



Helium Spectrophotometer

User Guide













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1. Introduction

Product Technology

DeNovix Helium Spectrophotometers are compact, stand-alone instruments with a high-resolution touchscreen. EasyApps™ software utilizes application-specific icons along with a custom operating system to provide an intuitive, easy-to-use instrument for absorbance measurements.

Sample volumes as low as one microliter can be accurately quantified. SmartPath™ technology automatically adjusts the microvolume pathlengths to enable sample measurements across a broad concentration range. Bridge Testing™ verification identifies and adjusts in real-time for a broken sample column, ensuring accurate measurements of difficult to pipette, low surface tension samples like proteins.

Absorbance range specifications are published on the DeNovix website.

Key Features

- Stand-Alone Design, no-PC Required
- Hi-Definition 7-inch Touchscreen Interface
- Simple, Intuitive EasyApps™ Software
- 1 µL UV Analysis
- USB and Label Printer Export

Please refer to our website for a list of supported label printers.

2. Set Up and Safety

- 1. Remove packaging, including the covers between the arm and the top of the instrument.
- 2. Plug the unit into a 100-240 VAC, 50-60/Hz receptacle using the grounded power supply (Globtek part number TR9CE3000C95CP-NR6B / RoHS Compliant) included with each unit. Input Rating: 100-240V~, 50-60 Hz, IEC 60320/C14 , Output Rating: 36W, 12V@3A
- 3. Turn on the instrument by toggling the power switch on the rear to the on position. The instrument will boot up in under one minute.
- 4. Important: Ensure that the arm is in the down position. Follow the onscreen instructions to complete the system Initialization each time the instrument is restarted.
- 5. Turn off the power switch to shut down the instrument.

Cautions

DO NOT REMOVE COVER

No operator serviceable components inside. Refer servicing to qualified personnel.

The Helium Spectrophotometer is designed for indoor use under the following conditions:

Temperature: 15° to 35° C Humidity: 35 to 65 % Elevation: 2000m or less Pollution: Pollution degree 2



Use only the power supply provided with the instrument. Use of the instrument in any manner not specified by the manufacturer may impair the protection provided by the supplied power cord and power supply.

NE PAS OUVRIR L'APPAREIL.

Aucun composant réparable par un utilisateur est inclus. Confiez l'entretien à du personnel qualifié .

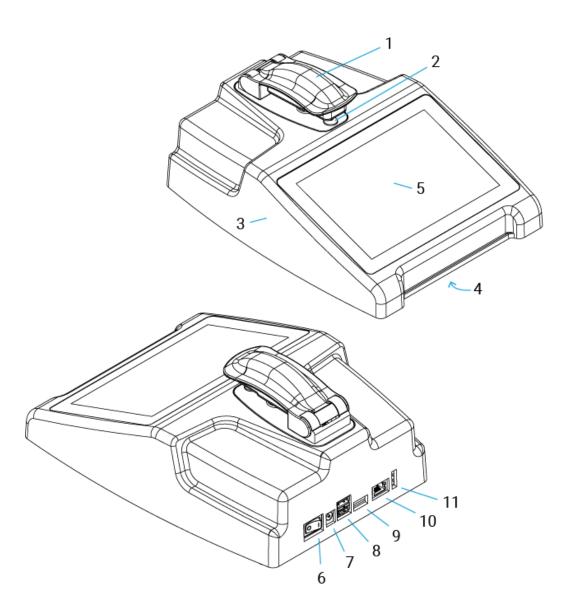
Le Helium est conçu pour une utilisation en intérieur dans les conditions suivantes :

Température : 15° à 35°C Humidité:35 à 65% Altitude : 2 000 m ou moins Pollution : Degré de pollution 2

Utilisez uniquement le bloc d'alimentation inclus avec l'instrument. Utiliser l'appareil dans des conditions non spécifiées par le fabricant peut compromettre la protection offerte par le cable d'alimentation et le bloc d'alimentation fournis avec l'appareil.

