PacBio

Vega™ system

Operations guide
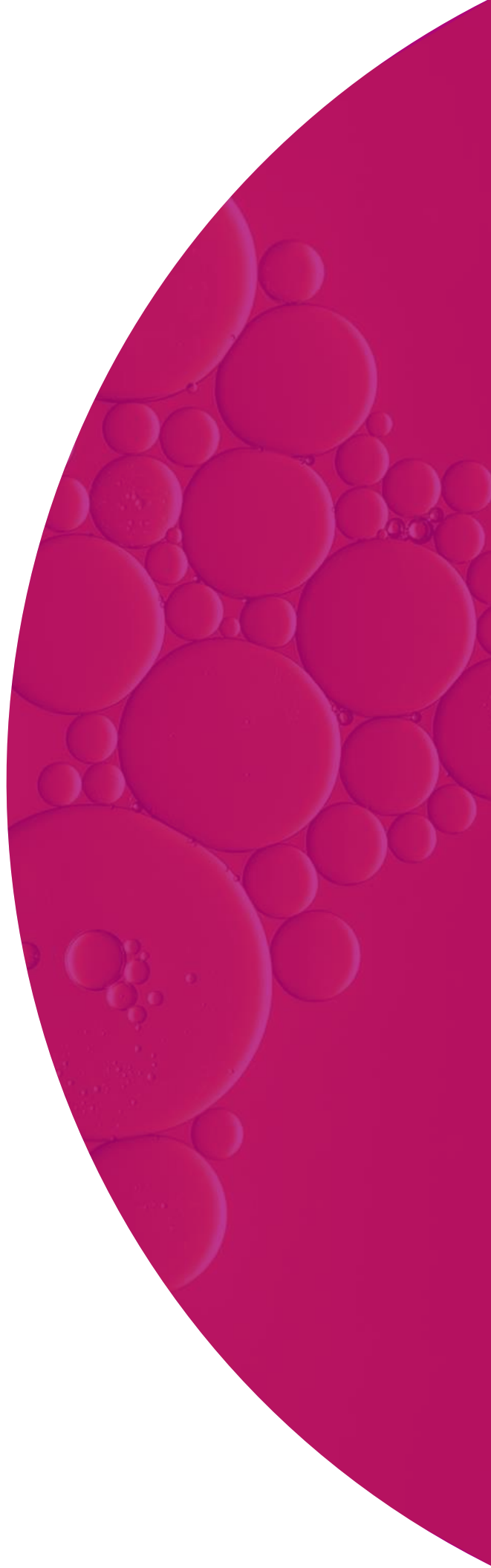
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Pacific Biosciences of California, Inc.
1305 O'Brien Dr.
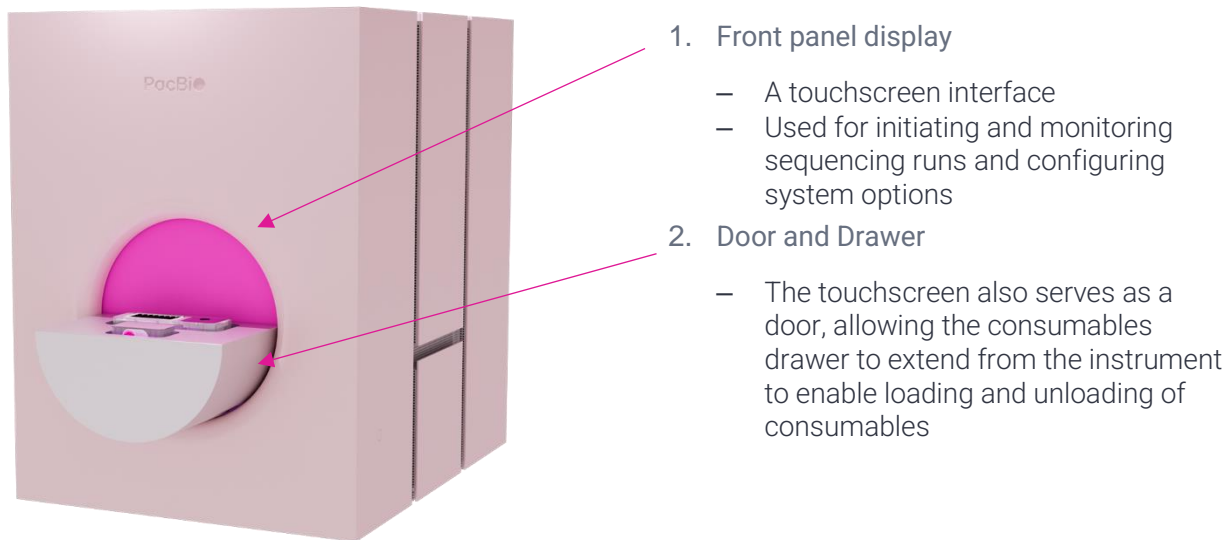Menlo Park, CA 94025
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Introduction

