



Cary 5000 UV-Vis-NIR Operating Instructions

Purpose

The Cary 5000 is a UV-Vis-NIR spectrophotometer. It shines selected wavelengths of ultraviolet (UV), visible (Vis), and near-infrared (NIR) light through or onto a sample and measures how much light the sample absorbs or transmits. The supplied facility procedure documents a 200–3300 nm spectral range and use with solid and liquid samples. This makes the instrument useful for collecting spectra that show how a material interacts with light across wavelengths.

This document provides operator instructions for the Cary 5000 UV-Vis-NIR spectrophotometer.

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1. Before You Begin

1. Inspect the sample compartment before power-on. READY → The compartment is clean and clear; there is no spilled liquid, loose sample material, or object blocking the holder/optical area. The sliding lid closes normally without force. NOT READY → STOP. Do not power on. Do not touch the optics. Contact Nanofab staff for spills, contamination, damage, or anything you are unsure how to handle.
2. Identify the installed holder/accessory and match it to the measurement.

Standard solid/film transmission or absorbance

→ Use the solid-sample holder / aperture setup.

Visual reference — solid/film holder on this Cary 5000:

Standard liquid transmission or absorbance

→ Use the cuvette / cell holder with a compatible non-frosted cuvette.

See the sample-loading images and cuvette/cell-holder definition later in this SOP.

UMA measurement

→ The UMA must be physically installed and the UMA method/settings must be used.

Visual reference — Cary Universal Measurement Accessory:

DRA reflectance measurement

→ The DRA must be physically installed and the DRA reflectance settings must be used.

Visual reference — Cary 5000 fitted with a DRA-2500 diffuse-reflectance accessory:

DOES NOT MATCH / UNSURE → STOP. Do not start the measurement or change hardware by guessing.

3. If an accessory is installed, inspect its required cable connection. READY → The required cable is fully seated and undamaged. NOT READY → If the cable is loose, disconnected, damaged, or you cannot identify where it belongs, STOP. The Cary 5000 must be OFF before any accessory connection is made or changed.

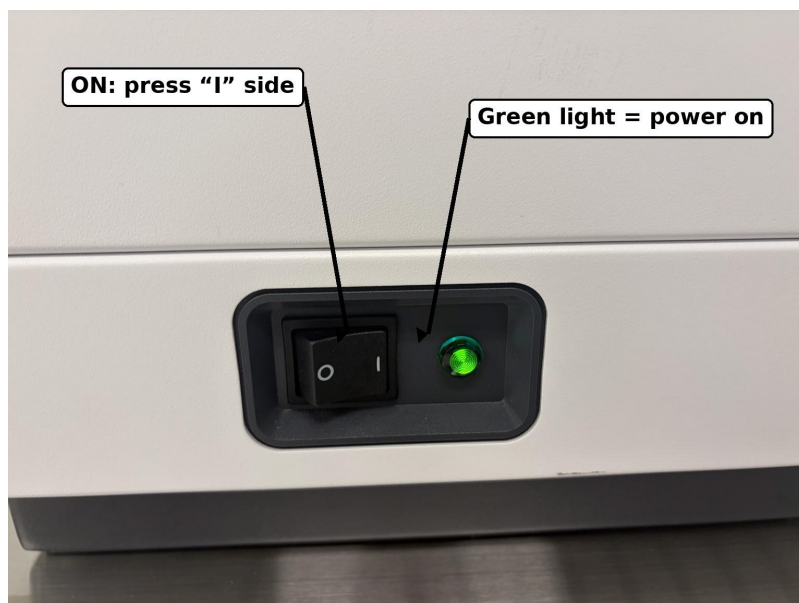
4. Final check before startup.

CONTINUE → You can identify the sample type, installed holder/accessory, and mounting method from this SOP.

STOP → If any of these are unfamiliar or unclear, do not improvise a mount or begin a measurement.

2. Instrument Startup

1. Press the “I” side of the rocker switch on the lower front-left of the Cary 5000.
2. Look at the green power indicator. GREEN LIGHT ON → continue. GREEN LIGHT OFF → STOP; do not continue startup and contact Nanofab staff.



3. Allow at least 20 minutes of warm-up before routine measurements. The supplied Nanofab Cary manual specifies at least 20 minutes.
4. At the instrument computer, log in and wait for the desktop to load normally. If the computer or login does not work normally, STOP and contact Nanofab staff.
5. On the instrument computer, open the Cary WinUV program/application needed for the measurement. Wait for it to finish opening before selecting a method or starting a measurement. If the software displays an error or does not open normally, STOP and contact Nanofab staff.
6. Before continuing, there should be no repeated software error, abnormal grinding/screeching noise, visible leak/damage, or loose/misaligned holder/accessory. If any of these occurs, STOP and contact Nanofab staff.



