

Controlled Rate Freezer

CRF-1 Series

Operating Manual

EN

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1. Introduction

1.1. Product Description

The Grant CRF-1 series is a unique liquid nitrogen-free controlled rate freezer for biological material preservation. It is pre-loaded with configuration specific freezing programs suitable for the freezing of embryos, oocytes, blastocysts, stem cells and spermatozoa.

The CRF-1 is not a Medical Device as defined by European Directive 2007/47/EC.

1.2. Product Accessories

The following optional extras may also be supplied with the CRF-1 series freezer:

CRF-1 CRYOPEN	Cryopen nucleating tool
CRF-1 CARTRIDGE	Cryopen spare cartridges

Brief operating instructions for these product accessories are given in Appendix 2. Please refer to accessory manuals for full operating instructions.

2. How to use this Operating Manual

This operating manual will allow you to unpack, correctly set-up and operate the CRF-1 series controlled rate freezer both efficiently and safely. Important safety information is listed in the following section. The technical specification is included in section 4.

If there is a technical matter that this operating manual does not address or any other question concerning the CRF-1 product please do not hesitate to contact Grant Instruments (Cambridge) Ltd.

The general details of your warranty for the CRF-1 series freezer are detailed in section 11.

3. Safety Information

3.1. CRF-1 Series Controlled Rate Freezer

The following symbols mean:



Caution: Read these operating instructions fully before use and pay particular attention to sections containing the symbol.



Read this manual before using the CRF-1 nitrogen-free controlled rate freezer. Particular attention should be paid to the Installation section and all safety precautions for the unit.

Always observe the following safety precautions:

⚠ The unit is designed to be used by a professional person with the appropriate knowledge to prevent damage to the user or the specimens

⚠ Use only as specified by the operating instructions

⚠ If the unit is used with accessories not provided or recommended by the manufacturer or in a manner not specified by the manufacturer, the protection provided by the unit may be impaired

⚠ After transport or storage in humid conditions, allow the unit to dry out before connecting it to the supply voltage

⚠ Only connect the unit to the mains supply using the power supply unit and cables provided

⚠ Ensure suitable protection is worn when accessing the freezing plate, as temperatures down to -100ºC can be reached during and after the freezing cycle

⚠ Ensure that the mains switch and isolating device (power supply connector) are easily accessible during use

⚠ Ensure the unit has been disconnected at the power supply socket before moving

⚠ The unit should not be used in hazardous atmospheres

⚠ The freezer should be switched off immediately if the plate temperature exceeds 30ºC

⚠ Do not check the plate temperature by touch.

⚠ The device contains a pressurised container and should not be incinerated.

3.2. Cryopen Nucleating Tool

When using the cryopen nucleating tool (if supplied) read the instruction manual and safety instructions supplied with the product before use.

⚠ Ensure that suitable safety precautions are taken during use of the Cryopen and never point the device towards the face.

4. Technical Specification

4.1. Operating Conditions

The CRF-1 is designed for operation indoors in a standard laboratory environment with temperature and humidity control, with an ambient temperature between 20°C and 30°C at altitudes up to 2000m.

4.2. Electrical Details

Mains Supply:	100-240V, 50-60Hz 2.5A (max)
Unit Supply:	24V DC
Unit Power Consumption:	150W (max)
Pollution degree:	2
Installation Category:	II

4.3. Models Available

Model	Description	Compatible Vessels
CRF-1 H00	Plate for 16 0.5ml CBS straws	Cryo Bio Systems straws (014650)
CRF-1 H01	Plate for 18 0.3ml IMV straws	IMV Technologies straws (Paillette fine)
CRF-1 H02	Plate for 55 1.8ml cryovials *Max working volume per vial is 0.5 ml*	NUNC cryovials (363401 – round) NUNC cryovials (377267 – starfoot)
CRF-1 H03	Flat plate	
CRF-1 H05	Plate for 1 SBS microplate	NUNC cryobank, 96 2D tube system (374084) FluidX data matrix tube 96 (65-52319)
CRF-1 H06	Plate for 55 1.8ml cryovials *Max working volume per vial is 1.0 ml*	NUNC cryovials (363401 – round) NUNC cryovials (377267 – starfoot)
CRF-1 H07	Plate for 10 0.5ml CBS straws & 12 1.8ml cryovials *Max working volume per vial is 0.5 ml*	Freezing containers as H00 & H02 above
CRF-1 H08	Plate for 12 0.3ml IMV straws & 12 1.8ml cryovials *Max working volume per vial is 0.5 ml*	Freezing containers as H01 & H02 above

4.4. Performance

Max Plate Temperature[2]:	30ºC (all models)
Min Plate Temperature[2]:	Dependent on model type
H00, H01, H03, H07, H08	-100ºC
H02, H05, H06	-80ºC (typical)
Max Cooling Rate[2]:	10ºC/min (dependent on model & plate temperature)
Max Temperature Stabilisation Time[1]	5 minutes (all models)
Plate Temperature Accuracy[3]	±0.5ºC (all models)
Plate Temperature Stability[2]	±0.1ºC (all models)
Plate Temperature Uniformity[2]	< ±1.0ºC (all models)

1. Time to achieve a stable 20ºC start temperature from switch on
2. Unloaded plate
3. Calibrated accuracy taken as an average of 4 points at defined locations on an unloaded plate

5. Installation

5.1. Unpacking

Standard equipment includes:

- CRF-1 controlled rate freezer
- Power supply unit with UK and Euro mains lead adaptor
- Serial communications lead
- Serial to USB adapter
- Software on USB memory stick
- Operating manual
- Unpacking instructions

If the cryopen nucleating tool has been ordered in addition to the CRF-1 freezer it will also be included.

Read and carefully follow the unpacking instructions included with the unit to ensure the unit is ready for installation and operation.

Remove packing materials carefully, and retain them for future shipment or storage of the equipment. Inspect the unit to ensure that no damage has occurred during transit. If the unit shows any sign of damage, or any items are missing, please contact Grant Instruments immediately.

When lifting the freezer place hands underneath the unit and do not move by pressing on the casework.

5.2. Freezer Position

Ensure the freezer is positioned on a flat, stable surface so that the vents and the air outlet at the rear of the freezer unit are not blocked. Leave a clearance of at least 200mm all around the unit. Ensure that the unit is not placed directly in line with a heating or air conditioning vent, is protected from draughts from fans, doors or windows, is not near a radiant heater and is not in direct sunshine.

5.3. Electrical Connection

For normal connection to the mains, plug the external power supply into the 24VDC socket at the rear of the freezer unit (
Figure 1) and connect the mains plug into an appropriate single-phase mains supply which has an earth contact. If a UPS is being used, please read the instruction manual supplied with the UPS before use. If the mains power supply fails then the UPS should allow the CRF-1 to complete any freezing cycle that is running.

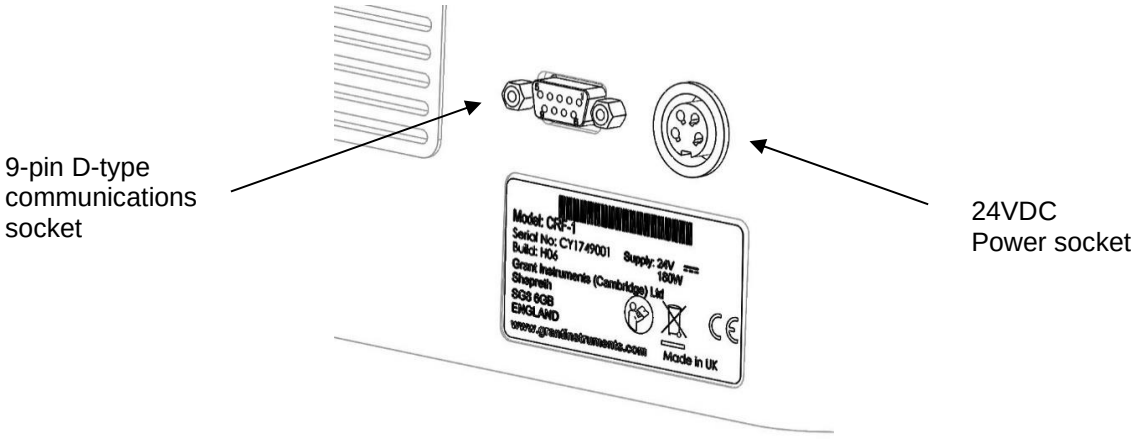


Figure 1: Rear panel of freezer unit

5.4. PC Setup

The CRF-1 user software can be run on any laptop or desktop computer using Windows 7 or Windows 10. However, there are several elements that must be considered before starting any data run.

Ensure any laptop used has sufficient power to maintain its powered-on state during the whole run. For complete security, the laptop should be powered directly from the mains supply.

To ensure run data is not lost, check the power settings do not allow the laptop to go into standby or hibernation mode.

Do not close the laptop during a data run as this could cause a loss of run data.

5.5. Switching the Freezer On

Before switching on the unit, connect the serial to USB communications lead (9-pin D-type) between a USB port of the computer and the back of the freezer.

Switch on the freezer unit using the power switch on the front panel of the unit (Figure 2). The switch illuminates red when the power is switched on.



Figure 2: Front panel of freezer unit (keypad and power switch)

6. Grant CRF Software

The software described in this manual is Grant CRF Software version 1.0.1

Grant CRF Software is available free of charge from your main distributor or from Grant Instruments (Cambridge) Ltd.

6.1. Installation and set up

6.1.1.1. Loading the software

The installation requires Windows 7 or Windows 10.

In addition to loading the Grant CRF software you will also be loading the drivers for the USB to serial converter (FTDI). In the case of Windows 7 only, you will be loading Microsoft .NET 4.5 framework if this is not already installed. This is detected automatically.

Administrator rights on the computer are needed to install the software requisites, software and USB to serial converter driver.

Insert the Grant CRF Software USB stick into the computer. Locate Setup.exe on the USB drive and double click to open.

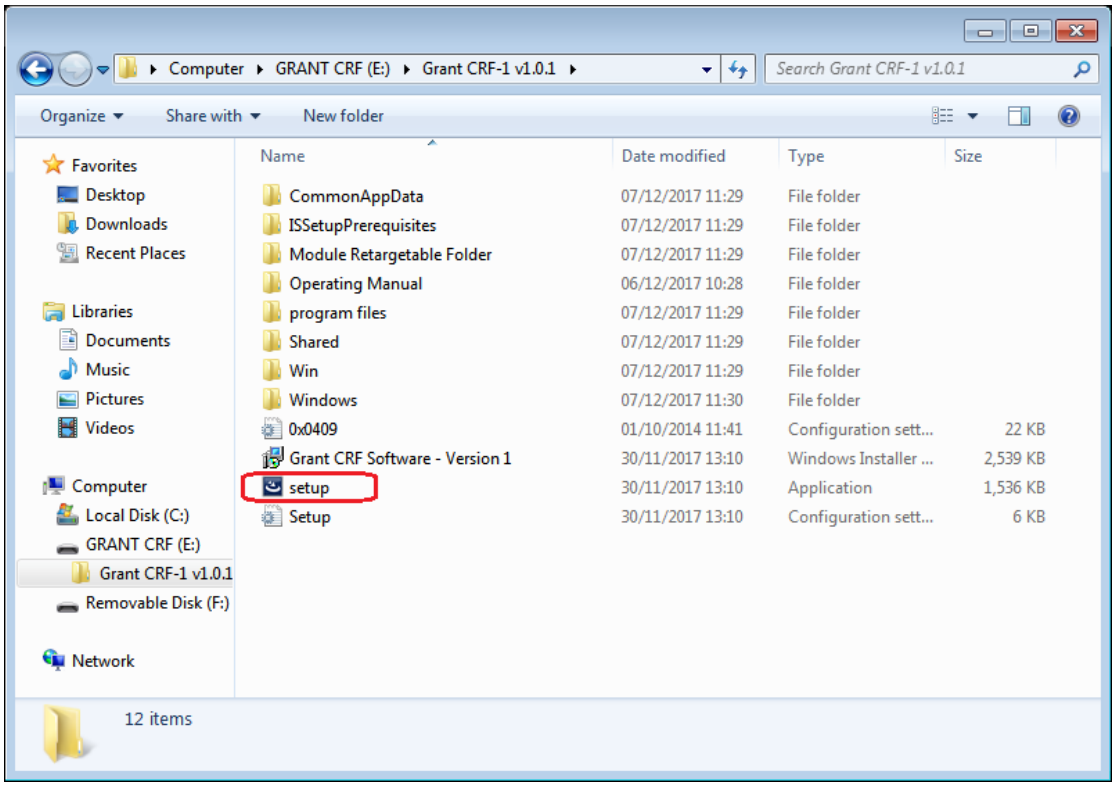


Figure 3: Software: Installation

6.1.1.2. .NET framework (Windows 7 only)

The installation will detect whether .Net requires to be installed. If it is already installed or if the operating system is Windows 10, then this step is skipped. Select "Install".

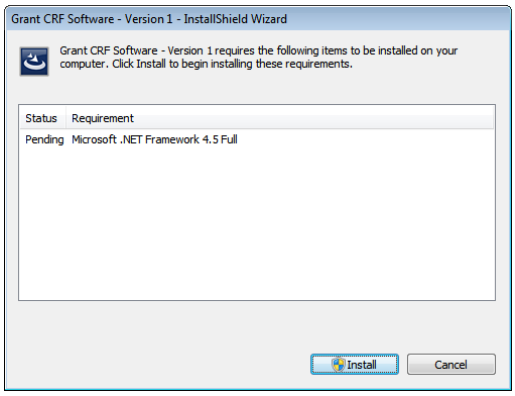


Figure 4: Software: .NET Framework Installation

You will also be prompted by the user account control. Select "Yes".

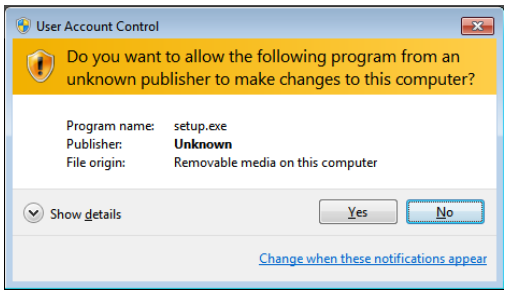


Figure 5: Software: .NET Framework Installation User Account Control

Installation of the .NET framework takes several minutes. To complete the installation of .NET your computer will restart automatically and then continue with the installation of the next part. You will need to log back in during this process.

6.1.1.3. Continue install and FTDI drivers

Installation will continue, or with Windows 10 this will be the first screen you will see. Select "Next" to move on to the Ready to Install window. Select "Install".

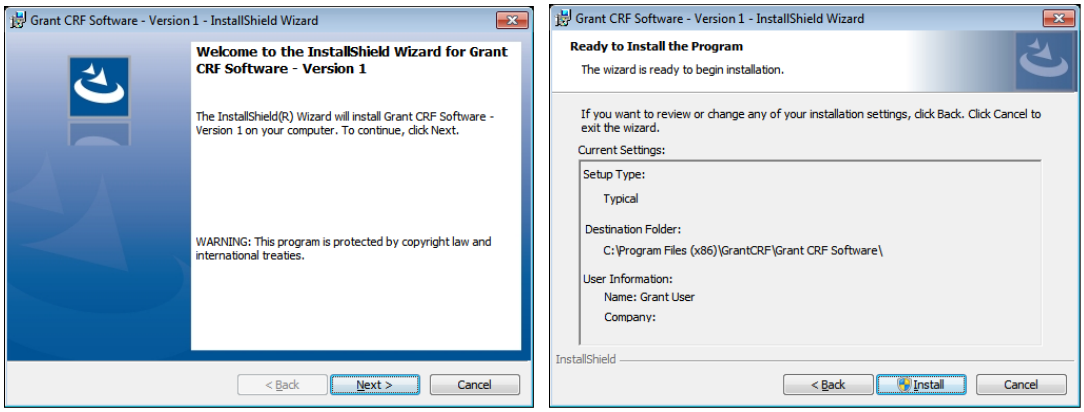


Figure 6: Software: Install Wizard

The software will start installing. User Account Control will ask if you wish to make changes. Select "Yes". Installation will continue.

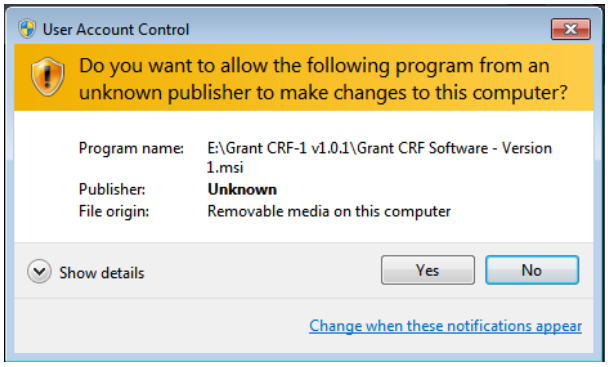


Figure 7: Software: Grant CRF Software Installation User Account Control

You will then be asked if you wish to extract the FTDI drivers. These are required for the USB to serial converter that you will use to make the hardware connection to the CFR. Select "Extract". Then on the following Device Driver Installation Wizard select "Next".

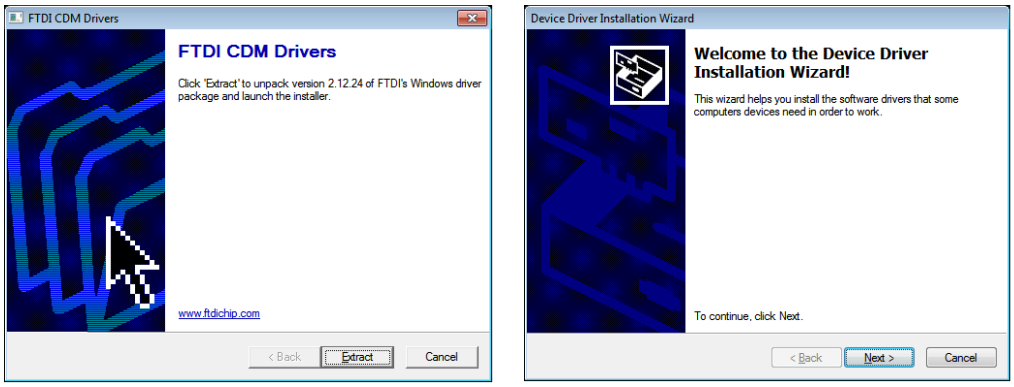


Figure 8: Software: FTDI CDM Drivers Extraction

You will be asked to read and agree to the licence terms. Then select "I accept this agreement" as shown. Select "Next". Driver installation will complete within a few seconds. Select "Finish".

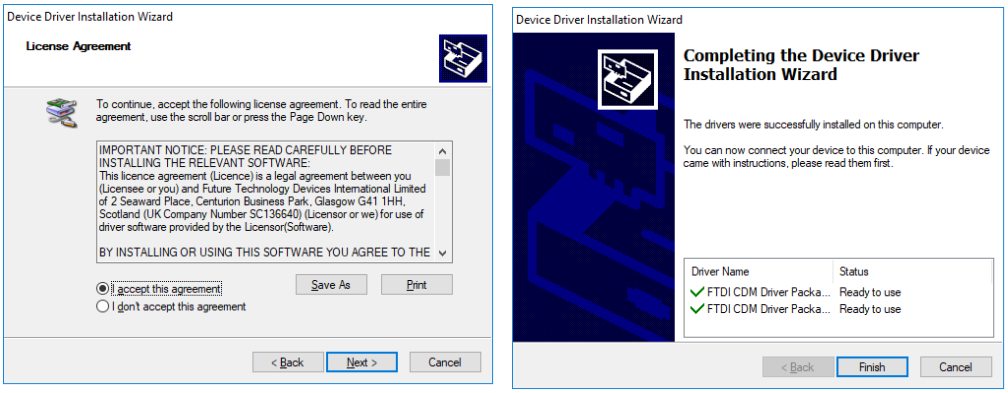


Figure 9: Software: FTDI CDM Drivers Installation

Installation will now complete in a few second. Select "Finish".