This guide explains how to operate the Vega system. **For information on instrument settings, sample setup, run design, user management, and Admin users, see the** *SMRT® Link user guide*.

External system overview

1. Front panel display

   – A touchscreen interface
   – Used for initiating and monitoring sequencing runs and configuring system options

2. Door and Drawer

   – The touchscreen also serves as a door, allowing the consumables drawer to extend from the instrument to enable loading and unloading of consumables

System power

To power up the instrument, ensure the main power switch at the rear of the instrument is in the On position, then press the power button located on the right-side panel of the instrument. Note that it may take a few minutes for the instrument to power up.

DANGER! ELECTRICAL SHOCK AND LASER HAZARD

Installation, maintenance, and repair are only allowed for authorized service personnel. Do **not** remove the panels of the Vega systems. Please refer to *Safety guide - Vega system* for more information.

Verifying instrument software version

1. Select the **menu** icon on the touchscreen:
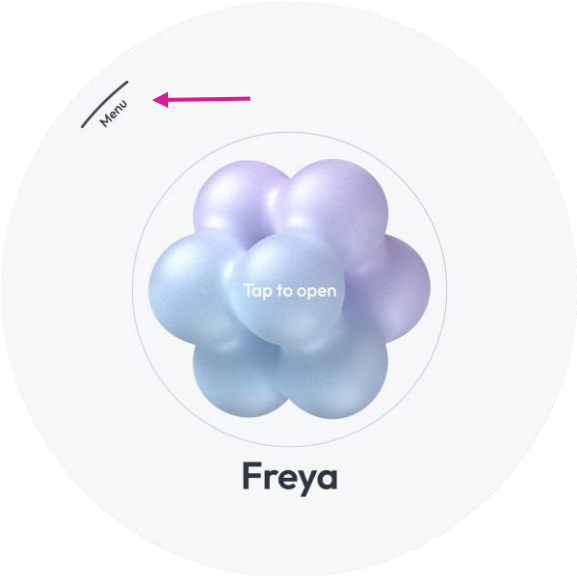
2. Select "Instrument detail":

A screen displaying the following will then appear:

- Instrument name and serial number
- SMRT Link address
- Data path
- Instrument software
- Chemistry version
- Option to edit the user avatar
- Option to change server configuration
- Option to change network

Preparing to load the instrument

The following materials and components are needed before sequencing on the Vega systems:

Materials for sequencing

Materials	Quantity	Source
Vega SMRT® Cell tray + tip	1	PacBio
Vega sequencing plate (and accompanying foil strip)	1	PacBio

Prepare the Vega sequencing plate for sequencing

- Thaw the sequencing plate. Remove the sequencing plate from the mylar bag. You can locate the lot number, serial number and expiration dates on the sequencing plate label. The mylar bag also contains a QR code card. You can scan the QR code on the sequencing plate label by using a laptop or webcam camera, then clicking the Scan button. This automatically fills in the Lot, Serial and Expiry fields in SMRT Link.
- Place the sequencing plate in a room temperature (~22°C) water bath covered with foil to protect the plate from light for 60 minutes.
- Remove the sequencing plate from the water bath and invert, tap, and observe the sequencing plate to look for any remaining frozen or precipitated materials. Repeat this 5 times to ensure sufficient observation. If wells remain frozen, return to the water bath for additional time.
- Once thawed, mix by vortexing at 1000 rpm for 1 minute.
- Spin down the sequencing plate for 1 minute at 150 rcf to ensure that the reagents are in the bottom of the plate.
- Before loading on the instrument, wipe any moisture or contaminants from the top of the sequencing plate.
- Note that the sample should have already been prepared and diluted according to instructions from SMRT Link Sample Setup.

