Genus**	**Species**
-	*BVAB-1*	*Kocuria*	*rhizophila*	*Sneathia*	*amnii*
	BVAB-3	***Lactobacillus***	*acetotolerans*		*sanguinegens*
Acinetobacter	*baumannii*		***acidophilus***	*Streptococcus*	*agalactiae*
	calcoaceticus		*amylophilus*		*mitis*
Actinomyces	*israelii*		*animalis*		*mutans*
	pyogenes		*coleohomonis*		*salivarius*
Aerococcus	*viridans*		***delbrueckii subsp. lactis***		*thermophilus*
Alcaligenes	*faecalis (subsp. faecalis)*		*fornicalis*	*Treponema*	*pallidum*
Anaerococcus	*tetradius*		*gasseri*	*Veillonella*	*atypica*
Atopobium	*minutum*		*iners*		*parvula*
	parvulum		*johnsonii*	*Vibrio*	*parahaemolyticus*
	rimae		*pontis*	*Yersinia*	*enterocolitica*
Bacillus	*subtilis*		*sharpeae*	***YEASTS***	
Bacteroides	*caccae*		*vaginalis*	*Candida*	*catenulata*
	fragilis	*Legionella*	*pneumophila subsp. pneumophila*		*famata*
	stercoris	*Listeria*	*monocytogenes*		***guilliermondii***
Bifidobacterium	*adolescentis*	*Megaspheara*-2	*Megasphaera* Type-2		***haemulonii***
	breve	*Mobiluncus*	*curtisii*		*inconspicua*
	coryneforme		*mulieris*		*intermedia*
	longum	*Moraxella*	*catarrhalis*		*kefyr*
	minimum	*Morganella*	*morganii subsp. morganii*		*lusitaniae*
Brevibacterium	*linens*	*Mycobacterium*	*smegmatis*		*norvegica*
Burkholderia	*cepacia*	*Mycoplasma*	*genitalium*		***orthopsilosis***
Campylobacter	*jejuni*		*hominis*		*rugosa*

Chlamydia	*trachomatis*	*Neisseria*	*gonorrhoeae*		*utilis*
Citrobacter	*freundii*	***Olsenella***	***uli***	*Issatchenkia*	*occidentalis*[2]
Clostridium	*perfringens*	*Pantoea*	*agglomerans*	*Kodamaea*	*ohmeri*[1]
Corynebacterium	*genitalium*	*Peptostreptococcus*	*anaerobius*	*Pichia*	***fermentans***
Dialister	*microaerophilus*	*Plesiomonas*	*shigelloides*		*norvegensis*[3]
Eikenella	*corrodens*	*Porphyromonas*	*asaccharolytica*	*Saccharomyces*	*cerevisiae*
Enterobacter	*aerogenes*	*Prevotella*	*melaninogenica*	**VIRUSES**	
Enterococcus	*faecalis*		*oris*	HBV	Human herpesvirus 2
	faecium	*Propionibacterium*	*acnes*	HIV	HPV
Erysipelothrix	*rhusiopathiae*	*Proteus*	*mirabilis*	HSV type 1	Varicella-zoster virus Ellen
Escherichia	*coli GC10*	*Providencia*	*stuartii*	Hepatitis C Virus	
	coli top 10	*Pseudomonas*	*aeruginosa*	**PARASITES**	
Fusobacterium	*nucleatum subsp. nucleatum*	*Salmonella*	*typhimurium*	*Pentatrichomonas*	*hominis*
Gemella	*haemolysans*	*Serratia*	*marcescens*	***Trichomonas***	***tenax***
Kingella	*denitrificans*	*Shigella*	*flexneri*		
Klebsiella	*pneumoniae*	*Staphylococcus*	*aureus*		

[1]Also reported as *Pichia ohmeri, C. guilliermondii*
[2]Also reported as *C. sorbosa*
[3]Also reported as *C. norvegensis*

The following additional unexpected detections were observed in the study. Repeat testing indicated that these organisms do not cross-react with the BD MAX Vaginal Panel targets.

- A single replicate each containing *Lactobacillus delbrueckii subsp. lactis or Chlamydia trachomatis* initially generated a false positive result for Cgroup. Repeat testing generated 10/10 expected negative results for Cgroup for both of these organisms
- A single replicate each containing *Bifidobacterium breve, E. coli GC10* or *Lactobacillus acetotolerans* initially *generated* a false positive result for the *A. vaginae* signal. Repeat testing generated 10/10 expected negative results for *A. vaginae* for these three organisms

h. Evaluation of Potentially Interfering Substances/Organisms

A study was performed to evaluate potentially interfering biological and chemical substances that may be present in vaginal specimens. Exogenous (e.g., prescription and Over-the-Counter drugs, creams and/or gels) and endogenous (e.g., blood, hormones, mucus) substances were evaluated in samples spiked with the highest concentration expected to be present in vaginal specimens. Each potentially interfering substance was evaluated in both negative and low positive samples. Samples for targeted vaginitis analytes were spiked with low concentrations (<2x LoD*) of Candida albicans, Candida glabrata, Candida krusei* or *Trichomonas vaginalis*. Positive BV samples were spiked with organism compositions designed to generate results near the assay BD MAX Vaginal Panel cutoffs for BV (i.e., C_{95}).

KY Jelly Personal Lubricant and Whole Blood were found to interfere at levels above >12.5 µL/mL (1.25% V/V). Zovirax Acyclovir 5 % Cream and VCF Contraceptive Foam were found to interfere at levels above > 3.1 µL/mL. Preparation H Hemorrhoidal

Cream was found to interfere above > 0.8 µL/mL. Interference with the following substances was observed at all tested levels: Conceptrol Vaginal Contraceptive Gel, Clotrimazole Vaginal Cream, Monistat 3 Cream, Vagisil Cream, Replens Vaginal Moisturizing Gel, Metronidazole, Leukocytes. Table 15 shows results for the potentially interfering substances evaluated in the study. Substances that demonstrated interference may result in unresolved, indeterminate or false negative results. A limitation is included in the package insert listing all substances that demonstrated interference with the BD MAX Vaginal Panel.

Table 15: Exogenous and Endogenous Substances Tested for Interference[a]

	No Interference Observed		Interference Observed	
	Substance		Substance	Level Below Which No Interference Observed µL/mL)
Exogenous	Tioconzole Ointment, 6.5%		VCF Contraceptive Foam	≤ 3.1
	VCF Contraceptive Film		Zovirax, Acyclovir 5% Cream	≤ 3.1
	Summer's Eve Douche		Preparation H Hemorrhoidal Cream	≤ 0.8
	FDS Feminine Deodorant Spray		KY Jelly Personal Lubricant	≤ 12.5
	Progesterone		Conceptrol Vaginal Contraceptive Gel	Interference observed at each level evaluated
	Estradiol		Clotrimazole Vaginal Cream, USP 2%	
			Monistat 3 Cream, Miconazole Nitrate, 4%	
			Vagisil, Benzocaine 20%, Resorcinol 3%	
			Replens Vaginal Moisturizing Gel	
			Metronidazole 0.75% Gel	
Endogenous	Mucus (Bovine Cervical, 5% v/v)		Whole Blood	≤ 12.5 (1.25% v/v)
	Semen (5% v/v)		Leukocytes	Interference observed at each level evaluated

[a] In total, with the BD MAX Vaginal Panel in the presence of potentially interfering substances, 2672 samples were tested for vaginosis targets and 3252 for vaginitis targets. For vaginitis targets, rates of 8.27% IND and 8.52% UNR results were recorded. For BV, rates of 9.92% IND and 1.83% UNR results were recorded.

Additional testing was performed to evaluate potential interference from microorganisms included in probiotic formulations. A total of 14 probiotic Lactobacillus species listed in Table 17 were evaluated at high concentrations (> 6.7 x 10^5 CFU/mL of Sample Buffer) in combination with low positive vaginitis analytes, low positive BV samples as well as negative samples containing no targeted analytes.

