510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION MEMORANDUM

A. 510(K) Number:

K190452

B. Purpose for Submission:

To obtain a substantial equivalence determination for the Aptima BV assay.

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. gasseri. L. crispatus* and *L. jensenii*)
    - *Gardnerella vaginalis*
    - *Atopobium vaginae*

D. Type of Test:

The Aptima BV assay, performed on the automated Panther system, is a nucleic acid-based test for the detection of the above listed bacteria in vaginal specimens obtained from symptomatic patients.

E. Applicant:

Hologic, Inc.

F. Proprietary and Established Names:

Aptima BV Assay

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, NSU, PMN

4. Panel:

   83 - Microbiology

H. Indication(s) for Use:

1. Indications for Use(s):

   The Aptima BV assay is an in vitro nucleic acid amplification test that utilizes real time transcription-mediated amplification (TMA) for detection and quantitation of ribosomal RNA from bacteria associated with bacterial vaginosis (BV), including *Lactobacillus* (*L. gasseri, L. crispatus*, and *L. jensenii*), *Gardnerella vaginalis*, and *Atopobium vaginae*. The assay reports a qualitative result for BV and does not report results for individual organisms. The assay is intended to aid in the diagnosis of BV on the automated Panther system using clinician-collected and patient-collected vaginal swab specimens from females with a clinical presentation consistent with vaginitis and/or vaginosis.

3. Special conditions for use statement(s):

   For *in vitro* diagnostic use only

   For Prescription Use Only

4. Special instrument requirements:

   The Aptima BV assay is performed on the Panther System.

I. Device Description:

The Aptima BV assay is an *in vitro* nucleic acid amplification test for the detection and quantitation of rRNA from bacteria associated with bacterial vaginosis in women with a clinical presentation consistent with vaginitis/vaginosis. The Aptima BV assay utilizes the automated Panther system to provide qualitative results to aid in the diagnosis of bacterial vaginosis.

Materials provided in each Aptima BV assay kit:

The Aptima BV assay is provided as a 100-test kit. The Aptima BV assay master kit contains 8 reagents, 1 calibrator, and 2 controls required for sample processing. There are 4 boxes that make up the assay master kit. Boxes 1 and 2 contain the Aptima BV assay reagents packaged

according to storage conditions. Box 3 contains the calibrator, and Box 4 contains the controls when provided as part of the master kit. The Aptima BV Calibrator and Controls kit may also be procured separately if customers need additional calibrators or controls. A listing of the components that are required to perform the Aptima BV assay are detailed in Table 1. In addition, there is one ancillary kit required to run the assay, and one collection kit utilized for collection of specimens (Table 2).

Table 1: Reagents Required to Perform the Aptima BV Assay

Box	Components Description
1	Amplification Reagent
	Enzyme Reagent
	Promoter Reagent
	Internal Control
2	Amplification Reconstitution Solution
	Enzyme Reconstitution Solution
	Promoter Reconstitution Solution
	Target Capture Reagent
3	Positive Calibrator
4	Negative Control
	Positive Control

Table 2: Ancillary and Collection Kits Required to Perform the Aptima BV Assay

Aptima Assay Fluids Kit
Aptima Multi-test Swab Specimen Collection Kit

J. Substantial Equivalence Information:

1. Predicate device name(s):

   BD MAX Vaginal Panel

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

   DEN160001

3. Comparison with predicate:

Similarities		
Item	**New Device: Aptima BV Assay (K190452)**	**Predicate: BD MAX Vaginal Panel (DEN160001)**
Indication for Use	The Aptima BV assay is an *in vitro* nucleic acid amplification test that utilizes real time transcription mediated amplification (TMA) for detection and quantitation of ribosomal RNA from bacteria associated with bacterial vaginosis (BV), including *Lactobacillus* (*L. gasseri, L. crispatus,* and *L. jensenii*), *Gardnerella vaginalis,* and *Atopobium vaginae*. The assay reports a qualitative result for BV and does not report results for individual organisms. The assay is intended to aid in the diagnosis of BV on the automated Panther system using clinician-collected and patient-collected vaginal swab specimens from females with a clinical presentation consistent with vaginitis and/or vaginosis.	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. 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.
Patient Population	Women with a clinical presentation of vaginitis/vaginosis	Same
Specimen Types	Vaginal swabs in patients who are symptomatic for vaginitis/vaginosis	Same
Assay Controls	Incorporates an Internal Control in every test. Uses external positive and negative controls.	Same
Test Interpretation	Automated test interpretation and report generation.	Same

Similarities		
Item	**New Device: Aptima BV Assay (K190452)**	**Predicate: BD MAX Vaginal Panel (DEN160001)**
Result Type	Qualitative	Same
User Complexity	Moderate	Same

Differences		
Item	**New Device: Aptima BV Assay (K190452)**	**Predicate: BD MAX Vaginal Panel (DEN160001)**
Organisms Detected	*Lactobacillus* (*L. gasseri*, *L. crispatus*, and *L. jensenii*), *Gardnerella vaginalis*, and *Atopobium vaginae*	*Lactobacillus* (*L. crispatus*, and *L. jensenii*), *Lactobacillus* (*L. gasseri*, *L. crispatus*, and *L. jensenii*), *Atopobium vaginae*, Bacterial Vaginosis Associated Bacteria-2 (BVAB-2), Megasphaera-1, *Candida* (*C. albicans*, *C. tropicalis*, *C. parapsilosis*, *C. dubliniensis*), C*andida glabrata, Candida krusei*, *Trichomonas vaginalis*
Technological Principles	Real-time Transcription Mediated Amplification (TMA)	Real-time polymerase chain reaction (PCR)
Instrumentation	Panther System	BD Max System
Analyte	Ribosomal RNA	DNA

K. Standard/Guidance Document Referenced:

- CLSI EP6-A - Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Guideline
- Approach; Approved
- Guidance for Industry and FDA Staff -Nucleic Acid Based In Vitro Diagnostic Devices for Detection of Microbial Pathogens (2005)
- Guidance for Industry and FDA Staff -Design Considerations for Pivotal Clinical Investigations for Medical Devices (2013)
- Guidance for Industry and FDA Staff -Collection of Race and Ethnicity Data in Clinical Trials (2016)

L. Test Principle:

The Aptima BV assay involves three main steps, all of which take place in a single tube on the Panther system: target capture, target amplification by TMA, and detection of the amplification products (amplicon) by fluorescent labeled probes (torches). The assay

incorporates an internal control (IC) in every test to monitor nucleic acid capture, amplification and detection.

