Table of Content

Introduction

NxTAG® Respiratory Pathogen Panel + SARS-CoV-2 (NxTAG RPP + SARS-CoV-2) for use on Luminex® MAGPIX® instru-ment is a multiplexed nucleic acid RT-PCR test intended for the qualitative detection of nucleic acid from multiple respiratory viral and bacterial organisms, including the SARS-CoV-2 in upper respiratory tract specimens.

NxTAG RPP + SARS-CoV-2 detects and differentiates nucleic acids from SARS-CoV-2 and the following organism types and subtypes: Influenza A, Influenza A H1, Influenza A H3, Influenza B, Influenza A 2009 H1N1, Legionella pneumophila, Respiratory Syncytial Virus A, Respiratory Syncytial Virus B, Coronavirus 229E, Coronavirus OC43, Coronavirus NL63, Coronavirus HKU1, Human Metapneumovirus, Rhinovirus/Enterovirus, Adenovirus, Parainfluenza virus 1, Parainfluenza virus 2, Parainfluenza virus 3, Parainfluenza virus 4, Human Bocavirus, Chlamydophila pneumoniae, and Mycoplasma pneumoniae.

Targets Probed by the NxTAG™ RPP

Viral Targets	**Bacterial Targets**
Influenza A	*Chlamydophila pneumoniae*
Influenza A – H1 subtype	*Mycoplasma pneumoniae*
Influenza A – H3 subtype	*Legionella pneumophila*
Influenza A -2009 H1N1	
Influenza B	
Respiratory Syncytial Virus A	
Respiratory Syncytial Virus B	
Coronavirus 229E	
Coronavirus OC43	
Coronavirus NL63	
Coronavirus HKU1	
Human Metapneumovirus	
Rhinovirus/Enterovirus	
Adenovirus	
Parainfluenza 1	
Parainfluenza 2	
Parainfluenza 3	
Parainfluenza 4	
Human Bocavirus	
SARS CoV-2 (ORF1 ab)	
SARS CoV-2 (M)	

Method Overview

The NxTAG® Respiratory Pathogen Panel + SARS-CoV-2 (NxTAG RPP + SARS-CoV-2) incorporates multiplex Reverse Transcriptase Polymerase Chain Reaction (RT-PCR) with the Luminex® proprietary universal tag sorting system on the Luminex platform to detect respiratory pathogen targets. Extracted total nucleic acid is added to pre-plated, Lyophilized Bead Reagents (LBRs), and mixed to resuspend the reaction reagents. The reaction is amplified via RT-PCR and the reaction product undergoes near simultaneous microsphere hybridization within the sealed reaction well. The hybridized, tagged microspheres are then sorted and read on the MAGPIX® instrument. The generated signals are analyzed using the NxTAG Respiratory Pathogen Panel + SARS-CoV-2 Assay File for SYNCT™ Software, providing a reliable, qualitative call for each of the 23 targets and internal controls within each reaction well.

MAGPIX® Technology

The MAGPIX® system operates by using magnetic beads (microspheres) that are coated with a reagent specific to a particular bioassay, enabling the capture and detection of specific analytes from a sample. The sample mixture is aspirated by the sample probe and conveyed via Drive Fluid into the camera chamber, where the beads are pulled down into a monolayer by the magnet, immobilized, and imaged. Within the chamber, beads are exposed to a red LED and a green LED, which excite both the internal dyes that identify each bead's color signature and the reporter fluorescence from the surface of the beads. The red LED is responsible for classifying the beads. The CL1 and CL2 filters function to categorize the beads based on color signature and place them properly on the bead map as well as throw out any doublets that may exist. The green LED with the RP1 filter excites the reporter fluorescence, which identifies the quantity of analyte captured for each bead region. The beads are then flushed to the waste container, clearing room for the next sample.

Calibration is important to ensure that the optical system functions effectively and that different Luminex® MAGPIX systems report similar results. Calibrating the MAGPIX system normalizes the settings for the classification channels (CL1 and CL2) and the reporter channel (RP1). Use the Luminex MAGPIX Calibration Kit to accomplish this.

Following calibration, use the Luminex MAGPIX Performance Verification Kit to check all of the optical channels in the system for correct calibration. It is essential to verify every time you calibrate. If there is a problem with optical integrity or fluidics, MAGPIX may pass calibration but fail performance verification. The Luminex MAGPIX Performance Verification Kit includes reagents to verify the calibration and optical integrity for the Luminex MAGPIX system as well as reagents to verify the fluidics channels using observations of bead count and well-to-well carryover.

Materials

Kit Provided: NxTAG™ Respiratory Pathogen Panel + SARSCoV-2

Reagents	Volume for 96 Tests	Storage Conditions
NxTAG™ Respiratory Pathogen Panel + SARS-CoV-2 plate	1-96 well plate containing 2 Lyophilized Bead Reagents per well	Store at 2 ° C to 8° C in the resealable pouch provided: avoid exposure to light and moisture
MS2	1.5 mL X 2vials	Store at -25° C to 8° C
Foil Seals	8 pieces X 1 case	Store at 2° C to 30° C Store at 15° C to 30° C after first use.

Equipment

- Computer
- Luminex® instrument (MAGPIX®) including xPONENT® Software, calibrators, verifiers, and controls
- Multichannel pipette or single channel pipette (10 µL to 200 µL, 1000-1200 µL)
- PCR cooler rack (Eppendorf® 022510509) or equivalent
- Nucleic acid extraction system-. NucliSENS® easyMAG® System with Generic protocol 2.0.1
- CFX 96 BioRad Thermal Cycler

Consumables

- Drive Fluid – Store at 15-30 ° C
- DNase/RNase-Free Water
- 0.1N NaOH
- 70% alcohol
- 0.1% Bleach
- NxTAG™ Probe Adjustment Strip (Cat # C000Z0452)
- Skirted Plate (Cat # C000Z0455) (96-well in white frame)

Specimen Collection and Nucleic Acid Extraction

Specimens can be stored between 2°C and 8°C for up to 7 days after collection in Universal Transport Media (UTM™) or equivalent. If the specimen is not going to be tested within 7 days of collection, then it should be stored at -80°C.

Pretreatment

Samples should be preheated at 65C for 15-30 minutes.

Nucleic Acid Extraction

- Please refer to Nucleic Acid Extraction-Biomerieux NucliSENS easyMAG
- For each run, extract 2 positive external controls (one positive for para or meta and one positive for covid) and 2 negative external controls (PCR degree water)
- Must name negative external controls as nc1 and nc2 (lower case)

Procedure

Luminex Daily Workflow

1. Prepare BioRad Thermocycler
2. Inoculate Strips - Keep everything cold
3. Load Strips into BioRad Thermocyler
4. Perform Daily Maintenance-pre assay
5. Create Batches (worklist)
6. Load strips into Magpix and read after BioRad PCR done
7. View Results in SYNCT
8. Perform Daily Maintenance-post assay

Prepare the BioRad Thermocycler

a. On the main screen of the Thermocycler choose Saved Files
b. Choose LMNX_RPP_PCR
c. Choose Run

   Sample volume 35 µL.
d. Press ok
e. Status will be in infinite hold. Do this before loading the plate as the Thermocycler needs to be preheating.

Inoculation of Strips

If frozen, thaw the extracted nucleic acid samples to be used for the run and place on ice or using a PCR cooler. If fresh, place extracted nucleic acid samples on a cold block and add directly to the plate setup.

1. Once the samples are thawed, briefly vortex the samples followed by a quick spin to collect the samples to the bottom. Set the samples back on ice or using a PCR cooler. Remove the assay plate from its storage pouch.
   NOTE: Protect the assay plate from excessive light.

2. Place the required number of vessels into a PCR set-up plate (skirted Plate for Bio-Rad® thermal cyclers). Place strips toward the center of the plate first as this remains coldest longest. Firmly press down on the strips to snap into place, ensuring they are flush with the plate surface. If using more than one strip, leave a blank row between strips. Return unused vessels to the pouch in the original tray and seal the zip lock of the silver pouch. Store the unused vessels at recommended condition.
3. If the Lyophilized Bead Reagents are not at the bottom of the wells, tap the plate once on the bench. The pink foam can be used to reduce static holding the lyophilized pellets at the top. Rub the strip on the pink foam to reduce static.

4. Place the plate on ice or using a PCR cooler and keep the plate cold during the setup.

5. Use the end-tabs to peel the clear release liner prior to the sample addition.

6. Perform the sample addition on ice or using a PCR cooler.

7. Using a precision pipette, add 35 µL of samples to each PCR vessel by piercing with the pipette tip through the foil at an angle. When piercing the foil be sure to NOT touch the black adhesive.
   a. Insert the tip a third to halfway down into the vessel.

   b. Dispense the sample into the vessel and wait 1 to 2 seconds while maintaining the pipette tip inside the vessel.

   c. Push the tips close to (but not touching) the bottom of the vessel and pipette up and down at least three times to reconstitute the pre-plated Lyophilized Bead Reagents.

8. Luminex® recommends using DNase/RNase-Free Water as negative control. Add 35 µL DNase/RNase-Free Water to a PCR vessel and pipette up and down at least 3 times to reconstitute the pre-plated Lyophilized Bead Reagents.

9. Reseal the plate after the sample addition using the foil provided. Apply the foil(s) directly on top of the plate and press firmly on and around the wells to ensure a tight seal.
   NOTE: Ensure the foil covers the wells and surrounding black adhesive.

10. Place the foil sealed plate in the pre-programmed and pre-heated thermal cycler.

Load strips into BioRad Thermocycler

1. Open lid of Thermocycler.
2. Load plate and close lid.
3. At the thermocycler screen press **skip step**.
4. Press Yes to “Are you sure you want to Skip test”
5. The run is 2 hours and 40 minutes (ensure that the thermocycler is counting down).
6. Proceed to section on Instrument Preparation for Data Acquisiton at least half an hour before the run is complete.
7. When the run is complete it will be in **infinite hold** on the Thermocycler screen.
8. Choose cancel on the BioRad screen to exit program.

9. Open and close lid as above instructions.

After the cycling program is complete, do not remove the seal. Transfer foil-sealed plate directly to the preheated MAGPIX®. The plate should be read by the MAGPIX instrument immediately after the end of cycle.

NOTE: Before you remove your sample from the PCR, prepare the instrument by turning on the heater and setting up your batch as stated in section "Instrument Preparation for Data Acquisition".

Perform Daily Maintenance – Pre Assay

1. **Check fluids level**

   Maintaining Fluids

   The MAGPIX® has a built-in compartment to hold a single-use disposable Drive Fluid container and a reusable waste fluid container.

   Monitor fluid levels daily. Replace the empty Drive Fluid container as needed. If the MAGPIX operates with an empty Drive Fluid container, the lack of Drive Fluid can interrupt a sample and prevent further samples from being collected.

   CAUTION: Only use the xMAP® Drive Fluid. Use of any other Drive Fluid constitutes improper use and can void the warranty provided by Luminex®, its authorized partner, or both.

   Empty the waste fluid container whenever the container is full. Use the following guidelines:

   - Replace the newly emptied waste fluid container with the second dry waste fluid container so the moisture remaining in the first waste fluid container does not cause a "waste bottle full" message.
   - Before removing the waste fluid container, make certain all other fittings and tubes are firmly attached to avoid any contamination from dripping waste fluid.