Interference was not observed for detection of *Candida albicans, Candida glabrata, Candida krusei,* or *Trichomonas vaginalis* in samples spiked with each of the probiotic organisms. False negative results for BV were observed in the presence of the following probiotic organisms: *Lactobacillus amylovorus, Lactobacillus delbrueckii subsp. bulgaricus, Lactobacillus kefirgranum* and *Lactobacillus helveticus*. The probiotic organisms evaluated are shown in Table 16.

Table 16: Interference Testing: Probiotic Microorganisms

No Interference Observed			Interference Observed
Lactobacillus plantarum	*Lactobacillus casei*		*Lactobacillus delbrueckii subsp. bulgaricus*
Lactobacillus reuteri	*Lactobacillus fermentum*		*Lactobacillus amylovorus*
Lactobacillus rhamnosus	*Lactobacillus paracasei*		*Lactobacillus helveticus*
Lactobacillus salivarius subsp. salivarius	*Bifidobacterium animalis subsp lactis*		*Lactobacillus kefirgranum*
Lactobacillus brevis	*Bifidobacterium longum subsp infantis*		

i. Matrix Equivalence Study

Because the BV analytes detected by the BD MAX Vaginal Panel are present in normal vaginal flora, it was necessary to use a simulated vaginal matrix for preparation of samples for some analytical studies. Equivalence between the simulated matrix and natural vaginal matrices was assessed using data generated in the LoD confirmation study for *Candida* spp. and *T. vaginalis*. In this study, LoD values initially determined using samples prepared in simulated vaginal matrix were confirmed in the presence of both simulated and real vaginal matrices. A minimum of 24 sample replicates for each organism evaluated (six different *Candida* species and one *T. vaginalis strain*) were tested in both simulated and real vaginal matrices, at the LoD (95% concentration) previously determined in in simulated matrix. All targets evaluated generated 100% positive results in both matrices except for *C. krusei* which generated only 79.2% positive results for samples prepared in natural vaginal matrix.

To further evaluate differences for detection of *C. krusei* in natural and simulated matrices, higher concentrations were tested in natural matrix, resulting in 91.6% of positive results obtained at 1.99x and 2.5x LoD. These study results together with *C. krusei* results from contrived clinical specimens prepared in natural vaginal matrices (i.e., 50/50 specimens with *C. krusei* at 1.99x LoD generated positive results) demonstrated that the assay LoD for *C. krusei* in natural matrix was ~1.99 x the LoD for this target in simulated matrix.

In summary, the matrix equivalency study results substantiated equivalence between the simulated vaginal matrix and natural vaginal matrices for all analytes evaluated with the exception of *C. krusei*, which demonstrates a higher LoD in natural matrix. This difference was deemed to be acceptable because analytical study samples for *C. krusei* were prepared with concentrations based on the applicable LoD for the matrix used.

j. Assay Cut-off

Assay cut-offs for the BD MAX Vaginal Panel were initially determined in pre-clinical studies. Data collected in the multi-site prospective clinical study was subsequently used to validate these cut-offs. For this validation, PCR metrics from vaginitis analytes and results generated by the BV call algorithm were graphically and statistically analyzed in comparison to results from applicable reference methods. ROC curve analysis was

performed to confirm the optimal cutoffs for each vaginitis analyte as well for the cutoffs used to determine results for bacterial vaginosis.

2. Clinical Studies:

Clinical performance characteristics for the BD MAX Vaginal Panel were evaluated in a prospective clinical study performed at 10 geographically diverse specimen collection sites. Of the 10 collection sites, seven sites performed specimen collection only and three sites performed both specimen collection as well as testing with the BD MAX Vaginal Panel.

For consented adult female subjects presenting with symptoms of vaginitis or bacterial vaginosis, one self-collected and one clinician-collected vaginal swab were collected using the BD MAX UVE Specimen Collection Kit and tested independently with the BD MAX Vaginal Panel. Three additional vaginal swabs were collected for reference method testing.

The following reference methods were performed for each patient:

- BV status was determined using a combination of Nugent Score and Amsel's criteria. Specimens with normal flora as per the Nugent Score were considered negative; those positive for BV flora were considered positive while those with intermediate BV flora were segregated into positive or negative categories using Amsel's criteria. Samples positive for 2 out of the 3 following criteria were considered Amsel's positive: vaginal pH > 4.5, presence of clue cells and positive Whiff test.
- *Candida* spp. status was determined by selective (Candida) chromogenic medium and Sabouraud Dextrose Emmons plate cultures. PCR amplification targeting the its2 gene was performed followed by bi-directional sequencing to identify all yeast isolates recovered by culture.
- *Trichomonas vaginalis* status was determined by a composite of microscopic visualization of motile trichomonads in saline wet mounts of vaginal secretion and by culture. A positive result either by wet mount or by culture was sufficient to categorize the patient as positive for Trichomonas vaginalis.

A total of 1763 subjects were enrolled in the prospective clinical study. Of those, 1740 subjects were compliant and 23 were found non-compliant as per protocol criteria. For clinician-collected specimens, the numbers of compliant specimens with reportable reference method and BD MAX Vaginal Panel results were 1559 for bacterial vaginosis, 1618 for *Candida* and 1600 for *Trichomonas vaginalis*. For self-collected specimens, the numbers of compliant specimens with reportable reference method and BD MAX Vaginal Panel results were 1582 for bacterial vaginosis, 1628 for *Candida* and 1610 for *Trichomonas vaginalis*.

BV Performance

Table 17 includes overall and per site performance for reporting of BV as observed in the prospective clinical study. The sensitivity and specificity for BV were 90.5% and 85.8 %

respectively for clinician-collected vaginal swabs, and 90.7% and 84.5 % respectively for self-collected vaginal swabs. For the population tested, this resulted in Positive Predictive Values (PPV) of 89.0 and 88.1 % for clinician-collected and self-collected specimens, respectively. Negative Predictive Values (NPV) of 87.7 % and 87.8% were obtained for clinician-collected and self-collected specimens, respectively. BV prevalence was 55.8% for patients with compliant reference method results.

Table 17: BV Performance by Collection Type and Collection Site

Site	Clinician-collected		Self-collected	
	Sensitivity	**Specificity**	**Sensitivity**	**Specificity**
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95%CI)
1	76.5 26/34 (60.0, 87.6)	96.6 113/117 (91.5, 98.7)	80.0 28/35 (64.1, 90.0)	94.1 111/118 (88.3, 97.1)
2	92.3 48/52 (81.8, 97.0)	78.9 30/38 (63.7, 88.9)	88.5 46/52 (77.0, 94.6)	76.9 30/39 (61.7, 87.4)
3	92.3 36/39 (79.7, 97.3)	81.0 17/21 (60.0, 92.3)	92.3 36/39 (79.7, 97.3)	70.0 14/20 (48.1, 85.5)
4	92.3 12/13 (66.7, 98.6)	66.7 4/6 (30.0, 90.3)	84.6 11/13 (57.8, 95.7)	66.7 4/6 (30.0, 90.3)
5	89.6 199/222 (84.9, 93.0)	87.9 131/149 (81.7, 92.2)	89.1 197/221 (84.4, 92.6)	88.0 139/158 (82.0, 92.2)
6	87.1 81/93 (78.8, 92.5)	87.8 72/82 (79.0, 93.2)	88.3 83/94 (80.2, 93.3)	85.4 70/82 (76.1, 91.4)
7	95.7 44/46 (85.5, 98.8)	84.8 28/33 (69.1, 93.3)	100.0 47/47 (92.4, 100.0)	80.0 28/35 (64.1, 90.0)
8	93.4 198/212 (89.2, 96.0)	75.0 87/116 (66.4, 82.0)	93.5 201/215 (89.4, 96.1)	78.5 95/121 (70.4, 84.9)
9	96.0 144/150 (91.5, 98.2)	77.6 52/67 (66.3, 85.9)	97.3 145/149 (93.3, 99.0)	73.5 50/68 (62.0, 82.6)
10	45.0 9/20 (25.8, 65.8)	98.0 48/49 (89.3, 99.6)	45.0 9/20 (25.8, 65.8)	96.0 48/50 (86.5, 98.9)
Overall	**90.5** **797/881** **(88.3, 92.2)**	**85.8** **582/678** **(83.0, 88.3)**	**90.7** **803/885** **(88.6, 92.5)**	**84.5** **589/697** **(81.6, 87.0)**

Tables 18, 19 and 20 include BV performance for clinician-collected and self-collected vaginal specimens stratified respectively by age group, ethnicity and patient clinical condition.