3. Instrument Performance Check (Validate)

1. Before running Validate, check the local instrument status or session instructions. If they tell you to run Validate, continue with this section. If they do not tell you whether Validate is needed, ask Nanofab staff before starting a Validate test. Do not invent a test schedule.
2. In Cary WinUV, open Validate.
3. Select Tests → Instrument Performance Tests.
4. Select Start to begin the Validate run.
 - 4 Read the prompt before touching the sample compartment. Validate may run some tests automatically, while other tests may display a Cell Loading Guide or another dialog asking you to load a specific validation solution, standard, or filter.
 - 4 If the screen asks you to load an item, open the sample compartment and place exactly the validation item named by the software in exactly the position shown. Close the sample compartment before selecting OK.
 - 4 If the screen asks you to remove or change a filter/standard, wait for that prompt, open the sample compartment, remove the item currently being used, and replace it only with the item and position shown by the Cell Loading Guide. Close the compartment, then select OK. Do not guess which filter is next and do not substitute a different filter, solution, sample, or position.
 - 4 Continue following the prompts until the selected performance tests are complete. Select Cancel if the run must be stopped.
5. Wait until the Validate test run is complete.
6. Look at the result shown for EACH test in the Validate report.
 - PASS → The test passed. Check the next test.
 - FAIL → STOP. Do not measure samples. Save/record the result and contact Nanofab staff.
7. Use this checklist when reviewing the completed Validate report. Check each row that actually appears in the report; the exact test set can depend on the selected Validate suite.

VALIDATE REPORT CHECKLIST

- Wavelength accuracy / wavelength performance — PASS
- Wavelength reproducibility — PASS, if listed separately
- Baseline flatness — PASS
- Photometric noise — PASS
- Photometric accuracy — PASS
- Photometric linearity — PASS, if listed separately
- Stray light — PASS
- Spectral resolution — PASS
- Any additional test shown in the report — PASS

EVERY ROW SHOWS PASS → continue to Step 4 — Measurement Setup. ANY ROW SHOWS FAIL → STOP. Do not measure samples. Save/record the result and contact Nanofab staff.

IMPORTANT: This checklist is a review aid, not a command to manually add tests. Follow the test suite shown by Validate. The supplied Nanofab Cary manual does not establish the exact required Validate suite or whether every listed check is required every session.

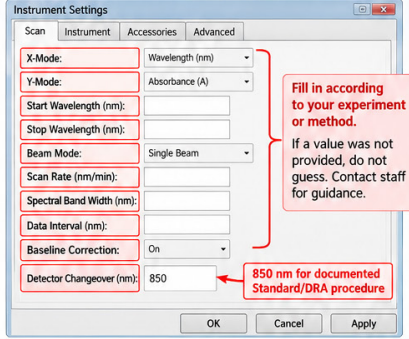
4. Measurement SetupSoftware workflow visual — illustrative guide; use the written settings in this SOP, not example values shown in the graphic.

Cary WinUV – Software Workflow Visual Guide

ILLUSTRATIVE / RECREATED — NOT AN ACTUAL FACILITY SCREENSHOT
The layout and appearance may differ from the software installed at your facility.

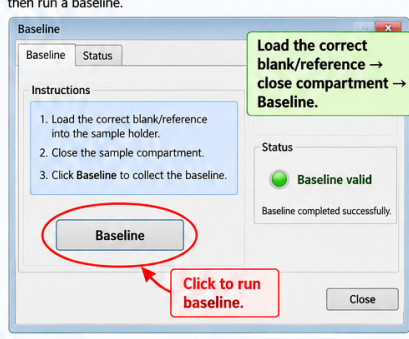
1 Instrument Settings / Setup

Enter your measurement parameters in the Instrument Settings window.



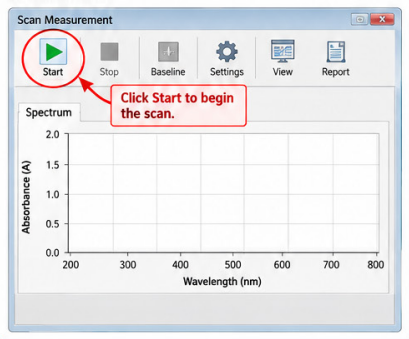
2 Baseline Workflow

Load the correct blank/reference, close the compartment, then run a baseline.



3 Start Measurement (Scan)

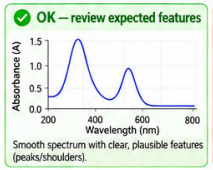
After setup and baseline/control are complete, click Start to begin the measurement.



4 Spectrum Quality Examples

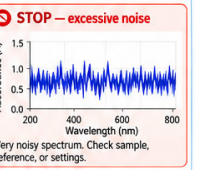
Check the spectrum after the scan completes. Here are examples of common results:

OK — review expected features



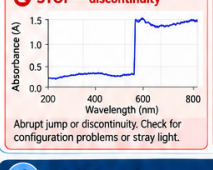
Smooth spectrum with clear, plausible features (peaks/shoulders).

STOP — excessive noise



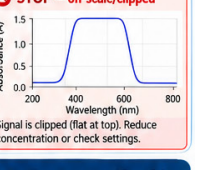
Very noisy spectrum. Check sample, reference, or settings.

STOP — discontinuity



Abrupt jump or discontinuity. Check for configuration problems or stray light.

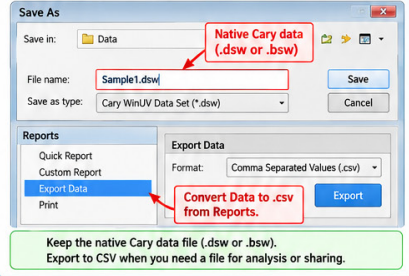
STOP — off-scale/clipped



Signal is clipped (flat at top). Reduce concentration or check settings.

5 Save / Export Data

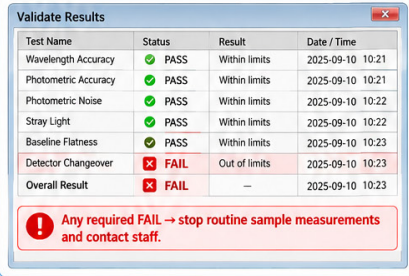
Save your data in the native Cary format and export a CSV file when needed.