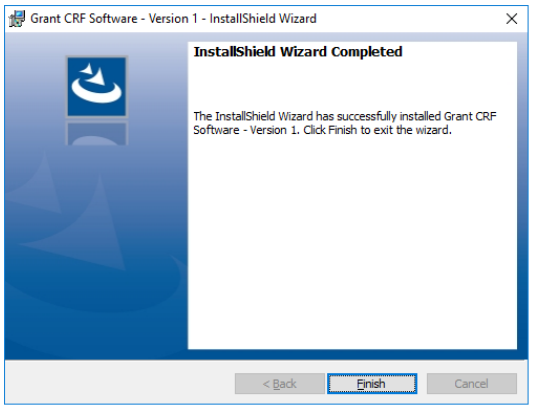


Figure 10: Software: Install Wizard Completed

6.1.2. Logging in

Start the software by either double-clicking on the shortcut icon on your desktop (Figure 11) or by selecting 'Grant CRF Software' from the program list in the start menu on your computer.



Figure 11: Desktop shortcut icon

When the software is run for the first time the default User ID is Admin and the default Password is Admin1 (see Figure 12). The Admin user is an administrator level user and has access to all software options. Other levels of access are available to Standard and Advanced users – these user levels can be set up via the Users page (see Section 6.8).

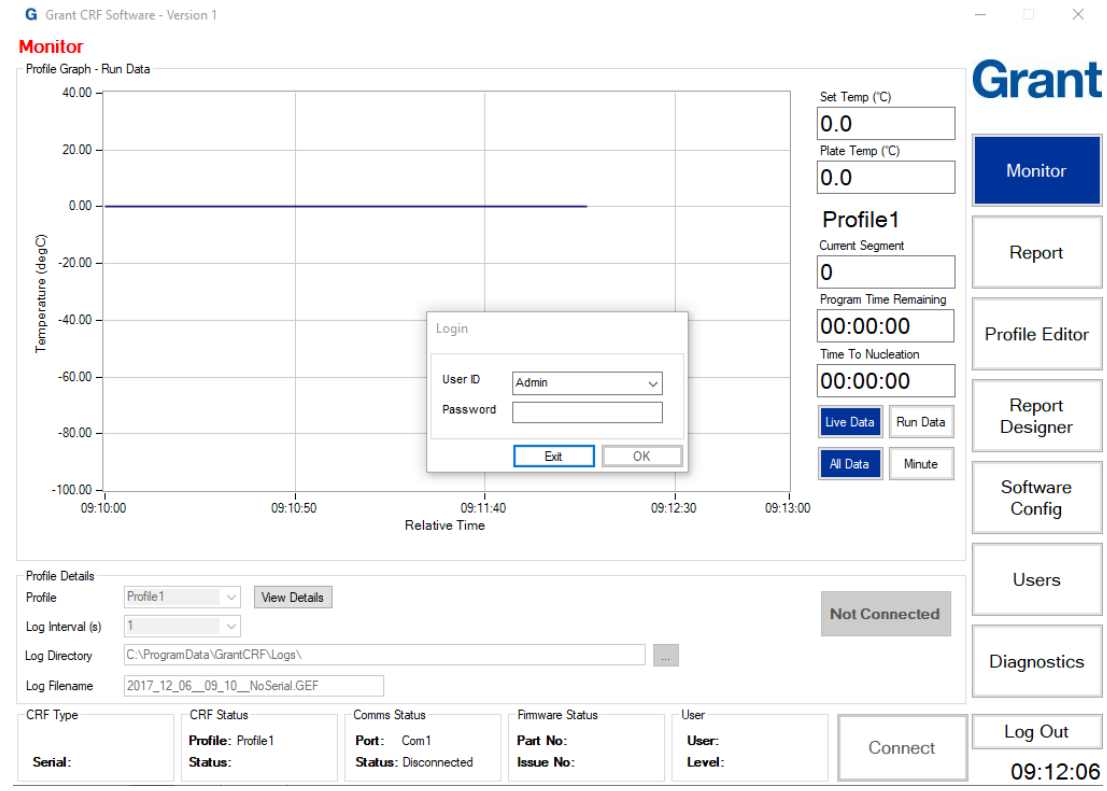


Figure 12: Software: Log In

6.1.3. CRF-1 connection & synchronisation

Wait at least 15 seconds after turning the CRF-1 on before trying to connect. To establish the connection, click the ***Connect*** button at the bottom of the software (see Figure 13). The CRF-1 and software will synchronise with each other. While this occurs, two bars show the progress. When the synchronisation is complete, the ***Connect*** button will change to ***Disconnect*** and the software will show ***Status: Connected*** (see Figure 13).



Figure 13: Software: Connected

As soon as the software is connected to the CRF-1, all parameters will be constantly read from the CRF-1 and updated. The CRF-1 will either be stable at the profile start temperature or changing towards the ***Set Temp (ºC)***. While the CRF-1 is changing it is not possible to run a temperature profile and the ***Start*** button and ***Profile Details*** are disabled.

When the value in the ***Plate Temp (ºC)*** box reaches the ***Set Temp (ºC)*** box value and is sufficiently stable then the ***Start*** button and ***Profile Details*** are enabled and the CRF-1 is ready to run a profile.

6.1.4. Software overview

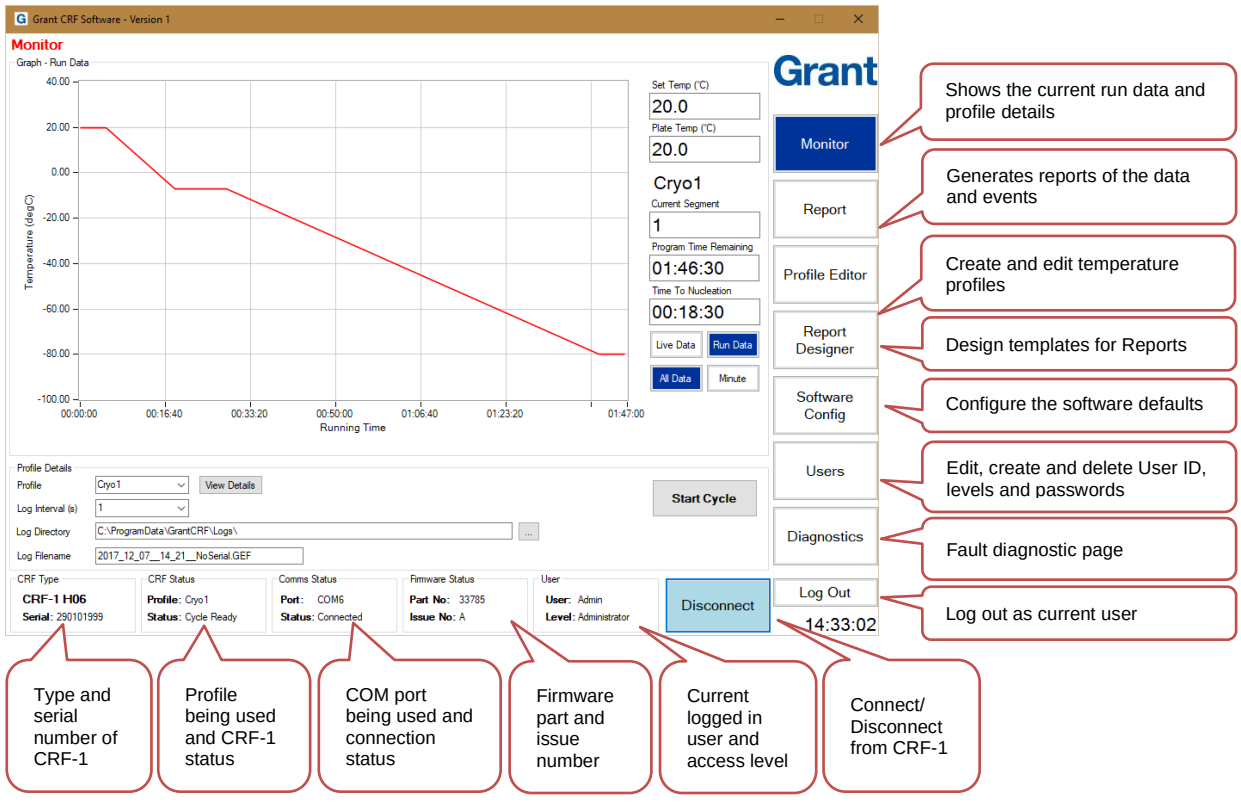


Figure 14: User Software Overview

6.2. Establishing communications with the freezer unit

Ensure the computer is connected to the unit using the serial communications lead or USB to Serial converter.

On the Monitor panel, click on the ‘Connect’ button as shown below. The ‘Comms Status’ at the bottom of the panel will change to ‘Status: Connected’. Other details such as the unit model and serial number will also be displayed. If the software fails to connect to the unit, see Appendix A1.3 for help.

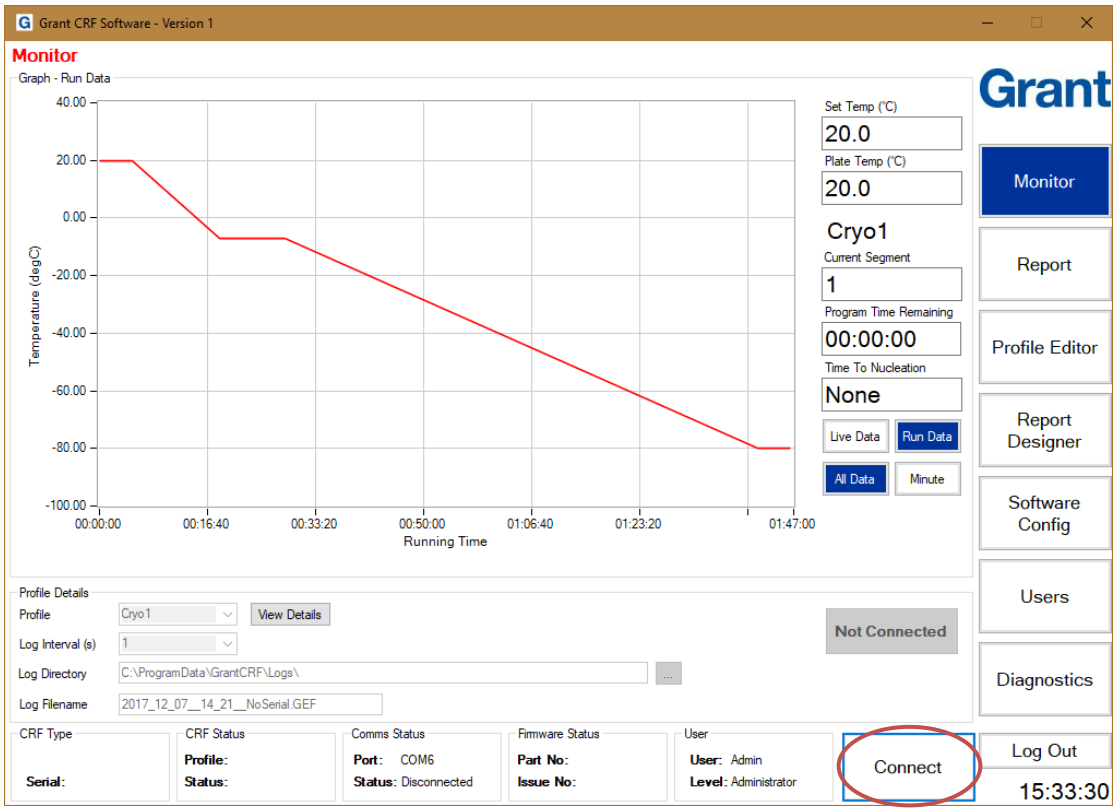


Figure 15: Software: Connect

The freezer unit will remain connected until the Disconnect button is pressed or the unit is switched off.

The system will be ready to use when the freezer unit plate temperature has stabilised at the Start Temperature. This may take approximately 5 minutes if the unit has only just been switched on

When the freezer has reached a stable temperature the Cryocooler Status will display ‘Status: Ready’. See figure below. The ‘Ready’ green light will also illuminate on the freezer front panel.

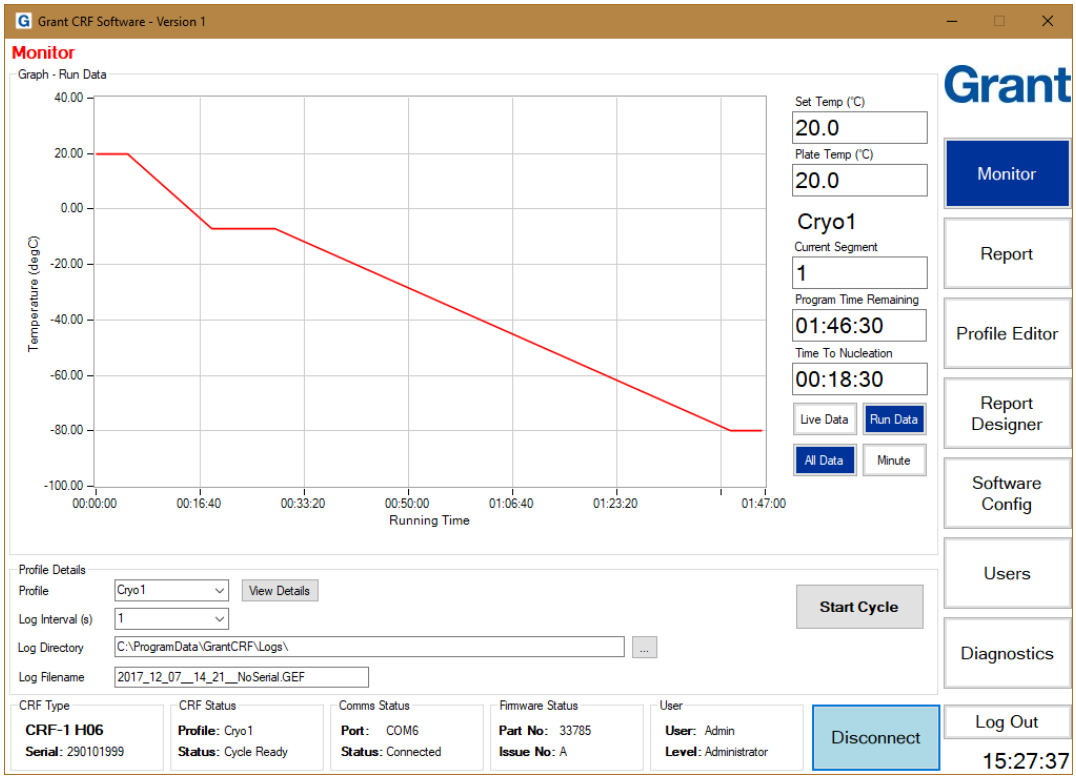


Figure 16: Software: Status bar

The software suite can now be used to control the freezing cycle as well as configure the freezer, edit and create new freezing cycles, as well as generate reports with graphs and data of each freezing cycle run.

6.3. Running a profile

Profiles can be set up and run using the ***Monitor*** page.

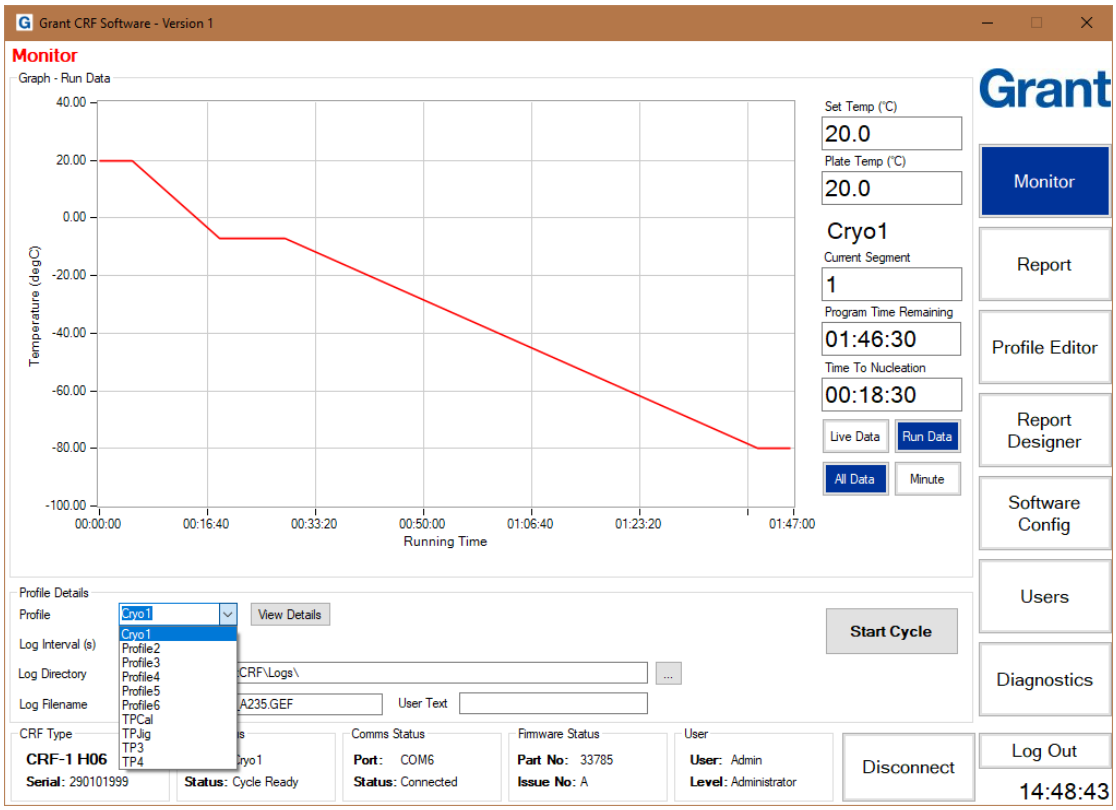


Figure 17: Software: Choosing Profile

6.3.1. Selecting the profile & details

Before running a cooling profile check the following are correct:

- The cooling profile
- The logging interval
- The name and storage folder of the log file

6.3.1.1. Choosing the profile to run

In the ***Profile Details*** window choose a profile from those loaded on the CRF-1 from the ***Profile*** options (e.g. Cryo1 in Figure 17). The specific temperature details of the chosen profile can be viewed by clicking on the ***View Details*** box. This will help ensure you have chosen the correct profile before it is run.

6.3.1.2. Setting the log file parameters

Set the ***Log Interval***, ***Log Directory*** and ***Log Filename*** in the ***Profile Details*** window. The log interval is normally set at 2 seconds for typical cryopreservation applications with run lengths up to approximately three hours (e.g. stem cell, blastocysts, mouse embryo, somatic cells etc).

Note: Shorter logging intervals and long cryopreservation profiles result in very large log files that may not provide any more information. For applications that need prolonged freezing stages, the interval can be increased to up to 60 minutes between logging points dependent on application.

The location and filename of the directory where the log files are stored should be confirmed before starting the run.

Note: The default settings for the profile details can be changed using the Software Config page (see Section 6.7 for further details)

6.3.2. Running the profile

When all of the settings are correct and the CRF-1 is at the start temperature (indicated by the 'Ready' light being on) you are ready to perform a cooling run. Load the samples onto the cooling plate according to the appropriate protocol and click ***Start Profile***.

Note: Clicking the Stop Profile button will abort the run and the CRF-1 will return to the start temperature.

6.4. Creating reports

6.4.1. Generating a run report

6.4.1.1. Report formats

The software stores log files and an event log in an encrypted GEF file format. This ensures that source files cannot be altered without detection and are suitable for validation and archiving purposes.

Four reports can be produced from the encrypted log file using the ***Report*** page:

- ***PDF Detailed Report:*** Includes a graph of the actual and set temperatures, a profile summary section, a notes box for handwritten information and a quality control box which gives a detailed analysis of each profile segment.
- ***PDF Standard Report:*** As the PDF Detailed Report except with no quality control analysis box.
- ***TSV Detailed Report:*** Includes a profile summary section and a quality control box which gives a detailed analysis of each profile segment. Suitable for a program capable of displaying TSV (Tab Separated Variable) data such as Excel.
- ***TSV Standard Report:*** As the TSV Detailed Report except with no quality control analysis box.

