Guilford County's General Terms and Conditions

1. This bid package serves as official notice that GUILFORD COUNTY is soliciting and will receive bids for the item(s) and/or service(s) stated on the event cover page and outlined in the Bid specifications. Bids shall be submitted electronically via the Purchasing Department's Strategic Sourcing website: www.co.guilford.nc.us/sourcing **by the event close date and time specified.**

2. All addenda to this bid package will be issued electronically. No oral changes by anyone shall affect this bid package.

3. The official bid price, quote, response for RFP, RFQ, or otherwise instructed; shall be signed by a duly authorized person acknowledging full understanding of the bid information and all addenda. The signature shall be witnessed and the Corporate Seal affixed if a corporation. The exact legal name of the corporation or other entity shall be provided

4. Price quotes shall be net, to include all discounts and delivery charges to GUILFORD COUNTY. In cases of difference between unit price and total price, unit price shall prevail unless otherwise noted.

5. Bid event submittal schedules are fixed and will not be amended unless Guilford County determines the County has given cause to extend the event.

6. Items and services bid are for delivery or completion as soon as possible unless otherwise stated. Delivery or completion dates could therefore be important in making the award.

7. With the exception of construction bids, state and local sales taxes are not to be included in quotes, but they are to be added later to all invoices shown as a separate line item for payment. Federal sales-excise) taxes, where applicable, are to be included in quotes as they are a part of the purchase price. See the construction bid specifications in the event for construction and repair sales tax instructions.

8. All Formal Bids will be publicly opened and recorded at the date and time specified by and in the Purchasing Department. It is GUILFORD COUNTY's policy to announce the award electronically. All other information, except that specifically noted by the Supplier as being of a Confidential nature, becomes public record in accordance with GS 132 and other applicable North Carolina laws. All interested parties are invited to attend any Formal Bid opening.

9. GUILFORD COUNTY will have a period of thirty (30) days, unless otherwise stated, after opening to analyze and award to lowest responsive and responsible bidder based on service, quality, delivery date, performance data and price. The successful supplier shall promptly enter into a contract acceptable to Guilford County.

10. All Events/Bids in the Formal Range require the final approval of the GUILFORD COUNTY Board of Commissioners who normally meet in open session two times each month, the first and third Thursday at 5:30 PM. Everyone is invited to attend those meetings.

11. A Bid Deposit may be required. If this is the case, it will be clearly stated in the Event specifications for each package. If a bid deposit is required, it should be no less than 5% of the total bid in cash, cashier's check, certified check, or a Bid Bond. The checks shall be drawn on a bank or trust company insured by the Federal Deposit Insurance Corporation; and, the bond shall be a corporate surety licensed under the State of North Carolina. The obligee in either check or bond shall be Guilford County.

12. If Bid Deposit Checks are received, they will be returned to all suppliers when the

successful supplier has been selected and the contract awarded by Guilford County. The successful deposit check will be returned when the required contract has been executed.

13. In addition to the bid deposit or bid bond, some supplier's may require a separate Performance Bond and/or laborers-materials men's bond as provided by law in the amount of the contract by the awarded supplier(s). If this is required, it will be clearly stated in the bid specifications. In place of a bond; cash, cashiers check, certified check or government securities shall be acceptable.

14. Guilford County reserves the right to reject any or all bids if in the best interest of the County.

15. In case of default by the Supplier, Guilford County shall retain the Bid Deposit or call upon the Bid Bond surety unless otherwise provided by Law.

16. Guilford County's policy is normally Net 30 days upon completion and acceptance. In the case of some longer term projects, Guilford County may choose to release partial payments to the supplier each month based on 90% of the estimated value of the work completed. The final payment will be released within thirty (30) days or less after the satisfactory completion of all work, its acceptance by Guilford County and the settlement of all other claims and accounts.

17. In the case of Continuing Service Type Contracts, payment will be made monthly or as otherwise agreed upon.

18. It is Guilford County's Purchasing Policy to conduct all purchasing within the North Carolina Laws and Guilford County Purchasing Policy, to provide each supplier/contractor an equal opportunity to participate, and to award on a best value basis. In order to accomplish our policy, we intend to make every supplier/contractor aware of each purchasing opportunity. Contracts shall be awarded to the lowest responsive and responsible bidder(s) based on quality, performance and the time specified in the proposal for the performance of the contract. Suppliers/contractors should register online at www.co.guilford.nc.us/sourcing.

19. A Material Safety Data Sheet (MSDS) shall be furnished to Guilford County for all products purchased that contain hazardous material and/or components.

20. Any supplier/contractor performing work on GUILFORD COUNTY property is required to have adequate Liability and Workers Compensation Insurance that will fully protect GUILFORD COUNTY from any damages to property and/or persons caused by the supplier/contractor.

21. The successful supplier shall be required (and is responsible) to take Affirmative Action to employ Disabled Veterans and Veterans of the Viet Nam era, including listing vacancies with the North Carolina Employment Security Commission, under 42 US Code 4212 and applicable regulations thereafter.

    The successful supplier shall be required to employ in the workforce only those laborers whose employment is consistent with all applicable State and Federal Laws. The successful supplier, and each subcontractor, shall prior to performance of the work receive clear written evidence from each laborer that said laborer may lawfully be employed. Said evidence shall immediately be submitted to the County. Failure of said Supplier or Subcontractor to receive, retain and/or provide to the County such evidence shall constitute a material breach of the Contract with the County.

22. The Supplier shall take Affirmative Action in complying with all Federal and State requirements concerning fair employment without regard to discrimination by reason of race, color, religion, sex, national origin or physical handicap.

23. The successful Supplier is responsible for compliance with all applicable Local, State and Federal Laws, including all state and local permits, licenses and fees.

24. If the Supplier/Contractor should undergo merger, acquisition or any change in their ownership or their name for any reason, the provider shall immediately notify Guilford County in writing of these changes and provide Guilford County with legal documentation supporting these changes, such as an Assumption Agreement, Bill of Sale, Articles of Incorporation, Articles of Amendment, sales contract, merger documents, etc. Further, the Supplier/Contractor shall submit the name and address of their registered agent for Service of Process and/or all notices required under the contract(s). This contract shall not be assumed or otherwise transferred to another party by the Supplier/Contractor without the express written consent of Guilford County, which said consent will be evidenced by acceptance memo, letter or e-mail from the Guilford County Manager, or designee, to the original Supplier/Contractor under the contract and the assuming Supplier/Contractor.

25. Provider shall operate as an independent contractor for all purposes. The Parties agree to each be solely responsible for their own acts or omissions in the performance of each of their individual duties hereunder, and shall be financially and legally responsible for all liabilities, costs, damages, expenses and attorney fees resulting from, or attributable to any and all of their individual acts or omissions to the extent allowable by law.

26. Guilford County and the awarded Vendor shall comply with Equal Employment Opportunities (EEO) requirements, and to take affirmative action to ensure that all individuals have an equal opportunity for employment without regard to race, color, religion, sex, sexual orientation, gender identity, national origin, age, disability, genetic information, or veteran status under the Guilford County EEO Plan, as amended, implemented pursuant to 41 CFR Part 60-2.10(a)(3), 41 CFR §60-741.44(a) and 41 CFR §60-300.44(a), and in accordance with the following laws, as amended: Title VII and Title IX of the Civil Rights Act of 1964; The Equal Pay Act of 1963; Executive Order 11246; the Age Discrimination in Employment Act of 1967; the Rehabilitation Act of 1973, as amended (Section 503); the Americans with Disabilities Act of 1990; the Vietnam Era Veterans' Readjustment Assistance Act of 1974 (VEVRAA); the Civil Rights Restoration Act of 1988; NC General Statutes Chapters 116 and 126 and Title II of the Genetic Information Nondiscrimination Act of 2008, the North Carolina Equal Employment Opportunity Policy effective June 1, 2015, along with all other applicable federal and state laws governing equal employment opportunities.

27. This contract is governed by the Laws of the State of North Carolina.

Guilford County Department of Health & Human Services
Public Health Division
Greensboro and High Point, North Carolina

Request for Proposal
Event #647

Nucleic Acid Application Testing for
N. gonorrhoeae, C trachomatis & Trichomonas

February 2019

Introduction

The Guilford County Department of Health & Human Services - Public Health Division ("HEALTH DEPARTMENT") is seeking proposals for nucleic acid amplification test for the detection of *Chlamydia trachomatis* (CT*)*, *Neisseria gonorrhoeae* (GC) and Trichomonas (TV). The Health Department is seeking a five-year reagent rental agreement. The Health Department has two laboratory sites that are high complexity testing facilities. The department is currently using a system to perform NAAT testing for CT/GC and TV from male and female urine, endocervical, urethral and extragenital collection sites. One is in Greensboro, North Carolina and the other one is in High Point, North Carolina. Our intent is to have all CT and *GC* specimens to be sent to the 1100 E. Wendover, Greensboro site for analysis and as such specimens must be stable for a minimum of 48 - 72hrs. If needed, assistance with validation and training of staff, at no expense to the County, will be required. System selected must accommodate direct tube sampling, have barcode capabilities and must be able to be interfaced with the Harvest Orchard LIS. In addition, remote troubleshooting is a desired feature. A complete list of specifications is provided in the following pages of this proposal.

Projected annual volume of testing is:

CT	20,000 patient samples per year
GC	20,000 patient samples per year
TV	10,000 patient samples per year

Anticipated Event Schedule

Bid opens by	3/1/19
Q&A Opens	3/1/19
Q&A Closes	3/14/19
Bid Closes	3/21/19
Finalize recommendation and submit contract to legal	3/26/19
Agenda Deadline- Department puts agenda item in Legistar and collaborates on final draft w/Purchasing	Completed by 4/3/19
Board Meeting	4/18/19
Contract executed	5/1/19

Contract Award: The contract will be awarded to one supplier who meets all the specification requirements of this bid event. Guilford County reserves the right to reject any and all proposals that is in the best interest of the County. Final selection will be based on the lowest, responsive, responsible bidder considering the quality, performance, and the time specified in proposals for performance of the contract.

Specifications

1. Assay must be a FDA approved nucleic amplification assay kit for the *in vitro* detection of *Chlamydia trachomatis* (CT,) *Neisseria gonorrhoeae* (GC) and Trichomonas (TV) from symptomatic and asymptomatic patients collected using swabs, urine, and liquid-based cytology specimens.
2. Assay must allow dual testing for both CT and GC simultaneously.
3. Assay system must have an automated nucleic acid extraction system specific enough so that naturally occurring components in patient samples such as blood or mucous and man-made components such as vitamins or gynecological products do not interfere in the performance of the assay.
4. System must accommodate direct tube sampling.
5. Assay must be such that, at a minimum, two hundred samples can be tested within a four to six-hour shift.
6. Tests kits/reagents must have a minimum of a two-month expiration date when received in the lab.
7. All reagent and equipment must be provided on a reagent rental basis.
8. The Health Department does not have a dock and inside delivery is required for all deliveries. Equipment/supplies must be such that, once received, can be delivered to the lab located on the second floor.
9. Technical support must be available by phone, Monday through Friday. If on-site in-house troubleshooting does not resolve the problem, on-site technical support should be provided within 24 working hours or if it is determined that equipment has failed, such equipment must be repaired or replaced within 24 working hours of the reported failure.
10. The vendor will assist the laboratory in performing any required method validation that meets CLIA-88 verification requirements. Specimen collection kits will be provided at no charge during the validation process.
11. If requested, vendor must provide on-site training to staff.
12. Contract Term shall be in effect for five (5) years, beginning May 1, 2019 and ending on April 30, 2024. This contract can be cancelled by either party upon a ninety (90) day written notice from either party to the other party. The terms of this contract may only be amended with a written Contract Amendment executed by both parties.

The following items should be addressed in your proposal.

1. Name of system.
2. Is the proposed system a fully automated instrument platform?
3. Describe any user interventions needed.
4. Does the system provide primary tube sampling with no cap removal?
5. Does the system have bi-directional interface capability with the Orchard Harvest LIS system?
6. Is there an interface allowance included in this proposal?
7. Can the system run samples for GC/CT simultaneously or must they be run separately?
8. How does the system account for potential contamination and is a 'clean room' or multiple testing rooms required for performance and utilization? Explain.
9. In the event a contamination event occurs, explain the decontamination process.
10. What specimen types have been cleared by the FDA to run on the system?
11. Is your system approved for use in testing extragenital site testing such as the pharynx or rectum? If not, is this under development?
12. What are the dimensional space requirements (LxWxH) for the instruments and ancillary operating systems? Specify the counter/floor space requirements.
13. What are the ambient temperature and humidity requirements for the system?
14. Describe the electrical and plumbing requirements for your instrument. Is access to a sink/drain required?
15. Does the system have onboard barcode scanning capabilities for sample or reagent identification integrity?
16. Can the system read barcode labels generated by the Orchard Harvest LIS system?
17. Describe the frequency and type of routine/operator maintenance, i.e. daily, weekly, monthly, etc., and the time it takes to complete each task.
18. Is vendor provided maintenance included in this proposal? If yes, describe the frequency, time required, and type of maintenance included.
19. If vendor provided maintenance is not included in the proposal, describe the frequency and cost for each required type of maintenance system.
20. What is the average response time for on-site attention to unscheduled instrument maintenance calls?
21. Provide warranty information for the instrument.
22. What is the system throughput?
23. What is the hands-on time for a batch of 100 specimens?

24. What are the information technology requirements for your system (such as modem or network connection requirements for operation)? If a computer needs to be supplied by the Health Department, what are the operating system requirements?

25. Are the reagents ready to use and in single unit dose?

26. Does the instrument have a mechanism for tracking reagent lot numbers?

27. Does the system alert when reagents or other disposables are getting low?

28. Describe the technical support available for the system.

29. Can vendor support connect remotely to the instrument for problem investigation? What are the requirements for remote connectivity?

30. What are the storage conditions required for your reagents?

31. What are the FDA approved specimen types tested by your system?

32. What are the specimen transport requirements and sample stability for each of the specimen types?
    Endocervical swabs
    Vaginal swabs (Provider/Self-collected)
    Male urethral swabs
    Male urine
    Female urine
    Male Urine Preservative Transport device
    Female Urine Preservative Transport device
    Liquid-base Cytology media

33. Are there any sample manipulations needed? If yes, explain.

34. Is there an internal control for the assay?

35. Provide a list of other FDA approved assays that can be run on the instrumentation.

36. Describe the sensitivity, specificity, and predictive values for each assay as compared to a patient infected standard. Which test(s) were used as comparators?

37. What are the QC requirements for each assay?

38. Are QC-acceptance and rejection criteria built in to the system?

39. Can the instrument retransmit or reprint results?

40. Is there any clinically relevant cross reactivity? At what level? Describe.

41. List the type, duration, cost (if any), and number of staff that will be offered clinical training on-site.

42. List the type, duration, cost (if any), and number of staff that will be offered clinical training off-site.

43. Based on an estimated volume of 20,000 GC/CT samples and 10,000 TV samples annually, provide an itemized cost for each reagent/supply needed to perform the GC/CT and TV testing for one year. In addition to the itemized list, provide an estimated annual cost for performing the 20,000 CT/GC and 10,000 TV samples.

44. Provide an itemized list of supplies/items that are not included in this proposal but are required. If supplies need to be purchased from specific vendors, list vendors and estimated quantity and cost to perform 20,000 CT/GC and 10,000 TV samples annually.

45. Provide an itemized list of supplies/items that are included at no charge in the performance of the requested testing.

46. Indicate cost per sample tested for GC, CT and TV.

47. How are supplies typically shipped? Does cost of reagent/supplies include shipping? If not, how are shipping charges handled and what are the estimated shipping charges per delivery?

48. From the time an order is placed for supplies, what is the average time for supplies/reagents to arrive?

49. What is the average and median outdate from the time of manufacture of the kits and reagents?

50. Describe the installation procedures and estimated time of installation from the time the RFP is awarded.

51. Explain assistance provided during the method validation process. Are supplies provided at no cost during the validation process and will validation samples be provided?

52. Is your system capable of testing for pathogens other than GC/CT/TV? If yes, list. Include any that are under development and expected release date.

53. Describe in detail, other value-added services available to the Health Department but not necessarily addressed. Supplier may offer additional products or services. The Health Department shall determine which value-added service options shall be most beneficial from both a cost and service standpoint, and may further negotiate these options to include or omit dependent on the needs of the Health Department.

54. Provide a current user reference list of users. Include local and state Health Department lab users in the list.

Nucleic Acid Application Testing Evaluation

Evaluation Criteria	Max. Score	Score	Comments Supporting Score
1. Pricing: Cost for performing samples annually. CT 20,000 patient samples per year - 20 pts. GC 20,000 patient samples per year - 20 pts. TV 10,000 patient samples per year - 10 pts. Total Max Score 50 pts.	50		
2. Qualifications Ability to meet the current and requirements specifications.	25		
3. Experience Experience in providing a system to perform NAAT testing for high complexity testing sites and strength of current references.	25		
Evaluator: Date:	100		

Bid Submittal Instructions

1. **All suppliers, who plan to submit a bid must register as a supplier in the Guilford County eProcurement Supplier Portal.**

2. Through the Supplier Portal, suppliers may browse to the open event or search for an event name or number.

3. **Suppliers sign in to begin submitting their response to an event. Click on the event to open it, then click on respond now and follow the instructions for each tab.**

4. All bids must be submitted electronically via the following website www.guilfordcountync.gov/sourcing by the event date and close time. There will be no exceptions.

5. All questions related to this event must be submitted online using the Question & Answer Forum from **Event #647 Friday, March 1, 2019 at 3:00pm through Thursday, March 14, 2019 at 3:00 pm.** Each question asked will be answered online for any/all vendors to view. No questions will be considered after the Q&A close date and time. The system cannot accept late submittals. **No exceptions.**

6. All official County responses to prospective Bidder/Supplier's questions and official addenda regarding bid document revisions or additional information will be communicated by the County through the Lawson Strategic Sourcing Q/A function tab within this event.

7. To complete the lines portion of a submittal in the Strategic Sourcing Event, open the lines tab to enter pricing for each line.

8. Use the provided line description and quantity for each line to complete the entries for each line. Upload all additional documentation required in the bid document as an attachment or attachments.

9. Suppliers are responsible for checking the event for any addendums prior to completion and submission of their response.

10. To complete an electronic submittal, be sure to click the "Submit" button. Clicking the "Done" button puts the response into a draft state that can be completed at a later time, but will not be part of the submitted responses until submitted via the "Submit" button.

11. Suppliers are strongly encouraged to submit their bids with all required documentation at least 24 hours in advance. Guilford County will not be responsible for any technical difficulties that may occur and result in the inability to submit.

12. For questions related to the Minority and Women Business Enterprise Program contact Cynthia Barnes-Phipps at 336-641-4565 or cbarnes1@guilfordcountync.gov.

13. For technical assistance to register as a supplier or submit a bid in the Guilford County Electronic Procurement System, please contact the buyer listed on this event. For immediate assistance, please contact the Purchasing Department at 336-641-3314 and ask to speak to an available Buyer.

Guilford County Department of Health & Human Services
Public Health Division
Greensboro and High Point, North Carolina

Request for Proposal
Event #647

Nucleic Acid Application Testing for N. gonorrhoeae, C trachomatis & Trichomonas

February 2019

Introduction

The Guilford County Department of Health & Human Services - Public Health Division ("HEALTH DEPARTMENT") is seeking proposals for nucleic acid amplification test for the detection of *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae* (GC) and Trichomonas (TV). The Health Department is seeking a five-year reagent rental agreement. The Health Department has two laboratory sites that are high complexity testing facilities. The department is currently using a system to perform NAAT testing for CT/GC and TV from male and female urine, endocervical, urethral and extragenital collection sites. One is in Greensboro, North Carolina and the other one is in High Point, North Carolina. Our intent is to have all CT and *GC* specimens to be sent to the 1100 E. Wendover, Greensboro site for analysis and as such specimens must be stable for a minimum of 48 - 72hrs. If needed, assistance with validation and training of staff, at no expense to the County, will be required. System selected must accommodate direct tube sampling, have barcode capabilities and must be able to be interfaced with the Harvest Orchard LIS. In addition, remote troubleshooting is a desired feature. A complete list of specifications is provided in the following pages of this proposal.

Projected annual volume of testing is:

CT 20,000 patient samples per year
GC 20,000 patient samples per year TV
10,000 patient samples per year

Anticipated Event Schedule

Bid opens by	3/1/19
Q&A Opens	3/1/19
Q&A Closes	3/14/19
Bid Closes	3/21/19
Finalize recommendation and submit contract to legal	3/26/19
Agenda Deadline- Department puts agenda item in Legistar and collaborates on final draft w/Purchasing	Completed by 4/3/19
Board Meeting	4/18/19
Contract executed	5/1/19

Contract Award: The contract will be awarded to one supplier who meets all the specification requirements of this bid event. Guilford County reserves the right to reject any and all proposals that is in the best interest of the County. Final selection will be based on the lowest, responsive, responsible bidder considering the quality, performance, and the time specified in proposals for performance of the contract.

Specifications

1. Assay must be a FDA approved nucleic amplification assay kit for the *in vitro* detection of *Chlamydia trachomatis* (CT,) *Neisseria gonorrhoeae* (GC) and Trichomonas (TV) from symptomatic and asymptomatic patients collected using swabs, urine, and liquid-based cytology specimens.

   Hologic's Aptima Combo 2 Assay for the detection of CT/GC may be used to test the following specimens from symptomatic and asymptomatic individuals: clinician-collected endocervical, vaginal and male urethral swab specimens, clinician-collected gynecological specimens collected in the PreservCyt® Solution, patient-collected vaginal swab specimens, and female and male urine specimens.

   Hologic's Aptima Trichomonas Assay may be used to test the following specimens from symptomatic or asymptomatic women: clinician-collected endocervical swabs, clinician-collected vaginal swabs, and specimens collected in PreservCyt Solution.

2. Assay must allow dual testing for both CT and GC simultaneously.

   Yes, Hologic's Aptima Combo 2 Assay meets this specification. The Aptima Combo 2 Assay is a target amplification nucleic acid probe test that utilizes target capture for the *in vitro* qualitative detection and differentiation of ribosomal RNA (rRNA) from *Chlamydia trachomatis* (CT) and/or *Neisseria gonorrhoeae* (GC) to aid in the diagnosis of chlamydial and/or gonococcal urogenital disease using the Panther System as specified.

3. Assay system must have an automated nucleic acid extraction system specific enough so that naturally occurring components in patient samples such as blood or mucous and man-made components such as vitamins or gynecological products do not interfere in the performance of the assay.

   Yes, Hologic's Panther system meets this specification. Blood interference was evaluated on the Panther System and the results of this testing indicated that blood does not interfere with Aptima Combo 2 Assay performance.

   The Aptima Combo 2 Assay performance in the presence of potentially interfering substances was tested on DTS Systems. The following interfering substances were individually spiked into swab and PreservCyt Solution liquid Pap specimens: 10% blood, contraceptive jelly, spermicide, moisturizer, hemorrhoidal anesthetic, body oil, powder, anti-fungal cream, vaginal lubricants, feminine spray, and leukocytes (1.0×10^6 cells/mL). All were tested for potential assay interference in the absence and presence of CT and GC at the estimated rRNA equivalent of 1.0 CT IFU/assay (5 fg/assay) and 50 GC cells/assay (250 fg/assay). The rRNA equivalents were calculated based on the genome size and estimated DNA:RNA ratio/cell of each organism.

   No interference was observed with any of the tested substances. No inhibitors of amplification were observed in the Aptima Combo 2 Assay. See Package Insert for more detail.

The Aptima Combo 2 Assay combines the technologies of target capture, TMA, and DKA.

Specimens are collected and transferred into their respective specimen transport tubes. The transport solutions in these tubes release the rRNA targets and protect them from degradation during storage. When the Aptima Combo 2 Assay is performed in the laboratory, the target rRNA molecules are isolated from specimens by use of capture oligomers via target capture that utilizes magnetic microparticles. The capture oligomers contain sequences complementary to specific regions of the target molecules as well as a string of deoxyadenosine residues. A separate capture oligomer is used for each target. During the hybridization step, the sequence specific regions of the capture oligomers bind to specific regions of the target molecules. The capture oligomer: target complex is then captured out of solution by decreasing the temperature of the reaction to room temperature. This temperature reduction allows hybridization to occur between the deoxyadenosine region on the capture oligomer and the poly-deoxythymidine molecules that are covalently attached to the magnetic particles. The microparticles, including the captured target molecules bound to them, are pulled to the side of the reaction vessel using magnets and the supernatant is aspirated. The particles are washed to remove residual specimen matrix that may contain amplification reaction inhibitors. After the target capture steps are completed, the specimens are ready for amplification.

4. System must accommodate direct tube sampling. Yes, the Panther system uses direct tube sampling.
5. Assay must be such that, at a minimum, two hundred samples can be tested within a four to six-hour shift. The Panther system has throughput of 275 samples with both a CT and GC result, within one eight hour shift using only one person with total hands on time of less than 25 minutes. 275 samples in 8 hours, 500 samples in 12 hours. See Exhibit C – Slide for more detail.
6. Tests kits/reagents must have a minimum of a two-month expiration date when received in the lab. Yes, Hologic assays meet this specification.
7. All reagent and equipment must be provided on a reagent rental basis. Yes, Hologic will provide the equipment and reagents on a reagent purchase and equipment rental basis.
8. The Health Department does not have a dock and inside delivery is required for all deliveries. Equipment/supplies must be such that, once received, can be delivered to the lab located on the second floor. Acknowledged.
9. Technical support must be available by phone, Monday through Friday. If on-site in-house troubleshooting does not resolve the problem, on-site technical support should be provided within 24 working hours or if it is determined that equipment has failed, such equipment must be repaired or replaced within 24 working hours of the reported failure. Hologic offers standard service at no additional cost for the term of the agreement with a reagent rental. Hologic's standard service includes preventative maintenance and access to Hologic's Technical Support telephone support, Monday through Friday, 5:00 a.m. to 5:00 p.m. Pacific Standard Time

(excluding Hologic holidays) by calling Technical Support at 888-484-4747. Additional/enhanced service options are also available. Please refer to Hologic's Panther Service Feature Sheet for options and rates. Hologic also offers Pro360°, a remote service management tool, at no extra charge. Pro360° allows for enhanced troubleshooting by allowing a Technical Support representative to log into the Customer's network remotely to troubleshoot and diagnose potential system problems quickly and accurately. If the Customer opts for Pro360°, a representative will be on site within 24 hours (Monday – Friday) if PRO360° Remote Diagnostics Management is installed. If Pro360 is not installed, a representative will be on site within 48 hours (Monday – Friday).

10. The vendor will assist the laboratory in performing any required method validation that meets CLIA-88 verification requirements. Specimen collection kits will be provided at no charge during the validation process. Hologic will provide enough supplies (tips, collection devices, and reagents) at no charge during the verification process.
11. If requested, vendor must provide on-site training to staff. Yes, Hologic will provide on-site training to staff upon request.
12. Contract Term shall be in effect for five (5) years, beginning May 1, 2019 and ending on April 30, 2024. This contract can be cancelled by either party upon a ninety (90) day written notice from either party to the other party. The terms of this contract may only be amended with a written Contract Amendment executed by both parties. Acknowledged.

The following items should be addressed in your proposal.

1. Name of system. Panther system
2. Is the proposed system a fully automated instrument platform? Yes, the Panther System is a fully automated system.
3. Describe any user interventions needed. The Panther system is created to be a "sample-in-result-out" instrument and eliminates batch processing and automates all aspects of nucleic acid testing on a single, integrated platform. As a feature, the Panther instrument also offers random access and full automation for true walkaway freedom.
4. Does the system provide primary tube sampling with no cap removal? Yes.
5. Does the system have bi-directional interface capability with the Orchard Harvest LIS system? The Panther system has been successfully interfaced with a wide variety of laboratory information systems including but not limited to systems offered from Orchard. Interface documentation is available for the lab IT contact and the LIS contact/vendor. At this time, the Panther system is already interfaced with Orchard Harvest LIS at the Guilford County lab.
6. Is there an interface allowance included in this proposal?
   There is currently one (1) Panther system installed and operational at the Guilford County lab that is interfaced with an LIS. If another Panther system is installed, Hologic will reimburse LIS vendor for the expenses associated with the procurement of an LIS interface for the System, up to $10,000 per unit upon receipt of an invoice from LIS vendor.
7. Can the system run samples for GC/CT simultaneously or must they be run separately? The Panther can run samples simultaneously. The Aptima Combo combines both GC/CT in one test.
8. How does the system account for potential contamination and is a 'clean room' or multiple testing rooms required for performance and utilization? Explain. There is no clean room or multiple rooms required for the Panther System.
   - The cotton on penetrable caps prevents splashing mitigating sample handling issues.
   - Use of Buffered Bleach during regularly scheduled maintenance deactivates amplified material.
   - The Panther instrument is a "Closed system" further mitigating the opportunity for contamination.
9. In the event a contamination event occurs, explain the decontamination process.

   There are many laboratory-specific factors that may contribute to contamination, including testing volume, workflow, disease prevalence and various other laboratory activities. These factors should be taken into consideration when contamination monitoring frequency is being established. Intervals for contamination monitoring should be established based on each laboratory's practices and procedures.

   The lab is directed to contact Hologic Molecular Technical Support in the case of a contamination event. The following general principles are considered in the event of contamination:

- Confirm contamination,
- Identify extent of contamination,
- Identify and isolate (if possible) the source of the contamination,
- Decontaminate the lab and address source of contamination,
- Perform protocols to ensure contamination has been resolved
- Retrain laboratory personnel if indicated,
- Monitor for recurrence based on Lab Contamination Monitoring Protocol for the Panther System, if indicated.

10. What specimen types have been cleared by the FDA to run on the system?
The Aptima Combo 2 Assay may be used to test the following specimens from symptomatic and asymptomatic individuals: clinician-collected endocervical, vaginal and male urethral swab specimens, clinician-collected gynecological specimens collected in the PreservCyt® Solution, patient-collected vaginal swab specimens, and female and male urine specimens.

Hologic's Aptima Trichomonas Assay may be used to test the following specimens from symptomatic or asymptomatic women: clinician-collected endocervical swabs, clinician-collected vaginal swabs, and specimens collected in PreservCyt Solution

11. Is your system approved for use in testing extragenital site testing such as the pharynx or rectum? If not, is this under development?

Hologic recognizes the importance of extragenital site testing for CT/GC. Extragenital site testing claims for the Aptima Combo 2 assay are currently under review with FDA. These claims are considered under development until cleared by FDA.

12. What are the dimensional space requirements (LxWxH) for the instruments and ancillary operating systems? Specify the counter/floor space requirements.

The Panther instrument is a stand-alone instrument that requires the space listed below.

Component Dimensions (w x d x h)
Panther System . . . . . 122 cm (48 in) x 81.5 cm (32 in) x 175 cm (69 in)
UPS (Optional) 21.4 cm (8.4in) x 41.0 cm (16.1in) x 32.5 cm (12.8in)

Weight
Panther System . . . . . 363 kg (800 lbs)
UPS (Optional) .....34.5 kg (76 lbs)

13. What are the ambient temperature and humidity requirements for the system?

Environmental Requirements
Ambient Temperature . . . . . 15 - 30°C
Relative Humidity . . . . . 20 - 85%

14. Describe the electrical and plumbing requirements for your instrument. Is access to a sink/drain required?

Yes, access to one (1) sink is required.

Electrical Requirements
Electrical Input . . . . . 100-230 +/- 10% vac , 50-60Hz, single phase
Current Input . . . . . Minimum of 15 amp circuit (dedicated)
20 amp circuit (dedicated) if used with optional UPS
Current Draw . . . . . Instrument and ancillaries draw 400 watts in power-save mode to 1400 watts peak
- 100 vac circuit draws 14 amps
- 230 vac circuit draws 6 amps

System Output
Heat Dissipation . . . . . 550 watts (1878 btu/hour) during steady state
Drain Output . . . . . None: waste fluids drain to removable containers

15. Does the system have onboard barcode scanning capabilities for sample or reagent identification integrity? Yes.

16. Can the system read barcode labels generated by the Orchard Harvest LIS system?

    Yes, the Panther can read the following standard barcode types: Code 39, Code 93, Code 128 (ISBT 128), Interleaved 2 of 5, and Codabar.

17. Describe the frequency and type of routine/operator maintenance, i.e. daily, weekly, monthly, etc., and the time it takes to complete each task.

    There are no daily maintenance tasks required for the system. Up to four maintenance tasks need to be completed weekly: PC Reboot, Clean Sample Shield, Mag Wash Clean and replacing the cleaning solution bottle. All but one, the Mag Wash Clean, can be completed with less than one minute of hands-on time. The PC Reboot is completed automatically by the system after initiating the maintenance task through the software and the two others involve the simple replacement of single items onboard. The Mag Wash clean can be scheduled to run automatically at a time that is best for the site. Scheduling the task in this way means that no time is taken away from operators or system processing time. The single required monthly task, Monthly Maintenance cleaning, is estimated to take one operator 30 minutes to complete.

18. Is vendor provided maintenance included in this proposal? If yes, describe the frequency, time required, and type of maintenance included. Yes, preventative maintenance is provided according to Operator's manual and is included at no additional cost with a standard service on a reagent rental.

19. If vendor provided maintenance is not included in the proposal, describe the frequency and cost for each required type of maintenance system. N/A. See answer to Specification #18.

20. What is the average response time for on-site attention to unscheduled instrument maintenance calls?

    Representative on site within 24 hours (Monday – Friday) if PRO360° Remote Diagnostics Management is installed. Representative on site within 48 hours (Monday – Friday) if PRO360° Remote Diagnostics is not installed. Service response times are predicated upon the Equipment operator being willing and able to transfer Equipment log files to Hologic when

instructed by Hologic Technical Support using the protocol described in the Equipment Operator's Manual. Please refer to Exhibit B – Description of Services for additional details.

21. Provide warranty information for the instrument.

Hologic Gen-Probe warrants that the Reagents shall meet the required performance specifications to perform the desired tests as described in the Package Inserts. The extent of Hologic Gen-Probe's liability under this warranty is limited to replacing any defective Reagent. Hologic Gen-Probe does not manufacture the System. The System is warranted through manufacturers as described in the Operator's Manual provided to Customer and such warranties extend to Hologic Gen-Probe's customers. Hologic Gen-Probe may, at its option, repair or replace any defective System. The foregoing warranty shall not apply in the event that: (a) Customer has not used and maintained the System in accordance with the guidelines set forth in the Operator's Manual provided to Customer; (b) if Customer has used the System with reagents and supplies not expressly authorized by Hologic Gen-Probe; (c) if the System is repaired or altered by a party other than Hologic Gen-Probe without Hologic Gen-Probe's prior written approval; or (d) if the System has been subject to misuse, negligence, or accident.

22. What is the system throughput? **TMA Assays**: 3.5 hours to first result; up to 275 samples processed in 8 hrs. **RT-TMA Assays**: 2.7 hours to first result, up to 320 samples processed in 8 hours, up to 750 samples processed in 15.2 hours.

23. What is the hands-on time for a batch of 100 specimens? See Exhibit D

24. What are the information technology requirements for your system (such as modem or network connection requirements for operation)? If a computer needs to be supplied by the Health Department, what are the operating system requirements? The Health Department is not required to supply a computer. If the lab chooses to run LIS and Remote Diagnostic Pro360, a network connection is needed. At this time, the Lab is already connected to these two services.

25. Are the reagents ready to use and in single unit dose? Lyophilized reagents come prepared to mix with liquid with minimal hands on time.

26. Does the instrument have a mechanism for tracking reagent lot numbers? Yes, the Panther System uses RFID technology that automatically tracks all reagent lot numbers.

27. Does the system alert when reagents or other disposables are getting low? Yes.

28. Describe the technical support available for the system. Hologic has live technical support associates available M-F 5 a.m. – 5 p.m. Pacific Standard Time (excluding Hologic holidays). Additional service options are available. Please refer to the Panther Service Feature Sheet enclosed for rates and details.

29. Can vendor support connect remotely to the instrument for problem investigation? What are the requirements for remote connectivity? Yes. The Pro360 remote service management tool allows for enhanced troubleshooting by Hologic Technical Support personnel and is highly recommended. Pro360 is provided with the Panther at no additional cost.

