Agilent BioTek Synergy HTX

Multi-Mode Microplate Reader

INSTRUCTIONS FOR USE

ERRATA NOTICE: This document contains references to BioTek. Please note that BioTek is now Agilent. For more information, go to www.agilent.com/lifesciences/biotek

Document part number 1341004I
Revision D
Agilent Technologies, Inc.
August 2022

For In Vitro Diagnostic Use

Contents

Preface

Copyright

Contact Information

Agilent Technologies, Inc.
5301 Stevens Creek Blvd.
Santa Clara, CA 95051

Worldwide Sales and Support

www.agilent.com/en/contact-us/page

Technical Support and Service

www.agilent.com/en/support

Email: bio.tac@agilent.com

Instrument service and repair is available worldwide at one of our international service centers and in the field at your location.

Customer Care

Email: bio.CustomerCare@agilent.com

European Coordination Center

EC REP

Agilent Technologies Denmark ApS
Produktionsvej 42, 2600 Glostrup, Denmark

UK Responsible Person (UKRP)

Agilent LDA UK Ltd.
5500 Lakeside
Cheadle Royal Business Park
Cheadle, Cheshire SK8 3GR

Intended Use Statement

The Synergy HTX is a multi-mode microplate reader and intended to be used for the examination of clinical specimens to analyze their characteristics in relation to a variety of analytes including in human serum and cells.

Incident Reporting

Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority, or appropriate regulatory body, in the country or region in which the user is established.

Quality Control

It is considered good laboratory practice to run laboratory samples according to instructions and specific recommendations included in the assay package insert for the test to be conducted. Failure to conduct Quality Control checks could result in erroneous test data.

Safety Notices

Pay special attention to the following safety notices in all product documentation.

WARNING A WARNING notice denotes a hazard. It calls attention to an operating procedure, practice, or the like that, if not correctly performed or adhered to, could result in personal injury or death. Do not proceed beyond a WARNING notice until the indicated conditions are fully understood and met.

CAUTION A CAUTION notice denotes a hazard. It calls attention to an operating procedure, practice, or the like that, if not correctly performed or adhered to, could result in damage to the product or loss of important data. Do not proceed beyond a CAUTION notice until the indicated conditions are fully understood and met.

Warnings and Precautions

Electrical Hazards

WARNING **Internal Voltage.** Always turn off the power switch and unplug the power supply before cleaning the outer surface of the instrument.

WARNING **Power Rating.** The instrument's power supply or power cord must be connected to a power receptacle that provides voltage and current within the specified rating for the system. Use of an incompatible power receptacle may produce electrical shock and fire hazards.

WARNING **Electrical Grounding.** Never use a plug adapter to connect primary power to the external power supply. Use of an adapter disconnects the utility ground, creating a severe shock hazard. Always connect the power cord directly to an appropriate receptacle with a functional ground.

WARNING **Service.** Only qualified technical personnel should perform service procedures on internal components.

CAUTION **Power Supply.** Use only the power supply shipped with the instrument and operate it within the range of line voltages listed on it.

Chemical/Environmental

WARNING **Potential Biohazards.** Some assays or specimens may pose a biohazard. Adequate safety precautions should be taken as outlined in the assay's package insert. Always wear safety glasses and appropriate protective equipment, such as chemical-resistant rubber gloves and apron.

WARNING **Liquids.** Avoid spilling liquids on the instrument; fluid seepage into internal components creates a potential for shock hazard or instrument damage. If a spill occurs while a program is running, stop the program and turn off the instrument. Wipe up all spills immediately. Do not operate the instrument if internal components have been exposed to fluid.

CAUTION **Liquids.** Do not immerse the instrument, spray it with liquid, or use a dripping-wet cloth on it. Do not allow water or other cleaning solution to run into the interior of the instrument. If this happens, contact Technical Support.

CAUTION **Environmental Conditions.** Do not expose the instrument to temperature extremes. For proper operation, temperature near the instrument should remain within the range in the ***Specifications*** section of this document. Performance may be adversely affected if temperatures fluctuate above or below this range.

CAUTION **Sodium Hypochlorite.** Do not expose any part of the instrument to the recommended diluted sodium hypochlorite solution for more than 20 minutes. Prolonged contact may damage the instrument surfaces. Be certain to rinse and thoroughly wipe all surfaces.

CAUTION **Lubricants.** Do not apply lubricants to moving parts. Lubricant on components in the carrier compartment will attract dust and other particles, which may cause the instrument to produce an error.

CAUTION **DMSO Concentration.** Dimethyl sulfoxide (DMSO) vapor can coat optical surfaces, which can trigger instrument self-test errors. Using DMSO assay concentrations of 2% or below is recommended. Limit long exposure in kinetic assays or incubated assays when possible. Agilent recommends increasing the frequency of Preventive Maintenance visits by a certified service technician to every six months and minimally every year when running assays with DMSO, especially if the concentration is higher than 2%.

CAUTION Agilent BioTek instruments are designed for use in standard benchtop conditions. Operation in corrosive, caustic, or abrasive surroundings, like anaerobic chambers, can negatively affect performance and require increased service frequency, i.e., higher frequency of service than is covered by the instrument warranty.

Components

WARNING

Pinch Hazard. Some areas of the external dispense module can present pinch hazards when the instrument is operating. Keep hands and fingers clear of these areas when the instrument is operating.

WARNING

Hot Surface. The fluorescence lamp assembly is hot when the instrument is turned on. Turn off the reader and allow the bulb to cool for at least 15 minutes before attempting to replace it.

WARNING **Accessories.** Only accessories that meet the manufacturer's specifications shall be used with the instrument.

CAUTION **Fluorescence lamp assembly.** Do not touch the glass lenses. Fingerprints on the condenser lens or heat absorber may negatively affect performance.

CAUTION **Shipping Hardware.** All shipping hardware must be removed before operating the instrument and reinstalled before repackaging the instrument for shipment.

CAUTION **Filters.** The Synergy HTX is shipped with a set of excitation and emission filters installed. The reader's onboard software is preconfigured with the filter values and their locations.

If you change the contents of a filter wheel, you must update Gen5's filter table and then download the information to the reader. The Synergy HTX does not automatically detect which filters are installed.

When changing, cleaning, or replacing filters, it is critical that the filters be placed in the filter wheel in the correct orientation.

When removing or replacing a filter or C-clip filter retainer, do not use a sharp instrument! Use several layers of lens paper and your finger to remove and replace filters and clips. Do not touch the filters with your bare fingers.

CAUTION **Spare Parts.** Only approved spare parts should be used for maintenance. The use of unapproved spare parts and accessories may result in a loss of warranty and potentially impair instrument performance or cause damage to the instrument.

CAUTION **Service.** Only qualified technical personnel should perform service procedures on internal components.

Intended Product Use

WARNING **Software Quality Control.** The operator must follow the manufacturer's assay package insert when modifying software parameters and establishing reading methods. It is considered good laboratory practice to run laboratory samples according to instructions and specific recommendations included in the assay package insert for the test to be conducted. Failure to conduct quality control checks could result in erroneous test data.

WARNING **Data Reduction.** No limits are applied to the raw measurement data. Data exported via computer control must be analyzed by the operator. The performance characteristics of the data reduction software have not been established with any laboratory diagnostic assay. Users must evaluate this instrument and PC-based software in conjunction with their specific assay(s). This evaluation must include the confirmation that performance characteristics for the specific assay(s) are met.

WARNING **Unspecified Use.** Failure to operate equipment according to the guidelines and safeguards specified in the product user documentation could result in a hazardous condition.

CAUTION Use of labware other than described in this document can result in positioning errors during program execution.

Symbols

	Caution
	Caution
	Warning; Biological hazard
	Warning; Pinch hazard
	Warning; Hot surface
	Disposal Notice: Dispose of the instrument according to Directive 2012/19/EU, "on waste electrical and electronic equipment (WEEE)" or local ordinances
	Consult instructions for use or consult electronic instructions for use

IVD	*In vitro* diagnostic medical device
CE	CE Marking — indicates compliance with the requirements of the In Vitro Diagnostic Regulation (2017/746)
EC REP	Authorized representative in the European Community/European Union
	Manufacturer
	Date of manufacture
REF	Catalogue number
SN	Serial number
TÜV SÜD NRTL C US	TÜV SÜD Certification Mark – Type tested; production monitored
IP20	Ingress Protection - Product protected against solid objects up to 12 millimeters. Not protected from liquids.
40	This product complies with environmental protection use period as defined in People's Republic of China Electronic Industry Standard SJ/T11364-2006. Toxic or hazardous substances will not leak or mutate under normal operating conditions for 40 years.
UK CA	UK Conformity Assessed marking is a certification mark that indicates conformity with the applicable requirements for products sold within Great Britain.
	Temperature limit
	Humidity limitation
UDI	Unique device identifier
#	Model number
	Importer

Conformance to Standards

The Synergy HTX meets the requirements of the following standards:

2014/35/EU – Low Voltage Directive

2014/30/EU – EMC Directive

2017/746 – In Vitro Diagnostic Regulation

2011/65/EU (with exemptions) and (EU) 2015/863 – RoHS Directives

2012/19/EU – WEEE Directive as amended by (EU) 2018/849

2006/42/EC of the European Parliament and of the Council of 17 May 2006 on machinery

Standard	Description
IEC QC 080000	IEC Quality Assessment System for Electronic Components (IECQ System) - Hazardous Substance Process Management (HSPM) System Requirements
UL 61010-1	UL Standard for Safety Electrical Equipment For Measurement, Control, and Laboratory Use; Part 1: General Requirements
EN 61010-1	Safety Requirements for Electrical Equipment For Measurement, Control, and Laboratory Use – Part 1: General Requirements
EN 61010-2-010	Safety Requirements for Electrical Equipment For Measurement, Control, and Laboratory Use – Part 2-010: Particular requirements for laboratory equipment for the heating of materials
EN 61010-2-081	Safety Requirements for Electrical Equipment For Measurement, Control, and Laboratory Use – Part 2-081: Particular requirements for automatic and semi-automatic laboratory equipment for analysis and other purposes
EN 61010-2-101	Safety Requirements for Electrical Equipment For Measurement, Control, and Laboratory Use – Part 2-101: Particular requirements for in vitro diagnostic (IVD) medical equipment
CAN/CSA C22.2 No. 61010-1	Safety Requirements for Electrical Equipment For Measurement, Control, and Laboratory Use – Part 1: General Requirements
CAN/CSA C22.2 No. 61010-2-010	Safety Requirements for Electrical Equipment For Measurement, Control, and Laboratory Use – Part 2-010: Particular requirements for laboratory equipment for the heating of materials

CAN/CSA C22.2 No. 61010-2-081	Safety Requirements for Electrical Equipment For Measurement, Control, and Laboratory Use – Part 2-081: Particular requirements for automatic and semi-automatic laboratory equipment for analysis and other purposes
CAN/CSA C22.2 No. 61010-2-101	Safety Requirements for Electrical Equipment For Measurement, Control, and Laboratory Use – Part 2-101: Particular requirements for in vitro diagnostic (IVD) medical equipment

EMC Information and Technical Description

The Synergy HTX conforms to:

Emissions:

EN55011/CISPR 11, Class A
CFR Title 47 FCC Part 15 Subpart B, Class A
ICES-001, Issue 5, Class A (CAN ICES-001(A)/NMB-001(A))
ACMA AS/NZS CISPR 11, Class A

Immunity:

EN/IEC 61326-1 and 61326-2-6
ELECTRICAL EQUIPMENT FOR MEASUREMENT, CONTROL AND LABORATORY USE
PART 2-6: PARTICULAR REQUIREMENTS FOR (IVD) MEDICAL EQUIPMENT

Ingress Protection Code

IP 20. Protected against solid foreign objects of 12.5 mm diameter and greater. No protection against water.

Disposal

Dispose of the instrument according to Directive 2012/19/EU, “on waste electrical and electronic equipment (WEEE)” or local ordinances.

Installation

Important Information

CAUTION **Shipping Hardware.** All shipping hardware must be removed before operating the instrument and reinstalled before repackaging the instrument for shipment.

- This chapter contains installation and setup tasks for the Synergy HTX and accessories. Perform the tasks in the order presented.
- Save all packaging materials. Be sure to use packaging materials supplied by the manufacturer when shipping the reader. Using other forms of commercially available packaging, or failing to follow the repackaging instructions, may void your warranty.
- During the unpacking process, inspect the packaging, reader, and accessories for shipping damage. If the reader is damaged, notify the carrier and your Agilent representative. Keep the shipping boxes and the packaging materials for the carrier's inspection.

