

How to run cryopreservation protocols using a VIA Freeze™ system

CANCER RESEARCH ★

TRANSLATIONAL MEDICINE

Joe Zitelli, Cytiva

Jessica Remy, Cytiva

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Abstract

The purpose of this protocol is to give the reader a comprehensive understanding of how to set up and operate the Cytiva VIA Freeze™ Uno, VIA Freeze™ Duo, and VIA Freeze™ Quad units.

This protocol applies to anyone performing a procedure with the VIA Freeze™ units. Once training on this document is complete the user should be able to set up and run their units to process samples.

Introduction

This protocol outlines how to use your VIA Freeze™ Uno, VIA Freeze™ Duo, and VIA Freeze™ Quad units, from unpacking to operation. You can use the instructions here to set up your units and begin processing samples.

Reagents and Equipment

Equipment

Item	Supplier	Code	Calibration Due Date
VIA Freeze™ Freezer Unit	Cytiva		Annually
SBS plates	Cytiva		None
USB drive (optional, not supplied)			
USB keyboard (optional, not supplied)			
USB mouse (optional, not supplied)			

Material (not supplied)

Reagent
Cryoprotectant solution

Cryobag or cryovials

Personal Protective Equipment (not supplied)

1. Laboratory coat
2. Safety goggles
3. Gloves

VIA Freeze™ instrument introduction

The VIA Freeze™ range of controlled-rate freezers are liquid nitrogen-free. They combine customizable freezing profiles, electronic control systems, and a conduction cooling method to optimize GMP compliant cryopreservation.

- **Controlled cooling:** Maintain optimal cell function and viability with customizable freeze profiles and precise temperature control.
- **Consistent quality:** Remove heat evenly by conduction. Apply stored freezing profiles to standardize cryopreservation of every sample.
- **Clean room compatibility:** Streamline manufacturing processes. Eliminate risks and infrastructure costs associated with liquid nitrogen.
- **Supports cGMP compliant cryopreservation:** Ready to integrate with cGMP process validation procedures. Controls designed to meet 21 CFR Part 11 regulations.
- **Versatility:** Three models for different processing requirements and scales. Options for 24–192 cryogenic vials or cryobags up to 250 mL.

Unit weights and handling

The operation and user interface are identical for all VIA Freeze™ units. The only difference is the capacity or number of samples of each unit.



- Take care when lifting and carrying all VIA Freeze™ freezers. Ensure that all precautions are taken to lift in a safe manner and follow your company's lifting and handling procedures.
- Below is a table of the weights associated with each piece of equipment.

System	VIA Freeze™ Uno	VIA Freeze™ Duo	VIA Freeze™ Quad
Weight	14 kg (31 lb)	40 kg (88 lb)	64 kg (141 lb)

Note: The VIA Freeze™ Uno unit is relatively light and may be lifted by one person.

Note: The VIA Freeze™ Duo and VIA Freeze™ Quad units are heavy instruments and are not suitable to be lifted and carried by a single person. When moving these units, make sure that one person grips each of the large blue handles at the sides.

- Place the unit on a surface capable of holding its weight.

Operating conditions

- The units are designed to operate in ambient air temperature of 0 to 30 degrees Celsius and from 0% to 90% humidity.
- Place the unit at least 10 cm (4 in) from any wall or obstruction. Do not run the system on a soft surface such as a carpet.
- If networking or remote access is required, then plug an Ethernet cable into the RJ45 socket at the back of the unit.

VIA Freeze™ unit disposal

- Each unit contains a Stirling engine with a small amount of pressurized helium. Do not dispose of the units in general waste.
- Instead, contact a Cytiva representative.

Procedure

Unit setup and main menu

After you plug in and turn on the unit, it will boot up. The initial screen will be the user login screen.

