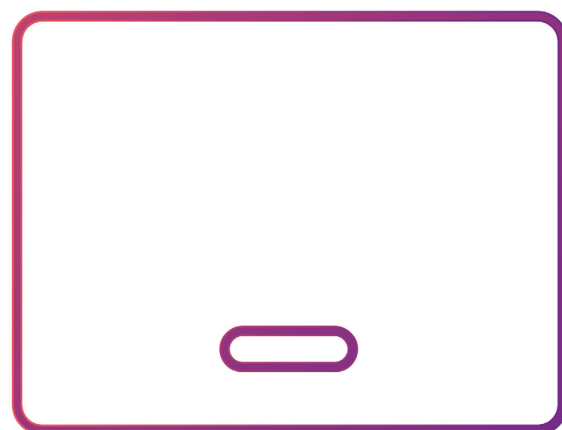


April 2023



Chronos™ Dx

Real-time PCR plateforme

USER MANUAL

BforCure Chronos™ Dx

Chronos Dx™ A1_A12

User Manual - English

D_A1_47_A2
April 2023



Any serious incident occurring in connection with the device shall be notified to the manufacturer and to the competent authority of the Member State in which the user and/or patient is established.

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Software user manual to be read carefully



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Utilisation In Vitro du Dispositif
For In Vitro Diagnostic use only



Lire le manuel utilisateur
Please read the user manual before use













Chronos™ Dx A1_A12

List of symbols

For safe use of Chronos Dx, users must comply with the provisions of this user manual. Chronos Dx is compliant with the regulations for Diagnostic Medical Devices *in vitro*,

but cannot be used by an untrained user. This user's manual warns the operator of possible dangers or points of attention with the following symbols (Table 1):

	Attention	Indicates that the user should check for precautionary information that may not be included in this manual.
	<i>In vitro</i> diagnostics	Indicates a medical device for <i>In vitro</i> Diagnosis.
	Electrical device	Indicates that the device is an electrical instrument. Risk of electric shock.
	Protection against moisture and liquid	Indicates that the device must be protected from all forms of liquid and moisture.
	CE marking	CE mark of conformity
	Do not throw in the garbage	Indicates that the device cannot be disposed of in the trash.
	Fragile	Indicates that the device should be handled with care.
	User information	Indicates that the user manual must be read carefully before use.
	Humidity limit	Indicates that the product can operate in an environment with a humidity level between 20 and 65% (non condensing).
	Maximum temperature limit	Indicates that the product can operate in an environment where the ambient temperature does not exceed the listed high temperature.






	Ref	Indicates the reference number of the device
	SN	Indicates the serial number of the device.
IP	IP	Indicates the IP address of the device.
	Date of manufacture	Indicates the date of manufacture of the device.
	Manufacturer	Indicates the address of the device manufacturer.
	QR-Code	This QR-Code links to Instructions for Use of the device.

Table 1 - List of symbols found on the device nameplate and in the user manual, as well as on the Chronos Chips packaging.



In order to use Chronos Dx in the best conditions, it is imperative that the operator uses only consumables whose expiration date has not passed. Please refer to the manufacturer's instructions.

Compliance with standards and hazards

Compliance with standards

Chronos Dx complies with the following standards:

Electrical Safety

EN 61010-1/A1: 2016 - Safety requirements for electrical equipment for measurement, control and laboratory use - Part 1: General requirements

EN 61010-2-101: 2017 - Safety requirements for electrical equipment for measurement, control and laboratory use - Part 2-101: Particular requirements for *in vitro* diagnostic (IVD) medical equipment.

Electromagnetic Compatibility

EN 61326-1: 2013 - Electrical equipment for measurement, control and laboratory use - EMC requirements - Part 1: General requirements

EN 61326-2-6: 2013 - Electrical equipment for measurement, control and laboratory use - EMC requirements - Part 2-6: Particular requirements - *In vitro* diagnostic medical equipment (IVD)



It is the responsibility of the operator to ensure that the device is used in accordance with the environmental and safety requirements described.

