Saphyr® System User Guide

DOCUMENT NUMBER: CG-30247

DOCUMENT REVISION:

Effective Date: 09/20/2023

For Research Use Only. Not for use in diagnostic procedures.

Table of Contents

Table of Contents
Revision History
Overview
Introduction
Features
Workflow
Prepare Sample7
Getting Started7
Start the Saphyr Instrument7
Laser Safety Light
Instrument Status Light
Sign in to the Saphyr Instrument Control Software
ICS Home Screen Options9
Configure Analysis Settings in Bionano Access9
Load a Saphyr Chip with Sample 10
Prerequisites
Loading Samples into the Chip Flowcells11
Placing Clips and/or Plugs onto the Chips13
G2.2 or G2.3 Chips 14
G1.2 Chip
Perform Run
Insert Saphyr Chip [®] Consumable16
Configure Run

Chip and Flowcell States	5
Pausing a Run to Insert a New Chip and/or Remove a Completed Chip	6
Pausing the Run to Rehydrate the Chip29	9
Stopping the Run	9
Remove Chip(s)	D
Analyze Data	1
Data Transfer for Analysis	1
Saphyr Assure	1
Updating Software	2
Logging off and System Administration	3
Maintenance	3
Clean the Saphyr Clip Plugs	3
Clean the Saphyr Seal	3
Clean the Objective Lens	4
Air Filter	5
Appendix A	6
Troubleshooting	6
Export Tier 1 Run Diagnostics 40	D
Export Tier 2 Run Diagnostics	3
Export / Upload Data Configuration 44	4
Appendix B 45	5
Rerunning a Chip	5
Example scenarios for re-running a chip46	6
Appendix C 49	9
Instrument Control Software (ICS) Icons 49	9

Run Info 50
Bionano Access Connection Status 50
Saphyr Assure Status
Remote Support 51
Appendix D
Instrument Initialization
Appendix E
Data Administration
Insufficient Disk Space53
Delete Runs
Appendix F
Upload Run to Access
Appendix G
Instrument Controller Operations55
Appendix H56
Instrument Disposal
Additional Resources
Technical Assistance
Legal Notice

Revision History

REVISION	NOTES					
A	nitial Release					
В	Revised to reflect the changes associated with the release of Instrument Control Software v4.9					
B1	Added missing image to page 17.					
B2	Fixed the incorrect chip names					
с	Updated to reflect increase in DNA output to 5 Tbp					
D	Updated to reflect changes in ICS v5.1					
E	 Updated to reflect changes in ICS v5.2 Updated Perform Run procedure section Updated Pause Run procedure section; Included updating Software section Updated Maintenance Section 					
F	Updated guidance on pipetting samples into chips and on loading two chips on the instrument					
G	Fix errors in rev F					
н	Revise Chapter 4 for new G3.3 Chip.					
1	Updated for objective front lens cleaning. Streamlined the sample loading section.					

Overview

Introduction

CAUTION: Prior to operating the Saphyr® instrument, please review the Saphyr System Safety Guide (CG-30144 or 30253) for all safety and compliance information. The Saphyr system has been tested to the IEC 61010-1:2010 standard.

The Saphyr system (Saphyr instrument with Instrument Controller and Bionano Access[™] server) together with the Saphyr Compute and Bionano Compute servers provides rapid, high-throughput, long-range optical genome mapping for *de novo* assembly of genome maps, rare variant analysis, hybrid scaffolding of NGS, and structural variation analysis.

Features

- Easy load-and-go workflow The adaptive loading feature in the Instrument Control Software (ICS) ensures that the optimal conditions for DNA molecule loading are maintained throughout the scanning. The initial hands-on-time is approximately 5 minutes.
- **Convenient setup and flow** Bionano Access software allows users to design experiments, track run performance, perform downstream analysis, and share data.
- **Real-time analysis** Bionano Access software enables users to view real-time data and performance metrics from any internet browser with connection to your Access Server as the run progresses.
- **Saphyr Assure software –** Optional automated system health monitoring service that receives run metrics (including data quality and instrument health and performance metrics) after each Run. Additionally, this provides a streamlined diagnostic workflow for customers to interact with Bionano support.

Workflow

To perform a run on the Saphyr instrument, use the following workflow example (Figure 1) as a guide.



Figure 1. Workflow

- 1. Perform DNA isolation and labeling of samples.
- 2. Set up an experiment and specify run parameters in Bionano Access.
- 3. Turn the Saphyr instrument on and sign into the Instrument Control Software.
- 4. Load the samples on a Saphyr Chip consumable and attach the Saphyr Clip to the chip. **NOTE:** Ensure that the correct Chip clip is used with the correct chip.
- 5. Insert the Saphyr Chip consumable into the instrument.
- 6. Setup a run.
- 7. Track run performance and analyze data in Bionano Access software.
- 8. Remove the chip from the instrument.

Prepare Sample

For details on DNA isolation and labeling of samples, visit the Bionano Support page.

Getting Started

Start the Saphyr Instrument

- 1. Always using gloves, locate the power switch on the back of the instrument.
- 2. Toggle the power switch (**Figure 2**) to turn the instrument on.



Figure 2. Power switch

3. Boot the Saphyr Instrument Controller. Systems that are in "kiosk" mode will boot directly to the ICS. Kiosk mode allows the users to interact with ICS only, while keeping the instrument controller in a secure environment. Systems that are not in kiosk mode will require the user to log into a Windows user session, using the password that was provided by the Bionano Field Service Engineer (FSE) or Field Application Scientist (FAS), then double click on the ICS icon to launch the software. If the username or password are forgotten, please contact Bionano Support.

Laser Safety Light

IMPORTANT: Before lifting the sample door to insert or remove the Saphyr Chip[®] consumable, ensure the laser safety light is blue (Figure 3 below, left). If the laser safety light is red (Figure 3 below, right), do not open the sample door and contact <u>Bionano Support</u> for assistance; users may be at risk for laser radiation exposure if the sample door is opened. For additional information, please refer to the <u>Saphyr System Safety</u> <u>Guide (CG-30144, 30253, or 30302)</u>.



Figure 3. Instrument lighting

Instrument Status Light

The instrument status light indicates the following (Table 1):

Color	Description			
Light off	ne instrument is powered off.			
Flashing yellow	The instrument is initializing.			
Green	The instrument is ready to use.			
Flashing green	The instrument is running, or the workflow has been paused.			
Red	The instrument encountered an error. NOTE: This does not present a safety issue to the user.			

Sign in to the Saphyr Instrument Control Software

1. When the software is launched, the Saphyr system will initialize. This can be seen by clicking on the **Status** icon. A description of all icons can be found in *Appendix C* and a description of the initialization screen can be found in *Appendix D*.

Enter the username and password. This is the username and password that has been set up in Bionano Access.

2. Click Logon.

ICS Home Screen Options

The following options appear as a menu on the **Home** screen (**Table 2**). Not all options are always available, depending on the state of the instrument.

Option	Description				
Insert Chip	Allows users to insert the Saphyr $\operatorname{Chip}^{\otimes}$ consumable into the Saphyr instrument.				
Configure Run	lows users to set up a run. This option is only available if a chip is loaded.				
Data Administration	Allows users to delete images from a Run where all images have been saved. See <i>Appendix E</i> for details.				
Remove Chip	Allows users to remove the chip from the Saphyr instrument. This option is only available when a chip is loaded.				

Table 2. Home screen menu items

Configure Analysis Settings in Bionano Access

The Instrument Control Software and Bionano Access server interact to allow users to track run progress and view results in real time. Prior to performing the run on the Saphyr instrument, an experiment must be set up in Bionano Access 1.8. For details on how to do this, as well as all Bionano Access analysis and visualization features, see the *Bionano Access Software User Guide* (CG-30142).

IMPORTANT: Using **Table 3** to track chips, make sure that samples are loaded in the same flowcells on the chip as the ones set up in Bionano Access described above. Also, note the serial number of the chip so it can be associated with the correct experiment.

