



Food and Drug Administration
10903 New Hampshire Avenue
Document Control Center – WO66-G609
Silver Spring, MD 20993-0002

Cepheid
Scott Campbell, PhD, MBA
Executive Director, Clinical Affairs
904 Caribbean Drive
Sunnyvale, CA 94089

December 3, 2015

Re: K151226

Trade/Device Name: Xpert[®] Flu+RSV Xpress, Xpert[®] Nasopharyngeal Sample Collection Kit, GeneXpert Xpress System (GX-I)

Regulation Number: 21 CFR 866.3980

Regulation Name: Respiratory viral panel multiplex nucleic acid assay

Regulatory Class: II

Product Code: OCC, OOI, JSM

Dated: May 5, 2015

Received: May 8, 2015

Dear Dr. Campbell:

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. Please note: CDRH does not evaluate information related to contract liability warranties. We remind you, however, that device labeling must be truthful and not misleading.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to 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 Parts 801 and 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); 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.

If you desire specific advice for your device on our labeling regulations (21 CFR Parts 801 and 809), please contact the Division of Industry and Consumer Education at its toll-free number (800) 638 2041 or (301) 796-7100 or at its Internet address <http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm>. Also, please note the regulation entitled, “Misbranding by reference to premarket notification” (21 CFR Part 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to <http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm> for the CDRH’s Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Industry and Consumer Education at its toll-free number (800) 638-2041 or (301) 796-7100 or at its Internet address <http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm>.

Sincerely yours,


Uwe Scherf -S

Uwe Scherf, M.Sc., Ph.D.
Director
Division of Microbiology Devices
Office of In Vitro Diagnostics
and Radiological Health
Center for Devices and Radiological Health

Enclosure

Indications for Use

510(k) Number (if known)
K151226

Device Name
Xpert Flu+RSV Xpress
Xpert® Nasopharyngeal Sample Collection Kit

Indications for Use (Describe)

The Cepheid Xpert Flu+RSV Xpress Assay, performed on the GeneXpert Xpress System, is an automated, multiplex real-time, reverse transcriptase polymerase chain reaction (RT-PCR) assay intended for the in vitro qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA. The Xpert Flu+RSV Xpress Assay uses nasopharyngeal swab specimens collected from patients with signs and symptoms of respiratory infection. The Xpert Flu+RSV Xpress Assay is intended as an aid in the diagnosis of influenza and respiratory syncytial virus infections in conjunction with clinical and epidemiological risk factors.

Negative results do not preclude influenza virus or respiratory syncytial virus infection and should not be used as the sole basis for treatment or other patient management decisions.

Performance characteristics for influenza A were established during the 2014-2015 influenza season. When other novel influenza A viruses are emerging, performance characteristics may vary.

If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health department for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.

The Xpert® Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay or the Xpert Flu/RSV XC Assay. The Xpert® Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu+RSV Xpress Assay.

Type of Use (Select one or both, as applicable)

Prescription Use (Part 21 CFR 801 Subpart D)

Over-The-Counter Use (21 CFR 801 Subpart C)

CONTINUE ON A SEPARATE PAGE IF NEEDED.

This section applies only to requirements of the Paperwork Reduction Act of 1995.

DO NOT SEND YOUR COMPLETED FORM TO THE PRA STAFF EMAIL ADDRESS BELOW.

The burden time for this collection of information is estimated to average 79 hours per response, including the time to review instructions, search existing data sources, gather and maintain the data needed and complete and review the collection of information. Send comments regarding this burden estimate or any other aspect of this information collection, including suggestions for reducing this burden, to:

Department of Health and Human Services
Food and Drug Administration
Office of Chief Information Officer
Paperwork Reduction Act (PRA) Staff
PRASStaff@fda.hhs.gov

"An agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a currently valid OMB number."

5.0 510(k) Summary

As required by 21 CFR Section 807.92(c).

Submitted by: Cepheid
904 Caribbean Drive
Sunnyvale, CA 90489
Phone number: (847) 228-3299
Fax number: (847) 593-0233

Contact: Scott A. Campbell, PhD, MBA

Date of Preparation: May 5, 2015

Device:

Trade name: Xpert[®] Flu+RSV Xpress

Common name: Xpert Flu+RSV Xpress Assay

Type of Test: Automated, multiplex real-time reverse transcription-polymerase chain reaction (RT-PCR) assay intended for the *in vitro* qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus.

Regulation number/
Classification name/ 866.3980/Respiratory viral panel multiplex nucleic acid assay/
866.2570/Instrumentation for clinical multiplex test systems

Product code(s): 866.2390/Transport culture medium
OCC, OOI, JSM

Classification Class II

Advisory Panel Microbiology (83)

Prescription Use Yes

Predicate Devices
Name(s): 1) *For the detection and differentiation of influenza A, influenza B, and RSV viral RNA in nasopharyngeal swab specimens:*
Cepheid Xpert Flu/RSV XC [510(k) #K142045]; and,

2) *For the Sample Collection Kits:*
Cepheid Xpert Nasopharyngeal Sample Collection Kit [510(k) #K142045]

Device Description:

The Xpert Flu+RSV Xpress Assay is an automated *in vitro* diagnostic test for qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV). The assay is performed on the Cepheid GeneXpert Xpress System (GeneXpert Dx System, GX-I). The GeneXpert Xpress System platform automates and integrates sample extraction, purification, amplification, and detection of the target sequence in simple or complex samples using real-time PCR and reverse transcriptase PCR (RT-PCR) assays. The systems require the use of single-use disposable cartridges (the Xpert Flu+RSV Xpress cartridges) that hold the PCR reagents and host the PCR process. Because the cartridges are self-contained and specimens never come into contact with working parts of the instrument modules, cross-contamination between samples is minimized.