Hazards

Chronos Dx has been designed for safe use according to the instructions in the user's manual. BforCure shall not be liable for any damage caused by unauthorized use or modifications.

Biological hazards

Chronos Dx is used in the laboratory for the analysis of *in vitro* samples. The samples can be potentially dangerous for :

- The safety of the user,
- Contamination of the analysis resulting in poor detection,
- Environmental contamination.

No emission of hazardous substances occurs during normal use of the device.

Chemical hazards

Chronos Dx does not contain any hazardous chemicals.

Fire and exposure hazards

Chronos Dx presents no explosion, fire or burn hazard to the operator when used in accordance with BforCure's instructions.

Usage

Intended use

Chronos Dx is a laboratory system designed to perform rapid fluorescence PCR to detect and quantify nucleic acid sequences. The system and software are intended for use for *in vitro* diagnostics by qualified laboratory technicians.

The user manual describes the implementation and use of the Chronos Dx device used for the detection of genes of interest by real-time PCR (or RT-PCR). The user manual does not allow for the interpretation of the results obtained. The clinical interpretation of the results of each analysis must be performed by a laboratory specialist.



Sampling and sample preparation kits are not covered by this manual. Refer to the associated manufacturer's instructions for the sample collection and preparation kit.

To use Chronos Dx with Chronos Control Test or any other kit of the Bfast range, please refer to their instructions.

Packaging

For an optimal use, Chronos Dx is delivered with different accessories listed in the following table:

Pack reference	Pack	Reference Article	Article	Quantity
PK010007	Chronos Dx Pack+	Chronos Dx- A1_A12	Chronos Dx	1
			Laptop with Chronos Soft software installed	1
			Laptop power supply	1
			Chronos Dx power supply	1
			Ethernet cable	1
		PK9900003 Chronos Set-up Pack+	Thermalization liquid (1 liter)	1
			Plug connected to hoses	1
			Filter connected to hoses	1
			Replacement filters	3
Chronos Dx can also be delivered with accessories to perform weekly control tests				
PK010002	Chronos Dx/Dx+ Starter Pack	A5_C1	Chronos Chips - Control Standard - 1 chamber (Box of 25 Chronos -Control chips)	1
		B4_A2	Chronos Control Test (PCR reagents for 10 tests)	1

Table 2- List of accessories supplied with the Chronos Dx- A1_A12 device

Always use the Chronos Dx instrument with the accessories provided by BforCure.



Refer to the Chronos Control Test manual for their description, receipt, use and storage .



Refer to the Chronos Chips manual for their description, storage and use.



Thermalization liquid supplied with Chronos Dx.

Product description

Chronos Dx is a real-time quantification platform for genes of interest, featuring the Fastgene™ module. Fastgene is a microfluidic module whose technology allows Chronos Dx to achieve very fast temperature changes and therefore to have an ultra-fast PCR output.

The Chronos Dx instrument is controlled by the Chronos Soft software, installed in the supplied laptop.



For more information, refer to the Chronos Soft user manual.

The compact design of Chronos Dx makes it easy to integrate and is perfect for small lab benches.

Chronos Dx Presentation

Specification table



Technical specifications (at sample level)

Temperature range	50 - 100°C
Speed of temperature change (90% of set point)	< 3 seconds
Resolution of the temperature setting	≤ 0,1°C
Stability of the temperature	≤ 0,5°C
Temperature stabilization time (< 0.6°C of the set point)	< 90 seconds
Absolute temperature accuracy	< 0,8°C
Maximum temporal resolution of acquisition	0,4 seconds

Operating time

Getting started	< 3 minutes
Time between 2 PCR or RT-PCR tests	< 1 minute

Physical specifications

Dimensions	360 x 256 x 271 mm (W x H x D)
Weight	7 kg

Interface

Ethernet port	✓
USB port	✗
4G dongle	With / Without
Bar code and QR-Code reader	✓

Software functions

Remote update	✓ integrated into the PC (available later)
Integrated server storage capacity	> 1000 experiences
Sequence programming	By the menu or the configuration file

Fluorophore compatibility

Cy3™
Cy5™
FAM™
SYBR Green™
HEX™
Texas Red™
JOE™
VIC™
TET™
Quasar 670™

Detailed presentation

Chronos Dx allows the realization of PCR (or RT-PCR) in ultra fast real time, thanks to the use of specific consumables, which are the Chronos Chips (see Chronos Chips User Manual) and the Chronos Soft calculation software (preinstalled in the laptop provided) necessary for the control of the Chronos Dx real-time PCR instrument.

