5215436 A

Sierra Diagnostics 510(k): Urine Collection, Preservation and Transport System

IMAGE

CDRH Document Imaging System

User: *SHS: Shelley, Shiela H. (OSM/DID)*
Note: *FOI REDACTED*

Folder: *K013819*
Pages: *Entire Folder (2 documents, 185 pages)*

Pages Printed: *116*

Date Requested: *Wed Mar 10 15:54:37 2002*
Date Printed: *Wed Mar 10 15:54:49 2004*
Printer: *OK4IMG01*

DEPARTMENT OF HEALTH & HUMAN SERVICES

Food and Drug Administration
2098 Gaither Road
Rockville MD 20850

FEB 2 8 2002

Sierra Diagnostics, L.L.C.
c/o Donald R. Stone, Esq.
Kirkpatrick and Lockhart, LLP
1800 Massachusetts Avenue, NW
Suite 200
Washington, DC 20036-1221

Re: k013819
Trade/Device Name: Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System
Regulation Number: 21 CFR 866.2900
Regulation Name: Microbiological Specimen Collection and Transport System
Regulatory Class: Class I
Product Code: JTW
Dated: February 11, 2002
Received: February 12, 2002

Dear Mr. Stone:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 898. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Part 801); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820); and if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR 1000-1050.

This letter will allow you to begin marketing your device as described in your 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific advice for your device on our labeling regulation (21 CFR Part 801 and additionally 809.10 for in vitro diagnostic devices), please contact the Office of Compliance at (301) 594-4588. Additionally, for questions on the promotion and advertising of your device, please contact the Office of Compliance at (301) 594-4639. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21CFR 807.97). Other general information on your responsibilities under the Act may be obtained from the Division of Small Manufacturers International and Consumer Assistance at its toll-free number (800) 638-2041 or (301) 443-6597 or at its internet address "http://www.fda.gov/cdrh/dsma/dsmamain.html".

Sincerely yours,

Steven Gutman

Steven I. Gutman, M.D., M.B.A.
Director
Division of Clinical Laboratory Devices
Office of Device Evaluation
Center for Devices and
Radiological Health

Enclosure

510(k) Number:

Device Name: Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System

Indications for Use:

The Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System is indicated for use in the collection, preservation, and transportation of urine specimens at temperatures not exceeding 60°C for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays.

PLEASE DO NOT WRITE BELOW THIS LINE. CONTINUE ON ANOTHER PAGE IN NEEDED

Concurrence of CDRH, Office of Device Evaluation (ODE)

Prescription Use ✓
(Per 21 CFR 801.109)

(Division Sign-Off)
Division of Clinical Laboratory Devices

OR

Over-The Counter Use _____

510(k) Number K013819

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service
Food and Drug Administration

Memorandum

From: Reviewer(s) - Name(s) Patricia M. Beverly February 20, 2002

Subject: 510(k) Number **K013819 / S1**

To: The Record - It is my recommendation that the subject 510(k) Notification:

☐ Refused to accept.

☐ Requires additional information (other than refuse to accept).

✓ Is substantially equivalent to marketed devices. per 21 CFR 866.2900
Specimen Collection and Transport System

☐ NOT substantially equivalent to marketed devices.

De Novo Classification Candidate? ☐ YES ☐ NO

☐ Other (e.g., exempt by regulation, not a device, duplicate, etc.)

Is this device subject to Postmarket Surveillance?	☐ YES	✓ NO
Is this device subject to the Tracking Regulation?	☐ YES	✓ NO
Was clinical data necessary to support the review of this 510(k)?	✓ YES	☐ NO
Is this a prescription device?	✓ YES	☐ NO
Was this 510(k) reviewed by a Third Party?	☐ YES	✓ NO
Special 510(k)?	☐ YES	✓ NO
Abbreviated 510(k)? Please fill out form on H Drive 510k/boilers	☐ YES	✓ NO

This 510(k) contains:

Truthful and Accurate Statement ☐ Requested ✓ Enclosed
(required for originals received 3-14-95 and after)

✓ A 510(k) summary OR ☐ A 510(k) statement

☐ The required certification and summary for class III devices

☐ The indication for use form (required for originals received 1-1-96 and after)

Material of Biological Origin ☐ YES ✓ NO

The submitter requests under 21 CFR 807.95 (doesn't apply for SEs):

☐ No Confidentiality ☐ Confidentiality for 90 days ☐ Continued Confidentiality exceeding 90 days

Predicate Product Code with class: JTW / System, Transport, Aerobic / CLASS I

Additional Product Code(s) with panel (optional):

Review: Freddie M. Poole (Branch Chief) BMCB (Branch Code) 2/26/02 (Date)

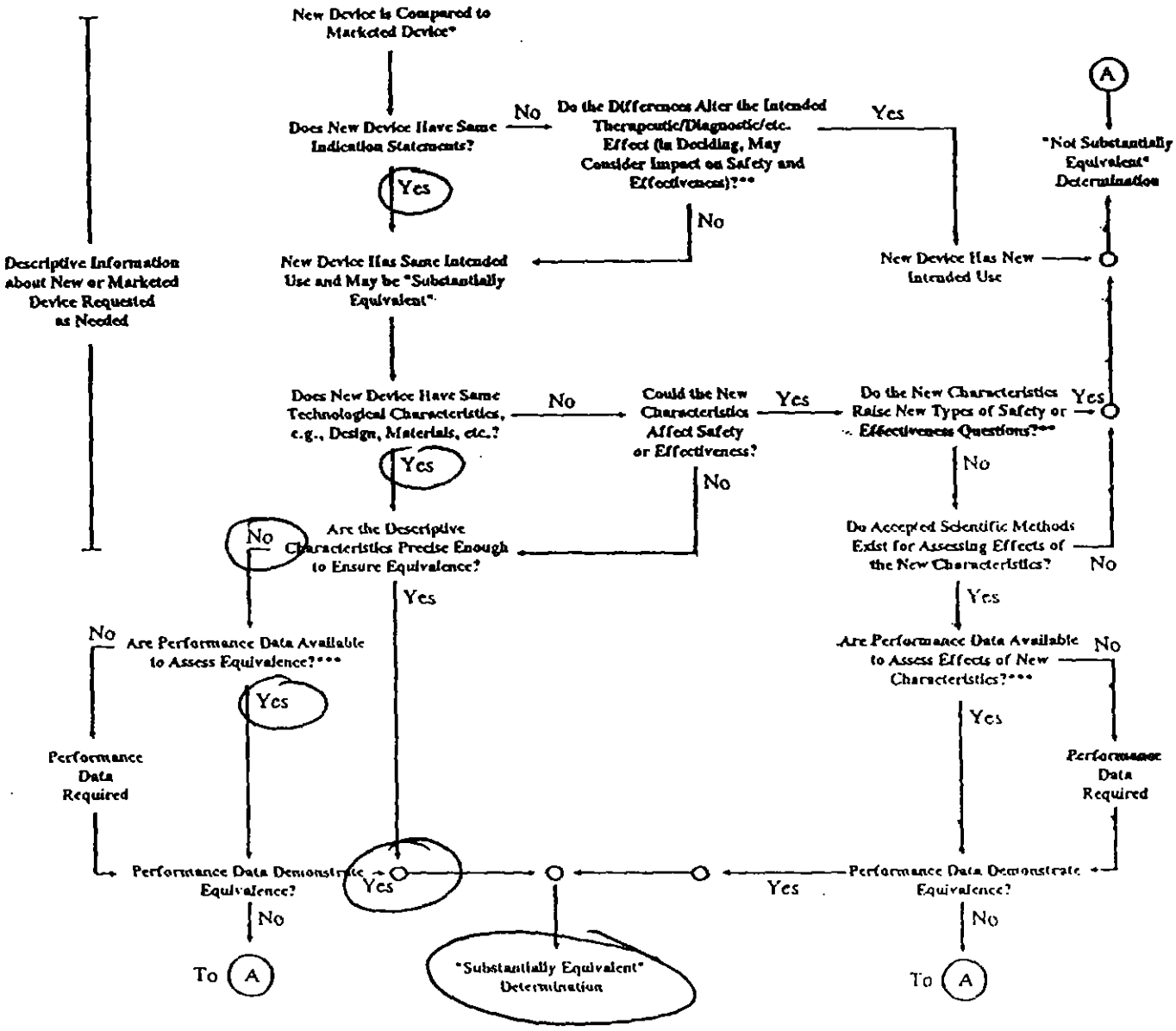
Final Review: S. Gutman (Division Director) 2/27/02 (Date)

Revised:8/17//99

4

510(k) "SUBSTANTIAL EQUIVALENCE" DECISION-MAKING PROCESS (DETAILED)

K013819



* 510(k) submissions compare new devices to marketed devices. FDA requests additional information if the relationship between marketed and "predicate" (pre-Amendments or reclassified post-Amendments) devices is unclear.

** This decision is normally based on descriptive information alone, but limited testing information is sometimes required.

*** Data may be in the 510(k), other 510(k)s, the Center's classification files, or the literature.

THE 510(K) DOCUMENTATION FORMS ARE AVAILABLE ON THE LAN UNDER 510(K) BOILERPLATES TITLED "DOCUMENTATION" AND MUST BE FILLED OUT WITH

EVERY FINAL DECISION (SE, NSE, NOT A DEVICE, ETC.).

"SUBSTANTIAL EQUIVALENCE" (SE) DECISION MAKING DOCUMENTATION

K013819

Reviewer: Patricia M. Beverly
Division/Branch: DCLD-Microbiology
Device Name: Urine Collection, Preservation and Transport System, Sierra Diagnostics L.L.C.
Product To Which Compared (510(K) Number If Known): Abbott LCx ® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assay, accessory item, Standard Collection Cup, K935833/K934622

	YES	NO	
1. Is Product A Device	✓		If **NO** = Stop
2. Is Device Subject To 510(k)?	✓		If **NO** = Stop
3. Same Indication Statement?	✓		If **YES** = Go To 5
4. Do Differences Alter The Effect Or Raise New Issues of Safety Or Effectiveness?			If **YES** = Stop **NE**
5. Same Technological Characteristics?	✓		If **YES** = Go To 7
6. Could The New Characteristics Affect Safety Or Effectiveness?			If **YES** = Go To 8
7. Descriptive Characteristics Precise Enough?		✓	If **NO** = Go To 10 If **YES** = Stop **SE**
8. New Types Of Safety Or Effectiveness Questions?			If **YES** = Stop **NE**
9. Accepted Scientific Methods Exist?			If **NO** = Stop **NE**
10. Performance Data Available?	✓		If **NO** = Request Data
11. Data Demonstrate Equivalence?	✓		Final Decision: **SE**

Note: In addition to completing the form on the LAN, "yes" responses to questions 4, 6, 8, and 11, and every "no" response requires an explanation.

1. Intended Use:
The Urine Collection System is intended for use for the collection, preservation, and transportation of urine specimens at temperatures not exceeding 60°C for testing with the Abbott LCx® Neisseria gonorrhoeae and Chlamydia trachomatis assays. The system contains a nucleic acid preservative which inactivates a broad range of heat catalyzed enzyme systems that are responsible for the degradation of the nucleic acid targets. This slows the nucleic acid degradation process and therefore allows urine specimens to be stored at room temperature for 6 days prior to testing with the LCx® assay. This system's preservative prevents the log phase growth of bacteria that may cause a shift in the urine pH. The device also [redacted] optimal conditions for the LCx® assay.

2. Device Description: Provide a statement of how the device is either similar to and/or different from other marketed devices, plus data (if necessary) to support the statement. Does the device design use software? Provide a summary about the devices design, materials, physical properties and toxicology profile if important.

The Urine Collection System is similar to the Standard Collection Cup that is already 510(k) cleared. The Urine Collection System is similar in that they both collect, and transport urine specimens while preserving the nucleic acid targets. Both systems preserve the nucleic acids in the specimen but they differ in that the Urine Collection System contains a chemical preservative and the standard urine cup is refrigerated. Storage for the Urine collection system is ≤ 60° C and the predicate device storage is 2-8° C.

The Urine Collection system consists of a sterile urine cup that contains a preservative and [redacted] The beads in the cup serve as an indicator that the preservative is present in the specimen. The urine collection cup is marked with "maximum" and "minimum" fill lines. The patient fills the cup with first stream urine to a level between the two lines that are marked on the cup. This ensures that the preservative-to-urine ratio in the specimen will be within the effective dilution range for the preservative. After the patient closes and labels the cup, the specimen is transported to the laboratory without refrigeration.

EXPLANATIONS TO "YES" AND "NO" ANSWERS TO QUESTIONS ON PAGE 1 AS NEEDED

7. Explain how descriptive characteristics are not precise enough:
The descriptive characteristics are not precise enough. Comparative studies for determination of the preservation nucleic acids needs to be demonstrated because the manufacturer's assay does not recommend use of preservatives.

11. Explain how the performance data demonstrates that the device is or is not substantially equivalent:

In this multi-center comparison study, of 582 patients, 80 subjects were positive for *Chlamydia* and *Neisseria gonorrhea,* and 502 subjects were negative. All testing was performed with the LCx® assay. The study was performed at five sites. At four of the study sites, a preservative-to-urine ratio of 1:10 (2ml preservative to 20 ml of urine) were

7

used, while a ratio of 1:5 (4ml of preservative to 20 ml of urine) was used at one study site. Specimens collected in the predicate device were refrigerated within one hour of collection. Testing on these specimens began within 24 hours of collection. Specimens collected in the Urine Collection System were transported and stored at ambient temperature (21° C) for 6 days prior to testing. The firm reported 100% correlation to the standard urine collection cup.

Recovery studies were performed with fresh urine spiked with chalamydial and gonococcal DNA targets. The chlamydia targets were from the nine individual serovars that occur in cervical infections. The gonococcal targets represented 18 individual PIA serovars and 25 PIB serovars that comprise pathogenic Neisseria gonorrhea. Fresh urine samples were frozen at –70° C and the preserved specimens were stored at [redacted] for 6 days. Control tubes without DNA targets were prepared in an identical manner. The firm reported 100% correlation between the refrigerated and preserved specimens.

An analytical study was performed using two concentrations (1:10 to 1:15) of the preservative of Sierra. To confirm the sensitivity of the assay on the LCx®, ten gonococcal serovars were serially diluted and fresh urine was spiked with a gonococcal culture of less than [redacted] Non-preserved specimens were tested within 1 day (stored at –70° C) and preserved specimens were tested at 6 days (stored at 25° C). Tests results demonstrated that the device could effectively preserve nucleic acid targets to the LCx® assay level of detection.

The FDA Guidance Document "Review Criteria for Assessment of *In-Vitro* Diagnostic Devices For Direct Detection Of Chlamydia in Clinical Specimens", "Review Criteria For Nucleic Acid Amplification-Based *In-Vitro* Diagnostic Devices For Direct Detection Of Infectious Microorganisms" and FDA recommendations made were followed. Included in the package insert are the performance characteristics and the correlation data.

All the review criteria were satisfied, and the package insert and labeling format conforms to 21 CFR § 809.10. The studies demonstrated that the Urine Collection Transport System to be substantially equivalent to other legally marketed devices performing the same testing such as the predicate device.

8

DEPARTMENT OF HEALTH AND HUMAN SERVICES
Food and Drug Administration

ADDENDUM TO MEMORANDUM

To: FILE, K013819

From: Reviewer, Bacteriology Branch, Division of Clinical Laboratory Devices, Office of Device Evaluation

Date: February 15, 2002

Re: **Sierra Diagnositics, L.L.C. – Urine Collection, Preservation and Transport System**

ADDENDUM

Donald Stone, Kirkpatrick & Lockhart, LLP, counsel for Sierra, submitted all the additional information requested by telephone on February 12, 2002. Responses to the requested concerns have been addressed and placed in the original document K013819.

The review of the submission is now complete. I recommend a substantial equivalent determination.

Patricia M. Beverly 2/15/02
Patricia M. Beverly
Reviewer

Freddie M. Poole
Freddie M. Poole, Chief

DEPARTMENT OF HEALTH AND HUMAN SERVICES Public Health Service

Food and Drug Administration
Center for Devices and
Radiological Health
Office of Device Evaluation
Document Mail Center (HFZ-401)
9200 Corporate Blvd.
Rockville, Maryland 20850

February 12, 2002

SIERRA DIAGNOSTIC, L.L.C.
C/O KIRKPATRICK & LOCKHART LLP
1800 MASSACHUSETTS AVE., NW
SUITE 200
WASHINGTON, DC 20036
ATTN: DONALD R. STONE

510(k) Number: K013819
Product: URINE COLLECTION, PRESERVATION AND TRANSPORT SYSTEM

The additional information you have submitted has been received.

We will notify you when the processing of this submission has been completed or if any additional information is required. Please remember that all correspondence concerning your submission MUST be sent to the Document Mail Center (HFZ-401) at the above letterhead address. Correspondence sent to any address other than the one above will not be considered as part of your official premarket notification submission. Because of equipment and personnel limitations we cannot accept telefaxed material as part of your official premarket notification submission, unless specifically requested of you by an FDA official.

The Safe Medical Devices Act of 1990, signed on November 28, states that you may not place this device into commercial distribution until you receive a letter from FDA allowing you to do so. As in the past, we intend to complete our review as quickly as possible. Generally we do so 90 days. However, the complexity of a submission or a requirement for additional information may occasionally cause the review to extend beyond 90 days. Thus, if you have not received a written decision or been contacted within 90 days of our receipt date you may want to check with FDA to determine the status of your submission.

If you have procedural or policy questions, please contact the Division of Small Manufacturers International and Consumer Assistance (DSMICA) at (301) 443-6597 or at their toll-free number (800) 638-2041, or contact me at (301) 594-1190.

Sincerely yours,

Marjorie Shulman
Supervisory Consumer Safety Officer
Premarket Notification Section
Office of Device Evaluation
Center for Devices and
Radiological Health

10

K 013819/S1

Kirkpatrick & Lockhart LLP

1800 Massachusetts Avenue, NW
Suite 200
Washington, DC 20036-1221
202.778.9000
www.kl.com

February 11, 2002

Donald R. Stone
202.778.9067
Fax: 202.778.9100
dstone@kl.com

VIA FEDERAL EXPRESS

Food and Drug Administration
Document Mail Center (HFZ-401)
Center for Devices and Radiological Health
Office of Device Evaluation
9200 Corporate Boulevard
Rockville, MD 20850

RE: 510(k) No. K013819 (Sierra Diagnostics, LLC) – Response to Request for Additional Information

Dear Sir or Madam:

On behalf of Sierra Diagnostics, LLC ("Sierra"), and in response to requests received from Ms. Patricia Beverly of the Office of Device Evaluation ("ODE"), we have enclosed the following information for inclusion in the file for 510(k) No. K013819:

1. The line (raw) data concerning the clinical study at 5 sites in tabular form (Exhibit 1).

2. The line (raw) data concerning the analytical studies in tabular form (Exhibit 2).

3. The raw absorbance data from the analytical studies (Exhibit 3).

4. A new Truthful and Accurate Statement on a separate page from all other information (Exhibit 4).

5. The appropriate Financial Disclosure form concerning the clinical investigators and their relation to and remuneration by the company (Exhibit 5).

6. Revised device labeling (Exhibit 6).

In accordance with 21 C.F.R. § 807.90, the enclosed information has been provided in duplicate with the original and two copies of this cover letter. Please note that **Exhibits 1, 2, and 3** of this submission contain trade secret and confidential commercial information and, therefore, are **exempt from public disclosure** under the Freedom of Information Act (5 U.S.C. § 552(b)(4)) and the applicable Food and Drug Administration ("FDA") regulations (21 C.F.R. § 20.61).

We believe that the enclosed information completely responds to the requests noted in the FDA's January 29, 2002 memorandum. Therefore, we request that 510(k) No. 013819 be immediately removed from "hold" status.

Thank you for your prompt attention to this matter.

Sincerely,

Donald R. Stone

Donald R. Stone

Enclosure(s)
cc: Michael H. Hinckle, Esq., w/ encl.
Mr. Robert Koch – Sierra Diagnostics, LLC, w/ encl.
Mr. Tony Baker – Sierra Diagnostics, LLC, w/ encl.

EXHIBIT 1

This page represents 29 whole-page redactions.

EXHIBIT 4

PREMARKET NOTIFICATION

TRUTHFUL AND ACCURATE STATEMENT

[As Required by 21 CFR 807.87(k)]

I certify that, in my capacity as Chief Technical Officer for Sierra Diagnostics, L.L.C., I believe to the best of my knowledge, that all data and information submitted in premarket notification number K013819 are truthful and accurate and that no material fact has been omitted.

Tony K. Baker
Chief Technical Officer
Sierra Diagnostics, L.L.C.

Date: 28 January 02

EXHIBIT 5

This page represents 1 whole-page redactions.

EXHIBIT 6

[PROPOSED PACKAGE INSERT]

SIERRA DIAGNOSTICS, L.L.C.
URINE COLLECTION, PRESERVATION AND TRANSPORT SYSTEM
For use with the Abbott LCx® Assays for *Neisseria gonorrhoeae* and *Chlamydia trachomatis*

Indications for Use:
The Sierra Diagnostics L.L.C. Urine Collection, Preservation, and Transport System is indicated for use in the collection, preservation, and transportation of urine specimens at temperatures not exceeding [ ] for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays.

Warning: Abbott recommends that a preservative-free, sterile urine collection cup be used with the LCx® assays. This system contains preservatives and indicator beads. Users should perform their own validation studies for the use of this system with the LCx® assays.

Caution: Abbott recommends that urine specimens stored at room temperature should not be used for testing. The labeling of the LCx® assays state specimens should be: (1) shipped to the testing laboratory either at 2-8°C or frozen so as to arrive within 24 hours of shipment; (2) stored by the testing site at either 2-8°C or -20°C and processed within 4 days of collection if held at 2-8°C, or within 60 days if held at -20°C; and (3) if frozen, used immediately upon thawing. As evidenced by the data and instructions provided below, compliance with these precautions is not required when using this collection system.

Precautions:
Specimens should not be collected from patients who have urinated one hour prior to the collection.

Instruction for Use:
The patient should be instructed to fill the collection cup with first pass urine (the first part of the stream) to a level between the two lines printed on the outside of the cup. After the urine is collected, verify that the cup is securely closed and labeled with the patient's ID number and date of collection. The red indicator beads serve as a reminder that the preservative is present in the specimen. Transport the specimen to the laboratory for testing. There is no need to refrigerate or freeze the specimen. The specimen will be suitable for LCx® testing for 144 hours (6 days) provided it is stored at temperatures not exceeding 60°C (140°F).

Storage:
The Urine Collection, Preservation and Transport System should be stored at room temperature until the expiration date. Use prior to expiration date printed on outside of package.

Performance and Correlation Data

1. "Spiked" Urine Recovery Studies (*N. gonorrhea*)

* S/CO = LCx® Sample Rate/Cutoff Value

120 hrs. @ +30°C

	Positive	Negative	Total
Frozen (Mean S/CO*)	10 (2.460)	3 (0.03)	13
Preserved (Mean S/CO*)	10 (2.393)	3 (0.03)	13
Difference	0 (0.067)	0 (0)	---

100% Correlation

144 hrs. @ +30°C

	Positive	Negative	Total
Frozen (Mean S/CO*)	10 (2.594)	3 (0.11)	13
Preserved (Mean S/CO*)	10 (2.741)	3 (0.11)	13
Difference	0 (0.147)	0 (0)	—

100% Correlation

120 hrs. @ +60°C

	Positive	Negative	Total
Frozen (Mean S/CO*)	10 (2.465)	3 (0.03)	13
Preserved (Mean S/CO*)	10 (2.489)	3 (0.13)	13
Difference	0 (0.024)	0 (0.10)	---

100% Correlation

144 hrs. @ +60°C

	Positive	Negative	Total
Frozen (Mean S/CO*)	10 (2.441)	3 (0.02)	13
Preserved (Mean S/CO*)	10 (2.540)	3 (0.02)	13
Difference	0 (0.099)	0 (0)	---

100% Correlation

2. "Spiked" Urine Recovery Studies (*C. trachomatis*)

* S/CO = LCx® Sample Rate/Cutoff Value

96 hrs. @ +30°C

	Positive	Negative	Total
Frozen (Mean S/CO*)	10 (2.441)	3 (0.02)	13
Preserved (Mean S/CO*)	10 (2.504)	3 (0.02)	13
Difference	0 (0.063)	0 (0)	—

100% Correlation

120 hrs. @ +30°C

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	—

100% Correlation

144 hrs. @ +30°C

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

100% Correlation

144 hrs. @ +60°C

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

100% Correlation

3. Clinical Correlation Study Data (5 Clinical Sites)

Analysis of Positive LCx® Results (C = *C. trachomatis;* N = *N. gonorrhea)*

	Site 1[1]		Site 2[1]		Site 3[1]		Site 4[1]		Site 5[2]	
	C	N	C	N	C	N	C	N	C	N
Unpreserved*	17	4	13	2	14	5	9	1	15	0
Preserved**	17	4	13	2	14	5	9	1	15	0
Correlation (%)	100	100	100	100	100	100	100	100	100	100

* 2-8°C – 24 hrs.
** Ambient Temp. – 144 hrs.
[1] Preservative to urine ratio = 1:10
[2] Preservative to urine ratio = 1:5
(No specimens were both C and N positive)

[PROPOSED PACKAGE LABEL]

Cup Label (4.0 ml preservative)

URINE COLLECTION, PRESERVATION AND TRANSPORT SYSTEM **4.0 ML LOT #** _____ **EXP. DATE** _____ **STORE AT ROOM TEMP.** **FOR IN VITRO DIAGNOSTIC USE ONLY** **WARNING: Contains sodium thiocynate and EDTA as preservatives**	Patient _____ ID _____ Date _____ Time _____ Physician _____ Phone _____ **SIERRA DIAGNOSTICS, L.L.C.** **SONORA, CA. 95370**

Sterile Label for Cup (attaches between cup and cap)

STERILE

DEPARTMENT OF HEALTH AND HUMAN SERVICES Public Health Service

Food and Drug Administration
Center for Devices and
Radiological Health
Office of Device Evaluation
Document Mail Center (HFZ-401)
9200 Corporate Blvd.
Rockville, Maryland 20850

January 31, 2002

SIERRA DIAGNOSTIC, L.L.C.
C/O KIRKPATRICK & LOCKHART LLP
1800 MASSACHUSETTS AVE., NW
SUITE 200
WASHINGTON, DC 20036
ATTN: DONALD R. STONE

510(k) Number: K013819
Product: URINE COLLECTION, PRESERVATION AND TRANSPORT SYSTEM

We are holding your above-referenced Premarket Notification (510(k)) for 30 days pending receipt of the additional information that was requested by the Office of Device Evaluation. Please remember that all correspondence concerning your submission MUST cite your 510(k) number and be sent in duplicate to the Document Mail Center (HFZ-401) at the above letterhead address. Correspondence sent to any address other than the one above will not be considered as part of your official premarket notification submission. Because of equipment and personnel limitations, we cannot accept telefax material as part of your official premarket notification submission unless specifically requested of you by an FDA official.