6.4.1.2. Generating PDF reports

Click ***PDF Standard Report*** or ***PDF Detailed Report*** to produce a PDF report. Browse to an GEF log file using the dialog box which appears then click ***Open*** (see **Figure 18**). The relevant report will appear in the viewing area. The report can be saved or printed by clicking on the ***Save*** or ***Print*** buttons at the foot of the page.

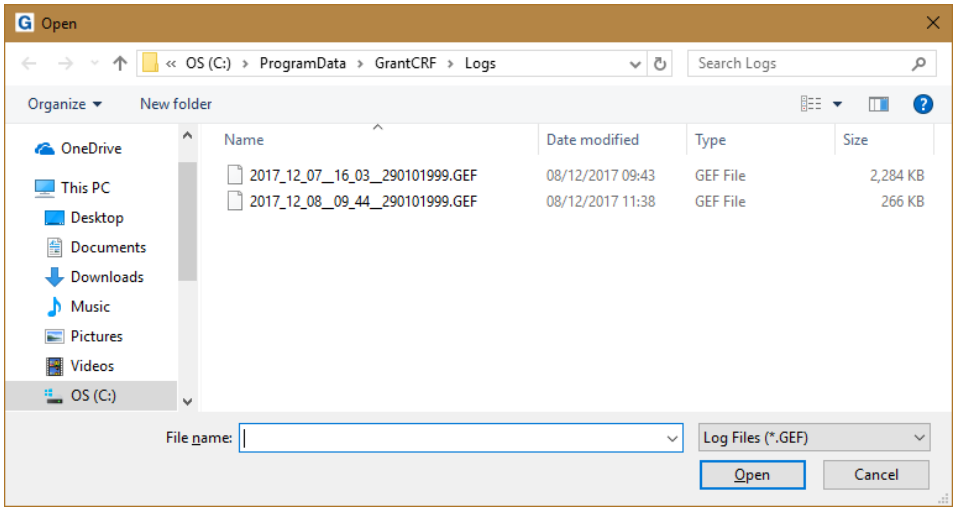


Figure 18: Software: Generating PDF Reports

6.4.1.3. Generating TSV reports

Click ***TSV Standard Report*** or ***TSV Detailed Report*** to produce a TSV report. Browse to an GEF log file using the dialog box which appears then click ***Open***. The relevant report should be saved in a suitable location using the dialog box which appears (see Figure 19).

Note: The TSV will not appear in the viewing area.

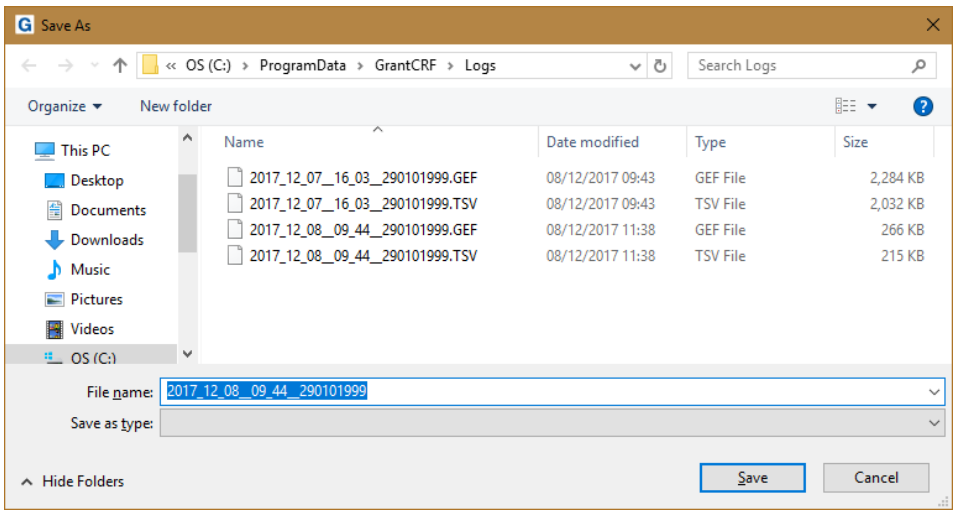


Figure 19: Software: Generating TSV Report

6.4.2. Running a TSV log file conversion

Click ***GEF to TSV*** in the ***Encryption Conversion*** window on the ***Report*** page. Highlight the GEF file to convert and click ***Open***. The TSV file should be saved in a suitable location using the dialog box which appears.

Note: The conversion provides a TSV file of the data log file (all data points recorded) rather than a summary of the profile run.

6.4.3. Generating an event log report

The software automatically creates a log of all key events as an encrypted GEF file. This information includes every user interaction, condition and profile status. Click ***PDF Event Log*** in the ***Reports*** window on the ***Report*** page. Highlight the event log GEF file and click ***Open***. The PDF report will appear in the viewing area. The report can be saved or printed by clicking on the ***Save*** or ***Print*** buttons at the foot of the page.

A TSV format report can also be generated using the above method.

Note: The location of the event log can be changed according to your file system (see Section 6.7 for details).

6.5. Editing and creating profiles

The CRF-1 is configured with a standard set of profiles specific to the model type but the software provides a great degree of flexibility in programming new profiles.

Note: Do not press the "Start" button on the CRF while editing profiles. This may cause corruption of profiles.

Note: Some CRF-1 'special' models may have been configured with application specific profile(s) upon installation

Note: The CRF-1 is designed for use by a professional user, it is the user's responsibility to ensure the profile created or used meets their needs.

The ***Profile Editor*** page contains three windows:

Profile Management – provides backup and restore functions for profiles
Profile Graph – provides a graph of the selected profile
Profile Editor – shows all the parameter values of the selected profile and enables editing of a profile or the creation of a new one

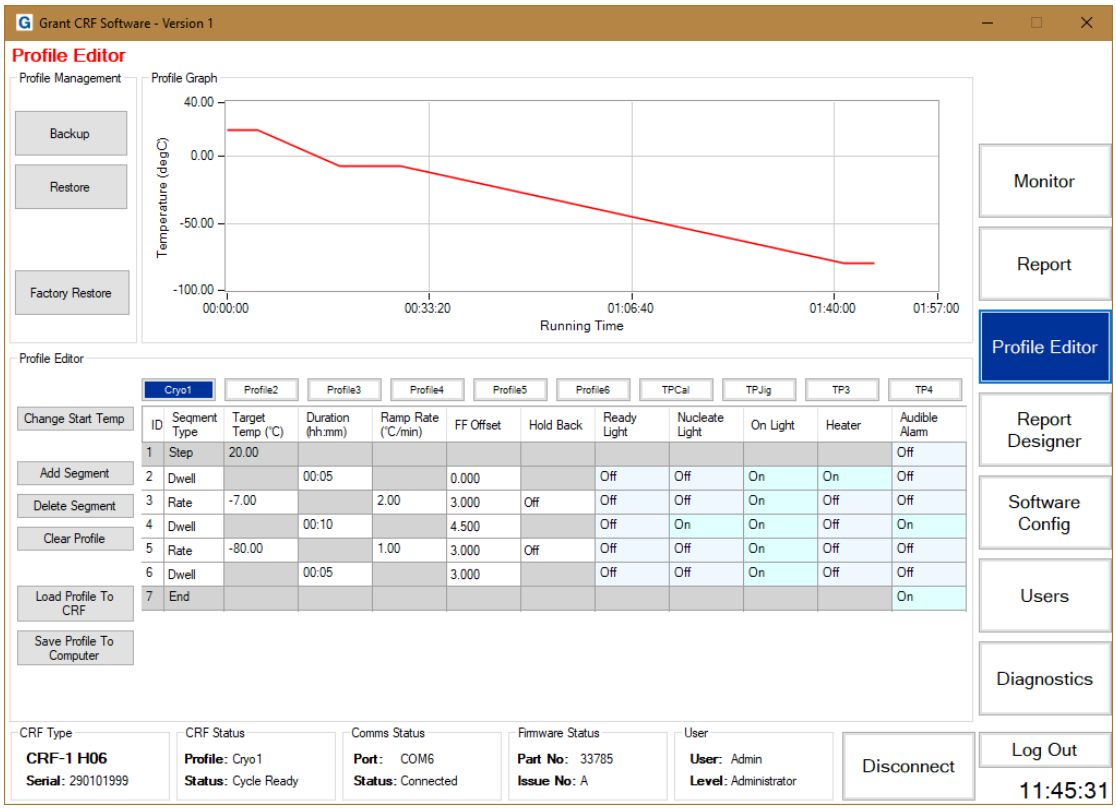


Figure 20: Software: Profile Editor

6.5.1. Editing an existing profile

The following sections provide information about how to change cooling profiles.

Note: When connected to a CRF-1 the changes made in the software are applied immediately to the cryocooler. Backups of all profiles or individual profiles should be created before making any significant changes.

6.5.1.1. Changing the start temperature

Note: It is only possible to have one start temperature for all profiles and changing this value will update all profiles. Care and consideration should be taken before changing this parameter.

Click ***Change Start Temp*** to change the universal profile start temperature. Update the ***Start Temp*** box with the required value and click ***Set Temp*** in the ***Set Start Temp*** dialog box. Click ***OK*** in the ***Confirm*** dialog box to apply the change to all profiles.

6.5.1.2. Adding a segment

Click the segment ***ID*** box to highlight the segment position where the new segment is to be placed. Click ***Add Segment***. A new ***Rate*** segment will always be added after the segment that is highlighted (see Figure 21) except when the last segment is selected, in which case the new segment is added before the end. The segment type can be changed after the segment has been added.

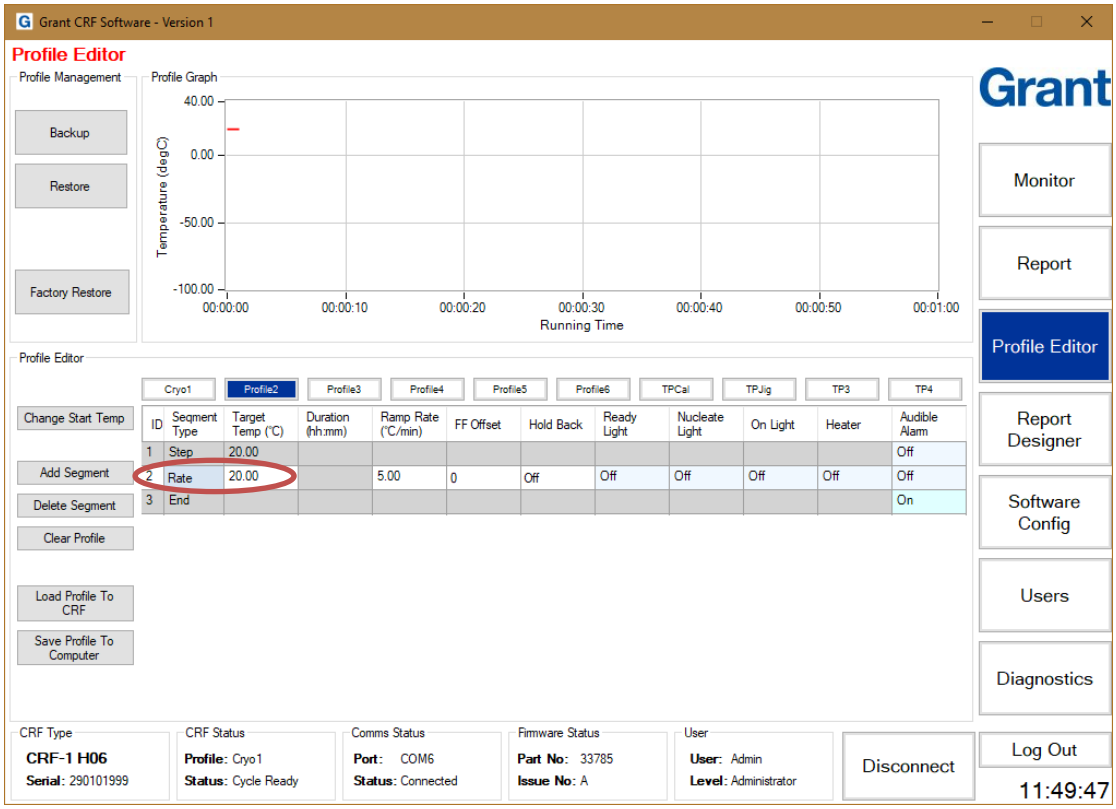


Figure 21: Software: Profile Editor – Adding Segment

6.5.1.3. Deleting a segment

Click the ***ID*** box of the segment to be deleted. The number is highlighted in light blue to show it has been selected. Click the ***Delete Segment*** button. The segment is removed and the ID numbers for the other segments are adjusted.

6.5.1.4. Clearing a profile

Clearing a profile will remove all segments. Click ***Clear Profile*** button. A ***Profile Warning*** dialog box opens up to confirm the deletion of the selected profile (see Figure 22 – the profile name highlighted in blue – in this example this is 'StemT2').

Note: Do not delete the profiles with names beginning "TP" – these are factory test profiles.

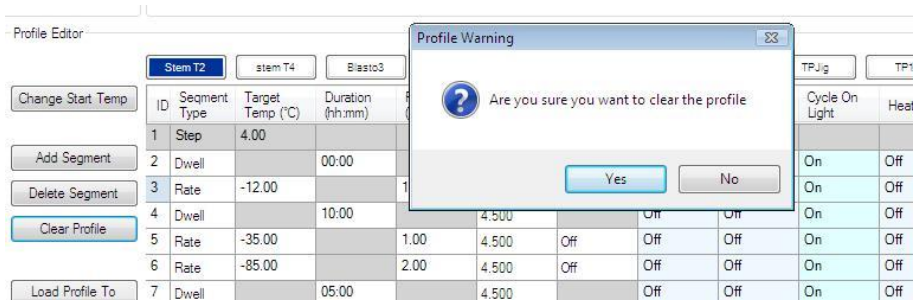


Figure 22: Software: Profile Editor – Clearing Profile

6.5.2. Setting the parameters of a profile

The segments of all cooling profiles can be modified to create variations of standard profiles or to create completely new profiles. Editing segments within existing profiles is permanent and should only be done with care and consideration. It is recommended that profile backups are taken prior to any profile editing.

Note: The CRF-1 is designed for common cryopreservation applications and most profile parameters can be easily met. The maximum or minimum parameter values for a segment represent the highest or lowest value that can normally be achieved. Any profile created or modified should be tested to confirm it is suitable for the given application. If the maximum or minimum rate cannot be achieved in practice, then the CRF-1 will reproducibly deliver the fastest or slowest value that can be achieved within the specific set up.

6.5.2.1. Changing a profile name

To change a profile name, double click on the name box of the profile to be edited. The name box turns light blue and the ***Profile Graph*** window will show the profile curve. Type in the new profile name (up to 8 characters only) and press 'enter'. The profile name will change and the box will turn dark blue to show it is still selected.

6.5.2.2. Segment ID

The ID is the numerical position of the segment in a profile. It is automatically adjusted by the software.

6.5.2.3. Changing the segment type

Click once in the ***Segment Type*** box of the segment to be changed. The ***Segment Type Entry*** dialog box will appear (see Figure 23). Click the appropriate segment type to select it. If no change is required, click on the X in the top right hand corner of the ***Segment Type Entry*** box to close and keep the original segment type.

Each segment can be one of three types:

Rate – precisely controls the rate of temperature change in °C/min

Dwell – temperature is maintained at the target temperature for a defined time
Step – makes a step change of the temperature, the CRF-1 will react as fast as it is able

Note: Segment 1 will always be automatically set as a Step segment. The parameter values for this segment cannot be changed except the Audible Alarm. The Target Temp for this first Step segment will be the same as the Start Temperature.

Note: The last segment will always be automatically set as an End segment. The End segment will maintain the final Target Temp until the profile is stopped or the CRF-1 is switched off.

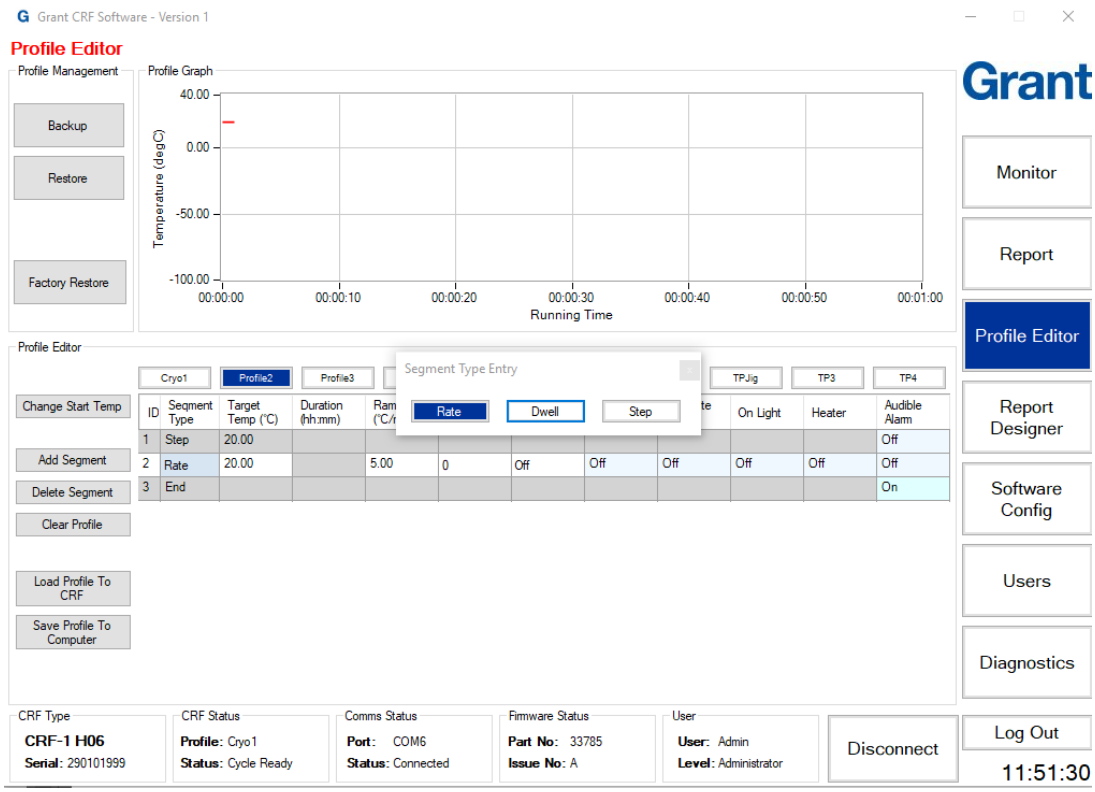


Figure 23: Software: Profile Editor – Segment Type

6.5.2.4. Setting the segment target temp (rate only)

Click once in the ***Target Temp (ºC)*** box of the segment to be changed. The background will change from white to light blue to indicate it has been selected. Type in the new desired target temperature and press 'enter' to store the value.

Note: The target temperature of an individual segment must be lower than the previous segment and higher than the following segment. If multiple new segments have been added to a profile then it may appear that they cannot be changed (since the target temperature defaults to the previous segment value). If this happens, then start by changing the target temperature of the last segment and work backwards to the first segment.

6.5.2.5. Setting the segment duration (dwell only)

Click once in the ***Duration (hh:mm)*** box of the segment to be changed. The ***Duration Editor*** dialog box will appear (see Figure 24). Click on the arrows in the ***New*** boxes to increase or decrease the duration and then click ***Accept***.