Table 18: BV Performance Stratified By Age Group

Age Group	Clinician-collected		Self-collected	
	Sensitivity	Specificity	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
18 - 29	90.3 531/588 (87.6, 92.4)	84.2 341/405 (80.3, 87.4)	91.2 539/591 (88.6, 93.2)	83.0 347/418 (79.1, 86.3)
30 - 39	91.0 182/200 (86.2, 94.2)	86.7 130/150 (80.3%, 91.2)	89.9 179/199 (85.0, 93.4)	85.0 130/153 (78.5, 89.8)
40 - 49	94.8 73/77 (87.4, 98.0)	89.8 79/88 (81.7, 94.5)	94.9 75/79 (87.7, 98.0)	87.8 79/90 (79.4, 93.0)
50 and over	68.8 11/16 (44.4, 85.8)	91.4 32/35 (77.6, 97.0)	62.5 10/16 (38.6, 81.5)	91.7 33/36 (78.2, 97.1)

Table 19: BV Performance Results Stratified by Ethnicity

Ethnicity	Prevalence[a]	Clinician-collected Specimens				Self-collected Specimens			
		Sensitivity	Specificity	PPV	NPV	Sensitivity	Specificity	PPV	NPV
		Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
Asian	50.9% 29/57	79.3 23/29 (61.6, 90.2)	100.0 26/26 (87.1, 100.0)	100.0 (87.3, 100.0)	82.4 (70.8, 92.4)	79.3 23/29 (61.6, 90.2)	88.9 24/27 (71.9, 96.1)	88.1 (73.3, 97.0)	80.6 (68.3, 91.1)
Black or African American	65.2% 559/857	91.9 502/546 (89.4, 93.9)	79.1 223/282 (74.0, 83.4)	89.2 86.9, 91.3	84.0 (79.8, 87.6)	92.5 506/547 (90.0, 94.4)	77.0 224/291 (71.8, 81.4)	88.3 (86.0, 90.4)	84.6 (80.4, 88.2)
Hispanic/Latino	39.5% 58/147	83.9 47/56 (72.2, 91.3)	84.9 73/86 (75.8, 90.9)	78.3 (69.1, 86.5)	89.0 (82.5, 94.2)	83.9 47/56 (72.2, 91.3)	87.5 77/88 (79.0, 92.9)	81.4 (72.2, 89.2)	89.3 (82.9, 94.4)
White (not Hispanic/Latino)	41.3% 164/397	90.7 146/161 (85.2, 94.3)	92.0 207/225 (87.7, 94.9)	88.9 (84.0, 92.8)	93.3 (89.9, 96.0)	90.2 148/164 (84.7, 93.9)	90.4 207/229 (85.9, 93.6)	86.9 (81.9, 91.0)	92.9 (89.5, 95.7)
Others/Mixed/ Unknown	58.6% 89/152	88.8 79/89 (80.5, 93.8)	89.8 53/59 (79.5, 95.3)	92.5 (86.0, 96.9)	85.0% (76.7, 91.8)	88.8 79/89 (80.5, 93.8)	91.9 57/62 (82.5, 96.5)	94.0 (87.8, 97.8)	85.3 (77.0, 91.8)

[a] Prevalence was calculated for specimens with compliant reference method results.

Table 20: BV Performance Stratified by Clinical Condition

Subgroup	Clinician-collected		Self-collected	
	Sensitivity	Specificity	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
Pregnant patients	88.9 8/9 (56.5, 98.0)	90.9 10/11 (62.3, 98.4)	88.9 8/9 (56.5, 98.0)	90.0 9/10 (59.6, 98.2)
Patients with estrogen therapy	86.4 57/66 (76.1, 92.7)	84.3 75/89 (75.3, 90.4)	91.0 61/67 (81.8, 95.8)	82.0 73/89 (72.8, 88.6)
Patients using anti-fungals	80.4 45/56 (68.2, 88.7)	93.8 75/80 (86.2, 97.3)	80.0 44/55 (67.6, 88.4)	90.9 80/88 (83.1, 95.3)
Patients with unprotected intercourse in the last 24 h	89.7 61/68 (80.2, 94.9)	68.3 28/41 (53.0, 80.4)	89.6 60/67 (80.0, 94.8)	69.8 30/43 (54.9, 81.4)
Patients with recurrent symptoms	87.6 162/185 (82.0, 91.6)	87.1 155/178 (81.4, 91.2)	86.1 161/187 (80.4, 90.3)	85.9 159/185 (80.2, 90.2)
Patients using oral antibiotics	82.3 79/96 (73.5, 88.6)	93.8 76/81 (86.4, 97.3)	84.4 81/96 (75.8, 90.3)	83.3 70/84 (73.9, 89.8)
Patients with menses	83.3 40/48 (70.4, 91.3)	86.5 32/37 (72.0, 94.1)	85.7 42/49 (73.3, 92.9)	86.8 33/38 (72.7, 94.2)
Patients without menses	90.8 754/830 (88.7, 92.6)	85.7 546/637 (82.8, 88.2)	91.0 758/833 (88.9, 92.8)	84.3 552/655 (81.3, 86.9)

Cgroup Performance

Table 21 includes overall and per site performance for detection of Cgroup (*Candida albicans, Candida tropicalis, Candida parapsilosis* and/or *Candida dubliniensis*) as observed in the prospective clinical study. The sensitivity and specificity were 90.9 and 94.1 % respectively for clinician-collected vaginal swabs, and 92.2 and 91.9 % respectively for self-collected vaginal swabs. For the population tested, this resulted in a PPV of 87.8 and 84.1 % for clinician-collected and self-collected specimens, respectively. NPV's of 95.7 and 96.2 % were obtained for clinician-collected and self-collected vaginal swabs, respectively. The prevalence of these *Candida* species combined was 31.6% for patients with compliant reference method results.

Table 21: Cgroup Performance per Collection Type and Collection Site

Site	Clinician-collected		Self-collected	
	Sensitivity	**Sensitivity**	**Specificity**	**Specificity**
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
1	96.4 53/55 (87.7, 99.0)	97.0 98/101 (91.6, 99.0)	98.2 54/55 (90.4, 99.7)	93.1 95/102 (86.5, 96.6)
2	82.8 24/29 (65.5, 92.4)	93.9 62/66 (85.4, 97.6)	93.1 27/29 (78.0, 98.1)	93.9 62/66 (85.4, 97.6)
3	61.5 8/13 (35.5, 82.3)	89.1 41/46 (77.0, 95.3)	83.3 10/12 (55.2, 95.3)	91.3 42/46 (79.7, 96.6)
4	100.0 3/3 (43.9, 100.0)	100.0 17/17 (81.6, 100.0)	100.0 3/3 (43.9, 100.0)	94.1 16/17 (73.0, 99.0)
5	96.1 99/103 (90.4, 98.5)	94.4 268/284 (91.0, 96.5)	91.3 95/104 (84.4, 95.4)	90.9 259/285 (87.0, 93.7)
6	91.9 57/62 (82.5, 96.5)	96.6 114/118 (91.6, 98.7)	85.2 52/61 (74.3, 92.0)	91.6 109/119 (85.2, 95.4)
7	90.9 30/33 (76.4, 96.9)	93.9 46/49 (83.5, 97.9)	91.2 31/34 (77.0, 97.0)	87.8 43/49 (75.8, 94.3)
8	95.4 104/109 (89.7, 98.0)	93.9 214/228 (90.0, 96.3)	96.4 107/111 (91.1, 98.6)	91.0 212/233 (86.6, 94.0)
9	86.4 70/81 (77.3, 92.2)	89.1 131/147 (83.1, 93.2)	90.0 72/80 (81.5, 94.8)	93.9 138/147 (88.8, 96.7)
10	70.0 14/20 (48.1, 85.5)	100.0 54/54 (93.4, 100.0)	90.5 19/21 (71.1, 97.3)	96.3 52/54 (87.5, 99.0)
Overall	**90.9** **462/508** **(88.1, 93.1)**	**94.1** **1045/1110** **(92.6, 95.4)**	**92.2** **470/510** **(89.5, 94.2)**	**91.9** **1028/1118** **(90.2, 93.4)**

Table 22 includes Cgroup performance stratified by each applicable *Candida* species identified by the reference culture and sequencing of the *its*2 gene.

