Luminex

Package Insert | IVD

ARIES® Flu A/B & RSV Assay

IVD For *In Vitro* Diagnostic Use.

For Use With the ARIES® Systems.

Rx Only

Luminex Corporation

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ARIES® Flu A/B & RSV Assay Package Insert

89-30000-00-106 Rev. A

Assay Kit Part Number: 50-10020

September 2016

EC REP

WMDE

Bergerweg 18

6085 AT Horn

The Netherlands

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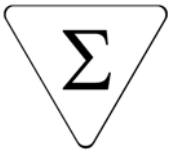
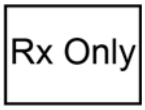
Key to Symbols

5.1.4* 	Use-by date Indicates the date after which the medical device is not to be used.	5.3.7* 	Temperature Limit Indicates the temperature limits to which the medical device can be safely exposed.
5.1.5* 	Batch Code Indicates the manufacturer's batch code so that the batch or lot can be identified.	5.4.2* 	Do not reuse Indicates a medical device that is intended for one use, or for use on a single patient during a single procedure.
5.1.6* 	Catalog(ue) Number Indicates the manufacturer's catalogue number so that the medical device can be identified.	5.4.4* 	Caution. Indicates the need for the user to consult the instructions for use for important cautionary information such as warnings and precautions that cannot, for a variety of reasons, be presented on the medical device itself.
5.1.1* 	Manufacturer Indicates the medical device manufacturer, as defined in EU Directives 90/385/EEC, 93/42/EEC and 98/79/EC.	5.5.5* 	Contains Sufficient for <n> Tests Indicates the total number of IVD tests that can be performed with the IVD kit reagents.
5.4.3* 	Consult instructions for use.Indicates the need for the user to consult the instructions for use.	5.4.1* 	Biological risks Indicates that there are potential biological risks associated with the medical device.
BC	Build Code	GHS02~ 	Highly flammable liquid and vapor
5.2.8* 	Do not use if package is damaged. Indicates a medical device that should not be used if the package has been damaged or opened.	5.5.1* 	*In vitro* diagnostic medical device Indicates a medical device that is intended to be used as an in vitro diagnostic medical device.

% 	Caution: Federal Law restricts this device to sale by or on the order of a licensed practitioner (U.S. Only). 21 CFR 809 (FDA Code of Federal Regulations)	# 	Conformite Europeenne (EU CE Marking of Conformity) Council Directive 98/79/ EC on In Vitro Diagnostic Medical Devices (IVDMD) (1998)
5.1.2* 	Authorized representative in the European Community Indicates the Authorized representative in the European Community		

* ANSI/AAMI/ISO 15223-1:2012, Medical devices—Symbols to be used with medical devices labels, labeling, and information to be supplied—Part 1: General requirements.

~ ST/SG/AC.10/30/Rev.6 Globally Harmonized System of Classification and Labeling of Chemicals (GHS) Sixth revised edition.

Council Directive 98/79/EC on In Vitro Diagnostic Medical Devices (IVDMD) (1998)

% 21 CFR 809 (FDA Code of Federal Regulations)

Standard Terms and Conditions For Use of Assay Product

By opening the packaging containing this assay product or cassette ("Product") or by using such Product in any manner, you are consenting and agreeing to be bound by the following terms and conditions. You are also agreeing that the following terms and conditions constitute a legally valid and binding contract that is enforceable against you. If you do not agree to all of the terms and conditions set forth below, you must promptly return the Product for a full refund prior to using them in any manner.

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strict compliance with the United States Food and Drug Administration and all applicable domestic and international laws and regulations, now and hereinafter enacted.

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Buyer has the responsibility and hereby expressly assumes the risk to verify the hazards and to conduct any further research necessary to learn the hazards involved in using the Product. Buyer also has the duty to warn Buyer's customers, employees, agents, assigns, officers, successors and any auxiliary or third party personnel (such as freight handlers, etc.) of any and all risks involved in using or handling the Product. Buyer agrees to comply with instructions, if any, furnished by Seller or Luminex relating to the use of the Product and to not misuse the Product in any manner. Buyer shall not reverse engineer, decompile, disassemble or modify the Product. Buyer acknowledges that Luminex retains ownership of all patents, trademarks, trade secrets and other proprietary rights relating to or residing in the Product and Buyer receives no rights to such intellectual property rights by virtue of its purchase of Product other than as expressly set forth herein. Buyer shall have no right to use any trademarks owned or licensed to Luminex without the express written permission of Luminex.

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89-30000-00-187 Rev C

Table of Contents

Intended Use

The ARIES® Flu A/B & RSV Assay is a polymerase chain reaction (PCR) based qualitative *in vitro* diagnostic test for the direct detection and differentiation of influenza A virus, influenza B virus, and respiratory syncytial virus (RSV) nucleic acid in nasopharyngeal swab (NPS) specimens from patients with signs and symptoms of respiratory tract infection in conjunction with clinical and laboratory findings. The test is intended for use as an aid in the differential diagnosis of Influenza A, Influenza B, and RSV in humans and is not intended to detect Influenza C.

Negative results do not preclude influenza virus or RSV infection and should not be used as the sole basis for diagnosis, treatment, or other management decisions. Conversely, positive results do not rule-out bacterial infection or co-infection with other viruses.

The agent detected may not be the definite cause of disease. The use of additional laboratory testing (e.g. bacterial culture, immunofluorescence, X-ray findings) and clinical presentation must be taken into consideration in order to obtain the final diagnosis of respiratory viral infection.

Performance characteristics for Influenza A were established during the 2014-2015 and the 2015-2016 influenza seasons when influenza A/H3N2 and A/H1N1 pandemic were the predominant influenza A viruses in circulation. When other 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.

The ARIES® Flu A/B & RSV Assay is indicated for use with the ARIES® Systems.

Summary and Explanation of the Test

Influenza A, influenza B, and respiratory syncytial virus (RSV) are contagious human viral pathogens that are transmitted from person to person primarily by aerosolized virus-containing droplets (influenza) or contaminated secretions (RSV) which can result in respiratory disease. Infections occur in all age groups, and cause significant health / financial concerns worldwide.

Of all the viruses that infect the respiratory tract, influenza, which belongs to the Orthomyxoviridae family of segmented, negative-strand enveloped RNA viruses, causes the largest number of serious acute illnesses. Three influenza virus types (A, B, and C) have been identified, of which influenza A causes the most human infections, with influenza C the least. Influenza A is known to infect humans, pigs, birds, and horses, while influenza B primarily infects humans; most epidemics and pandemics occur due to influenza A. Symptoms of influenza infection include sore throat, cough, fever, muscular pains, and weakness. The incubation period is brief (from 1 to 3 days), and the onset is usually sudden, with chills, fever, and general discomfort. There are doctor prescribed antiviral medications that can be used to treat influenza illness, but timing of administration is critical for optimal treatment. Additionally, bacterial pneumonia may occur among high-risk patients, as in the elderly, the very young, and people who have chronic diseases of the lungs. Annual vaccination with the current, prevalent strains of influenza virus is recommended, and is the primary line of defense, followed by antiviral drugs. Seasonal epidemics of influenza can involve 10% or more of the entire population each winter, with a toll and cost much higher during a pandemic.

Respiratory syncytial virus (RSV) is classified within the Paramyxoviridae family of non-segmented, negative-strand, enveloped RNA viruses. There are two RSV subgroups, A and B. Most RSV infections are acquired by contact with contaminated secretions in droplets; aerosol transmission is infrequent. Almost all children have been infected by 3 years of age due to exposure of successive outbreaks; transmission within households is common. RSV causes epidemics of bronchitis, pneumonia, and the common cold in children less than 5 years of age. It causes bronchitis and mild upper respiratory tract infections in adults. Symptoms of infection with this virus include fever, cough, and severe tiredness; some infants can die.