The deficiencies identified represent the issues that we believe need to be resolved before our review of your 510(k) submission can be successfully completed. In developing the deficiencies, we carefully considered the statutory criteria as defined in Section 513(i) of the Federal Food, Drug, and Cosmetic Act for determining substantial equivalence of your device. We also considered the burden that may be incurred in your attempt to respond to the deficiencies. We believe that we have considered the least burdensome approach to resolving these issues. If, however, you believe that information is being requested that is not relevant to the regulatory decision or that there is a less burdensome way to resolve the issues, you should follow the procedures outlined in the "A Suggested Approach to Resolving Least Burdensome Issues" document. It is available on our Center web page at: http://www.fda.gov/cdrh/modact/leastburdensome.html

If after 30 days the requested information, or a request for an extension of time, is not received, we will discontinue review of your submission and proceed to delete your file from our review system. Pursuant to 21 CFR 20.29, a copy of your 510(k) submission will remain in the Office of Device Evaluation. If you then wish to resubmit this 510(k) notification, a new number will be assigned and your submission will be considered a new premarket notification submission.

Please remember that the Safe Medical Devices Act of 1990 states that you may not place this device into commercial distribution until you receive a decision letter from FDA allowing you to do so.

If you have procedural or policy questions, please contact the Division of Small Manufacturers International and Consumer Assistance (DSMICA) at (301) 443-6597 or at their toll-free number (800) 638-2041, or contact me at (301) 594-1190.

Sincerely yours,

Marjorie Shulman
Supervisor Consumer Safety Officer
Premarket Notification Section
Office of Device Evaluation
Center for Devices and
Radiological Health

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service
Food and Drug Administration

Memorandum

From: Reviewer(s) - Name(s) Patricia M. Beverly January 29, 2002

Subject: 510(k) Number **K013819**

To: The Record - It is my recommendation that the subject 510(k) Notification:

☐ Refused to accept.

✓ Requires additional information (other than refuse to accept). **Telephone Hold**

☐ Is substantially equivalent to marketed devices.

☐ NOT substantially equivalent to marketed devices.

De Novo Classification Candidate? ☐ YES ☐ NO

☐ Other (e.g., exempt by regulation, not a device, duplicate, etc.)

Is this device subject to Postmarket Surveillance?	☐ YES	☐ NO
Is this device subject to the Tracking Regulation?	☐ YES	☐ NO
Was clinical data necessary to support the review of this 510(k)?	☐ YES	☐ NO
Is this a prescription device?	☐ YES	☐ NO
Was this 510(k) reviewed by a Third Party?	☐ YES	☐ NO
Special 510(k)?	☐ YES	☐ NO
Abbreviated 510(k)? Please fill out form on H Drive 510k/boilers	☐ YES	☐ NO

This 510(k) contains:

Truthful and Accurate Statement ☐ Requested ☐ Enclosed
(required for originals received 3-14-95 and after)

☐ A 510(k) summary OR ☐ A 510(k) statement

☐ The required certification and summary for class III devices

☐ The indication for use form (required for originals received 1-1-96 and after)

Material of Biological Origin ☐ YES ☐ NO

The submitter requests under 21 CFR 807.95 (doesn't apply for SEs):

☐ No Confidentiality ☐ Confidentiality for 90 days ☐ Continued Confidentiality exceeding 90 days

Predicate Product Code with class: Additional Product Code(s) with panel (optional):

Review: Judith M. Pool BACB 1/29/02
(Branch Chief) (Branch Code) (Date)

Final Review: _____
(Division Director) (Date)

d:8/17//99

54

****
*** TX REPORT ***
****

TRANSMISSION OK

TX/RX NO	1813
CONNECTION TEL	912027789100
SUBADDRESS	
CONNECTION ID	KIRKPATRICK & LO
ST. TIME	01/29 14:21
USAGE T	01'08
PGS.	3
RESULT	OK

FDA'S OFFICE OF DEVICE EVALUATION
DIVISION OF CLINICAL LABORATORY DEVICES
MICROBIOLOGY BRANCH
FAXING

2098 Gaither Road, HFZ-440
Rockville, Maryland 20850

Phone No.: (301) 594-2096

Fax No.: (301) 594-5940 or (301) 594-5941

DATE: January 29, 2002 Fax No.: 202-778-9100

TO: Donald R. Stone
Michael H. Hinckle
Kirkpatrick & Lockhart, LLP

FROM: Patricia Beverly

No. of pages (including cover sheet): 3

55

COMMENTS: Sierra Diagnostics, L.L.C- Urine Collection, Preservation and Transport System

FDA'S OFFICE OF DEVICE EVALUATION
DIVISION OF CLINICAL LABORATORY DEVICES
MICROBIOLOGY BRANCH
FAXING

2098 Gaither Road, HFZ-440
Rockville, Maryland 20850

Phone No.: (301) 594-2096

Fax No.: (301) 594-5940 or (301) 594-5941

DATE: January 29, 2002 Fax No.: 202-778-9100

TO: Donald R. Stone
Michael H. Hinckle
Kirkpatrick & Lockhart, LLP

FROM: Patricia Beverly

No. of pages (including cover sheet): 3

COMMENTS: Sierra Diagnostics, L.L.C- Urine Collection, Preservation and Transport System

NOTE: Please advise if transmission is illegible.

Thanks

56

This page represents 3 whole-page redactions.

The submission will be complete when the above concerns are addressed. Mr. Hinkle was informed that this submission would be placed on hold as of today's date.

Patricia M. Beverly
Review Scientist Date: 1/29/02

Kirkpatrick & Lockhart LLP

1800 Massachusetts Avenue, NW
Second Floor
Washington, DC 20036-1800
202.778.9000
Fax: 202.778.9100
Fax: 202.778.9200

FAX

Date • January 28, 2002

No. of Pages, including coversheet • 32

Transmit To •

Name	Company	Phone	Fax
Pat Beverly	FDA	301-594-2096	301-594-5940

From • Donald R. Stone — **Phone •** 202-778-9067

Secretary • Anne Picciano — **Phone •** 202-778-9407

Client/Matter Name	Client/Matter Number	Attorney Number
Sierra Diagnostics	0306475-0100	3051

COMMENTS: Please see attached.

When you are sending to us, please be sure to include a cover sheet with your transmittal and a telephone number where you can be contacted in case of equipment malfunction.

Transmitted by: **Time:**

61

This page represents 2 whole-page redactions.

EXHIBIT 1

This page represents 9 whole-page redactions.

Kirkpatrick & Lockhart LLP

1800 Massachusetts Avenue, NW
Second Floor
Washington, DC 20036-1800
202.778.9000
Fax: 202.778.9100
Fax: 202.778.9200

FAX

Date • January 28, 2002

No. of Pages, including coversheet • 32

Transmit To •

Name	Company	Phone	Fax
Pat Beverly	FDA	301-594-2096	301-594-5940

From • Donald R. Stone — **Phone •** 202-778-9067

Secretary • Anne Picciano — **Phone •** 202-778-9407

Client/Matter Name	Client/Matter Number	Attorney Number
Sierra Diagnostics	0306475-0100	3051

COMMENTS: Please see attached.

When you are sending to us, please be sure to include a cover sheet with your transmittal and a telephone number where you can be contacted in case of equipment malfunction.

Transmitted by: **Time:**

Kirkpatrick & Lockhart LLP

1800 Massachusetts Avenue, NW
Suite 200
Washington, DC 20036-1221
202.778.9000
www.kl.com

January 28, 2002

Donald R. Stone
202.778.9067
Fax: 202.778.9100
dstone@kl.com

Food and Drug Administration
Document Mail Center (HFZ-401)
Office of Device Evaluation
Center for Devices and Radiological Health
9200 Corporate Blvd.
Rockville, MD 20850

Attn: Ms. Patricia Beverly

Re: 510(k) Premarket Notification No. K013819: Sierra Diagnostics, L.L.C. – Urine Collection, Preservation and Transport System

Dear Ms. Beverly:

In a telephone conversation on Friday, January 25, 2002, you requested the following items regarding the above referenced 510(k). Those items were:

1. The line (raw) data concerning the clinical study at 5 sites in tabular form.
2. The line (raw) data concerning the analytical studies in tabular form.
3. A new Truthful and Accuracy Certification on a separate page from all other information.
4. The appropriate Financial Disclosure forms concerning the clinical investigators and their relation to and remuneration by the company.

Accordingly, enclosed as Exhibits 1 and 2, respectively, please find the requested raw data for the clinical studies and *in vitro* studies conducted by Sierra Diagnostics, L.L.C. ("Sierra") concerning the company's Urine Collection, Preservation, and Transport System (the "Urine Collection System"). Please note that, with respect to the clinical studies, data were collected only for specimens that tested "positive" at time zero in the unpreserved container. The Urine Collection System is designed to preserve nucleic acids for detection with the LCx® assays. When initial LCx® testing of an "unpreserved" specimen indicated that no target nucleic acids were present (i.e., a "negative" result), there was no scientific rationale for testing the corresponding

Kirkpatrick & Lockhart LLP

"preserved" specimens because it was presumed that there were no nucleic acids to preserve. This rationale was further buttressed by Sierra's "spiked" urine testing, which confirmed the lack of false positives in specimens preserved with the Urine Collection System. Based on this reasoning, a 100% correlation of negative test results for the LCx® assays and the Urine Collection System was inferred in the tables in the original 510(k).

Additionaly, the requested "Truthful and Accurate Statement" and "Certification: Financial Interests and Arrangements of Clinical Investigators" are enclosed as Exhibits 3 and 4.

Sincerely,

Donald R. Stone

Donald R. Stone

Enclosures

This page represents 27 whole-page redactions.

PREMARKET NOTIFICATION

TRUTHFUL AND ACCURATE STATEMENT

[As Required by 21 CFR 807.87(k)]

I certify that, in my capacity as Chief Technical Officer for Sierra Diagnostics, L.L.C., I believe to the best of my knowledge, that all data and information submitted in premarket notification number K013819 are truthful and accurate and that no material fact has been omitted.

Tony K. Baker
Chief Technical Officer
Sierra Diagnostics, L.L.C.

Date: 28 January 02

EXHIBIT 4

This page represents 5 whole-page redactions.

SCREENING CHECKLIST
FOR ALL PREMARKET NOTIFICATION [510(k)] SUBMISSIONS

510(k) Number: K013819

The cover letter clearly identifies the type of 510(k) submission as (Check the appropriate box):

☐ Special 510(k) - Do Sections 1 and 2

☐ Abbreviated 510(k) - Do Sections 1, 3 and 4

☒ Traditional 510(k) or no identification provided - Do Sections 1 and 4

Section 1: Required Elements for All Types of 510(k) submissions:

	Present	Inadequate or Missing
Cover letter, containing the elements listed on page 3-2 of the Premarket Notification [510)] Manual.	✓	
Table of Contents.	✓	
Truthful and Accurate Statement.	✓	✓ p46
Device's Trade Name, Device's Classification Name and Establishment Registration Number.	✓	
Device Classification Regulation Number and Regulatory Status (Class I, Class II, Class III or Unclassified).	✓	
Proposed Labeling including the material listed on page 3-4 of the Premarket Notification [510)] Manual.	✓	
Statement of Indications for Use that is on a separate page in the premarket submission.	✓	
Substantial Equivalence Comparison, including comparisons of the new device with the predicate in areas that are listed on page 3-4 of the Premarket Notification [510)] Manual.	✓	
510(k) Summary or 510(k) Statement.	✓	
Description of the device (or modification of the device) including diagrams, engineering drawings, photographs or service manuals.	✓	
Identification of legally marketed predicate device. *	✓	
Compliance with performance standards. * [See Section 514 of the Act and 21 CFR 807.87 (d).]		
Class III Certification and Summary. **		
Financial Certification or Disclosure Statement for 510(k) notifications with a clinical study. * [See 21 CFR 807.87 (i)]		✓
510(k) Kit Certification ***		

* - May not be applicable for Special 510(k)s.
** - Required for Class III devices, only.
*** - See pages 3-12 and 3-13 in the Premarket Notification [510)] Manual and the Convenience Kits Interim Regulatory Guidance.

1/1

Section 2: Required Elements for a SPECIAL 510(k) submission:

	Present	Inadequate or Missing
Name and 510(k) number of the sponsor's own, unmodified predicate device.		
A description of the modified device and a comparison to the sponsor's predicate device.		
A statement that the intended use(s) and indications of the modified device, as described in its labeling, are the same as the intended uses and indications for the sponsor's unmodified predicate device.		
A statement that the modification has not altered the fundamental technology of the sponsor's predicate device.		
A Design Control Activities Summary that includes the following elements (a-e):		
a. Identification of Risk Analysis method(s) used to assess the impact of the modification on the device and its components, and the results of the analysis.		
b. Based on the Risk Analysis, an identification of the required verification and validation activities, including the methods or tests used and the acceptance criteria to be applied.		
c. A Declaration of Conformity with design controls that includes the following statements:		
A statement that, as required by the risk analysis, all verification and validation activities were performed by the designated individual(s) and the results of the activities demonstrated that the predetermined acceptance criteria were met. This statement is signed by the individual responsible for those particular activities.		
A statement that the manufacturing facility is in conformance with the design control procedure requirements as specified in 21 CFR 820.30 and the records are available for review. This statement is signed by the individual responsible for those particular activities.		

Section 3: Required Elements for an ABBREVIATED 510(k)* submission:

	Present	Inadequate or Missing
For a submission, which relies on a guidance document and/or special control(s), a summary report that describes how the guidance and/or special control(s) was used to address the risks associated with the particular device type. (If a manufacturer elects to use an alternate approach to address a particular risk, sufficient detail should be provided to justify that approach.)		
For a submission, which relies on a recognized standard, a declaration of conformity [For a listing of the required elements of a declaration of conformity, SEE **Required Elements for a Declaration of Conformity to a Recognized Standard,** which is posted with the 510(k) boilers on the **H drive.**]		
For a submission, which relies on a recognized standard without a declaration of conformity, a statement that the manufacturer intends to conform to a recognized standard and that supporting data will be available before marketing the device.		
For a submission, which relies on a non-recognized standard that has been historically accepted by FDA, a statement that the manufacturer intends to conform to a recognized standard and that supporting data will be available before marketing the device.		
For a submission, which relies on a non-recognized standard that has not been historically accepted by FDA, a statement that the manufacturer intends to conform to a recognized standard and that supporting data will be available before marketing the device and any additional information requested by the reviewer in order to determine substantial equivalence.		
Any additional information, which is not covered by the guidance document, special control, recognized standard and/or non-recognized standard, in order to determine substantial equivalence.		

* - When completing the review of an abbreviated 510(k), please fill out an Abbreviated Standards Data Form (located on the H drive) and list all the guidance documents, special controls, recognized standards and/or non-recognized standards, which were noted by the sponsor.

Section 4: Additional Requirements for ABBREVIATED and TRADITIONAL 510(k) submissions (If Applicable):

	Present	Inadequate or Missing
a) Biocompatibility data for all patient-contacting materials, OR certification of identical material/formulation:		
b) Sterilization and expiration dating information:		
i) sterilization process		
ii) validation method of sterilization process		
iii) SAL		
iv) packaging		
v) specify pyrogen free		
vi) ETO residues		
vii) radiation dose		
c) Software Documentation:		

Items with checks in the "Present but Deficient" column require additional information from the sponsor. Items with checks in the " Missing" column must be submitted before substantive review of the document.

Passed Screening ✓ Yes ____ No
Reviewer: Lisa King
Concurrence by Review Branch: Patricia M. Beverly
Date: 1/25/07

The deficiencies identified above represent the issues that we believe need to be resolved before our review of your 510(k) submission can be successfully completed. In developing the deficiencies, we carefully considered the statutory criteria as defined in Section 513(i) of the Federal Food, Drug, and Cosmetic Act for determining substantial equivalence of your device. We also considered the burden that may be incurred in your attempt to respond to the deficiencies. We believe that we have considered the least burdensome approach to resolving these issues. If, however, you believe that information is being requested that is not relevant to the regulatory decision or that there is a less burdensome way to resolve the issues, you should follow the procedures outlined in the "A Suggested Approach to Resolving Least Burdensome Issues" document. It is available on our Center web page at: http://www.fda.gov/cdrh/modact/leastburdensome.html

Internal Administrative Form

	YES	NO
1. Did the firm request expedited review?		✓
2. Did we grant expedited review?		✓
3. Have you verified that the Document is labeled Class III for GMP purposes? N/A		
4. If, not, has POS been notified?		
5. Is the product a device?	✓	
6. Is the device exempt from 510(k) by regulation or policy?		✓
7. Is the device subject to review by CDRH?	✓	
8. Are you aware that this device has been the subject of a previous NSE decision?		✓
9. If yes, does this new 510(k) address the NSE issue(s), (e.g., performance data)? N/A		
10. Are you aware of the submitter being the subject of an integrity investigation?		✓
11. If, yes, consult the ODE Integrity Officer.	[redacted]	
12. Has the ODE Integrity Officer given permission to proceed with the review? (Blue Book Memo #I91-2 and Federal Register 90N0332, September 10, 1991. N/A		

DEPARTMENT OF HEALTH AND HUMAN SERVICES Public Health Service

Food and Drug Administration
Center for Devices and
Radiological Health
Office of Device Evaluation
Document Mail Center (HFZ-401)
9200 Corporate Blvd.
Rockville, Maryland 20850

November 16, 2001

SIERRA DIAGNOSTIC, L.L.C.
C/O KIRKPATRICK & LOCKHART LLP
1800 MASSACHUSETTS AVE., NW
SUITE 200
WASHINGTON, DC 20036
ATTN: DONALD R. STONE

510(k) Number: K013819
Received: 16-NOV-2001
Product: URINE COLLECTION, PRESERVATION AND TRANSPORT SYSTEM

The Center for Devices and Radiological Health (CDRH), Office of Device Evaluation (ODE), has received the Premarket Notification you submitted in accordance with Section 510(k) of the Federal Food, Drug, and Cosmetic Act(Act) for the above referenced product. We have assigned your submission a unique 510(k) number that is cited above. Please refer prominently to this 510(k) number in any future correspondence that relates to this submission. We will notify you when the processing of your premarket notification has been completed or if any additional information is required. YOU MAY NOT PLACE THIS DEVICE INTO COMMERCIAL DISTRIBUTION UNTIL YOU RECEIVE A LETTER FROM FDA ALLOWING YOU TO DO SO.

As a reminder, we would like to mention that FDA requires all 510(k) submitters to provide an indications for use statement on a separate page. If you have not included this indications for use statement in addition to your 510(k) summary (807.92), or a 510(k) statement (807.93), and your Truthful and Accurate statement, please do so as soon as possible. If the above mentioned requirements have been submitted, please do not submit them again. There may be other regulations or requirements affecting your device such as Postmarket Surveillance (Section 522(a)(1) of the Act) and the Device Tracking regulation (21 CFR Part 821). Please contact the Division of Small Manufacturer International and Consumer Assistance (DSMICA) at the telephone or web site below for more information.

The Clinical Laboratory Improvement Amendments of 1988 (CLIA) requires the categorization of commercially marketed test systems by level of complexity. If your device is a test system that requires categorization you will be notified of your complexity as an enclosure with any clearance letter.

Please remember that all correspondence concerning your submission MUST besent to the Document Mail Center (DMC)(HFZ-401) at the above letterhead address. Correspondence sent to any address other than the DMC will not be considered as part of your official premarket notification submission. Because of equipment and personnel limitations, we cannot accept telefaxed material as part of your official premarket notification submission, unless specifically requested of you by an FDA official. Any telefaxed material must be followed by a hard copy to the DMC (HFZ-401).

You should be familiar with the manual entitled, "Premarket Notification 510(k) Regulatory Requirements for Medical Devices" available from the DSMICA. If you have other procedural or policy questions, or want information on how to check on the status of your submission (after 90 days from the receipt date), please contact the DSMICA at (301) 443-6597 or its toll-free number (800) 638-2041, or at their Internet address http://www.fda.gov/cdrh/dsmamain.html or me at (301) 594-1190.

Sincerely yours,

Marjorie Shulman
Consumer Safety Officer

1/6

Kirkpatrick & Lockhart LLP

1800 Massachusetts Avenue, NW
Suite 200
Washington, DC 20036-1221
202.778.9000
www.kl.com

1C013819

November 14, 2001

Donald R. Stone
202.778.9067
Fax: 202.778.9100
dstone@kl.com

VIA FEDERAL EXPRESS

Food and Drug Administration
Document Mail Center (HFZ-401)
Center for Devices and Radiological Health
Office of Device Evaluation
9200 Corporate Boulevard
Rockville, MD 20850

Re: 510(k) Premarket Notification: Sierra Diagnostics, L.L.C. – Urine Collection, Preservation and Transport System

Dear Sir or Madam:

Pursuant to Section 510(k) of the Federal Food, Drug, and Cosmetic Act and 21 C.F.R. § 807.7, enclosed please find a 510(k) premarket notification for the Sierra Diagnostics, L.L.C. Urine Collection, Preservation and Transport System (for use with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays), which Kirkpatrick & Lockhart LLP is forwarding as counsel to the submitter, Sierra Diagnostics, L.L.C., Senora, California ("Sierra").

In accordance with 21 C.F.R. § 807.90(c), we have enclosed the original and one copy of the premarket notification submission, as well as the original and two copies of this cover letter.

Please note that **Exhibits 6, 7, and 8** of Sierra's 510(k) submission contain trade secret and confidential commercial information and, therefore, are **exempt from public disclosure** under the Freedom of Information Act (5 U.S.C. § 552(b)(4)) and the applicable Food and Drug Administration regulations (21 C.F.R. § 20.61).

SKIF 117

Kirkpatrick & Lockhart LLP

1800 Massachusetts Avenue, NW
Suite 200
Washington, DC 20036-1221
202.778.9000
www.kl.com

November 14, 2001

Donald R. Stone
202.778.9067
Fax: 202.778.9100
dstone@kl.com

VIA FEDERAL EXPRESS

Food and Drug Administration
Document Mail Center (HFZ-401)
Center for Devices and Radiological Health
Office of Device Evaluation
9200 Corporate Boulevard
Rockville, MD 20850

Re: 510(k) Premarket Notification: Sierra Diagnostics, L.L.C. – Urine Collection, Preservation and Transport System

Dear Sir or Madam:

Pursuant to Section 510(k) of the Federal Food, Drug, and Cosmetic Act and 21 C.F.R. § 807.7, enclosed please find a 510(k) premarket notification for the Sierra Diagnostics, L.L.C. Urine Collection, Preservation and Transport System (for use with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays), which Kirkpatrick & Lockhart LLP is forwarding as counsel to the submitter, Sierra Diagnostics, L.L.C., Senora, California ("Sierra").

In accordance with 21 C.F.R. § 807.90(c), we have enclosed the original and one copy of the premarket notification submission, as well as the original and two copies of this cover letter.

Please note that **Exhibits 6, 7, and 8** of Sierra's 510(k) submission contain trade secret and confidential commercial information and, therefore, are **exempt from public disclosure** under the Freedom of Information Act (5 U.S.C. § 552(b)(4)) and the applicable Food and Drug Administration regulations (21 C.F.R. § 20.61).

118

If you have any questions concerning this notification, please contact either me at (202) 778-9067 or Michael Hinckle at (202) 778-9296.

Sincerely,

Donald R. Stone

Donald R. Stone

Enclosure(s)
cc: Michael H. Hinckle
Sierra Diagnostics, L.L.C.

SIERRA DIAGNOSTICS, L.L.C.

PREMARKET NOTIFICATION
URINE COLLECTION, PRESERVATION AND TRANSPORT SYSTEM

TABLE OF CONTENTS

I. SUBMITTER'S INFORMATION 1
- A. Address 1
- B. Manufacturing Site 1
- C. Contact Person 1
- D. Establishment Registration Number 1

II. PRODUCT CLASSIFICATION 1
- A. Device Class 1
- B. Classification Regulations 2

III. ACTION TAKEN TO COMPLY WITH SECTION 513 OR 514 OF THE FEDERAL FOOD, DRUG, AND COSMETIC ACT 2

IV. PROPOSED PRODUCT LABELING 2

V. INTENDED USE 2

VI. 510(k) SUMMARY 2

VII. LEGALLY MARKETED DEVICE TO WHICH SIERRA DIAGNOSTICS, L.L.C. CLAIMS EQUIVALENCE 2

VIII. DESCRIPTION OF THE DEVICE 3

IX. STATEMENT OF SIMILARITIES TO, AND/OR DIFFERENCES FROM, THE PREDICATE DEVICE 4

X. COMPARISON TABLE OF THE DEVICE TO THE PREDICATE DEVICE 5

XI. PERFORMANCE STUDIES 5

XII. BIOCOMPATIBILITY INFORMATION 7

XIII. STERILIZATION INFORMATION 7

XIV. SPECIFIC GUIDANCE DOCUMENT 7

XV. TRUTHFUL AND ACCURATE STATEMENT 7

EXHIBITS

Title	Exhibit No.
Proposed Product Labeling	1
Indications for Use Statement	2
510(k) Summary	3
Abbott LCx® *Neisseria gonorrhoeae* Assay Product Labeling	4
Abbott LCx® *Chlamydia trachomatis* Assay Product Labeling	5
Device Components	6
Performance Study Data Tables	7
Sterilization Information	8

121

PREMARKET NOTIFICATION ("510(K)")
Sierra Diagnostics, L.L.C.
Urine Collection, Preservation and Transport System
(For Use with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays)

The following information is being submitted in accordance with 21 C.F.R. § 807.87:

I. SUBMITTER'S INFORMATION

A. Address

Sierra Diagnostics, L.L.C.
21109 Longeway #C
Sonora, CA 95370
Tele: (209) 536-0886
Fax: (209) 536-0853

B. Manufacturing Site

Same as above.