Table 22: Cgroup Performance Stratified by Candida Species

Species (*its*2 gene ID)	Sensitivity	
	Clinician-collected	Self-collected
	Estimate 95% CI	
Candida albicans	91.0% 445/489 (88.1%, 93.2%)	92.0% 451/490 (89.3%, 94.1%)
Candida albicans (co-infected with *C. glabrata*)	92.3% 12/13 (66.7%, 98.6%)	100% 13/13 (77.2%, 100.0%)
Co-infection *Candida albicans* and *Candida tropicalis*	100.0% 1/1 (20.7%, 100.0%)	100.0% 1/1 (20.7%, 100.0%)
Candida dubliniensis	100.0% 3/3 (43.9%, 100.0%)	100.0% 3/3 (43.9%, 100.0%)
Candida tropicalis	50.0% 1/2 (9.5%, 90.5%)	66.7% 2/3 (20.8%, 93.9%)
Overall	90.9 462/508 (88.1, 93.1)	92.2 470/510 (89.5, 94.2)

Tables 23, 24 and 25 include Cgroup performance for clinician-collected and self-collected vaginal specimens stratified respectively by age group, ethnicity and patient clinical condition.

Table 23: Cgroup Performance Stratified by Age Group

Age Group	Clinician-collected		Self-collected	
	Sensitivity	Specificity	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
18 - 29	90.6 326/360 (87.1, 93.2)	93.5 618/661 (91.4, 95.1)	91.2 331/363 (87.8, 93.7)	91.3 608/666 (88.9, 93.2)
30 - 39	93.9 92/98 (87.3, 97.2)	94.3 250/265 (90.9, 96.5)	96.8 92/95 (91.1, 98.9)	91.7 244/266 (87.8, 94.5)
40 - 49	90.2 37/41 (77.5, 96.1)	94.5 120/127 (89.1, 97.3)	88.4 38/43 (75.5, 94.9)	93.0 119/128 (87.2, 96.3)
50 and over	77.8 7/9 (45.3, 93.7)	100.0 57/57 (93.7, 100.0)	100.0 9/9 (70.1, 100.0)	98.3 57/58 (90.9, 99.7)

Table 24: Cgroup Performance Stratified by Ethnicity

Ethnicity	Prev.[a]	Clinician-collected Specimens				Self-collected Specimens			
		Sensitivity	Specificity	PPV	NPV	Sensitivity	Specificity	PPV	NPV
		Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
Asian	20.0% 12/60	90.9 10/11 (62.3, 98.4)	97.8 45/46 (88.7, 99.6)	91.3 (67.5, 99.7)	97.7 (90.6, 99.9)	90.9 10/11 (62.3, 98.4)	93.8 45/48 (83.2, 97.9)	78.4 (57.1, 93.9)	97.6 (90.5, 99.9)
Black or African American	32.5% 287/884	91.3 253/277 (87.4, 94.1)	92.8 542/584 (90.4, 94.6)	85.9 (82.1, 89.3)	95.7 (93.9, 97.1)	92.4 257/278 (88.7, 95.0)	92.3 541/586 (89.9, 94.2)	85.3 (81.5, 88.6)	96.2 (94.5, 97.6)
Hispanic/Latino	36.1% 53/147	92.2 47/51 (81.5, 96.9)	96.7 88/91 (90.8, 98.9)	94.0 (85.3, 98.6)	95.6 (90.3, 98.7)	92.3 48/52 (81.8, 97.0)	92.4 85/92 (85.1, 96.3)	87.2 (78.0, 94.0)	95.5 (90.2, 98.6)
White (not Hispanic/Latino)	30.9% 126/408	88.8 111/125 (82.1, 93.2)	95.0 264/278 (91.7, 97.0)	88.7 (82.9, 93.2)	95.0 (92.3, 97.1)	92.0 115/125 (85.9, 95.6)	90.4 253/280 (86.3, 93.3)	81.0 (75.1, 86.3)	96.2 (93.6, 98.0)
Others/Mixed/ Unknown	28.7% 45/157	93.2 41/44 (81.8, 97.7)	95.5 106/111 (89.9, 98.1)	89.3 (79.1, 95.9)	97.2 (92.9, 99.4)	90.9 40/44 (78.8, 96.4)	92.9 104/112 (86.5, 96.3)	83.6 (73.2, 91.7)	96.2 (91.7, 98.9)

[a] Prevalence was calculated for specimens with compliant reference method results.

Table 25: Cgroup Performance Stratified by Health Condition

Subgroup	Clinician-collected		Self-collected	
	Sensitivity	Specificity	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
Pregnant patients	100.0 11/11 (74.1, 100.0)	88.9 8/9 (56.5, 98.0)	90.0 9/10 (59.6, 98.2)	88.9 8/9 (56.5, 98.0)
Patients with estrogen therapy	96.3 52/54 (87.5, 99.0)	91.7 100/109 (85.0, 95.6)	98.1 51/52 (89.9, 99.7)	90.9 100/110 (84.1, 95.0)
Patients using anti-fungals	89.8 44/49 (78.2, 95.6)	89.7 87/97 (82.1, 94.3)	92.0 46/50 (81.2, 96.8)	86.0 86/100 (77.9, 91.5)
Patients with unprotected intercourse in the last 24 h	94.7 36/38 (82.7, 98.5)	93.1 67/72 (84.8, 97.0)	95.0 38/40 (83.5, 98.6)	93.1 67/72 (84.8, 97.0)
Patients with recurrent symptoms	90.3 93/103 (83.0, 94.6)	92.7 253/273 (89.0, 95.2)	93.5 100/107 (87.1, 96.8)	91.2 248/272 (87.2, 94.0)
Patients using oral antibiotics	94.9 56/59 (86.1, 98.3)	92.2 119/129 (86.3, 95.7)	96.6 57/59 (88.5, 99.1)	86.8 112/129 (79.9, 91.6)
Patients with menses	85.7 18/21 (65.4, 95.0)	94.0 63/67 (85.6, 97.7)	95.5 21/22 (78.2, 99.2)	95.5 64/67 (87.6, 98.5)
Patients without menses	91.3 443/485 (88.5, 93.5)	94.2 978/1038 (92.6, 95.5)	92.2 448/486 (89.4, 94.3)	91.7 959/1046 (89.9, 93.2)

***Candida glabrata* Performance**

Table 26 includes overall and per site performance for detection of *C. glabrata* as observed in the prospective clinical study. The sensitivity and specificity were 75.9 and 99.7 % respectively for clinician-collected vaginal swabs and 86.7 and 99.6 % respectively for self-collected vaginal swabs. For the population tested, this resulted in PPV of 81.6 and 81.0 % for clinician-collected and self-collected specimens, respectively. NPV of 99.6 and 99.8 % were obtained for clinician-collected and self-collected specimens, respectively. The prevalence of *C. glabrata* was 1.8% for patients with compliant reference method results.