Chronos Dx is presented as follows:

QR-code and barcode reader

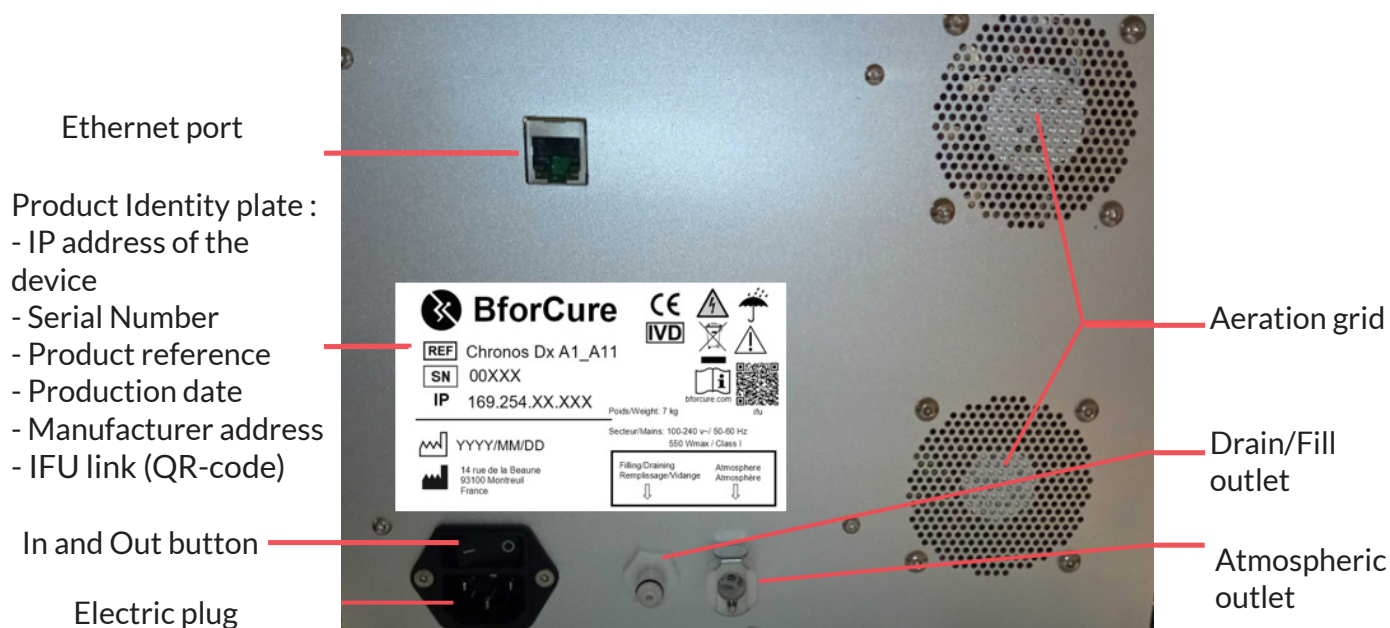
Chronos™Dx chip insertion slot and LED display



Front view of the Chronos Dx real-time PCR instrument.