C. Contact Person

Donald R. Stone, Esq.
Michael H. Hinckle, Esq.
Kirkpatrick & Lockhart, LLP
1800 Massachusetts Ave., NW
Washington, DC 20036
Tele: 202 778-9067 / 202 778-9296
Fax: 202 778-9100

D. Establishment Registration Number

2953142

II. PRODUCT CLASSIFICATION

A. Device Class

Class II – Accessory to Neisseria spp. and Chlamydia serological reagents.

122

Neisseria gonorrhoeae and *Chlamydia trachomatis* assays and referenced in the package inserts for the LCx® devices (See 510(k) Nos. K935833 (*Neisseria gonorrhoeae*) and K934622 (*Chlamydia trachomatis*)). Copies of the package inserts for the Abbott LCx® gonococcal and chlamydial assays are included in this submission as Exhibits 4 and 5, respectively.

VIII. DESCRIPTION OF THE DEVICE

Abbott's LCx® gonococcal and chlamydial assays have shown excellent sensitivity and specificity in clinical trials using urine as the primary specimen. However, the major disadvantage to the use of urine as a test specimen is the need to (1) immediately refrigerate the specimen at 2-8°C, and (2) transport the specimen to the testing laboratory within 24 hours of collection. These precautions are required because the nucleic acid targets that the LCx® assays detect degrade over time due to the enzyme activity that naturally occurs in urine.

The Urine Collection System acts to slow the nucleic acid degradation process and therefore allows urine specimens to be stored at room temperature (up to 60°C) for 6 days prior to testing with the appropriate LCx® assay. The device achieves this result by:

1. Inactivating a broad range of heat catalyzed enzyme systems that are responsible for the degradation of the nucleic acid targets;
2. Preventing the log phase growth of bacteria that can result in a shift in the urine pH; and
3. Buffering the specimen to stabilize the pH at optimal conditions for the LCx® assay.

The Urine Collection System is comprised of a sterile urine cup that contains a preservative and three red indicator beads. The beads in the cup serve as an indicator that the preservative is present in the specimen. A quantitative list of the proprietary components of the preservative is provided in Exhibit 6.

The urine collection cup is marked with "maximum" and "minimum" fill lines. In use, the patient is instructed to the fill the cup with first stream urine to a level between the two lines that are marked on the cup. This ensures that the preservative-to-urine ratio in the specimen will be within the effective dilution range for the preservative (i.e., 1:10 to 1:15).

Sierra proposes to market the Urine Collection System in any one or more of the following five package sizes:

Volume of Preservative	Collection Cup Size
	40 ml
	40 ml
	90 ml
	90 ml
	90 ml

To use the Urine Collection System, the patient fills the collection cup with first stream urine to a level between the maximum and minimum levels. After the patient closes and labels the cup, the specimen is ready to be transported to the laboratory without the need for refrigeration. Based on the data provided in this submission, Sierra has determined that the preserved urine should be tested within 6 days of collection and should be stored at temperatures not exceeding 60°C (the preserved urine may also be frozen).

Lastly, the Urine Collection System will be packaged in a heat sealed bag, and will be labeled with a 24 month expiration date.

IX. STATEMENT OF SIMILARITIES TO, AND/OR DIFFERENCES FROM, THE PREDICATE DEVICE

The Urine Collection System is identical to the standard collection cup referenced in the LCx package insert with the exception of the addition of a preservative and indicator beads. The basic intended use of both devices is the same (i.e., to collect, store, and transport urine specimens while preserving the nucleic acid targets). The only significant differences between Sierra's device and the predicate device are: (1) the predicate device uses refrigeration to preserve the nucleic acids in the specimen while Sierra's device uses a chemical preservative; and (2) the Sierra device is specifically labeled for use with the LCx assays.

As evidenced by the comparative studies contained in this submission, Sierra's Urine Collection System is as safe and effective for the collection and preservation of urine specimens for LCx testing as the predicate device.

X. COMPARISON TABLE OF THE DEVICE TO THE PREDICATE DEVICE

	DNA/RNA Protect System	Standard Collection Cup
Intended Use	Collection, Preservation, and Transport of Urine Specimens for LCx testing	Collection, Preservation, and Transport of Urine Specimens for LCx testing
Storage Requirements	[redacted]	4 days at 2-8°C
Preservative	Chemical Preservation	None, relies on refrigeration
Sterile	Yes	Yes

XI. PERFORMANCE STUDIES

Sierra has confirmed the effectiveness of its Urine Collection System through a variety of test methods. As described in greater detail below and in the attached tables, Sierra has compared its device to a standard collection cup (with refrigeration) to demonstrate that urine specimens collected and preserved in the Urine Collection System will provide LCx® test results for *N. gonorrhea* and *C. trachomatis* that are identical to those of specimens collected and preserved with the predicate device.

A. Spiked Urine Studies

Sierra spiked fresh urine with chlamydial and gonococcal DNA targets [redacted]. The chlamydial targets were from the 9 individual chlamydial serovars (types B, D, E, F, G, H, I, J, K) that occur most frequently in cervical infections, and the gonococcal targets represented the 18 individual PIA serovars and the 25 individual PIB serovars that comprise pathogenic *Neisseria gonorrhea.*

The spiked urine samples were divided in half and either frozen at -70°C or preserved with the Sierra preservative at a ratio of 1:10 (preservative to urine). Control tubes without DNA targets were prepared in an identical manner.

The urine specimens preserved with the Sierra Urine Collection System were stored at [redacted] for 160 hours, while the "fresh" specimens were stored at -70°C. Each specimen was then tested and the results tabulated. A summary of the test results is enclosed at Exhibit 7, Tables 1A – 1E and 2A – 2G. These data demonstrate that the Sierra Urine Collection System is equivalent to the predicate device in its ability to preserve chlamydial and gonococcal DNA targets in urine for detection with the LCx® assays.

B. Multi-Site Clinical Study

In order to demonstrate that the Sierra's Urine Collection System is substantially equivalent to the predicate device in a clinical setting, Sierra conducted a multi-site clinical study. Subjects at each site (male and female) were instructed to fill two cups with urine to a premarked volume of 20 ml. The first approximately 20 ml of urine was always added to the predicate device while the second approximately 20 ml was added to the Sierra Urine Collection System cup. This design was used in order to eliminate any potential wash out effect and ensure that, were any wash out to occur, the predicate device would have the greater number of potential DNA targets. At four of the study sites, a preservative-to-urine ratio of 1:10 (2 ml preservative to 20 ml of urine) was used, while a ratio of 1:5 (4 ml of preservative to 20 ml of urine) was used at one study site.

The specimens collected in the predicate device were refrigerated (2-8°C) within one hour of collection and transported (packed with ice packs) to the laboratory for LCx® testing within 24 hours of collection. The specimens collected in the Sierra Urine Collection System were transported to the laboratory at ambient temperature and stored at ambient temperature for 144 hours prior to testing.

Test results for the preserved specimens were correlated against the refrigerated specimens. All results correlated at 100%. Summaries of the data from all sites are enclosed at Exhibit 7, Tables 3A – 3E.

Also enclosed as Tables 4A and 4B of Exhibit 7, are the presentations of the clinical study data from females and males demonstrating that the Urine Collection System effectively preserves specimens from both symptomatic and asymptomatic patients.

C. Spiked Urine Sensitivity Testing At Varying Preservative-to-Urine Ratios

To ensure that the Urine Collection System did not have an adverse effect on the sensitivity of the LCx® assay, and to demonstrate the range of effective preservative-to-urine ratio of the device, Sierra conducted an analytical sensitivity test using two concentrations of Sierra's preservative. According to the LCx® package insert, the sensitivity of the assay is 10 cfu of *N. gonorrhea* organisms. Thus, Sierra confirmed the effectiveness of its Urine Collection System by preparing serial dilutions of 10 gonococcal serovars (1A-13, 1A-20, 1A-5, 1A-2, 1A-3, 1B-17, 1B-2, 1B-18, 1B-7, 1B-5), and spiking fresh urine specimens with a gonococcal culture of less than 10 cfu. LCx® test results were recorded from unpreserved spiked specimens (stored at -70°C, tested within 24 hours) and spiked specimens preserved with either a 1:10 or 1:15 concentration of Urine Collection System preservative (stored at 25°C and tested after 144 hours).

127

The data from the analytical sensitivity testing, which is enclosed as Table 5 of Exhibit 7, demonstrates that preserving a urine specimen with Sierra's Urine Collection System does not affect the sensitivity of the LCx® assay, and that the preservative is effective when used in a dilution range of 1:10 to 1:15.

D. Stability Study

To confirm that the stability of the Urine Collection System throughout its labeled expiration dating period, Sierra compared the nuclear magnetic resonance (NMR) and elemental analysis test results obtained from a 33 month old device with a newly manufactured device. Those data confirmed that the system is stable throughout the labeled 24-month expiration period.

I. BIOCOMPATIBILITY INFORMATION

The Urine Collection System is not intended to contact the body of the user and therefore does not present any biocompatibility issues.

II. STERILIZATION INFORMATION

The interior of the Urine Collection System cup and its contents (i.e., preservative and indicator beads) will be sold sterile. A description of the sterilization process is contained in Exhibit 8.

III. SPECIFIC GUIDANCE DOCUMENT

Not applicable.

IV. TRUTHFUL AND ACCURATE STATEMENT

I certify that, in my capacity as Chief Technical Officer for Sierra Diagnostics, L.L.C., I believe to the best of my knowledge, that all data and information submitted in this premarket notification are truthful and accurate and that no material fact has been omitted.

Tony K. Baker
Chief Technical Officer
Sierra Diagnostics, L.L.C.

November 13, 2001

EXHIBIT 1

PROPOSED PRODUCT LABELING

[PROPOSED PACKAGE INSERT]

SIERRA DIAGNOSTICS, L.L.C.
URINE COLLECTION, PRESERVATION AND TRANSPORT SYSTEM
For use with the Abbott LCx® Assays for *Neisseria gonorrhoeae* and *Chlamydia trachomatis*

Indications for Use

The Sierra Diagnostics L.L.C. Urine Collection, Preservation, and Transport System is indicated for use in the collection, preservation, and transportation of urine specimens at temperatures not exceeding 60°C for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays.

Precautions

Specimens should not be collected from patients who have urinated one hour prior to the collection.

Instruction for Use

The patient should be instructed to fill the collection cup with first pass urine (the first part of the stream) to a level between the two lines printed on the outside of the cup. After the urine is collected, verify that the cup is securely closed and labeled with the patient's ID number and date of collection. The red indicator beads serve as a reminder that the preservative is present in the specimen. Transport the specimen to the laboratory for testing. There is no need to refrigerate or freeze the specimen. The specimen will be suitable for LCx® testing for 144 hours (6 days) provided it is stored at temperatures not exceeding 60°C (140°F).

Storage

The Urine Collection, Preservation and Transport System should be stored at room temperature prior to use. Use prior to expiration date printed on outside of package.

[PROPOSED PACKAGE LABEL]

Cup Label (2.0 ml preservative)

URINE COLLECTION, PRESERVATION AND TRANSPORT SYSTEM **2.0 ML LOT # _____** **EXP. DATE _____** **STORE AT ROOM TEMP.** **FOR IN VITRO DIAGNOSTIC USE ONLY** **SIERRA DIAGNOSTICS, L.L.C.** **SONORA, CA. 95370**	Patient _____ ID _____ Date _____ Time _____ Physician _____ Phone _____

Sterile Label for Cup (attaches between cup and cap)

STERILE

EXHIBIT 2

INDICATIONS FOR USE STATEMENT

510(k) Number:

Device Name: Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System

Indications for Use:

The Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System is indicated for use in the collection, preservation, and transportation of urine specimens at temperatures not exceeding 60°C for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays.

PLEASE DO NOT WRITE BELOW THIS LINE. CONTINUE ON ANOTHER PAGE IN NEEDED

Concurrence of CDRH, Office of Device Evaluation (ODE)

Prescription Use _____ OR Over-The Counter Use _____
(Per 21 CFR 801.109)

EXHIBIT 3

510(k) SUMMARY

If you have any questions concerning this notification, please contact either me at (202) 778-9067 or Michael Hinckle at (202) 778-9296.

Sincerely,

Donald R. Stone

Donald R. Stone

Enclosure(s)
cc: Michael H. Hinckle
Sierra Diagnostics, L.L.C.

510(k) Summary

I. General Information on Submitter

Name:	Sierra Diagnostics, L.L.C.
Address:	21109 Longeway #C Sonora, CA 95370
Telephone:	(209) 536-0886
Fax:	(209) 536-0853
Contact Person:	Tony Baker
Date Prepared:	October __, 2001

II. General Information on Device

Name:	Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System
Classification Name:	Accessory to Neisseria spp. and Chlamydia serological reagents

III. Predicate Device

The standard urine collection cup used to collect specimens for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays and referenced in the package inserts for the LCx® devices (See 510(k) Nos. K935833 (*Neisseria gonorrhoeae*) and K934622 (*Chlamydia trachomatis*)).

IV. Description of Device

The device is comprised of a urine collection cup containing of a nucleic acid chemical preservative. The device allows urine specimens for LCx® gonococcal or chlamydial testing to be preserved for up to 6 days at temperatures not to exceed 60°C. Inert indicator beads are included in the urine cup as an indicator that a preservative is present in the sample.

V. Intended Use

The Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System is intended for use in the collection, preservation, and transportation of urine specimens at temperatures not exceeding 60°C for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays.

VI. Technological Characteristics of Device Compared to Predicate Device

The Sierra Urine Collection, Preservation, and Transport System and the predicate device share the same technological characteristics with the exception of the method of preservation. The predicate device employs a temperature preservation method while the Sierra device uses chemical preservation.

VII. Summary of Performance Data

The effectiveness of the Sierra Urine Collection, Preservation, and Transport System was established by the comparative testing of fresh and preserved urine spiked with gonococcal and chlamydial DNA. LCx® testing of samples that were preserved through refrigeration for 24 hours were compared with results for specimens preserved with the Sierra device and tested after being held for 144 hrs. at 60°C. There was a 100% correlation between the refrigerated and preserved samples.

Effectiveness was further established by a multi-site clinical study. The results of this study demonstrated that the device effectively preserved gonococcal and chlamydial nucleic acid targets in urine specimens from symptomatic and asymptomatic males and females.

The effective preservative concentration range and effect on LCx® sensitivity was established by a study using urine specimens spiked with less than 10 cfu of 10 different gonococcal serovars. Results from this test proved that Sierra's device effectively preserved nucleic acid targets down to the LCx® level of detection with a preservative to urine ratio ranging from 1:10 to 1:15.

137

EXHIBIT 4

Abbott LCx® *Neisseria gonorrhoeae* Assay Product Labeling

ABBOTT LCx® PROBE SYSTEM

Neisseria gonorrhoeae Assay

Note Changes Highlighted

Customer Support Center (USA)
1-800-527-1869
Outside of USA –
Contact your local Customer Service Center
66-8477/R1

139

ABBOTT LABORATORIES
Diagnostics Division
Abbott Park, IL 60064

August, 1996

List No. 8A68-81
Printed in U.S.A.

INTENDED USE

The LCx® *Neisseria gonorrhoeae* Assay uses LCR™ (Ligase Chain Reaction) amplification technology in the LCx Probe System for the direct, qualitative detection of a specific target nucleic acid sequence in the Opa gene of *Neisseria gonorrhoeae* in female endocervical and male urethral swab specimens or in male and female urine specimens from symptomatic and asymptomatic males and females.

SUMMARY AND EXPLANATION OF THE TEST

Gonorrhea is one of the most commonly reported sexually transmitted diseases in the United States with nearly 700,000 reported cases per year. *N. gonorrhoeae* is the etiologic agent of gonorrhea. In men, gonorrhea infection usually results in acute anterior urethritis accompanied by a purulent exudate[1]. In women, the infection is most often found in the cervix, but the vagina and uterus also may be infected. Frequently the infection is asymptomatic, especially in women. Without treatment, local complication of gonococcal infection can occur including pelvic inflammatory disease (PID) or acute salpingitis for women and epididymitis for men[1].

N. gonorrhoeae is a Gram-negative, oxidase-positive diplococcus without flagellae[1]. The gold standard for the detection of gonorrhea is the culture of *N. gonorrhoeae*[2]. Presumptive diagnosis of gonorrhea is based on the morphological examination, Gram stain, and oxidase measurement of the culture isolate. Confirmation procedures have been used for definitive identification of *N. gonorrhoeae* including sugar fermentation, fluorescent antibody staining, nucleic acid hybridization and agglutination[2,3].

The LCx *Neisseria gonorrhoeae* Assay uses the nucleic acid amplification method LCR to detect the presence of *N. gonorrhoeae* DNA directly in clinical specimens[4]. The four oligonucleotide probes in the LCx assay recognize and hybridize to a specific target sequence within the Opa gene of *N. gonorrhoeae* DNA. The oligonucleotides are designed to be complementary to the target sequence so that in the presence of target, the probes will bind adjacent to one another. They can be enzymatically joined to form the amplification product which subsequently serves as an additional target sequence during further rounds of amplification. The product of the LCR reaction is detected on the Abbott LCx Analyzer.

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

Specimen Preparation

Specimen preparation is the first step of the LCx *Neisseria gonorrhoeae* Assay during which the DNA of the organism is released and made accessible to the enzymes and other components of the LCR reaction. This entails disrupting the cells and separating the strands of the DNA within the specimen. This is accomplished by heating the clinical specimen in Swab Specimen Transport Buffer for endocervical and male urethral swabs, or LCx Urine Specimen Resuspension Buffer for urine specimens.

Amplification

The LCR target is located in the Opa genes of *N. gonorrhoeae*. The 48 base pair sequence was selected as the target DNA because it is conserved in all strains of *N. gonorrhoeae* thus studied, and it is the most specific to *N. gonorrhoeae*[5]. Up to 11 copies of the Opa gene are found per cell[6].

In the DNA amplification step, the prepared sample is added to the LCR reaction mixture consisting of four oligonucleotide probes, thermostable ligase and polymerase, and individual nucleotides in buffer.

The four oligonucleotide probes are designed in pairs that hybridize to complementary single-stranded *N. gonorrhoeae* target sequences exposed in sample preparation. When a pair of probes has hybridized to a target sequence on a single strand of DNA, there is a gap of a few nucleotides between the probes. Polymerase acts to fill in this gap with the nucleotides in the LCR reaction mixture. Once the gap is filled, ligase can covalently join the pair of probes to form an amplification product that is complementary to the original target sequence and can itself serve as a target in subsequent rounds of amplification. Amplification occurs when the LCR reaction mixture and sample are incubated in a DNA thermal cycler.

During thermal cycling, the temperature is raised above the melting point of the hybridized amplification product causing it to dissociate from the original target sequence. Lowering the temperature allows more of the oligonucleotide probes to hybridize to the targets now available. The temperature continues to be cycled in this manner until sufficient numbers of target amplification product have accumulated and can be detected by Microparticle Enzyme Immunoassay (MEIA). Because the number of targets increases exponentially, forty thermal cycles are sufficient to achieve up to a billion-fold amplification in the number of target sequences.

Detection

The two pairs of oligonucleotide probes in the LCx *Neisseria gonorrhoeae* Assay are labeled with immunoreactive chemical groups called haptens. Each individual probe has either a capture hapten (recognized by an antibody attached to the MEIA microparticles) or a detection hapten (recognized by an antibody conjugated to alkaline phosphatase). The probes are labeled such that, when they are joined during the LCR reaction, the amplification product has the capture hapten at one end and the detection hapten at the other. In the LCx Analyzer, a sample of the amplification product is automatically transferred to an incubation well[7]. Here the microparticles coated with anti-capture hapten (Rabbit) bind the amplification product as well as any unligated probes carrying the capture hapten. The reaction mixture is then automatically transferred to a glass fiber matrix to which the microparticle complexes bind irreversibly. A wash step removes the unligated probes having only the detection hapten. The bound microparticle complexes are then incubated with anti-detection hapten (Rabbit): Alkaline Phosphatase conjugate which binds to the detection haptens. This antibody conjugate binds only to amplification product. The antibody conjugate can then be detected by addition of the substrate, 4-Methylumbelliferyl Phosphate, which is dephosphorylated by alkaline phosphatase to produce a fluorescent molecule, 4-Methylumbelliferone, that is measured by the MEIA optical assembly.

Prevention of DNA Contamination

Amplification reactions such as LCR are sensitive to accidental introduction of product from previous amplification reactions. False-positive results could occur if either the clinical specimen or the LCx reagents used in the amplification step become contaminated by accidental introduction of even a few molecules of amplification product. Measures to reduce the risk of DNA contamination in the laboratory include physically separating activities involved in performing LCR, chemically inactivating amplification product and complying to good laboratory practice.

Dedicated Laboratory Areas

Sample preparation must be completed in a dedicated area (Area 1) of the laboratory. Area 1 is used for processing of samples (specimens, LCx Gonorrhea Negative Control and Calibrator) and the addition of processed samples to LCx Gonorrhea Amplification Vials. Amplification and detection of amplification product is completed in a second dedicated area (Area 2).

Aerosol Containment

To reduce the risk of DNA contamination due to aerosols formed during pipetting, pipettors with aerosol barrier pipette tips must be used for all pipetting except for one step of the Urine Specimen Preparation protocol. This protocol allows for fine-tipped, single-use, plastic disposable pipettes. Refer to Specimen Preparation, Urine Specimen Preparation in this assay package insert.

Inactivation of Amplification Product

To reduce the risk of amplification product contamination, at the end of the LCx *Neisseria gonorrhoeae* Assay, amplification product is automatically inactivated using a two-reagent, chemical inactivation system. Both reagents (a chelated metal complex and an oxidizing agent) are delivered into the LCx Reaction Cells by the LCx Analyzer after the amplification product has been detected. The ensuing reaction results in the nearly complete destruction of any nucleic acid present. This effectively reduces the risk of contamination of the laboratory by amplification product.

REAGENTS

LCx STD Swab Specimen Collection and Transport Kit

(100 individually-wrapped sterile Collection Systems)

Each Collection System contains: one capped transport tube with 0.5 mL Swab Specimen Transport Buffer, one large-tipped cleaning swab, and one small-tipped specimen swab. Swab Specimen Transport Buffer contains ≥50 mM $MgCl_2$. Preservative: Sodium Azide.

140

LCx® Urine Specimen Preparation Kit

(100 tests, 4 bottles of LCx Urine Specimen Resuspension Buffer, and 100 LCx Urine Specimen Microfuge Tubes with Cap Locks)

LCx Urine Specimen Resuspension Buffer contains ≥50 mM $MgCl_2$ and detergent. Preservative: Sodium Azide.

LCx Gonorrhea Amplification Kit[a]

LCx Gonorrhea Amplification Vials

(96 vials, 0.090 mL per vial)

Four oligonucleotide probes each at $>10^{10}$ molecules per reaction, enzymes (≥1 unit thermostable DNA polymerase and ≥10,000 units thermostable DNA ligase), ≥3 µM dNTP, ≥20 µM NAD, and stabilizers in a buffered solution. Preservative: Sodium Azide.

LCx Gonorrhea Negative Control, Calibrator and Activation Reagent

(8 sets of each)

LCx Gonorrhea Negative Control (N)

(0.48 mL per bottle)

≥1.8 µg/mL Salmon Testes DNA in a buffered solution. Preservative: Sodium Azide.

LCx Gonorrhea Calibrator (C)

(0.48 mL per bottle)

Extracted DNA from inactivated *N. gonorrhoeae* at approximately 540 CFU/mL in a buffered solution. Preservative: Sodium Azide.

LCx Gonorrhea Activation Reagent (A)

(0.7 mL per bottle)

≥300 mM $MgCl_2$, dye red (FD&C Red No. 2) and stabilizers in a buffered solution. Preservative: Sodium Azide.

LCx Gonorrhea Detection Reagent Pack[a]

(100 tests)[b]

1. 1 bottle, (6 mL)
   Anti-Capture Hapten (Rabbit) coated Microparticles, >0.025% solids in buffered solution. Preservative: Sodium Azide.
2. 1 bottle, (8 mL)
   Anti-Detection Hapten (Rabbit): Alkaline Phosphatase Conjugate, >0.01 µg/mL, in buffered solution with stabilizers and antimicrobials.
3. 1 bottle, (7 mL)
   4-Methylumbelliferyl phosphate, 1.2 mM, in buffered solution. Preservative: Sodium Azide.
4. 1 bottle, (25 mL)
   Metal chelate, >5 mM, in buffered solution.

[a]The LCx Gonorrhea Amplification Kit and the LCx Gonorrhea Detection Reagent Pack are packaged as a set which must be used together. Verify that the lot numbers are identical before use.

[b]There are 100 tests provided in the LCx Gonorrhea Detection Reagent Pack. Ninety-six tests are provided for the LCx *Neisseria gonorrhoeae* Assay. An additional 4 tests remain, of which 3 tests are provided for the purpose of troubleshooting the detection portion of the assay. See the LCx Analyzer Operations Manual, Section 10.

LCx Inactivation Diluent (1)

(2 bottles, 900 mL per bottle)

An aqueous solution of 6-7.9% hydrogen peroxide.

LCx System Diluent (2)

(4 bottles, 1000 mL per bottle)

Tris-Acetate Buffer, >0.01 M. *Preservative: Sodium Azide.*

WARNINGS AND PRECAUTIONS

For *In Vitro* Diagnostic Use.

The LCx *Neisseria gonorrhoeae* Assay is only for use with female endocervical and male urethral swabs, or male and female urine specimens, and is limited to specimens collected, transported and stored according to instructions in the SPECIMEN COLLECTION AND TRANSPORT TO TEST SITE and SPECIMEN PREPARATION sections.

Use only the LCx STD Swab Specimen Collection and Transport Kit for collection of swab specimens.

LCx STD Swab Specimen Collection Systems in which transport media has spilled out should not be used.

The LCx STD Swab Specimen Collection and Transport Kit is intended to be used only in LCx Assays which require its use. No other intended use is applicable.

Use only the LCx Urine Specimen Preparation Kit for urine specimen processing.

The LCx Gonorrhea Amplification Kit and the LCx Gonorrhea Detection Reagent Pack are packaged as a set which must be used together. Verify that the lot numbers are identical before use.

Do not mix reagents from different lots. Do not pool reagents.

Use two dedicated areas within the laboratory for performing the LCx *Neisseria gonorrhoeae* Assay: Area 1 and Area 2.