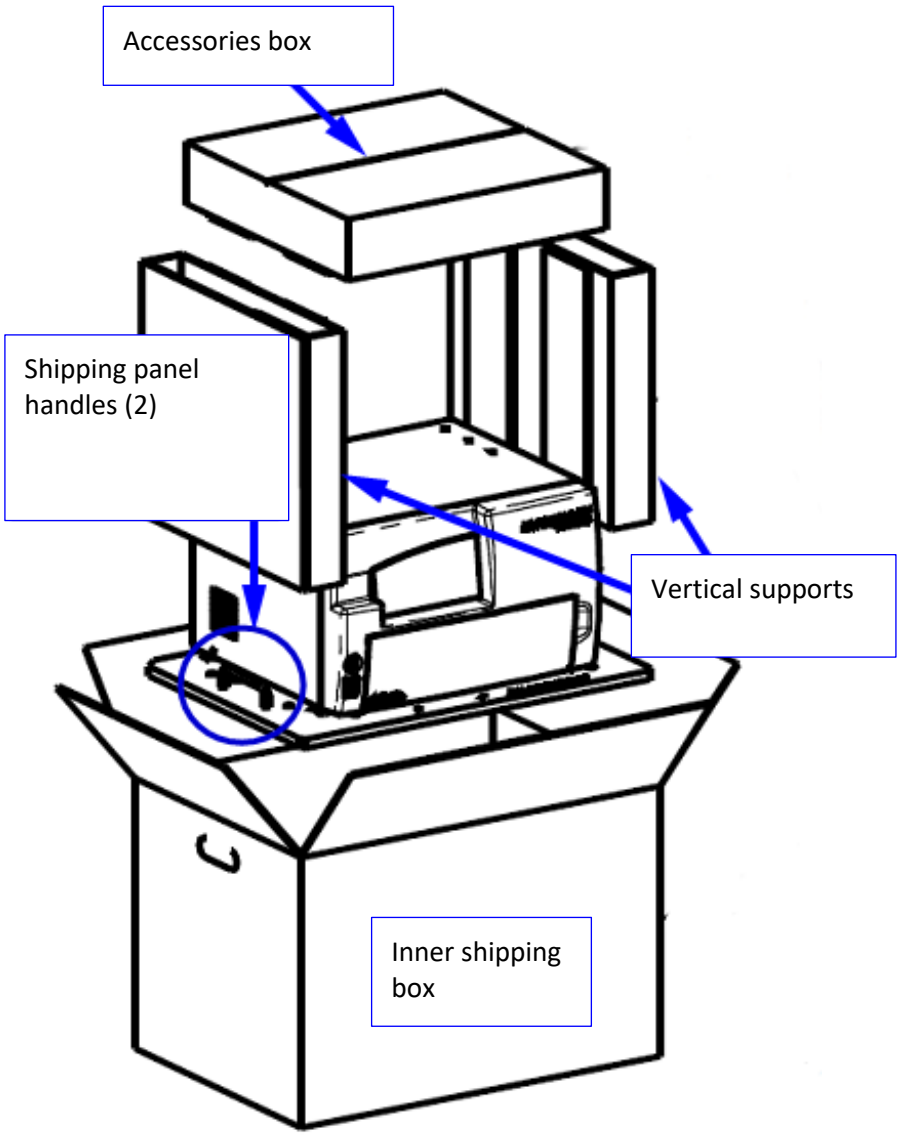
Package Contents

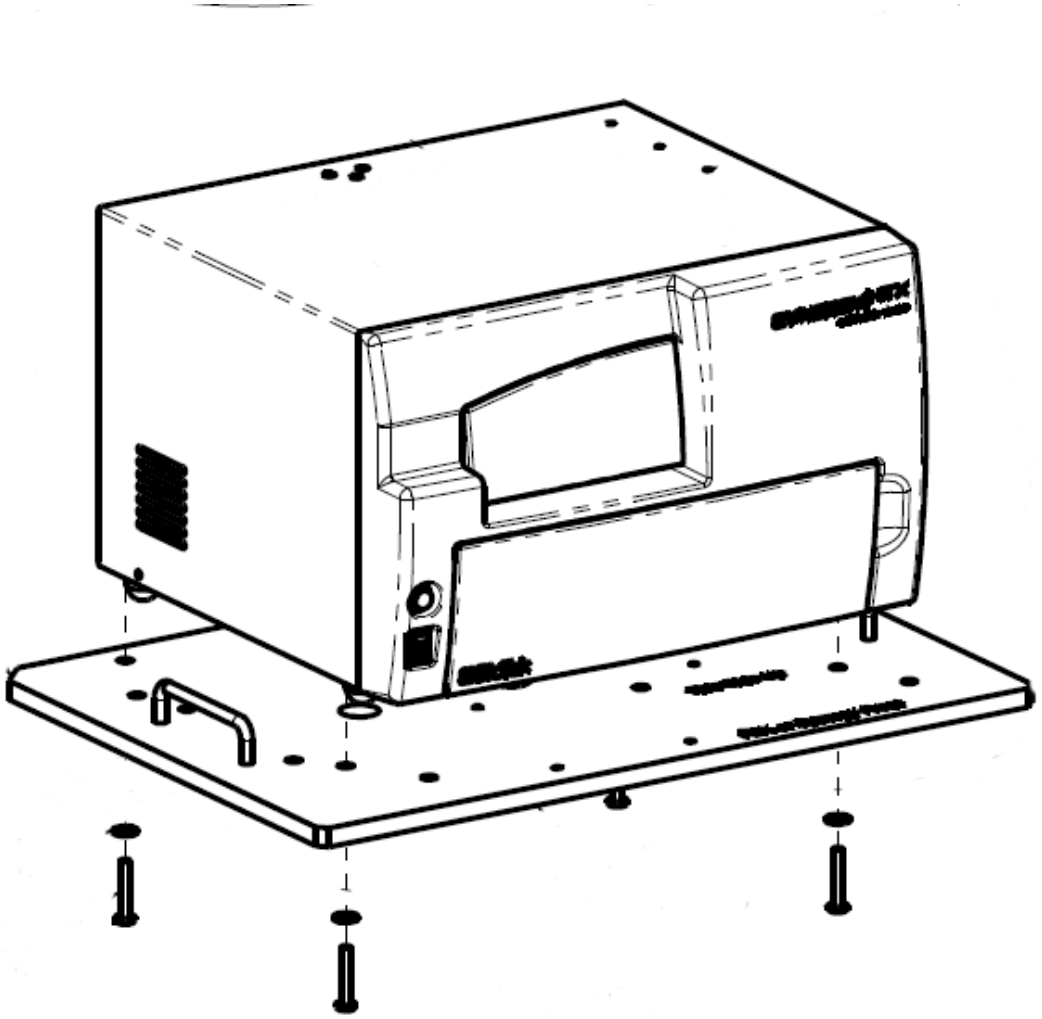
- Synergy HTX instrument model per the sales order
- *Synergy HTX User Manual* on USB flash drive
- Power supply
- Power cord
- USB and serial cables
- Fluorescence lamp assembly
- Time-Resolved Fluorescence cartridge assembly (“T” models)
- Filter plugs
- Wrench
- Optional accessories per the sales order, unless shipped separately

Models

Part Number	Absorbance	Fluorescence	Luminescence	Alpha	Incubation	Shaking	Dispense Ready	TRF (secondary mode)
S1L-SI			√		√	√	√	
S1A-SI	√				√	√	√	
S1LA-SI	√		√		√	√	√	
S1LF-SI		√	√	√	√	√	√	
S1LFA-SI	√	√	√	√	√	√	√	
S1LFTA-SI	√	√	√	√	√	√	√	√

Unpack the Box and Remove the Shipping Panel





Save the packaging and panel in case you need to ship the instrument for service/repair.

Select an Appropriate Location

Install the reader on a level, stable surface in an area where an operating temperature between 18°C and 40°C can be maintained.

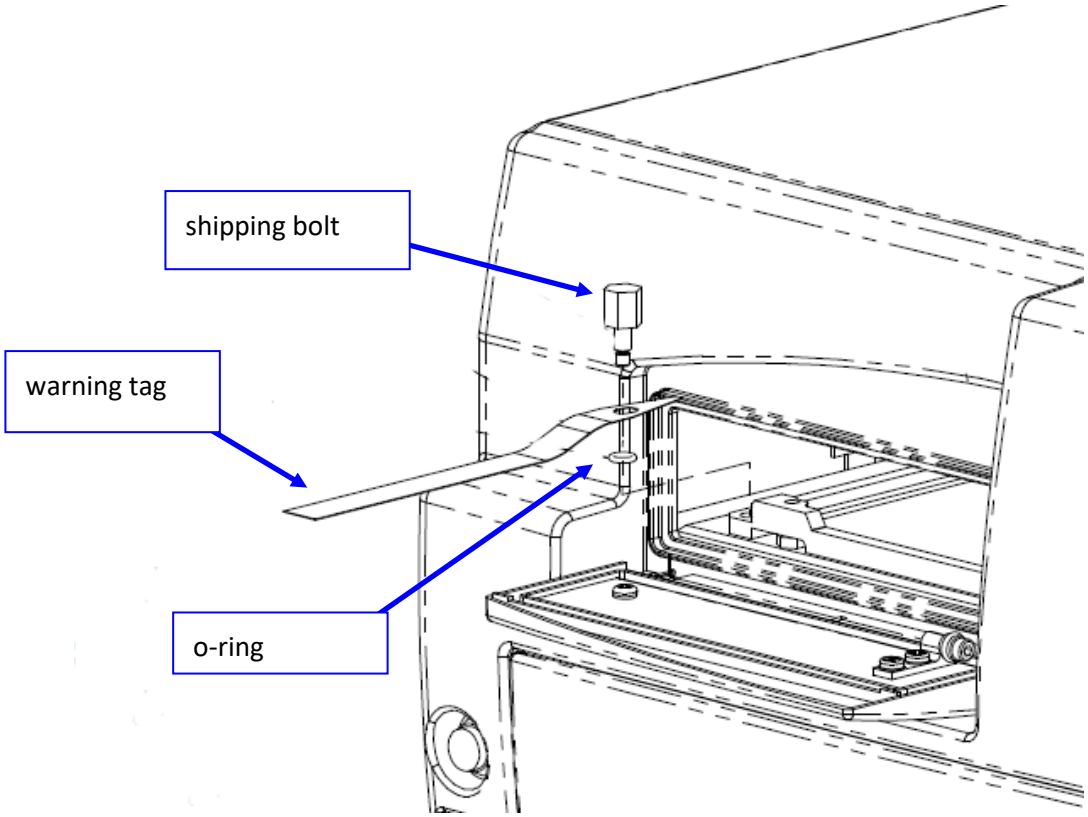
The reader is sensitive to extreme environmental conditions. Avoid:

- Excessive humidity. Condensation directly on the sensitive electronic circuits can cause the instrument to fail internal self-checks. The humidity must be in the range of 10–85%, non-condensing.
- Excessive ambient light. Bright light may affect the reader's optics and readings, reducing its linear range.
- Dust. Readings may be affected by extraneous particles in the microplate wells. A clean work area is necessary to ensure accurate readings.

Remove the Shipping Hardware

CAUTION **Shipping Hardware.** All shipping hardware must be removed before operating the instrument and reinstalled before repackaging the instrument for shipment.

1. Pull down the microplate loading door on the front of the reader.
2. Using the supplied wrench, remove the carrier shipping bolt with its o-ring and warning tag.
3. Store the wrench, bolt, o-ring, and tag in the supplied plastic tool storage bag.



Install the Fluorescence Lamp Assembly

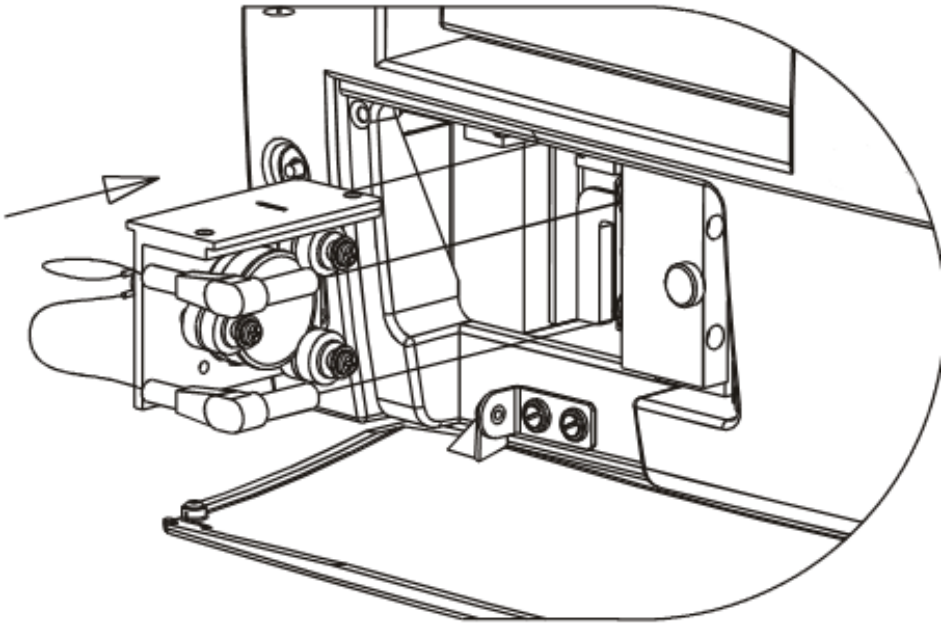
WARNING

Hot Surface. The fluorescence lamp assembly is hot when the instrument is turned on. Turn off the reader and allow the bulb to cool for at least 15 minutes before attempting to replace it.

CAUTION

Fluorescence lamp assembly. Do not touch the glass lenses. Fingerprints on the condenser lens or heat absorber may negatively affect performance.

1. Locate the lamp assembly (replacement part number 7080500) in the accessories box. The lamp is attached to a bracket that also holds a condenser lens and a heat absorber. Two cables are attached to the back of the lamp.
2. Open the reader's hinged door. The lamp compartment is on the left.
3. Orient the lamp assembly as shown below and slide it all the way into the compartment.
4. Plug the lamp cables into the power source located to the right of the lamp. Either cable can be plugged into either socket.



Install the Power Supply

WARNING **Power Rating.** The instrument's power supply or power cord must be connected to a power receptacle that provides voltage and current within the specified rating for the system. Use of an incompatible power receptacle may produce electrical shock and fire hazards.

WARNING **Electrical Grounding.** Never use a plug adapter to connect primary power to the external power supply. Use of an adapter disconnects the utility ground, creating a severe shock hazard. Always connect the power cord directly to an appropriate receptacle with a functional ground.

CAUTION **Power Supply.** Use only the power supply shipped with the instrument and operate it within the range of line voltages listed on it.

1. Plug the power supply's cord into the power inlet on the rear of the reader.
2. Connect the power cord to the power supply.
3. Plug the power cord into an appropriate power receptacle.

Install the Dispense Module (if applicable)

Applies only to models equipped with injectors

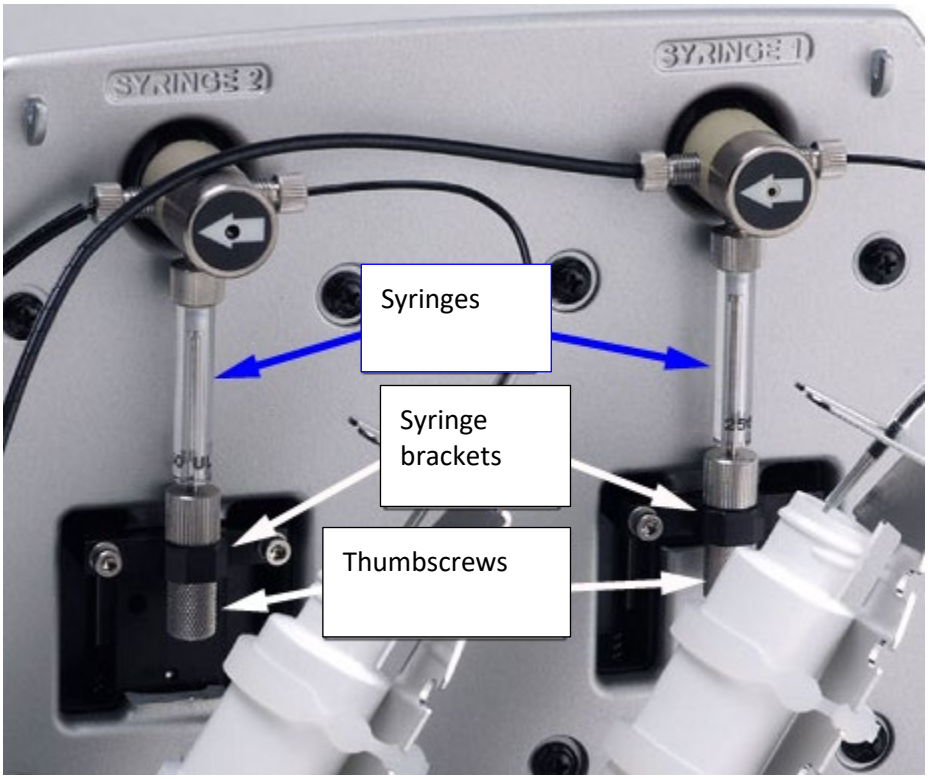
1. Place the dispense module to the left side or on top of the reader.
2. On the rear panel of the reader, identify the SYRINGE 1 and SYRINGE 2 tubing ports. Remove the nylon screws from both ports.
3. Open two of the plastic bags containing the outlet tubes. Remove the clear plastic shrouds from the tubes. Put the other two bags in a safe place; they are spares.
4. Place the nylon screws and the shrouds in the plastic tool storage bag. Use the supplied fastener strips to attach the bag to the rear panel of the dispense module.
5. Remove the two inlet tubes from their canisters.
6. Identify the two syringe valves on the dispense module. Each is labeled with a left-pointing arrow.

When installing the tubes, do not use any tools. Finger-tighten only.

7. Screw the fitting of one inlet tube into the right side of the Syringe 1 valve.
8. Screw one end of one outlet tube into the left side of the Syringe 1 valve.
9. Screw the other end of the outlet tube into the SYRINGE 1 port on the reader.
10. Repeat these steps to attach the inlet and outlet tubing for Syringe 2.
11. Seat the outlet tubes in the clip to the left of the Syringe 2 valve.

Be sure to correctly connect the outlet tubes between the syringe valves and the ports on the instrument's rear panel. Otherwise, injected fluid may miss the intended well.

12. Remove the two syringes from their boxes. They are identical and interchangeable. Each should already be assembled in one piece, but if for some reason there are two separate pieces, assemble them now: insert the white tip of the syringe plunger into the barrel of the syringe and gently push it all the way into the barrel.
13. Install the syringes:
    - Hold the syringe vertically with the threaded end at the top.
    - Screw the top of the syringe into the bottom of the syringe valve. Finger-tighten only.
    - Carefully pull down the bottom of the syringe until it rests inside the hole in the bracket.
    - Pass a thumbscrew up through this hole and thread it into the bottom of the syringe. Hold the syringe to prevent it from rotating while tightening the thumbscrew. Finger-tighten only.



14. Locate the dispense module cable. Plug one end into the port on the left side of the dispense module. Plug the other end into the "Dispenser Port" on the rear of the reader.
15. Locate the injector-tip-cleaning stylus, packaged in a small cylinder. Attach the cylinder to the back of the dispense module for storage.

Connect the Host Computer

The Synergy HTX is equipped with two communication ports, "USB" and "RS232" (serial), located on the back of the reader. Connect one end of the supplied communication cable to the appropriate port on the reader, and the other end to an appropriate port on the host computer.

Install Gen5 Software

The Synergy HTX is controlled by Gen5 software running on a host computer. Follow instructions supplied with Gen5 to install the necessary software.

- Ensure the computer meets the minimum system requirements as described in the *Gen5 Instructions for Use*.
- You must have administrator privileges to install Gen5. Log in to Windows as "Administrator" or consult your IT department for assistance.

Turn on the Reader

1. If Gen5 is open, close it now.
2. Turn on the Synergy HTX. The reader performs a system test. When the test is completed, the reader extends the microplate carrier. If the test fails, contact Technical Support.

Establish Communication

1. Start Gen5 and log in if prompted. The default System Administrator password is **admin**.
2. From the main screen, select **System > Instrument Configuration**.
3. Click **Add Reader** and select **Synergy HTX**. Click **OK**.
4. Perform one of the following steps, as applicable:
   - Select **Plug & Play**. (A reader must be connected to the computer and turned on to appear in the Available Plug & Play Readers list.)
   - Select **Com Port** and select the computer's COM port to which the reader is connected. (If using the USB cable, the information can be found via Windows Control Panel, under Ports in the Hardware/Device Manager area of System Properties.)
5. Click the **Test Comm** button. Gen5 attempts to communicate with the reader. If the communication attempt is successful, return to Gen5's main screen.