Target Capture:
Specimens are collected in a tube containing specimen transport media (STM) that lyses the cells, releases the RNA, and protects it from degradation during storage. When the Aptima BV assay is performed, capture oligonucleotides hybridize to highly conserved regions of the target RNA, if present, in the test specimen. The hybridized target is then captured onto magnetic microparticles that are separated from the specimen in a magnetic field. Wash steps remove extraneous components from the reaction tube.

Target Amplification by TMA:
Target amplification occurs via TMA, a transcription-based nucleic acid amplification method that utilizes two enzymes, Moloney murine leukemia virus (MMLV) reverse transcriptase and T7 RNA polymerase. The reverse transcriptase is used to generate a DNA copy of the target RNA sequence, adding a promoter sequence for T7 RNA polymerase. T7 RNA polymerase produces multiple copies of RNA amplicon from the DNA copy template.

Detection:
Detection is achieved using single-stranded nucleic acid "torches" that are present during the amplification of the target and hybridize specifically to the amplicon in real time. Each torch has a fluorophore and a quencher. The quencher suppresses the fluorescence of the fluorophore when the torch is not hybridized to the amplicon. When the torch binds to the amplicon, the fluorophore is separated from the quencher and emits a signal at a specific wavelength when excited by a light source. The Panther system detects and discriminates between four fluorescent signals corresponding to *Lactobacillus* group, *Atopobium vaginae*, *Gardnerella vaginalis* and IC amplification products. The Panther system software compares signal emergence times for each target organism to calibration information in order to determine the BV Positive or Negative status of each sample.

Test Interpretation:
Test results are automatically determined by the assay software. Table 3 below shows the possible results reported in a valid run and result interpretations. Samples with invalid test results should be retested.

Table 3: Aptima BV Assay Result Interpretation

BV Result	Interpretation
Positive	Positive for BV
Negative	Negative for BV
Invalid	Invalid Test

M. Performance Characteristics:

1. Analytical Performance:

a. Reproducibility:

Aptima BV assay reproducibility was evaluated at three US sites using seven panel members. Testing was performed using one lot of assay reagents and six operators (two at each site). At each site, testing was performed for at least six days. Each run had three replicates of each panel member.

The panel members were made using a simulated vaginal swab matrix ('SVSM', which contains specimen transport media (STM) spiked with simulated vaginal fluid) negative for *Lactobacillus* species, *G. vaginalis*, and *A. vaginae*. Six panel members contained cell lysates of at least 1 of the following organisms: *L. crispatus*, *L. jensenii*, *G. vaginalis*, or *A. vaginae*; different bacterial combinations were prepared to represent the variety of targeted BV organism combinations present in vaginal specimens. One negative panel member contained only the matrix with no added target analytes. Results are shown in the Table 4 below.

Table 4: Agreement of Aptima BV Assay Results With Expected Results

Panel Description	Expected Results	N	Agreement, % (95% CI)
True Neg	Negative	36/36	100 (96.6-100)
BV Neg[1]	Negative	36/36	100 (96.6-100)
Gvag Low Pos	Positive	36/36	100 (96.6-100)
Avag Low Pos	Positive	36/36	100 (96.6-100)
BV Low Pos[1]	Positive	36/36	100 (96.6-100)
Gvag Mod Pos	Positive	36/36	100 (96.6-100)
Avag MosPos	Positive	36/36	100 (96.6-100)

Avag = *A. vaginae*, CI = Score confidence interval, Gvag = *G. vaginalis*, Mod=moderate, Neg = negative, Pos = positive

[1] Panel member contains 2 different organisms.

Across organisms/panel members, the total %CV values ranged from 4.21% to 4.76%; total SD values were ≤1.12. For most panel members, the "between sites," "between operators," and "between runs" factors were the largest contributors to total variability; for all other sources of variation, SD values were ≤0.18 (%CV values were ≤1.13%), as shown in Table 5 below.

Table 5: Signal Variability of the Aptima BV Assay by Analyte-positive Panel Member

			Between Sites		Between Operators		Between Days		Between Runs		Within Runs		Total	
Panel Description	**N**	**Mean T Time[1]**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**
Lcrisp BV Neg[2]	108	19.73	0.30	1.53	0.61	3.07	0.13	0.64	0.63	3.17	0.12	0.62	0.94	4.76
Ljen Low Pos[2]	108	24.31	0.00	0.00	0.77	3.16	0.00	0.00	0.80	3.28	0.15	0.62	1.12	4.60
Gvag Low Pos	108	15.69	0.35	2.26	0.40	2.52	0.00	0.00	0.38	2.43	0.15	0.96	0.67	4.28
Gvag Mod Pos	108	14.33	0.30	2.07	0.37	2.58	0.00	0.00	0.35	2.41	0.14	0.98	0.60	4.21
Avag BV Neg[2]	108	18.01	0.39	2.15	0.44	2.46	0.08	0.45	0.47	2.59	0.18	0.97	0.78	4.30
Avag Low Pos	108	14.95	0.38	2.52	0.41	2.75	0.00	0.00	0.39	2.61	0.14	0.93	0.69	4.64
Avag Low Pos[2]	108	14.94	0.41	2.76	0.37	2.51	0.00	0.00	0.37	2.45	0.17	1.13	0.69	4.60
Avag Mod Pos	108	13.99	0.29	2.08	0.36	2.60	0.03	0.18	0.39	2.82	0.14	1.00	0.63	4.48

Avag = *A. vaginae*, CV = coefficient of variation, Gvag = *G. vaginalis*, Lcrisp = *L. crispatus*, Ljen = *L. jensenii*, Mod = moderate, Neg = negative, Pos = positive, SD = standard deviation, TTime = emergence time of a signal (above a specific threshold)

Note: If variability from a factor was numerically negative, SD and CV are shown as 0.0.

[1] The assay reports TTime for each assay analyte separately; the mean and signal variability reported are for the TTime corresponding to the analyte(s) present in each panel member.