The Xpert Flu+RSV Xpress Assay includes reagents for the detection and differentiation of influenza A, influenza B, and RSV viral RNA directly from nasopharyngeal (NP) swab specimens collected from patients with signs and symptoms of respiratory infection. A Sample Processing Control (SPC) and a Probe Check Control (PCC) are also included in the cartridge. The SPC is present to control for adequate processing of the target viruses and to monitor the presence of inhibitors in the PCR reaction. The Probe Check Control (PCC) verifies reagent rehydration, PCR tube filling in the cartridge, probe integrity, and dye stability.

The single-use, multi-chambered fluidic cartridges are designed to complete sample preparation and real-time RT-PCR for detection and differentiation of influenza A, influenza B and RSV viral RNA in approximately 60 minutes. The GeneXpert Xpress System, comprised of the GeneXpert Dx System GX-I, has one module that is capable of performing separate sample preparation and real-time PCR and RT-PCR tests. Each module contains a syringe drive for dispensing fluids (i.e., the syringe drive activates the plunger that works in concert with the rotary valve in the cartridge to move fluids between chambers), an ultrasonic horn for lysing cells or spores, and a proprietary I-CORE[®] thermocycler for performing real-time PCR and RT-PCR and detection.

Specimens are collected following the instructions for collecting NP swab specimens provided in Xpert Flu+RSV Xpress Assay package insert for influenza and RSV testing. The Cepheid Xpert Nasopharyngeal Sample Collection Kit (Cepheid catalog #SWAB/B-100) is required but not provided for use with the assay. The NP swab specimen is placed in the Xpert viral transport medium and sent to the GeneXpert[®] Xpress testing area for processing. When stored in the transport medium, the NP swab specimen is stable for up to 24 hours at 2–30 °C or up to seven days at 2–8 °C. When ready to test the specimen, the user briefly mixes the specimen by inverting the tube five times, transfers the eluted material to the sample chamber in the top of the disposable fluidic cartridge. The user initiates a test from the system user interface and places the cartridge into the GeneXpert Xpress instrument platform, which performs hands-off real-time, multiplex polymerase chain reaction (PCR) for detection of RNA. The results are automatically generated at the end of the process in a report that can be viewed and printed.

Device Intended Use:

Xpert Flu+RSV Xpress Assay:

The Cepheid Xpert[®] Flu+RSV Xpress Assay, performed on the GeneXpert[®] Xpress System, is an automated, multiplex real-time, reverse transcriptase polymerase chain reaction (RT-PCR) assay intended for the *in vitro* qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA. The Xpert Flu+RSV Xpress Assay uses nasopharyngeal swab specimens collected from patients with signs and symptoms of respiratory infection. The Xpert Flu+RSV Xpress Assay is intended as an aid in the diagnosis of influenza and respiratory syncytial virus in conjunction with clinical and epidemiological risk factors.

Negative results do not preclude influenza virus or respiratory syncytial virus infection and should not be used as the sole basis for treatment or other patient management decisions.

Performance characteristics for influenza A were established during the 2014-2015 influenza season. When other novel influenza A viruses are emerging, performance characteristics may vary.

If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health departments for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.

Ancillary Collection Kits' Indications for Use (the expanded indication is shown in bold):

Xpert Nasopharyngeal Sample Collection Kit

The Xpert[®] Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay or the Xpert Flu/RSV XC Assay. **The Xpert[®] Nasopharyngeal Sample Collection Kit is designed to collect, preserve, and transport nasopharyngeal swab specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu+RSV Xpress Assay.**

Substantial Equivalence:

The Xpert Flu+RSV Xpress Assay is substantially equivalent to the Cepheid Xpert Flu/RSV XC Assay [510(k) #K142045]. The Xpert Flu+RSV Xpress Assay and the Xpert Flu/RSV XC Assay detect influenza A, influenza B, and RSV from NP swab specimens. Both assays utilize the same technology by determining the presence of the target organisms through real-time RT-PCR amplification and fluorogenic target-specific hybridization detection. A multi-

center clinical study was conducted to determine the performance characteristics of the device with collection of NP swab specimens from each subject relative to a comparator device, the ProFlu+ Assay which is FDA cleared for NP swab specimens. Discordant results between the Xpert Flu+RSV Xpress Assay and the ProFlu+ Assay were analyzed by sequencing using primers different from those used in the Xpert Flu+RSV Xpress Assay. The study results showed the Xpert Flu+RSV Xpress Assay is acceptable for its intended use with inexperienced lab users.

Table 5-1 shows the similarities and differences between the Xpert Flu+RSV Xpress Assay and the predicate assay, Xpert Flu/RSV XC Assay.

Table 5-1: Comparison of Similarities and Differences of the Xpert Flu+RSV Xpress Assay with the Predicate Device

Similarities		
Item	Device	Predicate Device
	Cepheid Xpert Flu+RSV Xpress Assay	Cepheid Xpert Flu/RSV XC Assay
510(k) Number	K151226	K142045
Regulation	866.3980	866.3980
Product Code	OCC, OOI	OCC, OOI
Device Class	Same	II
Technology Principle of Operation	Same	Multiplex real time RT-PCR

