

Quick Reference Cards | CG000543 | Rev G

Visium CytAssist

for Instrument Operation

For use with:

Visium CytAssist: Includes Accessory & Training Kit, 12-Month Warranty, PN-1000441

Visium CytAssist: Includes Accessory & Training Kit Kit, 24-Month Warranty, PN-1000442

Visium CytAssist, Refurbished, PN-1000432R



Visium CytAssist Document Revision Summary



Document Number

CG000543 | Rev G

Title

Visium CytAssist Quick Reference Cards - for Instrument Operation

Revision

Rev F to Rev G

Revision Date

April 2025

Description of Changes

• Updated steps for "Load Tissue Slides onto Tissue Slide Stage" to account for stronger clips on the Tissue Slide Stage on page 7

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Visium CytAssist Step 1: Installation



Before unboxing the instrument, choose a location that:

- Is not in direct sunlight.
- Includes a level sturdy laboratory bench that is not subject to movement or vibration.
- Provides maximum bench space (front to back).
- Allows access to the wall plug and to the detachable power cord at the rear.
- Provides a minimum of 4" (10 cm) clearance around unit to allow free air circulation.

Unbox

- **a.** Open box according to box label "this side up." Carefully cut or remove the tape on the top of the box and open the top flaps. Ensure that cut is not too deep as it may damage the Instrument Accessory Kit box.
- **b.** Remove the Instrument Accessory Kit box, documents, and packaging material.
- **c.** Lift the instrument from the sides. Place on bench top. Carefully remove all packaging materials/tape from the exterior of the instrument. Retain all packaging materials in the event that the instrument needs to be moved.
- **d.** Remove Shipping Bolt from the back of the instrument. Retain for future use.
- e. Open the lid. Carefully remove tape from the four Tissue Slide Clips and the Visium Slide Lock.
- **f.** Remove glass protective films from the alignment windows on both stages. Close lid (clicking sound indicates closure).
- **g.** Within Instrument Accessory Kit, locate the universal power cord and plug adaptors. Attach the correct country adaptor to the universal adaptor.
- **h.** Plug the Visium CytAssist into a properly grounded outlet of proper voltage. If using an Ethernet cable (not provided), connect cable directly to the instrument.



Retain packaging for future transportation







Visium CytAssist Step 2: First-Time Use



The instrument will walk through a first-time setup protocol. Setup requires the following:

- 10x Genomics Account
 Onscreen instruction will guide account setup.
 Alternatively, visit cloud.10xgenomics.com/instrument
 (also accessible by scanning the QR code from the
 "Register" screen) to create an account beforehand.
- Instrument serial number and instrument ID Located onscreen during setup, and also on the label at the back of the instrument
- Network settings
- A computer with Internet access
- Internet connection to the instrument Wi-Fi configuration may not be available. At present, only 5 GHz Wi-Fi connections are supported. Ethernet is recommended for optimal performance.

Turn on the System

- Power on the instrument using the power switch at the back of the unit.
 If using Ethernet, connect the cable to the instrument Ethernet port.
- **b.** Follow onscreen instruction for Network Setup (via ethernet or Wi-Fi). *Refer to the CytAssist Network Connectivity Technical Note (CG000653) for more details.*
- c. Set the Date and Time.
- **d.** Cloud Registration: On a networked device, login to your 10x Genomics account at <u>register.10xgenomics.com</u>. Enter the serial number + instrument ID. Read and accept the legal agreement.
- e. Update the firmware. After registration, the instrument will automatically search for the latest firmware update. Confirm settings for instrument diagnostics and automatic firmware download.
- f. Reboot by clicking the "Restart Instrument" button to complete installation.DO NOT use the power switch at the back of the unit.



•		
Networ	k Setup	
For the simplest firmware update and support experience, connect your instrument to a network.		
Connect an Ethernet cable to	o the instrument to continue.	
Ethernet status	Not Connected	
Setup Wi-Fi	Skip Network Setup	
register:10xgenomics.com		
register.ioxg	enomics.com	Ð
	enomics.com	G
Scan the OR code or visit the instrument. When prompt	enomics.com	G
Scan the OR code or visit the instrument. When prompt	enomics.com	0
Scan the OR code or visit the instrument. When prompt Once registration is comp Serial number ABCD1234	enomics.com	0

g. To level the instrument, turn the adjustable feet at the front of the instrument.