[2] Panel member contains 2 different organisms; results are shown for only the component shown.

For Aptima BV assay controls and positive calibrators, the total %CV values ranged from 4.47% to 5.36%; total SD values were ≤1.11 (see Tables 6 and 7, respectively).

Table 6: Signal Variability of the Aptima BV Assay Positive Controls

			Between Sites		Between Operators		Between Days		Within Runs		Total	
Control	**N**	**Mean Ct**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**
Neg - Lcrisp	36	20.34	0.24	1.16	0.74	3.63	0.00	0.00	0.71	3.48	1.05	5.16
Pos - Gvag	36	17.52	0.05	0.28	0.60	3.41	0.00	0.00	0.52	2.96	0.79	4.52
Pos - Avag	36	13.79	0.27	1.97	0.38	2.77	0.00	0.00	0.41	2.97	0.62	4.51

Avag = *A. vaginae*, CV = coefficient of variation, Gvag = *G. vaginalis*, Lcrisp = *L. crispatus*, Ljen = *L. jensenii*, Mod = moderate, Neg = negative, Pos = positive, SD = standard deviation, TTime = emergence time of a signal (above a specific threshold)

Table 7: Signal Variability of the Aptima BV Assay Positive Calibrators

			Between Sites		Between Operators		Between Days		Between Runs		Within Runs		Total	
Analyte	**N**	**Mean T Time**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**
Lcrisp	108	20.75	0.00	0.00	0.78	3.77	0.00	0.00	0.78	3.75	0.13	0.64	1.11	5.36
Gvag	108	19.03	0.34	1.78	0.61	3.19	0.00	0.00	0.56	2.96	0.15	0.79	0.91	4.76
Avag	108	18.46	0.41	2.23	0.48	2.62	0.08	0.42	0.51	2.76	0.11	0.58	0.83	4.47

Avag = *A. vaginae*, CV = coefficient of variation, Gvag = *G. vaginalis*, Lcrisp = *L. crispatus*, SD = standard deviation, TTime = emergence time of a signal (above a specific threshold)

b. Precision:

Within-laboratory precision was evaluated for the Aptima BD assay on three Panther systems at one site. Three operators performed testing across 21 days and three reagent lots. Each operator performed two runs per day using an 11 member panel. Each run consisted of three replicates of each panel member. The panel members were made using SVSM negative for *Lactobacillus* species, *G. vaginalis*, and *A. vaginae*. Ten panel members contained cell lysates of at least 1 of the following organisms: *L. crispatus, L. jensenii, G. vaginalis*, or *A. vaginae*; different bacterial combinations were prepared to represent the variety of targeted BV organism combinations present in vaginal specimens. Ten panel members targeted BV Negative (<5% BV Positive), BV High Negative (20-80% BV positive), BV Low Positive (~95% BV positive) and BV Moderate Positive (100% BV positive) results. One negative panel member contained matrix with no added target analytes.

BV percent positive results for each panel are presented in Table 8. Signal variability (TTime) of the Aptima BV assay was calculated for each target in analyte positive panel members. Variability calculated between operators, between instruments, between days, between lots, between runs, within run, and overall, is shown in Tables 9 through 11.

Table 8: BV Positivity of Precision Panels

Panel Description	BV Positive/ Total n	Expected BV Positivity	BV Positivity (95% CI)
SVSM	0/168	0%	0 (0.0-1.6)
L. crispatus, A. vaginae BV Negative	0 /168	<5%	0 (0.0-1.6)
L. crispatus, G. vaginalis BV High Negative	76 /168	20-80%	45.2 (37.9-52.8)
L. crispatus, G. vaginalis, A. vaginae BV High Negative	131/165[1]	20-80%	79.4 (72.6-84.9)
G. vaginalis BV Low Positive	168/168	≥95%	100 (98.4-100.0)
A. vaginae BV Low Positive	168/168	≥95%	100 (98.4-100.0)
L. jensenii, A. vaginae BV Low Positive	168/168	≥95%	100 (98.4-100.0)
G. vaginalis, A. vaginae BV Low Positive	168/168	≥95%	100 (98.4-100.0)
L. crispatus, G. vaginalis, A. vaginae BV Low Positive	168/168	≥95%	100 (98.4-100.0)
G. vaginalis BV Mod Positive	168/168	100%	100 (98.4-100.0)

A. vaginae BV Mod Positive	168/168	100%	100 (98.4-100.0)

[1] Three invalid results were excluded from the analysis.

Table 9: Signal Variability of *Lactobacillus* Panel Members

			Between Operators		Between Instruments		Between Days		Between Lots		Between Runs		Within Run		Total	
Panel Description	**N**	**Mean T Time[1]**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**
L. crispatus BV Negative[2]	168	19.87	0.10	0.49	0.16	0.80	0.14	0.71	1.03	5.18	0.17	0.09	0.18	0.93	1.08	5.46
L. crispatus BV High Negative[2]	168	23.95	0.11	0.47	0.12	0.52	0.19	0.79	1.22	5.11	0.18	0.77	0.28	1.15	1.29	5.40
L. crispatus BV High Negative[3]	165[4]	22.40	0.09	0.40	0.17	0.74	0.20	0.87	1.22	5.47	0.09	0.39	0.27	1.21	1.29	5.74
L. jensenii BV Low Positive[2]	168	24.80	0.10	0.38	0.14	0.57	0.14	0.57	1.33	5.35	0.17	0.69	0.25	1.01	1.38	5.56
L. crispatus BV Low Positive[3]	168	23.51	0.15	0.63	0.09	0.40	0.17	0.73	1.36	5.77	0.10	0.44	0.31	1.31	1.42	6.02

CV = Coefficient of variation

[1]TTime is shown for *Lactobacillus* only

[2]Panel member contains 2 different organisms: results are shown for only the *Lactobacillus* component.

[3]Panel member contains different organisms: results are shown for only the *Lactobacillus* component.

[4] Three invalid results were excluded from the analysis.

Note: Variability from some factors may be numerically negative. This can occur if the variability due to those factors is very small. In these cases, SD and CV are shown as 0.00.