The ARIES® Flu A/B & RSV Assay uses Luminex Corporation's PCR chemistry in combination with the ARIES® Systems. The ARIES® Systems are capable of automated nucleic acid extraction and purification, real-time PCR detection of nucleic acid sequences, and data analysis. The ARIES® Flu A/B & RSV Assay can directly detect and differentiate three respiratory pathogens: influenza A virus (H1N1 (seasonal and pandemic 2009 (pdm09) and H3N2 (seasonal and variant (H3N2v)), influenza B virus (Yamagata and Victoria), and respiratory syncytial virus (RSV-A and RSV-B) from nucleic acid in nasopharyngeal swab (NPS) specimens from patients with signs and symptoms of respiratory tract infection.

Principles of the Procedure

Primary sample (NPS specimen in universal transport medium) is added directly to the ARIES® Flu A/B & RSV Assay cassette sample chamber. The cassette is then placed into an ARIES® magazine which can hold up to six cassettes. The magazine is inserted into an ARIES® instrument. A barcode on top of the ARIES® Flu A/B & RSV Assay cassette is automatically scanned by the ARIES® instrument, associating a preloaded ARIES® Flu A/B & RSV Assay protocol file with the cassette. The ARIES® Flu A/B & RSV Assay protocol file contains the necessary parameters to run the cassette, analyze data, and generate reports.

Once a run is started, the Sample Processing Control (SPC) is automatically added to the sample chamber of the cassette to control for sample lysis, recovery of extracted nucleic acid, the detection of inhibitory substances, and confirmation of PCR reagent integrity. Sample and SPC lysis, as well as isolation and purification of nucleic acids, are automated within the ARIES® Systems and the ARIES® Flu A/B & RSV Assay cassette. Purified nucleic acids are automatically transferred to the cassette's PCR tube that contains the lyophilized Influenza A/B & RSV Master Mix for the PCR amplification step. The lyophilized Influenza A/B & RSV Master Mix contains four sets of primers: one primer set for the SPC target, and an independent primer set for each pathogen. Total assay time, including extraction and PCR cycling, takes approximately two hours.

Materials Provided

The ARIES® Flu A/B & RSV Assay (Part Number 50-10020) contains 24 assay cassettes.

The assay protocol file, package insert, and *ARIES® Quick Guide* ship separately on a USB as part of the ARIES® Flu A/B & RSV Assay Protocol File Kit (CN-0335-01).

TABLE 1. **ARIES® Flu A/B & RSV Assay Contents Provided By Luminex**

Item	Part Number	Description
ARIES® Flu A/B & RSV Assay Cassette Kit	50-10020	24 ARIES® Flu A/B & RSV Assay cassettes which contain necessary reagents for sample extraction, nucleic acid purification, and amplification.
ARIES® Flu A/B & RSV Assay Protocol File Kit	CN-0335-01	An assay protocol file, a package insert, and an *ARIES® Quick Guide* containing instructions for use are provided on a USB.

Materials Required But Not Provided

Reagents for sample collection:

- Nasopharyngeal swab (NPS) (flocked, polyester, or rayon swab)
- Universal Transport Medium (UTM)

Equipment:

- -65°C to -95°C freezer
- 2°C to 8°C refrigerator
- Luminex® ARIES® Systems (either an ARIES® System or an ARIES® M1 System can be used) and accessories

- ARIES® magazines
- Sample Prep Tray
- Hand-held barcode reader

- Vortex mixer
- Appropriately sized pipettor

Plasticware and Consumables:

- Nuclease-free aerosol-barrier pipette tips

Warnings and Precautions

1. For *In Vitro* Diagnostic Use Only.
2. For prescription use only.
3. Handle all samples as if infectious using safe laboratory procedures such as those outlined in CDC/NIH *Biosafety in Microbiological and Biomedical Laboratories*, and in the CLSI Document M29 *Protection of Laboratory Workers from Occupationally Acquired Infections*.
4. Thoroughly clean and disinfect all surfaces with 10% bleach.
5. Avoid contamination from positive controls and samples by following good laboratory practices.
6. Avoid contamination by using a new nuclease-free aerosol barrier tip to add an individual sample aliquot to each cassette.
7. Wear appropriate personal protective equipment (PPE), including a lab coat and disposable gloves, when performing procedures. Wash your hands thoroughly after performing the test.
8. Follow your institution's safety procedures for working with chemicals and handling biological samples.
9. Do not use cassettes, kits, or reagents beyond their expiration date.
10. The cassettes are single-use. Do not reuse cassettes.
11. Store cassettes at the temperatures recommended on the cassette label. Do not freeze.
12. Only use the extraction protocol file provided by Luminex on the USB drive.
13. Only use the procedures described in this package insert. Any deviation from the outlined procedures can result in extraction failure or cause erroneous results.
14. Only use ARIES® Systems that have been properly maintained according to the manufacturer's recommendations.
15. ARIES® cassettes contain guanidinium thiocyanate. Refer to the Safety Data Sheet (SDS) regarding safe handling practices for any spills.
16. In the event that a PCR tube falls off the cassette or a cassette leaks inside the ARIES® instrument, you should perform appropriate decontamination procedures to reduce the risk of contamination. Immediately clean all surfaces of the ARIES® magazine and the surrounding bench top with water. Wipe the surfaces with a lint-free cloth. Follow that with a fresh 10% bleach solution. Allow the bleach solution to sit for a minimum of 10 minutes. Thoroughly rinse bleached surfaces with deionized water. Dispose of all lint-free cloths in the appropriate waste container. Immediately contact Luminex Technical Support in order to retrieve the PCR tube from the ARIES® instrument. Do not throw away the cassette before you contact Technical Support. Do not attempt to retrieve the tube or put your hands inside the ARIES® instrument at any time. Do not proceed with additional testing until the PCR tube has been removed from the ARIES® instrument. Discard the cassette in accordance with the procedures defined by appropriate biohazard safety guidelines or regulations.
17. Refer to the appropriate ARIES® system operation manual for electrical warnings.
18. Do not let the ARIES® Systems get wet or allow standing water to pool under the instrument.
19. Safety Data Sheets (SDS) are available by contacting Luminex Corporation.

Reagent Storage, Handling, and Stability

ARIES® Flu A/B & RSV Assay cassettes are shipped refrigerated. Store at room temperature (15°C to 30°C) after receipt.

Always check the expiration date on the kit box and cassettes.

Sample Handling and Storage

Sample Collection

Nasopharyngeal swab (NPS) samples should be collected with a synthetic tip (e.g. flocked, polyester, or rayon) following the user institution's standard procedures and placed into 3 mL of Universal Transport Medium (UTM).

Sample Transport

When transporting biological samples, ensure that all applicable regulations for the transport of etiologic agents are met.

Transport samples refrigerated at 2°C to 8°C. If there will be a long delay before sample processing (greater than three days from the date of collection), samples should be frozen at -70°C or colder for up to 8 months and transported on dry ice.

Sample Storage

Samples can be stored refrigerated at 2°C to 8°C for up to three days from the date of collection. If samples will be used after three days from the date of collection, store frozen at -70°C or colder for up to 8 months.

Store left-over samples at -70°C or colder for up to 8 months.

Assay Procedure

Adding Assay Files to the ARIES® Systems

The ARIES® Flu A/B & RSV Assay protocol file is provided on the USB flash drive. The assay protocol file only needs to be imported to the ARIES® Systems once. To import the assay protocol file, complete the following:

1. Insert the USB flash drive into one of the five USB connectors (one in the front and four in the back).
2. Select  in the upper left-hand corner of the screen and navigate to **Assay Management**.
3. Select 

from the Page Action bar. The **Import File** dialog box displays.
4. Choose the **Location** and **File Name** of the assay file. Select **OK**.