Keep the native Cary data file (.dsw or .bsw).
Export to CSV when you need a file for analysis or sharing.

6 Validate (Instrument Performance Check)

Run the Validate tests as required by the facility. All required tests must PASS.



Test Name	Status	Result	Date / Time
Wavelength Accuracy	PASS	Within limits	2025-09-10 10:21
Photometric Accuracy	PASS	Within limits	2025-09-10 10:21
Photometric Noise	PASS	Within limits	2025-09-10 10:22
Stray Light	PASS	Within limits	2025-09-10 10:22
Baseline Flatness	PASS	Within limits	2025-09-10 10:23
Detector Changeover	FAIL	Out of limits	2025-09-10 10:23
Overall Result	FAIL	—	2025-09-10 10:23

Any required FAIL → stop routine sample measurements and contact staff.



⚠ ILLUSTRATIVE / RECREATED CARY WINUV BUTTON GUIDE — VISUAL REFERENCE ONLY

This is not a screenshot of the facility computer. Use it only to locate controls. Displayed values and prompts are examples, not UArizona/Nanofab-authoritative settings. Follow the written facility-derived settings below.

What this section means: the supplied Nanofab Cary manual documents three configurations. First choose the experiment type. Then use **ONLY** the settings for that configuration. “Documented standard method” below means the STANDARD NO-UMA configuration written in the supplied Nanofab Cary 5000 manual; it does NOT mean one universal Agilent method for every experiment.

1. CHOOSE THE EXPERIMENT TYPE

A. STANDARD TRANSMISSION / ABSORBANCE — NO UMA

Use for ordinary liquid measurements in cuvettes and supported solid/film transmission or absorbance.

Open: noUMA_BaseMethod.

B. UMA EXPERIMENT

Use only when the UMA hardware is installed for the documented UMA experiment.

Open: UMA_BaseMethod.

C. DRA DIFFUSE-REFLECTANCE EXPERIMENT

Use when the DRA hardware is installed and the measurement is %R.

Open: noUMA_BaseMethod, then use the DRA-specific settings below.

2. Open Cary WinUV → Setup → Summary → Instrument Settings. You should see the Instrument Settings fields. If this window does not open, STOP rather than changing unrelated controls.

3. SET EACH FIELD FOR YOUR EXPERIMENT



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X-Mode — horizontal-axis unit

STANDARD: If your instructions give wavelengths in nm, select Nanometers. Use Wavenumber or Angstroms only when your experiment specifically asks for that unit.

DRA: If your instructions give wavelengths in nm, select Nanometers. Use another X-Mode only when your experiment specifically asks for it.

UMA: The supplied UMA section does not give a separate X-Mode instruction.

WHEN TO CHANGE IT: Change X-Mode when the experiment requires a different horizontal-axis unit. If the experiment is described in wavelength and no other unit is specified, use Nanometers (nm).

Y-Mode — what optical response is measured/plotted

STANDARD: Absorbance for an absorbance experiment; %Transmission for a transmission experiment.

DRA: %R.

UMA: Use the Y-Mode already specified for your UMA experiment/method. If no Y-Mode has been specified, STOP and ask before scanning.

WHEN TO CHANGE IT: Change Y-Mode when switching between absorbance, transmission, and DRA reflectance. If you do not know which measurement is intended, STOP before scanning.

Starting Range — wavelength interval scanned

STANDARD: Variable. The facility manual gives 800–250 nm only as an EXAMPLE.

DRA: Variable; the documented DRA procedure gives a maximum range of 900–200 nm.

UMA: Enter the wavelength range specified for your UMA experiment. If no range has been specified, STOP and ask before scanning.

WHEN TO CHANGE IT: Change this field to the wavelength range required by the experiment. Do not automatically copy 800–250 nm.

Beam Mode

STANDARD: Double.

DRA: Double.

UMA: The supplied UMA section does not give a separate Beam Mode instruction.

WHAT “DOUBLE” MEANS: The Cary uses both optical beam paths—the sample path and the reference path—rather than operating as a single-beam measurement.

WHEN TO CHANGE IT: For the documented STANDARD or DRA procedure, it should read Double. If it does not, set Double.



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DRA: 0.5–1 nm.

WHEN TO CHANGE IT: Change it when the DRA experiment requires the documented 0.5–1 nm interval.

Baseline Correction

STANDARD: On.

DRA: On.

UMA: Zero/Baseline Correction On.

WHEN TO CHANGE IT: For these documented procedures, correction should be On. If it is Off, turn it On.

UV-Vis Detector Changeover Wavelength

STANDARD: 850 nm — IMPORTANT: keep at 850 nm.

DRA: 850 nm — IMPORTANT: keep at 850 nm.

UMA: Leave the detector-changeover field at the value already stored in UMA_BaseMethod. Do not type a new value.

WHEN TO CHANGE IT: Do NOT choose this based on the sample. For STANDARD/DRA, if it is not 850 nm, set 850 nm. For UMA, keep the preset value in the UMA method.

UMA-ONLY SETTINGS

Positive/Negative Angle Measurement: Off in the supplied UMA procedure.

Polarizer: Use the setting specified for your UMA experiment. If no polarizer setting has been provided, do not guess; ask before scanning.