Table 10: Signal Variability of *G. vaginalis* Panel Members

			Between Operators		Between Instruments		Between Days		Between Lots		Between Runs		Within Run		Total	
Panel Description	**N**	**Mean T Time[1]**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**
G. vaginalis BV High Negative[2]	168	17.11	0.00	0.00	0.18	1.08	0.17	0.99	0.47	2.75	0.17	0.96	0.16	0.94	0.58	3.39
G. vaginalis BV High Negative[3]	165[4]	15.71	0.00	0.00	0.19	1.19	0.18	1.12	0.48	3.05	0.11	0.72	0.12	0.79	0.57	3.62
G. vaginalis BV Low Positive	168	15.80	0.00	0.00	0.16	1.00	0.14	0.89	0.43	2.70	0.15	0.97	0.15	0.92	0.52	3.30

G. vaginalis BV Mod Positive	168	14.46	0.00	0.00	0.17	1.18	0.05	0.35	0.38	2.63	0.16	1.09	0.18	1.25	0.48	3.35
G. vaginalis BV Low Positive[2]	168	15.01	0.00	0.00	0.14	0.93	0.14	0.91	0.40	2.67	0.16	1.08	0.13	0.86	0.49	3.28
G. vaginalis BV Low Positive[3]	168	14.06	0.00	0.00	0.16	1.11	0.15	1.09	0.39	2.75	0.14	0.99	0.16	1.16	0.49	3.51

CV = Coefficient of variation, Mod = moderate
[1]TTime is shown for *G. vaginalis* only.
[2]Panel member contains 2 different organisms: results are shown for only the *G. vaginalis* component.
[3]Panel member contains 3 different organisms: results are shown for only the *G. vaginalis* component.
[4]Three invalid results were excluded from the analysis.
Note: Variability from some factors may be numerically negative. This can occur if the variability due to those factors is very small. In these cases, SD and CV are shown as 0.00.

Table 11: Signal Variability of *A. vaginae* Panel Members

			Between Operators		Between Instruments		Between Days		Between Lots		Between Runs		Within Run		Total	
Panel Description	**N**	**Mean TTime[1]**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**	**SD**	**CV (%)**
A. vaginae BV Negative[2]	168	18.20	0.02	0.11	0.25	1.36	0.15	0.84	0.58	3.17	0.19	1.02	0.19	1.05	0.70	3.84
A. vaginae BV High Negative[3]	165[4]	16.56	0.00	0.00	0.25	1.53	0.18	1.11	0.56	3.38	0.13	0.79	0.12	0.70	0.67	4.02
A. vaginae BV Low Positive	168	15.11	0.00	0.00	0.19	1.25	0.15	0.97	0.51	3.40	0.12	0.82	0.12	0.78	0.59	3.92
A. vaginae BV Low Positive[2]	168	15.13	0.00	0.00	0.20	1.30	0.12	0.80	0.51	3.34	0.14	0.89	0.16	1.07	0.59	3.92
A. vaginae BV Mod Positive	168	14.13	0.08	0.54	0.21	1.50	0.17	1.21	0.51	3.63	0.08	0.57	0.20	1.40	0.62	4.41
A. vaginae BV Low Positive[2]	168	15.78	0.03	0.16	0.17	1.09	0.10	0.65	0.50	3.17	0.16	1.00	0.12	0.75	0.57	3.64
A. vaginae BV Low Positive[3]	168	15.61	0.00	0.00	0.23	1.47	0.15	0.94	0.51	3.29	0.10	0.66	0.18	1.15	0.62	3.95

CV = Coefficient of variation, Mod = moderate
[1]TTime is shown for *A. vaginae* only
[2]Panel member contains 2 different organisms: results are shown for only the *A. vaginae* component.
[3]Panel member contains 3 different organisms: results are shown for only the *A. vaginae* component.
[4]Three invalid results were excluded from the analysis.
Note: Variability from some factors may be numerically negative. This can occur if the variability due to those factors is very small. In these cases, SD and CV are shown as 0.00.

c. Linearity/Assay Reportable Range:

A study was performed to demonstrate linearity for the individual organisms detected in the Aptima Bacterial Vaginosis assay using dilutions of one representative strain for each bacterial species *Atopobium vaginae, Gardnerella vaginalis, Lactobacillus crispatus, Lactobacillus gasseri* and *Lactobacillus jensenii* in simulated vaginal swab matrix. The linearity of each bacterial species was demonstrated by testing cell lysate at multiple dilutions with concentrations known relative to each other and performing a polynomial regression analysis to determine an acceptable degree of non-linearity (±0.5 log CFU/mL). Dilutions spanned the clinically relevant range of the assay. Experimental design was based on CLSI EP6-A.

Atopobium vaginae, *Gardnerella vaginalis*, *Lactobacillus crispatus*, *Lactobacillus gasseri* and *Lactobacillus jensenii* tested as lysate in simulated vaginal swab matrix quantified linearly within their clinically relevant range. All five organisms quantified linearly with a criteria of ≤0.25 log CFU/mL for repeatability and ≤0.5 log CFU/mL for nonlinearity.

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

Internal Control

An internal control consisting of a non-infectious RNA transcript is added to each reaction via the Target Capture Reagent. The IC monitors nucleic acid capture, amplification, detection, and operator or instrument error. During sample processing, IC acceptance criteria are automatically verified by the Panther System software. If an IC result is invalid, the sample result is invalidated. Every sample with an invalid IC result is retested.

External Assay Controls

External controls are not provided with the Aptima BV assay; however, testing of external positive and negative controls are recommended in the assay labeling. External controls should be tested in conformance with local, state, and/or federal regulations or accreditation requirements and each laboratory's standard quality control procedures.

Calibrators:

A positive and negative calibrator are run in duplicate each time a reagent kit is loaded on the Panther System. Assay calibration is required to generate valid results. The Aptima BD assay calibration is valid for up to 48 hours. Software on the Panther System alerts the operator when a new calibrator set should be run.

Specimen Stability

Evaluation of specimen stability was performed to establish the stability of vaginal swab specimens using the Aptima BV assay on Panther. The study combined different storage conditions in a nested design.