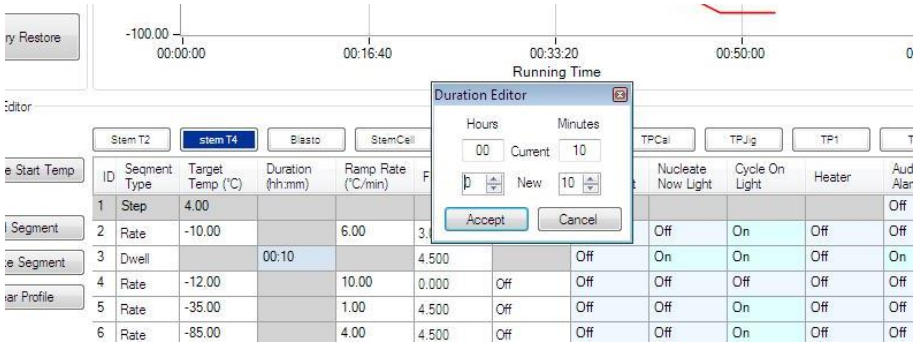


Figure 24: Software: Profile Editor – Segment Duration

Note: The maximum dwell time that can be set is 10 hours and 59 minutes. The minimum dwell time is one minute.

6.5.2.6. Setting the segment ramp rate (rate only)

Click once in the ***Ramp rate (ºC/min)*** box of the segment to be changed. The background will change from white to light blue to indicate it has been selected. Type in the new desired ramp rate (to one decimal place) and press 'enter' to store the value.

Note: The ramp rate is limited to a range of values depending on the model type. Values entered outside the range will default to the highest or lowest acceptable rates.

Note: Although a valid ramp rate is chosen the unit may still not be able to follow it, please refer to the Troubleshooting section of the CRF-1 Operating Instructions for more information.

6.5.2.7. Setting the segment FF offset

This value is predefined and should not need to be altered.

Note: If the value displayed for a new segment is very different from the existing values please contact your distributor or Grant for support and advice.

6.5.2.8. Setting the segment hold back

This value is predefined and should be set to ***Off***.

6.5.2.9. Setting the indicator light configuration & heater state

Click once in any of ***Ready Light***, ***Nucleate Light, On Light*** or ***Heater*** boxes of the segment to be changed. The ***Profile Output Editor*** dialog box opens (see Figure 25) which allows all of these parameters to be adjusted.

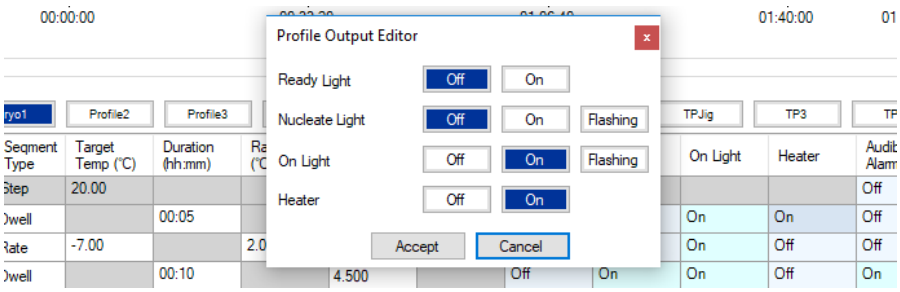


Figure 25: Software: Profile Editor – Profile Output Editor

The ***Ready Light*** is always used to indicate that the CRF-1 is stable at the start temperature. Depending on the ambient conditions it will take approximately 10 minutes for the CRF-1 to

become stable at the start temperature. During this phase the temperature may 'overshoot' but will quickly stabilise. The ***Ready Light*** can be used to indicate when a specific stage of a profile is reached but this is normally rare.

The ***Nucleate Light*** is normally used to indicate when a nucleation phase occurs and when there is a need for manual intervention (i.e. to initiate the nucleation or 'seeding'). The 'Nucleate' light on the front of the CRF-1 is controlled by this parameter. The light can be switched on or off or set to flash as required.

The ***On Light*** is normally used to show a profile is running and to deter a user from using the CRF-1 until the profile has finished. The 'On' light on the front of the CRF-1 is controlled by this parameter. The light can be switched on or off or set to flash as needed.

The ***Heater*** is used to allow temperature control around or above ambient temperature. The heater should be switched off unless:

- The profile is maintaining a dwell temperature of 0°C or greater
- The profile is cooling at a controlled rate down to 0°C

Note: The heater should not be set to on during a cooling segment below 0°C for any segment type.

6.5.2.10. Setting the audible alarm

Click once in the ***Audible Alarm*** box of the segment to be changed. The ***Alarm Editor*** dialog box will appear. Choose whether to set the alarm to sound or not. The 'Mute' button on the front of the CRF-1 is used to silence the audible alarm that is triggered by this parameter

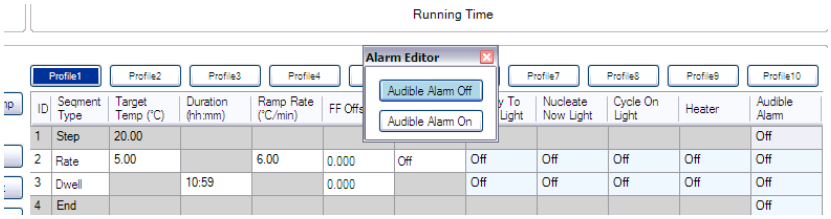


Figure 26: Software: Profile Editor – Audible Alarm

Note: The audible alarm is often linked to a nucleation phase to warn the user that manual intervention is needed at that time. It can also be used to indicate the unit has finished running a profile, or in other segments if required. Note the buzzer is triggered by the control going from low to high. It must be set to off in the previous segment for it to operate.

6.5.3. Saving & loading profiles

Profiles created can be saved onto the PC such that they can be restored in the future. This also allows many profiles to be created and stored without exceeding the limit of 10 profiles available in the CRF-1. Saved profiles can be loaded back into the CRF-1 in place of any other profile.

6.5.3.1. Saving profiles to the computer

Click on the name box of the profile to be saved. The name box turns dark blue and the ***Profile Graph*** window will show the profile curve. Click ***Save Profile To Computer***. The ***Save As*** dialog box opens (see Figure 27) with the default file name. Click ***Save*** to save with the default file name.

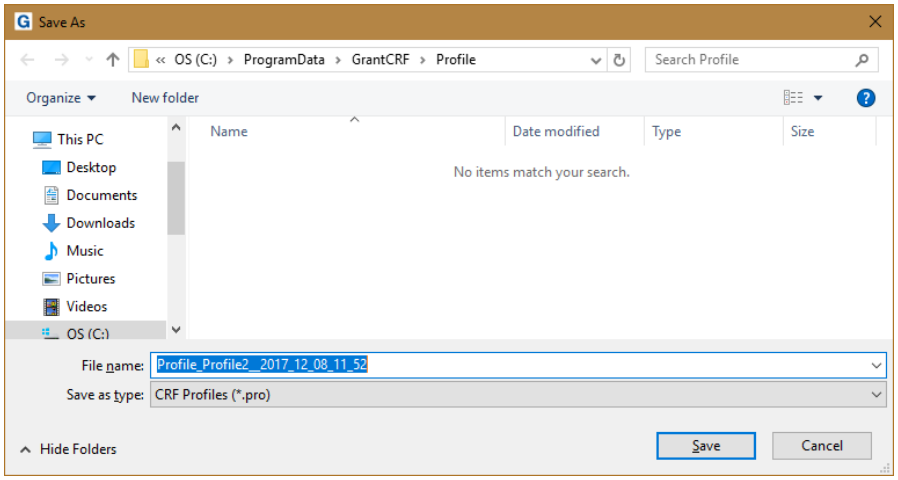


Figure 27: Software: Saving Profiles

Note: If the user input name (Profile2 in Section 6.5.3.1) is changed when saving the profile (e.g. to Profile3) the filename will use the new name BUT the name of the profile (as it appears in the name box on the Profile Editor page) will still be Profile2. It is therefore recommended that the default profile filename is used to avoid confusion.

6.5.3.2. Loading profiles to the CRF-1

Note: The CRF-1 must be connected and synchronised before profiles can be loaded to it.

Click on the name box of the profile from the 10 displayed on the screen where the profile is to be loaded. The name box turns dark blue.

Note: When a profile is loaded into a specific slot it will overwrite (permanently) the previous profile stored. Care and consideration should be taken before loading profiles as information can be lost.

Click ***Load Profile To CRF-1***. The ***Open*** dialog box opens in the default profiles directory and shows all saved profiles. Select the profile then click ***Open*** to load the profile. The profile is loaded to the CRF-1 and the name box initially highlighted will now display the name of the new profile.

6.5.4. Creating & restoring backups

A back up of all profiles can be saved onto the PC such that they can be restored in the future if necessary.

Note: To create a backup of profiles that are installed in the CRF-1 then the unit MUST be switched on and connected. Taking a backup when not the CRF-1 is not connected will only back up offline profiles.

6.5.4.1. Creating a backup

Click on ***Backup*** in the ***Profile Management*** window to create a backup of all of the profiles. A ***Save As*** dialog box opens (see Figure 28). Edit the backup name if required and click ***Save***.

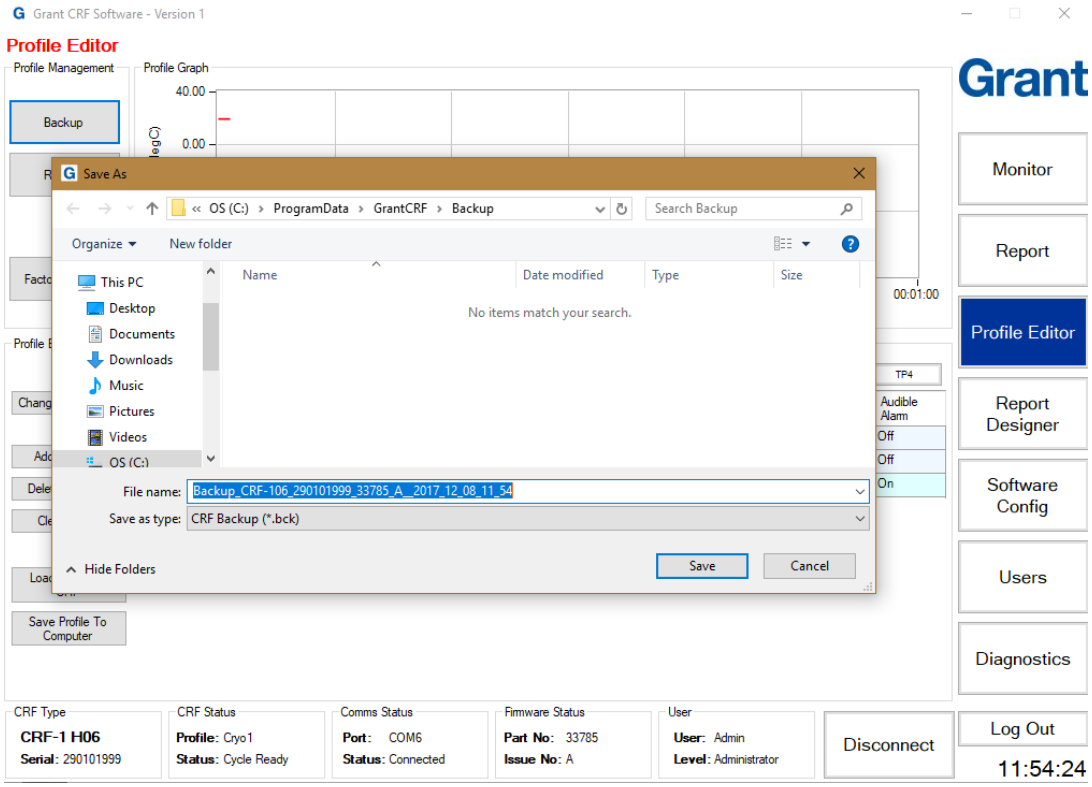


Figure 28: Software: Creating backup

6.5.4.2. Restoring a backup

Click on ***Restore*** in the ***Profile Management*** window to restore a backup of all of the profiles. Select the backup to restore from the list in the ***Open*** dialog box and click ***Open***. The ***Restore Profile Selection*** dialog box opens (see Figure 29). Tick the checkboxes of the individual profiles that you wish to restore. Leave all checkboxes ticked to restore all profiles. Click ***Accept*** to confirm the restore.

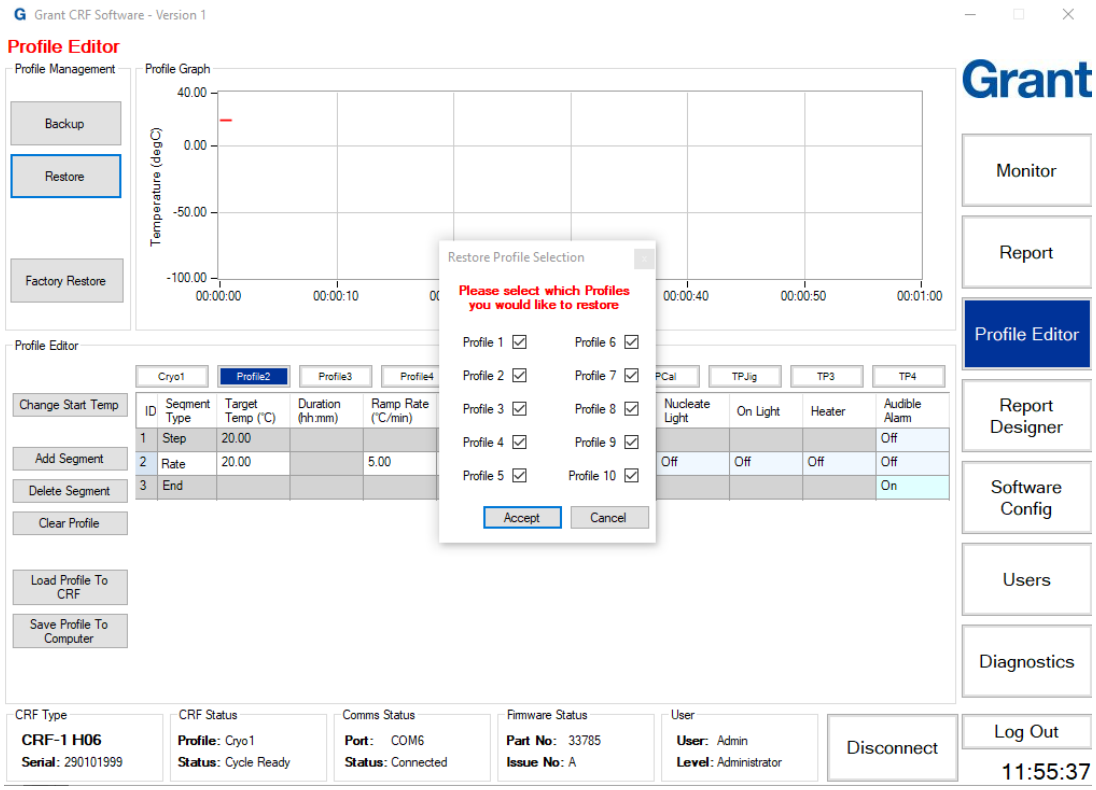


Figure 29: Software: Restoring backup

6.5.4.3. Completing a factory restore

It is possible to restore the CRF-1 back to its original factory configuration. Prior to completing a factory restore please contact your main distributor or Grant Instruments for technical support.

6.6. Designing and configuring reports

The reports created are based on a standard template that can be customised via the ***Report Designer*** page.

6.6.1. User defined report header

The ***Report Template*** has two header fields at the top of the report which can be determined by the user. The header fields on the top left and top right can be set to either an image or text using the ***Text*** box or the ***Logo*** box. Figure 30 shows an example where '***Grant.jpg'*** has been added to the left hand ***Logo*** box and ***'Program Series A1'*** text added to the right-hand ***Text*** box.

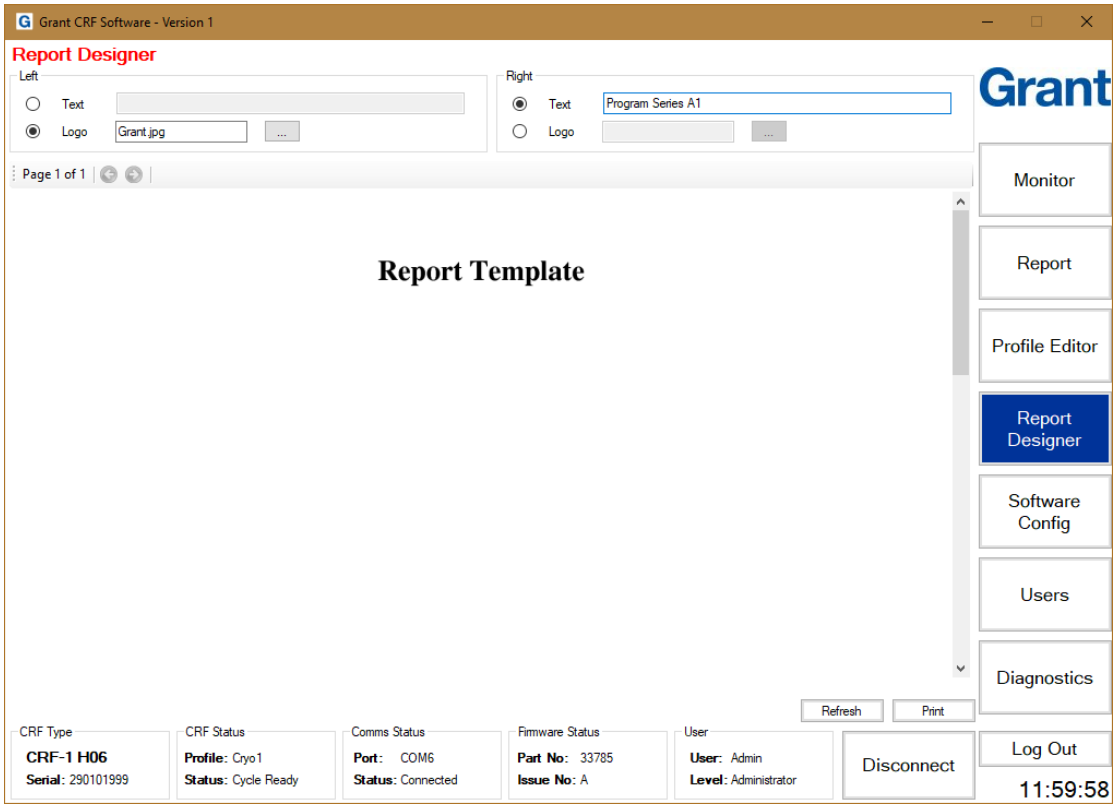


Figure 30: Software: User Defined Report Header

These user defined conditions will appear at the top of a generated report (see Figure 31)

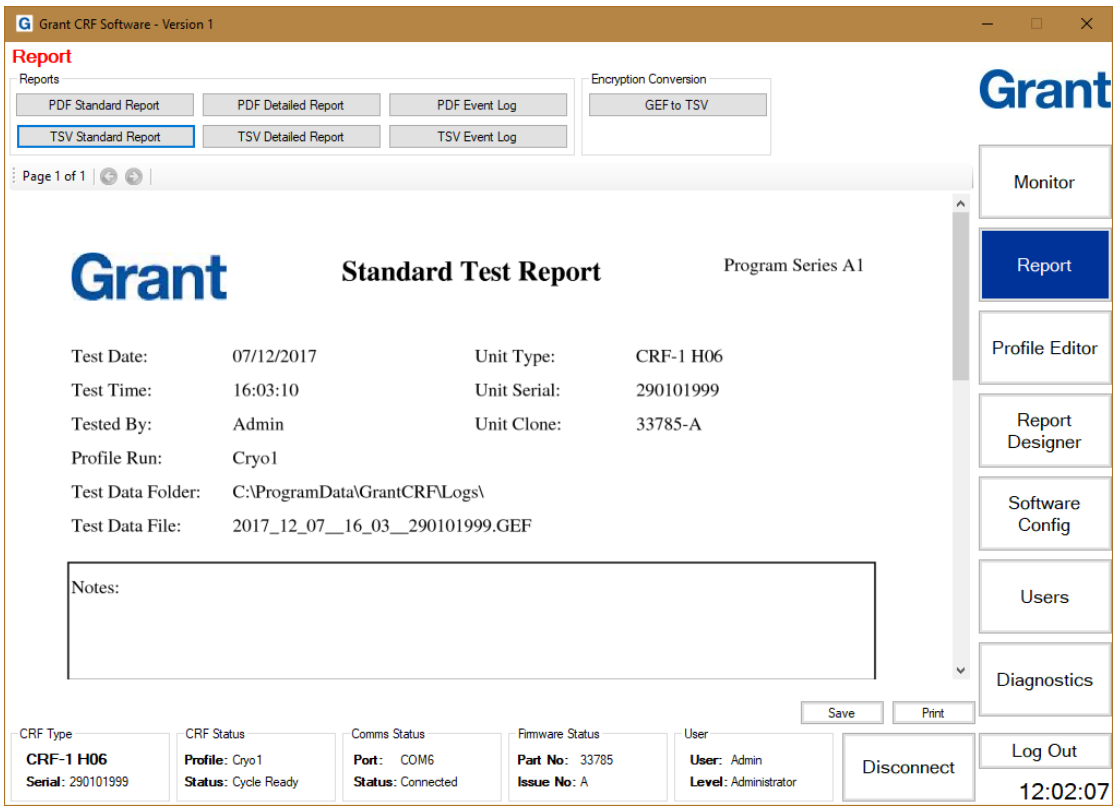


Figure 31: Software: User Defined Report

6.6.2. Report template storage location

The default storage location of the report template is defined in the ***Report*** window on the ***Software Config*** page. This location can be changed if required.