Measurement Angles: Enter the angle(s) listed in your UMA experiment instructions. If no angle has been provided, STOP and ask before scanning.

Do not change these for a standard or DRA run merely because they appear in the software.

4. QUICK CONFIGURATION CHECK

STANDARD NO-UMA

noUMA_BaseMethod | X/Y/range = experiment-dependent | Double | Reduced | ~600 nm/min |

Use UMA Off | SBW 4.0 nm | Data Interval 1 nm | Baseline Correction On | Detector

Changeover 850 nm.



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UMA

UMA_BaseMethod | Use UMA On | SBW 4.0 nm | NIR Data Interval 1 nm | Zero/Baseline Correction On | Positive/Negative Angle Measurement Off | Polarizer/angles as required | detector changeover stays at preset value.

DRA

noUMA_BaseMethod | Y-Mode %R | range max 900–200 nm | Double | Reduced | scan rate <600 nm/min | Use UMA Off | SBW 2.0 nm | Data Interval 0.5–1 nm | Baseline Correction On | Detector Changeover 850 nm.

5. Reports → select Convert Data to .csv if you need a CSV file for Excel or other data-analysis software. The native Cary file is saved separately in Section 8.
6. Re-read ONLY the configuration for your experiment. If every fixed field matches and all experiment-dependent values are known, select OK. If a required value is unknown, STOP rather than guessing.

5. Load the Sample and Reference

1. Identify the holder BEFORE loading anything.

Solid/film → solid-sample/aperture holder.

Liquid → cuvette/cell holder.

UMA → UMA setup.

DRA → DRA setup.

CANNOT IDENTIFY HOLDER → STOP. Do not load the sample.

2. If the correct holder is already installed → go to Step 3.

If the holder must be changed:

- a. Identify the holder from Step 1 above: solid/film → solid-sample/aperture holder; liquid → cuvette/cell holder; UMA → UMA setup; DRA → DRA setup. If you still cannot identify it, STOP.

- b. Unlock the stage using the front lever.
- c. Remove the front panel.
- d. Lift the current holder straight up and out.
- e. Align the replacement holder's three holes with the three mounting pegs.
- f. Lower it into position without force.
- g. Relock the stage.
- h. Replace the front panel.

3. Prepare the sample and required reference/blank. Liquid →
use a chemically compatible, non-frosted cuvette.

Film → use the documented aperture-plate setup.

REFERENCE/BLANK NOT IDENTIFIED → STOP. For a liquid measurement, use the solvent/blank specified by the method. For DRA, use the specified reflective standard. For other cases, do not invent a reference.

4. Place the SAMPLE in the FRONT position.

5. For the documented standard double-beam setup, place the required REFERENCE/BLANK in the BACK position. If your method does not call for a reference, do not add one just because the holder is empty.

6. Before closing the lid, check:

- ☐ Sample/reference seated securely
- ☐ Nothing tilted or loose
- ☐ No leaking liquid
- ☐ Nothing can fall into the optical area

ANY PROBLEM → remove/correct the setup before continuing.



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7. Close the sample-compartment lid.

CLOSES NORMALLY → continue.

DOES NOT CLOSE → STOP. Do not force it. Recheck the setup.

6. Run the Baseline

1. Use the baseline/control procedure for the configuration selected in Section 4. STANDARD NO-UMA → Load the blank/reference configuration required by the method, then select Baseline in Cary WinUV. UMA → The supplied facility procedure calls for a control experiment at the angle(s) of interest before inserting the sample, followed by a second experiment with the sample and manual subtraction of the control data. If you have not been trained on this UMA control/subtraction workflow, STOP and ask Nanofab staff. DRA → Use the appropriate approximately 100% reflective standard for the control/baseline. If the required reflective standard is not identified for your session, STOP and ask Nanofab staff.

2. For a STANDARD baseline, read the Cary WinUV prompt before selecting OK. Compare the requested configuration with what is physically loaded.

MATCH → close the compartment and select OK.

DOES NOT MATCH / PROMPT IS UNCLEAR → STOP. Do not select OK.

3. Wait for the baseline/control scan to finish. Do not open the compartment or start the sample measurement while it is running.

4. Inspect the completed baseline/control data. Look for unexplained large drift, abrupt jumps/breaks, excessive jagged noise, or clipped/off-scale regions.

NONE OF THESE PROBLEMS → continue to Section 7.

ANY PROBLEM PRESENT → STOP. Do not measure/use the sample result until the cause is resolved.

7. Run and Review the Measurement

1. BEFORE selecting Start, confirm ALL of the following:

- Correct method/settings displayed
- Required baseline/control measurement finished
- Sample is in FRONT position
- Reference/blank is in BACK position, if required
- Sample-compartment lid is completely closed

ALL CHECKED → continue.

ANY ITEM NOT CHECKED → STOP and correct it.

2. Select Start when it becomes available. START NOT AVAILABLE → do not click other controls at random. Recheck the selected configuration, required baseline/control measurement, and setup. If the cause is unclear, contact Nanofab staff.

3. When prompted to save:

a. Select the intended folder for your Nanofab session.

4. While the scan is running, watch for:

- Software errors
- Abnormal mechanical noise
- Leakage
- Sample movement

ANY OCCURS → STOP the run and contact Nanofab staff.

5. When the scan finishes, inspect the spectrum.

Look for:

- Excessive jagged noise
- Sudden jumps/breaks
- Clipped/off-scale regions
- Features also present in the baseline/reference

NONE PRESENT → continue.