Table 26: *Candida glabrata* Performance by Collection Type and Collection Site

Site	Clinician-collected		Self-collected	
	Sensitivity	**Specificity**	**Sensitivity**	**Specificity**
	Percent (95% CI [a])	Percent (95% CI [a])	Percent (95% CI [a])	Percent (95% CI [a])
1	100.0 3/3 (43.9, 100.0)	100.0 153/153 (97.6, 100.0)	100.0 3/3 (43.9, 100.0)	100.0 154/154 (97.6, 100.0)
2	0.0 0/1 (0.0, 79.3)	100.0 94/94 (96.1, 100.0)	0.0 0/1 (0.0, 79.3)	100.0 94/94 (96.1, 100.0)
3	100.0 1/1 (20.7, 100.0)	100.0 58/58 (93.8, 100.0)	100.0 1/1 (20.7, 100.0)	100.0 57/57 (93.7, 100.0)
4	100.0 1/1 (20.7, 100.0)	100.0 19/19 (83.2, 100.0)	100.0 1/1 (20.7, 100.0)	100.0 19/19 (83.2, 100.0)
5	100.0 5/5 (56.6, 100.0)	99.7 381/382 (98.5, 100.0)	100.0 5/5 (56.6, 100.0)	99.2 381/384 (97.7, 99.7)
6	40.0 2/5 (11.8, 76.9)	100.0 175/175 (97.9, 100.0)	83.3 5/6 (43.6, 97.0)	100.0 174/174 (97.8, 100.0)
7	No data for Sensitivity calculation	100.0 82/82 (95.5, 100.0)	No data for Sensitivity calculation	98.8 82/83 (93.5, 99.8)
8	60.0 3/5 (23.1, 88.2)	99.1 329/332 (97.4, 99.7)	60.0 3/5 (23.1, 88.2)	99.4 337/339 (97.9, 99.8)
9	100.0 6/6 (61.0, 100.0)	99.5 221/222 (97.5, 99.9)	100.0 6/6 (61.0, 100.0)	100.0 221/221 (98.3, 100.0)
10	50.0 1/2 (9.5, 90.5)	100.0 72/72 (94.9, 100.0)	100.0 2/2 (34.2, 100.0)	100.0 73/73 (95.0, 100.0)
Overall	**75.9** **22/29[b,c]** **(57.9, 87.8)**	**99.7** **1584/1589** **(99.3, 99.9)**	**86.7** **26/30[d,e]** **(70.3, 94.7)**	**99.6** **1592/1598** **(99.2, 99.8)**

[a] CI: Confidence interval

[b] Out of 7 *C.glabrata* false negative results, 6 showed chromagar results consistent with low *C.glabrata* load (1+ to 2+ growth level) and 1 showed chromagar result consistent with high *C.glabrata* load (3+ growth level)

[c] The BD MAX Vaginal Panel detected BV and/or Cgroup signals in 6 out of 7 specimens with *C.glabrata* false negative results

[d] Out of 4 *C.glabrata* false negative results, 3 showed chromagar results consistent with low *C.glabrata* load (1+ to 2+ growth level) and 1 showed chromagar result consistent with high *C.glabrata* load (3+ growth level)
[e] The BD MAX Vaginal Panel detected BV and/or Cgroup signals in the 4 specimens with *C.glabrata* false negative results

Tables 27, 28 and 29 include *Candida glabrata* performance for clinician-collected and self-collected vaginal specimens stratified respectively by age group, ethnicity and pregnancy status.

Table 27: *Candida glabrata* Performance Stratified by Age Group

Age Group	Clinician-collected		Self-collected	
	Sensitivity	Specificity	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
18 - 29	73.7 14/19 (51.2, 88.2)	99.6 998/1002 (99.0, 99.8)	78.9 15/19 (56.7, 91.5)	99.7 1007/1010 (99.1, 99.9)
30 - 39	100.0 1/1 (20.7, 100.0)	100.0 362/362 (98.9, 100.0)	100.0 1/1 (20.7, 100.0)	99.4 358/360 (98.0, 99.8)
40 - 49	83.3 5/6 (43.6, 97.0)	99.4 161/162 (96.6, 99.9)	100.0 7/7 (64.6, 100.0)	99.4 163/164 (96.6, 99.9)
50 and over	66.7 2/3 (20.8, 93.9)	100.0 63/63 (94.3, 100.0)	100.0 3/3 (43.9, 100.0)	100.0 64/64 (94.3, 100.0)

Table 28: *Candida glabrata* Performance Stratified by Ethnicity

Ethnicity	Prev.[a]	Clinician-collected Specimens				Self-collected Specimens			
		Sens	Spec	PPV	NPV	Sens	Spec	PPV	NPV
		Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
Asian	3.3% 2/60	50.0 (1/2) (9.5, 90.5)	100.0 (55/55) (93.5, 100.0)	100.0 (6.1, 100.0)	98.3 (96.7, 100.0)	50.0 (1/2) (9.5, 90.5)	100.0 (57/57) (93.7, 100.0)	100.0 (6.3, 100.0)	98.3 (96.7, 100.0)
Black or African American	1.6% 14/884	78.6 (11/14) (52.4, 92.4)	99.6 (844/847) (99.0, 99.9)	78.1 (54.0, 94.1)	99.7 (99.2, 99.9)	85.7 (12/14) (60.1, 96.0)	99.5 (846/850) (98.8, 99.8)	74.6 (53.5, 91.4)	99.8 (99.3, 100.0)
Hispanic/Latino	2.0% 3/147	66.7 (2/3) (20.8, 93.9)	100.0 (139/139) (97.3, 100.0)	100.0 (28.4, 100.0)	99.3 (98.1, 100.0)	100.0 (3/3) (43.9, 100.0)	100.0 (141/141) (97.3, 100.0)	100.0 (44.0, 100.0)	100.0 (98.5, 100.0)
White (not Hispanic/ Latino)	1.7% 7/408	66.7 (4/6) (30.0, 90.3)	99.7 (396/397) (98.6, 100.0)	82.2 (41.4, 99.2)	99.4 (98.7, 99.9)	85.7 (6/7) (48.7, 97.4)	99.7 (397/398) (98.6, 100.0)	85.6 (51.0, 99.4)	99.8 (99.0, 100.0)
Others/Mixed/ Unknown	2.5% 4/157	100.0 4/4 (51.0, 100.0)	99.3 150/151 (96.3, 99.9)	79.8 (39.7, 99.4)	100.0 (98.4, 100.0)	100.0 4/4 (51.0, 100.0)	99.3 151/152 (96.4, 99.9)	79.9 (39.9, 99.4)	100.0 (98.4, 100.0)

[a] Prevalence was calculated for specimens with compliant reference method results.

Table 29: *Candida glabrata* Performance in Pregnant Patients

Subgroup	Clinician-collected		Self-collected	
	Sensitivity	Specificity	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
Pregnant patients	No data for Sensitivity calculation	100.0 20/20 (83.9, 100.0)	No data for Sensitivity calculation	100.0 19/19 (83.2, 100.0)

Due to the low prevalence of *Candida glabrata* observed in the prospective clinical study, evaluation of contrived specimens was performed to supplement the clinical data collected. Contrived specimens were prepared by spiking 50 different *Candida glabrata* strains into individual negative vaginal matrices. True negative specimens, containing vaginal matrix only, were interspersed with positive specimens and all specimen identities were blinded to the user. Strains were spiked at various clinically relevant organism concentrations and randomly distributed among three clinical testing sites for BD MAX Vaginal Panel testing. The study results demonstrated 100% positive agreement for all contrived positive specimens evaluated. Results for contrived specimens are presented in Table 30.

Table 30: *Candida glabrata* Contrived Specimens Results

Candida glabrata		Percent Agreement
Category	Load (x LoD)	Percent (95% CI)
High Positive	≥10 and <20	100.0 (5/5) (56.6, 100.0)
Moderate Positive	≥2 and <10	100.0 (20/20) (83.9, 100.0)
Low Positive	≥1 and <2	100.0 (25/25) (86.7, 100.0)
True Negative	No organisms	100.0 (50/50) (92.9, 100.0)

Candida krusei Performance

Performance of the BD MAX Vaginal Panel for detection of *Candida krusei* is presented in Table 31. No *Candida krusei* positive specimens were identified in the prospective study by the reference method; thus no data is available for sensitivity calculation. The specificity was 99.8 and 100.0 % for clinician-collected and self-collected vaginal swabs respectively.

Table 31: *Candida krusei* Performance Results

Collection Type	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)
Clinician-collected	No data for Sensitivity calculation	99.8 1614/1618 (99.4, 99.9)
Self-collected	No data for Sensitivity calculation	100.0 1628/1628 (99.8, 100.0)

Due to the lack of positive results for *C. krusei* observed in the prospective clinical study, evaluation of contrived specimens was performed to supplement the clinical data collected. Contrived specimens were prepared by spiking 50 different *Candida krusei* strains into individual negative vaginal matrices. True negative specimens, containing vaginal matrix only, were interspersed with positive specimens and all specimen identities were blinded to the user. Strains were spiked at various clinically relevant organism concentrations and randomly distributed among three clinical testing sites for BD MAX Vaginal Panel testing. The study results demonstrated 100% positive agreement for all contrived positive specimens evaluated. Results for contrived specimens are presented in Table 32.