30. What are the storage conditions required for your reagents?

Aptima Combo 2 Assay (CT/GC)
Reagent Storage and Handling Requirements

A. The following reagents are stable when stored at 2°C to 8°C (refrigerated):
Aptima Combo 2 Amplification Reagent
Aptima Combo 2 Enzyme Reagent
Aptima Combo 2 Probe Reagent
Aptima Combo 2 Target Capture Reagent B
Aptima Positive Control, CT / Negative Control, GC
Aptima Positive Control, GC / Negative Control, CT
B. The following reagents are stable when stored at 2°C to 30°C:
Aptima Combo 2 Amplification Reconstitution Solution
Aptima Combo 2 Enzyme Reconstitution Solution
Aptima Combo 2 Probe Reconstitution Solution
Aptima Combo 2 Selection Reagent
C. The following reagents are stable when stored at 15°C to 30°C (room temperature):
Target Capture Reagent
Aptima Wash Solution
Aptima Buffer for Deactivation Fluid
Aptima Oil Reagent
D. Working Target Capture Reagent (wTCR) is stable for 30 days when stored at 15°C to 30°C. Do not refrigerate.
E. After reconstitution, the Enzyme Reagent, Amplification Reagent, and Probe Reagent are stable for 30 days when stored at 2°C to 8°C.
F. Discard any unused reconstituted reagents and wTCR after 30 days or after the Master Lot expiration date, whichever comes first.
G. Controls are stable until the date indicated on the vials.
H. Reagents stored on-board the Panther System have 72 hours of on-board stability.
I. The Probe Reagent and Reconstituted Probe Reagent are photosensitive. Store the reagents protected from light. The specified reconstituted stability is based on 12 hours exposure of the Reconstituted Probe Reagent to two 60W fluorescent bulbs, at a distance of 17 inches (43 cm), and temperature less than 30°C. Light exposure of the Reconstituted Probe Reagent should be limited accordingly.
J. Upon warming to room temperature, some control tubes may appear cloudy or contain precipitates. Cloudiness or precipitation associated with controls does not affect control performance. The controls may be used whether they are clear or cloudy/precipitated. If clear controls are desired, solubilization may be expedited by incubating them at the upper end of the room temperature range (15°C to 30°C).
K. Do not freeze the reagents.

Aptima Trichomonas Vaginalis
Reagent Storage and Handling Requirements

A. The following reagents are stable when stored at 2ºC to 8ºC:
- Aptima *Trichomonas vaginalis* Amplification Reagent
- Aptima *Trichomonas vaginalis* Enzyme Reagent
- Aptima *Trichomonas vaginalis* Probe Reagent
- Aptima *Trichomonas vaginalis* Assay Target Capture Reagent B
- Aptima *Trichomonas vaginalis* Controls B. T

B. The following reagents are stable when stored at room temperature (15°C to 30°C):
- Aptima *Trichomonas vaginalis* Amplification Reconstitution Solution
- Aptima *Trichomonas vaginalis* Enzyme Reconstitution Solution
- Aptima *Trichomonas vaginalis* Probe Reconstitution Solution
- Aptima *Trichomonas vaginalis* Target Capture Reagent

Aptima *Trichomonas vaginalis* Selection Reagent C.

C. After reconstitution, Amplification Reagent, Enzyme Reagent, and Probe Reagent are stable for 60 days when stored at 2°C to 8°C.
D. Working Target Capture Reagent (wTCR) is stable for 60 days when stored at 15°C to 30°C. Do not refrigerate.
E. Discard any unused reconstituted reagents and wTCR after 60 days, or after the Master Lot expiration date, whichever comes first.
F. Controls are stable until the date indicated on the vials.
G. Reagents stored on-board the Panther System have 72 hours of on-board stability.
H. Avoid cross-contamination during reagent handling and storage. Recap all reconstituted reagents with new reagent caps each time prior to storage.
I. The Probe Reagent and Reconstituted Probe Reagent are photosensitive. Store the reagents protected from light.
J. **Do not freeze reagents**.

31. What are the FDA approved specimen types tested by your system?
Hologic's Aptima Combo 2 Assay for the detection of CT/GC may be used to test the following specimens from symptomatic and asymptomatic individuals: clinician-collected endocervical, vaginal and male urethral swab specimens, clinician-collected gynecological specimens collected in the PreservCyt® Solution, patient-collected vaginal swab specimens, and female and male urine specimens.

Hologic's Aptima Trichomonas Assay may be used to test the following specimens from symptomatic or asymptomatic women: clinician-collected endocervical swabs, clinician-collected vaginal swabs, and specimens collected in PreservCyt Solution.

32. What are the specimen transport requirements and sample stability for each of the specimen types?

Endocervical swabs
Vaginal swabs (Provider/Self-collected)
Male urethral swabs
Male urine
Female urine
Male Urine Preservative Transport device
Female Urine Preservative Transport device
Liquid-base Cytology media

Aptima Combo 2 Assay (CT/GC)

A. Endocervical Swab Specimens
Data to support the recommended shipping and storage conditions for endocervical swab samples were generated with pooled negative swab samples. Five pooled samples were spiked with CT and GC at final concentrations of 10 IFU and 100 CFU per reaction, respectively. The spiked samples were held at -70°C, -20°C, 4°C, and 30°C. Samples were tested in duplicate at days 0, 20, 35, 60, and 90. All test conditions were positive for both CT and GC at all times and temperatures.

B. PreservCyt Solution Liquid Pap Specimens
Data to support the recommended shipping and storage conditions for PreservCyt Solution liquid Pap samples were generated with pooled negative PreservCyt Solution liquid Pap

samples. Four pooled samples were spiked with CT and GC at final concentrations of 10 IFU and 100 CFU per reaction, respectively. The PreservCyt Solution liquid Pap samples were placed at 30°C for 7 days, after which 1.0 mL of the sample was added to an Aptima Transfer Tube. The spiked samples were held at 4°C, 10°C and 30°C. Samples stored at 4°C and 10°C were tested in duplicate at days 0, 6, 13, 26, 30 and 36. Samples stored at 30°C were tested in duplicate at days 0, 5, 8, 14 and 17. Four spiked PreservCyt Solution liquid Pap sample pools were added to Aptima Transfer Tubes and placed at 30°C for 14 days before being stored at either -20°C or -70°C. The -20°C samples and the -70°C samples were tested in duplicate after 0, 30, 60, 90 and 106 days of storage. All test conditions were positive for both CT and GC at all times and temperatures.

C. Vaginal Swab Specimens
Data to support the recommended shipping and storage conditions for vaginal swab samples were generated with pooled negative swab samples. Fifteen vaginal swab pools were spiked with CT and GC at final concentrations of 1.0 IFU and 50 CFU per reaction, respectively. The spiked samples were held at -70°C, -20°C, 4°C, and 30°C. Samples were tested using one aliquot at days 0, 20, 36, 73, and 114. All test conditions were positive for both CT and GC at all times and temperatures.

D. Urine Specimens
Data to support the recommended shipping and storage conditions for urine samples were generated with ten female and ten male negative urine samples. The urine samples were spiked with CT and GC at final concentrations of 10 IFU and 100 IFU per reaction, respectively. Two sets of the spiked urine samples were held at 4°C and 30°C for 24 hours prior to being added to the Urine Transport Media (UTM). The two sets of UTM samples then were held at 4°C and 30°C, and tested in triplicate at days 0, 1, 5, 20, and 35. All samples were positive for both CT and GC when the urine samples were held at 4°C prior to addition of the UTM. When the urine samples were held at 30°C prior to addition of the UTM, all of the samples were positive for CT and 95% of the samples were positive for GC at Day 35. These same samples were tested after 116 days of storage at -20°C and -70°C. All samples were positive for both CT and GC under both storage conditions.

E. Additional Frozen (at -20°C) Specimen Stability Study
Data to support the recommended storage condition at -20°C for endocervical swab, urethral swab, vaginal swab, and PreservCyt Solution liquid Pap specimens were generated using 90 specimens for each type with negative result, where 30 specimens were spiked with CT and GC at 1.0 IFU and 50 CFU per reaction, respectively; 30 specimens were spiked at 0.1 IFU and 5 CFU per reaction, respectively; and 30 specimens were unspiked. The specimens were stored at -20°C and were tested at days 0, 200, and 400 days.

Aptima Trichomonas Assay
PreservCyt liquid Pap specimens were generated with negative clinical specimens spiked with *T. vaginalis*. Greater than 98% positivity was observed in all matrices (vaginal swab and PreservCyt liquid Pap) at all times and temperatures tested confirming the validity of the maximum storage times and temperatures described in *Specimen Collection and Storage*.

33. Are there any sample manipulations needed? If yes, explain. There are no sample manipulations required for the CT/GC or ATV assay.

34. Is there an internal control for the assay?

An inhibition control is not necessary for the Aptima Combo 2 Assay when the assay is run according to the package insert. Various documentation, referenced upon request, demonstrates that the inhibition rate in the Aptima Combo 2 Assay is negligible and an internal control is not necessary. An inhibition control is also not necessary for the ATV assay.

35. Provide a list of other FDA approved assays that can be run on the instrumentation.

Aptima Combo 2 Assay (CT/GC)
Aptima HPV Assay
Aptima HPV 16 18/45 Assay
Aptima Trichomonas Assay
Aptima Zika Virus Assay (EUA*)
Aptima HSV 1 & 2 Assay
Aptima HIV-1 Quant Assay
Aptima HCV Quant Assay
Aptima HBV Quant Assay
Aptima Mycoplasma genitalium Assay
In addition, Hologic now offers the Panther Fusion which can be an upgrade to the lab's existing Panther instrument. The Panther Fusion's menu includes the following assays:
Flu A/B/RSV
Paraflu
AdV/hMPV/RV
GBS

The Panther Fusion also includes Open Access™. Open Access is functionality to run lab developed tests.

36. Describe the sensitivity, specificity, and predictive values for each assay as compared to a patient infected standard. Which test(s) were used as comparators?

The prevalence of CT and GC in patient populations depends on risk factors such as age, gender, the presence or absence of symptoms, the type of clinic, and the sensitivity of the test used to detect infections. A summary of the prevalence of three CT and GC disease outcomes, as determined by the Aptima Combo 2 Assay on the Panther System in the clinical trial, is shown in Tables 1, 2, and 3 by specimen type and clinical site of the Package Insert, pages 21 – 39.

Estimates of the prevalence of *T. vaginalis* in different populations depend on the sensitivity of the test in detecting the infection and on patient risk factors such as age, lifestyle, and the presence or absence of symptoms. A summary of the prevalence of *T. vaginalis*, by specimen type, as determined by the Aptima Trichomonas vaginalis Assay during the Panther System clinical study is shown in Table 1 on page 17 of the Package Insert. More detailed information regarding sensitivity, specificity, and prevalence values can be found in Tables 2-5 on pages 18-22 of the Package Insert.

37. What are the QC requirements for each assay? Please refer to Package Insert pages 16 – 18 for Aptima Combo 2 and pages 14-16 for Aptima Trichomonas.

38. Are QC-acceptance and rejection criteria built in to the system? Yes.

39. Can the instrument retransmit or reprint results? Yes, it can print and also transmit electronically.

40. Is there any clinically relevant cross reactivity? At what level? Describe.
No, The Aptima Combo 2 assay on the Panther System does not have any clinically significant cross reactivity. Analytical Sensitivity and Specificity and the list of organisms tested can be found in the Package Insert, Pages 40-43. For the Aptima Trichomonas Assay, Analytical Sensitivity and Specificity can be found on pages 25-27 of the Package Insert.

41. List the type, duration, cost (if any), and number of staff that will be offered clinical training on-site. There is no charge for onsite training—typically we like to train two (2) operators on the Panther system.

42. List the type, duration, cost (if any), and number of staff that will be offered clinical training off- site. Hologic provides a three (3) day off-site training for two (2) key operators, per testing instrument, at Hologic's training facility to include roundtrip airfare, ground transportation, hotel accommodations and meals.

43. Based on an estimated volume of 20,000 GC/CT samples and 10,000 TV samples annually, provide an itemized cost for each reagent/supply needed to perform the GC/CT and TV testing for one year. In addition to the itemized list, provide an estimated annual cost for performing the 20,000 CT/GC and 10,000 TV samples.

Product Number	Product Description	Price Per Kit	Price Per Test	No. of Samples	Total
302923	Aptima Combo 2, 100-Test Kit, Panther	$725.00	$7.25	20,000	$145,000.00
303536	Aptima Trichomonas vaginalis Assay	$600.00	$6.00	10,000	$60,000.00
301041*	Kit,APTIMA COMBO 2 Swab Spec Coll	$62.50	$1.25	10,000	$12,500.00
301040*	Kit,APTIMA COMBO 2 Urine Spec Coll	$62.50	$1.25	10,000	$12,500.00
				Estimated Annual Cost:	$230,000.00

**Items 301040 & 301041 will have an aggregate total volume of 20,000. Only one device is needed per CT/GC test at $1.25 per device. Individual volume will depend on ordering pattern.*

The price per test for CT/GC (Aptima Combo 2) is $7.25. One collection device is needed per test at a cost of $1.25 per test for a combined price per test of $8.50 per CT/GC test. The price per test of a TV test is $6.00. Please read our response to #44 below for information on TECAN tips. The estimated annual cost to run 20,000 GC/CT samples and 10,000 TV samples is $230,000.

In the Guilford County eProcurement Supplier portal, we split the $7.25 price per test by Line Item ($3.62 for CT and $3.63 for GC) due to portal set up of individual tests. However, Hologic's Aptima Combo 2 detects both CT and GC combined.

44. Provide an itemized list of supplies/items that are not included in this proposal but are required. If supplies need to be purchased from specific vendors, list vendors and estimated quantity and cost to perform 20,000 CT/GC and 10,000 TV samples annually.

TECAN tip (catalog # 10612513) are the only tips that Hologic Gen-Probe has validated for use on the Panther System ("System"). Hologic Gen-Probe does not support the use of non-TECAN tips on the Panther system as stated in the System Operator's Manual and pursuant to the terms of the warranty for the System. The TECAN tip (catalog #10612513) can be directly ordered from TECAN U.S. at 800-352-5128 for an estimated cost of $814.24 per case (9600 tips per case). Pricing for TECAN Tips are subject to change by TECAN. To run 30,000

tests annually, 6.875 cases will be needed (2.2 tips needed per test, 9600 tips per case) at an estimated cost of $5,597.90.

With the addition of TECAN tips purchased by the lab from TECAN, the annual all-inclusive cost to run 20,000 CT/GC tests and 10,000 TV tests is $235,597.90.

45. Provide an itemized list of supplies/items that are included at no charge in the performance of the requested testing.

Product No.	Description
301110	Aptima Combo 2 Controls Kit
302807	Aptima Trichomonas Controls Kit
303096	Run Kit, Panther
303085	Advanced Cleaning Solution
CL0041	Caps, AMP/P.R.S.(CL0045)DIAG.
CL0040	Caps, TCR/SEL.(CL0038) DIAG.
501604	Spare Caps, PP, 60mL, TCR APTIMA 2x50
501616	Spare Caps,30mL tube (501213)Diagnostics

46. Indicate cost per sample tested for GC, CT and TV. The all-inclusive price of a CT/GC test is $8.69. This includes the price per test of $7.25, collection device of $1.25, and 2.2 Tecan tips at $0.08 each. The all-inclusive price of a TV test is $6.19. This includes the price per test of $6.00 and 2.2 Tecan tips at $0.08 each.

47. How are supplies typically shipped? Does cost of reagent/supplies include shipping? If not, how are shipping charges handled and what are the estimated shipping charges per delivery? Reagents are shipped FOB Destination, prepaid and included.

48. From the time an order is placed for supplies, what is the average time for supplies/reagents to arrive? Approximately seven (7) days from date order is placed.

49. What is the average and median outdate from the time of manufacture of the kits and reagents? 12 months

50. Describe the installation procedures and estimated time of installation from the time the RFP is awarded. Hologic has a detailed and thorough implementation plan---which includes delivery, install, training, proficiencies, and assistance with verification (if needed). It takes 2-3 days to install the system, 2-3 days for on-site training, and approximately a week to finish verification (though this is lab dependent).

51. Explain assistance provided during the method validation process. Are supplies provided at no cost during the validation process and will validation samples be provided? The Account Executive and Molecular Application specialist work together to provide supplies for verification for 250 tests (tips, collection devices, and reagents) at no charge.

Is your system capable of testing for pathogens other than GC/CT/TV? If yes, list. Include any that are under development and expected release date. Yes.
Aptima HPV Assay
Aptima HPV 16 18/45 Assay
Aptima Zika Virus Assay (EUA*)
Aptima HSV 1 & 2 Assay

Aptima HIV-1 Quant Assay
Aptima HCV Quant Assay
Aptima HBV Quant Assay
Aptima Mycoplasma genitalium Assay

In addition, Hologic now offers the Panther Fusion which can be an upgrade to the lab's existing Panther instrument. The Panther Fusion's menu includes the following assays:

Flu A/B/RSV
Paraflu
AdV/hMPV/RV
GBS

Currently under development is CMV, Bacterial Vaginosis, and Candida/Trichomoniasis.

52. Describe in detail, other value-added services available to the Health Department but not necessarily addressed. Supplier may offer additional products or services. The Health Department shall determine which value-added service options shall be most beneficial from both a cost and service standpoint, and may further negotiate these options to include or omit dependent on the needs of the Health Department.

The Panther Fusion® system
The Panther Fusion system represents the next step in the legacy of Hologic molecular testing innovation that began with the Panther system and depending on volume and the lab's testing menu, can be an upgrade to the lab's existing Panther system. The Panther Fusion module and Panther Fusion assays bring polymerase chain reaction (PCR) capabilities to the Panther system. Combining PCR with our powerful TMA technology on a single platform allows for multiple chemistries and even more flexibility on a consolidated system. The Panther Fusion system also offers **Open Access™**. Integrating this software into the Panther Fusion system opens many new possibilities. Being able to incorporate IVDs and LDTs into your routine workflow gives you the freedom to consolidate your menu. The ability to run 16 LDTs at once and 3 different PCR reactions from a single sample extraction increases your lab's testing capacity, all without adding an inch to the instrument footprint.

53. Provide a current user reference list of users. Include local and state Health Department lab users in the list.

Institution	Contact	Contract Information
Moses Cone	Amy Lax SCT, ASCP Operations Manager Cytopathology	Cone Health 1200 N Elm Street Greensboro, NC Phone: 336-832-7542 Fax: 336-832-8247 Email: amy.lax@conehealth.com
NC State Lab	Mark A. Turner Bacterial STD Laboratory Supervisor North Carolina State Laboratory of Public Health- Division of Public Health	4312 District Drive Raleigh, NC 27607-5490 Phone: 919-807-8865 Fax: 919-715-7700 Email: mark.a.turner@dhhs.nc.gov

Institution	Contact	Contract Information
Durham County Health Department	Katie J. Mallette, MLS (ASCP) Division Director Laboratory and Pharmacy	Durham County Department of Public Health Human Services Building 414 East Main Street Durham, North Carolina 27701 Phone: (919) 560-7692 Fax: (919) 560-7834 Email: kmallette@dconc.gov

Nucleic Acid Application Testing Evaluation

Evaluation Criteria	Max. Score	Score	Comments Supporting Score
1. **Pricing: Cost for performing samples annually.** CT 20,000 patient samples per year - 20 pts. GC 20,000 patient samples per year - 20 pts. TV 10,000 patient samples per year - 10 pts. Total Max Score 50 pts.	50		
2. **Qualifications** Ability to meet the current and requirements specifications.	25		
3. **Experience** Experience in providing a system to perform NAAT testing for high complexity testing sites and strength of current references.	25		
Evaluator: Date:	100		

Bid Submittal Instructions

1. **All suppliers, who plan to submit a bid must register as a supplier in the Guilford County eProcurement Supplier Portal.**
2. Through the Supplier Portal, suppliers may browse to the open event or search for an event name or number.
3. **Suppliers sign in to begin submitting their response to an event. Click on the event to open it, then click on respond now and follow the instructions for each tab.**
4. All bids must be submitted electronically via the following website www.guilfordcountync.gov/sourcing by the event date and close time. There will be no exceptions.
5. All questions related to this event must be submitted online using the Question & Answer Forum from **Event #647 Friday, March 1, 2019 at 3:00pm through Thursday, March 14, 2019 at 3:00 pm.** Each question asked will be answered online for any/all vendors to view. No questions will be considered after the Q&A close date and time. The system cannot accept late submittals. **No exceptions.**
6. All official County responses to prospective Bidder/Supplier's questions and official addenda regarding bid document revisions or additional information will be communicated by the County through the Lawson Strategic Sourcing Q/A function tab within this event.
7. To complete the lines portion of a submittal in the Strategic Sourcing Event, open the lines tab to enter pricing for each line.
8. Use the provided line description and quantity for each line to complete the entries for each line. Upload all additional documentation required in the bid document as an attachment or attachments.
9. Suppliers are responsible for checking the event for any addendums prior to completion and submission of their response.
10. To complete an electronic submittal, be sure to click the "Submit" button. Clicking the "Done" button puts the response into a draft state that can be completed at a later time, but will not be part of the submitted responses until submitted via the "Submit" button.
11. Suppliers are strongly encouraged to submit their bids with all required documentation at least 24 hours in advance. Guilford County will not be responsible for any technical difficulties that may occur and result in the inability to submit.
12. For questions related to the Minority and Women Business Enterprise Program contact Cynthia Barnes-Phipps at 336-641-4565 or cbarnes1@guilfordcountync.gov.
13. For technical assistance to register as a supplier or submit a bid in the Guilford County Electronic Procurement System, please contact the buyer listed on this event. For immediate assistance, please contact the Purchasing Department at 336-641-3314 and ask to speak to an available Buyer.

March 20, 2019

Guilford County Department of Health
Attn: Ms. Shelia Reaves-Willett
Guilford County Purchasing Department
301 West Market Street
Greensboro NC 27401

Re: Event 647- Nucleic Acid Application Testing

Dear Ms. Reaves-Willett:

Gen-Probe Sales & Service, Inc., a subsidiary of Hologic, Inc. ("Hologic") is pleased to submit its response to the above -mentioned solicitation to provide Nucleic Acid Application testing to the Guilford County Department of Health.

Hologic is a global leader in the development, manufacture and marketing of rapid, accurate and cost-effective molecular diagnostic products and services that are used primarily to diagnose human diseases. Hologic has nearly three decades of molecular diagnostics expertise and its subsidiary, Gen-Probe Sales & Service, Inc., received the 2004 National Medal of Technology, America's highest honor for technological innovation for developing molecular assays and systems that help safeguard the blood supply. Hologic has a proven track record in providing technically superior products of the highest quality to test for various infectious diseases.

Based on Hologic's expertise in hardware and software engineering, we have developed instrument platforms that offer superior automation and workflow for our customers. Since 2014, Hologic has provided the equipment and reagents for CT/GC testing to the Guilford County Department of Health and will meet the future testing requirements as outlined in the solicitation.

Our highly trained sales, technical support, instrumentation service, customer service, and marketing staff are dedicated to satisfying the needs of our customers on an ongoing basis. We are focused on all aspects of the customer experience - compliance, customer satisfaction and resolution.

Please see our account management and staffing contact information listed below:

Scott Loudenback
Director or Public Health and Dept. of Defense
Mobile: (904) 710-8255
Email: scott.loudenback@hologic.com

Ernesto Roman
Account Executive
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Customer Service
Telephone: (800) 442-9892
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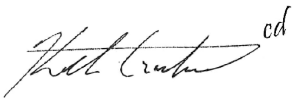
Technical Support
Telephone: (888) 484-4747
Fax: (858) 410-8250
Email: molecularsupport@hologic.com

We look forward to continue providing the Guilford County Public Health Department with our Aptima assays on the Panther Instrument System. Please feel free to contact Ernesto Roman should you have any questions.

Regards,



Keith Gantner
SVP, Group Sales & Commercial Excellence

Panther Instrument System

Hologic offers the **Panther Instrument System ("Panther")**. The Panther provides unprecedented control of workflow, driven by random-access and continuous loading of molecular samples, reagents and consumables. This innovative design enables your lab to improve productivity and accelerate results, ultimately helping enhance patient care, while reducing lab costs.

Created to be a fully automated sample-to-answer instrument, the Panther eliminates the need for batch processing and automates all aspects of nucleic acid testing on a single, integrated platform. This Panther combines true walkaway freedom with intuitive design for ease of use. With its extraordinary level of automation, the Panther improves laboratory productivity and operational efficiencies, promising to transform the world of molecular testing as we know it today.

The Panther provides unique, innovative and streamlined features. The only system that gives you control to run:

- Samples as you choose, random access or batch.
- Multiple assays on the same patient sample.
- HPV, CT/GC and trichomonas simultaneously on a single platform.

Integrated automation, enabling you to maximize productivity and reduce costs:

- True walkaway freedom — sample-to-answer automation.
- Run multiple tests from a single sample anytime.
- Bi-directional LIS interface for streamlined data management.
- Automated scheduled maintenance, reducing hands-on time.

Frees your lab from the confines of batch testing with:

- Random access and continuous sample/reagent loading to enable flexible workflow.
- Load-and-go with unattended results processing.
- Auto-reflexing for HPV genotyping.
- Controls or calibrators that are run only once in 24 hours.

Have confidence in your results with:

- Sample chain-of-custody control (PosID).
- In-process checks, which reduce operator and system errors.
- Liquid-level detection and reagent dispense verification.
- Contamination control.
- Quality control (Levey-Jennings and prevalence) reporting.

Aptima Assays

Hologic also offers the **Aptima Combo 2 Assay**. The Aptima Combo 2 Assay is a target amplification nucleic acid probe test that utilizes target capture for the *in vitro* qualitative detection and differentiation of ribosomal RNA (rRNA) from *Chlamydia trachomatis* (CT) and/or *Neisseria gonorrhoeae* (GC) to aid in the diagnosis of chlamydial and/or gonococcal urogenital disease using the Panther System as specified. On the Panther System, the Aptima Combo 2 assay has been FDA cleared and may be used to test the following specimens from symptomatic and asymptomatic individuals: clinician-collected endocervical,

vaginal and male urethral swab specimens, clinician-collected gynecological specimens collected in the PreservCyt Solution, patient-collected vaginal swab specimens, and male and female urine specimens.

The **Aptima Trichomonas vaginalis Assay** is an in vitro qualitative nucleic acid amplification test (NAAT) for the detection of ribosomal RNA (rRNA) from Trichomonas vaginalis to aid in the diagnosis of trichomoniasis using the Panther System. The assay may be used to test the following specimens from symptomatic or asymptomatic women: clinician-collected endocervical swabs, clinician-collected vaginal swabs, and specimens collected in PreservCyt Solution.

For more details on the Panther and Aptima assay, please see our responses to the solicitation and the Package Inserts.

Aptima® Trichomonas vaginalis Assay (Panther® System)

For *in vitro* diagnostic use.

General Information

Intended Use

The Aptima Trichomonas vaginalis Assay is an *in vitro* qualitative nucleic acid amplification test (NAAT) for the detection of ribosomal RNA (rRNA) from *Trichomonas vaginalis* to aid in the diagnosis of trichomoniasis using the Panther System.

The assay may be used to test the following specimens from symptomatic or asymptomatic women: clinician-collected endocervical swabs, clinician-collected vaginal swabs, and specimens collected in PreservCyt Solution.

Summary and Explanation of the Test

Trichomonas vaginalis (TV) is the most common curable sexually transmitted disease (STD) agent in the United States, with an estimated 7.4 million new cases occurring annually (1, 2).

Infections in women cause vaginitis, urethritis, and cervicitis. Discharge and small hemorrhagic lesions may be present in the genitourinary tract. Complications can include premature labor, low-birth-weight offspring, premature rupture of membranes, and post-abortion or post-hysterectomy infection. An association with pelvic inflammatory disease, tubal infertility, and cervical cancer with previous episodes of trichomoniasis has been reported. Symptomatic women with trichomoniasis usually complain of vaginal discharge, vulvovaginal soreness, and/or irritation. Dysuria is also common. However, it has been estimated that 10 to 50% of *T. vaginalis* infections in women are asymptomatic, and in men the proportion may even be higher (3, 4, 5).

Detection of *T. vaginalis* with traditional culture methods is technically challenging and requires up to 7 days. Immediate inoculation into the media is preferred, and proper incubation conditions are required in addition to frequent microscopic examinations of the media to successfully culture the protozoa. The sensitivity of culture has been estimated to range from 38% to 82% when compared to molecular methods due to problems visualizing low numbers of the organisms or the motility of the protozoa (6, 7).

T. vaginalis may also be detected using “wet-mount” preparation by mixing vaginal secretions with saline on a slide and examining the slide under a microscope. However, the wet-mount method is only 35% to 80% sensitive compared with culture (7). The sensitivity of the wet-mount method is highly dependent on the experience of the microscopist as well as the time of specimen transport to the laboratory.

The Aptima Trichomonas vaginalis Assay is a nucleic acid test that utilizes Target Capture, Transcription-Mediated Amplification (TMA), and Hybridization Protection Assay (HPA) technologies.

Principles of the Procedure

The Aptima Trichomonas vaginalis Assay involves the technologies of target capture, transcription-mediated amplification (TMA), and hybridization protection assay (HPA).

Specimens are collected and transferred into their respective specimen transport tubes. The transport solution in these tubes releases the rRNA target and protects it from degradation during storage. When the Aptima Trichomonas vaginalis Assay is performed in the laboratory, the target rRNA is isolated from the specimens by the use of a specific capture oligomer and magnetic microparticles in a method called target capture. The capture oligomer contains a

sequence complementary to a specific region of the target molecule as well as a string of deoxyadenosine residues. During the hybridization step, the sequence-specific region of the capture oligomer binds to a specific region of the target molecule. The capture oligomer:target complex is then captured out of solution by decreasing the temperature of the reaction to room temperature. This temperature reduction allows hybridization to occur between the deoxyadenosine region on the capture oligomer and the poly-deoxythymidine molecules that are covalently attached to the magnetic particles. The microparticles, including the captured target molecule bound to them, are pulled to the side of the reaction vessel using magnets and the supernatant is aspirated. The particles are washed to remove residual specimen matrix that may contain amplification inhibitors. After the target capture steps are completed, the specimens are ready for amplification.

Target amplification assays are based on the ability of complementary oligonucleotide primers to specifically anneal and allow enzymatic amplification of the target nucleic acid strands. The Hologic TMA reaction amplifies a specific region of the small ribosomal subunit from *T. vaginalis* via DNA and RNA intermediates and generates RNA amplicon molecules. Detection of the rRNA amplification product sequences is achieved using nucleic acid hybridization (HPA). A single-stranded chemiluminescent DNA probe, which is complementary to a region of the target amplicon, is labeled with an acridinium ester molecule. The labeled DNA probe combines with amplicon to form stable RNA:DNA hybrids. The Selection Reagent differentiates hybridized from unhybridized probe, eliminating the generation of signal from unhybridized probe. During the detection step, light emitted from the labeled RNA:DNA hybrids is measured as photon signals in a luminometer and are reported as Relative Light Units (RLU).

Warnings and Precautions

A. For *in vitro* diagnostic use.

B. For additional specific warnings and precautions, refer to the *Panther System Operator's Manual*.

Laboratory Related

C. Use only supplied or specified disposable laboratory ware.

D. Use routine laboratory precautions. Do not eat, drink or smoke in designated work areas. Wear disposable, powderless gloves, protective eye wear, and laboratory coats when handling specimens and kit reagents. Wash hands thoroughly after handling specimens and kit reagents.

E. **Warning: Irritants and Corrosives.** Avoid contact of Auto Detect 1 and Auto Detect 2 with skin, eyes and mucous membranes. If these fluids come into contact with skin or eyes, wash with water. If these fluids spill, dilute the spill with water before wiping dry.

F. Work surfaces, pipettes, and other equipment must be regularly decontaminated with 2.5% to 3.5% (0.35M to 0.5M) sodium hypochlorite solution.

Specimen Related

G. Expiration dates for the specimen transfer kits pertain to the collection/transfer of specimens and not to specimen testing. Specimens collected/transferred any time prior to these expiration dates are valid for testing provided they have been transported and stored in

accordance with the package insert, even if the expiration date on the transfer tube has passed.

H. Specimens may be infectious. Use Universal Precautions when performing this assay. Proper handling and disposal methods should be established by the laboratory director. Only personnel adequately trained in handling infectious materials should be permitted to perform this diagnostic procedure.

I. Avoid cross-contamination during the specimen handling steps. Specimens can contain extremely high levels of organisms. Ensure that specimen containers do not contact one another, and discard used materials without passing over any container. Change gloves if they come in contact with specimen.

J. Upon piercing, liquid can discharge from Aptima transfer tube caps under certain conditions. Refer to the appropriate *Test Procedure* for more information.

K. Maintain proper storage conditions during specimen shipping to ensure the integrity of the specimen. Specimen stability under shipping conditions other than those recommended has not been evaluated.

L. If the lab receives a swab specimen transport tube with no swab, two swabs, a cleaning swab, or a swab not supplied by Hologic, the specimen must be rejected.

Assay Related

M. Store reagents at the specified temperatures. Performance of the assay may be affected by use of improperly stored reagents.

N. Use Universal Precautions when handling controls.

O. Avoid microbial and ribonuclease contamination of reagents.

P. Do not use kit after its expiration date.

Q. Do not interchange, mix, or combine assay reagents from kits with different lot numbers. Controls and assay fluids may be interchanged.

Reagent Storage and Handling Requirements

A. The following reagents are stable when stored at 2ºC to 8ºC:

Aptima *Trichomonas vaginalis* Amplification Reagent

Aptima *Trichomonas vaginalis* Enzyme Reagent

Aptima *Trichomonas vaginalis* Probe Reagent

Aptima *Trichomonas vaginalis* Assay Target Capture Reagent B

Aptima *Trichomonas vaginalis* Controls

B. The following reagents are stable when stored at room temperature (15°C to 30°C):

Aptima *Trichomonas vaginalis* Amplification Reconstitution Solution

Aptima *Trichomonas vaginalis* Enzyme Reconstitution Solution

Aptima *Trichomonas vaginalis* Probe Reconstitution Solution

Aptima *Trichomonas vaginalis* Target Capture Reagent

Aptima *Trichomonas vaginalis* Selection Reagent

C. After reconstitution, Amplification Reagent, Enzyme Reagent, and Probe Reagent are stable for 60 days when stored at 2ºC to 8ºC.

D. Working Target Capture Reagent (wTCR) is stable for 60 days when stored at 15ºC to 30ºC. Do not refrigerate.

E. Discard any unused reconstituted reagents and wTCR after 60 days, or after the Master Lot expiration date, whichever comes first.

F. Controls are stable until the date indicated on the vials.

G. Reagents stored on-board the Panther System have 72 hours of on-board stability.

H. Avoid cross-contamination during reagent handling and storage. Recap all reconstituted reagents with new reagent caps each time prior to storage.

I. The Probe Reagent and Reconstituted Probe Reagent are photosensitive. Store the reagents protected from light.

J. **Do not freeze reagents**.

Specimen Collection and Storage

The Aptima Trichomonas vaginalis Assay is designed to detect the presence of *T. vaginalis* in clinician-collected endocervical and vaginal swab specimens and PreservCyt liquid Pap specimens. Performance with specimens other than those collected with the following specimen collection kits has not been evaluated:

- Aptima Unisex Swab Specimen Collection Kit for Endocervical and Male Urethral Swab Specimens
- Aptima Vaginal Swab Specimen Collection Kit
- Aptima Specimen Transfer Kit (for use with gynecological samples collected in PreservCyt Solution)

A. Instructions for collection

1. Refer to the appropriate specimen collection kit package insert for specific collection instructions.

B. Specimen transport and storage before testing:

1. Swab specimens
   a. After collection, transport and store the swab in the swab specimen transport tube at 2°C to 30°C until tested.
   b. Assay specimens within 60 days of collection. If longer storage is needed, freeze the specimen transport tube at ≤ –20°C for up to 24 months.
2. Specimens collected in PreservCyt Solution
   a. Transport and store the PreservCyt Solution specimen at 2°C to 30°C for up to 30 days.
   b. Specimens collected in PreservCyt Solution must be transferred into an Aptima specimen transfer tube according to the instructions in the Aptima Specimen Transfer kit package insert.
   c. After transfer to an Aptima specimen transfer tube, specimens may be stored an additional 14 days at 15°C to 30°C or 30 days at 2°C to 8°C.
   d. If longer storage is needed, the PreservCyt Solution specimen or the PreservCyt Solution liquid Pap specimen diluted into the specimen transfer tube may be stored at ≤ –20°C for up to 24 months after transfer.