Similarities		
Item	Device	Predicate Device
	Cepheid Xpert Flu+RSV Xpress Assay	Cepheid Xpert Flu/RSV XC Assay
Intended Use	<p>The Cepheid Xpert Flu+RSV Xpress Assay, performed on the GeneXpert Xpress System, is an automated, multiplex real-time, reverse transcriptase polymerase chain reaction (RT-PCR) assay intended for the <i>in vitro</i> qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA. The Xpert Flu+RSV Xpress Assay uses nasopharyngeal swab specimens collected from patients with signs and symptoms of respiratory infection. The Xpert Flu+RSV Xpress Assay is intended as an aid in the diagnosis of influenza and respiratory syncytial virus in conjunction with clinical and epidemiological risk factors.</p> <p>Negative results do not preclude influenza virus or respiratory syncytial virus infection and should not be used as the sole basis for treatment or other patient management decisions.</p> <p>Performance characteristics for influenza A were established during the 2014-2015 influenza season. When other novel influenza A viruses are emerging, performance characteristics may vary.</p>	<p>The Cepheid Xpert Flu/RSV XC Assay is an automated, multiplex real-time, reverse transcriptase polymerase chain reaction (RT-PCR) assay intended for the <i>in vitro</i> qualitative detection and differentiation of influenza A, influenza B, and respiratory syncytial virus (RSV) viral RNA. The Xpert Flu/RSV XC Assay uses nasopharyngeal swab and nasal aspirate/wash specimens collected from patients with signs and symptoms of respiratory infection in conjunction with clinical and epidemiological risk factors. The Xpert Flu/RSV XC Assay is intended as an aid in the diagnosis of influenza and respiratory syncytial virus.</p> <p>Negative results do not preclude influenza virus or respiratory syncytial virus infection and should not be used as the sole basis for treatment or other patient management decisions.</p> <p>Performance characteristics for influenza A were established during the 2013-2014 influenza season. When other novel influenza A viruses are emerging, performance characteristics may vary.</p>

Similarities		
Item	Device	Predicate Device
	Cepheid Xpert Flu+RSV Xpress Assay	Cepheid Xpert Flu/RSV XC Assay
Intended Use	If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health department for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.	If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health department for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.
Indication for Use	Same	Patients with signs and symptoms of respiratory infection in conjunction with clinical and epidemiological risk factors.
Assay Targets	Same	Influenza A Virus, Influenza B Virus, and RSV viral RNA
Specimen Types	Nasopharyngeal (NP) swab specimens	Nasopharyngeal (NP) swab specimens and Nasal aspirate/wash (NA/W) specimens
Nucleic Acid Extraction	Yes	Yes
Extraction Methods	Sample preparation integrated in GeneXpert Cartridge and GeneXpert Xpress System	Sample preparation integrated in GeneXpert Cartridge and GeneXpert Instrument System
Assay Results	Same	Qualitative
Instrument System	Cepheid GeneXpert Xpress System (instrument model GX-I) ; same Cepheid I-core technology	Cepheid GeneXpert Instrument Systems (various instrument models including instrument model GX-I); Cepheid I-core technology

Similarities		
Item	Device	Predicate Device
	Cepheid Xpert Flu+RSV Xpress Assay	Cepheid Xpert Flu/RSV XC Assay
Assay Controls	Same	Encapsulated (armored) RNA pseudovirus as a sample processing control. Available but not provided are inactivated virus controls for influenza A/B and RSV as external positive controls, and Coxsackie virus as an external negative control.
Time to obtain test results	Approximately 60 minutes for sample preparation and real-time RT-PCR.	Approximately 60 minutes or less for sample preparation and real-time RT-PCR.
Primers and probes	Same	Primers and probes to detect the presence of nucleic acid sequences of influenza A, influenza B, and RSV.

Primary Differences (Differences are also Captured in the Similarities Table above)		
	New Device	Predicate Device
Item	Cepheid Xpert Flu+RSV Xpress Assay	Cepheid Xpert Flu/RSV XC Assay
Instrument System	Cepheid GeneXpert Xpress System	Cepheid GeneXpert Dx Systems and GeneXpert Infinity Systems
Laboratory Users	Untrained operators with no clinical lab experience in a CLIA-waiver environment.	Operators with no clinical lab experience to experienced clinical laboratory technologists.
Combinatorial Assay Selections	No combinatorial assay selections are available.	Yes, user may select combined assay with all targets or a Flu only assay or a RSV only assay.
Early assay termination function	No early assay termination function is available.	Yes, on Flu only or RSV only assay selections.

The Xpert Flu+RSV Xpress Assay has the same general intended use as the predicate device and has the same technological characteristics as the predicate device. The differences between the Xpert Flu+RSV Xpress Assay and the predicate device do not raise different questions of safety and effectiveness. The clinical study demonstrates that the Xpert Flu+RSV Xpress Assay is acceptable for its intended use with inexperienced laboratory users and is substantially equivalent to the predicate device described above.

Xpert Nasopharyngeal Sample Collection Kit

The predicate device for the ancillary specimen collection kit, the Xpert[®] Nasopharyngeal Sample Collection Kit, is the Cepheid Nasopharyngeal Sample Collection Kit, [510(k) # K042970]. The similarities and differences are shown in Table 5-2.

Table 5-2: Comparison of Similarities and Differences of the Xpert Nasopharyngeal Sample Collection Kit with the Predicate Device

Similarities		
Item	Device	Predicate
	Xpert Nasopharyngeal Sample Collection Kit	Xpert Nasopharyngeal Sample Collection Kit
Intended Use (Similarities)	<p>For collection, preservation and transport of nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay and the Xpert Flu/RSV XC Assay.</p> <p>For collection, preservation and transport of nasopharyngeal swab specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu+RSV Xpress Assay.</p>	<p>For collection, preservation and transport of nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay and the Xpert Flu/RSV XC Assay.</p>
Single-use Device	Yes	Yes
Medium Formulation	Same	Hank's Balanced Salt Solution Bovine Serum Albumin L-cysteine Gelatin Sucrose L-glutamic acid HEPES buffer Vancomycin Amphotericin B Colistin Phenol red
pH	Same	7.3 ± 0.2

Similarities		
Item	Device	Predicate
	Xpert Nasopharyngeal Sample Collection Kit	Xpert Nasopharyngeal Sample Collection Kit
Storage Temperature	Same	2 - 25°C (refrigerated and room temperature)
Volume	Same	3 ml
Glass Beads	Same	3 x 3 mm
Container	Same	Plastic (medical-grade polypropylene)
Product Configuration	Same	Medium Tube in Kit with individually-wrapped sterile swab.
Differences		
Item	Device	Predicate
Intended Use (differences)	For collection, preservation and transport of nasopharyngeal swab specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu+RSV Xpress Assay.	For collection, preservation and transport of nasopharyngeal swab specimens and to preserve and transport nasal aspirate/wash specimens containing viruses from patients with signs and symptoms of respiratory infection prior to analysis with the Xpert Flu Assay and the Xpert Flu/RSV XC Assay.