3. Instrument Diagram

Helium Component Details



- 1. Arm
- 2. Sample surface
- 3. Cover
- 4. Serial number & regulatory info (underside)
- 5. Touchscreen
- 6. Power switch

- 7. DC power in from power supply
- 8. USB ports (2)
- 9. Unused
- 10. Ethernet port (unused)
- 11. LED indications



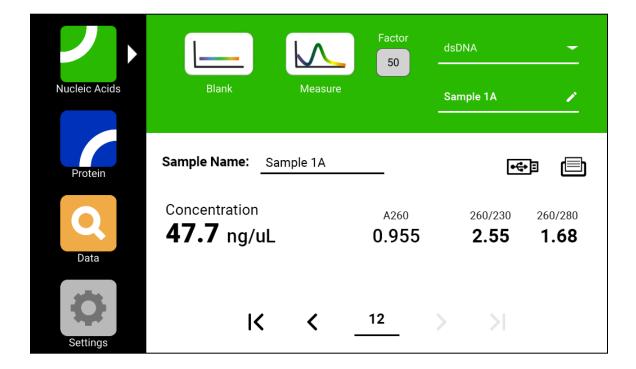
4. EasyApps™ Software Quick Guide

27	Apps are launched using the Application Menu icons.
	The Blank button is used to establish a reference absorbance within each measurement app.
	The Measure button is used for sample measurements. The button is inactive until a Blank is complete.
	Fields with Dropdown Menus are indicated by triangles located on the right.
ř	Text fields with the pencil icon are editable by the user.
•⇔]3	Export to USB.
	Print to a compatible label printer.
i	Delete
::	Select All
I< >I	Move to the first or last saved result.
< >	Move to the previous or next saved result.

5. Run Screen

Measurement applications provide all of the relevant information for a given sample on a single screen. Inputs such as sample name can be added prior to measurement. Measurement data for concentration, absorbance and purity ratios are clearly displayed after data collection.

Run Screen



- Blank and Measure: Use the **Blank** button when blanking with the reference solution and the **Measure** button for samples.
- Sample Type: Select from DNA, RNA or ssDNA in the Nucleic acids App or select the protein of interest in the Protein App using the dropdown menu.
- Factor: Used to calculate the concentration of dsDNA (50), RNA (40) or ssDNA (33) from the measured absorbance.
- Sample Name (optional): Tap the input box to display an onscreen keyboard or use an Opticon OPI 3601 USB scanner to enter sample names.
- Printing and Data Export. All measurements are automatically saved to the instrument.
 Data can be exported to USB or Label Printer on the Run Screen or in the Data App.
 - o Run Screen: A screenshot of the current on-screen result is exported.
 - Data app: Search for and select specific results for export. A .csv file and pdf report of the selected results are exported.

Microvolume Measurements

- 1. Ensure both top and bottom sample surfaces are clean.
- 2. Pipette 1 μ L of the blank solution onto the lower sample surface. Lower the top arm and tap the **Blank** button.
- 3. Remove the solution from both sample surfaces using a clean, dry lab wipe.
- 4. Pipette 1 μ L of the sample solution onto the lower sample surface.
- 5. Lower the arm and tap the **Measure** button. All microvolume measurements are reported in 10 mm equivalent absorbance values.

6. Best Practices

Absorbance Measurement Best Practices

- Clean both sample measurement surfaces prior to making the Blank measurement.
- Use dH₂O, not detergents or alcohol, for routine sample measurement surface cleaning.
- Use a dry lab wipe to remove measurement liquids from both the top and bottom surfaces immediately after each measurement.
- Use 1 µL samples for microvolume measurements.
- Use a fresh aliquot for each measurement.
- Use a fresh tip to deliver each sample aliquot.
- Avoid introducing bubbles when pipetting samples onto the measurement surfaces.
- Ensure sample concentrations fall within absorbance limits of the instrument for accurate results.
- Use the same buffer a sample is suspended in for the Blank measurements.
- Avoid using buffers such as RIPA buffers that contain components with strong absorbances at the wavelength of interest.
- Measure a fresh aliquot of the buffer using the Measure button and ensure the measured result is near zero before measuring samples.

7. Tech Team Tips

Initialization

- Before the system is powered on, always remove all packaging or protective materials from between arm and sample surfaces.
- Follow onscreen instructions to clean the sample surfaces.
- Lower the arm and Press OK to complete the system Initialization.
- Although there is no harm in doing so, there is no need to shut down the instrument each day. The backlight for the screen will automatically turn off to protect the screen and will switch on when touched.