   To empty the waste fluid container:

   a. Open the fluid compartment at the bottom front of the MAGPIX.
   b. Disconnect the orange waste fluid line from the waste fluid container.
   c. Carefully remove the waste fluid container from its tray.

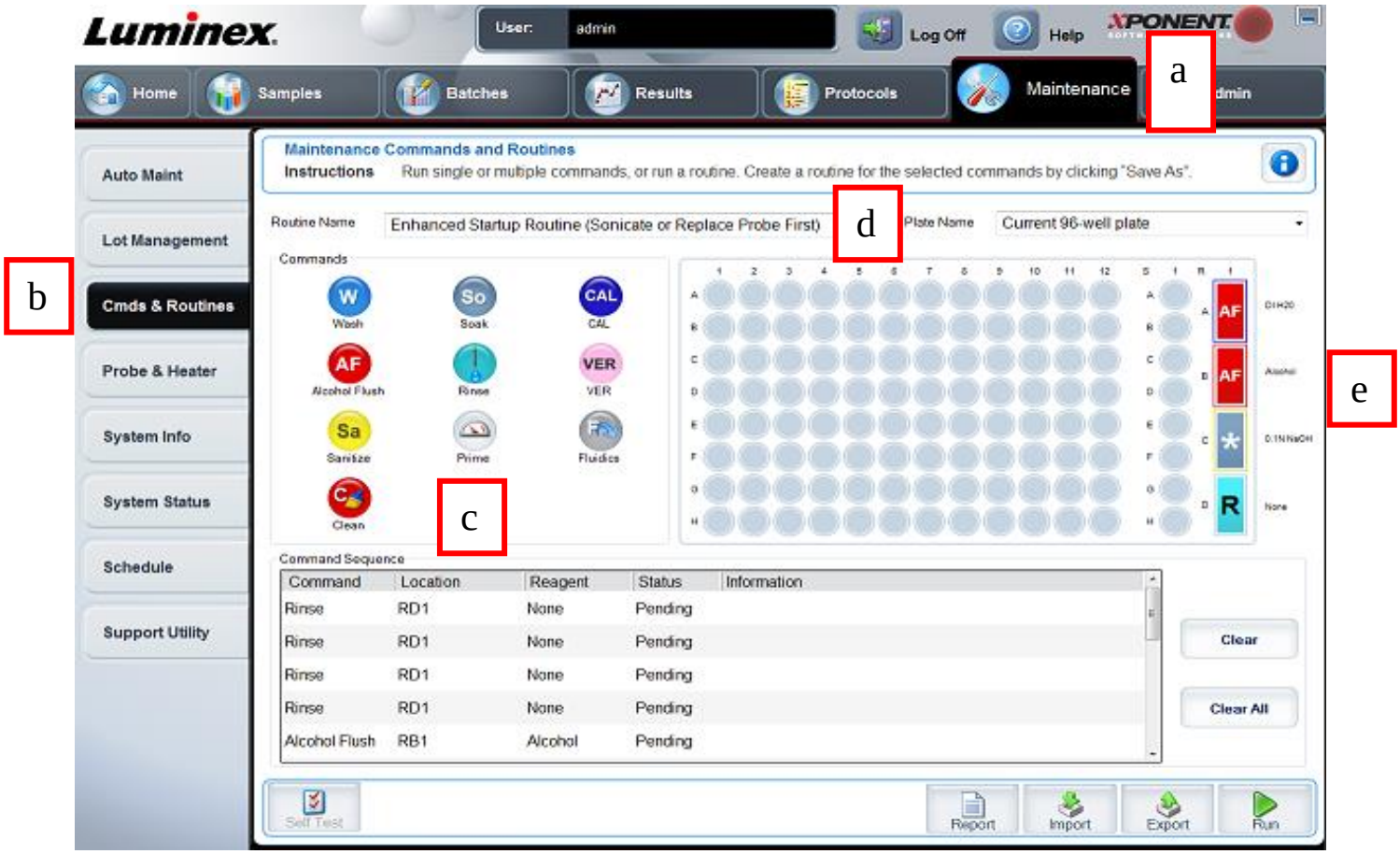
d. Unscrew the cap on top of the luminex waste fluid container and pour into the large liquid waste container located in the sink. The waste fluid should be disposed of as bioharzard waste (i.e. treated with javex overnight before pouring down drain). Rinse the luminex waste container with water and let dry.

e. Insert the second dry waste fluid container in the fluid compartment.

2. **Enhanced Startup Routine**

Double click Xponent icon

To login to XPonent : user ID: admin



a. Select **Maintenance** tab (horizontal bar)

b. Click **Cmds & Routines** (side bar)

c. Click **Prime** 3 times to prime the system

d. Under **Routine name** , select **Enhanced Startup Routine (Sonicate or Replace Probe First)** from the drop down menu.

e. Fill the reservoir as per screen.

(for 0.1N NaOH dilute 50mL of 1M NaOH in 450 mL of DI water. The 1M or 1N NaOH is found in the acid cabinet in the media room)

f. Click Eject to open door.

g. Load the reservoir as the instructions on the screen.

h. Click Retract to close door.

i. Click Run

j. When the Enhanced Startup Routine is complete, click OK.

3. **Set Probe Height (Procedure takes a few seconds to complete)**

a. Select **Maintenance** tab (horizontal bar)

b. Select **Probe and Heater** from the side bar.

c. Under plate name select **"current 96-well plate**" from drop down menu.

d. Click **Eject** to open the door

e. Place empty cal/ver strip to Strip wells

f. Place a white strip in a skirted tray and load the tray. Press the tray firmly down.

g. Put a bead in well D6

h. Click on the well 1,D6

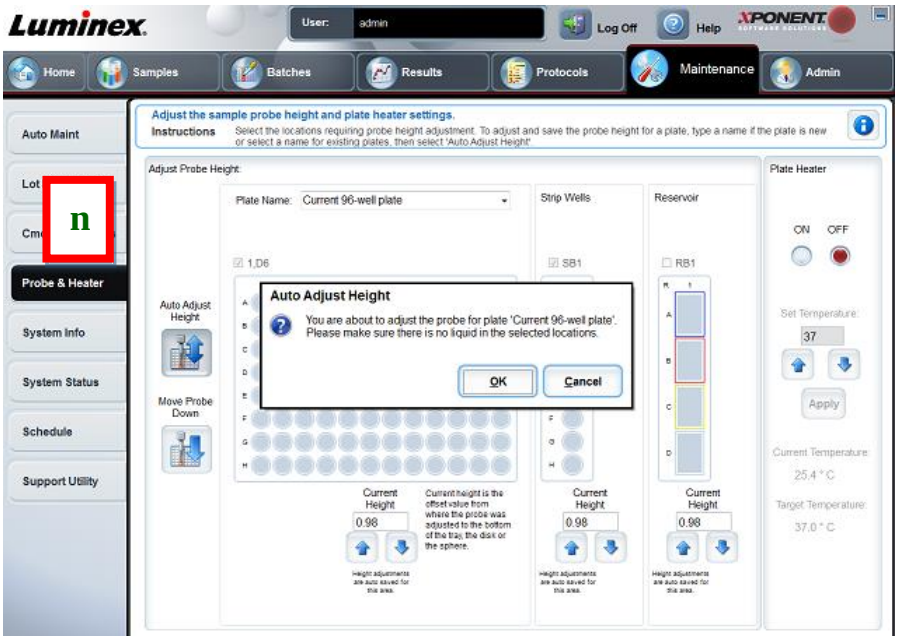
i. Choose and Check **SB1**

j. Choose and Check **RB1**

k. Click **Eject** to open door.

l. Click **Retract** to close door.

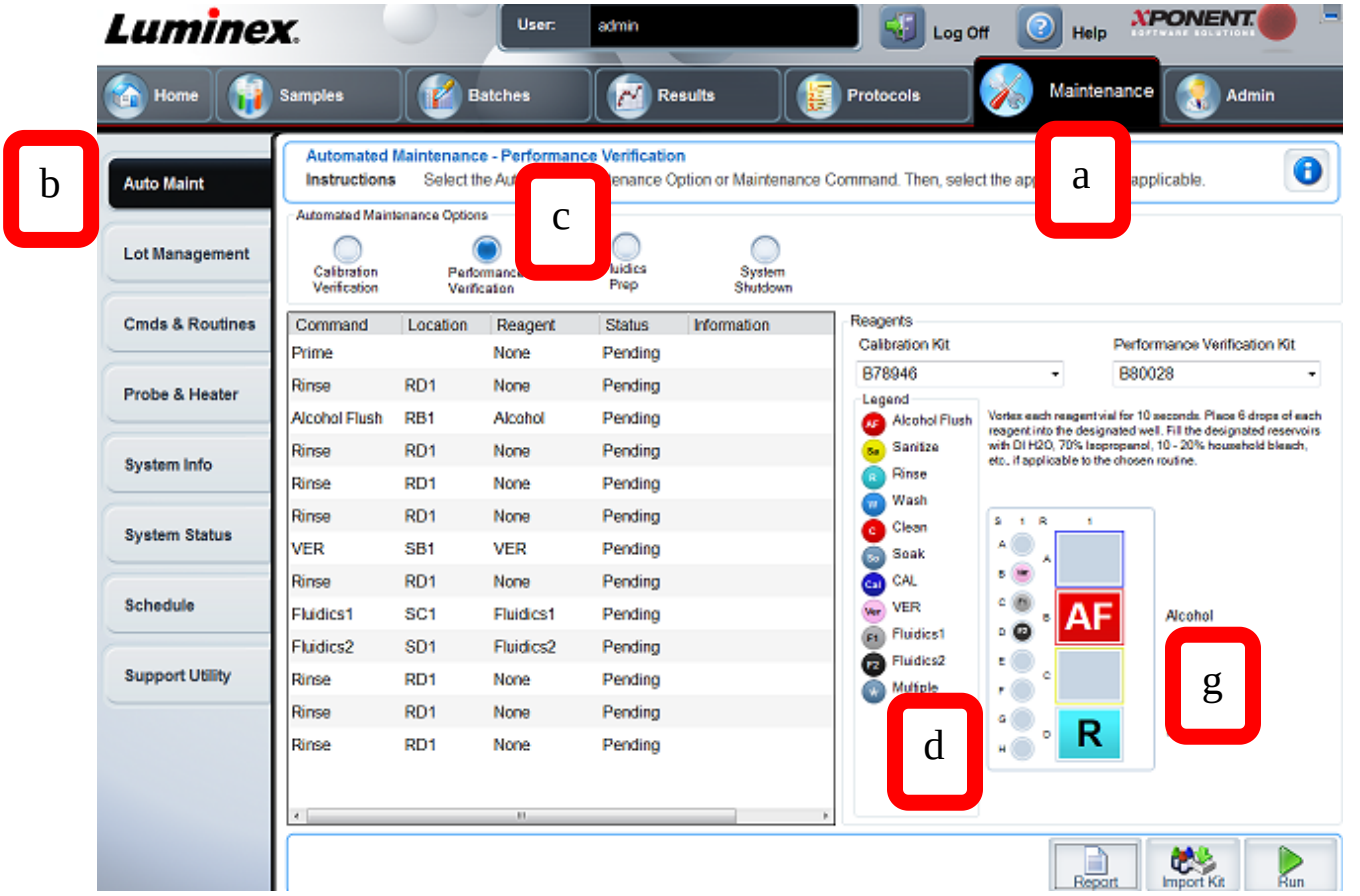
m. Click **Auto Adjust Height**



n. Auto Adjust Height window pops up, click **OK**

o. Select **Yes** to "Overwrite existing plate…. "

4. **Perform Verification daily (4 min) Calibration Verification (weekly)**



a. Click the **Maintenance** tab.

b. Click **Auto Maint** from the side bar.

c. Select **Performance Verification**.

d. Fill wells supplied with verification kit and reservoirs as per screen.

e. Vortex vials for 10 secs.

f. Add 6 drops to each of Ver (Daily), F1 (Daily), F2 (Daily) in the wells (provided in the Verification kit).

g. Fill the 2nd reservoir with 70% alcohol.