Table 32: *Candida krusei* Contrived Specimens Results per Category

Candida krusei		Percent Agreement
Category	Load (x LoD)	Percent (95% CI)
High Positive	≥10 and <20	100.0 5/5 (56.6, 100.0)
Moderate Positive	≥2 and <10	100.0 20/20 (83.9, 100.0)
Low Positive	≥1 and <2	100.0 25/25 (86.7, 100.0)
True Negative	0	100.0 50/50 (92.9, 100.0)

Trichomonas vaginalis Performance

Table 33 includes overall and per site performance for detection of *T. vaginalis* as observed in the prospective clinical study. The assay sensitivity and specificity were 93.1 and 99.3 % respectively for clinician-collected vaginal swabs and 93.2 and 99.3 % respectively for self-collected vaginal swabs. For the population tested, this resulted in PPV of 91.8% and NPV of 99.4% for both collection types. The prevalence of *T. vaginalis* was 8.2% for patients with compliant reference method results.

Table 33: *Trichomonas vaginalis* Performance by Collection Type and Collection Site

Site	Clinician-collected		Self-collected	
	Sensitivity	**Specificity**	**Sensitivity**	**Specificity**
	Percent (95% CI[a])	Percent (95% CI[a])	Percent (95% CI[a])	Percent (95% CI[a])
1	No data for Sensitivity calculation	100.0 168/168 (97.8, 100.0)	No data for Sensitivity calculation	100.0 169/169 (97.8, 100.0)
2	100.0 4/4 (51.0, 100.0)	97.9 92/94 (92.6, 99.4)	100.0 5/5 (56.6, 100.0)	98.9 92/93 (94.2, 99.8)
3	100.0 17/17 (81.6, 100.0)	97.8 45/46 (88.7, 99.6)	100.0 17/17 (81.6, 100.0)	97.8 44/45 (88.4, 99.6)
4	60.0 3/5 (23.1, 88.2)	100.0 15/15 (79.6, 100.0)	60.0 3/5 (23.1, 88.2)	100.0 15/15 (79.6, 100.0)
5	86.7 26/30 (70.3, 94.7)	99.4 308/310 (97.7, 99.8)	86.7 26/30 (70.3, 94.7)	99.0 309/312 (97.2, 99.7)
6	90.9 10/11 (62.3, 98.4)	98.3 170/173 (95.0, 99.4)	100.0 12/12 (75.8, 100.0)	98.3 169/172 (95.0, 99.4)
7	100.0 6/6 (61.0, 100.0)	100.0 67/67 (94.6, 100.0)	100.0 6/6 (61.0, 100.0)	100.0 68/68 (94.7, 100.0)
8	93.1 27/29 (78.0, 98.1)	99.4 320/322 (97.8, 99.8)	90.0 27/30 (74.4, 96.5)	99.4 326/328 (97.8, 99.8)
9	100.0 27/27 (87.5, 100.0)	99.5 206/207 (97.3, 99.9)	100.0 27/27 (87.5, 100.0)	99.5 205/206 (97.3, 99.9)
10	100.0 1/1 (20.7, 100.0)	100.0 68/68 (94.7, 100.0)	100.0 1/1 (20.7, 100.0)	100.0 69/69 (94.7, 100.0)
Overall	**93.1** **121/130[b]** **(87.4, 96.3)**	**99.3** **1459/1470[c]** **(98.7, 99.6)**	**93.2** **124/133[b]** **(87.6, 96.4)**	**99.3** **1466/1477[c]** **(98.7, 99.6)**

[a] CI: Confidence interval
[b] 9 false-negative results were recorded. Of those, 7 were found negative with an FDA-cleared molecular method.
[c] 11 false-positive results were recorded. Of those, 10 were found positive with an FDA-cleared molecular method.

Tables 34, 35 and 36 include *Candida glabrata* performance for clinician-collected and self-collected vaginal specimens stratified respectively by age group, ethnicity and patient clinical condition.

Table 34: *Trichomonas vaginalis* Performance Stratified by Age Group

Age Group	Clinician-collected		Self-collected	
	Sensitivity	Specificity	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
18 - 29	94.9 74/78 (87.5, 98.0)	99.6 924/928 (98.9, 99.8)	95.0 76/80 (87.8, 98.0)	99.5 928/933 (98.8, 99.8)
30 - 39	96.3 26/27 (81.7, 99.3)	98.2 326/332 (96.1, 99.2)	96.3 26/27 (81.7, 99.3)	98.2 325/331 (96.1, 99.2)
40 - 49	93.8 15/16 (71.7, 98.9)	99.3 150/151 (96.3, 99.9)	94.1 16/17 (73.0, 99.0)	100.0 153/153 (97.6, 100.0)
50 and over	66.7 6/9 (35.4, 87.9)	100.0 59/59 (93.9, 100.0)	66.7 6/9 (35.4, 87.9)	100.0 60/60 (94.0, 100.0)

Table 35: *Trichomonas vaginalis* Performance Results Stratified by Ethnicity

Ethnicity	Prevalence[a]	Clinician-collected Specimens				Self-collected Specimens			
		Sensitivity	Specificity	PPV	NPV	Sensitivity	Specificity	PPV	NPV
		Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
Asian	1.6% 1/61	100.0 (1/1) (20.7, 100.0)	98.2 (56/57) (90.7, 99.7)	48.7 (2.8, 97.4)	100.0 (98.4, 100.0)	100.0 (1/1) (20.7, 100.0)	100.0 (59/59) (93.9, 100.0)	100.0 (6.3, 100.0)	100.0 (98.4, 100.0)
Black or African American	11.8% 105/887	94.1 (95/101) (87.6, 97.2)	99.2 (756/762) (98.3, 99.6)	94.1 (88.4, 97.6)	99.2 (98.3, 99.7)	94.2 (97/103) (87.9, 97.3)	99.3 (759/764) (98.5, 99.7)	95.1 (89.5, 98.3)	99.2 (98.4, 99.7)
Hispanic/Latino	3.5% 5/141	100.0 (5/5) (56.6, 100.0)	99.2 (130/131) (95.8, 99.9)	82.8 (46.5, 99.5)	100.0 (98.1, 100.0)	100.0 (5/5) (56.6, 100.0)	99.2 (132/133) (95.9, 99.9)	83.0 (46.9, 99.5)	100.0 (98.1, 100.0)
White (not Hispanic/Latino)	3.4% 14/412	100.0 (13/13) (77.2, 100.0)	99.5 (392/394) (98.2, 99.9)	87.4 (65.9, 98.4)	100.0 (99.1, 100.0)	100.0 (14/14) (78.5, 100.0)	99.2 (392/395) (97.8, 99.7)	82.2 (61.5, 95.7)	100.0 (99.2, 100.0)
Others/Mixed/ Unknown	7.3% 10/137	70.0 7/10 (39.7, 89.2)	99.2 125/126 (95.6, 99.9)	87.4 (55.4, 99.5)	97.7 (95.1, 99.5)	70.0 7/10 (39.7, 89.2)	98.4 124/126 (94.4, 99.6)	77.6 (48.1, 97.0)	97.7 (95.1, 99.4)

[a] Prevalence was calculated for specimens with compliant reference method results.

Table 36: Trichomonas vaginalis Performance Stratified by Health Condition

Subgroup	Clinician-collected		Self-collected	
	Sensitivity	Specificity	Sensitivity	Specificity
	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)	Percent (95% CI)
Pregnant patients	100.0 1/1 (20.7, 100.0)	100.0 18/18 (82.4, 100.0)	100.0 1/1 (20.7, 100.0)	100.0 17/17 (81.6, 100.0)
Patients with recurrent symptoms	92.0 23/25 (75.0, 97.8)	99.7 338/339 (98.3, 99.9)	92.3 24/26 (75.9, 97.9)	99.4 338/340 (97.9, 99.8)

Multi-Analyte Detection Rates

The rates of multi-analyte detections by the BD MAX Vaginal Panel observed in the prospective clinical study are presented in Table 37. The data presented includes specimens with compliant results for all targets by both the BD MAX Vaginal Panel and reference methods. The most prevalent multi-analyte detection was a combination of BV and Cgroup with 13.9% and 15.3% for clinician and self-collected specimens respectively. In total, 21.5% of clinician-collected and 23.3% of self-collected specimens resulted in more than one BD MAX Vaginal Panel analyte or result reported.