C. Specimen storage after testing:

1. Specimens that have been assayed must be stored upright in a rack.
2. The specimen transport tubes should be covered with a new, clean plastic film or foil barrier.
3. If assayed samples need to be frozen or shipped, remove penetrable cap and place new non-penetrable caps on the specimen transport tubes. If specimens need to be shipped for testing at another facility, recommended temperatures must be maintained. Prior to uncapping, specimen transport tubes must be centrifuged for 5 minutes at 420 RCF (Relative Centrifugal Force) to bring all of the liquid down to the bottom of the tube. **Avoid splashing and cross-contamination.**

Note: *Specimens must be shipped in accordance with applicable national and international transportation regulations.*

Panther System

Reagents for the Aptima Trichomonas vaginalis Assay are listed below for the Panther System. Reagent Identification Symbols are also listed next to the reagent name.

Reagents and Materials Provided

Note: *For information on any hazard and precautionary statements that may be associated with reagents, refer to the Safety Data Sheet Library at www.hologic.com/sds.*

Aptima Trichomonas vaginalis Assay (Panther System) Kit

250 tests (2 boxes and 1 Controls kit) (Cat. No. 303537)

100 tests (2 boxes and 1 Controls kit) (Cat. No. 303536)

Aptima Trichomonas vaginalis Assay Refrigerated Box (Box 1 of 2)
(store at 2°C to 8°C upon receipt)

Symbol	Component	Quantity	
		250-test kit	100-test kit
A	**Aptima *Trichomonas vaginalis* Amplification Reagent** *Primers and nucleotides dried in buffered solution containing < 5% bulking agent.*	1 vial	1 vial
E	**Aptima *Trichomonas vaginalis* Enzyme Reagent** *Reverse transcriptase and RNA polymerase dried in HEPES buffered solution containing < 10% bulking reagent.*	1 vial	1 vial
P	**Aptima *Trichomonas vaginalis* Probe Reagent** *Chemiluminescent DNA probes dried in succinate buffered solution containing < 5% detergent.*	1 vial	1 vial
TCR-B	**Aptima *Trichomonas vaginalis* Assay Target Capture Reagent B** *Buffered solution containing < 5% detergent.*	1 x 0.56 mL	1 x 0.30 mL

Aptima Trichomonas vaginalis Assay Room Temperature Box (Box 2 of 2)
(store at room temperature, 15°C to 30°C upon receipt)

Symbol	Component	Quantity	
		250-test kit	100-test kit
AR	**Aptima *Trichomonas vaginalis* Amplification Reconstitution Solution** *Aqueous solution containing preservatives.*	1 x 27.7 mL	1 x 11.9 mL
ER	**Aptima *Trichomonas vaginalis* Enzyme Reconstitution Solution** *HEPES buffered solution containing a surfactant and glycerol.*	1 x 11.1 mL	1 x 6.3 mL
PR	**Aptima *Trichomonas vaginalis* Probe Reconstitution Solution** *Succinate buffered solution containing < 5% detergent.*	1 x 35.4 mL	1 x 15.2 mL
S	**Aptima *Trichomonas vaginalis* Selection Reagent** *600 mM borate buffered solution containing surfactant.*	1 x 108 mL	1 x 43.0 mL
TCR	**Aptima *Trichomonas vaginalis* Target Capture Reagent** *Buffered solution containing capture oligomers and magnetic particles.*	1 x 54.0 mL	1 x 26.0 mL
	Reconstitution Collars	3	3
	Master Lot Barcode Sheet	1 sheet	1 sheet

Aptima Trichomonas vaginalis Controls Kit
(store at 2°C to 8°C upon receipt)

Symbol	Component	Quantity
NC	**Aptima *Trichomonas vaginalis* Negative Control** *Non-infectious non-target nucleic acid in a buffered solution containing < 5% detergent.*	5 x 1.7 mL
PC	**Aptima *Trichomonas vaginalis* Positive Control** *Non-infectious Trichomonas vaginalis organisms in buffered solution containing < 5% detergent.*	5 x 1.7 mL

Materials Required But Available Separately

Note: *Materials available from Hologic have catalog numbers listed, unless otherwise specified.*

	Cat. No.
Panther System	303095
Aptima Assay Fluids Kit *(Aptima Wash Solution, Aptima Buffer for Deactivation Fluid, and Aptima Oil Reagent)*	303014 (1000 tests)
Aptima Auto Detect Kit	303013 (1000 tests)
Multi-tube units (MTUs)	104772-02
Panther Waste Bag Kit	902731
Panther Waste Bin Cover	902714
Or Panther Run Kit *contains MTUs, waste bags, waste bin covers, assay fluids, and auto detects*	303096 (5000 tests)
Tips, 1000 µL conductive, liquid sensing	10612513 (Tecan)
Aptima Specimen Transfer Kit *for use with specimens in PreservCyt Solution*	301154C
Aptima Vaginal Swab Specimen Collection Kit	301162
Aptima Unisex Swab Specimen Collection Kit for Endocervical and Male Urethral Swab Specimens	301041
Bleach, 5% to 7% (0.7 M to 1.0 M) sodium hypochlorite solution	—
Disposable gloves	—
SysCheck calibration standard	301078
Aptima penetrable caps	105668
Replacement non-penetrable caps	103036A
Replacement Caps for the 250-test kits *Amplification and Probe reagent reconstitution solutions* *CL0041 (100 caps)* *Enzyme Reagent reconstitution solution* *501616 (100 caps)* *TCR and Selection reagent* *CL0040 (100 caps)*	—
Replacement Caps for 100-test kits *Amplification, Enzyme, and Probe reagent reconstitution solutions* *CL0041 (100 caps)* *TCR and Selection reagent* *501604 (100 caps)*	—

Optional Materials

	Cat. No.
Aptima Trichomonas vaginalis Controls Kit	302807
Hologic Bleach Enhancer for Cleaning *for routine cleaning of surfaces and equipment*	302101

Panther System Test Procedure

Note: *See the Panther System Operator's Manual for additional Panther System procedural information.*

A. Work Area Preparation

1. Clean work surfaces where reagents and samples will be prepared. Wipe down work surfaces with 2.5% to 3.5% (0.35 M to 0.5 M) sodium hypochlorite solution. Allow the sodium hypochlorite solution to contact surfaces for at least 1 minute and then follow with a water rinse. Do not allow the sodium hypochlorite solution to dry. Cover the bench surface on which the reagents and samples will be prepared with clean, plastic-backed absorbent laboratory bench covers.

B. Reagent Reconstitution/Preparation of a New Kit

Note: *Reagent reconstitution should be performed prior to beginning any work on the Panther System.*

1. To reconstitute Amplification, Enzyme, and Probe Reagents, combine the bottles of lyophilized reagent with the reconstitution solution. If refrigerated, allow the reconstitution solutions to reach room temperature before use.
   a. Pair each reconstitution solution with its lyophilized reagent. Ensure that the reconstitution solution and reagent have matching label colors before attaching the reconstitution collar.
   b. Check the lot numbers on the Master Lot Barcode Sheet to ensure that the appropriate reagents are paired.
   c. Open the lyophilized reagent vial and firmly insert the notched end of the reconstitution collar into the vial opening (Figure 1, Step 1).
   d. Open the matching reconstitution solution bottle, and set the cap on a clean, covered work surface.
   e. While holding the reconstitution solution bottle on the bench, firmly insert the other end of the reconstitution collar into the bottle opening (Figure 1, Step 2).
   f. Slowly invert the assembled bottles. Allow the solution to drain from the bottle into the glass vial (Figure 1, Step 3).
   g. Gently swirl the solution in the bottle to mix. Avoid creating foam while swirling the bottle (Figure 1, Step 4).
   h. Wait for the lyophilized reagent to go into solution, then invert the assembled bottles again, tilting at a 45° angle to minimize foaming (Figure 1, Step 5). Allow all of the liquid to drain back into the plastic bottle.
   i. Remove the reconstitution collar and glass vial (Figure 1, Step 6).
   j. Recap the plastic bottle. Record operator initials and reconstitution date on the label (Figure 1, Step 7).
   k. Discard the reconstitution collar and glass vial (Figure 1, Step 8).

Warning: *Avoid creating foam when reconstituting reagents. Foam compromises the level-sensing in the Panther System.*

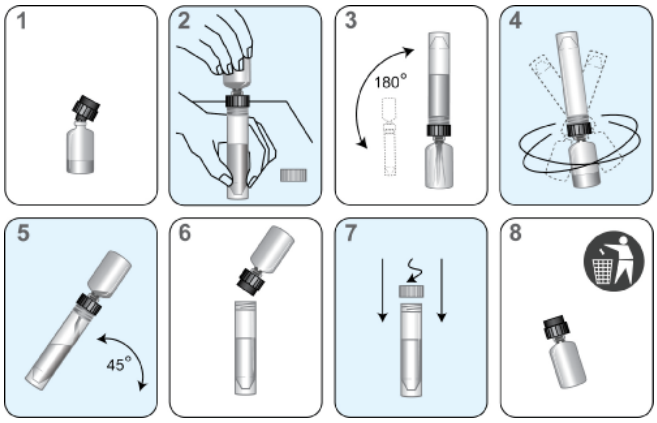


Figure 1. Panther System Reconstitution Process

2. Prepare Working Target Capture Reagent (wTCR)
   a. Pair the appropriate bottles of TCR and TCR-B.
   b. Check the reagent lot numbers on the Master Lot Barcode Sheet to make sure that the appropriate reagents in the kit are paired.
   c. Open the bottle of TCR, and set the cap on a clean, covered work surface.
   d. Open the bottle of TCR-B and pour the entire contents into the bottle of TCR. Expect a small amount of liquid to remain in the TCR-B bottle.
   e. Cap the bottle of TCR and gently swirl the solution to mix the contents. Avoid creating foam during this step.
   f. Record operator initials and the current date on the label.
   g. Discard the TCR-B bottle and cap.
3. Prepare Selection Reagent
   a. Check the lot number on the reagent bottle to make sure that it matches the lot number on the Master Lot Barcode Sheet.
   b. Record operator initials and the current date on the label.

Note: *Thoroughly mix by gently inverting all reagents prior to loading on the system. Avoid creating foam during inversion of reagents.*

C. Reagent Preparation for Previously Reconstituted Reagents

1. Previously reconstituted Amplification, Enzyme, and Probe Reagents must reach room temperature (15°C to 30°C) prior to the start of the assay.
2. If reconstituted Probe Reagent contains precipitate that does not return to solution at room temperature, heat the capped bottle at a temperature that does not exceed 62°C for 1 to 2 minutes. After this heat step, the Probe Reagent may be used even if residual precipitate remains. Mix Probe Reagent by inversion, being careful not to induce foam, prior to loading onto the system.
3. Thoroughly mix each reagent by gently inverting prior to loading on the system. Avoid creating foam during inversion of reagents.
4. Do not top off reagent bottles. The Panther System will recognize and reject bottles that have been topped off.

D. Specimen Handling

1. Allow the controls and specimens to reach room temperature prior to processing.
2. **Do not vortex specimens.**
3. Visually confirm that each specimen tube meets one of the following criteria:
   a. The presence of a single blue Aptima collection swab in a unisex swab specimen transport tube.
   b. The presence of a single pink Aptima collection swab in a vaginal swab specimen transport tube.
   c. The absence of a swab in the Aptima specimen transport tube for PreservCyt Solution liquid Pap specimens.
4. Inspect specimen tubes before loading into rack:
   a. If a specimen tube contains bubbles in the space between the liquid and the cap, centrifuge the tube for 5 minutes at 420 RCF to eliminate the bubbles.
   b. If a specimen tube has a lower volume than typically observed when collection instructions have been followed, centrifuge the tube for 5 minutes at 420 RCF to ensure that no liquid is in the cap.

Note: *Failure to follow Steps 4a-4b may result in liquid discharge from the specimen tube cap.*

Note: *Up to 3 separate aliquots can be tested from each specimen tube. Attempts to pipette more than 3 aliquots from the specimen tube can lead to processing errors.*

E. System Preparation

1. Set up the system according to the instructions in the *Panther System Operator's Manual* and *Procedural Notes*.
2. Load samples.

Procedural Notes

A. Controls

1. To work properly with the Panther Aptima Assay software, one pair of controls is required. The Aptima Positive Control for Trichomonas and Aptima Negative Control for Trichomonas can be loaded in any rack position or in any Sample Bay Lane on the Panther System. Patient specimen pipetting will begin when one of the following two conditions has been met:
   a. A pair of controls is currently being processed by the system.
   b. Valid results for the controls are registered on the system.
2. Once the control tubes have been pipetted and are processing for a specific reagent kit, patient specimens can be run with the associated kit up to 24 hours **unless:**
   a. Controls results are invalid.
   b. The associated assay reagent kit is removed from the system.
   c. The associated assay reagent kit has exceeded stability limits.
3. Each Aptima control tube can be tested once. Attempts to pipette more than once from the tube can lead to processing errors.

B. Temperature

Room temperature is defined as 15°C to 30°C.

C. Glove Powder

As in any reagent system, excess powder on some gloves may cause contamination of opened tubes. Powderless gloves are recommended.

D. Lab Contamination Monitoring Protocol for the Panther System

There are many laboratory-specific factors that may contribute to contamination, including testing volume, workflow, disease prevalence and various other laboratory activities. These factors should be taken into consideration when contamination monitoring frequency is being established. Intervals for contamination monitoring should be established based on each laboratory's practices and procedures.

To monitor for laboratory contamination, the following procedure may be performed using the Aptima Unisex Swab Specimen Collection Kit for Endocervical and Male Urethral Swab Specimens:

1. Label swab transport tubes with numbers corresponding to the areas to be tested.
2. Remove the specimen collection swab (blue shaft swab with green printing) from its packaging, wet the swab in the swab transport medium, and swab the designated area using a circular motion.
3. Immediately insert the swab into transport tube.
4. Carefully break the swab shaft at the score line; use care to avoid splashing of the contents.
5. Recap the swab transport tube tightly.
6. Repeat Steps 2 to 5 for each area to be swabbed.
7. Test samples with the Aptima Trichomonas vaginalis Assay on the Panther System.
8. Further investigation should be performed if any samples yield a positive result.

For test interpretation, see *Test Interpretation — QC/Patient Results*. For additional Panther System-specific contamination monitoring information, contact Hologic Technical Support.

Test Interpretation — QC/Patient Results

A. Test Interpretation

Assay test results are automatically interpreted by the Panther System Aptima Trichomonas Assay software. A test result may be negative, positive, or invalid as determined by total RLU in the detection step (see below). A test result may be invalid due to RLU values outside the normal expected ranges. Initial invalid test results should be retested. Report the first valid result.

Test Interpretation	Total RLU (x1000)
Negative	0* to < 100
Positive	100 to < 2400
Invalid	0* or ≥ 2400

*If the RLU measured on the Panther System is between 0 and 999, a result of "0" is reported in the "Total RLU (000s)" column in the run report. Measured RLU values less than 690 are reported as invalid. RLU values between 690 and 999 are reported as valid.

B. Quality Control Results and Acceptability

The Aptima Negative Control for Trichomonas, which is labeled "NC CONTROL – TRICH," and the Aptima Positive Control for Trichomonas, which is labeled "PC CONTROL + TRICH," act as controls for the target capture, amplification, and detection steps of the assay. In accordance with guidelines or requirements of national, regional, and/or local regulations or accrediting organizations, additional controls for cell lysis and RNA stabilization may be included. The Aptima Positive Control for Trichomonas which is labeled "PC CONTROL + TRICH" contains non-infectious *T. vaginalis* rRNA.

The Aptima Trichomonas vaginalis Controls must produce the following test results:

Control	Total RLU (x1000)	*T. vaginalis* Result
NC Control – TRICH	0* and < 20	Negative
PC Control + TRICH	≥ 500 and < 2400	Positive

*If the RLU measured on the Panther System is between 0 and 999, a result of "0" is reported in the "Total RLU (000s)" column in the run report. Measured RLU values less than 690 are reported as invalid. RLU values between 690 and 999 are reported as valid.

Each laboratory should implement appropriate control procedures to satisfy the requirements of CLIA regulations (section 493.1256).

Note: *For assistance with out-of-range controls, contact Hologic Technical Support.*

Limitations

A. Use of this assay is limited to personnel who have been trained in the procedure. Failure to follow the instructions given in this insert may result in erroneous results.

B. The effects of tampon use, douching, and specimen collection variables have not been assessed for their impact on the detection of *Trichomonas vaginalis*.

C. TV-positive mucoid samples may exhibit decreased RLU values. To ensure proper endocervical sampling, excess mucus should be removed.

D. Vaginal swab and PreservCyt Solution liquid Pap specimen sampling is not designed to replace cervical exams and endocervical specimens for diagnosis of female urogenital infections. Patients may have cervicitis, urethritis, urinary tract infections, or vaginal infections due to other causes or concurrent infections with other agents.

E. This assay has been tested using only the specimen types indicated. Performance with other specimen types has not been evaluated.

F. Reliable results are dependent on adequate specimen collection. Because the transport system used for this assay does not permit microscopic assessment of specimen adequacy, training of clinicians in proper specimen collection techniques is necessary. See *Specimen Collection and Storage* for instructions. For detailed information, refer to the appropriate instructions for use.

G. Therapeutic failure or success cannot be determined with the Aptima Trichomonas vaginalis Assay since nucleic acid may persist following appropriate antimicrobial therapy.

H. Results from the Aptima Trichomonas vaginalis Assay should be interpreted in conjunction with other clinical data available to the clinician.

I. A negative result does not preclude a possible infection because results are dependent on adequate specimen collection. Test results may be affected by improper specimen collection, technical error, specimen mix-up, or target levels below the assay limit of detection.

J. A negative result does not preclude a possible infection because the presence of *Trichomonas tenax* or *Pentatrichomonas hominis* in a specimen may affect the ability to detect *T. vaginalis* rRNA. See *Cross-Reactivity in the Presence of Microorganisms* for details.

K. The Aptima Trichomonas vaginalis Assay provides qualitative results. Therefore, a correlation cannot be drawn between the magnitude of a positive assay signal and the number of organisms in a specimen.

L. The Aptima Trichomonas vaginalis Assay has not been validated for use with vaginal swab specimens collected by patients.

M. Performance has not been evaluated in pregnant women.

N. Performance has not been evaluated in women less than 14 years of age.

O. The performance of the Panther System has not been determined at altitudes above 2000 m (6561 feet).

P. If a specimen has a small number of *T. vaginalis* organisms, uneven distribution of these trichomonads may occur, which may affect the ability to detect *T. vaginalis* rRNA in the collected material. If negative results from the specimen do not fit with the clinical impression, a new specimen may be necessary.

Q. Customers must independently validate an LIS transfer process.

Expected Values

Prevalence

Estimates of the prevalence of *T. vaginalis* in different populations depend on the sensitivity of the test in detecting the infection and on patient risk factors such as age, lifestyle, and the presence or absence of symptoms. A summary of the prevalence of *T. vaginalis*, by specimen type, as determined by the Aptima Trichomonas vaginalis Assay during the Panther System clinical study is shown in Table 1.

Table 1: Prevalence of T. vaginalis as Determined by the Aptima Trichomonas vaginalis Assay by Specimen Type and Collection Site

Specimen Type	% (# positive / # tested)									
	All Sites	Site 1	Site 2	Site 3	Site 4	Site 5	Site 6	Site 7	Site 8	Site 9
CVS	11.8 (80/678)	17.0 (9/53)	7.7 (4/52)	16.7 (2/12)	19.5 (8/41)	0.7 (1/145)	16.0 (12/75)	12.0 (21/175)	15.0 (12/80)	24.4 (11/45)
ES	11.2 (80/713)	20.4 (11/54)	8.9 (5/56)	12.5 (2/16)	17.1 (7/41)	0.6 (1/162)	20.2 (18/89)	9.1 (15/164)	13.3 (11/83)	20.8 (10/48)
PCyt	11.8 (93/790)	18.3 (11/60)	7.4 (5/68)	17.6 (3/17)	18.6 (8/43)	0.6 (1/167)	22.1 (23/104)	11.2 (22/197)	10.5 (9/86)	22.9 (11/48)

CVS = clinician-collected vaginal swab, ES = endocervical swab, PCyt = PreservCyt Solution liquid Pap.

Positive and Negative Predictive Values for Hypothetical Prevalence Rates

The estimated positive predictive value (PPV) and negative predictive value (NPV) of the Aptima Trichomonas vaginalis Assay across different hypothetical prevalence rates are shown for each specimen type in Table 2. These calculations are based on the overall estimated sensitivity and specificity for each specimen type in the Panther System clinical study.

Table 2: Hypothetical PPV and NPV of the Aptima Trichomonas vaginalis Assay by Specimen Type

Specimen Type	Prevalence (%)	PPV (%)	NPV (%)
CVS	1	35.4	100
	2	52.6	100
	5	74.1	100
	10	85.8	100
	15	90.6	100
	20	93.1	100
	25	94.8	100
ES	1	34.8	100
	2	51.8	100
	5	73.5	100
	10	85.4	100
	15	90.3	100
	20	93.0	100
	25	94.6	100
PCyt	1	41.1	100
	2	58.5	100
	5	78.4	100
	10	88.5	100
	15	92.4	100
	20	94.5	100
	25	95.8	100

CVS = clinician-collected vaginal swab, ES = endocervical swab, PCyt = PreservCyt Solution liquid Pap. The PPV and NPV are derived for different hypothetical prevalence rates using the sensitivity and specificity estimates from the clinical performance study. Sensitivity was 100% in vaginal swab, endocervical swab, and PreservCyt Solution liquid Pap specimens. Specificity was 98.2% in vaginal swab specimens, 98.1% in endocervical swab specimens, and 98.6% in PreservCyt Solution liquid Pap specimens.

Panther System Clinical Performance

Clinical Study

Clinical performance of the Aptima Trichomonas vaginalis Assay on the Panther System was evaluated using leftover specimens collected from consenting subjects during a previous, prospective, multicenter clinical study of the Aptima Trichomonas vaginalis Assay on the Tigris DTS System. Symptomatic and asymptomatic women were enrolled from 9 US clinical sites, including obstetrics and gynecology, family planning, and STD clinics. Three (3) vaginal swab, 1 endocervical swab, and 1 PreservCyt Solution liquid Pap specimen were collected from each subject. All specimens were clinician-collected.

PreservCyt liquid Pap specimens were collected with a broom-type device or a spatula and cytobrush. Two of the vaginal swab specimens were tested with a commercially available culture system and wet mount microscopic examination to establish infected status. The remaining specimens were prepared for Aptima Trichomonas vaginalis Assay testing in accordance with the appropriate Aptima specimen collection kit package insert instructions.

Panther System testing with the Aptima Trichomonas vaginalis Assay was conducted at 3 sites (2 external laboratories and Hologic) in accordance with package insert instructions.

Performance characteristics of the Aptima Trichomonas vaginalis Assay were estimated by comparing results to a patient infected status algorithm. In the algorithm, the designation of a subject as being infected or non-infected with *T. vaginalis* was based on results from vaginal swab specimens tested by culture and/or wet mount microscopic examination. At least one of the reference test results was required to be positive to establish an infected patient status. Both reference tests were required to be negative to establish a non-infected patient status.

Twenty-three (23) Aptima Trichomonas vaginalis Assay runs were initiated on the Panther System. Of these 23 runs, 1 (4.3%, 1/23) was aborted due to a fatal hardware error that led to a software failure. Specimens tested in the aborted run were retested. A total of 689 vaginal swab, 737 endocervical swab, and 791 PreservCyt Solution liquid Pap specimens were tested in the 22 valid runs. Of these specimens, 12 vaginal swab (1.7%, 12/689), 24 endocervical swab (3.3%, 24/737), and 29 PreservCyt Solution liquid Pap (3.7%, 29/791) specimens had initial invalid results due to hardware or software errors. Specimens with initial invalid results were retested. Eleven (11) vaginal swab (1.6%, 11/689), 24 endocervical swab (3.3%, 24/737), and 1 PreservCyt Solution liquid Pap (0.1%, 1/791) specimens had final invalid results due to hardware or software errors; these specimens were excluded from the analyses.

Table 3 shows the sensitivity, specificity, PPV, and NPV of the Aptima Trichomonas vaginalis Assay on the Panther System and the prevalence of *T. vaginalis* (based on the infected status) in each specimen type by symptom status and overall. Subjects were classified as symptomatic if symptoms were reported by the subject. Subjects were classified as asymptomatic if the subject did not report symptoms. Prevalence was higher in symptomatic women.

Table 3: Performance Characteristics of the Aptima Trichomonas vaginalis Assay by Symptom Status

Specimen Type	Symptom Status	n	TP	FP[1]	TN	FN	Prev %	Sensitivity % (95% CI)[2]	Specificity % (95% CI)[2]	PPV % (95% CI)[3]	NPV % (95% CI)[3]
CVS	Asymptomatic	274	12	7[a]	255	0	4.4	100 (75.8-100)	97.3 (94.6-98.7)	63.2 (45.8-80.9)	100 (98.8-100)
	Symptomatic	393	57	4[b]	332	0	14.5	100 (93.7-100)	98.8 (97.0-99.5)	93.4 (84.9-98.1)	100 (98.9-100)
	All	667	69	11[c]	587	0	10.3	100 (94.7-100)	98.2 (96.7-99.0)	86.3 (77.9-92.6)	100 (99.4-100)
ES	Asymptomatic	309	16	5[d]	288	0	5.2	100 (80.6-100)	98.3 (96.1-99.3)	76.2 (58.1-90.8)	100 (98.9-100)
	Symptomatic	391	51	7[e]	333	0	13.0	100 (93.0-100)	97.9 (95.8-99.0)	87.9 (78.1-94.7)	100 (99.0-100)
	All	700	67	12[f]	621	0	9.6	100 (94.6-100)	98.1 (96.7-98.9)	84.8 (76.3-91.5)	100 (99.4-100)
PCyt	Asymptomatic	333	19	2[g]	312	0	5.7	100 (83.2-100)	99.4 (97.7-99.8)	90.5 (72.6-98.7)	100 (98.9-100)
	Symptomatic	441	64	8[h]	369	0	14.5	100 (94.3-100)	97.9 (95.9-98.9)	88.9 (80.4-94.9)	100 (99.1-100)
	All	774	83	10[i]	681	0	10.7	100 (95.6-100)	98.6 (97.4-99.2)	89.2 (82.0-94.5)	100 (99.5-100)

CI = confidence interval, CVS = clinician-collected vaginal swab, ES = endocervical swab, FN = false negative, FP = false positive, PCyt = PreservCyt Solution liquid Pap, Prev = prevalence, TN = true negative, TP = true positive.

[1]Specimens were also tested by an alternative *T. vaginalis* NAAT assay with the following results (# positive results / # samples tested): a: 4/7, b: 3/4, c: 7/11, d 1/5, e: 2/7, f: 3/12, g: 0/2, h: 3/8, i: 3/10.

[2]Score confidence interval.

[3]PPV 95% confidence interval computed from the exact 95% confidence interval for the positive likelihood ratio, NPV 95% confidence interval computed from the exact 95% confidence interval from the negative likelihood ratio.

Table 4 shows the sensitivity, specificity, PPV, and NPV of the Aptima Trichomonas vaginalis Assay on the Panther System and the prevalence of *T. vaginalis* (based on the infected status) in each specimen type by collection site. For each specimen type, performance was similar across collection sites. Prevalence varied across collection sites, as expected.

Table 4: Performance Characteristics of the Aptima Trichomonas vaginalis Assay by Collection Site

Site	Specimen Type	n	TP	FP	TN	FN	Prev %	Sensitivity (95% CI)[1]	Specificity (95% CI)[1]	PPV % (95% CI)[2]	NPV % (95% CI)[2]
1	CVS	52	8	1	43	0	15.4	100 (67.6-100)	97.7 (88.2-99.6)	88.9 (60.2-99.7)	100 (93.7-100)
	ES	53	9	2	42	0	17.0	100 (70.1-100)	95.5 (84.9-98.7)	81.8 (56.9-97.4)	100 (93.5-100)
	PCyt	59	11	0	48	0	18.6	100 (74.1-100)	100 (92.6-100)	100 (75.6-100)	100 (93.9-100)
2	CVS	52	3	1	48	0	5.8	100 (43.9-100)	98.0 (89.3-99.6)	75.0 (28.5-99.2)	100 (95.8-100)
	ES	56	4	1	51	0	7.1	100 (51.0-100)	98.1 (89.9-99.7)	80.0 (40.5-99.4)	100 (95.6-100)
	PCyt	68	5	0	63	0	7.4	100 (56.6-100)	100 (94.3-100)	100 (58.3-100)	100 (96.0-100)
3	CVS	12	2	0	10	0	16.7	100 (34.2-100)	100 (72.2-100)	100 (32.1-100)	100 (85.6-100)
	ES	16	2	0	14	0	12.5	100 (34.2-100)	100 (78.5-100)	100 (31.5-100)	100 (89.3-100)
	PCyt	17	2	1	14	0	11.8	100 (34.2-100)	93.3 (70.2-98.8)	66.7 (19.9-98.8)	100 (89.5-100)
4	CVS	41	7	1	33	0	17.1	100 (64.6-100)	97.1 (85.1-99.5)	87.5 (57.3-99.6)	100 (92.2-100)
	ES	41	7	0	34	0	17.1	100 (64.6-100)	100 (89.8-100)	100 (66.7-100)	100 (92.2-100)
	PCyt	43	7	1	35	0	16.3	100 (64.6-100)	97.2 (85.8-99.5)	87.5 (57.2-99.6)	100 (92.6-100)
5	CVS	145	1	0	144	0	0.7	100 (20.7-100)	100 (97.4-100)	100 (6.4-100)	100 (99.3-100)
	ES	162	1	0	161	0	0.6	100 (20.7-100)	100 (97.7-100)	100 (6.4-100)	100 (99.4-100)
	PCyt	167	1	0	166	0	0.6	100 (20.7-100)	100 (97.7-100)	100 (6.4-100)	100 (99.4-100)
6	CVS	67	10	2	55	0	14.9	100 (72.2-100)	96.5 (88.1-99.0)	83.3 (59.2-98.2)	100 (94.8-100)
	ES	80	13	4	63	0	16.3	100 (77.2-100)	94.0 (85.6-97.7)	76.5 (57.1-92.2)	100 (95.3-100)
	PCyt	92	20	3	69	0	21.7	100 (83.9-100)	95.8 (88.5-98.6)	87.0 (70.4-97.0)	100 (95.5-100)
7	CVS	173	18	3	152	0	10.4	100 (82.4-100)	98.1 (94.5-99.3)	85.7 (67.7-96.7)	100 (97.9-100)
	ES	161	12	3	146	0	7.5	100 (75.8-100)	98.0 (94.2-99.3)	80.0 (58.3-95.4)	100 (97.9-100)
	PCyt	194	18	4	172	0	9.3	100 (82.4-100)	97.7 (94.3-99.1)	81.8 (64.1-94.3)	100 (98.1-100)
8	CVS	80	10	2	68	0	12.5	100 (72.2-100)	97.1 (90.2-99.2)	83.3 (59.0-98.2)	100 (95.8-100)
	ES	83	9	2	72	0	10.8	100 (70.1-100)	97.3 (90.7-99.3)	81.8 (56.3-97.4)	100 (96.1-100)
	PCyt	86	9	0	77	0	10.5	100 (70.1-100)	100 (95.2-100)	100 (71.4-100)	100 (96.2-100)
9	CVS	45	10	1	34	0	22.2	100 (72.2-100)	97.1 (85.5-99.5)	90.9 (65.7-99.7)	100 (91.9-100)
	ES	48	10	0	38	0	20.8	100 (72.2-100)	100 (90.8-100)	100 (74.0-100)	100 (92.5-100)
	PCyt	48	10	1	37	0	20.8	100 (72.2-100)	97.4 (86.5-99.5)	90.9 (65.6-99.7)	100 (92.5-100)

CI = confidence interval, CVS = clinician-collected vaginal swab, ES = endocervical swab, FN = false negative, FP = false positive, PCyt = PreservCyt Solution liquid Pap, Prev = prevalence, TN = true negative, TP = true positive.

[1]Score confidence interval.

[2]PPV 95% confidence interval computed from the exact 95% confidence interval for the positive likelihood ratio, NPV 95% confidence interval computed from the exact 95% confidence interval from the negative likelihood ratio.

Table 5 shows the sensitivity, specificity, PPV, and NPV of the Aptima Trichomonas vaginalis Assay on the Panther System and the prevalence of *T. vaginalis* (based on the infected status) in PreservCyt Solution liquid Pap specimens by cervical collection device. For PreservCyt Solution liquid Pap specimens, performance was similar across collection devices.

Table 5: Performance Characteristics of the Aptima Trichomonas vaginalis Assay in PreservCyt Solution Liquid Pap Specimens by Collection Device Type

Collection Device	n	TP	FP	TN	FN	Prev %	Sensitivity (95% CI)[1]	Specificity (95% CI)[1]	PPV % (95% CI)[2]	NPV % (95% CI)[2]
Broom-type Device	414	54	5	355	0	13.0	100 (93.4-100)	98.6 (96.8-99.4)	91.5 (82.4-97.1)	100 (99.0-100)
Spatula/Cytobrush	360	29	5	326	0	8.1	100 (88.3-100)	98.5 (96.5-99.4)	85.3 (71.5-94.7)	100 (99.0-100)

CI = confidence interval, FN = false negative, FP = false positive, Prev = prevalence, TN = true negative, TP = true positive.

[1]Score confidence interval.

[2]PPV 95% confidence interval computed from the exact 95% confidence interval for the positive likelihood ratio, NPV 95% confidence interval computed from the exact 95% confidence interval from the negative likelihood ratio.

Agreement of Aptima Trichomonas vaginalis Assay Results on the Panther System and the Tigris DTS System

It is recognized that device performance in an asymptomatic population is essential since the majority of individuals infected with *Trichomonas vaginalis* do not have symptoms. To further characterize performance of the assay in asymptomatic subjects, agreement between Aptima Trichomonas vaginalis Assay results on the Panther System and the Tigris DTS System was assessed using prospectively collected specimens from asymptomatic subjects. Women were enrolled from 6 US clinical sites, including obstetrics and gynecology, family planning, and STD clinics. One (1) vaginal swab, 1 endocervical swab, and 1 PreservCyt Solution liquid Pap specimen were collected from each subject. All specimens were clinician-collected. PreservCyt liquid Pap specimens were collected with a broom-type device or a spatula and cytobrush.

Aptima Trichomonas vaginalis Assay testing was conducted in accordance with the package insert instructions. Panther System testing was conducted at 3 sites (2 external laboratories and Hologic). Tigris DTS System testing was conducted at Hologic.

Eighteen (18) Aptima Trichomonas vaginalis Assay runs were initiated on the Panther System; all were valid. A total of 227 vaginal swab, 227 endocervical swab, and 227 PreservCyt Solution liquid Pap specimens were tested. Of these specimens, 1 vaginal swab specimen (0.4%, 1/227) had an initial invalid result due to hardware error. The specimen with an initial invalid result was retested and had a valid result.

Of the samples with final valid Aptima Trichomonas vaginalis Assay results on the Panther System, 227 vaginal swab, 227 endocervical swab, and 226 PreservCyt Solution liquid Pap specimens had valid, paired results on the Tigris DTS System.

Table 6 shows positive and negative percent agreements of Aptima Trichomonas vaginalis Assay results on the Panther System and the Tigris DTS System in each specimen type for asymptomatic subjects.

Table 6: Agreement between Aptima Trichomonas vaginalis Assay Results on the Panther System and the Tigris DTS System in Asymptomatic Subjects

Specimen Type	n	System					% Positive Agreement (95% CI)[2]	% Negative Agreement (95% CI)[2]
		Tigris + Panther +	Tigris - Panther +	Tigris - Panther -	Tigris + Panther -	Tigris Positivity		
CVS[1]	227	29	5	191	2	13.7	93.5 (79.3-98.2)	97.4 (94.2-98.9)
ES	227	28	1	198	0	12.3	100 (87.9-100)	99.5 (97.2-99.9)
PCyt	226	26	1	199	0	11.5	100 (87.1-100)	99.5 (97.2-99.9)

+ = positive, - = negative, CI = confidence interval, CVS = clinician-collected vaginal swab, ES = endocervical swab, PCyt = PreservCyt Solution liquid Pap,

[1]The 2 vaginal swab samples with positive Aptima Trichomonas vaginalis Assay results on the Tigris DTS System and negative results on the Panther System were from subjects whose other samples had negative results on both the Panther System and the Tigris DTS System.