ANY PRESENT → do not assume the spectrum is valid. See the Step 7 explanation before using the data.

6. After the scan is finished AND the spectrum has been inspected, select Finish.

8. Save and Export Data

1. If CSV export was selected, open the save location.

.csv FILE PRESENT → ✓

.csv FILE NOT PRESENT → STOP. Return to Cary WinUV and save/export it before closing the software.

Save the native Cary file using File → Save As. The supplied procedure identifies .dsw and .bsw as Cary save options.

3. Open the save location and confirm the native Cary file is present.

FILE PRESENT → ✓

FILE MISSING → save it before continuing.

4. Do not delete the native Cary file just because a CSV was created.

5. Use the file name and storage location specified for your Nanofab session.

If no naming/storage instruction has been provided, use a clear descriptive filename and save to the session location you are authorized to use; do not overwrite another user's data.

9. Shutdown

1. Remove the sample and reference.

Look inside the sample compartment:

- Sample removed
- Reference/blank removed
- Cuvette removed
- No loose material
- No spilled liquid



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SPILL/MATERIAL PRESENT → STOP. Do not wipe or clean optical components yourself. Contact Nanofab staff for the correct cleanup method.

Confirm your required files have been saved. If a save prompt appears, do not discard data unless it is intentionally unwanted.

3. Close Cary WinUV.

4. Turn OFF the Cary 5000 using the main rocker switch.

Check the green power indicator.

GREEN LIGHT OFF → ✓

GREEN LIGHT STAYS ON / abnormal shutdown → contact Nanofab staff.

5. Complete the instrument log book as required by the supplied facility shutdown procedure. Record any problem or failed run that occurred.

6. Log off the instrument as required by the supplied facility shutdown procedure.

7. Before leaving, confirm:

- Sample/reference removed
- Sample compartment left clean
- Required files saved
- ☐ Cary WinUV closed
- ☐ Cary 5000 powered off
- ☐ Instrument log completed
- ☐ Instrument logged off

ALL CHECKED → shutdown complete.

10. Explanations by Step

Step 1 — Before You Begin

What is the sample compartment?

The sample compartment is the covered measurement area in the center/top of the Cary 5000. It is where the light beam interacts with the sample. Open the sliding top lid to access it. In the standard double-beam setup, the front position is used for the sample and the rear position is used for the reference when a reference is required.

What does “installed holder/accessory” mean?

A holder is the hardware inside the sample compartment that physically supports the sample in the light path. For example, the solid-sample holder/aperture setup holds a film or other supported solid, while a cell/cuvette holder holds a liquid-filled cuvette. An accessory is additional measurement hardware used when the standard holder arrangement cannot perform the required measurement. The facility procedure documents the UMA and DRA configurations.

How do I know whether the installed setup matches the measurement?

Start with what you are trying to measure:

- Light passing THROUGH a supported solid or film (transmission/absorbance) → use the standard solid-sample holder/aperture setup.
- Light passing THROUGH a liquid (transmission/absorbance) → use the standard cuvette/cell holder.
- The facility UMA procedure, including its documented opaque/external diffuse-reflectance or kinetic configuration → use the UMA and UMA_BaseMethod.
- The facility DRA reflectance procedure → use the DRA and its documented %R settings.

If the hardware in the compartment does not match the method you intend to run, do not start the measurement.

What does “clean, clear, and ready” mean?

- The Cary 5000 exterior and sample compartment look intact rather than cracked, broken, or visibly damaged.
- The sample compartment is clear and clean; there is no spilled liquid, loose sample material, or object blocking the optical/sample area.
- The holder/accessory sits securely in its intended mounting position rather than being loose, tilted, or obviously misaligned.
- The sliding sample-compartment door can close normally.
- Required accessory cables are connected, and those connections were made while the instrument was OFF.
- There is no obvious evidence of leakage or contamination.

After startup, normal operation also means the green power indicator illuminates and the instrument/software initializes without repeated errors or abnormal mechanical noise.

DO NOT plug/unplug, install, or remove an accessory while the instrument is powered on. DO NOT lift/mount the UMA alone; the supplied procedure directs users to obtain assistance for mounting it.

Step 2 — Instrument Startup

Instrument computer used to open Cary WinUV:



Step 2.3 (“Allow at least 20 minutes of warm-up”) means the Cary 5000 should remain powered on and idle for at least 20 minutes before routine sample measurements. Do not turn it on and immediately run a sample. This warm-up period allows the instrument/light sources to stabilize before data collection. The supplied Nanofab Cary manual establishes a minimum 20-minute warm-up; any different performance-test warm-up requirement must come from the applicable performance-test procedure.

The green indicator confirms that the instrument is receiving power. Examples of an error/abnormal condition include a failed initialization, repeated software error, unexpected grinding/screeching or other abnormal mechanical noise, failure of the green power indicator, visible damage or contamination, a holder/accessory that will not seat normally, or another condition that prevents normal measurement.

Step 3 — Instrument Verification / Validate

SOURCE NOTE — Validate details in this SOP are supplemental Cary WinUV/Agilent guidance, not requirements established by the supplied Nanofab Cary manual. Nanofab must confirm when Validate is required and which test suite/standards are approved. Validate is the Cary WinUV application used to run instrument performance tests. These tests check the instrument itself; they are not measurements of your sample.

Validate is an instrument-performance check, not a sample measurement. Run it when your local instrument status/session instructions call for it. If the Cary is behaving abnormally or a previous performance test failed, stop routine measurements and contact Nanofab staff rather than deciding on your own that the instrument is acceptable.