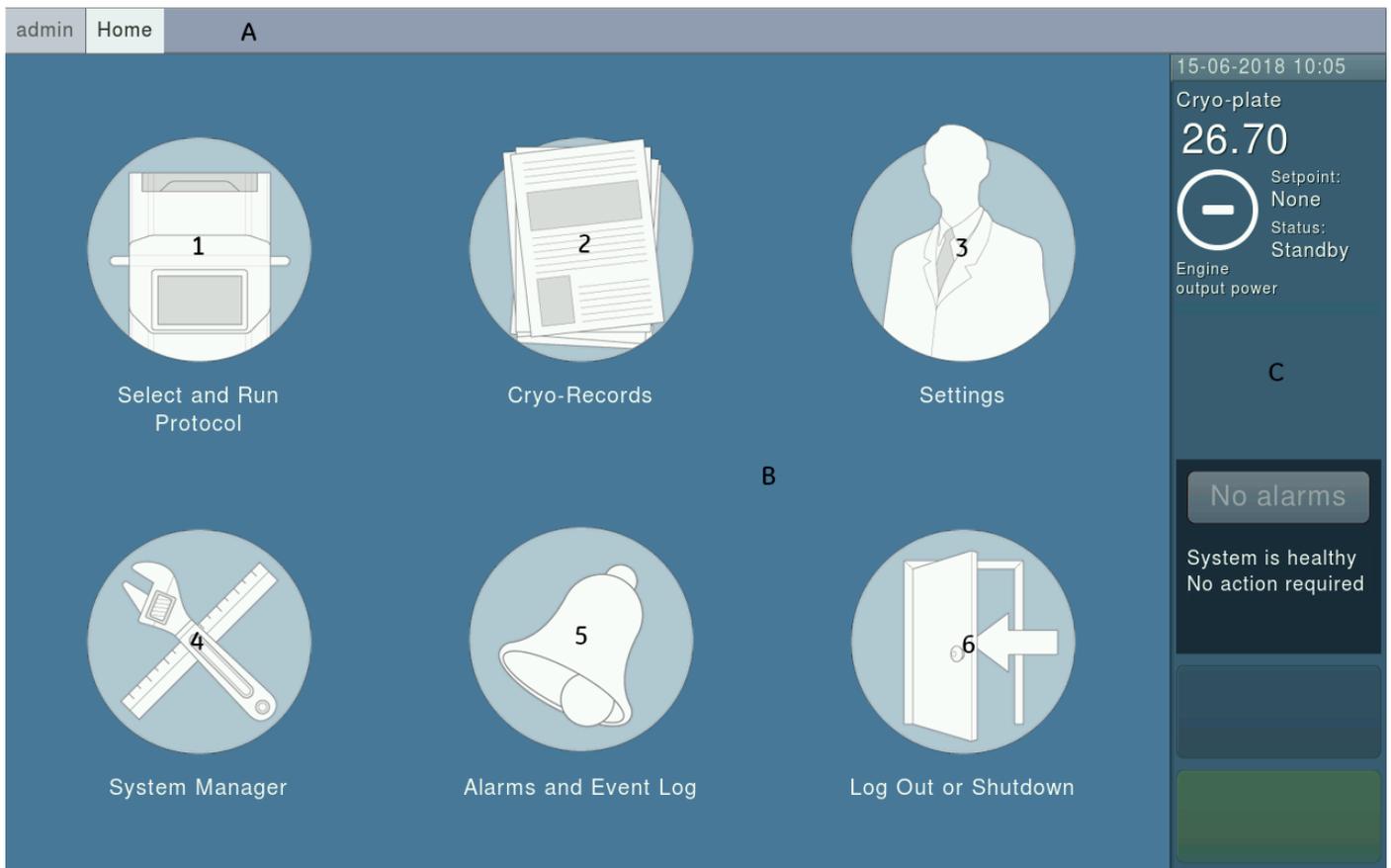
Note: There are two types of user accounts: User and Admin. Both user types can log in from the user login screen. User and Admin accounts can access different areas. For example, only an Admin can create new users.