Both devices have the same general intended use and use the same technology to collect, store, and transport clinical specimens, including viruses, to the laboratory for further testing. The differences between the two devices do not raise new or different questions of safety and effectiveness. The multi-center clinical study of the Xpert Flu+RSV Xpress Assay was conducted using Xpert Nasopharyngeal Sample Collection Kit and demonstrated that the assay and its collection kit are acceptable for their intended use with inexperienced lab users and substantially equivalent to the predicate device.

Non-Clinical Studies:

Analytical Sensitivity (Limit of Detection)

Studies were performed to determine the analytical limit of detection (LoD) of the Xpert Flu+RSV Xpress Assay with two lots of reagents across three testing days. The higher LoD observed per strain and per lot was selected for verification. Verification of the estimated LoD claim was performed on one reagent lot across a minimum of three testing days. LoD was established using two influenza A H3N2 strains, two influenza A 2009 H1N1 strains, two influenza B strains, two respiratory syncytial virus A (RSV A) strains, two respiratory syncytial virus B (RSV B) strains, and one influenza A H7N9 strain diluted into a negative pooled clinical matrix. The LoD is defined as the lowest concentration (tissue culture infective dose, TCID₅₀/mL) per sample that can be reproducibly distinguished from negative samples with 95% confidence or the lowest concentration at which 19 of 20 replicates were positive. Each strain was tested in replicates of 20 per concentration of virus.

The LoD was determined empirically as the first concentration that had 19/20 or 20/20 positive results. The LoD point estimates for each strain tested are summarized in Tables 5-3 to 5-8.

Table 5-3: Confirmed LoD (TCID₅₀/mL): Influenza A 2009 H1N1

Strain ID	Confirmed LoD (TCID₅₀/mL) (at least 19/20 positive)
Influenza A/California/7/2009	0.3 (20/20)
Influenza A/Florida/27/2011	16.0 (20/20)

Table 5-4: Confirmed LoD (TCID₅₀/mL): Influenza A H3N2

Strain ID	Confirmed LoD (TCID₅₀/mL) (at least 19/20 positive)
Influenza A/Perth/16/2009	0.3 (20/20)
Influenza A/Victoria/361/2011	0.8 (20/20)

Table 5.5: Confirmed LoD (TCID₅₀/mL): Influenza B

Strain ID	Confirmed LoD (TCID₅₀/mL) (at least 19/20 positive)
Influenza B/Massachusetts/2/2012	0.5 (20/20)
Influenza B/Wisconsin/01/2011	0.6 (20/20)

Table 5-6: Confirmed LoD (TCID₅₀/mL): Respiratory Syncytial Virus A

Strain ID	Confirmed LoD (TCID ₅₀ /mL) (at least 19/20 positive)
RSV A/2/Australia/61	1.2 (20/20)
RSV A/Long/MD/56	1.0 (20/20)

Table 5-7: Confirmed LoD (TCID₅₀/mL): Respiratory Syncytial Virus B

Strain ID	Confirmed LoD (TCID ₅₀ /mL) (at least 19/20 positive)
RSV B/Washington/18537/62	1.8 (20/20)
RSV B/9320/Massachusetts/77	2.0 (20/20)

Table 5-8: Confirmed LoD (TCID₅₀/mL): Influenza A H7N9

Strain ID	Confirmed LoD (TCID ₅₀ /mL) (at least 19/20 positive)
Influenza A/Anhui/1/2013	0.8 (19/20)

Although this test has been shown to detect the novel avian influenza A (H7N9) cultured material, the performance characteristics of this device with clinical specimens that are positive for the novel avian influenza A (H7N9) virus have not been established. The Xpert Flu+RSV Xpress Assay can distinguish between influenza A and B viruses, but it cannot differentiate influenza A subtypes.

Analytical Specificity (Exclusivity)

The analytical specificity of the Xpert Flu+RSV Xpress Assay was evaluated by testing a panel of 44 cultures consisting of 16 viral, 26 bacterial, and two yeast strains representing common respiratory pathogens or those potentially encountered in the nasopharynx.

Three replicates of all bacterial and yeast strains were tested at concentrations of $\geq 10^6$ CFU/mL with the exception of one strain which was tested at 10^5 CFU/mL (*Chlamydia pneumoniae*). Three replicates of each virus were tested at concentrations of $\geq 10^5$ TCID₅₀/mL. The analytical specificity was 100%. Results are shown in Table 5-9.