   If the communication attempt is not successful, try the following:

   - Is the reader connected to the power supply and turned on?

- Is the communication cable firmly attached to both the reader and the computer?
- Did you select the correct Reader Type in Gen5?
- Try a different COM port.
- If using the USB cable, did you install the driver software?

If you remain unable to get Gen5 and the reader to communicate with each other, contact Technical Support.

Set Dispenser Calibration Values (if applicable)

Applies only to models equipped with injectors

Before using the external dispense module with the Synergy HTX, you must set its calibration values in Gen5.

The calibration values for both dispensers (#1 and #2) are printed on labels affixed to the dispense module. Each label lists six target calibration values (e.g., 200, 80, 40) with their actual measured values (e.g., 199.3, 79.7, 39.9). You will enter the measured calibration values into Gen5.

1. If you have not already done so, turn on the instrument and establish communication with Gen5.
2. In Gen5, go to **System > Instrument Configuration**, select the **Synergy HTX**, and click **View/Modify**.
3. Click **Setup** and select the **Dispenser 1** tab.
4. Press **CTRL+SHIFT+M** to enter maintenance mode for the Dispenser 1 window.
5. Enter the syringe calibration values from the corresponding label on the rear of the dispense module.
6. Click **Send Volumes** and then **Get Volumes** to verify that the entered values were sent to the reader.
7. Select the **Dispenser 2** tab and repeat steps 4–6 for Dispenser 2.

Run a System Test

Running a System Test will confirm that the reader is set up and running properly or will provide an error message if a problem has been detected.

If applicable, adjust Gen5's Absorbance Wavelengths table to values that will confirm operation of the reader at its limits. We recommend 200 and 999 nm (the lower and upper limits of the monochromator), and four wavelengths in between that best represent your assays and/or the lowest and highest values typically used in your lab.

1. Turn on the incubator:

- In Gen5, select **System > Instrument Control > Synergy HTX**.
- Click the **Pre-Heating** tab. Enter a requested temperature of at least 37°C and then click **On**.
- Wait until the incubator reaches the set point before continuing.

2. Select **System > Diagnostics > Run System Test**. Select your reader if prompted and click **OK**.
3. When the test is complete, a dialog requesting additional information appears. Enter the information (if required) and click **OK**.
4. The results report appears. The text should read "SYSTEM TEST PASS." If it shows "SYSTEM TEST FAIL" contact Technical Support.
5. Turn off the incubator.

Test the Injection System

Applies only to models equipped with injectors

1. If necessary, press the carrier eject button to eject the microplate carrier.
2. Place the tip priming trough in its pocket in the carrier.
3. Place the priming plate on the carrier.
4. Fill the two reagent bottles with distilled or deionized water. Place the bottles in their holders and place the holders directly in front of the syringes. Insert the inlet tubes into the bottles.
5. In Gen5, select **System > Instrument Control > Synergy HTX** and click the **Prime** tab.
6. With Dispenser set to 1, set the Volume to 5000 µL and click **Prime**. The syringe should move down and up repeatedly, drawing fluid from the bottle and pumping it through the tubing and into the priming plate. Examine the fittings; no leaks should be detected. If leaks are detected, tighten all fittings and repeat the prime. If leaks are still detected, contact Technical Support.
7. When finished, set the Volume to 2000 µL and click **Purge** to clear the fluid lines.
8. Set the Dispenser to 2 and repeat steps 6 and 7.
9. Remove and empty the priming plate.

Verify Performance

As applicable for your Synergy HTX model, refer to the *Instrument Testing* section for test procedures for:

- Absorbance Tests
- Fluorescence Tests
- Luminescence Test
- Injection System Tests

Repackaging and Shipping Instructions

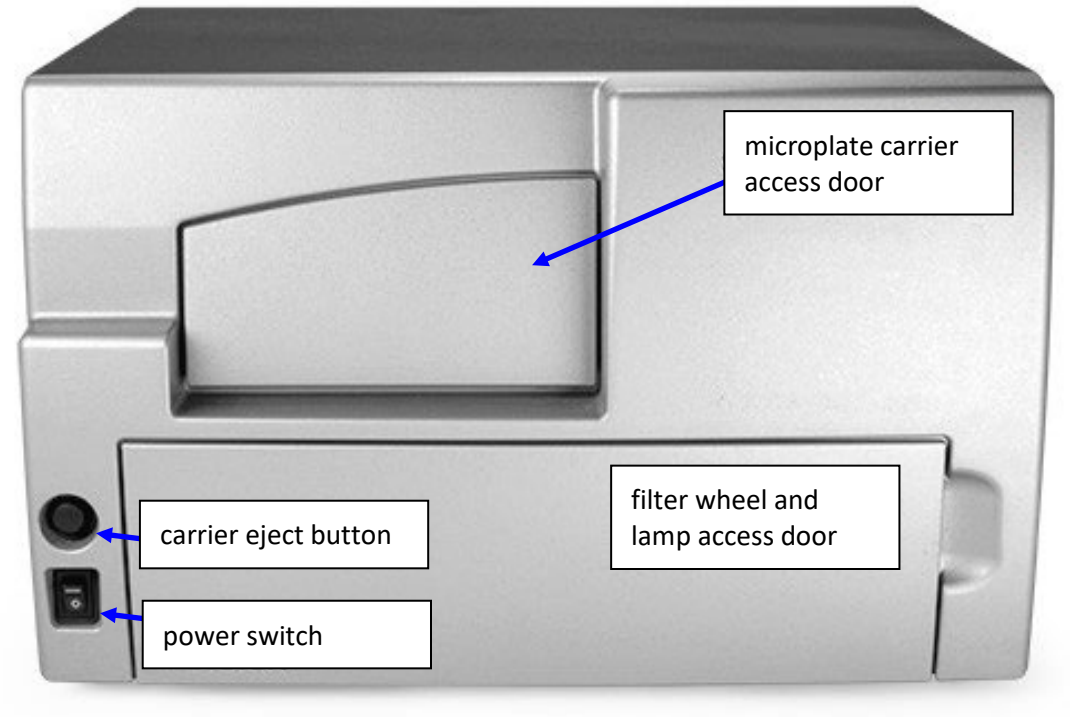
Please read the information provided below before preparing the Synergy HTX for shipment.

- Contact Technical Support before returning equipment for service.
- Decontamination prior to shipment is required by the U.S. Department of Transportation regulations.
- If the reader has been exposed to potentially hazardous material, decontaminate it to minimize the risk to all who come in contact with the instrument during shipping, handling, and servicing. The Maintenance chapter contains decontamination instructions.
- Ensure the microplate carrier is empty. Spilled fluids can contaminate the optics and damage the instrument.
- Install the shipping hardware.
- The instrument's packaging design is subject to change. If the instructions in this document do not apply to the packaging materials you are using, contact Technical Support for guidance.
- Be sure to use packaging materials supplied by the manufacturer. Other forms of commercially available packaging are not recommended and can void the warranty.
- If the packaging materials have been damaged or lost, or if the same set has been used more than four times, order replacement materials.

1. Decontaminate the reader and, if attached, the dispense module. Disconnect the dispense module.
2. If applicable, remove the tip priming trough from the microplate carrier.
3. Retract the microplate carrier. Turn off and unplug the reader.
4. Remove the lamp assembly and pack it in bubble wrap.
5. Replace the microplate carrier shipping bolt.
6. Tip the reader onto its back. Attach the shipping panel to the bottom of the reader using the four screws and washers.
7. Wrap the plastic bag around the reader and shipping panel.
8. Locate the original outer shipping box. Place four foam blocks in the four bottom corners of the box. Place the inner shipping box inside the outer box.
9. Grasp the handles on the shipping panel and carefully lower the reader into the inner shipping box.
10. Slide the foam vertical supports into place around the reader. Place the accessories box on top.
11. Close and seal the inner box with tape.
12. Place four foam corner blocks around the inner shipping box. Close and seal the outer box with tape.

Getting Started

Main Components



- The power switch contains an LED, which is illuminated green when the power is on.
- The carrier eject button can be used to move the microplate carrier into or out of the measurement chamber and also to stop the instrument from “beeping” when it encounters an error.
- The microplate carrier supports microplates and adapter plates as described in the Specifications appendix. The plate is positioned so that well A1 is in the left rear corner of the carrier.

For fluorescence and luminescence reading modes, the height of the top optic probe can be adjusted. Use the Read Height option to define how far the top probe should be offset from the top surface of the plate during the read.

Lamp Assembly and Filter Wheel Access

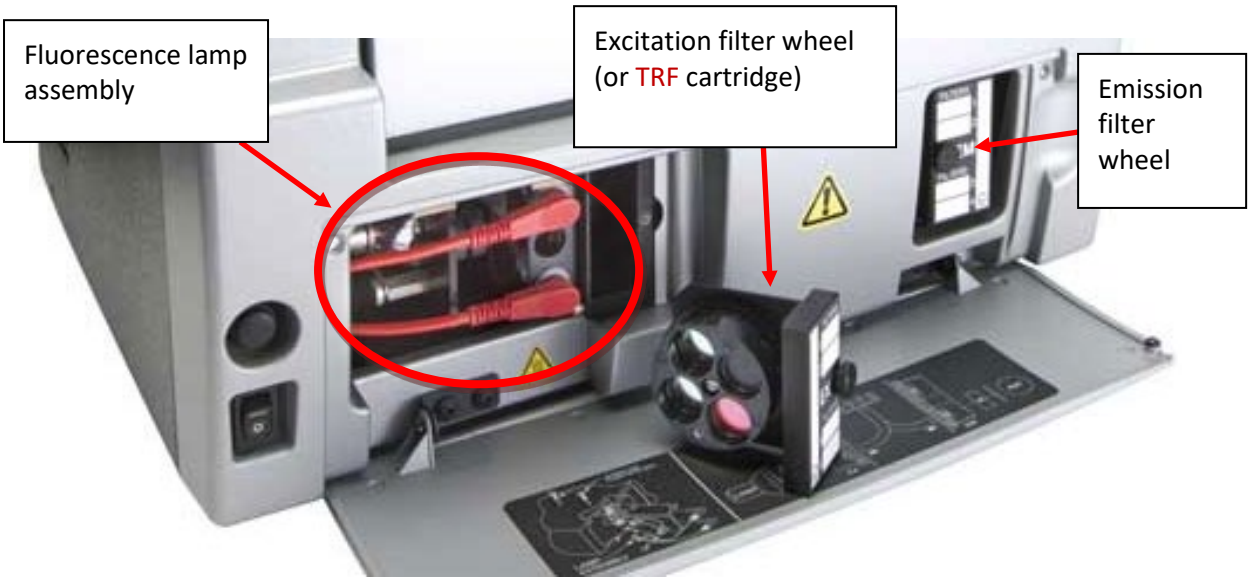
Applies only to models with fluorescence and luminescence capability.

WARNING

Hot Surface. The fluorescence lamp assembly is hot when the instrument is turned on. Turn off the reader and allow the bulb to cool for at least 15 minutes before attempting to replace it.

CAUTION

Fluorescence lamp assembly. Do not touch the glass lenses. Fingerprints on the condenser lens or heat absorber may negatively affect performance.



The Synergy HTX has two lamps:

- **Standard Fluorescence** (shown above). This lamp's part number is 7080500.
- **Absorbance and Time-Resolved Fluorescence.** This lamp is not user replaceable.

Excitation and Emission Filter Wheels

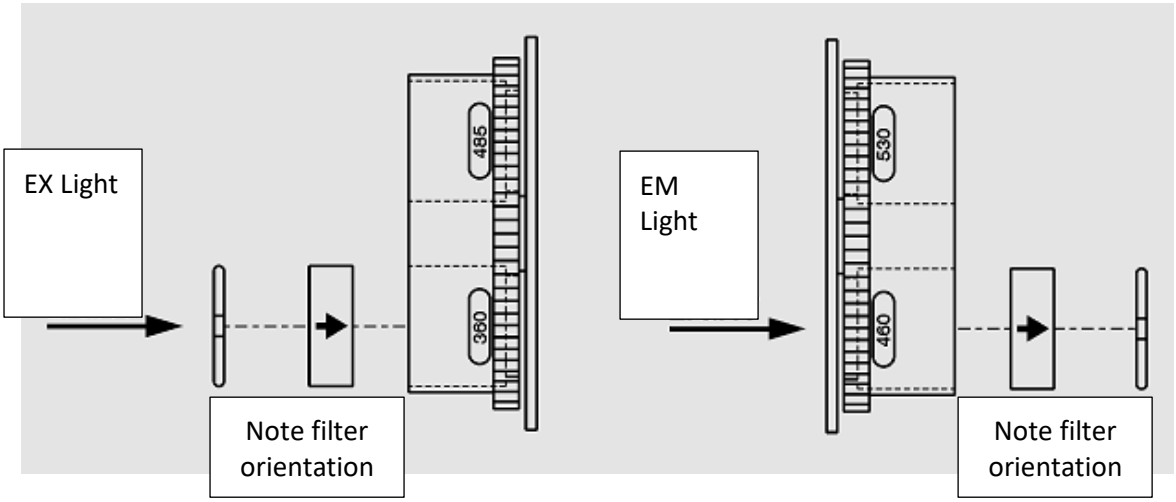
CAUTION

Filters. The Synergy HTX is shipped with a set of excitation and emission filters installed. The reader's onboard software is preconfigured with the filter values and their locations.

If you change the contents of a filter wheel, you must update Gen5's filter table and then download the information to the reader. The Synergy HTX does not automatically detect which filters are installed.

When changing, cleaning, or replacing filters, it is critical that the filters be placed in the filter wheel in the correct orientation.

When removing or replacing a filter or C-clip filter retainer, do not use a sharp instrument! Use several layers of lens paper and your finger to remove and replace filters and clips. Do not touch the filters with your bare fingers.



Synergy HTX models with fluorescence capability are equipped with one excitation filter wheel and one emission filter wheel; readers with luminescence capability use only an emission filter wheel.

The excitation and emission filter wheels are not interchangeable and are labeled as follows: EX = Excitation, EM = Emission. (TR = Time-Resolved Cartridge)

Filter direction within a filter wheel is important. The direction differs depending on the filter wheel. Refer to the diagram on the inside of the front panel door.

Each filter is marked with an arrow indicating the proper direction of light. Refer to the figure on the previous page for proper filter orientation.

Recommendation: Placing filters in the wheels in ascending wavelength order from position 1 to 4 (no holes in EX2 or EM3), particularly if the reader has generated a 4E18 (saturation) error.

Installing the Time-Resolved Cartridge

The cartridge must be installed in place of the excitation filter wheel before a TRF assay can be run. The reader automatically detects the presence of the TR cartridge. At the start of a time-resolved fluorescence assay, Gen5 will prompt you to install the TR cartridge if it is missing.

Configuring the System for Luminescence Measurements

CAUTION **Filters.** If you change the contents of a filter wheel, you must update Gen5's filter table and then download the information to the reader. The Synergy HTX does not automatically detect which filters are installed.

When changing, cleaning, or replacing filters, it is critical that the filters be placed in the filter wheel in the correct orientation.

When removing or replacing a filter or C-clip filter retainer, do not use a sharp instrument! Use several layers of lens paper and your finger to remove and replace filters and clips. Do not touch the filters with your bare fingers.