Table 3. Chip tracking table

Chip SN	
Flowcell 1 Sample :	
Flowcell 2 Sample :	
Flowcell 3 Sample :	

Load a Saphyr Chip with Sample

Prerequisites

- Equilibrate the labeled samples and Saphyr Chip consumable (sealed in pouch) to room temperature for <u>at least</u> <u>thirty (30) minutes</u> prior to use.
 - IMPORTANT: Do not open the pouch containing the Saphyr Chip consumable when the chip is cold. Wait until the pouch is at room temperature before removing the chip. Opening a chip pouch while still cold may damage the chip and cause performance degradation. Always use gloves.
- When loading sample, ensure there is no airflow around the chip to avoid sample dehydration.
- Proceed through the following steps carefully, without interruption to minimize sample evaporation.
- Use gloves (powder free) for this procedure.
- The instructions below are applicable for loading a sample in any Saphyr Chip. However, it is important to use the appropriate clip with its companion chip as follows:
 - G3 Chip Clip (with its Chip Seal and Plugs) with G3.3 Chips (Figure 4A)
 - G2 Chip Clip (with its plugs) with G2.3 or G2.2 Chips (Figure 4B)
 - G1 Chip Clip will only fit G1.2 Chips (Figure 4C).



Figure 4. **A)** Chip Clip G3 and Saphyr Chip G3.3 **B)** Chip Clip G2 and Saphyr Chip G2.3 **C)** Chip Clip G1 and Saphyr Chip G1.2

Loading Samples into the Chip Flowcells

NOTE: A video of how to load the sample onto the Saphyr Chip[®] consumable on a G3.3 Chip (no intended audio) can be found at https://bionano.com/training-videos/saphyr-g3-3-chip-loading-training/

NOTE: A video of how to load the sample onto the Saphyr Chip[®] consumable on a G2.3 Chip can be found at https://bionano.com/training-videos/loading-saphyr-chip-g2-3-pn-20366/

- 1. Open the pouch containing the Saphyr Chip consumable.
- 2. Place the chip onto lint-free paper and a clean and level lab bench.
- 3. With a 10 μ l pipette, press the plunger down and hold at the first stop.
- 4. Position the pipette tip near the middle or bottom of the sample and slowly aspirate 8.5 µl of the sample.
- 5. Begin loading the sample in the inlet well of flowcell 1.

Place the pipette tip into the flowcell 1 inlet well (**Figure 5** for G3.3, G2.3 and G2.2, **Figure 6** for G1.2) on the right of the chip, and then slowly dispense the sample until the bottom of the inlet well is coated with sample solution. Continue to dispense, adjusting the rate as the DNA travels slowly into the flowcell fingers, until the entire volume of 8.5 μ l is in the well.

IMPORTANT: The fluid level in the inlet well should be flat. If the sample overflows the well and out onto the top of the chip or onto the plastic, the flowcell is not usable. Always maintain positive pressure on the plunger to avoid bubble formation.



Figure 6. Outlet and Inlet wells and fingers for Chip G1.2 but no reservoir.

- 6. Repeat steps 3 through 5 for flowcell 2 and flowcell 3 (as applicable) inlet wells.
- 7. Set a timer for two minutes to allow the sample to wet through the nanochannels.
- After two minutes, with a 20 µl pipette positioned near the middle or bottom of the sample, slowly aspirate 11 µl of the sample, then slowly dispense into the outlet well of flowcell 1. Repeat for flowcell 2 and flowcell 3.
- 9. For flowcell 1, add 5 µl of nuclease-free water to the hydration reservoir on the outlet side of the chip (see OUTLET on the chip label). Do not add water to the reservoir on the inlet side of the chip as this may lead to problems with the chip run. NOTE: There are no hydration reservoirs on G1.2 Chips. This step can therefore be skipped.

Add the necessary volume of nuclease-free water to the inlet and outlet wells of both flowcells to replace sample solution that has evaporated. To lower the risk of evaporation, add water to make the fluid level slightly convex relative to the top of the chip. See **Figure 7** and **Figure 8** below and the instructional videos listed.



Figure 8. Well fill level.

IMPORTANT: The inlet and outlet wells (see Figure 5 and Figure 6) should be filled with sample solution during the initial load. Do not compensate for lack of sample solution by adding nuclease-free water or buffer. Nuclease-free water should only be added after all wells are loaded to replace volume lost by evaporation.

<u>For G2.2, G2.3 and G3.3 Chips only</u>: as soon as a flowcell is loaded with sample and the eventual sample evaporation has been compensated for with nuclease-free water, proceed with loading. Attaching the plug to the flowcell limits evaporation occurring while other flowcells are loaded.

Placing Clips and/or Plugs onto the Chips

G3.3 CHIP

1. Align the plug (with the electrodes facing down) to flowcell 1 and gently insert fully into the chip. **NOTE:** It is highly recommended that all flowcells in the chip are used in the same run. Once the chip has been removed from the original sealed pack, the performance will degrade over time. See **Figure 9**.



Figure 9. Saphyr G3.3 Chip with samples loaded; one plug closing each flowcell

- 2. Repeat steps 9-10 in the Loading Samples into the Chip Flowcells section for flowcells 2 and 3.
- 3. Place the Saphyr Clip Seal on top of the Saphyr Chip consumable by aligning the six holes in the Saphyr Clip Seal with the top of the six electrode posts on the plugs. Lightly press down on the surface of the Saphyr Clip Seal to ensure uniform contact with the Saphyr Chip consumable and plug surfaces.
- 4. Align the Saphyr Clip onto the Saphyr Chip consumable, as seen in Figure 10. Verify correct orientation such that the electrode posts on the Teflon plugs fit in the opening in the clip. Using both hands, simultaneously apply an even downward force on the right and left side of the clip to attach. A click can be heard on each side of the clip when it is engaged with the chip. Once the clip is attached to the chip, do not remove it unless necessary. If any of the plugs are removed, rehydrate the chip inlets and outlets with nuclease-free water to replace any sample solution that has evaporated so the fluid level in the wells is flat.



Figure 10. Saphyr G3.3 Chip closed with Clip, after Chip Seal addition

IMPORTANT: Even if all flowcells do not have sample, all three plugs must be inserted into the chip (**Figure 10**) to ensure proper evaporation control from the clip. <u>Use gloves when handling samples, chips, and plugs.</u>

5. The chip and sample can be left out for up to one (1) hour at room temperature with the clip engaged before starting the run.

G2.2 or G2.3 Chips

 Align the plug (with the electrodes facing down) to flowcell 1. Use a slight to-and-away rocking motion on the plug to gently insert the plug fully into the chip (Figure 11). NOTE: It is highly recommended that all flowcells in the chip are used in the same run. Once the chip has been removed from the original sealed back, the performance will degrade over time.



Figure 11. Saphyr Chip G2.3 with one Plug engaged.

2. Repeat steps 9-10 in the Loading Samples into the Chip Flowcells section for flowcells 2 and 3.

3. Align the Saphyr Clip onto the Saphyr Chip[®] consumable. Verify correct orientation such that the electrode posts on the Teflon plugs fit in the opening in the clip. Using both hands, simultaneously apply an even downward force on the right and left side of the clip to attach. A click can be heard on each side of the clip when it is attached as seen in **Figure 12** Once the clip is attached to the chip, do not remove it unless necessary. If any of the plugs are removed, rehydrate the chip inlets and outlets with nuclease-free water to replace any sample solution that has evaporated so the fluid level in the wells is flat.



IMPORTANT: Even if all flowcells do not have samples, all three plugs must be inserted into the chip (**Figure 12**) to ensure proper evaporation control from the clip. <u>Use gloves when handling samples, chips, and plugs.</u>



Figure 12. Saphyr G2.3 Chip closed by Clip, after flowcells closed by the three plugs.

4. The chip and sample can be left out for up to one (1) hour at room temperature with the clip engaged before starting the run.

G1.2 Chip

- 1. Align the clip on the chip by verifying the appropriate clip orientation. Make sure the clip is aligned correctly, as depicted in **Figure 13**; putting the clip on backwards may break the chip.
- 2. Place the clip over the chip.
- 3. Using both hands, apply an even downward force on the right and left side of the clip to attach. **NOTE:** downward pressure should be applied to the clip, not the chip.
- 4. Users will hear a click when the clip is attached. Once the clip is attached to the chip (**Figure 14**), do not remove it unless necessary. If the clip is removed, rehydrate the chip inlets and outlets with nuclease-free water to replace any sample solution that has evaporated.
- 5. The chip can be left out for up to 1 hour at room temperature before starting the run.



Figure 13. Saphyr G1.2 Chip Clip alignment



Figure 14. Saphyr Chip G1.2 with Chip Clip attached

Perform Run

Insert Saphyr Chip[®] Consumable

Before inserting the Saphyr Chip consumable into the Saphyr instrument, carefully check the underside of the chip for dust or debris without turning the chip upside down. If there is dust or debris, gently wipe the underside of the chip with lens paper. Avoid touching the underside of the chip with anything other than lens paper.