Table 5-9: Analytical Specificity of Xpert Flu+RSV Xpress Assay

Organism	Concentration	Influenza A	Influenza B	RSV
No Template Control		NEG	NEG	NEG
Adenovirus Type 1	1.12x10 ⁷ TCID ₅₀ /mL	NEG	NEG	NEG
Adenovirus Type 7	1.87x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
Human coronavirus OC43	2.85x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
Human coronavirus 229E	1x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
Cytomegalovirus	7.24x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
Echovirus	3.31x10 ⁷ TCID ₅₀ /mL	NEG	NEG	NEG
Enterovirus	1x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
Epstein Barr Virus	7.16x10 ⁷ TCID ₅₀ /mL	NEG	NEG	NEG
HSV	8.9x10 ⁶ TCID ₅₀ /mL	NEG	NEG	NEG
Measles	6.3x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
Human metapneumovirus	3.8x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
Mumps virus	6.31x10 ⁶ TCID ₅₀ /mL	NEG	NEG	NEG
Human parainfluenza Type 1	1.15x10 ⁶ TCID ₅₀ /mL	NEG	NEG	NEG
Human parainfluenza Type 2	1x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
Human parainfluenza Type 3	3.55x10 ⁷ TCID ₅₀ /mL	NEG	NEG	NEG
Rhinovirus Type 1A	1.26x10 ⁵ TCID ₅₀ /mL	NEG	NEG	NEG
<i>Acinetobacter baumannii</i>	>1x10 ⁶ CFU/mL	NEG ^a	NEG	NEG
<i>Burkholderia cepacia</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Candida albicans</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Candida parapsilosis</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Bordetella pertussis</i>	1x10 ⁸ CFU/mL	NEG	NEG	NEG
<i>Chlamydia pneumoniae</i>	3.16x10 ⁵ CFU/mL	NEG	NEG	NEG

Organism	Concentration	Influenza A	Influenza B	RSV
<i>Citrobacter freundii</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Corynebacterium sp.</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Escherichia coli</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Enterococcus faecalis</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Haemophilus influenzae</i>	1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Lactobacillus reuter</i>	1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Legionella spp.</i>	1x10 ⁸ CFU/mL	NEG	NEG	NEG
<i>Moraxella catarrhalis</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Mycobacterium tuberculosis (avirulent)</i>	1.15x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Mycoplasma pneumoniae</i>	1x10 ⁷ CFU/mL	NEG	NEG	NEG
<i>Neisseria meningitidis</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Neisseria mucosa</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Propionibacterium acnes</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Pseudomonas aeruginosa</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Staphylococcus aureus</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Staphylococcus epidermidis</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Staphylococcus haemolyticus</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Streptococcus agalactiae</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Streptococcus pneumoniae</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Streptococcus pyogenes</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Streptococcus salivarius</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG
<i>Streptococcus sanguinis</i>	>1x10 ⁶ CFU/mL	NEG	NEG	NEG

Analytical Reactivity (Inclusivity)

The analytical reactivity of the Xpert Flu+RSV Xpress Assay was evaluated against multiple strains of influenza A H1N1 (seasonal pre-2009), influenza A H1N1 (pandemic 2009), influenza A H3N2 (seasonal), avian influenza A (H5N1, H5N2, H6N2, H7N2, H7N3, H2N2, H7N9, and H9N2), influenza B (representing strains from both Victoria and

Yamagata lineages), and respiratory syncytial virus subgroups A and B (RSV A and RSV B) at levels near the analytical LoD. A total of 64 strains including 54 influenza viruses and 10 RSV strains were tested in this study with the Xpert Flu+RSV Xpress Assay.

Three replicates were tested for each strain. Results are shown in Table 5-10.

Table 5-10: Analytical Reactivity (Inclusivity) of Xpert Flu+RSV Xpress Assay

Virus	Strain	Concentration	Result		
			Flu A	Flu B	RSV
No Template Control			NEG	NEG	NEG
Influenza A H1N1 (pre- 2009)	A/swine/Iowa/15/30	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/WS/33	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/PR/8/34	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/Mal/302/54	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/Denver/1/57	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/New Jersey/8/76	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/New Caledonia/20/1999	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/New York/55/2004	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/Soloman Islands/3/2006	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/Taiwan/42/06	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/Brisbane/59/2007	32.0 TCID ₅₀ /mL	POS	NEG	NEG
Influenza A H1N1 (pdm2009)	A/California/7/2009	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/swine/NY/02/2009	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/Florida/27/2011	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/Colorado/14/2012	32.0 TCID ₅₀ /mL	POS	NEG	NEG
	A/Washington/24/2012	80.0 ^a TCID ₅₀ /mL	POS	NEG	NEG
Influenza A H3N2 (Seasonal)	A/Aichi/2/68	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/HongKong/8/68	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Port Chalmers/1/73	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Hawaii/15/2001	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Wisconsin/67/05	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Brisbane/10/2007	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Perth/16/2009	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Minnesota/11/2010 (H3N2)v	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Indiana/08/2011 (H3N2)v	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Victoria/361/2011	1.6 TCID ₅₀ /mL	POS	NEG	NEG
	A/Texas/50/2012	1.6 TCID ₅₀ /mL	POS	NEG	NEG

Virus	Strain	Concentration	Result		
			Flu A	Flu B	RSV
Avian influenza A	A/duck/Hunan/795/2002 (H5N1)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/chicken/Hubei/327/2004 (H5N1)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/Anhui/01/2005 (H5N1)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/Japanese white eye/HongKong/1038/2006 (H5N1)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/mallard/WI/34/75 (H5N2)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/chicken/CA431/00 (H6N2)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/duck/LTC-10-82743/1943 (H7N2)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/chicken/NJ/15086-3/94 (H7N3)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/Anhui/1/2013 (H7N9)	N/A ^c	POS	NEG	NEG
	A/Shanghai/1/2013 (H7N9)	N/A ^c	POS	NEG	NEG
	A/chicken/Korea/38349-p96323/ 1996 (H9N2)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
	A/mallard/NY/6750/78 (H2N2)	$\leq 1\text{pg}/\mu\text{L}^b$	POS	NEG	NEG
Influenza B	B/Lee/40	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Allen/45	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/GL/1739/54	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Maryland/1/59	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Panama/45/90 ^d	3.0 TCID ₅₀ /mL ^e	NEG	POS	NEG
	B/Florida/07/2004 ^f	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Florida/02/06 ^d	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Florida/04/06 ^f	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Wisconsin/01/2011 ^d	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Massachusetts/2/2012 ^f	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Hong Kong/5/72	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Wisconsin/01/2010 ^f	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Malaysia/2506/04 ^d	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Taiwan/2/62	1.2 TCID ₅₀ /mL	NEG	POS	NEG
	B/Brisbane/60/2008 ^d	1.2 TCID ₅₀ /mL	NEG	POS	NEG
RSV A	RSV-A/Long/MD/56	2.4 TCID ₅₀ /mL	NEG	NEG	POS