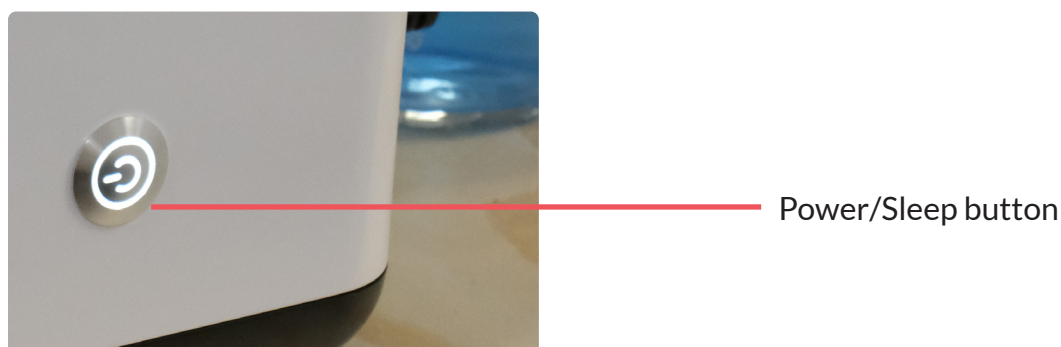


The front panel (black part) is not a touch screen.



Back view of the Chronos Dx real-time PCR instrument.

On the back of the Chronos Dx device are displayed its IP address, the manufacturer's address and the production date as well as important pictograms. The arrangement of these elements on the company plate can be different.



View of the button on the right side of the Chronos Dx real-time PCR instrument.

Precautions for use and safety instructions

General precautions

- Wear protective clothing, gloves and eye protection.
- Change gloves regularly. If indirect contact with a potentially contaminated biological sample, change gloves immediately and wash hands thoroughly.
- Protect any cuts or abrasions from contact with infected or hazardous materials.
- Remove any jewelry that could be contaminated (pendant, ring, watch...).
- Do not touch eyes, nose or mouth.
- Store materials, which may be affected, in dedicated containers.
- Clean, decontaminate locations and non-disposable instruments after use, following laboratory good practice.
- Avoid the crossing of contaminated materials with uncontaminated materials.
- Do not expose materials (e.g. notebook, paper...) that cannot be decontaminated due to potential contamination.
- Do not touch personal belongings or any other non-dedicated surface with gloves that are potentially contaminated.
- Provide a place to handle biological samples and use Chronos Dx with a computer (provided).
- Remove protective gear (gown, gloves, protective clothing) before leaving the laboratory.
- Wash hands thoroughly after contact with biological samples.







Specific IVD precautions

- For *in vitro* diagnostic (IVD) use only.
- For use by trained health professionals only.
- To avoid any risk of contamination on Chronos Dx, it is not recommended to install it in a room dedicated to the preparation of biological samples, extraction and purification of nucleic acids etc.
- Any biological sample should be considered potentially infectious and should be treated with appropriate safety precautions.
- No biohazardous substances are emitted if the precautions for use are observed.

Product specific precautions



Read the information in “Compliance with Standards” and “Hazards” before using Chronos Dx.

Symbols	Environmental conditions	Criteria - Ranges
NA	Power supply	100-240 Vac / 50Hz, 550W max
NA	Overvoltage category	Cat III
	Protection class	IP = 30 IK = 08
NA	Altitude	Up to 3000 meters
	Operating temperature	15-40°C for PCR temperatures of at least 60°C 15-33°C for PCR temperatures of at least 50°C
	Fragile	Chronos Dx should be handled with care when transported out of its packaging.
	User information	Indicates that the user manual should be read carefully before use.
	Humidity limit	Indicates that the product can operate in an environment with a humidity level between 20 and 65% (non condensing).
	Storage temperature	+10°C to +50°C (filled material) +10°C to +60°C (empty material)
NA	Degree of Pollution	Level 2
NA	Distance to surrounding objects	10 cm distance to surrounding objects



Do not obstruct the air vents of Chronos Dx. Keep away from any liquid not required for the operation of the instrument.

Chronos Dx installation and startup

Install Chronos Dx

After unpacking, Chronos Dx must be installed on a flat, clean and rigid surface with free air circulation to ensure proper operation of the device. A place must also be provided for the supplied laptop, which must be installed near Chronos Dx.

Make sure to:

- Leave at least 10 cm of free space behind the Chronos Dx device for free air circulation and good cooling of the system,
- Do not place the Chronos Dx device under a sunny window (heat source) as this may damage the electrical components,
- Leave unobstructed access to the power switch and power cord of the unit.