1. From the Home screen, click Insert Chip, as seen in Figure 15.



Figure 15. Insert chip.

Users will hear the stage moving outward, which can take about 10 seconds. When the stage has reached the proper position to allow for insertion of the chip, the **Chip is Inserted** option will activate on the screen.

2. Manually lift the sample door of the instrument, shown in Figure 16.



Figure 16. Sample door open with Chip inserted and with bundle arm lowered.

3. Lift the bundle arm:

For a dual chip instrument, press the button that says **Press** to release the bundle arm (**Figure 17**). The bundle gently rises to a vertical orientation, thereby presenting the two positions where the chip(s) can be inserted into the instrument.



Figure 17. Press button.

For a single chip instrument, manually lift the bundle arm, as seen in Figure 18.





4. Insert one or two chips (if applicable) into the instrument and ensure they rest snugly on the stage. If only inserting one chip, either platform can be used.

NOTE: The clip is designed to provide evaporation control for up to 65 hours. If inserting two chips into the instrument, the second chip will need to be complete within 65 hours.

TIP: If loading two chips, record which chip is on which platform, using **Table 4**. The ICS will ask which experiment to associate with each chip (See the *Configure Run* section).

Table 4. Chip position table.

Stage Position	1 (left)	2 (right)
Chip SN :		

- 5. Gently lower the bundle arm onto the Saphyr Clip(s), and then close the sample door.
 - In the Home screen, click Chip is Inserted. The instrument will read the barcode to identify the chip(s). If the instrument is unable to identify the chip barcode, an error message will appear. Remove the chip and inspect the barcode to see if it is damaged. If not, reseat the chip(s), making sure that it is centered on the stage mounts. If there are further issues, contact Bionano Support.
- 6. Proceed to the *Configure Run* section.

Configure Run

From the Home screen, click Configure Run. The Configure Run screen appears, as seen in Figure 19. If two chips were inserted, the user will be prompted to configure the chip on platform 1 first (Chip Location: P1). Platform 1 is on the left side of the stage.

FOR INSTRUMENTS CONNECTED TO ACCESS 1.8 AND NEWER VERSION

Configur	e Rı	ın						
Chip Location:								
Serial Number:		KWNPMXAł						
Part Info:								
Chip Name:			+	\$				
User:	bionan	oadmin	Char	nge				
 Advanced 								
				Sk	ip Current C	hip	Remove Chips	

Figure 19. Configure Run screen for Access 1.8.

2. At the **Chip Name** field, select the chip setup to associate with the chip from the drop-down list. See the *Bionano Access User Guide* (CG-30142) for details on how to create an experiment.

[Optional] Click to create a new chip setup. This will bring up the Bionano Access software within the Instrument Control Software. See the *Bionano Access Software User Guide* (CG-30142) for instructions on setting up a chip.

[Optional] Click with to update the list.

FOR INSTRUMENTS CONNECTED TO ACCESS 1.6 AND OLDER VERSION

Configure Run				
Chip Location:				
Serial Number:				
Part Info:				
Experiment:	← Refresh			
Chip Setup:				
User:	pionanoadmin			
▶ Advanced				
Next	Skip Current Chip Remove Chips			

Figure 20. Configure Run screen for Access 1.6.

1. At the **Experiment** field, select the experiment to associate with the chip from the drop-down list. See the *Bionano Access Software User Guide* (CG-30142) for details on how to create an experiment.

[Optional] Click **Refresh** to update the list.

2. If applicable, at the **Chip Setup** field (**Figure 20**), select the chip setup from the drop-down list. **NOTE:** This step is only applicable if there is more than one chip associated with the experiment and will take the user to the expanded **Configure Run** screen, seen in **Figure 21**.

FOR ALL ACCESS VERSIONS



Figure 21. Expanded Configure Run screen.

- 1. Review the runtime and throughput target parameters for each of the flowcells. These values will default to the values that were entered into the Bionano software during setup. The user may decrease the runtime or throughput as desired. **NOTE:** If the chip has been run previously, the runtime limit and throughput target may be modified. See *Appendix B* for details.
- 2. [Optional] Prior to clicking **Accept** or **Next**, the option is to click on either **Skip Current Chip** or **Remove Chips.**
- 3. Skip Current Chip This will allow users to disregard the chip in this platform. The chip will not be run. See the *Chip and Flowcell States* section below for more details.
- 4. Remove Chips This will allow users to stop configuring runs and remove the chip(s), if needed. After chips are removed, users will return to the **Home** screen.
- 5. [Optional] Clicking on the arrow next to **Advanced...** will give users the option to save the images that are captured during the run. By default, images are not saved. If users choose to save images, see *Appendix E* on managing that data. This option is only available for customers who are not running in kiosk mode.
- 6. If two chips have been inserted into the instrument, click Next and repeat steps 2-7.

NOTE: If the second chip belongs to a different user, change users before starting the second chip configuration. Individuals with "User" privileges can only run experiments that they have created

- 7. Click Accept.
- 8. If two chips have been inserted, users will have the option of which chip to run first. Click **Start Run** on the chip to be run first (**Figure 22**).

Select First Run	
LOCATION P1 SERIAL NUMBER RI5K2UWNPOAWNNWU CHIP NAME Demo Test	LOCATION P2 SERIAL NUMBER NM53CHWNPPMO6HHG CHIP NAME Demo Test 2
Start Run	Start Run
Car	ncel

Figure 22. Select First Run screen.

- 9. Saphyr starts the following process:
 - Chip Registration (approximately 5 minutes)
 - DNA Loading Optimization (approximately 1 hour)
 - Scanning
- 10. During scanning, the user can view the run metrics and most recent images on the **Run** dashboard, as seen in **Figure 23**. Run metrics are also available to view in Bionano Access. **NOTE:** The color of the **Progress** bar indicates the information seen in **Table 5**.



Table 5. Progress bar indicators.

Color	Description
Blue	Flowcell is running
Green	Flowcell has completed running
Yellow	Flowcell has been cancelled and is stopping
Red	Flowcell has stopped due to an error

1. Clicking on one of the flowcells expands the details of the run metrics for that flowcell. In the expanded view, users can see the per-scan throughput and map rate metrics, as shown in **Figure 24**.

saphyr		
Run > Scanning		
CHIP SERIAL NUMBER VW7UCBGNPNWR3NWU CHIP NAME VW7U	Prep 1 SAMPLE: VW7U ENZYME1: DLE-1 + Green 01 FN RATE: 9.4% FP RATE: 4.1%	« FC1
52 FC1 REM: 48.8hr 988 N50: 294 LD: 15.8 MR: 88.5%	60 50 40 30 20 10 10	-
44 FC2 % REM: 49.1hr 789 N50: 286 LD: 15.8 MR: 88.4%	$ \begin{array}{c} \overrightarrow{b} & \overrightarrow$	 17
47 FC3 % REM: 49.5hr 329 N50: 304 LD: 16.2 MR: 83.1%	100 100 100 100 100 100 100 100	

Figure 24. Run > Scanning screen.

2. When the chip run is complete, if there is another chip ("pending") in the instrument, it will automatically start running. The pending chip will be shown on the bottom left of the screen, as in *Figure 25*.

Pending Run:		



3. Once all chips on the instrument have been completed ICS will return to the **Home** screen. Check the **Run Info** icon **for** run details.

Chip and Flowcell States

Depending on the situation, the chip(s) and their respective flowcells may be in various states, shown in **Table 6**.

Table 6. Chip states.

Chip State	Description
Running	A chip in the instrument is currently running (data is being collected on one or more flowcells)
Complete	A chip has finished its run. This means that all the associated flowcells have either reached their throughput or time limit, an issue has occurred (see flowcell states), or the user has stopped the run.
Pending	A chip is ready to run (loaded into the instrument, and the user has configured a run), but is awaiting the completion of a currently running chip.
Unscheduled	A chip is loaded into the instrument, but the user has not configured a run.

When a chip is running, the flowcells associated with the chip can be in any of the following states (Table 7):

Flowcell State	Description	lcon
Loading	Sample is being loaded in preparation for scanning.	4
Scanning	The flowcell is being imaged.	\mathbf{O}
Cleaning	Post-scanning cleaning is taking place.	*
Complete	Flowcell has achieved its throughput target or runtime limit as defined in the Run Configuration.	\checkmark
Evaporated	The instrument is unable to load DNA in the flowcell due to loss of contact. See the Pausing a Run to Rehydrate the Chip section below for more information.	₩
Error	An error has occurred that has prevented the instrument from collecting data.	
Cancelled	The flowcell was cancelled by the user and will stop when possible.	8
Paused	The flowcell is temporarily paused by user request.	-
Waiting	The flowcell is waiting for instrument resources to become available to load or scan.	