Entering Orders

Sample barcodes are scanned to associate them with an order. An assay cassette is also then scanned to specify the assay and associate the cassette with a specific sample. Refer to *"Running an Assay"* on page 8 for more information.

The Sample ID is required on all orders and is the link between sample and cassette. The Accession ID and Requisition Number can also perform this role and associate the cassette to the sample, but are optional unless otherwise chosen to be required by the user. You can set requirement options in the Sample Options dialog box located on the Order Management Settings page.

Enabling the Automatic Print and Export Results Options

The Auto Print and Auto Export options are settings that need to be enabled prior to starting the run on the ARIES® instrument. Results can also be printed and exported manually after a run. Refer to *"Manually Printing Reports"* on page 10 for more information.

To enable the Auto Print feature, complete the following:

1. Select  in the upper left-hand corner of the screen and navigate to **Results > Settings**.
2. Toggle the **Generate Reports After Run** button to **Yes.**
3. For the **Sample Reports to Printer** option, select **Default** or **All**.

To export results automatically after a run, complete the following:

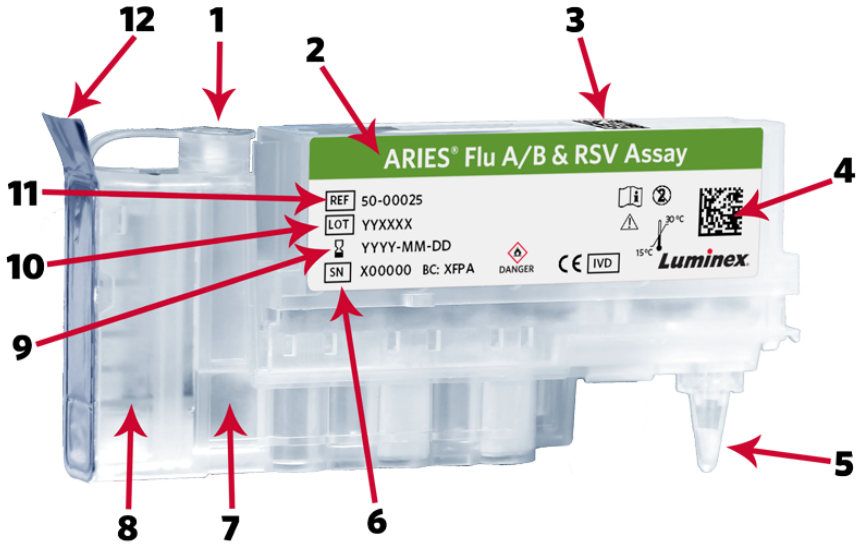
1. Select  in the upper left-hand corner of the screen and navigate to **Results > Settings**.
2. Toggle the **Summary Report as CSV** button to **Yes.**
3. Enter the **CSV Output Location** by selecting the folder icon in the upper right-hand corner of the **CSV Output location** box. The **Select Folder** dialog box opens.
   a. Choose the **Location** for the export.
   b. Select **OK**.

To automatically export LIS results as either HL7 or CSV, complete the following:

1. Select  in the upper left-hand corner of the screen and navigate to **Administration > LIS Settings.**
2. Toggle the **Enable Export to LIS** button to **Yes**.
3. Toggle the **Auto Export to LIS** button to **Yes.**
4. Enter the **LIS Export Location** by selecting the folder icon in the upper right-hand corner of the **LIS Export Location** box. The **Select Folder** dialog box opens.
   a. Choose the **Location** for the export.
   b. Select **OK**.

CAUTION: Validation of LIS compatibility must be performed by the user.

FIGURE 1. **ARIES® Cassette**



1. Cassette cap	7. Sample chamber
2. Assay type	8. Side cassette
3. Cassette barcode (top)	9. Cassette expiration date
4. Cassette barcode (side)	10. Cassette lot number
5. PCR tube	11. Cassette part number
6. Cassette serial number	12. Back seal

Entering Orders on the ARIES® Systems

When entering orders, the Sample ID and Assay are required for an order to be valid.

1. Select ☰ in the upper left-hand corner of the screen and navigate to **Order Management > Sample Orders**.
2. Select New Order from the Page Action bar. The **New Order** dialog box displays.
3. Remove the assay cassette from its packaging and visually inspect the cassette for any damage.

   CAUTION: If the cassette(s) appears damaged in any way or if you see any leaks, DO NOT USE THE CASSETTE. Immediately contact Luminex Technical Support to report the damage.
4. Close the cassette cap to seal the cassette sample chamber.
5. Scan the barcode on the top (or side) of the cassette with the hand-held barcode reader or enter the required cassette information manually. A touch screen keyboard or a drop-down menu displays.

   NOTE: If the keyboard does not automatically appear, toggle the keyboard icon to **Yes**. The keyboard will appear when you click in a field.

   NOTE: If manually entering the **Cassette Lot Expiration**, select the calendar icon and choose the date using the calendar. The date is shown in the YYMMDD format.

   a. If applicable, to add a control, choose **Control** in the **Sample Type** drop-down menu.
   b. In the **Control** field, click the magnifying glass to select a control from the **Controls** dialog box.
   c. Select the type of control in the **Control Type** drop-down menu.

NOTE: You can define the controls on the **Assay Management > Controls** page. Refer to the appropriate ARIES® system operation manual for more information on controls.

6. Scan or enter the Sample ID.
7. Optionally, select the **Selected Tests** field. The **Test Selection** dialog box opens.
   a. Choose **Selected** or **Masked** for each test shown. Reports and results will not include data for any masked tests.

      NOTE: At least one test must remain unmasked. You cannot mask all tests.

   b. Select **Close**.
8. Scan the **Data Matrix** barcode on the screen next to **Save**, or manually select **Save**.

Adding Samples to the Cassettes

1. Place the sample tube in the Sample Prep Tray.
2. Pull the tab to remove the foil seal from the cassette.

   CAUTION: Use caution when pulling the back seal off the cassettes. The foil is sharp and may cause injury.

3. Place the cassette in the Sample Prep Tray next to the sample.

4. Vortex the sample for 5 to 10 seconds to homogenize the mixture.
5. Open the cassette cap to access the cassette sample chamber.
6. Using an appropriately sized pipettor and aerosol barrier pipette tip, aspirate 200 µL of sample from the sample tube.

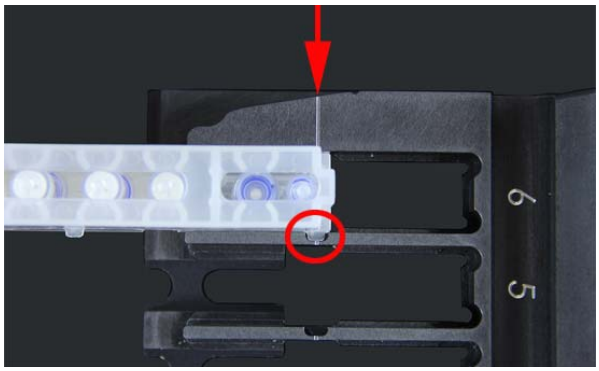
CAUTION: Ensure that the correct amounts of sample are used.

CAUTION: Use care to avoid contamination of the pipettor during transfer of the sample from the sample tube to the cassette.

7. Place the sample in the cassette sample chamber by inserting the pipette tip near the bottom of the chamber before expelling the sample.
8. Close the cassette cap to seal the cassette sample chamber.

WARNING: Failure to ensure that the cassette cap is fully closed may cause a delay or failure in results and expose you to biohazards.