An instrument performance test compares a measured instrument value with an acceptance specification. Depending on the selected Validate test set, checks can include wavelength accuracy/reproducibility, baseline flatness, photometric noise, photometric accuracy/linearity, stray light, and spectral resolution.



What happens during a Validate run?

After Start is selected, Validate controls the sequence of the selected performance tests. For a manual run, tests that need you to physically change something are normally arranged early in the run. When the software needs your help, it displays a dialog telling you what to load or change. A Cell Loading Guide, when shown, identifies the specified validation solutions and/or filters and where they must be placed. Load exactly what the screen shows, close the compartment, and select OK. When no physical change is required, the test can proceed automatically. Results appear in the Report area as tests finish. The software will indicate when your intervention is no longer required.

Important: the supplied facility Cary procedure does not identify the exact validation standards/filters that this Nanofab keeps for every Validate test. Therefore, the SOP should not invent a universal filter or solution; use the exact item identified by the installed Validate method/on-screen loading guide.

How to read a Validate result:

1. Find the test name.
2. Find the measured/result value. This is what the Cary actually measured.
3. Find the specification, limit, or allowed range. This is the criterion the measured value must satisfy.
4. Read the reported status:
 - PASS = the measured value meets the test's acceptance criterion.
 - FAIL = the measured value does not meet the acceptance criterion.
5. Do not judge a failed result by whether the graph "looks okay." The acceptance criterion controls the result.
6. If ANY required test says FAIL, do not run routine samples. Record/save the result and contact Nanofab staff.

The operator does not decide whether a measured value is "close enough." Use the PASS/FAIL status reported by Validate.

Step 4 — Measurement Setup

Front accessory connection panel (visual reference):



What is a method?

A method is a saved Cary WinUV setup containing instrument settings. The supplied facility procedure provides noUMA_BaseMethod for the standard/no-UMA configuration and UMA_BaseMethod when the UMA is installed.

When do I use the standard holder, UMA, or DRA?

The three configurations serve different measurement geometries. The standard holder is for the documented transmission/absorbance measurements of liquids and supported solids/films. The UMA is an alternate configuration documented for external diffuse reflectance of opaque substances and kinetic studies. The DRA is the documented diffuse-reflectance configuration reported as %R. The operating choices and exact settings are kept in Section 4 above rather than repeated here.

What “without the UMA” means

“Without the UMA” means the Cary 5000 is being used in its normal/standard sample-compartment configuration rather than with the Universal Measurement Accessory installed. For the documented standard solid/film or liquid transmission/absorbance measurements, open noUMA_BaseMethod and keep Use UMA set to Off. If the UMA hardware is installed for an UMA experiment, use the UMA procedure and UMA_BaseMethod instead.

Step 4.3 — What each Instrument Setting means

- X-Mode — Chooses the horizontal-axis quantity/unit for the scan. The facility procedure lists Nanometers, Wavenumber, Angstroms, etc.; nm is the common wavelength display.
- Y-Mode — Chooses what optical response is plotted on the vertical axis. Use Absorbance when the experiment is intended to show light attenuation/absorption; use %Transmission when the experiment is intended to show the percentage of light passing through the sample. DRA reflectance is a separate documented configuration using %R.
- Starting Range — Sets the wavelength interval to scan. The facility procedure gives 800–250 nm only as an example; choose the range required by the approved experiment rather than treating that example as universal.
- Beam Mode: Double — Uses the Cary's sample and reference optical paths during the measurement.
- Slit Height: Reduced — Uses the documented reduced slit-height setting for the standard method. Keep the supplied method setting unless an approved method requires otherwise.
- Scan Rate — Controls how quickly the instrument moves through the wavelength range. The supplied standard procedure documents approximately 600 nm/min.
- Use UMA: Off — Tells the software that the Universal Measurement Accessory is not being used. This must match the physical hardware configuration.
- Spectral Band Width (SBW): 4.0 nm — Controls the wavelength band passed by the monochromator at one time. The supplied standard method documents 4.0 nm.
- Data Interval: 1 nm — Controls the spacing between recorded wavelength data points; at 1 nm, the software records a data point at 1 nm intervals.
- Baseline Correction: On — Applies the measured instrument/reference background correction to the sample scan.
- UV-Vis Detector Changeover Wavelength: 850 nm — Sets the wavelength at which the instrument changes detector regions. Leave the documented standard setting at 850 nm; it is not a sample-dependent value.

Accessory connectors visible inside the Cary 5000 sample-compartment area:

What are the accessory connection ports?

The Cary 5000 has dedicated accessory connections; they are not general-purpose computer ports. Agilent documentation for the Cary 4000/5000/6000i family describes three low-voltage/digital accessory connectors in the sample compartment and one accessory connector on the front of the instrument. It also documents a separate D-range connector containing high-voltage lines for DRA detector use. The front connector visible behind the ACCESSORY flap in the facility photo is therefore an accessory-control connection, not a place to connect an arbitrary cable or device.

Examples of compatible accessory hardware documented for the Cary 5000 include the UMA, external/internal DRA configurations, and other Agilent Cary accessories such as motorized

sample-positioning hardware when the specific accessory is supported. The exact connector used depends on that accessory's installation instructions. Do not choose a port by connector shape alone.