Table 7. Flowcell states.

NOTE: When all flowcells cease to be in a Loading, Scanning, Cleaning, or Waiting state, the chip state then changes from **Running** to **Complete**.

Pausing a Run to Insert a New Chip and/or Remove a Completed Chip

NOTE: For Instruments with Dual Chip capability only

To support continuous data collection, a run may be paused to swap out a chip that has already completed with a new chip.

1. Press the Pause icon at the top left of the user interface (Figure 26).



Figure 26. Pause icon.

2. When asked whether a run is desired to be paused, click **Yes** (**Figure 27**). The current scan will complete before the run is paused. This may take up to 15 minutes.



Figure 27. Pause Run screen.

3. Users will be prompted to insert and remove chips. Open the sample door to add and/or remove chips as described in steps 2-7 of the *Insert Saphyr Chip Consumable* section above.

WARNING: Do not remove the chip that is currently running if it is desired to continue running. Press **Resume** when the new chip has been inserted (see **Figure 28**).

Insert/Remove Chi	ps	
LOCATION PI PI SINGL LAMPE RISKQUWNPOAMNMUL CHIP WAT Demo Tast SINT Running FC1 6.544 NG 247 LC 1152 MC 722% FC2 7.544 NG 272 LC 1151 MC 90.54		LOCATORI P2 CHP SEAL RUMBIR None State: Empty
	Resun	ne

Figure 28. Insert/Remove Chips screen

• If the Active Chip is removed (from the instrument or moved to another platform) ICS will detect the move and will prompt a warning indicating that the current run will be stopped if the user continues (**Figure 29**).



Figure 29. Chip run will be stopped screen

- In the Configure Run screen (Figure 30 and Figure 31), confirm the correct user in the User field; otherwise update the username. Individuals with "User" privileges can only run experiments that they have created. To update the username, click on Change next to the current user. Then enter the correct username and password. This is the same username and password used to log in to Bionano Access.
- 2. For systems connected to Access 1.7 and newer version, select the **Chip Name** desired to run from the dropdown list.

Chip Location:				
Serial Number:				
Part Info:				
Chip Name:		+ \$]	
Jser:	bionanoadmin	Change		
Advanced				
				Remove Chips

Figure 30. Configure Run Screen in Access 1.6.

3. For systems connected to Access 1.6 and older version, select the **Experiment** desired to run from the dropdown list (**Figure 31**).

Chip Location:			
Serial Number:			
Part Info:			
Experiment:		• Refr	esh
Chip Setup:			•
User:	bionanoadmin	Char	nge
Advanced			
			Remove Chips

Figure 31. Configure Run Screen in Access 1.7.

4. Review the runtime and throughput target parameters for each of the flowcells. These values will default to the values that were entered into Bionano Access during the experiment setup. The user may decrease the runtime or throughput as desired, as seen in **Figure 34**.

NOTE: If the chip has been run previously, the runtime limit and throughput target may be modified. See *Appendix B* for details.

Configure Run				
Chip Location: P1	✓ Flowcell 1	✓ Flowcell 2	✓ Flowcell 3	
Serial Number: ZPLEXKWNPMXAHNWU	Molecule Set: Sample: Blank	Molecule Set: Sample: Blank 3	Molecule Set: Sample: Blank 3	
Part Info: 20366	Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	
Chip Name: Example • +	Completion Criteria:	Completion Criteria:	Completion Criteria:	
User: bionanoadmin Cha	nge 65 hr 🗘 Runtime Limit	65 hr 🗘 Runtime Limit	65 hr 🗘 Runtime Limit	
Advanced	1,500 Gbp 🗘 Throughput Target	1,500 Gbp 🗘 Throughput Target	1,500 Gbp 🗘 Throughput Target	
	Projected Total: 1,500 Gbp	Projected Total: 1,500 Gbp	Projected Total: 1,500 Gbp	
			<u> </u>	
	Flowcell 1 Molecule Set: Sample: Blank Enzyme: DLE-1- Backbone: Backb	Green 01 one Blue 01		
	Molecule Set: Sample: Blank Enzyme: DLE-1	one Blue 01		
	Molecule Set: Sample: Blank Enzyme: DLE-1- Backbone: Backb Completion Crit	one Blue 01		
	Molecule Set: Sample: Blank Enzyme: DLE-1 Backbone: Backb Completion Crit 65 hr	one Blue 01 teria:		

Figure 32. Expanded Configure Run screen

- 5. Click Accept.
- 6. The current run will resume, first verifying chip alignment of the chip that was previously running. The newly placed chip will be put into a pending state.

Pausing the Run to Rehydrate the Chip

If the sample level becomes too low during a run, the ICS will notify the user that there is a sample contact problem and will not continue to run that flowcell (**Figure 33**).



Figure 33. User Notification screen.

- 1. To pause a run that has flowcell(s) with a sample contact problem, follow the instructions above in the *Pausing a Run to Insert a New Chip and/or Remove a Completed Chip* section.
- 2. Remove the chip from the instrument and top off ALL flowcells with nuclease-free water. This includes adding water to the hydration reservoir if the water there has evaporated completely.

NOTE: Make sure to keep track of which plugs are in which flowcells to prevent cross-contamination.

3. Resume the run by placing the chip back into the same position in the instrument and following the instructions in the *Pausing a Run to Insert a New Chip and/or Remove a Completed Chip* section.

NOTE: If all the flowcells have a sample contact issue, then the instrument will move on to the next chip if one is available. If no chips are pending, the instrument will return to the start menu. Once the chip has been re-hydrated, the chip can be rerun up to the runtime limit by starting a new run. A new experiment in Access does not need to be defined; however, in Access a new chip run will be created that is associated with that experiment. This may require the user to merge the molecules files. See the *Bionano Access Software User Guide* (CG-30142) for more information.

Stopping the Run

 To stop the instrument run before it reaches the limits that were set at the beginning of the run, press the Stop icon at the top left of the user interface (Figure 34). Otherwise, the run will continue until the throughput or time limit is met.



Figure 34. Stop icon.

2. When this button is pressed, users will be asked to confirm, as seen in **Figure 37**.



Figure 35. Confirm Stop screen.

3. If there is no pending run, then selecting yes will stop the run on that chip. If there is a pending run, users will be asked to confirm if all chips or just the one currently running should be stopped (**Figure 36**).

STOP ALL?					
Do you	also want t	o stop the p	ending run?		
	yes	no	cancel		

Figure 36. Stop All? Screen.

4. Select **Yes** to stop both chips. Select **No** to stop the current chip but allow the pending chip to begin. Select **Cancel** to return to the current chip run.

Remove Chip(s)

In addition to the *Pausing a Run to Insert a New Chip and/or Remove a Completed Chip* section above, users can remove the Saphyr Chip[®] consumable from the Saphyr instrument when the run is successfully complete or when the run is stopped.

- 1. From the Home screen, click Remove Chip.
- 2. Once the Wait icon is no longer showing, manually lift the sample door of the instrument.
- 3. Press the **Press** button to release the bundle arm.
- 4. Remove the chip with the clip attached.
- 5. Lower the bundle arm and close the sample door after removing the chip.
- 6. From the Main Menu, click Chip is Removed.
- 7. Close the Instrument Control Software program.
- 8. [Optional] Turn off the instrument.

The Saphyr instrument is designed to run continuously. To set up another chip run, repeat these tasks:

- Create an Experiment
- Insert Saphyr Chip[®] consumable
- Configure Run
- 9. After removing the chip, detach and clean the Saphyr clip and plugs as soon as possible. See the *Maintenance* section for details.

Analyze Data

Data Transfer for Analysis

After the instrument scans the chip for about 2-3 minutes, users can monitor the run progress and view quality metrics in real time in Bionano Access.

Following the completion of a run, users can perform various analyses. For more details on Bionano Access software analysis and visualization features, see the *Bionano Access Software User Guide* (CG-30142).