Area 1 is to be used for the processing of samples (specimens, LCx Gonorrhea Negative Control and Calibrator), and the addition of samples to the LCx Gonorrhea Amplification Vials. Use of a biosafety hood or glove box equipped for UV irradiation is recommended. **All reagents and equipment used in Area 1 (such as pipettors, microcentrifuge and the Abbott LCx Dry Bath) should remain in this dedicated area at all times. Area 1 items should never be used when working with amplification product. Do not bring amplification product into Area 1.** Specimens, and activated Negative Control and Calibrator should be stored separately from Amplification Vials. All pipetting should be performed with aerosol barrier pipette tips except for a disposable pipette which may be used for one step in the Urine Specimen Preparation protocol. Swab specimens must only use extended-length (≥75 mm in length) aerosol barrier pipette tips to aspirate specimen from transport tubes. Cap locks must be placed on LCx Urine Specimen Microfuge tubes before specimens are heated in the LCx Dry Bath.

Area 2 is dedicated to amplification by thermal cycling and detection of the amplification product. **All equipment, such as the Abbott LCx Thermal Cycler, the LCx Analyzer, and all the accessories for the LCx Analyzer must be kept in this area. The LCx Gonorrhea Detection Reagent Pack must be stored at 2-8°C. Care must be taken to separate the detection reagent pack that is in use from *direct contact with samples* and other LCx reagents.** During execution of the detection protocol, aerosols from amplification product may potentially contaminate any surface within the closed LCx Analyzer. Everything inside the LCx Analyzer (including the detection reagent pack and the carousel) must be considered potential sources of DNA contamination and must be kept away from other LCx reagents, Negative Controls, Calibrators and specimens.

The LCx Calibrator contains extracted DNA from *N. gonorrhoeae* that have been inactivated by a heat and chemical treatment process.

All specimens and reagents should be handled as potentially infectious materials using universal precautions as specified in the *OSHA Standard*[5] *on bloodborne pathogens, December, 1991*, or other applicable biosafety guidelines.[9,11] This includes, but is not limited to, the use of eye protection, lab coat and disposable gloves. Wash hands thoroughly after handling kit reagents and specimens. Do not pipette by mouth. Do not smoke, eat or drink in areas in which specimens or kit reagents are handled.

To reduce the risk of DNA contamination, clean and disinfect all spills of specimens and reagents using 1% (v/v) sodium hypochlorite solution, followed with 70% (v/v) ethanol. **Note: Chlorine solutions may pit equipment and metal. Use sufficient amounts or repeated applications of 70% ethanol until chlorine residue is no longer visible.**

Dispose of all materials that have come into contact with specimens and reagents in accordance with local, state and federal regulations. Solid waste may be incinerated or autoclaved for an appropriate period of time. Due to variations among autoclaves and in waste configuration, each user must verify the effectiveness of the decontamination cycle using biological indicators.

This product contains sodium azide as a preservative.

Sodium azide has been reported to form lead or copper azide in laboratory plumbing. These azides may explode on percussion, such as hammering. To prevent formation of lead or copper azide, flush drains thoroughly with water after disposing of solutions containing sodium azide.

To remove contamination from old drains suspected of azide accumulation, the National Institute for Occupational Safety and Health recommends the following: (1) siphon liquid from trap using a rubber or plastic hose, (2) fill with 10% sodium hydroxide solution, (3) allow to stand for 16 hours, and (4) flush well with water.

141

Avoid contact of the skin or mucus membranes with the metal chelate solution or with deposits around the wash station of the LCx® Analyzer. The LCx Inactivation Diluent (1) can irritate skin. If contact occurs with either solution, wash immediately with soap and large amounts of water.

Do not use kits or reagents beyond the expiration date.

Use accurately calibrated equipment.

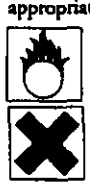
Failure to adhere to assay package insert instructions may result in erroneous results.

If the Step-Cycle run of the thermal cycler is interrupted or aborted, the run is invalid. Do not continue to process these samples. Make sure that the amplification vial caps are tightly closed. Remove carefully to a biohazard bag and seal the bag. Dispose of according to the procedure of waste disposal in the LCx Thermal Cycler Operations Manual, Section 8: Hazards, Biosafety.

Some components of this product contain Sodium Azide. For a specific listing, refer to the REAGENTS section of this package insert. The components containing Sodium Azide are classified per applicable European Economic Community (EEC) Directive as: Harmful (Xn). The following are the appropriate Risk (R) and Safety (S) phrases.

R22	Harmful if swallowed.
R32	Contact with acids liberates very toxic gas.
S2	Keep out of reach of children.
S13	Keep away from food, drink and animal feedingstuffs.
S36	Wear suitable protective clothing.
S46	If swallowed, seek medical advice immediately and show this container or label.

The LCx Inactivation Diluent (1) contains Hydrogen Peroxide and is classified per applicable European Economic Community (EEC) Directives as: Oxidizing (O) and Irritant (Xi). The following are the appropriate Risk (R) and Safety (S) phrases.



R8	Contact with combustible material may cause fire.
R36/R38	Irritating to eyes and skin.
S1/2	Keep locked up and out of reach of children.
S36/39	Wear suitable protective clothing and eye/face protection.
S26	In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
S14l	Keep away from direct sunlight.

STORAGE INSTRUCTIONS

1. LCx Reaction Cells may be stored at 15-30°C until the expiration date.
2. The LCx Gonorrhea Amplification Vials, LCx Gonorrhea Activation Reagent, Negative Control, and Calibrator must be stored at 2-8°C until the expiration date.
3. The LCx Gonorrhea Detection Reagent Pack must be refrigerated at 2-8°C when not in use. Care must be taken to separate the LCx Gonorrhea Detection Reagent Pack that is in use from direct contact with samples and other LCx kit reagents. The detection reagents must not be frozen.
4. The LCx System Diluent (2) and the LCx Inactivation Diluent (1) may be stored at 15-30°C until the expiration date. The LCx Inactivation Diluent (1) must be kept away from direct sunlight.

SPECIMEN COLLECTION AND TRANSPORT TO TEST SITE

For domestic or international shipments, specimens should be packaged and labeled in compliance with applicable state, federal and international regulations covering the transport of clinical specimens and etiologic agents/infectious substances.

Time and temperature conditions for storage must be adhered to during transport. See Swab Specimen Transport and Urine Specimen Collection and Transport Sections for storage conditions.

For swab specimen collection, use only the LCx STD Swab Specimen Collection and Transport Kit. (No. 3B15 or No. 6C94).

Note: Do Not Use The Large-tipped Cleaning Swab For Specimen Collection.

Note: Swab or urine specimens that are moderately bloody (greater than approximately 0.5% (v/v)) or grossly mucoid (greater than approximately 5% (w/v)) should not be tested since they may cause inhibition in the LCx *Neisseria gonorrhoeae* Assay.

Endocervical Swab Specimen Collection

1. Remove excess mucus from the exocervix with the large-tipped cleaning swab provided in the LCx STD Swab Specimen Collection System and discard.
2. Insert the small-tipped, specimen swab into the endocervix and rotate the swab for 15 to 30 seconds to ensure adequate sampling.
3. Verify that all Swab Specimen Transport Buffer is at the bottom of the tube. If necessary, tap or shake the solution down to the bottom of the tube. Unscrew the cap of the transport tube, insert the swab into the transport tube and break the swab at the score line. Replace the cap securely making sure the swab fits into the cap and then screw on the cap until it clicks into place.
4. Label the transport tube with the patient's ID number and date of collection.

Male Urethral Swab Specimen Collection

1. Insert the small-tipped, specimen swab 2 to 4 cm into the urethra and rotate the swab for 3-5 seconds to ensure adequate sampling.
2. Verify that all the Swab Specimen Transport Buffer is at the bottom of the tube. If necessary, tap or shake the solution down to the bottom of the tube. Unscrew the cap of the transport tube, insert the swab into the transport tube and break the swab at the score line. Replace the cap securely making sure the swab fits into the cap and then screw on the cap until it clicks into place.
3. Label the transport tube with the patient's ID number and date of collection.

Swab Specimen Transport

1. Swab specimens can be shipped to the laboratory or testing site at 2-30°C or frozen. Swab specimens must arrive at the testing site within 24 hours of shipment or be frozen.
2. Upon receipt in the laboratory or testing site, the swab specimens may be stored at 2-30°C, otherwise store at -20°C or below until processed. Store specimens at 2-30°C if testing is performed within 4 days of collection. If specimens are shipped frozen, maintain them at -20°C or below until testing.
3. All swab specimens stored at -20°C or below must be processed within 60 days of specimen collection.

Urine Specimen Collection and Transport

1. Collect specimen in a plastic, preservative-free, sterile urine specimen collection cup from patients who have not urinated within one hour prior to collection.
2. The patient should collect the first 15-20 mL of voided urine (the first part of the stream).
3. Verify the cup is securely closed and label the collection cup with the patient's ID number and date of collection.
4. Refrigerate the specimen immediately at 2-8°C or store at -20°C or below.

Note: Urine specimens must not be transported or stored at 15-30°C.

Caution: Urine Specimens stored at Room Temperature should not be used for testing.

1. Urine specimens can be shipped to the laboratory or testing site at 2-8°C or frozen. Urine specimens must arrive at the test site within 24 hours of shipment.
2. Upon receipt in the laboratory or testing site, the urine specimen may be stored at 2-8°C or -20°C or below until processed.
3. All urine specimens stored at 2-8°C must be processed within 4 days of specimen collection.
4. All urine specimens stored at -20°C or below must be processed within 60 days of specimen collection. Once frozen specimens should not be thawed until ready for testing.

MATERIALS PROVIDED

[redacted]

A. LCx Gonorrhea Amplification Kit

LCx Gonorrhea Amplification Vials
LCx Gonorrhea Negative Control, Calibrator and Activation Reagent

142

B. LCx® Gonorrhea Detection Reagent Pack

*No. 8A48-87 includes an LCx Gonorrhea Amplification Kit (96 tests), LCx Gonorrhea Detection Reagent Pack (100 tests), and LCx Reaction Cells, List No. 9A48-01 (96 cells). List No. 8A48-81 includes these items for international shipments.

MATERIALS REQUIRED BUT NOT PROVIDED

The LCx Probe System for the LCx *Neisseria gonorrhoeae* Assay consists of one LCx Analyzer, one LCx Thermal Cycler, and one LCx Dry Bath.

COLLECTION SITE

LCx STD Swab Specimen Collection and Transport Kit

(No. 3B15-24 or No. 6C94-24)
100 Collection Systems
Specimen tubes containing 0.5 mL Swab Specimen Transport Buffer

Sterile Urine Specimen Collection Cup

Plastic, preservative-free sterile cup with a secure, screw-top cap capable of holding at least 25 mLs of urine.

AREA 1 (Specimen Preparation Area)

LCx Urine Specimen Preparation Kit

(No. 3B21-24)
100 tests for processing urine specimens

Specimen Tube Racks

Precision Pipettors

100 µL, with aerosol barrier pipette tips (standard length) Swab specimens require extended-length pipette tips (≥75 mm in length)
1.0 mL, with aerosol barrier pipette tips.

Fine-tipped, Single-use, Plastic Disposable Pipette (optional)

X SYSTEMS® Centrifuge (No. 9527) or Equivalent Laboratory Microcentrifuge

(speed ≥9,000 x g) for centrifuging urine specimens and pulse centrifuging amplification vials if required prior to addition of specimens, Calibrator or Controls.

Note: Some centrifuges may require adapters for centrifuging amplification vials.

Abbott LCx Dry Bath

(No. 8B23)
For heat processing of specimen.

Swab Tube Closures

(No. 3B55-30, quantity 500)
For resealing Swab Transport Tubes after processing.

Vortex Mixer

AREA 2 (Amplification and Detection Area)

Abbott LCx Thermal Cycler

(No. 8B24)
Software version 2.1 or higher.

Abbott LCx Analyzer

(No. 9A40)
The LCx Analyzer and accessories used for detection.
LCx System Module, version 1.0 or higher.
LCx Assay Module 1, version 2.0 or higher.
Assay Activation as described in the LCx Analyzer Operations Manual is required to initiate use of the LCx *Neisseria gonorrhoeae* Assay.

LCx Inactivation Diluent (1)

(No. 7B15-04)
2 x 900 mL bottles

LCx System Diluent (2)

(No. 7B14-04)
4 x 1000 mL bottles

X SYSTEMS Centrifuge (No. 9527) or Equivalent Laboratory Microcentrifuge

Note: This must be a separate unit other than the one in Area 1.

(speed ≥9,000 x g) for pulse centrifuging amplification vials before placing into the LCx Reaction Cells.

Note: Some centrifuges may require adapters for centrifuging amplification vials.

SPECIMEN PREPARATION

The use of the LCx Gonorrhea Negative Control and Calibrator is integral to the performance of this LCx assay. These reagents must be prepared in conjunction with specimens to be tested. Refer to the Quality Control Procedures section for details.

All specimen storage and processing must take place in the dedicated Specimen Preparation Area (Area 1).

The LCx Dry Bath will require 20-40 minutes to heat up from a cold start. Confirm the dry bath has reached 97°C (±2°C) before proceeding.

Swab Specimen Preparation

1. Allow specimen to completely thaw if frozen.
2. Insert specimen tubes into wells of preheated dry bath and allow the heat block temperature to stabilize to 97°C(±2°C).
3. After the temperature of the heat blocks is stabilized at 97°C, heat specimens for 15 minutes (±1 minute). Failure to reach 97°C (±2°C) could impair release of the DNA in the specimen and may result in false negative results.
4. Remove specimen from the dry bath and allow to cool at room temperature for 15 minutes (±5 minutes).
5. After cooling, unscrew the cap and express the specimen swab along the inside of the tube so that liquid drains back into the sample solution at the bottom of the tube. The expressed swab and original closure should be discarded, and new Swab Tube Closure (No. 3B55) should be screwed on until it clicks in place.
6. Test the processed swab specimen immediately or store at -20°C or below for up to 90 days. If the processed specimen is stored frozen, it must be completely thawed prior to addition to the LCx Gonorrhea Amplification Vial.
7. Before opening the LCx Gonorrhea Amplification Vials, verify by visual inspection that no liquid is in the cap of the vial. The amplification reagent level should measure approximately two-thirds of the conical part of the vial. If necessary, the vial may be pulse centrifuged in a microcentrifuge for 10-15 seconds.
8. Using a pipettor and extended-length pipette tips (≥75 mm in length) with aerosol barriers, add 100 µL of each processed specimen to the appropriately labeled LCx Gonorrhea Amplification Vial and make sure each vial is securely closed. Transfer the vials to Area 2 and immediately place in the LCx Thermal Cycler for amplification. See LCx Amplification.

Urine Specimen Preparation

1. Allow urine specimen to completely thaw if frozen. Mix urine in the urine collection cup by swirling to resuspend any settled material. It is not necessary for all particulate matter to be fully dissolved.
2. Using a pipettor with aerosol barrier pipette tips, transfer 1 mL of mixed urine into the LCx Urine Specimen Microfuge Tube from the LCx Urine Specimen Preparation Kit (No. 3B21).
3. Centrifuge at ≥9,000 x g for 15 minutes (±2 minutes) in a microcentrifuge.
4. Using a fine-tipped, plastic disposable pipette, gently aspirate the urine supernatant off. **Be cautious not to contact or dislodge the pellet, which may be translucent.** The time between centrifugation and removal of supernatant must not exceed 15 minutes.
5. Using a pipettor with aerosol barrier pipette tips, add 1.0 mL of LCx Urine Specimen Resuspension Buffer. Close the lid of the microfuge tube and resuspend the pellet by vortexing until the pellet is off the bottom of the tube.
6. Secure tube closure with a cap lock until it clicks into place.
7. Insert specimen tubes into wells of preheated dry bath and allow the heat block temperature to stabilize to 97°C (±2°C).
8. After the temperature of the heat blocks is stabilized at 97°C, heat specimens for 15 minutes (±1 minute). Failure to reach 97°C (±2°C) could limit release of the DNA in the specimen and may result in false negative results.
9. Remove the specimen from the dry bath and allow to cool at room temperature for 15 minutes (±5 minutes). Remove cap lock and discard.
10. Pulse centrifuge the processed urine specimen in a microcentrifuge for 10-15 seconds.

/ 93

11. Test the processed urine specimen immediately, or store for up to 60 days at -20°C or below prior to testing. The processed urine specimen must be completely thawed prior to addition to the LCx® Gonorrhea Amplification Vial.
12. Before opening the LCx Gonorrhea Amplification Vials, verify by visual inspection that no liquid is in the cap of the vial. The amplification reagent level should measure approximately two-thirds of the conical part of the vial. If necessary, the vial may be pulse centrifuged in a microcentrifuge for 10-15 seconds.
13. Using a pipettor with aerosol barrier pipette tips, add 100 µL of each processed urine specimen to the appropriately labeled LCx Gonorrhea Amplification Vial and make sure each vial is securely closed. Transfer the vials to Area 2 and immediately place in the LCx Thermal Cycler for amplification. See procedure under LCx Amplification.

PROCEDURE

Procedural Precautions

1. Work in a laboratory using DNA amplification methods should always flow in a one-way direction beginning in the Specimen Preparation Area (Area 1), then moving to the Amplification and Detection Area (Area 2). Do not bring any materials from Area 2 into Area 1.
2. **Surface cleaning using a 1% (v/v) sodium hypochlorite solution followed by 70% (v/v) ethanol should be performed on bench tops and pipettors at least once per day prior to beginning an LCx Assay.**

   Note: Chlorine solutions may pit equipment and metal. Use sufficient amounts or repeated applications of 70% ethanol until chlorine residue is no longer visible.
3. Monthly monitoring procedures for the presence of DNA can be found in the Quality Control Section of this package insert. In addition, if the LCx Negative Control consistently fails the NEG HIGH or NEG AVE HIGH specifications for the LCx *Neisseria gonorrhoeae* Assay Parameters, laboratory contamination may be suspect. If this occurs contact LCx CSC.
4. The LCx *Neisseria gonorrhoeae* Assay is designed to be detected only on an Abbott LCx Analyzer.
5. All plastic materials coming into contact with the specimen should be free of any residue from previous specimens, reagents, or cleaning compounds.
6. During the sample (specimens, Calibrators or Negative Controls) addition step, only one LCx Gonorrhea Amplification Vial should be open at any given time. After this step, the vials should remain closed throughout the thermal cycling and detection procedures. This aids in the prevention of cross-contamination.
7. Only one bottle of Negative Control or Calibrator should be open at any one time.

LCx Amplification

1. Refer to the LCx Thermal Cycler Operations Manual for detailed instructions on thermal cycler operation. Turn the LCx Thermal Cycler on for at least 15 minutes prior to use.
2. Collect all LCx Gonorrhea Amplification Vials containing samples, Negative Control and Calibrator from Area 1 and transfer to Area 2 for thermal cycling.
3. LCx thermal cycling conditions should be edited to the following amplification parameters described below.

   Assay Step-Cycle File:

Segment 1	93°C for 1 second
Segment 2	59°C for 1 second
Segment 3	62°C for 1 minute 10 seconds
Cycle count	40 cycles

   The amplification run time is approximately 2 hours.

   The Assay Step-Cycle File is "Linked to" the Soak File at 25°C, indefinitely.
4. Place the amplification vials into the thermal cycler, and initiate run. After completion of the thermal cycler run, amplification product may remain at 15-30°C for up to 72 hours prior to LCx detection.

LCx Detection and Inactivation of Amplification Product

LCx *Neisseria gonorrhoeae* Assay Parameters

The following LCx *Neisseria gonorrhoeae* Assay parameters have been factory set in the Assay Module and may not be edited. These parameters can be printed and displayed according to the procedure in your LCx Analyzer Operations Manual, Section 5.

Assay 12 Gonorrhea LCR

3.	SAMPLE REP	1
32.	MAX NRM	0.500
33.	MIN CORR	0.950
34.	MAX INTRCPT	12000.0
45.	CAL HIGH	2230.00
46.	CAL LOW	370.00
47.	CAL AVE HIGH	2000.00
48.	CAL AVE LOW	600.00
54.	NUM POS CNTL	0
55.	NUM NEG CNTL	2
61.	% CUT OFF	25.000
83.	NUM CAL	2
85.	NEG HIGH	145.00
86.	NEG LOW	0.00
87.	NEG AVE HIGH	100.00
88.	NEG AVE LOW	0.00

1. Refer to your LCx Analyzer Operations Manual for detailed instrument operation procedures. Before running the LCx Analyzer, check to see that LCx Inactivation Diluent (1) contains a minimum of 100 mL and the LCx System Diluent (2) contains a minimum of 250 mL.
2. Remove the LCx Gonorrhea Amplification Vials from the LCx Thermal Cycler.
3. Place LCx Reaction Cells into an MEIA Carousel; lock the carousel.
4. Pulse centrifuge the LCx Gonorrhea Amplification Vials in a microcentrifuge for 10-15 seconds before placing into the LCx Reaction Cells.
5. Place the amplification vials into the LCx Reaction Cells in the following order: Negative Controls in positions 1 and 2, Calibrators in positions 3 and 4, and specimens in the remaining positions.
6. Place the carousel into the LCx Analyzer.
7. Lock the Amplification Vial Retainer by turning the handle counterclockwise.
8. Remove the LCx Gonorrhea Detection Reagent Pack from 2-8°C storage, gently invert it 5 times, and open the reagent pack bottles in the numeric order: 1, 2, 3, 4.
9. Look for any film that may have formed over the openings of the reagent bottles. If present, remove using a long, clean pipette tip or a wooden applicator stick for each bottle.
10. Place the LCx Gonorrhea Detection Reagent Pack into the LCx Analyzer.
11. Press RUN on the LCx Analyzer control panel. Final assay results will be printed in approximately 60 minutes.
12. Remove the assay results printout from the LCx Analyzer.
13. After completion of the detection procedure, remove the LCx Gonorrhea Detection Reagent Pack, and close the caps in the numeric order: 4,3,2,1. Store the detection reagent pack at 2-8°C.
14. Unlock the Amplification Vial Retainer by turning the handle clockwise until it is no longer over the MEIA carousel.
15. Remove the MEIA Carousel, individually remove the LCx Reaction Cells, and dispose appropriately.
16. Review the assay results and record patient results.

QUALITY CONTROL PROCEDURES

Negative Control and Calibrator Preparation

All Calibrator and Negative Control preparation must take place in the dedicated Specimen Preparation Area (Area 1).

1. The LCx *Neisseria gonorrhoeae* Assay procedure requires that the LCx Gonorrhea Negative Control and the Calibrator be run in duplicate with each MEIA carousel of clinical specimens.

144

2. The LCx® Gonorrhea Negative Control and Calibrator are activated by the addition of 100 µL of LCx Gonorrhea Activation Reagent. After addition, the contents of the bottles are then recapped and vortexed for 2-5 seconds. Use activated Negative Control and Calibrator immediately or store at 2-8°C for up to 8 hours.

Not adding activation reagent or adding incorrect volumes of activation reagent may give erroneous results and the run may be invalid.

3. Before opening the LCx Gonorrhea Amplification Vials, verify by visual inspection that no liquid is in the cap of the vial. The amplification reagent level should measure approximately two-thirds of the conical part of the vial. If necessary, the vials may be pulse centrifuged in a microcentrifuge for 10-15 seconds.
4. Using a pipettor with aerosol barrier pipette tips, add 100 µL each of the activated LCx Gonorrhea Negative Control and Calibrator to the appropriately labeled LCx Gonorrhea Amplification Vials and make sure each vial is securely closed. Transfer the vials to Area 2 and immediately place in the LCx Thermal Cycler for amplification. See LCx Amplification.

Positive Control

A positive control that monitors the entire assay procedure including the specimen processing step should be tested in accordance with requirements of appropriate accrediting organizations. For such a control, a known positive urine specimen can be processed in parallel and tested with unknown specimens. Specimens used as processing controls must be stored per the Specimen Collection and Transport to Test Site section in this assay package insert.

Alternatively, *N. gonorrhoeae* cells (available from ATCC, Catalogue No.27631) may be subcultured or an existing subculture of *N. gonorrhoeae* may be used as a positive control.

Preparing the Cell Stock and the Positive Control

1. Thaw lyophilized stock, or vial of stock culture at room temperature and inoculate a chocolate agar plate within 30 minutes of thawing cells.
2. Incubate the inoculated plate at 37°C with 5% CO_2 for 24 to 36 hours until growth is noted.
3. Suspend and serially dilute cells in phosphate buffered saline to determine CFU/mL by routine laboratory procedures.
4. Dilute cell stock to approximately 1×10^7 CFU/mL in trypticase soy broth containing 20% glycerol. To store stock, aliquot in 100 µL to 500 µL volumes and keep at -20°C or below.
5. Preparing the Positive Control: Serially dilute cell stock to approximately 1×10^3 CFU/mL in LCx Urine Resuspension Buffer. Aliquot 1.0 mL volume in LCx Urine Specimen Microfuge Tubes and use or store at -20°C or below.
6. Thaw the aliquot, if frozen. Heat the aliquot in the LCx Dry Bath at 97°C (±2°C) for 15 minutes (±1 minute). Note: **Place Cap Locks on the microfuge tubes before heating. Allow tubes to cool to room temperature before handling.**
7. Perform amplification and detection in parallel with unknown specimens.

The positive control should give a positive assay value (S/CO ratio ≥1.00). Each laboratory should establish a target value and limits for each control batch using statistical control rules. (See NCCLS C-24A.) These target values and limits should be maintained throughout storage.

Assay Validity

Validity of the LCx Gonorrhea Negative Control and Calibrator assay results are automatically assessed by the LCx Analyzer before proceeding to analyze specimen assay results.

The LCx Analyzer first verifies that the assay results of the Negative Control and Calibrator are within the specified ranges of the LCx *Neisseria gonorrhoeae* Assay Parameters by comparing the assay results of the Negative Control and Calibrator to the values listed in the assay parameters. A run is valid when the individual and average results are within the values listed for CAL HIGH, CAL LOW, CAL AVE HIGH, CAL AVE LOW, NEG LOW, NEG HIGH, NEG AVE HIGH, and NEG AVE LOW parameters in the LCx *Neisseria gonorrhoeae* Assay Parameters.

In the event of an invalid Negative Control or Calibrator assay result, the assay results printout will identify the out-of-range result, the S/CO ratio of the specimens will NOT be calculated and a flag indicating an invalid result will occur in the NOTE column of the printout next to the specimen assay results. If an out-of-range result is identified on the printout, refer to the LCx Analyzer Operations Manual, Section 10: Troubleshooting and Diagnostics for an explanation of the error message. Instructions for troubleshooting the detection portion of the assay can also be found under General Troubleshooting Procedures, LCx MEIA Performance Troubleshooting.

The LCx Analyzer does not calculate imprecision between Negative Control or Calibrator replicate values.