Virus	Strain	Concentration	Result		
			Flu A	Flu B	RSV
	RSV-A/2/Australia/61	2.4 TCID ₅₀ /mL	NEG	NEG	POS
	RSV-A/NY (Clinical unknown)	2.4 TCID ₅₀ /mL	NEG	NEG	POS
	RSV-A/WI/629-8-2/2007	2.4 TCID ₅₀ /mL	NEG	NEG	POS
	RSV-A/WI/629-11-1/2008	2.4 TCID ₅₀ /mL	NEG	NEG	POS
	RSV B				
	RSV-B/Wash/18537/62	4.0 TCID ₅₀ /mL	NEG	NEG	POS
	RSV-B/9320/MA/77	4.0 TCID ₅₀ /mL	NEG	NEG	POS
	RSV-B/WV14617/85	4.0 TCID ₅₀ /mL	NEG	NEG	POS
	RSV-B/CH93(18)-18	20.0 TCID ₅₀ /mL ^g	NEG	NEG	POS
	RSV-B/WI/629-5B/0607	4.0 TCID ₅₀ /mL	NEG	NEG	POS

^aInfluenza A/Washington/24/2012 was tested at 5X LoD (80.0 TCID₅₀/mL) to obtain 3 of 3 Flu A POSITIVE result calls.

^bPurified viral RNA in simulated background matrix was used for avian influenza A viruses due to biosafety regulations.

^cInactivated avian influenza A (H7N9) viruses without viral titer was diluted 100,000 fold in simulated background matrix and tested due to biosafety regulations.

^dKnown Victoria lineage.

^eInfluenza B/Panama/45/90 was tested at 5X LoD (3.0 TCID₅₀/mL) to obtain 3/3 Flu B POSITIVE result calls.

^fKnown Yamagata lineage.

^gRSV-B/CH93(18)-18 was tested at 10X LoD (20.0 TCID₅₀/mL) to obtain 3/3 RSV POSITIVE result calls.

Potentially Interfering Substances

In a non-clinical study, potentially interfering substances that may be present in the nasopharynx were evaluated directly relative to the performance of the Xpert Flu+RSV Xpress Assay. Potentially interfering substances in the nasopharynx may include, but are not limited to: blood, nasal secretions or mucus, and nasal and throat medications used to relieve congestion, nasal dryness, irritation, or asthma and allergy symptoms, as well as antibiotics and antivirals. Negative samples (n = 8) were tested per each substance to determine the effect on the performance of the sample processing control (SPC). Positive samples (n = 8) were tested per substance with six influenza (four influenza A and two influenza B) and four RSV (two RSV A and two RSV B) strains spiked at 2X the analytical LoD determined for each strain. All results were compared to positive and negative Universal Transport Medium (UTM) controls.

These evaluated substances are listed in Table 5.11 with active ingredients and concentrations tested shown. There was no assay interference in the presence of the substances at the concentrations tested in this study. All positive and negative replicates were correctly identified using the Xpert Flu+RSV Xpress Assay.

FluMist vaccine samples were correctly reported as **Flu A POSITIVE; FLU B POSITIVE; RSV NEGATIVE** as expected. Samples containing FluMist may cause false positive results. This is addressed in the device labeling Section 17, Limitations.

Table 5-11. Potentially Interfering Substances in Xpert Flu+RSV Xpress Assay

Substance/Class	Description/Active Ingredient	Concentration Tested
Beta-adrenergic bronchodilator	Albuterol Sulfate	0.83 mg/mL (equivalent to 1 dose per day)
Blood	Blood (Human)	2% (v/v)
BD™ Universal Viral Transport System	Transport Media	100% (v/v)
Remel M4	Transport Media	100% (v/v)
Remel M4RT	Transport Media	100% (v/v)
Remel M5®	Transport Media	100% (v/v)
Throat lozenges, oral anesthetic and analgesic	Benzocaine, Menthol	1.7 mg/mL
Mucin	Purified Mucin protein (Bovine or porcine submaxillary gland)	2.5% (w/v)
Antibiotic, nasal ointment	Mupirocin	10 mg/mL
Saline Nasal Spray	Sodium Chloride (0.65%)	15% (v/v)
Anefrin Nasal	Oxymetazoline, 0.05%	15% (v/v)

Substance/Class	Description/Active Ingredient	Concentration Tested
Spray		
Nasal Drops	Phenylephrine, 0.5%	15% (v/v)
Tamiflu [®] /Anti-viral drugs	Zanamivir	7.5 mg/mL
Antibacterial, systemic	Tobramycin	4 µg/mL
Zicam [®] /Nasal Gel	Luffa operculata, Galphimia glauca, Histaminum hydrochloricum Sulfur	15% (w/v)
FluMist [®]	Live intranasal influenza virus vaccine	6.7% (v/v)
Nasal corticosteroid	Fluticasone Propionate	5 µg/mL

Carry-Over Contamination

A study was conducted to demonstrate that single-use, self-contained GeneXpert cartridges prevent carry-over contamination in negative samples run following very high positive samples in the same GeneXpert module. The study consisted of a negative sample processed in the same GeneXpert module immediately following a very high influenza A sample (approximately 10^6 TCID₅₀/test) or a very high RSV A sample (approximately 10^6 TCID₅₀/test). This testing scheme was repeated 20 times on two GeneXpert modules for a total of 82 runs resulting in 40 positive and 42 negative specimens for each virus type. All 40 positive samples were correctly reported as **Flu A POSITIVE; Flu B NEGATIVE; RSV NEGATIVE** or **Flu A NEGATIVE; Flu B NEGATIVE; RSV POSITIVE**. All 42 negative samples were correctly reported as **Flu A NEGATIVE; Flu B NEGATIVE; RSV NEGATIVE**.