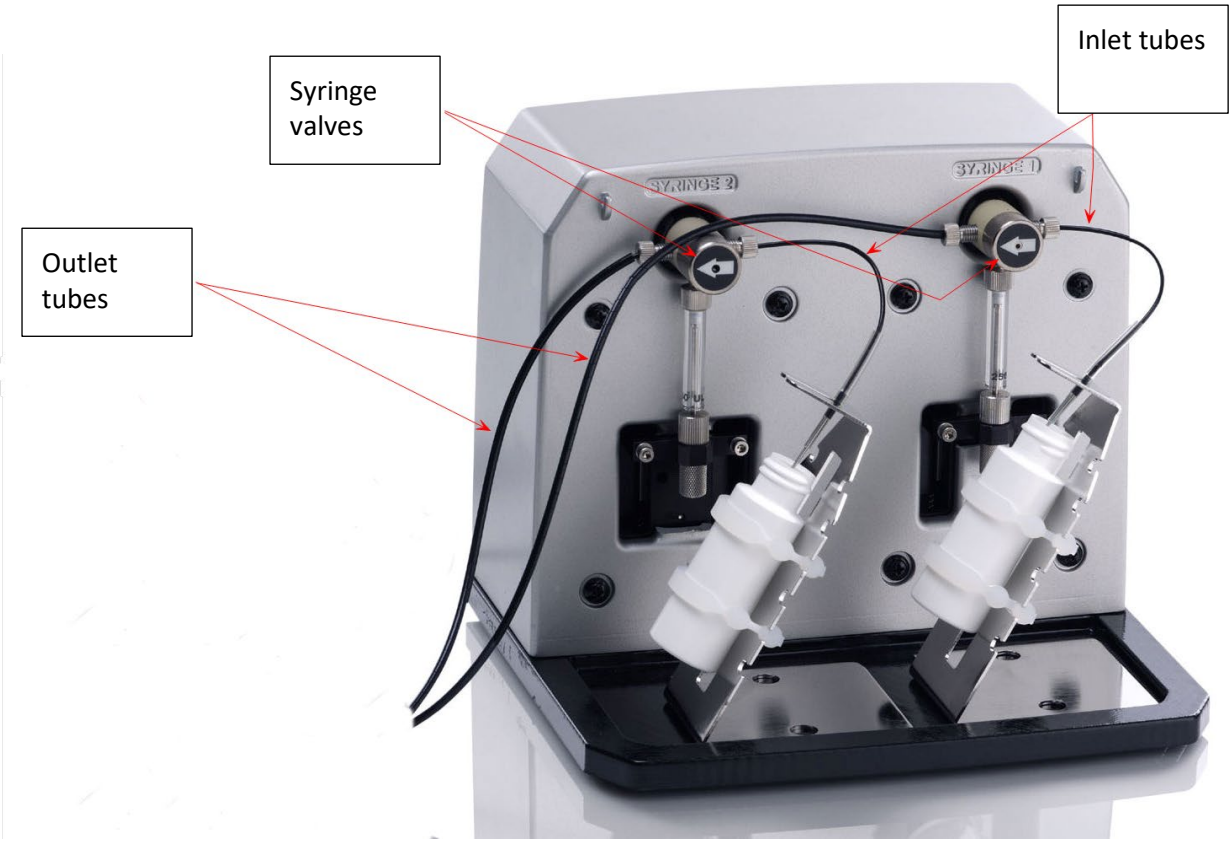
For best results when taking luminescence measurements, the excitation filter wheel should have no empty locations, and it should have at least one "plug."

If your tests require that the light emitted from the samples remain unfiltered, the emission filter wheel should have an empty location in it.

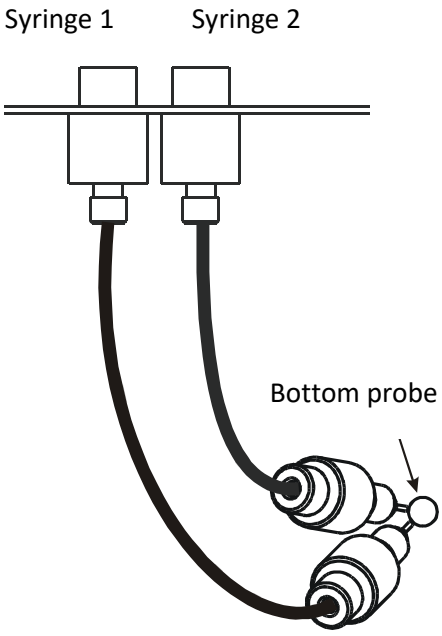
Injection System (if equipped)

External Dispense Module

The dispense module pumps fluid from the supply bottles to injector heads located inside the instrument. Fluid is injected into one well at a time.



Internal Tubing

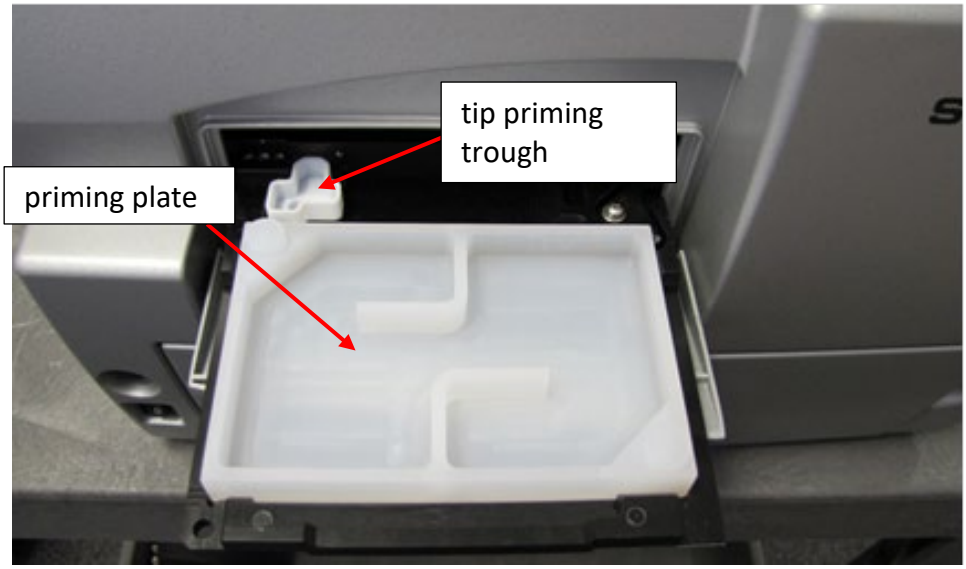


Priming the Injection System

Before running a dispense assay, prime the system. Tip priming can be performed at the start of the assay, and sometimes, just before each dispense to a well.

Both types of primes require a fluid reservoir to be present on the microplate carrier:

- The priming plate is placed on the microplate carrier for a Prime operation (to prime the dispense system with fluid).
- The tip priming trough is a small, removable priming cup located in the left rear of the carrier. The trough holds up to 1.5 mL of liquid and must be periodically emptied and cleaned by the user.



Gen5 Software

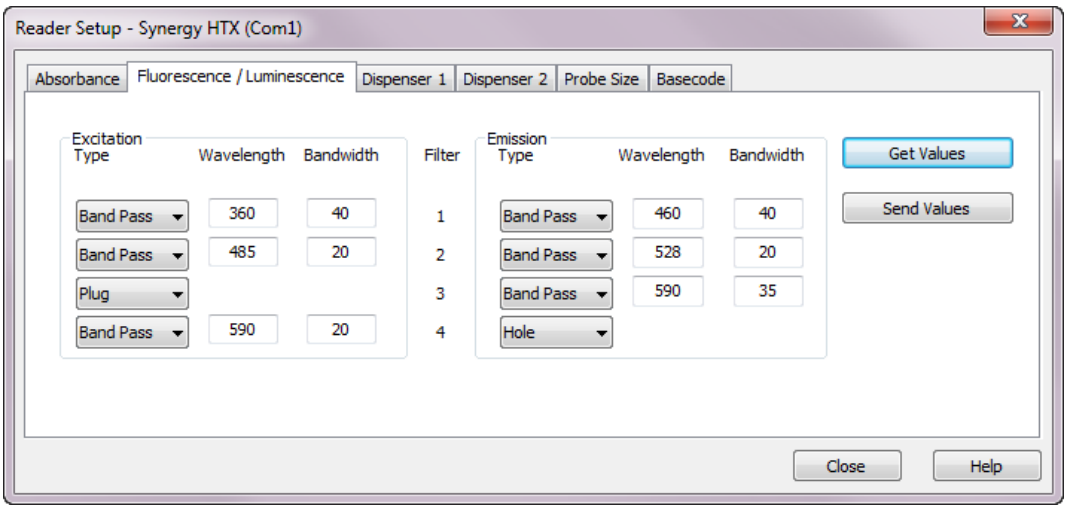
Gen5 supports all Synergy HTX reader models. Use Gen5 to control the reader, the dispense module (if equipped), and BioStack (if equipped); perform data reduction and analysis on the measurement values; print/export results; and more. This section provides brief instructions for working with Gen5 go create protocols and experiments and read plates. Refer to the *Gen5 Instructions for Use* and the Gen5 Help system for more information.

Viewing and Modifying Filter Wheel Information

CAUTION **Filters.** If you change the contents of a filter wheel, you must update Gen5's filter table and then download the information to the reader. The Synergy HTX does not automatically detect which filters are installed.

When changing, cleaning, or replacing filters, it is critical that the filters be placed in the filter wheel in the correct orientation.

To view or modify the information, select **System > Instrument Configuration**, highlight the **Synergy HTX** reader, and click **View/Modify**. Click **Setup** and then click the **Fluorescence/Luminescence** tab.



To change the settings and download them to the instrument:

1. Select **Band Pass**, **Plug**, or **Hole** for the excitation and emission filter wheels.
2. For each filter type, enter the wavelength value and its accompanying bandwidth. The bandwidth is printed on the side of each filter.
3. When finished, click **Send Values** to download the information to the reader. (Clicking **Get Values** uploads information from the reader.)
4. Click **OK** to save the settings and close this dialog. The settings become available for selection in the Read step dialog in a Procedure.

Creating Protocols and Experiments

In Gen5, a protocol contains instructions for controlling the reader and (optionally) for analyzing the data retrieved from the reader. At a minimum, a protocol must specify the procedure for the assay you wish to run. After creating a protocol, create an experiment that references the protocol. You'll run the experiment to read plates and analyze the data.

The instructions below briefly describe how to create a simple protocol in Gen5. See the Gen5 Help system for complete instructions.

1. In the Gen5 Task Manager, select the **Protocols** icon and click **Create New**.
2. Open the Procedure dialog (double-click **Procedure** in the menu tree).
3. Select an appropriate Plate Type.
4. Add steps to shake or heating the plate, dispense fluid, read the plate, and more.
5. Click **Validate** to verify that the attached reader supports the defined steps, and then click **OK**.
6. Optionally, perform the next steps to analyze and report the results:
   - Open the Plate Layout dialog and assign blanks, samples, controls, and/or standards to the plate.
   - Open the Data Reduction dialog to add data reduction steps. Categories include Transformation, Well Analysis, Curve Analysis, and more.
   - Create a report or export template, via one of the Report/Export Builder options.
7. Select **File > Save** and give the protocol an identifying name.

The instructions below briefly describe how to create an experiment and then read a plate in Gen5. See the Gen5 Help system for complete instructions.

1. In the Gen5 Task Manager, select the **Experiments** icon and click **Create using an existing protocol**.
2. Select the desired protocol and click **OK**.
3. Select a plate in the menu tree and select **Plate > Read Plate #** or click the **Read New** icon.
4. When the read is complete, measurement values appear in Gen5.
5. Select **File > Save** and give the experiment an identifying name.

Dispense Module Control

Gen5 is used to perform several dispense functions, such as initialize, dispense, prime, and purge. The Prime and Purge functions are introduced here; refer to the Gen5 Help system for additional information.

Prime

Before running an experiment with a Dispense step, prime the system with the fluid to be used.

1. Place the priming plate on the carrier.
2. Fill the supply bottle with a sufficient volume of the fluid to be used for the prime and the assay. Insert the appropriate inlet tube into the bottle.
3. Select **System > Instrument Control > Synergy HTX** and click the **Prime** tab.
4. Select the Dispenser number (1 or 2) associated with the supply bottle.
5. Enter the Volume to be used for the prime. The minimum recommended prime volume is 2000 µL.
6. Select a prime Rate, in µL/second.
7. Click **Prime** to start the process. When finished, carefully remove the priming plate from the carrier and empty it.

If the priming plate is empty, the prime volume was too low.

Purge

To conserve reagent, Gen5 provides the option to purge fluid from the system back into the supply bottle.

1. Select **System > Instrument Control > Synergy HTX** and click the **Prime** tab.
2. Select the Dispenser number (1 or 2) associated with the supply bottle.
3. Enter the desired purge Volume in µL (e.g., 2000).
4. Select a prime Rate in µL/sec.
5. Click **Purge** to start the process.

Plate Shaking Options

The Synergy HTX supports multiple plate shaking options. Shaking is controlled using Gen5 by adding a Shake step to a protocol's procedure.

Mode	Speed	Amplitude (in 1-mm steps)	Frequency
Linear	---	1 mm to 6 mm	18 Hz to 6 Hz
Orbital	Slow	1 mm to 6 mm	10 Hz to 3 Hz
Orbital	Fast	1 mm to 6 mm	14 Hz to 5 Hz

Maintenance

Warnings and Precautions

WARNING **Internal Voltage.** Always turn off the power switch and unplug the power supply before cleaning the outer surface of the instrument.

WARNING **Liquids.** Avoid spilling liquids on the instrument; fluid seepage into internal components creates a potential for shock hazard or instrument damage. If a spill occurs while a program is running, stop the program and turn off the instrument. Wipe up all spills immediately. Do not operate the instrument if internal components have been exposed to fluid.

CAUTION **Liquids.** Do not immerse the instrument, spray it with liquid, or use a dripping-wet cloth on it. Do not allow water or other cleaning solution to run into the interior of the instrument. If this happens, contact Technical Support.

CAUTION **Lubricants.** Do not apply lubricants to moving parts. Lubricant on components in the carrier compartment will attract dust and other particles, which may cause the instrument to produce an error.

CAUTION **DMSO Concentration.** Dimethyl sulfoxide (DMSO) vapor can coat optical surfaces, which can trigger instrument self-test errors. Using DMSO assay concentrations of 2% or below is recommended. Limit long exposure in kinetic assays or incubated assays when possible. Agilent recommends increasing the frequency of Preventive Maintenance visits by a certified service technician to every six months and minimally every year when running assays with DMSO, especially if the concentration is higher than 2%.

WARNING **Potential Biohazards.** Wear protective gloves when handling contaminated instruments. Gloved hands should be considered contaminated at all times; keep gloved hands away from eyes, mouth, nose, and ears.

Mucous membranes are considered prime entry routes for infectious agents. Wear eye protection and a surgical mask when there is a possibility of aerosol contamination. Intact skin is generally considered an effective barrier against infectious organisms; however, small abrasions and cuts may not always be visible. Wear protective gloves when handling contaminated instruments.

Recommended Maintenance Schedule

Task	Daily	Quarterly	As Needed
All models:			
Clean exposed surfaces			√
Inspect/clean excitation and emission filters (if equipped)		√	
Decontaminate the instrument	*before shipment or storage*		
Models with injectors and an external dispense module:			
Flush/purge the fluid path	√		
(Optional) Run a Dispense protocol			√
Empty/clean tip prime trough	√		
Clean priming plate			√
Clean internal components		√	√
Clean tubing and injector heads		√	√
Clean optical probes		√	√
Clean internal surfaces		√	√

Clean Exposed Surfaces

Exposed surfaces may be cleaned (not decontaminated) with a cloth moistened (not soaked) with water or water and a mild detergent, and then water.

Models with injectors: If the tip priming trough overflows, wipe the carrier and the surface beneath the carrier with a dry cotton cloth. If overflow is significant, you may have to remove the shroud of the instrument to better access the surface beneath the carrier.

Inspect/Clean Excitation and Emission Filters

Agilent recommends inspecting the filters for dust and other debris every three months. To clean them, you will need:

- Isopropyl, ethyl, or methyl alcohol
- 100% pure cotton balls or high-quality lens-cleaning tissue
- Cloth gloves
- Magnifying glass

Do not touch the filters with your bare fingers.

1. Inspect the glass filters for speckled surfaces or a “halo” effect. This may indicate deterioration due to moisture. If you have any concerns, contact Technical Support.

2. Using cotton balls or lens-cleaning tissue moistened with a small amount of high-quality alcohol, clean each filter by lightly stroking its surface in one direction. Ensure that the filters remain in their current locations.
3. Use a magnifying glass to inspect the surface; remove any loose threads left by the cotton ball.

Maintenance for Models with Injectors

Flush/Purge the Fluid Path

At the end of each day that the dispense module is in use, flush the fluid path using the Gen5 priming utility. Leave the fluid to soak overnight or over a weekend, and then purge the fluid before using the instrument again.

This flushing and purging routine is also recommended before disconnecting the outlet tubes from the rear of the reader, and before decontamination to remove any assay residue prior to applying isopropyl alcohol or sodium hypochlorite.

1. Fill two supply bottles with deionized or distilled water. Insert the supply (inlet) tubes into the bottles.
2. Place the priming plate on the carrier.
3. Select **System > Instrument Control > Synergy HTX**.
4. Click the **Prime** tab and select Dispenser 1.
5. Set the Volume to 5000 µL. Keep the default prime rate.
6. Click **Prime** to start the process. When the process is complete, carefully remove the priming plate from the carrier and empty it.
7. Repeat the process for Dispenser 2.