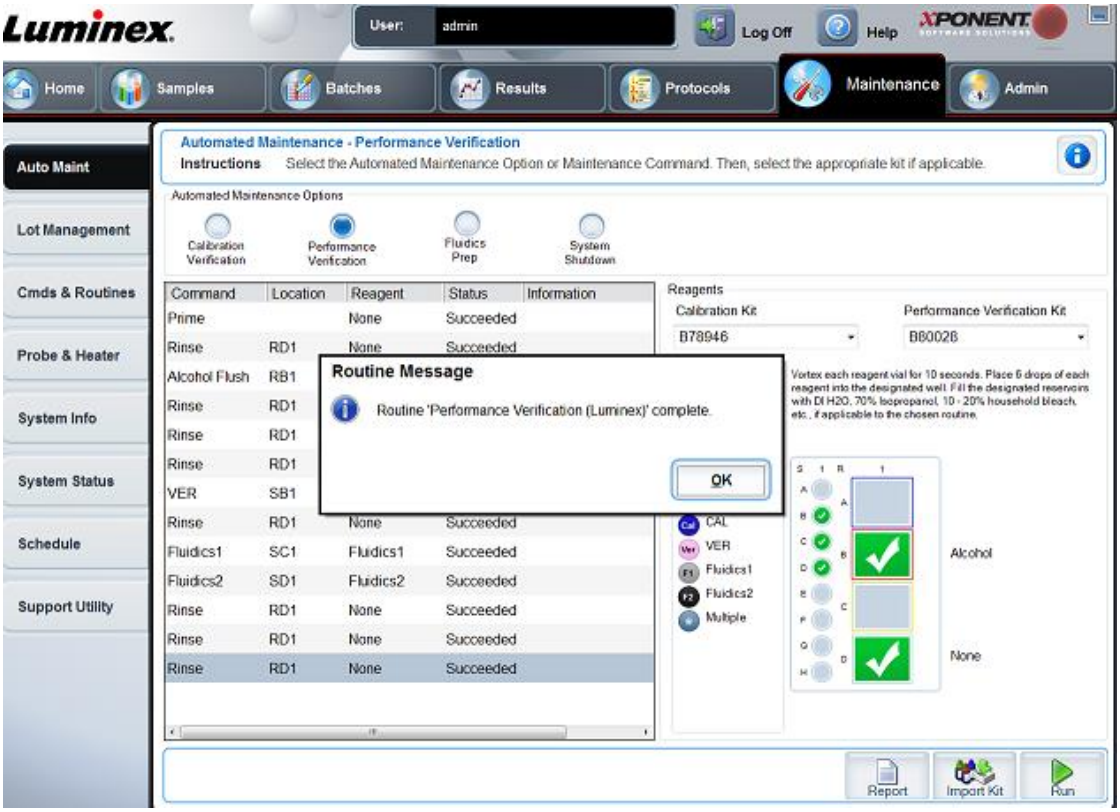
h. Click **Eject** to open door.

i. Load the wells in the space adjacent to the alcohol reservoir.

j. Click **Retract** to close door.

k. Press **Run**.

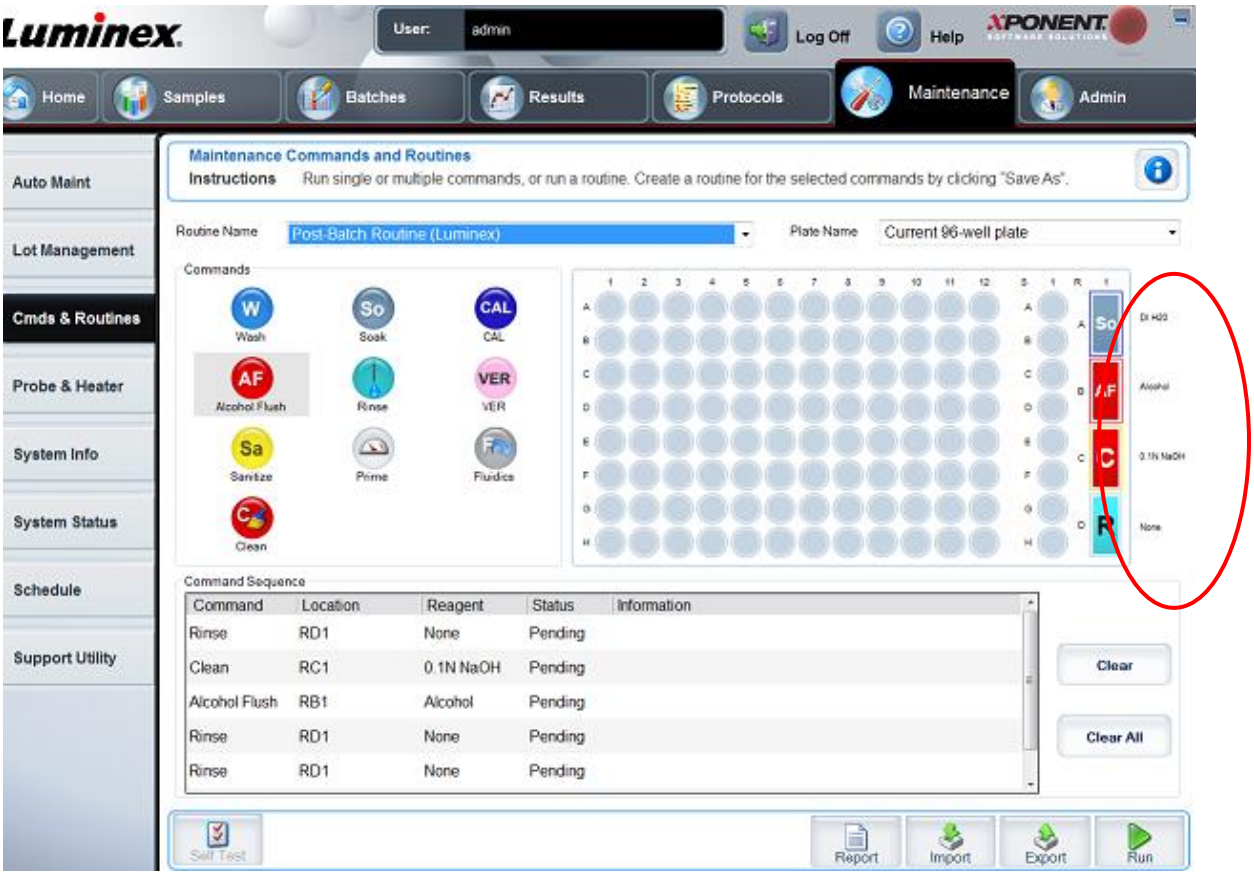
l. For calibration follow the same procedure but add 6 drops of CAL in the first well.



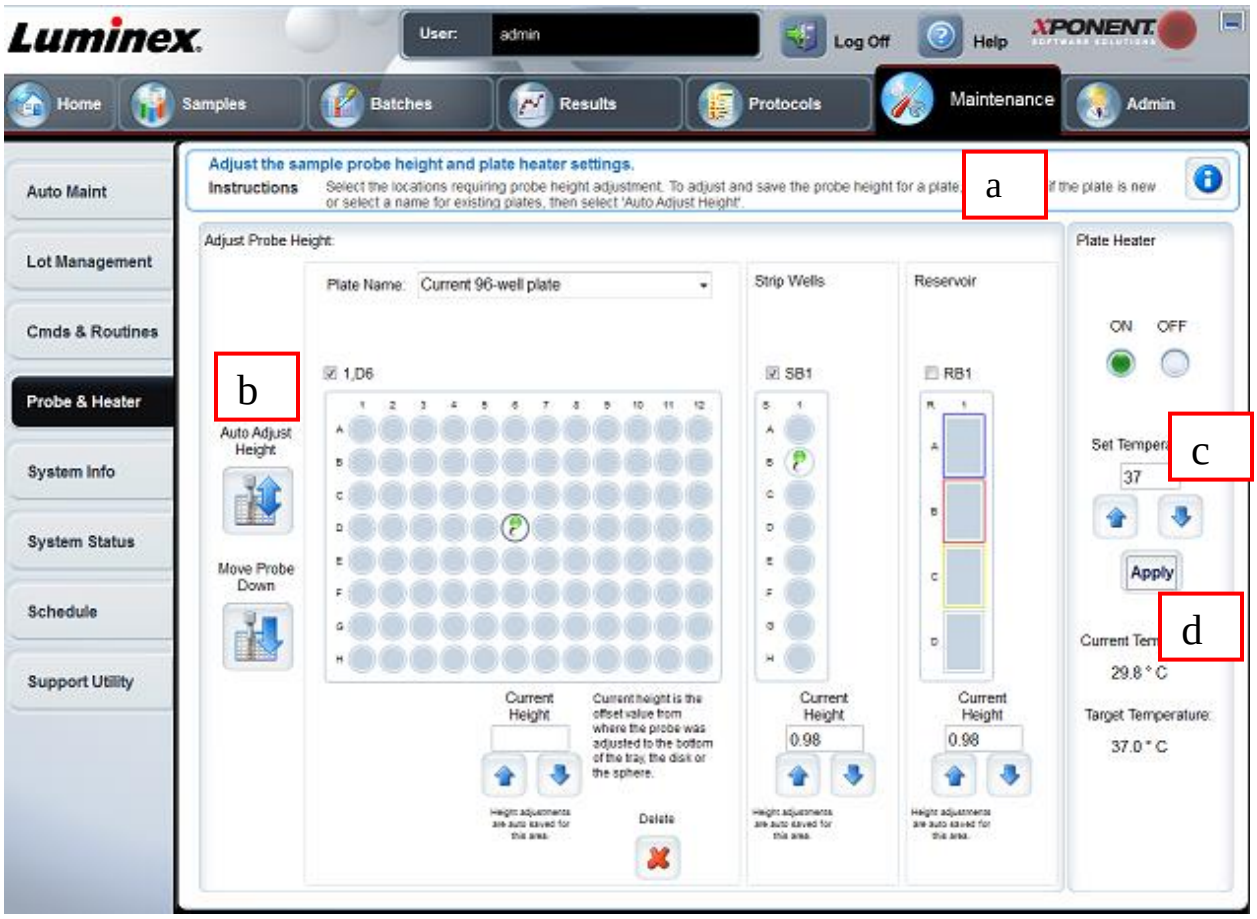
5. Load Post-Batch Reagents

Navigate to Maintenance page > Cmds & Routines tab>Post Batch Routine. Add appropriate reagents to the off-plate reagent reservoirs as specified by the Post-Batch Routine indicated in the software.