Fresh vs. Frozen Sample Equivalency Study

Fresh and frozen specimen equivalency in the Xpert Flu+RSV Xpress Assay was evaluated by testing individual influenza and RSV strains at three different concentrations representing low positives (2X LoD), moderate positives (5X LoD), and high positives (10X LoD) in simulated background matrix. Negative samples consisted of simulated background matrix only. Fresh and frozen specimen equivalency was determined using one seasonal Flu A H3N2 strain (A/Victoria/361/2011), one Flu B strain (B/Wisconsin/01/11), one RSV A strain (RSV A/Long/MD/56), and one RSV B strain (RSV B/9320/MA/77). Replicates of 20 were tested for each specimen type and concentration. All positive and negative specimens were tested fresh, after one freeze-thaw cycle, and after two freeze-thaw cycles.

There was no statistically significant effect in the performance of the Xpert Flu+RSV Xpress Assay between fresh virus dilutions and two sequential freeze thaw cycles for positive and negative samples. All positive and negative replicates were correctly identified using the Xpert Flu+RSV Xpress Assay.

Linearity

Not applicable, the Xpert Flu+RSV Xpress Assay is a qualitative assay.

Clinical Studies

Clinical Comparison Study

Performance characteristics of the Xpert Flu+RSV Xpress Assay were evaluated at 12 institutions in the U.S. during the 2014-2015 influenza season. Due to the low prevalence of Flu B and RSV during the sampling time frame of this study, supplementation with pre-selected archived NP swab specimens known to be positive for Flu B or RSV was required to meet the sample size for these targets.

Subjects included individuals with signs or symptoms of respiratory infection and whose routine care called for collection of NP swab specimens for influenza and/or RSV testing. For eligible subjects, NP swab specimens were obtained for testing with the Xpert Flu+RSV Xpress Assay and the comparator assay, and patient management continued at the site per their standard practice.

The Xpert Flu+RSV Xpress Assay performance was compared to a FDA-cleared molecular comparator assay. Xpert Flu+RSV Xpress Assay results from NP swab specimens were compared to the molecular comparator assay result from the same swab specimens. Bi-directional sequencing was performed on specimens where the Xpert Flu+RSV Xpress Assay and the comparator assay were discrepant, and is provided for informational purposes only.

NP Swab Specimens

A total of 2435 NP swab specimens were tested for influenza A, influenza B and RSV by the Xpert Flu+RSV Xpress Assay and the comparator assay. Of the 2435 NP swab specimens 2176 were fresh, prospectively collected and 259 were pre-selected frozen, archived specimens.

On fresh, prospectively collected NP swab specimens, the Xpert Flu+RSV Xpress Assay demonstrated a positive percent agreement (PPA) and negative percent agreement (NPA) for detection of influenza A of 100% and 94.8%, respectively, relative to the comparator assay (Table 5-12). The Xpert Flu+RSV Xpress Assay PPA and NPA for influenza B were 100% and 99.5%, respectively (Table 5-12). The Xpert Flu+RSV Xpress Assay PPA and NPA for RSV were 96.9% and 99.6%, respectively (Table 5-12).

On pre-selected frozen, archived NP swab specimens, the Xpert Flu+RSV Xpress Assay demonstrated an NPA for detection of influenza A of 98.5% relative to the comparator assay (Table 5-12). The Xpert Flu+RSV Xpress Assay PPA and NPA for influenza B were 100% and 98.7%, respectively (Table 5-12). The Xpert Flu+RSV Xpress Assay PPA and NPA for RSV were 97.6% and 99.5%, respectively (Table 5-12).

Table 5-12: Xpert Flu+RSV Xpress Assay Performance on NP Swab Specimens

Specimen Type	Target	n	TP	FP	TN	FN	PPA % (95 CI)	NPA % (95 CI)
Fresh	Flu A	2176	250	101 ^a	1825	0	100 (98.5-100)	94.8 (93.7-95.7)

	Flu B	2176	63	10 ^b	2103	0	100 (94.3-100)	99.5 (99.1-99.8)
	RSV	2176	125	8 ^c	2039	4 ^d	96.9 (92.3-99.1)	99.6 (99.2-99.8)
Pre-selected Frozen	Flu A	259	0	4 ^e	255	0	NA	98.5 (96.1-99.6)
	Flu B	259	100	2 ^f	157	0	100 (96.4-100)	98.7 (95.5-99.8)
	RSV	259	40	1 ^g	217	1 ^h	97.6 (87.1-99.9)	99.5 (97.5-100)

- a. Testing results by sequencing: 92 of 101 were Flu A positive; 8 of 101 failed to sequence; 1 of 101 insufficient remaining volume for sequencing.
- b. Testing results by sequencing: 9 of 10 were Flu B positive; 1 of 10 insufficient remaining volume for sequencing.
- c. Testing results by sequencing: 7 of 8 were RSV positive; 1 of 8 was RSV negative.
- d. Testing results by sequencing: 3 of 4 were RSV positive; 1 of 4 was RSV negative.
- e. Testing results by sequencing: 4 of 4 insufficient remaining volume for sequencing.
- f. Testing results by sequencing: 2 of 2 insufficient remaining volume for sequencing.
- g. Testing results by sequencing: 1 of 1 insufficient remaining volume for sequencing.
- h. Testing results by sequencing: 1 of 1 insufficient remaining volume for sequencing.