Leave the water in the system overnight or until the instrument will be used again. Purge the fluid from the system (see below) and then prime with the dispense reagent before running an assay.

To purge the fluid from the system:

1. Place the inlet tubes in empty supply bottles or a beaker.
2. Select **System > Instrument Control > Synergy HTX**.
3. Click the **Prime** tab and select Dispenser 1.
4. Set the Volume to 2000 µL.
5. Click **Purge** to start the process.

When the purge is complete, repeat the process for Dispenser 2.

After purging the system, you may wish to run a quick Dispense protocol to visually verify the dispense accuracy.

Empty/Clean the Tip Priming Trough

1. Extend the microplate carrier and carefully remove the tip priming trough from its pocket in the left rear of the carrier.
2. Wash the trough in hot, soapy water. Use a small brush to clean in the corners.
3. Rinse the trough thoroughly and allow it to dry completely.
4. Replace the trough in the microplate carrier.

Clean the Priming Plate

Clean the priming plate regularly to prevent bacteria growth and residue buildup. Wash the plate in hot soapy water, using a small brush to clean in the corners if necessary. Rinse thoroughly and allow it to dry completely.

Clean the Internal Components

Required Materials

For all tasks:

- Protective gloves
- Safety glasses

For removing the shroud and some of the internal components:

- Screwdriver
- 1/8" hex key
- 3/32" hex key

For cleaning the internal dispense tubes and injector heads, as well as for wiping the surface under the plate carrier:

- Mild detergent
- Clean, lint-free cotton cloths
- Deionized or distilled water
- Stylus (stored in a plastic cylinder affixed to the rear of the dispense module or reader) (Part Number 2872304)

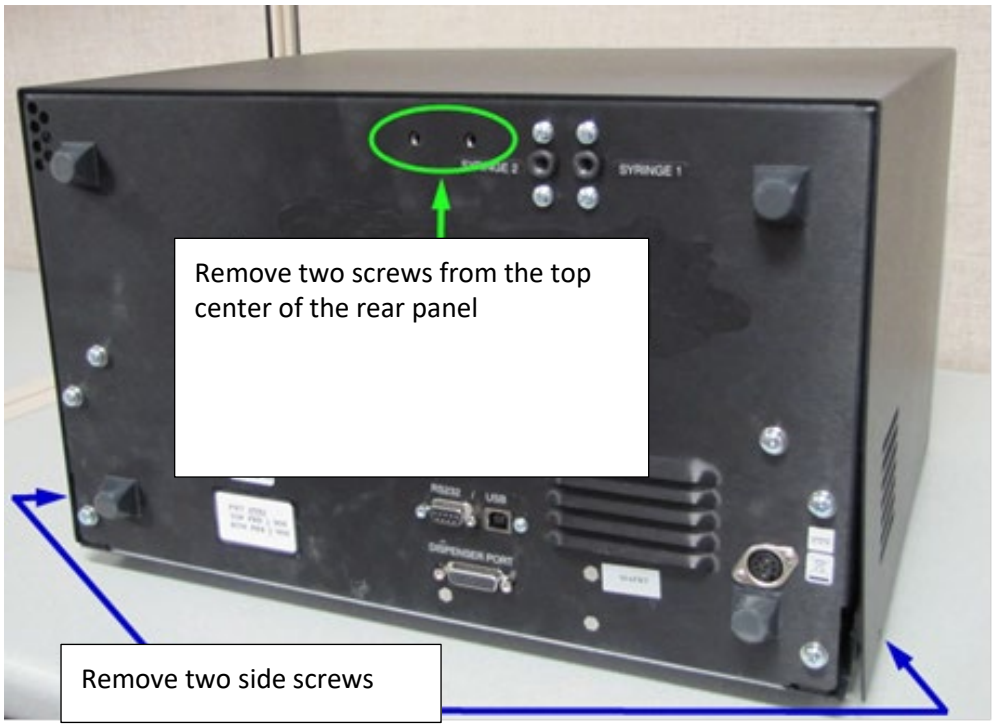
For cleaning the optical probes:

- Clean cotton swabs
- Isopropyl alcohol
- Lens-cleaning tissue

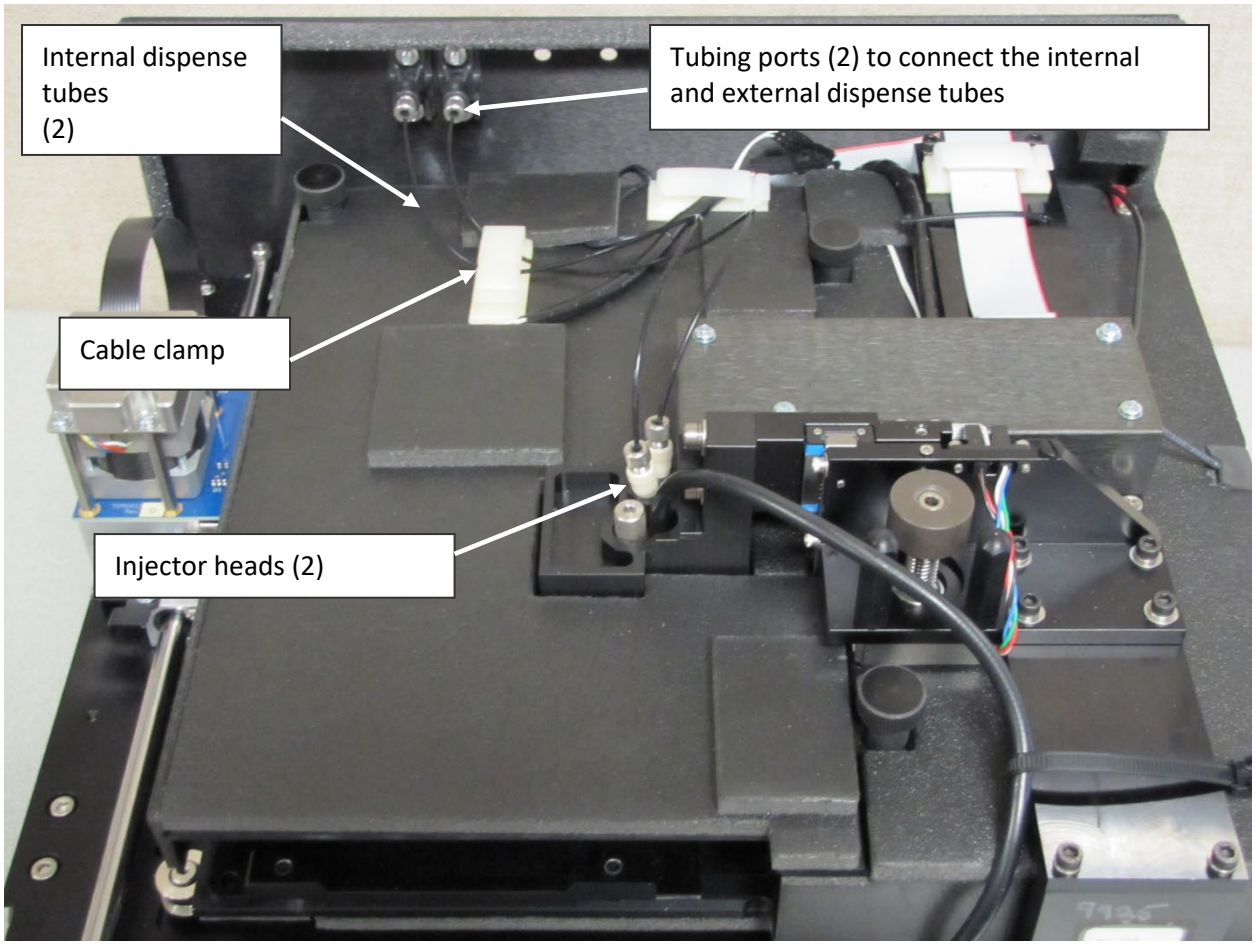
Remove the Reader's Shroud

1. Purge the injection system of all fluid.
2. Disconnect power and all cables. Set the external dispense module aside.
3. Remove four mounting screws: one at the bottom-rear corner on each side, and two at the top center of the rear panel.

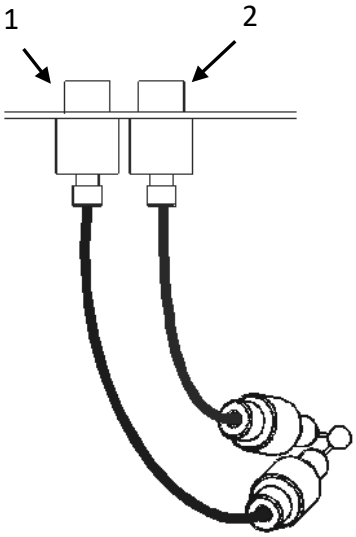
4. Stand facing the front of the instrument. Grasp both sides of the shroud, slide it toward you, and pull it straight off the instrument. Set the shroud aside.

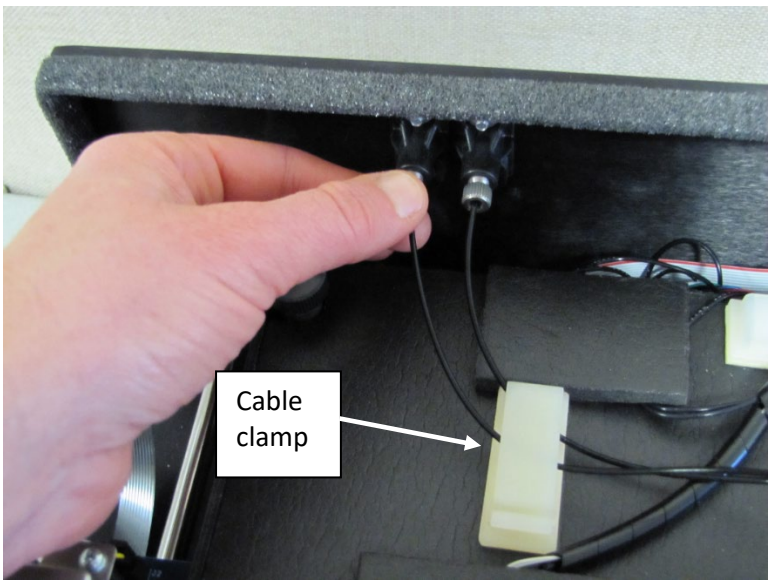


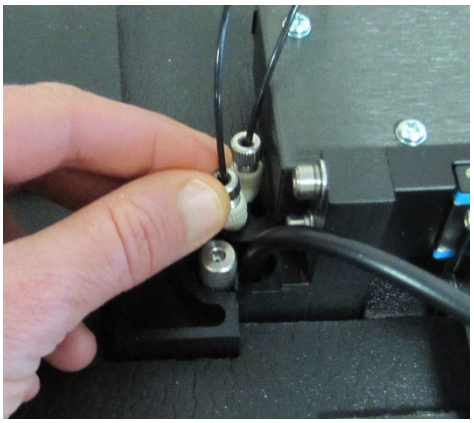
Removing the Internal Tubes and Injector Heads

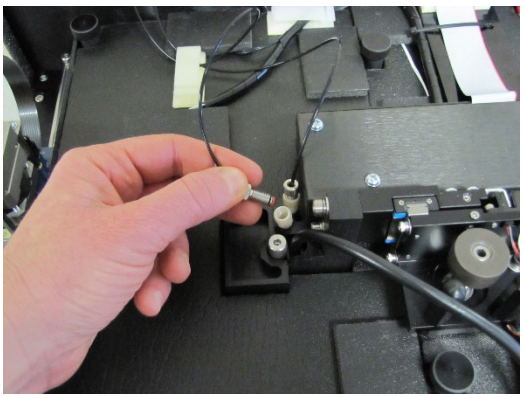


When reinstalling the internal dispense tubes, be sure to align the tubing ports with the injector heads as shown in this diagram. Look for the **SYRINGE 1** and **SYRINGE 2** labels on the instrument's rear panel.









Be sure to seat the injector tips securely when reinstalling.

Cleaning the Internal Tubes and Injector Heads

To clean the tubes:

1. Soak the internal tubes in hot, soapy water to soften and dissolve any hardened particles.
2. Flush each tube by holding it vertically under a stream of water from a faucet.

To clean the injector heads:

Do not remove the o-ring from the injector head.

1. Gently insert the stylus into each injector head pipe to clear any blockages. (The stylus should be stored in a plastic cylinder affixed to the rear of the dispense module or reader.)
2. Stream water from a faucet through the pipe to be sure it is clean. If the water does not stream out, try soaking the heads in hot, soapy water and then reinserting the stylus.

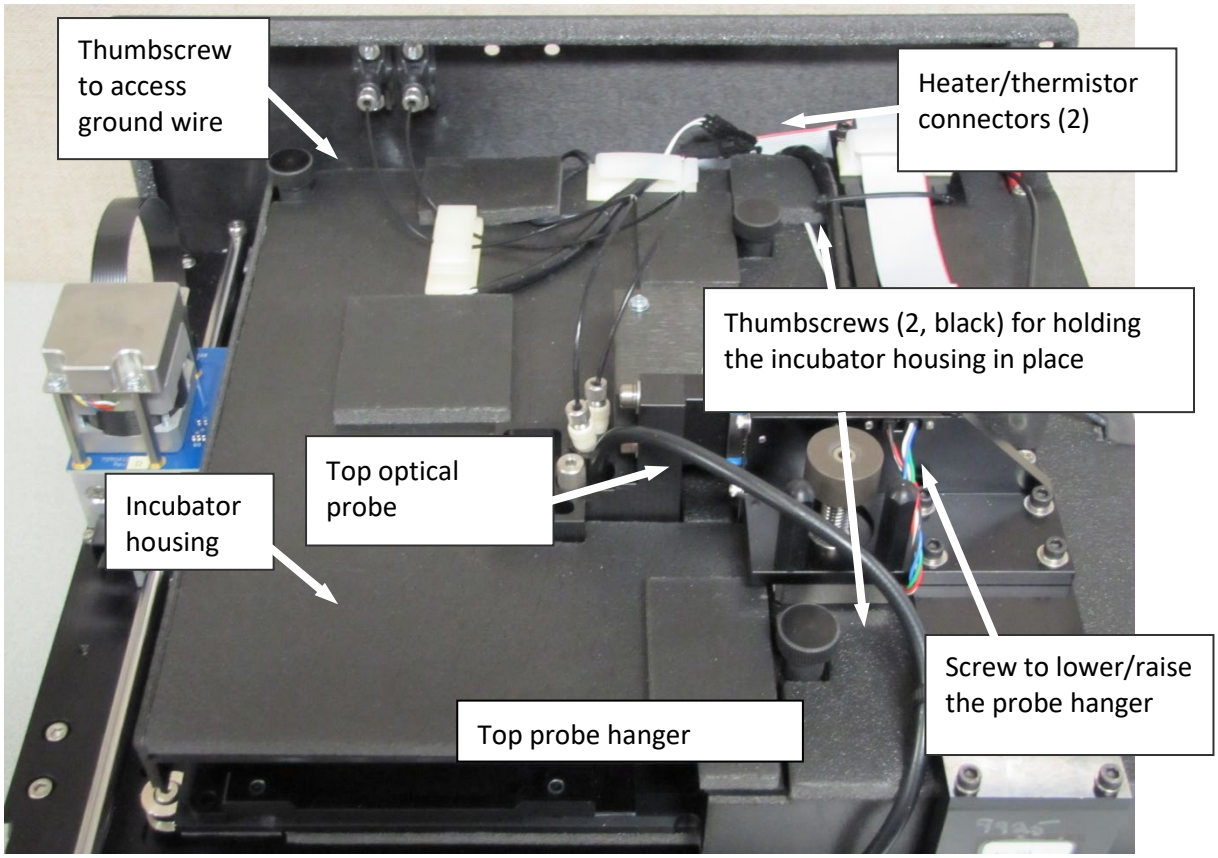
Cleaning the Optical Probes and Internal Surfaces

The optical probes should be cleaned if reagent has spilled and/or if an unusually high background signal has been flagged by the assay controls.

Contaminated probes can lead to a loss of sensitivity.