(for 0.1N NaOH dilute 50mL of 1M NaOH in 450mL of DI water.)



6. **Turn on Plate Heater to 37º C**



a. In Xponent choose Maintenance.

b. Select **Probe and Heater** from side bar.

c. Select **ON** under Plate Heater and enter 37 in the Set Temperature field to heat the MAGPIX® heater plate to 37°C.

d. Click **Apply**

Creating Batches

On main screen click on xPONENT icon.

Click **Login**

Click **Batches** Tab (3rd Tab from left)

Click **Create a New Batch from Existing Protocol**

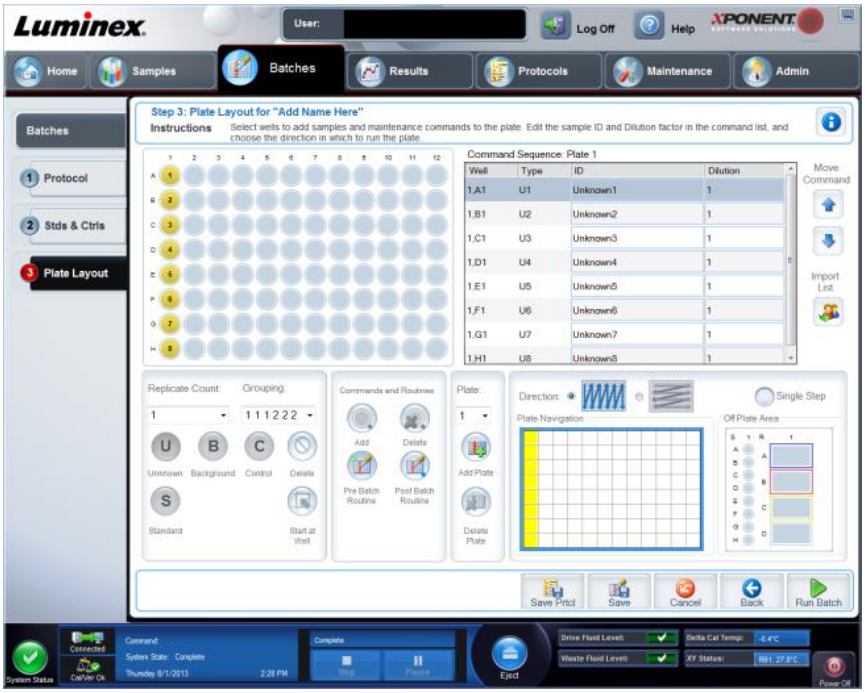
Choose **NxTAG RPP + SARS-CoV-2 26plex**

Next to Batch name: type in your run name (yearmonthday) no slashes

Click **Next**.

Highlight (by clicking or dragging) the appropriate wells where the samples will be analyzed.

Click **Unknown**.



Scan the sample's LIS numbers.

For the amplification negative controls (s) enter as nc1 and nc2

Click Save. The batch is now saved as a pending batch and ready to run.

To import the worklist from USB

Highlight the number of wells to be analyzed, click on import worklist.

From the USB, open the text file and check if the sample numbers correlate to your plate map.

Click **Save**

Load the plate into Magpix

Remove the plate from the Biorad thermocycler. Cancel Run on Thermocycler.

Immediately after completion of the cycling, analyze plate.

To open MagPix door, click Eject.

Place the plate on the prepared (37degrees) MAGPIX heater block.

If the probe height was adjusted with the skirted plate, ensure you put the sample on the skirted plate before placing on the heater block.

NOTE: Be sure to leave the seal in place.

NOTE: When placing the plate on the heater block, ensure that the numbers are on the left side and the letters are closest to you.

From the Batches page choose the batch from the pending batches list and click Run to start data acquisition. Verify information in warning dialog boxes and click OK.

After the last sample is read, navigate to Home > click Probe and Heater > click Eject to remove the plate from the heater block and turn OFF the heater.

Carefully discard the test vials to avoid aerosolization of the amplicons. If re-using the Skirted Plate, clean by soaking in a 10% bleach solution for 15 minutes. Rinse the Skirted Plate under running tap water to remove bleach, and air dry on paper towels or wipe with 70% alcohol for fast drying, if necessary.

Log off of Xponent and go to Main screen.

View Results in SYNCT

The Import Raw Data function allows a raw data (CSV) file from xPONENT® software to be imported. To manually import the xPONENT raw data into the SYNCT Software, perform the following steps:

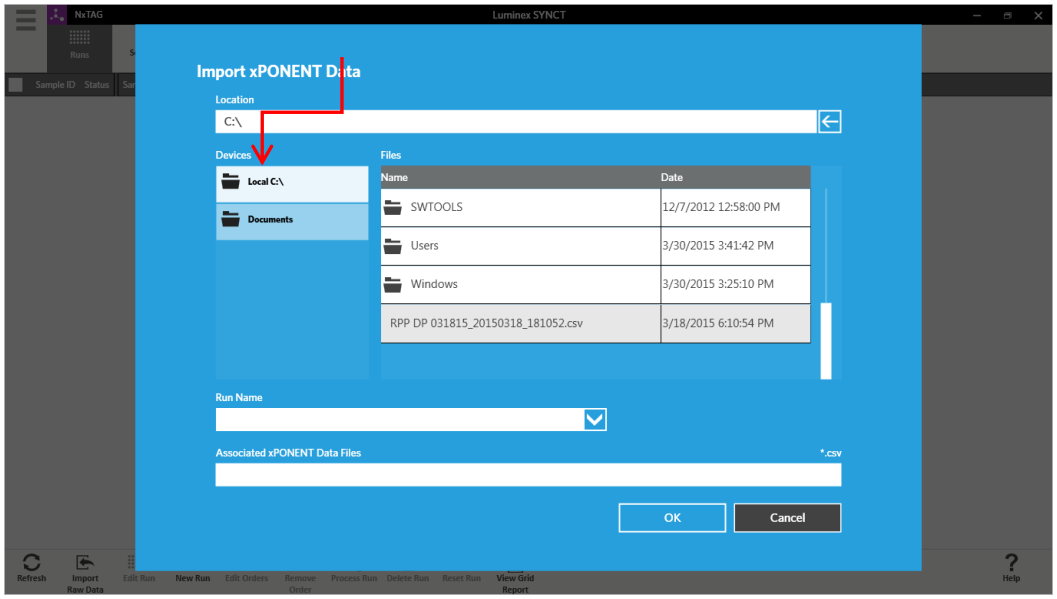
Choose the SYNCT icon on the Desktop

Username: admin

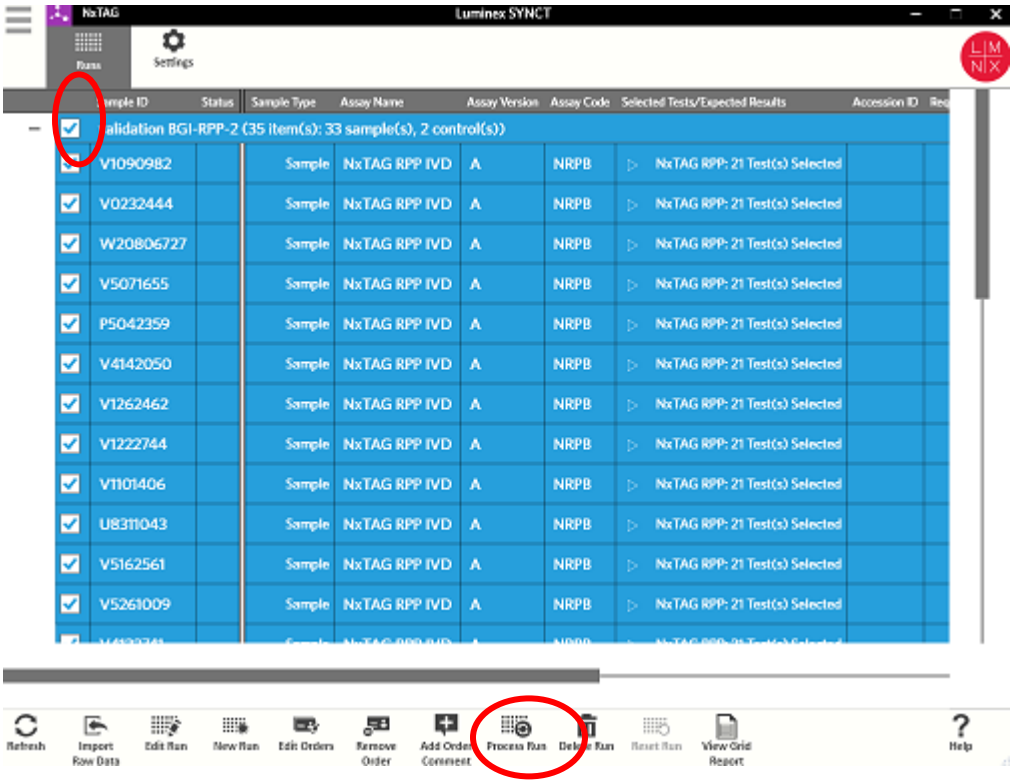
Password: password

Click Login

1. Click ☰ on the upper left-hand corner of the screen and navigate to **NxTAG**
2. Click Import Raw Data from the Page Action bar at bottom of page.
3. The Import xPONENT Data window displays
4. Choose the Location : C:\ProgramData\Luminex\xPONENT42\Output (should be there)



5. Under Files: Choose the run by scrolling down. The Run Name field is automatically populated with the Batch name from the xPONENT file
6. Press the + of the run file. Run will open
7. Click inside the box of the desired run.
8. Choose Process Run from the bottom menu-7th icon.