1. Pierce column 1 well A
   - Using a pipettor, secure a 200 µL tip.
   - Press the pipette tip to the center of the well gently until the seal breaks. Touch the bottom of the well with the tip. Do not attempt to open the foil completely to the edge of the well by following the perimeter of the well with the tip.

2. Discard the tip, then using a new tip, add the sample to the well.
3. Peel the adhesive foil strip (included with the Vega sequencing plate) from its backing. Use both hands (with gloves) to hover the seal over column 1 and firmly seal the pierced well (being careful to not cover any wells in column 2). Do not attempt to peel and reseat seal.
4. Spin down the sequencing plate for 1 minute at 150 rcf.
5. Leave the plate on ice until ready to load on to the instrument.

   Caution: Only the foil strips included with the Vega sequencing plate should be used on the sequencing plate. Other seals may cause failures.
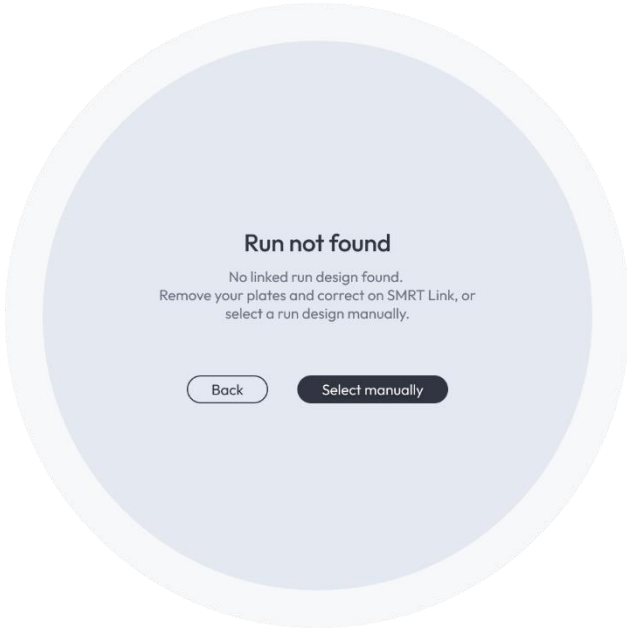
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Starting a run

1. To start a run when the system is idle, touch the "Tap to open" text on the home screen. The screen will raise partially to allow the drawer to slide out and the waste door will automatically open. Always keep hands free of the area while the screen or drawer are in motion.

2. The screen will then instruct you to place the Vega SMRT Cell and tips consumable, and the Vega sequencing plate in their respective slots on the deck. The consumables should be placed with their labels facing the user; registration features will prevent them from seating if incorrectly oriented.