Table 37: BD MAX Vaginal Panel Multi-Analyte Detection Rates

Analytes Detected	Clinician-collected	Self-collected
BV and Cgroup	13.9% 205/1471	15.3% 229/1494
BV and TV	4.9% 72/1471	4.6% 68/1494
BV and Cgroup and TV	1.4% 21/1471	1.5% 23/1494
BV and Cgroup and Cgla	0.5% 7/1471	0.6% 9/1494
BV and Cgla	0.2% 3/1471	0.5% 7/1494
Cgroup and TV	0.3% 4/1471	0.3% 5/1494
Cgroup and Cgla	0.2% 3/1471	0.4% 6/1494
BV and Cgroup and Ckru	0.1% 2/1471	0.0% 0/1494
BV and Cgla and TV	0.0% 0/1471	0.1% 1/1494
Total	21.5% 317/1471	23.3% 348/1494

A comparison of multi-analyte detections based on all reportable specimen results is presented in Table 38 for clinician and self-collected specimens. Bolded entries represent multi-analyte detection events with concordant reference method and BD MAX Vaginal Panel results. Non-bolded entries represent specimens with discordant results. Concordant single detections are not represented.

Table 38: Multi-Analyte Detections Observed in the Prospective Clinical Study

	Total Number of Occurrences Clinician collected / Self- collected Reference Method														
	Organism Detections	BV	BV, Cgroup	BV, C.glabrata	BV, Cgroup C. glabrata	BV, TV	BV, Cgroup TV	BV, C. glabrata, TV	Cgroup	Cgroup, C.glabrata	Cgroup, TV	Cgroup, C. glabrata, TV	C. glabrata	TV	Negative
BD MAX Vaginal Panel	BV		16/17	2/1	-	2/2	-	-	-	-	-	-	-	-	-
	BV, Cgroup	37/51	**139/143**	0/1	1/1	-	0/1	-	36/42	2/1	-	-	-	-	3/3
	BV, C.glabrata	1/2	-	**1/1**	-	-	-	-	-	-	-	-	1/4	-	-
	BV, Cgroup, C.glabrata	0/1	2/1	-	**4/4**	-	-	-	1/0	1/3	-	0/1	-	-	-
	BV, Cgroup, C.krusei	1/0	1/0	-	-	-	-	-	-	-	-	-	-	-	-
	BV, TV	7/7	1/0	-	-	**48/47**	5/5	-	0/1	-	2/1	-	-	16/11	1/0
	BV, Cgroup TV	-	-	-	-	3/2	**17/18**	-	-	-	1/3	-	-	0/2	-
	BV, C.glabrata, TV	-	-	-	-	-	-	**0/1**	-	-	-	-	-	-	-
	Cgroup	-	33/32	-	-	-	1/0	-		1/0	1/0	-	-	-	-
	Cgroup, C.glabrata	-	-	-	1/1	-	-	-	-	**1/2**	-	1/0	0/2	-	0/1
	Cgroup, TV	-	-	-	-	-	-	-	2/2	-	**1/3**	-	-	1/0	-
	TV	-	0/1	-	-	10/12	-	-	-	-	1/0	-	-		-
	Negative	-	2/2	-	-	1/1	-	-	-	1/0	-	-	-	-	

Non-Reportable Rates

Of all specimens initially evaluated with the BD MAX Vaginal Panel in the prospective clinical study, 3.0 and 2.9 % initially reported as Unresolved for clinician and self-collected specimens, respectively. Following a valid repeat test, 1.3 and 0.6% remained Unresolved for clinician and self-collected specimens, respectively. Of all specimens initially evaluated with the BD MAX Vaginal Panel, 3.7 and 2.7 % initially reported as Indeterminate for clinician and self-collected specimens, respectively. Following a valid repeat test, 0.8 and 0.6 % remained Indeterminate for clinician and self-collected specimens, respectively. Of all specimens initially evaluated with the BD MAX Vaginal Panel, 1.4% initially reported as Incomplete for both collection types. Following a valid repeat test, 0.2 % remained Incomplete for both collection types. The total rates of non-reportable results were 8.1 and 7.0% for clinician and self-collected specimens, respectively. Following a valid repeat test, 2.2 and 1.4 % remained non-reportable for clinician and self-collected specimens, respectively. Results are presented in Table 39.

Table 39: Non-reportable Rates

Collection Type	Unresolved Rate		Indeterminate Rate		Incomplete Rate		Total Rate	
	Initial Percent (95% CI)	**Final**[a] Percent (95% CI)	**Initial** Percent (95% CI)	**Final**[a] Percent (95% CI)	**Initial** Percent (95% CI)	**Final**[a] Percent (95% CI)	**Initial Percent (95% CI)**	**Final**[a] **Percent (95% CI)**
Clinician-collected	3.0 52/1734 (2.3, 3.9)	1.3 22/1725 (0.8, 1.9)	3.7 64/1734 (2.9, 4.7)	0.8 13/1725 (0.4, 1.3)	1.4 24/1734 (0.9, 2.1)	0.2 3/1725 (0.1, 0.5)	**8.1** 140/1734 (6.9, 9.5)	**2.2** 38/1725 (1.6, 3.0)
Self-collected	2.9 50/1736 (2.2, 3.8)	0.6 11/1733 (0.4, 1.1)	2.7 47/1736 (2.0, 3.6)	0.6 10/1733 (0.3, 1.1)	1.4 24/1736 (0.9, 2.0)	0.2 4/1733 (0.1, 0.6)	**7.0** 121/1736 (5.9, 8.3)	**1.4** 25/1733 (1.0, 2.1)

[a] The final rate is calculated with valid repeats only

Evaluation of the BD MAX Vaginal Panel in Asymptomatic Women

Although the BD MAX Vaginal Panel is not intended for testing specimens from asymptomatic women, presence of *Candida* species, *T. vaginalis* and BV markers has been reported in this population. The BD MAX Vaginal Panel was evaluated for detection of *Candida* species, *T. vaginalis* and BV markers with vaginal specimens collected from 202 asymptomatic women. BD MAX Vaginal Panel vaginitis targets were detected with rates varying from 1.5% for *C. krusei* to 20.8% for Cgroup. Positive BV results were generated for 34.2% of asymptomatic women. Results from the study are presented in Table 40 which also includes results for the most prevalent ethnic groups enrolled. BV, Cgroup and *T. vaginalis* were detected in all ethnic categories.

Table 40: BD MAX Vaginal Panel Positive Rates in Asymptomatic Women

Target	Overall	By Ethnic Group		
		Black/African American	White (not Hispanic)	Others[a]
BV	34.2% 69/202	40.4% (38/94)	28.8% (23/80)	28.6% (8/28)
Cgroup	20.8% 42/202	22.3% (21/94)	16.3% (13/80)	28.6% (8/28)
C. glabrata	5.9% 12/202	11.7% (11/94)	0.0% (0/80)	3.6% (1/28)
C. krusei	1.5% 3/202	1.1% (1/94)	2.5% (2/80)	0.0% (0/28)
T. vaginalis	11.4% 23/202	22.3% (21/94)	1.3% (1/80)	3.6% (1/28)

[a] Including: American Indian or Alaska natives, Asian, Mixed Ethnicity and Unknown

3. Clinical Cutoff:

See Assay Cut-off Section L.1.j above.

4. Expected Values:

The incidence of each BD MAX Vaginal Panel result as observed in the prospective clinical study is presented in Table 41, stratified by clinic type and specimen type.