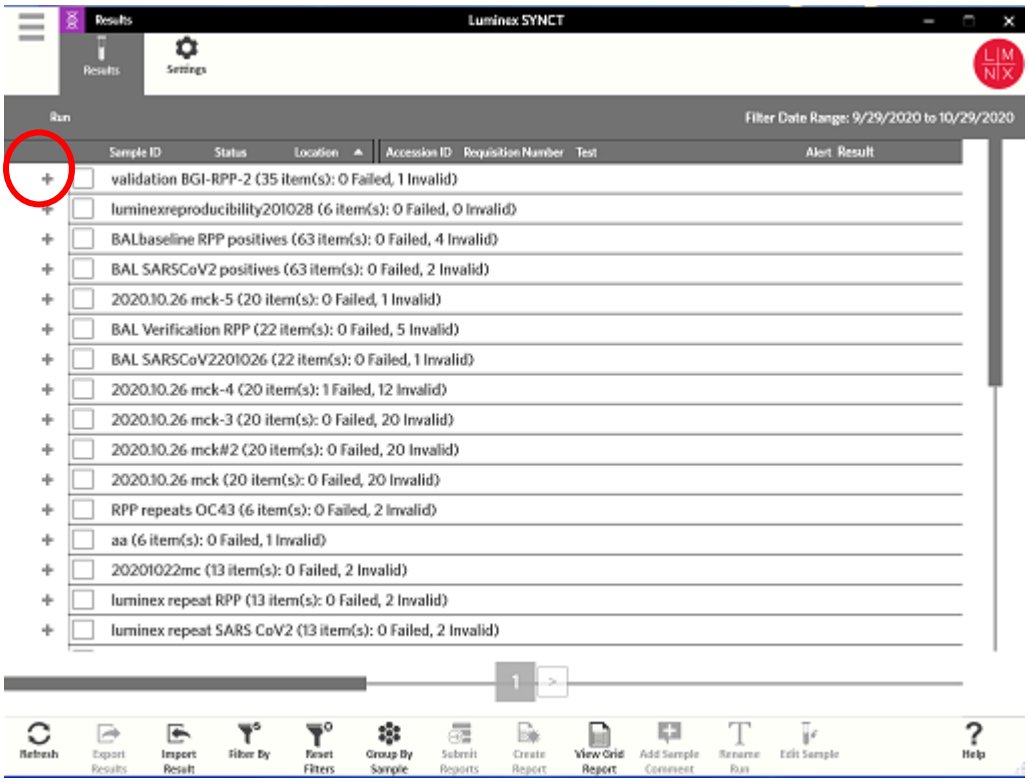


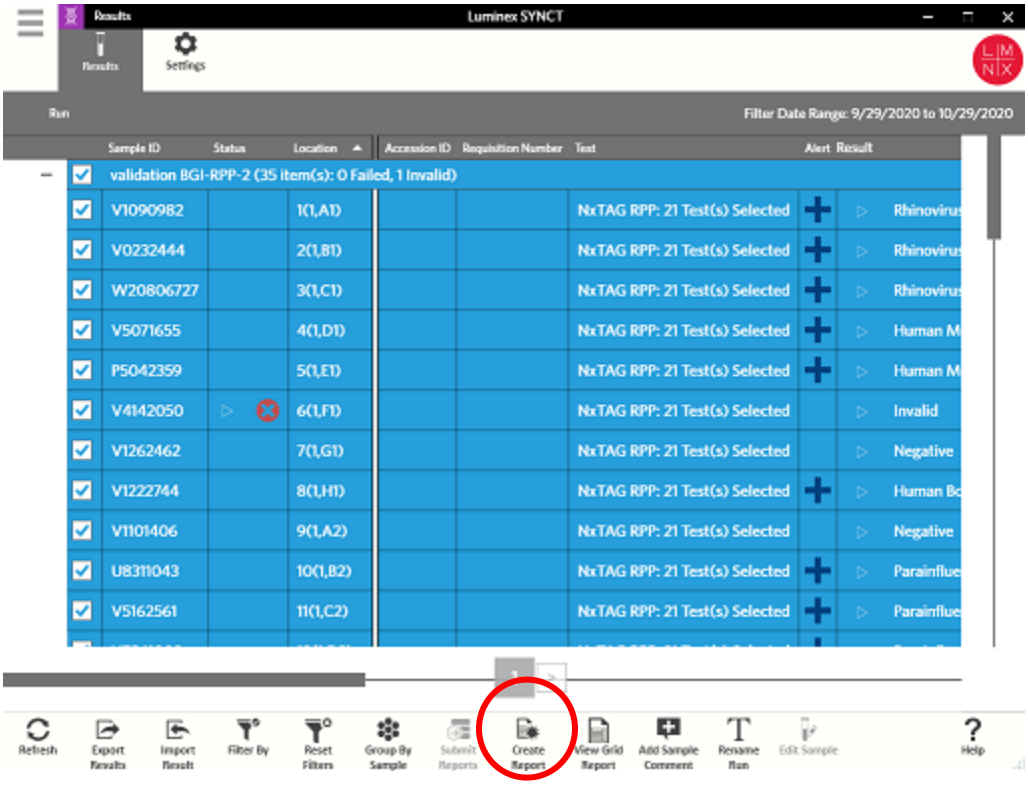
Pop-up "Run ……. analyzed successfully. Click OK.

Click on the menu.

Click **Results**. Runs will display.

Click on the + of your run.


Results
Luminex SYNCT
Results
Settings
Run
Filter Date Range: 9/29/2020 to 10/29/2020
Sample ID
Status
Location
Accession ID
Requisition Number
Test
Alert
Result
validation BGI-RPP-2 (35 item(s): 0 Failed, 1 Invalid)
luminexreproducibility201028 (6 item(s): 0 Failed, 0 Invalid)
BALbaseline RPP positives (63 item(s): 0 Failed, 4 Invalid)
BAL SARSCoV2 positives (63 item(s): 0 Failed, 2 Invalid)
2020.10.26 mck-5 (20 item(s): 0 Failed, 1 Invalid)
BAL Verification RPP (22 item(s): 0 Failed, 5 Invalid)
BAL SARSCoV2201026 (22 item(s): 0 Failed, 1 Invalid)
2020.10.26 mck-4 (20 item(s): 1 Failed, 12 Invalid)
2020.10.26 mck-3 (20 item(s): 0 Failed, 20 Invalid)
2020.10.26 mck#2 (20 item(s): 0 Failed, 20 Invalid)
2020.10.26 mck (20 item(s): 0 Failed, 20 Invalid)
RPP repeats OC43 (6 item(s): 0 Failed, 2 Invalid)
aa (6 item(s): 0 Failed, 1 Invalid)
20201022mc (13 item(s): 0 Failed, 2 Invalid)
luminex repeat RPP (13 item(s): 0 Failed, 2 Invalid)
luminex repeat SARS CoV2 (13 item(s): 0 Failed, 2 Invalid)
1
Refresh
Export Results
Import Result
Filter By
Reset Filters
Group By Sample
Submit Reports
Create Report
View Grid Report
Add Sample Comment
Rename Run
Edit Sample
Help



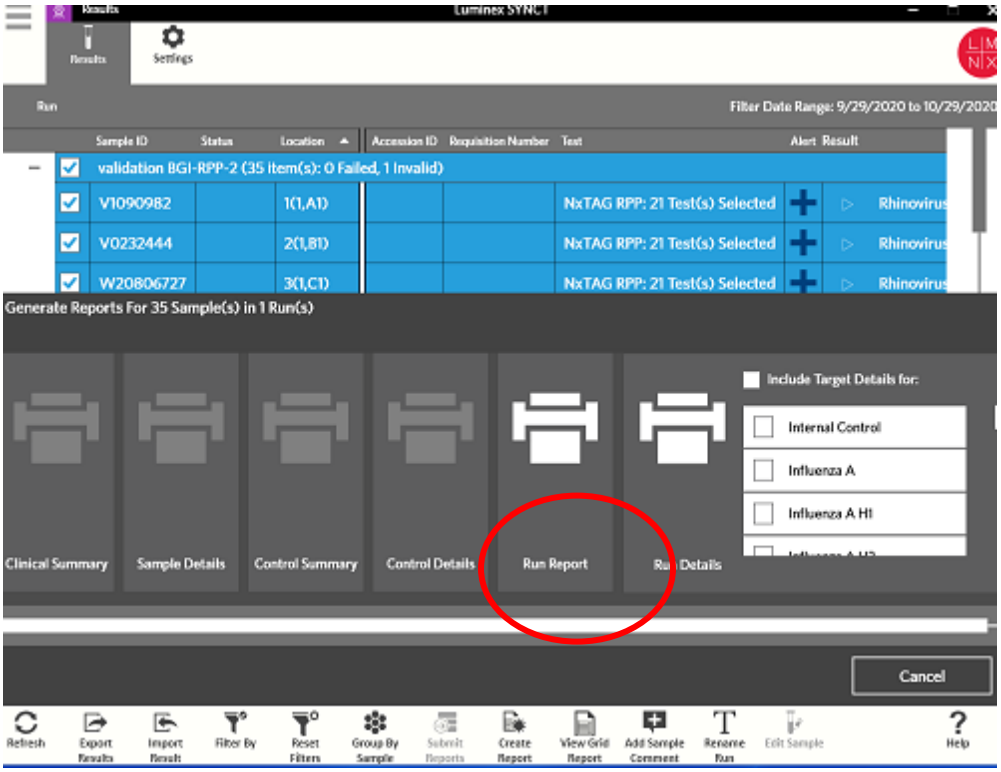
The status column indicates whether there are errors, warnings, info messages or user comments for a sample.

The Alert column indicates if any test has a positive result. If the positive the Alert column will display a + for that sample.

The Alert column indicates if a control has failed with an exclamation mark for that control. (!)

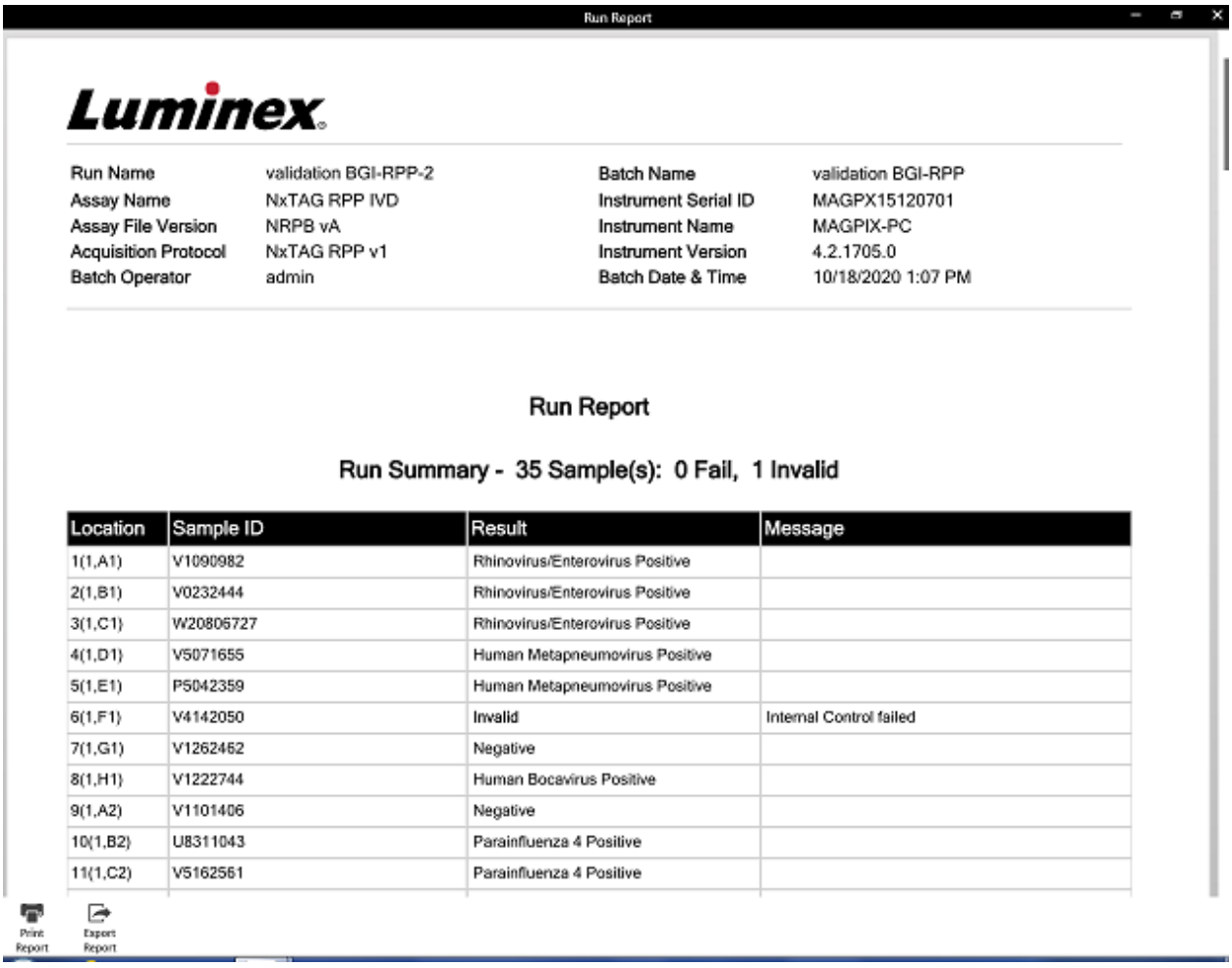
Click on the box before the run file.