Connectivity and Data Export

- Use the USB icon to export results to a USB drive, or the Printer icon export the result to a label printer.
- Helium can store up to 2,000 results. Once the limit has been reached, the system will
 operate using a "First In, First Out" method and by deleting the oldest result when a
 new measurement is made.
- Results are automatically saved on the device until deleted using the Data app.
- Export individual results from the measurement app screen, or use the Data App to search for and export results later.

Software Updates

 Visit the DeNovix website to download the update package to a FAT32 formatted USB drive. Insert the USB drive and then select the Updater tab from the Settings App.

Technical Support

 Visit the DeNovix website for instructional videos, Technical Notes and troubleshooting tips. Contact your DeNovix Distributor or email techsupport@denovix.com for additional support.

8. Nucleic Acid App







The Helium Nucleic Acids App is used to measure common nucleic acid types. DNA, RNA and ssDNA can be selected from the dropdown menu to ensure the correct extinction coefficient is used in calculations.

Quick Protocol

- 1. Launch the desired application from the Application Menu.
- 2. Select the Nucleic Acid being measured from the dropdown menu.
- 3. Establish a Blank using the appropriate buffer.
- 4. Enter a sample name (optional).
- 5. Measure a fresh aliquot of sample using the **Measure** button.

Notes:

- Blank with TE if the sample is dissolved in TE.
- Ensure that the sample isolation procedure is optimized and that samples are purified prior to making absorbance measurements.
- Ensure all solutions are homogenous and well-mixed prior to sampling.
- Use fresh aliquots for each microvolume measurement.

dsDNA, RNA and ssDNA Measurements

The Nucleic Acid App is optimized for the measurement parameters used to calculate sample concentrations of dsDNA, RNA and ssDNA.

Experiment information

- Sample Type: The sample type dropdown menu is used to select the correct nucleic acid type. This determines which ng-cm/µL factor is used to calculate the concentration.
- Factor: The default factors used for each app are as follows:
 - dsDNA: 50 ng-cm/µL RNA: 40 ng-cm/µL ssDNA: 33 ng-cm/µL
- Concentration Results: Concentration results are reported in units of ng/µL.

- A260: Reports the baseline-corrected 260 nm absorbance value.
- 260/230: Displays the ratio of the 260 nm and 230 nm absorbance values.
- 260/280: Displays the ratio of the 260 nm and 280 nm absorbance values.

Note: Absorbance measurements at 260 nm do not distinguish between the nucleic acid types within a sample. It is important to optimize the isolation protocol to ensure accurate quantitation.

Nucleic Acid Concentration Calculations

A modified Beer-Lambert equation is used to calculate concentrations as follows:

$$c = (A * e)/b$$

c = the nucleic acid concentration in ng/microliter

A = the absorbance in AU

e = the wavelength-dependent factor in ng-cm/microliter

b= the pathlength in cm (10 mm)

Baseline Correction

Baseline correction is an important pre-processing technique used to separate true spectroscopic signals from background effects, stains, or traces of compounds. Helium automatically determines and applies a correction to every measurement. An optimized baseline absorbance value is determined and applied in the region of 350 nm.

Purity Ratios

- The generally accepted 260/280 values are ~1.8 for pure DNA and ~2.0 for RNA.
- The 260/230 values for nucleic acids usually range between 1.8 and 2.2.