Note: Ensure the LCx Negative Controls and Calibrators are in the correct order on the MEIA carousel to avoid an invalid run.

If an amplification vial opens during thermal cycling, the sample is invalid and should not be used. Make sure that the amplification vial caps are tightly closed. Remove carefully to a biohazard bag and seal the bag. Dispose of according to procedure of waste disposal in the LCx Thermal Cycler Operations Manual Section 8: Hazards, Biosafety.

Displayed and Printed Error Codes

If a displayed or printed error code appears, refer to the LCx Analyzer Operations Manual, Section 10.

Monitoring the Laboratory for the Presence of Amplification Product

It is recommended that this test be done at least once a month to monitor laboratory surfaces and equipment for contamination by amplification product.

Using the small-tipped specimen swab from an LCx STD Swab Specimen Collection System, insert the swab into the tube of the Swab Specimen Transport Buffer. Allow the tip to become saturated with the buffer. When the tip is saturated, remove from the container and wipe the desired area using a broad sweeping motion. Replace the swab into the transport tube and break at the scored mark. Process following the LCx *Neisseria gonorrhoeae* Assay procedure. It is very important to be sure to test all areas that may have been exposed to samples and/or amplification product. This includes pipettors, pipettor handles, LCx Analyzer function keys, LCx Thermal Cycler function keys, bench surfaces, microcentrifuges and centrifuge adapters. If positive results (S/CO ratio ≥1.00) on surfaces are observed, clean the contaminated areas with 1% (v/v) sodium hypochlorite solution, followed by 70% (v/v) ethanol. Follow the appropriate operations manual for cleaning and decontamination equipment if positive results are observed. Note: **Chlorine solutions may pit equipment and metal. Use sufficient amounts or repeated applications of 70% ethanol until chlorine residue is no longer visible.** Repeat this cleaning procedure until the results are negative (S/CO ratio <1.00).

RESULTS

CALCULATIONS

The LCx *Neisseria gonorrhoeae* Assay uses MEIA detection on the LCx Analyzer to detect *N. gonorrhoeae* DNA. All calculations are performed automatically.

The presence or absence of *N. gonorrhoeae* is determined by relating the LCx Assay results for the specimen to the Cutoff value. The Cutoff value is the mean RATE (c/s/s) of the LCx calibrator duplicates multiplied by 0.25.

Calculation of the Cutoff value:

Cutoff value = 0.25 x (Mean of LCx Gonorrhea Calibrator RATES)

The S/CO value is determined by calculating a ratio of the sample RATE to the Cutoff value.

$$\frac{S}{CO} = \frac{\text{Sample RATE}}{\text{Cutoff Value}}$$

145

INTERPRETATION OF RESULTS

Patient specimens results should be interpreted and reported as follows:

S/CO Ratio	Result	Report
≥1.20	LCx® Positive	*N. gonorrhoeae* DNA detected, and positive for *N. gonorrhoeae* by LCR™ amplification and MEIA detection.
<0.80	LCx Negative	*N. gonorrhoeae* DNA not detected, and presumptively negative for *N. gonorrhoeae* by LCR amplification and MEIA detection. Specimen may not contain *N. gonorrhoeae* or may be negative due to inhibitors in the specimen.
0.80-1.20	LCx Equivocal	Repeat LCx test. If repeat test S/CO ratio is greater than or equal to 1.00, *N. gonorrhoeae* DNA detected, and positive for *N. gonorrhoeae* by LCR amplification and MEIA detection. If the repeat test is less than S/CO ratio 1.00, *N. gonorrhoeae* DNA not detected and presumptively negative for *N. gonorrhoeae* by LCR amplification and MEIA detection.

Note: Check the printout for initial results with S/CO values of 0.80-1.20 which require repeat testing.

LIMITATIONS OF THE PROCEDURE

1. Optimal performance of this test requires adequate specimen collection (sampling columnar epithelial cells) and handling (see Specimen Collection section). The assay should be performed only on swab samples from the endocervix and male urethra or on urine from males and females. The use of specimens other than those listed has not been validated. Specimen adequacy can only be assessed by microscopic visualization of columnar epithelial cells in the swab specimens.
2. A negative result does not exclude the possibility of infection because results are dependent on adequate specimen collection and absence of inhibitors. The presence of LCR inhibitors may cause false negative results with this product. The effect of freezing samples on inhibition or on the ability to detect low levels of organisms has not been determined. Results from the LCx *N. gonorrhoeae* Assay should be interpreted in conjunction with other laboratory and clinical data available to the clinician.
3. A positive result for the presence of *N. gonorrhoeae* nucleic acids in the endocervical specimens does not establish the causative agent for salpingitis or PID. A negative result for the *N. gonorrhoeae* nucleic acids in the endocervical specimen does not exclude gonococcal infection as a cause of ascending infection.
4. The LCx *Neisseria gonorrhoeae* Assay is not intended to replace culture and other methods for diagnosis of gonococcal infection. Symptomatic patients may have cervicitis, urinary tract infections, or vaginal infections due to other causes or concurrent infections with other agents. Assessment of antimicrobial susceptibility, auxotype or serotype is dependent on culture isolation.
5. Use of the LCx *Neisseria gonorrhoeae* Assay is limited to personnel who have been trained in the procedures of an LCx Assay and the LCx Analyzer.
6. The LCx Inactivation procedure reduces the risk of contamination by amplification product. However, DNA contamination from the Calibrator or clinical specimens must be controlled by good laboratory practice and careful adherence to the procedures specified in the package insert.
7. Therapeutic success or failure cannot be determined as *N. gonorrhoeae* nucleic acids may persist following appropriate antimicrobial therapy.
8. The LCx *Neisseria gonorrhoeae* Assay should not be used for the evaluation of suspected sexual abuse or for other medico-legal indications. Verification by an alternate method is recommended when results may have psycho-social impact.
9. The LCx *Neisseria gonorrhoeae* Assay provides qualitative results. No correlation can be drawn between the magnitude of a positive LCx *Neisseria gonorrhoeae* Assay signal and the number of *Neisseria gonorrhoeae* cells within an infected specimen.
10. Some spermicidal agents and feminine powder sprays interfere with the assay and should therefore not have been used prior to collection of specimens for the assay.
11. Swab or urine specimens that are moderate bloody (greater than approximately 0.5% (v/v)) should not be tested since they may cause inhibition in the LCx *Neisseria gonorrhoeae* Assay.
12. Swab or urine specimens that are grossly mucoid (greater than approximately 5% (w/v)) should not be tested since they may cause inhibition in the LCx *Neisseria gonorrhoeae* Assay. Therefore, it is important that the exocervix be wiped free of mucus prior to collection of the swab specimen to insure optimal specimen condition.
13. The LCx *Neisseria gonorrhoeae* Assay for male and female urine testing must be performed on first catch urine specimens (defined as the first 15-20 mL of the urine stream). The effects of other variables such as first-catch vs. mid-stream, post douching, etc. have not been determined.
14. The effects of other potential variables such as vaginal discharge, use of tampons, douching, etc., and specimen collection variables have not been determined.
15. The predictive value of an assay will depend on disease prevalence in any particular population. See Table 2 for calculated predictive values at different hypothetical prevalence rates that were derived from LCx sensitivity and specificity results.

EXPECTED VALUES

The LCx *Neisseria gonorrhoeae* Assay was tested on a total of 3362 specimens obtained from patients attending sexually transmitted disease clinics or OB/GYN clinics. For endocervix and male urethra testing, one swab specimen was taken to streak the culture plate and then the same swab specimen was used for the LCx assay. For urine testing, a urine specimen was collected after the endocervical or male urethral swab was taken. LCx Assay urine results were compared to the endocervical or male urethral culture. Discrepant specimen pairs with culture-negative and LCx-positive results were re-analyzed by LCR using another *N. gonorrhoeae* specific probe set targeting the Pilin gene.

Prevalence

The prevalence of positive *N. gonorrhoeae* specimen results in patient populations varies depending on population characteristics such as age, sex and risk factors, and can vary depending on testing methodology. During clinical trials, prevalence using the LCx *Neisseria gonorrhoeae* Assay was observed to range from 3.5% to 35.6% as shown in Table 1.

Table 1
Prevalence of LCx Positive Results by Sample Type and Test Site

Sample Type	Test Site	No. Pos/ No. Tested	% PREV
Female Endocervical	1	28/142	19.7
	2	71/492	14.4
	3*	13/189	6.9
	4	12/342	3.5
Female Urine	1	22/119	18.5
	2	52/340	15.3
	3*	21/121	17.4
Male Urethral	1	73/205	35.6
	2	128/429	29.8
	3*	17/116	14.7
Male Urine	1	57/163	35.0
	2	168/590	28.5
	3*	11/114	9.6

*In-house testing of specimens collected from sites 1, 2 and 4.

Positive and Negative Predictive Values

The calculated positive and negative predictive values (PPV and NPV) for different hypothetical prevalence rates using overall LCx sensitivity and specificity of 97.5% (626/642) and 98.3% (2673/2720), respectively, are shown in Table 2.

146

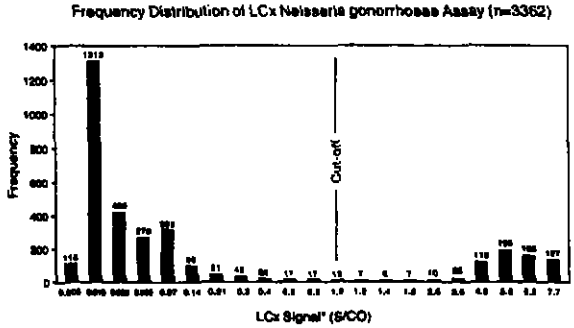
Table 2

LCx® *Neisseria gonorrhoeae* Assay Calculated Predictive Values at Hypothetical Prevalence Rates

Prevalence Rate (%)	Sensitivity (%)	Specificity (%)	Positive Predictive Value (%)	Negative Predictive Value (%)
5	97.5	98.3	75.1	99.9
10	97.5	98.3	86.4	99.7
15	97.5	98.3	91.0	99.6
20	97.5	98.3	93.5	99.4

Frequency Distribution

A total of 3362 male and female specimens were assayed at four clinical sites. The frequency of S/CO values (sample c/s/s to cutoff c/s/s) of the specimens is illustrated in Figure 1. The distribution of S/CO values is as follows: 79.54% of the S/CO values were <0.80, 19.78% were ≥1.20, and 0.68% were between 0.80 and 1.20. Six of the 23 specimens with S/CO values between 0.80 and 1.20 were culture positive. Of the 17 remaining culture negative specimens, 13 were tested by LCR probe set targeting the Pilin gene and seven were positive by this LCR test.



*Value under each column is the midpoint LCx signal for that cell.

SPECIFIC PERFORMANCE CHARACTERISTICS

The performance characteristics of the LCx *Neisseria gonorrhoeae* Assay established during clinical trials were generated on an earlier version of the LCx instrument. Reproducibility studies, however, were generated using the current version of the LCx instrument. Performance characteristics were determined by comparing LCx assay results to culture results for *N. gonorrhoeae*.

The results of the clinical studies are shown in Tables 3A, 3B, and 3C. For the clinical specimens tested, there were 642 (486 frozen and 156 fresh) that were culture positive. Of the 642 culture positive specimens, the LCx assay was negative for 16 (12 frozen and 4 fresh). The reason for these results could be inhibition, collection variables, or other factors. The LCx assay was positive for an additional 47 specimens that were culture negative (39 frozen and 8 fresh), 41 of which (33 frozen and 8 fresh) were positive by an alternate target test (specimens with LCx positive/culture negative discordant results were tested by LCR with a different probe set targeting the Pilin gene region of *N. gonorrhoeae*). There were 15 specimens with S/CO values between 0.80 and 0.99. Twelve of the 15 were culture negative. Eight of the 12 culture negative specimens were tested by LCR Pilin assay and 4 were positive by this test.

Specificity

The following list identifies the bacteria, parasites, viruses, yeast and fungi that have been tested in the LCx *Neisseria gonorrhoeae* Assay. All isolates were tested using the equivalent of at least 10^7 copies of genomic DNA per test except as indicated. The tested organisms include those that are commonly found in the urogenital tract as well as others. All gave negative values.

Acinetobacter calcoaceticus
Actinomyces israelii
Adenovirus*
Aeromonas hydrophila
Alcaligenes faecalis
Bacillus subtilis
Bacillus thuringiensis
Bacteroides fragilis
Bifidobacterium longum
Branhamella catarrhalis
Candida albicans
Candida glabrata
Chlamydia trachomatis
Citrobacter freundii
Clostridium sporogenes
Corynebacterium hoffmanni
Corynebacterium renale
Cryptococcus laurentii
Cytomegalovirus*
Edwardsiella tarda
Enterobacter aerogenes
Enterobacter cloacae
Enterococcus faecalis
Enterococcus faecium
Epstein-Barr Virus*
Escherichia coli
Ewingella americana
Flavobacterium odoratum
Fusobacterium nucleatum
Gardnerella vaginalis
Haemophilus influenzae
Haemophilis parainfluenzae
Hafnia alvei
Helicobacter pylori
Herella vaginicola
Hepatitis B Virus*
Herpes Simplex Virus I*
Herpes Simplex Virus II*
Human Herpes Virus*
Human Immunodeficiency Virus type 1*
Human Papilloma Virus 16*
Human Papilloma Virus 18*
Human T-Cell Lymphotropic Virus type I*
Klebsiella pneumoniae
Lactobacillus casei
Lactobacillus plantarum
Mima polymorpha
Moraxella lacunata
Morganella morganii
Moraxella osloensis
Mycobacterium avium
Mycobacterium gordonae
Mycoplasma hominis
Neisseria catarrhalis
Neisseria cinerea
Neisseria elongata
Neisseria flava
Neisseria flavescens
Neisseria lactamica
Neisseria meningitidis
Neisseria mucosa
Neisseria perflava
Neisseria polysaccharea
Neisseria sicca
Neisseria subflava
Peptostreptococcus productus
Plesiomonas shigelloides
Propionibacterium acnes
Proteus mirabilis
Proteus vulgaris
Providencia stuartii
Pseudomonas aeruginosa
Saccharomyces cerevisiae
Salmonella enteritidis
Salmonella minnesota
Salmonella typhimurium
Serratia marcescens
*Shistosoma haematobium**
Shigella sonnei
Staphylococcus aureus
Staphylococcus epidermidis
Streptococcus agalactiae
Streptococcus faecalis
Streptococcus mutans
Streptococcus pyogenes
Treponema pallidum
*Trichomonas vaginalis**
Ureaplasma urealyticum
Varicella-zoster Virus*
Veillonella parvula
Yersinia enterocolitica

* Testing using equivalent of 10^6 copies of genomic DNA per test.

Analytical Sensitivity

The analytical sensitivity of this assay (limit of detection) is 10 Colony Forming Units (CFU) of any of the 6 auxotrophs of *Neisseria gonorrhoeae*. The analytical sensitivity of this assay was determined by a serial dilution study on all 6 auxotrophs (auxotype 1, 5, 9, 12, 16, and 22). Each auxotroph was diluted to less than 10 CFU per reaction and tested in the LCx *Neisseria gonorrhoeae* Assay. In all cases, each replicate of a dilution giving 10 CFU per test (100 µL specimen volume) was positive by the LCx *Neisseria gonorrhoeae* Assay.

Inactivation Efficiency of Amplification Product

A 10^7 fold reduction of the amplification product concentration is achieved in overamplified samples by the inactivation chemistry used in the LCx *Neisseria gonorrhoeae* Assay. For clinical specimens, 40 positive urine and swab specimens were used to evaluate the inactivation efficiency. Amplified specimens were diluted 100-fold and then inactivated using the LCx assay procedure before re-testing with the LCx *Neisseria gonorrhoeae* Assay. All specimens showed negative results (S/CO ratio <1.00) after inactivation.

Reproducibility

Assay reproducibility of the LCx *Neisseria gonorrhoeae* Assay was demonstrated by testing a 5-member panel consisting of 4 dilutions of *N. gonorrhoeae* in a specimen matrix. This reproducibility panel was run at three different sites in triplicate twice a day for three days. The clinical trials were all run in laboratories with a high degree of expertise in DNA amplification assays. It was important to demonstrate the reproducibility obtained by technicians who had no prior experience with this type of assay. Therefore, the technicians at two of the sites in which the reproducibility panel was run had been instructed in the LCx *Neisseria gonorrhoeae* Assay but had not previously run DNA amplification assays. The results of the average within-run, between run and between day and total reproducibility of the LCx *Neisseria gonorrhoeae* Assay are shown in Table 4. The variance estimates presented in Table 4 were calculated from the variance components obtained using SAS's® PROC NESTED procedure. Variance estimates are cumulative from left to right, i.e., "Within Run" variability is included in the "Between Run" term, etc.

147

Coefficients of Variation (CV) for Panel Member A and the Negative Control are not presented, instead the Standard Deviation (SD) is presented as the measure of variability. Results are presented according to the National Committee for Clinical Laboratory Standards (NCCLS).

Negative Control and Calibrator Performance

The performance of the Calibrator used during the clinical trials was evaluated by calculating the overall mean and the percent Coefficient of Variation (%CV) across the valid runs. (See Table 5). The overall %CV was calculated without regard to replicates. The average within run %CV is the %CV of each Calibrator replicate averaged over 212 runs. For the Negative Control, the overall Standard Deviation (SD) is presented as the measure of variability. These results are similar to the results obtained in Table 4, Reproducibility.

Proficiency

In order to determine the proficiency of the LCx® Neisseria gonorrhoeae Assay, a 24-member panel consisting of 24 unique samples at various LCx signals in urine and swab samples were tested. This panel was run at three different sites using three separate instruments and each sample was analyzed once. The technicians at all sites were inexperienced users and received training on the LCx Assay just prior to beginning the proficiency study. The results of this study demonstrated 100% agreement on this panel across all three test sites.

Table 3A
Performance Summary Compared to Culture: Frozen Specimens

Sample Type	Symptomatology	Total	LCx	Culture	Pos Pos	Pos Neg	Neg Pos	Neg Neg	Sensitivity (95% C.I.)	Specificity (95% C.I.)
Female Endocervical	Asymptomatic	623			24	11	3	585	88.9% (24/27) (70.8-97.6)	98.2% (585/596) (96.7-99.1)
	Symptomatic	332			47	8	0	277	100.0% (47/47) (92.5-100.0)	97.2% (277/285) (94.5-98.8)
Female Urine *	Asymptomatic	140			23	1	2	114	92.0% (23/25) (74.0-99.0)	99.1% (114/115) (95.3-100.0)
	Symptomatic	237			43	0	2	192	95.6% (43/45) (84.9-99.5)	100.0% (192/192) (98.1-100.0)
Male Urethral	Asymptomatic	172			6	2	1	163	85.7% (6/7) (42.1-99.6)	98.8% (163/165) (95.7-99.9)
	Symptomatic	412			155	6	2	249	98.7% (155/157) (95.5-99.8)	97.6% (249/255) (94.9-99.1)
Male Urine *	Asymptomatic	234			6	1	1	226	85.7% (6/7) (42.1-99.6)	99.6% (226/227) (97.6-100.0)
	Symptomatic	466			170	10	1	285	99.4% (170/171) (96.8-100.0)	96.6% (285/295) (93.9-98.4)
Total	Asymptomatic	1,169			59	15	7	1,088	89.4% (59/66) (79.4-95.6)	98.6% (1088/1103) (97.8-99.2)
	Symptomatic	1,447			415	24	5	1,003	98.8% (415/420) (97.2-99.6)	97.7% (1003/1027) (96.5-98.5)

Table 3B
Performance Summary Compared to Culture by Site: Frozen Specimens

Sample Type	Site	Total	LCx	Culture	Pos Pos	Pos Neg	Neg Pos	Neg Neg	Sensitivity (95% C.I.)	Specificity (95% C.I.)
Female Endocervical	Site 1	142			25	3	0	114	100.0% (25/25) (86.3-100.0)	97.4% (114/117) (92.7-99.5)
	Site 2	282			30	7	2	243	93.8% (30/32) (79.2-99.2)	97.2 (243/250) (94.3-98.9)
	Site 3 (In-house)	189			9	4	1	175	90.0% (9/10) (55.5-99.7)	97.8 (175/179) (94.4-99.4)
	Site 4	342			7	5	0	330	100.0% (7/7) (59.0-100.0)	98.5 (330/335) (96.6-99.5)
Female Urine *	Site 1	119			21	1	1	96	95.5% (21/22) (77.2-99.9)	99.0% (96/97) (94.4-100.0)
	Site 2	137			24	0	2	111	92.3% (24/26) (74.9-99.1)	100.0% (111/111) (96.7-100.0)
	Site 3 (In-house)	121			21	0	1	99	95.5% (21/22) (77.2-99.9)	100.0% (99/99) (96.3-100.0)
Male Urethral	Site 1	205			70	3	0	132	100.0% (70/70) (94.9-100.0)	97.8% (132/135) (93.6-99.5)
	Site 2	263			76	3	2	182	97.4% (76/78) (91.0-99.7)	98.4% (182/185) (95.3-99.7)
	Site 3 (In-house)	116			15	2	1	98	93.8% (15/16) (69.8-99.8)	98.0% (98/100) (93.0-99.8)
Male Urine *	Site 1	163			54	3	0	106	100.0% (54/54) (93.4-100.0)	97.2% (106/109) (92.2-99.4)
	Site 2	423			114	5	0	304	100.0% (114/114) (96.8-100.0)	98.4% (304/309) (96.3-99.5)
	Site 3 (In-house)	114			8	3	2	101	80.0% (8/10) (44.4-97.5)	97.1% (101/104) (91.8-99.4)

* Comparison cultures for these specimens were performed on endocervical and male urethral swab specimens, respectively.

148

Table 3C
Performance Summary Compared to Culture: Fresh Specimens (Site 2)

Sample Type	Symptomatology	Total	LCx® Culture	Pos Pos	Pos Neg	Neg Pos	Neg Neg	Sensitivity (95% C.I.)	Specificity (95% C.I.)
Female Endocervical	Asymptomatic	78		10	3	0	65	100.0% (10/10) (69.2-100.0)	95.6% (65/68) (87.6-99.1)
	Symptomatic	132		20	1	0	111	100.0% (20/20) (83.2-100.0)	99.1% (111/112) (95.1-100.0)
Female Urine *	Asymptomatic	74		9	1	0	64	100.0% (9/9) (66.4-100.0)	98.5% (64/65) (91.7-100.0)
	Symptomatic	129		17	1	3	108	85.0% (17/20) (62.1-96.8)	99.1% (108/109) (95.0-100.0)
Male Urethral	Asymptomatic	74		1	0	0	73	100.0% (1/1) (2.5-100.0)	100.0% (73/73) (95.1-100.0)
	Symptomatic	92		47	1	0	44	100.0% (47/47) (92.5-100.0)	97.8% (44/45) (88.2-99.9)
Male Urine *	Asymptomatic	74		1	0	0	73	100.0% (1/1) (2.5-100.0)	100.0% (73/73) (95.1-100.0)
	Symptomatic	93		47	1	1	44	97.9% (47/48) (88.9-99.9)	97.8% (44/45) (88.2-99.9)
Total	Asymptomatic	300		21	4	0	275	100.0% (21/21) (83.9-100.0)	98.6% (275/279) (96.4-99.6)
	Symptomatic	446		131	4	4	307	97.0% (131/135) (92.6-99.2)	98.7% (307/311) (96.7-99.6)

Although it was not included in the analysis described in Tables 3A, 3B and 3C, discrepant specimen pairs with culture-negative and LCx-positive results were re-analyzed by an LCx assay using an alternate *N. gonorrhoeae* probe set targeting the Pilin gene. Of the 47 LCx-positive, culture-negative specimen pairs, 41 were found to be positive with the Pilin assay including 18/23 endocervical specimens, 3/3 female urine specimens, 9/9 male urethral specimens, and 11/12 male urine specimens. These data indicate that these apparent false positive specimens, may have been, in fact, true positives that were missed by culture. The LCx Assay was negative for 16 of 642 culture positive specimens. The reason for these results could be inhibition, collection variables or other factors.

* Comparison cultures for these specimens were performed on endocervical and male urethral swab specimens, respectively.

Table 4
Reproducibility

Panel Member	N	Mean S/CO	Within Run (SD)	Within Run (%CV)	Between Run (SD)	Between Run (%CV)	Between Day (SD)	Between Day (%CV)	Total (SD)	Total (%CV)
A	54	0.02	0.004	NA	0.004	NA	0.004	NA	0.006	NA
B	54	2.01	0.293	14.5	0.332	16.5	0.461	22.9	0.576	28.6
C	54	4.07	0.221	5.4	0.397	9.8	0.397	9.8	0.638	15.7
D	54	5.33	0.129	2.4	0.300	5.6	0.431	8.1	0.581	10.9
E	54	5.77	0.220	3.8	0.389	6.7	0.603	10.5	0.675	11.7

Panel Member	N	Mean RATE	Within Run (SD)	Within Run (%CV)	Between Run (SD)	Between Run (%CV)	Between Day (SD)	Between Day (%CV)	Total (SD)	Total (%CV)
Negative	54	6.8	2.73	NA	2.73	NA	2.78	NA	2.92	NA
Calibrator	54	1292.9	60.06	4.6	115.05	8.9	161.40	12.5	161.40	12.5

NA = Not Applicable

Table 5
Reproducibility of LCx Gonorrhea
Negative Control and Calibrator Values (212 Runs)

Sample	Calibrator	Negative Control
Mean (c/s/s)	1171.5	8.6
SD	219.8	6.3
%CV		
Average Within Run	5.5	NA
Overall	18.8	NA
Range (c/s/s)	565.1 - 1589.5	3.8 - 66.8

BIBLIOGRAPHY

1. Bovre K: Family VIII. Neisseriaceae Prevot 1933; 119. Krieg, N.R. and Holt, J.G. (Eds): Bergey's Manual of Systematic Bacteriology. Williams and Wilkins, Baltimore, 1984; 288-296.
2. Morello JA, Janda WM, and Doern GV: Neisseria and Branhamella *Manual of Clinical Microbiology* 1991; Balows A, Hausler WJ, and Herrmann KL. (Eds), 258-261.
3. Hale YM, Melton ME, Lewis JS, and Willis DE: Evaluation of the PACE 2 *Neisseria gonorrhoeae* assay by three public health laboratories. *Journal of Clinical Microbiology* 1993; 31: 451-453.
4. Burczak JD, Ching SF, Hu HY and Lee H: Ligase Chain Reaction for the Detection of Infectious Agents. *Molecular Methods for Virus Detection* 1995; Wiebrauk D, and Farkas DH (Eds); Academic Press Inc, 315-328.