Create report.



Click on the Run Report -> Print.

Report will look like this.

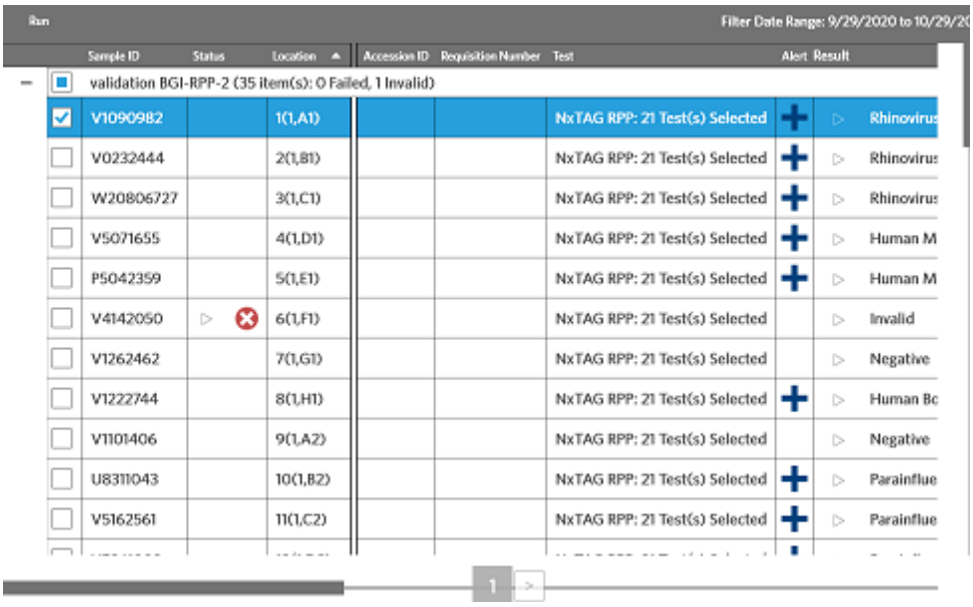


Print Report.

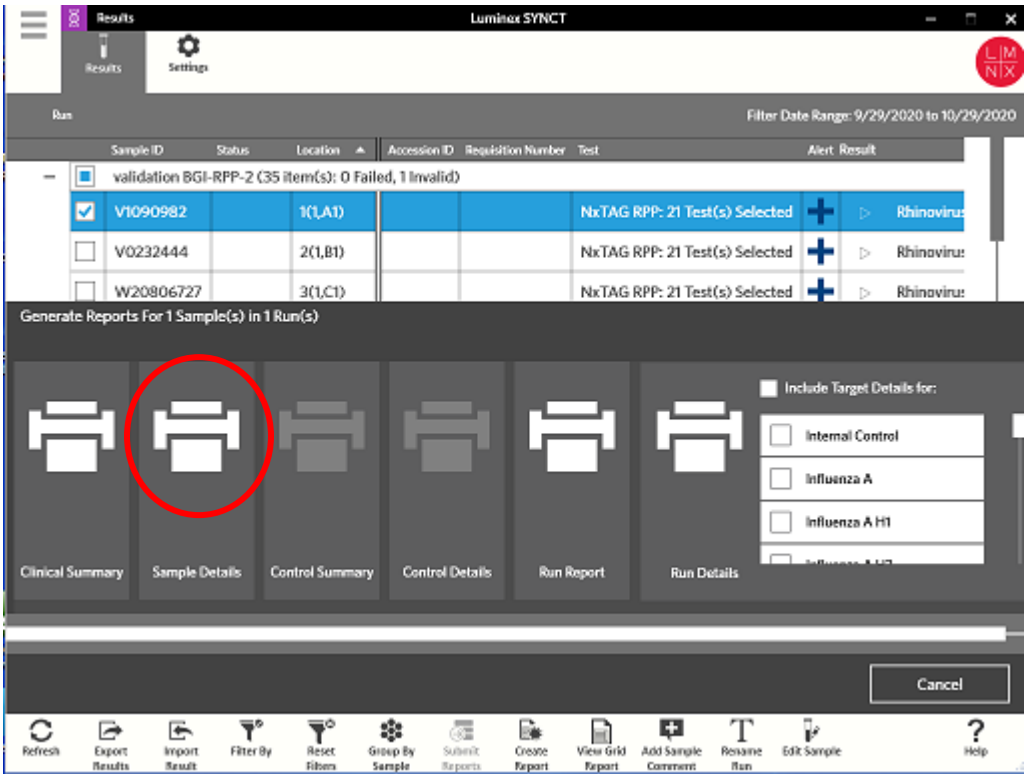
Exit.

Check the print outs for all positives and invalids.

Go back to SYNCT and uncheck the box of the run file and check box of the individual positives one by one to print individual reports.