6.7. Configuring the software

All parameters and defaults can be changed in the ***Software Config*** page.

6.7.1. Log file configuration options

The default directory where the log files are stored is not normally in the folder where the main software is located. The default log file location is:

C:\ProgramData\GrantCRF\Logs;

The log file location can be varied to suit your file set up and can be a network share. Click on the “Browse For Folder” button next to the ***Default Dir*** option to choose a new location see Figure 32.

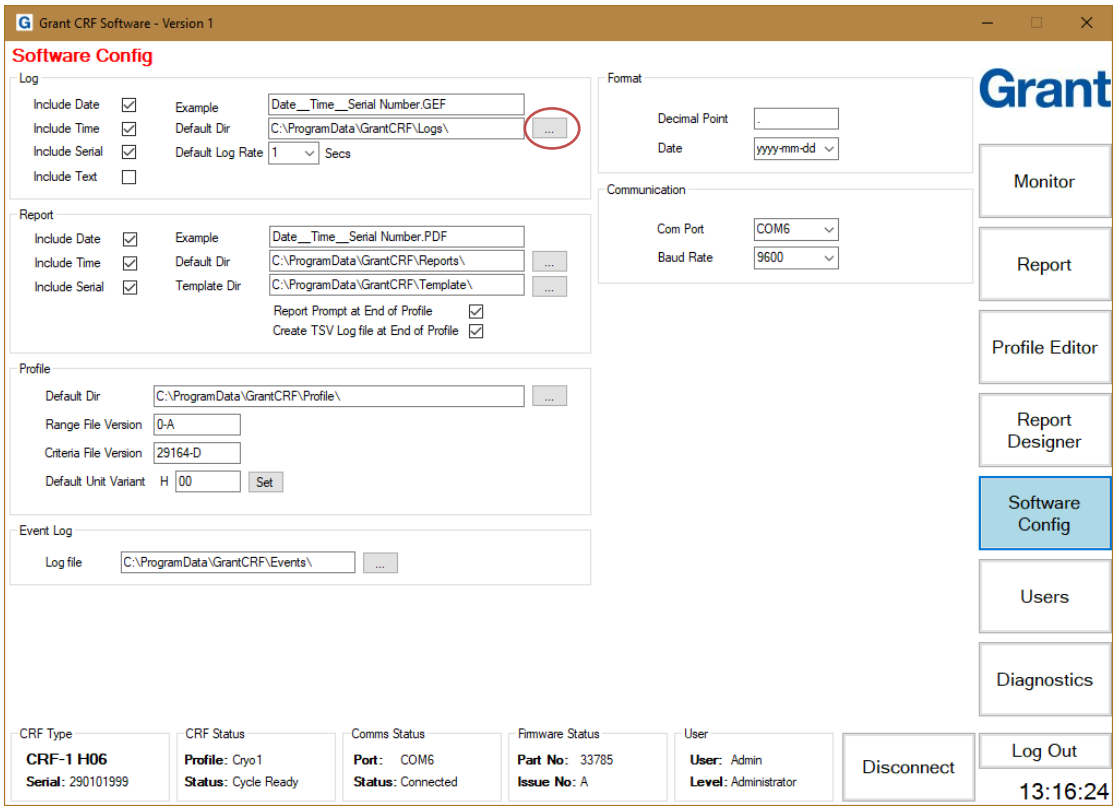


Figure 32: Software: Log File Configuration Options

Note: Ensure that the Windows User has WRITE permissions to the folder where the log file is stored otherwise data may be lost.

By default, each log file will be named based on a combination of ***date*** **_ time _ serial number.*GEF.*** The GEF file is an encrypted format that can be automatically converted to .PDF or .TSV (Tab Separated Variable) by the reporting function.

If date, time or CRF-1 serial number is not required in the log file name, then the default situation can be overwritten. Figure 33 shows the ***Include Date*** box unchecked and no date field appearing in the ***Example*** log file name box.

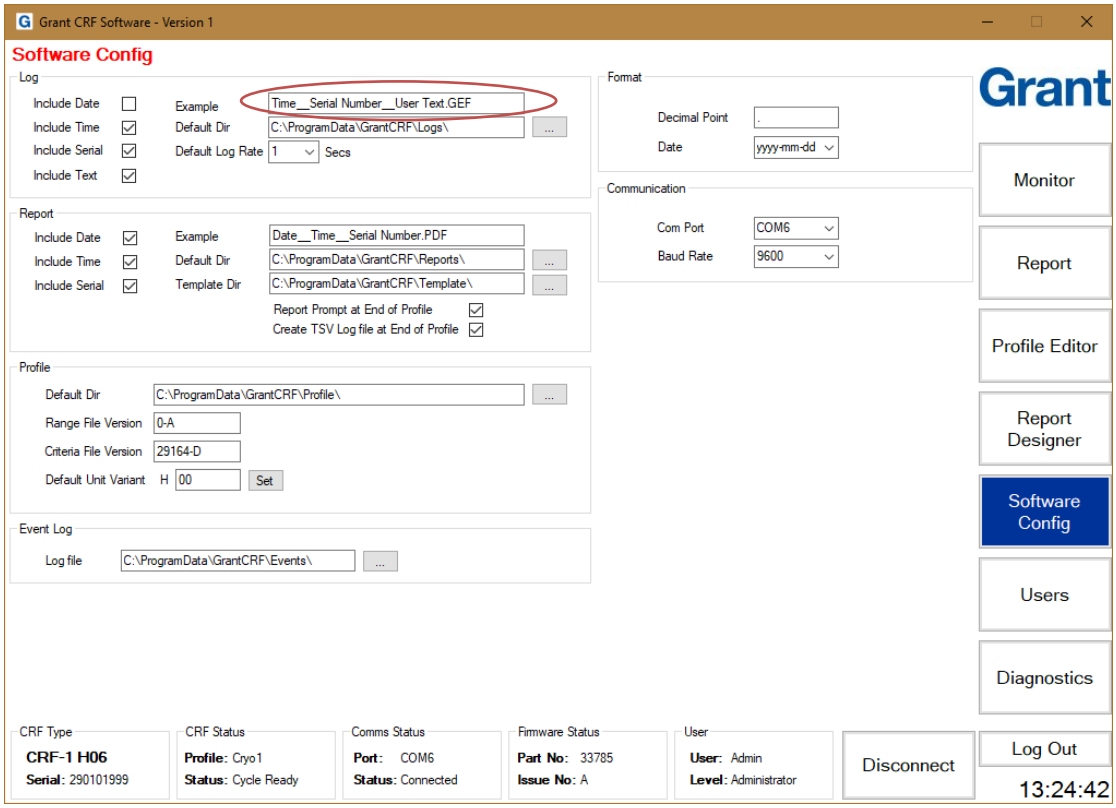


Figure 33: Software: User Defined Conditions

To add user specific text to the file name, such as an assay name, ensure that the ***Include Text*** box in the ***Log*** window on the ***Software Config*** page is checked (see Figure 34)

When the ***Include Text*** box is checked – a ***User Text*** box next to the ***Log Filename*** box on the **Monitor** page becomes active, see Figure 34 (with example user text of '***Test A235'*** in the ***User Text*** box which then appears in the name of the file in the ***Log Filename*** box)

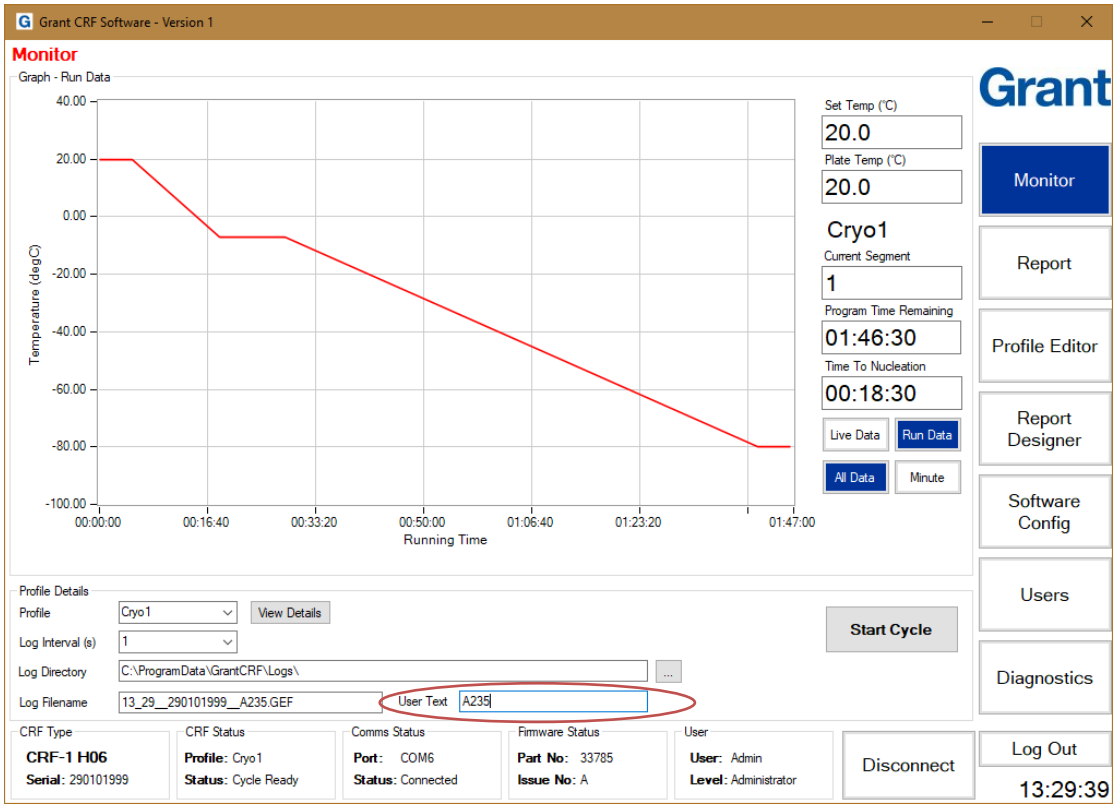


Figure 34: Software: User Text

6.7.2. Report configuration options

The default storage location and naming format for reports is defined in the ***Report*** window on the ***Software Config*** page. The default report name is composed of three fields ***Date_Time_Serial Number*** in a similar manner to the log file naming. If any of these fields is not required then the relevant box (***Include Date, Include Time, Include Serial***) should be unchecked.

Note: The report name can be changed when it is being saved if required

6.7.3. Profile configuration options

The ***Default Dir*** box in the ***Profile*** window defines the default storage folder for saved profiles. This location can be varied to suit your file set up and can be a network share. Click on the “Browse For Folder” button next to the ***Default Dir*** box to choose a new location.

Note: Other software config options in the profile window should not be altered.

6.7.4. Event log configuration options

The ***Log file*** box in the ***Event Log*** window defines the default storage folder for the event logs. The event log location can be varied to suit your file set up and can be a network share. Click on the “Browse For Folder” button next to the ***Log file*** box to choose a new location.

6.7.5. Format configuration options

The ***Decimal Point*** indicator and ***Date*** boxes allow specific formats to be set. The defaults are a ‘full stop’ for decimal point and yyyy-mm-dd for the date. Three formats are available for the date format.

6.7.6. Communication configuration options

The appropriate ***Com Port*** that is connected to the CRF-1 can be set using this option. ***Baud Rate*** should always be set to 9600.

6.8. Users

The software is programmed with one administrator level user: User ID: ***Admin***; Password: ***Admin1***. It is recommended that this user is employed as the main administrator and is not deleted. The password can be changed if required using the notes below.

6.8.1. Creating new users

New users can only be created when logged on as ***Admin*** or another administrator level user. Click ***Users*** to open the ***Users*** page. The first user in the list will be highlighted in blue and the ***User Details*** for that user will be shown (see Figure 35).

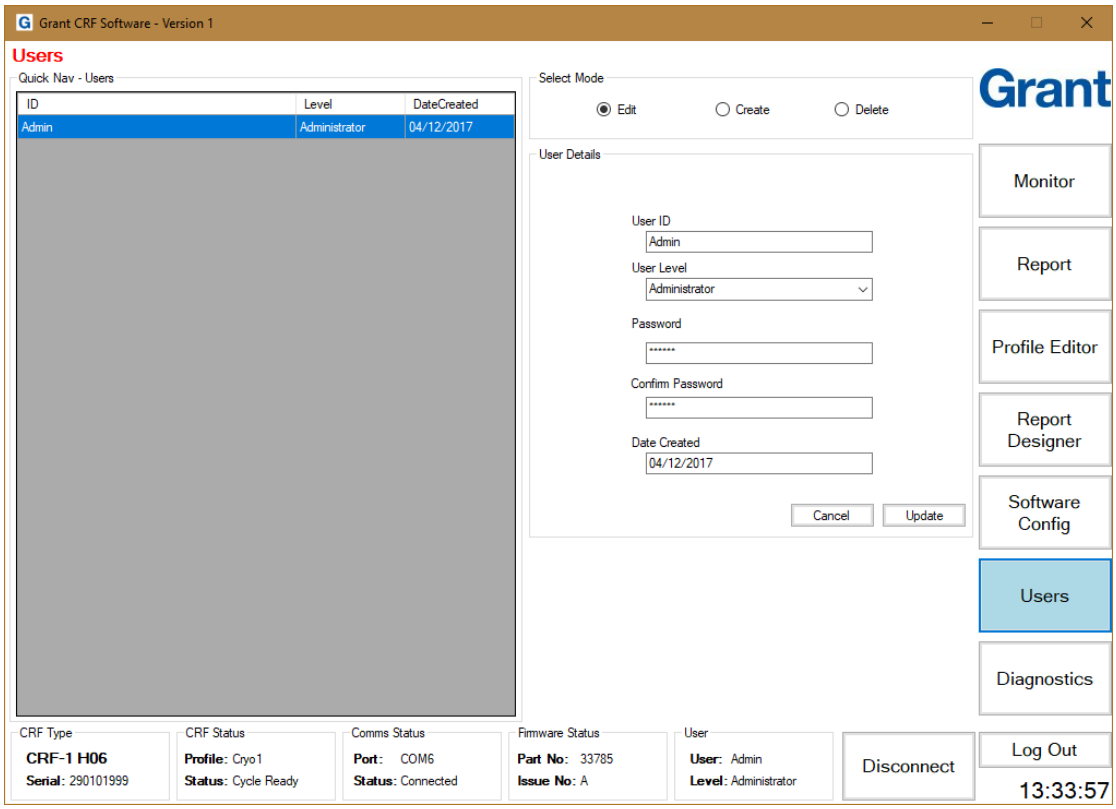


Figure 35: Software: User Details

Select ***Create*** from the ***Select Mode*** window. The fields in the ***User Details*** become available to enter user data. Enter the required information and click ***Update***. The new user appears in the ***Quick Nav – Users*** window.

The ***User Levels*** are defined as follows:
Administrator – has access to all areas of the software with the ability to make changes.
Advanced – has access to Monitor, Reports, Profile Editor and Report Designer only.
Standard – has access to Monitor and Reports only.

6.8.2. Editing existing users

Click the user that needs to be changed in the ***Quick Nav – Users*** window. Select ***Edit*** from the ***Select Mode*** window. The fields in the ***User Details*** are updated to show the selected users information. Update the required information and click ***Update***.

Be sure not to edit the Administrator's User Level. If there is no user with Administrator permissions it will be necessary to reinstall the software to make significant changes.

6.8.3. Deleting existing users

Click the user that needs to be deleted in the ***Quick Nav – Users*** window. Select ***Delete*** from the ***Select Mode*** window. Click ***Update*** to remove the user.

Note: Deleting a user cannot be undone and the user that is currently logged on cannot be deleted.

7. Operating Procedure for the CRF-1 Freezer

7.1. Pre-operation Checks

Before operating, ensure that:

The freezer has been installed and switched on (Section 5)
The software suite has been opened and communications established (Sections 6)

It is recommended that an initial test run is completed using the maximum volume of liquid allowed at the fastest cooling rate to ensure that everything has been set up correctly. This should be done before first use or if the unit has been transported to a different location and is especially important when using the H03 flat plate where the mass of the user supplied vessel and the effectiveness of its thermal coupling to the plate could adversely affect performance. It is also recommended that this test is repeated periodically to ensure consistency of results.

7.2. Nucleating Tool Preparation

The cryopen is a medical device that can be used to nucleate the samples in a sterile environment. If the cryopen has been ordered with the freezer, please refer to Appendix 2 for instructions on how to prepare and use it.

Alternative tools for nucleating (e.g. forceps) may be used if sterility of the operating environment is not essential.

7.3. Sample Preparation

Before preparing samples, remove the lid and check that the sample plate is dry. If there is any condensation on the plate, gently dry it with a tissue or soft cloth. The sample plate should never be dried using a hot-air gun as this could result in damage to the freezer. Care should also be taken not to damage or scratch the surface of the plate during drying or cleaning.

Samples should not be added to the plate until the ‘Ready’ green light on the front of the freezer unit has illuminated

7.3.1.1. CRF-1 H00 – 0.5ml CBS Straws

Load up to 16 high security straws with samples for freezing and seal as recommended by the manufacturer. Thoroughly dry the outside of the straws after preparation to prevent them from freezing to the sample plate.

Position the straws into the sample plate and fully press them into the grooves (Figure 36). Place the lid over the sample plate checking that it is located correctly. Straws should be positioned such that the samples are in the centre of the freezing plate; the label section of the straw can overhang the freezing plate if required.

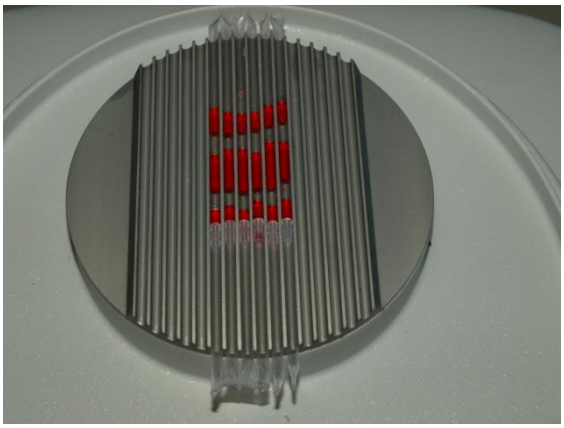


Figure 36: 0.5ml CBS straws positioned on sample plate

7.3.1.2. CRF-1 H01 – 0.25ml IMV Straws

The CRF-1 H01 is designed for the controlled rate freezing of 0.25ml capacity straws, for example the CBS Paillette Cristal. Load up to 18 straws as normal and seal with a straw plug, cristaseal etc. Thoroughly dry the outside of the straws after preparation to prevent them from freezing to the sample plate.