149

5. Stern A, Brown M, Nickel P, and Meyer TF: Opacity genes in *Neisseria gonorrhoeae*: Control of phase and antigenic variation. *Cell* 1986; 47: 61-71.
6. Meyer TF, Gibbs CP, and Haas R: Variation and control of protein expression in Neisseria. *Annual Review of Microbiology* 1990; 44: 451-477.
7. Fiore M, Mitchell J, Doan T, Nelson R, Winter G, et al.: The Abbott IMx automated benchtop immunochemistry analyzer system. *Clinical Chemistry* 1988; 34: 1726-1732.
8. US Department of Labor, Occupational Safety and Health Administration, 29 CFR Part 1910.1030, *Occupational Exposure to Bloodborne Pathogens*, final rule. Fed Register, 1991; 56:No. 235;64175-82.
9. U.S. Department of Health and Human Services, Biosafety in Microbiological and Biomedical Laboratories. *HHS Publication number* (CDC) 93-8395.
10. World Health Organization. Laboratory Biosafety Manual. Geneva, World Health Organization, 1993.
11. National Committee for Clinical Laboratory Standards. Protection of Laboratory Workers for Infectious Disease Transmitted by Blood, Body Fluids, and Tissue: Tentative Guidelines. NCCLS Document M29-T2. Villanova, Pa: NCCLS, 1991

150

EXHIBIT 5

Abbott LCx® *Chlamydia trachomatis* Assay Product Labeling

ABBOTT LCx® PROBE SYSTEM

Chlamydia trachomatis Assay

Customer Support Center (U.S.A.)
Contact your local Customer Service Center
(LCx CSC) 1-800-527-1869

69-3134/R2

List No. 9B11-91
Printed in U.S.A.

INTENDED USE

The LCx® *Chlamydia trachomatis* Assay uses LCR™ (Ligase Chain Reaction) amplification technology in the LCx Probe System for the direct, qualitative detection of plasmid DNA of *C. trachomatis* in female endocervical and male urethral swab specimens or in male and female urine specimens from symptomatic and asymptomatic males and females.

SUMMARY AND EXPLANATION OF THE TEST

Chlamydia are non-motile, Gram-negative, obligate intracellular parasites of eukaryotic cells. They form inclusions in the cytoplasm of the host cell. *Chlamydia trachomatis*, one of three chlamydial species, is known to be a major etiologic agent of urogenital infections associated with salpingitis, ectopic pregnancies and tubal factor infertility in women as well as nongonococcal urethritis and epididymitis in men[1-3]. The genital site most commonly infected in women is the cervix, but the infection can be asymptomatic and, if untreated, is likely to ascend to the uterus, fallopian tubes and ovaries and may result in pelvic inflammatory disease (PID)[4]. Neonates born of infected mothers can contract inclusion conjunctivitis, nasopharyngeal infections and pneumonia due to *C. trachomatis*[5]. Infection by *C. trachomatis* in men is also often asymptomatic and, if untreated, may lead to epididymitis, a major complication[3].

The LCx *Chlamydia trachomatis* Assay uses the nucleic acid amplification method LCR to detect the presence of *C. trachomatis* plasmid DNA directly in clinical specimens. The four oligonucleotide probes in the LCx assay recognize and hybridize to a specific target sequence within the *C. trachomatis* plasmid DNA. The oligonucleotides are designed to be complementary to the target sequence so that in the presence of target, the probes will bind adjacent to one another. They can then be enzymatically joined to form the amplification product which subsequently serves as an additional target sequence during further rounds of amplification. The product of the LCR reaction is detected on the Abbott LCx Analyzer.

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

Specimen Preparation

Specimen preparation is the first step of the LCx *Chlamydia trachomatis* Assay, during which the DNA of the organism is released and made accessible to the enzymes and other components of the LCR reaction. This entails disrupting the cells and separating the strands of the DNA within the specimen. This is accomplished by heating the clinical specimen in Swab Specimen Transport Buffer for endocervical and male urethral swabs, or LCx Urine Specimen Resuspension Buffer for urine specimens.

Amplification

In addition to its chromosomal DNA, *C. trachomatis* harbors a cryptic plasmid, which is found in all serovars at approximately 10 copies per elementary body or reticulate body[6]. The LCR target is located within this plasmid and is a short sequence which is highly conserved among all the serovars of *C. trachomatis* but is not found in other species[7].

In the DNA amplification step, the prepared sample is added to the LCR reaction mixture consisting of four oligonucleotide probes, thermostable ligase and polymerase, and individual nucleotides in buffer.

The four oligonucleotide probes are designed in pairs that hybridize to complementary single-stranded *C. trachomatis* target sequences exposed in sample preparation. When a pair of probes has hybridized to a target sequence on a single strand of DNA, there is a gap of a few nucleotides between the probes. Polymerase acts to fill in this gap with the nucleotides in the LCR reaction mixture. Once the gap is filled, ligase can covalently join the pair of probes to form an amplification product that is complementary to the original target sequence and can itself serve as a target in subsequent rounds of amplification. Amplification occurs when the LCR reaction mixture and sample are incubated in a DNA thermal cycler.

During thermal cycling, the temperature is raised above the melting point of the hybridized amplification product causing it to dissociate from the original target sequence. Lowering the temperature allows more of the oligonucleotide probes to hybridize to the targets now available. The temperature continues to be cycled in this manner until sufficient numbers of target amplification product have accumulated and can be detected by Microparticle Enzyme Immunoassay (MEIA). Because the number of targets increases exponentially, forty thermal cycles are sufficient to achieve up to a billion-fold amplification in the number of target sequences.

Detection

The two pairs of oligonucleotide probes in the LCx *Chlamydia trachomatis* Assay are labeled with immunoreactive chemical groups called haptens. Each individual probe has either a capture hapten (recognized by an antibody attached to the MEIA microparticles) or a detection hapten (recognized by an antibody conjugated to alkaline phosphatase). The probes are labeled such that, when they are joined during the LCR reaction, the amplification product has the capture hapten at one end and the detection hapten at the other. In the LCx Analyzer, a sample of the amplification product is automatically transferred to an incubation well.[8] Here the microparticles coated with anti-capture hapten (Rabbit) bind the amplification product as well as any unligated probes carrying the capture hapten. The reaction mixture is then automatically transferred to a glass fiber matrix to which the microparticle complexes bind irreversibly. A wash step removes the unligated probes having only the detection hapten. The bound microparticle complexes are then incubated with anti-detection hapten (Rabbit): Alkaline Phosphatase conjugate which binds to the detection haptens. This antibody conjugate binds only to amplification products. The antibody conjugate can then be detected by addition of the substrate, 4-Methylumbelliferyl Phosphate, which is dephosphorylated by alkaline phosphatase to produce a fluorescent molecule, 4-Methylumbelliferone, that is measured by the MEIA optical assembly.

Prevention of DNA Contamination

Amplification reactions such as LCR are sensitive to accidental introduction of product from previous amplification reactions. False-positive results could occur if either the clinical specimen or the LCx reagents used in the amplification step become contaminated by accidental introduction of even a few molecules of amplification product. Measures to reduce the risk of DNA contamination in the laboratory include physically separating activities involved in performing LCR, chemically inactivating amplification product and complying to good laboratory practice.

Dedicated Laboratory Areas

Sample preparation must be completed in a dedicated area (Area 1) of the laboratory. Area 1 is used for processing of samples (specimens, LCx Chlamydia Negative Control and Calibrator) and the addition of processed samples to LCx Chlamydia Amplification Vials. Amplification and detection of amplification product is completed in a second dedicated area (Area 2).

153

Aerosol Containment

To reduce the risk of DNA contamination due to aerosols formed during pipetting, pipettors with aerosol barrier pipette tips must be used for all pipetting except for one step of the Urine Specimen Preparation protocol. This protocol allows for fine-tipped, single-use, plastic disposable pipettes. Refer to Specimen Preparation, Urine Specimen Preparation in this assay package insert.

Inactivation of Amplification Product

To reduce the risk of amplification product contamination, at the end of the LCx *Chlamydia trachomatis* Assay, amplification product is automatically inactivated using a two-reagent, chemical inactivation system. Both reagents (a chelated metal complex and an oxidizing agent) are delivered into the LCx Reaction Cells by the LCx Analyzer after the amplification product has been detected. The ensuing reaction results in the nearly complete destruction of any nucleic acid present. This effectively reduces the risk of contamination of the laboratory by amplification product.

REAGENTS

LCx STD Swab Specimen Collection and Transport Kit

(100 individually-wrapped sterile Collection Systems)

Each Collection System contains: one capped transport tube with 0.5 mL Swab Specimen Transport Buffer, one large-tipped cleaning swab, and one small-tipped specimen swab. Swab Specimen Transport Buffer contains ≥ 50 mM $MgCl_2$. Preservative: Sodium Azide.

LCx Urine Specimen Preparation Kit

(100 tests, 4 bottles of LCx Urine Specimen Resuspension Buffer, and 100 LCx Urine Specimen Microfuge Tubes with cap locks)

LCx Urine Specimen Resuspension Buffer contains ≥ 50 mM $MgCl_2$ and detergent. Preservative: Sodium Azide.

LCx Chlamydia Amplification Kit[a]

LCx Chlamydia Amplification Vials
(96 vials, 0.090 mL per vial)

Four oligonucleotide probes each at >10^{10} molecules per reaction, enzymes (≥ 1 unit thermostable DNA polymerase and ≥ 10,000 units thermostable DNA ligase), ≥ 3 µM each of two dNTPs, ≥ 20 µM NAD, and stabilizers in a buffered solution. Preservative: Sodium Azide.

LCx Chlamydia Negative Control, Calibrator, and Activation Reagent
(6 sets of each)

LCx Chlamydia Negative Control (N)
(0.48 mL per bottle)

≥ 1.8 µg/mL Salmon Testes DNA in a buffered solution. Preservative: Sodium Azide.

LCx Chlamydia Calibrator (C)
(0.48 mL per bottle)

Extracted DNA from inactivated *C. trachomatis* elementary bodies at approximately 20 IFU/mL in a buffered solution. Preservative: Sodium Azide.

LCx Chlamydia Activation Reagent (A)
(0.7 mL per bottle)

≥ 300 mM $MgCl_2$, dye: red (FD&C Red No. 2) and stabilizers in buffered solution. Preservative: Sodium Azide.

LCx Chlamydia Detection Reagent Pack[a] (100 tests)[b]

1. 1 Bottle, (6 mL)

   Anti-Capture Hapten (Rabbit) coated Microparticles, > 0.025% solids in buffered solution. Preservative: Sodium Azide.

2. 1 Bottle, (8 mL)

   Anti-Detection Hapten (Rabbit): Alkaline Phosphatase Conjugate > 0.01 µg/mL, in buffered solution with stabilizers and antimicrobials.

3. 1 Bottle, (10 mL)

   4-Methylumbelliferyl phosphate (MUP) 1.2 mM, in buffered solution. Preservative: Sodium Azide.

4. 1 Bottle, (25 mL)

   Metal chelate, > 13.5 mM, in buffered solution.

[a] The LCx Chlamydia Amplification Kit and the LCx Chlamydia Detection Reagent Pack are packaged as a set which must be used together. Verify that the lot numbers are identical before use.

[b] There are 100 tests provided in the LCx Chlamydia Detection Reagent Pack. Ninety-six tests are provided for the LCx *Chlamydia trachomatis* Assay. An additional 4 tests remain, of which 3 tests are provided for the purpose of troubleshooting the detection portion of the assay. See the LCx Analyzer Operations Manual, Section 10.

LCx Inactivation Diluent (1)

(2 Bottles, 900 mL per bottle)

An aqueous solution of 6-7.9% hydrogen peroxide.

LCx System Diluent (2)

(4 Bottles, 1000 mL per bottle)

Tris-Acetate Buffer, > 0.01 M. Preservative: Sodium Azide.

WARNINGS AND PRECAUTIONS

For *In Vitro* Diagnostic Use.

The LCx *Chlamydia trachomatis* Assay is only for use with female endocervical and male urethral swabs, or male and female urine specimens, and is limited to specimens collected, transported and stored according to instructions in the SPECIMEN COLLECTION AND TRANSPORT TO TEST SITE and SPECIMEN PREPARATION sections.

Use only the LCx STD Swab Specimen Collection and Transport Kit for collection of swab specimens.

LCx STD Swab Specimen Collection Systems in which transport media has spilled out should not be used.

The LCx STD Swab Specimen Collection and Transport Kit is intended to be used only in LCx Assays which require its use. No other intended use is applicable.

Use only the LCx Urine Specimen Preparation Kit for urine specimen processing.

The LCx Chlamydia Amplification Kit and the LCx Chlamydia Detection Reagent Pack are packaged as a set which must be used together. Verify that the lot numbers are identical before use.

Do not mix reagents from different lots. Do not pool reagents.

Use two dedicated areas within the laboratory for performing the LCx *Chlamydia trachomatis* Assay: Area 1 and Area 2.

Area 1 is to be used for the processing of samples (specimens, LCx Chlamydia Negative Control and Calibrator), and the addition of samples to the LCx Chlamydia Amplification Vial. Use of a biosafety hood or glove box equipped for UV irradiation is recommended. All reagents and equipment used in Area 1 (such as pipettors, microcentrifuge and the Abbott LCx Dry Bath) should remain in this dedicated area at all times. Area 1 items should never be used when

154

working with amplification product. Do not bring amplification product into Area 1. Specimens, activated Negative Control and Calibrator should be stored separately from Amplification Vials. All pipetting should be performed with aerosol barrier pipette tips except for a disposable pipette which may be used for one step in the Urine Specimen Preparation protocol. Swab specimens must only use extended-length (≥ 75 mm in length) aerosol barrier pipette tips to aspirate specimen from transport tubes. Cap locks must be placed on LCx Urine Specimen Microfuge tubes before specimens are heated in the LCx Dry Bath.

Area 2 is dedicated to amplification by thermal cycling and detection of the amplification product. All equipment, such as the Abbott LCx Thermal Cycler, the LCx Analyzer and all the accessories for the LCx Analyzer must be kept in this area. The LCx Chlamydia Detection Reagent Pack must be stored at 2-8°C. Care must be taken to separate the detection reagent pack that is in use from direct contact with samples and other LCx reagents. During execution of the detection protocol, aerosols from amplification product may potentially contaminate any surface within the closed LCx Analyzer. Everything inside the LCx Analyzer (including the detection reagent pack and the carousel) must be considered potential sources of DNA contamination and must be kept away from other LCx reagents, Negative Controls, Calibrators and specimens.

The LCx Calibrator contains extracted DNA from *C. trachomatis* elementary bodies that have been inactivated by a heat and chemical treatment process. The LCx Calibrator contains potentially infectious components. No known test method can offer complete assurance that products derived from inactivated microorganisms will not transmit infection. It is recommended that the LCx Calibrator and human specimens be handled in accordance with the OSHA Standard on Bloodborne Pathogens.[9] Biosafety Level 2[10] or other appropriate biosafety practices[11,12] should be used for materials that contain or are suspected of containing infectious agents. These precautions include, but are not limited to the following:

1. Wear gloves when handling specimens or reagents.
2. Do not pipette by mouth.
3. Do not eat, drink, smoke, apply cosmetics, or handle contact lenses in areas where these materials are handled.
4. Clean and disinfect all spills of specimens or reagents using a tuberculocidal disinfectant such as 1% sodium hypochlorite, or other suitable disinfectant.[13,14] Note: Chlorine solutions may pit equipment and metal. Use sufficient amounts or repeated applications of 70% ethanol until chlorine residue is no longer visible.
5. Decontaminate and dispose of all specimens, reagents and other potentially contaminated materials in accordance with local, state and federal regulations.[15,16]

To reduce the risk of DNA contamination, clean and disinfect all spills of specimens and regents using 1% (v/v) sodium hypochlorite solution, followed with 70% (v/v) ethanol. Note: Chlorine solutions may pit equipment and metal. Use sufficient amounts or repeated applications of 70% ethanol until chlorine residue is no longer visible.

This product contains sodium azide as a preservative.

Sodium azide has been reported to form lead or copper azide in laboratory plumbing. These azides may explode on percussion, such as hammering. To prevent formation of lead or copper azide, flush drains thoroughly with water after disposing of solutions containing sodium azide.

To remove contamination from old drains suspected of azide accumulation, the National Institute for Occupational Safety and Health recommends the following: (1) siphon liquid from trap using a rubber or plastic hose, (2) fill with 10% sodium hydroxide solution, (3) allow to stand for 16 hours, and (4) flush well with water.

The LCx STD Swab Specimen Collection and Transport Kit, LCx Urine Specimen Preparation Kit, LCx Chlamydia Negative Control, LCx Chlamydia Calibrator, LCx Chlamydia Activation Reagent, Microparticles, 4-Methylumbelliferyl phosphate (MUP) and LCx System Diluent (2) contain Sodium Azide and are classified per applicable European Community (EC) Directives as: Harmful (Xn). The following are the appropriate Risk (R) and Safety (S) phrases.

R22	Harmful if swallowed.
R32	Contact with acids liberates very toxic gas.
S2	Keep out of the reach of children.
S13	Keep away from food, drink and animal feedingstuffs.
S36	Wear suitable protective clothing.
S46	If swallowed, seek medical advice immediately and show this container or label.

The LCx Inactivation Diluent (1) contains Hydrogen Peroxide and is classified per applicable European Community (EC) Directives as: Irritant (Xi). The following are the appropriate Risk (R) and Safety (S) phrases.

R36/38	Irritating to eyes and skin.
S2	Keep out of the reach of children.
S26	In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
S36/39	Wear suitable protective clothing and eye/face protection.
S46	If swallowed, seek medical advice immediately and show this container or label.

Do not use kit or reagents beyond expiration date.

Use accurately calibrated equipment.

Failure to adhere to assay package insert instructions may result in erroneous results.

If the Step-Cycle run of the thermal cycler is interrupted or aborted, the run is invalid. Do not continue to process these samples. Make sure that the amplification vial caps are tightly closed. Remove carefully to a biohazard bag and seal the bag. Dispose of according to procedure of waste disposal in the LCx Thermal Cycler Operations Manual, Section 8: Hazards, Biosafety.

STORAGE INSTRUCTIONS

1. LCx Reaction Cells may be stored at 15-30°C until the expiration date.
2. The LCx Chlamydia Amplification Vials, LCx Chlamydia Activation Reagent, Negative Control, and Calibrator must be stored at 2-8°C until the expiration date.
3. The LCx Chlamydia Detection Reagent Pack must be refrigerated at 2-8°C when not in use. Care must be taken to separate the LCx Chlamydia Detection Reagent Pack that is in use from direct contact with samples and other LCx kit reagents. The detection reagents must not be frozen.
4. The LCx System Diluent (2) and LCx Inactivation Diluent (1) may be stored at 15-30°C until the expiration date. The LCx Inactivation Diluent (1) must be kept away from direct sunlight.
5. The LCx Urine Specimen Preparation Kit may be stored at 15-30°C until the expiration date.
6. The LCx STD Swab Specimen Collection and Transport Kit may be stored at 15-30°C until the expiration date.

155

SPECIMEN COLLECTION AND TRANSPORT TO TEST SITE

For domestic or international shipments, specimens should be packaged and labeled in compliance with applicable state, federal and international regulations covering the transport of clinical specimens and etiologic agents/infectious substances.

Time and temperature conditions for storage must be adhered to during transport. See Swab Specimen Transport and Urine Specimen Collection and Transport Sections for storage conditions.

For swab specimen collection, use only the LCx STD Swab Specimen Collection and Transport Kit (No. 6C94).

Note: Do Not Use The Large-tipped Cleaning Swab For Specimen Collection.

Note: Swab or urine specimens that are moderately bloody (greater than approximately 0.5% (v/v)) or grossly mucoid (greater than approximately 10% (w/v)) should not be tested since they may cause inhibition in the LCx *Chlamydia trachomatis* Assay.

Endocervical Swab Specimen Collection

1. Remove excess mucus from the exocervix with the large-tipped cleaning swab provided in the LCx STD Swab Specimen Collection System and discard.
2. Insert the small-tipped, specimen swab into the endocervix and rotate the swab for 15 to 30 seconds to ensure adequate sampling.
3. Verify that all Swab Specimen Transport Buffer is at the bottom of the tube. If necessary, tap or shake the solution down to the bottom of the tube. Unscrew the cap of the transport tube, insert the swab into the transport tube and break the swab at the score line. Replace the cap securely making sure that the swab fits into the cap and then screw on the cap until it clicks into place.
4. Label the transport tube with the patient's ID number and date of collection.

Male Urethral Swab Specimen Collection

1. Insert the small-tipped, specimen swab 2 to 4 cm into the urethra and rotate the swab for 3-5 seconds to ensure adequate sampling.
2. Verify that all Swab Specimen Transport Buffer is at the bottom of the tube. If necessary, tap or shake the solution down to the bottom of the tube. Unscrew the cap of the transport tube, insert the swab into the transport tube and break the swab at the score line. Replace the cap securely making sure that the swab fits into the cap and then screw on the cap until it clicks into place.
3. Label the transport tube with the patient's ID number and date of collection.

Swab Specimen Transport

1. Swab specimens can be shipped to the laboratory or testing site at 2-30°C or frozen. Swab specimens must arrive at the test site within 24 hours of shipment or be frozen.
2. Upon receipt in the laboratory or testing site, the swab specimens may be stored at 2-30°C, otherwise store at -20°C or below until processed. Store specimens at 2-30°C if testing is performed within 4 days of collection. If specimens are shipped frozen, maintain them at -20°C or below until testing.
3. All swab specimens stored at -20°C or below must be processed within 60 days of specimen collection.

Urine Specimen Collection and Transport

1. Collect specimen in a plastic, preservative-free, sterile urine specimen collection cup from patients who have not urinated within one hour prior to collection.
2. **The patient should collect the first 15-20 mL of voided urine (the first part of the stream).**
3. Verify the cup is securely closed and label the collection cup with the patient's ID number and date of collection.
4. Refrigerate the specimen immediately at 2-8°C, or store at -20°C or below.

Note: **Urine specimens must not be transported or stored at 15-30°C.**

Caution: Urine Specimens stored at Room Temperature should not be used for testing.

1. Urine specimens can be shipped to the laboratory or testing site at 2-8°C or frozen. Urine specimens must arrive at the test site within 24 hours of shipment.
2. Upon receipt in the laboratory or testing site, the urine specimen may be stored at 2-8°C or -20°C or below until processed.
3. All urine specimens stored at 2-8°C must be processed within 4 days of specimen collection.
4. All urine specimens stored at -20°C or below must be processed within 60 days of specimen collection. Once frozen specimens should not be thawed until ready for testing.

MATERIALS PROVIDED

LCx *Chlamydia trachomatis* Assay
(No. 9B11-91)*
96 Tests

A. LCx Chlamydia Amplification Kit

1.Cx Chlamydia Amplification Vials

LCx Chlamydia Negative Control, Calibrator and Activation Reagent

B. LCx Chlamydia Detection Reagent Pack

*** No. 9B11-97 includes an LCx Chlamydia Amplification Kit (96 tests), LCx Chlamydia Detection Reagent Pack (100 tests), and LCx Reaction Cells, List No. 9A48-01 (96 cells).**

MATERIALS REQUIRED BUT NOT PROVIDED

The LCx Probe System for the LCx *Chlamydia trachomatis* Assay consists of one LCx Analyzer, one LCx Thermal Cycler, and one LCx Dry Bath.

COLLECTION SITE

LCx STD Swab Specimen Collection and Transport Kit

(No. 6C94-24)

100 Collection Systems

Each Collection System contains: one capped transport tube with 0.5 mL Swab Specimen Transport Buffer, one large-tipped cleaning swab, and one small-tipped specimen swab. Swab Specimen Transport Buffer contains ≥ 50 mM $MgCl_2$. Preservative: Contains Sodium Azide.

Sterile Urine Specimen Collection Cup

Plastic, preservative-free sterile cup with a secure, screw-top cap capable of holding at least 25 mLs of urine.

AREA 1 (Specimen Preparation Area)

LCx Urine Specimen Preparation Kit

(No. 3B21-24)
100 Tests for processing urine specimens

156

Specimen Tube Racks

Precision Pipettors

100 µL, with aerosol barrier pipette tips (standard length)

Swab specimens require extended-length pipette tips (≥ 75 mm in length).

1.0 mL, with aerosol barrier pipette tips

Fine-tipped, Single-use, Plastic Disposable Pipette (optional)

X SYSTEMS® Centrifuge (No. 9527) or Equivalent Laboratory Microcentrifuge

(Speed ≥ 9,000 x g) for centrifuging urine specimens and pulse centrifuging of amplification vials if required prior to addition of specimens, Calibrator or Controls.

Note: Some centrifuges may require adapters for centrifuging amplification vials.

Abbott LCx Dry Bath

(No. 8B23)
For heat processing of specimen.

Swab Tube Closures

(No. 3B55-30, quantity 500)
For resealing Swab Transport Tubes after processing.

Vortex Mixer

AREA 2 (Amplification and Detection Area)

Abbott LCx Thermal Cycler

(No. 8B24)
Software version 2.1 or higher

Abbott LCx Analyzer

(No. 9A40)

The LCx Analyzer and accessories used for detection.

LCx System Module, version 3.0 or higher.

LCx Assay Module 2, version 2.0 or higher.

Assay Activation as described in the LCx Analyzer Operations Manual is required to initiate use of the LCx *Chlamydia trachomatis* Assay.

LCx Inactivation Diluent (1)

(No. 7B15-04)
2 x 900 mL bottles

LCx System Diluent (2)

(No. 7B14-04)
4 x 1000 mL bottles

X SYSTEMS Centrifuge (No. 9527) or Equivalent Laboratory Microcentrifuge

Note: This must be a separate unit other than the one in Area 1.

(Speed ≥ 9,000 x g) for pulse centrifuging of amplification vials before placing into the LCx Reaction Cells.

Note: Some centrifuges may require adapters for centrifuging amplification vials.

SPECIMEN PREPARATION

The use of the LCx Chlamydia Negative Control and Calibrator is integral to the performance of this LCx assay. These reagents must be prepared in conjunction with specimens to be tested. Refer to the Quality Control Procedures section for details.

All specimen storage and processing must take place in the dedicated Specimen Preparation Area (Area 1).

The LCx Dry Bath will require 20-40 minutes to heat up from a cold start. Confirm the dry bath has reached 97°C (±2°C) before proceeding.