Saphyr Assure

Saphyr Assure is a remote service provided by Bionano that enables the ICS to transmit instrument health metrics, run diagnostics and configuration information to enable Bionano to proactively provide support to customers. Saphyr Assure enables users to install software updates for Saphyr ICS, allowing for quick and easy updates. Saphyr Assure also manages Windows updates, checking to ensure the system is always running with the latest validated updates.

To enable the instrument health monitoring feature of Saphyr Assure, the Bionano Access administrator is required to opt-in. The administrator has the option to enable the Saphyr Assure and Compute on Demand service, or if the user is not using Compute On Demand, they can opt-in to only the Saphyr Assure service. To opt-in, log into Bionano Access as an administrator, click **Settings** and then click **System Settings** (**Figure 37**).

bionano GENOMICS (ome > Settings > System Se	attings	Access
	System Level Services	Use
	Compute On Demand & Saphyr Assure The Consult On Demand service provides pay per use eletitic computing resources to supplement and accelerate your analysis needs. Together with the Saphyr Asare service you can monitor the health of your entire workflow. This is the recommended option.	
		Pas
	Saphyr Assure Only Use this option if you want to ensure optimal performance of your instrument, but are not	
	Interested in the Compute On Demand service.	p

Figure 37. System Settings window.

Updating Software

For systems that are on ICS 5.1 or above, software can be updated through the ICS User Interface (UI). When a software update is available, the **Saphyr Assure** icon will turn orange

- 1. Click on the **Saphyr Assure** icon
- 2. Select Saphyr ICS Updates, shown in Figure 38.



Figure 38. ICS System Updates.

- 3. Select Install.
- 4. The system will alert the user that a restart is needed to complete the installation. Select **Yes** to proceed.

Logging off and System Administration

To either restart the Instrument Controller 🔁 or to turn off the Instrument Controller 😃, click the applicable

icon at the top right of the screen. Users with System Administrator credentials will also see an icon **that will** allow them access to the **System Settings** menu as well as the Windows login.

Maintenance

Clean the Saphyr Clip Plugs

Clean the clip plugs before and after each use. Use gloves when cleaning clips.

- 1. Use a lint-free paper saturated with 70% alcohol to remove any visible dirt or debris on all sides of the clip plugs.
- 2. Air dry the clip plugs before using them again. Do not use canned air. Always position clip inserts on their side or upside down, as shown in Error! Reference source not found.. Do not place them on the bench with the electrodes down.
- 3. Store the clip in a clean case with a cover when not in use.





Figure 39. Electrodes air-drying after cleaning.

Clean the Saphyr Seal

Clean the seal before and after each use. Use gloves when cleaning seals.

- 1. Use a lint-free swab saturated with 70% ethanol to remove any visible dirt or debris on all sides of the seal. If a lint-free swab is not available, an alternative solution is to spray with 70% ethanol.
- 2. Air dry the seal before using it again. Do not use canned air.
- 3. Store the seal in a clean case with a cover when not in use.

Clean the Objective Lens

We recommend that users clean the Objective lens every 3 months or whenever a spill occurs on the instrument stage.

- 1. Launch the Instrument Control Software.
- 2. Click the Help icon at the top right of the screen.
- 3. In the Help screen, click Clean Objective, then click the Clean Objective button as seen in Figure 40.



Figure 40. Help screen.

4. The instrument will move the stage into a position that will allow the objective to be accessible. Once this has completed, click **Begin Cleaning**. A message will appear, as seen in **Figure 41**. Click **OK**.





- 5. The instrument will power down; manually lift the sample door. **NOTE:** Users will need to apply some force to lift the sample door.
- Apply one or two drops of pure methanol (or ethanol, recommend 100%) to the head of a cleaning swab or the tip of folded optical paper provided with the Saphyr Accessories or Saphyr Preventive Maintenance kit.
- 7. Clean the objective's front lens in one motion while lightly applying some weight to the swab or the optical paper to allow the flat head to thoroughly clean the objective (**Figure 42** and **Figure 43**).
- 8. If additional cleaning is required, use a new swab or a new optical paper each time.



Figure 42. Lens cleaning using folded optical paper.



Figure 43. Lens cleaning using swabs.

Air Filter

The air filter should be replaced every 3 months. Air filters are included in the Accessory Kit that comes with the Saphyr instrument. Additional air filters are supplied to the customer when the annual preventative maintenance is performed.

- 1. Turn off the instrument.
- 2. On the back of the instrument pull the air filter cover (**Figure 44**) firmly away from the instrument to release the magnets that hold the cover in place.



Figure 44. Air filter cover.

- 3. Remove the air filter from its cover.
- 4. Insert a new air filter (Figure 45). Make sure the **Air Flow** arrow symbols are facing up (air flows into instrument).



Figure 45. Replacing the air filter.

5. Attach the cover with the new air filter to the instrument.

Appendix A

Troubleshooting

1. Issue: Unable to log into ICS

Possible causes:

- Incorrect password was entered.
- The Access server is not active.

Solution:

- · Check if you can log into Access.
- Refer to page 8 of the Bionano Access Software User Guide if you forgot your password.
- Check whether the Access service status is active. If you can open the Bionano Access login page from a web browser the service is active.
- Reboot the Instrument Controller.
- 2. Issue: Data service is unable to connect to the Bionano Access[®] server.


Solution: Check if the Access webserver is active. Confirm that the cable connection between the instrument controller and Access is secure. If the issue is not resolved, contact Bionano Support at support@bionano.com.

3. Issue: Instrument does not initialize (Figure 46).



Figure 46. Image of initialization status of each instrument component.

Possible cause: This is more likely a hardware issue.

Solution:

- Make sure to close ICS first.
- Switch off the instrument and wait for at least 15 sec before switching back on.
- Wait approximately 30 seconds. Restart ICS and wait for system to re-initialize.
- If this does not resolve the issue, contact Bionano at <u>support@bionano.com</u>.
- 4. Issue: Barcode Read Error (Barcode Cannot be Read)

Possible causes:

- Chip not well seated.
- Barcode (Q-code) is damaged.
- Barcode reader failure
- Barcode reader is misaligned.

Solution:

- Reseat the chip.
- Ensure there is no physical damage to the chip that may prevent it from fitting well into the chip slot.

- Retry Barcode scan. If it fails again, then try the following:
- Remove the chip. Ensure there is no physical damage to the chip that may prevent it from fitting well into the chip slot.
- Ensure that the chip sits flat and fits within the groove on the platform. Restart the run.
- Contact Bionano at support@bionano.com if this happens with more than one chip.
- 5. Issue: Disk Full, or not enough disk space (Figure 47).



Figure 47. Image of error indicating that the disk space is full.

Possible cause: The instrument control PC hard drive only holds one run. If you try to save images on a run and have not transferred or removed the images from the previous run, you will see this error message.

Solution: The software will prompt you to delete the previously saved run if you start a new run.

- 1. Go to the main menu of ICS and select 'Data administration.
- Use the tick box (right-hand side) to select run(s) to delete from the run list and click the 'Delete' button.
- 6. Issue: No electrical contact could be established. ICS message: "Check sample fluid level."

Possible cause: Sample Evaporation

Solution:

- Pause the run; Remove the chip; Remove the clip from the chip.
- Rehydrate the chip with nuclease-free water.
- Attach the clip to the chip; Reinsert the chip into the instrument; Resume the run.
- 7. Issue: Bionano Access dashboard is not being updated in real time as the run proceeds

Possible cause:

• Connection is lost between the Bionano Access Server and the Saphyr Instrument Controller.

Solution:

• Confirm that the cable linking both components is plugged in at both ends.

- Refresh Bionano Access webpage (allow some time for the run data to populate in Access).
 NOTE: When loss of connection occurs, the instrument keeps running and data is stored locally on the Instrument Controller. Transfer will restart when the connection is restored.
- 8. **Issue:** Map rate is zero

Possible causes:

- Reference not provided.
- Wrong reference provided.
- Wrong labelling enzyme selected.

Solution:

- Wait for the run to complete and for the sample's molecules object to appear in the corresponding project (do not stop the run).
- Make sure the correct reference is loaded in the Bionano Access software. If it is not, refer to the *Edit Job* section in the *Bionano Access*[®] *Software User Guide* (CG-30142) to change the reference.
- 9. Issue: No throughput

Possible causes:

• Evaporation might have occurred.

Solution:

- Remove the chip.
- · If there is no liquid, re-hydrate the chip and re-start the run.
- If the issue is not resolved, contact Bionano at support@bionano.com.