Accessory installation — Do not install or reconnect unfamiliar accessory hardware by guessing. The Cary 5000 must be OFF before accessory electrical connections are made or changed. For the UMA, the supplied procedure specifically says to get assistance when mounting it. For the DRA, get help before the first configuration.

“Setup → Summary → Instrument Settings” is a navigation path inside Cary WinUV. It opens the scan-setting fields explained in Step 4.

X-Mode controls the horizontal-axis quantity/unit. Y-Mode controls what is reported on the vertical axis. Beam Mode: Double uses the sample and reference optical paths. Spectral Band Width (SBW) is the wavelength band passed by the monochromator at one time. Data Interval is the wavelength spacing between recorded data points. Baseline Correction corrects for the measured instrument/reference background. The documented UV-Vis detector changeover setting is 850 nm.

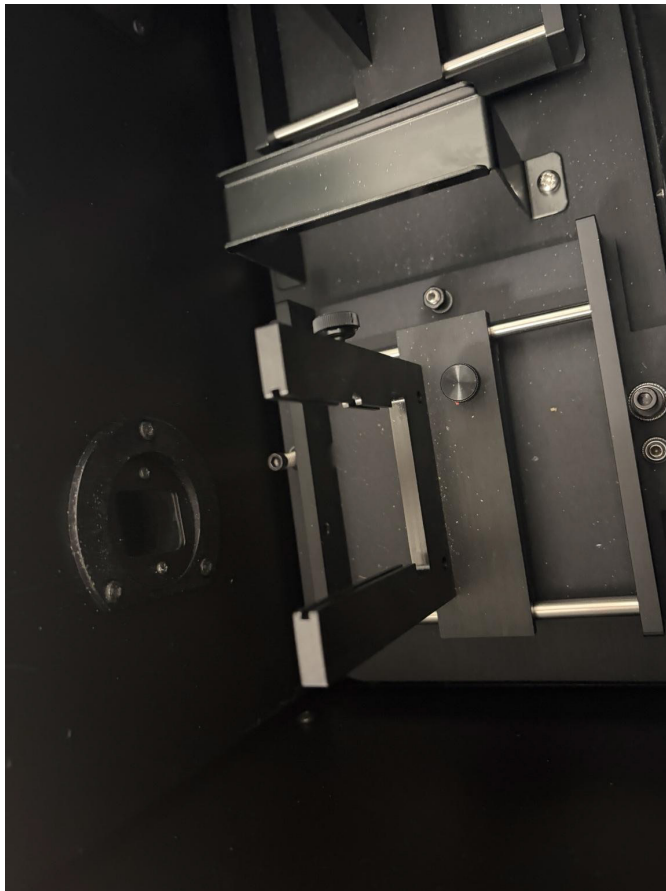
What is the 850 nm detector changeover wavelength?

The Cary 5000 uses different detectors to cover its wide UV-Vis-NIR wavelength range. The detector changeover wavelength is the point in the scan where the instrument changes from the detector used for the shorter-wavelength region to the detector used for the longer-wavelength/NIR region. In the supplied facility procedure, this changeover is set to 850 nm and is specifically marked “Important: keep at 850 nm.” Treat 850 nm as part of the documented instrument method, not as a value to choose based on the sample.

For the documented standard and DRA configurations, keep the detector changeover at 850 nm as specified by the supplied facility procedure.

Step 5 — Sample and Reference

Sample-holder area inside the sample compartment



:

The sample compartment is the enclosed measurement area under the sliding top door. In the standard configuration described by the supplied procedure, the FRONT holder is the sample position and the BACK holder is the reference position.

What is the reference? The reference is the comparison or “control” used to represent everything you do NOT want counted as the sample’s optical response. The correct reference depends on what is being measured. For a liquid measurement, this commonly means a blank containing the solvent/matrix without the analyte. For a film or solid, an appropriate substrate/background may be used when the method requires it. The supplied facility procedure tells the user to place a reference in the back holder “if necessary,” but it does not specify one universal reference material for every sample. The reference must therefore be chosen for the specific method/sample rather than assuming one material works for everything.

How do you know whether a sample is approved? The supplied procedure explicitly documents solid and liquid samples, films mounted to an aperture plate, and liquids in compatible non-frosted cuvettes. It also warns that the liquid must be compatible with the cuvette material; plastic/polystyrene can absorb some organic liquids such as toluene and leak. Liquid and/or volatile samples are to be handled in a fume hood. UMA and DRA measurements are also documented when those accessories are correctly installed. The supplied sources do not provide a complete chemical-by-chemical or sample-size approval list, so a sample outside these documented configurations should not be assumed approved.

The correct sample holder is the holder intended for the sample form and method: use the documented solid-sample/aperture holder for supported films/solids and the cuvette holder for liquids. The holder should seat correctly without force and allow the compartment to close.

DO NOT use a leaking, incompatible, damaged, or improvised cuvette/holder. DO NOT force a holder into place. DO NOT allow a liquid sample to leak into the sample compartment. DO NOT plug/unplug or install/remove accessories while the instrument is powered on.

Step 6 — Baseline

A baseline establishes the instrument/reference response before the sample spectrum is collected. A questionable baseline may result from an incorrect blank/reference, dirty or mismatched cuvettes, incorrect sample/reference mounting, an open sample compartment, or an instrument/performance problem. Do not simply continue with a questionable baseline.

Step 7 — How to Read the Measurement

First identify the axes:

- X-axis = wavelength, or the X-Mode selected in Setup.
- Y-axis = the measurement mode selected in Setup.