Log in using your account credentials.

Note: By default, the initial log in when setting up the unit for the first time is Admin without password. For security and traceability reasons, change this default account.

Note: If the login screen is inactive for more than 15 minutes, you will be logged out. If the session expires you will be prompted to log in again.

Here is a screen shot showing the Main Menu screen after you log in. The Main Menu has three distinct areas: The navigation bar (A), the menu window (B) and the status bar (C).



A The navigation bar shows the menus and windows that are currently active. You can navigate backwards through active windows by tapping each menu.

B The menu window is the main area where you to interact with the unit, and it changes according to the selected menu.

1 **Select and Run Protocol** provides access to the select protocol type menu via which you start and control a run.

2 **Cryo-Records** lists all the previous process records to review and export.

3 **Settings** allows a logged-in Admin to access settings such as password control and/or email address management.

Note: if you are logged in as a User, you will not be able to see the settings.

4 **System Manager** allows you to access the system manager submenu, which provides the protocol editor, user management controls, and service menu.

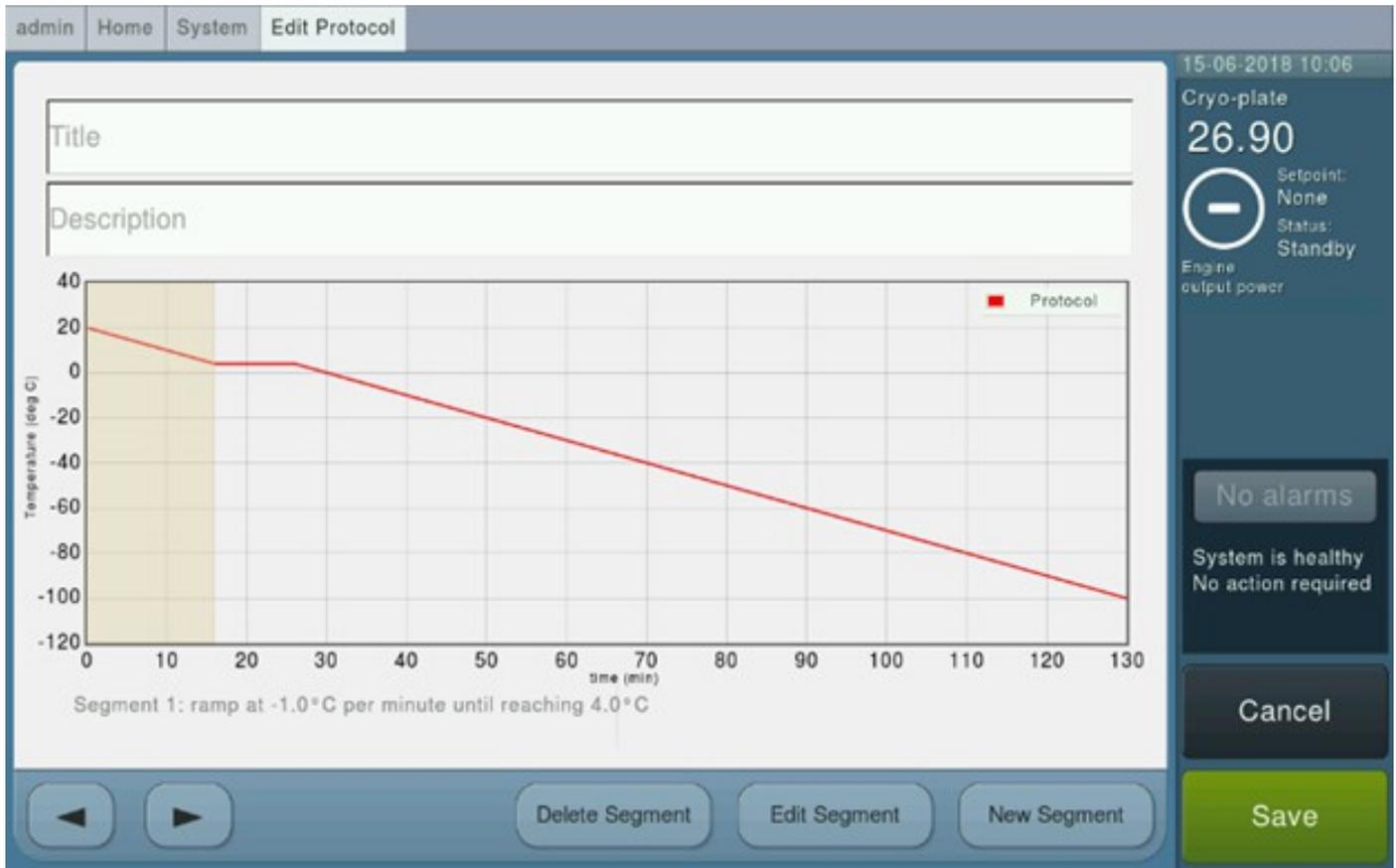
5 **Alarms and Events Log** lists all the previous alarms and events to review and export. These can then be exported the same way as explained later.