The study was conducted using twenty individual clinical vaginal swab pools that are negative for *L. crispatus*, *G. vaginalis*, and *A. vaginae* were evaluated. Each pool was split into four aliquots and spiked with lysate of either *Lactobacillus crispatus*, (3xLOD), *A. vaginae* (3xC95), *G. vaginalis* (3xC95) or a mixture of all three at a BV low positive level. Each aliquot was split to create three sample tubes for testing under various storage conditions.

The results of this study demonstrated that vaginal swab specimen are stable after storage up to 30 days at 30°C, 60 days at -20°C, 60 days at 2 to 8°C, after three cycles of freeze/thaw, or 60 days at 2 to 8°C followed by 61 days at -20°C when tested with the Aptima BV assay.

The study data provided support the specimen handling recommendations described in the Aptima BV assay package insert.

e. Limit of Detection:

The analytical sensitivity (Limit of Detection or LoD) and BV positivity limits of the Aptima BV assay were determined by testing a series of panels consisting of *L. crispatus, L. gasseri, L. jensenii, G. vaginalis*, or *A. vaginae* cell lysates diluted into simulated vaginal swab matrix. A minimum of 20 replicates of each panel member were tested with each of two reagent lots for a minimum of 40 replicates per panel member to determine the assay LoD for each target (i.e., organism concentration at which >95% of replicates are detected). The predicted detection limits for each organism calculated using Probit analysis are shown in Table 12 below.

Table 12: Limit of Detection Results

Organism	Predicted Detection Limit	CFU/mL
Atopobium vaginae	95%	290[1]
Gardnerella vaginalis	95%	55[1]
Lactobacillus crispatus	95%	143
Lactobacillus gasseri	95%	2207
Lactobacillus jensenii	95%	10

[1]Predicted BV Positivity Limits (C95) for *A. vaginae* and *G. vaginalis* in the Aptima BV assay are approximately 5.10 log CFU/mL and 4.86 log CFU/mL, respectively.

f. Analytical Inclusivity:

An analytical inclusivity study was conducted to evaluate the Aptima BV assay 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 *G. vaginalis, A. vaginae,* and *Lactobacillus* species (*L. crispatus, L. gasseri,* and *L. jensenii*) in simulated vaginal swab matrix. Samples were inoculated at 3x LoD of the corresponding reference strain evaluated in the LoD study. The Aptima BV assay correctly identified all five strains of *G. vaginalis* and *A. vaginae* at 3X C95. All five strains of *L. crispatus* and *L. gasseri* were detected at 3X LoD. Three of the five strains of *L. jensenii* were detected at 3X LoD, and the remaining two strains were detected at 10X LoD.

g. Analytical Specificity/Cross-reactivity and Microbial Interference:

Cross-reactivity and microbial interference with the Aptima BV assay was evaluated in the presence of non-targeted organisms. A panel consisting of 62 organisms (Table 13 below) was tested in simulated vaginal swab matrix in the absence or in the presence of *L. crispatus* at 3X LoD, *G. vaginalis* at 3X C95, or *A. vaginae* at 3X C95. No cross-

reactivity or microbial interference was observed for any of the 62 organisms tested in the Aptima BV assay at the following concentrations.

Table 13: Cross-Reactivity and Microbial Interference Panel

Microorganism	Concentration	Microorganism	Concentration
Acinetobacter lwoffii	$1x10^6$ CFU/mL	Herpes simplex virus I	$1x10^4$ $TCID_{50}$/mL
Actinomyces israelii	$1x10^6$ CFU/mL	Herpes simplex virus II	$1x10^4$ $TCID_{50}$/mL
Alcaligenes faecalis	$1x10^6$ CFU/mL	HIV	$1x10^5$ copies/mL
Atopobium minutum	$1x10^6$ CFU/mL	*Klebsiella pneumoniae*	$1x10^6$ CFU/mL
Atopobium parvulum	$1x10^6$ CFU/mL	*Lactobacillus acidophilus*	$1x10^3$ CFU/mL[2]
Atopobium rimae	$1x10^6$ CFU/mL	*Lactobacillus iners*	$1x10^6$ CFU/mL
Bacteroides fragilis	$1x10^6$ CFU/mL	*Lactobacillus mucosae*	$1x10^6$ CFU/mL
Bifidobacterium adolescentis	$1x10^6$ CFU/mL	*Leptotrichia buccalis*	$1x10^6$ CFU/mL
Bifidobacterium breve	$1x10^6$ CFU/mL	*Listeria monocytogenes*	$1x10^6$ CFU/mL
BVAB-1[1]	$1x10^6$ copies/mL	*Megasphaera Type 1*[1]	$1x10^6$ copies/mL
BVAB-2[1]	$1x10^6$ copies/mL	*Mobiluncus curtisii*	$1x10^6$ CFU/mL
Campylobacter jejuni	$1x10^6$ CFU/mL	*Mycoplasma genitalium*	$1x10^6$ CFU/mL
Candida albicans	$1x10^6$ CFU/mL	*Mycoplasma hominis*	$1x10^6$ CFU/mL
Candida dubliniensis	$1x10^6$ CFU/mL	*Neisseria gonorrhoeae*	$1x10^6$ CFU/mL
Candida glabrata	$1x10^6$ CFU/mL	*Pentatrichomonas hominis*	$1x10^5$ cells/mL
Candida krusei	$1x10^6$ CFU/mL	*Peptostreptococcus magnus*	$1x10^6$ CFU/mL
Candida lusitaniae	$1x10^6$ CFU/mL	*Pichia fermentans*	$1x10^6$ CFU/mL
Candida orthopsilosis	$1x10^6$ CFU/mL	*Prevotella bivia*	$1x10^6$ CFU/mL
Candida parapsilosis	$1x10^6$ CFU/mL	*Propionibacterium acnes*	$1x10^6$ CFU/mL
Candida tropicalis	$1x10^6$ CFU/mL	*Proteus vulgaris*	$1x10^6$ CFU/mL
Chlamydia trachomatis	$1x10^6$ IFU/mL	SiHa cells	$1x10^4$ cells/mL
Clostridium difficile	$1x10^6$ CFU/mL	*Sneathia amnii*	$1x10^6$ CFU/mL
Corynebacterium genitalium	$1x10^6$ CFU/mL	*Staphylococcus aureus*	$1x10^6$ CFU/mL
Cryptococcus neoformans	$1x10^6$ CFU/mL	*Staphylococcus epidermidis*	$1x10^6$ CFU/mL
Eggerthella lenta	$1x10^6$ CFU/mL	*Streptococcus agalactiae*	$1x10^6$ CFU/mL
Enterobacter cloacae	$1x10^6$ CFU/mL	*Streptococcus pyogenes*	$1x10^6$ CFU/mL
Enterococcus faecalis	$1x10^6$ CFU/mL	*Treponema pallidum*[1]	$1x10^6$ copies/mL
Escherichia coli	$1x10^6$ CFU/mL	*Trichomonas tenax*	$1x10^5$ cells/mL
Fusobacterium nucleatum	$1x10^6$ CFU/mL	*Trichomonas vaginalis*	$1x10^5$ cells/mL
Haemophilus ducreyi	$1x10^6$ CFU/mL	*Ureaplasma parvum*	$1x10^6$ CFU/mL
HeLa cells	$1x10^4$ cells/mL	*Ureaplasma urealyticum*	$1x10^6$ CFU/mL