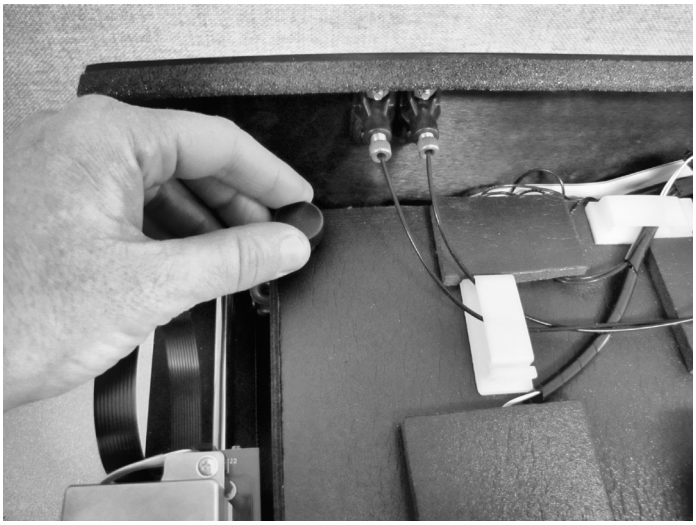
Supplies:

- Small container of isopropyl alcohol
- Small container of deionized or distilled water
- Lens-cleaning tissue
- Cotton swabs



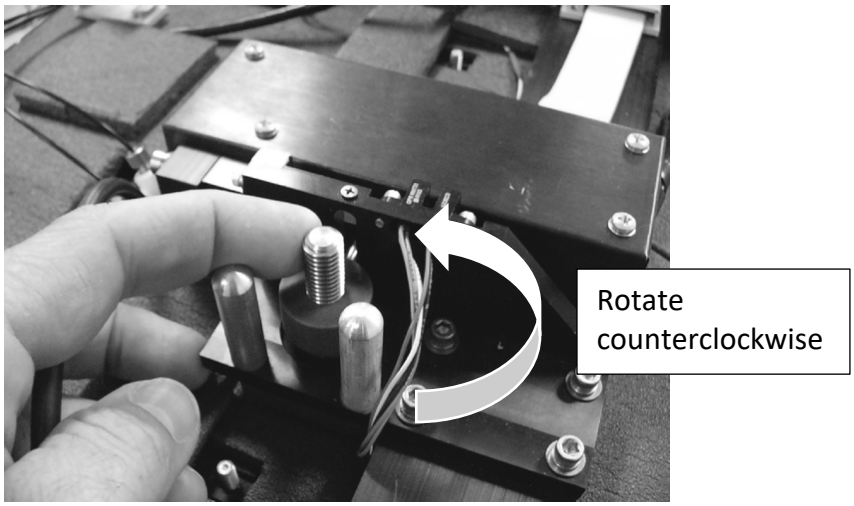
1. Disconnect the heater and thermistor wires. To do this, depress the small tab (pictured below) and separate the connectors.

2. Remove the thumbscrew located in the left rear of the instrument and set it aside. This exposes the ground wire.



3. Lift the ground wire and move it to the side.

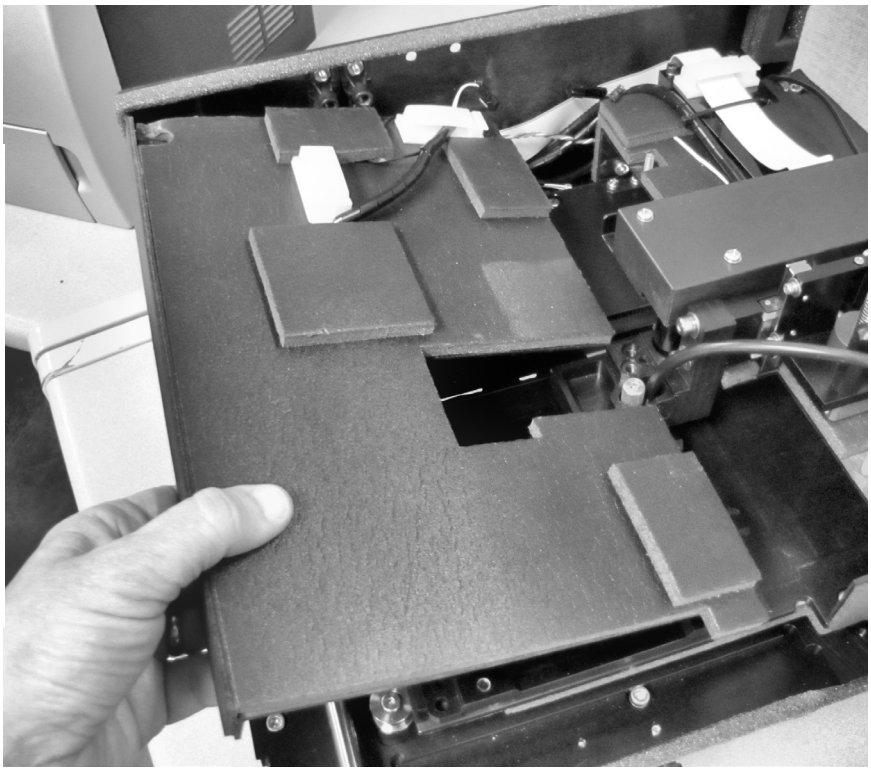
4. Remove the two black thumbscrews that hold the incubator housing in place and set them aside.
5. Turn the top probe screw counterclockwise to lower the probe hanger all the way to the bottom. (Rotate the screw, not the ring around it.)



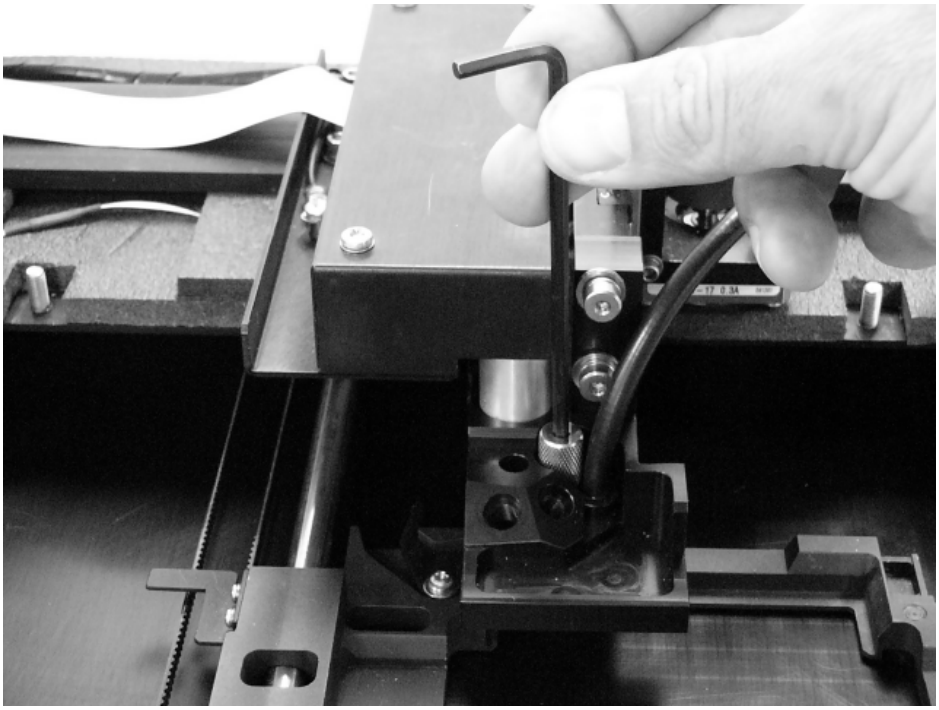
6. Gently lift the left side of the incubator housing and carefully slide it out.

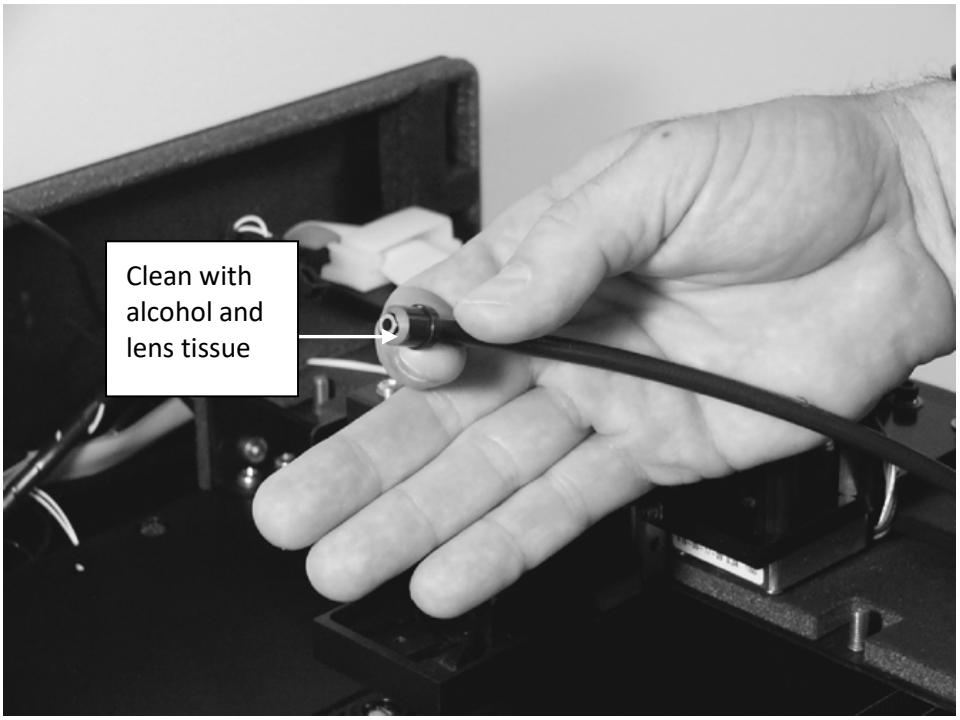
When replacing the incubator housing, the two “forks” on its right side should wrap around the holding screws.

The forks should not slide under the fixed foam housing.

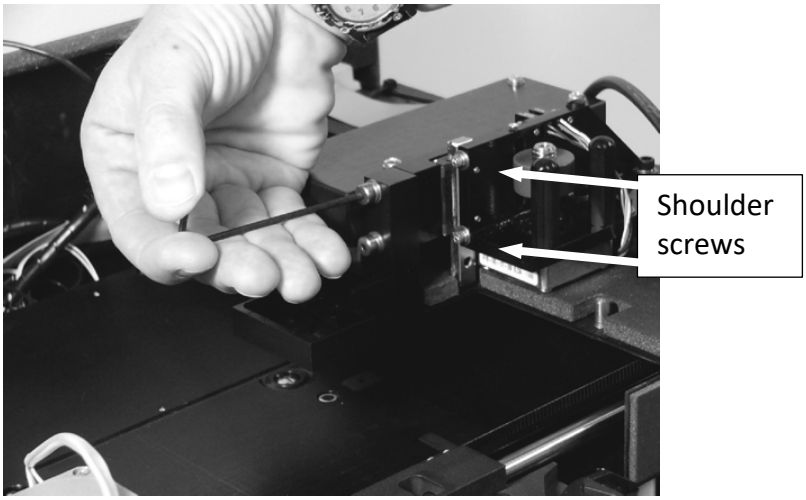


7. Use a 1/8" hex key to remove the top optical probe's holding screw.

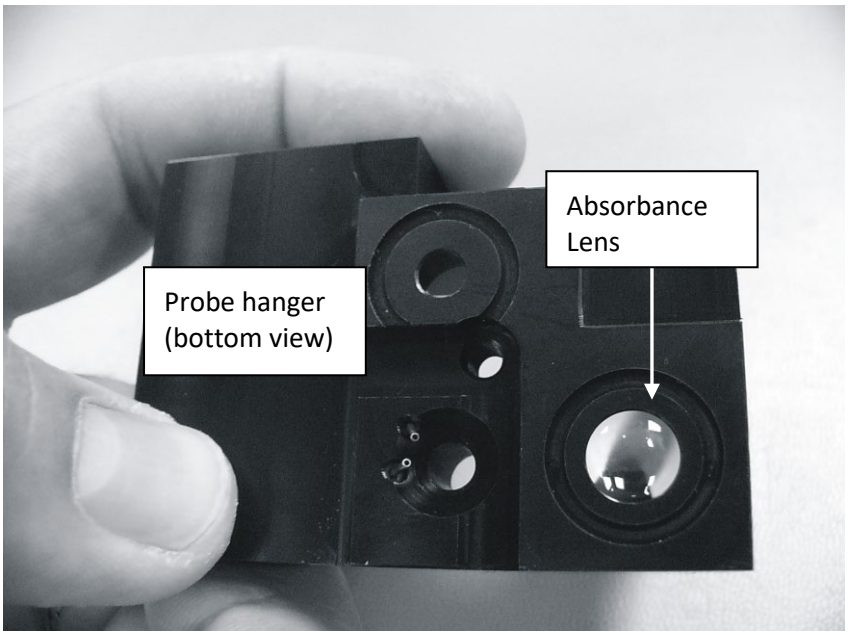




8. Use a 3/32" hex key to remove the two shoulder screws securing the top probe hanger. Remove the screws and set them aside.



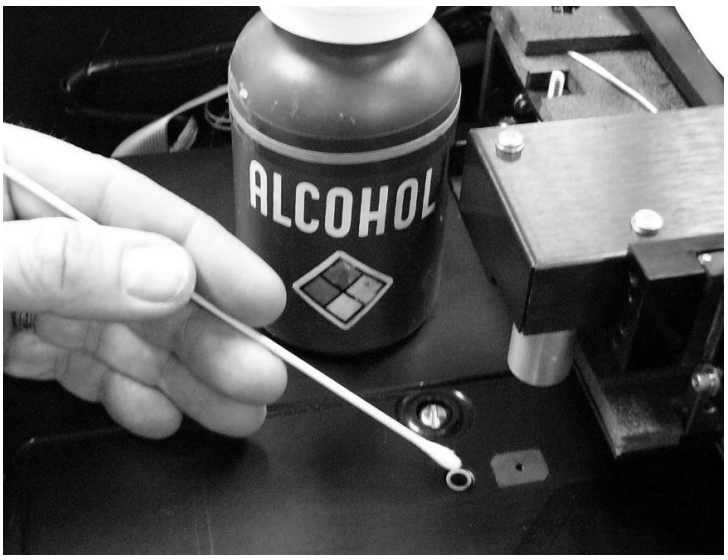
9. Drop the top probe hanger down and slide to the left to remove it. Turn the hanger upside down to clean the absorbance lens. **Do not touch the lens with your fingers!** Inspect the block for spills or other contamination. Carefully clean with mild detergent if necessary.



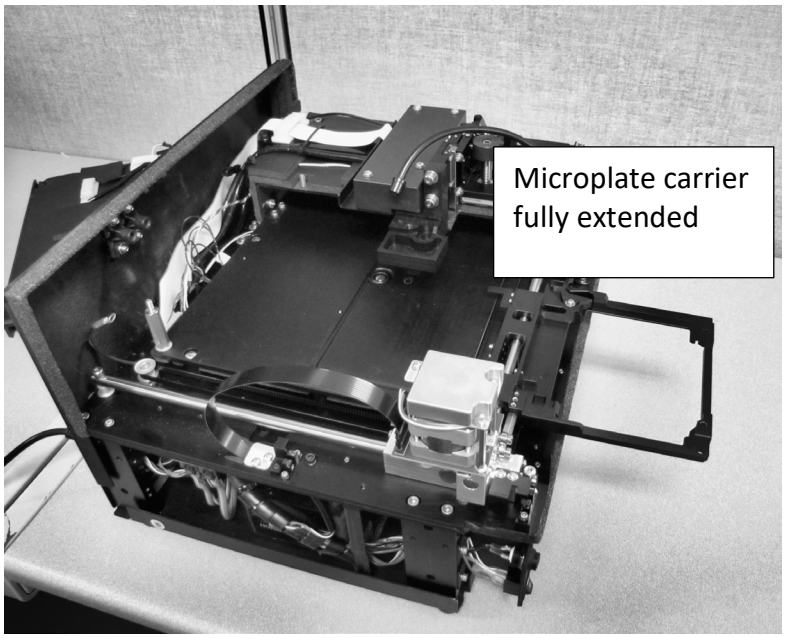
When cleaning the absorbance lens with the swab, apply very little pressure to the lens. Applying too much pressure can push the lens out of its holder; reinstallation must be performed by Agilent service personnel. If the lens does fall out, contact Technical Support.

10. Use a cotton swab moistened with alcohol to **gently** clean the lens on the top probe hanger.

11. Slide the microplate carrier out of the way. Use a cotton swab moistened with alcohol to clean the lens on the instrument surface.



Cleaning Internal Surfaces



1. Moisten (**do not soak**) a clean cotton cloth with alcohol, water, or with water and mild detergent. Wipe all sides of the plate carrier. Wipe the instrument's horizontal surface.

2. If detergent was used, wipe the surfaces with a cloth moistened with water.
3. Use a clean, dry, lint-free cloth to dry all wet surfaces.

Reassemble the Components

Perform these steps in the order listed to reassemble the components.