Position the straws into the sample plate and fully press them into the grooves (Figure 37). Place the lid over the sample plate checking that it is located correctly. Straws should be positioned such that the samples are in the centre of the freezing plate; the straw plug should overhang the freezing plate if required.



Figure 37: 0.25ml IMV straws positioned on sample plate

7.3.1.3. CRF-1 H02 – 1.8ml Cryovials, 0.5ml max fill

The CRF-1 H02 is designed for the controlled rate freezing of 12.0mm diameter cryovials including Nunc 363401 and Nunc 340711. The maximum fill for each cryovial is 0.5ml. Load up to 55 cryovials on the sample plate. Cryovials should always be loaded starting at the centre of the freezing plate. Thoroughly dry the outside of the cryovials after preparation to prevent them from freezing to the sample plate.

Position the cryovials (Figure 38). Place the lid over the sample plate checking that it is located correctly.

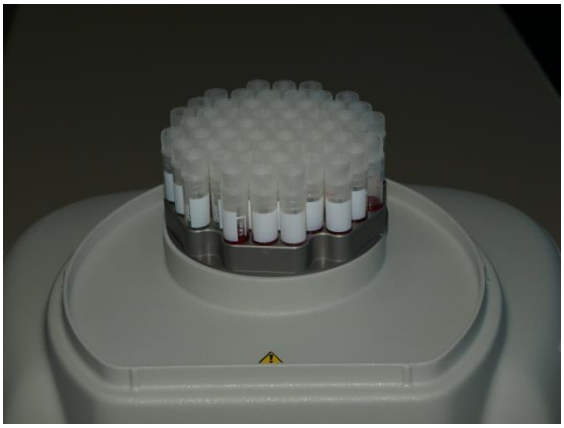


Figure 38: Cryovials positioned on sample plate

7.3.1.4. CRF-1 H03 – Flat Plate

The CRF-1 H03 has been designed with a flat plate to accommodate non-standard vessels.

Note that it is recommended that the user performs one or more test runs to ascertain that the required cooling rate can be achieved with the intended vessel and volume of liquid, see Appendix 1 for more help.

Place the vessel at the centre of the plate such that contact with the plate is maximised to achieve the best thermal transfer required for freezing. Once in position, place the lid over the sample plate checking that it is located correctly.

7.3.1.5. CRF-1 H05 - 1 SBS microplate

The CRF-1 H05 is designed for the controlled rate freezing of a single SBS type microplate including the NUNC cryobank (374084) and FluidX data matrix tube (65-52319). The plate matches the footprint of the microplate exactly. Position the microplate such that the rack is securely located and the tube bottoms are in good thermal contact with the freezing plate (Figure 39: SBS microplate positioned on sample plate).

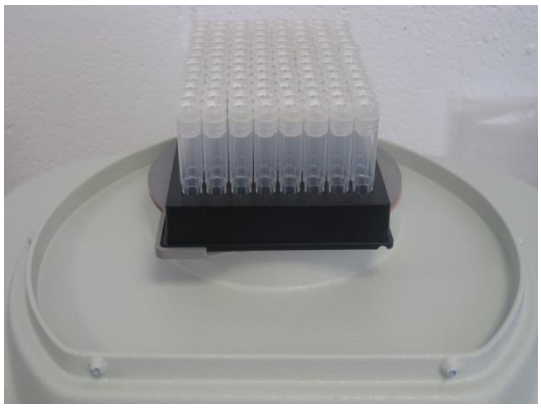


Figure 39: SBS microplate positioned on sample plate

Once in position, place the lid over the sample plate checking that it is located correctly.

7.3.1.6. CRF-1 H06 – 1.8ml Cryovials. 1.0ml max fill

The CRF-1 H06 is designed for the controlled rate freezing of 12.0mm diameter cryovials including Nunc 363401 and Nunc 340711. The maximum fill for each cryovial is 1.0ml. Load up to 55 cryovials on the sample plate. Cryovials should always be loaded starting at the centre of the freezing plate. Thoroughly dry the outside of the cryovials after preparation to prevent them from freezing to the sample plate.

Position the cryovials. Place the lid over the sample plate checking that it is located correctly.

7.3.1.7. CRF-1 H07 & H08 – Combination plates

The CRF-1 H07 and H08 plates are designed for the freezing of 2 different straws or cryovials within the same plate.

CRF-1 H07 – Plate for 10 0.5ml CBS straws & 12 1.8ml cryovials.
CRF-1 H08 – Plate for 12 0.3ml IMV straws & 12 1.8ml cryovials.

For each of these plates, only straws or cryovials can be frozen in one operation. It is not possible to freeze straws and cryovials at the same time. Straws or cryovials should be positioned on the sample plate as defined in previous sections 7.3.1.1, 7.3.1.2 and 7.3.1.3.

7.4. Starting the Freezing Cycle

When the samples have been positioned correctly, put the lid back on. A few more minutes may be required for the plate temperature to stabilise again. When it is ready to start, the Cryocooler status will show ‘Cycle Ready’

Follow the instructions below to start the freezing cycle profile.

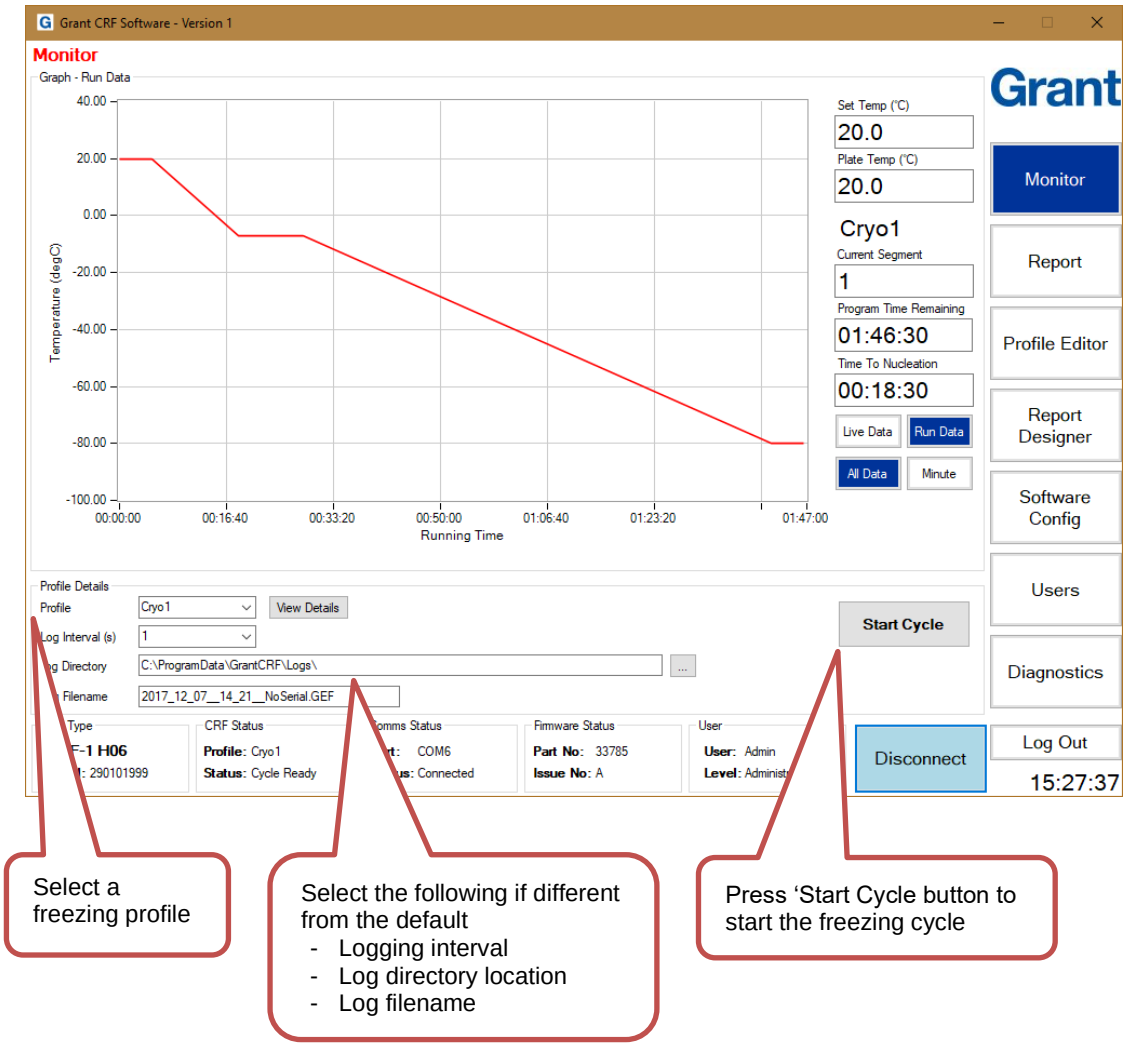


Figure 40: Software: Monitor Panel. Ready to start

Once started, the Cryocooler Status will change to ‘Cycle Running’ and the plate temperature data is plotted on the graph.

During the cycle, the temperature of the plate is logged automatically in an encrypted format. The data is stored in the file location shown on the Monitor panel. At the end of the cycle the data can be used to create a report or analysed for quality control purposes.

Note that the ‘Ready’ light on the freezer unit may remain on for a short time at the same time as the ‘Cycle’ light. It will switch off automatically when the temperature starts to decrease.

7.5. Sample Nucleation

The Monitor Panel constantly shows the time remaining before the Profile reaches the programmed dwell temperature for nucleating samples.

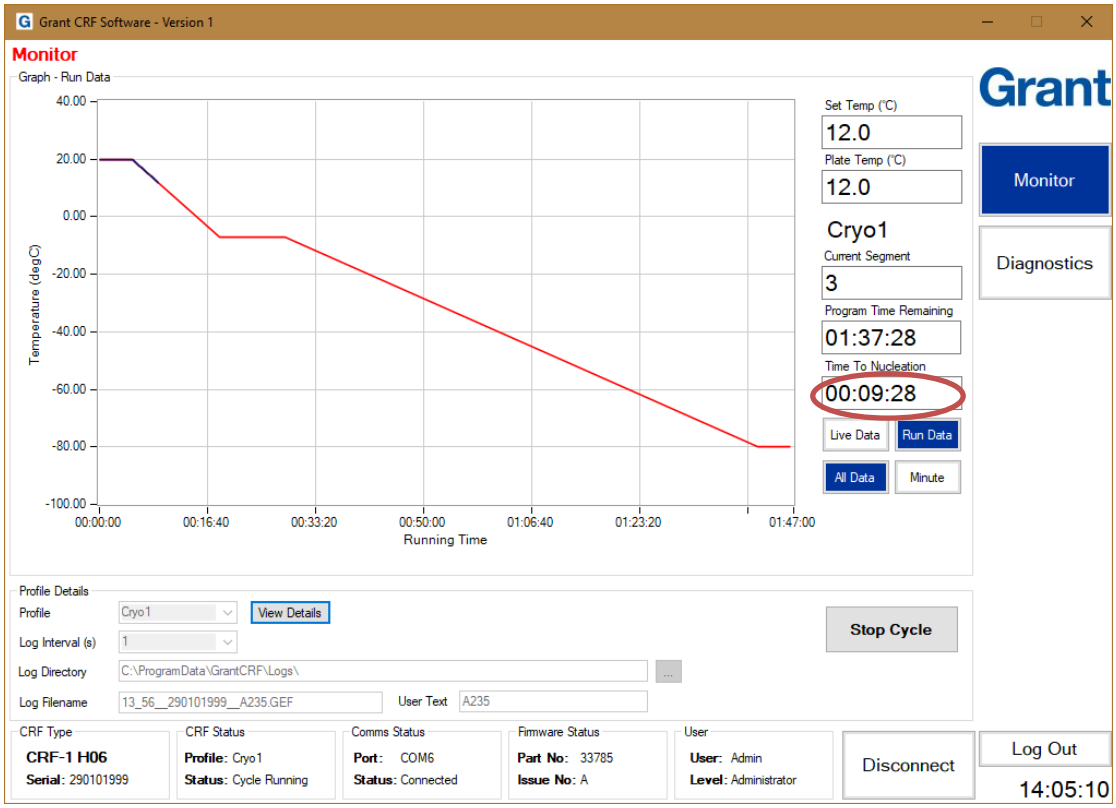


Figure 41: Monitor Panel. Time to Nucleation

When the samples reach the nucleation plateau temperature, the time displayed changes to ‘Nucleate’. The freezer unit buzzer sounds and the yellow ‘Nucleate’ light on the freezer unit illuminates. Nucleation should be done at this time. The programs normally allow a 10 minute period for the samples to be nucleated, unless otherwise specified.

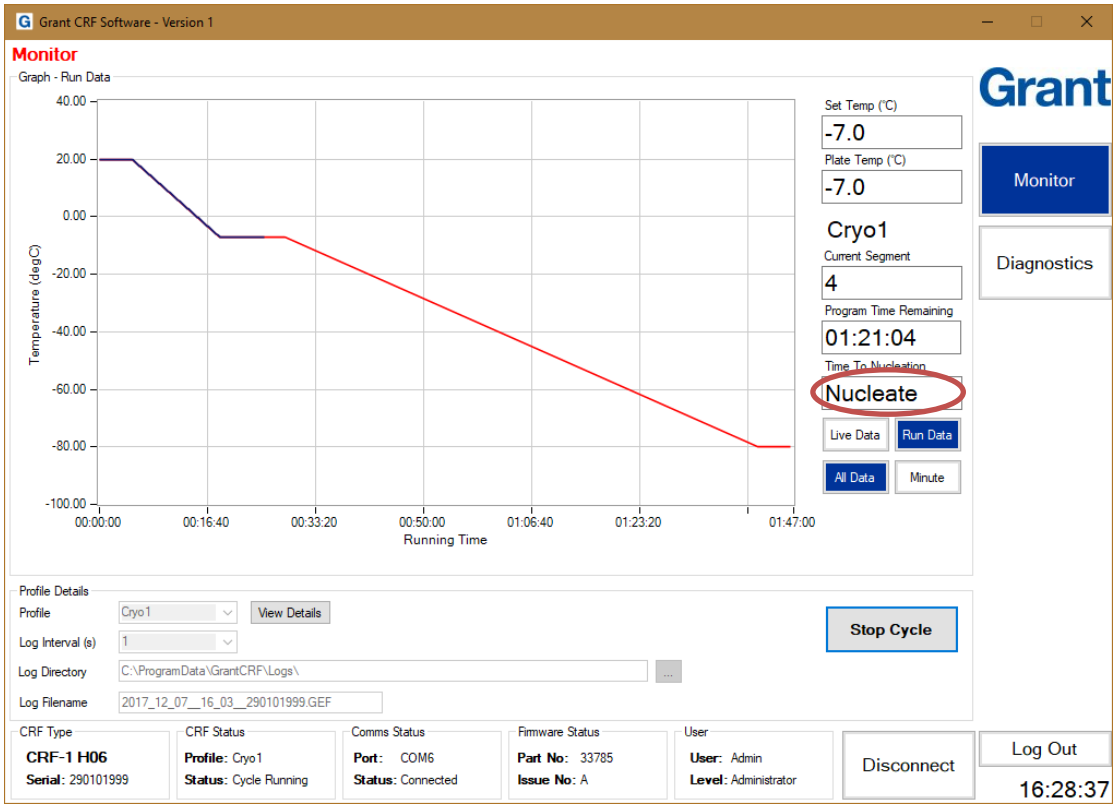


Figure 42: Monitor Panel. Nucleate

Remove the lid of the freezer completely. Turn off the buzzer by pressing the ‘Buzzer Off’ button on the keypad. Touch the nucleating tool (if using the cryopen please refer to the operating instructions for safe use) to the first sample for approximately one second (Figure 43, Figure 44 & Figure 45). Repeat with the remaining samples and replace the lid of the freezer. Note that other methods of ice nucleation may also be employed dependent on experimental procedure.

Inspect the samples approximately one minute after nucleation to verify that there is some ice-growth. If ice has not formed in any of the samples repeat nucleation.

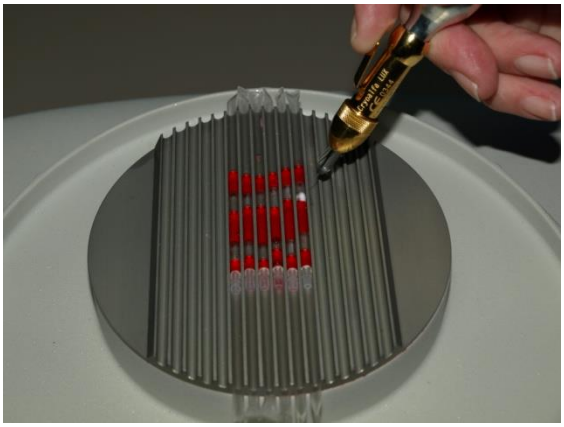


Figure 43: Nucleation of CBS straws (CRF-1 H00)

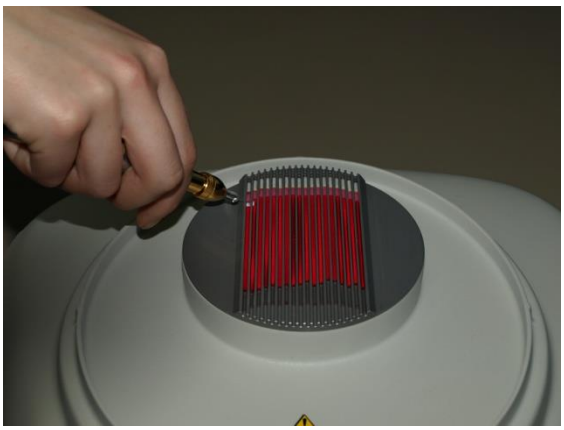


Figure 44: Nucleation of IMV straws (CRF-1 H01)

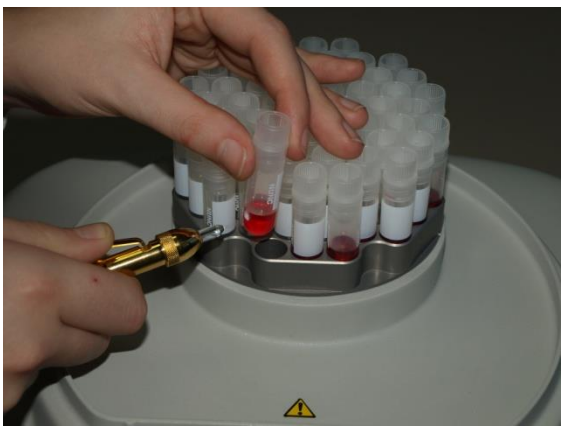


Figure 45: Nucleation of cryovials (CRF-1 H02)

7.6. End of Freezing Profile

When the freezing profile is complete the ‘Program Time Remaining’ will show ‘Complete’ on the Monitor panel. On the freezer unit the yellow ‘Complete’ light will illuminate and the buzzer will sound. The samples can now be removed from the freezer and placed in long term storage. The freezer will hold at the final temperature of the cycle until the ‘Stop Cycle’ button is pressed or the freezer is switched off. Plate temperature data will continue to be logged.

7.7. Stopping the Cycle and Generating a Report.

When all of the vessels have been transferred, click on the ‘Stop Cycle’ button on the software Monitor panel. Temperature logging will stop and the unit will return to the start temperature. The Cryocooler Status changes to ‘Temp Stabilization.’

A message box will appear asking if a report is required. Click ‘Yes’ to generate a report in pdf or TSV (tab separated value) format. Click ‘No’ if a report is not required immediately. It is recommended that a report is generated and checked after each run.