Absorbance tells you how strongly the sample attenuates light at each wavelength. Use Absorbance when the experiment is concerned with where/how strongly a sample absorbs light or when an absorbance-based method is being used. Higher absorbance means less of the incident light is transmitted. An absorbance peak is a wavelength region where absorbance is higher than the surrounding spectrum.

%Transmission tells you what percentage of the incident light passes through the sample. Use %Transmission when the experiment is concerned directly with how transparent/transmitting the sample is. Higher %T means more light passes through; a dip/lower %T means less light passes through.

Absorbance and transmittance describe the same light interaction in different ways, so choose the Y-Mode required by the experiment rather than treating one as automatically “better.”



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What abnormal-looking data means:

- Excessive noise = the trace looks unusually jagged/fuzzy instead of showing a stable trend; many small rapid fluctuations appear from one data point to the next.
- Sudden discontinuity = the trace abruptly jumps, drops, or breaks rather than changing smoothly with wavelength. A discontinuity can indicate a measurement/instrument transition or problem and should not automatically be interpreted as a real sample feature.
- Saturated / off-scale = the signal has reached or exceeded the usable measurement range, so the displayed value may no longer represent the sample accurately. A clipped/flattened region at an extreme value is a warning sign.
- Feature also present in the baseline/reference = a peak, dip, or shape seen in both the sample result and the background/reference may come from the blank, substrate, cuvette, background, or instrument rather than from the sample itself.
- Poor repeatability = a feature disappears, moves, or changes greatly when the same measurement is repeated or the sample is repositioned. Treat the feature as questionable until the cause is understood.

How to decide whether the spectrum is usable:

1. Compare the sample spectrum with the baseline/reference.
2. Check for excessive noise, discontinuities, off-scale regions, and background/reference features.
3. Repeat an unexpected result.
4. If the repeated spectrum agrees and the feature is not explained by the baseline/reference, it is more likely to represent the sample; interpretation still depends on the experiment.
5. Do not identify a material from a peak alone unless the method provides an appropriate comparison or identification basis.
6. If measurements remain inconsistent or a required Validate test has failed, do not report the data as a valid routine measurement until the problem is resolved.

Step 8 — Saving and Exporting

CSV is a portable text-based table of the measured data for analysis outside Cary WinUV. The supplied facility procedure instructs users to select Convert Data to .csv under Reports for automatic CSV conversion. The same procedure identifies .dsw and .bsw as Cary Save As options. Validate report files are separate from sample-scan data.

Step 9 — Shutdown

The supplied facility procedure ends with four actions: turn off the Cary 5000, close the software, sign the log book, and log off the instrument. Follow the local logoff screen/session process you were trained to use; do not shut down or disconnect additional equipment unless instructed.

11. Definitions

Absorbance (Abs) — A logarithmic measure of light attenuation by a sample. Higher absorbance means less light is transmitted.

Baseline — A measurement used to establish the instrument/reference background before measuring the sample.

Aperture Plate / Solid-Sample Aperture Setup — Hardware used to support a film or solid sample while defining the opening through which the instrument's light beam passes.

Cuvette / Cell — A small optical sample container used for liquid measurements.

Cuvette / Cell Holder — Hardware inside the sample compartment that holds a cuvette in the instrument's light path.

Cary 5000 — UV-Vis-NIR spectrophotometer used to measure wavelength-dependent absorption/transmission of supported samples.

Cary WinUV — Agilent software used to control the Cary 5000, configure measurements, collect spectra, and save/export data.

CSV — Comma-separated values; a text-based tabular export that can be opened in spreadsheet/data-analysis software.

Data Interval — The wavelength spacing between recorded data points.

Double Beam — A measurement configuration using sample and reference optical paths.

DRA — Diffuse Reflectance Accessory; the accessory used for the facility's documented diffuse-reflectance measurement, reported as %R.

Instrument Performance Test — A test that compares a measured instrument property with a specified acceptance criterion.

Instrument Performance Test Procedure — A Cary WinUV Validate procedure that checks instrument performance against acceptance criteria; it is separate from a sample measurement. Use the installed Validate test suite/on-screen instructions rather than creating your own test sequence.

Method — A saved Cary WinUV setup containing measurement/instrument settings. The supplied facility procedure uses noUMA_BaseMethod and UMA_BaseMethod as starting configurations.

NIR — Near-infrared light, beyond the visible region.

Reference / Blank — The comparison or “control” optical path/material used to account for background contributions that should not be attributed to the sample.

Sample Compartment — The enclosed measurement area under the Cary 5000 sliding top door that contains the sample/reference positions and installed holder/accessory.

Spectrophotometer — An instrument that measures how much light a sample absorbs or transmits at different wavelengths. The Cary 5000 does this across the UV, visible, and near-infrared regions to produce a spectrum.

SBW / Spectral Band Width — The wavelength band passed by the monochromator at one time.

Transmittance (%T) — The percentage/fraction of incident light transmitted through a sample. Higher %T means more light passes through.

UMA — Universal Measurement Accessory; an alternate Cary 5000 configuration documented by the supplied facility procedure for external diffuse-reflectance measurements of opaque substances and for kinetic studies.

UV — Ultraviolet light, below the visible region.

Vis — Visible light.

UV-Vis-NIR — Ultraviolet, visible, and near-infrared spectral regions measured by the instrument. UV-Vis-NIR measurements can be used to examine how a material's optical behavior changes with wavelength.

Validate — Cary WinUV application used for instrument performance testing.