CFU = Colony Forming Units; IFU = Inclusion Forming Units; $TCID_{50}$ = Median Tissue Culture Infectious Dose
[1]*In Vitro* Transcript tested.
[2]*Lactobacillus acidophilus* affects BV positivity at $1x10^4$ CFU/mL or higher.

h. Interfering Studies

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 (3x LoD*)* of *L. crispatus, G. Vaginalis* and *A. vaginae.* No interference was observed in the presence of the following exogenous and endogenous substances tested at the concentrations listed in Table 14 below.

Table 14: Exogenous and Endogenous Substances Tested for Interference

Substance	Final Concentration[1]
Whole Blood	5% V/V
Leukocytes	$1x10^6$ cells/mL
Mucus[2]	1.5% V/V
Seminal Fluid	5% V/V
Contraceptive Foam	5% W/V
Contraceptive Film	5% W/V
Tioconazole[3]	1% W/V
Douche	5% W/V
Progesterone	5% W/V
Estradiol	5% W/V
Acyclovir	5% W/V
Metronidazole	5% W/V
Hemorrhoidal Cream	5% W/V
Vaginal Moisturizing Gel[4]	0.4% W/V
Lubricant	5% V/V
Spermicide	5% W/V
Anti-fungal	5% W/V
Deodorant/Spray	5% W/V
Glacial Acetic Acid	5% V/V
Vagisil Cream	5% W/V

W/V = weight by volume; V/V = volume by volume

[1] Final Concentration represents final concentration in the sample when tested on the Panther instrument.

[2] Interference was observed with Mucus at ≥2% W/V and not observed at 1.5% V/V.

[3]Interference was observed with Tioconazole 6.5% Ointment at 5% W/V and not observed at 1% W/V.

[4] Interference was observed with Vaginal Moisturizing Gel at ≥0.5% W/V and not observed at 0.4% W/V.

i. Matrix Equivalence Study

Because the BV analytes detected by the Aptima BV assay are present in normal vaginal flora, it was necessary to use a simulated vaginal matrix for preparation of samples for

some analytical studies. The simulated vaginal swab matrix consists of Specimen Transport Media (STM) combined with a simulated vaginal swab matrix (SVSM).

The LoD was determined by testing a series of panels consisting of *L. crispatus, L. gasseri, L. jensenii, G. vaginalis,* or *A. vaginae* cell lysates diluted into SVSM. A minimum of 20 replicates of each panel member were tested with each of two reagent lots for a minimum of 40 replicates per panel member.

j. Assay Cut-off

Assay cut-offs for the Aptima BV assay were initially determined in a pre-clinical training set (pCTS). The pCTS data were generated during feasibility or development from specimens from a total of 1204 unique symptomatic subjects (drawn from four multi-site pre-clinical collections). Patient infected status (BV Positive, BV Negative) was established using Nugent scores, with intermediates determined by Amsel criteria.

2. Clinical Performance:

Clinical performance characteristics for the Aptima BV assay were evaluated in a prospective clinical study performed using prospectively-collected patient- and clinician collected vaginal swab samples from women ≥14 years who were asymptomatic for or who exhibited signs and/or symptoms of vaginitis (i.e., symptomatic). Twenty-one participating geographically diverse US private and academic family practice, obstetric-gynecologic, family planning, public health, STI, medical group clinics, and clinical research centers obtained vaginal swab samples from 1519 symptomatic women, 174 asymptomatic women, and 4 women with unknown symptom status.

For each symptomatic subject, two vaginal swab samples (one patient-collected, one clinician collected) were tested with the investigational Aptima BV assay. The clinician-collected vaginal swab sample was also used for Nugent score evaluation, and modified Amsel criteria if necessary, to determine BV infection status. A Nugent interpretation established positive and negative BV reference status, except in cases of intermediate determinations. For intermediate Nugent interpretations, BV reference status was established using modified Amsel criteria. For each asymptomatic subject, one clinician-collected vaginal swab sample was collected and tested with the investigational Aptima BV assay.

The clinical performance of the Aptima BV assay in symptomatic subjects (i.e., the intended use population) was estimated relative to the BV infection status; sensitivity, specificity, PPV, and NPV were calculated for each Aptima sample type, along with corresponding 2-sided 95% CIs. Positivity rates were calculated for asymptomatic subjects.

Of the 1519 symptomatic subjects enrolled, 102 were not evaluable due to withdrawal (n = 17) or unknown BV infection status (n = 85). The remaining 1417 symptomatic subjects were evaluable for at least one of the sample types. Of the 174 asymptomatic

subjects, 2 were not evaluable due to withdrawal; the remaining 172 asymptomatic subjects were evaluable. Table 15 shows the demographic and baseline clinical characteristics of evaluable subjects.

Of the 1502 non-withdrawn symptomatic subjects, 1417 subjects were evaluable for the performance analyses for BV detection; results from 1405 patient-collected Aptima vaginal swab samples (99.2%, 1405/1417) and 1413 clinician-collected Aptima vaginal swab samples (99.7%, 1413/1417) were included in the performance analyses. All 172 non-withdrawn asymptomatic subjects were evaluable for the performance analyses for BV detection; results from the 172 clinician-collected Aptima vaginal swab samples were included in the positivity analysis.