[2]Score confidence interval.

RLU Distribution of Aptima Trichomonas vaginalis Controls

The distribution of the RLU values for the Aptima Trichomonas vaginalis Negative Control and the Aptima Trichomonas vaginalis Positive Control from all valid Aptima Trichomonas vaginalis Assay runs performed during the clinical performance study of the Aptima Trichomonas vaginalis Assay on the Panther System is presented in Table 7.

Table 7: RLU Distribution of Aptima Trichomonas vaginalis Negative and Positive Controls

Control	Statistic	Total RLU (x1000)
Negative	N	22
	Mean	1.3
	SD	0.99
	Median	1.0
	Minimum	0
	Maximum	5
	CV%	75.5
Positive	N	22
	Mean	1262.3
	SD	45.89
	Median	1276.0
	Minimum	1168
	Maximum	1322
	CV%	3.6

RLU = relative light unit.

Note: The RLU value reported by the software was the basis for analysis. The reported RLU value is the total measured RLU divided by 1000 with the digits after the decimal point truncated.

Reproducibility Study

Aptima Trichomonas vaginalis Assay reproducibility was evaluated on the Panther System at two external US laboratories and at Hologic. Testing was performed using two lots of assay reagents and a total of six operators (two at each site). At each site, testing was performed over at least 6 days.

Reproducibility panel members were created by using negative PreservCyt Solution liquid Pap specimens with specimen transport medium. The positive panel members were created by spiking the PreservCyt Solution liquid Pap matrix with the appropriate amount of *T. vaginalis* lysate. Final *T. vaginalis* concentrations ranged from 0.003 trichomonads/mL to 1 trichomonads/mL.

Table 8 presents, for each panel member, RLU data in terms of mean, standard deviation (SD), and coefficient of variation (CV) between sites, between operators, between lots, between runs, within runs, and overall (Total). Percent agreement with expected results is also shown. Samples with valid results were included in the analyses.

Table 8: Aptima Trichomonas vaginalis Assay Reproducibility Study

Conc	Target Conc (TV/mL)	N	Agmt (%)	Mean RLU	Between Sites		Between Operators		Between Lots		Between Runs		Within Runs		Totals	
					SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)	SD	CV (%)
Neg	N/A	108	99.1	23.5	0.0	0.0	2.7	11.6	0.0	0.0	0.0	0.0	37.5	159.7	37.6	160.1
HNeg	0.003	108	90.7	69.3	5.0	7.3	4.5	6.5	6.1	8.8	14.8	21.4	16.0	23.1	23.6	34.1
MPos	0.02	108	97.2	348.1	30.3	8.7	33.1	9.5	33.1	9.5	77.0	22.1	62.9	18.1	114.0	32.8
HPos	1.00	108	100	1185.5	0.0	0.0	17.0	1.4	0.0	0.0	28.0	2.4	34.2	2.9	47.4	4.0

Agmt = agreement, Conc = concentration, CV = coefficient of variation, HNeg = high negative, HPos = high positive, MPos = moderate positive, Neg = negative, RLU = relative light units, SD = standard deviation.

Note: The RLU value reported by the software is the total measured RLU divided by 1000 with the digits after the decimal point truncated.

Variability from some factors may have been numerically negative. This occurred if the variability due to those factors was very small. In these cases, SD and CV are shown as 0.

Panther System Analytical Performance

Analytical Sensitivity

Sensitivity panels were prepared with two strains of *T. vaginalis* (one Metronidazole-susceptible strain and one Metronidazole-resistant strain). Testing showed greater than 95% positivity in both strains of *T. vaginalis* for panels containing 0.01 TV/mL in PreservCyt liquid Pap specimen matrix and panels containing 0.003 TV/mL in swab specimen matrix.

Cross-Reactivity in the Presence of Microorganisms

Specificity

Specificity of the Aptima Trichomonas vaginalis Assay was evaluated by testing various microorganisms, including common flora of the genitourinary tract, opportunistic organisms, and closely related organisms. Testing was conducted in specimen transport medium (STM) and PreservCyt in STM with 25 replicates of each isolate. The list of organisms and the concentrations tested are provided in Table 9. No cross-reactivity or significant effect on Aptima Trichomonas vaginalis Assay specificity was observed with any of the organisms tested.

Sensitivity

Sensitivity of the Aptima Trichomonas vaginalis Assay was evaluated by testing the same organisms (Table 9) in STM and PreservCyt in STM spiked with *T. vaginalis* lysate to a final concentration of 0.01 TV/mL (25 replicates of each isolate). Sensitivity of the Aptima Trichomonas vaginalis Assay was not significantly affected by the presence of the microorganisms tested, except in the presence of *Trichomonas tenax* and *Pentatrichomonas hominis* (where lower signal outputs were observed). *T. tenax* is a commensal of the oral cavity and *Pentatrichomonas hominis* is a commensal of the large intestine.

Table 9: Microorganisms Tested in the Aptima Trichomonas vaginalis Assay

Microorganism	Concentration	Microorganism	Concentration
Acinetobacter Iwoffi	$1x10^6$ CFU/mL	HPV 16	$2.5x10^6$ copies/mL
Actinomyces israelii	$1x10^6$ CFU/mL	HPV 6	$2.5x10^6$ copies/mL
Atopobium vaginae	$1x10^6$ CFU/mL	*Klebsiella pneumoniae*	$1x10^6$ CFU/mL
Bacteroides fragilis	$1x10^6$ CFU/mL	*Lactobacillus acidophilus*	$1x10^6$ CFU/mL
Bifidobacterium adolescentis	$1x10^6$ CFU/mL	*Lactobacillus crispatus*	$1x10^6$ CFU/mL
Campylobacter jejuni	$1x10^6$ CFU/mL	*Listeria monocytogenes*	$1x10^6$ CFU/mL
Candida albicans	$1x10^6$ CFU/mL	*Mobiluncus curtisii*	$1x10^6$ CFU/mL
Chlamydia trachomatis	$1x10^6$ IFU/mL	*Mycoplasma genitalium*	$2.5 x10^6$ copies/mL
Clostridium difficile	$1x10^6$ CFU/mL	*Mycoplasma hominis*	$1x10^6$ CFU/mL
Corynebacterium genitalium	$1x10^6$ CFU/mL	*Neisseria gonorrhoeae*	$1x10^6$ CFU/mL
Cryptococcus neoformans	$1x10^6$ CFU/mL	*Pentatrichomonas hominis*	$1x10^6$ cells/mL
Cytomegalovirus	$2x10^5$ $TCID_{50}$/mL	*Peptostreptococcus magnus*	$1x10^6$ CFU/mL
Dientamoeba fragilis	$1x10^6$ CFU/mL	*Prevotella bivia*	$1x10^6$ CFU/mL
Enterobacter cloacae	$1x10^6$ CFU/mL	*Propionibacterium acnes*	$1x10^6$ CFU/mL
Enterococcus faecalis	$1x10^6$ CFU/mL	*Proteus vulgaris*	$1x10^6$ CFU/mL
Escherichia coli	$1x10^6$ CFU/mL	*Pseudomonas aeruginosa*	$1x10^6$ CFU/mL
Gardnerella vaginalis	$1x10^6$ CFU/mL	*Staphylococcus aureus*	$1x10^6$ CFU/mL
Haemophilus ducreyi	$1x10^6$ CFU/mL	*Staphylococcus epidermidis*	$1x10^6$ CFU/mL
Herpes simplex virus I	$2x10^5$ $TCID_{50}$/mL	*Streptococcus agalactiae*	$1x10^6$ CFU/mL
Herpes simplex virus II	$2x10^5$ $TCID_{50}$/mL	*Trichomonas tenax*	$1x10^6$ cells/mL
HIV-1	$2.5x10^6$ copies/mL	*Ureaplasma urealyticum*	$1x10^6$ CFU/mL

Interference

The following substances were individually spiked into STM and PreservCyt in STM for a final concentration of 1% (vol/vol or wt/vol): personal lubricants, personal deodorants, spermicides, anti-fungals, intravaginal hormones, porcine gastric mucus, seminal fluid from 25 donors, and whole blood (10% final concentration). Glacial acetic acid was tested by spiking into PreservCyt-STM (10% final concentration). Samples with each interfering substance alone as well as samples spiked with *T. vaginalis* lysate to a final concentration of 0.01 TV/mL were tested.

Testing results yielded no false positive results for all substances tested (100% specificity).

No interference with the detection of *T. vaginalis* at concentrations of 0.01 TV/mL (≥ 95% sensitivity) was observed with any of the substances tested with the exception of Astroglide personal lubricant, porcine gastric mucus, and glacial acetic acid. Astroglide personal lubricant and glacial acetic acid did not interfere with the detection of *T. vaginalis* when tested at a concentration of 0.3 TV/mL (100% sensitivity). Porcine gastric mucus did not interfere with the detection of *T. vaginalis* when tested at a concentration of 1 TV/mL (100% sensitivity).

Carryover

To establish that the Panther System minimizes the risk of false positive results arising from carryover contamination, a multi-day analytical study was conducted using spiked panels on three Panther Systems with one lot of Aptima Trichomonas vaginalis Assay reagents. The study used > 20% high-target *T. vaginalis* samples containing 10,000 TV/mL, which were placed among negative samples containing STM. Over the course of the study, 698 high-target samples

and 2,266 negative samples were tested across the three Panther Systems. There were 0 false positive results for a 0% carryover contamination rate. These results demonstrate that carryover contamination is minimized on the Panther System.

Specimen Stability

Data to support the recommended shipping and storage conditions for the vaginal swab and PreservCyt liquid Pap specimens were generated with negative clinical specimens spiked with *T. vaginalis*. Greater than 98% positivity was observed in all matrices (vaginal swab and PreservCyt liquid Pap) at all times and temperatures tested confirming the validity of the maximum storage times and temperatures described in *Specimen Collection and Storage*.

Bibliography

1. **Weinstock, H., S. Berman, and W. Cates Jr.** 2004. Sexually transmitted diseases among American youth: incidence and prevalence estimates, 2000. Perspect. Sex. Reprod. Health **36**(1):6-10.
2. **Soper, D.** 2004. Trichomoniasis: under control or undercontrolled? Am. J. Obstet. Gynecol. **190**(1):281-290.
3. **Cotch, M. F., J. G. Pastorek II, R. P. Nugent, S. L. Hillier, R. S. Gibbs, D. H. Martin, et al.** 1997. *Trichomonas vaginalis* associated with low birth weight and preterm delivery. The Vaginal Infections and Prematurity Study Group. Sex. Transm. Dis. **24**:353-360.
4. **Sorvillo, F. J., A. Kovacs, P. Kerndt, A. Stek, L. Muderspach, and L. Sanchez-Keeland.** 1998. Risk factors for trichomoniasis among women with HIV infection at a public clinic in Los Angeles County; Implications for HIV prevention. Am. J. Trop. Med. Hyg. **58**:495-500.
5. **Niccolai, L. M., J. J. Kopicko, A. Kassie, H. Petros, R. A. Clark, and P. Kissinger.** 2000. Incidence and predictors of reinfection with *Trichomonas vaginalis* in HIV-infected women. Sex. Transm. Dis. **27**:284-288.
6. **Nye, M. B., J. R. Schwebke, and B. A. Body.** 2009. Comparison of Aptima Trichomonas vaginalis transcription-mediated amplification to wet mount microscopy, culture, and polymerase chain reaction for diagnosis of trichomoniasis in men and women. Am. J. Obstet. Gynecol. **200**:188.e1-188.e7.
7. **Wendel, K. A., E. J. Erbelding, C. A. Gaydos, and A. M. Rompalo.** 2002. *Trichomonas vaginalis* polymerase chain reaction compared with standard diagnostic and therapeutic protocols for detection and treatment of vaginal trichomoniasis. Clin. Infect. Dis. **35**(5):576-580

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2016-03

PANTHER DESCRIPTION OF SERVICE OPTIONS

STANDARD SERVICE OPTION

SERVICES INCLUDED. The services included under the Standard Service option are the following:

- Labor, travel expenses, and any necessary replacement parts (excluding disposables which include, but are not limited to, tips, MTU's, TTU's, waste bags, and bench covers), during normal business hours. Normal working hours are defined as Monday-Friday, 8:30 a.m. - 5:30 p.m. local time (excluding Hologic holidays).
- Preventative maintenance by Hologic service technician according to operator's or user's manual (normal working hours Monday through Friday).
- Equipment repair for reasons other than those listed below under "Services Excluded".
- Access to Hologic Technical Support telephone support, Monday through Friday, 5:00 a.m. to 5:00 p.m. Pacific Standard Time (excluding Hologic holidays).
- Telephone number for Technical Support: 888-484-4747
- Factory authorized updates or modifications, including parts.
- Up to (2) Pro360 and/or LIS configuration changes
  Exception: If service technician already on-site or has service scheduled, the LIS and/or Pro360 can be modified at that time at no charge and can exceed the 2 allotted.

Service Representative Dispatch and PRO360º REMOTE DIAGNOSTICS

- Representative on site within 24 hours (Monday – Friday) if PRO360º Remote Diagnostics Management is installed.
- Representative on site within 48 hours (Monday – Friday) if PRO360º Remote Diagnostics is not installed. Service response times are predicated upon the Equipment operator being willing and able to transfer Equipment log files to Hologic when instructed by Hologic Technical Support using the protocol described in the Equipment Operator's Manual.

SERVICES EXCLUDED. The services excluded under the Standard Service option are the following:

- Any repair required because of causes other than use of the Equipment pursuant to the operator's or user's manual. Such causes include, but are not limited to: misuse, abuse, improper use, casualty loss, neglect, reprogramming error, malfunction or failure of environmental control Equipment, electrical Equipment malfunction or failure, repair maintenance, modification, relocation, or reinstallation by other than Hologic authorized personnel, installation of commercial or non-Equipment software, use of any other tips on the Equipment other than TECAN Tips, or acts of God, fire, flood, earthquake, or other natural causes.
- Routine tasks, other than those performed by Hologic during preventative maintenance visits, covered in the operator's or user's manual, such as cleaning and maintenance.
- Supply items (including, but not limited to, those items listed in the package insert or manual as "materials required but not provided," TECAN Tips, bleach, squirt bottles, paper towels, and other such items that are needed for general use but not specifically by the Equipment) and consumable items.
- Relocation of Equipment. All equipment to be moved during normal working hours.
- LIS and Pro360 configuration changes which exceed the above allotted (2) (e.g. urgent requests to change Pro360/LIS).

  Note: Labor, travel, and material charges for all of the excluded services will be billed at rates prevailing at the time of service.

CUSTOMER OBLIGATIONS. Prior to any shipment of repair parts or visit by Hologic service representative, Customer must perform all pertinent diagnostic programs, tests, simple/ basic troubleshooting and provide an accurate description of the failure/error.

REPLACED OR REMOVED PARTS. All parts replaced or removed under this Exhibit become the property of Hologic.

Service Plans

PANTHER®

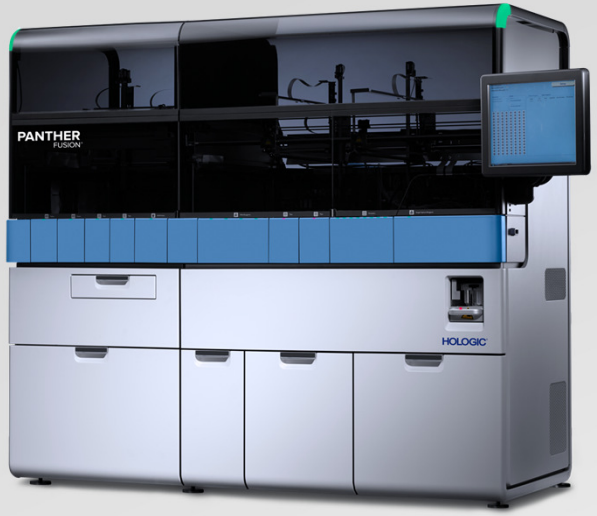
	On-Demand Service	PM only Service Plan	Standard Service Plan	Standard Plus Service Plan	Premium Service Plan	Premium Plus Service Plan
Technical Support Phone	Monday-Friday 5:00 am - 5:00 pm PT (Excluding company holidays)	Monday-Friday 5:00 am - 5:00 pm PT (Excluding company holidays)	Monday-Friday 5:00 am - 5:00 pm PT (Excluding company holidays)	24/7	24/7	24/7
Pro36o™ Remote Support	Monday-Friday 5:00 am - 5:00 pm PT (Excluding company holidays)	Monday-Friday 5:00 am - 5:00 pm PT (Excluding company holidays)	Monday-Friday 5:00 am - 5:00 pm PT (Excluding company holidays)	24/7	24/7	24/7
On-site Service (with Pro360)	$5,000/visit plus parts* After hrs/wknds $7,500 plus parts	$5,000/visit plus parts* After hrs/wknds $7,500 plus parts	On-site within 24 hours*	On-site within 24 hours*	On-site within 18 hours	On-site within 18 hours
On-site Application Service	$5,000/visit plus parts* After hrs/wknds $7,500 plus parts	$5,000/visit plus parts* After hrs/wknds $7,500 plus parts	On-site within 48 hours*	On-site within 48 hours*	On-site within 48 hours	On-site within 48 hours
Pro360 & LIS Updates	$1,800/service plus parts*	$1,800/service plus parts*	2x per system*	2x per system*	Unlimited*	Unlimited*
Customer Care Call	$5,000/visit	$5,000/visit	2x per year* (Provided)	2x per year* (Provided)	2x per year* (Provided)	2x per year* (Provided)
Preventive Maintenance	$8,000 per PM parts included 2 PM visits in 12 months	2x per year* (Provided)	2x per year* (Provided)	2x per year* (Provided)	2x per year* (Provided)	2x per year* (Provided)
Assay Verification Support	$5,000	$5,000	$5,000	$5,000	$5,000	Included
Additional Operator Training	$5,000	$5,000	$5,000	$5,000	$5,000	Included
Factory Authorized Updates/Modifications	Included Additional updates $4,500 plus parts	Included Additional updates $4,500 plus parts	Included	Included	Included	Included
System Relocation	Within facility $4,500* New facility $5,000* After hrs/wknds add $2,500	Within facility $4,500* New facility $5,000* After hrs/wknds add $2,500	Within facility $4,500* New facility $5,000* After hrs/wknds add $2,500	Within facility $4,500* New facility $5,000* After hrs/wknds add $2,500	Within facility $4,500* New facility $5,000* After hrs/wknds add $2,500	Within facility $4,500* New facility $5,000* After hrs/wknds add $2,500
Purchased Systems	Rates as listed	$14,500 per year	$20,500 per year	$23,000 per year	$29,500 per year	$38,000 per year
Reagent Rentals			Included	$2,500 per year	$9,000 per year	$17,500 per year

Please contact your Hologic representative for more information on service plans.

*On-site service available Monday-Friday, 8:30 am - 5:30 pm (local time) excluding company holidays.

The Panther Fusion® system offers even more options for consolidation

PANTHER FUSION®



Panther® Assays (TMA/RT TMA)

Panther Fusion® Assays (RT PCR)

CURRENT TARGETS

CT/NG
HPV
HPV 16 18/45
Trichomoniasis
Zika virus (EUA)
HSV 1 & 2
HIV-1 Quant
HCV Quant Dx
HBV Quant
Mycoplasma genitalium

Flu A/B/RSV
Paraflu
AdV/hMPV/RV
GBS

Open Access™ functionality to run lab developed tests

CURRENTLY IN DEVELOPMENT*

CMV
Bacterial Vaginosis
Candida/Trichomoniasis

Bordetella
GI Panel

POTENTIAL FUTURE TARGETS

Infectious diseases
Respiratory pathogens
GC resistance and
Mycoplasma genitalium
resistance

Assay consolidation on a fully automated platform allows ***greater # of tests per FTE***

*Under development and not currently for sale in U.S.

The Aptima Zika Virus assay: This test has not been FDA cleared or approved; this test has been authorized by FDA under an EUA for use by authorized laboratories; this test has been authorized only for the detection of RNA from Zika virus and diagnosis of Zika virus infection, not for any other viruses or pathogens; and this test is only authorized for the duration of the declaration that circumstances exist justifying the authorization of the emergency use of in vitro diagnostic tests for detection of Zika virus and/or diagnosis of Zika virus infection under section 564(b)(1) of the Act, 21 U.S.C.§ 360bbb-3(b)(1), unless the authorization is terminated or revoked sooner.

Aptima Combo 2® Assay (Panther® System)

For *in vitro* diagnostic use.

Rx only.

General Information

Intended Use

The Aptima Combo 2® Assay is a target amplification nucleic acid probe test that utilizes target capture for the *in vitro* qualitative detection and differentiation of ribosomal RNA (rRNA) from *Chlamydia trachomatis* (CT) and/or *Neisseria gonorrhoeae* (GC) to aid in the diagnosis of chlamydial and/or gonococcal urogenital disease using the Panther® System as specified.

On the Panther System, the assay may be used to test the following specimens from symptomatic and asymptomatic individuals: clinician-collected endocervical, vaginal and male urethral swab specimens, clinician-collected gynecological specimens collected in the PreservCyt® Solution, patient-collected vaginal swab specimens,[1] and female and male urine specimens.

[1]Patient-collected vaginal swab specimens are an option for screening women when a pelvic exam is not otherwise indicated. The vaginal and multitest swab specimen collection kits are not for home use.

Summary and Explanation of the Test

Chlamydia trachomatis (CT) and *Neisseria gonorrhoeae* (GC) infections are two of the most common sexually transmitted infections worldwide. In the United States alone, an estimated 1,598,354 (497 cases per 100,000 population) new cases of CT and 468,514 (146 per 100,000 population) new cases of GC infections were reported to the Centers for Disease Control in 2016 (6).

Chlamydiae are nonmotile, gram-negative, obligate intracellular bacteria. The CT species is comprised of fifteen serovars (A, B, Ba, C, D, E, F, G, H, I, J, K, L1, L2 and L3) that can cause disease in humans (31). The serovars D through K are the major cause of genital chlamydial infections in men and women (23). *C. trachomatis* can cause nongonococcal urethritis, epididymitis, proctitis, cervicitis, acute salpingitis, and Pelvic Inflammatory Disease (PID) (3, 16, 25, 26). *C. trachomatis* infections are often asymptomatic in both males and females. Children born to infected mothers are at significantly higher risk for inclusion conjunctivitis and chlamydial pneumonia (1, 12, 24).

Historically, several methods for CT detection have been utilized in the clinical laboratory, including cell culture, direct fluorescent antibody testing, and enzyme immunoassay. More recent methodologies for CT detection include direct DNA probe assays and nucleic acid amplification test (NAAT) DNA probe assays. Cell culture was once considered to be the “gold standard” for detection of CT. Culture is quite specific, but scientific publications have demonstrated that the NAAT DNA probe technologies have a higher clinical sensitivity than culture (2, 10, 18, 27). Due to its lower clinical sensitivity and variable performance between laboratories, culture has been replaced in many laboratories by direct DNA probe assays and NAATs.

N. gonorrhoeae is the causative agent of gonorrheal disease. *N. gonorrhoeae* are non-motile, gram-negative diplococci. The majority of gonorrheal infections are uncomplicated lower genital tract infections and may be asymptomatic. However, if left untreated in women, infections can ascend and cause PID, which can manifest as endometritis, salpingitis, pelvic peritonitis, and tubo-ovarian abscesses. A smaller percentage of persons with gonococcal infections may develop Disseminated Gonococcal Infection (DGI) (15, 21).

Conventional diagnosis of GC infection requires isolation of the organism on selective media or the observation of diplococci in Gram stained smears (17). Culture methods can have good clinical sensitivity, but are highly dependent on proper specimen handling. Improper specimen

storage and transport can result in the loss of organism viability and yield false negative results. Poor sampling technique, toxic sampling materials, and the inhibition of growth by components of body secretions can also result in false negative results (8, 19). Commonly used non-culture methods for GC detection include direct DNA probe tests and NAATs.

First generation NAATs for CT and GC have technological issues that have limited their performance. These issues include cumbersome specimen processing and specimen inhibition that can yield false negative results (7, 11, 14, 20, 22, 28, 29, 30). The Aptima Combo 2 Assay is a second generation NAAT that utilizes target capture, Transcription-Mediated Amplification (TMA), and Dual Kinetic Assay (DKA) technologies to streamline specimen processing, amplify target rRNA, and detect amplicon, respectively. Studies comparing performance and specimen inhibition of various amplification systems have demonstrated the benefits of target capture, TMA®, and DKA technologies (9, 13). The Aptima Combo 2 Assay on the Panther System qualitatively detects CT and/or GC rRNA in clinician-collected endocervical, vaginal, and male urethral swab specimens, patient-collected vaginal swab specimens, PreservCyt Solution liquid Pap specimens, and female and male urine specimens from symptomatic and asymptomatic individuals.

Principles of the Procedure

The Aptima Combo 2 Assay combines the technologies of target capture, TMA, and DKA.

Specimens are collected and transferred into their respective specimen transport tubes. The transport solutions in these tubes release the rRNA targets and protect them from degradation during storage. When the Aptima Combo 2 Assay is performed in the laboratory, the target rRNA molecules are isolated from specimens by use of capture oligomers via target capture that utilizes magnetic microparticles. The capture oligomers contain sequences complementary to specific regions of the target molecules as well as a string of deoxyadenosine residues. A separate capture oligomer is used for each target. During the hybridization step, the sequence specific regions of the capture oligomers bind to specific regions of the target molecules. The capture oligomer:target complex is then captured out of solution by decreasing the temperature of the reaction to room temperature. This temperature reduction allows hybridization to occur between the deoxyadenosine region on the capture oligomer and the poly-deoxythymidine molecules that are covalently attached to the magnetic particles. The microparticles, including the captured target molecules bound to them, are pulled to the side of the reaction vessel using magnets and the supernatant is aspirated. The particles are washed to remove residual specimen matrix that may contain amplification reaction inhibitors. After the target capture steps are completed, the specimens are ready for amplification.

Target amplification assays are based on the ability of complementary oligonucleotide primers to specifically anneal and allow enzymatic amplification of the target nucleic acid strands. The Aptima Combo 2 Assay replicates a specific region of the 23S rRNA from CT and a specific region of the 16S rRNA from GC via DNA intermediates. A unique set of primers is used for each target molecule. Detection of the rRNA amplification product sequences (amplicon) is achieved using nucleic acid hybridization. Single-stranded chemiluminescent DNA probes, which are complementary to a region of each target amplicon, are labeled with different acridinium ester molecules. The labeled DNA probes combine with amplicon to form stable RNA:DNA hybrids. The Selection Reagent differentiates hybridized from unhybridized probe, eliminating the generation of signal from unhybridized probe. During the detection step, light emitted from the labeled RNA:DNA hybrids is measured as photon signals in a luminometer, and are reported as Relative Light Units (RLU). In DKA, differences in the kinetic profiles of the CT and GC labeled probes allow for the differentiation of signal; kinetic profiles are derived from measurements of

photon output during the detection read time. The chemiluminescent detection reaction for CT signal has very rapid kinetics and has the “flasher” kinetic type. The chemiluminescent detection reaction for GC signal is relatively slower and has the “glower” kinetic type. Assay results are determined by a cut-off based on the total RLU and the kinetic curve type.

Warnings and Precautions

A. For *in vitro* diagnostic use.

B. For additional specific warnings, precautions and procedures to control contamination for the Panther System, consult the *Panther System Operator’s Manual*.

Laboratory Related

C. Use only supplied or specified disposable laboratory ware.

D. Use routine laboratory precautions. Do not eat, drink or smoke in designated work areas. Wear disposable, powderless gloves, protective eye wear, and laboratory coats when handling specimens and kit reagents. Wash hands thoroughly after handling specimens and kit reagents.

E. **Warning: Irritant and Corrosive:** Avoid contact of Auto Detect 2 with skin, eyes and mucous membranes. If this fluid comes into contact with skin or eyes, wash the affected area with water. If this fluid spills, dilute the spill with water before wiping it dry.

F. Work surfaces, pipettes, and other equipment must be regularly decontaminated with 2.5% to 3.5% (0.35M to 0.5M) sodium hypochlorite solution.

Specimen Related

G. This assay has been cleared for the following specimens on the Panther System:

- Clinician-collected endocervical, vaginal, and male urethral swab specimens
- Female and male urine specimens
- Clinician-collected PreservCyt Solution liquid Pap specimens
- Patient-collected vaginal swab specimens

Only specimens collected with the following specimen collection kits have been cleared on the Panther System:

- Aptima® Unisex Swab Specimen Collection Kit for Endocervical and Male Urethral Swab Specimens
- Aptima Urine Collection Kit for Male and Female Urine Specimens
- Aptima Vaginal Swab Specimen Collection Kit
- Aptima Multitest Swab Specimen Collection Kit
- Aptima Specimen Transfer Kit (for use with gynecologic samples collected in PreservCyt Solution)

Gynecologic samples collected for preparation using the ThinPrep® 2000 System should be collected using broom-type or endocervical brush/plastic spatula combination collection devices.

H. Expiration dates listed on the collection kits pertain to the collection site and not the testing facility. Samples collected any time prior to the expiration date of the collection kit, and transported and stored in accordance with the package insert, are valid for testing even if the expiration date on the collection tube has passed.

I. The PreservCyt Solution has been validated as an alternative medium for testing with Aptima Combo 2 Assay. PreservCyt Solution liquid Pap specimens processed using the ThinPrep 3000 Processor or other instruments have not been evaluated to test for *Chlamydia trachomatis* and *Neisseria gonorrhoeae* using the Aptima Combo 2 Assay.

J. After urine has been added in the urine transport tube, the liquid level must fall between the two black indicator lines on the tube label. Otherwise, the specimen must be rejected.

K. Maintain proper storage conditions during specimen shipping to ensure the integrity of the specimen. Specimen stability under shipping conditions other than those recommended has not been evaluated.

L. Specimens may be infectious. Use Universal Precautions when performing this assay. Proper handling and disposal methods should be established by the laboratory director. Only personnel adequately trained in handling infectious materials should be permitted to perform this diagnostic procedure.

M. Avoid cross-contamination during the specimen handling steps. Specimens can contain extremely high levels of organisms. Ensure that specimen containers do not contact one another, and discard used materials without passing over open containers. Change gloves if they come in contact with specimen.

N. If the lab receives a swab specimen transport tube with no swab, two swabs, a cleaning swab, or a swab not supplied by Hologic, the specimen must be rejected. Prior to rejecting a swab transport tube with no swab, verify that it is not an Aptima Specimen Transfer Tube as this specimen transport tube will not contain a swab.

O. For PreservCyt Solution liquid Pap specimens, collect according to the manufacturer's instructions. Aliquots subsequently removed from the PreservCyt vial for testing by the Aptima Combo 2 Assay should be processed using only the Aptima Specimen Transfer Kit.

P. Upon piercing, liquid can discharge from Aptima transport tube caps under certain conditions. Follow instructions in the *Panther System Test Procedure* to prevent this occurrence.

Assay Related

Q. Do not use this kit after its expiration date.

R. Do not interchange, mix, or combine assay reagents from kits with different lot numbers. Aptima controls and assay fluids (Panther System) can be from different lot numbers.

S. Some reagents of this kit are labeled with risk and safety symbols.

Note: *For hazard communication information, refer to the Safety Data Sheet Library at www.hologicsds.com.*

US Hazard Information	
	**Selection Reagent** ***BORIC ACID 1-5%*** **WARNING** H315 - Causes skin irritation H319 - Causes serious eye irritation P264 - Wash face, hands and any exposed skin thoroughly after handling P280 - Wear protective gloves/protective clothing/eye protection/face protection P305 + P351 + P338 - IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing P337 + P313 - If eye irritation persists: Get medical advice/attention P302 + P352 - IF ON SKIN: Wash with plenty of soap and water P332 + P313 - If skin irritation occurs: Get medical advice/attention P362 - Take off contaminated clothing and wash before reuse

Reagent Storage and Handling Requirements

A. The following reagents are stable when stored at 2°C to 8°C (refrigerated):

Aptima Combo 2 Amplification Reagent

Aptima Combo 2 Enzyme Reagent

Aptima Combo 2 Probe Reagent

Aptima Combo 2 Target Capture Reagent B

Aptima Positive Control, CT / Negative Control, GC

Aptima Positive Control, GC / Negative Control, CT

B. The following reagents are stable when stored at 2°C to 30°C:

Aptima Combo 2 Amplification Reconstitution Solution

Aptima Combo 2 Enzyme Reconstitution Solution

Aptima Combo 2 Probe Reconstitution Solution

Aptima Combo 2 Selection Reagent

C. The following reagents are stable when stored at 15°C to 30°C (room temperature):

Target Capture Reagent

Aptima Wash Solution

Aptima Buffer for Deactivation Fluid

Aptima Oil Reagent

D. Working Target Capture Reagent (wTCR) is stable for 30 days when stored at 15°C to 30°C. Do not refrigerate.

E. After reconstitution, the Enzyme Reagent, Amplification Reagent, and Probe Reagent are stable for 30 days when stored at 2°C to 8°C.

F. Discard any unused reconstituted reagents and wTCR after 30 days or after the Master Lot expiration date, whichever comes first.

G. Controls are stable until the date indicated on the vials.

H. Reagents stored on-board the Panther System have 72 hours of on-board stability.

I. The Probe Reagent and Reconstituted Probe Reagent are photosensitive. Store the reagents protected from light. The specified reconstituted stability is based on 12 hours exposure of the Reconstituted Probe Reagent to two 60W fluorescent bulbs, at a distance of 17 inches (43 cm), and temperature less than 30°C. Light exposure of the Reconstituted Probe Reagent should be limited accordingly.

J. Upon warming to room temperature, some control tubes may appear cloudy or contain precipitates. Cloudiness or precipitation associated with controls does not affect control performance. The controls may be used whether they are clear or cloudy/precipitated. If clear controls are desired, solubilization may be expedited by incubating them at the upper end of the room temperature range (15°C to 30°C).

K. Do not freeze the reagents.

Specimen Collection and Storage

The Aptima Combo 2 Assay is designed to detect the presence of CT and GC in the following specimens: endocervical and male urethral swab specimens, vaginal swab specimens, PreservCyt Solution liquid Pap specimens, and female and male urine specimens.

A. Instructions for collection:

Refer to the appropriate specimen collection kit package insert for collection instructions.

B. Specimen transport and storage before testing:

1. Swab specimens:
   a. After collection, transport and store the swab in the swab specimen transport tube at 2°C to 30°C until tested. Specimens must be assayed with the Aptima Combo 2 Assay within 60 days of collection. If longer storage is needed, freeze at -20°C to -70°C for up to 12 months after collection (see *Specimen Stability Studies*).
2. Urine specimens:
   a. Urine samples that are still in the primary collection container must be transported to the lab at 2°C to 30°C. Transfer the urine sample into the Aptima urine specimen transport tube within 24 hours of collection. Store at 2°C to 30°C and test within 30 days of collection.
   b. After collection, transport the processed urine specimens in the Aptima urine specimen transport tube at 2°C to 30°C and store at 2°C to 30°C until tested. Processed urine specimens should be assayed with the Aptima Combo 2 Assay within 30 days of collection. If longer storage is needed, freeze at -20°C to -70°C for up to 116 days after collection (see *Specimen Stability Studies*).
3. PreservCyt Solution liquid Pap specimens:
   a. PreservCyt Solution liquid Pap specimens intended for CT and/or GC testing must be processed for cytology and/or transferred to an Aptima Specimen Transfer tube within 30 days of collection when stored at 2°C to 30°C (see *Specimen Stability Studies*).
   b. If the ThinPrep Aliquot Removal procedure will be used, refer to the *ThinPrep 2000 or ThinPrep 3000 Processor Operator's Manual - Addendum* for instructions on aliquot

removal. Transfer 1 mL of the removed aliquot into an Aptima Specimen Transfer tube according to the instructions in the Aptima Specimen Transfer Kit package insert.

c. If testing the specimen after processing using the ThinPrep 2000 Processor, process the PreservCyt Solution liquid Pap specimen in accordance with the *ThinPrep 2000 Processor Operator's Manual* and the Aptima Specimen Transfer Kit package insert. Transfer 1 mL of the fluid remaining in the PreservCyt Solution vial into an Aptima Specimen Transfer tube according to the instructions in the Aptima Specimen Transfer Kit package insert.

d. Once the PreservCyt Solution liquid Pap specimen is transferred to the Aptima Specimen Transfer tube, the specimen must be assayed with the Aptima Combo 2 Assay within 30 days when stored at 2°C to 8°C or 14 days when stored at 15°C to 30°C. If longer storage is needed, freeze at -20°C to -70°C for up to 12 months after transfer (see *Specimen Stability Studies*).