10. Issue: Chip Throughput Max Reached (Figure 48).

 ✓ Flowcell 1 Molecule Set: Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01 Completion Criteria: 65 hr ⁺ Runtime Limit 1,000 Gbp ⁺ Throughput Target Projected Total: 5,000 Gbp 	 ✓ Flowcell 2 Molecule Set: Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01 Completion Criteria: 65 hr ⁺, Runtime Limit 500 Gbp ⁺, Throughput Target Projected Total: 5,000 Gbp 	 Flowcell 3 Molecule Set: Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01 Completion Criteria: 65 hr \$ Runtime Limit 1 Gbp \$ Throughput Target Projected Total: 5,000 Gbp Flowcell 3 reached maximum throughput of 5,000.
---	---	---

Figure 48. Error message showing flowcell 3 has reached maximum throughput.

Solution:

- Uncheck the box next to the flowcell that reached max throughput.
- Continue to run the other flowcells to collect more throughput, if desired.

Export Tier 1 Run Diagnostics

If there is an issue with a flowcell run that needs to be addressed by Bionano Support for troubleshooting, a small diagnostic package can be generated as Tier 1 Run Diagnostics. If you are connected to the Saphyr Assure service, these diagnostic files can be transmitted directly to Bionano Support through the ICS.

- 1. At the top of the window, click on the icon.
- 2. A list of recent chip runs will be displayed. Select the serial number of the chip associated with the run containing the diagnostic information you would like to export (**Figure 49**).

	ZZK6CAWNPMQFNNWU 11/13/2020 4:30 PM to 11/13/2020 11:47 PM	
	Flowcell 1 11/13/2020 4:30 PM Request Support 💿	
	Flowcell 1 scan 13 reached target throughput of 400. Collected 429.3 Gbp.	
	Flowcell 2 11/13/2020 4:30 PM Request Support	
	Flowcell 2 scan 13 reached target throughput of 400. Collected 419.4 Gbp.	
	Flowcell 3 11/13/2020 4:30 PM Request Support	
	Flowcell 3 scan 12 reached target throughput of 400. Collected 401.0 Gbp.	
	• 76KMCIWNPMTVNNWU 11/13/2020 9:22 AM to 11/13/2020 4:29 PM	
\sim	L2JGC2WNPO2FNNWU 11/12/2020 2:55 PM to 11/12/2020 3:36 PM	
nto	L2JGC2WNPO2FNNWU 11/12/2020 2:47 PM to 11/12/2020 2:54 PM	
an Into	L2JGC2WNPO2FNNWU 11/10/2020 8:22 PM to 11/11/2020 7:10 AM	
Y	Acknowledge Errors Cancel	

Figure 49. Chip serial number.

3. To troubleshoot, click on the arrow to the right of **Request Support** for the flowcell desired (**Figure 50** and **Figure 51**).





	ZZK6CAWNPMQFNNWU 11/*	3/2020 4:30 PM to	11/13/2020 11:47	PM 🔺
	Flowcell 1 11/13/2020 4:30 PM		Request Support	•
	Contact name (requred)			
	Contact email (required)			- 1
	Contact phone (optional)			
	Operator name (optional)			
	Description (required)			
Jfo				
Run Info				-
Ru	Acknowledge Errors Cancel			

Figure 51. Request Support form.

- 4. In the dropdown that appears, fill in the applicable information and press Submit Request. This will transmit the diagnostic information to Bionano Support. The Contact Name, Contact Email, and Description fields are required for submission. The Operator Name will be automatically populated with the User ID of the person logged into ICS.
- 5. If the submission begins but does not complete due to a network connection problem, it will automatically be re-submitted later.

For users who have not opted in to Saphyr Assure and do not have network access to enable connection to Saphyr Assure, the diagnostic package must be saved and emailed to Bionano support. Follow steps 1 and 2 above, then do the following:

- 6. For systems in kiosk mode:
 - Click on the arrow to the right of Export Diagnostics for the flowcell desired to troubleshoot (Figure 52).



Figure 52. Export Diagnostics.

- Insert exactly one USB flash drive into the Instrument Controller and wait 30 seconds for the device to be recognized.
- In the dropdown that appears, fill in the applicable information and press Export (Figure 53).

				물
			•	
	Flowcell 1 10/29/2020 5:14 PM	Export Diagnostics		
	Contact name (requred)		ш	
	Contact Name		Ш	
	Contact email (required)			
	ContactEmail@Domain.com			
	Contact phone (optional)			
	555-555-5555			
	Operator name (optional)			
	Operator Name			
	Description (required)			
	Help Needed because			
Jfc				
un Info	Submit Request Export			
L L				



7. ICS will generate the diagnostic file and save it into the root of the USB drive. ICS indicates that the export is complete by displaying a green checkbox (**Figure 54**).



Figure 54. Export complete indicator.

- Remove the USB drive, insert it into another computer and email to <u>support@bionano.com</u>. The file size is intended to be small enough to email as an attachment using most standard email service providers.
- 8. For systems not in kiosk mode:
 - Click on the arrow to the right of **Export Diagnostics** for the flowcell desired to troubleshoot.
 - Select the location to save the Saphyr Archive (.sar) file, and then click OK.
 - In the dropdown that appears, fill in the applicable information and press **Export**.
 - When ICS completes compressing the files, navigate to the location of the file, then send it to <u>Bionano</u> <u>Support (support@bionano.com)</u>. The file size is intended to be small enough to email as an attachment using most standard email service providers.
- 9. Once the file has been transmitted, delete the exported file to prevent filling the hard drive.

Export Tier 2 Run Diagnostics

If the Tier 1 diagnostic package is insufficient to address the issue, Bionano will need to collect additional data from the run. For users connected to Saphyr Assure, this will occur without any intervention from the user.

Users not connected to Saphyr Assure will be instructed by Bionano Support to assist in providing the additional information. The user will be provided a Diagnostic Request File (.drf) from Bionano Support.

For systems in kiosk mode:

- 1. Save the .drf file on a USB stick at the top level (root) of the directory structure. The USB stick may only have one .drf file in this location. Attach only one USB stick at a time to the Instrument Controller.
- 2. Click on the **Run Info** icon. Select **Open Diagnostic Request** to import a .drf file.
- 3. Once the file is imported, ICS will generate a .sar file and save it to the USB stick. When the export is complete, remove the USB stick from the ICS Controller and transfer the .sar file to Bionano Support.

For systems not in kiosk mode:

- 1. Save the .drf file in the instrument hard drive, and double click the .drf file. This will open ICS (if it is not already open) and prompt the user to save a .sar file.
- 2. Choose a location on the Instrument Controller and save the file.
- 3. Once the file is saved, transfer it to Bionano Support.
- 4. Once the file is transmitted, delete the .sar file to ensure that enough hard drive space remains on the Instrument Controller for normal operation.

Export / Upload Data Configuration

Users can export the instrument data configuration to their computer, such as log and calibration data from the instrument, and then send to <u>Bionano Support</u> for troubleshooting. For customers who are connected to Saphyr Assure, the configuration can upload directly to Saphyr Assure.

1. From the **Home** screen, click the **Help** icon at the top-right corner of the screen. A dialog box appears (**Figure 55**).



Figure 55. Export Configuration.

- 2. For users not connected to Assure, click **Export Configuration**. For users connected to Assure, skip to number 9.
- 3. [Optional] Select the **Full configuration** checkbox to collect additional system information. This option increases the export file size and the time it takes to compress the files.
- 4. Click Compress and Export Configuration.
- 5. Type the file name.
- 6. Select the location to save the ZIP file. If your system is in kiosk mode, you will not be prompted to select the location, the file will be saved directly to a USB device that has been inserted into the Instrument Controller. Only one USB drive may be inserted into the Instrument Controller.
- 7. Click Save.
- 8. When ICS completes compressing the files, navigate to location of the ZIP file, and then send it to Bionano Support.
- 9. For users connected to Assure, click **Upload Configuration**. Fill out the optional and required information and click **Compress and Upload Configuration**.

Appendix B

Rerunning a Chip

ICS allows for user flexibility in collecting data to accommodate various workflow issues and scenarios. Each flowcell in the chip has an associated maximum accumulated throughput and maximum accumulated runtime. The chip can be re-run as many times as desired to collect data up until either of these limits are reached. See the *Chip Specification Document* (CG-30256 and 30264) for the maximum accumulated throughput and accumulated runtime associated for a particular chip part number.