6 **Log Out or shutdown** allows both a safe logout from one user to another, and power off the unit. Always power down the unit via **Log out or shutdown**.

- C The status column is a vertical column located on the right side of the menu screen. It displays live data from the Stirling engine. If you are using a Quad unit, you will see two displays.

Create or edit a freezing profile

1. From the Main Menu, touch the **System Manager** button.
2. Select either **Create New Protocol** or **Edit Existing Protocol**. Open the Edit Protocol window as shown in the image. From here, you can delete a segment, edit a segment, or add a new segment.



1. Enter the title and description of the cooling profile.

Note: When you edit a profile, you can only change the description.

1. Use the arrows to move between the segments.
2. For each segment (edit or new), you can choose different options.

- **Ramp** allows you to ramp down the temperature to reach a targeted temperature (min -100°C) with a defined rate from 0.01°C/min to 2.00°C/min.

Note: The targeted temperature cannot be positive relative to the room temperature.

- **Dwell** allows you to keep the temperature for a defined time.
 1. At the end of the last segment, the hold function is activated by default (even if the hold at end of this segment shall be disabled).
 2. Select **Save** and validate the message.
 3. Select **Back** to return to the Main Menu.

Note: The range is from 00:00:00 to 99:59:59 (hh:mm:ss).

- **Hold at end of this segment** allows you to pause the procedure at the end of a segment. To resume the procedure, you must touch the **Resume** button to continue to the next segment.
- **Lid alarm active** allows you to activate an alarm if the lid is opened during the current segment.

Perform a procedure

1. From the Main Menu, touch the **Select and Run Protocol** button. This brings up the protocol screen.
2. The protocol screen lets you select the protocol you want to run. Up to six protocols are displayed on each screen. Use the right arrow at the bottom of the screen to see the next six protocols available.
3. After you select a protocol it is displayed in the Main Menu. Use the arrows at the bottom of the screen to inspect the protocol, which is the cooling profile that the unit will follow.

Note: The protocol consists of ramps, dwells, and holds. Ramps are parts of the protocol where cooling happens. Dwells are periods where the temperature is held for a determined amount of time. Holds are points where the temperature is held until a user resumes the run.

1. Once you have inspected the protocol either select **Back** to choose a different protocol or **Select protocol** to begin the run.
2. Before you start a run, you will see a window that shows a record details screen. Here you can input information about the run. You can also use a scanner to input barcode information.
3. Start the run by touching the **Start** button. When the run starts the window displays a live graph showing temperature on the vertical axis vs time on the horizontal axis. The cryoplate temperature is displayed as a blue line, and the setpoint is displayed as a yellow line.

Note: At any point during the run, you can cancel the run by touching the **Abort** button.

Note: At any point during the run, you can hold the temperature by touching the **Hold** button. Continue the run by selecting **Resume**.