In order to avoid any risk of contamination of Chronos Dx, it is strongly recommended not to install it in a room dedicated to the preparation of biological samples, extraction and purification of nucleic acids.

The first step consists in taking out of the box all the elements that make up the Chronos Dx Pack+ (see paragraph “Packaging the product”).

Chronos Dx installation procedure :

1. Connect the Chronos Dx instrument power cable to the back of Chronos Dx and to a power outlet (preferably an inverter)
2. Connect the Ethernet cable between Chronos Dx and the computer provided.
3. Connect the computer's power cable to a power outlet (if necessary).
4. Remove the cap and put the cap with the two hoses on the thermalization bottle (Fig. 1).
5. Take out the filter connected to its two hoses (Fig. 2) and connect the elements according to the pictures on the next page:

Port A
To screw
with port B



Connector X
For the Atmo-
spheric outlet

Fig.1 The bottle screwed with the cap and its two hoses



Port B

Connector Z
for the Drain/Fill
outlet

Fig.2 The filter connected to its hoses



Atmospheric outlet port

Drain/Fill outlet

Fig.3 The two outlets at the back of Chronos Dx

- **Screw port A with port B (Fig. 1 and 2),**
- **Press the Z connector into the drain/fill outlet on the back of Chronos Dx until it clicks (Fig. 2 and 3),**
- **Press the X connector to the atmospheric outlet port on the back of Chronos Dx until it clicks (Fig. 1 and 3).**

This picture shows a final connection:



Once the installation procedure is completed, Chronos Dx is ready for start-up. Please refer to the following graph "Starting up Chronos Dx".



Be careful not to introduce dust or particles into the thermalization liquid during step 4. Do not leave the plug uncapped.

Be careful not to spill the bottle once it is connected, as the cap is not waterproof.

The thermalization bottle must be changed after 6 months (refer to the date of manufacture on the bottle label).

Chronos Dx startup



Refer to the Chronos Soft user manual by clicking on the BforCure web link, accessible via “Help”, then “About”.

Once Chronos Dx is installed, it is ready to be powered up:

1. Turn on the power using the switch on the back of the instrument.
2. Press the right side button of Chronos Dx once.
3. The instrument initializes.
4. Wait until the LED in the insertion slot turns solid white.
5. Turn on the computer. Designate the password provided in the delivery if requested. Log in to the computer with the password provided in the delivery.
6. Open the Chronos Soft software by clicking on the icon representing the BforCure logo in the left sidebar of the screen (installed in the computer provided).
7. A splash screen appears briefly and then the user interface appears on the home screen.
8. Log in as a user by clicking on the “Connect” button (located at the top right of the Chronos software interface).
9. Enter the factory username and password supplied on delivery. If necessary, select the Chronos Dx instrument to be connected (by entering the IP address written on the back of the machine), then click on the “Connect” button.
10. The Chronos Dx real-time PCR instrument is ready to use.

Right side



LED of the insertion slot.





When initializing the Chronos Dx real-time PCR instrument, various noises are heard.

The IP address of the Chronos Dx instrument, which is later required for connection to the Chronos software (see Chronos Soft User Manual), is preconfigured at the factory and is written on the rear panel. In case this address needs to be changed by the operator, contact BforCure technical support.

Once this procedure has been carried out, Chronos Dx is ready for use. At the very first ignition, or at subsequent ignitions, the instrument may need to perform a filling procedure by the user, via the Maintenance tab.

Chronos Dx is equipped with a microfluidic system, based on Fastgene's patented and proprietary technology, at the heart of the device. This technology works with demineralized and filtered thermal fluid.



To switch off Chronos Dx, proceed when the device is in the “IDLE” state.

Chronos Dx can be turned off in two ways:

- by clicking on the “Turn off” button in the Chronos Soft software.
- or by briefly pressing the right side button of Chronos Dx.

Then wait at least 3 minutes until the two front side LEDs are off, and press the power switch on the back of Chronos Dx.