Table 15: Summary of Demographics of Evaluable Subjects in the Aptima BV Assay Evaluation

	Symptomatic	**Asymptomatic**
Total, N	1417	172
Age, years		
Mean ± SD	34.7 ± 11.11	41.1 ± 13.22
Median	33.0	40.0
Range	14-75	18-73
Age category (years), n (%)		
14-17	4 (0.3)	0 (0.0)
18-29	537 (37.9)	42 (24.4)
30-39	469 (33.1)	42 (24.4)
40-49	235 (16.6)	36 (20.9)
>50	172 (12.1)	52 (30.2)
Race/Ethnicity, n (%)		
Asian	67 (4.7)	5 (2.9)
Black or African American	731 (51.6)	75 (43.6)
White (Hispanic or Latino)	248 (17.5)	41 (23.8)
White (Not Hispanic or Latino)	307 (21.7)	44 (25.6)
Other[1]	64 (4.5)	7 (4.1)

[1]Includes patient-reported other, mixed, and unknown races

Performance characteristics for detection of BV infection for patient-collected and clinician collected vaginal swab samples from symptomatic subjects were calculated overall (see Table 16), by race/ethnicity (see Table 17), and by clinical condition (see Table 18).

Table 16: Aptima BV Assay Performance Relative to BV Infection Status in Symptomatic Subjects

Specimen Type	N	TP	FP	TN	FN	Prevalence[1] (95% CI)[2]	Sensitivity (95% CI)[2]	Specificity (95% CI)[2]	PPV (95% CI)[3]	NPV (95% CI)[3]
Patient collected	1405	673	101[4]	612	19[5]	49.3	97.3 (95.8-98.2)	85.8 (83.1-88.2)	87.0 (84.8-88.9)	97.0 (95.5-98.1)
Clinician collected	1413	660	75[6]	643	35[7]	49.2	95.0 (93.1-96.4)	89.6 (87.1-91.6)	89.8 (87.7-91.7)	94.8 (93.1-96.3)

FN = false negative, FP = false positive, NPV = negative predictive value, PPV = positive predictive value, TP = true positive, TN = true negative

[1]Study prevalence reported.

[2]Score CI.

[3]PPV 95% CI computed from the exact 95% CI for positive likelihood ratio; NPV 95% CI computed from the 95% for the negative likelihood ratio

[4] Of the 101 false positive results, 55 subjects were Nugent intermediates and had BV infection status determined by Amsel criteria, and 9 were positive by Amsel.

[5] Of the 19 false negative results, 6 subjects were Nugent intermediates and had BV infection status determined by Amsel criteria, and 7 were negative by Amsel.

[6] Of the 75 false positive results, 46 subjects were Nugent intermediates and had BV infection status determined by Amsel criteria, and 6 were positive by Amsel.

[7] Of the 35 false negative results, 10 subjects were Nugent intermediates and had BV infection status determined by Amsel criteria, and 15 were negative by Amsel.

Table 17: Aptima BV Assay Performance Relative to BV Infection Status in Symptomatic Subjects, by Race/Ethnicity

Race/Ethnicity Patient-collected	N	TP	FP	TN	FN	Prev[1] (%)	Sensitivity (95% CI)[2]	Specificity (95% CI)[2]	PPV (95% CI)[3]	NPV (95% CI)[3]
Asian	65	19	6	39	1	30.8	95.0 (76.4-99.1)	86.7 (73.8-93.7)	76.0 (61.6-88.7)	97.5 (89.3-99.9)
Black / African American	727	434	43	239	11	61.2	97.5 (95.6-98.6)	84.8 (80.1-88.5)	91.0 (88.6-93.1)	95.6 (92.6-97.7)
White (Hispanic/Latino)	246	112	22	111	1	45.9	99.1 (95.2-99.8)	83.5 (76.2-88.8)	83.6 (78.0-88.6)	99.1 (95.6- 100)
White (Not Hispanic/Latino)	303	81	27	189	6	28.7	93.1 (85.8-96.8)	87.5 (82.4-91.3)	75.0 (68.1-81.5)	96.9 (94.0-98.8)
Other[4]	64	27	3 3	34	0	42.2	100 (87.5-100)	91.9 (78.7-97.2)	90.0 (76.9-97.7)	100 (91.3- 100)
Clinician-collected										
Asian	67	20	4	42	1	31.3	95.2 (77.3-99.2)	91.3 (79.7-96.6)	83.3 (68.2-94.3)	97.7 (89.8-99.9)
Black / African American	729	425	31	253	20	61.0	95.5 (93.2-97.1)	89.1 (84.9-92.2)	93.2 (90.9-95.2)	92.7 (89.4-95.2)
White (Hispanic/Latino)	247	110	18	115	4	46.2	96.5 (91.3-98.6)	86.5 (79.6-91.3)	85.9 (80.3-90.8)	96.6 (92.3-99.0)
White (Not Hispanic/Latino)	306	78	18	200	10	28.8	88.6 (80.3-93.7)	91.7 (87.3-94.7)	81.3 (73.9-87.5)	95.2 (92.1-97.5)
Other[4]	64	27	4	33	0	42.2	100 (87.5-100)	89.2 (75.3-95.7)	87.1 (74.2-96.0)	100 (91.1- 100)

FN = false negative, FP = false positive, NPV = negative predictive value, PPV = positive predictive value, Prev = prevalence TP = true positive, TN = true negative

[1] Study prevalence reported.

[2] Score CI.

[3] PPV 95% CI computed from the exact 95% CI for positive likelihood ratio; NPV 95% CI computed from the 95% for the negative likelihood ratio.

[4] Includes patient-reported other, mixed, and unknown races.