9. Place the cassette into the magazine by lining the cassette up with the first notch (a tab on the cassette fits into the notch).



NOTE: The PCR tube must face toward the numbers on the magazine.

10. Gently insert the cassette into the magazine.
11. Gently slide the cassette all the way back toward the numbers. Repeat for all other cassettes.

WARNING: Do not use your index finger to push the cassette into the magazine. You may indirectly dispense the reagent. Luminex recommends using the palm of your hand, or holding the cassette and sliding the cassette into proper position.

Running an Assay

1. Select ☰ in the upper left-hand corner of the screen and navigate to **Run**.
   a. Insert the magazine into the ARIES® instrument. The ARIES® instrument automatically scans the barcode printed on the top of the ARIES® Flu A/B & RSV Assay cassettes, identifies associated orders and the proper assay protocol files before starting the run.

NOTE: Ensure that the **Auto run upon Magazine Insertion** is toggled to **Yes** in the **Run Options** dialog box, located on the **Run Settings** page. The instrument automatically scans the cassettes once the magazine is inserted and starts the run.

2. If there are any errors, the ARIES® instrument displays the specific error (for example, cassettes that cannot be run together, cassette IDs that have not been read, or assay files not loaded on to the ARIES® instrument). These errors must be corrected in order for the run to begin.
   If there are no errors, and the auto run option is selected, the color indicator of the cassette turns purple, the magazine state indicator indicates **PLEASE DO NOT REMOVE THE MAGAZINE** and an orange lock icon displays on the left-hand side of the magazine state indicator. The Run Status bar, located at the bottom of the **Run** page, displays an orange progress bar next to the estimated time to completion, colored purple. If you do not have the Auto Run feature enabled, you can start the run manually by selecting 

   from the Page Action bar.

   NOTE: If you are using an ARIES® System with two modules, highlight the module you want before selecting **Start Run**.

Monitoring the Run

From the Run page, select 

on the Page Action bar to display the status of the magazine(s), the estimated time to completion, and the customizable name of the ARIES® instrument. This status screen is intended to be visible from across the room, allowing you to monitor your runs while you are working on other projects.

TIP: On the Run > Settings page, you can customize whether the estimated completion time or estimated time remaining displays.

Reviewing, Printing, and Exporting Run Results

When the ARIES® Flu A/B & RSV Assay run finishes successfully, the cassettes are colored green on the Run page. See *Table 2,* on page 9 for other color indicators. Refer to the appropriate ARIES® system operation manual for more color definitions.

TABLE 2. **Color Indicators**

Color	Reason
Red	Cassettes contain errors, were not scanned successfully, require additional information, or the run failed or was aborted. Contact Luminex Technical Support for assistance.
Yellow	Information was manually entered on the Run page or cassette is expired.
Green	Run finished successfully, the cassettes were scanned with no errors.
Blue	Magazine is inserted and a cassette is detected for this slot.
Purple	Module is currently running: the magazine slot is in use.
White	Empty module, no magazine is inserted or no cassette is detected.

The Run page includes visual indicators such as a status bar, an estimated time to completion indicator, and a Run Complete notification once the run has completed.

Automatically Printing and Exporting Results

NOTE: To ensure that the LIS Reporting (Auto Print) feature is enabled, check that Sample Reports to Printer is set to All or Default in the Export Settings dialog box located on the Results > Settings page.

When the run finishes successfully, the result reports are automatically printed at the default printer and exported in .csv and .pdf format to the designated location. Refer to *"Enabling the Automatic Print and Export Results Options"* on page 5.

Manually Printing Reports

To manually select a report to print, complete the following:

1. Select  in the upper left-hand corner of the screen and navigate to **Results**. Regardless of the type of report you want to view, select only one result. Otherwise, the **Create Report** icon grays out.
2. Select **Create Report** from the Page Action bar. Choose the type of report you want to view from the drop-down menu. There are three options: **Run Report**, **Detailed Report**, and **Summary Report**.

   NOTE: Selecting a single result gives you the option to generate a **Run Report**, **Summary Report**, or a **Detailed Report**. You cannot select more than one result and run a **Summary Report** or a **Detailed Report** -- the **Create Report** icon grays out. When generating a **Run Report**, you can select multiple results from the same run and still use the **Create Report** icon. With **Run Report**, the **Create Report** icon is disabled only when results from multiple runs are selected. Once the report opens, choose to export 

   or print the result 

   .

   - The Run Report displays the run results for all samples in the run and any comments or logs associated with that run.
   - The Summary Report displays the run result for one individual sample and any comments or logs associated with that sample.
   - The Detailed Report displays all cassette information for one individual sample, the Amplification and Melt Graphs, and any comments or logs associated with that sample. Access to run this report is restricted to users with administrative rights.

Manually Exporting Results

To manually export results, complete the following:

1. Select the result(s) to export on the **Results** page and select 

   from the Page Action bar.
2. Choose the **Location** and the **File Name** for the export in the **Export File** dialog box and select **OK**.

   NOTE: You can only export to a USB drive or a mapped network drive.

Interpretation of Sample Results

The ARIES® software determines results for the sample and the sample processing control (SPC) based on the amplification cycle (Ct) value and the melting temperature (T_m) value provided in the assay protocol file. All assay outcomes are listed in *Table 3,* on page 11.

TABLE 3. **Interpretation of Sample Results**

Example*	SPC		Influenza A		Influenza B		RSV		Call
	T_m Value	Ct Value	T_m Value	Ct Value	T_m Value	Ct Value	T_m Value	Ct Value	
1	+	N/A	+	+	+	>	+	>	Influenza A Positive, Influenza B Negative, RSV Negative
2	+	N/A	+	+	-	N/A	-	N/A	
3	+	N/A	+	>	+	+	+	>	Influenza A Negative, Influenza B Positive, RSV Negative
4	+	N/A	-	N/A	+	+	-	N/A	
5	+	N/A	+	>	+	>	+	+	Influenza A Negative, Influenza B Negative, RSV Positive
6	+	N/A	-	N/A	-	N/A	+	+	
7	+	N/A	+	+	+	+	+	>	Influenza A Positive, Influenza B Positive, RSV Negative**
8	+	N/A	+	+	+	+	-	N/A	
9	+	N/A	+	+	+	>	+	+	Influenza A Positive, Influenza B Negative, RSV Positive
10	+	N/A	+	+	-	N/A	+	+	
11	+	N/A	+	>	+	+	+	+	Influenza A Negative, Influenza B Positive, RSV Positive
12	+	N/A	-	N/A	+	+	+	+	
13	+	N/A	+	+	+	+	+	+	Influenza A Positive, Influenza B Positive, RSV Positive**
14	+	+	+	>	+	>	+	>	Influenza A Negative, Influenza B Negative, RSV Negative
15	+	+	-	N/A	-	N/A	-	N/A	

* All scenarios not captured above will be called “Invalid”. In case of an “Invalid” result, re-test the sample with a new assay cassette. If the problem is unresolved, please contact Luminex Technical Support.

** Dual infections of Influenza A and Influenza B are rare. If the test result is “Influenza A Positive” and “Influenza B Positive”, the assay should be repeated with the same patient specimen, or if possible, with a newly collected specimen

Legend			
+	Indicates that a valid Ct value is present.	-	Indicates that a valid Ct value is not present.
>	Indicates that the Ct value obtained is above the Ct cutoff.	N/A	Not applicable. All possible outcomes will result in the same call.

Quality Control

Quality control procedures intended to monitor the ARIES® Systems and assay performance are outlined in *Table 4,* on page 12.

TABLE 4. **Controls to Monitor Quality**

Control Type	Use
Sample Processing Control	Verifies proper sample lysis and nucleic acid extraction, and proper reagent, cassette, ARIES® instrument, and assay protocol performance.