Troubleshooting Purity Ratios

Issue	Probable Causes/ Solutions
	Dirty measurement surfaces or improper Blank. Clean the surfaces, measure a new Blank using fresh source of dH ₂ O or sample buffer. Use the same solution (water or buffer) the sample is in for the blank measurement.
Low 260/230	Presence of residual extraction reagent (ex. Carbohydrates, Chaotropic salts, phenol). Re-purify the sample, then remeasure. Contact the manufacturer of the extraction kit for guidance on how to optimize the procedure.
High 260/230	Dirty measurement surfaces or improper Blank. Clean the surfaces, measure a new Blank using fresh source of dH ₂ 0 or sample buffer. Use the same solution (water or buffer) the sample is in for the blank measurement.
Low 260/280	Dirty measurement surfaces or improper Blank. Clean the surfaces, measure a new Blank using fresh source of dH ₂ 0 or sample buffer. Use the same solution (water or buffer) the sample is in for the blank measurement.
LOW 200/280	Presence of residual extraction reagent (ex. protein, phenol). Re-purify the sample, then remeasure. Contact the manufacturer of the extraction kit for guidance on how to optimize the procedure.
High 260/230	Dirty measurement surfaces or improper Blank. Clean the surfaces, measure a new Blank using fresh source of dH ₂ O or sample buffer. Use the same solution (water or buffer) the sample is in for the blank measurement.

9. Protein App



Protein

Quick Protocol

- 1. Launch the Protein App application from the Application Menu.
- 2. Establish a **Blank** using the appropriate buffer.
- 3. Enter a sample name (optional).
- 4. Select a sample type. Additional user input is required for some options.
- 5. Measure a fresh aliquot of sample using the **Measure** button.

Notes:

- If the buffer exhibits significant absorbance at 280 nm, use alternative methods to quantitate proteins.
- Ensure all solutions are homogenous and well-mixed prior to sampling.
- Use fresh aliquots for each microvolume measurement.

Protein Measurements

The Protein application is used to quantify protein samples based upon absorbance values at 280 nm.

Experiment Information

 Sample Type: The sample type and associated mass extinction coefficients options include:

Sample Type	E1% at 280 nm for 10 mm pathlength	
BSA	6.67	
1A=1mg/mL	10	
IgG	13.7	
E1%	User-defined mass extinction coefficient (L gm ⁻¹ cm ⁻¹) for a 10 mg/mL (1%) solution	
MW & Ext Coeff	User-defined MW (Daltons) and molar extinction coefficient (M ⁻¹ cm ⁻¹)	

Use the 1A=1mg/mL option when neither the E1% nor Ext Coeff and MW for the purified protein sample are known.

- Concentration Results: Results are reported in units of mg/mL.
- A280: Reports the baseline-corrected 280 nm absorbance value.
- 260/280: Displays the ratio of the 260 and 280 nm absorbance values. An ideal ratio for common proteins is 0.6. Higher ratios may indicate the contamination of isolated proteins with DNA.

Baseline Correction

Baseline correction is an important pre-processing technique used to separate true spectroscopic signals from background effects, stains, or traces of compounds. Helium automatically determines and applies a correction to every measurement. An optimized baseline absorbance value is determined and applied in the region of 350 nm.

Protein and Concentration Calculations

The Beer-Lambert equation is used to calculate concentrations as follows:

$$A = E1\% * b * c$$

c = the protein concentration in g/100 mL

A = the absorbance in 10 mm equivalent

E1%= the percent extinction coefficient (L gm⁻¹ cm⁻¹) for a 10 mg/mL (1%) solution b= the pathlength in cm (10 mm)

The following equation is used to convert g/100 mL to mg/mL concentrations:

The relationship between percent extinction coefficient (E1%) and molar extinction coefficient (Ext. Coeff) is:

E1% = (Ext. Coeff *10) / molecular weight of protein (Daltons)

10. Data App



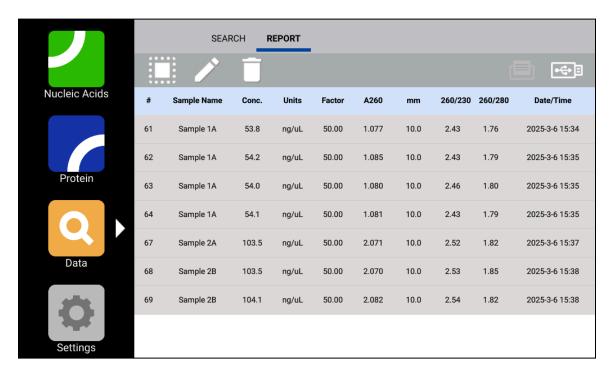
This app allows the user to search for past data by date or application. Data can then be added to the Report screen for review, export or deletion.

All results are automatically saved to Helium's onboard storage at the time of measurement. The device can store up to 2,000 results. Once the limit has been reached, the system will operate using a "First In, First Out" method by deleting the oldest result when a new measurement is made.