1. Slide the microplate carrier back into the instrument.
2. Insert the two injector heads into their sockets in the top probe hanger. Do not touch the absorbance lens with your fingers! Ensure that the injector heads are properly seated in the hanger. The knurled plastic should sit flush against the hanger surface.
3. Attach the two internal dispense tubes to the injector heads. Do not overtighten the thumbscrews!
4. Replace the top probe hanger and shoulder screws (using the 3/32" hex key).
5. Insert the top optic probe into its socket and replace its holding screw (using the 1/8" hex key).
6. Replace the incubator housing and two thumbscrews. Do not slide the two “forks” on the housing’s right side under the fixed foam housing.
7. Replace the ground wire and its thumbscrew.
8. Reconnect the heater and thermistor wires. Be sure to connect wires of the same color.
9. Attach the two internal dispense tubes to the tubing ports, taking care to align the correct port with the correct injector head.
10. Slide the two internal dispense tubes into the cable clamp and close the clamp.
11. Review the steps you just performed to make sure the components have been properly reassembled.
12. Slide the shroud onto the instrument.
13. Replace the four screws to securely attach the shroud to the base.

Performance Check

After reassembling the instrument, plug the instrument in and turn it on; allow its run-time system test to complete. Run a system test through Gen5.

Decontamination

WARNING **Internal Voltage.** Always turn off the power switch and unplug the power supply before cleaning the outer surface of the instrument.

CAUTION **Liquids.** Do not immerse the instrument, spray it with liquid, or use a dripping-wet cloth on it. Do not allow water or other cleaning solution to run into the interior of the instrument. If this happens, contact Technical Support.

- The Synergy HTX requires decontamination prior to shipping, storage, and disposal.
- Decontamination is required by the U.S. Department of Transportation regulations.
- Persons performing the decontamination process must be familiar with the basic setup and operation of the instrument.
- Agilent recommends the use of the following decontamination solutions and methods based on our knowledge of the instrument and recommendations of the Centers for Disease Control and Prevention (CDC). Neither Agilent nor the CDC assumes any liability for the adequacy of these solutions and methods. Each laboratory must ensure that decontamination procedures are adequate for the biohazard(s) they handle.

Required Materials

- Sodium hypochlorite (NaClO)
- 70% isopropyl alcohol (as an alternative to sodium hypochlorite)
- Deionized or distilled water
- Safety glasses
- Surgical mask
- Protective gloves
- Lab coat
- Biohazard trash bags
- 125-mL beakers
- Clean, lint-free cotton cloths

Additional Materials for Models with Injectors

- Screwdriver
- Small brush for cleaning the tip priming trough and priming plate
- (Optional) Mild detergent

Procedure for Models without Injectors

1. Turn off and unplug the reader.
2. Prepare a disinfecting solution: An aqueous solution of 0.5% sodium hypochlorite. If the effects of sodium hypochlorite are a concern, 70% isopropyl alcohol may be used.

3. Moisten a clean, lint-free cloth with the disinfecting solution, then thoroughly wring it out so that liquid does not drip from it. Do not soak the cloth.
4. Wipe the plate carrier and all exposed surfaces of the instrument.
5. Wait 20 minutes.
6. Moisten a cloth with deionized or distilled water and wipe all surfaces of the instrument that have been cleaned with the disinfecting solution.
7. Use a clean, dry lint-free cloth to wipe all wet surfaces.
8. Discard the used gloves and cloths, using a biohazard trash bag and an approved biohazard container.

Procedure for Models with Injectors

If disinfecting with sodium hypochlorite, be sure to flush repeatedly with deionized water to ensure that no sodium hypochlorite is carried over. After disinfecting with sodium hypochlorite, perform the rinse procedure.

If disinfecting with alcohol, do not immediately prime with deionized water, because the drying effect of the alcohol is an important aspect of its disinfectant properties.

1. Follow steps 1–7 of the procedure for models without the dispense module.
2. If the dispense module is installed, detach the outlet tubes from the rear panel of the instrument. If it is not installed, attach just the dispense module's communication cable to the instrument. Remove the supply bottles and their holders.
3. Perform the procedures described next to decontaminate the fluid lines in the dispense module, the internal tubing and injector heads, and the tip priming trough and priming plate.

Decontaminate the Fluid Lines

1. Place a beaker with 20 mL of 0.5% sodium hypochlorite solution or 70% isopropyl alcohol near SYRINGE 1 on the dispense module.
2. Place the SYRINGE 1 inlet tube in the beaker.
3. If you have not already done so, detach the dispense module's outlet tubes from the instrument's rear panel. Place the ends of the outlet tubes in an empty beaker and set the beaker on the work surface.
4. Launch Gen5, select **System > Instrument Control**, and click the **Prime** tab.
5. Select Dispenser 1, enter a Volume of 5000 µL, and keep the default dispense Rate.
6. Place the priming plate on the carrier (it is not used, but the reader requires its presence).
7. Run two prime cycles, for a total of 10,000 µL.

8. Pause for 20 to 30 minutes to allow the solution to disinfect the tubing.
9. Remove the inlet tube from the beaker of disinfectant solution.
10. From the Reader Control dialog, change the Volume to 1000 µL.
11. Run one prime cycle, to flush the disinfectant out of the fluid lines.
12. Empty the beaker containing the outlet tubes. Put the tubes back in.
13. If sodium hypochlorite was used, perform *Rinse the Fluid Lines*, below.

    Otherwise, (or after performing the Rinse procedure), repeat steps 1–13 for SYRINGE 2 / Dispenser 2.

Rinse the Fluid Lines

Perform this procedure only if decontamination was performed using sodium hypochlorite.

1. Place a beaker containing at least 30 mL of deionized water on the dispense module.
2. Place the SYRINGE 1 or 2 inlet tube in the beaker.
3. If you have not already done so, place the outlet tubes in an empty beaker.
4. From the Reader Control dialog, select **Dispenser 1** or **2**, set the Volume to 5000 µL, and keep the default dispense Rate.
5. Run five prime cycles, for a total of 25000 µL.
6. Pause for 10 minutes and then run one prime cycle with 5000 µL. This delay will allow any residual sodium hypochlorite to diffuse into the solution and be flushed out with the next prime.
7. Empty the beaker containing the outlet tubes.
8. Wipe all surfaces with deionized water.
9. Discard the used gloves and cloths using a biohazard trash bag and an approved biohazard container.

Clean the Internal Tubing and Injector Heads

Perform the procedures described earlier to access, remove, and clean the internal tubing and injectors. When finished, replace the internal components and the reader's shroud.

Clean the Tip Priming Trough and Priming Plate

1. Remove the tip priming trough from the left rear of the instrument's microplate carrier (see below).
2. Wash the tip priming trough and priming plate in hot, soapy water. Use a small brush or cloth to clean the corners of the trough and plate.

3. To decontaminate, soak the trough and plate in a container of 0.5% sodium hypochlorite or 70% isopropyl alcohol for 20 to 30 minutes.
4. If decontaminating in sodium hypochlorite solution, remove the trough and plate, and thoroughly rinse with deionized water.

   If decontaminating with alcohol, remove the trough and plate and let them air-dry.
5. Discard the used gloves and cloths using a biohazard trash bag and an approved biohazard container.

Instrument Testing

Recommended Qualification Schedule

The following schedule is recommended for a Synergy HTX used two to five times per week:

Tasks/Tests	IQ	OQ	PQ	
	Initially	Initially/ Annually	Monthly	Quarterly
All models:				
Installation, setup, and configuration of the reader, host computer, and Gen5	√			
System Test	√	√	√	
Models with absorbance capability:				
Absorbance Plate Test		√	√	
Absorbance Liquid Test 1 or Liquid Test 2*		√		√
(Optional) Absorbance Liquid Test 3 *or* 340 nm Absorbance Plate Test (using part number #7260551)		√		√
Models with fluorescence capability:				
Corners, Sensitivity, Linearity (FI) Tests		√	√	
Models with luminescence capability:				
Luminescence Test		√	√	
Models with injectors and an external dispense module:				
Installation and setup of external dispense module	√			
Injection System Test	√			
Dispense Accuracy and Precision Test		√		√

* If you have Absorbance Test Plate part number #7260522, perform Liquid Test 1. Otherwise, perform Liquid Test 2.

System Test

Each time the Synergy HTX is turned on, it performs a series of tests. If any test fails, the reader beeps repeatedly and the LED on the power switch flashes. If this occurs, press the carrier eject button to stop the beeping.

1. Turn on the reader and launch Gen5.
2. If necessary, set Gen5's wavelength table to the six wavelengths you most frequently use.
3. If your assays use incubation, we recommend enabling temperature control and allowing the incubator to reach its set point before running the system test. To access this feature, select **System > Instrument Control > Synergy HTX** and click the **Pre-Heating** tab.
4. Select **System > Diagnostics > Run System Test**.
5. When the test is complete, a dialog will appear, requesting additional information. Enter the information (if desired) and then click **OK**.
6. The test report will appear; it should show "SYSTEM TEST PASS". If it shows "SYSTEM TEST FAIL" contact Technical Support.
7. Print the report if required.
8. Turn off the incubator.

Absorbance Tests

Absorbance Test Plate Part Number 7260522 uses NIST-traceable neutral density filters to confirm absorbance specifications in the visible range (400–800 nm). This test plate also contains precision-machined holes to verify mechanical alignment.

Absorbance Test Plate Part Number 7260551 uses NIST-traceable neutral density filters to confirm absorbance specifications in the UV range (340 nm).

Define the Absorbance Test Plate Parameters

Before the Absorbance Plate Test can be performed, the wavelength settings and the calibration data for each wavelength selected must be initially entered into Gen5. Use the data sheet included with the Absorbance Test Plate for the following:

- Select **System > Diagnostics > Test Plates > Add/Modify Plates**, then click **Add**. Click **Help** for guidance when setting the wavelengths and entering the OD and peak wavelength values.

Run the Absorbance Plate Test

1. Select **System > Diagnostics > Test Plates > Run**. If prompted, select the desired Test Plate and click **OK**.
2. When the Absorbance Test Plate Options dialog appears, enter any required information, and, if applicable, select **Perform Peak Wavelength Test**.

3. Highlight the wavelength(s) to be included in this test. Select only those wavelengths most appropriate for your use of the reader.
4. (Optional) Enter a comment.
5. Click **Start Test**.
6. Place the Test Plate in the microplate carrier, with well A1 in the proper location.
7. Click **OK** to run the test.
8. When the test completes, the results report will appear. Scroll down through the report; every result should show "PASS."

If any result shows "FAIL", try the following and rerun the test. If the test continues to fail, contact Technical Support.

- Make sure the information entered into Gen5 matches the Test Plate's Certificate.
- Verify that the Test Plate is within its calibration certification period. If it is out of date, contact Agilent to schedule a recertification.
- Ensure that the Test Plate is correctly seated in the microplate carrier.
- Check the alignment (corner) holes on the Test Plate to ensure they are clear of debris.
- Check the filters on the Test Plate to ensure they are clean. If necessary, clean them with lens paper. Do not remove the filters from the test plate, and do not use alcohol or other cleaning agents.

Fluorescence Tests

For models with fluorescence capability, a solid-state Fluorescence Test Plate (Part Number 1400006) is used to test the fluorescence system.

The Fluorescence Test Plate simplifies the process of conducting fluorescence intensity, fluorescence polarization, and time-resolved fluorescent tests. The test plate is solid and immune to the pipetting errors, evaporation issues, and costs experienced with conventional liquid tests.

The test plate package includes Synergy HTX-specific Gen5 protocols designed for use with the test plate. The protocols include embedded Microsoft Excel spreadsheets to automatically calculate results and determine pass/fail.

The package also contains a user manual that describes the test methods, helps you get started with using the plate, and provides information for cleaning and maintaining the test plate. The manual also provides troubleshooting tips and information on the annual recalibration program.

Results Analysis

Refer to the *Fluorescence Test Plate User Manual* for descriptions of the data reduction calculations. The tests must meet the following criteria to pass:

Corners Test %CV	< 3.0

Sensitivity Tests	
Sodium Fluorescein analogue	
Top optics	Detection Limit <= 53 pM
Bottom optics	Detection Limit <= 30 pM
Methylumbelliferone analogue	
Top optics	Detection Limit <= 160 pg/mL
Bottom optics	Detection Limit <= 160 pg/mL
Linearity Test	R2 >= 0.9500

Luminescence Test

For models with luminescence capability, a Harta Luminometer Reference Microplate is used to test the luminescence system. The test plate is LED-based and NIST-traceable.

Materials

- Harta Luminometer Reference Microplate, Part Number 8030015 (includes adapter Part Number 8032028 for this reader)
- Gen5 protocol described below
- A plug in the excitation filter wheel
- An Open position (Hole) in the emission filter wheel

Procedure

1. Turn on the Harta reference plate using the I/O switch on the back of the plate.
2. Check the plate's battery by pressing the test button on the back of the plate and ensuring that the test light turns on. If the light does not turn on, replace the battery.
3. Place the adapter on the reader's microplate carrier, and then place the Harta reference plate on top of the adapter.
4. In Gen5, create an experiment based on **Synergy HTX LumTest_Harta.prt** protocol, and read the plate.
5. Calculate and evaluate results as described under **Results Analysis** below.
6. When finished, turn off the Harta reference plate to preserve battery life.

Results Analysis

1. Determine if the plate's battery is still functioning properly. If A8 > (0.2 * A7), the battery is good. Otherwise, it requires replacement.
2. On the Harta plate's calibration certificate, locate the NIST measurement for the A2 position and convert to attomoles: (A2 NIST measurement*0.02884)
3. Calculate the signal-to-noise ratio:
   (A2–Mean of the buffer cells)/(3 * Standard deviation of buffer cells)
4. Calculate the detection limit:
   A2 NIST measurement in attomoles/signal-to-noise ratio
   - If the reader is equipped with the low-noise PMT, the detection limit must be **<= 60 amol** to pass.
   - If the reader is equipped with the red-shifted PMT, the detection limit must be **<= 500 amol** to pass.

Gen5 Protocol Reading Parameters

Parameter	Setting
Plate Type:	“8030015 Harta - with 8032028 adapter”
Delay Step:	3 minutes
Read Step 1:	
Detection Method:	Luminescence
Read Type:	Endpoint
Optics Type	Filters
Step Label:	“Reference well A2”
Read Well:	A2
Filter Sets:	1
Excitation:	Plug
Emission:	Hole
Optics Position:	Top
Gain:	150
Integration Time:	0:10.00 MM:SS.ss
Delay After Plate Movement:	100 msec
Dynamic Range:	Standard
Read Height:	1.00 mm
Read Step 2:	
Detection Method:	Luminescence
Read Type:	Endpoint
Optics Type:	Filters
Step Label:	“Background”
Read Wells:	F1–G12
Filter Sets:	1
Excitation:	Plug
Emission:	Hole
Optics Position:	Top
Gain:	150
Integration Time:	0:10.00 MM:SS.ss
Delay After Plate Movement:	100 msec
Dynamic Range:	Standard

Read Height:	1.00 mm
Read Step 3:	
Detection Method:	Luminescence
Read Type:	Endpoint
Step Label:	“Battery Check”
Read Wells:	A7–A8
Filter Sets:	1
Excitation:	Plug
Emission:	Hole
Optics Position:	Top
Gain/Sensitivity:	50
Integration Time:	0:01.00 MM:SS.ss
Delay After Plate Movement:	100 msec
Dynamic Range:	Standard
Read Height:	1.00 mm

Injection System Tests

For models equipped with injectors and an external dispense module, the tests describe below assure that the injection system performs to specification.

Test Method

The Accuracy Test is a measure of the mean volume per well for multiple dispenses. The actual weight of the dispensed fluid is compared to the expected weight and must be within a certain percentage to pass. Pass/Fail criteria depends on the per-well volume dispensed: 2.0% for 80 µL, 5.0% for 20 µL, and 20.0% for 5 µL.

The Precision Test is a measure of the variation among volumes dispensed to multiple wells and uses the green test dye solution. For each volume dispensed (80 µL, 20 µL, and 5 µL) to four columns, the %CV of 32 absorbance readings is calculated. Pass/Fail criteria depends on the per-well volume dispensed: 2.0% for 80 µL, 7.0% for 20 µL, and 10.0% for 5 µL. Columns 1–4 are read at 405/750 nm and columns 5–12 at 630/750 nm.

Gen5 Parameters

The information in this section represents the recommended reading parameters for the referenced Gen5 protocol(s). It is possible that your tests will require modifications to some of these parameters, such as the Plate Type.

The Plate Type setting in each Gen5 protocol should match the actual plate in use.