Each ARIES® Flu A/B & RSV Assay cassette contains a Sample Processing Control, which is processed with the sample and analyzed during the amplification reaction.

External controls should be used in accordance with local, state, federal accrediting organizations, as applicable. For example, reference influenza A, influenza B, and RSV strains or well characterized influenza A, influenza B, and RSV clinical isolates may be used as positive controls; universal transport medium may be used as a negative control.

Limitations

1. The detection of viral nucleic acids depends on proper sample collection, handling, transportation, storage, and preparation (including extraction). Failure to observe proper procedures in any one of these steps can lead to an incorrect result.
2. There is a risk of false negative results due to improperly collected, transported, or handled samples.
3. There is a risk of false negative results due to the presence of sequence variants in the targets of the assay, procedural errors, amplification inhibitors in samples, or inadequate numbers of organism(s) for amplification.
4. There is a risk of false negative RSV results when at low concentration and in the presence of co-infection with high concentration of influenza A.
5. There is a risk of false positive results due to potential cross-contamination by target organism(s), their nucleic acid or amplified product, or from non-specific signals in the assay.
6. Analyte targets (viral nucleic acid) may persist *in vivo*, independent of virus viability. Detection of analyte target(s) does not imply that the corresponding virus(es) are infectious, or are the causative agents of the clinical symptoms.
7. This test has been evaluated for use with human specimen material only.
8. This test is a qualitative test and does not provide quantitative values of the detected organism.
9. This test has not been evaluated in patients without signs and symptoms of a respiratory tract infection.
10. The effect of interfering substances has been evaluated only for those listed within the labeling. Interference by substances other than those described can lead to erroneous results.
11. Cross-reactivity with respiratory tract organisms other than those tested can lead to erroneous results.
12. Recent patient exposure to FluMist® or other live attenuated influenza vaccines may cause inaccurate positive results.
13. This test cannot rule out diseases caused by other bacterial or viral pathogens.
14. For use only on the ARIES® System or ARIES® M1 System.

Disposal

Dispose of hazardous or biologically contaminated materials according to the practices of your institution.

Clinical Studies

Prospective specimens

The clinical performance of the ARIES® Flu A/B & RSV Assay was evaluated using leftover, de-identified, nasopharyngeal swab (NPS) specimens prospectively collected from pediatric or adult patients suspected of having a respiratory tract infection during the 2014/2015 and 2015/2016 flu seasons. In the first phase of the prospective study (2014/2015 flu season) specimens were collected and tested at 3 clinical sites located in the United States and Canada from 18-January-2015 to 20-March-2015. Clinical specimens accrued during the second phase of the prospective study (2015/2016 flu season) were collected from 02-November-2015 to 29-February-2016 at 4 clinical sites located in the United States and Canada. The clinical specimen collection sites were chosen based on the types of patients usually referred to them, and the prevalence of respiratory pathogens in order to ensure a broad coverage of respiratory organisms, patient ages, and geographical regions.

A total of 2504 nasopharyngeal swab specimens were collected in the prospective study. Twenty-five (25) specimens were excluded based on inclusion/exclusion criteria or protocol deviation leaving a total of 2479 eligible unique specimens for subsequent data analysis. Of these, 1017 were collected during the 2014/2015 Flu season while the remaining 1462 specimens were enrolled during the 2015/2016 Flu season. *Table 5,* on page 13 provides a summary of the general demographic information of the prospectively collected nasopharyngeal swabs that were included in the analysis (combined 2015/2016 and 2014/2015 prospective data set). Demographic information is also presented by clinical site.

TABLE 5. **General Demographic Details for the Combined Prospective Dataset (N=2479)**

GENDER	Site 1	Site 2	Site 3	Site 4	All Sites
Male	375 (46.2%)	134 (41.0%)	200 (51.7%)	459 (48.1%)	1168 (47.1%)
Female	436 (53.8%)	193 (59.0%)	187 (48.3%)	495 (51.9%)	1311 (52.9%)
Total	**811**	**327**	**387**	**954**	**2479**
AGE (yrs)					
0 – 1	37 (4.6%)	32 (9.8%)	102 (26.4%)	263 (27.6%)	434 (17.5%)
>1 – 5	29 (3.6%)	29 (8.9%)	62 (16.0%)	103 (10.8%)	223 (9.0%)
>5 – 21	29 (3.6%)	49 (15.0%)	64 (16.5%)	103 (10.8%)	245 (9.9%)
>21 – 65	316 (39.0%)	131 (40.1%)	124 (32.0%)	274 (28.7%)	845 (34.1%)
>65	400 (49.3%)	86 (26.3%)	35 (9.0%)	211 (22.1%)	732 (29.5%)
Total	**811**	**327**	**387**	**954**	**2479**

All 2479 eligible prospective clinical specimens were tested by an FDA-cleared molecular comparator assay and the ARIES® Flu A/B & RSV Assay. The comparator assay was performed in accordance with manufacturer's instructions at a centralized testing facility. Clinical runs and re-runs using ARIES® Flu A/B & RSV Assay were carried out by trained operators at the testing sites on clinical specimens that were either stored frozen at -80°C (N=1316; 53.1%) or kept refrigerated at 4°C to 8°C (N=1163; 46.9%) prior to testing. Out of the 2479 clinical specimens included in the prospective analysis, 2458 (2458/2479; 99.2%) generated valid results with ARIES® Flu A/B & RSV Assay during initial testing. There were 21 specimens (21/2479, 0.8%) that were re-tested with ARIES® Flu A/B & RSV Assay because they yielded initial invalid results. All 21 specimens in question generated valid ARIES® results upon repeat testing.

The performance of the assay was evaluated by calculating the percent agreement of the results with the comparator results. Positive and Negative Percent Agreement were based on the fraction of comparator positive (or negative) results which were also positive (or negative) by the ARIES® Flu A/B & RSV Assay. Positive Percent Agreement (PPA) was calculated by dividing the total number of "true positive" ARIES® Flu A/B & RSV Assay results (TP) by the sum of the TP and "false negative" (FN) ARIES® Flu A/B & RSV Assay results. Negative Percent Agreement (NPA) was calculated by dividing the total number of "true negative" ARIES® Flu A/B & RSV Assay results (TN) by the sum of the TN and "false positive" (FP) ARIES® Flu A/B & RSV Assay results. An ARIES® Flu A/B & RSV Assay result was considered to be a TP or TN result only in the event that it agreed with the comparator method result for the analyte in question. Ninety-five percent (95%) confidence intervals were calculated using the Exact (Clopper-Pearson) method. Discordant results between the ARIES® Flu A/B & RSV Assay and the FDA-cleared molecular comparator assay were further analyzed by bi-directional sequencing using analytically validated primers that targeted genomic regions distinct from the ARIES® Flu A/B & RSV Assay Assay. Results from discordant testing analysis were not included in the calculation of Positive Percent Agreement and Negative Percent Agreement for each target. However, these results are included as footnotes in the performance evaluation table.

The clinical performance of the ARIES® Flu A/B & RSV Assay in the prospective study (N=2479) is summarized for each individual target probed by the ARIES® Flu A/B & RSV Assay in *Table 6,* on page 14.