3. Press down on the consumables until they 'click' into place on both the left and right sides.

4. Empty any waste from the waste receptacle, replace the bin and close the waste door. As you complete these steps, solid check marks will appear next to the respective instructions.
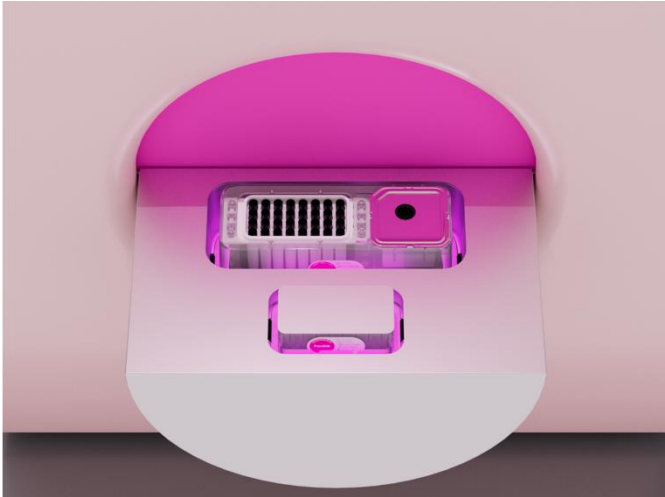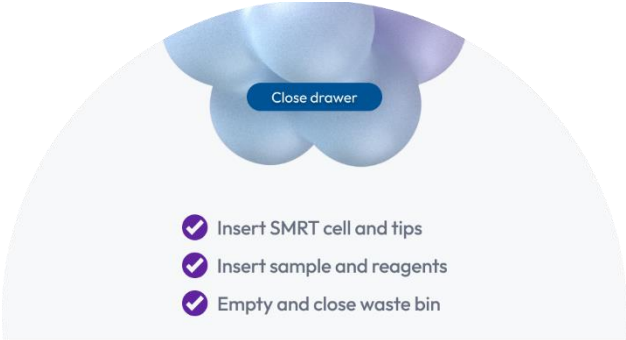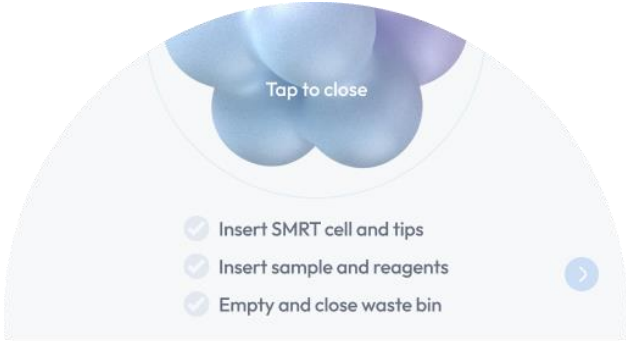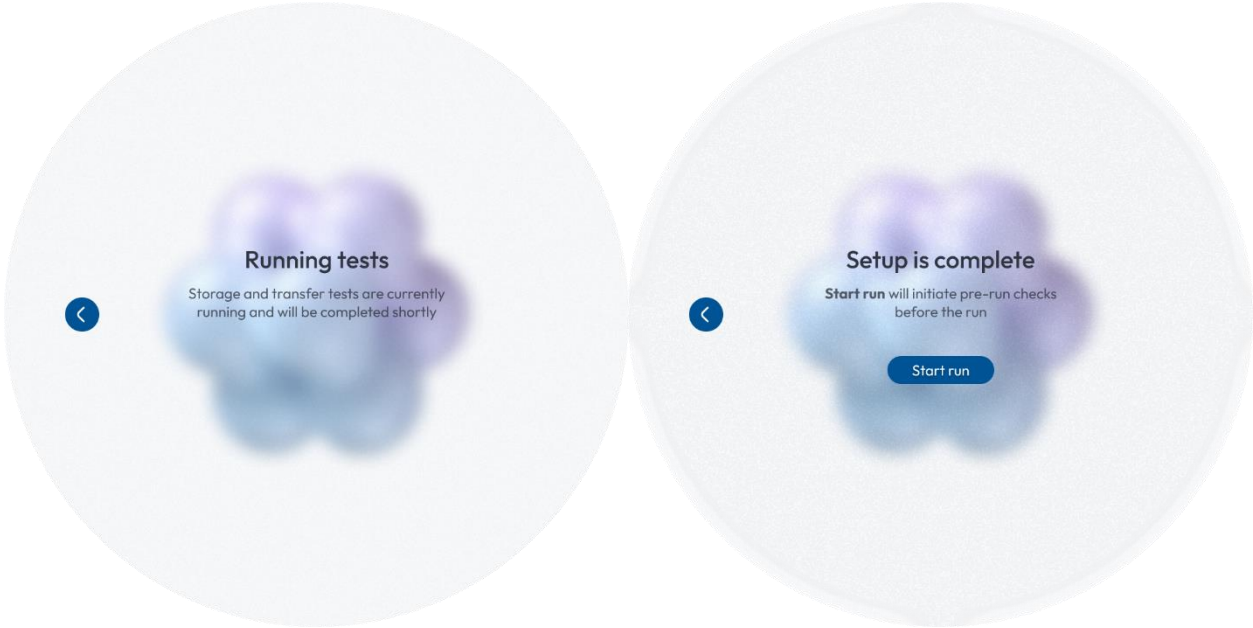
5. Press the Close drawer button to proceed.