Table 18: Aptima BV Assay Performance Relative to BV Infection Status in Symptomatic Subjects, by Clinical Condition

Clinical Condition	N	TP	FP	TN	FN	Prev[1] (%)	Sensitivity (95% CI)[2]	Specificity (95% CI)[2]	PPV (95% CI)[3]	NPV (95% CI)[3]
Patient-collected										
Use of antibiotics	3	1	0	2	0	33.3	100 (20.7-100)	100 (34.2-100)	100 (7.8- 100)	100 (45.1- 100)
Use of antifungals	8	2	0	6	0	25.0	100 (34.2-100)	100 (61.0-100)	100 (33.3- 100)	100 (77.4- 100)
Use of estrogen therapy	2	0	0	2	0	0.0	NC	100 (34.2-100)	NC	100 (NC)
Recurrent symptoms of vaginitis in the last 12 months	828	405	62	353	8	49.9	98.1 (96.2-99.0)	85.1 (81.3-88.2)	86.7 (83.9-89.3)	97.8 (95.9-99.0)
Unprotected intercourse in the last 24 hours	94	53	10	30	1	57.4	98.1 (90.2-99.7)	75.0 (59.8-85.8)	84.1 (76.5-91.0)	96.8 (85.6-99.9)
Pregnant	20	9	1	10	0	45.0	100 (70.1-100)	90.9 (62.3-98.4)	90.0 (66.5-99.7)	100 (77.9- 100)
With Menses	109	52	9	48	0	47.7	100 (93.1-100)	84.2 (72.6-91.5)	85.2 (76.6-92.4)	100 (93.6- 100)
Without Menses	1175	579	85	496	15	50.6	97.5 (95.9-98.5)	85.4 (82.3-88.0)	87.2 (84.9-89.3)	97.1 (95.4-98.3)
Post-menopausal	121	42	7	68	4	38.0	91.3 (79.7-96.6)	90.7 (82.0-95.4)	85.7 (75.5-93.2)	94.4 (88.1-98.3)
Clinician-collected										
Use of antibiotics	3	1	0	2	0	33.3	100 (20.7-100)	100 (34.2-100)	100 (7.8- 100)	100 (45.1- 100)
Use of antifungals	8	2	0	6	0	25.0	100 (34.2-100)	100 (61.0-100)	100 (33.3- 100)	100 (77.4- 100)
Use of estrogen therapy	2	0	0	2	0	0.0	NC	100 (34.2-100)	NC	100 (NC)
Recurrent symptoms of vaginitis in the last 12 months	832	394	47	371	20	49.8	95.2 (92.7-96.9)	88.8 (85.4-91.4)	89.3 (86.6-91.8)	94.9 (92.5-96.7)
Unprotected intercourse in the last 24 hours	94	50	6	34	4	57.4	92.6 (82.4-97.1)	85.0 (70.9-92.9)	89.3 (81.2-95.4)	89.5 (78.4-96.6)
Pregnant	20	9	0	11	0	45.0	100 (70.1-100)	100 (74.1-100)	100 (74.2- 100)	100 (78.4- 100)
With Menses	111	50	8	51	2	46.8	96.2 (87.0-98.9)	86.4 (75.5-93.0)	86.2 (77.6-93.1)	96.2 (88.7-99.5)
Without Menses	1177	569	62	520	26	50.6	95.6 (93.7-97.0)	89.3 (86.6-91.6)	90.2 (88.0-92.2)	95.2 (93.3-96.8)
Post-menopausal	125	41	5	72	7	38.4	85.4 (72.8-92.8)	93.5 (85.7-97.2)	89.1 (79.1-95.8)	91.1 (84.7-95.9)

FN = false negative, FP = false positive, NC = not calculable, NPV = negative predictive value, PPV = positive predictive value, Prev = prevalence TP = true positive, TN = true negative

[1] Study prevalence reported.

[2] Score CI.

[3] PPV 95% CI computed from the exact 95% CI for positive likelihood ratio; NPV 95% CI computed from the 95% for the negative likelihood ratio.

Evaluation of the Aptima BV assay in Asymptomatic Women

The Aptima BV assay was evaluated with clinician-collected vaginal swab specimens collected from 172 asymptomatic women. Results from the study are presented in Table 19 below which also includes results for the most prevalent ethnic groups enrolled.

Table 19: Positivity of BV Infection as Determined by the Aptima BV Assay in Asymptomatic Subjects by Race and Ethnicity

Race and Ethnicity	% Positivity (# positive/# tested with valid results)
All	40.7% (70/172)
Asian	40.0% (2/5)
Black/African American	52.0% (39/75)
White (Hispanic/Latino)	43.9% (18/41)
White (Not Hispanic/Latino)	15.9% (7/44)
Other	57.1% (4/7)

3. Clinical Cutoff:

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

4. Expected Values:

The observed expected values (number of positive results detected by the Aptima BV Assay) in this prospective clinical study were 52.0% (735/1413) in clinician-collected vaginal swab samples and 55.1% (774/1405) in patient-collected vaginal swab samples.

M. Instrument Name

Panther system

N. System Descriptions:

1. Modes of Operation:

The Panther instrument provides automation of all assay steps including sample processing, amplification of nucleic acid, detection, data reduction and amplicon inactivation. The "Panther Instrument" refers to the analyzer and associated hardware combined with the software. The "Panther system" refers to the Panther Instrument operated in conjunction with the Aptima BV assay.

The Panther system is a fully automated nucleic acid testing system of diagnostic assays. The two main components of the Panther instrument are the analyzer and the computer

workstation. The analyzer holds all of the fluids, reagents, and consumables needed to perform the assay. The analyzer contains an embedded Central Organization Processor (COP) module and additional control modules for all of the major sub-components of the analyzer. The COP and control modules contain the firmware responsible for controlling the analyzer.

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

Yes ___X___ or No _____

Level of Concern:

Moderate

Software Description:

There are two software components that comprise the Panther System which is required to perform the Aptima BV Assay:

- System Software: System software includes the Master Controller software and instrument firmware. This software is assay independent and does not include assay specific processing parameters. The System software communicates with the Assay software to access assay specific parameters.

- Assay Software: Assay software contains information that is specific to a given assay and provides assay-specific configuration information to the instrument. Assay software includes assay specific parameters for reagent volumes, incubation times, incubation temperatures, and other assay parameters, such as the sequence of steps taken.

3. Specimen Identification:

Patient ID/Sample ID is labeled with a unique barcode, which is tracked by the software

to prevent re-use and track positive sample identification.

4. Specimen Sampling and Handling:

   Not Applicable

5. Calibration:

   The Panther System is calibrated upon installation by Hologic Inc. Field service engineers as well as during preventive maintenance as scheduled.

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. Patient Perspectives:

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

R. Conclusion:

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