Search Options

- 1. Choose the type of data to review Nucleic Acids or Protein.
- 2. Filter by date if desired.
- Tap the Search button at the bottom of the screen. Tap any column header to sort the results list.
- 4. Highlight the sample(s) of interest and tap the **Add to Report** button to include the samples on the Report screen. Blank measurements may be included in the list but will not display any absorbance data.
- 5. Only data from the same app may be viewed together.
- To clear previous search results, return to the Search tab, choose new search criteria and touch Search.

Data App Report Screen



Exporting Data

- Once records have been added to the report from the Search tab, switch to the Report tab.
- 2. Insert a USB drive or connect a supported label printer.
- 3. Select the records to export.
- 4. Use the USB icon to export results to a USB drive. A pdf summary report and a .csv file for the results selected will be saved to the USB drive in a Helium directory. The exported file names include a time stamp to prevent accidentally overwriting a previous record.
- 5. Tap the Printer icon to export a table of results to a compatible label printer. See the DeNovix website for a list of supported printers.

Deleting Data

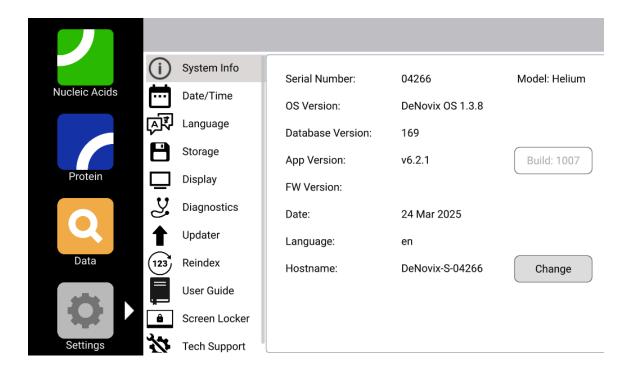
- The storage capability of Helium is sufficient for 2,000 samples. When the limit is reached, the software will automatically begin to replace the oldest data with new results.
- 2. To delete results, select the record(s) of interest and tap the delete icon.

WARNING – Records will be permanently deleted from the device!

11. Settings App



Settings



System Info

View software and firmware versions, language options and hostname.

Date/Time

Select time zone and adjust the date/time.

Language

Select the language of operation for the device. Not all languages are fully translated.

Sound

Play a test sound.

Storage

Displays current storage statistics.

Display

Includes controls for screen brightness and when the screen sleeps.

Diagnostics

Run Self Test and Verification tests on the device. See details in the following **Diagnostics** section of this guide.

Updater

To update system software and firmware to the most current available versions, updates must be downloaded from denovix.com to a FAT32 or exFAT formatted USB drive. Download the update file to the root directly of the USB drive. Do not change the file name or unzip the file.

Insert the USB drive in the device. Select the Settings app Updater tab. When the update file is recognized on the USB drive, the USB update button will be active. Touch the USB button to start the update.

Note: The update process takes three to ten minutes and may require one or more automatic system reboots.

Reindex

Reindex will reset the sample numbers displayed on the bottom of the measurement applications. Helium stores up to 2,000 results. If results have been deleted, reindexing will sequentially renumber all remaining results starting with the number one.

User Guide

View the User Guide document.

Screen Locker

This function allows users to temporarily lock the screen for ease of cleaning.

Tech Support

Export a DeNovix Tech Support utility file to a USB drive.

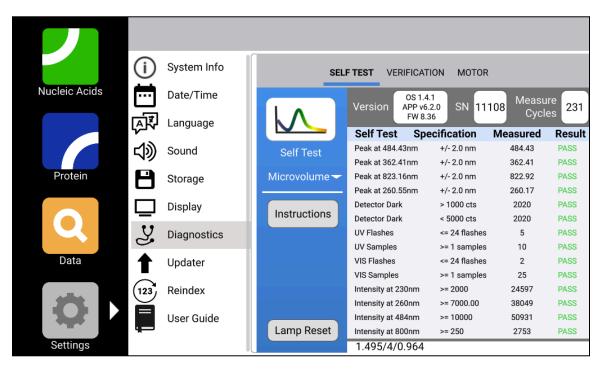
12. Diagnostics



Although the Helium Spectrophotometer proprietary SmartPath™ technology enables accurate microvolume pathlength control and eliminates the need for routine recalibration, labs may choose to run the Diagnostics app to verify that the instrument is working within specifications.