Of the Xpert Flu+RSV Xpress Assay runs performed with eligible specimens, 95.0% (2335/2459) of these specimens were successful on the first attempt. The initial invalid rate was 5.0% (95% CI 4.2-6.0%). One-hundred twenty-four gave invalid results on the first attempt (121 NO RESULT-REPEAT TEST and 3 INSTRUMENT ERROR). One-hundred eighteen of the 124 specimens were retested, of which 107 yielded valid results after a single retest. There were 17 NP swab specimens with invalid results upon retest which were excluded in the analyses.

Reproducibility Study

A panel of 10 specimens with varying concentrations of influenza A, influenza B, and RSV was tested on ten different days by three different operators, at each of three sites (10 specimens x 1 time/day x 10 days x 3 operators x 3 sites). One lot of Xpert Flu+RSV Xpress Assay cartridges was used at each of the 3 testing sites. The Xpert Flu+RSV Xpress Assay was performed according to the Xpert Flu+RSV Xpress Assay procedure. Results are summarized in Table 5-13.

Table 5-13: Summary of Reproducibility Results

Sample ID	Site 1			Site 2			Site 3			% Total Agreement by Sample ^a
	Op 1	Op 2	Op 3	Op 1	Op 2	Op 3	Op 1	Op 2	Op 3	
Negative	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (9/9) ^b	100% (9/9) ^b	100% (10/10)	100% (10/10)	100% (88/88) ^b

Sample	Site 1			Site 2			Site 3			% Total
	Flu A-High Neg	0.0% (0/10)	20.0% (2/10)	60.0% (6/10)	20.0% (2/10)	10.0% (1/10)	50.0% (5/10)	75.0% (6/8) ^b	20.0% (2/10)	
Flu A-Low Pos	100% (10/10)	100% (10/10)	90.0% (9/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	98.9% (89/90)
Flu A-Mod Pos	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (90/90)
Flu B-High Neg	50.0% (5/10)	0.0% (0/10)	50.0% (5/10)	20.0% (2/10)	10.0% (1/10)	55.6% (5/9) ^b	80.0% (8/10)	50.0% (5/10)	20.0% (2/10)	37.1% (33/89) ^b
Flu B-Low Pos	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	90.0% (9/10)	100% (10/10)	100% (10/10)	98.9% (89/90)
Flu B-Mod Pos	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (90/90)
RSV-High Neg	60.0% (6/10)	30.0% (3/10)	66.7% (6/9) ^b	50.0% (5/10)	60.0% (6/10)	50.0% (5/10)	90.0% (9/10)	33.3% (3/9) ^b	50.0% (5/10)	54.5% (48/88) ^b
RSV-Low Pos	88.9% (8/9)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	90.0% (9/10)	100% (10/10)	100% (10/10)	97.8% (87/89) ^c
RSV-Mod Pos	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (10/10)	100% (90/90)

a. Percent of samples yielding expected results – negative for Neg and High Neg samples; positive for Low Pos and Mod Pos samples.

b. Seven samples (2 Neg, 2 Flu A High Neg, 1 Flu B High Neg, and 2 RSV High Neg) were indeterminate upon initial and retest.

c. One RSV Low Pos sample was inadvertently not tested.

The reproducibility of the Xpert Flu+RSV Xpress Assay was also evaluated in terms of the fluorescence signal expressed in Ct values for each target detected. The mean, standard deviation (SD), and coefficient of variation (CV) between-sites, between-days, and between-operators for each panel member are presented in Table 5-14. One replicate was performed per day per operator, therefore, operator and assay (within-run) precision are confounded.

Table 5-14: Summary of Reproducibility Data

Sample	Assay Channel (Analyte)	N ^a	Mean Ct	Between-Site		Between-Day		Between-Operator + Within Assay		Total	
				SD	CV (%) ^b	SD	CV (%) ^b	SD	CV (%) ^b	SD	CV (%) ^b
Neg	SPC	88	31.4	0.3	0.9	0.0	0.0	1.5	4.7	1.5	4.8
Flu A – High Neg	Flu A1	62	36.9	0.1	0.3	0.6	1.6	1.6	4.4	1.7	4.7
Flu A – Low Pos	Flu A1	89	34.5	0.2	0.7	0.0	0.0	1.6	4.5	1.6	4.5
Flu A – Mod Pos	Flu A1	90	32.3	0.0	0.0	0.7	2.2	1.2	3.7	1.4	4.2
Flu B – High Neg	Flu B	56	34.6	0.0	0.0	1.4	4.0	2.1	6.0	2.5	7.2
Flu B – Low Pos	Flu B	89	32.6	0.6	2.0	0.7	2.1	1.7	5.1	1.9	5.9
Flu B – Mod Pos	Flu B	90	30.4	0.0	0.0	0.0	0.0	1.0	3.3	1.0	3.3
RSV – High Neg	RSV	40	36.9	1.2	3.1	0.0	0.0	1.7	4.6	2.1	5.6
RSV – Low Pos	RSV	87	35.0	0.7	1.9	0.0	0.0	1.6	4.6	1.7	5.0
RSV – Mod Pos	RSV	90	32.6	0.0	0.0	0.1	0.4	1.0	3.0	1.0	3.0

a. Results with non-zero Ct values out of 90.

b. (%) is contribution of variance component to overall CV.

Conclusions

The results of the nonclinical analytical and clinical performance studies summarized above demonstrate that the Xpert Flu+RSV Xpress Assay performs comparably to the predicate device for its intended use with inexperienced users in a CLIA waived environment and is substantially equivalent to the predicate device.