Table 41: BD MAX Vaginal Panel Positivity Rate by Clinic Type

Collection Type	Clinic Type	Bacterial Vaginosis	Cgroup[a]	Candida glabrata	Candida krusei	Trichomonas vaginalis
Clinician-collected	STD / HIV	72.7% (224/308)	34.0% (105/309)	2.6% (8/309)	0.3% (1/309)	15.9% (49/309)
	Family Planning	60.7% (683/1125)	33.3% (379/1138)	1.2% (14/1138)	0.1% (1/1138)	7.5% (85/1138)
	OB/Gyn	20.6% (52/252)	29.6% (75/253)	2.0% (5/253)	0.8% (2/253)	0.4% (1/253)
	Total	56.9% (959/1685)	32.9% (559/1700)	1.6% (27/1700)	0.2% (4/1700)	7.9% (135/1700)
Self-collected	STD / HIV	74.6% (229/307)	33.9% (104/307)	2.3% (7/307)	0.0% (0/307)	16.0% (49/307)
	Family Planning	60.1% (687/1143)	35.1% (403/1147)	1.7% (19/1147)	0.0% (0/1147)	7.7% (88/1147)
	OB/Gyn	22.0% (56/255)	34.5% (88/255)	2.4% (6/255)	0.0% (0/255)	0.4% (1/255)
	Total	57.0% (972/1705)	34.8% (595/1709)	1.9% (32/1709)	0.0% (0/1709)	8.1% (138/1709)

[a] *Candida albicans, Candida tropicalis, Candida parapsilosis* and/or *Candida dubliniensis*

M. Instrument Name

BD MAX System

N. System Descriptions:

1. Modes of Operation:

   The BD MAX System fully automates cell lysis, nucleic acid extraction, PCR set-up, target amplification and detection. For the BD MAX Vaginal Panel, the system can process and analyze up to 12 specimens in one cartridge with two cartridges running simultaneously on the instrument. The system includes external and internal barcode reading, ensuring traceability throughout the extraction and PCR processes. The system includes a heater module, temperature sensors, and a fluorescence detection system with six optical channels.

2. Software:

   FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

   Yes ___X___ or No _____

3. Specimen Identification:

   Specimens are identified via barcode.

4. Specimen Sampling and Handling:

   Sample Buffer Tubes containing vaginal swab specimens are vortexed for one minute on the Multi-tube Vortexer, after which the user uncaps each specimen, removes the excess

fluid from the swab, discards the swab and then recaps the tube with a blue septum cap. Specimens are then loaded into the BD MAX System Rack on the BD MAX System after which all additional specimen handling steps are automated.

5. Calibration:

   The system is calibrated by the manufacturer on-site as part of the installation procedure as well as during biannual preventive maintenance.

6. Quality Control:

   See Quality Control Section above (Section L.1.c)

O. Other Supportive Instrument Performance Characteristics Data Not Covered In The "Performance Characteristics" Section above:

Not Applicable

P. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Parts 801 and 809 and the specials controls for this device type.

Q. Identified Risks to Health and Identified Mitigations:

Identified Risks to Health	Identified Mitigations
Incorrect identification or lack of identification of a pathogenic microorganism by the device can lead to improper patient management	General controls and special controls (1), (2), (3), and (4)
Failure to correctly interpret test results	General controls and special controls (5), (6), (7), and (8)

R. Benefit/Risk Analysis:

Summary	
Summary of the Benefit(s)	• The BD MAX Vaginal Panel detects nucleic acids from microorganisms associated with bacterial vaginosis, candidiasis and trichomoniasis from a clinician or self-collected vaginal swab to aid in the diagnosis of bacterial vaginosis, vulvovaginal candidiasis and trichomoniasis. • The BD MAX Vaginal Panel provides molecular detection of analytes that are typically diagnosed clinically and/or with microscopy, which may reduce human error from microscopy or clinical diagnosis of signs and symptoms. • The BD MAX Vaginal Panel may reduce operator error and provide for more uniform diagnosis of bacterial vaginosis, vulvovaginal candidiasis and trichomoniasis.
Summary of the Risk(s)	• False positives and false negative results are the primary risks associated with the BD MAX Vaginal Panel. • False positive results would result in an unnecessary course of oral or topical antimicrobials. • False negative results could result in delayed diagnosis of bacterial vaginosis or trichomoniasis. Patients who remain symptomatic are likely to be retested, or receive empiric therapy.
Summary of Other Factors	None.
Conclusions Do the probable benefits outweigh the probable risks?	The probable benefits of the BD MAX Vaginal Panel outweigh the potential risks in light of the established special controls and applicable general controls, including design controls. The BD MAX Vaginal Panel is the first multiplex panel using nucleic acid amplification to detect multiple microorganisms associated with bacterial vaginosis, vulvovaginal candidiasis and trichomoniasis. Potential risks include false positive and false negative results, but it is highly unlikely that a patient would suffer a serious adverse event as a result of an erroneous result from the BD MAX Vaginal Panel given the clinical performance demonstrated in the prospective clinical trial and the special controls established for this device. Risks are further mitigated by the use of the device in association with clinical assessment, supplemental laboratory testing, natural progression of clinical symptoms and clinical judgment. Ultimately, the majority of risks associated with the BD MAX Vaginal Panel may be minimized with appropriate precautions and the BD MAX Vaginal Panel may better standardize the assessment of vaginosis/vaginitis.

Patient Perspectives:

This submission did not include specific information on patient perspectives for this device.

S. Conclusion:

The information provided in this *de novo* submission is sufficient to classify this device into class II under regulation 21 CFR 866**.**3975. FDA believes that the stated special controls, and applicable general controls, including design controls, provide reasonable assurance of the safety and effectiveness of the device type. The device is classified under the following:

Product Code: PQA, OUY, OOI, NSU

Device Type: Device that detects nucleic acid sequences from microorganisms associated with vaginitis and bacterial vaginosis

Class: II (special controls)

Regulation: 21 CFR 866.3975

(a) Identification. A device that detects nucleic acid sequences from microorganisms associated with vaginitis and bacterial vaginosis is a qualitative *in vitro* device intended for the detection of microbial nucleic acid sequences in vaginal specimens collected from patients with signs and symptoms of vaginitis or bacterial vaginosis. This device is intended to aid in the diagnosis of vaginitis or bacterial vaginosis when used in conjunction with clinical signs and symptoms and other laboratory findings.

(b) Classification. Class II (special controls). A device that detects nucleic acid sequences from microorganisms associated with vaginitis and bacterial vaginosis is subject to the following special controls:

1) Premarket notification submissions must include a detailed device description of the following:
   (i) Device components;
   (ii) Ancillary reagents required but not provided; and
   (iii) Explanation of the methodology including primer/probe sequence, design, and rationale for sequence selection.

2) Premarket notification submissions must include information that demonstrates the performance characteristics of the device, including:
   (i) Limit of Detection;
   (ii) Precision (reproducibility);
   (iii) Analytical specificity;
   (iv) Analytical reactivity (inclusivity);
   (v) Specimen stability; and
   (vi) Effects of interfering substances.

3) Premarket notification submissions must include detailed documentation from a prospective clinical study. As appropriate to the intended use, the prospective clinical study must be performed on an appropriate study population including women of various ages and ethnicities. The prospective clinical study must

compare the device performance to results obtained from well-accepted comparator methods.

4) Premarket notification submissions must include detailed documentation for device software, including, but not limited to, software applications and hardware-based devices that incorporate software.

5) A detailed explanation of the interpretation of results and acceptance criteria must be included in the device's 21 CFR 809.10(b)(9) compliant labeling.

6) For indications for use that include detection of nucleic acid sequences from bacteria associated with bacterial vaginosis, the 21 CFR 809.10(b)(12) compliant labeling must include clinical performance stratified by patient demographics such as race, ethnicity, age, and pregnancy status.

7) For indications for use that include detection of nucleic acid sequences from bacteria associated with bacterial vaginosis, the 21 CFR 809.10(b)(12) compliant labeling must include a summary of device results in an asymptomatic population with demographic characteristics appropriate to the intended use population.

8) For indications for use that include detection of either Candida species or bacteria associated with bacterial vaginosis, the 21 CFR 809.10 compliant labeling must include a limitation that *Candida* species and bacterial compositions associated with bacterial vaginosis can be present as part of normal vaginal flora and results should be considered in conjunction with available clinical information.