Turn feet clockwise for decreasing and counterclockwise for increasing height. The feet will come off the instrument if continued to be turned counterclockwise.

Acceptable leveling ranges are $\pm 3^{\circ}$ from left to right and $\pm 1^{\circ}$ from front to back.

See Troubleshooting section of the Visium CytAssist Instrument User Guide (CG000542) if an error occurs during the first-time setup protocol.

Visium CytAssist Step 3: Readiness Test



The Readiness Test calibrates the imaging parameters and verifies that all systems are working optimally. No reagents are used. Wear gloves.

a. During first-time setup, follow onscreen instruction to launch the Readiness Test (~40 min).

To initiate a Readiness Test at any other time, navigate through the menu to System > Alternate Modes > Readiness Test.

OR press the blue Run Start Button on the touchscreen to initiate a run. Enter the Readiness Test serial number (located on the Calibration Slide label).

- **b.** Locate two blank slides and the Visium CytAssist Calibration Slide in the Instrument Accessory Kit. Ensure slides are clean.
- **c.** Enter the serial number on the calibration slide (using a different serial number could result in poor calibration).
- d. Open Lid.
- e. Follow onscreen instruction to load slides.
- f. Press the blue play button to start the test.
- **g.** A successful Readiness Test marks the end of setup. Follow onscreen instruction in the event of a failed or incomplete test.
- **h.** Upon test completion, follow onscreen instruction to unload slides.
- i. Store the Calibration Slide away from sunlight and extreme temperature, in the original packaging bag. Retain for future use.
- **j.** Wipe down the Tissue Slide Stage and the Visium Slide Stage with laboratory wipes, a mixture of mild detergent and distilled water.
- **k.** Close lid. Wipe down the base and lid of the instrument.

For optimal performance, a test is recommended during first-time use, after any update, and after moving the instrument.

The instrument requires a **~20 min cool down** between runs.

Operation Screens

After the Readiness Test, the instrument is ready for use. Representative instrument operation screens shown below.



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Visium CytAssist Step 4: Main Operation



Main Operation Overview

This section is meant as a general overview of instrument operation and its placement within the steps of the Visium CytAssist-enabled assay. When performing the protocol, follow the steps listed in the appropriate assay kit user guide and use this section only as a reference.

Before proceeding with the appropriate Visium CytAssist-enabled assay:

Select two tissue slides containing sections of interest, and verify that the tissue section is located in an area that results in successful analyte-derived molecule transfer and imaging. Overlay the slides on the diagrams (instructions for 25 mm or 26 mm width) in the Visium CytAssist Quick Reference Cards – for Accessory Kit Instruction (CG000548).

If only one tissue on one slide will be loaded, use a blank slide for the second position on the Tissue Slide Stage.

Follow the protocol in the appropriate assay kit user guide for preliminary steps and preparation prior to using the Visium CytAssist instrument.

- **a.** Gather two tissue slides, a Visium slide and reagents as listed in the appropriate assay kit user guide.
- **b.** Enter new run information, including:
 - Visium Slide serial number Once entered, editing is limited—the first digits (for slide type) cannot be changed. This prevents the instrument from running an incompatible program. To input a different full serial number, exit the screen, return to the home screen, and start over.
 - Custom run name
 - Any change from default temperature and time (37 °C for 30 min)
 - Sample names; the 6.5 mm capture areas are designated **A1** and **D1**. The 11 mm capture areas are designated **A** and **B**.

- **c.** Open the lid.
- **d.** Follow onscreen instruction and load the blank slides and Visium Slide onto the instrument. See Load Slides section for details.
- e. Load reagents to each spacer when instructed in the appropriate assay kit user guide (be mindful of time-sensitive reagents).
- f. Close the lid.
- **g.** The home screen will now display a play symbol and run information along the bottom of the screen. (Use button in upper right corner of the screen to abort).
- **h.** Press the play button to start the run.
- i. At the end of a run, the button will display "Done" and a "Run Info" tab at the bottom of the screen.
 - Green: a successfully completed run
 - Red: a failed run/error
 - Yellow: a canceled run
- **j.** Click the "Done" button and open the lid. DO NOT power off the instrument at this time, as it needs to process Support Data.
- **k. IMMEDIATELY** remove the Visium slide from the instrument, and proceed with the protocol workflow as specified in the appropriate assay kit user guide (save cleanup for later).