If Chronos Dx is not in “IDLE” status, click on “Cancel run” in the “Experiment” tab. This will then allow the device to be turned off.

Start a test on Chronos Dx



BforCure commercializes reagents compatible with Chronos Dx.

The reagents must be inserted in the Chronos Chips corresponding to the chosen test.

The choice of the test is made on the configuration tab of the Chronos Soft software.

To perform an analysis on Chronos Dx, refer to the user manual of the Chronos Soft software, the Chronos Chips IVD and the reagents used.

Chronos Control Test

Chronos Dx is delivered with PCR reagents to ensure the proper functioning of the device: Chronos Control Test.

On the Chronos Soft software, the configuration to be selected in the “Configuration” tab is “Service - Test control”, and the chips are the Chronos - Control chips (Chronos Chips - Control Standard).

This control test should be performed once a week when using Chronos Dx to validate *in vitro* diagnostic (IVD) tests.



For more information, please refer to the Chronos Control Test manual, the Chronos Chips and the Chronos Soft user manual.

Maintenance of Chronos Dx

In order to ensure the reliability of the results obtained during the experiments with Chronos Dx (free of any external contamination) it is necessary to carry out a periodic maintenance:

1. Switch off the device via the switch and unplug the power cable from the mains socket.
2. Wait until the system is completely cool.
3. Clean only the external surfaces of the device with a slightly damp cloth. Do not allow water to get into the air vents to prevent damage to the device's functional parts.
4. Dry the cleaned parts with a soft, dry cloth.



Never use gasoline, solvents or abrasive cleaners, metal objects or hard brushes to clean Chronos Dx as this may damage it.



Never soak Chronos Dx or the power cord in water or any other liquid. Before performing any maintenance, switch off and disconnect Chronos Dx to avoid any risk of electric shock.



Avoid spraying liquids through the air vents on Chronos Dx as this may damage the device.

Chronos Dx maintenance: filling and emptying procedures

The level of thermalization liquid in the Chronos Dx microfluidic core may vary during use of the instrument and require refilling. In the same way, it is necessary to change the liquid level before transporting or storing the Chronos Dx instrument on a long term basis.

These maintenance operations are accessible via the **“Maintenance”** tab in the Chronos Soft software and cannot be done without the Chronos Soft software.

These operations are carried out with the “Chronos Dx+ Set-Up Pack” supplied with Chronos Dx, the elements of which are presented in the table below:

Element	Lifetime	Quantity
Thermalization Liquid (1 liter)	6 months	1
Plug with its two hoses		1
Filter connected to its two hoses	to change if the filter is clogged (see more details in the filling section)	1
Replacement filters		3

Table 3- Chronos Dx+ Set-Up Pack required for filling and emptying the Chronos™ Dx microfluidic system.



Refer to the Chronos Soft user manual for more details.

Manual filling of Chronos Dx

The user has the possibility to perform a fill by going to the maintenance tab of the Chronos software and clicking on “Fill the instrument”, provided that the instrument is in “Idle”.



Follow the order of the steps described below.
Check that the thermalization liquid bottle is filled to no more than 1/4 of its capacity and is properly connected.

The manual filling procedure is as follows:

1. Open the Chronos software and connect the instrument to the software.
2. Check that the instrument is in “IDLE”. If the instrument is in use, press “CANCEL RUN” in the EXPERIMENT tab.
3. Click on the “MAINTENANCE” tab.
4. Click on the “Fill the instrument” button on the Chronos Soft interface.
5. A window appears indicating the filling of the device and Chronos Dx beeps. Click on OK.
6. The status changes to “Filling” and the LED at the insertion slot turns purple and flashes. Chronos Dx fills up and the liquid level in the thermalization bottle decreases.
7. The device goes into the initialization state and excess water can be expelled into the thermalization bottle at this time.
8. The instrument is ready for use when it returns to “IDLE” and when the buttons on the “SERVICE” button turns blue again.
9. Once the filling procedure is completed, Chronos Dx can be used.