6. The system will attempt to find a linked run design in SMRT Link.

7. If a linked run design is found, the system will show you a run summary, including estimated run duration and completion time. Press the Select different run button to select a different run design if desired, or press the right arrow to proceed. Press the Open drawer button if you wish to back up and remove or replace consumables.
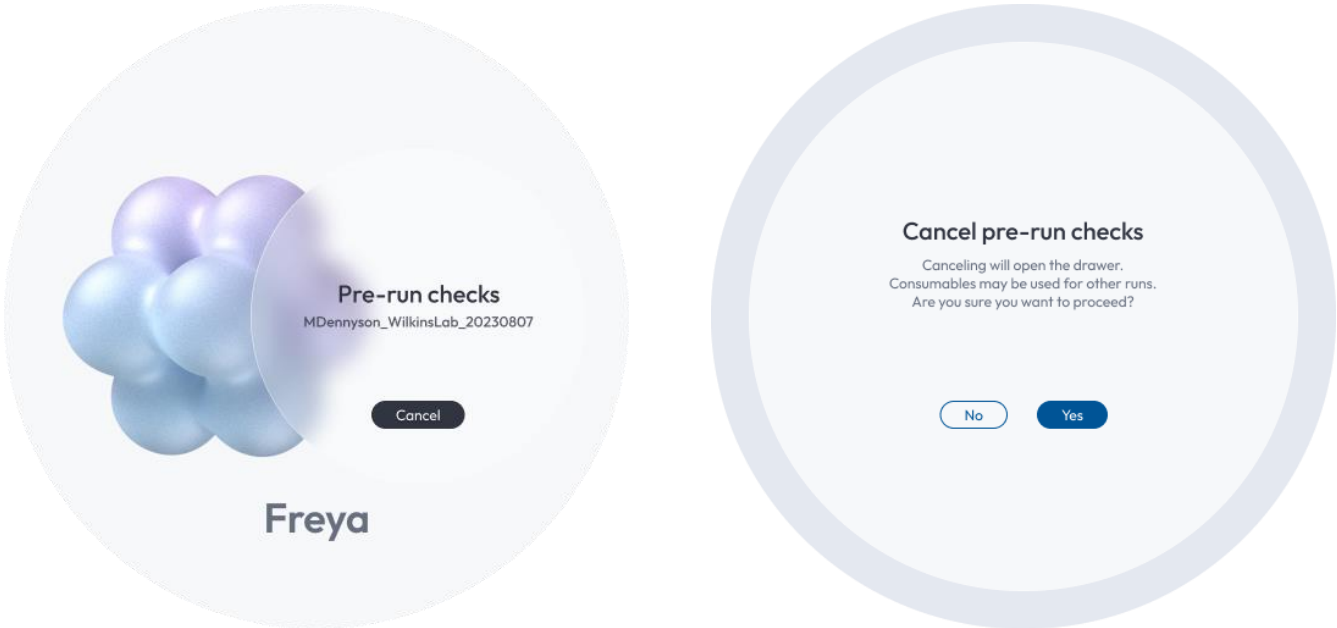

8. If a run is not found, or if you elect to select a different run, you can manually choose any available run design previously defined in SMRT Link. You can sort by date created, author, or run name.

9. One the run is selected and you press the right button to proceed (as in Step 4).

Running tests
Storage and transfer tests are currently running and will be completed shortly

Setup is complete
Start run will initiate pre-run checks before the run

Start run

10. The system will now commence pre-run checks. Allow the system to proceed, or hit cancel if you wish to cancel the run.

Pre-run checks
MDennyson_WilkinsLab_20230807

Cancel

Freya

Cancel pre-run checks
Canceling will open the drawer.
Consumables may be used for other runs.
Are you sure you want to proceed?