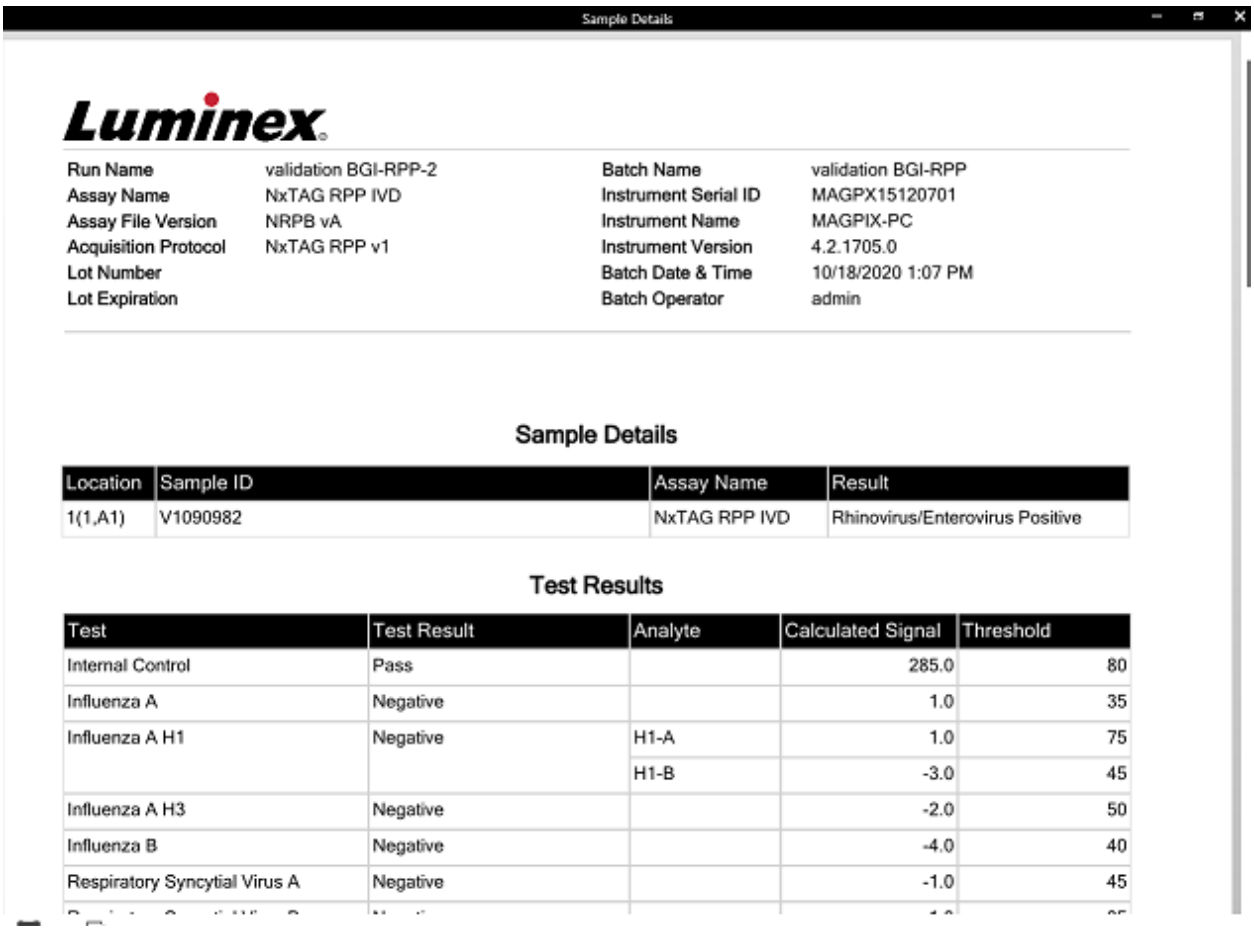


Create report



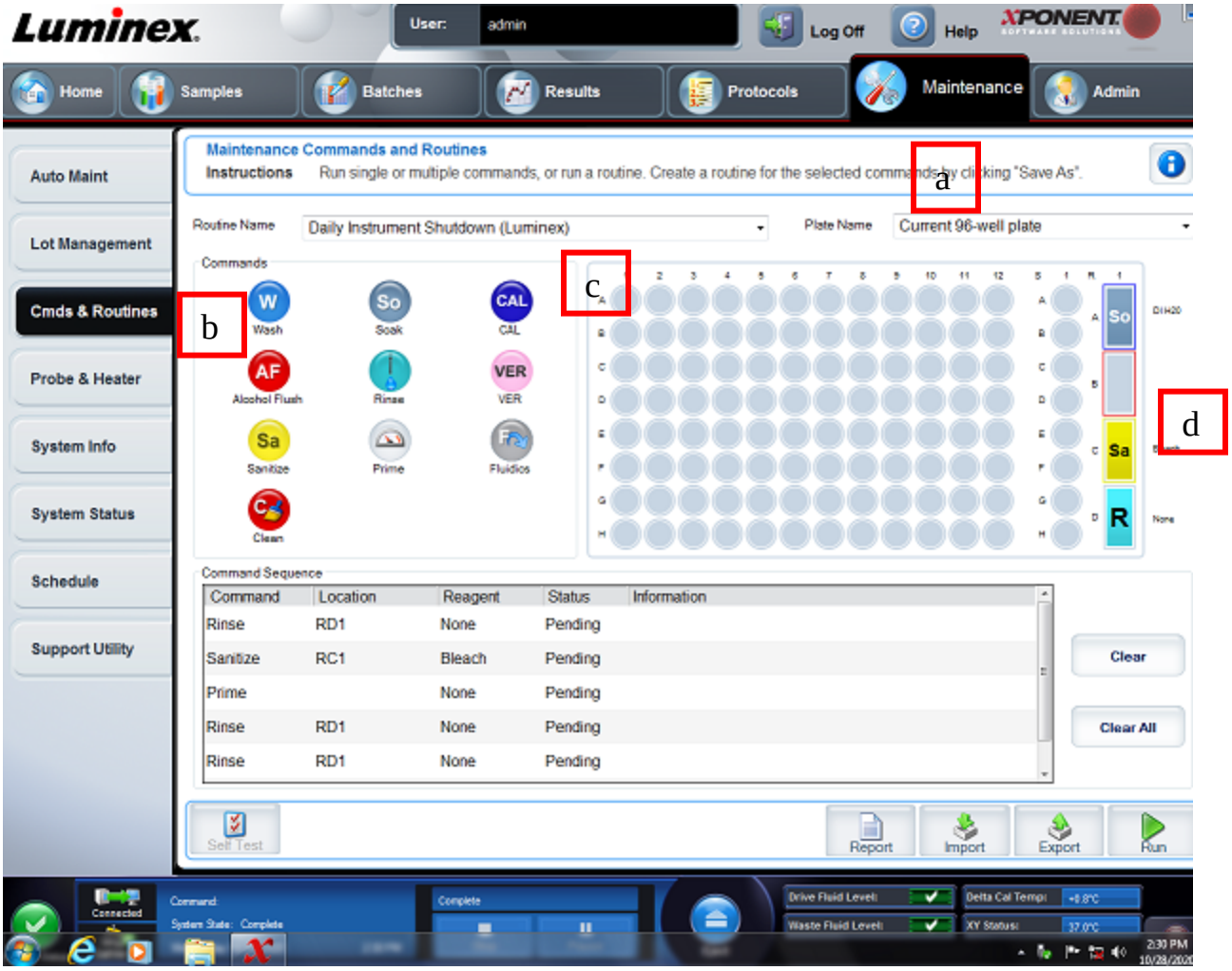
Click Sample details.

Will show individual targets



Perform Daily Maintenance - Post Assay

This procedure is for shutting down the MAGPIX and includes sanitize, clean (with 0.1 N NaOH), and soak routines.



a. Click maintenance tab (horizontal bar)

b. Click Cmds and routines (side bar)

c. Under Routine Name: choose Daily Instrument shut down from drop-down menu

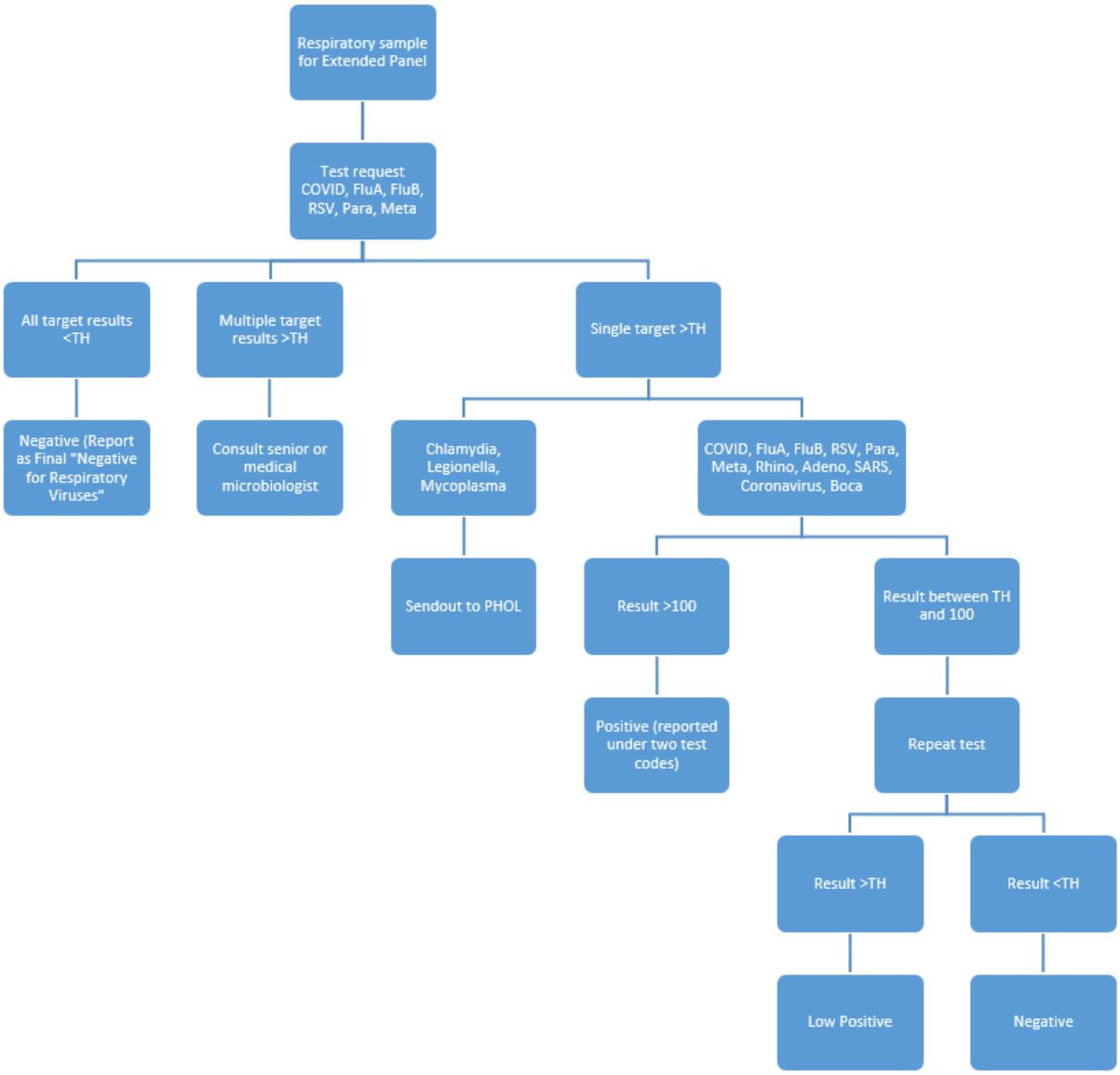
d. Fill reservoir as per screen.

e. After Shutdown procedure, empty reservoirs, rinse reservoirs with water and return to MagPix.

f. Log off all software.

g. Turn off Computer and Turn off MagPix.

Interpretation Chart



TH = Threshold (determined by Luminex for each target)

Talk to a Senior Technologist if results are questionable.

COVID can be reported positive based on either M gene or orf1ab gene >100 bead count.

Invalids are repeated. If a run has high number of invalids talk to a senior.

Reporting

- Inform all positive results according to Isolate notification and freezing document

1. **Negative report:**

```
 * SARS-CoV-2:                                    Not detected
 * Adenovirus:                                    Not detected
 * Coronavirus 229E                               Not detected
 * Coronavirus HKU1:                              Not detected
 * Coronavirus NL63:                              Not detected
 * Coronavirus OC43:                              Not detected
 * Human Bocavirus:                               Not detected
 * Human Metapneumovirus:                         Not detected
 * Influenza virus A:                             Not detected
 * Influenza virus A - subtype H1:                Not detected
 * Influenza virus A - subtype H3:                Not detected
 * Influenza virus B:                             Not detected
 * Parainfluenza 1:                               Not detected
 * Parainfluenza 2:                               Not detected
 * Parainfluenza 3:                               Not detected
 * Parainfluenza 4:                               Not detected
 * Respiratory Syncytial Virus A:                 Not detected
 * Respiratory Syncytial Virus B:                 Not detected
 * Rhinovirus/Enterovirus:                        Not detected
 * Chlamydophila pneumoniae:                      Not detected
 * Mycoplasma pneumoniae:                         Not detected
 * Method:                                        *NOTE*
   Testing performed using the Luminex NxTAG Respiratory
    Pathogen Panel (RPP) + CoV Assay. This assay detects
    SARS-CoV-2 as well as 18 non-SARS-CoV-2 respiratory
    viruses and 2 respiratory bacteria.
    NOTE:  The Luminex NxTAG Respiratory Pathogen Panel (RPP)
    + CoV Assay has been approved by Health Canada For
    Emergency Use Access (EUA) and has been verified by the
University Health Network/Sinai Health Microbiology
    Laboratory.
```

```
* Outbreak No.                                    N/A
* Specimen Source:                                BAL
```

2. **Positive report:**

```
* SARS-CoV-2:                                     DETECTED
* Adenovirus:                                     Not detected
* Coronavirus 229E                                Not detected
* Coronavirus HKU1:                               Not detected
* Coronavirus NL63:                               Not detected
* Coronavirus OC43:                               Not detected
* Human Bocavirus:                                Not detected
* Human Metapneumovirus:                          Not detected
* Influenza virus A:                              Not detected
* Influenza virus A - subtype H1:                 Not detected
* Influenza virus A - subtype H3:                 Not detected
* Influenza virus B:                              Not detected
* Parainfluenza 1:                                Not detected
* Parainfluenza 2:                                Not detected
* Parainfluenza 3:                                Not detected
* Parainfluenza 4:                                Not detected
* Respiratory Syncytial Virus A:                  Not detected
* Respiratory Syncytial Virus B:                  Not detected
* Rhinovirus/Enterovirus:                         DETECTED
* Chlamydophila pneumoniae:                       Not detected
* Mycoplasma pneumoniae:                          Not detected
* Method:                                         *NOTE*
  Testing performed using the Luminex NxTAG Respiratory
   Pathogen Panel (RPP) + CoV Assay. This assay detects
   SARS-CoV-2 as well as 18 non-SARS-CoV-2 respiratory
   viruses and 2 respiratory bacteria.
   NOTE:  The Luminex NxTAG Respiratory Pathogen Panel (RPP)+ CoV Assay has
  been approved by Health Canada For
   Emergency Use Access (EUA) and has been verified by the
   University Health Network/Sinai Health Microbiology
   Laboratory.
* Outbreak No.                                    N/A
* Specimen Source:                                Nasopharyngeal
```

For Influenza A: report subtype H3 if detected.

If subtypes H1 or H1N1 detected, send to PHOL for confirmation.

Please document any repeat testing result for Luminex RPP/COVID panel in workup.

- Include initial calculated signal, reason for repeat
- Record what the repeat result is.

If there is high positivity rate of certain virus/es e.g. Human Metapneumovirus, Enterovirus/ Rhinovirus in the run talk to a Senior Technologist or Microbiologist on call.

Weekly Maintenance Procedures

1. **Cleaning the Sample Probe**

   To clean the sample probe:

   1. Execute STOP if a plate is running. Refer to the software manual for instructions.
   2. Turn off the MAGPIX and unplug the power cord.
   3. Remove the sample probe.
      a. Open the side access door of the MAGPIX (use key taped to top of MagPix).
      b. Unscrew the probe fitting on top of the probe completely.
      c. Grasp the probe gently and push up.
      d. Lift the probe out of the top of the probe holder.