C. Specimen storage after testing:

1. Specimens that have been assayed must be stored upright in a rack.
2. The specimen transport tubes should be covered with a new, clean plastic film or foil barrier.
3. If assayed samples need to be frozen or shipped, remove penetrable cap and place new non-penetrable caps on the specimen transport tubes. If specimens need to be shipped for testing at another facility, recommended temperatures must be maintained. Prior to uncapping previously tested and recapped samples, specimen transport tubes must be centrifuged for 5 minutes at 420 Relative Centrifugal Force (RCF) to bring all of the liquid down to the bottom of the tube. **Avoid splashing and cross-contamination**.

Note: *Specimens must be shipped in accordance with applicable national and international transportation regulations.*

Panther System

Reagents for the Aptima Combo 2 Assay for CT and GC are listed below for the Panther System. Reagent Identification Symbols are also listed next to the reagent name.

Reagents and Materials Provided

Aptima Combo 2 Assay Kit

100 tests (2 boxes and 1 Controls kit) (Cat. No. 302923)

250 tests (2 boxes and 1 Controls kit) (Cat. No. 303094)

Aptima Combo 2 Refrigerated Box (Box 1 of 2)
(store at 2°C to 8°C upon receipt)

Symbol	Component	Quantity 250 test kit	Quantity 100 test kit
A	**Aptima Combo 2 Amplification Reagent** *Non-infectious nucleic acids dried in buffered solution containing < 5% bulking agent.*	1 vial	1 vial
E	**Aptima Combo 2 Enzyme Reagent** *Reverse transcriptase and RNA polymerase dried in HEPES buffered solution containing < 10% bulking reagent.*	1 vial	1 vial
P	**Aptima Combo 2 Probe Reagent** *Non-infectious chemiluminescent DNA probes dried in succinate buffered solution containing < 5% detergent.*	1 vial	1 vial
TCR-B	**Aptima Combo 2 Target Capture Reagent B** *Non-infectious nucleic acid in a buffered solution containing < 5% detergent.*	1 x 0.61 mL	1 x 0.30 mL

Aptima Combo 2 Room Temperature Box (Box 2 of 2)
(store at 15°C to 30°C upon receipt)

Symbol	Component	Quantity 250 test kit	Quantity 100 test kit
AR	**Aptima Combo 2 Amplification Reconstitution Solution** *Aqueous solution containing preservatives.*	1 x 27.7 mL	1 x 11.9 mL
ER	**Aptima Combo 2 Enzyme Reconstitution Solution** *HEPES buffered solution containing a surfactant and glycerol.*	1 x 11.1 mL	1 x 6.3 mL
PR	**Aptima Combo 2 Probe Reconstitution Solution** *Succinate buffered solution containing < 5% detergent.*	1 x 35.4 mL	1 x 15.2 mL
S	**Aptima Combo 2 Selection Reagent** *600 mM borate buffered solution containing surfactant.*	1 x 108 mL	1 x 43.0 mL

Aptima Combo 2 Room Temperature Box (Box 2 of 2) (Continued)
(store at 15°C to 30°C upon receipt)

Symbol	Component	Quantity 250 test kit	Quantity 100 test kit
TCR	**Aptima Combo 2 Target Capture Reagent** *Buffered salt solution containing solid phase and capture oligomers.*	1 x 54 mL	1 x 26.0 mL
	Reconstitution Collars	3	3
	Master Lot Barcode Sheet	1 sheet	1 sheet

Aptima Controls Kit
(store at 2°C to 8°C upon receipt)

Symbol	Component	Quantity
PCT/NGC	**Aptima Positive Control, CT / Negative Control, GC** *Non-infectious CT nucleic acid in a buffered solution containing < 5% detergent. Each 400 µL sample contains the estimated rRNA equivalent of 1 CT IFU (5 fg/assay*).*	5 x 1.7 mL
PGC/NCT	**Aptima Positive Control, GC / Negative Control, CT** *Non-infectious GC nucleic acid in a buffered solution containing < 5% detergent. Each 400 µL sample contains the estimated rRNA equivalent of 50 GC cells (250 fg/assay*).*	5 x 1.7 mL

*The rRNA equivalents were calculated based on the genome size and estimated DNA:RNA ratio/cell of each organism.

Materials Required But Available Separately

Note: *Materials available from Hologic have catalog numbers listed, unless otherwise specified.*

	Cat. No.
Panther System	303095
Aptima Assay Fluids Kit *(Aptima Wash Solution, Aptima Buffer for Deactivation Fluid, and Aptima Oil Reagent)*	303014 (1000 tests)
Aptima Auto Detect Kit	303013 (1000 tests)
Multi-tube units (MTUs)	104772-02
Panther Waste Bag Kit	902731
Panther Waste Bin Cover	504405
Or Panther Run Kit *contains MTUs, waste bags, waste bin covers, assay fluids, and auto detects*	303096 (5000 tests)
Tips, 1000 µL conductive, liquid sensing	10612513 (Tecan)
Aptima Specimen Transfer Kit *for use with specimens in PreservCyt Solution*	301154C
Aptima Vaginal Swab Specimen Collection Kit	301162

	Cat. No.
Aptima Multitest Swab Specimen Collection Kit[1]	PRD-03546
Aptima Unisex Swab Specimen Collection Kit for Endocervical and Male Urethral Swab Specimens	301041
Aptima Urine Specimen Collection Kit for Male and Female Urine Specimens	301040
Aptima Urine Specimen Transport Tubes for Male and Female Urine Specimens	105575
Bleach, 5% to 7% (0.7M to 1.0M) sodium hypochlorite solution	—
Disposable gloves	—
SysCheck calibration standard	301078
Aptima penetrable caps	105668
Replacement non-penetrable caps	103036A
Replacement Caps for the 250-test kits	—
Amplification and Probe reagent reconstitution solutions	*CL0041 (100 caps)*
Enzyme Reagent reconstitution solution	*501616 (100 caps)*
TCR and Selection reagent	*CL0040 (100 caps)*
Replacement Caps for the 100-test kits	—
Amplification, Enzyme, and Probe reagent reconstitution solutions	*CL0041(100 caps)*
TCR and Selection reagent	*501604 (100 caps)*

[1] For Aptima Combo 2 Assay, the Aptima Multitest Swab Specimen Collection Kit has been validated for the collection of vaginal swab specimens.

Optional Materials

	Cat. No.
Aptima Controls Kit	301110
Hologic Bleach Enhancer for Cleaning *for routine cleaning of surfaces and equipment*	302101
Tube rocker	—

Panther System Test Procedure

Note: *See the Panther System Operator's Manual for additional Panther System procedural information.*

A. Work Area Preparation

Clean work surfaces where reagents and samples will be prepared. Wipe down work surfaces with 2.5% to 3.5% (0.35M to 0.5M) sodium hypochlorite solution. Allow the sodium hypochlorite solution to contact surfaces for at least 1 minute and then follow with a water rinse. Do not allow the sodium hypochlorite solution to dry. Cover the bench surface on which the reagents and samples will be prepared with clean, plastic-backed absorbent laboratory bench covers.

B. Reagent Reconstitution/Preparation of a New Kit

Note: *Reagent reconstitution should be performed prior to beginning any work on the Panther System.*

1. To reconstitute Amplification, Enzyme, and Probe Reagents, combine the bottles of lyophilized reagent with the reconstitution solution. If refrigerated, allow the reconstitution solutions to reach room temperature before use.
   a. Pair each reconstitution solution with its lyophilized reagent. Ensure that the reconstitution solution and reagent have matching label colors before attaching the reconstitution collar.
   b. Check the lot numbers on the Master Lot Barcode Sheet to ensure that the appropriate reagents are paired.
   c. Open the lyophilized reagent vial and firmly insert the notched end of the reconstitution collar into the vial opening (Figure 1, Step 1).
   d. Open the matching reconstitution solution, and set the cap on a clean, covered work surface.
   e. While holding the reconstitution solution bottle on the bench, firmly insert the other end of the reconstitution collar into the bottle opening (Figure 1, Step 2).
   f. Slowly invert the assembled bottles. Allow the solution to drain from the bottle into the glass vial (Figure 1, Step 3).
   g. Thoroughly mix the solution in the glass vial by swirling (Figure 1, Step 4).
   h. Wait for the lyophilized reagent to go into solution, then invert the assembled bottles again, tilting at a 45° angle to minimize foaming (Figure 1, Step 5). Allow all of the liquid to drain back into the plastic bottle.
   i. Remove the reconstitution collar and glass vial (Figure 1, Step 6).
   j. Recap the plastic bottle. Record operator initials and reconstitution date on the label (Figure 1, Step 7).
   k. Discard the reconstitution collar and glass vial (Figure 1, Step 8).

      Option: Additional mixing of the Amplification, Enzyme and Probe Reagents using a tube rocker is allowed. The reagents may be mixed by placing the recapped plastic bottle on a tube rocker set to 20 RPM (or equivalent) for a minimum of 5 minutes.

Warning: *Avoid creating foam when reconstituting reagents. Foam compromises the level-sensing in the Panther System.*

Warning: *Adequate mixing of the reagents is necessary to achieve expected assay results.*

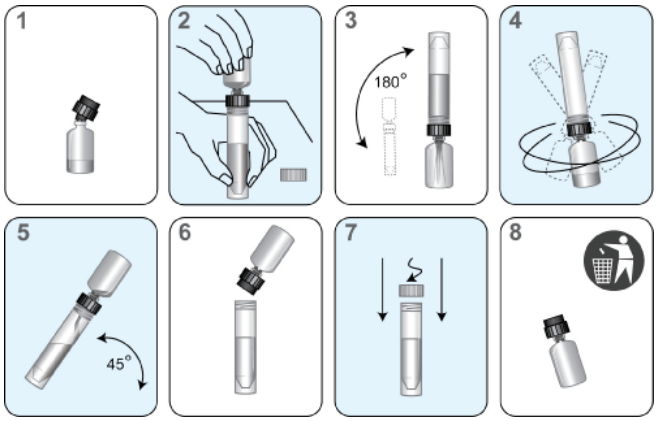


Figure 1. Panther System Reconstitution Process

2. Prepare Working Target Capture Reagent (wTCR)
   a. Pair the appropriate bottles of TCR and TCR-B.
   b. Check the reagent lot numbers on the Master Lot Barcode Sheet to make sure that the appropriate reagents in the kit are paired.
   c. Open the bottle of TCR, and set the cap on a clean, covered work surface.
   d. Open the bottle of TCR-B and pour the entire contents into the bottle of TCR. Expect a small amount of liquid to remain in the TCR-B bottle.
   e. Cap the bottle of TCR and gently swirl the solution to mix the contents. Avoid creating foam during this step.
   f. Record operator initials and the current date on the label.
   g. Discard the TCR-B bottle and cap.
3. Prepare Selection Reagent
   a. Check the lot number on the reagent bottle to make sure it matches the lot number on the Master Lot Barcode Sheet.
   b. Record operator initials and the current date on the label.

Note: *Thoroughly mix by gently inverting all reagents prior to loading on the system. Avoid creating foam during inversion of reagents.*

C. Reagent Preparation for Previously Reconstituted Reagents

1. Previously reconstituted Amplification, Enzyme, and Probe Reagents must reach room temperature (15°C to 30°C) prior to the start of the assay.

   Option: The reagents may be brought to room temperature by placing the reconstituted Amplification, Enzyme and Probe Reagents on a tube rocker set to 20 RPM (or equivalent) for a minimum of 25 minutes.

2. If reconstituted Probe Reagent contains precipitate that does not return to solution at room temperature, heat the capped bottle at a temperature that does not exceed 62°C for 1 to 2 minutes. After this heat step, the Probe Reagent may be used even if residual precipitate remains. Mix Probe Reagent by inversion, being careful not to induce foam, prior to loading onto the system.

3. Thoroughly mix each reagent by gently inverting prior to loading on the system. Avoid creating foam during inversion of reagents. This step is not required if reagents are loaded onto the system directly after mixing on the tube rocker.
4. Do not top off reagent bottles. The Panther System will recognize and reject bottles that have been topped off.

Warning: *Adequate mixing of the reagents is necessary to achieve expected assay results.*

D. Specimen Handling

1. Allow the controls and specimens to reach room temperature prior to processing.
2. **Do not vortex specimens.**
3. Visually confirm that each specimen tube meets one of the following criteria:
   a. The presence of a single blue Aptima collection swab in a unisex swab specimen transport tube.
   b. The presence of a single pink Aptima collection swab in a multitest or vaginal swab specimen transport tube.
   c. A final volume of urine between the black fill lines of a urine specimen transport tube.
   d. The absence of a swab in the Aptima specimen transport tube for PreservCyt Solution liquid Pap specimens.
4. Inspect specimen tubes before loading into rack:
   a. If a specimen tube contains bubbles in the space between the liquid and the cap, centrifuge the tube for 5 minutes at 420 RCF to eliminate the bubbles.
   b. If a specimen tube has a lower volume than typically observed when collection instructions have been followed, centrifuge the tube for 5 minutes at 420 RCF to ensure that no liquid is in the cap.
   c. If the liquid level in a urine specimen tube is not between the two black indicator lines on the label, the specimen must be rejected. Do not pierce an overfilled tube.
   d. If a urine specimen tube contains precipitate, heat the specimen at 37°C for up to 5 minutes. If the precipitate does not go back into solution, visually ensure that the precipitate does not prevent delivery of the specimen.

Note: *Failure to follow Steps 4a–c may result in liquid discharge from the specimen tube cap.*

Note: *Up to 3 separate aliquots can be tested from each specimen tube. Attempts to pipette more than 3 aliquots from the specimen tube can lead to processing errors.*

E. System Preparation

1. Set up the system according to the instructions in the *Panther System Operator's Manual* and *Procedural Notes*. Make sure that the appropriately sized reagent racks and TCR adapters are used.
2. Load samples.

Procedural Notes

A. Controls

1. To work properly with the Aptima Assay software for the Panther System, one pair of controls is required. The Positive Control, CT / Negative Control, GC and the Positive Control, GC / Negative Control CT tubes can be loaded in any rack position or in any Sample Bay Lane on the Panther System. Patient specimen pipetting will begin when one of the following two conditions has been met:
   a. A pair of controls is currently being processed by the system.
   b. Valid results for the controls are registered on the system.
2. Once the control tubes have been pipetted and are processing for a specific reagent kit, patient specimens can be run with the associated kit up to 24 hours unless:
   a. Controls results are invalid.
   b. The associated assay reagent kit is removed from the system.
   c. The associated assay reagent kit has exceeded stability limits.
3. Each Aptima control tube can be tested once. Attempts to pipette more than once from the tube can lead to processing errors.

B. Temperature

Room temperature is defined as 15°C to 30°C.

C. Glove Powder

As in any reagent system, excess powder on some gloves may cause contamination of opened tubes. Powderless gloves are recommended.

D. Lab Contamination Monitoring Protocol for the Panther System

There are many laboratory-specific factors that may contribute to contamination, including testing volume, workflow, disease prevalence and various other laboratory activities. These factors should be taken into consideration when contamination monitoring frequency is being established. Intervals for contamination monitoring should be established based on each laboratory's practices and procedures.

To monitor for laboratory contamination, the following procedure may be performed using the Aptima Unisex Swab Specimen Collection Kit for Endocervical and Male Urethral Swab Specimens:

1. Label swab transport tubes with numbers corresponding to the areas to be tested.
2. Remove the specimen collection swab (blue shaft swab with green printing) from its packaging, wet the swab in the specimen transport medium (STM), and swab the designated area using a circular motion.
3. Immediately insert the swab into transport tube.
4. Carefully break the swab shaft at the score line; use care to avoid splashing of the contents.
5. Recap the swab transport tube tightly.
6. Repeat Steps 2 to 5 for each area to be swabbed.

If the results are CT or GC positive or equivocal, see *Test Interpretation — QC/Patient Results*. For additional Panther System-specific contamination monitoring information, contact Hologic Technical Support.

Test Interpretation — QC/Patient Results

A. Test Interpretation

Assay test results are automatically interpreted by the Aptima Assay software, using the Aptima Combo 2 protocol, and presented as individual CT and GC test results. A test result may be a negative, equivocal, positive, or invalid as determined by the kinetic type and total RLU in the detection step (see below). A test result may be invalid due to a parameter outside the normal expected ranges. Initial equivocal and invalid test results should be retested.

Kinetic Type	Total RLU (x1000) to give CT Result		
	Negative	Equivocal	Positive
CT only	1 to < 25	25 to < 100	100 to < 4,500
CT and GC	1 to < 85	85 to < 250	250 to < 4,500
CT indeterminate	1 to < 85	85 to < 4,500	N/A

Kinetic Type	Total RLU (x1000) to give GC Result		
	Negative	Equivocal	Positive
GC only	1 to < 60	60 to < 150	150 to < 4,500
GC and CT	1 to < 85	85 to < 250	250 to < 4,500
GC indeterminate	1 to < 85	85 to < 4,500	N/A

B. Quality Control Results and Acceptability

The Positive Control, CT / Negative Control, GC and the Positive Control, GC / Negative Control, CT act as controls for the target capture, amplification, and detection steps of the assay. In accordance with guidelines or requirements of local, state, and/or federal regulations or accrediting organizations, additional controls for cell lysis and RNA stabilization may be included. The Positive Control, CT / Negative Control, GC serves as the negative control for the GC test results. The Positive Control, GC / Negative Control, CT serves as the negative control for the CT test results. If desired, a dual negative control furnished by the user can be added to monitor assay background. Correct preparation of specimens is confirmed visually by the presence of a single Aptima collection swab in a swab specimen transport tube, a final volume of urine in between the black fill lines of a urine specimen transport tube, or the absence of a swab in an Aptima specimen transfer tube for PreservCyt liquid Pap specimens.

The Positive Controls must produce the following test results:

Control	Total RLU (x1000)	CT Result	GC Result
Positive Control, CT / Negative Control, GC	≥ 100 and < 3,000	Positive	Negative
Positive Control, GC / Negative Control, CT	≥ 150 and < 3,000	Negative	Positive

1. The Aptima Assay software automatically evaluates the controls according to the above criteria and will report the Run Status as PASS if the run control criteria are met, and FAIL if the run control criteria are not met.
2. If the Run Status is FAIL, all test results in the same run are invalid and must not be reported.
3. Each laboratory should implement appropriate control procedures to satisfy the requirements of CLIA regulations (section 493.1256).

4. Negative controls may not be effective in monitoring random carryover. See *Panther System Analytical Performance* for results from a high-target analytical carryover study that was performed to demonstrate control of carryover on the Panther System.

C. Specimen Preparation Control (Optional)

The Positive Control, CT / Negative Control, GC and the Positive Control, GC / Negative Control, CT provided in the kit act as controls for the target capture, amplification, and detection steps of the assay and must be included in each assay run. If desired, controls for cell lysis and RNA stabilization in appropriate transport media (PreservCyt Solution, STM) can be tested in accordance with the requirements of appropriate accrediting organizations or individual laboratory procedures. Known positive specimens can serve as controls by being prepared and tested in conjunction with unknown specimens. Specimens used as preparation controls must be stored, handled, and tested according to the package insert. Specimen preparation controls should be interpreted in the same manner as described for patient test specimens. See *Test Interpretation — QC/Patient Results*.

D. Patient Test Results

1. If the controls in any run do not yield the expected results, test results on patient specimens in the same run must not be reported.
2. Swab, PreservCyt Solution liquid Pap, and urine specimen results (see Notes below).
   a. Initial results

CT Pos	Positive for CT rRNA.
CT Neg	Presumed negative for CT rRNA.
CT Equiv	Sample should be retested.
GC Pos	Positive for GC rRNA.
GC Neg	Presumed negative for GC rRNA.
GC Equiv	Sample should be retested.
Invalid	Sample should be retested.

   b. Retest results

CT Pos	Positive for CT rRNA.
CT Neg	Presumed negative for CT rRNA.
CT Equiv	Indeterminate, a new specimen should be collected.
GC Pos	Positive for GC rRNA.
GC Neg	Presumed negative for GC rRNA.
GC Equiv	Indeterminate, a new specimen should be collected.
Invalid	Indeterminate, a new specimen should be collected

Notes

- Careful consideration of performance data is recommended for interpreting Aptima Combo 2 Assay results for asymptomatic individuals or any individuals in low prevalence populations.
- The first valid result for each analyte is the result that should be reported.
- A negative result does not preclude the presence of a CT or GC infection because results are dependent on adequate specimen collection, absence of inhibitors, and sufficient rRNA to be detected. Test results may be affected by improper specimen collection, improper specimen storage, technical error, or specimen mix-up.

- As is true for all non-culture methods, a positive specimen obtained from a patient after therapeutic treatment cannot be interpreted as indicating the presence of viable CT or GC.
- Testing of an endocervical specimen is recommended for female patients who are clinically suspected of having a chlamydial or gonococcal infection. If both a Pap and endocervical swab are collected, the PreservCyt Solution liquid Pap specimen must be collected before the endocervical swab specimen.

Limitations

A. Use of this assay is limited to personnel who have been trained in the procedure. Failure to follow the instructions given in this package insert may result in erroneous results.

B. The effects of tampon use, douching, and specimen collection variables have not been assessed for their impact on the detection of CT or GC.

C. The presence of mucus in endocervical specimens does not interfere with the detection of CT or GC by the Aptima Combo 2 Assay. However, to ensure collection of cells infected with CT, columnar epithelial cells lining the endocervix should be sampled. If excess mucus is not removed, sampling of these cells is not ensured.

D. Vaginal swab and PreservCyt Solution liquid Pap specimen sampling is not designed to replace cervical exams and endocervical specimens for diagnosis of female urogenital infections. Patients may have cervicitis, urethritis, urinary tract infections, or vaginal infections due to other causes or concurrent infections with other agents.

E. The Aptima Combo 2 Assay is not intended for the evaluation of suspected sexual abuse or for other medico-legal indications. For those patients for whom a false positive result may have adverse psycho-social impact, the CDC recommends retesting (4).

F. Reliable results are dependent on adequate specimen collection. Because the transport system used for this assay does not permit microscopic assessment of specimen adequacy, training of clinicians in proper specimen collection techniques is necessary. Refer to the package insert of the appropriate Hologic specimen collection kit.

G. Therapeutic failure or success cannot be determined with the Aptima Combo 2 Assay since nucleic acid may persist following appropriate antimicrobial therapy.

H. Results from the Aptima Combo 2 Assay should be interpreted in conjunction with other laboratory and clinical data available to the clinician.

I. A negative result does not preclude a possible infection because results are dependent on adequate specimen collection. Test results may be affected by improper specimen collection, technical error, specimen mix-up, or target levels below the assay limit of detection.

J. The Aptima Combo 2 Assay provides qualitative results. Therefore, a correlation cannot be drawn between the magnitude of a positive assay signal and the number of organisms in a specimen.

K. Performance of the Aptima Specimen Transfer kit was not evaluated for testing the same PreservCyt Solution liquid Pap specimen both before and after ThinPrep Pap processing.

L. PreservCyt Solution liquid Pap specimens processed with instruments other than the ThinPrep 2000 processor have not been evaluated for use in Aptima Assays.

M. Patient-collected vaginal swab specimens are an option for screening women when a pelvic exam is not otherwise indicated.

N. The patient-collected vaginal swab specimen application is limited to health care facilities where support/counseling is available to explain procedures and precautions.

O. The Aptima Combo 2 Assay has not been validated for use with vaginal swab specimens collected by patients at home.

P. The performance of the Aptima Combo 2 Assay has not been evaluated in adolescents less than 14 years of age.

Q. The performance of the Panther System has not been evaluated at altitudes above 6561 feet (2000 m).

R. There is no evidence of degradation of nucleic acids in PreservCyt Solution. If a PreservCyt Solution liquid Pap specimen has small numbers of CT and GC cellular material, uneven distribution of this cellular material may occur. Also, when compared to direct sampling with the Aptima Specimen Transport Medium, the additional volume of PreservCyt Solution results in greater dilution of the sample material. These factors may affect the ability to detect small numbers of organisms in the collected material. If negative results from the specimen do not fit with the clinical impression, a new specimen may be necessary.

S. Customers must independently validate an LIS transfer process.

T. First catch female urine specimens are acceptable but may detect up to 10% fewer CT/GC infections when compared with vaginal and endocervical swab specimens (5).

Panther System Expected Values

Prevalence

The prevalence of CT and GC in patient populations depends on risk factors such as age, gender, the presence or absence of symptoms, the type of clinic, and the sensitivity of the test used to detect infections. A summary of the positivity of three CT and GC disease outcomes, as determined by the Aptima Combo 2 Assay on the Panther System, is shown in Tables 1, 2, and 3 for three multi-center clinical studies by clinical site and overall.

Table 1: Clinical Study 1. Positivity of CT and GC Infections as Determined by the Aptima Combo 2 Assay in Male Urethral Swab, Vaginal Swab, PreservCyt Solution Liquid Pap, and Endocervical Swab Samples by Clinical Site

	Positivity % (# positive/# tested with valid results)											
Site	**MS**			**CVS/PVS**			**PCyt**			**FS**		
	CT+/GC-	**CT-/GC+**	**CT+/GC+**	**CT+/GC-**	**CT-/GC+**	**CT+/GC+**	**CT+/GC-**	**CT-/GC+**	**CT+/GC+**	**CT+/GC-**	**CT-/GC+**	**CT+/GC+**
1	0 (-)	0 (-)	0 (-)	9.9 (21/212)	3.3 (7/212)	3.8 (8/212)	8.9 (20/225)	2.7 (6/225)	3.1 (7/225)	10.4 (20/193)	3.1 (6/193)	3.6 (7/193)
2	13.9 (28/202)	5.9 (12/202)	3.0 (6/202)	8.3 (19/230)	3.9 (9/230)	1.3 (3/230)	8.8 (21/239)	4.6 (11/239)	0.8 (2/239)	8.2 (19/231)	4.8 (11/231)	0.9 (2/231)
3	1.3 (1/76)	1.3 (1/76)	0.0 (0/76)	2.7 (6/222)	0.5 (1/222)	0.0 (0/222)	3.1 (7/226)	0.4 (1/226)	0.0 (0/226)	2.7 (6/223)	0.4 (1/223)	0.0 (0/223)
4	24.4 (33/135)	1.5 (2/135)	4.4 (6/135)	11.7 (40/342)	1.5 (5/342)	1.2 (4/342)	10.2 (35/342)	1.5 (5/342)	0.9 (3/342)	11.3 (38/337)	1.8 (6/337)	0.9 (3/337)
5	0 (-)	0 (-)	0 (-)	4.5 (1/22)	0.0 (0/22)	0.0 (0/22)	4.8 (1/21)	0.0 (0/21)	0.0 (0/21)	4.3 (1/23)	0.0 (0/23)	0.0 (0/23)
6	21.5 (28/130)	5.4 (7/130)	0.8 (1/130)	11.9 (13/109)	3.7 (4/109)	0.9 (1/109)	8.7 (10/115)	1.7 (2/115)	0.9 (1/115)	8.8 (10/114)	1.8 (2/114)	0.9 (1/114)
7	16.7 (1/6)	0.0 (0/6)	0.0 (0/6)	3.2 (5/157)	2.5 (4/157)	0.6 (1/157)	2.5 (4/161)	2.5 (4/161)	0.6 (1/161)	2.6 (4/152)	2.6 (4/152)	0.7 (1/152)
All	16.6 (91/549)	4.0 (22/549)	2.4 (13/549)	8.1 (105/1294)	2.3 (30/1294)	1.3 (17/1294)	7.4 (98/1329)	2.2 (29/1329)	1.1 (14/1329)	7.7 (98/1273)	2.4 (30/1273)	1.1 (14/1273)

CVS = clinician-collected vaginal swab, FS = female endocervical swab, MS = male urethral swab, PCyt = PreservCyt Solution liquid Pap, PVS = patient-collected vaginal swab.

Table 2: Clinical Study 1 and Clinical Study 2. Positivity of CT and GC Infections as Determined by the Aptima Combo 2 Assay in Male Urine Samples by Clinical Site

Site	Positivity % (# positive/# tested with valid results)		
	CT+/GC-	CT-/GC+	CT+/GC+
1	6.0 (6/100)	0.0 (0/100)	0.0 (0/100)
2	3.0 (2/67)	3.0 (2/67)	0.0 (0/67)
3	0.0 (0/109)	0.9 (1/109)	0.0 (0/109)
4	13.0 (13/100)	3.0 (3/100)	1.0 (1/100)
5	13.6 (17/125)	5.6 (7/125)	0.0 (0/125)
6	15.1 (43/284)	7.0 (20/284)	2.1 (6/284)
7	1.4 (3/212)	0.9 (2/212)	0.0 (0/212)
8	1.3 (1/75)	0.0 (0/75)	0.0 (0/75)
9	16.7 (42/251)	5.2 (13/251)	3.2 (8/251)
10	20.5 (17/83)	1.2 (1/83)	0.0 (0/83)
11	4.1 (6/146)	0.7 (1/146)	0.7 (1/146)
12	14.3 (16/112)	4.5 (5/112)	2.7 (3/112)
13	8.9 (10/112)	2.7 (3/112)	2.7 (3/112)
14	7.7 (2/26)	0.0 (0/26)	0.0 (0/26)
All	9.9 (178/1802)	3.2 (58/1802)	1.2 (22/1802)

Note. CT and GC prevalence was estimated using symptomatic male urine samples from Clinical Study 2 and asymptomatic male urine samples from both studies.

Table 3: Clinical Study 3. Positivity of CT and GC Infections as Determined by the Aptima Combo 2 Assay in Female Urine Samples by Clinical Site

Site	Positivity % (# positive/# tested with valid results)		
	CT+/GC-	CT-/GC+	CT+/GC+
1	14.8 (23/155)	3.2 (5/155)	1.9 (3/155)
2	2.5 (5/199)	0.0 (0/199)	0.0 (0/199)
3	2.0 (4/199)	0.0 (0/199)	0.0 (0/199)
4	6.3 (5/79)	0.0 (0/79)	0.0 (0/79)
5	5.1 (5/99)	0.0 (0/99)	0.0 (0/99)
6	9.8 (15/153)	2.0 (3/153)	2.0 (3/153)
7	7.3 (18/247)	0.0 (0/247)	0.0 (0/247)
8	7.4 (14/189)	1.1 (2/189)	0.0 (0/189)
9	6.7 (6/90)	0.0 (0/90)	1.1 (1/90)
10	6.1 (6/99)	0.0 (0/99)	0.0 (0/99)
11	3.2 (3/93)	0.0 (0/93)	0.0 (0/93)
12	0.0 (0/97)	0.0 (0/97)	0.0 (0/97)
13	8.7 (26/299)	1.0 (3/299)	0.3 (1/299)
14	4.6 (9/196)	0.0 (0/196)	0.0 (0/196)
15	5.0 (5/100)	0.0 (0/100)	0.0 (0/100)
16	8.8 (23/261)	1.5 (4/261)	0.8 (2/261)
17	20.0 (5/25)	4.0 (1/25)	0.0 (0/25)
All	6.7 (172/2580)	0.7 (18/2580)	0.4 (10/2580)

Positive and Negative Predictive Values for Hypothetical Prevalence Rates

The estimated positive and negative predictive values (PPV and NPV) of the Aptima Combo 2 Assay for different hypothetical prevalence rates are shown for each specimen type in Table 4. For each specimen type, the PPV and NPV are derived for different hypothetical prevalence rates using the sensitivity and specificity estimates from two multi-center clinical studies (see Tables 5 and 9).

Table 4: Positive and Negative Predictive Values for Hypothetical Prevalence Rates by Specimen Type

Specimen Type	Hypothetical Prevalence (%)	CT Detection		GC Detection	
		PPV (%)	NPV (%)	PPV (%)	NPV (%)
Clinician-Collected Vaginal Swab/Patient-Collected Vaginal Swab	1	38.9	100	70.6	100
	2	56.3	99.9	82.9	100
	5	76.8	99.9	92.6	99.9
	10	87.5	99.7	96.3	99.7
	15	91.7	99.5	97.7	99.6
	20	94.0	99.3	98.3	99.4
	25	95.5	99.1	98.8	99.2
PreservCyt Solution Liquid Pap	1	100	100	100	100
	2	100	100	100	100
	5	100	99.9	100	100
	10	100	99.8	100	100
	15	100	99.7	100	100
	20	100	99.6	100	100
	25	100	99.4	100	100
Female Endocervical Swab	1	58.5	100	85.8	100
	2	74.0	99.9	92.4	100
	5	88.0	99.9	96.9	100
	10	93.9	99.7	98.5	100
	15	96.1	99.5	99.1	100
	20	97.2	99.3	99.3	100
	25	97.9	99.1	99.5	100
Male Urethral Swab	1	53.1	100	100	100
	2	69.6	100	100	100
	5	85.5	100	100	100
	10	92.6	100	100	100
	15	95.2	100	100	100
	20	96.6	100	100	100
	25	97.4	100	100	100
Male Urine	1	83.6	100	77.4	100
	2	91.2	99.9	87.4	100
	5	96.4	99.7	94.7	99.9
	10	98.2	99.5	97.4	99.9
	15	98.9	99.2	98.4	99.8
	20	99.2	98.8	98.8	99.7
	25	99.4	98.4	99.1	99.6

Note. Aptima Combo 2 Assay performance was estimated using vaginal swab, PreservCyt Solution liquid Pap, female endocervical swab, and male urethral swab sample results from Clinical Study 1, symptomatic male urine samples from Clinical Study 2, and asymptomatic male urine samples from Clinical Studies 1 and 2.

Panther System Clinical Performance

Three clinical studies were performed. Aptima Combo 2 Assay clinical performance was estimated with male urethral swab, vaginal swab, PreservCyt Solution liquid Pap, and endocervical swab specimens in Clinical Study 1, with male urine specimens in Clinical Study 2, and with female urine specimens in Clinical Study 3.

Clinical Study 1. Vaginal Swab, PreservCyt Solution Liquid Pap, Female Endocervical Swab, and Male Urethral Swab Specimen Clinical Study[2]

A prospective, multi-center clinical study was conducted to establish the performance characteristics of the Aptima Combo 2 Assay on the Panther System. Specimens were collected from symptomatic and asymptomatic men (n=580) and women (n=1332) enrolled from 7 geographically and ethnically diverse US clinical sites, including obstetrics and gynecology, family planning, public health, and STD clinics. Subjects were classified as symptomatic if symptoms were reported by the subject. Subjects were classified as asymptomatic if the subject did not report symptoms. Of the 580 male subjects, none were <18 years of age, 72 were 18 to 20 years of age, 201 were 21 to 25 years of age, and 307 were >25 years of age. Of the 1332 female subjects, 11 were 14 to 15 years of age, 59 were 16 to 17 years of age, 319 were 18 to 20 years of age, 401 were 21 to 25 years of age, and 542 were >25 years of age.

Up to 2 specimens were collected from each male subject (1 urethral swab and 1 first-catch urine, in that order) and up to 4 specimens were collected from each female subject (1 first-catch urine, 1 vaginal swab, 1 PreservCyt Solution liquid Pap specimen, and 1 endocervical swab, in that order). All specimens were clinician-collected except urine specimens and approximately half of the vaginal swab specimens, which were collected by the subject at the clinic. Approximately half of the PreservCyt Solution liquid Pap specimens were collected with a broom-type device and half were collected with a spatula and cytobrush. Samples were prepared for Aptima testing in accordance with the appropriate Aptima specimen collection kit package insert instructions.

All evaluable samples (567 male urethral swab, 580 male urine, 1319 vaginal swab, 1330 PreservCyt Solution liquid Pap, and 1310 endocervical swab samples) were tested with the Aptima Combo 2 Assay on the Panther System in accordance with package insert instructions. The samples were split among three laboratories (two external laboratories and in-house). Samples with initial invalid, equivocal, or error results were retested. Eighteen (18) male urethral swab, 25 vaginal swab, 1 PreservCyt Solution liquid Pap, and 37 endocervical swab samples had final invalid results and were excluded from the analyses. Most of the invalid results were due to insufficient sample volume. One vaginal swab and 1 endocervical swab had final CT equivocal results and 1 PreservCyt Solution liquid Pap sample and 1 endocervical swab had final GC equivocal results and were excluded from the analyses.