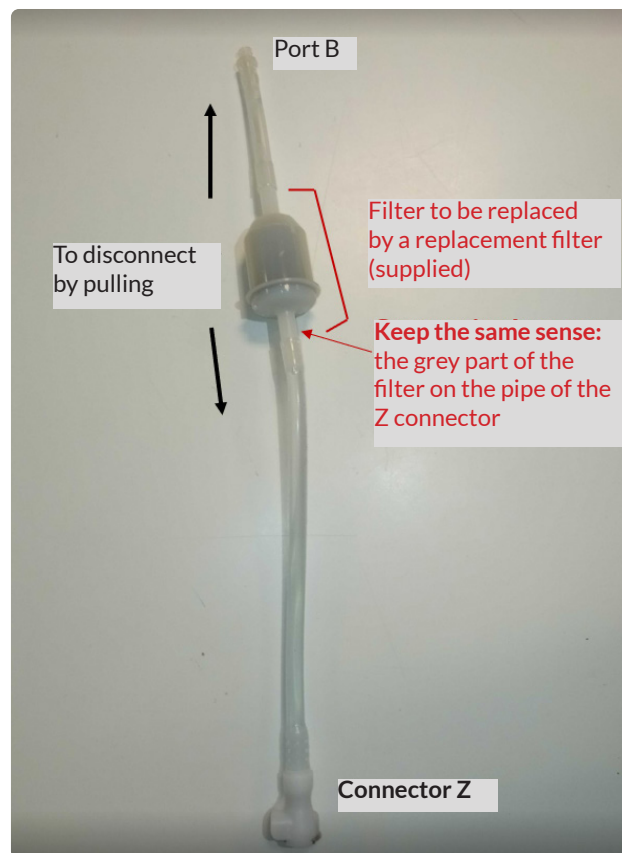
Filter to be changed :

It is possible that the fillings are not done correctly (repeated demanding of Chronos Dx to fill the instrument).



If the thermalization bottle is properly connected and filled (more than a quarter of its capacity), the filter may be clogged.

The filter must be changed with a replacement filter supplied with Chronos Dx. Proceed when Chronos Dx is switched off. Remove the two hoses at the ends of the filter and replace it, keeping the same direction, then click on OK .



Chronos Dx is a microfluidic system and the use of unsuitable filling fluids may cause irreversible damage to the device. BforCure will not be held responsible for any malfunction in the event of improper use of a filling fluid.



When filling, do not splash any liquid on the back of Chronos Dx, especially on the air vents and the switch.

Emptying Chronos Dx

In the case of long-term storage or before transporting the Chronos Dx device, an emptying procedure must be performed. To do this, the fluidic circuit must be drained using the maintenance kit (Table 3). The use of Chronos Soft software is imperative. Follow the sequence of steps described below.



- Follow the order of the steps described below.
- The thermalization bottle must be connected as indicated in the “Installing Chronos Dx” graph.
- Make sure that the thermalization liquid bottle is not filled to more than 3/4 of its capacity, otherwise there is a risk of overflowing when emptying.

The procedure for emptying is as follows:

1. Open the Chronos software and connect the instrument to the software.
2. Check that the instrument is in “IDLE”. If the instrument is in use, press “CANCEL RUN” in the EXPERIMENT tab.
3. Click on the “MAINTENANCE” tab.
4. Click on the “Empty the instrument” button on the Chronos Soft interface.
5. A window appears indicating the emptying of the device and Chronos Dx emits beeps. Validate the choice by clicking on “OK”.
6. The status changes to “Emptying” and the LED at the insertion slot turns purple and flashes. When emptying is complete, the device returns to “IDLE” status and the re- buttons turn blue.
7. Once the emptying procedure is completed, Chronos Dx can be stored for a long time or transported.

Bubbles may form in the thermalization bottle at the end of the emptying process.

After emptying Chronos Dx, it is advisable to change the filter, as explained in the graph on the previous page.



The drained liquid does not contain anything harmful to the environment and can therefore be disposed of in the sewage system.