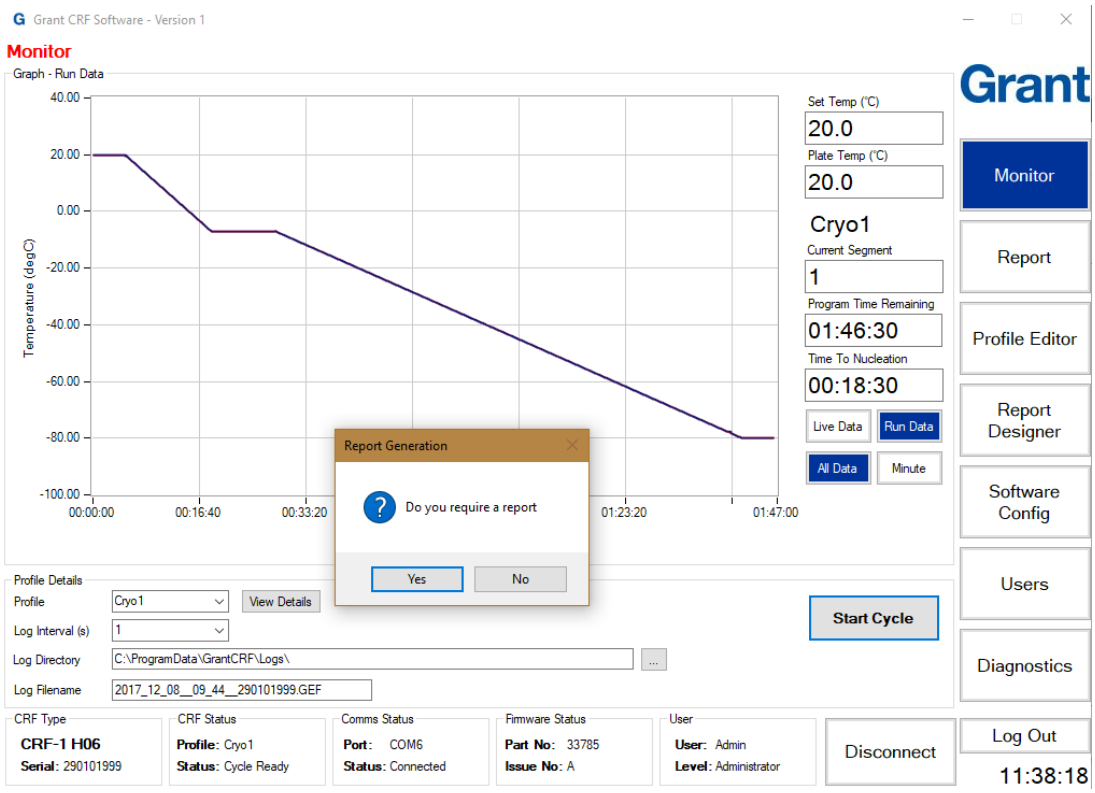


Figure 46: Monitor Panel. End of Cycle

Reports can still be generated at any time after the cycle has been stopped using the Report Panel.

7.8. Starting a New Cycle

Once the freezer has returned to and stabilised at the start temperature, the Cryocooler Status will change to ‘Cycle Ready’. The ‘Start Cycle’ button will no longer be greyed out and more samples can be loaded in preparation for another freezing cycle.

Remember to dry the sample plate between cycles to prevent vessels sticking during the freezing cycle.

8. Freezing Profiles

The CRF-1 series freezers are pre-loaded with specific cooling profiles.

8.1. Embryo Cryopreservation

Profile name: Embryo	Applicable model H00, H01, H03, H07, H08
Start temperature	20°C
Cool	from 20°C to -7°C at 2°C min^{-1}
Hold for nucleation	at -7°C for 10 minutes
Cool	from -7°C to -30°C at 0.3°C min^{-1}
Cool	from -30°C to -33°C at 3°C min^{-1}
Cool	from -33°C to -39°C at 6°C min^{-1}
Cool	from -39°C to -100°C at 10°C min^{-1}
Hold	at -100°C
Transfer to storage	

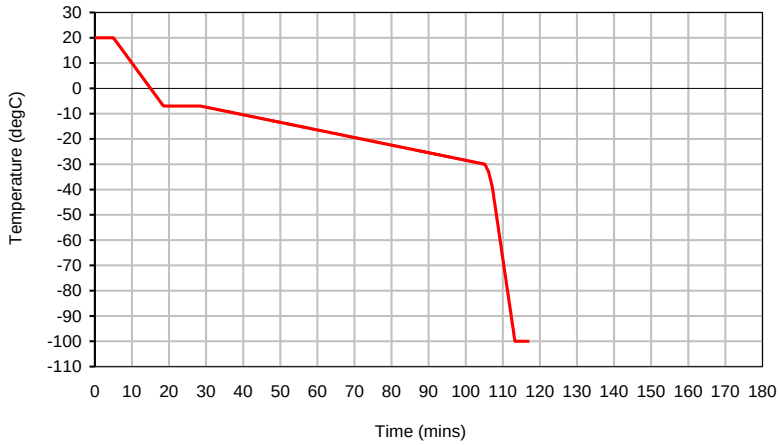


Figure 47: Pre-programmed embryo freezing cycle

8.2. Oocyte Cryopreservation

Profile name: Oocyte	Applicable models: H00, H01, H03, H07, H08
Start temperature	20°C
Cool	from 20°C to -8°C at 2°C min^{-1}
Hold for nucleation	at -8°C for 10 minutes
Cool	from -8°C to -30°C at 0.3°C min^{-1}
Cool	from -30°C to -33°C at 3°C min^{-1}
Cool	from -33°C to -39°C at 6°C min^{-1}
Cool	from -39°C to -100°C at 10°C min^{-1}
Hold	at -100°C
Transfer to storage	

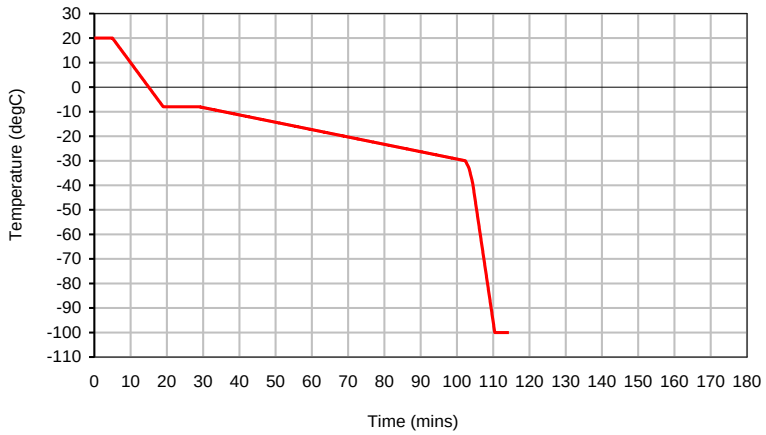


Figure 48: Pre-programmed oocyte freezing cycle

8.3. Blastocyst Cryopreservation

Profile name: Blasto	Applicable models: H00, H01, H03, H07, H08
Start temperature	20°C
Cool	from 20°C to -7°C at 1°C min^{-1}
Hold for nucleation	at -7°C for 10 minutes
Cool	from -7°C to -30°C at 0.3°C min^{-1}
Hold	at -30°C
Transfer rapidly into liquid nitrogen	

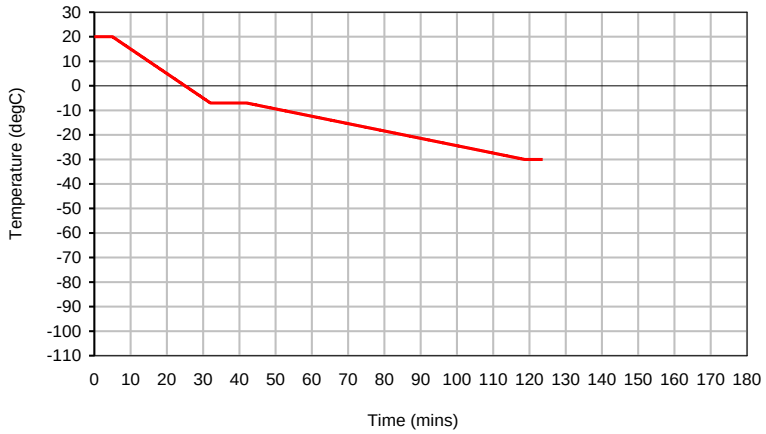


Figure 49: Pre-programmed blastocyst freezing cycle

8.4. Embryonic Stem Cell Cryopreservation

Profile name: Stem Cell	Applicable models: H00, H01, H03, H07, H08
Start temperature	20°C
Cool	from 20°C to -7°C at 2°C min^{-1}
Hold for nucleation	at -7°C for 10 minutes
Cool	from -7°C to -80°C at 1°C min^{-1}
Hold	at -80°C
Transfer to storage	

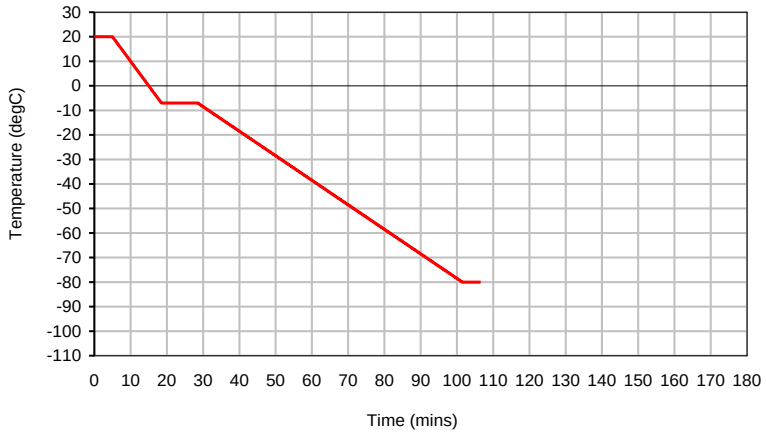


Figure 50: Pre-programmed embryonic stem cell freezing cycle

8.5. Spermatozoa Cryopreservation – Linear Cooling

Profile name: Sperm	Applicable models: H00, H01, H03, H07, H08
Start temperature	20°C
Cool	from 20°C to -7°C at 2°C min^{-1}
Hold for nucleation	at -7°C for 10 minutes
Cool	from -7°C to -100°C at 10°C min^{-1}
Hold	at -100°C
Transfer to storage	

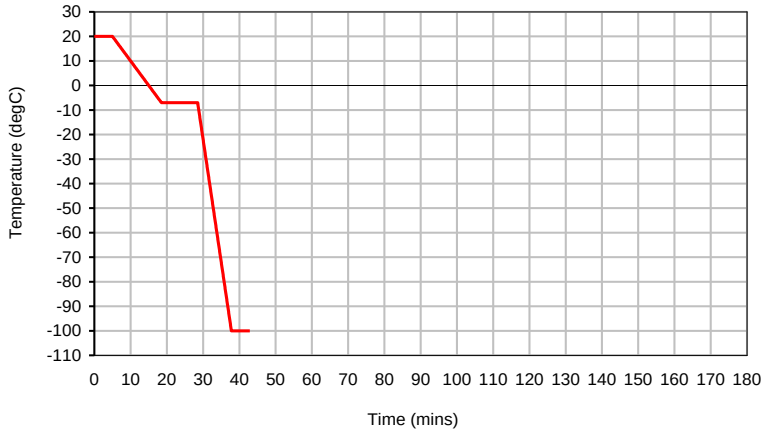


Figure 51: Pre-programmed sperm - linear freezing cycle

8.6. Oocyte – Non-Linear Cooling Cryopreservation

Profile name: OocyteNL	Applicable models: H00, H01, H03, H07, H08
Start temperature	20°C
Cool	from 20°C to -7°C at 2°C min^{-1}
Hold for nucleation	at -7°C for 10 minutes
Cool	from -7°C to -18.1°C at 0.5°C min^{-1}
Cool	from -18.1°C to -22.2°C at 0.4°C min^{-1}
Cool	from -22.2°C to -25.3°C at 0.3°C min^{-1}
Cool	from -25.3°C to -27.8°C at 0.2°C min^{-1}
Cool	from -27.8°C to -31°C at 0.1°C min^{-1}
Cool	from -31°C to -34°C at 3°C min^{-1}
Cool	from -34°C to -40°C at 6°C min^{-1}
Cool	from -40°C to -100°C at 10°C min^{-1}
Hold	at -100°C
Transfer to storage	

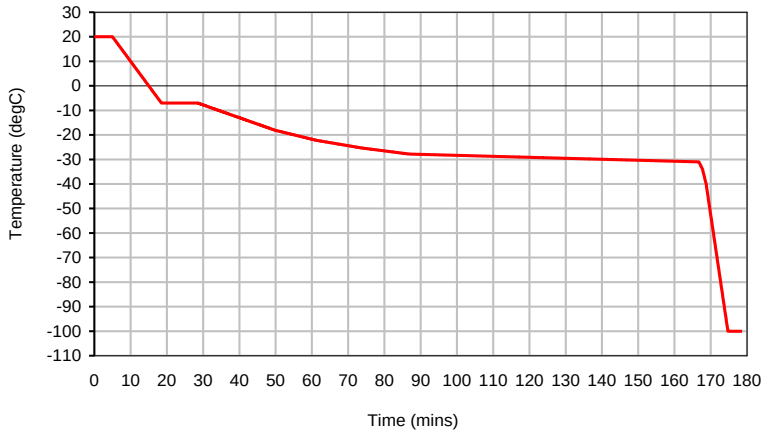


Figure 52: Pre-programmed oocyte – non-linear freezing cycle

8.7. Cryovial Cryopreservation

Profile name: Cryo1	Applicable models: H02, H05, H06
Start temperature	20°C
Cool	from 20°C to -5°C at 2°C min^{-1}
Cool	from -5°C to -7°C at 1°C min^{-1}
Hold for nucleation	at -7°C for 10 minutes
Cool	from -7°C to -80°C at 1°C min^{-1}
Hold	at -80°C
Transfer to storage	

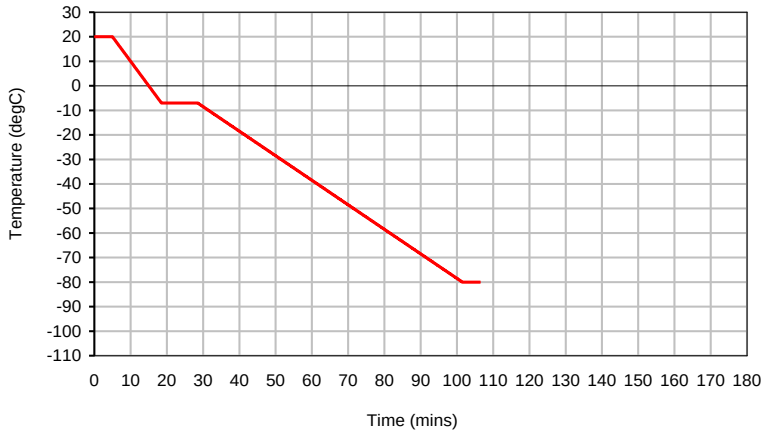


Figure 53: Pre-programmed cryovial freezing cycle

8.8. Test and Calibration profiles

Profile name: TCal, TP1, TP2	Applicable models: H00, H01, H03, H07, H08
Profile name: TP3, TP4	Applicable models: H02, H05, H06

These profiles are for testing and calibrating the freezer unit. Please do not edit or remove these profiles. For further information on how to test and calibrate your freezer unit contact Grant Instruments.

9. Scientific References

Kim SJ, Park JH, Lee JE, Kim JM, Lee, JB, Moon, SY, Roh SI, Kim CG and Yoon, HS (2004) Effects of type IV collagen and laminin on the cryopreservation of human embryonic stem cells. Stem Cells 22, 950-961.

Lasalle P, Testart J and Renard JP (1985) Human embryo features that influence the success of cryopreservation with the use of 1, 2 propanediol. Fertility and Sterility 44 645-651.

Morris GJ, Acton E, and Avery S (1999) A novel approach to sperm cryopreservation. Human Reproduction 14 1013-1021.

10. Maintenance and Service

No routine maintenance is required except for cleaning, there are no user serviceable parts inside the CRF-1 unit.

Warning: Attempted disassembly could cause damage that could seriously affect the operation of the unit and its ability to follow the desired temperature profile.

10.1. Cleaning

The freezer unit and sample plate is designed to be easily cleaned and sterilised. It is important to use a cleaner which does not leave a residue and will not attack the sample plate (chlorine based products must not be used).

We recommend the use of Klercide™ Sporicidal Low Residue Peroxide (formerly known as Premier Klercide™ -CR Sterile Filtered Biocide C) which meets these requirements. Before using any other cleaning or decontamination method, check with Grant Instruments to ensure that the proposed method will not damage the equipment.

Side and rear vents should be kept clean and free of dust to ensure good ventilation.

If a potentially hazardous liquid is spilt onto the equipment, disconnect it from the power supply and have it checked by a competent person. It is the user's responsibility to carry out appropriate decontamination if hazardous material is spilt on the equipment.

10.2. Replacement parts

The following is a list of purchasable spare parts:

04619	Cordset 230V UK straight 10A
04620	Cordset 230V Euro straight
28729	Transit Bolt Thumbscrew
29130	Transit bolt
29131	Plate transit Bolt Blanking
29159	Packing strap
33251	Transit case
33789	Standard black lid
40638	External PSU Assembly
LC71	Cable, Connecting, 9/9 Way "D"
USB2SERIAL	USB to Serial converter lead

10.3. Service & Calibration

If any unit servicing is required, switch off the unit and contact Grant Instruments.

Grant recommends the CRF-1 is calibrated annually.

Servicing and calibration should only be completed by Grant Instruments Service Department or via a Grant Instruments worldwide distributor.

A service contract for the CRF-1 series freezers is available. For full details of the service contract, calibration and verification please contact Grant Instruments.

Service Department
Grant Instruments (Cambridge) Ltd
Shepreth, Cambridgeshire
SG8 6GB
England.

Tel: +44 (0) 1763 260 811
E-mail: service@grantinstruments.com

11. Warranty

The warranty period for the CRF-1 series freezers is **three years**. Please visit www.grantinstruments.com/support to register your warranty.

Additional extended warranties are available from Grant Instruments and their distributors. These warranties are chargeable and will be added onto your standard free warranty period. A warranty certificate will be supplied. This should be retained and submitted should a repair be necessary during the extended warranty period.

For details of extended warranties please contact the sales department at Grant Instruments. salesdesk@grantinstruments.com

12. Product Compliance

12.1. Disposal & WEEE

In Europe, at the end of its life the CRF-1 must be disposed of in accordance with the WEEE directive. For information regarding WEEE collections in the UK please contact our B2B Compliance Scheme directly on +44 1691 676 124.

Grant Instruments complies fully with the EU Waste Electrical & Electronic Equipment (WEEE) regulations 2006. We are a member of the B2B compliance scheme (Scheme Approval Number WEE/MP3338PT/SCH), which handle our WEEE obligations on our behalf. Grant Instruments have been issued with a unique registration number by the Environmental Agency; this reference number is WEE/GA0048TZ.

For other countries please contact your equipment supplier.

For General WEEE information please visit: www.b2bcompliance.org.uk



Please note that because it contains a pressure vessel the CRF-1 series freezer is treated as “Special Waste”.

12.2. RoHS Directive

This product complies with the requirements of the UK Restriction of the Use of Certain Substances in Electrical and Electronic equipment Regulations 2012 and the EU RoHS Directive (2011/65/EC including 2015/863). This means the products are free of Lead and other hazardous substances covered by the directive.

12.3. Electrical safety and Electromagnetic compatibility

This product complies with the requirements of the UK Electrical Equipment (Safety) Regulations 2016 and the European Union Low Voltage Directive (2014/35/EC) for Electrical safety.
This product complies with the requirements of the UK Electromagnetic Compatibility Regulations 2016 and the European Union EMC directive (2014/30/EC).

12.4. REACH Regulation

This product does not contain any substances of very high concern (SVHCs) at greater than 0.1% that must be identified in accordance with Regulation (EC) No 1907/2006 and therefore does not have an entry in the SCIP database.

Appendix 1 Troubleshooting

A1.1 Loud Mechanical Clicking or Tapping Noise or Unit Fails to Follow Cooling Ramp

If there is a loud noise then switch the power off immediately as permanent damage can occur. The noise is caused by the pistons in the Free Piston Stirling Compressor hitting each other, usually because the system is overloaded.