Helium Self Test

- Ensure that both the upper and lower microvolume measurement surfaces are clean prior to starting the Self Test microvolume mode assessment.
- 2. Tap "Instructions" and follow the steps provided.
- 3. A pass/fail result will be provided once the test has completed.



Lamp Reset

The Lamp Rest function should only be used when the device will not pass the Diagnostics Self Test function. Carefully follow **all** on-screen instructions to execute the lamp reset.

Microvolume Pathlength Verification

The DeNovix LC-NA verification fluid is supplied as single use vials composed of a non-toxic solution of aqueous nicotinic acid. The solution is used to verify that the microvolume pathlengths are within specifications.

Procedure

- 1. Ensure both sample measurement surfaces are clean. Refer to the general cleaning instructions in the chapter entitled "Maintenance" for more information.
- 2. From the **Diagnostics** tab, select the **Self Test** tab. Ensure the Self Test passes prior to proceeding.
- 3. With all Self Test results reporting PASS, select the **Verification** tab.
- 4. Enter the lot specific Target Absorbance stated on the LC-NA ampule in the appropriate field.
- 5. Add 1 μ L of dH₂O to the pedestal, lower the arm and click **Blank**.
- 6. Wipe the upper and lower pedestals using a dry, lint-free laboratory wipe.
- 7. Vigorously shake the nicotinic acid ampule to thoroughly mix the solution. Ensure all of the solution is in the bottom portion of the ampule before opening.
- 8. Use a lab wipe as protection from sharp edges when snapping off the top of the LC-NA ampule to open it.
- 9. Pipette 1 µL of the solution onto the sample surface, lower the arm and click **Measure**.
- 10. Wipe the sample off both the top and bottom sample surfaces.
- 11. Repeat steps 9 and 10 for a total of 5 measurements using fresh aliquots for each measurement.

LC-NA Single Use Ampules

- The LC-NA solution is supplied in single use ampules that should be used immediately upon opening. Significant changes in concentration and possible verification check failures may occur if the ampule is opened for longer than one hour prior to use.
- Store unopened ampules in a dark, room temperature environment.
- Always use a fresh vial for each verification check procedure.
- Do not use parafilm or other methods of sealing an ampule as evaporation may still occur and result in verification test result errors.
- Note: Only the LC-NA standard available from DeNovix and its authorized distributors should be used for the verification check. No other sources of nicotinic acid are tested and validated for use with the Helium verification procedure.

Results

A Pass or Fail message will appear in the results box at the bottom of the screen. If the instrument fails, please clean the surfaces and repeat the test with a fresh aliquot of LC-NA.

Verification Check Troubleshooting

Issue	Probable Causes/ Solutions
Low Absorbance Values	Dirty measurement surfaces or improper Blank. Clean the surfaces, measure a new Blank using fresh source of dH_20 and restart the procedure using a new ampule of LC-NA.
	Multiple measurements made using the same aliquot. Use a fresh aliquot for each measurement.
High	Pipette tips not changed between aliquots. Use a fresh tip to pipette each aliquot onto the sample surface.
Absorbance Values	The sample surfaces are not adequately cleaned between measurements. Wipe both the top and bottom surfaces with a dry lab wipe.
	The solution has concentrated due to prolonged exposure. Use a fresh ampule of solution.
	Multiple measurements made using a single 1 ul aliquot. Use a fresh aliquot for each measurement.
High Standard	Pipette tips not changed between aliquots. Use a fresh tip to pipette each aliquot onto the sample surface.
Deviations	The sample surfaces are not adequately cleaned between measurements. Clean both the top and bottom surfaces between measurements.