At this point it is no longer possible to perform experiments with Chronos Dx. To do this, it must be refilled with thermalization liquid. To do this, follow the filling procedure described in the section “Filling of Chronos Dx”.

Transport of Chronos Dx



Before moving Chronos Dx, carry out a maintenance and emptying procedure (see Care of Chronos Dx and Maintenance of Chronos Dx). Chronos Dx can be stored (long-term in a dry place) or transported, once it has been drained.



Use dedicated transport cases (not supplied) to protect the Chronos Dx system and the associated notebook from possible damaging shocks. Chronos Dx should be handled with care when moved from its packaging.

Description of the different states of Chronos Dx

During the use of the Chronos Dx real-time PCR instrument with Chronos Soft software, different light and sound states can be observed and heard, mainly at the front of the device. These different states are informative for the operator.

State	Description	Color and light condition	Noise
Starting	The device has just been put into service and starts its self-test cycle.	White - Flashing	No
Idle (Rest)	The unit is switched on and ready, but no cycle is running.	White - Fixed	No
Filling	Thermalization liquid is introduced into the system and the instrument is switched on in this sequence to fill it.	Purple - Flashing	Yes - Once
Emptying	The reservoir and the microfluidic circuit of the device is being drained.	Purple - Flashing	Yes - Once
Pre-Filling	The device is being initialized. It performs a pre-filling step of the microfluidic circuit to remove any air bubbles.	Green - Flashing	No
Adjusting Temperature	The unit is adjusting the temperature to reach the rated operating temperature.	Green - Flashing	No
Insert Chip (ready temperature)	The instrument has reached the set temperature for the programmed experiment. The chip containing the reaction mixture can be inserted into the instrument.	Green - Fixed	Yes - Once
Waiting for Config	The instrument is at the set temperature for the previous experiment and is waiting for a new configuration.	Yellow - Fixed	Yes - Once

Loading Chip	The chip containing the reaction mixture is inserted into the device. The motorized optical module is put in place.	Cyan - Flashing	No
Running Sequence	The experiment sequence is currently being carried out.	Cyan - Fixed	No
Unloading Chip	The experiment sequence is finished. The chip can be removed from the device.	Cyan - Flashing	Yes - Once
Cooling	The unit is now cooling down before returning to the Idle state.	White - Fixed	No
ERROR	An error has occurred. A window with details of the errors that have occurred since the device was last switched on appears on the screen. If necessary, the window can be recalled by clicking on the "Error" status panel.	Red - Flashing	Yes - Several times

Table - Light states at the Chronos Dx insertion slot

Troubleshooting

What to do if:

Chronos Dx does not turn on	Check that the power outlet is properly plugged in and that there is a power supply.
Starting an impossible experiment	Make sure that the reaction chip is inserted into the device until it clicks. Check the system status. Check that the temperature set points are reached.
Data stored in Chronos Dx lost	The Chronos Dx system can store up to 1000 experiments internally. But this number can vary depending on the size of each experiment. Therefore, it is important to make regular backups of the data on an external device (such as an external hard disk). BforCure cannot be held responsible for any loss of data. Refer to the Chronos Soft user manual for more details on data backup.
Not enough space in the Chronos Dx memory	Contact BforCure's customer service.

If the instructions in the table do not solve the problem, or if none of these cases correspond to the problem encountered,

contact the After-sales service by sending an e-mail with details of the problem encountered and photos, if possible.



For safety reasons, never attempt to work inside the Chronos Dx system. Doing so may damage the device and may cause personal injury.

BforCure accepts no liability for any damage (in contract or tort, including negligence) that may arise from failure to comply with these warnings and instructions.

Disposal and recycling

Disposal of biohazardous materials

Dispose of the following wastes separately according to the laboratory recommendations and the regulations in force:

- Biological samples,
- PCR reagents,
- Reaction chips,
- Packaging,
- Other consumables that may be contaminated.

Chronos Dx contains electrical and electronic components that under the European Directive 2012/19/EU on Electrical and Electronic Waste. Under this directive, the device must follow a recycling path.

Consult the regulations in force at your place of use.