There are a few situations where a chip may need to be re-run. These include failure to successfully achieve the desired throughput target in the initial run due to:

- Loss of power
- Instrument Control Software or operating system crash.
- Loss of electrode contact to all flowcells from evaporation, resulting in ICS stopping the chip run prematurely.
- Data collection rate insufficient to achieve target within the allocated time.

In addition, if the desired throughput was collected, the user can rerun the chip to collect more data on the desired flowcells provided the maximum accumulated throughput and maximum accumulated runtime have not yet been achieved.

Notes on safe handling the chip for re-running:

- Re-hydrate the flowcells prior to re-running chip (see steps 9-13 of the appropriate section in *Prepare Chip*.
- It's recommended that the chip be re-run immediately after the initial run, within 8 hours of the previous run being completed.

NOTE: If the same chip is run multiple times, it will have multiple chip runs and multiple molecules objects in the Bionano Access software. See the *Bionano Access Software User Guide* (CG-30142) for instructions.

Example scenarios for re-running a chip

NOTE: Actual Saphyr Chip[®] consumable specifications are provided on the product specifications for your chip. The following numbers are for example only.

Scenario 1: Desired throughput targets not achieved in the initial run due to insufficient data generation rate (**Table 8**).

	Run Conditions Set by the User in the First Run		What Actually Happened Durir	ng First Run
Flowcell	Desired Throughput Target	Desired Runtime Limit	Throughput Collected During the Run	Total Flowcell Runtime
1	5000 Gbp	65 hr	4000 Gbp	65 hr
2	5000 Gbp	65 hr	4500 Gbp	65 hr
3	5000 Gbp	65 hr	5000 Gbp	65 hr

Table 8. In this example, only flowcell 3 was able to achieve its desired throughput during the allocated runtime:

If the user desires to re-run the chip to collect more data on flowcells 1 and 2, the **Configure Run** screen will appear as in **Figure 56**. Here, the user can re-run the chip to collect an additional 1000 Gbp and 500 Gbp in flowcells 1 and 2, respectively. Since flowcell 3 has achieved the maximum accumulated throughput limit of 5000 Gbp, it can no longer be run.

✓ Flowcell 1	✓ Flowcell 2	Flowcell 3
Molecule Set:	Molecule Set:	Molecule Set:
Sample: Sample Green	Sample: Sample Green	Sample: Sample Green
Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01
Backbone, Backbone Blue of	backbone, backbone blac of	Backbone, Backbone Blac of
Completion Criteria:	Completion Criteria:	Completion Criteria:
65 hr 🗘 Runtime Limit	65 hr 🌲 Runtime Limit	65 hr 🗘 Runtime Limit
1,000 Gbp 🗘 Throughput Target	500 Gbp 🌲 Throughput Target	1 Gbp 🗘 Throughput Target
Projected Total: 5,000 Gbp	Projected Total: 5,000 Gbp	Projected Total: 5,000 Gbp



Scenario 2: Desired throughput targets not achieved in the initial run due to loss of contact on all flowcells (Table 9).

 Table 9. In this example, all three flowcells independently lost contact, resulting in ICS stopping the chip run prematurely. ICS will only stop the chip run when all flowcells have lost contact.

Run Conditions Set by the User in the First Run		What Actually Happened During First Run		
Flowcell	Desired Throughput Target	Desired Runtime Limit	Throughput Collected During the Run	Total Flowcell Runtime
1	1300 Gbp	65 hr	1200 Gbp	28 hr (Loss of contact)
2	1300 Gbp	65 hr	1220 Gbp	32 hr (Loss of contact)
3	1300 Gbp	65 hr	1190 Gbp	25 hr (Loss of contact)

If the user desires to re-run the chip to collect more data, the **Configure Run** screen will appear as in **Figure 57**. Here the user can re-run the chip to collect additional data on all three flowcells.

✓ Flowcell 1	✓ Flowcell 2	✓ Flowcell 3	
Molecule Set:	Molecule Set:	Molecule Set:	
Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	
Completion Criteria:	Completion Criteria:	Completion Criteria:	
65 hr 🗘 Runtime Limit	65 hr 🗘 Runtime Limit	65 hr 🗘 Runtime Limit	
100 Gbp 🗘 Throughput Target	80 Gbp 🗘 Throughput Target	110 Gbp 🗘 Throughput Target	
Projected Total: 1,300 Gbp	Projected Total: 1,300 Gbp	Projected Total: 1,300 Gbp	

Figure 57. Configure Run screen.

Scenario 3: Desired throughput target insufficient (Table 10).

Table 10. In this example, the user determines that the amount of data collected was insufficient and wants to collect more.

 This is allowed provided the total accumulated throughput is within the allowed limits. For example, the initial run completed with the following:

Flowcell	Run Conditions Set by the User in the First Run		What Actually Happened During First Run	
	Desired Throughput Target	Desired Runtime Limit	Throughput Collected During the Run	Total Flowcell Runtime
1	320 Gbp	65 hr	320 Gbp	12 hr
2	320 Gbp	65 hr	320 Gbp	13 hr
3	320 Gbp	65 hr	320 Gbp	11 hr

However, after the run, the user desires to collect a total of 400 Gbp on each flowcell. In this case the user would fill out the **Configure Run** screen as follows (note that ICS provides a projected total of 400 Gbp for each flowcell) (**Figure 58**):

✓ Flowcell 1	✓ Flowcell 2	✓ Flowcell 3
Molecule Set:	Molecule Set:	Molecule Set:
Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01	Sample: Sample Green Enzyme: DLE-1+Green 01 Backbone: Backbone Blue 01
Completion Criteria:	Completion Criteria:	Completion Criteria:
65 hr 🗘 Runtime Limit	65 hr 🗘 Runtime Limit	65 hr 🗘 Runtime Limit
80 Gbp 🗘 Throughput Target	80 Gbp 🗘 Throughput Target	80 Gbp 🗘 Throughput Target
Projected Total: 400 Gbp	Projected Total: 400 Gbp	Projected Total: 400 Gbp

Figure 58. Configure Run screen.

Appendix C

Instrument Control Software (ICS) Icons

The icons at the top right-corner of the **ICS** screen, described in **Table 11**, indicate the Saphyr instrument status. **Table 12** indicates the status of recent Runs.

lcon	Color	Description
	Blue	The instrument is working normally.
	Flashing Green	The instrument is initializing.
	Flashing Red	The instrument encountered an error. Contact Bionano Support for assistance.
	Flashing Bright Blue	The instrument door is open. If the instrument door is closed, there is an issue with the safety interlocks, please contact Bionano Support.
	Flashing Amber	The instrument is not receiving power or is not turned on.
	Purple	The instrument hardware is not correctly configured. Contact Bionano Support for assistance.
	Flashing Dark Orange	There is an interlock fault. Contact Bionano Support before attempting to start the run.

Table 11. The Saphyr instrument icon indicates the status of the Saphyr instrument.

Run Info

Table 12. The	run info icon	indicates the s	tatus of recent runs.
---------------	---------------	-----------------	-----------------------

Icon	Color	Description
	Blue	There are no run errors, or any previous run errors have been acknowledged.
	Red	There is run error that has not yet been acknowledged.

Bionano Access Connection Status

The **Bionano Access Connection** icon indicates that there is a network connection between the Saphyr Instrument Controller and the Bionano Access server (**Table 13**).

Table 13. The instrument must be connected to the Bionano Access server before it can begin a run. If connection fails while the instrument is scanning, ICS will cache run data locally and will automatically resume once the connection is reestablished.

lcon	Color	Description
모	Blue	The instrument control software is connected to the Bionano Access server.
문	Amber	The instrument control software is unable to connect to Bionano Access server.

Saphyr Assure Status

Table 14. The Saphyr Assure Status icon indicates the status of the Saphyr Assure connection and any available updates.

lcon	Color	Description
	Green	Saphyr Assure is connected and health monitoring is enabled.
⊜	Blue	Saphyr Assure is connected and health monitoring is disabled.
⊠	Red	There is a connection error between ICS and Saphyr Assure.
	Amber	Update(s) are available for either ICS or Windows

lcon	Color	Description
$\overline{\mathbf{x}}$	Blue	Disabled / Establishing Connection

Remote Support

Table 15. The Remote Support icon indicates whether the system is connected to remote support.

lcon	Color	Description
₽	Green	Remote support is enabled.
÷	Blue	Remote support is disabled.