No   Yes

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11. Once pre-run checks are complete, the system will advance to processing the run. The screen will show the progress of the run and the estimated time of completion. Press the 'I' button to view more information about the run in progress.
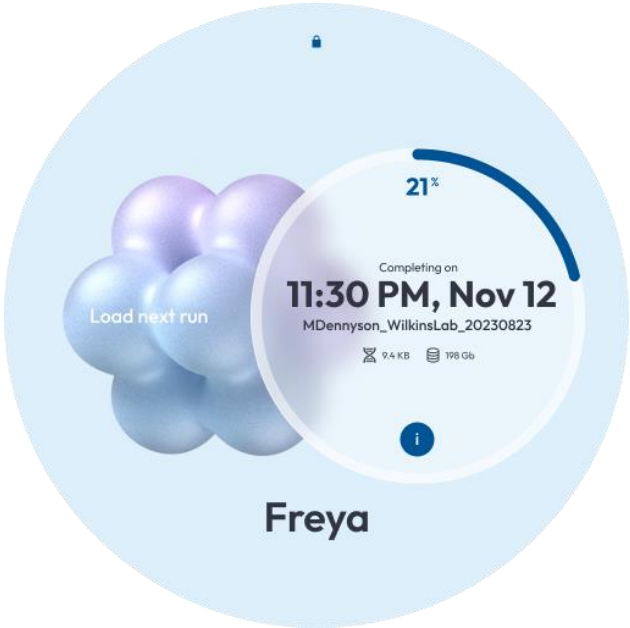

12. When the run has finished, the display will indicate that the run is complete and will prompt you to open the draw to remove used consumables. When all cleanup steps are complete, you can tap to close the drawer.

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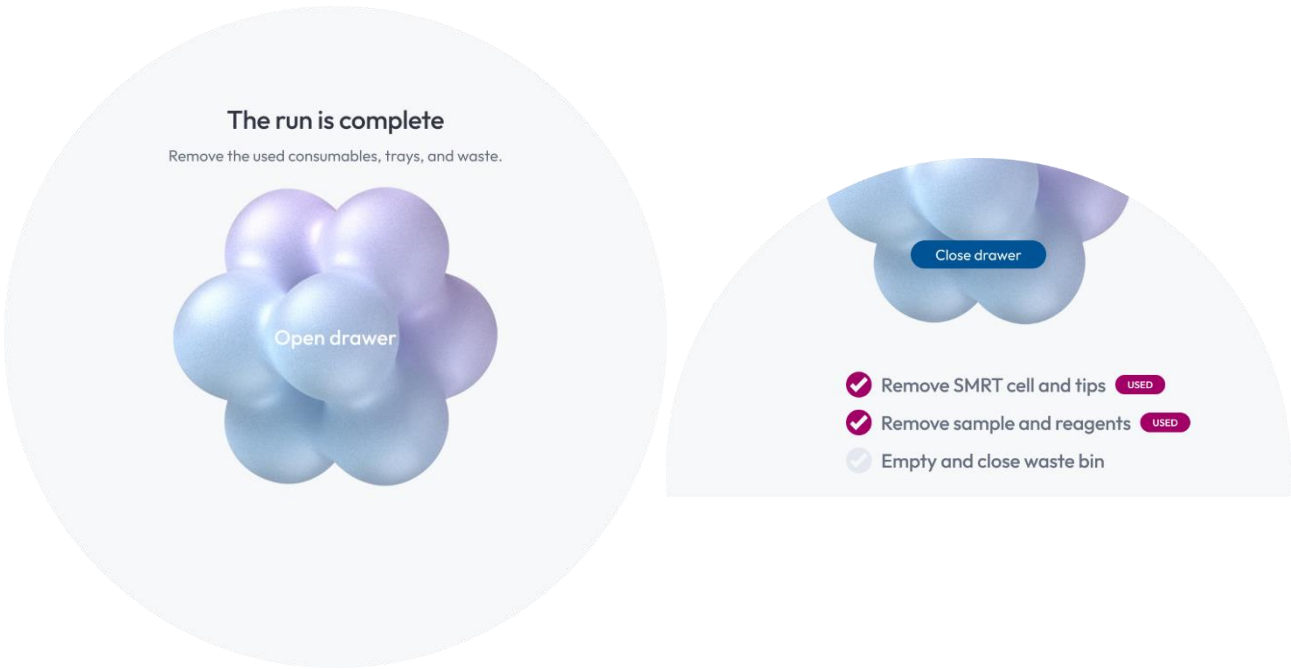
Setting up a run while the instrument is sequencing

1. During portions of an active run, the system will display "Load next run" on the screen. This functionality allows you to remove spent consumables and queue your next run to be automatically started once the current run has completed data acquisition. This process is nearly identical to setting up a run on an idle instrument, with the following differences.