Some causes and solutions are listed below:

Too large a mass on the cooling plate – reduce the total number of containers or reduce the volume in each container if using a standard cooling plate, do not use straws and vials at the same time on the H07 or H08 plate or reduce the mass of or volume of liquid in a non-standard vessel used with the H03 flat plate.

Too high a cooling rate has been selected – either reduce the mass on the cooling plate as indicated above or reduce the cooling rate.

The unit is in a high ambient temperature– ensure that the unit is operating in an ambient of less than 30°C, is not placed directly in line with a heating or air conditioning vent or radiant heater and is not in direct sunshine.

A1.2 No Power To Unit

The power switch is switched to the OFF position
Switch on. The power switch will illuminate red.

The power lead is not properly connected to the unit
Check the power lead is properly connected into the rear of the unit.

There is no power to the power lead
Check the power lead is plugged into a socket and that the socket is switched on.

The protective fuse in the CRF-1 unit has blown
Contact Grant Service Department.

A1.3 The Software Suite will not Communicate with the Freezer Unit.

Communications lead between the CRF-1 unit and the computer is not properly connected
Check the connections at both ends.

The Software is using the wrong COM port.
On the Software Config Panel, check the correct COM port is set.
If connected directly (no USB to serial converter) this is likely to be COM1.

When using the USB to serial converter this should be set to the COM port allocated to the USB port. To find this out, use the computers Device Manager, open Ports (COM & LPT1) and see which COM port number has been allocated to the USB Serial Port. The Software suite should then be set to use this port.

CRF-1 unit not switched on
Check power switch is illuminated and switched to the ON position.

A1.4 Straws Stuck To Sample Plate

Freezing plate not dried properly before samples placed in grooves
The plate may be damp and as the samples fit tightly into the plate they may have frozen onto the plate. Ensure the plate is dried before freezing (Section 7.3). Samples can be removed with careful use of tweezers.

Samples not dried before placing in grooves

The samples may be damp and as they fit tightly into the plate they may have frozen onto the plate. Ensure the samples are dry before freezing (Section 7.3). Samples can be removable with careful use of tweezers.

Appendix 2 Cryopen Nucleating Tool Operation

Please read the instruction manual supplied with the cryopen before use. The cryopen is a medical device that can be used to nucleate the samples. Connect the cartridge to the cryopen according to the cryopen instruction manual.

Appendix 3 Comms set-up when using the USB to Serial converter

The USB to Serial Port converter supplied with the unit has a device driver that must be installed on your computer before it can be used. Follow the instructions below.

Take the mini-CD supplied with the converter and insert it into the CD/DVD drive of your PC. Follow the on-screen instructions.

Insert the converter into a USB socket.

To find out which Serial port has been allocated to the USB converter, open Device Manager on your computer and expand the Ports (COM & LPT1).

In the example below, Figure 54, COM6 has been allocated.

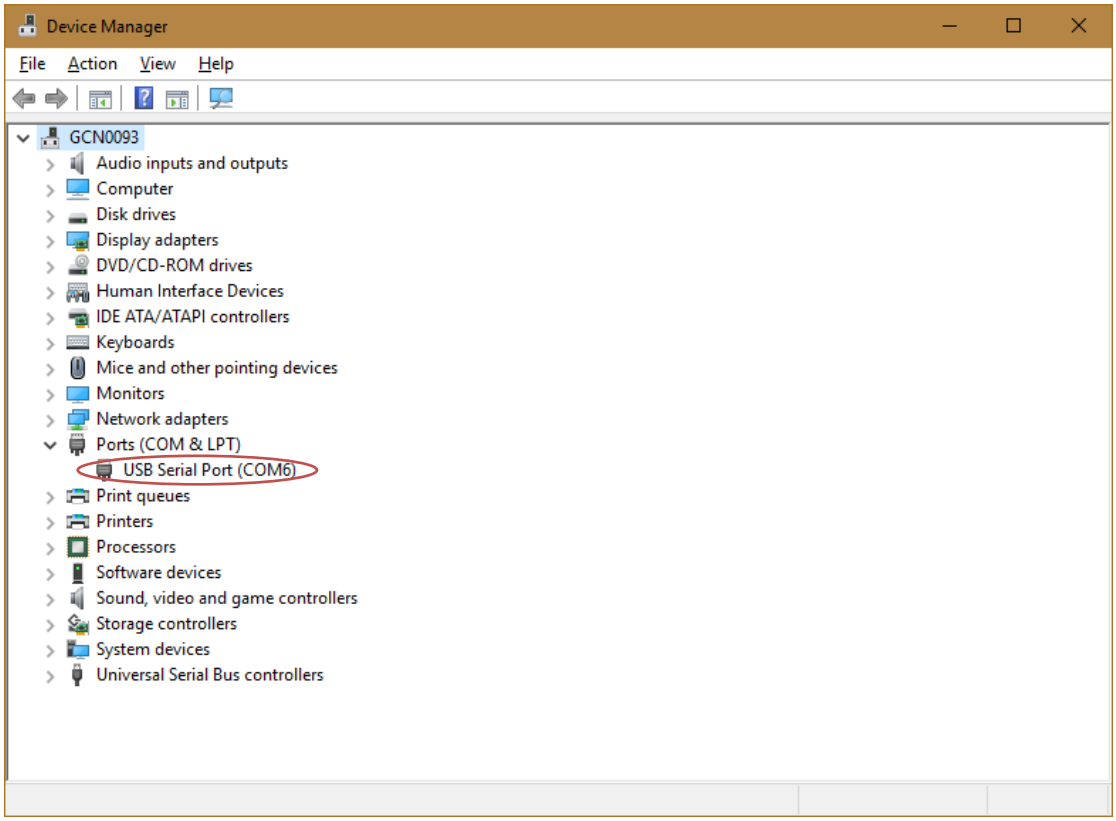


Figure 54: Allocated Com Port for USB-to-Serial Converter

This should be used when setting the COM port in the Software Config panel of the User Software Suite. See Figure 55.

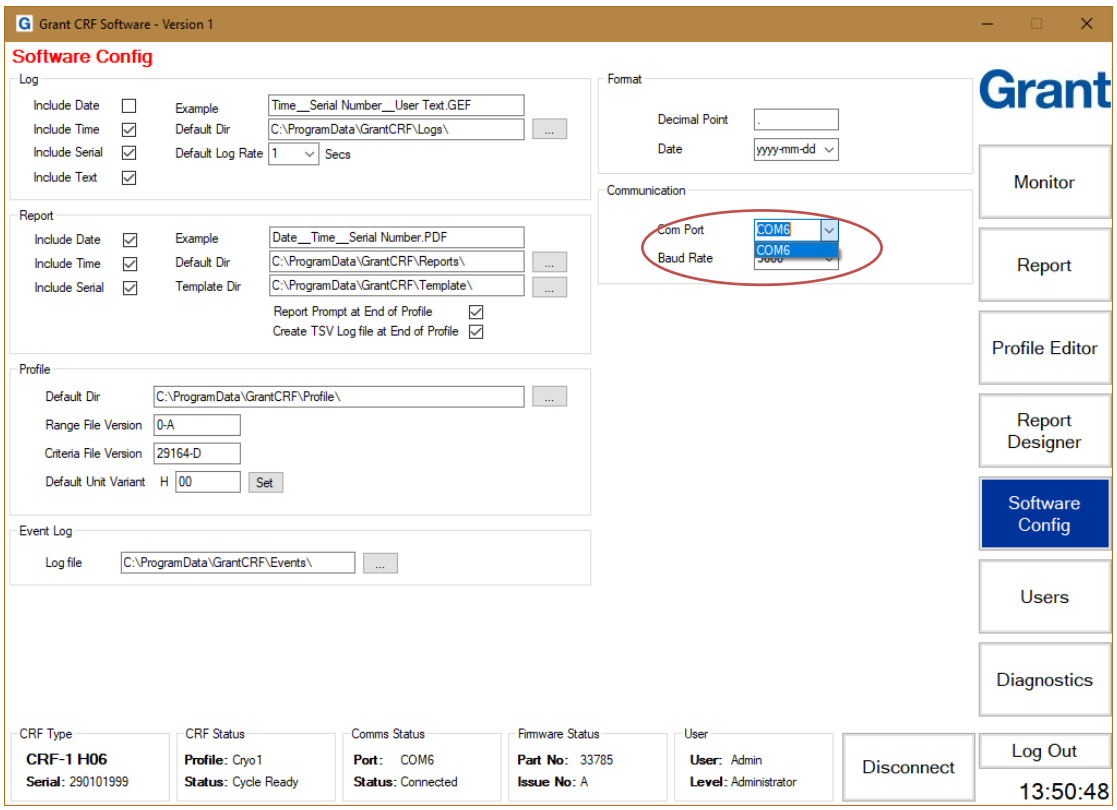
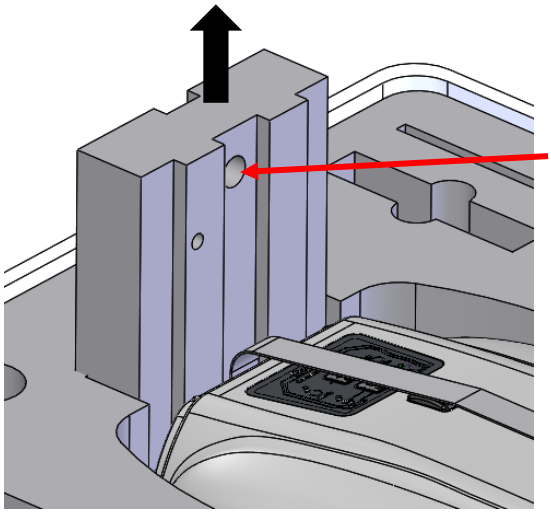


Figure 55: Configuring the Com Port on the User Software

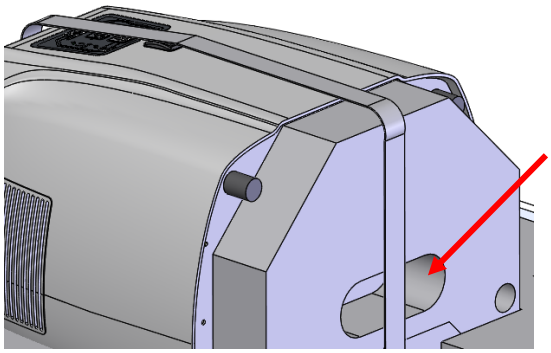
Appendix 4 Unpacking instructions

THE CRF-1 IS A DELICATE SCIENTIFIC INSTRUMENT AND SHOULD BE HANDLED WITH CARE AT ALL TIMES. THESE INSTRUCTIONS **MUST** BE FOLLOWED TO UNPACK THE UNIT.

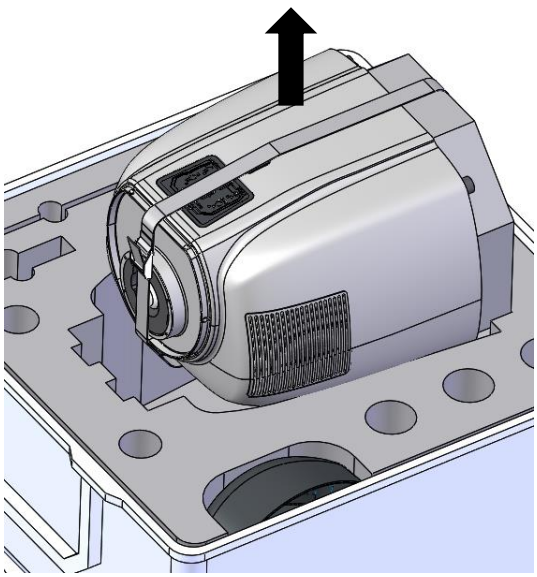
WARNING: ENSURE TRANSIT BOLT IS REMOVED BEFORE OPERATING THE UNIT, SEE TRANSIT BOLT INSTRUCTIONS BELOW.



1. Remove the Top foam by inserting a finger into the top hole and pulling it gently but firmly upwards.



2. Once the top foam is removed, the unit can be gripped by the bottom foam hand-hold and with one hand on the sample plate.



3. Grasp the unit securely and draw it from the case.

4. Carefully place the unit upright on a firm surface. Undo the strap and remove the unit from the bottom foam.

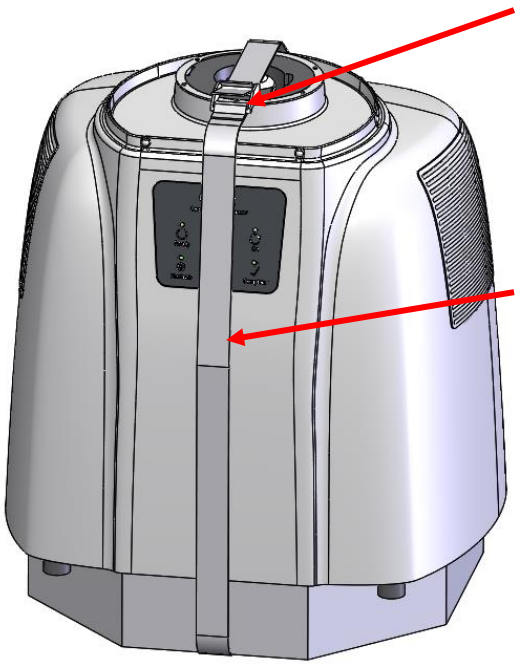
5. Remove the unit from the bag, and consult the separate instructions to remove the transit bolt.

6. RETAIN ALL PACKAGING AS IT **MUST** BE USED TO REPACK THE UNIT FOR FURTHER TRANSPORTATION. SEE OVER FOR PACKING INSTRUCTIONS.

Appendix 5 Packing instructions

THE CRF-1 IS A DELICATE SCIENTIFIC INSTRUMENT. FAILURE TO CORRECTLY PACK THE UNIT MAY RESULT IN DAMAGE TO THE UNIT AND RENDER THE WARRANTY VOID.

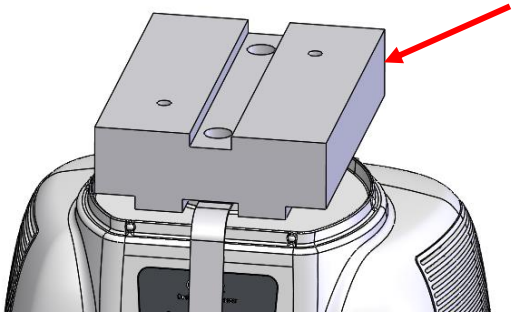
WARNING: ENSURE TRANSIT BOLT IS IN PLACE BEFORE PACKING THE UNIT, SEE SEPARATE TRANSIT BOLT INSTRUCTIOS BELOW.



1.Place CRF-1 Unit into a large polythene bag (Grant part no. 00402).

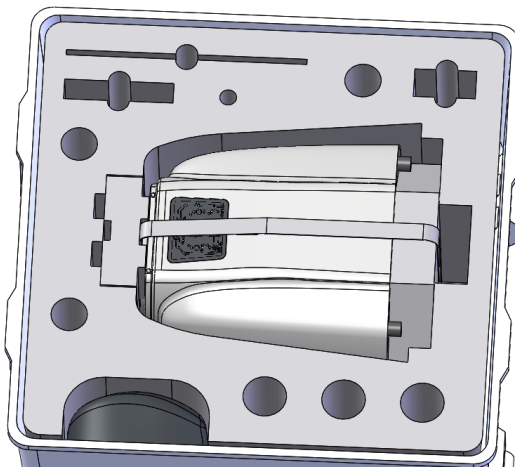
2. Run the packing strap (Grant part no. 29159) under the bottom foam piece and sit the unit on the foam with the cut corners of the foam facing forwards as shown.

3. Tighten the strap until it is taut. Place the buckle as shown. Avoid the buckle touching the unit.



4. Fit top foam piece as shown for ALL models other than CRF-1 H02 and H06. For CRF-1 H02 fit top foam the other way up. For CRF-1 H06 please use the alternative thinner flat packing foam supplied.

5.With one finger through the rear finger hole in the top foam to support the plate, gently roll the unit backwards. Grasp the hand-hold in the bottom foam and lower the CRF-1 into the packing case.



6. Fit the power supply, leads, lid and other accessories into their appropriate slots.

7. Close the lid and securely fasten all 6 integral catches.

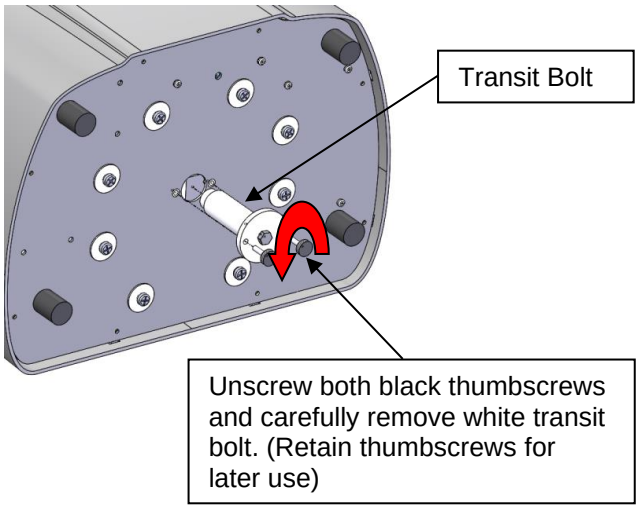
Appendix 6 Transit Bolt

THE CRF-1 CONTROLLED RATE FREEZER IS A DELICATE SCIENTIFIC INSTRUMENT AND SHOULD BE HANDLED WITH CARE AT ALL TIMES. THESE INSTRUCTIONS MUST BE FOLLOWED TO REMOVE THE TRANSIT BOLT ATTACHED TO THE UNIT BASE PLATE.

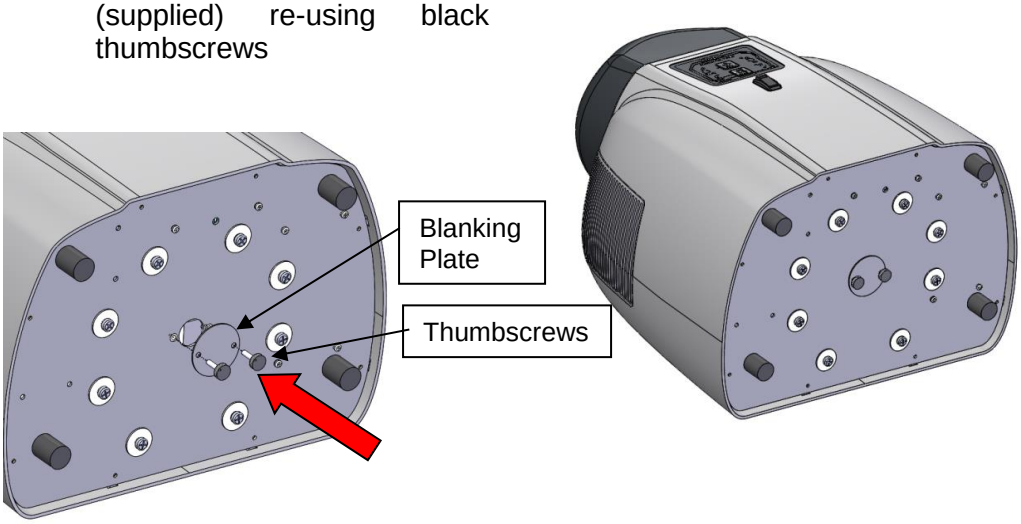
FAILURE TO REMOVE THE TRANSIT BOLT BEFORE OPERATION MAY RESULT IN UNIT DAMAGE AND MAY VOID THE PRODUCT WARRANTY

1. Place unit carefully on a secure surface with the switch and keypad facing upwards to remove the transit bolt





2. Attach blanking plate (supplied) re-using black thumbscrews



These instructions MUST be followed in reverse for transit bolt attachment prior to further transportation. The product warranty will be void if the unit has not been packed in accordance with these instructions.



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