Note: If you touch the **Hold** or **Abort** button, a message is displayed asking you to confirm the action. If you do not validate or cancel the action after 15 seconds, then another message is displayed stating that the operation was canceled automatically. This means that the procedure continues.

1. When the process is complete, touch the **Finish** button to confirm the end of it.
2. The unit will prompt you to remove the samples.

Note: Always use appropriate gloves when handling frozen samples in the working area.

1. Once the process completion is confirmed the internal heating mechanism will begin to heat the unit, to eliminate residual moisture.

Note: Keep the lid open and remove the plate during this heating.

Note: The heating will automatically end after the unit reaches the target temperature and length of time prescribed by the unit. Then, the main screen is displayed.

Export data

1. When a run is complete all the data is stored permanently on the unit. You can export the data as a .pdf or .csv file onto a USB drive, email transfer to an FTP server, or via Chronicle™ software.

Note: To export to a USB drive, you must insert a USB drive into any of the three USB locations in the back of the unit.

Note: To set up the email transfer, refer to the Operator Manual or contact your local Cytiva representative.

1. From the Main Menu, touch the **Cryo-records** button.
2. Select the data to be exported and touch **Export**.
3. A window displays asking you to choose an export location. Select either the USB drive name or an email address and touch **Confirm**.

Note: It may take several minutes to generate the report and export it.

1. After the export is complete, the 'Cryo-records' menu displays again.

Note: If using an USB drive, select **Eject USB** before removing the drive.

1. Select **Back** to return to the Main Menu.

Notes and Comments

Cleaning and maintenance

- Regularly clean the unit to reduce the risk of sample contamination.
- Clean using any of the following reagents: mild detergent, Isopropanol, ethanol, mild bleach, warm water.
- Specialist cleaning products from STERIS™ have also been tested and approved for cleaning of VIA Freeze™ units. These products include: Environ™ Vesphene™ st - diluted 1:128, Environ™ LpH™ st - diluted 1:125, Spor-Klenz™ RTU.
- Do not use acetone to clean the system as it will damage the paint.
- Before cleaning, make sure the heating cycle has been completed and the unit is unplugged.
- Dampen a cleaning cloth with the desired cleaning product. Do not spray directly or over-dampen the towel.
- Remove dust or debris near the air vents using a conventional vacuum cleaner.
- After you complete the cleaning, turn on the unit and complete a heating cycle.
- See the User Manual for detailed instructions on replacing the heat transfer pads. Replacing the heat transfer pads can optimize performance, and this is recommended whenever they become dirty or damaged.
- Routine maintenance is performed annually by a trained Cytiva representative. This includes an annual calibration and maintenance to make sure the unit is working as expected.

Software update and factory reset

For any software update or factory reset, please contact your local Cytiva representative.

Learning outcomes

After reviewing this document you should be familiar with setup, login, main menu, protocol creation, and maintenance for VIA Freeze™ UNO, VIA Freeze™ Duo, and VIA Freeze™ Quad units.

Related Policies/Forms/References

VIA Freeze™ User Manual

VIA Freeze™ Accessory Catalog

Recommendations

1. If using a cryobag, remove the air.
2. If using an overwrap bag, remove the air while sealing it.
3. Homogenize the cells well to facilitate uniform cryoprotectant repartition.
4. Flatten the cryobag when you place it inside the unit.
5. Wipe any liquid from the surface of vials or cryobags before starting to cool, to avoid frost formation.

Safety precautions

Power and voltage

1. All VIA Freeze™ units operate using mains voltage of between 100 and 240V with a standard IEC C14 power socket on its rear.
2. Make sure you always have access to the power switch or the outlet power source when operating the unit.

Waste disposal

Dispose of waste from this process via the appropriate waste stream.

Please refer to the User Manual for additional information on unpacking, safety, and disposal – and for detailed diagrams for the menu structure and size dimensions of the different VIA Freeze™ units.

References

Not available