2. Tap the "Load next run" button on the homescreen to access the consumables drawer. The screen will raise partially to allow the drawer to slide out. Always keep hands free of the area while the screen or drawer are in motion.

3. The LED lights will be illuminated in white, indicating the presence of used consumables. The IUI will also indicate that the consumables are used, and will prompt you to remove them. Once this is done, you can replace new consumables.
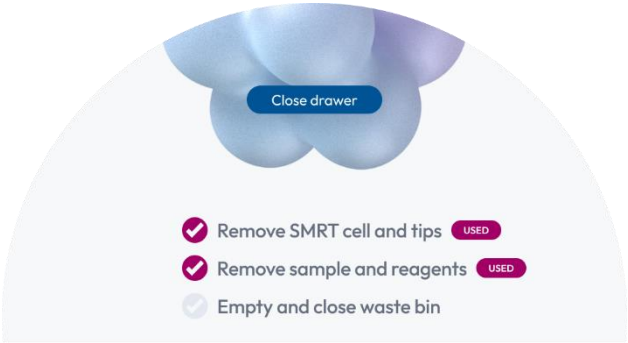
4. Once used consumables are removed, you can proceed with loading consumables and selecting or confirming a run design just as you would when setting up a run on an idle instrument. Once you have loaded new consumables and the system has validated them, press Close to continue and proceed from step 4 of Starting a run.
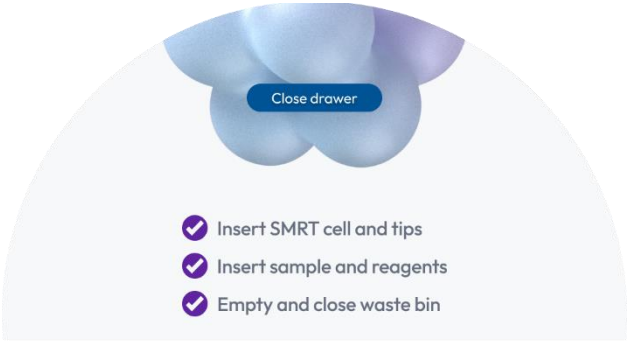
5. Alternatively, you can close the drawer with no consumables loaded, in which case the system will return to displaying the current run status.

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Viewing or canceling a pending run

1. If you pre-load a subsequent run, the run status screen will indicate that a run is pending.

2. Pressing the small circular status button will bring the pending run to the foreground of the run status screen. There, you can press the 'i' (information) button to get details about the pending run.

3. If you wish to cancel a pending run, press Cancel from the information screen. The pending run will be canceled, and you will be prompted to remove and store the unused consumables.

Expired consumables

4. If you load expired consumables, the system will detect them and alert you. Runs performed with expired consumables are not covered for reimbursement in the event of under performance or failure, but you may elect to proceed with using them at your own risk.
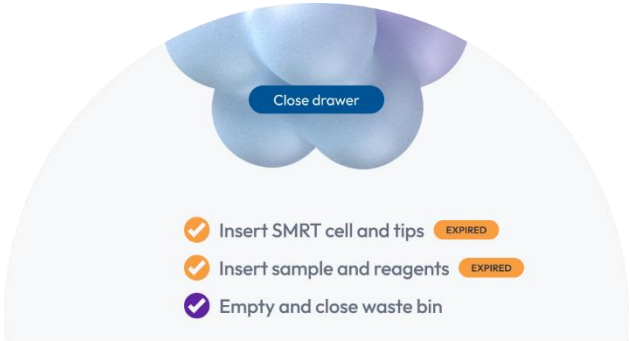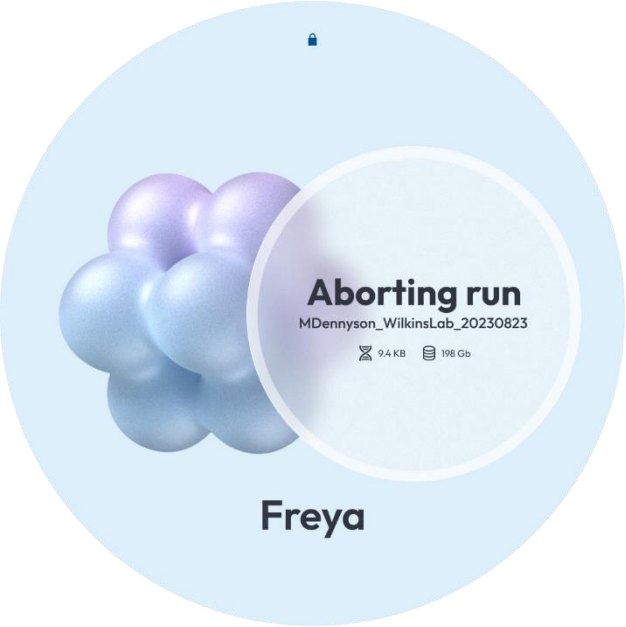
Aborting a run