13. Maintenance

Cleaning

	1. Pipette 2 μ L of dH $_2$ 0 onto the bottom sample surface. Lower the upper arm. Wait 1 - 2 minutes.
Routine	2. Wipe away the water from both the upper and lower sample surfaces with a dry, lint-free lab wipe.
Between Measurements	Use a dry, lint-free lab wipe to remove samples from both the lower and upper sample surfaces.
	Use the following procedure to remove sticky solutions (e.g. proteins) that may have dried down on the sample surfaces.
Additional	1. Pipette 2 μL of 0.5 M HCl onto the bottom sample surface. Lower the upper arm. Wait 2 - 3 minutes.
Cleaning	2. Wipe away the HCl from both the upper and lower sample surfaces with a dry, lint-free lab wipe.
	3. Repeat steps 1 - 2 with 2 μ L of dH ₂ 0.

Guidelines

- 1. Use only a dry, soft, lint-free cloth to clean the front screen.
- 2. Do not use a spray bottle to apply water or any other solutions onto any surface of the instrument as the liquid may damage internal components.
- 3. The use of detergents or isopropyl alcohol as cleaning solvents is not recommended as they may temporarily alter the hydrophobic nature of the sample surfaces.

Solvent Compatibility

The Helium Spectrophotometer microvolume sample surfaces are compatible with most solvents typically used in life science laboratories.

Exception: Do not use Hydrofluoric Acid (HF) as the fluoride ion will dissolve the quartz fiber optic cable.

14. Troubleshooting

Quick Help

Most issues concerning accuracy, reproducibility, negative results and low nucleic acid purity ratios are sample or technique related and are resolved by following the suggestions below:

- 1. Establish a new Blank using the appropriate buffer.
- 2. Ensure that the sample isolation procedure is optimized and that samples are purified when required prior to making absorbance measurements.
- 3. Ensure all solutions are homogenous and well-mixed prior to sampling.
- 4. Ensure sample concentrations fall within the absorbance limits of the instrument.

Microvolume Measurements

- 1. Ensure both top and bottom microvolume measurement surfaces are clean prior to making the Blank measurement.
- 2. Always remove sample solutions from both the top and bottom measurement surfaces using dry, lint-free wipe immediately after each measurement is complete.
- 3. Use calibrated pipets and properly fitting tips to ensure a full 1 μ L aliquot is delivered to the sample measurement surface. Protein samples sometimes wick up on the outside of the tip and may not be properly dispensed.
- 4. Use a clean pipette tip and a new aliquot of sample for each microvolume measurement.

Software Errors

Pop-up messages will provide guidance for software-based errors or invalid user actions. In most cases, simply relaunching the app or restarting the instrument will allow the user to continue to make measurements. Update the software via the Settings app if experiencing a reoccurring software error.

Printer and Barcode Scanner

Helium is compatible with selected label printers and Opticon OPI 3601 USB barcode scanners. For accessory product troubleshooting and warranty information, please contact the manufacturer of the product. DeNovix support is limited to the specific use of the product with a Helium instrument.

Please refer to the DeNovix website for a list of supported label printers.

15. Customer Support

DeNovix Inc. 3411 Silverside Road, Hanby Building Wilmington, DE USA 19810 302-442-6911 techsupport@denovix.com

Please include the serial number of your instrument and a data export (.csv) file of any relevant data when contacting Customer Support by email.

Outside of the US, please contact your local distributor for assistance.

Warranty

All product(s) and accessories sold by DeNovix Inc. are under warranty against manufacturing defects in parts.

Disclaimer

All information in this document is for reference purposes only. DeNovix Inc. makes no claims that this document is complete or error-free and assumes no responsibility and will not be liable for any errors, omissions, damage or loss that might result from any use of this document. The information in this document is updated frequently and may be updated at any time. Visit denovix.com to obtain the latest version.

Instructions for Disposal of Packaging and Waste Equipment

DeNovix encourages customers to locally recycle shipping and packaging materials. Corrugated cardboard and paper can be recycled wherever it is accepted locally (e.g., curbside bins, recycling facilities, etc.). For foam and plastics, please contact your local recycling office for information on how to recycle.

When an instrument has reached its end of life, DeNovix recommends that customers responsibly dispose of the equipment in line with local regulations. Material construction is available upon request to assist in this process. Visit denovix.com/sustainability for more information.

Research Use Only

DeNovix Inc. products are for research use only.