Male urethral swab, male and female urine, and PreservCyt Solution liquid Pap samples were tested with cleared nucleic acid amplification tests (NAATs) to establish the infected status. The infected status algorithm used results from two specimen types and two reference NAATs. Subjects were categorized as infected if a positive result occurred in each of the two reference NAATs (see Tables 13, 14, 17, and 18 for the infected status algorithms). For female subjects, if the positive NAAT results occurred only in the urine specimens and not in the PreservCyt Solution liquid Pap specimens, the subject was categorized as infected; however, for the

2 This study included testing of male urine samples with the Aptima Combo 2 Assay on the Panther System that were not included in the original performance results due to the low prevalence of GC in the study population.

evaluation of the non-urine specimen types, the specimens were considered non-infected. Subjects that could not be categorized as infected or not infected were excluded from the performance analyses.

In addition, male urine samples tested with the Aptima Combo 2 Assay on the Panther System were excluded from the performance analyses due to the low prevalence of GC in the study population, particularly in the asymptomatic subjects.

Clinical Study 2. Male Urine Specimen Clinical Study

A prospective, multi-center clinical study was conducted to establish the performance characteristics of the Aptima Combo 2 Assay on the Panther System in male urine specimens. Specimens were collected from symptomatic and asymptomatic men (n=1492) enrolled from 13 geographically and ethnically diverse US clinical research sites, and family planning, public health, men's health, and STD clinics. Subjects were classified as symptomatic if symptoms were reported by the subject. Subjects were classified as asymptomatic if the subject did not report symptoms. Of the 1492 subjects enrolled, 14 were withdrawn.

Two specimens were collected from each subject (1 urethral swab and 1 first-catch urine, in that order). The urethral swab specimens were clinician-collected, and urine specimens were collected by the subject at the clinic. Urine specimens from each subject were processed into multiple samples for CT/GC testing with different NAATs in accordance with the instructions in the appropriate specimen collection kit package insert. The male urine samples for Aptima Combo 2 Assay testing on the Panther System were split among three external laboratories.

All 1478 male urine samples from non-withdrawn subjects were tested with the Aptima Combo 2 Assay on the Panther System in accordance with the Aptima Combo 2 Assay package insert instructions. Samples with initial invalid, equivocal, or error results were retested. One male urine sample had a final invalid result and was excluded from the analyses. The invalid result was due to insufficient sample volume. Of the remaining 1477 evaluable male subjects, 46 were 16 to 17 years of age, 155 were 18 to 20 years of age, 524 were 21 to 30 years of age, 279 were 31 to 40 years of age, and 473 were >40 years of age.

Male urethral swab and urine samples were tested with cleared NAATs to establish the infected status (see Tables 15 and 19 for the infected status algorithms). The infected status algorithm used urethral swab and urine sample results from one reference CT and GC NAAT and urine sample results from two additional reference CT and GC NAATs to generate four reference results for each analyte. Subjects were categorized as infected if a positive result occurred in at least two of the reference NAATs. Subjects that could not be categorized as infected or not infected were excluded from the performance analyses; 1 subject had an indeterminate CT infected status and was excluded from the performance analyses for detection of CT.

Clinical Study 3. Female Urine Specimen Clinical Study

A retrospective study that used results and remnant female urine samples from a previously completed prospective, multi-center clinical study was conducted to establish the performance characteristics of the Aptima Combo 2 Assay on the Panther System in female urine specimens. Specimens were collected from symptomatic and asymptomatic women (n=2640) enrolled from 17 geographically and ethnically diverse US clinical sites, including family planning clinics, academic center clinics, and public health clinics. Subjects were classified as symptomatic if symptoms were reported by the subject. Subjects were classified as asymptomatic if the subject did not report symptoms. Of the 2640 subjects enrolled, 42 were withdrawn.

Three specimens were used from each subject (1 first-catch urine and 2 vaginal swabs, in that order). The urine specimens were collected by the subject at the clinic and the vaginal swab specimens were clinician-collected. Urine specimens from each subject were processed into multiple samples for CT/GC testing with different NAATs in accordance with the instructions in the appropriate specimen collection kit package insert. The female urine samples for Aptima Combo 2 Assay testing on the Panther System were split among three external laboratories.

Female urine samples were tested with cleared NAATs to establish a composite comparator algorithm (CCA) result (see Tables 16 and 20). The CCA used urine sample results from up to three reference CT and GC NAATs to generate reference results for each analyte. Subjects were categorized as positive if 2 out of 3 reference NAAT results were positive and negative if 2 out of 3 reference NAAT results were negative. Subjects who could not be categorized as CCA-positive or CCA-negative were excluded from the performance analyses.

Of the 2598 non-withdrawn subjects, 2581 had urine samples tested with the Aptima Combo 2 Assay on the Panther System in accordance with the Aptima Combo 2 Assay package insert instructions. Seventeen subjects had urine samples that were withdrawn or not collected (missing both CT and GC Aptima Combo 2 Assay [Panther System] results). Samples with initial invalid, equivocal, or error results were retested. All 2581 samples had final valid results after required retesting. One sample had a repeat CT equivocal result and one sample had a repeat GC equivocal result.

Of the 2581 subjects that had valid Aptima Combo 2 Assay (Panther System) results, 2580 subjects had a conclusive CT and/or GC composite comparator status and were evaluable for performance; one subject had unknown composite comparator status for both CT and GC and was not evaluable. One evaluable subject had a final equivocal CT result (negative GC result), and one evaluable subject had a final equivocal GC result (negative CT result). Of the 2580 evaluable subjects, 47 were 16 to 17 years of age, 346 were 18 to 20 years of age, 1350 were 21 to 30 years of age, 550 were 31 to 40 years of age, and 287 were >40 years of age.

Of the 2580 evaluable subjects, 2572 subjects were evaluable for performance analyses for CT detection (including one with a final equivocal result). The remaining 8 subjects had an unknown composite comparator status for CT. Of the 2580 evaluable subjects, 2579 subjects were evaluable for performance analyses for GC detection (including one with a final equivocal result). The remaining subject had an unknown composite comparator status for GC. Samples with final equivocal results were categorized as false negative relative to the CCA result (32).

In addition, female urine detected 8.3% fewer CT infections than vaginal and endocervical swab specimens and 12.9% fewer GC infections than vaginal swab specimens and 15.2% fewer GC infections than endocervical swab specimens when compared using the patient infected status (PIS) algorithm.

Chlamydia trachomatis Performance Results

Performance characteristics of the Aptima Combo 2 Assay for CT detection were estimated for each specimen type and are displayed in Tables 5 and 6 combining data from the three clinical studies. Performance was calculated by comparing Panther System results to different infected status algorithms in Clinical Studies 1 and 2 and to a composite comparator algorithm (CCA) in Clinical Study 3 (see Tables 13 through 16 for the CT infected status algorithms). Table 5 shows the sensitivity, specificity, PPV, and NPV of the Aptima Combo 2 Assay for CT detection and the prevalence of CT (based on the infected status) in male urine samples and urethral swab specimens, and in female vaginal swab, endocervical swab, and PCyt specimens.

Table 6 shows the positive percent agreement (PPA) and negative percent agreement (NPA) of the Aptima Combo 2 Assay for CT detection based on the CCA in female urine samples.

Table 5: Performance Characteristics of the Aptima Combo 2 Assay for CT Detection in Female and Male Specimens

Specimen Type[1]	n	TP	FP	TN	FN	Prev %	Sensitivity % (95% CI)[2]	Specificity % (95% CI)[2]	PPV % (95% CI)[3]	NPV % (95% CI)[3]
CVS/PVS	1274	104	18	1149	3	8.4	97.2 (92.1-99.0)	98.5 (97.6-99.0)	85.2 (78.8-90.5)	99.7 (99.3-99.9)
PCyt	1311	112	0	1197	2	8.7	98.2 (93.8-99.5)	100 (99.7-100)	100 (96.9-100)	99.8 (99.4-100)
FS	1254	104	8	1139	3	8.5	97.2 (92.1-99.0)	99.3 (98.6-99.6)	92.9 (87.1-96.7)	99.7 (99.3-99.9)
MS	549	100	4	445	0	18.2	100 (96.3-100)	99.1 (97.7-99.7)	96.2 (90.8-98.9)	100 (99.2-100)
MU	1799	197	3	1589	10	11.5	95.2 (91.3-97.4)	99.8 (99.4-99.9)	98.5 (95.8-99.7)	99.4 (98.9-99.7)

CI = confidence interval, CVS = clinician-collected vaginal swab, FN = false negative, FP = false positive, FS = female endocervical swab, MS = male urethral swab, MU = male urine, NPV = negative predictive value, PCyt = PreservCyt Solution liquid Pap, PPV = positive predictive value, Prev = prevalence, PVS = patient-collected vaginal swab, TN = true negative, TP = true positive.

[1] Male urethral swab, vaginal swab, PreservCyt Solution liquid Pap, and endocervical swab sample results are from Clinical Study 1. Symptomatic male urine sample results are from Clinical Study 2, and asymptomatic male urine sample results are from Clinical Studies 1 and 2.

[2] Score CI.

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

Table 6: Performance Characteristics of the Aptima Combo 2 Assay for CT Detection in Female Urine Samples

Specimen Type[1]	n	CCA+ AC2+	CCA- AC2+	CCA- AC2-	CCA+ AC2-[2]	PPA % (95% CI)[3]	NPA % (95% CI)[3]
FU	2572	174	5	2391	2	98.9 (96.0-99.7)	99.8 (99.5-99.9)

AC2 = Aptima Combo 2 Assay, CCA = composite comparator algorithm, CI = confidence interval, FU = female urine, NPA = negative percent agreement, PPA = positive percent agreement.

[1] Symptomatic and asymptomatic female urine sample results are from Clinical Study 3.

[2] Includes equivocal results from Panther AC2 testing. Equivocal results from AC2 testing are considered indeterminate; a new specimen should be collected.

[3] Score CI.

Table 7 shows the sensitivity, specificity, PPV, and NPV of the Aptima Combo 2 Assay for CT detection and the prevalence of CT (based on the infected status) in male urine samples and urethral swab specimens, and in female vaginal swab, endocervical swab, and PCyt specimens by symptom status. CT prevalence was higher in symptomatic men and women, compared to asymptomatic subjects.

Table 8 shows the PPA and NPA of the Aptima Combo 2 Assay for CT detection based on the CCA in female urine samples by symptom status.

Table 7: Performance Characteristics of the Aptima Combo 2 Assay for CT Detection by Symptom Status in Female and Male Specimens

Specimen Type[1]	Symptom Status	n	TP	FP	TN	FN	Prev %	Sensitivity % (95% CI)[2]	Specificity % (95% CI)[2]	PPV % (95% CI)[3]	NPV % (95% CI)[3]
CVS/PVS	Sym	810	73	8	729	0	9.0	100 (95.0-100)	98.9 (97.9-99.4)	90.1 (82.3-95.5)	100 (99.5-100)
	Asym	464	31	10	420	3	7.3	91.2 (77.0-97.0)	97.7 (95.8-98.7)	75.6 (63.1-86.2)	99.3 (98.1-99.8)
PCyt	Sym	838	76	0	762	0	9.1	100 (95.2-100)	100 (99.5-100)	100 (95.4-100)	100 (99.5-100)
	Asym	473	36	0	435	2	8.0	94.7 (82.7-98.5)	100 (99.1-100)	100 (91.1-100)	99.5 (98.5-99.9)
FS	Sym	794	71	5	718	0	8.9	100 (94.9-100)	99.3 (98.4-99.7)	93.4 (85.9-97.8)	100 (99.5-100)
	Asym	460	33	3	421	3	7.8	91.7 (78.2-97.1)	99.3 (97.9-99.8)	91.7 (79.9-98.0)	99.3 (98.1-99.8)
MS	Sym	238	59	1	178	0	24.8	100 (93.9-100)	99.4 (96.9-99.9)	98.3 (91.5-100)	100 (98.0-100)
	Asym	311	41	3	267	0	13.2	100 (91.4-100)	98.9 (96.8-99.6)	93.2 (82.5-98.5)	100 (98.7-100)
MU	Sym	497	85	1	406	5	18.1	94.4 (87.6-97.6)	99.8 (98.6-100)	98.8 (94.1-100)	98.8 (97.3-99.6)
	Asym	1302	112	2	1183	5	9.0	95.7 (90.4-98.2)	99.8 (99.4-100)	98.2 (94.1-99.8)	99.6 (99.1-99.9)

Asym = asymptomatic, CI = confidence interval, CVS = clinician-collected vaginal swab, FN = false negative, FP = false positive, FS = female endocervical swab, MS = male urethral swab, MU = male urine, NPV = negative predictive value, PCyt = PreservCyt Solution liquid Pap, PPV = positive predictive value, Prev = prevalence, PVS = patient-collected vaginal swab, Sym = symptomatic, TN = true negative, TP = true positive.

[1] Male urethral swab, vaginal swab, PreservCyt Solution liquid Pap, and endocervical swab sample results are from Clinical Study 1. Symptomatic male urine sample results are from Clinical Study 2, and asymptomatic male urine sample results are from Clinical Studies 1 and 2.

[2]Score CI.

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

Table 8: Performance Characteristics of the Aptima Combo 2 Assay for CT Detection by Symptom Status in Female Urine Samples

Specimen Type[1]	Symptom Status	n	CCA+ AC2+	CCA- AC2+	CCA- AC2-	CCA+ AC2-[2]	PPA % (95% CI)[3]	NPA % (95% CI)[3]
FU	Sym	1379	109	2[4]	1267[5]	1	99.1 (95.0-99.8)	99.8 (99.4-100)
	Asym	1193	65	3[6]	1124[7]	1[2]	98.5 (91.9-99.7)	99.7 (99.2-99.9)

AC2 = Aptima Combo 2 Assay, Asym = asymptomatic, CCA = composite comparator algorithm, CI = confidence interval, FU = female urine, NPA = negative percent agreement, PPA = positive percent agreement, Sym = symptomatic.

[1] Symptomatic and asymptomatic female urine sample results are from Clinical Study 3.

[2] Includes equivocal results from Panther AC2 testing. Equivocal results from AC2 testing are considered indeterminate; a new specimen should be collected.

[3] Score CI.

[4] 2/2 subjects had positive CT vaginal swab sample results in both reference NAATs.

[5] 38/1267 subjects had at least one positive CT vaginal swab sample result by a reference NAAT; one or more vaginal swab sample reference results were not available 11/1267 subjects; 1218/1267 subjects had negative vaginal swab sample reference results.

[6] 1/ 3 subject had positive CT vaginal swab sample results in both reference NAATs; 2/3 subjects had negative vaginal swab sample reference results.

[7] 20/1124 subjects had at least one positive CT vaginal swab sample result by a reference NAAT; one or more vaginal swab sample reference results were not available for 11/1124 subjects; 1093/1124 subjects had negative vaginal swab sample reference results.

Neisseria gonorrhoeae Performance Results

Performance characteristics of the Aptima Combo 2 Assay for GC detection were estimated for each specimen type and are displayed in Tables 9 and 10 combining data from the three clinical studies. The infected status algorithm differed among the three clinical studies (see Tables 17 through 20 for the GC infected status algorithms). Table 9 shows the sensitivity, specificity, PPV, and NPV of the Aptima Combo 2 Assay for GC detection and the prevalence of GC (based on the infected status) in male urine samples and urethral swab specimens, and in female vaginal swab, endocervical swab, and PCyt specimens.

Table 10 shows the PPA and NPA of the Aptima Combo 2 Assay for GC detection based on the CCA in female urine samples.

Table 9: Performance Characteristics of the Aptima Combo 2 Assay for GC Detection in Female and Male Specimens

Specimen Type[1]	n	TP	FP	TN	FN	Prev %	Sensitivity % (95% CI)[2]	Specificity % (95% CI)[2]	PPV % (95% CI)[3]	NPV % (95% CI)[3]
CVS/PVS	1258	42	5	1210	1	3.4	97.7 (87.9-99.6)	99.6 (99.0-99.8)	89.4 (78.6-96.1)	99.9 (99.6-100)
PCyt	1293	43	0	1250	0	3.3	100 (91.8-100)	100 (99.7-100)	100 (92.1-100)	100 (99.7-100)
FS	1238	42	2	1194	0	3.4	100 (91.6-100)	99.8 (99.4-100)	95.5 (85.4-99.4)	100 (99.7-100)
MS	546	34	0	512	0	6.2	100 (89.8-100)	100 (99.3-100)	100 (90.2-100)	100 (99.3-100)
MU	1797	75	5	1716	1	4.2	98.7 (92.9-99.8)	99.7 (99.3-99.9)	93.8 (86.7-97.8)	99.9 (99.7-100)

CI = confidence interval, CVS = clinician-collected vaginal swab, FN = false negative, FP = false positive, FS = female endocervical swab, MS = male urethral swab, MU = male urine, NPV = negative predictive value, PCyt = PreservCyt Solution liquid Pap, PPV = positive predictive value, Prev = prevalence, PVS = patient-collected vaginal swab, TN = true negative, TP = true positive.

[1] Vaginal swab, PreservCyt Solution liquid Pap, endocervical swab, and male urethral swab sample results are from Clinical Study 1. Symptomatic male urine sample results are from Clinical Study 2, and asymptomatic male urine sample results are from Clinical Studies 1 and 2.

[2] Score CI.

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

Table 10: Performance Characteristics of the Aptima Combo 2 Assay for GC Detection in Female Urine Samples

Specimen Type[1]	n	CCA+ AC2+	CCA- AC2+	CCA- AC2-	CCA+ AC2-[2]	PPA % (95% CI)[3]	NPA % (95% CI)[3]
FU	2579	28	0	2550	1	96.6 (82.8-99.4)	100 (99.8-100)

AC2 = Aptima Combo 2 Assay, CCA = composite comparator algorithm, CI = confidence interval, FU = female urine, NPA = negative percent agreement, PPA = positive percent agreement.

[1] Symptomatic and asymptomatic female urine sample results are from Clinical Study 3.

[2] Includes equivocal results from Panther AC2 testing. Equivocal results from AC2 testing are considered indeterminate; a new specimen should be collected.

[3] Score CI.

Table 11 shows the sensitivity, specificity, PPV, and NPV of the Aptima Combo 2 Assay for GC detection and the prevalence of GC (based on the infected status) in male urine samples and urethral swab specimens, and in female vaginal swab, endocervical swab, and PCyt specimens by symptom status. GC prevalence was higher in symptomatic men but similar in symptomatic and asymptomatic women.

Table 12 shows the PPA and NPA of the Aptima Combo 2 Assay for CT detection based on the CCA in female urine samples by symptom status.

Table 11: Performance Characteristics of the Aptima Combo 2 Assay for GC Detection by Symptom Status in Female and Male Specimens

Specimen Type[1]	Symptom Status	n	TP	FP	TN	FN	Prev %	Sensitivity % (95% CI)[2]	Specificity % (95% CI)[2]	PPV % (95% CI)[3]	NPV % (95% CI)[3]
CVS/PVS	Sym	802	27	4	771	0	3.4	100 (87.5-100)	99.5 (98.7-99.8)	87.1 (72.6-96.1)	100 (99.6-100)
	Asym	456	15	1	439	1	3.5	93.8 (71.7-98.9)	99.8 (98.7-100)	93.8 (74.0-99.8)	99.8 (98.9-100)
PCyt	Sym	829	27	0	802	0	3.3	100 (87.5-100)	100 (99.5-100)	100 (88.0-100)	100 (99.6-100)
	Asym	464	16	0	448	0	3.4	100 (80.6-100)	100 (99.1-100)	100 (81.3-100)	100 (99.3-100)
FS	Sym	785	26	1	758	0	3.3	100 (87.1-100)	99.9 (99.3-100)	96.3 (82.4-99.9)	100 (99.5-100)
	Asym	453	16	1	436	0	3.5	100 (80.6-100)	99.8 (98.7-100)	94.1 (74.3-99.8)	100 (99.3-100)
MS	Sym	236	31	0	205	0	13.1	100 (89.0-100)	100 (98.2-100)	100 (89.5-100)	100 (98.3-100)
	Asym	310	3	0	307	0	1.0	100 (43.9-100)	100 (98.8-100)	100 (44.4-100)	100 (99.3-100)
MU	Sym	497	66	1	430	0	13.3	100 (94.5-100)	99.8 (98.7-100)	98.5 (92.3-100)	100 (99.2-100)
	Asym	1300	9	4	1286	1	0.8	90.0 (59.6-98.2)	99.7 (99.2-99.9)	69.2 (45.6-91.7)	99.9 (99.7-100)

Asym = asymptomatic, CI = confidence interval, CVS = clinician-collected vaginal swab, FN = false negative, FP = false positive, FS = female endocervical swab, MS = male urethral swab, MU = male urine, NPV = negative predictive value, PCyt = PreservCyt Solution liquid Pap, PPV = positive predictive value, Prev = prevalence, PVS = patient-collected vaginal swab, Sym = symptomatic, TN = true negative, TP = true positive.

[1] Vaginal swab, PreservCyt Solution liquid Pap, endocervical swab, and male urethral swab sample results are from Clinical Study 1. Symptomatic male urine sample results are from Clinical Study 2, and asymptomatic male urine sample results are from Clinical Studies 1 and 2.

[2] Score CI.

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

Table 12: Performance Characteristics of the Aptima Combo 2 Assay for GC Detection by Symptom Status in Female Urine Samples

Specimen Type[1]	Symptom Status	n	CCA+ AC2+	CCA- AC2+	CCA- AC2-	CCA+ AC2-[2]	PPA % (95% CI)[3]	NPA % (95% CI)[3]
FU	Sym	1383	19	0	1363[4]	1	95.0 (76.4-99.1)	100 (99.7-100)
	Asym	1196	9	0	1187[5]	0	100 (70.1-100)	100 (99.7-100)

AC2 = Aptima Combo 2 Assay, Asym = asymptomatic, CCA = composite comparator algorithm, CI = confidence interval, FU = female urine, NPA = negative percent agreement, PPA = positive percent agreement, Sym = symptomatic.

[1] Symptomatic and asymptomatic female urine sample results are from Clinical Study 3.

[2] Includes equivocal results from Panther AC2 testing. Equivocal results from AC2 testing are considered indeterminate; a new specimen should be collected.

[3] Score CI.

[4] 5/1363 subjects had at least one positive GC vaginal swab sample result by a reference NAAT; one or more vaginal swab sample reference results were not available for 11/1363 subjects; 1347/1363 subjects had negative vaginal swab sample reference results.

[5] 6/1187 subjects had at least one positive GC vaginal swab sample result by a reference NAAT; one or more vaginal swab sample reference results were not available for 11/1187 subjects; 1170/1187 asymptomatic subjects had negative vaginal swab sample reference results.

Chlamydia trachomatis Infected Status Tables

The frequency of test outcomes from reference NAAT and investigational Panther System testing is summarized in Tables 13 through 16 for CT.

Table 13: Clinical Study 1. CT Infected Status for Performance Evaluation in Female Vaginal Swab, PreservCyt Solution Liquid Pap, and Endocervical Swab Samples

CT Infected Status	Assay Results							Symptom Status	
	AC2 Tigris		ACT Tigris		AC2 Panther				
	PCyt	FU	PCyt	FU	CVS/PVS	PCyt	FS	Sym	Asym
Infected	+	+	+	+	+	+	+	62	26
Infected	+	+	+	+	+	+	-	0	1
Infected	+	+	+	+	+	+	NA	3	0
Infected	+	+	+	+	+	-	+	0	2
Infected	+	+	+	+	-	+	+	0	1
Infected	+	+	+	+	NA	+	+	1	1
Infected	+	+	+	+	NA	+	NA	2	1
Infected	+	-	+	+	+	+	+	4	1
Infected	+	-	+	+	NA	+	NA	0	1
Infected	+	-	+	-	+	+	+	4	0
Infected	+	-	+	-	-	+	-	0	1
Infected	+	-	+	-	NA	+	+	0	1
Infected	+	NA	+	NA	+	+	+	0	1
Infected	+	NA	+	NA	-	+	-	0	1
Infected[1]	-	+	-	+	+	-	+	1	0
Infected[1]	-	+	-	+	+	-	-	2	0
Infected[1]	-	+	-	+	-	-	-	1	1
Not Infected	+	-	-	-	-	-	-	0	2
Not Infected	-	+	-	-	-	-	-	1	0
Not Infected	-	-	+	-	+	-	+	0	1
Not Infected	-	-	+	-	-	-	-	5	0
Not Infected	-	-	-	+	+	-	-	0	1
Not Infected	-	-	-	+	+	-	NA	0	1
Not Infected	-	-	-	+	-	-	-	1	3
Not Infected	-	-	-	-	+	-	+	1	0
Not Infected	-	-	-	-	+	-	-	2	7
Not Infected	-	-	-	-	+	-	NA	2	0
Not Infected	-	-	-	-	-	-	+	2	2
Not Infected	-	-	-	-	-	-	-	680	396
Not Infected	-	-	-	-	-	-	NA	29	8
Not Infected	-	-	-	-	-	NA	-	1	0
Not Infected	-	-	-	-	NA	-	-	17	4
Not Infected	-	-	-	-	NA	-	NA	8	1
Not Infected	-	NA	-	-	-	-	-	8	6
Not Infected	-	NA	-	-	-	-	NA	0	1
Not Infected	NA	-	-	-	-	-	-	0	1
Not Infected	NA	-	-	-	-	-	NA	1	0
Not Infected	NA	-	-	-	NA	-	+	1	0

AC2 = Aptima Combo 2 Assay, ACT = Aptima CT Assay, Asym = asymptomatic, CVS = clinician-collected vaginal swab, FS = female endocervical swab, FU = female urine, NA = result not available, Panther = Panther System, PCyt = PreservCyt Solution liquid Pap, PVS = patient-collected vaginal swab, Sym = symptomatic, Tigris = Tigris DTS System.

[1] For the evaluation of the non-urine specimen types, the specimens were considered non-infected.

Table 14: Clinical Study 1. CT Infected Status for Performance Evaluation in Male Urethral Swab Samples

CT Infected Status	Assay Results					Symptom Status	
	AC2 DTS		ACT Tigris		AC2 Panther		
	MS	MU	MS	MU	MS	Sym	Asym
Infected	+	+	+	+	+	50	37
Infected	+	+	+	+	NA	4	1
Infected	+	+	+	-	+	2	0
Infected	+	-	+	+	+	4	2
Infected	+	-	+	-	+	3	2
Not Infected	+	+	-	-	-	0	1
Not Infected	+	-	-	-	+	0	1
Not Infected	+	-	-	-	-	1	1
Not Infected	-	-	+	-	-	3	2
Not Infected	-	-	-	+	-	1	1
Not Infected	-	-	-	-	+	1	2
Not Infected	-	-	-	-	-	173	262
Not Infected	-	-	-	-	NA	10	9
Not Infected	NA	-	-	-	NA	1	2

AC2 = Aptima Combo 2 Assay, ACT = Aptima CT Assay, Asym = asymptomatic, DTS = DTS Systems, MS = male urethral swab, MU = male urine, NA = result not available, Panther = Panther System, Sym = symptomatic, Tigris = Tigris DTS System.

Table 15: Clinical Study 1 and Clinical Study 2. CT Infected Status for Performance Evaluation in Male Urine Samples

CT Infected Status	Assay Results							Symptom Status	
	AC2[1]		ACT Tigris		NAAT 1[3]	NAAT 2[3]	AC2 Panther		
	MS	MU	MS	MU	MU	MU	MU	Sym	Asym
Clinical Study 1									
Infected	+	+	+	+			+		38
Infected	+	-	+	+			+		2
Infected	+	-	+	-			-		2
Clinical Study 2									
Infected	+	+			+	+	+	73	66
Infected	+	+			+	+	-	2	1
Infected	+	+			+	-	+	0	1
Infected	+	+			+	NA	+	0	1
Infected	+	+			-	+	+	3	0
Infected	+	+			-	+	-	0	1
Infected	+	-			+	+	+	4	0
Infected	+	-			+	+	-	3	0
Infected	+	=			-	+	-	0	1
Infected	-	+			+	+	+	5	4
Clinical Study 1									
Not Infected	+	+	-	-			-		1
Not Infected	+	-	-	-			-		2
Not Infected	-	-	+	-			-		2
Not Infected	-	-	-	+			+		1

Table 15: Clinical Study 1 and Clinical Study 2. CT Infected Status for Performance Evaluation in Male Urine Samples (Continued)

CT Infected Status	Assay Results							Symptom Status	
	AC2[1]		ACT Tigris		NAAT 1[3]	NAAT 2[3]	AC2 Panther		
	MS	MU	MS	MU	MU	MU	MU	Sym	Asym
Not Infected	-	-	-	-			-		273
Not Infected	NA	-	-	-			-		2
Clinical Study 2									
Not Infected	+	-			-	-	-	1	6
Not Infected	-	+			-	-	+	0	1
Not Infected	-	-			+	-	+	1	0
Not Infected	-	-			+	-	-	0	2
Not Infected	-	-			-	-	-	388	874
Not Infected	-	-			-	=	-	0	1
Not Infected	-	-			-	NA	-	10	18
Not Infected	-	-			NA	-	-	1	2
Not Infected	-	NA			-	-	-	2	0
Not Infected	NA	-			-	-	-	4	0

AC2 = Aptima Combo 2 Assay, ACT = Aptima CT Assay, Asym = asymptomatic, MS = male urethral swab, MU = male urine, NA = result not available, Panther = Panther System, Sym = symptomatic, Tigris = Tigris DTS System.

The equal symbol (=) represents an equivocal result.

[1] Male urethral swab and male urine samples were tested with the Aptima Combo 2 Assay on the DTS Systems in Clinical Study 1 and on the Tigris DTS System in Clinical Study 2.

[2] Male urethral swab and male urine samples were tested with the Aptima CT Assay on the Tigris DTS System in Clinical Study 1.

[3] Male urine samples were tested with two FDA-cleared CT NAATs in Clinical Study 2.

Note. Data from asymptomatic men in Clinical Study 1 are combined with data from Clinical Study 2.

Table 16: Clinical Study 3. CT Composite Comparator Status for Performance Evaluation in Female Urine Samples

Composite Comparator Status	Assay Results				Symptom Status	
	NAAT 1	NAAT 2	NAAT 3	AC2 Panther	Sym	Asym
	FU	FU	FU	FU		
Positive	+	+	NR	+	101	61
Positive	+	+	NR	-	1	0
Positive	+	+	NR	=	0	1
Positive	+	-	+	+	4	4
Positive	-	+	+	+	3	0
Positive	=	+	+	+	1	0
Negative	-	+	-	+	1	0
Negative	-	+	-	-	3	1
Negative	-	-	NR	+	1	3
Negative	-	-	NR	-	1261	1119
Negative	-	NA	-	-	1	1
Negative	NA	-	-	-	2	3

Asym = asymptomatic, FU = female urine, NA = result not available, NR = not required, AC2 Panther = Aptima Combo 2 assay on the Panther system, Sym = symptomatic.

The equal symbol (=) represents final equivocal result.

Neisseria gonorrhoeae Infected Status Tables

The frequency of test outcomes from reference NAAT and investigational Panther System testing is summarized in Tables 17 through 20 for GC.

Table 17: Clinical Study 1. GC Infected Status for Performance Evaluation in Female Vaginal Swab, PreservCyt Solution Liquid Pap, and Endocervical Swab Samples

GC Infected Status	Assay Results							Symptom Status	
	AC2 Tigris		AGC Tigris		AC2 Panther				
	PCyt	FU	PCyt	FU	CVS/PVS	PCyt	FS	Sym	Asym
Infected	+	+	+	+	+	+	+	22	10
Infected	+	+	+	+	+	+	NA	1	0
Infected	+	+	+	-	+	+	+	1	0
Infected	+	+	+	=	+	+	+	0	1
Infected	+	-	+	-	+	+	+	3	3
Infected	+	-	+	-	-	+	+	0	1
Infected	+	NA	+	NA	+	+	+	0	1
Not Infected	+	NA	-	-	-	=	-	0	1
Not Infected	-	-	NA	NA	+	-	+	0	1
Not Infected	-	-	NA	NA	+	-	-	3	0
Not Infected	-	-	NA	NA	+	-	NA	1	0
Not Infected	-	-	NA	NA	-	-	+	1	0
Not Infected	-	-	NA	NA	-	-	-	736	429
Not Infected	-	-	NA	NA	-	-	=	1	0
Not Infected	-	-	NA	NA	-	-	NA	32	9
Not Infected	-	-	NA	NA	-	NA	-	1	0
Not Infected	-	-	NA	NA	NA	-	-	18	6
Not Infected	-	-	NA	NA	NA	-	NA	10	3

AC2 = Aptima Combo 2 Assay, AGC = Aptima GC Assay, Asym = asymptomatic, CVS = clinician-collected vaginal swab, FS = female endocervical swab, FU = female urine, NA = result not available, Panther = Panther System, PCyt = PreservCyt Solution liquid Pap, PVS = patient-collected vaginal swab, Sym = symptomatic, Tigris = Tigris DTS System.

The equal symbol (=) represents an equivocal result on repeat testing.

Table 18: Clinical Study 1. GC Infected Status for Performance Evaluation in Male Urethral Swab Samples

GC Infected Status	Assay Results					Symptom Status	
	AC2 DTS		AGC DTS		AC2 Panther		
	MS	MU	MS	MU	MS	Sym	Asym
Infected	+	+	+	+	+	30	2
Infected	+	+	+	+	NA	0	1
Infected	+	-	+	-	+	1	1
Infected	NA	+	NA	+	NA	1	0
Not Infected	-	-	NA	NA	-	205	307
Not Infected	-	-	NA	NA	NA	14	9

AC2 = Aptima Combo 2 Assay, AGC = Aptima GC Assay, Asym = asymptomatic, DTS = DTS Systems, MS = male urethral swab, MU = male urine, NA = result not available, Panther = Panther System, Sym = symptomatic.

Table 19: Clinical Study 1 and Clinical Study 2. GC Infected Status for Performance Evaluation in Male Urine Samples

GC Infected Status	Assay Results							Symptom Status	
	AC2[1]		AGC DTS[2]		NAAT 1[3]	NAAT 2[3]	AC2 Panther		
	MS	MU	MS	MU	MU	MU	MU	Sym	Asym
Clinical Study 1									
Infected	+	+	+	+			+		3
Infected	+	-	+	-			-		1
Clinical Study 2									
Infected	+	+			+	+	+	63	4
Infected	+	+			+	NA	+	1	1
Infected	-	+			+	-	+	0	1
Infected	NA	+			+	+	+	2	0
Clinical Study 1									
Not Infected	-	-	NA	NA			+		2
Not Infected	-	-	NA	NA			-		314
Clinical Study 2									
Not Infected	+	-			-	-	-	2	4
Not Infected	-	+			-	-	+	0	1
Not Infected	-	-			+	-	-	6	2
Not Infected	-	-			-	+	-	1	0
Not Infected	-	-			-	-	+	1	1
Not Infected	-	-			-	-	-	407	945
Not Infected	-	-			-	NA	-	9	19
Not Infected	-	-			NA	-	-	1	2
Not Infected	-	NA			-	-	-	2	0
Not Infected	NA	-			-	-	-	2	0

AC2 = Aptima Combo 2 Assay, AGC = Aptima GC Assay, Asym = asymptomatic, DTS = DTS Systems, MS = male urethral swab, MU = male urine, NA = result not available, Panther = Panther System, Sym = symptomatic.

[1] Male urethral swab and male urine samples were tested with the Aptima Combo 2 Assay on the DTS Systems in Clinical Study 1 and on the Tigris DTS System in Clinical Study 2.

[2] Male urethral swab and male urine samples were tested with the Aptima GC Assay on the DTS Systems in Clinical Study 1.

[3] Male urine samples were tested with two FDA-cleared GC NAATs in Clinical Study 2.

Note. Data from asymptomatic men in Clinical Study 1 are combined with data from Clinical Study 2.