Synergy HTX Disp 1 Test.prt and **Synergy HTX Disp 2 Test.prt** (for use with models with Absorbance capability)

Parameter	Default Setting
Plate Type	96 WELL PLATE
Dispense Step	Dispenser <1 or 2> Wells A1–H4 Tip prime before this dispense step, 20 µL Dispense 80 µL at 275 µL/sec
Plate Out,In	Comment: Weigh the plate (80 uL test). RECORD the weight, TARE the balance. Place the plate back on the carrier. Click OK to continue.
Dispense Step	Dispenser <1 or 2> Wells A5–H8 Tip prime before this dispense step, 20 µL Dispense 20 µL at 250 µL/sec
Plate Out,In	Comment: Weigh the plate (20 uL test). RECORD the weight, TARE the balance. Place the plate back on the carrier. Click OK to continue.
Dispense Step	Dispenser <1 or 2> Wells A9–H12

	Tip prime before this dispense step, 5 µL Dispense 5 µL at 225 µL/sec
Plate Out,In	Comment: Weigh the plate (5 uL test). RECORD the weight, TARE the balance. PIPETTE 150 µL/well of DI water into all 12 columns. Place the plate back on the carrier. Click OK to perform the Read steps.
Shake Step	Linear, 15 seconds, default frequency
Read Step	Detection Method: Absorbance Read Type: Endpoint Optics Type: Monochromator Step label: 80 ul Read_Disp <1 or 2> Wells: A1–H4 Wavelengths, 2: 405 nm, 750 nm Speed: Normal
Read Step	Detection Method: Absorbance Read Type: Endpoint Optics Type: Monochromator Step label: 20 and 5 ul Read_Disp <1 or 2> Wells: A5–H12 Wavelengths, 2: 630 nm, 750 nm Speed: Normal
Data Reduction	Define two Delta OD transformations: 405–750 nm for the 80 uL Read step, columns 1–4 630–750 nm for the 20 and 5 uL Read step, columns 5-12

Synergy HTX Disp 1 Test No Read.prt and **Synergy HTX Disp 2 Test No Read.prt** (for use with models without Absorbance capability)

Parameter	Default Setting
Plate Type	96 WELL PLATE
Dispense Step	Dispenser <1 or 2> Wells A1–H4 Tip prime before this dispense step, 20 µL Dispense 80 µL at 275 µL/sec
Plate Out,In	Comment: Weigh the plate (80 uL test). RECORD the weight, TARE the balance. Place the plate back on the carrier. Click OK to continue.
Dispense Step	Dispenser <1 or 2> Wells A5–H8 Tip prime before this dispense step, 20 µL Dispense 20 µL at 250 µL/sec
Plate Out,In	Comment: Weigh the plate (20 uL test). RECORD the weight, TARE the balance. Place the plate back on the carrier. Click OK to continue.

Dispense Step	Dispenser <1 or 2> Wells A9–H12 Tip prime before this dispense step, 5 µL Dispense 5 µL at 225 µL/sec
Plate Out,In	Comment: Weigh the plate (5 uL test). RECORD the weight, TARE the balance. PIPETTE 150 µL/well of DI water into all 12 columns. Place the plate back on the carrier. Click OK to perform the Read steps.
Read Step	*Define a brief Read step for a single well. The measurement value will not be used. The step is only necessary because Gen5 requires a Read step with dispense protocols.*

Synergy HTX Disp Test Other Reader.prt (for use with an Agilent BioTek absorbance-capable reader other than Synergy HTX)

Parameter	Default Setting
Shake Step	<medium intensity> for 15 seconds
Read Step	Detection Method: Absorbance Read Type: Endpoint Optics Type: <as appropriate for the reader type> Step label: 80 ul Read Wells: A1..H4 Wavelengths, 2: 405 nm, 750 nm Speed: Normal
Read Step	Detection Method: Absorbance Read Type: Endpoint Optics Type: <as appropriate for the reader type> Step label: 20 and 5 ul Read Wells: A5..H12 Wavelengths, 2: 630 nm, 750 nm Speed: Normal
Data Reduction	Define two Delta OD transformations: 405-750 nm for the 80 ul Read step, columns 1-4 630-750 nm for the 20 and 5 ul Read step, columns 5-12

Test Procedure

Materials

- Absorbance reader with capability of reading at 405, 630, and 750 nm. The reader must have an accuracy specification of ±1.0% ±0.010 OD or better and a repeatability specification of ±1.0% ±0.005 OD or better. The Synergy HTX may be used if it supports Absorbance and has passed the Absorbance Plate Test.
- Microplate shaker (if the absorbance reader does not support shaking)
- Precision balance with capacity of 100 g minimum and readability of 0.001 g
- 50–200 µL hand pipette and disposable tips
- Deionized water
- Supply bottles
- 250-mL beaker
- New 96-well, clear, flat-bottom microplates
- Green Test Dye Solution (BTI #7773003) undiluted
- Gen5 protocols listed below (as applicable for your reader model) and described in the previous section

  For models with Absorbance capabilities:

  Synergy HTX Disp 1 Test.prt
  Synergy HTX Disp 2 Test.prt

  For models without Absorbance capabilities:

  Synergy HTX Disp 1 Test No Read.prt
  Synergy HTX Disp 2 Test No Read.prt

  and, if you will use Gen5 with another Agilent BioTek absorbance-capable reader:

  Synergy HTX Disp Test Other Reader.prt

Test Procedure for Models with Absorbance Capability

1. Prime both dispensers with 4000 µL of deionized or distilled water.
2. Remove the inlet tubes from the supply bottles. Prime both dispensers with the Volume set to 2000 µL. This prevents the water from diluting the dye.
3. Fill a beaker with at least 20 mL of the green dye solution. Prime both dispensers with 2000 µL of the solution. When finished, remove the priming plate from the carrier.
4. Create an experiment based on the Synergy HTX Disp 1 Test protocol.
5. Place a new 96-well microplate on the balance and tare the balance.
6. Place the plate on the microplate carrier.

When each dispense step is finished, you will weigh the plate, record the weight, tare the balance with the plate on it, and then place the plate back on the carrier for the next step.

7. Initiate a plate read. Gen5 will prompt you to empty the tip priming trough.
8. When ready, proceed with the experiment. The sequence is as follows:
    - 80 µL/well is dispensed to columns 1–4.
    - When prompted, remove the plate and weigh it. Record the weight and tare the balance. Place the plate on the carrier.
    - 20 µL/well is dispensed to columns 5–8.
    - When prompted, remove the plate and weigh it. Record the weight and tare the balance. Place the plate on the carrier.
    - 5 µL/well is dispensed to columns 9–12.
    - When prompted, remove the plate and weigh it. Record the weight.
    - Manually pipette 150 µL of deionized or distilled water into all 12 columns, on top of the green test dye solution.
    - Place the plate on the carrier for the shake and read steps.
9. When the experiment is complete, save the file with an identifying name.
10. Remove the plate from the carrier and set it aside.
11. Repeat the procedure using the Synergy HTX Disp 2 Test protocol and a new microplate.
12. When the tests are complete:
    - Prime both dispensers with at least 5000 µL of deionized water to flush out the dye solution.
    - See the Results Analysis section.

Test Procedure for Models without Absorbance Capability

If you are not using an Agilent BioTek absorbance reader for this procedure, prepare your reader to perform two reads with the following characteristics:

	80 µL Read	20 and 5 µL Read
Primary Wavelength:	405 nm	630 nm
Reference Wavelength:	750 nm	750 nm
Plate Columns:	1–4	5–12

1. Prime both dispensers with 4000 µL of deionized or distilled water.
2. Remove the inlet tubes from the supply bottles. Prime both dispensers with the Volume set to 2000 µL. This prevents the water from diluting the dye.

3. Fill a beaker with at least 20 mL of the green dye solution. Prime both dispensers with 2000 µL of the solution. When finished, remove the priming plate from the carrier.
4. Create an experiment based on the Synergy HTX Disp 1 Test No Read protocol.
5. Place a new 96-well microplate on the balance and tare the balance.
6. Place the plate on the microplate carrier.

When each dispense step is finished, you will weigh the plate, record the weight, tare the balance with the plate on it, and then place the plate back on the carrier for the next step.

7. Initiate a plate read. Gen5 will prompt you to empty the tip priming trough.
8. When ready, proceed with the experiment. The sequence is as follows:
    - 80 µL/well is dispensed to columns 1–4.
    - When prompted, remove the plate and weigh it. Record the weight and tare the balance. Place the plate on the carrier.
    - 20 µL/well is dispensed to columns 5–8.
    - When prompted, remove the plate and weigh it. Record the weight and tare the balance. Place the plate on the carrier.
    - 5 µL/well is dispensed to columns 9–12.
    - When prompted, remove the plate and weigh it. Record the weight.
    - Manually pipette 150 µL of deionized or distilled water into all 12 columns, on top of the green test dye solution.
    - Carefully set the plate aside.
9. Close the experiment without saving it.

If you are not using an Agilent BioTek absorbance reader, read the plate using the parameters described in the table above. Perform the calculations and determine pass/fail according to the instructions in the Results Analysis section.

10. If you are using an Agilent BioTek absorbance reader, configure Gen5 to communicate with the reader.
11. Create an experiment based on the Other Reader protocol and read the plate.
12. When the experiment is complete, save the file with an identifying name.
13. Remove the plate from the carrier and set it aside.
14. Repeat the procedure using the Synergy HTX Disp 2 Test No Read protocol and a new microplate.
15. When the tests are complete:

- Prime both dispensers with at least 5000 µL of deionized water to flush out the dye solution.
- Refer to the instructions in the Results Analysis section.

Results Analysis

When the experiment for one injector is complete, 32 delta OD values are reported for each of the three dispense volumes. The pass/fail criteria for each set of 32 wells with the same dispense volume is based on the calculated coefficient of variation (% CV) and Accuracy % Error.

For each volume dispensed (80 µL, 20 µL, 5 µL), for each injector (1, 2):

1. Calculate the Standard Deviation of the 32 wells
2. Calculate the Mean of the 32 wells
3. Calculate the %CV: (Standard Deviation / Mean) x 100
4. Calculate the Accuracy % Error:

   ((ActualWeight–ExpectedWeight)/ExpectedWeight)* 100

Expected Weights for 32 wells: 80 µL (2.560 g), 20 µL (0.640 g), 5 µL (0.160 g). It is assumed that one gram is equal to one milliliter.

Dispense Volume	To pass, %CV must be:	To pass, Accuracy % Error must be:
80 µL	≤ 2.0%	≤ 2.0%
20 µL	≤ 7.0%	≤ 5.0%
5 µL	≤ 10.0%	≤ 20.0%

If any tests fail, prime the fluid lines and rerun the tests. If the tests fail again, the injectors may require cleaning. If tests continue to fail, contact Technical Support.

Specifications

General Specifications

Microplates	
All models accommodate standard 6-, 12-, 24-, 48-, 96-, and 384-well microplates with 128 x 86 mm geometry up to 28.575 mm high, and the Take3 and Take3 Trio Micro-Volume Plates.	
Hardware and Environmental	
Light Source	**Absorbance**: Xenon flash light source **Fluorescence**: Tungsten halogen, 20W power
Dimensions	40.6 cm x 40.6 cm x 25.4 cm
Weight	17 kg
Environment	Operational: Temperature range 18°C–40°C 10% to 85% relative humidity (non-condensing) Storage: Temperature range -25°C to 50°C 10% to 80% relative humidity (non-condensing)
Power Supply	24V external power supply compatible with 100-240 V~; +10% @50-60 Hz
Power Consumption	100 VA maximum, 130 VA maximum with injectors
Incubation	Temperature control range from 4°C over ambient to 50°C. Temperature variation ±0.5°C at 37°C, tested with Innovative Instruments, Inc. temperature test plate. Top and bottom incubation controlled via software-adjustable gradient.

Absorbance Specifications

Optics	
Wavelength range:	200 to 999 nm
Wavelength accuracy:	± 2 nm
Wavelength precision:	± 0.2 nm (standard deviation)
Wavelength bandpass:	2.4 nm
Resolution:	0.0001 OD
Increment:	1 nm

Performance

All qualifications were conducted using 96-/384-well, flat-bottom microplates. For the performance described here, the Gain on the Optics Test should be below 8.

Accuracy

96-well plate, normal read speed:

0.000 to 2.000 OD ±1.0% ±0.010 OD, Delay after plate movement: 100 ms
2.000 to 3.000 OD ±3.0% ±0.010 OD, Delay after plate movement: 100 ms

384-well plate, normal read speed:

0.000 to 2.000 OD ±2.0% ±0.010 OD, Delay after plate movement: 100 ms
2.000 to 2.500 OD ±3.0% ±0.010 OD, Delay after plate movement: 100 ms

96-well and 384-well plate, sweep read speed:

0.000 to 1.000 OD ±1.0% ±0.010 OD

Linearity

96-well plate, normal read speed:

0.000 to 2.000 OD ±1.0% ±0.010 OD, Delay after plate movement: 100 ms
2.000 to 3.000 OD ±3.0% ±0.010 OD, Delay after plate movement: 100 ms

384-well plate, normal read speed:

0.000 to 2.000 OD ±2.0% ±0.010 OD, Delay after plate movement: 100 ms
2.000 to 2.500 OD ±3.0% ±0.010 OD, Delay after plate movement: 100 ms

96-well and 384-well plate, sweep read speed:

0.000 to 1.000 OD ±1.0% ±0.010 OD

Repeatability

96-well and 384-well plate, normal read speed:

0.000 to 2.000 OD ±1.0% ±0.005 OD, Delay after plate movement: 100 ms
2.000 to 3.000 OD ±3.0% ±0.005 OD, Delay after plate movement: 100 ms

96-well and 384-well plate, sweep read speed:

0.000 to 1.000 OD ±2.0% ±0.010 OD

Read Timing

Minimum kinetic interval (450 nm): Sweep mode, < 20 seconds, 96-well plate

Plate in to plate out (450 nm): Sweep mode, <35 seconds, 96-well plate

Fluorescence Specifications

Optics

Optic Probes	Configuration is model-dependent Top, 3 mm probe Bottom, 5 mm probe
Detection	PMT, low-noise standard; red-shifted (850 nm) option available

Sensitivity (96-well, 200 µL, 50 measurements, standard PMT)

5 mm optical probe, bottom reading

DL Sodium Fluorescein in PBS, Excitation 485/20, Emission 528/20 <= 30 pM
3 mm optical probe, top reading

DL Sodium Fluorescein in PBS, Excitation 485/20, Emission 528/20 <= 53 pM
DL Methylumbelliferone in CBB, Excitation 360/40, Emission 460/40 <=0.16 ng/mL

Read Timing

Excitation Filter 485/20 nm, Emission Filter 528/20 nm; 10 measurements per data point; 100 ms delay after plate movement.

Minimum kinetic interval: <55 seconds

Time-Resolved Fluorescence

For "T" models
Delay: 0, or 20 to 2,000 µs
Integration Time: 20 to 2,000 µs
Granularity: 10-µs step

Luminescence Specifications

≤ 60 amol/well DL ATP in a 96-well plate (low-noise PMT), 20 amol typical
≤ 500 amol/well in a 96-well plate (red-shifted PMT)
10-second integration, PMT sensitivity 150, 16 blank wells

Dispense/Read Specifications

Applies only to models equipped with injectors

Plate Type	Dispenses to standard 6-, 12-, 24-, 48-, 96-, and 384-well microplates with 128 x 86 mm geometry
Detection Method	Absorbance, Fluorescence (including TRF), Luminescence
Volume Range	5–1000 µL with a 5–20 µL tip prime
Accuracy	±1 µL or 2.0%, whichever is greater
Precision	≤2.0% for volumes of 50–200 µL ≤4.0% for volumes of 25–49 µL ≤7.0% for volumes of 10–24 µL ≤10.0% for volumes of 5–9 µL
Injection Speeds	225, 250, 275, and 300 µL/sec

Delay between the end-of-dispense and start-of-read processes (96-/384-well plates, default probe heights only)

Absorbance: T ≤ 3 seconds
Top Filter Fluorescence: T ≤ 1 seconds
Bottom Filter Fluorescence: T ≤ 1 seconds
Luminescence: T ≤ 0.5 seconds

In This Book

This document contains installation, operation, maintenance, and qualification information for all models of the Synergy HTX.

Document Revision History			
Part Number	Revision	Date	Modifications
1341004I	D	August 2022	Added a CAUTION statement regarding DMSO Concentration. Added a CAUTION statement regarding the expected benchtop conditions.

Original Language – EN

Manufactured by Agilent Technologies, Inc.
5301 Stevens Creek Blvd.
Santa Clara, CA 95051