5. A run in progress can be aborted if desired. To abort a run, first press the 'i' button to access the Run information UI, then press Abort.
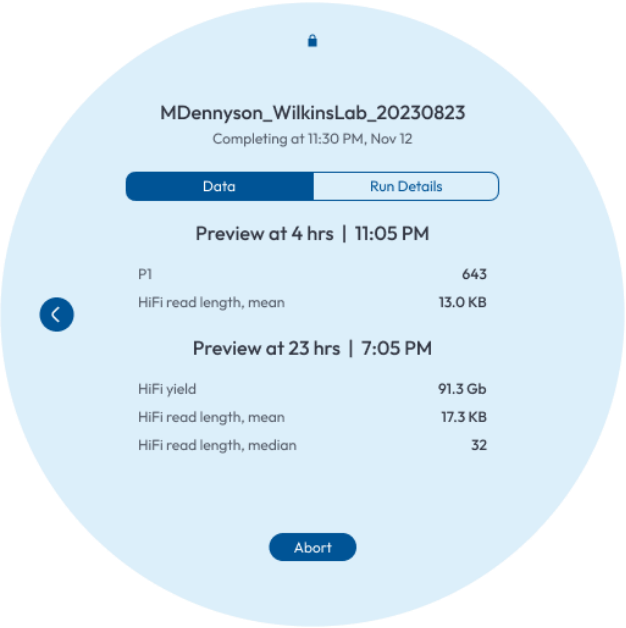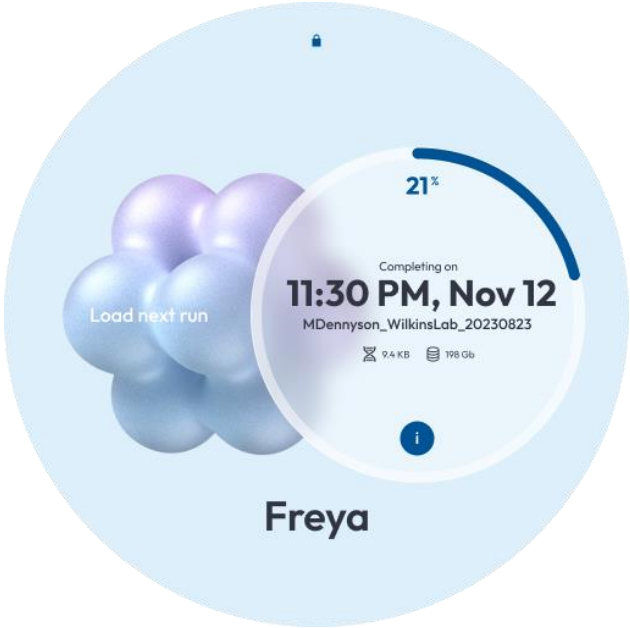
6. The system will ask you to confirm your intention to abort the run. Once confirmed, you have no option to cancel the abort process and run data will be lost.

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7. The system will ask you to confirm your intention to abort the run. Once confirmed, you have no option to cancel the abort process and run data will be lost. Once the abort process completes, the system will prompt you to open the drawer and remove used consumables.

Revision history (description)	Version	Date
Initial release.	01	December 2024

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