During next stopping point in the workflow:

- **1.** Clean the instrument after every run.
- **m.** Export the Run Data via USB or Network Shared Folder.
- **n.** View and verify the tissue samples were accurately aligned within the instrument alignment guides.



The instrument requires a **~20 min cool down** between runs.

Visium CytAssist Load Slides onto Slide Stage

Load Tissue Slides onto Tissue Slide Stage

To avoid slide damage during loading:

- DO NOT adjust slide while tissue clip is touching the slide
- DO NOT place clips on the edge of the slide
- DO NOT touch the tissue section
- DO NOT snap-release the clip (instead, gently decrease pressure to release)
- **a.** Press down fully on each clip to lift and rotate them to their outermost position. For stability, cradle the instrument lid while pressing.
- **b.** Lay tissue slide flat on the stage surface and against the center line. Rotate the slide 180° as needed to place off-center tissues closer to the center line (do not overlap center line).
- **c.** Use one hand to hold the slide in place. Use the other to rotate the clip over the slide.
- **d.** Hover clip over the slide while making fine adjustments to tissue alignment. Gently release clip.
- e. Continue holding slide in place while positioning the second clip. Use both clips whenever possible. Omit second clip if the tissue slide is beyond its reach.
- **f.** If only one tissue slide will be loaded, use a blank slide for the second stage position.

Load Visium Slide onto Visium Slide Stage

- **a.** Inspect the stage for dust/debris.
- **b.** Pivot the Visium Slide Lock with one finger.
- **c.** Line up the slide with the label face up and oriented to the right.
- **d.** Fit the slide within the raised grooves.
- e. Hold the slide in place while closing the Visium Slide Lock (slide label will be partially obscured).





INCORRECT Slide does not fit inside grooves. Capture areas do not line up with alignment windows.



INCORRECT Label is on the left. Capture areas do not line up with alignment windows.





Visium CytAssist Firmware Update Procedure



Consult the Visium CytAssist Instrument User Guide (CG000542) for additional details. Automatic downloads of updates are enabled after firmware updates to ensure the instrument is up-to-date with the latest assay products, features, and bug fixes.

Step 1: Download and install the update

Procedure with an instrument connected to the internet and auto-download enabled (recommended):

- **a.** A pop-up on the home screen will alert the user of available updates.
- **b.** Open pop-up and click to install.
- **c.** Once the update is complete, click the "Restart Instrument" button.

DO NOT skip this button and use the power switch at the back of the unit; this could result in lost data.

Procedure with an instrument connected to the internet, auto-download disabled:

- **a.** A pop-up on the home screen will alert the user of available updates. Open and click to download.
- **b.** Once the update has been downloaded, install the update.
- **c.** Once the update is complete, click the "Restart Instrument" button.



DO NOT skip this button and use the power switch at the back of the unit; this could result in lost data.

Step 2: Reinitialize the instrument

After installing an update, run a Readiness Test.

 a. To initiate a Readiness Test, navigate through the menu to System > Alternate Modes > Readiness Test. OR press the blue Run Start Button on the touchscreen.
 Enter the serial number located on the Visium CytAssist Calibration Slide label.

- **b.** Locate two blank slides and the Visium CytAssist Calibration Slide in the Instrument Accessory Kit. Ensure slides are clean.
- **c.** Follow onscreen instruction to load the blank slides and calibration slide onto the instrument.
- **d.** Press the blue play button to start the test.
- e. Upon test completion, follow onscreen instruction to unload slides. Follow onscreen instruction in the event of a failed or incomplete test.
- **f.** Store the slides in their original packaging, away from sunlight and extreme temperature. Retain for future use.
- **g.** If lab safety guidelines permit, a soft lab towel may be used to clean the instrument with a mixture of mild detergent and distilled water.

External instrument surfaces and slide stages can tolerate periodic additional cleaning with a 10% bleach solution followed by a 70% ethanol or 70% isopropanol wash. Follow the relevant lab safety requirements when cleaning the instrument. **Glass windows** may be cleaned with dry polyester swabs (such as Texwipe TX714A Cleanroom Swab, 0.5"L - EW-33677-62 from Cole-Parmer or equivalent). Use a bright light (e.g. flashlight) to inspect the glass windows for any smudges and remove them as described above.

- **h.** Close the lid (clicking sound indicates closure). Wipe down the base and lid of the instrument.
- i. The instrument is now ready for use.