Swab Specimen Preparation

1. Allow specimen to completely thaw if frozen.
2. Insert specimen tubes in wells of preheated dry bath and allow the heat block temperature to stabilize to 97°C (±2°C).
3. After the temperature of the heat blocks is stabilized at 97°C, heat specimens for 15 minutes (±1 minute). Failure to reach 97°C (±2°C) could impair release of the DNA in the specimen and may result in false negative results.
4. Remove specimen from the dry bath and allow to cool at room temperature for 15 minutes (±5 minutes).
5. After cooling, unscrew the cap and express the specimen swab along the inside of the tube so that liquid drains back into the sample solution at the bottom of the tube. The expressed swab and original closure should be discarded, and a new Swab Tube Closure (No. 3B55) should be screwed on until it clicks into place.
6. Test the processed swab specimen immediately or store at -20°C or below for up to 60 days. If the processed specimen is stored frozen, it must be completely thawed prior to addition to the LCx Chlamydia Amplification Vial.
7. Before opening the LCx Chlamydia Amplification Vials, verify by visual inspection that no liquid is in the cap of the vial. The amplification reagent level should measure approximately two-thirds of the conical part of the vial. If necessary, the vial may be pulse centrifuged in a microcentrifuge for 10-15 seconds.
8. Using a pipettor and extended-length pipette tips (≥ 75 mm in length) with aerosol barriers, add 100 µL of each processed specimen to the appropriately labeled LCx Chlamydia Amplification Vial and make sure each vial is securely closed. Transfer the vials to Area 2 and immediately place in the LCx Thermal Cycler for amplification. See LCx Amplification.

Urine Specimen Preparation

1. Allow urine specimen to completely thaw if frozen. Mix urine in the urine collection cup by swirling to resuspend any settled material. It is not necessary for all particulate matter to be fully dissolved.
2. Using a pipettor with aerosol barrier pipette tips, transfer 1 mL of mixed urine into the Urine Specimen Microfuge Tube from the Urine Specimen Preparation Kit (No. 3B21).
3. Centrifuge at ≥ 9,000 x g for 15 minutes (±2 minutes) in a microcentrifuge.
4. Using a fine-tipped, plastic disposable pipette, gently aspirate the urine supernatant completely off the pellet. **Be cautious not to contact or dislodge the pellet, which may be translucent.** The time between centrifugation and removal of supernatant must not exceed 15 minutes.
5. Using a pipettor with aerosol barrier pipette tips, add 1.0 mL of LCx Urine Specimen Resuspension Buffer. Close lid of microfuge tube and resuspend the pellet by vortexing until the pellet is off the bottom of the tube.
6. Secure the tube closure with a cap lock until it clicks into place.
7. Insert specimen tubes in wells of preheated dry bath and allow the heat block temperature to stabilize to 97°C (±2°C).

157

8. After the temperature of the heat blocks is stabilized at 97°C, heat specimens for 15 minutes (±1 minute). Failure to reach 97°C (±2°C) could limit release of the DNA in the specimen and may result in false negative results.
9. Remove the specimen from the dry bath and allow to cool at room temperature for 15 minutes (±5 minutes). Remove cap lock and discard.
10. Pulse-centrifuge the processed urine specimen in a microcentrifuge for a minimum of 10-15 seconds.
11. Test the processed urine specimen immediately, or store for up to 60 days at 2-8°C or -20°C or below prior to testing. If the processed urine specimen is stored frozen, it must be completely thawed prior to addition to the LCx Chlamydia Amplification Vial.
12. Before opening the LCx Chlamydia Amplification Vials, verify by visual inspection that no liquid is in the cap of the vial. The amplification reagent level should measure approximately two-thirds of the conical part of the vial. If necessary, the vial may be pulse centrifuged in a microcentrifuge for 10-15 seconds.
13. Using a pipettor with aerosol barrier pipette tips, add 100 µL of each processed urine specimen to the appropriately labeled LCx Chlamydia Amplification Vial and make sure each vial is securely closed. Transfer the vials to Area 2 and immediately place in the LCx Thermal Cycler for amplification. See procedure under LCx Amplification.

PROCEDURE

Procedural Precautions

1. Work in a laboratory using DNA amplification methods should always flow in a one-way direction beginning in the Specimen Preparation Area (Area 1), then moving to the Amplification and Detection Area (Area 2). **Do not bring any materials from Area 2 into Area 1.**
2. **Surface cleaning using a 1% (v/v) sodium hypochlorite solution followed by 70% (v/v) ethanol should be performed on bench tops and pipettors at least once per day prior to beginning an LCx Assay.**

   Note: Chlorine solutions may pit equipment and metal. Use sufficient amounts or repeated applications of 70% ethanol until chlorine residue is no longer visible.
3. Monthly monitoring procedures for the presence of DNA can be found in the Quality Control Section of this package insert. In addition, if the LCx Negative Control consistently fails the NEG HIGH or NEG AVE HIGH specifications for the LCx *Chlamydia trachomatis* Assay Parameters, laboratory contamination may be suspect. If this occurs contact LCx CSC.
4. The LCx *Chlamydia trachomatis* Assay is designed to be detected only on an Abbott LCx Analyzer.
5. All plastic materials coming into contact with the specimen should be free of any residue from previous specimens, reagents, or cleaning compounds.
6. During the sample (specimens, Calibrators or Negative Controls) addition step, only one LCx Chlamydia Amplification Vial should be open at any given time. After this step, the vials should remain closed throughout the thermal cycling and detection procedures. This aids in the prevention of cross-contamination.
7. Only one bottle of Negative Control or Calibrator should be open at any one time.

LCx Amplification

1. Refer to the LCx Thermal Cycler Operations Manual for detailed instructions on thermal cycler operation. Turn the LCx Thermal Cycler on for at least 15 minutes prior to use.
2. Collect all LCx Chlamydia Amplification Vials containing samples, Negative Control and Calibrator from Area 1 and transfer to Area 2 for thermal cycling.
3. LCx thermal cycling conditions should be edited to the following amplification parameters described below:

   Assay Step-Cycle File:

Segment 1	93°C for 1 second
Segment 2	59°C for 1 second
Segment 3	62°C for 1 minute 10 seconds
Cycle count	40 cycles

   The amplification run time is approximately 2 hours.

   The Assay Step-Cycle File is "Linked to" the Soak File at 25°C, indefinitely.
4. Place the amplification vials into the thermal cycler, and initiate run. After completion of the thermal cycler run, amplification product may remain at 15-30°C for up to 72 hours prior to LCx detection.

LCx Detection and Inactivation of Amplification Product

LCx *Chlamydia trachomatis* Assay Parameters

The following LCx *Chlamydia trachomatis* Assay parameters have been factory set in the Assay Module. These parameters can be printed, edited, and displayed as applicable, according to the procedure in your LCx Analyzer Operations Manual, Section 5.

ASSAY #16 CHLAMYDIA LCR

PARAMETER	DEFAULT	PARAMETER	DEFAULT
1 SPOOL LOCKOUT	1	34 MAX INTRCPT	12000.0
6 EQUIV ZONE LO*	0.80	45 CAL HIGH	2400.00
7 EQUIV ZONE HI*	1.00	46 CAL LOW	350.00
9 LS CHECK HI	115	47 CAL AVE HIGH	2200.00
10 AIR TEMP	35.0	48 CAL AVE LOW	550.00
11 AIR DEV	20.0	54 NUM POS CTRL	0
13 REAG TEMP	35.0	55 NUM NEG CTRL	2
14 REAG DEV	0.7	61 % CUTOFF	45.000
16 DIL TEMP	35.0	83 NUM CAL	2
17 DIL DEV	0.5	85 NEG HIGH	250.00
19 REMOTE TEMP	0.0	86 NEG LOW	0.00
20 REMOTE DEV	1.5	87 NEG AVE HIGH	150.00
32 MAX NRMSE	0.500	88 NEG AVE LOW	0.00
33 MIN CORR	0.950		

* These assay parameters are editable. Please refer to the LCx Analyzer Operations Manual, Section 5, for instruction on editing of assay parameters.

With the LCx Chlamydia Assay, patient results falling within the Equivocal Zone will be flagged as EQV in the NOTE column of the assay results printout. The Equivocal Zone Low and High parameters are set at default S/CO values of 0.80 and 1.00 respectively, which results in an S/CO Equivocal Zone of 0.80 - 0.99. These parameters may be edited if desired, but may only be edited to allow for the use of an expanded Equivocal Zone provided sufficient data has been generated by your laboratory. Please refer to Section 5 of the LCx Analyzer Operations manual for editing instructions. The EQUIV ZONE LO and EQUIV ZONE

158

HI parameters may be edited in the following ranges:

PARAMETER	DEFAULT	EDITABLE RANGE
6 EQUIV ZONE LO	0.80	0.00 - 0.80
7 EQUIV ZONE HI	1.00	1.00 - 2.00

1. *Refer to your LCx Analyzer Operations Manual for* detailed instrument operation procedures. Before running the LCx Analyzer, check to see that LCx Inactivation Diluent (1) contains a minimum of 100 mL and the LCx System Diluent (2) contains a minimum of 250 mL.
2. Remove the LCx Chlamydia Amplification Vials from the LCx Thermal Cycler.
3. Place LCx Reaction Cells into a MEIA Carousel; lock the carousel.
4. Pulse centrifuge the LCx Chlamydia Amplification Vials in a microcentrifuge for 10-15 seconds before placing into the LCx Reaction Cells.
5. *Place the amplification vials into the LCx Reaction Cells* in the following order: Negative Controls in positions 1 and 2, Calibrators in positions 3 and 4, and specimens in the remaining positions.
6. Place the carousel into the LCx Analyzer.
7. Lock the Amplification Vial Retainer by turning the handle counterclockwise.
8. Remove the LCx Chlamydia Detection Reagent Pack from 2-8°C storage, gently invert it 5 times, and open the reagent pack bottles in the numeric order: 1, 2, 3, 4.
9. Look for any film that may have formed over the openings of the reagent bottles. If present, remove using a long, clean pipette tip or a wooden applicator stick for *each bottle.*
10. Place the LCx Chlamydia Detection Reagent Pack into the LCx Analyzer.
11. Press RUN on the LCx Analyzer control panel. Final *assay results will be printed in approximately 60 minutes.*
12. Remove the assay printout results from the LCx Analyzer.
13. After completion of the detection procedure, remove the LCx Chlamydia Detection Reagent Pack, and close the caps in the numeric order: 4, 3, 2, 1. Store the detection reagent pack at 2-8°C.
14. Unlock the Amplification Vial Retainer by turning the handle clockwise until it is no longer over the MEIA carousel.
15. Remove the MEIA Carousel, individually remove the LCx Reaction Cells, and dispose appropriately.
16. Review the assay results and record patient results.

QUALITY CONTROL PROCEDURES

Negative Control and Calibrator Preparation

All Calibrator and Negative Control preparation must take place in the dedicated Specimen Preparation Area (Area 1).

1. The LCx *Chlamydia trachomatis* Assay procedure requires that the LCx Chlamydia Negative Control and the Calibrator be run in duplicate with each MEIA carousel of clinical specimens.
2. The LCx Chlamydia Negative Control and Calibrator are activated by the addition of 100 µL of LCx Chlamydia Activation Reagent. After addition, the *contents of the bottles are* then recapped and vortexed for 2-5 seconds. Each bottle of activated Negative Control or Calibrator is designed to be used up to 48 hours if stored at 2-8°C.

   Not adding activation reagent or adding incorrect volumes of activation reagent may give erroneous results and the run may be invalid.
3. Before opening the LCx Chlamydia Amplification Vials, verify by visual inspection that no liquid is in the cap of the vial. The amplification reagent level should measure approximately two-thirds of the conical part of the vial. If necessary, the vial may be pulse centrifuged in a microcentrifuge for 10-15 seconds.
4. Using a pipettor with aerosol barrier pipette tips, add 100 µL each of the activated LCx Chlamydia Negative Control and Calibrator to the appropriately labeled LCx Chlamydia Amplification Vial and make sure each vial is securely closed. Transfer the vials to Area 2 and immediately place in the LCx Thermal Cycler for amplification. See LCx Amplification.

Positive Control

A positive control that monitors the entire assay procedure including the specimen processing step should be tested in accordance with requirements of appropriate accrediting organizations.

Cells infected with *C. trachomatis* (available from ATCC, Catalogue No. VR-902B) may be processed and tested.

1. Pellet a single vial of cells by centrifugation.
2. Resuspend in 1.0 mL of LCx Urine Specimen Resuspension Buffer and serially dilute. It is recommended that a 10^{-5} to 10^{-6} dilution be used for testing.
3. Heat the diluted cell preparation (1.0 mL in LCx Urine Specimen Microfuge Tubes) in the LCx Dry Bath at 97°C (±2°C) for 15 minutes (±1 minute).

   Note: Place Cap Locks on the microfuge tubes before heating. Allow tubes to cool to room temperature before handling.
4. Perform amplification and detection in parallel with unknown specimens.

The positive control should give a positive assay value (S/CO ratio ≥ 1.00). Additional commercially available controls may be used.

Assay Validity

Validity of the LCx Chlamydia Negative Control and Calibrator assay results are automatically assessed by the LCx Analyzer before proceeding to analyze specimen assay results.

The LCx Analyzer first verifies that the assay results of the Negative Controls and Calibrator are within the specified ranges of the LCx *Chlamydia trachomatis* Assay Parameters by comparing the assay results of the Negative Control and Calibrator to the values listed in the assay parameters. A run is valid when the individual and average results are within the values listed for CAL HIGH, CAL LOW, CAL AVE HIGH, CAL AVE LOW, NEG LOW, NEG HIGH, NEG AVE HIGH, and NEG AVE LOW parameters in the LCx *Chlamydia trachomatis Assay* Parameters.

In the event of an invalid Negative Control or Calibrator assay result, the assay results printout will identify the out-of-range result, the S/CO ratio of the specimens will NOT be calculated and a flag indicating an invalid result will occur in the NOTE column of the printout next to the specimen assay results. If an out-of-range result is identified on the printout, refer to the LCx Analyzer Operations Manual, Section 10: Troubleshooting and Diagnostics for *an explanation of the error message. Instructions for* troubleshooting the detection portion of the assay can also be found under General Troubleshooting Procedures, LCx MEIA Performance Troubleshooting.

The LCx Analyzer does not calculate imprecision between Negative Control or Calibrator replicate values.

Note: Ensure the LCx Negative Controls and Calibrators are in the correct order on the MEIA carousel to avoid an invalid run.

If an amplification vial opens during thermal cycling, the sample is invalid and should not be used. Make sure that the amplification vial caps are tightly closed. Remove carefully to a biohazard bag and seal the bag. Dispose of according to procedure of waste disposal in the LCx Thermal Cycler Operations Manual, Section 8: Hazards, Biosafety.

Displayed and Printed Error Codes

If a displayed or printed error code appears, refer to the LCx Analyzer Operations Manual, Section 10.

Monitoring the Laboratory for the Presence of Amplification Product

It is recommended that this test be done at least once a month to monitor laboratory surfaces and equipment for contamination by amplification product.

Using the small-tipped specimen swab from an LCx Swab Specimen Collection System, insert the swab into the tube of Swab Specimen Transport Buffer. Allow the tip to become saturated with the buffer. When the tip is saturated, remove from the container and wipe the desired area using a broad sweeping motion. Replace the swab into the transport tube and break at the scored mark. Process following the LCx *Chlamydia trachomatis* Assay procedure. It is very important to be sure to test all areas that may have been exposed to samples and/or amplification product. This includes pipettors, pipettor handles, LCx Analyzer function keys, LCx Thermal Cycler function keys, bench surfaces, microcentrifuge and centrifuge adapters. If positive results (S/CO ratio ≥ 1.00) on surfaces are observed, clean the contaminated areas with 1% (v/v) sodium hypochlorite solution, followed by 70% (v/v) ethanol. Follow the appropriate operations manual for cleaning and decontaminating equipment if positive results are observed. **Note: Chlorine solutions may pit equipment and metal. Use sufficient amounts or repeated applications of 70% ethanol until chlorine residue is no longer visible. Repeat this cleaning procedure until the results are negative (S/CO ratio < 1.00).**

RESULTS

CALCULATIONS

The LCx *Chlamydia trachomatis* Assay uses MEIA detection on the LCx Analyzer to detect *C. trachomatis* plasmid DNA. All calculations are performed automatically.

The presence or absence of *C. trachomatis* is determined by relating the LCx Assay results for the specimen to the Cutoff value. The Cutoff value is the mean RATE (c/s/s) of the LCx calibrator duplicates multiplied by 0.45.

Calculation of the Cutoff value:

Cutoff value = 0.45 x (Mean of LCx Chlamydia Calibrator RATES)

The S/CO value is determined by calculating a ratio of the sample RATE to the Cutoff value.

$$\frac{S}{CO} = \frac{\text{Sample RATE}}{\text{Cutoff Value}}$$

INTERPRETATION OF RESULTS

Interpretation of specimen results is as follows:

S/CO RATIO	RESULT/REPORT
> or = EQUIV ZONE HI	LCx Positive. *C. trachomatis* plasmid DNA is detected, and positive for *C. trachomatis* by LCR amplification and MEIA detection.
< EQUIV ZONE LO	LCx Negative. *C. trachomatis* plasmid DNA is not detected and presumed negative for *C. trachomatis* by LCR amplification and MEIA detection.
> or = EQUIV ZONE LO and < EQUIV ZONE HI	LCx Equivocal. Repeat LCx test. If the repeat test S/CO ratio is greater than or equal to 1.00, *C. trachomatis* plasmid DNA is detected, and positive for *C. trachomatis* by LCR amplification and MEIA detection. If the repeat test is less than the S/CO ratio of 1.00, *C. trachomatis* plasmid DNA is not detected and presumed negative for *C. trachomatis* by LCR amplification and MEIA detection. **NOTE: On repeat test, the printout will display EQV for repeat test results falling in the Equivocal range; however, all repeat results should be interpreted per the above criteria.**

NOTE: A presumed negative result may be caused by possible inhibition of the LCx method, collection variables or other factors.

LIMITATIONS OF THE PROCEDURE

1. As with any diagnostic test, results from the LCx *Chlamydia trachomatis* Assay should be interpreted in conjunction with other clinical and laboratory findings.
2. The LCx *Chlamydia trachomatis* Assay will not detect plasmid-free variants of *C. trachomatis*.
3. Optimal performance of this test requires adequate specimen collection (sampling columnar epithelial cells) and handling (see Specimen Collection section). The assay should be performed only on swab samples from the endocervix and male urethra or on urine from males and females. The use of specimens other than those listed has not been validated. Specimen adequacy can only be assessed by microscopic visualization of columnar epithelial cells in the swab specimens.
4. A negative result does not exclude the possibility of infection because results are dependent on adequate specimen collection and absence of inhibitors. The presence of LCR inhibitors may cause false negative results with this product.
5. Use of the LCx *Chlamydia trachomatis* Assay is limited to personnel who have been trained in the procedures of an LCx Assay and the LCx Analyzer.
6. The LCx Inactivation procedure reduces the risk of contamination by amplification product. However,

160

DNA contamination from the Calibrator or clinical specimens must be controlled by good laboratory practice and careful adherence to the procedures specified in the package insert.

7. Therapeutic success or failure should not be determined as chlamydial nucleic acids may persist following appropriate antimicrobial therapy.[17]
8. Use of the LCx *Chlamydia trachomatis* Assay is not approved for the evaluation of suspected sexual abuse as well as for other medico-legal indications, or when positive results may have psycho-social impact.
9. The LCx *Chlamydia trachomatis* Assay provides qualitative results. No correlation can be drawn between the magnitude of a positive LCx *Chlamydia trachomatis* Assay signal and the number of *Chlamydia trachomatis* cells within an infected specimen. The assay detects only *C. trachomatis*, not *C. psittaci* or *C. pneumoniae*.
10. Some spermicidal agents and feminine powder sprays interfere with the assay and should therefore not have been used prior to collection of specimens for the assay.
11. Swab or urine specimens that are moderately bloody (greater than approximately 0.5% (v/v)) should not be tested since they may cause inhibition in the LCx *Chlamydia trachomatis* Assay.
12. Swab or urine specimens that are grossly mucoid (greater than approximately 10% (w/v)) should not be tested since they may cause inhibition in the LCx *Chlamydia trachomatis* Assay. Therefore, it is important that the exocervix be wiped free of mucus prior to collection of the swab specimen to ensure optimal specimen condition.
13. The LCx *Chlamydia trachomatis* Assay for male and female urine testing must be performed on first catch random urine specimens (defined as the first 15-20 mL of the urine stream). The effects of other variables such as first-catch vs. mid-stream, post douching, etc. have not been determined.
14. The effects of other potential variables such as vaginal discharge, use of tampons, douching, etc., and specimen collection variables have not been determined.
15. The predictive value of an assay will depend on disease prevalence in any particular population. See Table 2 for hypothetical predictive values at different prevalence rates that were derived from culture and DFA sensitivity and specificity results.
16. The LCx *Chlamydia trachomatis* Assay is not intended to replace cervical exam and endocervical sampling for diagnosis of urogenital infection. Patients may have cervicitis, urethritis, urinary tract infections, or vaginal infections due to other causes or concurrent infections with other agents.
17. Optimal assay performance and minimal inhibition are dependent upon removal of all urine supernatant from the pellet during urine processing.

EXPECTED VALUES

A total of 3281 male and female swab and urine specimens were obtained from patients attending six different hospitals or clinics including sexually transmitted disease clinics, family planning clinics and OB/GYN clinics.

Chlamydia trachomatis was detected by estimating the number of inclusion forming units found in culture of the male urethral or endocervical swabs. A specimen was determined to be positive if one or more inclusions were found. If cell culture was negative and the DFA test was positive, the specimen was considered positive for *C. trachomatis*.

Prevalence

The prevalence of positive *C. trachomatis* specimen results in patient populations varies depending on population characteristics such as age, sex and risk factors, and can vary depending on testing methodology. During clinical trials, prevalence using the LCx *Chlamydia trachomatis* Assay was observed to range from 0% to 25.4% as shown in Table 1.

Table 1

Prevalence of LCx positive results by Sample Type and Test Site

Sample Type	Test Site	No. Pos/ No. Tested	% PREV
Female Endocervical	1	23/196	11.7
	2	76/414	18.4
	3*	32/265	12.1
	4	11/202	5.4
	5	39/589	6.6
Female Urine	2	40/196	20.4
	6	18/231	7.8
	4	23/290	7.9
Male Urethral	1	30/217	13.8
	3*	25/111	22.5
	5	0/2	0.0
Male Urine	1	68/412	16.5
	6	15/59	25.4
	4	9/97	9.3

* In-house

Positive and Negative Predictive Values

The hypothetical positive and negative predictive values (PPV and NPV) for different prevalence rates using sensitivity and specificity of 93.1% and 97.1%, respectively, are shown in Table 2.

Table 2

LCx *Chlamydia trachomatis* Assay Hypothetical Predictive Values at Different Prevalence Rates

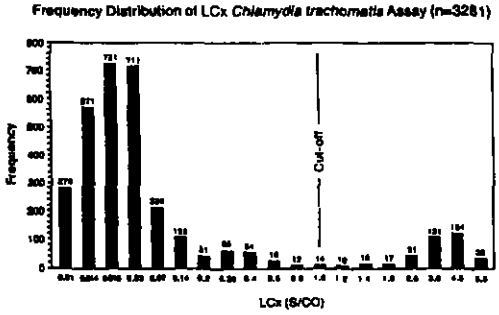
Prevalence Rate (%)	Sensitivity (%)	Specificity (%)	Positive Predictive Value (%)	Negative Predictive Value (%)
5	93.1	97.1	62.8	99.6
10	93.1	97.1	78.1	99.2
15	93.1	97.1	85.0	98.8
20	93.1	97.1	88.9	98.3

161

Frequency Distribution

A total of 3281 male and female specimens were assayed at six clinical sites. The frequency of S/CO values (sample c/s/s to cutoff c/s/s) of the specimens is illustrated in Figure 1. The distribution of the S/CO values is as follows: 87.1% of the S/CO values were <0.80, 12.5% were ≥1.0, and 0.4% were between 0.80 and 0.99. Three of the thirteen specimens with S/CO values between 0.80 and 0.99 were culture positive/LCx negative discordants. There were 19 LCx positive specimens with S/CO values between 1.0 and 1.3, of which 9 were either culture or DFA positive. Nine of the ten remaining specimens were positive when tested by LCR for the major outer membrane protein (MOMP) gene.

Figure 1



*Value under each column is the midpoint LCx S/CO for that cell.

SPECIFIC PERFORMANCE CHARACTERISTICS

The performance characteristics of the LCx *Chlamydia trachomatis* Assay were determined by comparing assay results to results of tissue culture for *C. trachomatis*; specimens with LCx positive/culture negative discordant results were retested by direct fluorescent monoclonal antibody (DFA) test. Because tissue culture and DFA may not detect all specimens with *C. trachomatis*, additional testing with a different probe set for the MOMP gene region of *C. trachomatis* was used for specimens that were culture negative/DFA negative/LCx positive.

The results of the clinical studies are shown in Tables 3 and 3A. For the clinical specimens tested, there were 252 that were culture positive. Of the 252 culture positive specimens, the LCx assay was negative for 24. The reason for these results could be inhibition, collection variables, or other factors. The LCx assay was positive for an additional 157 specimens, 73 of which were DFA positive. An additional 80 of the 84 remaining culture negative, LCx positive specimens that were not confirmed by DFA were positive by an alternate probe test (LCR assay run on the LCx Analyzer using a probe set targeting the MOMP gene region of *C. trachomatis*). This alternate target test was performed only on specimens that were positive in the LCx Assay, but negative by culture and DFA.

Specificity

The following list identifies the bacteria, parasites, viruses, yeast and fungi that have been tested in the LCx *Chlamydia trachomatis* Assay. All isolates were tested using the equivalent of at least 10^7 copies of genomic DNA except as indicated. The tested organisms include those that are commonly found in the urogenital tract as well as others. All gave negative values.