Appendix D

Instrument Initialization

The hardware components of the Saphyr instrument initialize upon startup of the Instrument Control Software. During initialization, the **Instrument Status** icon will flash green. Once initialization is complete, the icon will turn blue. If there is an issue with the initialization of any component, the icon will turn red. Clicking on the icon will show the initialization status of each component, as shown in **Figure 59**.



Figure 59. Instrument Status screen.

Appendix E

Data Administration

The Data Administration option from the **Home** screen allows users to manage run data that are on the Saphyr Instrument Controller. This is applicable only if you are using the option to save images.

Insufficient Disk Space

If you are using the option to save images, the data needs to be removed from the Instrument Controller before starting a new run. The Instrument Controller will only save one run at a time before prompting the user that there is not enough space remaining. When clicking **Accept** in the **Configure Run** screen, if there is a run that has not been removed, a message will appear ("Too much existing data to add another run") prompting you that you cannot proceed until data is removed.

Follow these steps:

- 1. Click OK.
- 2. At the **Configure Run** screen, click **Cancel**.
- 3. Copy off any images that you want to retain to a separate server.

NOTE: Bionano is not responsible for the integrity of the data transfer

NOTE: Do not cut or delete images from the Instrument Controller. Use the copy function only. Removing images from this directory will inhibit the ability to archive diagnostic information that may be useful for troubleshooting issues in the future. Remove images by following the instructions in the *Delete Runs* section below.

4. Once you have confirmed that your images have been transferred, proceed to the *Delete Runs* section below.

Delete Runs

After data analysis is complete, delete runs to clear space for a subsequent run. Runs that are currently in progress cannot be deleted.

WARNING: Once images have been deleted, they cannot be recovered.

- 1. From the Home screen, select Data Administration.
- 2. If there are images saved, the serial number of that chip will appear. Click Delete (Figure 60).

Saved Image Data
SERIAL NUMBER
KOB7XH6NPPJGLNWX
SIZE
39.50 GB
Delete

Figure 60. Saved Image Data screen.

- 3. When the process is complete, the screen will show no additional data that can be deleted.
- 4. Click the **Home** icon to return to the **Home** screen.

Appendix F

Upload Run to Access

The **Resubmit Run to Access** option from the **Help** screen allows users to resubmit a dataset for reanalysis. This may be used if an error in the initial analysis of the run data has occurred.

NOTE: If ICS and Access disconnect during a run, the run will continue. Access will automatically resume analysis once the connection is reestablished. In this case, there is no need to resubmit the data. The status of the data transfer to Access can be monitored using the **Data Service Status** icon.

- 1. Click **Help** in the upper right corner of the screen.
- 2. Select Resubmit Data to Access.
- 3. Select Upload Run to Access (Figure 61).

Upload Run to Access	
Submit	
Cancel	

Figure 61. Upload Run to Access screen

- 4. Select the run you wish to reanalyze from the dropdown list.
- 5. Click Submit.

6. The existing **Chip Run will be deleted from the Access database**, and a new run will be created in Access.

NOTE: The Chip Run ID will remain the same.

Appendix G

Instrument Controller Operations

- The Instrument Controller runs Windows Defender for anti-virus protection. It is recommended that no thirdparty anti-virus software (AVS) is installed. However, if a customer chooses to install a different AVS, it must only be run manually when the instrument is not running a chip. The stability of the Instrument Controller cannot be guaranteed if additional software is installed. Systems that have additional software added will not be supported.
- The Instrument Control Software (ICS) >5.1 includes an updater feature for the ICS software, Windows, and Windows Defender. Windows Updates must not be applied from within windows environment but only from the ICS feature.
- Customers should not delete any files from the Instrument Controller using Windows, as this may inhibit the Saphyr instrument's ability to archive diagnostics and may result in data loss. The only exception to this rule is the diagnostics packages generated by ICS after the packages have been safely copied off the instrument and sent to Bionano customer support.
- It is strongly recommended that the Instrument Controller is not added to the customer's Windows domain. For more information, please see CG-30292. This may cause the Windows security settings, power states, or other settings critical to the function of the instrument to be modified. Systems that have been added to a domain will not be supported
- It is strongly recommended that user only use the provided Windows User login.
- Please do not make any changes to the Instrument Controller system as any unexpected Windows configuration modification will void support, including:
 - Remove or modify any files in C:\Installs
 - Uninstall or install any programs
 - · Uninstall or install any drivers
 - Change any PC power settings
 - Change any BIOS settings
 - Change any Local Group Policy settings
 - · Add or remove user accounts
 - Change user account passwords
 - Change Ethernet adapter settings
 - Modify the Windows hosts file
 - Change any Windows Update settings

Appendix H

Instrument Disposal

This label indicates that the instrument meets the 2012/19/EU Directive on waste electrical and electronic equipment (WEEE).

Email support@bionano.com for instrument disposal in the EU.

WEEE Label



Additional Resources

The following documentation is available for download from the **Bionano Support** page.

Resource	Description
Saphyr System Safety Guides (CG-30144, 30253, 30302, 30467)	Provides information about the instrument safety considerations.
Saphyr Site Preparation Guide (CG-30145)	Provides specifications for laboratory space, electrical requirements, and environmental considerations.
Bionano Access Software User Guide (CG-30142)	Provides an overview of data analysis.
Saphyr Networking and Setup Guide (CG-30251)	Provides networking information for all Bionano computers.
Bionano Data Security Guidelines (CG-30292)	Provides information about how Bionano data is handled.

Technical Assistance

For technical assistance, contact Bionano Technical Support.

You can retrieve documentation on Bionano products, SDS's, certificates of analysis, frequently asked questions, and other related documents from the Support website or by request through e-mail and telephone.

ТҮРЕ	CONTACT
Email	support@bionano.com
Phone	Hours of Operation: Monday through Friday, 9:00 a.m. to 5:00 p.m., PST US: +1 (858) 888-7663
Website	www.bionano.com/support
Address	Bionano, Inc. 9540 Towne Centre Drive, Suite 100 San Diego, CA 92121

Legal Notice

For Research Use Only. Not for use in diagnostic procedures.

This material is protected by United States Copyright Law and International Treaties. Unauthorized use of this material is prohibited. No part of the publication may be copied, reproduced, distributed, translated, reverseengineered or transmitted in any form or by any media, or by any means, whether now known or unknown, without the express prior permission in writing from Bionano Genomics, Inc. ("Bionano Genomics"). Copying, under the law, includes translating into another language or format. The technical data contained herein is intended for ultimate destinations permitted by U.S. law. Diversion contrary to U.S. law prohibited. This publication represents the latest information available at the time of release. Due to continuous efforts to improve the product, technical changes may occur that are not reflected in this document. Bionano Genomics reserves the right to make changes in specifications and other information contained in this publication at any time and without prior notice. Please contact Bionano Genomics Customer Support for the latest information.

BIONANO GENOMICS DISCLAIMS ALL WARRANTIES WITH RESPECT TO THIS DOCUMENT, EXPRESSED OR IMPLIED, INCLUDING BUT NOT LIMITED TO THOSE OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE. TO THE FULLEST EXTENT ALLOWED BY LAW, IN NO EVENT SHALL BIONANO GENOMICS BE LIABLE, WHETHER IN CONTRACT, TORT, WARRANTY, OR UNDER ANY STATUTE OR ON ANY OTHER BASIS FOR SPECIAL, INCIDENTAL, INDIRECT, PUNITIVE, MULTIPLE OR CONSEQUENTIAL DAMAGES IN CONNECTION WITH OR ARISING FROM THIS DOCUMENT, INCLUDING BUT NOT LIMITED TO THE USE THEREOF, WHETHER OR NOT FORESEEABLE AND WHETHER OR NOT BIONANO GENOMICS IS ADVISED OF THE POSSIBILITY OF SUCH DAMAGES.

Patents

Products of Bionano Genomics may be covered by one or more U.S. or foreign patents.

Trademarks

The Bionano Genomics logo and names of Bionano Genomics products or services are registered trademarks or trademarks owned by Bionano Genomics in the United States and certain other countries.

Bionano[®], Saphyr, Saphyr Chip[®], Bionano Access, VIA[™] software, and Bionano EnFocus[™] are trademarks of Bionano Genomics. All other trademarks are the sole property of their respective owners.

No license to use any trademarks of Bionano Genomics is given or implied. Users are not permitted to use these trademarks without the prior written consent of Bionano Genomics. The use of these trademarks or any other materials, except as permitted herein, is expressly prohibited and may be in violation of federal or other applicable laws.

© Copyright 2023 Bionano Genomics, Inc. All rights reserved.