TABLE 6. **ARIES® Flu A/B & RSV Assay Clinical Performance (Prospective Sample Set)**

Target	PPA		95% CI	NPA		95% CI	"No Call" by Reference[g]
Influenza A[h]	299/ 312[a]	95.8%	93.0% - 97.8%	2131/ 2165[b]	98.4%	97.8% - 98.9%	2
Influenza B[h]	45/48[c]	93.8%	82.8% - 98.7%	2417/ 2431[d]	99.4%	99.0% - 99.7%	0
RSV	270/ 278[e]	97.1%	94.4% - 98.7%	2165/ 2201[f]	98.4%	97.7% - 98.9%	0

[a] Seven (7) ARIES® Flu A/B & RSV Assay negative specimens that were positive for influenza A by the reference method (that is, False Negative) tested negative by bi-directional sequencing analysis using analytically validated primers that targeted genomic regions distinct from the ARIES® Assay.

[b] Four (4) ARIES® Flu A/B & RSV Assay positive specimens for influenza A that were negative by the reference method (that is, False Positive) tested positive by bi-directional sequencing analysis using analytically validated primers that targeted genomic regions distinct from the ARIES® Assay.

[c] Two (2) ARIES® Flu A/B & RSV Assay negative specimens that were positive for influenza B by the reference method (that is, False Negative) tested negative by bi-directional sequencing analysis using analytically validated primers that targeted genomic regions distinct from the ARIES® Assay.

[d] Three (3) ARIES® Flu A/B & RSV Assay positive specimens for influenza B that were negative by the reference method (that is, False Positive) tested positive by bi-directional sequencing analysis using analytically validated primers that targeted genomic regions distinct from the ARIES® Assay.

[e] One (1) ARIES® Flu A/B & RSV Assay negative specimen that was positive for RSV by the reference method (that is, False Negative) tested negative by bi-directional sequencing analysis using analytically validated primers that targeted genomic regions distinct from the ARIES® Assay.

[f] Thirty-two (32) ARIES® Flu A/B & RSV Assay positive specimens that were negative for RSV by the reference method (that is, False Positive) tested positive by bi-directional sequencing analysis using analytically validated primers that targeted genomic regions distinct from the ARIES® Assay.

[g] Two (2) clinical specimens generated a "No Call" or "Invalid" result for influenza A by the comparator upon repeat testing. The results of both specimens were therefore excluded from assay performance calculation for this target.

[h] Three subjects had dual infections (Flu A & Flu B) by ARIES® Flu A/B & RSV Assay (all Flu A & Flu B dual infections were not confirmed by a comparator assay)

Pre-selected Clinical Specimens

Due to the low prevalence of Influenza B observed in the prospective study, the sample set was supplemented with 40 banked (pre-selected) Influenza B positive specimens collected at a single clinical laboratory. The presence of the expected target (Influenza B) in each of the pre-selected specimens was confirmed by testing with the comparator assay. In order to minimize bias, the pre-selected positive specimens were tested along with 40 unique negative clinical specimens in a randomized, blinded fashion at 4 external testing sites. Of the 80 Influenza B positive and negative specimens included in this study, 78 (78/80; 97.5%) generated valid results with ARIES® Flu A/B & RSV Assay on the first attempt. Invalid results were generated for 2 specimens (2/80; 2.5%) tested. Both specimens in question yielded valid ARIES results upon repeat testing.

The results from pre-selected specimens were analyzed separately from those of the prospective data set. ARIES® Flu A/B & RSV Assay accurately detected all 40 Influenza B positive specimens tested (100.0%; 95.0% confidence interval, 91.2% - 100.0%).

Analytical Performance of ARIES® Flu A/B & RSV Assay

Limit of Detection

Limit of Detection (LoD) was established for the ARIES® Flu A/B & RSV Assay using three influenza A strains, two influenza B strains, and two respiratory syncytial virus strains (1 RSV-A, and 1 RSV-B) diluted into a simulated nasal matrix (SNM) containing negative pooled human clinical matrix and Universal Transport Media (UTM). The LoD is defined as the lowest sample concentration ($TCID_{50}$/mL) that had a positivity rate of ≥ 95.0%. Serial dilutions of each quantified viral strain in SNM were initially tested in a range finding study where the preliminary LoD $TCID_{50}$/mL concentrations were determined. The preliminary LoD concentrations were then confirmed by testing 20 replicates of each strain.

The LoD for each viral strain was determined and confirmed empirically as the lowest concentration that had ≥ 95.0% positive results. The LoD concentrations for each viral strain are summarized in *Table 7,* on page 15.

TABLE 7. **Limit of Detection of the ARIES® Flu A/B & RSV Assay**

Assay Target	Strain	Concentration ($TCID_{50}$/mL or $CEID_{50}$/mL)	Positivity	95.0% Confidence Interval
Influenza A	PR/8/34	$1 \times 10^{-0.34}$	100.0%	83.2% - 100.0%
	Hong Kong/8/68	$1 \times 10^{2.40}$	100.0%	83.2% - 100.0%
	Mexico/4108/2009 (H1N1)pdm09	$1 \times 10^{1.45}$	95.0%	75.1% - 99.9%
Influenza B	Florida/04/06	$1 \times 10^{0.30}$	100.0%	83.2% - 100.0%
	Malaysia/2506/04	$1 \times 10^{1.05}$	100.0%	83.2% - 100.0%
RSV	A2 (VR-1540)	$1 \times 10^{-0.57}$	95.0%	75.1% - 99.9%
	WV/14617/85 (VR-1400)	$1 \times 10^{0.90}$	100.0%	83.2% - 100.0%

Competitive Interference / Co-infection

Competitive interference was evaluated to confirm the ability of the ARIES® Flu A/B & RSV Assay to detect influenza A, influenza B, RSV-A, or RSV-B in the presence of a co-infection. Competitive interference can occur when one organism is present in a specimen near the LoD and an additional organism that the

assay is intended to detect is present at high concentration. In various combinations, organisms were tested in triplicate at high concentrations (≥ 10^5 $TCID_{50}$/mL) mixed with low concentrations (1.5x LoD), as identified in *Table 8,* on page 16. All results were positive and in agreement with the expected results indicating the ARIES® Flu A/B & RSV Assay can detect influenza A, influenza B, and RSV co-infections.

TABLE 8. **Co-infection Test Cases**

Low Concentration Organism	High Concentration Organism	Co-infection Scenario
A/Hong Kong/8/68	B/Florida/04/06	Influenza A + Influenza B
	RSV A2	Influenza A + RSV A
	RSV B WV/14617/85	Influenza A + RSV B
B/Florida/04/06	A/Hong Kong/8/68	Influenza B + Influenza A
	RSV A2	Influenza B + RSV A
	RSV B WV/14617/85	Influenza B + RSV B
RSV A2	B/Florida/04/06	RSV A + Influenza B
RSV A2*	A/Hong Kong/8/68*	RSV A + Influenza A
RSV B WV/14617/85	A/Hong Kong/8/68	RSV B + Influenza A
	B/Florida/04/06	RSV B + Influenza B

*High concentration of influenza A had inhibitory effect on RSV A2 at a concentration lower than 3x LoD.

Interfering Substances

The potential inhibitory effect of non-microbial substances expected to be found in nasopharyngeal swab specimens was evaluated by testing within the ARIES® Flu A/B & RSV Assay. Three (3) replicates each of influenza A, influenza B, RSV-A, and RSV-B were tested at concentrations near the assay LoD with a clinically relevant concentration of each potentially interfering substance spiked into the reaction. All influenza A, influenza B, and RSV samples were 100.0% positive in the presence of a non-microbial substance at concentrations shown below; all negative samples containing only the non-microbial substance were 100.0% negative, with the exception of FluMist®. The interfering substances used in the study are shown in *Table 9,* on page 16.

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.

Positive influenza results obtained in a patient who received FluMist prior to sample collection may be due to detection of vaccine virus and may mask a true positive result caused by an influenza infection; additionally, FluMist may interfere with RSV detection due to high concentration of vaccine virus nucleic acid, causing a possible RSV false negative result.