Acinetobacter calcoaceticus
Actinomyces israelii
Adenovirus
Aeromonas hydrophila
Alcaligenes faecalis
Bacillus subtilis
Bacillus thuringiensis
Bacteroides fragilis
Bifidobacterium longum
Blastomyces dermatitidis
Branhamella catarrhalis
Candida albicans
Candida glabrata
Citrobacter freundii
Chlamydia pneumoniae
Chlamydia psittaci
Clostridium sporogenes
Corynebacterium renale
Cryptococcus laurentii
Cryptococcus neoformans
Cytomegalovirus
Edwardsiella tarda
Enterobacter aerogenes
Enterobacter cloacae
Enterococcus faecalis
Enterococcus faecium
Epstein-Barr Virus
Escherichia coli
Ewingella americana
Flavobacterium odoratum
Fusobacterium nucleatum
Gardnerella vaginalis
Haemophilus influenzae
Haemophilus ducreyi
Hafnia alvei
*Helicobacter pylori**
Hepatitis B Virus
Herpes Simplex Virus I
Herpes Simplex Virus II
Histoplasma capsulatum
Human Herpes Virus 6
Human Immunodeficiency Virus type 1
Human T-Cell Lymphotropic Virus type I
Klebsiella pneumoniae
Lactobacillus casei
Moraxella lacunata
Morganella morganii
Mycobacterium avium
Mycobacterium gordonae
Mycobacterium tuberculosis
Mycoplasma hominis
Neisseria gonorrhoeae
Neisseria lactamica
Neisseria meningitidis
Neisseria sicca
Human Papilloma Virus 16
Human Papilloma Virus 18
Pasteurella multocida
Peptostreptococcus asaccharolyticus
Peptostreptococcus productus
Plesiomonas shigelloides
Propionibacterium acnes
Proteus mirabilis
Providencia stuartii
Proteus vulgaris
Pseudomonas aeruginosa
Saccharomyces cerevisiae
Salmonella enteritidis
Salmonella minnesota
Salmonella typhimurium
Schistosoma haematobium
Shigella sonnei
Staphylococcus aureus
Staphylococcus epidermidis
Streptococcus agalactiae
Streptococcus mitis
Streptococcus mutans
Streptococcus pneumoniae
Streptococcus pyogenes
Streptomyces griseus
Treponema pallidum
Trichomonas vaginalis
Ureaplasma urealyticum
Varicella-zoster Virus
Veillonella caviae
Veillonella parvula
Veillonella species
Vibrio parahaemolyticus
Yersinia enterocolitica

* Tested using equivalent of 10^8 copies of genomic DNA.

Analytical Sensitivity

The analytical sensitivity of this assay (limit of detection) is 1 Inclusion Forming Unit (IFU) of any of the 15 serovars of *Chlamydia trachomatis*. The analytical sensitivity of this assay was determined by a serial dilution study on all 15 serovars of *Chlamydia trachomatis* (A, B, Ba, C, D, E, F, G, H, I, J, K, L1, L2, L3). Each serovar was diluted to less than 1 IFU per reaction and tested in the LCx *Chlamydia trachomatis* Assay. In all cases, each replicate of a dilution giving ≤ 1 IFU per test (100 µL specimen volume) was positive by the LCx *Chlamydia trachomatis* Assay.

162

Table 3

Performance Summary Compared to Culture and DFA Testing

Population By Sample Type		N	LCx Culture DFA	Pos Pos NA	Pos Neg Pos	Pos Neg Neg	Neg Pos NA	Neg Neg NA	Sensitivity (95% C.I.)	Specificity (95% C.I.)
Female Endocervical	Symptomatic	602		39	6	5	5	547	90.0% (45/50) (78.2-96.7)	99.1% (547/552) (97.9-99.7)
	Asymptomatic	1,064		72	34	25	7	926	93.8% (106/113) (87.7-97.5)	97.4% (926/951) (96.1-98.3)
Female Urine*	Symptomatic	393		20	8	7	1	357	96.6% (28/29) (82.2-99.9)	98.1% (357/364) (96.1-99.2)
	Asymptomatic	324		24	8	14	2	276	94.1% (32/34) (80.3-99.3)	95.2% (276/290) (92.0-97.3)
Male Urethral	Symptomatic	260		40	1	8	0	211	100.0% (41/41) (91.4-100)	96.3% (211/219) (92.9-98.4)
	Asymptomatic	70		5	0	1	1	63	83.3% (5/6) (35.9-99.6)	98.4% (63/64) (91.6-100)
Male Urine*	Symptomatic	354		41	9	20	6	278	89.3% (50/56) (78.1-96.0)	93.3% (278/298) (89.8-95.9)
	Asymptomatic	214		11	7	4	2	190	90.0% (18/20) (68.3-98.8)	97.9% (190/194) (94.8-99.4)
Total	Symptomatic	1,609		140	24	40	12	1,393	93.2% (164/176)	97.2% (1393/1433)
	Asymptomatic	1,672		112	49	44	12	1,455	93.1% (161/173)	97.1% (1455/1499)

Table 3A

Performance Summary Compared to Culture and DFA Testing by Site

Sample Type	Test Site	Total	LCx Culture DFA	Pos Pos NA	Pos Neg Pos	Pos Neg Neg	Neg Pos NA	Neg Neg NA	Sensitivity (95% C.I.)	Specificity (95% C.I.)
Female Endocervical	1	196		20	0	3	3	170	87.0% (20/23) (66.4-97.2)	98.3% (170/173) (95.0-99.6)
	2	414		38	23	15	2	336	96.8% (61/63) (89.0-99.6)	95.7% (336/351) (93.1-97.6)
	3 (In-house)	265		18	9	5	0	233	100.0% (27/27) (87.2-100.0)	97.9% (233/238) (95.2-99.3)
	4	202		9	1	1	1	190	90.9% (10/11) (58.7-99.8)	99.5% (190/191) (97.1-99.9)
	5	589		26	7	6	6	544	84.6% (33/39) (69.5-94.1)	98.9% (544/550) (97.6-99.6)
Female Urine*	2	196		21	6	13	2	154	93.1% (27/29) (77.2-99.2)	92.2% (154/167) (87.1-95.8)
	6	231		6	9	3	0	213	100.0% (15/15) (78.2-100.0)	98.6% (213/216) (96.0-99.7)
	4	290		17	1	5	1	266	94.7% (18/19) (74.0-99.9)	98.2% (266/271) (95.8-99.4)
Male Urethral	1	217		25	1	4	1	186	96.3% (26/27) (81.0-99.9)	97.9% (186/190) (94.7-99.4)
	3 (In-house)	111		20	0	5	0	86	100.0% (20/20) (83.2-100.0)	94.5% (86/91) (87.6-98.2)
	5	2		0	0	0	0	2		100.0% (2/2) (15.8-100.0)
Male Urine*	1	412		37	9	22	7	337	86.8% (46/53) (74.7-94.5)	93.9% (337/359) (90.9-96.1)
	6	59		8	7	0	0	44	100.0% (15/15) (78.2-100.0)	100.0% (44/44) (92.0-100.0)
	4	97		7	0	2	1	87	87.5% (7/8) (47.4-99.7)	97.8% (87/89) (92.1-99.7)

Although it was not included in the analysis described above, an LCx assay specific for sequences contained within the MOMP gene of *Chlamydia trachomatis* was used to examine the culture negative/LCx positive specimens which were not confirmed by DFA. Eighty of these 84 specimens were shown to be positive for *Chlamydia trachomatis* nucleic acid by the MOMP assay. These data indicate that these apparent false positive specimens may have been, in fact, true positives that were missed by culture and DFA. The LCx Assay was negative for 24 of 252 culture positive specimens. The reason for these results could be inhibition, collection variables or other factors.

*Comparison cultures for these specimens were performed on endocervical and male urethral swab specimens, respectively.

Table 4

Reproducibility

Panel Member	N	Mean S/CO	Within Run (SD)	Within Run (%CV)	Between Run (SD)	Between Run (%CV)	Between Day (SD)	Between Day (%CV)	Total (SD)	Total (%CV)
A	90	0.02	0.020	NA	0.021	NA	0.021	NA	0.022	NA
B	90	2.23	0.396	17.8	0.428	19.2	0.428	19.2	0.458	20.6
C	90	3.18	0.265	8.4	0.380	12.0	0.380	12.0	0.422	13.3
D	90	3.98	0.161	4.0	0.312	7.8	0.335	8.4	0.422	10.6
E	90	4.65	0.136	2.9	0.414	8.9	0.414	8.9	0.518	11.1
	N	Mean RATE	Within Run (SD)	Within Run (%CV)	Between Run (SD)	Between Run (%CV)	Between Day (SD)	Between Day (%CV)	Total (SD)	Total (%CV)
Negative Control	78	17.8	27.49	NA	27.49	NA	27.54	NA	28.35	NA
Calibrator	78	1089.8	97.69	9.0	103.71	9.5	127.64	11.7	142.97	13.1

*NA = Not Applicable

Table 5

LCx Chlamydia
Reproducibility of Negative Control and Calibrator Values (318 Runs)

Sample	Calibrator	Negative Control
Mean (c/s/s)	1219.5	16.3
SD	200.0	21.5
%CV		
Average Within Run	5.9	NA
Overall	16.4	NA
Range (c/s/s)	577.6 - 1960.5	0 - 222.2

Inactivation Efficiency of Amplification Product

A 10^7 fold reduction of the amplification product concentration is achieved in overamplified samples by the inactivation chemistry used in the LCx *Chlamydia trachomatis* Assay. For clinical specimens, 70 positive urine and swab specimens were used to evaluate the inactivation efficiency. Amplified specimens were diluted 100-fold and then inactivated using the LCx assay procedure before re-testing with the LCx *Chlamydia trachomatis* Assay. All specimens showed negative results (S/CO ratio < 1.00) after inactivation.

Reproducibility

Assay reproducibility of the LCx *Chlamydia trachomatis* Assay was demonstrated by testing a 5-member panel consisting of 4 dilutions of *Chlamydia trachomatis*-infected McCoy cells in a specimen matrix. This reproducibility panel was run at four different sites in triplicate twice a day for three days. The clinical trials were all run in laboratories with a high degree of expertise in DNA amplification assays. It was important to demonstrate the reproducibility obtained by technicians who had no prior experience with this type of assay. Therefore, the technicians at two of the sites in which the reproducibility panel was run had been instructed in the LCx *Chlamydia trachomatis* Assay but had not previously run DNA amplification assays. The results of the average within-run, between run, between day, and total reproducibility of the LCx *Chlamydia trachomatis* Assay are shown in Table 4. The variance estimates presented in Table 4 were calculated from the variance components obtained using SAS's® PROC NESTED procedure. Variance estimates are cumulative from left to right, i.e., "Within Run" variability is included in the "Between Run" term, etc. Coefficients of Variation (CV) for Panel Member A and the Negative Control are not presented, instead the Standard Deviation (SD) is presented as the measure of variability. Results are presented according to the National Committee for Clinical Laboratory Standards (NCCLS).

Negative Control and Calibrator Performance

The performance of the Calibrator used during the clinical trials was evaluated by calculating the overall mean and the percent Coefficient of Variation (%CV) across the valid runs. (See Table 5). The overall %CV was calculated without regard to replicates. The average within run %CV is the %CV of each Calibrator replicate averaged over 318 runs. For the Negative Control, the overall Standard Deviation (SD) is presented as the measure of variability. These results are similar to the results obtained in Table 4, Reproducibility.

Proficiency

In order to determine the proficiency of the LCx *Chlamydia trachomatis* Assay, a 20-member panel consisting of 20 unique samples at various LCx signals in urine and swab samples were tested. This panel was run at three different sites and each sample was analyzed once. The technicians at all sites were inexperienced users and received training on the LCx Assay just prior to beginning the proficiency study. The results of this study demonstrated 100% agreement on this panel across all three sites.

BIBLIOGRAPHY

1. Schachter J: Chlamydial infections. *Western Journal of Medicine* 1990; 153: 523-534.
2. Cates W Jr., and Wasserheit JN: Genital chlamydial infections: epidemiology and reproductive sequelae. *American Journal of Obstetrics and Gynecology* 1991; 164: 1771-1781.
3. Berger RE, Alexander ER, Harnisch JP, et al: Etiology, manifestation and therapy of acute epididymitis: prospective study of 50 cases. *Journal of Urology* 1979; 121: 750-754.
4. Brunham RC, Paavonen J, Stevens CE, Kiviat N, et al: Mucopurulent cervicitis the ignored counterpart in women of urethritis in men. *New England Journal of Medicine* 1984; 311: 1-6.
5. Alexander ER, and Harrison HR: Role of *Chlamydia trachomatis* in perinatal infection. *Review of Infectious Diseases* 1983; 5:713.
6. Palmer L, and Falkow S: A common plasmid of *Chlamydia trachomatis*. *Plasmid* 1986; 16:52-62.
7. Joseph T, Nano FE, Garon CF, et al: Molecular Characterization of *Chlamydia trachomatis* and *Chlamydia psittaci* plasmids. *Infection and Immunity* 1986; 51: 699 -703.
8. Burczak J, Ching S, Hu H, and Lee H: Ligase Chain Reaction for the Detection of Infectious Agents. *Molecular Methods For Virus Detection* 1995; (Wiedbrank, D. and Farkas, D., Editors) 315-328.
9. US Department of Labor, Occupational Safety and Health Administration, 29 CFR Part 1910.1030,

164

Occupational Exposure to Bloodborne Pathogens; Final Rule. *Federal Register* 1991;56(235):64175-82.

10. US Department of Health and Human Services. *Biosafety in Microbiological and Biomedical Laboratories*, HHS Publication No. (CDC) 93-8395. Washington, DC: US Government Printing Office, May 1999.
11. World Health Organization. *Laboratory Biosafety Manual.* Geneva: World Health Organization, 1993.
12. National Committee for Clinical Laboratory Standards. *Protection of Laboratory Workers from Instrument Biohazards and Infectious Disease Transmitted by Blood, Body Fluids, and Tissue;* Approved Guideline. NCCLS Document M29-A. Wayne, PA: NCCLS, 1997.
13. CDC. Recommendations for Prevention of HIV Transmission in Health-Care Settings. *MMWR* 1987; 36(2S):3S-18S.
14. Sehulster LM, Hollinger FB, Dreesman GR, et al. Immunological and Biophysical Alteration of Hepatitis B Virus Antigens by Sodium Hypochlorite Disinfection. *Appl Envir Microbiol* 1981;42(5):762-7.
15. National Committee for Clinical Laboratory Standards. *Clinical Laboratory Waste Management:* Approved Guideline. NCCLS Document GP5-A. Villanova, PA: NCCLS, 1993;13(22):1-18, 29-42.
16. US Environmental Protection Agency. *EPA Guide for Infectious Waste Management.* Publication No. EPA/530-SW-86-014. Washington, DC: US Environmental Protection Agency, 1986:1-1–5-5, R1–R3, A1–A24.
17. Gaydos C: Molecular Amplification Assays to Detect Chlamydial Infections in Urine Specimens from High School Female Students and to Monitor the Persistence of Chlamydial DNA after therapy. *The Journal of Infectious Diseases* 1998:177:417-24.

Notice:

The purchase of this product allows the purchaser to use it for amplification of nucleic acid sequences and for detection of nucleic acid sequences for human *In-Vitro* diagnostics in accordance with the stated intended use. No general patent or other license of any kind other than this specific right of use from purchase is granted hereby.

ABBOTT LABORATORIES
Diagnostics Division
Abbott Park, Illinois 60064

EXHIBIT 6

Device Components

This page represents 1 whole-page redactions.

EXHIBIT 7

Performance Study Data Tables

Tables 1A through 1E

Aged Urine Study Using LCx® Assay and Sierra Diagnostics Collection, Preservation, and Transport System With Urine Spiked With *N. gonorrhea*

Table 1A

Lot No. 06x—Frozen (-70°C); Preserved (120 hrs. @ +30°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Table 1B

Lot No. 08x—Frozen (-70°C); Preserved (144 hrs. @ +30°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Table 1C

Lot No. 011x—Frozen (-70°C); Preserved (120 hrs. @ +60°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	—

Correlation = 100%

169

Table 1D

Lot No. 015x—Frozen (-70°C); Preserved (144 hrs.

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Table 1E

Lot No. 016x—Frozen (-70°C); Preserved (160 hrs.

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Tables 2A through 2G
Aged Urine Study Using LCx® Assay and Sierra Diagnostics Collection, Preservation, and Transport System With Urine Spiked With *C. trachomatis*

Table 2A

Lot No. 04x—Frozen (-70°C); Preserved (96 hrs. @ +30°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Table 2B

Lot No. 06x—Frozen (-70°C); Preserved (120 hrs. @ +30°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Table 2C

Lot No. 08x—Frozen (-70°C); Preserved (144 hrs. @ +30°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Table 2D

Lot No. 09x—Frozen (-70°C); Preserved (144 hrs. @ +60°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Table 2E

Lot No. 011x—Frozen (-70°C); Preserved (160 hrs. @ +60°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

Table 2F

Lot No. 015x—Frozen (-70°C); Preserved (180 hrs. @ +60°C)

	Positive	Negative	Total
Frozen	10	3	13
Preserved	10	3	13
Difference	0	0	---

Correlation = 100%

172

Table 2G

Lot No. 016x—Frozen (-70°C); Preserved (166 hrs.

	Positive	Negative	Total
Frozen	**10**	**3**	**13**
Preserved	**10**	**3**	**13**
Difference	**0**	**0**	**---**

Correlation = 100%

Clinical Study Summary Data

Table 3A

Site 1

Preservative vol. = 2.0ml / Collection Cup Size = 90ml

Specimen*	Positive Results		Negative Results
	Chlamydia	Gonorrhea	
Unpreserved 2-8°C 24 hrs.	17	4	155
Preserved Ambient Temp.** 144 hrs.	17	4	155
Correlation	100%	100%	100%

* Average combined total volume (preserved and unpreserved) was44ml.
** Average transport temperature was 48°C. Ambient storage temperature was 21°C.

Table 3B

Site 2

Preservative vol. = 2.0ml / Collection Cup Size = 90ml

Specimen*	Positive Results		Negative Results
	Chlamydia	Gonorrhea	
Unpreserved 2-8°C 24 hrs.	13	2	145
Preserved Ambient Temp.** 144 hrs.	13	2	145
Correlation	100%	100%	100%

* Average combined total volume (preserved and unpreserved) was 52ml.
** Average transport temperature was 63.9°C. Ambient storage temperature was 21°C.

174

Table 3C

Site 3

Preservative vol. = 2.0ml / Collection Cup Size = 90ml

Specimen*	Positive Results		Negative Results
	Chlamydia	Gonorrhea	
Unpreserved 2-8°C 24 hrs.	14	5	80
Preserved Ambient Temp.** 144 hrs.	14	5	80
Correlation	100%	100%	100%

* Average combined total volume (preserved and unpreserved) was 43ml.
** Average transport temperature was 58.8°C. Ambient storage temperature was 21°C.

Table 3D

Site 4

Preservative vol. = 2.0ml / Collection Cup Size = 90ml

Specimen*	Positive Results		Negative Results
	Chlamydia	Gonorrhea	
Unpreserved 2-8°C 24 hrs.	9	1	52
Preserved Ambient Temp.** 144 hrs.	9	1	52
Correlation	100%	100%	100%

* Average combined total volume (preserved and unpreserved) was 52ml.
** Average transport temperature was 63.9°C. Ambient storage temperature was 21°C.

Table 3E

Site 5

Preservative vol. = 4.0ml / Collection Cup Size = 90ml

Specimen*	Positive Results		Negative Results
	Chlamydia	Gonorrhea	
Unpreserved 2-8°C 24 hrs.	15	0	70
Preserved Ambient Temp.** 144 hrs.	15	0	70
Correlation	100%	100%	100%

* Average combined total volume (preserved and unpreserved) was 49ml.
** Average transport temperature was 46.2°C. Ambient storage temperature was 25°C.

176

Symptomatic vs. Asymptomatic Results By Sex and Study Site

Table 4A
Female Subjects

	Symptomatic			Asymptomatic		
	Subjects	Positive	Negative	Subjects	Positive	Negative
Site 1	84	11	73	76	6	70
Site 2	53	10	43	87	3	84
Site 3	50	9	41	31	2	29
Site 4	34	6	28	18	3	15
Site 5	33	10	23	40	5	35
Total	254	46	208	252	19	233

Table 4B
Male Subjects

	Symptomatic			Asymptomatic		
	Subjects	Positive	Negative	Subjects	Positive	Negative
Site 1	26	3	23	0	--	--
Site 2	20	2	18	0	--	--
Site 3	14	3	11	5	2	3
Site 4	8	1	7	2	2	0
Site 5	3	2	1	9	1	8
Total	71	11	60	16	5	11

Table 5
LCx® Gonococcal Serovar Sensitivity At Varying Preservative to Urine Concentrations (Spiked Urine)

Serovar	Unpreserved (-70°C/24 hrs.)	Preserved (25°C/144 hrs.)	
	Result	Ratio	Result
1A-13	Positive	1:10	Positive
		1:15	Positive
1A-20	Positive	1:10	Positive
		1:15	Positive
1A-5	Positive	1:10	Positive
		1:15	Positive
1A2	Positive	1:10	Positive
		1:15	Positive
1A-3	Positive	1:10	Positive
		1:15	Positive
1B-17	Positive	1:10	Positive
		1:15	Positive
1B2	Positive	1:10	Positive
		1:15	Positive
1B-18	Positive	1:10	Positive
		1:15	Positive
1B-7	Positive	1:10	Positive
		1:15	Positive
1B-5	Positive	1:10	Positive
		1:15	Positive

EXHIBIT 8

Sterilization Information

Sierra Diagnostics, L.L.C.
Premarket Notification – Urine Collection, Preservation and Transport System

Sterilization Information

Sierra purchases the collection cup component of the Urine Collection System from Samco Scientific, who contracts with Steris Isomedix to have the cups sterilized by gamma irradiation prior to shipment to Sierra.

Upon receipt of the collection cups, Sierra aseptically fills the cups with the preservative and indicator beads. Prior to filling, Sierra sterilizes the preservative by passage through a 0.2 micron filter, and the indicator beads by steam autoclave. The sterility assurance level (SAL) for the interior of the finished device is 10^{-6}.

Sterility of the finished device is confirmed by conducting a USP sterility test on a statistically valid sample size of each manufactured lot in accordance with Sierra's standard operating procedures.

FEB 2 8 2002

510(k) Summary

K013819

I. General Information on Submitter

Name:	Sierra Diagnostics, L.L.C.
Address:	21109 Longeway #C Sonora, CA 95370
Telephone:	(209) 536-0886
Fax:	(209) 536-0853
Contact Person:	Tony Baker
Date Prepared:	October __, 2001

II. General Information on Device

Name:	Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System
Classification Name:	Accessory to Neisseria spp. and Chlamydia serological reagents

III. Predicate Device

The standard urine collection cup used to collect specimens for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays and referenced in the package inserts for the LCx® devices (See 510(k) Nos. K935833 (*Neisseria gonorrhoeae*) and K934622 (*Chlamydia trachomatis*)).

IV. Description of Device

The device is comprised of a urine collection cup containing of a nucleic acid chemical preservative. The device allows urine specimens for LCx® gonococcal or chlamydial testing to be preserved for up to 6 days at temperatures not to exceed 60°C. Inert indicator beads are included in the urine cup as an indicator that a preservative is present in the sample.

V. Intended Use

The Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System is intended for use in the collection, preservation, and transportation of urine specimens at temperatures not exceeding 60°C for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays.

VI. Technological Characteristics of Device Compared to Predicate Device

The Sierra Urine Collection, Preservation, and Transport System and the predicate device share the same technological characteristics with the exception of the method of preservation. The predicate device employs a temperature preservation method while the Sierra device uses chemical preservation.

VII. Summary of Performance Data

The effectiveness of the Sierra Urine Collection, Preservation, and Transport System was established by the comparative testing of fresh and preserved urine spiked with gonococcal and chlamydial DNA. LCx® testing of samples that were preserved through refrigeration for 24 hours were compared with results for specimens preserved with the Sierra device and tested after being held for 144 hrs. at 60°C. There was a 100% correlation between the refrigerated and preserved samples.

Effectiveness was further established by a multi-site clinical study. The results of this study demonstrated that the device effectively preserved gonococcal and chlamydial nucleic acid targets in urine specimens from symptomatic and asymptomatic males and females.

The effective preservative concentration range and effect on LCx® sensitivity was established by a study using urine specimens spiked with less than 10 cfu of 10 different gonococcal serovars. Results from this test proved that Sierra's device effectively preserved nucleic acid targets down to the LCx® level of detection with a preservative to urine ratio ranging from 1:10 to 1:15.

DEPARTMENT OF HEALTH & HUMAN SERVICES

Food and Drug Administration
2098 Gaither Road
Rockville MD 20850

FEB 2 8 2002

Sierra Diagnostics, L.L.C.
c/o Donald R. Stone, Esq.
Kirkpatrick and Lockhart, LLP
1800 Massachusetts Avenue, NW
Suite 200
Washington, DC 20036-1221

Re: k013819
Trade/Device Name: Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System
Regulation Number: 21 CFR 866.2900
Regulation Name: Microbiological Specimen Collection and Transport System
Regulatory Class: Class I
Product Code: JTW
Dated: February 11, 2002
Received: February 12, 2002

Dear Mr. Stone:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 898. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Part 801); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820); and if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR 1000-1050.

This letter will allow you to begin marketing your device as described in your 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific advice for your device on our labeling regulation (21 CFR Part 801 and additionally 809.10 for in vitro diagnostic devices), please contact the Office of Compliance at (301) 594-4588. Additionally, for questions on the promotion and advertising of your device, please contact the Office of Compliance at (301) 594-4639. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21CFR 807.97). Other general information on your responsibilities under the Act may be obtained from the Division of Small Manufacturers International and Consumer Assistance at its toll-free number (800) 638-2041 or (301) 443-6597 or at its internet address "http://www.fda.gov/cdrh/dsma/dsmamain.html".

Sincerely yours,

Steven Gutman

Steven I. Gutman, M.D., M.B.A.
Director
Division of Clinical Laboratory Devices
Office of Device Evaluation
Center for Devices and
Radiological Health

Enclosure

510(k) Number:

Device Name: Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System

Indications for Use:

The Sierra Diagnostics L.L.C. Urine Collection, Preservation and Transport System is indicated for use in the collection, preservation, and transportation of urine specimens at temperatures not exceeding 60°C for testing with the Abbott LCx® *Neisseria gonorrhoeae* and *Chlamydia trachomatis* assays.

PLEASE DO NOT WRITE BELOW THIS LINE. CONTINUE ON ANOTHER PAGE IN NEEDED

Concurrence of CDRH, Office of Device Evaluation (ODE)

Prescription Use ✓ (Per 21 CFR 801.109)

(Division Sign-Off)
Division of Clinical Laboratory Devices

Over-The Counter Use _____

510(k) Number K013819

foi

5215436 A

Quality Control Page