Table 20: Clinical Study 3. GC Composite Comparator Status for Performance Evaluation in Female Urine Samples

Composite Comparator Status	Assay Results				Symptom Status	
	NAAT 1 FU	NAAT 2 FU	NAAT 3 FU	AC2 Panther FU	Sym	Asym
Positive	+	+	NR	+	19	9
Positive	=	+	+	=	1	0
Negative	-	-	NR	-	1360	1183
Negative	-	NA	-	-	1	1
Negative	NA	-	-	-	2	3

Asym = asymptomatic, FU = female urine, NA = result not available, NR = not required, AC2 Panther = Aptima Combo 2 assay on the Panther system, Sym = symptomatic.
The equal symbol (=) represents final equivocal result.

RLU Distribution of Aptima Combo 2 Controls

The distribution of the RLU values for the Aptima Combo 2 controls is presented in Table 21 from all valid Panther System runs performed during Clinical Study 1, Clinical Study 2, and Clinical Study 3.

Table 21: RLU Distribution of Aptima Combo 2 Controls

Control	Statistic	Total RLU (x1000)		
		Clinical Study 1	Clinical Study 2	Clinical Study 3
Positive Control, CT/ Negative Control, GC	N	66	23	41
	Maximum	1335	1258	1577
	Median	1081.5	1135.0	1091.0
	Minimum	624	910	771
	CV%	11.2	7.5	13.5
Positive Control, GC/ Negative Control, CT	N	66	23	41
	Maximum	1241	1311	1308
	Median	1172.0	1174.0	1060.0
	Minimum	1063	1082	905
	CV%	3.2	4.9	8.9

Reproducibility Studies

Reproducibility of the Aptima Combo 2 Assay on the Panther System was evaluated in two different studies using panel members created with Specimen Transport Medium (STM) in Reproducibility Study 1 and using panel members created with clinical urine specimens in Reproducibility Study 2.

Reproducibility Study 1

Aptima Combo 2 Assay reproducibility was evaluated with panel members created using STM at three external US laboratories using the Panther System. Testing was performed using one lot of assay reagents and a total of six operators (two at each site). Testing was performed over at least 10 days at each site. The negative panel member consisted of STM and positive panel members were created by spiking STM with lysate from CT and/or GC organisms to result in panel members with expected targeted concentrations. Table 22 shows the CT and GC concentrations for each panel member and the mean, standard deviation (SD), and coefficient of variation (CV) of the RLU data for each panel member between-sites, between-operators, between-days, between-runs, within-runs, and overall. Percent agreement with expected results is also shown. Only samples with valid results were included in the analyses.

Table 22: Reproducibility Study 1 Data

Target Concentration		Agreed/N	Agmt (%)	Mean RLU (x1000)	Between-Sites		Between-Operators		Between-Days		Between-Runs		Within-Runs		Total	
CT (IFU/mL)	GC (CFU/mL)				SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)
0	0	180/180	100	6	1.0	17.5	0.5	8.1	0.2	3.7	0.5	8.2	1.5	24.4	1.9	32.4
0.25	0	180/180	100	1207	45.0	3.7	17.3	1.4	0.0	0.0	35.1	2.9	66.9	5.5	89.7	7.4
2.5	0	180/180	100	1272	41.3	3.2	19.2	1.5	0.0	0.0	31.0	2.4	36.8	2.9	66.3	5.2
25	0	180/180	100	1292	43.7	3.4	14.9	1.2	7.7	0.6	35.1	2.7	36.3	2.8	68.8	5.3
1000	0	180/180	100	1294	48.1	3.7	14.3	1.1	26.8	2.1	29.6	2.3	34.8	2.7	73.0	5.6
0	0.25	180/180	100	589	92.2	15.7	19.9	3.4	28.1	4.8	21.2	3.6	44.8	7.6	110.2	18.7
0	12.5	179/179	100	1251	163.5	13.1	0.0	0.0	15.1	1.2	31.5	2.5	29.8	2.4	169.8	13.6
0	125	180/180	100	1295	168.3	13.0	6.7	0.5	33.4	2.6	21.1	1.6	33.3	2.6	176.2	13.6
0	1250	180/180	100	1309	166.5	12.7	0.0	0.0	28.4	2.2	27.6	2.1	31.2	2.4	173.9	13.3
0	2500	179/179	100	1305	170.9	13.1	11.4	0.9	30.4	2.3	15.2	1.2	32.2	2.5	177.5	13.6
2.5	125	178/178	100	2513	123.9	4.9	24.6	1.0	24.0	1.0	57.5	2.3	52.4	2.1	150.3	6.0
2.5	2500	180/180	100	2515	123.5	4.9	6.5	0.3	33.8	1.3	39.3	1.6	59.4	2.4	146.6	5.8
1000	125	179/179	100	2524	117.4	4.6	35.2	1.4	52.1	2.1	28.9	1.1	54.7	2.2	146.8	5.8
1000	2500	180/180	100	2525	118.2	4.7	21.6	0.9	38.7	1.5	54.8	2.2	48.5	1.9	145.9	5.8

Agmt = agreement, CFU = colony-forming unit, CV = coefficient of variation, IFU = inclusion-forming unit, RLU = relative light unit, SD = standard deviation.

Note. Variability from some factors may be numerically negative, which can occur if the variability due to those factors is very small. When this occurs, the variability as measured with standard deviation and %CV is set to 0.

Reproducibility Study 2

Aptima Combo 2 Assay reproducibility was evaluated with panel members created using clinical urine specimens at two external US laboratories and in-house using the Panther System. Testing was performed using one lot of assay reagents and a total of six operators (two at each site). Testing was performed over at least 10 days at each site. The negative panel member consisted of negative urine and the positive panel members were created by spiking negative urine with lysate from CT and/or GC organisms to result in panel members with expected targeted concentrations. Table 23 shows the CT and GC concentrations for each panel member and the mean, SD, and CV of the RLU data for each panel member between-sites, between-operators, between-days, between-runs, within-runs, and overall. Percent agreement with expected results is also shown. Only samples with valid results were included in the analyses.

Table 23: Reproducibility Study 2 Data

Target Concentration		Agreed/N	Agmt (%)	Mean RLU (x1000)	Between-Sites		Between-Operators		Between-Days		Between-Runs		Within-Runs		Total	
CT (IFU/mL)	GC (CFU/mL)				SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)
0	0	178/180	98.9	6	1.2	19.0	0.0	0.0	0.0	0.0	0.0	0.0	8.2	131.7	8.3	133.0
0.25	0	180/180	100	1202	92.4	7.7	0.0	0.0	0.0	0.0	62.9	5.2	50.3	4.2	122.6	10.2
2.5	0	178/178	100	1185	90.9	7.7	0.0	0.0	0.0	0.0	53.8	4.5	34.6	2.9	111.1	9.4
25	0	180/180	100	1265	97.4	7.7	18.9	1.5	0.0	0.0	62.4	4.9	35.1	2.8	122.4	9.7
1000	0	180/180	100	1278	101.9	8.0	15.7	1.2	20.6	1.6	61.4	4.8	31.8	2.5	125.9	9.8
0	0.25	177/179	98.9	422	40.3	9.5	21.9	5.2	27.6	6.5	35.3	8.4	72.7	17.2	96.9	23.0
0	12.5	179/180	99.4	1142	11.9	1.0	0.0	0.0	44.4	3.9	37.3	3.3	75.8	6.6	96.2	8.4
0	125	180/180	100	1224	31.4	2.6	13.0	1.1	11.1	0.9	19.8	1.6	34.3	2.8	53.4	4.4
0	1250	180/180	100	1263	16.7	1.3	9.4	0.7	21.0	1.7	14.0	1.1	30.6	2.4	44.1	3.5
0	2500	180/180	100	1309	20.7	1.6	13.4	1.0	0.0	0.0	21.7	1.7	25.3	1.9	41.4	3.2
2.5	125	180/180	100	2468	71.9	2.9	31.5	1.3	21.7	0.9	64.8	2.6	44.4	1.8	113.1	4.6
2.5	2500	180/180	100	2453	76.2	3.1	30.9	1.3	0.0	0.0	62.5	2.5	51.6	2.1	115.4	4.7
1000	125	179/179	100	2504	74.0	3.0	38.5	1.5	0.0	0.0	59.1	2.4	39.1	1.6	109.4	4.4
1000	2500	180/180	100	2357	79.1	3.4	0.0	0.0	0.0	0.0	74.2	3.1	55.2	2.3	121.7	5.2

Agmt = agreement, CFU = colony-forming unit, CV = coefficient of variation, IFU = inclusion-forming unit, RLU = relative light unit, SD = standard deviation.

Note. Variability from some factors may be numerically negative, which can occur if the variability due to those factors is very small. When this occurs, the variability as measured with standard deviation and %CV is set to 0.

Panther System Analytical Performance

Analytical Sensitivity Study

Chlamydia trachomatis analytical sensitivity (limit of detection) was determined by testing dilutions of CT organisms in the Aptima Combo 2 Assay. The analytical sensitivity claim for the assay is 1 IFU/assay (7.25 IFU/swab, 9.75 IFU/mL PreservCyt Solution liquid Pap, 5.0 IFU/mL urine). However, dilutions of less than 1 IFU/assay tested positive in the Aptima Combo 2 Assay for the following 12 serovars: D, E, F, G, H, I, J, K, L1, L2, L2a, and L3 (≥95% positivity was observed in samples containing CT concentrations of 1.89 IFU/mL).

Neisseria gonorrhoeae analytical sensitivity (limit of detection) was determined by testing dilutions of GC organisms in the Aptima Combo 2 Assay. The analytical sensitivity claim for the assay is 50 cells/assay (362 cells/swab, 488 cells/mL PreservCyt Solution liquid Pap, 250 cells/mL urine). However, dilutions of less than 50 cells/assay tested positive in the Aptima Combo 2 Assay for 30 different strains of GC (≥95% positivity was observed in samples containing GC concentrations of 0.36 cells/mL).

Analytical Specificity

The Aptima Combo 2 Assay formulation for the Panther System has not changed from that used with DTS® Systems or the Tigris® DTS System. The analytical specificity study was conducted on DTS Systems. A total of 154 culture isolates were evaluated using the Aptima Combo 2 Assay. These isolates included 86 organisms that may be isolated from the urogenital tract and 68 additional organisms that represent a phylogenetic cross-section of organisms. The tested organisms included bacteria, fungi, yeast, parasites, and viruses. All organisms except *C. psittaci*, *C. pneumoniae*, and the viruses were tested at 1.0 x 10^6 cells/assay in STM. The Chlamydia and Neisseria organisms were tested in PreservCyt Solution medium. *C. psittaci* and *C. pneumoniae* were tested at 1.0 x 10^5 IFU/assay. The viruses were tested as follows: (a) herpes simplex viruses I and II: 2.5 x 10^4 $TCID_{50}$/assay, (b) human papilloma virus 16: 2.9 x 10^6 DNA copies/assay, and (c) cytomegalovirus: 4.8 x 10^5 infected cell culture cells/assay. Only CT and GC samples produced positive results in the Aptima Combo 2 Assay. The list of organisms tested is shown in Table 24.

Table 24: Analytical Specificity

Organism	Organism	Organism
Achromobacter xerosis	*Escherichia coli*	*Neisseria mucosa (3)*
Acinetobacter calcoaceticus	*Flavobacterium meningosepticum*	*Neisseria sicca (3)*
Acinetobacter lwoffi	*Fusobacterium nucleatum*	*Neisseria subflava (14)*
Actinomyces israelii	*Gardnerella vaginalis*	*Neisseria perflava*
Actinomyces pyogenes	*Gemella haemolysans*	*Neisseria polysaccharea*
Aerococcus viridans	*Haemophilus ducreyi*	*Paracoccus denitrificans*
Aeromonas hydrophila	*Haemophilus influenzae*	*Peptostreptococcus anaerobius*
Agrobacterium radiobacter	Herpes simplex virus I	*Peptostreptococcus productus*
Alcaligenes faecalis	Herpes simplex virus II	*Plesiomonas shigelloides*
Bacillus subtilis	Human papilloma virus 16	*Propionibacterium acnes*
Bacteriodes fragilis	*Kingella dentrificans*	*Proteus mirabilis*
Bacteriodes ureolyticus	*Kingella kingae*	*Proteus vulgaris*
Bifidobacterium adolescentis	*Klebsiella oxytoca*	*Providencia stuartii*

"(n)" represents the number of strains tested.

All organisms tested produced a negative result in the Aptima Combo 2 Assay based on kinetic profile type and RLU.

Table 24: Analytical Specificity (Continued)

Organism	Organism	Organism
Bifidobacterium brevi	*Klebsiella pneumoniae*	*Pseudomonas aeruginosa*
Branhamella catarrhalis	*Lactobacillus acidophilus*	*Pseudomonas fluorescens*
Brevibacterium linens	*Lactobacillus brevis*	*Pseudomonas putida*
Campylobacter jejuni	*Lactobacillus jensonii*	*Rahnella aquatilis*
Candida albicans	*Lactobacillus lactis*	*Rhodospirillum rubrum*
Candida glabrata	*Legionella pneumophila (2)*	*Saccharomyces cerevisiae*
Candida parapsilosis	*Leuconostoc paramensenteroides*	*Salmonella minnesota*
Candida tropicalis	*Listeria monocytogenes*	*Salmonella typhimurium*
Chlamydia pneumoniae	*Micrococcus luteus*	*Serratia marcescens*
Chlamydia psittaci (2)	*Moraxella lacunata*	*Staphylococcus saprophyticus*
Chromobacterium violaceum	*Moraxella osloensis*	*Staphylococcus aureus*
Citrobacter freundii	*Morganella morganii*	*Staphylococcus epidermidis*
Clostridium perfringens	*Mycobacterium smegmatis*	*Streptococcus agalactiae*
Corynebacterium genitalium	*Mycoplasma genitalium*	*Streptococcus bovis*
Corynebacterium xerosis	*Mycoplasma hominis*	*Streptococcus mitis*
Cryptococcus neoformans	*N. meningitidis* Serogroup A	*Streptococcus mutans*
Cytomegalovirus	*N. meningitidis* Serogroup B	*Streptococcus pneumoniae*
Deinococcus radiodurans	*N. meningitidis* Serogroup C (4)	*Streptococcus pyogenes*
Derxia gummosa	*N. meningitidis* Serogroup D	*Streptococcus salivarius*
Eikenella corrodens	*N. meningitidis* Serogroup Y	*Streptococcus sanguis*
Enterobacter aerogenes	*N. meningitidis* Serogroup W135	*Streptomyces griseinus*
Enterobacter cloacae	*Neisseria cinerea (4)*	*Trichomonas vaginalis*
Entercoccus avium	*Neisseria dentrificans*	*Ureaplasma urealyticum*
Entercoccus faecalis	*Neisseria elongata (3)*	*Vibrio parahaemolyticus*
Entercoccus faecium	*Neisseria flava*	*Yersinia enterocolitica*
Erwinia herbicola	*Neisseria flavescens (2)*	
Erysipelothrix rhusiopathiae	*Neisseria lactamica (9)*	

"(n)" represents the number of strains tested.

All organisms tested produced a negative result in the Aptima Combo 2 Assay based on kinetic profile type and RLU.

Interfering Substances

The Aptima Combo 2 Assay formulation for the Panther System has not changed from that used with DTS Systems or the Tigris DTS System. Blood interference was evaluated on the Panther System and the results of this testing indicated that blood does not interfere with Aptima Combo 2 Assay performance.

Aptima Combo 2 Assay performance in the presence of potentially interfering substances was tested on DTS Systems. The following interfering substances were individually spiked into swab and PreservCyt Solution liquid Pap specimens: 10% blood, contraceptive jelly, spermicide, moisturizer, hemorrhoidal anesthetic, body oil, powder, anti-fungal cream, vaginal lubricants, feminine spray, and leukocytes (1.0×10^6 cells/mL). All were tested for potential assay interference in the absence and presence of CT and GC at the estimated rRNA equivalent of 1.0 CT IFU/assay (5 fg/assay) and 50 GC cells/assay (250 fg/assay). The rRNA equivalents were calculated based on the genome size and estimated DNA:RNA ratio/cell of each organism.

No interference was observed with any of the tested substances. No inhibitors of amplification were observed in the Aptima Combo 2 Assay.

Within Laboratory Precision Study

Aptima Combo 2 Assay precision was evaluated at Hologic using the Panther System. Testing was performed using three Panther Systems and three lots of assay reagents. Testing was performed over 24 days.

Reproducibility panel members were created using negative PreservCyt Solution liquid Pap specimens and STM. The positive panel members were created by spiking CT and/or GC organisms to the targeted concentrations shown in Table 25.

For each panel member, Table 25 presents mean RLU, between-instrument, between-lot, between-run, within-run, and overall variation as SD and percent CV. Percent agreement with expected results is also shown.

Table 25: Within Laboratory Precision Data

Matrix	Target Concentration		Agreed/N	Agrmt (%)	Mean RLU (x1000)	Between-Instruments		Between-Lots		Between-Runs		Within-Runs		Total	
	CT (IFU/mL)	GC (CFU/mL)				SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)	SD (x1000)	CV (%)
STM	0	0	96/96	100	6	0.1	1.0	0.9	13.5	0.0	0.0	1.0	15.7	1.3	20.1
	0.25	0	95/95	100	1226	70.0	5.7	20.0	1.6	8.4	0.7	47.1	3.8	87.1	7.1
	2.5	0	96/96	100	1249	78.0	6.2	6.1	0.5	0.0	0.0	32.9	2.6	84.8	6.8
	25	0	95/95	100	1268	72.9	5.7	15.3	1.2	0.0	0.0	39.6	3.1	84.3	6.6
	0	12.5	96/96	100	1081	18.4	1.7	28.6	2.6	0.0	0.0	26.7	2.5	43.2	4.0
	0	125	96/96	100	1266	29.8	2.4	0.0	0.0	8.9	0.7	27.6	2.2	41.6	3.3
	0	1250	96/96	100	1309	29.4	2.2	0.0	0.0	9.8	0.8	31.8	2.4	44.4	3.4
	2.5	125	96/96	100	2456	86.6	3.5	0.0	0.0	0.0	0.0	53.0	2.2	101.5	4.1
	2.5	2500	96/96	100	2509	73.1	2.9	0.0	0.0	19.8	0.8	46.8	1.9	89.0	3.5
	1000	2500	96/96	100	2496	31.7	1.3	6.1	0.2	0.0	0.0	193.7	7.8	196.3	7.9
	1000	125	96/96	100	2471	83.6	3.4	9.4	0.4	0.0	0.0	52.4	2.1	99.1	4.0
PCyt	0	0	96/96	100	7	0.0	0.0	0.8	11.7	0.0	0.0	1.5	22.4	1.7	24.7
	0.25	0	96/96	100	1113	92.3	8.3	30.1	2.7	0.0	0.0	63.6	5.7	116.0	10.4
	2.5	0	96/96	100	1194	62.5	5.2	24.8	2.1	0.0	0.0	47.0	3.9	82.1	6.9
	25	0	95/95	100	1222	65.1	5.3	26.4	2.2	14.7	1.2	35.0	2.9	79.8	6.5
	0	12.5	93/93	100	994	33.3	3.3	36.9	3.7	16.0	1.6	26.2	2.6	58.4	5.9
	0	125	95/95	100	1189	40.1	3.4	4.5	0.4	10.9	0.9	21.4	1.8	47.0	4.0
	0	1250	95/95	100	1239	37.7	3.0	7.5	0.6	13.6	1.1	18.0	1.5	44.6	3.6
	2.5	125	95/95	100	2333	99.7	4.3	35.3	1.5	12.6	0.5	48.9	2.1	117.2	5.0

Agrmt = agreement, CFU = colony-forming unit, CV = coefficient of variation, IFU = inclusion-forming unit, N = number of samples, PCyt = PreservCyt Solution liquid Pap, RLU = relative light unit, SD = standard deviation, STM = specimen transport medium.
Note: Variability from some factors may be numerically negative, which can occur if the variability due to those factors is very small. When this occurs, the variability as measured with standard deviation and %CV is set to 0.

Carryover Studies for the Panther System

Two studies were conducted to evaluate carryover on the Panther System. In the first study, carryover was assessed in multiple runs on three Panther Systems with approximately 20% high titer GC samples dispersed between negative samples. The runs included clusters of high positive samples with clusters of negative samples as well as single high positives dispersed within the run. High titer samples were made using GC rRNA spiked into STM to give a final concentration equivalent to 2.5×10^5 CFU/mL. Five runs were performed on each of three Panther Systems. Carryover was calculated from a total of 2938 valid negative results. The overall carryover rate from this study was 0% with a 95% confidence interval of 0–0.1%.

The second carryover study was conducted on one Panther System with high titer GC positive samples (GC rRNA spiked into STM at the equivalent of 2.5×10^5 CFU/mL) alternately processed with negative samples in a checkerboard format. Five checkerboard runs were performed. The overall carryover rate from this study was 0.74% (1/135 negative samples).

Specimen Stability Studies

Specimen stability was evaluated using the DTS Systems and/or the Tigris DTS System.

A. Endocervical Swab Specimens

Data to support the recommended shipping and storage conditions for endocervical swab samples were generated with pooled negative swab samples. Five pooled samples were spiked with CT and GC at final concentrations of 10 IFU and 100 CFU per reaction, respectively. The spiked samples were held at -70°C, -20°C, 4°C, and 30°C. Samples were tested in duplicate at days 0, 20, 35, 60, and 90. All test conditions were positive for both CT and GC at all times and temperatures.

B. PreservCyt Solution Liquid Pap Specimens

Data to support the recommended shipping and storage conditions for PreservCyt Solution liquid Pap samples were generated with pooled negative PreservCyt Solution liquid Pap samples. Four pooled samples were spiked with CT and GC at final concentrations of 10 IFU and 100 CFU per reaction, respectively. The PreservCyt Solution liquid Pap samples were placed at 30°C for 7 days, after which 1.0 mL of the sample was added to an Aptima Transfer Tube. The spiked samples were held at 4°C, 10°C and 30°C. Samples stored at 4°C and 10°C were tested in duplicate at days 0, 6, 13, 26, 30 and 36. Samples stored at 30°C were tested in duplicate at days 0, 5, 8, 14 and 17. Four spiked PreservCyt Solution liquid Pap sample pools were added to Aptima Transfer Tubes and placed at 30°C for 14 days before being stored at either -20°C or -70°C. The -20°C samples and the -70°C samples were tested in duplicate after 0, 30, 60, 90 and 106 days of storage. All test conditions were positive for both CT and GC at all times and temperatures.

C. Vaginal Swab Specimens

Data to support the recommended shipping and storage conditions for vaginal swab samples were generated with pooled negative swab samples. Fifteen vaginal swab pools were spiked with CT and GC at final concentrations of 1.0 IFU and 50 CFU per reaction, respectively. The spiked samples were held at -70°C, -20°C, 4°C, and 30°C. Samples were tested using one aliquot at days 0, 20, 36, 73, and 114. All test conditions were positive for both CT and GC at all times and temperatures.

D. Urine Specimens

Data to support the recommended shipping and storage conditions for urine samples were generated with ten female and ten male negative urine samples. The urine samples were spiked with CT and GC at final concentrations of 10 IFU and 100 IFU per reaction, respectively. Two sets of the spiked urine samples were held at 4°C and 30°C for 24 hours prior to being added to the Urine Transport Media (UTM). The two sets of UTM samples then were held at 4°C and 30°C, and tested in triplicate at days 0, 1, 5, 20, and 35. All samples were positive for both CT and GC when the urine samples were held at 4°C prior to addition of the UTM. When the urine samples were held at 30°C prior to addition of the UTM, all of the samples were positive for CT and 95% of the samples were positive for GC at Day 35. These same samples were tested after 116 days of storage at -20°C and -70°C. All samples were positive for both CT and GC under both storage conditions.

E. Additional Frozen (at -20°C) Specimen Stability Study

Data to support the recommended storage condition at -20°C for endocervical swab, urethral swab, vaginal swab, and PreservCyt Solution liquid Pap specimens were generated using 90 specimens for each type with negative result, where 30 specimens were spiked with CT and GC at 1.0 IFU and 50 CFU per reaction, respectively; 30 specimens were spiked at 0.1 IFU and 5 CFU per reaction, respectively; and 30 specimens were unspiked. The specimens were stored at -20°C and were tested at days 0, 200, and 400 days. All spiked specimens met the acceptance criteria of 95% agreement with expected results.

Bibliography

1. **Beem, M. O., and E. M. Saxon**. 1977. Respiratory tract colonization and a distinctive pneumonia syndrome in infants infected with *Chlamydia trachomatis*. NEJM **296**:306-310.
2. **Buimer, M., G. J. J. Van Doornum, S. Ching, P. G. H. Peerbooms, P. K. Plier, D. Ram, and H. H. Lee**. 1996. Detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* by Ligase chain reaction-based assays with clinical specimens from various sites: implications for diagnostic testing and screening. J. Clin. Microbiol. **34**:2395-2400.
3. **Cates, Jr., W., and J. N. Wasserheit**. 1991. Genital chlamydia infections: epidemiology and reproductive sequelae. Am. J. Obstet. Gynecol. **164**:1771-1781.
4. **Centers for Disease Control and Prevention.** 2002. United States Morbid. and Mortal. Weekly Rep. 51 (RR-15).
5. **Centers for Disease Control and Prevention.** 2014. United States Morbid. and Mortal. Weekly Rep. 63 (No. 2).
6. **Centers for Disease Control and Prevention.** 2017. *Sexually Transmitted Disease Surveillance 2016*. Atlanta, GA: U.S. Department of Health and Human Services. September.
7. **Chernesky, M. A., D. Jang, J. Sellors, K. Luinstra, S. Chong, S. Castriciano, and J. B. Mahony**. 1996. Urinary inhibitors of polymerase chain reaction and Ligase chain reaction and testing of multiple specimens may contribute to lower assay sensitivities for diagnosing *Chlamydia trachomatis* infected women. Mol. Cell. Probes. **11**:243-249.
8. **Ching, S., H. Lee, E. W. Hook, III, M. R. Jacobs, and J. Zenilman.** 1995. Ligase chain reaction for detection of *Neisseria gonorrhoeae* in urogenital swabs. J. Clin. Microbiol. **33**:3111-3114.
9. **Chong, S., D. Jang, X. Song, J. Mahoney, A. Petrich, P. Barriga, and M. Chernesky.** 2003. Specimen processing and concentration of *Chlamydia trachomatis* added can influence false-negative rates in the LCx assay but not in the Aptima Combo 2 Assay when testing for inhibitors. J. Clin. Microbiol. **41**:778-782.
10. **Crotchfelt, K. A., B. Pare, C. Gaydos, and T. C. Quinn.** 1998. Detection of *Chlamydia trachomatis* by the Hologic AMPLIFIED Chlamydia Trachomatis assay (AMP CT) in urine specimens from men and women and endocervical specimens from women. J. Clin. Microbiol. **36**:391-394.
11. **Farrel, D. J**. 1999. Evaluation of AMPLICOR *Neisseria gonorrhoeae* PCR using cppB nested PCR and 16S rRNA PCR. J. Clin. Microbiol. **37**:386-390.
12. **Frommell, G. T., R. Rothenberg, S. Wang, and K. McIntosh**. 1979. Chlamydial infection of mothers and their infants. Journal of Pediatrics **95**:28-32.
13. **Gaydos, C. A., T.C. Quinn, D. Willis, A. Weissfeld, E. W. Hook, D. H. Martin, D. V. Ferraro, and J. Schachter.** 2003. Performance of the Aptima Combo 2 Assay for detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* in female urine and endocervical swab specimens. J. Clin. Microbiol. **41**:304-309.
14. **Goessens, W. H. F., J. W. Mouton, W. I. Van Der Meijden, S. Deelen, T. H. Van Rijsoort-Vos, N. L. Toom, H. Verbrugh, and R. P. Verkooyen.** 1997. Comparison of three commercially available amplification assays, AMP CT, LCx, and COBAS AMPLICOR, for detection of *Chlamydia trachomatis* in first-void urine. J. Clin. Microbiol. **35**:2628-2633.
15. **Holmes, K. K., G. W. Counts, and H. N. Beatz**. 1971. Disseminated Gonococcal infection. Ann. of Intern. Med. **74**:979-993.
16. **Holmes, K. K., H. H. Handsfield, S. P. Wang, B. B. Wentworth, M. Turck, J. B. Anderson, and E. R. Alexander**. 1975. Etiology of nongonococcal urethritis. NEJM **292**:1199-1205.
17. **Hook, E. W., III, and H. H. Handsfield.** 1999. Gonococcal infections in the adult. p. 458. *In* K. Holmes *et al.* (eds.) Sexually Transmitted Diseases. McGraw Hill, New York, NY.
18. **Jaschek, G., C. A. Gaydos, L. E. Welsh, and T. C. Quinn.** 1993. Direct detection of *Chlamydia trachomatis* in urine specimens from symptomatic and asymptomatic men by using a rapid polymerase chain reaction assay. J. Clin. Microbiol. **31**:1209-1212.
19. **Krauss, S. J., R. C. Geller, G. H. Perkins, and D. L. Rhoden.** 1976. Interference of *Neisseria gonorrhoeae* growth by other bacterial species. J. Clin. Microbiol. **4**:288-295.
20. **Mahony, J., S. Chong, D. Jang, K. Luinstra, M. Faught, D. Dalby, J. Sellors, and M. Chernesky**. 1998. Urine specimens from pregnant and nonpregnant women inhibitory to amplification of *Chlamydia trachomatis* nucleic acid by PCR, Ligase chain reaction, and transcription-mediated amplification: identification of urinary substances associated with inhibition and removal of inhibitory activity. J. Clin. Microbiol. **36**:3122-3126.

21. **Masi, A. T., and B. I. Eisenstein.** 1981. Disseminated Gonococcal Infections (DGI) and Gonococcal Arthritis (GCA): II Clinical Manifestations, Diagnosis, Complications, Treatment and Prevention. Semin. Arthritis Rheum. **10**:173.
22. **Peterson E. M., V. Darrow, J. Blanding, S. Aarnaes, and L. M. de La Maza.** 1997. Reproducibility problems with the AMPLICOR PCR *Chlamydia trachomatis* test, J. Clin. Microbiol. **35**:957-959.
23. **Schachter, J.** 1985. Chlamydiae (Psittacosis-Lymphogranuloma Venereum-Trachoma group), p. 856-862. *In* E. H. Lennette, et al. (ed.), Manual of Clinical Microbiology, 4th ed. American Society for Microbiology, Washington, D.C.
24. **Schachter, J., and M. Grossman.** 1981. chlamydial infections. Ann. Rev. Med. **32**:45-61.
25. **Schachter, J.** 1978. Medical progress: chlamydial infections (third of three parts). NEJM **298**:540-549.
26. **Schachter, J., E. C. Hill, E. B. King, V. R. Coleman, P. Jones, and K. F. Meyer.** 1975. Chlamydial infection in women with cervical dysplasia. Am. J. Obstet. Gynecol. **123**:753-757.
27. **Stary, A., E. Schuh, M. Kerschbaumer, B. Gotz, and H. Lee**. 1998. Performance of transcription-mediated amplification and Ligase chain reaction assays for detection of chlamydial infection in urogenital samples obtained by invasive and noninvasive methods. J. Clin. Microbiol. **36**:2666-2670.
28. **Toye, B., W. Woods, M. Bobrowska, and K. Ramotar.** 1998. Inhibition of PCR in genital and urine specimens submitted for *Chlamydia trachomatis* testing. J. Clin. Microbiol. **36**:2356-2358.
29. **Verkooyen, R. P., A. Luijendijk, W. M. Huisman, W. H. F. Goessens, J. A. J. W. Kluytmans, J. H. Rijsoort-Vos, and H. A. Verbrugh.** 1996. Detection of PCR inhibitors in cervical specimens by using the AMPLICOR *Chlamydia trachomatis* assay. J. Clin. Microbiol. **34**:3072-3074.
30. **Vincelette, J., J. Schirm, M. Bogard, A. Bourgault, D. Luijt, A. Bianchi, P. C. Van Voorst Vader, A. Butcher, and M. Rosenstraus.** 1999. Multicenter evaluation of the fully automated COBAS AMPLICOR PCR test for detection of *Chlamydia trachomatis* in urogenital specimens. J. Clin. Microbiol. **3**:74-80.
31. **Yuan, Y., Y-X. Zhang, N. G. Watkins, and H. D. Caldwell.** 1989. Nucleotide and deduced amino acid sequences for the four variable domains of the major outer membrane proteins of the 15 *Chlamydia trachomatis* serovars. Infect. Immun. **57**:1040-1049.
32. **U.S. Food and Drug Administration.** 2007. *Guidance for Industry and FDA Staff: Statistical Guidance on Reporting Results from Studies Evaluating Diagnostic Tests*.

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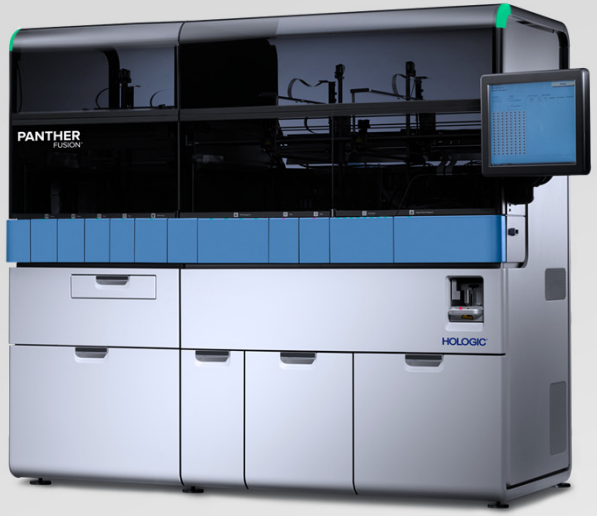
PANTHER FUSION®



Panther® Assays (TMA/RT TMA)

Panther Fusion® Assays (RT PCR)

CURRENT TARGETS

CT/NG
HPV
HPV 16 18/45
Trichomoniasis
Zika virus (EUA)
HSV 1 & 2
HIV-1 Quant
HCV Quant Dx
HBV Quant
Mycoplasma genitalium

Flu A/B/RSV
Paraflu
AdV/hMPV/RV
GBS

Open Access™ functionality to run lab developed tests

CURRENTLY IN DEVELOPMENT*

CMV
Bacterial Vaginosis
Candida/Trichomoniasis

Bordetella
GI Panel

POTENTIAL FUTURE TARGETS

Infectious diseases
Respiratory pathogens
GC resistance and
Mycoplasma genitalium
resistance

Assay consolidation on a fully automated platform allows ***greater # of tests per FTE***

*Under development and not currently for sale in U.S.

The Aptima Zika Virus assay: This test has not been FDA cleared or approved; this test has been authorized by FDA under an EUA for use by authorized laboratories; this test has been authorized only for the detection of RNA from Zika virus and diagnosis of Zika virus infection, not for any other viruses or pathogens; and this test is only authorized for the duration of the declaration that circumstances exist justifying the authorization of the emergency use of in vitro diagnostic tests for detection of Zika virus and/or diagnosis of Zika virus infection under section 564(b)(1) of the Act, 21 U.S.C.§ 360bbb-3(b)(1), unless the authorization is terminated or revoked sooner.