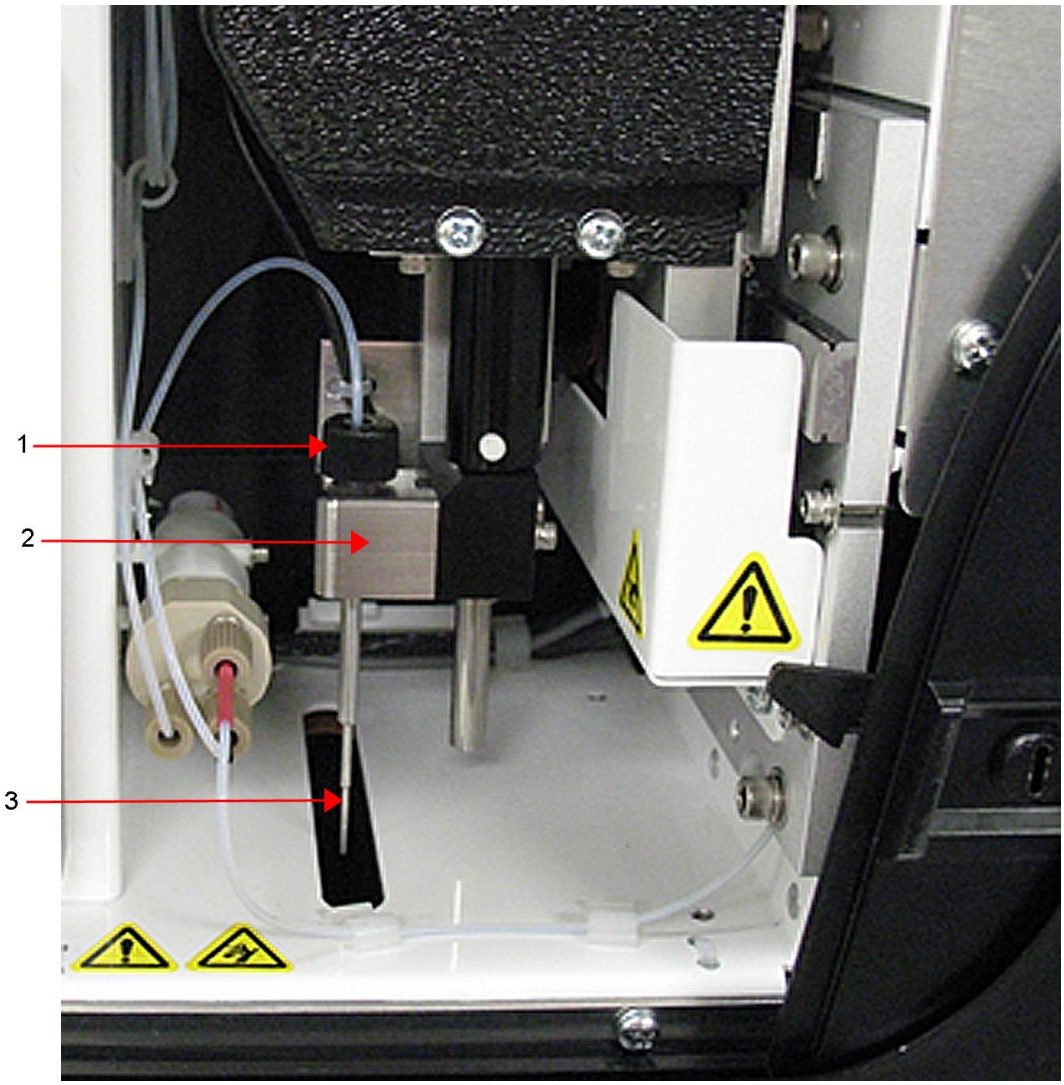


Diagram: 1. Probe fitting - Unscrew and remove

2. Probe holder

3. Probe - Push up gently and lift out of holder

4. Clean the sample probe using by immersion in Ultrasonic Cleaner for a few hours. Change the water after each use.

5. Replace the sample probe and tightly screw in the probe fitting until the fitting clicks.

6. Use the software to perform an automatic probe height adjustment.

NOTE: Perform an automatic probe height adjustment any time the probe is reinstalled after removal.

2. Performing a Visual Inspection

Inspect the MAGPIX® weekly. Make sure the instrument is idle, so there are no moving parts. Open the MAGPIX side access door and fluid compartment door to visually inspect for leaks, corrosion, and other signs of improper function. Check all visible tubing connections.

Load the strip in the space adjacent to the alcohol reservoir.

Click Retract to close door.

Press Run.

Entering New Lot #'s for Verification and Calibration Kits

1. Log into Xponent software.
2. Click Maintenance tab from the top bar.
3. Click Lot Management from side bar.
4. Insert Disc that comes with the Verification and Calibration Kits.
5. Close the Autoplay box that pops up.
6. Click Import button at bottom of screen.
7. Choose the ".lxl" file that is on the disc.
8. Choose Open.
9. Click OK

Monthly Maintenance Procedures

Cleaning the Exterior Surfaces

To clean exterior surfaces:

1. Turn off the MAGPIX® and unplug the power cord.

2. Clean all exterior surfaces with a mild detergent, then by a household bleach solution diluted to 10% to 20%, then by distilled water.

3. Open the side access door of the instrument.

4. Clean all accessible surfaces with a mild detergent, then by a household bleach solution diluted to 10% to 20%, then by distilled water.

5. Dry any unpainted metal surfaces to prevent corrosion.

6. Plug in the power cord and turn on the MAGPIX.

Semi-Annual Maintenance Procedures

Maintaining Air Filters

The MAGPIX® has two air filters, one on the bottom of the instrument and one on the back of the instrument. Every six months, remove these air filters, clean them, and reinstall them.

To clean the MAGPIX air filters:

1. Turn off the MAGPIX® and unplug the power cord.
2. Slide the back filter up out of its holder.
3. Lift or tilt forward the MAGPIX to slide the bottom filter out of its holder toward the front of the instrument.

CAUTION: Before removing the bottom air filter, remove both the waste fluid and Drive Fluid containers, the off-plate reagent block, and any microtiter plates in the instrument.

1. Clean the filters with a vacuum or with distilled water. Stand the filters upright to air dry.

CAUTION: Filters must be completely dry prior to reinstallation.

2. Locate the small incised arrow on the frame of the filter. This indicates air flow. The filter must be installed with the arrow pointing inward.

WARNING: Avoid contact with the tubing and electronic parts of the instrument.

FAILED VERIFICATION OR CLOG (TROUBLESHOOTING GUIDE)

Please perform a self-test (Maintenance -> Cmds & Routines -> click “Self Test” on lower left) and follow the steps below:

1. Run a Clean command with 0.1 N NaOH, then after the sample is taken in the chamber press the stop button under the progress bar.

2. Start the Clean command again, allow the sample to be taken in the chamber and press the soft power switch on the front of the instrument. This shuts down the instrument only (with 200 µL of NaOH in the chamber).
3. Let the chamber soak for 20 minutes.
4. Press the start button to turn on the instrument and allow connection to PC.
5. Run the Daily Fluidics Prep routine
6. Run Sanitize command with 10% bleach 3 times
7. Run 70% Alcohol Flush 2 times
8. Run Wash command with DI water 2 times.
9. Perform calibration and verification.

Troubleshooting

Software Result and Messages	Problem	Possible Cause(s)	Recommendation(s)
Result: Invalid *Message:* "Internal Control failed."	MS2 specific signal is below the positive call cutoff and none of the targets have a positive signal.	Extraction failure, or no MS2 was spiked into that sample. 1. Insufficient sample was added during setup. 2. Failed to fully resuspend Lyophilized Bead Reagents.	Re-extract the sample, making sure that MS2 is spiked into the sample. 1. Ensure the correct sample volume was added. 2. Ensure the Lyophilized Bead Reagents were fully resuspended.
Result: Invalid *Message:* "<Target Name>: non-specific signal detected in control sample"	An unexpected target was detected in a control sample.	Contamination may have occurred during extraction, with extraction reagents, or during sample addition.	Re-extract the samples, including the negative extraction control with new (un-used) reagents.
Result: Invalid *Message*:	More than 7 positive signals were detected	Contamination may have occurred during extraction, with	Re-extract the samples, including negative extraction control with

"Inconclusive results based on abnormal number of positive signals"	in a sample.	extraction reagents, or during sample addition.	new (un-used) reagents
Result: Invalid *Message:* "Run failed. All negative control samples have failed"	An instrument error occurred and all samples identified as negative controls are invalid.	Refer to *xPONENT®* *Software User Manual* for possible causes.	Re-run the sample.
Result: Invalid *Message:* "<Target Name>: invalid value encountered" OR "<Target Name>: low bead count"	The probe failed to acquire enough of the sample.	Low sample volume; probe height adjustment was not completed successfully. 1. Failed to fully resuspend Lyophilized Bead Reagents.	Repeat probe height adjustment procedure. Re-run the sample 1. Ensure the Lyophilized Bead Reagents were fully resuspended.
Result: Invalid *Message*: "<Target Name>: invalid negative control value"	Failed to acquire enough of target signal within all negative control samples	Probe height was not completed successfully; failed to fully-resuspend Lyophilized Bead Reagents.	Re-extract and re-run samples since you cannot rule out contamination for this target

Re-Test Recommendations After Data Acquisition.

FAILED VERIFICATION OR CLOG (TROUBLESHOOTING GUIDE)

Please perform a self-test (Maintenance -> Cmds & Routines -> click “Self Test” on lower left) and follow the steps below:

1. Run a Clean command with 0.1 N NaOH, then after the sample is taken in the chamber press the stop button under the progress bar.
2. Start the Clean command again, allow the sample to be taken in the chamber and press the soft power switch on the front of the instrument. This shuts down the instrument only (with 200 uL of NaOH in the chamber).
3. Let the chamber soak for 20 minutes.
4. Press the start button to turn on the instrument and allow connection to PC.
5. Run the Daily Fluidics Prep routine
6. Run Sanitize command with 10% bleach 3 times
7. Run 70% Alcohol Flush 2 times
8. Run Wash command with DI water 2 times.
9. Perform calibration and verification.

Luminex Technical Support

- Technical Support Contact Information
- Phone: 1 512-381-4397
- Toll Free: 1-877-785-2323
- International: +800-2939-4959
- Fax: 512-219-5114
- Email: support@luminexcorp.com

References

- Luminex Respiratory Pathogen Panel + SARSCoV2 Package Insert
- RUO NxTAG™
- xPONENT® 4.2 for MAGPIX® Software User Manual
- MAGPIX® Hardware Installation and User Manual *Installation and User Manual* | RUO MAGPIX® Hardware

Record of Edited Revisions

Manual Section Name: Standard Operation Procedure Template

Page Number / Item	Date of Revision	Edited by:
New	Dec 30, 2020	Dorna Zareianjahromi
Updated interpretation chart – a) boca verified and b) low pos ->repeat neg report as neg	Feb 18, 2021	Dorna Zareianjahromi
Added "COVID can be reported based on either M gene or orf1ab gene >100 bead count." To interpretation	May 14, 2021	Wayne Chiu
No longer need to confirm Flu A H3 from PHOL if we get the typing from the Magpix.	Jan 10, 2023	Wayne Chiu
Deleted MGI extraction procedure Minor formatting	Mar 30, 2023	Qin Liu
Removed Interpretation Chart 2 (all samples to be hinterpreted the same way)	February 14, 2024	Vanessa Allen