TABLE 9. **Interfering Substance Information**

Interfering Substance	Test Concentration
Benzocaine	2.5% w/v
Budesonide	25 mg/mL
Dexamethasone	3 mg/mL
FluMist®	0.5% v/v
Flunisolide	55 mg/mL

Menthol	1.7 mg/mL
Mometasone	2.5 mg/mL
Phenylephrine	0.5% w/v
Afrin® (Oxymetazoline)	15% v/v
Tobramycin	4 µg/mL
Mupirocin	6.6 mg/mL
Beconase AQ® (Beclomethasone)	5% v/v
Flonase® (Fluticasone)	5% v/v
Zanamivir	3.3 mg/mL
Tamiflu®	1 µM
Triamcinolone	5.5 mg/mL
Sodium chloride	0.65% v/v
Human Whole Blood	2% v/v
Mucin Protein	60 µg/mL
ZICAM® (Galphimia glauca, Histaminum hydrochloricum, Luffa operculata, Sulfur)	5% v/v

Analytical Specificity

Microbial interference for the ARIES® Flu A/B & RSV Assay was assessed with 32 potential cross reactive microorganisms evaluated in the cross reactivity study, and identified in *Table 10,* on page 17. Each potential interfering microorganism was spiked into a simulated nasal matrix (SNM) containing a representative strain of influenza A, influenza B, RSV-A, or RSV-B near the LoD; bacteria were spiked and tested at concentrations ≥ 10^6 CFU/mL and viruses were spiked and tested at concentrations ≥ 10^5 $TCID_{50}$/mL, or the highest available concentration for both types of potential cross reactive microorganisms. All prepared samples were tested in triplicate (n=3) on the ARIES® System. All positive results were in agreement with the expected results for influenza A, influenza B, and RSV demonstrating the microorganisms tested do not interfere with the ARIES® Flu A/B & RSV Assay at the concentrations tested.

TABLE 10. **Microorganism Information**

Microorganism		
Adenovirus type 1	*Hemophilus influenzae*	Parainfluenza type 1
Adenovirus 7a	*Mycoplasma pneumonia*	Parainfluenza type 2
Bordetella pertussis (A639)	*Mycobacterium tuberculosis*	Parainfluenza type 3
Chlamydia pneumoniae	*Lactobacillus plantarum (17-5)*	*Pseudomonas aeruginosa*
Coronavirus 229E	*Legionella longbeachae*	Rhinovirus type 1A
Coronavirus OC43	Measles	*Staphylococcus aureus (COL)*
Corynebacterium diphtheriae	Metapneumovirus	*Staphylococcus epidermidis*
Cytomegalovirus (CMV)	*Moraxella catarrhalis Ne 11*	*Streptococcus pneumoniae*
Enterovirus 71	Mumps	*Streptococcus pyogenes*
Epstein Barr virus	*Neisseria elongata*	*Streptococcus salivarius*
Escherichia coli O157	*Neisseria meningitidis*	

The analytical specificity of the ARIES® Flu A/B & RSV Assay was evaluated by testing the potential cross-reactivity of 32 microorganisms listed in *Table 10,* on page 17. The microorganisms tested consisted of 14 viral and 18 bacterial strains representing common respiratory pathogens, or those potentially encountered in the human nasopharynx region. The potential cross-reacting organisms were spiked into simulated nasal matrix (SNM) that was negative for influenza A, influenza B, and RSV and tested with the ARIES Flu A/B & RSV Assay in triplicate. Bacterial organisms were tested at concentrations ≥ 10^6 CFU/mL and viral organisms tested at ≥ 10^5 $TCID_{50}$/mL, or the highest available concentration for both types of potential cross-reactive microorganisms. The results were negative for all organisms and in 100.0% agreement with expected results demonstrating no cross-reactivity with the ARIES Flu A/B & RSV Assay.

Carry-Over and Cross Contamination

Carry-over and cross contamination for the ARIES® Flu A/B & RSV Assay were evaluated by testing 30 high concentration influenza A positive samples in a series alternating with 30 influenza A negative samples (UTM). The high concentration positive samples were run adjacent to negative samples in 10 consecutive runs on one ARIES® System. No carry-over or cross contamination was observed, and the overall percent agreement with expected results was 100.0% for positive and negative samples.

Analytical Reactivity (Inclusivity)

The analytical reactivity/inclusivity of the ARIES® Flu A/B & RSV Assay was evaluated with multiple strains of influenza A H1N1 (pre-2009), influenza A H1N1 (pandemic 2009), influenza A H3N2 (seasonal and variant), avian influenza A (H5N1, H7N2, H9N2), influenza B (Yamagata and Victoria lineage), and RSV (A and B subtypes). A total of 34 unique strains were tested in triplicate at concentrations near the LoD. All reactivity strains for all organisms were detected by the ARIES® Flu A/B & RSV Assay at the indicated concentrations. See *Table 11,* on page 18.

TABLE 11. **Analytical Reactivity for the ARIES® Flu A/B & RSV Assay**

Inclusivity Strains	Lowest Concentration Detected ($TCID_{50}$/mL)
Influenza A /Perth/16/2009 (H3N2)-like	$1x10^1$
Influenza A/Brisbane/10/07 H3	$1x10^1$
Influenza A/Brisbane/59/07 H1	$1x10^1$
Influenza A/Port Chalmers/1/73 H3N2	$1x10^1$
Influenza A/Solomon Island/03/06 H1	$1x10^1$
Influenza A/Swine H1N1/Iowa/15/1930	$1x10^1$
Influenza A/Taiwan/42/06 H1N1	$1x10^1$
Influenza A/Wisconsin/67/05 H3	$1x10^1$
Influenza A/California/7/2009-like (pH1N1)	$1x10^1$
Influenza A/Hong Kong/33982/2009 H9N2 x PR8-IDCDC-RG26	$1x10^1$
Influenza A/Indiana/08/2011 (H3N2)v	$1x10^1$
Influenza A/Texas/50/2012 H3N2	$1x10^1$
Influenza A/WS/33 H1N1	$1x10^1$
Influenza A/New Caledonia/20/99 H1N1	$1x10^2$
Influenza A/Swine H1N1/USA/1976/1931	$1x10^2$
Influenza A/California/07/2009 NYMC x-179A	$1x10^2$

Influenza A/Victoria/361/2011-like (H3N2)	$1x10^2$
Influenza A/Minnesota/11/2010 (H3N2)v	$1x10^3$
Influenza A/Ohio/02/2012 (H3N2)	$1x10^3$
Influenza A/Anhui/01/2005 (H5N1)	$1x10^1$
Influenza A/Anhui/1/2013 (H7N9)	$1x10^1$
Influenza A/Egypt/321/2007 (H5N1)	$1x10^1$
Influenza A/Shanghai/1/2013 (H7N9)	$1x10^1$
Influenza A/Vietnam/1194/2004 (H5N1)	$1x10^1$
Influenza B/Massachusetts/2/2012-like	$1x10^1$
Influenza B/Wisconsin/1/2010-like	$1x10^1$
Influenza B/Florida/02/2006 (Victoria)	$1x10^1$
Influenza B/Lee/40	$1x10^1$
Influenza B/Panama/45/90 (Yamagata)	$1x10^1$
Influenza B/Brisbane/60/2008	$1x10^1$
Influenza B/Florida/07/04 (Yamagata)	$1x10^2$
RSV A/Long	$1x10^1$
RSV B/9320	$1x10^1$
RSV B/Wash/18537/62	$1x10^1$

Reproducibility (Site-to-Site)

Reproducibility of the ARIES® Flu A/B & RSV Assay was evaluated by testing one lot of ARIES® Flu A/B & RSV Assay Cassettes on two ARIES® Systems by two operators at each of three sites on five non-consecutive days. A reproducibility panel was prepared containing a moderate positive (10x LoD), low positive (3x LoD), and high negative (0.2x LoD) independently for influenza A, influenza B, RSV-A, and RSV-B, as well as a negative sample. The reproducibility panels were created by an independent operator and blinded to the testing sites. The result of the reproducibility study is shown in *Table 12,* on page 20. Site-to-site Reproducibility Ct and T_m results are shown in *Table 13,* on page 21.

TABLE 12. **Site-to-Site Reproducibility for the ARIES® Flu A/B & RSV Assay**

Strain	Target Concentration	Agreement with Expected Results[a]					
		Site 1		Site 2		Site 3	
Influenza A	High Negative	27/30	90.0%	30/30	100.0%	26/30	86.7%
	Low Positive	30/30	100.0%	30/30	100.0%	30/30	100.0%
	Moderate Positive	30/30	100.0%	30/30	100.0%	30/30	100.0%
	Negative	1/30	3.3%	0/30	0.0%	0/30	0.0%
Influenza B	High Negative	9/30	30.0%	7/30	23.3%	15/30	50.0%
	Low Positive	29/30	96.7%	30/30	100.0%	30/30	100.0%
	Moderate Positive	30/30	100.0%	30/30	100.0%	30/30	100.0%
	Negative	0/30	0.0%	0/30	0.0%	0/30	0.0%
RSV[b]	High Negative	39/60	65.0%	50/60	83.3%	43/60	71.7%
	Low Positive	60/60	100.0%	60/60	100.0%	60/60	100.0%
	Moderate Positive	60/60	100.0%	60/60	100.0%	59/60	98.3%
	Negative	0/30	0.0%	0/30	0.0%	0/30	0.0%

[a] The expected result for: a moderate positive target was 100.0% positive, low positive target was approximately 95.0% positive, high negative was 20.0% to 80.0% positive, and negative was 0.0% positive.

[b] RSV-A and RSV-B are not differentiated by the ARIES Flu A/B & RSV Assay and, therefore, are combined and represented as RSV only.

TABLE 13. **Site-to-Site Reproducibility Ct and T_m Results for the ARIES® Flu A/B & RSV Assay**

		Site 1				Site 2				Site 3				Overall Results			
		Avg Ct	Ct %CV	Avg Tm	Tm %CV	Avg Ct	Ct %CV	Avg Tm	Tm %CV	Avg Ct	Ct %CV	Avg Tm	Tm %CV	Avg Ct	Ct% CV	Avg Tm	Tm %CV
Influenza A	Moderate Positive	31.5	9.84%	83.3	0.09%	32.2	2.39%	83.4	0.07%	32.2	2.28%	83.4	0.12%	32.0	5.94%	83.4	0.10%
	Low Positive	34.2	2.59%	83.4	0.11%	34.1	3.00%	83.4	0.11%	34.3	2.95%	83.3	0.10%	34.2	2.83%	83.4	0.11%
	High Negative	38.4	1.74%	83.2	0.12%	38.2	2.25%	83.3	0.11%	38.2	2.34%	83.2	0.10%	38.3	2.12%	83.2	0.11%
	Negative[a]	29.7	3.95%	78.5	0.27%	28.3	4.07%	78.8	0.31%	29.7	3.66%	78.4	0.38%				
Influenza B	Moderate Positive	35.0	2.30%	79.4	0.11%	35.2	1.97%	79.4	0.10%	35.1	1.70%	79.5	0.10%	35.1	2.00%	79.4	0.11%
	Low Positive	36.7	2.56%	79.5	0.10%	36.7	2.71%	79.4	0.12%	36.6	2.51%	79.4	0.10%	36.7	2.57%	79.4	0.11%
	High Negative	37.7	2.33%	79.4	0.13%	37.3	2.63%	79.4	0.10%	38.0	2.15%	79.4	0.07%	37.8	2.34%	79.4	0.09%
	Negative[a]	29.7	3.95%	78.5	0.27%	28.3	4.07%	78.8	0.31%	29.7	3.66%	78.4	0.38%				
RSV[b]	Moderate Positive	34.3	4.51%	75.9	0.34%	34.1	3.12%	75.9	0.36%	34.3	3.63%	75.9	0.33%	34.2	3.79%	75.9	0.34%
	Low Positive	36.0	3.92%	75.9	0.33%	35.6	3.49%	75.9	0.37%	35.8	4.05%	75.9	0.35%	35.8	3.83%	75.9	0.35%
	High Negative	37.3	4.65%	75.9	0.40%	37.0	4.21%	75.9	0.35%	37.3	4.00%	75.9	0.36%	37.2	4.27%	75.9	0.36%
	Negative[a]	29.7	3.95%	78.5	0.27%	28.3	4.07%	78.8	0.31%	29.7	3.66%	78.4	0.38%				
Flu A/B& RSV	Negative[a]													29.2	4.45%	78.5	0.38%

[a] Ct and T_m values for the Influenza A/B & RSV Negative reflects RNA SPC values.

[b] RSV-A and RSV-B are not differentiated by the ARIES Flu A/B & RSV Assay and, therefore, are combined and represented as RSV only.

Precision

Within Laboratory Precision of the ARIES® Flu A/B & RSV Assay was evaluated by two operators performing testing across multiple ARIES® Systems using multiple lots of ARIES® Flu A/B & RSV Assay cassettes. Testing was performed on 12 nonconsecutive days and included a total of 675 replicates of a representative reproducibility panel. The reproducibility panel contains influenza A, influenza B, RSV-A, and RSV-B representative viral cultures diluted to three concentrations: moderate positive (10x LoD), low positive (3x LoD), and high negative (0.125x LoD for influenza A, and 0.25x LoD for influenza B, RSV-A, and RSV-B). All dilutions were prepared in simulated nasal matrix (SNM); negative samples consisted of only SNM. See *Table 14,* on page 22.

TABLE 14. **Within Laboratory Precision Results for the ARIES® Flu A/B & RSV Assay**

Strain	Target Concentration	Agreement with Expected Results[a]	95.0% Confidence Interval
Influenza A Hong Kong/8/68	**Moderate Positive**	100.0% (48/48)	93.0% - 100.0%
	Low Positive	100.0% (48/48)	93.0% - 100.0%
	High Negative	79.0% (38/48)	65.0% - 90.0%
Influenza B Florida/04/06[b]	**Moderate Positive**	100.0% (48/48)	93.0% - 100.0%
	Low Positive	100.0% (48/48)	93.0% - 100.0%
	High Negative	81.0% (39/48)	67.0% - 91.0%
RSV A2	**Moderate Positive**	100.0% (48/48)	93.0% - 100.0%
	Low Positive	96.0% (46/48)	86.0% - 99.0%
	High Negative	48.0% (23/48)	33.0% - 63.0%
RSV B WV/14617/85[b]	**Moderate Positive**	100.0% (48/48)	93.0% - 100.0%
	Low Positive	100.0% (48/48)	93.0% - 100.0%
	High Negative	90.0% (43/48)	77.0% - 97.0%
Flu A/B & RSV Negative[c]	**Negative**	2.1% (2/96)	0.0% - 7.0%

[a] The expected result for: a moderate positive target was 100.0% positive, low positive target was approximately 95.0% positive, high negative was 20.0% to 80.0% positive, and negative was 0.0% positive.

[b] Influenza B/Florida/04/06 and RSV-B WV/14617/85 high negative samples generated positivity that exceeded the expected positivity results of 20.0% to 80.0%.

[c] Two Flu A/B & RSV negative replicates tested by different operators, on different test dates, on different instruments, with different cassette lots generated false influenza B positive results.

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