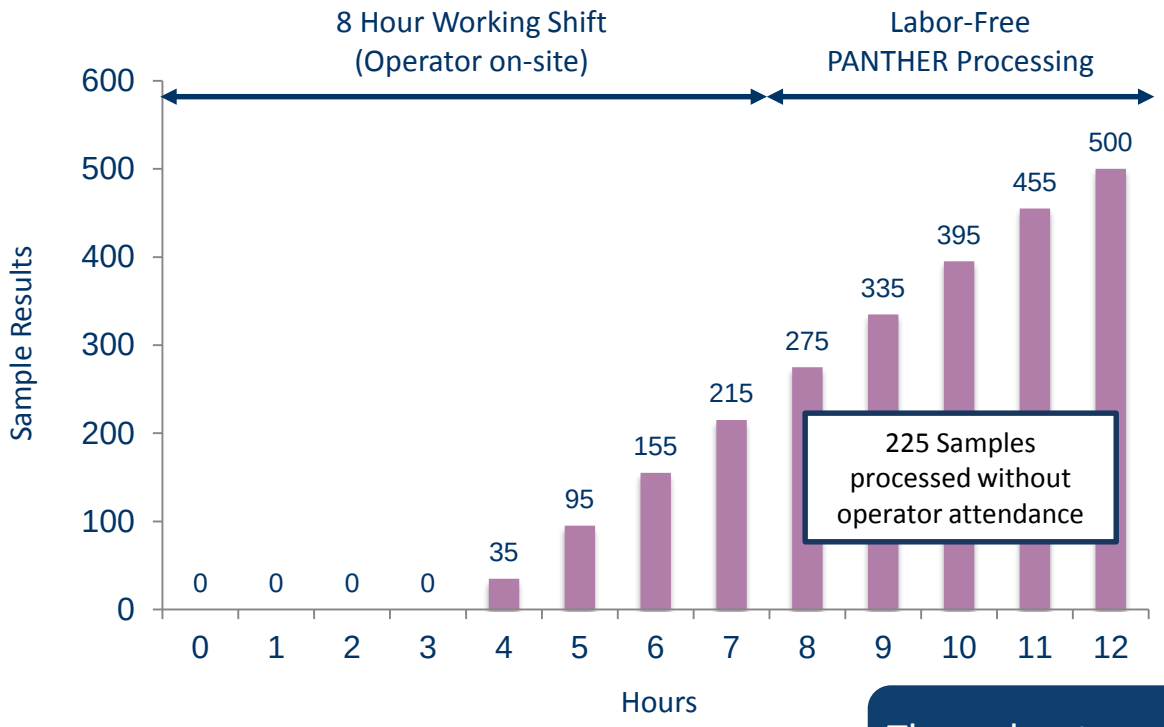
EXHIBIT C

ATTACHMENT B

Increase Productivity

Unattended Processing adds labor-free hours to your day

Maximize Productivity



Throughput

- 275 sample results in 8 hrs
- 500 sample results in 12 hrs

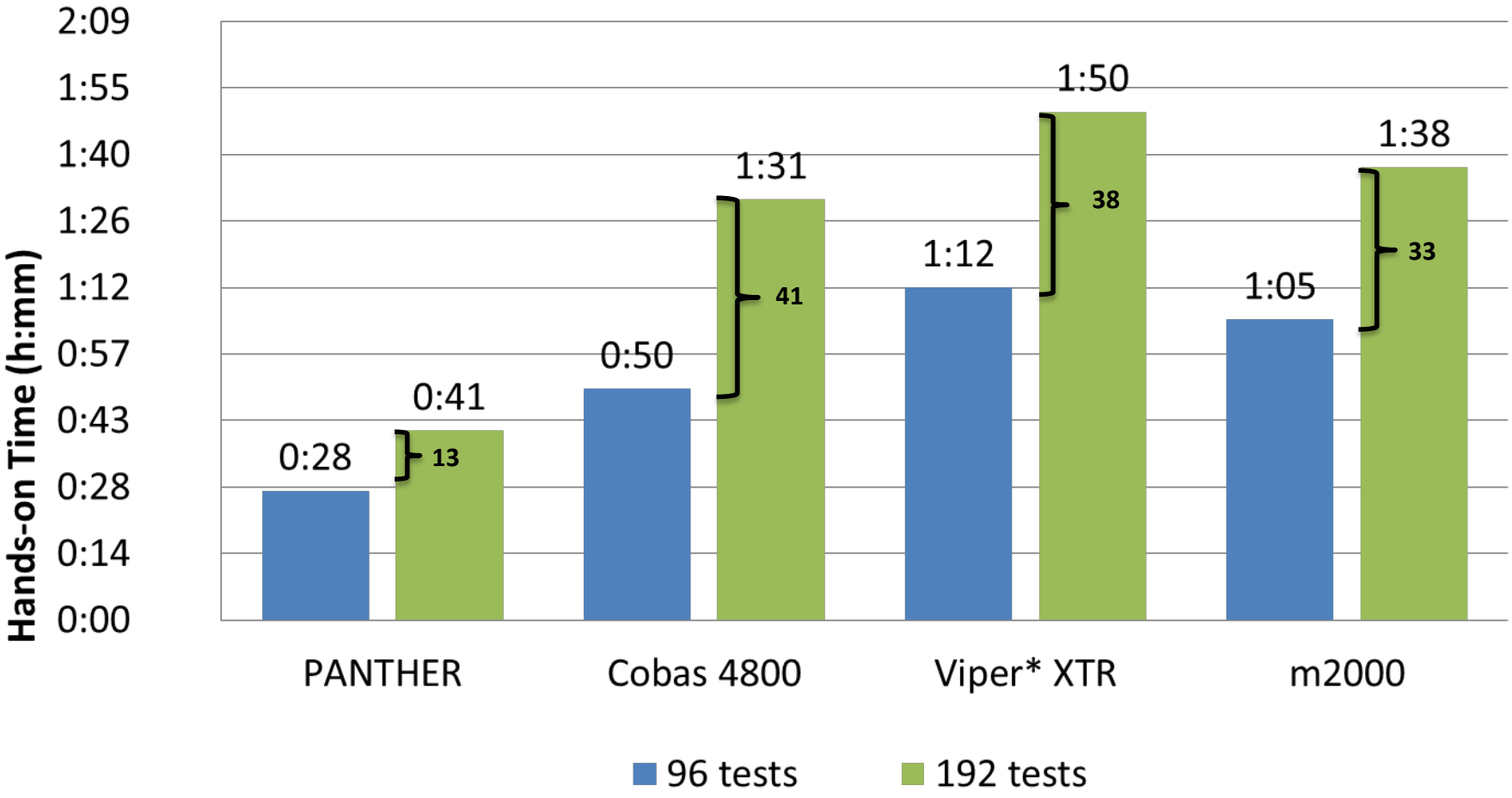
HOLOGIC® | GEN-PROBE

EXHIBIT D

ATTACHMENT B

Hands-on Time

Comparison for 96 and 192 Tests (CT/GC)



HOLOGIC | GEN-PROBE

*High throughput mode

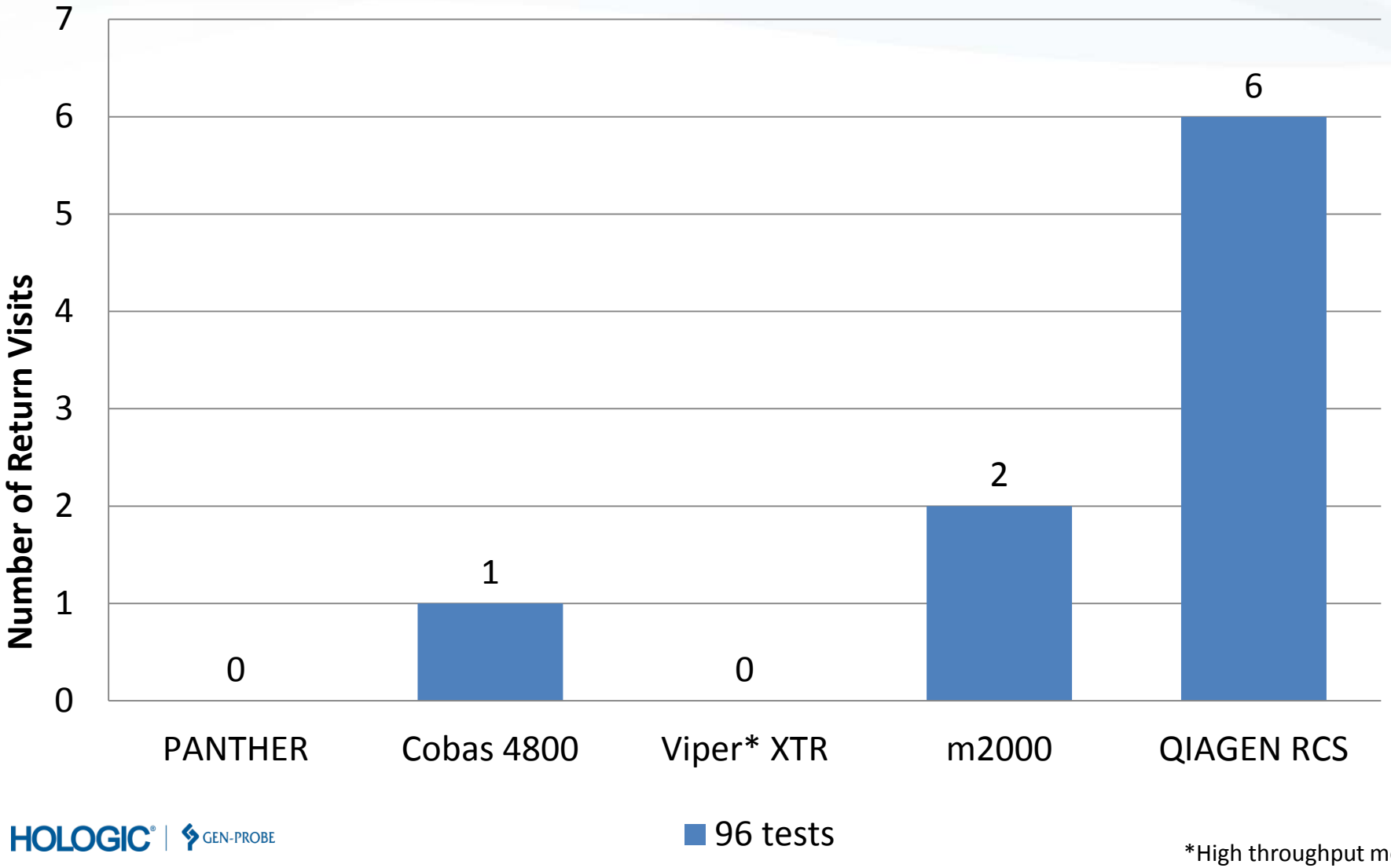
Source: Instrument Comparison: Nexus Global Solutions

EXHIBIT E

ATTACHMENT B

Operator Return Visits

96 Tests (CT/GC or HPV)



HOLOGIC | GEN-PROBE

*High throughput mode

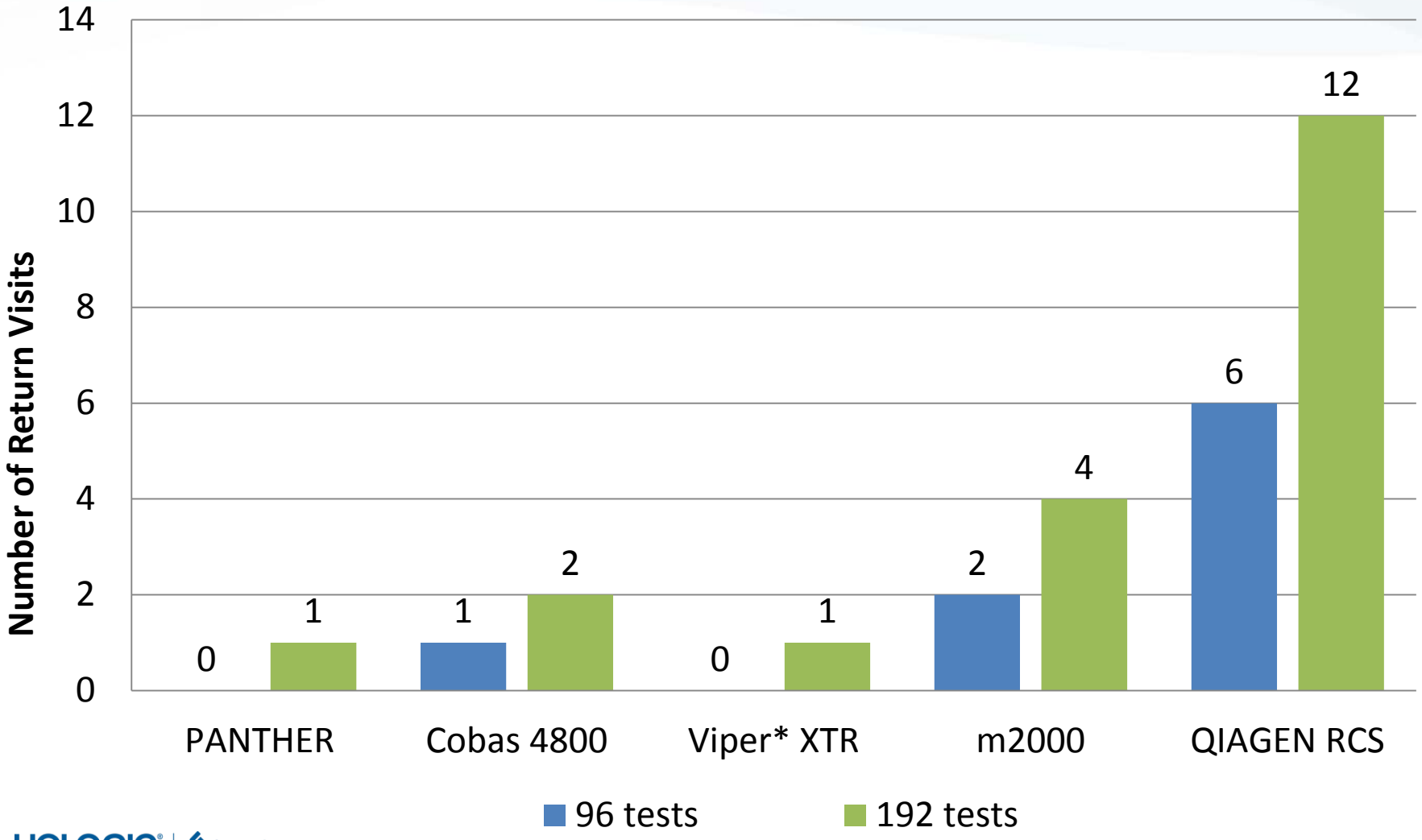
Source: Instrument Comparison: Nexus Global Solutions

EXHIBIT F

ATTACHMENT B

Operator Return Visits

Comparison for 96 and 192 Tests (CT/GC or HPV)



HOLOGIC | GEN-PROBE

*High throughput mode

Source: Instrument Comparison: Nexus Global Solutions

Response For Supplier: Gen-Probe Sales & Service, Inc

Event # : 647-0

Name: Nucleic Acid Application Testing

Reference: Nucleic Acid Application Testing

Description: The Guilford County Department of Health & Human Services - Public Health Division is seeking proposals for nucleic acid amplification tests for the detection of Chlamydia trachomatis (CT), Neisseria gonorrhoeae (GC) and Trichomonas (TV).

Preview Date:

Open Date: 03/01/2019 03:00:00 PM

Close Date: 03/21/2019 03:00:00 PM

Q & A Open Date: 03/01/2019 03:00:00 PM

Q & A Close Date: 03/14/2019 03:00:00 PM

Dispute Close Date:

Responded To: 3 Out of 3 Lines

Total Bid Amount: 13.25 USD

Response Attachments

Attachment

00-Hlx_Event 647_Executive Summary-signed.pdf

01-Hlx_Event 647 NAAT Testing Specification Responses FINAL.pdf

02-Hlx-GPS E-VERIFY AFFIDAVIT_3-20-19.pdf

03-Aptima Combo 2 Assay (Panther System) PI.pdf

04-Aptima Trichomonas Vaginalis Assay (Panther System).pdf

05-Panther - Description of Standard Services.pdf

06-Panther Service Feature Sheet 12.4.17.pdf

07-Exhibits C-E.pdf

08-PB-00552-001_003_Panther Fusion-Respiratory CVA.pdf

09-Panther solutions.pdf

Line Responses

Event # 647-0: Nucleic Acid Application Testing

Line 1: Nucleic Acid Application Test

Description: Cost per test based on annual volume of 20,000 Chlamydia trachomatis (CT) patients per year.

Item: NUCLEIC ACID TEST for CT

Commodity Code: 493 LABORATORY EQUIPMENT, ACCESSORIES, AND SUPPLIES: BIOCHEMIS

Quantity: 1.00 **UOM:** EA

Bid Quantity: 1.0000 **Unit Price:** 3.62000 **Extended Price:** 3.62

No Charge: No **No Bid:** No

Vendor Item: 302923 Aptima Combo 2, 100 Test Kit

Comments: The Aptima Combo 2 detects both CT/GC. The combined price is $7.25 per test. Hologic does not sell individual tests. The Aptima Combo 2, 100 Test Kit (PN 302923) for CT and GC is $725.00 per kit. See Question #43 for a detailed breakdown.

Line 2: Nucleic Acid Application Testing

Description: Cost per test based on annual volume of 20,000 Neisseria Gonorrhoeae (GC) patients per year.

Item: NUCLEIC ACID TEST for GC

Commodity Code: 493 LABORATORY EQUIPMENT, ACCESSORIES, AND SUPPLIES: BIOCHEMIS

Quantity: 1.00 **UOM:** EA

Bid Quantity: 1.0000 **Unit Price:** 3.63000 **Extended Price:** 3.63

No Charge: No **No Bid:** No

Vendor Item: 302923 Aptima Combo 2 - 100 Test Kit

Comments: The Aptima Combo 2 detects both CT/GC. The combined price is $7.25 per test. Hologic does not sell individual tests. The Aptima Combo 2, 100 Test Kit (PN 302923) for CT and GC is $725.00 per kit. See Question #43 for a detailed breakdown.

Line 3: Nucleic Acid Application Testing

Description: Cost per test based on annual volume of 10,000 Trichomonas (TV) patients per year.

Item: NUCLEIC ACID TEST for TV

Commodity Code: 493 LABORATORY EQUIPMENT, ACCESSORIES, AND SUPPLIES: BIOCHEMIS

Quantity: 1.00 **UOM:** EA

Event # 647-0: Nucleic Acid Application Testing

Bid Quantity: 1.0000 **Unit Price:** 6.00000 **Extended Price:** 6.00

No Charge: No **No Bid:** No

Vendor Item: 303536 Aptima Trichomonas Assay, 100T

Comments: The price per test is $6.00. Hologic does not sell individual tests. The Aptima Trichomonas Assay, 100 Test Kit (PN 303536) is $600.00 per kit. See Question #43 for a detailed breakdown.

Line Responses For Event # 647-0 (5,000 combined row record limit for lists)

Company: 1 **Event #:** 647-0 **Event Name:** Nucleic Acid Application Testing

Supplier Group: 1 **Supplier:** 5941 **Supplier Name:** Gen-Probe Sales & Service, Inc

Supplier Contact: 1 **Supplier Contact Name:** RFX Dept

Line Responses

Line #	Item	Description	Vendor Item	Vendor Item Desc	Line Quantity	Response Quantity	UOM	Unit Price	No Charge	Alternate Responses Only	No Bid	No Bid Reason	Response Exists	Questions Exist	Is Awarded	Alternate Responses
*1	NUCLEIC ACID TEST	for CT	302923	Aptima Combo 2, 100 Test Kit	1.00	1.0000	EA	3.62000	No	No	No		Yes	No	Yes	
*2	NUCLEIC ACID TEST	for GC	302923	Aptima Combo 2 - 100 Test Kit	1.00	1.0000	EA	3.63000	No	No	No		Yes	No	Yes	
*3	NUCLEIC ACID TEST	for TV	303536	Aptima Trichomonas Assay, 100T	1.00	1.0000	EA	6.00000	No	No	No		Yes	No	Yes	

ATTACHMENT B

EVENT # 647

ITEM DESCRIPTION	VENDOR ITEM	VENDOR ITEM DESCRIPTION	QUANTITY	UOM	UNIT PRICE	TOTAL
NUCLEIC ACID TEST FOR CT	302923	Aptima Combo 2, 100 Test Kit	20,000	EA	3.62	72,400.00
NUCLEIC ACID TEST FOR GC	302923	Aptima Combo 2 - 100 Test Kit	20,000	EA	3.63	72,600.00
NUCLEIC ACID TEST FOR TV	303536	Aptima Trichomonas Assay, 100T	10,000	EA	6.00	60,000.00
						205,000.00

ATTACHMENT B

ATTACHMENT C

HIPAA BUSINESS ASSOCIATE ADDENDUM

This Business Associate Addendum, effective 1st day of May, 2019, by and between **GUILFORD COUNTY, on behalf of the GUILFORD COUNTY DEPARTMENT OF HEALTH AND HUMAN SERVICES, DIVISION OF PUBLIC HEALTH,** a body politic and corporate of the State of North Carolina, hereinafter referred to as the **"COVERED ENTITY,"** and **GEN-PROBE SALES & SERVICE, INC., a subsidiary of Hologic, Inc."** hereinafter referred to as the **"BUSINESS ASSOCIATE,"** and also collectively referred to as the "Parties."

Definitions

Terms used, but not otherwise defined, in this Addendum shall have the same meaning as those terms in 45 CFR 160.103 and 164.501.

A. Individual. "Individual" shall have the same meaning as the term "individual" in 45 CFR 164.501 and shall include a person who qualifies as a personal representative in accordance with 45 CFR 164.502(g).

B. Privacy Rule. "Privacy Rule" shall mean the standards for privacy of individual identifiable health information at 45 CFR part 160 and part 164, subparts A and E.

C. Protected Health Information. "Protected Health Information" shall have the same meaning, as the term "protected health information" is 45 CFR 164.501, limited to the information created or received by the Business Associate from or on behalf of the Covered Entity.

D. Required by Law. "Required by Law" shall have the same meaning as the term "required by law" in 45 CFR 164.501.

E. Secretary. "Secretary" shall mean the Secretary of the Department of Health and Human Services or his/her designee.

F. Data Aggregation. "Data Aggregation" shall mean, with respect to Protected Health Information created or received by the Business Associate in its capacity as the business associate of the Covered Entity, the combining of such Protected Health Information by the Business Associate with the Protected Health Information received by the Business Associate in its capacity as a business associate of another covered entity, to permit data analyses that relate to the health care operations of the respective covered entities.

G. Designated Record Set. "Designated Record Set" shall mean a group of records maintained by or for the Covered Entity that is (i) the medical records and billing records about individuals maintained by or for the Covered Entity, (ii) the enrollment, payment, claims adjudication, and case or medical management record systems maintained by or for a health plan; or (iii) used, in whole or in part, by or for the Covered Entity to make decisions about individuals. As used herein the term "Record" means any item, collection,

or grouping of information that includes Protected Health Information and is maintained, collected, used, or disseminated by or for the Covered Entity.

H. Electronic Media. "Electronic Media" shall mean the mode of electronic transmissions. It includes the Internet, extranet (using Internet technology to link a business with information only accessible to collaborating parties), leased lines, dial-up lines, private networks and those transmissions that are physically moved from one location to another using magnetic tape, disk or compact disk media.

Recitals

A. The U.S. Department of Health and Human Services has issued regulations on "Privacy Standards for Individually Identifiable Health Information," implementing the Health Insurance Portability and Accountability Act of 1996 (the "Privacy Standards").

B. Covered Entity is a service provider. The U.S. Department of Health and Human Services has issued final regulations, pursuant to the Health Insurance Portability and Accountability Act of 1996 ("HIPAA"), governing the privacy of Individually Identifiable Health Information obtained, created or maintained by certain entities, including health care providers.

C. Business Associate either 1) performs certain functions for, or on behalf of the Covered Entity involving the disclosure of Protected Covered Entity Health Information ("PHI") by the Covered Entity to Business Associate, or the creation or receipt of PHI by Business Associate on behalf of the Covered Entity; or 2) provides legal, actuarial, accounting, consulting, data aggregation, management, accreditation, administrative or financial services for the Covered Entity involving the disclosure of Protected Health Information ("PHI") by the Covered Entity or another business associate of the Covered Entity.

D. The Parties of this Addendum agree to enter into this agreement to protect PHI, and to amend any agreements between them, whether oral or written, with the execution of this Addendum.

In consideration of the mutual promises and agreements below and in order to comply with all legal requirements for the protection of this information, the Parties agree as follows:

General Provisions

A. Effect. This Addendum supplements, modifies and amends any and all agreements, whether oral or written, between the Parties involving the disclosure of PHI by the Covered Entity to Business Associate, or the creation or receipt of PHI by Business Associate on behalf of the Covered Entity. The terms and provisions of the Addendum shall supersede any other conflicting or inconsistent terms and provisions in any agreements between the Parties, including all exhibits or other attachments thereto and all documents incorporated therein by reference. Without limitation of the foregoing, any limitation or exclusion of damages provisions shall not be applicable to this Addendum.

B. Amendment. Business Associate and the Covered Entity agree to amend this Addendum to the extent necessary to allow either Party to comply with the Privacy Standards, the

Standards for Electronic Transactions (45 CFR Parts 160 and 162) and the Security Standards (45 CFR Part 142) (collectively, the "Standards") promulgated or to be promulgated by the Secretary or other regulations or statutes. Business Associate agrees that it will fully comply with all such Standards and that it will agree to amend this Addendum to incorporate any material required by the Standards.

Obligations of Business Associate

A. **Use and Disclosure of Protected Health Information.** Business Associate may use and disclose Protected Health Information only as required to satisfy its obligations under the Agreement(s), as permitted herein, or required by law, but shall not otherwise use or disclose any Protected Health Information. Business Associate shall not, and shall ensure that its directors, officers, employees, contractors and agents do not, use or disclose Protected Health Information received from the Covered Entity in any manner that would constitute a violation of the Privacy Standards if so used or disclosed by the Covered Entity, except that Business Associate may use or disclose Protected Health Information (i) for Business Associate's proper management and administrative services, (ii) to carry out the legal responsibilities of Business Associate or (iii) to provide data aggregation services relating to the health care operations of the Covered Entity if required under the Agreement(s). Business Associate hereby acknowledges that, as between Business Associate and the Covered Entity, all Protected Health Information shall be and remain the sole property of the Covered Entity, including any and all forms thereof developed by Business Associate in the course of its fulfillment of its obligations pursuant to this Addendum. Business Associate further represents that, to the extent Business Associate requests that the Covered Entity disclose Protected Health Information to Business Associate, such a request is only for the minimum necessary Protected Health Information for the accomplishment of Business Associate's purpose.

B. **Safeguards Against Misuse of Information.** Business Associate agrees that it will use all appropriate safeguards to prevent the use or disclosure of Protected Health Information other than pursuant to the terms and conditions of this Addendum.

C. **Reporting of Disclosures of Protected Health Information.** Business Associate shall, within thirty (30) days of becoming aware of any use or disclosure of Protected Health Information in violation of this Addendum by Business Associate, its officers, directors, employees, contractors or agents or by a third party to which Business Associate disclosed Protected Health Information, report any such disclosure to the Covered Entity.

D. **Agreements by Third Parties.** Business Associate shall obtain and maintain an agreement with each agent or subcontractor that has or will have access to Protected Health Information, which is received from, or created or received by Business Associate on behalf of the Covered Entity, pursuant to which agreement such agent or subcontractor agrees to be bound by the same restrictions, terms and conditions that apply to Business Associate pursuant to this Addendum with respect to such Protected Health Information.

E. **Access to Information.** Within five (5) business days of a request by the Covered Entity for access to Protected Health Information about an individual contained in a Designated Record Set, Business Associate shall make available to the Covered Entity such Protected Health Information for so long as such information is maintained in the Designated Record

Set. In the event any individual requests access to Protected Health Information directly from Business Associate, Business Associate shall within five (5) business days forward such request to the Covered Entity. Any denials of access to the Protected Health Information requested shall be the responsibility of the Covered Entity. **[Not necessary if Business Associate does not have Protected Health Information in a Designated Record Set.**

F. Availability of Protected Health Information for Amendment. Within ten (10) business days of receipt of a request from the Covered Entity for the amendment of an individual's Protected Health Information or a record regarding an individual contained in a Designated Record Set (for so long as the Protected Health Information is maintained in the Designated Record Set), Business Associate shall provide such information to the Covered Entity for amendment and incorporate any such amendments in the Protected Health Information as required by 45 C.F.R. §164.526. **[Not necessary if Business Associate does not have Protected Health Information in a Designated Record Set.]**

G. Accounting of Disclosures. Within ten (10) business days of notice by the Covered Entity to Business Associate that it has received a request for an accounting of disclosures of Protected Health Information, other than related to the treatment of the patient, the processing of payments related to such treatment, or the operation of a Covered Entity or its Business Associate and not relating to disclosures made earlier than six (6) years prior to the date on which the accounting was requested, Business Associate shall make available to the Covered Entity such information as is in Business Associate's possession and is required for the Covered Entity to make the accounting required by 45 C.F.R. §164.528. At a minimum, Business Associate shall provide the Covered Entity with the following information: (i) the date of the disclosure, (ii) the name of the entity or person who received the Protected Health Information, and if known, the address of such entity or person, (iii) a brief description of the Protected Health Information disclosed, and (iv) a brief statement of the purpose of such disclosure which includes an explanation of the basis for such disclosure. In the event the request for an accounting is delivered directly to Business Associate, Business Associate shall within two (2) business days forward such request to the Covered Entity. Business Associate hereby agrees to implement an appropriate record keeping process to enable it to comply with the requirements of this Section.

H. Availability of Books and Records. Business Associate hereby agrees to make its internal practices, books and records relating to the use and disclosure of Protected Health Information received from, or created or received by Business Associate on behalf of, the Covered Entity available to the Secretary for purposes of determining the Covered Entity's and Business Associate's compliance with the Privacy Standards.

I. Indemnification. Business Associate hereby agrees to indemnify and hold the Covered Entity harmless from and against any and all liability and costs, including attorneys' fees, created by a breach of this Addendum by Business Associate, its agents or subcontractors, without regard to any limitation or exclusion of damages provision otherwise set forth in the Agreement(s) only to the extent permitted by NC Tort Claims Act without waiving sovereign immunity.

J. Insurance. Business Associate shall obtain and maintain during the term of this Addendum liability insurance covering claims based on a violation of the Standards or any applicable state law or regulation concerning the privacy of patient information and claims based on its obligations pursuant to this Addendum in an amount not less than $1,000,000 per claim. Such insurance shall be in the form of occurrence-based coverage and shall name the Covered Entity as an additional named insured. A copy of such policy or a certificate evidencing the policy shall be provided to the Covered Entity upon written request.

K. Notice of Request for Data. Business Associate agrees to notify the Covered Entity within five (5) business days of Business Associate's receipt of any request or subpoena for Protected Health Information. To the extent that the Covered Entity decides to assume responsibility for challenging the validity of such request, Business Associate agrees to cooperate fully with the Covered Entity in such challenge.

L. Injunction. Business Associate hereby agrees that the Covered Entity will suffer irreparable damage upon Business Associate's breach of this Addendum and that such damages shall be difficult to quantify. Business Associate hereby agrees that the Covered Entity may file an action for an injunction to enforce the terms of this Addendum against Business Associate, in addition to any other remedy the Covered Entity may have.

Term and Termination

A. Term. This Addendum shall become effective on the Effective Date and, unless otherwise terminated as provided herein, shall have a term that shall run concurrently with that of the last expiration date or termination of the Agreement(s).

B. Termination Upon Breach of Provisions Applicable to Protected Health Information. Any other provision of the Agreement(s) notwithstanding, this Addendum and the Agreement(s) may be terminated by the Covered Entity upon five (5) business days written notice to Business Associate in the event that the Business Associate breaches any provision contained in this Addendum and such breach is not cured within such five (5) business day period; provided, however, that in the event that termination of this Addendum and the Agreement(s) is not feasible, in the Covered Entity's sole discretion, Business Associate hereby acknowledges that the Covered Entity shall have the right to report the breach to the Secretary, notwithstanding any other provision of this Addendum or any Agreement(s) to the contrary.

C. Return or Destruction of Protected Health Information upon Termination. Upon termination of this Addendum, Business Associate shall either return or destroy all Protected Health Information received from the Covered Entity or created or received by Business Associate on behalf of the Covered Entity and which Business Associate still maintains in any form. Business Associate shall not retain any copies of such Protected Health Information. Notwithstanding the foregoing, to the extent that the Covered Entity agrees that it is not feasible to return or destroy such Protected Health Information, the terms and provisions of this Addendum shall survive such termination and such Protected Health Information shall be used or disclosed solely for such purpose or purposes which prevented the return or destruction of such Protected Health Information.

D. The Covered Entity's Right of Cure. At the expense of Business Associate, the Covered Entity shall have the right to cure any breach of Business Associate's obligations under this Addendum. The Covered Entity shall give Business Associate notice of its election to cure any such breach and Business Associate shall cooperate fully in the efforts by the Covered Entity to cure Business Associate's breach. All requests for payment for such services of the Covered Entity shall be paid within thirty (30) days except that Business Associate shall have five (5) business days as noted in Section B to cure such breach. Covered Entity may cure breach upon expiration of the 5th business day.

E. Transition Assistance. Following the termination of this Addendum and the Agreement(s) for any reason, Business Associate agrees to provide transition services for the benefit of the Covered Entity, including the continued provision of its services required under the Agreement(s) until notified by the Covered Entity that the alternative provider of services is able to take over the provision of such services and
the transfer of the Protected Health Information and other data held by the Business Associate related to its services under the Agreement(s).

(The remainder of this page is intentionally left blank.)
This Addendum in incorporated as Attachment C.

ATTACHMENT D

STATE OF NORTH CAROLINA

AFFIDAVIT

COUNTY OF GUILFORD

****

I, Peter P. Dunne (the individual attesting below), being duly authorized by and on behalf of Gen-Probe Sales & Service, Inc.* (the entity bidding on project hereinafter "Employer") after first being duly sworn hereby swears or affirms as follows:

1. Employer understands that E-Verify is the federal E-Verify program operated by the United States Department of Homeland Security and other federal agencies, or any successor or equivalent program used to verify the work authorization of newly hired employees pursuant to federal law in accordance with NCGS §64-25(5).

2. Employer understands that Employers Must Use E-Verify. Each employer, after hiring an employee to work in the United States, shall verify the work authorization of the employee through E-Verify in accordance with NCGS§64-26(a).

3. Employer is a person, business entity, or other organization that transacts business in this State and that employs 25 or more employees in this State. Mark "Yes" or "No":

   a. YES X; or, (59 Hologic company wide and 14 Diagnostics division)

   b. NO _____

4. Employer's subcontractors comply with E-Verify, and if Employer is the winning bidder on this project Employer will ensure compliance with E-Verify by any subcontractors subsequently hired by Employer. N/A, no subcontractors are used for this contract.

This 20th day of March, 2019.

_____

Signature of Affiant

Print or Type Name: Peter P. Dunne

State of North Carolina County of Guilford

Signed and sworn to (or affirmed) before me, this the _____

day of _____, 2013.

My Commission Expires:

_____. _____

Notary Public

See attached

(Affix Official/Notarial Seal)

*Gen-Probe Sales & Service, Inc. is a subsidiary of Hologic, Inc.

CALIFORNIA ALL-PURPOSE ACKNOWLEDGMENT **CIVIL CODE § 1189**

A notary public or other officer completing this certificate verifies only the identity of the individual who signed the document to which this certificate is attached, and not the truthfulness, accuracy, or validity of that document.

State of California)
County of San Diego)

On 3/20/19 (Date) before me, Claudia Deutsch, Notary Public (Here Insert Name and Title of the Officer),

personally appeared Peter P. Dunne (Name(s) of Signer(s)),

who proved to me on the basis of satisfactory evidence to be the person(s) whose name(s) is/are subscribed to the within instrument and acknowledged to me that he/she/they executed the same in his/her/their authorized capacity(ies), and that by his/her/their signature(s) on the instrument the person(s), or the entity upon behalf of which the person(s) acted, executed the instrument.

I certify under PENALTY OF PERJURY under the laws of the State of California that the foregoing paragraph is true and correct.

WITNESS my hand and official seal.

CLAUDIA DEUTSCH
Commission # 2141411
Notary Public - California
San Diego County
My Comm. Expires Feb 2, 2020

Signature C Claudia Deutsch
Signature of Notary Public

Place Notary Seal Above

OPTIONAL

Though this section is optional, completing this information can deter alteration of the document or fraudulent reattachment of this form to an unintended document.

Description of Attached Document

Title or Type of Document: E-Verify Affidavit Document Date: 3/20/19
Number of Pages: 1 Signer(s) Other Than Named Above: none

Capacity(ies) Claimed by Signer(s)

Signer's Name: Peter P. Dunne	Signer's Name:
☐ Corporate Officer — Title(s):	☐ Corporate Officer — Title(s):
☐ Partner — ☐ Limited ☐ General	☐ Partner — ☐ Limited ☐ General
☐ Individual ☐ Attorney in Fact	☐ Individual ☐ Attorney in Fact
☐ Trustee ☐ Guardian or Conservator	☐ Trustee ☐ Guardian or Conservator
☑ Other: VP, Finance & Accounting	☐ Other:
Signer Is Representing: Gen-Probe Sales & Service, Inc., a subsidiary of Hologic, Inc.	Signer Is Representing: