

BIOMETRA TRIO

Instruction Manual

Ver. 15/07



Please read these instructions carefully before using
this apparatus!

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This document describes the state at the time of publishing. It needs not necessarily agree with future versions.

Subject to change!

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2 Introducing the Biometra TRIO Thermal Cycler

2.1 Field of Applications

Developed in 1983 by Kary Mullis PCR (Polymerase Chain Reaction) is now a common and often indispensable technique used in medical and biological research labs for a variety of applications. The Biometra TRIO is an end-point Thermal Cycler and intended to amplify nucleic acids by repeated cycles of heating and cooling using DNA-Polymerases in a PCR reaction. The development of the instrument had the goal to combine latest technology with stylish design and user-friendly software. The user interface consists of a touchscreen with a graphical or tabular display that shows the time, status, and temperature profile for each run. A touchscreen keypad allows you to enter information into fields on the screen. The Biometra TRIO can run three independent PCR reactions simultaneously and is especially helpful when versatile samples are to be incubated using different PCR programs.

2.2 General Description

There are three models of the Biometra TRIO Thermal Cycler:

- Biometra TRIO 48 for 0.2 ml tubes, plates or strips
- Biometra TRIO 30 for 0.5 ml tubes
- Biometra TRIO combi for 0.2 ml tubes, plates or strips or 0.5 ml tubes

The instrument is controlled by a 7" touchscreen display and easy to use interface. PCR protocols and Run-Logfiles can be saved to a connected USB drive. By using the USB functions PCR protocols can be exchanged easily between instruments and Run-Logfiles can be stored for documentation reasons on PC.



Figure 1 The Biometra TRIO Thermal Cycler

The Biometra TRIO Thermal Cycler features an automatic restart. If a power failure occurs during the run the instrument will continue the run when the power returns. In case of long-term power failure (longer than 30 minutes) the instrument keeps the sample block at 4°C

(freeze step) and the user can decide to repeat the run with the same samples or to discard them.

This instruction manual provides information on how to use all models of the Biometra TRIO Thermal Cycler most effectively.

2.3 Special features

2.3.1 Thermal Block

The Biometra TRIO Thermal Cycler is available with different block formats. The 48 well version can be used with standard 0.2 ml tubes, strips or plates. With 30 well blocks the Biometra TRIO can be used with 0.5 ml tubes and the combi block version offers the flexibility to incubate 0.2 ml tubes, strips or plates or 0.5 ml tubes. With a sample capacity of up to 144 samples in parallel the Biometra TRIO 48 can also be used for high throughput applications.

By the rubber sealing at the lid an encapsulated space is formed when the lid is closed. The closed room serves to improve the temperature uniformity of the sample block and avoids formation of condensed water at the final PCR cooling step. Furthermore all blocks are perfectly sealed to prevent condensed water from penetrating the Peltier elements underneath the sample block and other parts of the electronics. By the perfect sealing the Peltier elements are protected and the instruments life time is expanded.

2.3.2 High Performance Smart Lid

The Biometra TRIO heated lid has been optimized to heat twice as fast as with older models and thus helps to shorten protocol run times. Moreover the heated lid serves for two additional essential functions: It prevents the formation of condensed water at the reaction tube portion located above the block surface level and it ensures reliable contact between samples and thermal block by applying constant pressure. By the integrated clutch mechanism the pressure applied to the reaction tubes is always same regardless of the plastic ware height. The combination of the heated lid shape and the reliable contact pressure ensure an even temperature distribution between samples thus significantly improves the temperature uniformity.

With one press on the front button, the lid gently swings open and arrests in its end position. By this mechanism the lid cannot fall down and the heated lid is out of the radius of action when the user is inserting tubes or plates. Thus the risk of injury by violent pressure or the risk of burn by hot surfaces is prevented.

2.3.3 Display

The Biometra TRIO Thermal Cycler features a 7" state of the art color touchscreen user interface. The display is built-in at a flat angle to ensure reflection-free viewing and ergonomic programming. For programming PCR protocols the software incorporates Biometras proven spreadsheet philosophy and alternatively offers graphical programming. One touch leads from the spreadsheet to the alternative graphical programming mode. This makes the creation of new or editing existing programs fast and easy.

2.3.4 User specific quick start function

Many other thermal cyclers offer a so called "latest program list". In the latest program list a certain number of PCR programs is collected and offered for quick start. The list is typically ordered only by date and the user has to search for the desired program. In contrast the Biometra TRIO software for each sample block creates a user specific list and only the latest programs started by the currently logged-in user are offered for quick start. Even a lab

member does not use the instrument for a longer time the information on the latest programs started by this user will not get lost.

For maximum convenience the Biometra TRIO software features additionally features the program preview. Before start of a program the protocol steps are summarized in a clearly arranged table by the program preview tool. The program preview therefore provides a comprehensive overview on the protocol structure without the need to access programming screens. The program preview is also available before quick start of programs.

2.3.5 Easy programming

Creating new PCR programs takes a lot of time if for each single step all parameters have to be set manually. The Biometra TRIO offers two options for easy protocol creation:

1. Pre-installed program templates
2. Protocol wizard

The Biometra TRIO software offers several pre-installed program templates for different applications. The program templates provide a general protocol structure and can be easily adapted for the current experiment.

The protocol wizard offers the comfort to create specific PCR programs by just a few inputs. The protocol wizard is based on program templates for specific polymerases. For up to eight different polymerases program templates can be saved in the Biometra TRIO software and just the annealing temperature, cycle number and product length has to be filled in to create a specific PCR program. Optionally two-step or three-step PCR programs can be created by the software. Furthermore the protocol wizard includes a primer annealing temperature calculator and the calculated T_a value can be used for protocol creation.

2.3.6 Temperature Optimisation Step (TOS)

Finding the best primer annealing temperature is crucial for the specificity and efficiency of PCR reactions. Often optimization of experiments is conducted only limited leading to unspecific byproducts or reduced PCR sensitivity. By using the Biometra TRIO Temperature Optimisation Step (TOS) function new primer pairs with unknown annealing temperatures can be tested quickly and optimized in very short time.

The Temperature Optimisation Step (TOS) function makes use of the three sample blocks and provides three different temperatures for a selected program step. Just set the temperature for sample block 2 and define the temperature difference (temperature increment) for block 1 and 3. Since the Biometra TRIO samples blocks work independent of each other the maximum programmable temperature difference between the sample blocks is at $\pm 47.5^\circ\text{C}$.

2.3.7 GLP compliance

The software can manage up to 30 user accounts. By default three different user-levels are available: Administrator, user with generic rights and user with limited rights. However, in a comfortable menu the administrator can configure the settings for each user individually by activating or inactivating single rights. By the user administration tool in combination with the password protection of user accounts access to the instrument can be limited to authorized persons and unwanted modifications to the system settings and PCR protocols prevented.

With each system start the Biometra TRIO performs an initial self-test and additionally the software offers an extended self-test that can be initiated by the user. The results of the extended self-test becomes are summarized in a protocol and are stored by the instrument. Additionally to the self-test functions the instruments creates and stores run-logfiles for each single run. For long-term storage run-logfiles and extended self-test protocols can be exported as proprietary file format.

2.3.8 Open system philosophy

The Biometra TRIO is an open system regarding consumables and reagents. Depending upon the sample block the system can be used with single tubes, strips or plates. PCR plates can be standard profile or low profile and they can be skirted, semi-skirted or non-skirted. The TAdvance can handle all kind of plates and the plastic ware be sealed with domed caps, flat caps, sealing foil or other technologies. Regardless of the used sealing method thanks to the HPSL technology (see chapter 2.3.2) always the same pressure is applied to the consumables serving for absolutely reproducible conditions.

The Biometra TRIO Thermal Cycler is not limited to reagents from a specific vendor and can be used with any kind of polymerase. Regardless if isothermal-, hot start- or non-hot start polymerases are used, the Biometra TRIO will produce reliable results. E.g. by programming a pause step for the initial denaturation the system can be used for manual hot start applications. After inserting samples into the hot block the PCR protocol can be started just by pressing “continue”.

2.4 Order numbers

Model	Block type	Order No.
Biometra TRIO 30	Aluminum	846-x-070-720
Biometra TRIO 48	Aluminum	846-x-070-723
Biometra TRIO combi	Aluminum	846-x-070-724

Abbreviations:

x = 2 for 230V EN/DE, 4 for 115V EN, 5 for 100V EN

2.5 Technical specifications

Order Number	846-x-070-720	846-x-070-723	846-x-070-724
Name	Biometra TRIO 30	Biometra TRIO 48	Biometra TRIO combi
Capacity	3 blocks for 30 x 0.5ml tubes each	3 blocks for 48 x 0.2ml tubes or 48 well microplates or 6 x strips of 8 each	3 combi blocks for 18 x 0.5ml tubes** or 48 x 0.2ml tubes, or 48 well microplates or 6 x strips of 8
Block	Aluminum	Aluminum	Aluminum
Block coating	Special Alloy	Special Alloy	Special Alloy
Max. Heating Rate*	4.0 °C/sec	5.0 °C/sec	3.0 °C/sec
Max. Cooling Rate*	3.6 °C/sec	4.2 °C/sec	2.7 °C/sec
Avg. Heating Rate*	3.6 °C/sec	4.5 °C/sec	2.7 °C/sec
Avg. Cooling Rate*	3.2 °C/sec	3.8 °C/sec	2.4 °C/sec
Multiblock tool	Temperature Optimisation Step (TOS)		
Temperature Uniformity	95°C	+/- 0.60 °C after 15 s	
	70°C	+/- 0.30 °C after 15 s	
	55°C	+/- 0.20 °C after 15 s	
Temperature Range	3 °C to 99 °C		
Control Accuracy	+/- 0.1 °C		
Software	User specific quick start of the last five programs, program preview prior to start, toggle between spreadsheet and graphical programming mode, Temperature Optimisation Step (TOS), Service Info Files (SINF), Extended Self-Test (EST), adjustable ramp rates, PC control via Ethernet, comprehensive user administration tool, protocol wizard, annealing temperature calculator		
Program memory	Total capacity of 350 programs in up to 30 user directories		
Display	7" Color Touchscreen		
Auto restart after power failure	Yes		
High Performance Smart Lid (HPSL) Technology	Yes		
Lid Temperature Range	30 °C to 110 °C		
Power consumption	1000 Watt		
Power Supply	100, 115, 230 Volt, 50-60 Hz		
Noise emission	Very low		
Interfaces ¹	USB A, Ethernet		
Working conditions	15 °C to 35 °C, 70 % air humidity, max 2000 m NN		
Dimensions (WxDxH)	30 cm x 41 cm x 25 cm		

Weight	17.3 kg
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* Measured inside the block

** Capacity increases to 35 x 0.5 ml tubes by use of small cap tubes

¹ The network cable must be at least performance class Cat 5e and the cable configuration has to be STP
x = 2 for 230V EN/DE, 4 for 115V EN, 5 for 100V EN

3 Legal Notes

3.1.1 Copyright

All rights reserved. It is not allowed to copy and publish the manual or parts of it in any form as copies, micro film or other methods without a written authorization from Biometra.

Biometra is pointing out that applied company and brand names are usually protected trademarks.

3.1.2 Liability





Biometra is not liable for damages and injuries caused by use not considering these operating instructions in parts or completely.

3.1.3 Meaning of instructions

Biometra recommends that you first read these instructions carefully. This operation instruction is part of the product and should be kept over the full life-time of the instrument. It should also be forwarded to subsequent owners and users. Make sure that additions and updates are inserted into the operation instructions.

4 Safety and Warning Notices

4.1 Definition of Symbols

<u>Symbol</u>	<u>Definition</u>
	Caution! Refer to instruction manual!
	Danger! High voltage!
	Fragile!
	Danger! Hot surface!

4.2 General Safety Instructions

Please read this manual carefully before starting operation of the Biometra TRIO Thermal Cycler. The Biometra TRIO Thermal Cycler is intended for sample incubation at varying temperatures.

General safety precautions for laboratory work must be observed when working with the instrument.

The Biometra TRIO Thermal Cycler does not produce a sound power level hazardous for the user.

The sample block and the heated lid will reach high temperatures during operation. Both sample block and heated lid can burn you. Do not touch the sample block or heated lid.



Rapid heating of the sample block can cause liquids to boil explosively. Always wear safety goggles during operation. Close the lid before starting a program.



Do not heat samples without having the lid locked securely.



Be aware that samples are reaching high temperatures. Do not touch or open hot tubes or microplates because hot liquid may quickly spill out.



Use only suited plastic ware in the Biometra TRIO Thermal Cycler. Tubes and plates must show good fit when placed in the sample block. Only use tubes that are suited for high temperatures (tight lids).



The Biometra TRIO Thermal Cycler contains no user serviceable parts. Do not open the housing instrument. Service and repair may only be carried out by the Biometra Service department or otherwise qualified technical personal.



Do not use the instrument when damages of the housing, block, cable or other parts are visible.



Prior to connecting the unit to the power source, please ensure that the voltage selector at the bottom of the instrument is set to the required voltage (see chapter 5.4).



Make sure that the main supply voltage is in accordance with the label above the power connection.



Unplug the power cable before you open the Biometra TRIO Thermal Cycler. Danger of electric shock!



Make sure that the appliance connector and the plug of the supply cord are accessible, so you can separate the instrument from the mains.



Connect the Biometra TRIO Thermal Cycler to a grounded socket.



When only few samples are put in the block place additional tubes in the four corner positions. This is to evenly distribute the lid pressure and prevents single tubes from excessive pressure. Use of few tubes may result in damage of the tubes by excessive pressure.



Appropriate safety regulations must be observed when working with infectious, pathogenic or radioactive material. Ask the responsible local safety inspector for details.



The Biometra TRIO Thermal Cycler must not be used with explosive, flammable or volatile liquids.



Do not place fingers between lid and housing when opening or closing the lid.



Before opening of the lid, release lid pressure (see section 6.3)



It is not necessary to apply oil into the opening of the block in order to improve the heat transfer between the block and the sample tubes.

If you still decide to use oil, do not use silicon oil. Mineral oil may be used.



Ensure that both the rear and bottom ventilation slits not clogged by dust or other material. Danger of overheating!



Let equilibrate the Biometra TRIO Thermal Cycler to room temperature before starting operation.



There must be sufficient distance between the ventilation slots on the rear side of the Thermal Cycler and a wall or another instrument (min 10 cm). Danger of overheating!



This instrument is designed and certified to meet EN 61010-1 safety standards. It should not be modified or altered in any way. Alteration of this instrument will void the warranty, void the EN61010-1 certification, and create a potential safety hazard.



Place the Biometra TRIO Thermal Cycler on a stable, non-flammable surface in a dry, safe environment. For details see working conditions in table “Technical specifications” (see chapter 2.4).



Do not use alcohol (e.g. methanol, ethanol), organic solvents or abrasives to clean the instrument.



For transport always use the original Biometra box.

5 Installation

5.1 Content of delivery

- 1) Thermal Cycler
- 2) Mains connector
- 3) Manual
- 4) Short Manual

Please keep the original packaging material for return shipment in case of servicing. The Biometra TRIO Thermal Cycler shipping box provides a specially developed system for contact-free transport of this electronic device.

5.2 Unpack and Check

Unpack and carefully examine the instrument. Report any damage to Biometra.

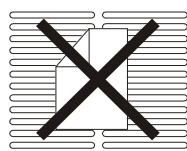
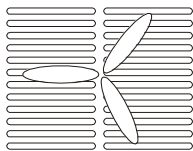


Do not attempt to operate this device if physical damage is present!

Please keep the original packing material for return shipment in case of service issues.

5.3 Installation Conditions

- Place the Biometra TRIO Thermal Cycler on a stable surface in a dry, safe environment. For details see working conditions in table “Technical specifications” (see chapter 2.4).
- Let equilibrate the Biometra TRIO Thermal Cycler to room temperature before starting operation (1 to 6h).
- Make sure that the appliance connector and the plug of the supply cord are accessible, so you can separate the instrument from the mains.
- Make sure that the ventilation slots on the bottom and the rear are not obstructed (see chapter 6.2). Make sure that there is no object underneath the Thermal Cycler that may block the ventilation slots at the bottom (e.g. a piece of paper etc.)
- There must be sufficient distance between the ventilation slots on the rear side of the Thermal Cycler and a wall or another instrument (min 10 cm).



Ensure that both the rear and bottom ventilation slits of the rear and bottom of the instrument are unobstructed.

Insufficient ventilation can cause overheating of the instrument.

- Make sure that the main supply voltage is in accordance with the label above the power connection (see chapter 5.4)
- Connect the Biometra TRIO Thermal Cycler to a grounded socket.

Prior to connecting the unit to the power source, ensure that the voltage selector at the back side of the instrument is set to the required voltage.



Danger of electric shock! Unplug the power cable before you open the Biometra TRIO Thermal Cycler!

- The display contrast can be adjusted to local light conditions (see chapter 12.1.5)

5.4 Operation Voltage

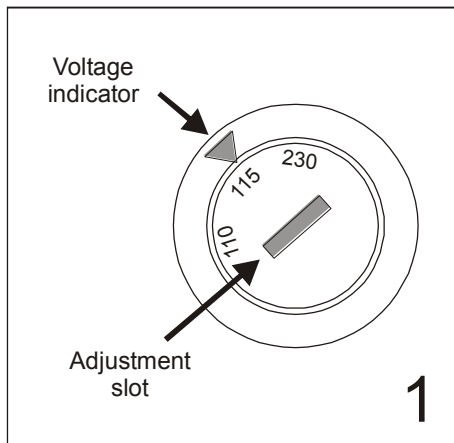
Important: Prior to connecting the Biometra TRIO Thermal Cycler to the mains, make sure that the setting of the Voltage selector is in accordance with your mains Voltage.

The Biometra TRIO Thermal Cycler can operate at 100, 115 or 230 Volt. The operation Voltage is shown on the Voltage selector which is located at the instrument bottom.

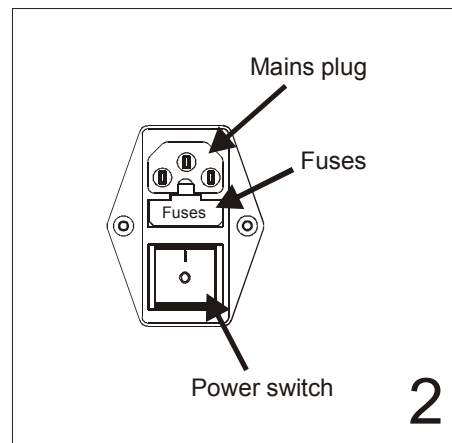
To change operation voltage, switch off the instrument and disconnect the mains plug.

Use a coin or another round shape item to turn the adjustment slot of the Voltage selector to the new Voltage.

Instrument bottom



Instrument backside



6 Operating elements

6.1 The Biometra TRIO front view



Figure 2 Biometra TRIO front view

6.2 The Biometra TRIO rear view

At the Biometra TRIO backside the Ethernet port, mains plug and mains switch are located.

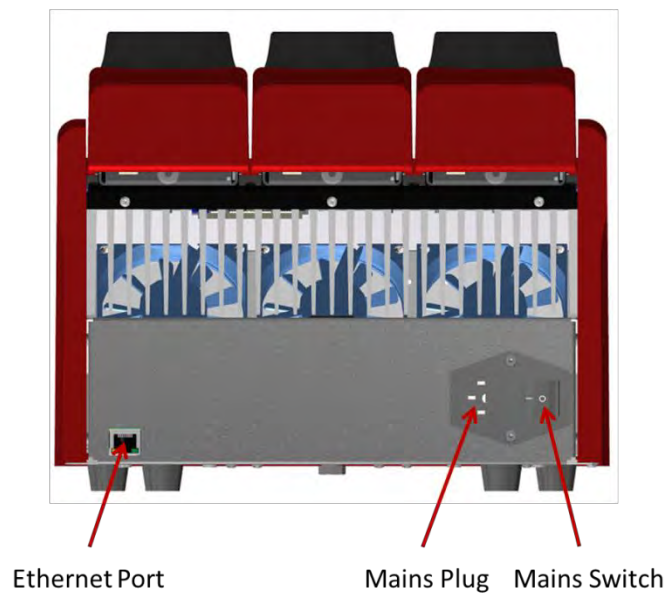


Figure 3 Biometra TRIO rear view

6.3 High Performance Smart Lid (HPSL)

To achieve optimum pressure on the tubes the Biometra TRIO Thermal Cycler is equipped with a height adjustable heated lid.

Close the lid:

After the samples have been placed in the block close the lid. Turn the wheel clockwise until you hear a clicking noise. In this mode the pressure will not increase further, even when you keep on turning the wheel.

Open the lid:

First: Release pressure by turning the wheel counter clockwise. As soon as there is no more resistance the pressure has been released.

Then: Open the lid with by pushing the front button.

Important: The lid should not be opened under pressure because this leads to damage of the locking mechanism.

6.4 Loading samples

The Biometra TRIO Thermal Cycler is an open system and compatible with strips, tubes or plates. Regardless if plates are skirted, semi-skirted or non-skirted, if tubes have flat or domed caps or if the plastic ware has 0.1 ml or 0.2 ml format, the Biometra TRIO accepts almost all formats. By the high performance smart lid (HPSL) technology with clutch mechanism the heated lid applies heat and force to the reaction vessels to prevent condensation of the reaction mix and reliable contact pressure between plastic ware and sample block.



After a run, the heated inner lid can remain hot. Use caution when opening and closing the lid.

1. To open a lid, press on the marker indicated in Figure 1.



Figure 4 Biometra TRIO lid open button

2. Release the pressure on the button when the lid becomes unlocked
3. The lid opens automatically and stays open without assistance
4. Load the samples

Note: The heated lid force has been optimized for a fully loaded block. If only very few tubes are loaded to the block place a dummy tubes each in the four corner positions to avoid damage of tubes by excessive pressure.

	1	2	3	4	5	6
A	●	○	○	○	○	●
B	○	○	○	○	○	○
C	○	○	○	○	○	○
D	○	○	○	○	○	○
E	○	○	○	○	○	○
F	○	○	○	○	○	○
G	○	○	○	○	○	○
H	●	○	○	○	○	●

5. To close the lid push it down until it becomes arrested by the locking mechanism.

6.5 The Biometra TRIO Software

The Biometra TRIO features a 7" color touch screen and a completely new software as user interface.

6.5.1 Keyboard

For some applications names, addresses or passwords have to be defined. For these purposes the Biometra TRIO software offers a keyboard to enter characters, digits and special characters.

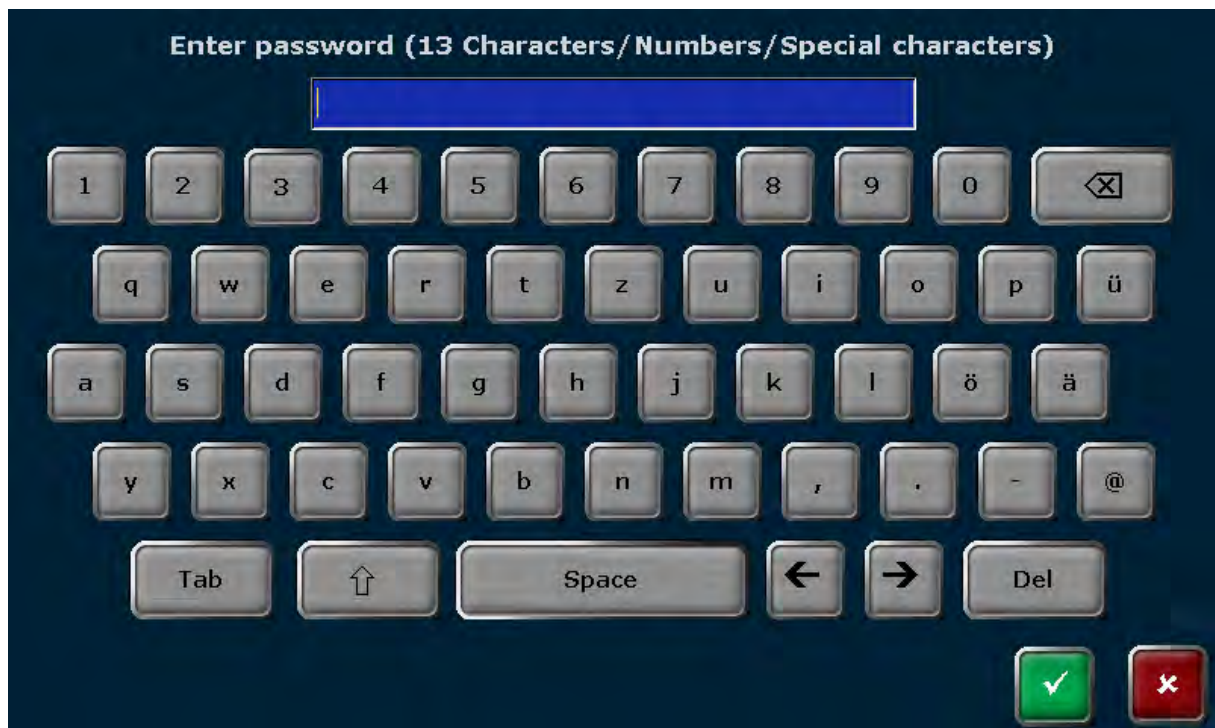








Figure 5 Biometra TRIO keyboard

The keyboard offers the following special keys:

Space

Space

Tab	
Shift	
Delete single character	
Delete all characters	
Back	
Forward	

Note: For some functions the number of maximum characters is limited. The user names and passwords can have maximal 13 characters, user initials maximal 3 characters and program names maximal 13 characters.





6.5.2 Keypad

In some screens of the Biometra TRIO software digits have to be entered. For this purpose the software features a virtual keypad integrated in some screens:



Figure 6 Biometra TRIO keypad

The keyboard offers to enter digits, dot and minus and infinite. Additionally the following special keys are available:

Delete single character	
Delete all characters	
Tab	
Infinite	

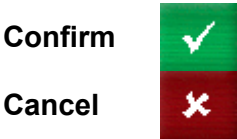
6.5.3 Cursor keys

The Biometra TRIO software uses lists, tables or graphical representations. Whenever the number of entries becomes higher than the maximum number of steps or entries that can be displayed, the software will show cursor keys for navigation.



6.5.4 Confirm or Cancel






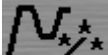
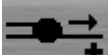
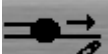
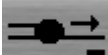











For many screens user entries have to be confirmed to avoid unintended operating errors. Press **Confirm** to accept settings/processes or press **Cancel** to refuse settings/processes.









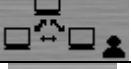






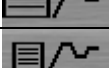
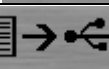
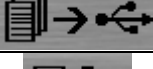
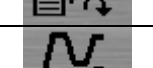







6.5.5 List of used commands and icons

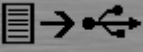
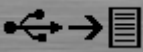



The Biometra TRIO thermal cycler software uses the following icons:

Command			Icon
Home			
Back			
Next			
Select sample block 1			1
Select sample block 2			2
Select sample block 3			3
	Right		>>
	Left		<<
	Up		^^
	Down		vv
		Confirm	
		Cancel	
Log in			

		User Management	
		User Management ON/OFF	
		New User	
		Delete User	
		Edit User	
Protocol Wizard			
	New Polymerase		
	Edit Polymerase		
	Delete Polymerase		
		Annealing temperature calculator	Calc T _a
		Edit primer concentration	
		Edit primer	ACGT 
		Calculate primer	
		Save annealing temperature T _a	T _a ✓
		Protocol Wizard Info	
Incubation			
	Start Incubation		
Programs			
	New Program		
	Open Template		
	Copy Program/Copy all Programs		
	Delete Program		

	Edit Program		
	Start Program		
	Choose block start program		
	Stop Program		
	Choose block stop program		
	Done		
	Save Program		
	Pause Program		
	Continue Program		
	Skip Step		
		Graphical view	
		Spreadsheet view	
		Temperature Optimisation Step view	
		Edit Step	
		Insert Step	
		Delete Step	
		Temperature Increment	ΔT
		Time Increment	Δt
		Ramp Rate	ΔR
		Temperature Optimisation Step (TOS)	TOS
		Preheating OFF	Off
		Preheating ON	On
Tools			

	Settings		
		Date and Time	
		Automatic user logout	
		Beeper	
		Network	
		Network Settings	
		Network User	
		Display brightness	
		Display darker	
		Display brighter	
		Touch screen calibration	
		Factory settings	
	Documentation		
		Run-Logfile	
		Overview Run-Logfiles	
		Save Run-Logfile to USB	
		Save all Run-Logfiles to USB	
		View Run-Logfile	
		View Program	
		Selftest Logfile	
		Error Logfile	
	Extended Self-Test		
		Start Extended Self-Test	
		Stop Extended Self-Test	

	Backup	Save Backup File	
		Load Backup File	
		Delete all Backup Files	
		Delete Backup File	
	Cycler Info		

7 Operating

7.1 Power On Self-Test

After the instrument is switched on the Biometra TRIO Thermal Cycler passes an initial self-test (power on self-test). While the instrument passes the test the start screen is shown

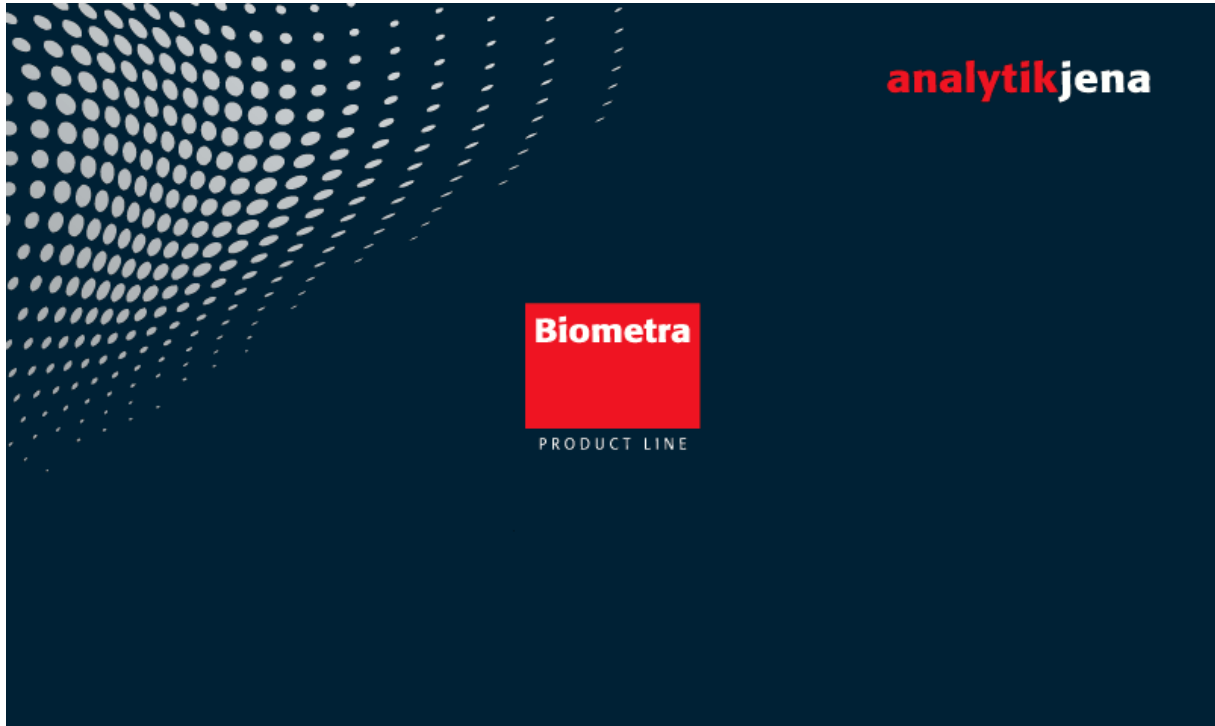


Figure 7 Biometra TRIO start screen

For the power on self-test a log file is created and stored in the Thermal Cycler memory (see chapter 12.3.2).

7.2 Log in Screen

After the instrument has passed the power on self-test the log in screen is displayed. The log in screen offers the following functions:

Command	Function	Chapter
Log in	Starts the log in dialog.	See chapter 7.3
Block	Shows the current block status. If a block is Free a touch on the button leads to a list of previously used programs for quick start for the selected sample block (not user specific before log in).	See chapter 7.5 and chapter 7.6
EN/DE	Changes the language setting for the log in screen.	For user-specific language settings see chapter 12.2.2

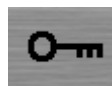


Figure 8 Biometra TRIO log in screen

7.3 Log in

After starting the Biometra TRIO Thermal Cycler the log in screen is displayed (see chapter 7.1). To log in as existing user press

Login



In the next screen press the button to selected a user.



Figure 9 Biometra TRIO select user screen

Note: The button position can change. The button for the latest logged in user is always shown at the top left position of the screen. All other buttons become arranged from top left to bottom right by the date and time of the latest user log in. If more than 6 user accounts are available you might need to use the cursor keys **Left** or **Right** to turn the page (see chapter 6.5.3).

Touch the input box “Password” and enter the password using the Biometra TRIO keyboard (see chapter 6.5.1). The default password for the administrator Admin is “Admin”. Please note that the password query is case sensitive.

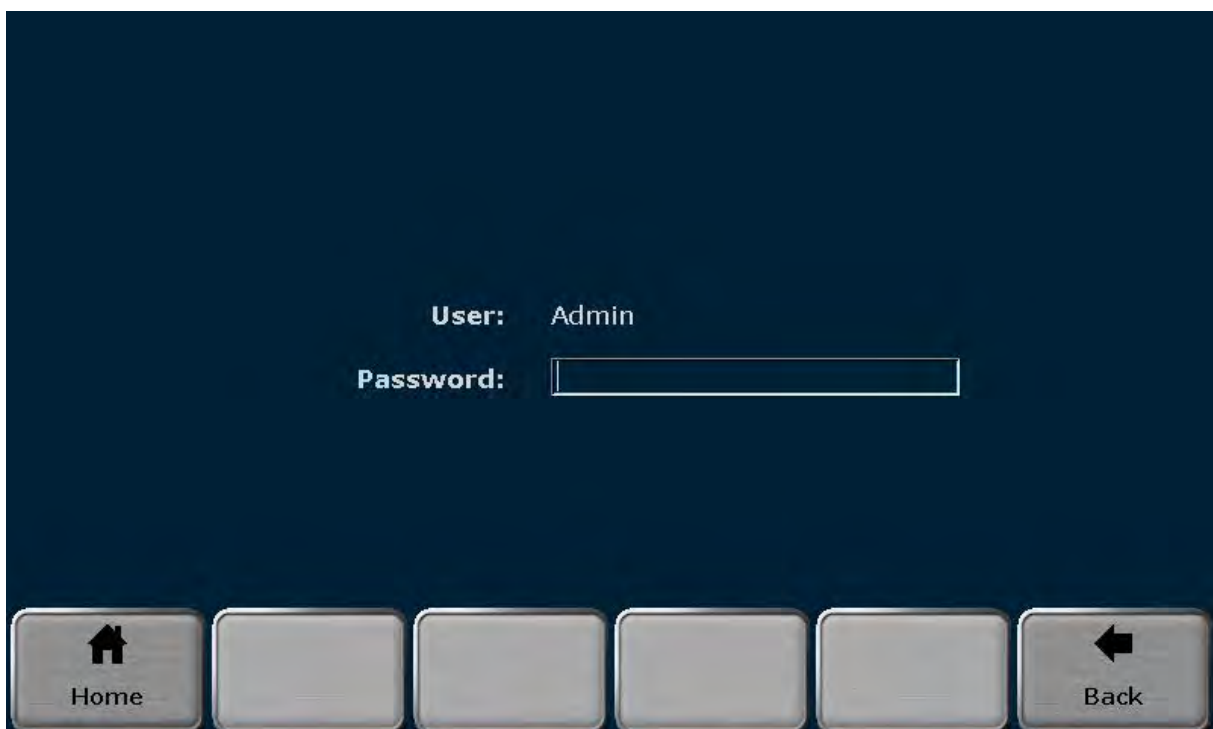


Figure 10 Biometra TRIO enter password screen

Note: If a user directory is not password protected the software skips the enter password screen (see Figure 10) and jumps directly into the home screen.

7.4 Home Screen

After user log in the home screen is shown by the Biometra TRIO software. The home screen offers the following functions:

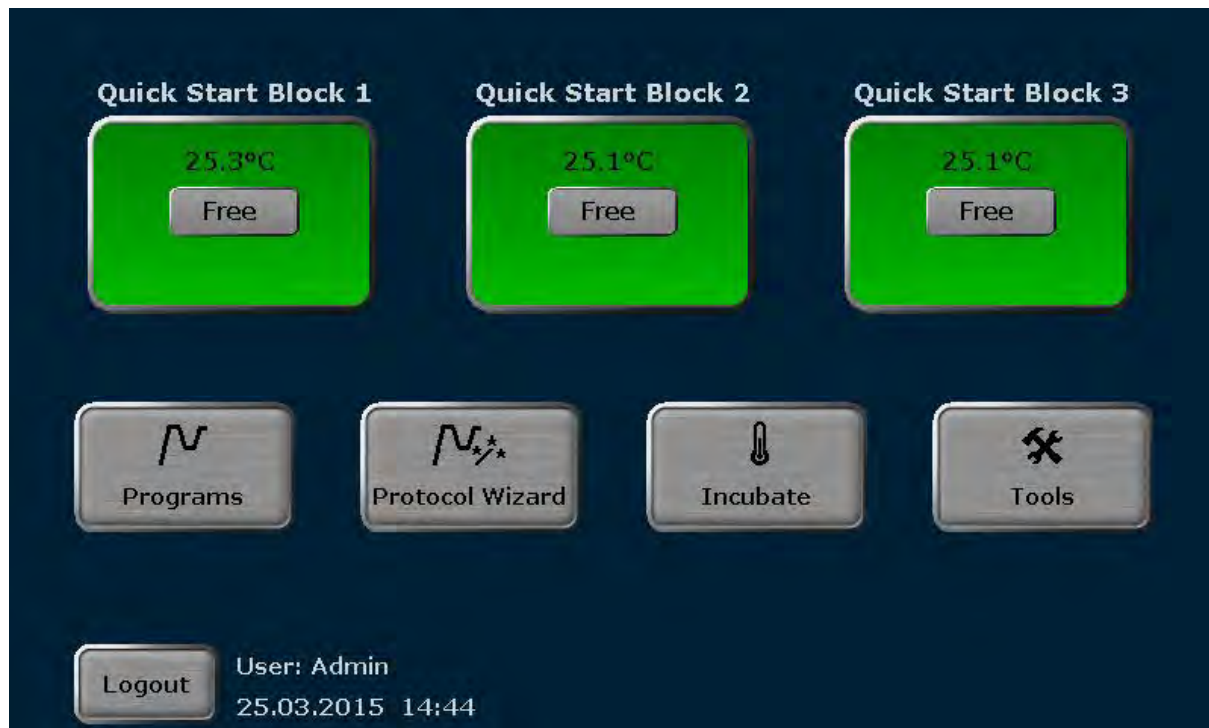


Figure 11 Biometra TRIO home screen

Command	Function	Chapter
Programs	Viewing, running and editing programs	See chapter 8
Protocol Wizard	Automatic creation of PCR programs	See chapter 11
Block	Shows the current block status. If a block is Free a touch on the button leads to a user-specific list of the previously used programs for quick start for the selected sample block	See chapter 7.5 and chapter 7.6
Incubate	For running a sample block at a constant temperature	See chapter 7.7
Tools	Settings, documentation, extended self-test, cyclor info and diagnosis mode	See chapter 11
Log out	User log out	

7.5 Block status

In the log in screen (see chapter 7.2) and the home screen (see chapter 7.4) a special button for the current sample block status is shown. The sample block status can be **Free**, **Run** or **Pause** and is indicated by the color of the button and the textbox in the middle of the button ②. In the line above the textbox the current sample block temperature is shown ①. If the temperature is higher than 70°C the characters are shown in red and the warning Hot plus the corresponding warning symbol are displayed. In the line below the textbox the remaining run time is shown ③. The block number is shown above the button.

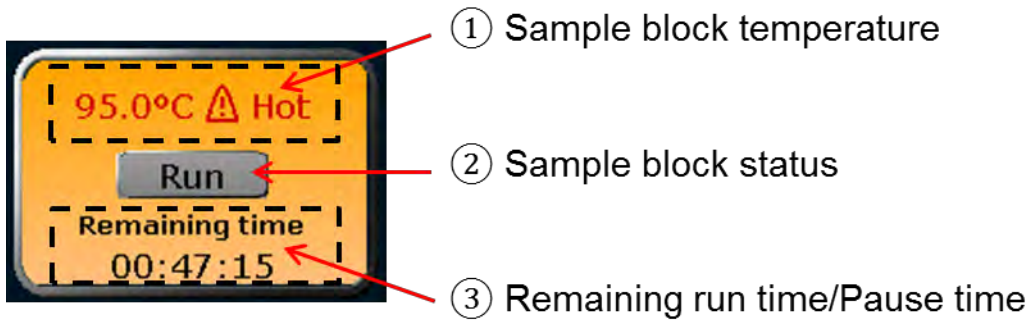


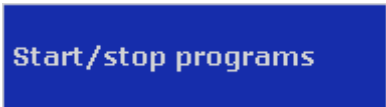
Figure 12 Biometra TRIO block status/quick start button

The following table summarizes the different button colors and sample block status. When the button is pressed, it depends on the sample block status which function is activated by the Biometra TRIO software. If the status is **Run** or **Pause**, a touch of the button will lead to the program overview (see chapter 0). If the status is **Free**, the program quick start function is activated (see chapter 7.6).

Button	Block Status	Color	Function
	Free	Green	Quick start function
	Run	Yellow	Program view
	Pause	Blue	Program view

7.6 Quick start function

Note: Programs can only be started by users with the right **Start/stop programs** activated (see chapter 12.2.2).



The Biometra TRIO software offers a quick start function for previously used programs. To use the quick start function, press the block status button in the log in screen (see chapter 7.2) or the home screen (see chapter 7.4).

Note: The quick start function can only be activated if the current block status is **Free** (see chapter 7.5). If the status is **Run** or **Pause** a touch of the button will lead to the program overview screen instead.

If the block status button is used in the log in screen, before a user is logged in, the software will show a list of latest used programs for quick start for the selected sample block. This list is not user-specific. If the block button is used in the home screen, after user log in, the software will show a user specific list of the latest used programs for quick start for the selected sample block.

- Log in screen** Latest program list
- Home screen** User specific latest program list

The latest used programs are summarized in a table.



Figure 13 Biometra TRIO quick start screen

To switch between the sample blocks press

- Sample block 1** 1
- Sample block 2** 2
- Sample block 3** 3

Select and touch a program in the table for quick start. Press

Start on block x



to start the selected program.

To start a program on multiple sample blocks press

Choose block start program



and by activating the checkboxes select the sample blocks to start.

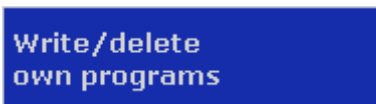


Figure 14 Biometra TRIO multiblock selection query

If necessary the program can be edited before or a new program can be created using a template (see chapter 8.1).

Note For programs including a TOS step (see chapter 8.6.5) all blocks are started simultaneously and therefore the block selection option is not offered.

Note: Programs offered for quick start can only be edited or created by users with the right **“Write/delete own programs”** activated (see chapter 12.2.1).

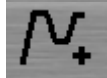


Press the corresponding button

Edit Program



New from template



to activate the programming mode or to select a program from the list of templates.

7.7 Incubation Mode

Note: Incubations can only be started or stopped by users with the right **Start/stop programs** activated (see chapter 12.2.2).

Start/stop programs

The incubation mode allows to incubate samples at a constant temperature for a defined time. Press

Incubate



and in the next screen set the block temperature, hold time, heated lid on or off, heated lid temperature and set preheating mode on or off using the virtual keypad and the corresponding buttons (see Figure 15).



Note: To set the time to endless use button

To select a sample block press

Sample block 1

1

Sample block 2

2

Sample block 3

3

To start the incubation press

Start incubation





Figure 15 Biometra TRIO incubation mode screen

During incubation the following commands can be used:

Stop Incubation

Choose block*

Pause Incubation

Continue Incubation

Skip Step



* the button "choose block stop program" for multiblock selection is only available if more than one sample block is running.

Note: To stop the incubation, use the corresponding command. Do not switch off the instrument while the incubation is running. The instrument will react like after a power failure and will restart the incubation step when it is switched on next time.

8 Creating, Editing and Saving programs

8.1 Programming modes

The Biometra TRIO software offers spreadsheet programming or graphical programming. Use the buttons

Graphical view



Spreadsheet view



to toggle between the two modes (see Figure 16). The following screenshots show the spreadsheet and graphical programming mode.

Spreadsheet programming

User: Admin 25.03.2015 17:12

Name: Lid: °C Preheat Lid:

Program Runtime: 01h 22m 51s

Step	Block Temp. (°C)	Hold Time (h:mm:ss)	Go To	Cycles	ΔT(°C)	Δt(s)	ΔR(°C/s)
1	95.0	0:05:00	--	--	--	--	6.0
2	95.0	0:00:30	--	--	--	--	6.0
30x	59.0 \uparrow 61.0	0:00:30	--	--	1.0	1	1.0
4	72.0	0:00:30	2	30	--	--	6.0
5	72.0	0:05:00	--	--	--	--	6.0
6	15.0	∞	--	--	--	--	6.0

Graphical programming

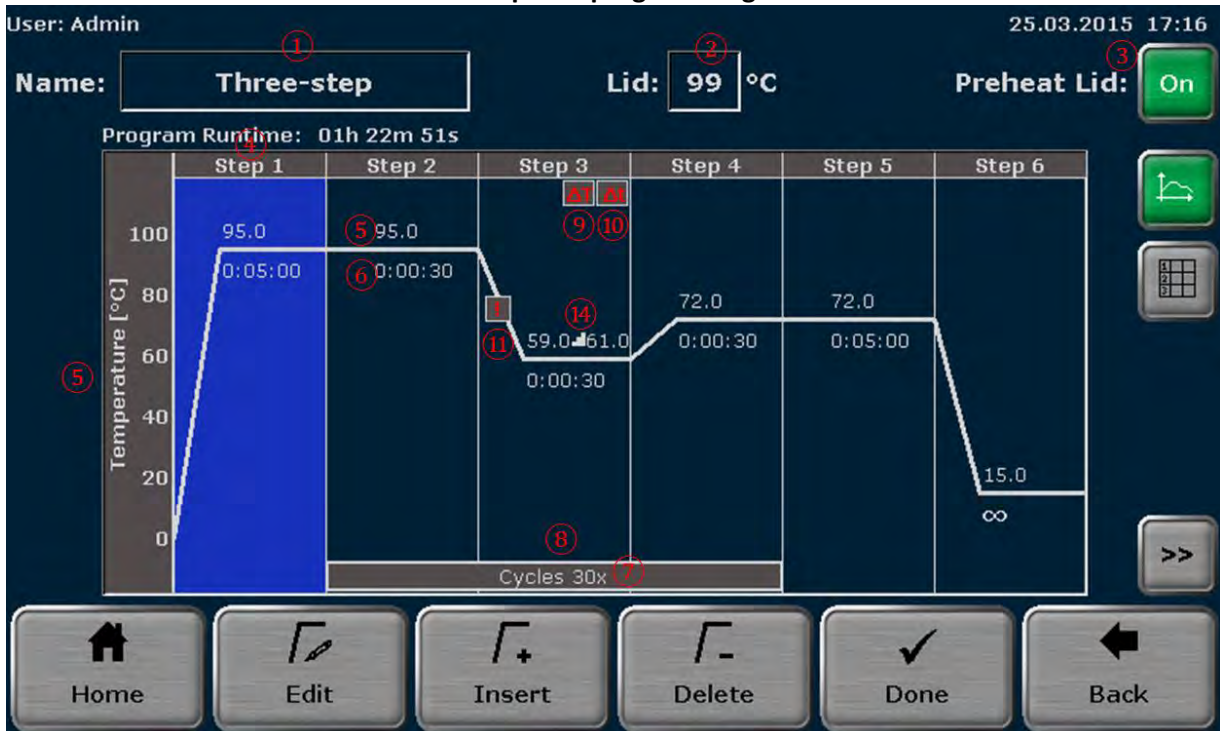


Figure 16 Biometra TRIO spreadsheet and graphical programming screen

Both programming modes show the same information but in different ways. In the spreadsheet mode all parameters are summarized in a table. In the graphical mode for some parameters symbols are displayed (see table below). Toggle to the spreadsheet mode to view the settings for these parameters.

Parameter	Spreadsheet mode	Graphical mode	Chapter
Program name	①	①	See chapter 8.3
Heated lid temperature [°C]	②	②	See chapter 8.4
Preheat lid mode	③	③	See chapter 8.5
Step number	④	④	See chapter 8.6
Temperature [°C]	⑤	⑤	See chapter 8.6
Holding time [hh:mm:ss]	⑥	⑥	See chapter 8.6
Goto	⑦, ⑫	⑦	See chapter 8.6.1
Cycles	⑧, ⑬	⑧	See chapter 8.6.1
Temperature increment [ΔT (°C)]	⑨	⑨ ΔT	See chapter 8.6.2
Time increment [Δt (s)]	⑩	⑩ Δt	See chapter 8.6.3
Ramping rate [ΔR (°C/s)]	⑪	⑪ !	See chapter 8.6.4
TOS step	⑭	⑭	See chapter 8.6.5

If a program has more than 6 steps, use the cursor keys to navigate in the spreadsheet or the graphical display (see chapter 6.5.3)

8.2 Creating a new program/Use program template

Note: New programs can only be created by users with the right **“Write/delete own programs”** activated (see chapter 12.2.1). Programs of other users can only be modified by users with the right **“Write/delete all programs”** activated

Write/delete
own programs

1. To create a new program or to open a program template, first log in as existing user (see chapter 7.1).
2. After log in press button

Programs



in the home screen (see chapter 7.3). The program overview screen opens:

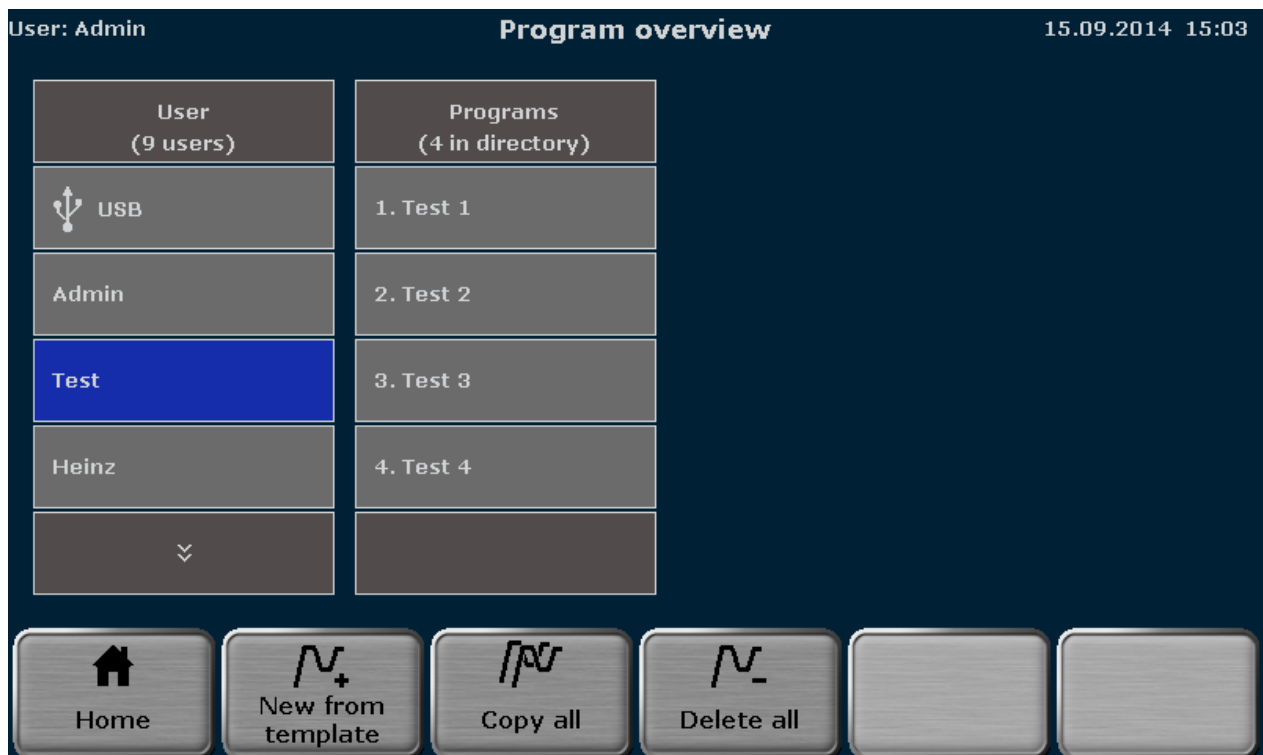
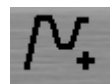


Figure 17 Biometra TRIO program overview screen

3. In the program overview screen press button

New from template



4. The program template screen opens (see Figure 19). Search for a program template using the cursor keys

Scroll up



Scroll down



and touch the button to select it.

Note: The Biometra TRIO offers several program templates for different purposes. You may use a template and modify the program as desired or you can select “Blank”. “Blank” is program with no pre-defined steps (see Figure 18).

Spreadsheet programming

Graphical programming



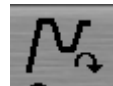
Figure 18 Biometra TRIO program template “Blank”.

Select “Blank” whenever a new program should be created and select another template from the list whenever an existing program should be edited.

The program structure can be controlled in the program preview. If a template is selected the program steps and some additional information are displayed in the program preview table. By using the program preview function the template can be checked before it is opened.

5. Press

Open template



to open a program template.

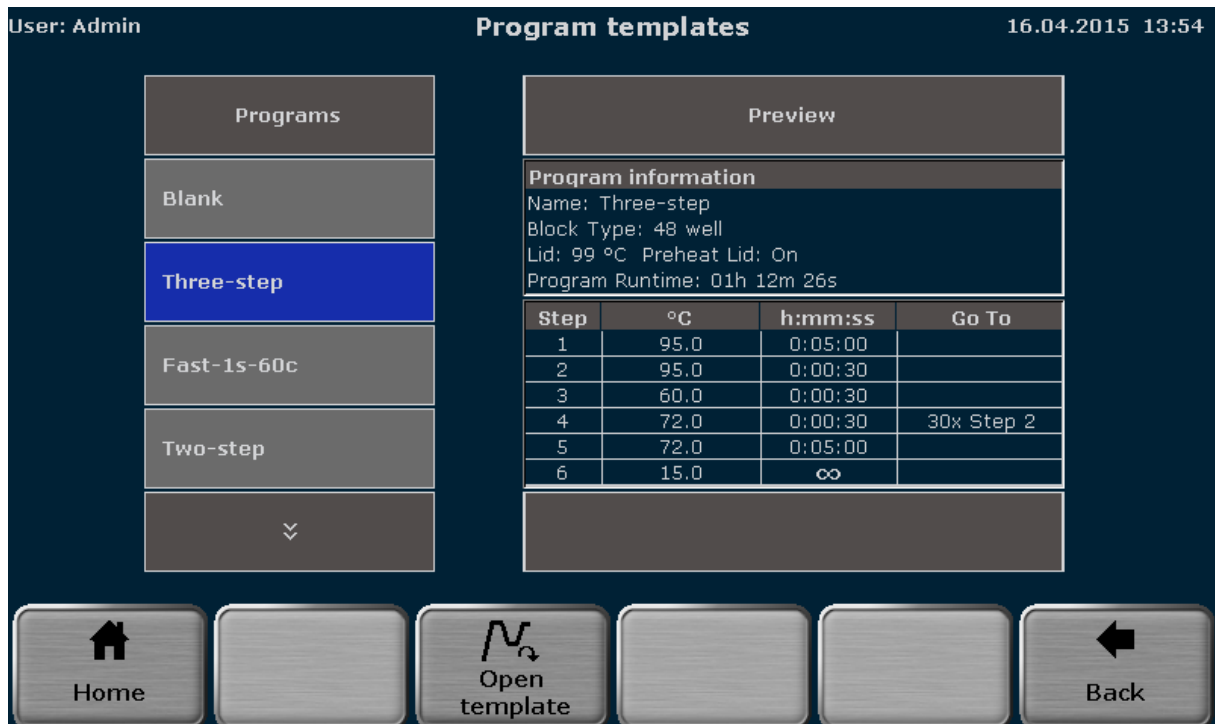


Figure 19 Biometra TRIO program template screen

8.3 Set program name

Each template has a specific name which is superimposed in the programming screen. To enter a new name touch the input box **Name** (see Figure 16) and enter a new name using the Biometra TRIO virtual keyboard (see chapter 6.5.1).

Note: If a character is pressed, the pre-displayed name will be deleted and a new program name can be set. If the program name should be modified please first press another button like delete or space.

8.4 Set heated lid temperature

To set the heated lid temperature, touch the input field **Lid** (see Figure 16) and in the next screen (see Figure 20) enter a value between 30°C and 110°C using the Biometra TRIO virtual keypad.

Note: The default setting for the heated lid temperature is 99°C. 99°C is sufficient for typical PCR programs.

To switch the heated lid on or off use the corresponding buttons.



Figure 20 Biometra TRIO lid temperature setting screen

8.5 Set preheating mode

The heated lid can be set to preheating ON or OFF using the corresponding buttons (see Figure 16):

Preheating OFF



Preheating ON

If the preheating mode is activated the heated lid is heated up to the target temperature while the sample block is held constant at 25°C. After the heated lid reaches the target temperature a 40 seconds equilibration phase follows. Then the sample block starts to heat up from 25°C to the first programmed temperature.

Note: For most applications it is recommendable to set the preheating function to “ON”.

8.6 Edit parameter or step

The software offers two different options to edit program steps or single parameters:




1. In the programming spreadsheet or the graphical view single parameters can be activated and values entered.
2. To edit all parameters in a program step the software offers the edit step screen (see Figure 21). Furthermore in this screen it can be switched between steps and parameters entered for multiple steps (Multi Step Programming).

Activate a parameter in the tabular or graphical view (see chapter 8.1) and enter the desired values or select a program step and press

Edit Step



The following parameters can be edited:

Parameter	Description and ranges	Chapter
Temperature	Temperature in °C for a step. The target temperature can be set between 3.0°C and 99.0°C in tenths of a degree.	See chapter 8.6
Hold time	Hold time for a step in hh:mm:ss. Between 0 and 8 hours, 0 and 59 minutes and 0 to 59 seconds can be set. To program a pause and set the holding time indefinitely, enter ∞ in one of the input boxes for hh:mm:ss.	See chapter 8.6
Goto	Defines the step number the program jumps back to.	See chapter 8.6.1
Cycles	Defines the number of repeats. The maximum number of repeats is 999.	See chapter 8.6.1
	Temperature increment to increase or decrease the set temperature with each cycle by the defined value. The increment can be set to ± 20°C.	See chapter 8.6.2
	Time increment (e.g. for long range PCR). Increases the set time with each cycle by the defined increment. The increment can be set from 0s to 240s.	See chapter 8.6.3
	Average ramp rate between steps. The average ramp rate can be set from 0.1°C/s to max in tenths of a degree (The maximum ramp rate is sample block specific). The ramp rate specifies the speed at which the selected step is reached with.	See chapter 8.6.4


	<p>Temperature Optimisation Step (TOS). For optimisation of new PCR protocols very often gradient enabled Thermocyclers are used. The Biometra TRIO offers no gradient function but by the temperature optimisation step (TOS) function programs making use of the three blocks and delivering different annealing temperatures at the defined step can be created.</p>	<p>See chapter 8.6.5</p>
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Figure 21 Biometra TRIO edit step screen

Note: To program a step at least the temperature and the hold time must be defined.

To set the temperature touch the corresponding input box and enter a value between 3.0°C and 99.0°C. Use the Tab button or touch the corresponding input box to activate the next input box and enter the time in h:mm:ss.

The Biometra TRIO software offers to program several steps subsequently. Use the buttons



to navigate between the steps. The currently activated step is highlighted in the program overview table (see Figure 21).

Note: By navigating between steps it is not necessary to confirm the settings for each single step and to leave the edit step screen but you can fill in the parameters for several steps and confirm the settings after all steps are programmed.

8.6.1 Set cycles

A typical PCR program consists of repeated steps for denaturation, annealing and extension. For the repetition of steps the Biometra TRIO software offers to program loops. For a cycle two parameters have to be set:

1. The step number to go back to (**Goto**)
2. The number of repeats (**Cycles**)

To program a loop select the **last step** of the cycle and enter the step number the program shall go back to in the input box "**Goto**" (see Figure 21). To define the number of repeats, enter a number in the input box "**Cycles**" (see Figure 21).

For example to program the cycles for the following protocol select step **4**, enter **2** in the input box "**Goto**" and enter **35** in the input box "**Cycles**".

Step No.	Temp.	Time	Cycles
1	94 °C	05:00	
2	94 °C	01:00	
3	55 °C	01:00	35x
4	72 °C	01:00	
5	72 °C	05:00	
6	16 °C	Pause	

In this example the instrument first executes step 1 to step 4, then repeats step 2 to step 4 for 34 times, performs the final extension step at 72°C and holds the temperature indefinitely at 16°C.

Note: The Biometra TRIO allows to set maximal two interlaced loops. It is possible to program a loop within a loop but it is not allowed to set loop in loop in a loop.


8.6.2 Set temperature increment

For some applications, like for example touch down PCR, temperature increments are used. The temperature is increased or decreased with each cycle by the set value. To program a temperature increment press

Temperature Increment (ΔT)



(see Figure 21) and fill in a value between +20°C and -20°C in the corresponding input box.

Note: Temperature increments only become active in repeated steps (cycles). If a temperature increment has been programmed, the small symbol  will be shown at the corresponding step in the graphical programming mode.


8.6.3 Set time increment

For some applications, like for example long range PCR, time increments are used. The time is increased with each cycle by the set value. To program a time increment press

Time Increment (Δt)



(see Figure 21) and fill in a value between 0s and 240s in the corresponding input box.

Note: Time increments only become active in repeated steps (cycles). If a time increment has been programmed, the small symbol  will be shown at the corresponding step in the graphical programming mode.


8.6.4 Adjust ramp rate

The average ramp rate can be adjusted for each step. The ramp rate adjustment can be useful if PCR programs are transferred from slower Thermal Cyclers the Biometra TRIO or for special PCR applications like for example telomerase PCR. The average ramp rate specifies the speed at which the selected step is reached with. To adjust the ramp rate press

Ramp Rate (ΔR)



(see Figure 21) and fill in a value between 0.1°C/s and max in tenths of a degree.

Note: The maximum average rate depends on the sample block installed in the Biometra TRIO Thermal Cycler. If the ramp rate has been adjusted, the small symbol  will be shown at the corresponding step in the graphical programming mode.

8.6.5 Set Temperature Optimisation Step (TOS)

For optimisation of new PCR protocols very often gradient enabled Thermocyclers are used. The Biometra TRIO offers no gradient function but by the temperature optimisation step (TOS) function programs making use of the three blocks and delivering different annealing temperatures at the defined step can be created.

Sample replicates are distributed across the sample blocks and are tested for the temperature giving the optimal result. To program a Temperature Optimisation Step press

Temperature Optimisation Step (TOS)



(see Figure 21), enter the desired annealing temperature (**Annealing Temp.**) and define a temperature increment (**Increment**) between the sample blocks. The effective temperatures for all sample blocks are displayed (see Figure 22).

Note: The annealing temperature is set for sample block 2 and the increment defines the temperatures for sample block 1 and 3. When setting a positive increment the temperature in sample block 1 is the lowest and the temperature in sample block 3 is the highest. Use the minus button on the keypad to enter a negative increment. When a negative value is used as increment the temperature in sample block 1 is the lowest and the temperature in sample block 3 is the highest.

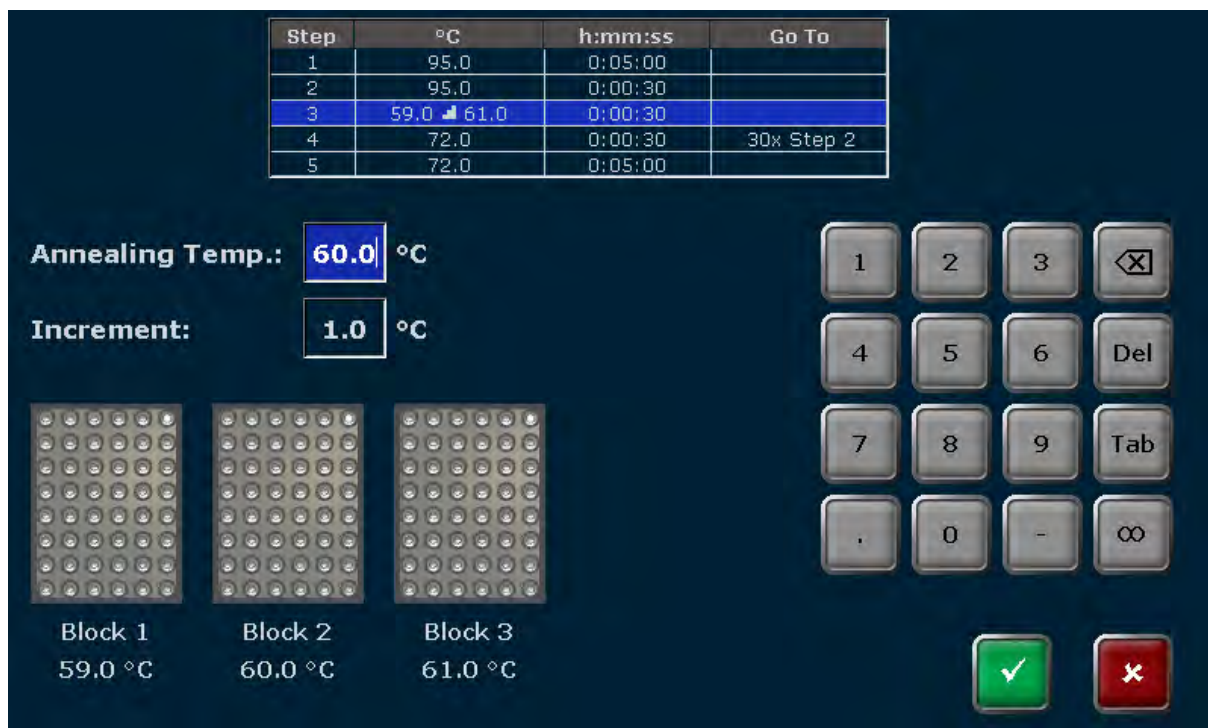


Figure 22 Biometra TRIO Temperature Optimisation Step screen

8.7 Insert step

To insert a new step, select a program step in the Biometra TRIO programming screen (see Figure 16) and press



By using the button **Insert Step** the **Edit Step** function (see chapter 8.6) is activated. Set the parameters for the new step.

Note: The new step is inserted at the selected program step. If for example step 3 is selected, the new step will be inserted at step 3. The existing step 3 is not overwritten, but instead step 3 and all following steps will be moved one position higher. This means, the existing step 3 will become step 4, step 4 will become step 5 and so on.

Note: If the new step is inserted to an existing loop, the number of steps within the loop increases, whereas the number of cycles remains the same.

8.8 Delete step

To delete a step, select a program step in the Biometra TRIO programming screen (see Figure 16) and press



Note: If a step is deleted, all following steps will be moved one position lower. If for example step 3 is deleted, step 4 will become step 3, step 5 will become step 4 and so on.

Note: If the step is deleted in an existing loop, the number of steps within the loop is reduced, whereas the number of cycles remains the same.

8.9 Save Program

To save a program press

Done



in the Biometra TRIO programming screen (see Figure 16). In the next screen (see Figure 23) touch the corresponding buttons to select a user directory and a storage place for the program. If the number of user directories or programs is higher than can be maximal displayed in the list, the buttons above and below the user and program list can be used to scroll up and down.

Note: The user directory of the logged in user is automatically pre-selected. Users with the right "Write/delete all programs" (see chapter 12.2.1) can also select directories of others to save programs.

Note: If a new program is to be saved, the first free storage place in the user directory of the logged in user is pre-selected. However, the storage place can be modified by using the cursor key function above and below the list. If the selected storage place is already in use, the stored program will not be overwritten, but instead the program stored at the selected storage place and all other following programs will be moved to a higher storage place. The storage place number for these programs will increase by +1 and the new program is saved at the selected storage place.

After a user directory and storage place are selected, press

Save



in the Biometra TRIO program overview screen (see Figure 23) to save the program.

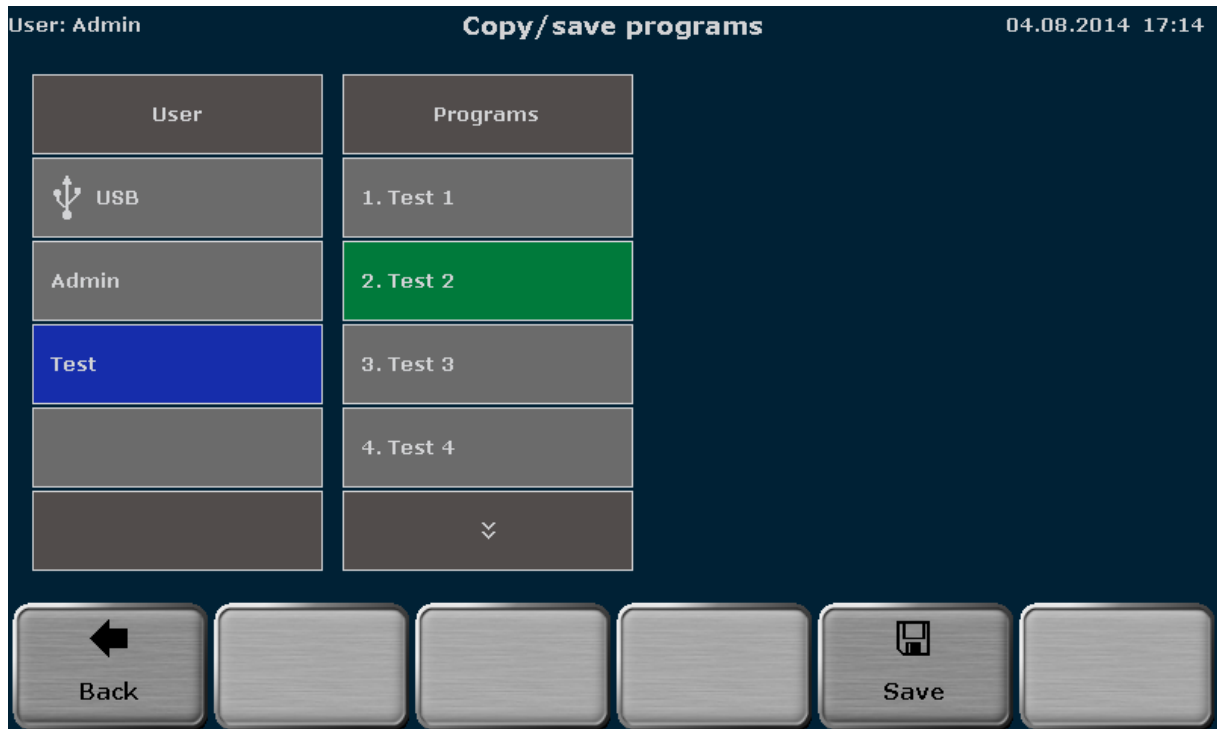


Figure 23 Biometra TRIO program overview screen

9 Starting, Copying and Deleting programs

9.1 Start program

Note: Programs can only be started by users with the right “**Start/stop programs**” activated (see chapter 12.2.2). Additionally the right “**Read all programs**” defines, if a user is allowed to read all programs or if the access is limited to own programs. A user who is not allowed to read all programs cannot start programs of others even if the right “**Start/stop programs**” is activated.



The Biometra TRIO software offers a quick start function for previously used programs. The quick start function is described in chapter 7.6. This chapter deals with the start of stored programs by selecting a program from a user account. To select a program for start, press

Programs



in the Biometra TRIO home screen (see chapter 7.4). The program overview screen opens with the user directory of the logged in user pre-selected (see Figure 24).

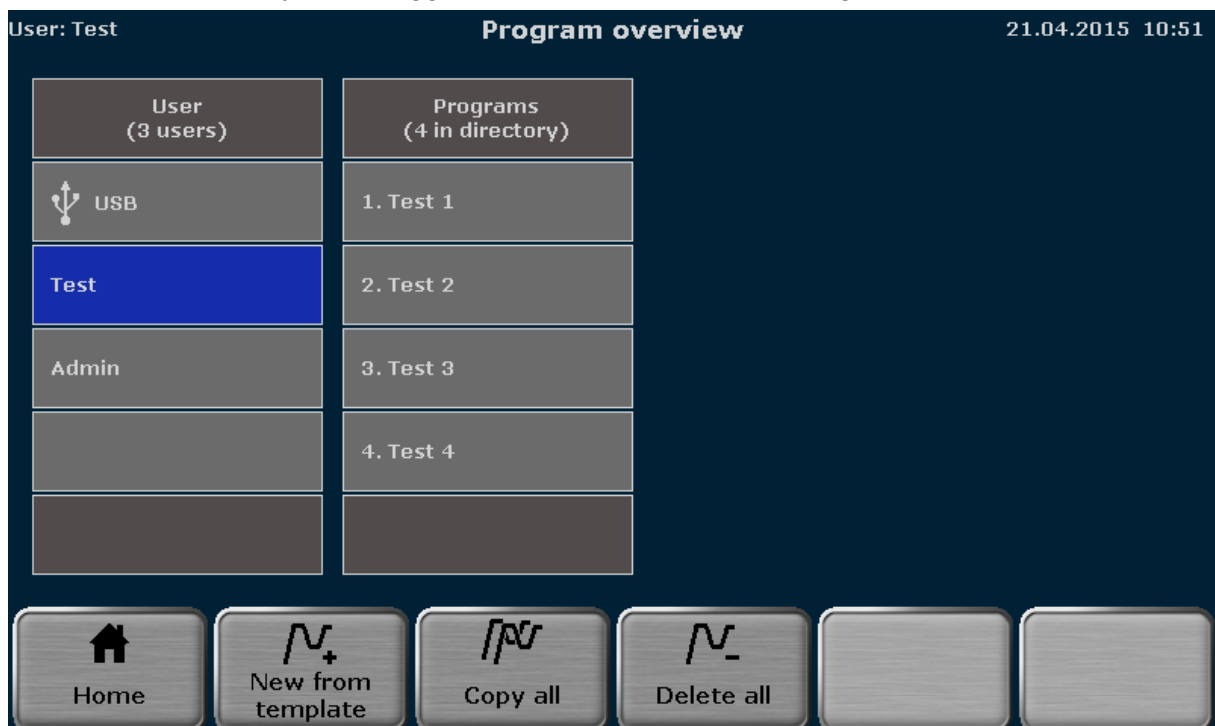


Figure 24 Biometra TRIO program overview screen

Touch the corresponding buttons to select a user directory and a stored program. If the number of user directories or programs is higher than can be maximal displayed in the list, the buttons above and below the user and program list can be used to scroll up and down.

For the selected program a preview opens with general information on the program and a list of the steps (see Figure 25). The program preview can be used to check the program prior to start. To start the selected program press

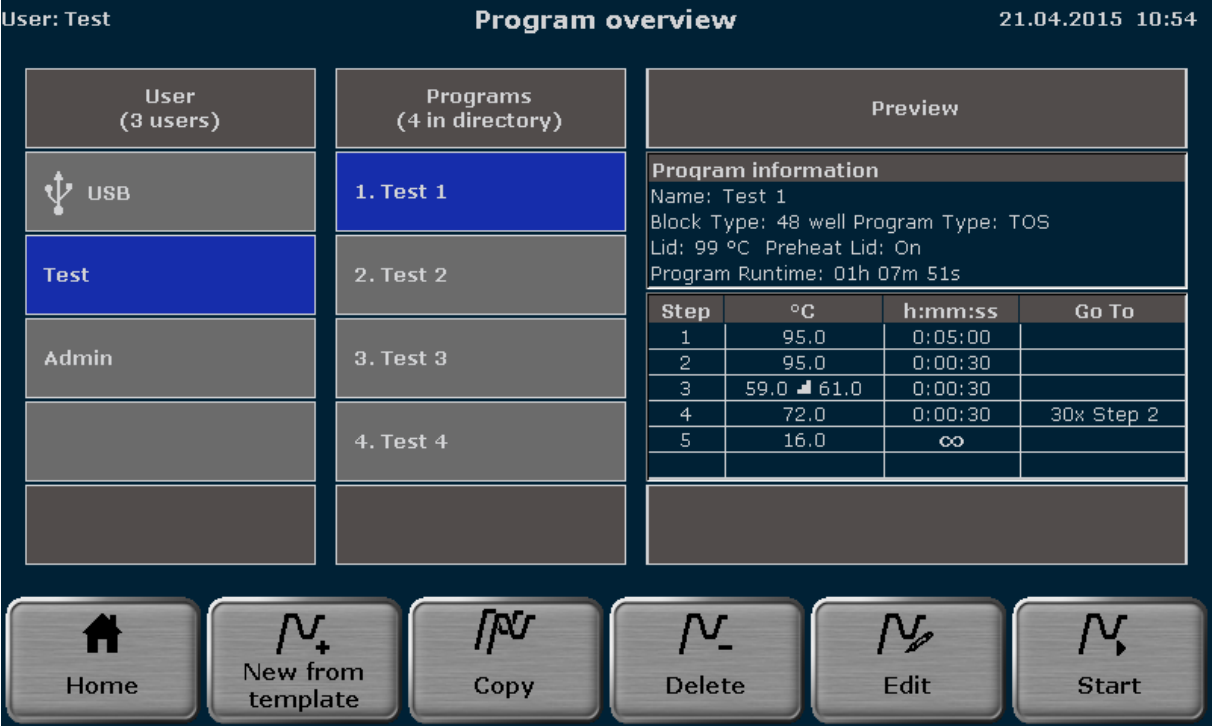


Figure 25 Biometra TRIO program overview screen with program preview

In the multiblock selection query select the sample block to start



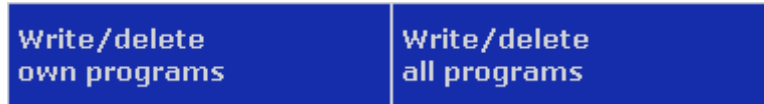
Figure 26 Biometra TRIO multiblock selection query

and confirm your selection. The program will be started on the selected sample blocks.

Note For programs including a TOS step (see chapter 8.6.5) all blocks are started simultaneously and therefore the block selection option is not offered.

9.2 Copy program

Note: Own programs can only be copied and saved by users with the right **“Write/delete own programs”** activated. Programs of others can be copied and saved with the right **“Write/delete all programs”** activated (see chapter 12.2.2).



To copy a program, touch the corresponding buttons to select a user directory and a stored program in the Biometra TRIO program overview screen (see Figure 24). If the number of user directories or programs is higher than can be maximal displayed in the list, the buttons above and below the user and program list can be used to scroll up and down. To copy the selected program press



Touch the corresponding buttons to select a user directory and a storage place in the Biometra TRIO program overview screen (see Figure 27). The program to be copied is highlighted in green. If the number of user directories or programs is higher than can be maximal displayed in the list, the buttons above and below the user and program list can be used to scroll up and down. To save the program press



Note: If the selected storage place is already in use, the stored program will not be overwritten, but instead the program stored at the selected storage place and all other following programs in the user directory will be moved to a higher storage place. The storage place number for these programs will increase by +1 and the program is copied to the selected storage place.

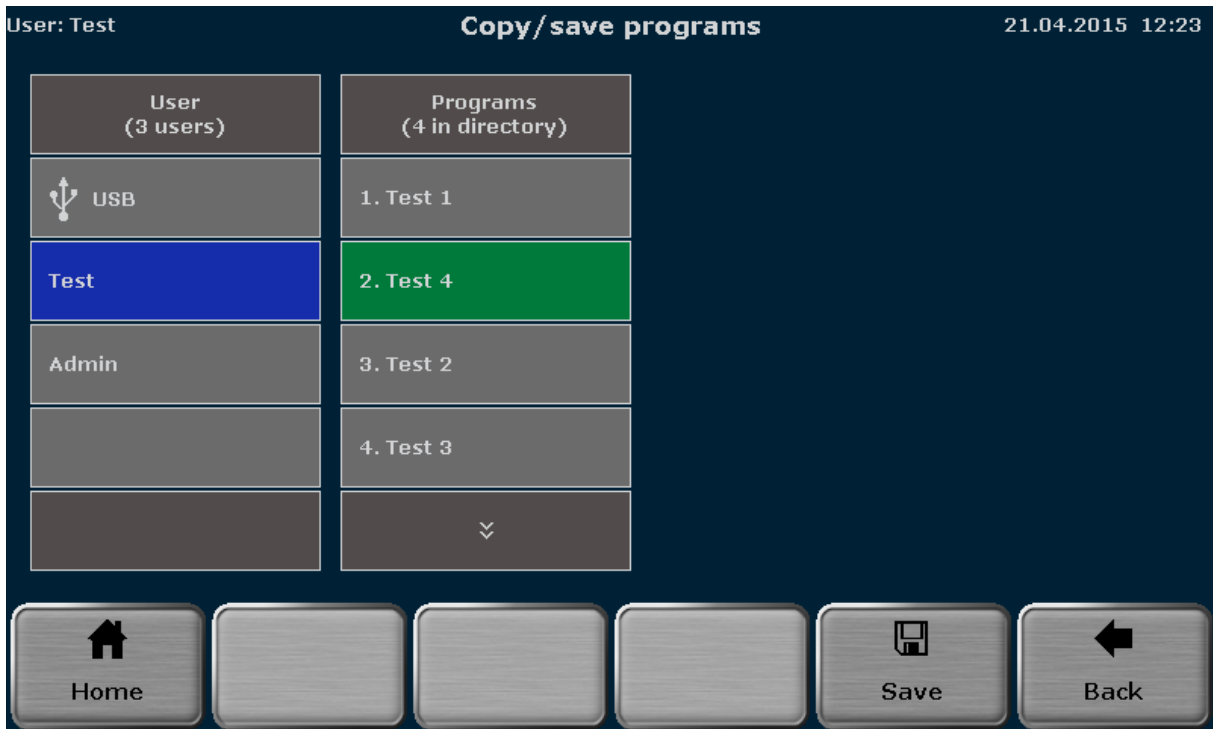


Figure 27 Biometra TRIO program overview screen

9.3 Copy all programs

Note: Own programs can only be copied and saved by users with the right **“Write/delete own programs”** activated. Programs of others can be copied and saved with the right **“Write/delete all programs”** activated (see chapter 12.2.2).



To copy all programs of a user directory, touch the corresponding button to select a user directory in the Biometra TRIO program overview screen (see Figure 24). If the number of user directories is higher than can be maximal displayed in the list, the buttons above and below the user list can be used to scroll up and down. To copy all programs press



Touch the corresponding button to select a user directory in the Biometra TRIO program overview screen (see Figure 28). If the number of user directories is higher than can be maximal displayed in the list, the buttons above and below the user list can be used to scroll up and down. To save the programs press



Note: The programs will be copied into the selected user directory beginning at the first free storage place.

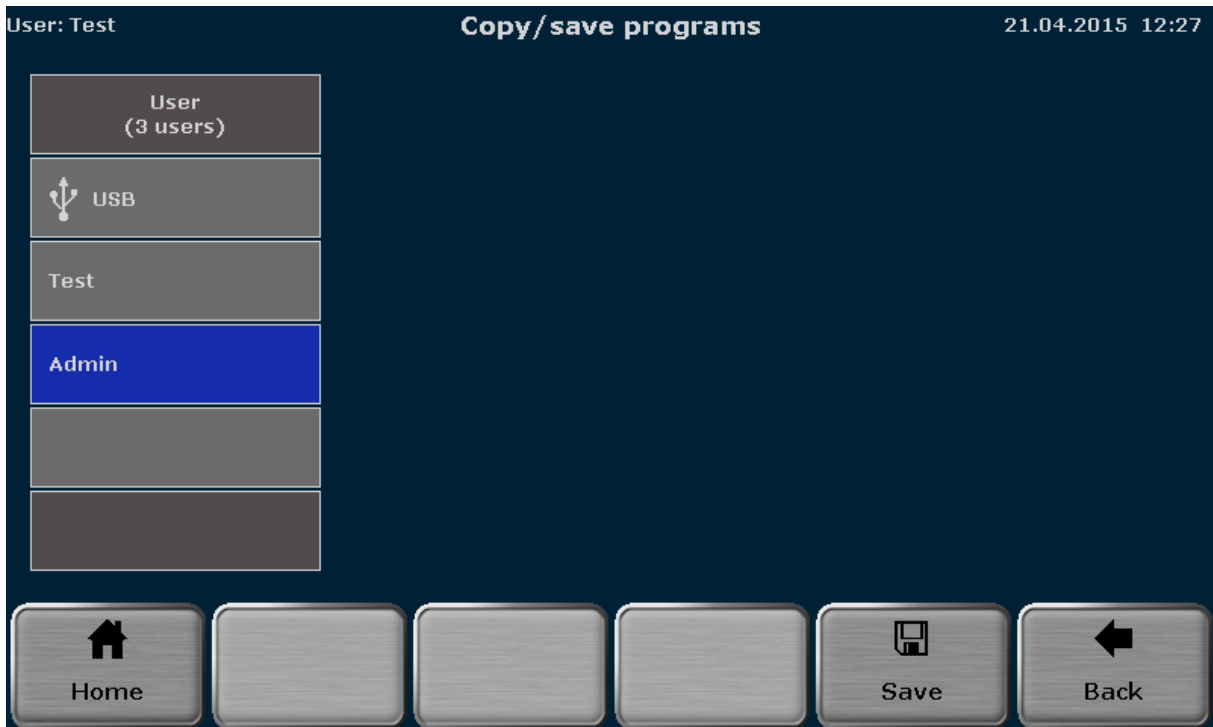
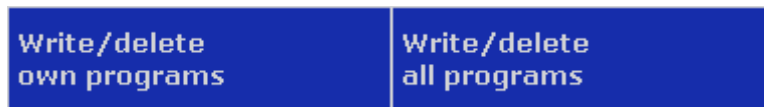


Figure 28 Biometra TRIO program overview screen

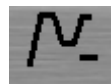
9.4 Delete program

Note: Own programs can only be deleted by users with the right **“Write/delete own programs”** activated. Programs of others can be deleted with the right **“Write/delete all programs”** activated (see chapter 12.2.2).



To delete a program, touch the corresponding buttons to select a user directory and a stored program in the Biometra TRIO program overview screen (see Figure 24). If the number of user directories or programs is higher than can be maximal displayed in the list, the buttons above and below the user and program list can be used to scroll up and down. To delete the selected program press

Delete



Confirm the security query to delete the program.

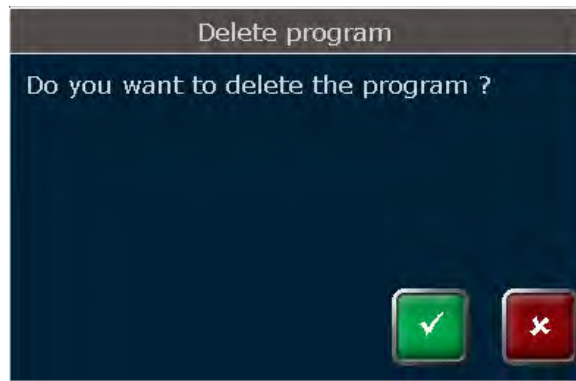
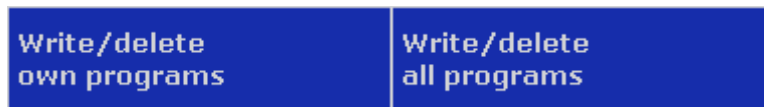


Figure 29 Biometra TRIO security query

Note: If a program is deleted, all other following programs in the user directory will be moved to a lower storage place. The storage place number for these programs will decrease by -1.

9.5 Delete all programs

Note: Own programs can only be deleted by users with the right **“Write/delete own programs”** activated. Programs of others can be deleted with the right **“Write/delete all programs”** activated (see chapter 12.2.2).



To delete all programs, touch the corresponding button to select a user directory in the Biometra TRIO program overview screen (see Figure 24). If the number of user directories is higher than can be maximal displayed in the list, the buttons above and below the user can be used to scroll up and down. To delete all programs in the user directory press



Confirm the security query to delete all programs.

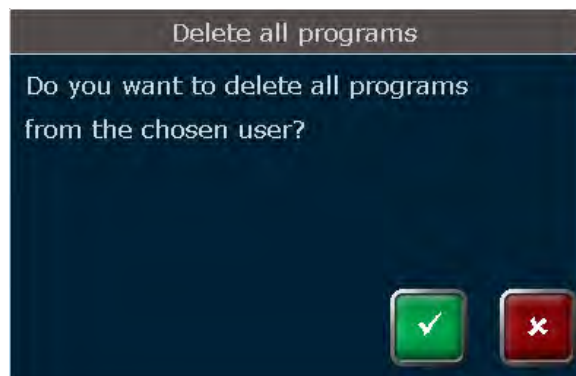
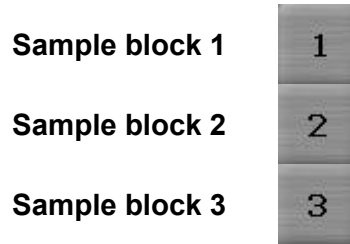


Figure 30 Biometra TRIO security query

10 Running, Pausing, Continuing and Stopping programs

To switch between the view for the sample blocks during run press



in the corresponding screen.

10.1 Display during run

After a program is started (see chapter 9.1), the Biometra TRIO software can display the running program in spreadsheet or graphical view. Use the buttons

Graphical view



Spreadsheet view



to toggle between both modes.

Graphical view



Spreadsheet view



Figure 31 Biometra TRIO run screen in graphical view and spreadsheet view.

The following parameters are summarized in the headline table:

- Program Name**
- Estimated End**
- Remaining Time**
- Sample Block Temperature**
- Lid Temperature**
- Step Number**

If the heated lid is set to preheating **ON** (see chapter 8.5), during the preheating of the heated lid the word “Preheating” will be displayed in the field **Step** in the headline table. After the heated lid has reached the target temperature and the 40s equilibration phase has passed, the first program step starts. During program run, the current step is highlighted in yellow in **Graphical view** and **Spreadsheet view**. In graphical view, each step is divided in heating phase and holding time and both parts are highlighted separately. Time and temperature increments and ramping rate adjustments cannot be shown in **Graphical view**. Instead the symbols ΔT, Δt or ! are displayed (see chapter 8.1).

10.2 Pause Program

To pause a program press



in the Biometra TRIO run screen (see Figure 31). The word “Pause” is shown and in the field remaining run time the pause time is counted.

Spreadsheet view

Graphical view

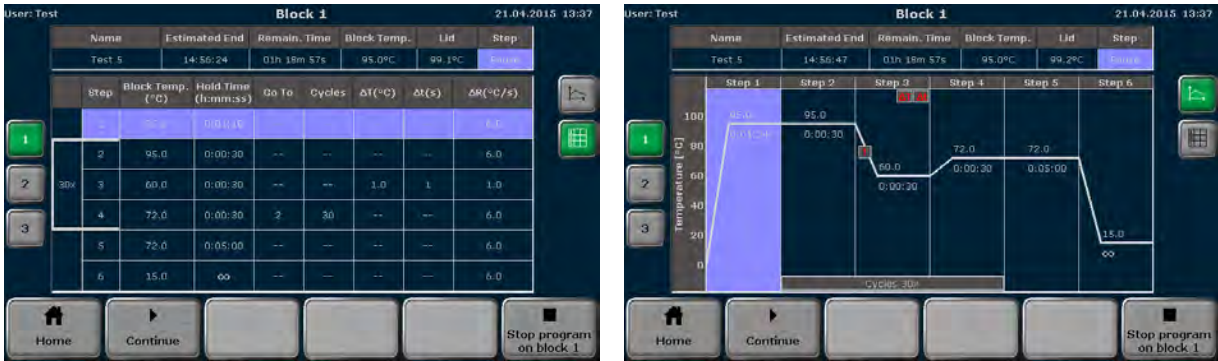


Figure 32 Biometra TRIO run screen during program pause

Note: To press the pause button requires manual interaction. The Biometra TRIO software also allows to program a pause step (see chapter 8.6), then the pause step is automatically started by the instrument.

Note For programs including a TOS step (see chapter 8.6.5) all blocks are paused simultaneously.

Note: If the instrument is switched off during a pause step, this will be recognized as a power failure (a referring error message is written to the Run-Logfile). For example, if the last step in a program is a pause, the program has to be stopped by pressing the **Stop** button (see chapter 10.5) before switching off the instrument.

10.3 Continue Program

To continue a paused program press

Continue



in the Biometra TRIO run screen during program pause (see Figure 32). The Biometra TRIO Thermal Cycler will continue the program exactly from the point when the pause was initiated.

Note For programs including a TOS step (see chapter 8.6.5) the program is continued on all blocks simultaneously.

10.4 Skip Step

To skip a step program press

Skip



in the Biometra TRIO run screen (see Figure 31). The Biometra TRIO Thermal Cycler will immediately continue with the next program step and skips the current step.

10.5 Stop Program

Note: Programs can only be stopped by users with the right “**Start/stop programs**” activated (see chapter 12.2.2).

Start/stop programs

To stop a program press

Stop program on block x



in the Biometra TRIO run screen (see Figure 31). If two or more sample blocks are running the Biometra TRIO thermal cycler offers the option to stop multiple sample blocks simultaneously. Press

Choose block stop program



in the Biometra TRIO run screen (see Figure 31) and activate the corresponding check boxes to select the sample blocks to stop.

* the button "choose block stop program" for multiblock selection is only available if more than one sample block is running.



Figure 33 Biometra TRIO query for multiblock selection

Confirm the block selection and the following security query to stop the running program(s).

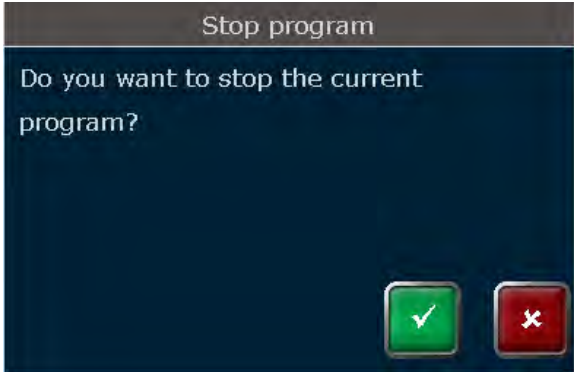


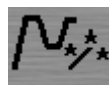
Figure 34 Biometra TRIO security query

The Biometra TRIO Thermal Cycler will immediately stop the running sample block(s).

11 Protocol Wizard

The protocol wizard is a helpful tool for automatic creation of PCR programs and primer annealing temperature calculation. To start the protocol wizard press

Protocol Wizard



in the Biometra TRIO home screen (see chapter 7.4).

11.1 Select Polymerase

The protocol wizard uses polymerase-specific program templates to create PCR programs. Program templates for up to 8 polymerases can be stored by the Biometra TRIO software.

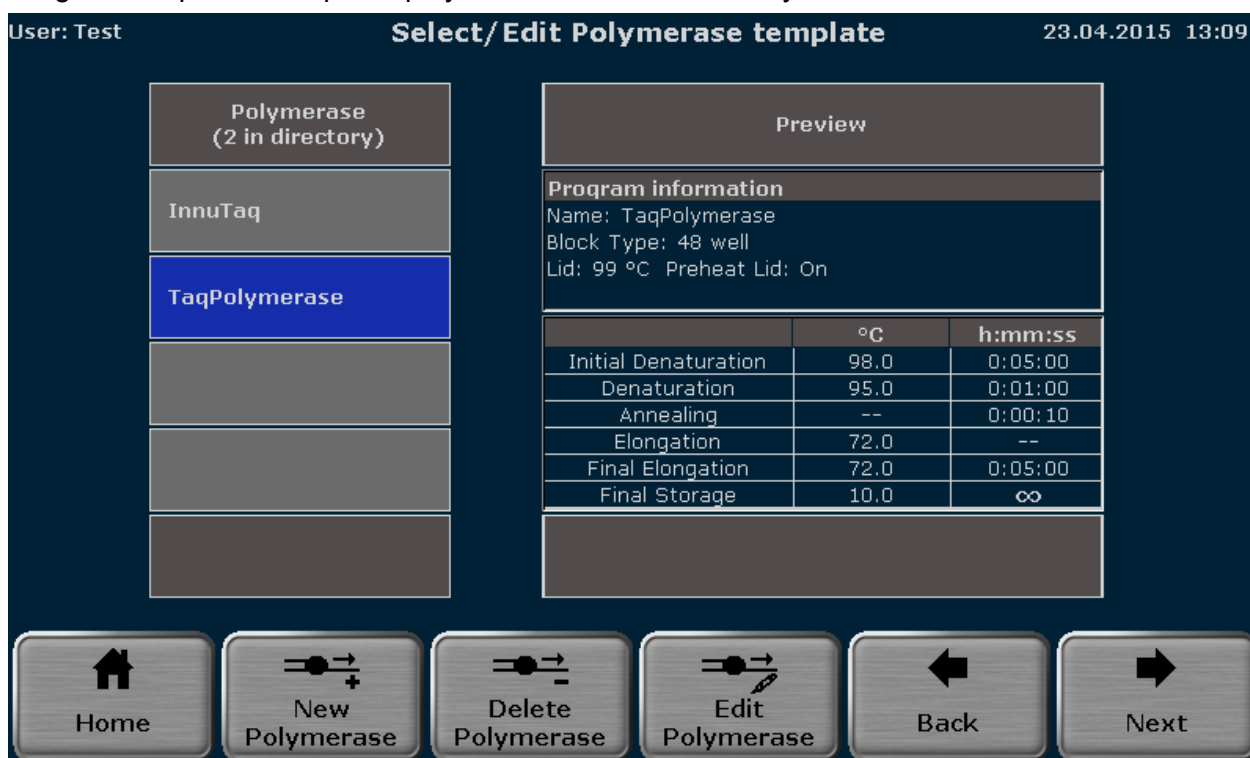


Figure 35 Biometra TRIO polymerase selection screen

Pre-installed in the software is the program template for Analytik Jena Innupure polymerases. To create a PCR program select a polymerase from the list and press

Next



In the next screen (see chapter 11.3) enter values for the requested parameters to continue with the automatic program creation.

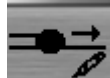
11.2 Enter New Polymerase or Edit Polymerase

To enter program templates for new polymerases or to edit preset program templates for polymerases press the corresponding button:

New Polymerase



Edit Polymerase



In the next screen (see figure Figure 36) touch the corresponding input boxes to enter a name for the polymerase and set parameters for all requested program steps. With two exceptions for most program steps the temperature and the holdtime must be defined:

1. The temperature for the annealing step depends on the calculated primer melting temperature (T_m -value) and has to be set individually for each primer pair (see chapter 11.4).
2. The holdtime for the elongation step becomes calculated by the expected product length (see chapter 11.4).

The corresponding input boxes for the annealing temperature and elongation time are shown in grey and cannot be activated.

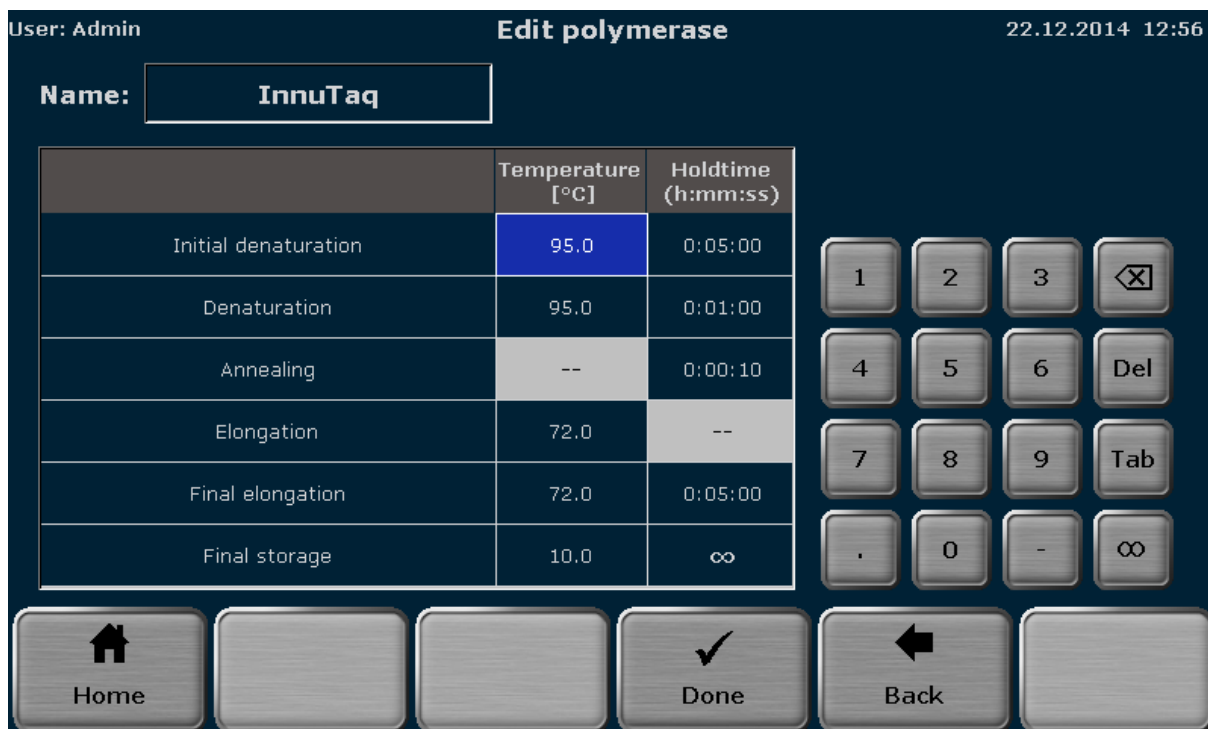


Figure 36 Biometra TRIO screen for editing program templates for polymerases

After creating the program template for a new polymerase or editing the program template of a polymerase press

Done



and in the next screen (see Figure 37) touch the corresponding button to select a storage place for the program template (polymerase). If the number of polymerases is higher than

can be maximal displayed in the list, the buttons above and below the polymerase list can be used to scroll up and down.

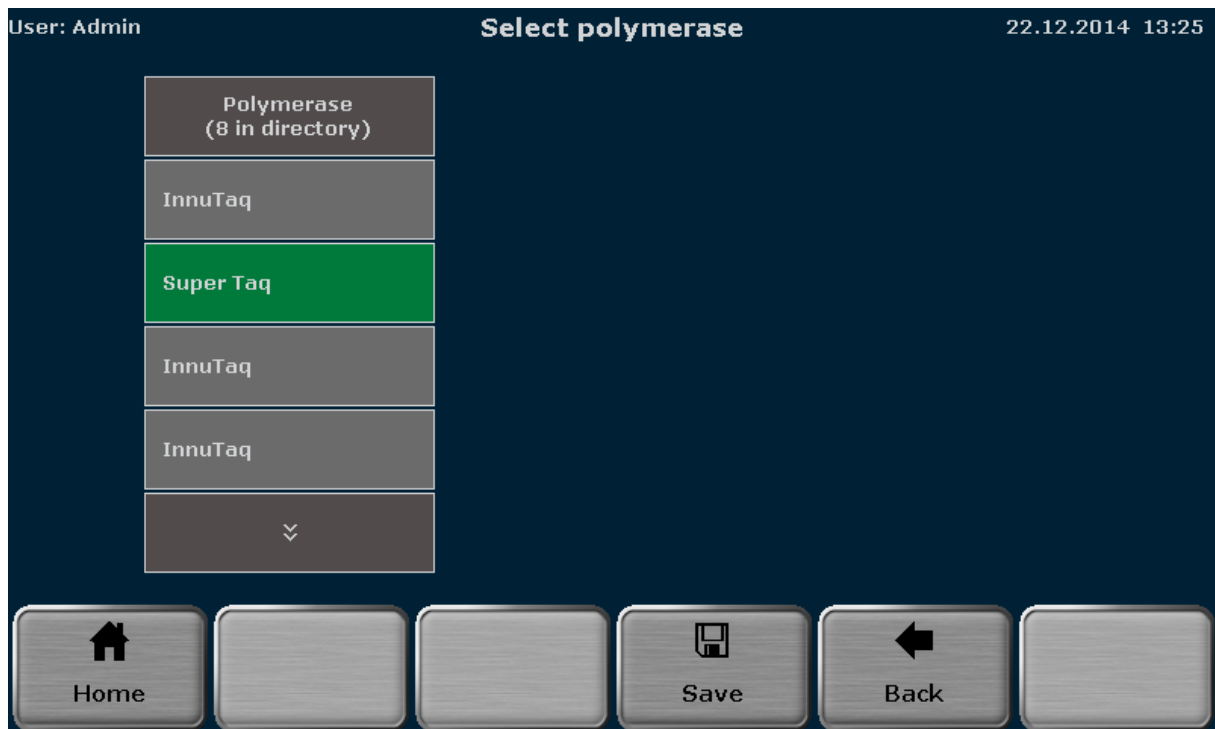


Figure 37 Biometra TRIO for saving program templates for polymerases

Note: If a new or edited polymerase is to be saved, the first free storage place in the polymerase list is pre-selected. However, the storage place can be modified by using the cursor key function above and below the list.

To save the new or edited polymerase press



Note: If the selected storage place is already in use, the stored polymerase will be overwritten. Confirm the security query to overwrite the existing polymerase:

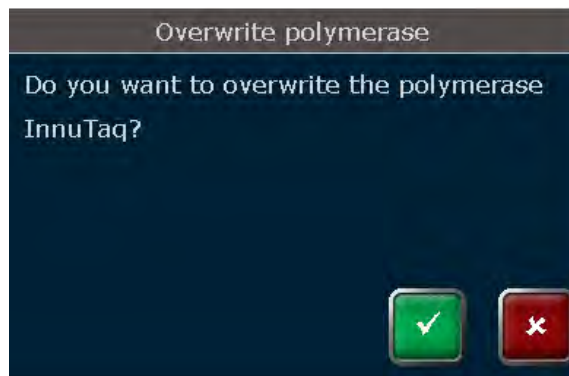


Figure 38 Biometra TRIO security query

11.3 Delete Polymerase

To delete a polymerase press

Delete Polymerase



Note: The Template “InnuTaq” cannot be deleted.

11.4 Protocol Wizard parameter screen

For automatic calculation of PCR programs in the protocol wizard parameter screen (see Figure 39) some parameters have to be set by the user:

1. Touch the corresponding checkbox to select between two step and three step programs.
Note: The protocol wizard offers to create two step or three step PCR programs. A three step program consists of separated steps for denaturation, annealing and extension. In a two step program the annealing and extension step are combined to one single step. In consequence for a three step protocol different temperatures for the annealing and extension step are used and for a two step program the primer annealing temperature T_a (see below) is used for the annealing and extension step.
2. Enter the PCR product length
Note: The product length is used by the software to calculate the elongation time. The software uses the following equation:

$$\text{Elongation time [s]} = \text{product length [bases]} / 1000 \text{ [bases]} * 60\text{s}$$
 For very short products the minimum elongation time is set to 6s.
3. Enter the primer annealing temperature
Note: In general the annealing temperature (T_a) should be set approximately 5°C below the calculated mean primer pair melting temperature (T_m). The theoretical T_a -value can also be calculated by the Biometra TRIO protocol wizard (see chapter 11.5).
4. Enter the number of cycles

To set the parameters touch the corresponding field in the protocol wizard parameter screen and enter the desired values.



Figure 39 Biometra TRIO protocol wizard parameter screen

After setting all program parameters press



The next screen lists the all steps of the program that will be created by the software:

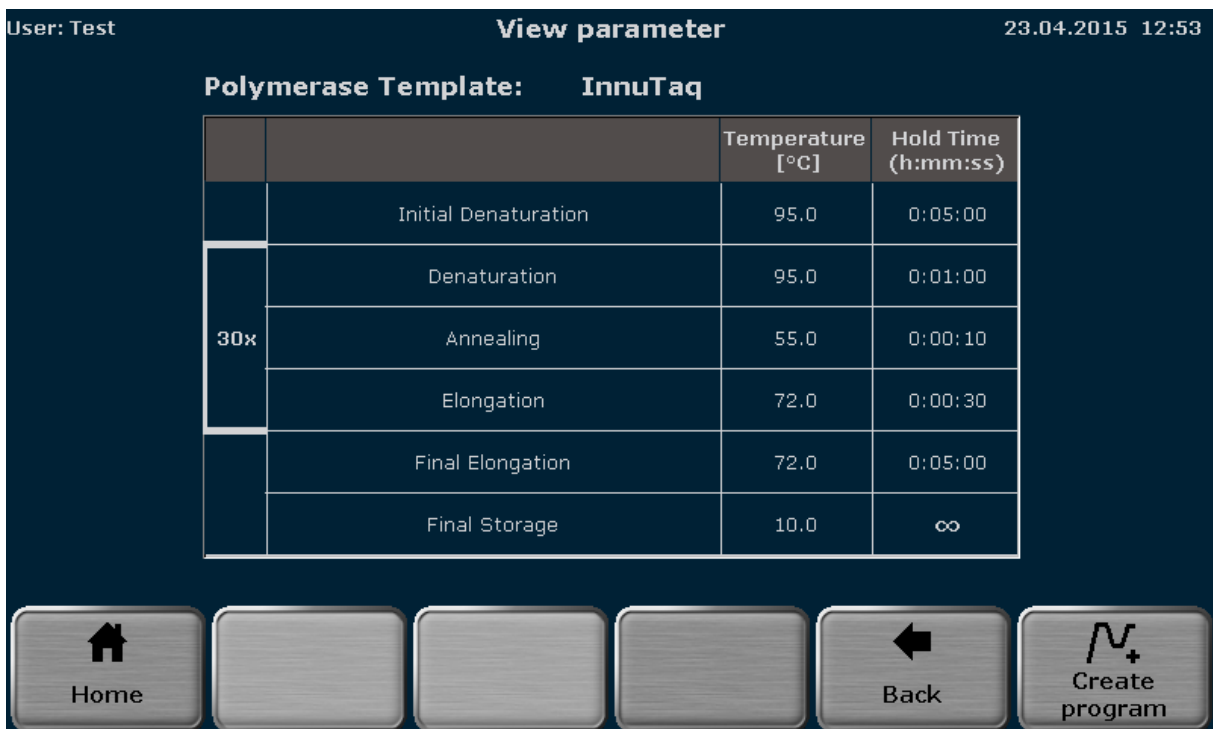
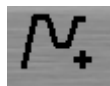


Figure 40 Biometra TRIO protocol wizard view parameter screen

Press

Create program



to enter the Biometra TRIO programming screen (see chapter 8). In the programming screen the program is displayed in graphical or spreadsheet mode and can be edited. To save the program created by the protocol wizard follow the instructions in chapter 8.9.

Note: Programs have to be saved before they can be started. If the program has been created but should not be saved confirm the following security query to discard the program:

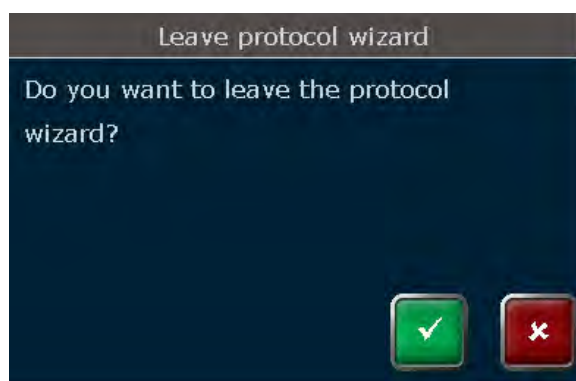


Figure 41 Biometra TRIO security query

11.5 Primer Annealing Temperature Calculator

Based on the primer sequences the Biometra TRIO protocol wizard can calculate the theoretical primer annealing temperature (T_a). Press button

Calc T_a



in the Biometra TRIO protocol wizard parameter screen (see Figure 39) to access the primer annealing temperature calculator screen (see Figure 42). In the annealing temperature calculator screen enter the sequences for the forward and reverse primer using the button below the input fields. The sequence length is counted by the software and displayed in brackets left to the input fields.

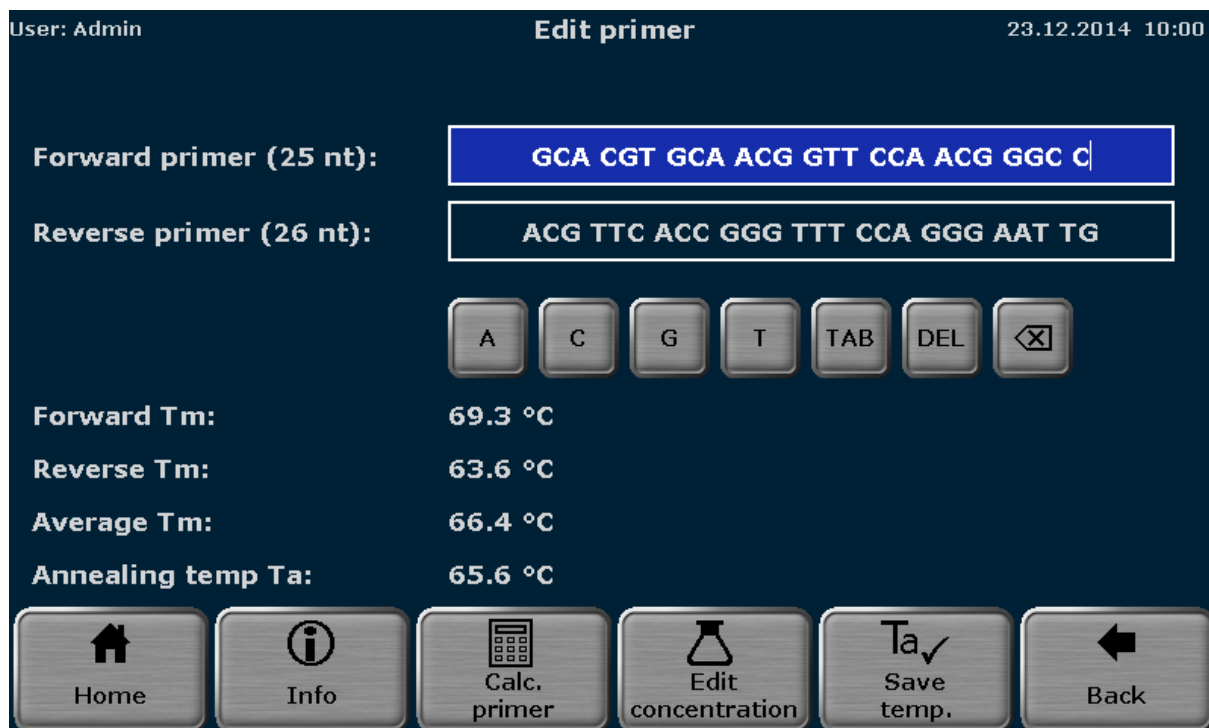


Figure 42 Biometra TRIO primer annealing temperature calculator screen

To adjust the primer or salt concentration press

Edit concentration



In the next screen the primer concentration, salt concentration and magnesium concentration can be adjusted. Touch the corresponding input field and enter values using the keypad. The concentrations have the following default values and ranges:

Function	Default value	Range
Primer concentration	200 nM	0-2.000 nM
Salt concentration	50 mM	0-200 mM
Magnesium concentration	0 mM	0-200 mM

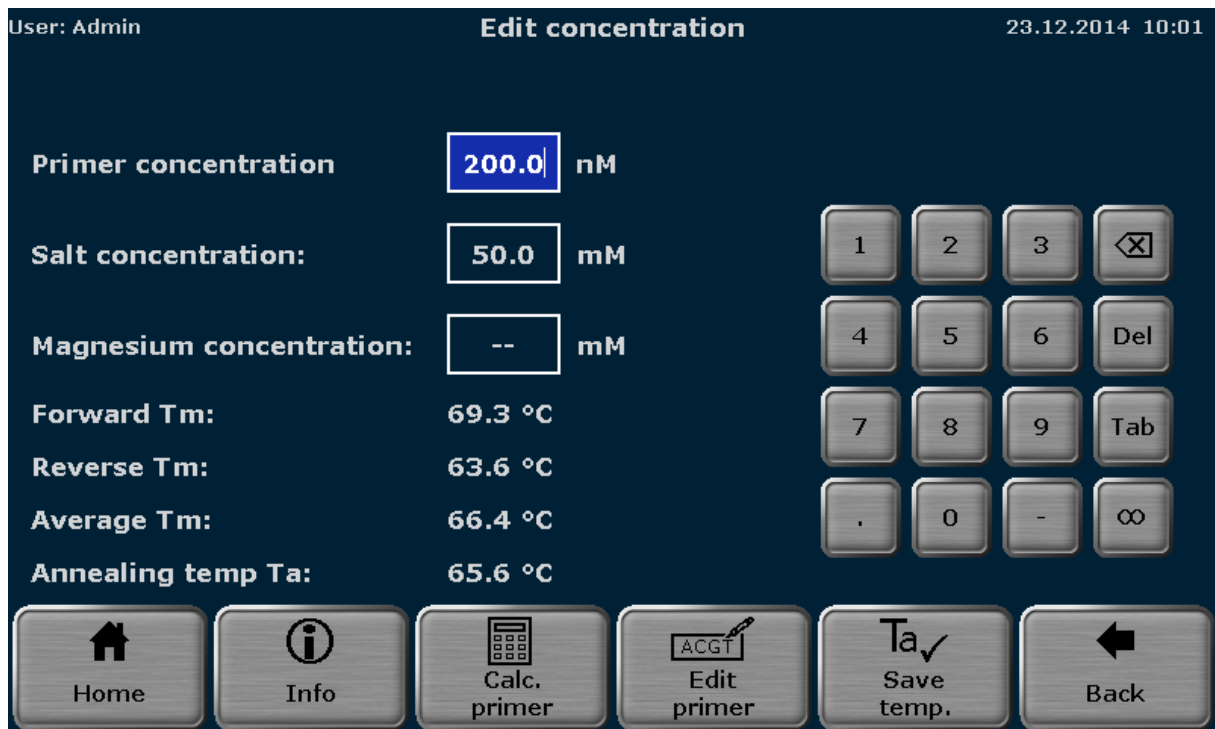


Figure 43 Biometra TRIO screen for primer concentration adjustment

Press

Edit primer



to toggle to the primer annealing temperature calculator screen (see Figure 42) or press

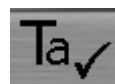
Calc. primer



to calculate the theoretical primer annealing temperature (T_a). The calculated values for the forward primer melting temperature (T_m), reverse primer melting temperature (T_m), mean primer melting temperature (T_m) and calculated annealing temperature (T_a) are displayed.

Press

Save temp.



to transfer the calculated primer annealing temperature to the protocol wizard (see Figure 39). The calculated T_a value will be displayed in the input box for the primer annealing temperature.

Press

Info



to get information on the algorithms used for annealing temperature calculations.

For short sequences of 14 nucleotides or less the software calculates the primer annealing temperature (T_a) based on the primer T_m values as follows:

1. For each primer the melting temperature is calculated using the following equation according to Wallace et al.¹:

$$T_m = ((w*A + x*T)*2) + ((y*G + z*C)*4)$$

where w, x, y and z are the number of the bases A, T, G and C in the primer sequence, respectively.

2. The average T_m value is calculated from the T_m values for both primer sequences
3. The annealing temperature T_a is calculated based on the average T_m value by applying the following rules:
 - If the difference between the primer T_m values is 4°C or less, the T_a is calculated as the average T_m value minus 5°C
 - If the difference between the primer T_m values is higher than 4°C, the T_a is calculated as the lower T_m value plus 2°C.

For long primer sequences of 15 nucleotides or more the software calculates the primer annealing temperature (T_a) based on the nearest neighbor method according to SantaLucia² and von Ahsen et al.³.

¹ Wallace et al., Nucleic Acids Res. 6, 3543, 1979

² SantaLucia, Proc Natl Acad Sci U S A. 1998 Feb 17;95(4):1460-5

³ von Ahsen et al., Clin Chem. 1999 Dec;45(12):2094-101

12 Tools

The Biometra TRIO software provides the following tools:

Settings	Define basic instrument settings	See chapter 12.1
User Management	Tool for the user management	See chapter 12.2
Documentation	Access to stored log files	See chapter 12.3
Extended Self-Test	Execute extended self-test	See chapter 12.4
Service Info File (SINF)	Create Service Info File	See chapter 12.5
Backup	Saves all folders, programs, users and user settings to USB	See chapter 12.6
Cycler Info	Provides general information on the instrument	See chapter 12.7
Contact	Biometra contact data	See chapter 12.8



Figure 44 Biometra TRIO tools screen

Press the corresponding button to access the desired function.

12.1 Settings

Note: System settings can only be modified by users with the right “**System configuration**” activated (see chapter 12.2.2). The system configuration affects all functions listed below expect of the user management.



The Biometra TRIO software offers the following settings tools:

Date and Time	Set date and time	See chapter 12.1.1
Automatic user logout	Switch automatic user logout on or off and time settings	See chapter 12.1.2

Beeper	Switch beeper on or off	See chapter 12.1.3
Network	Define network settings or list network users	See chapter 12.1.4
Display Brightness	Adjust display contrast	See chapter 12.1.5
Touch Screen Calibration	Calibrate touch screen	See chapter 12.1.6
Factory Settings	Reset the settings	See chapter 12.1.7



Figure 45 Biometra TRIO settings screen

Press the corresponding button to access the desired function.

12.1.1 Set Date and Time

Note: Date and time can only be edited by users with the right “**System configuration**” activated (see chapter 12.2.2).

System configuration

The Biometra TRIO software allows the user to set time and date. The date is defined in day:month:year and the time in hours:minutes:seconds.

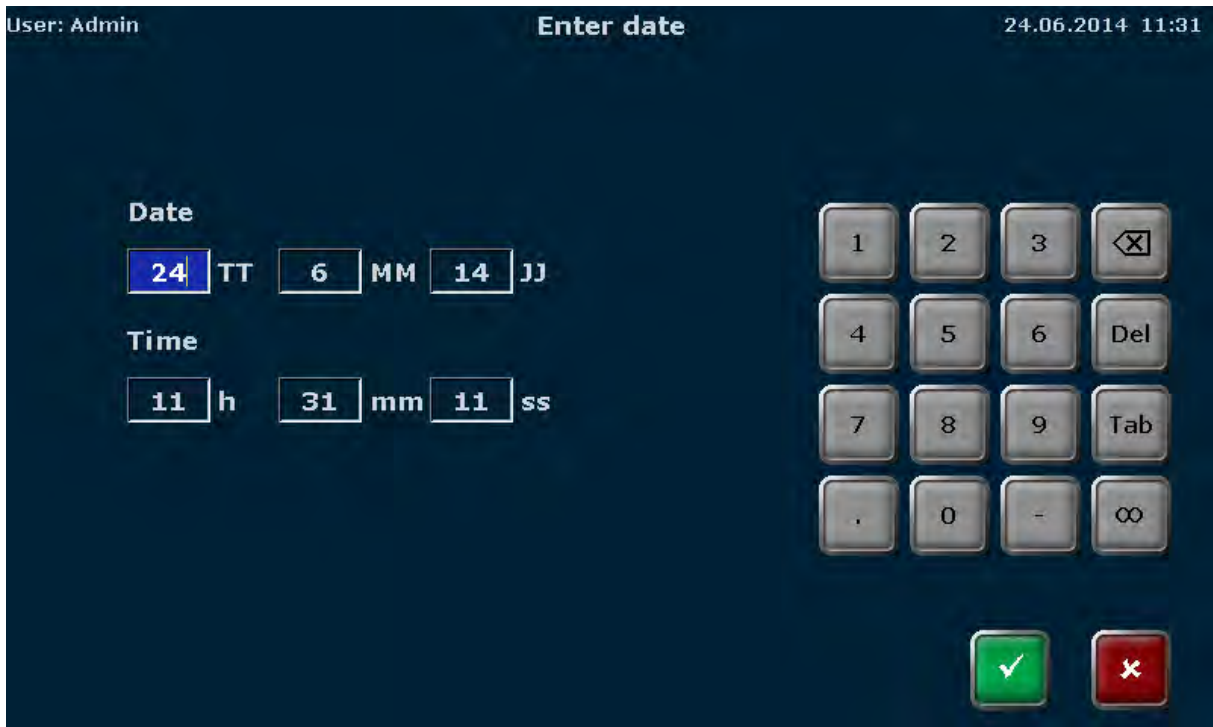


Figure 46 Biometra TRIO date and time screen

Activate the corresponding input box and use the keypad on the right side of the date and time screen to enter digits. Additionally the following keys might be helpful to fill out the input boxes:

Delete single character



Delete all characters

Jump to the next input box

Note: The following keys are inactive in the date and time screen and cannot be used:





12.1.2 Automatic user logout

Note: The automatic user logout can only be configured by users with the right “System configuration” activated (see chapter 12.2.2).



The Biometra TRIO Thermal Cycler features an automatic user logout. The automatic user logout can be switched ON or OFF by using the corresponding buttons:

Automatic user logout ON 
 Automatic user logout OFF 

If the automatic user logout is switched ON the time after that the user is automatically logged out can be set. You can enter values between 30 minutes and 60 minutes.

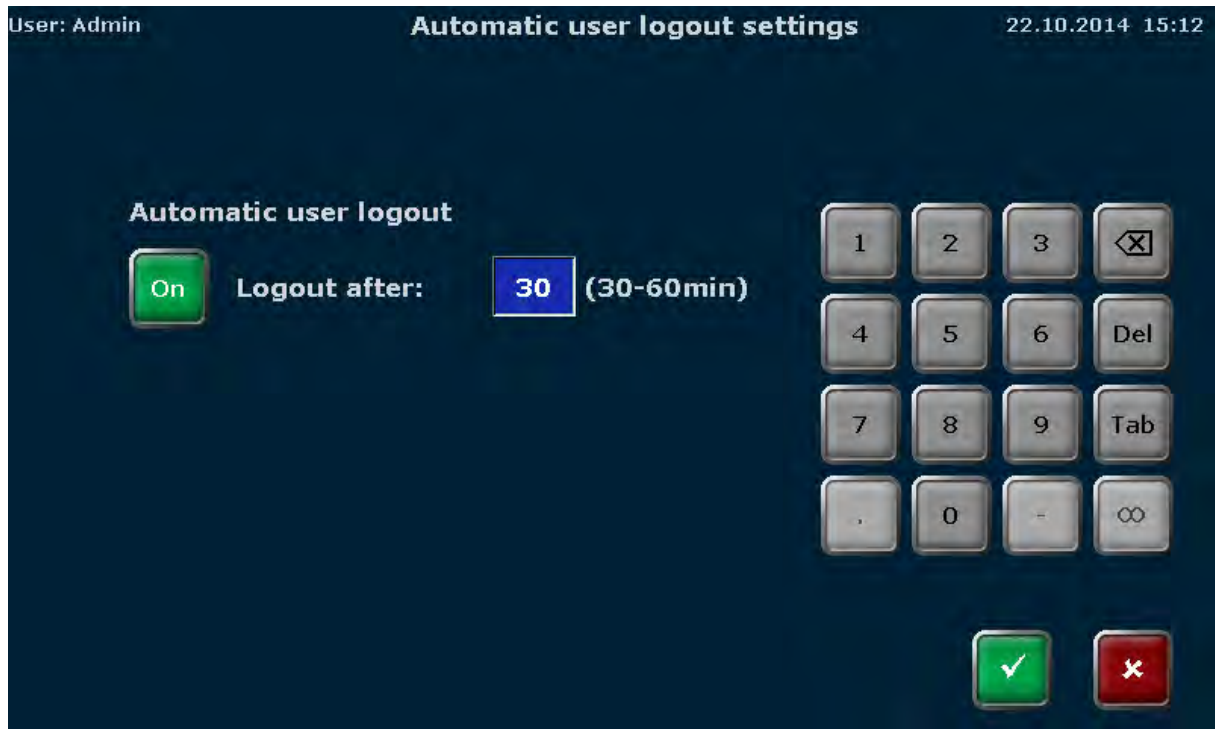
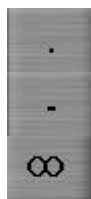


Figure 47 Biometra TRIO automatic user logout screen

Note: The standard setting is automatic user logout OFF.

Note: The following keys are inactive in the automatic user logout screen and cannot be used:



12.1.3 Configure Beeper

Note: The beeper can only be configured by users with the right “System configuration” activated (see chapter 12.2.2).

System configuration

The Biometra TRIO Thermal Cycler has a beeper and gives an acoustic signal if a PCR program is finished and the beeper is turned ON. The beeper can be switched ON or OFF by activating the corresponding check box.

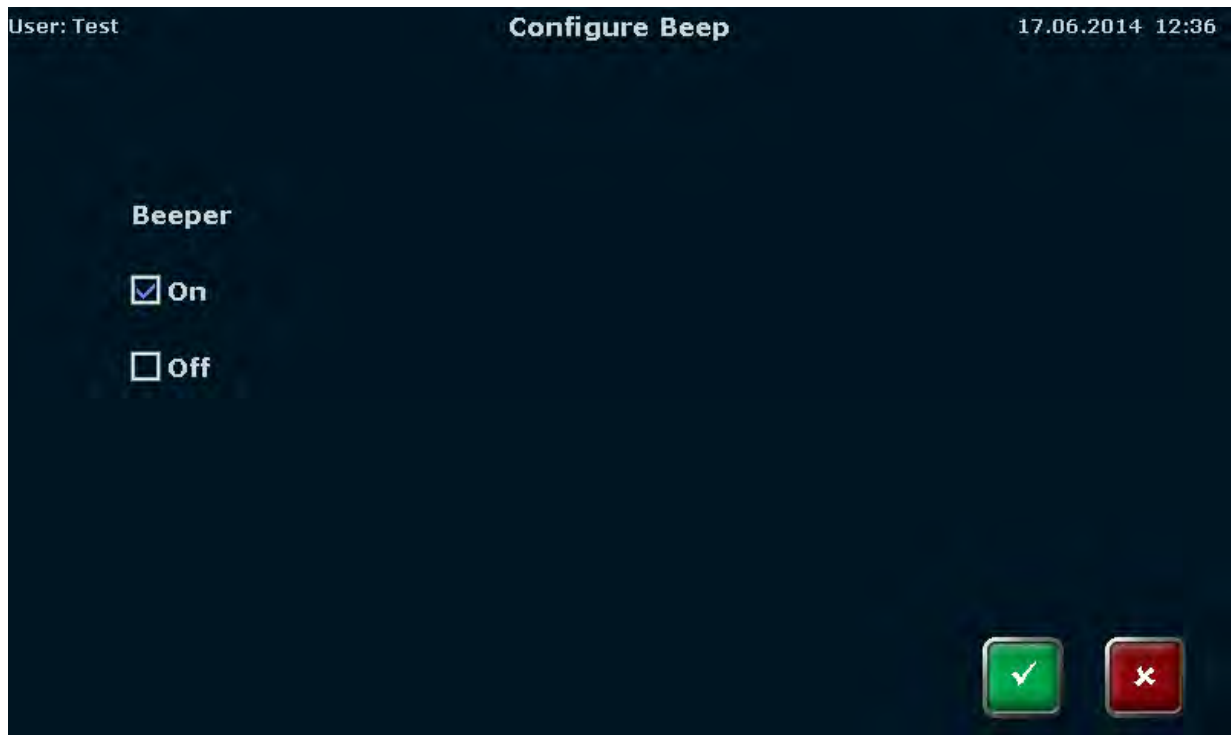


Figure 48 Biometra TRIO beeper configuration screen

12.1.4 Network

The menu item **Network** allows to configure the network settings and to show a list of network users. Press the corresponding button

Network Settings



Network User



to access a function.

12.1.4.1 Network settings

Note: The network settings can only be edited by users with the right “**System configuration**” activated (see chapter 12.2.2).

System configuration

The network settings allow to set a name for the thermal cycler used in the network. The default setting is the instruments serial number. Touch the input box and use the Biometra TRIO keyboard (see chapter 6.5.1) to enter a new name.

Note: The name can have a length up to 16 characters.

The Biometra TRIO Thermal Cycler can handle static and dynamic IP addresses. Activate the corresponding check box to choose between dynamic (DHCP) and static IP address. If DHCP is activated the instrument automatically receives the network configuration settings. If the check box static is activated you can enter the IP address, the subnet mask and the port by using the keypad on the right side of the screen.

The following keys might be helpful to fill out the input boxes:

Delete single character

Delete all characters

Jump to the next input box

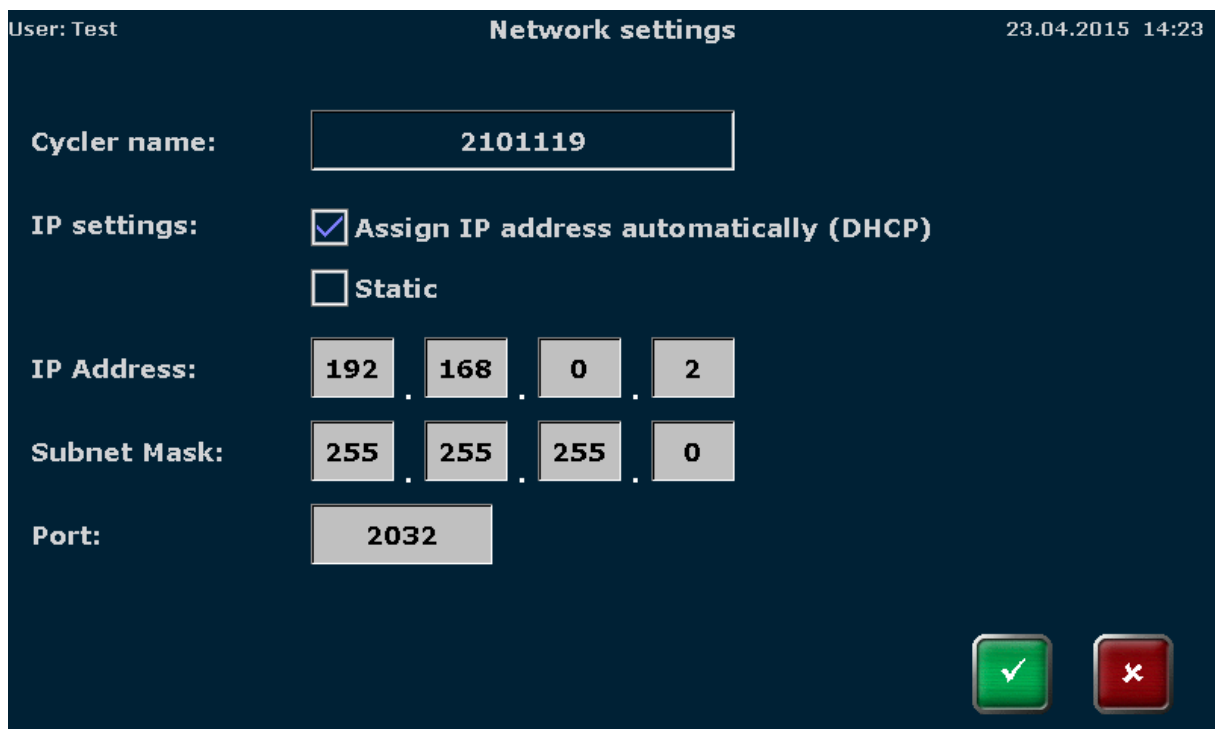


Figure 49 Biometra TRIO network settings screen

Note: The network cable must be at least performance class Cat 5e and the cable configuration has to be STP.

12.1.4.2 Network user

The software stores the network users and lists them in chronological order in a table. By the function it is possible to check if only authorized persons have network access to the instrument.

12.1.5 Display brightness

Note: The display brightness can only be edited by users with the right “**System configuration**” activated (see chapter 12.2.2).

System configuration

The display brightness can be adjusted to the local light conditions. To adjust the brightness use the corresponding buttons:

Darker



Brighter



or use the horizontal slider.

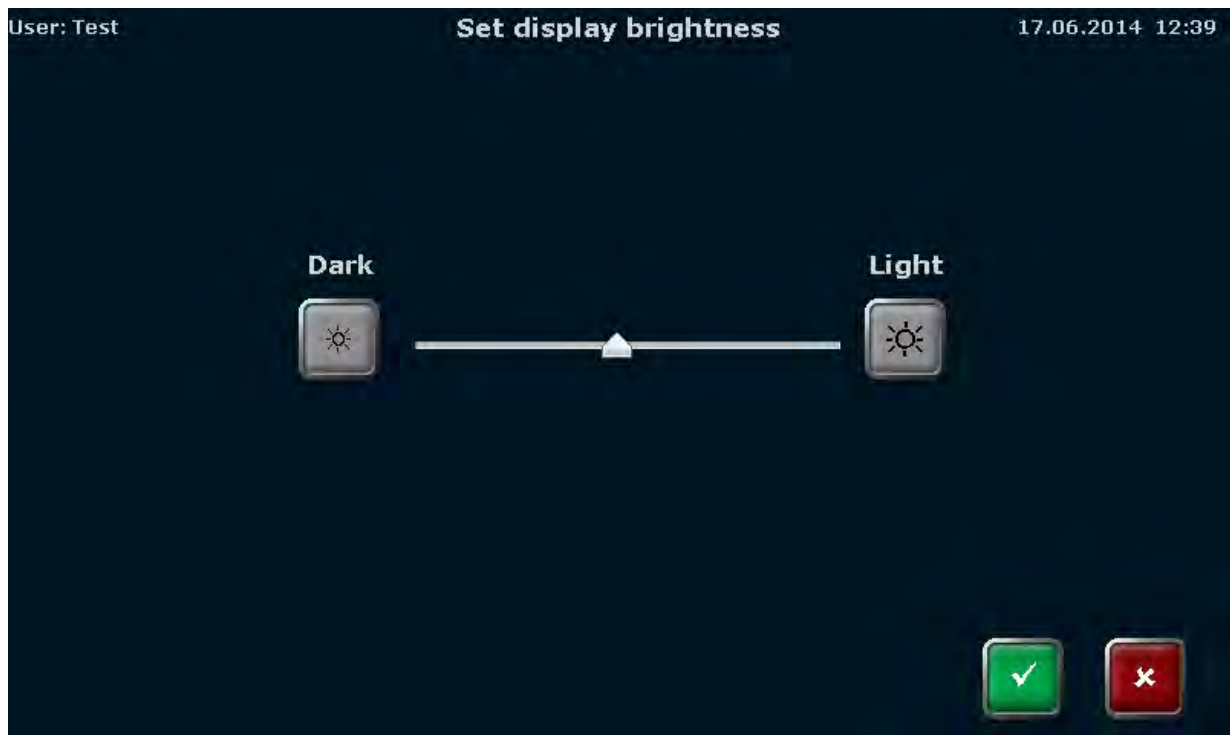


Figure 50 Biometra TRIO display brightness settings screen

12.1.6 Touch screen calibration

Note: The touch screen can only be calibrated by users with the right “**System configuration**” activated (see chapter 12.2.2).

System configuration

To calibrate the touch screen follow the instructions and touch the screen at the center of the displayed circle. The procedure is repeated three times at different positions of the display. If at the end of the procedure a small cross is shown within the circle the calibration is successful.

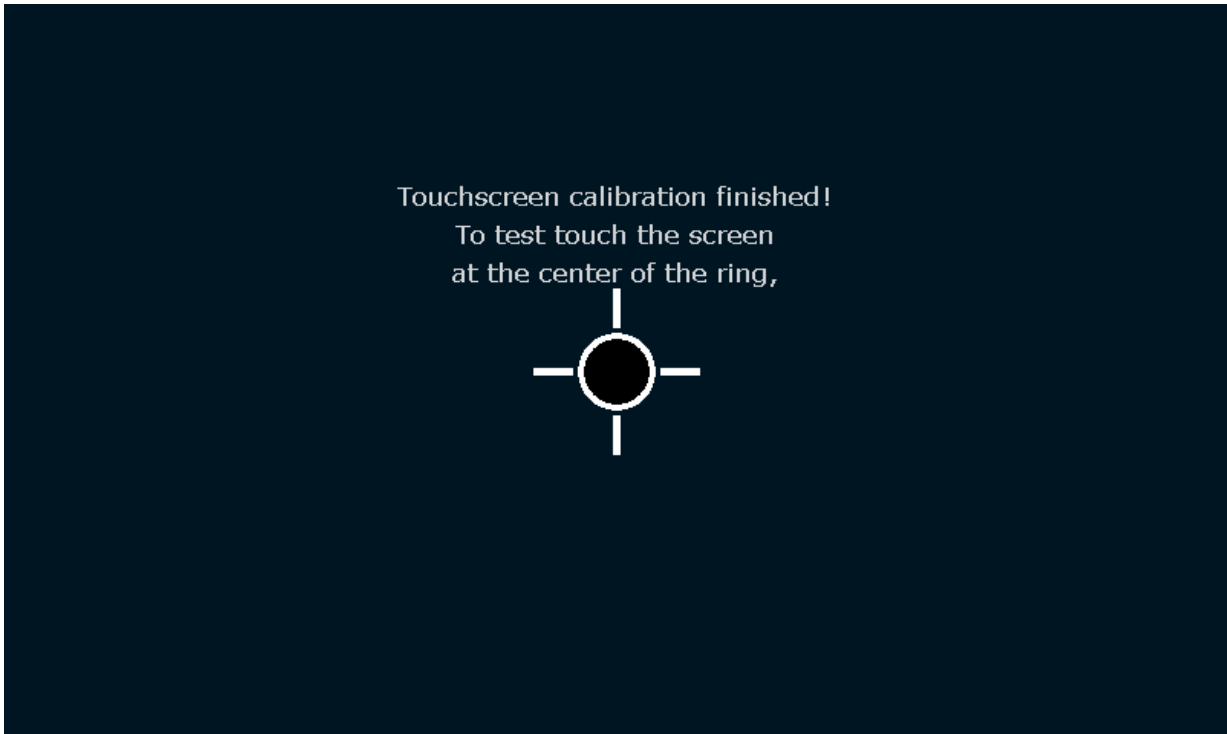
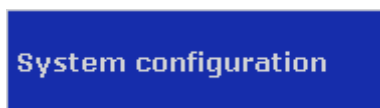


Figure 51 Biometra TRIO touch screen calibration screen

12.1.7 Factory settings

Note: The factory settings tool can only be executed by users with the right “**System configuration**” activated (see chapter 12.2.2).



Press the corresponding button and confirm the following security query:

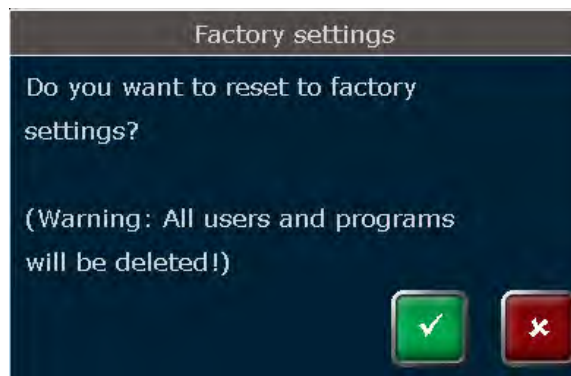


Figure 52 Biometra TRIO security query

By using the factory settings tool all user accounts and programs will be deleted!

Note: Use a backup file (see chapter 12.6) to restore the system.

12.2 User Management

The Biometra TRIO thermal cyclers manage up to 30 individual user accounts. The user management allows to create new users and to set rights for users. The user management is a complex tool. Please follow the general guidelines:

1. The factory setting for the administrator password is “Admin”. Please change the password after switching on user management to protect the system from unwanted modifications.
2. Use the backup tool to create backup files. Backup files can be used to restore a system or to synchronize the memory contents of Biometra TRIO thermal cyclers.
3. Do not switch off the user management if you are not absolutely sure. All rights settings will get lost and can only be restored if a backup file is available.

The user management can be switched on or off. By default the user management is switched off after delivery.

To set the user management on or off press

User Management



in the Biometra TRIO user management screen (see Figure 53).

Note: The button **User Management** is only available for the user group “Administrator”. Only an administrator can switch on or off the Biometra TRIO user management. Also with the user management switched off the administrator “Admin” has to log in.



Figure 53 Biometra TRIO user management screen

In the next screen activate the corresponding checkbox to configure the Biometra TRIO user management:

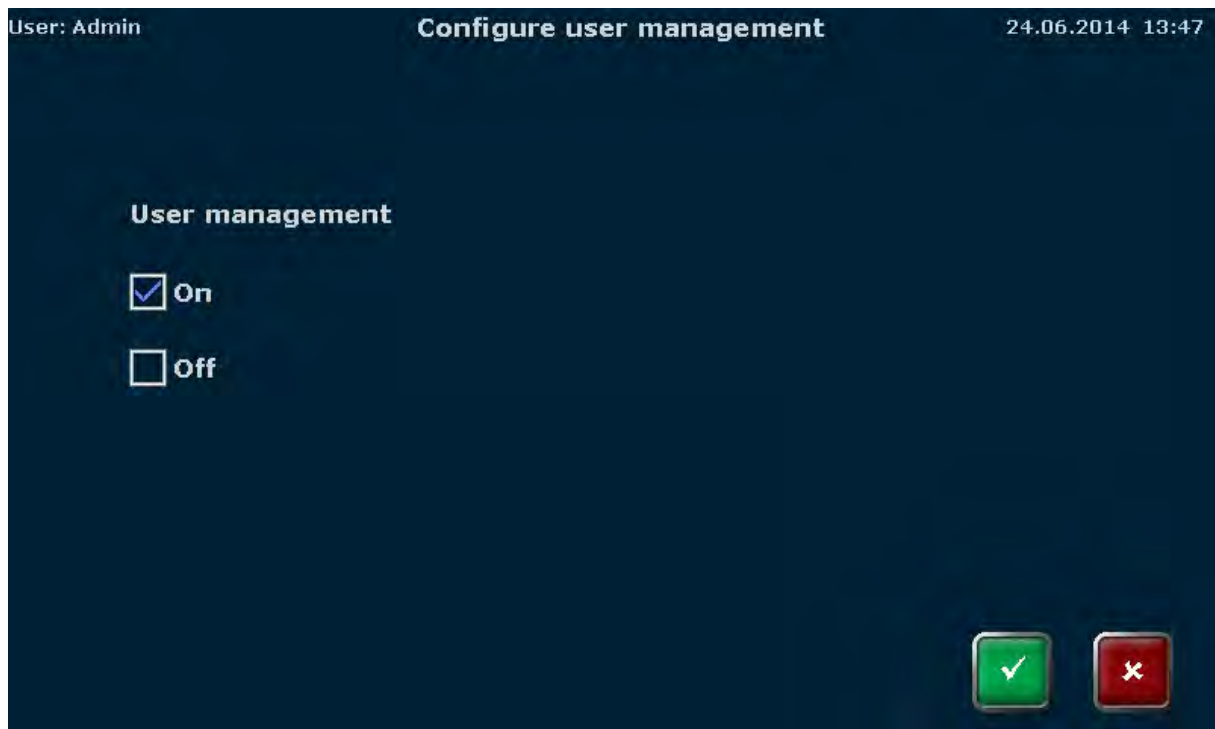


Figure 54 Biometra TRIO configure user management screen

If the user management is switched off, all new users have all rights and can access all software functions. With the user management switched on, the Biometra TRIO Thermal Cycler supports 3 different user groups with pre-defined rights:

	Administrator	User with generic rights	User with limited rights
System configuration	X		
Edit user	X		
Create user	X [#]	X [#]	
Delete user	X		
Write/delete all programs	X		
Write/delete own programs	X	X	
Read all programs	X	X	X
Start/stop programs	X	X	X

[#] an administrator can create users of all groups and modify their rights settings. A user with generic rights is only allowed to create users with generic or limited rights but no administrator. In addition, the user with generic rights cannot modify the rights settings of other users.

The settings for each user can be modified (see chapter 12.2.2) by the administrator and will be stored by the Biometra TRIO software.

Note: If the user management is switched off, user accounts and programs are not protected and can be modified or deleted by others.

Available user accounts are listed in the Biometra TRIO user management screen (see Figure 53) sorted by the date and time of the last log in and the user name, initials and group are shown. Depending on the settings a user can be grouped as

1. Administrator
2. User with generic rights
3. User with limited rights
4. User defined

If more than 8 user accounts are available you might need to scroll the table up or down to get see your desired user account (see Figure 53).

Scroll up



Scroll down



To select a user account press on the corresponding line in the table.

12.2.1 Create user

Note: New users can only be created by users with the right **Create User** activated (see chapter 12.2.2).



An administrator can create users of all groups and modify their rights settings. A user with generic rights is only allowed to create users with generic or limited rights but no administrator. In addition, the user with generic rights cannot modify the rights settings of other users.

To create a new user press

New User



In the next screen enter

- The user name (up to 13 characters or numbers)
- Initials (2 to 3 characters)
- Password (up to 13 characters, numbers or special characters)
- Repeat Password

using the Biometra TRIO keyboard (see chapter 6.5.1) and set the language by activating the corresponding check box.

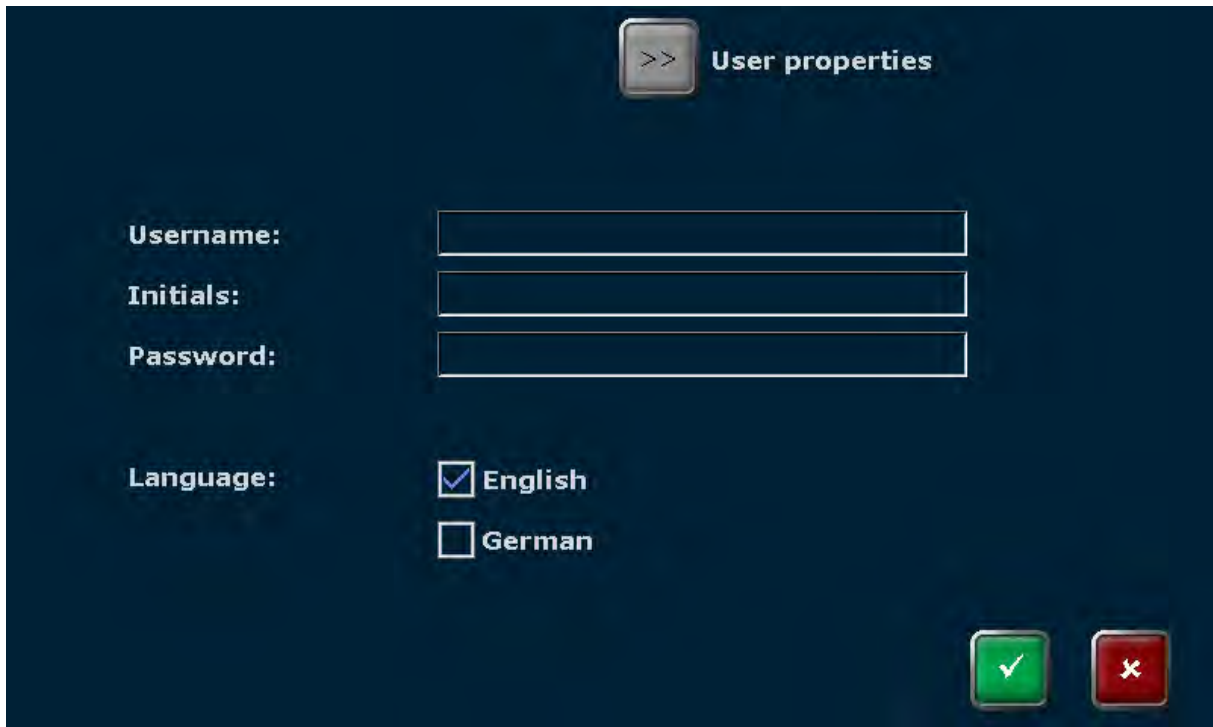


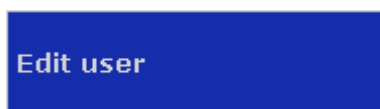
Figure 55 Biometra TRIO create user screen

Note: The password protection is optional. However, if no password is used, your account containing all of your programs might be modified or deleted by unauthorized persons.

Note: The language settings are user specific. For each user the language setting becomes saved by the instrument. The language setting can be changed during operation and the instrument does not be switched off and rebooted.

12.2.2 Edit user

Note: User rights can only be edited by users with the right “**Edit User**” activated (see chapter 12.2.2).



To edit the user properties select a user account in the user management screen (see chapter 12.2) and press the button



In the next screen the user data and the language settings can be changed.



Figure 56 Biometra TRIO user data screen

Note: For the administrator “Admin” the user name and initials “ADM” are fixed and cannot be modified. It is highly recommendable to change the password for the administrator „Admin“ after the first system start und to create a backup file. Directories, programs users and right settings can be restored from a backup file.

To modify the user rights press



In the next screen the user can be defined as

1. Administrator
2. User with generic rights
3. User with limited rights

For each user group templates defining the user rights are available. However, the pre-defined settings can be modified. You can activate or deactivate single rights. Activated rights are highlighted blue, deactivated rights are shown in grey. The settings for each user will be stored by the Biometra TRIO software and are shown in the Biometra TRIO user management screen (see chapter 12.2).



Figure 57 Biometra TRIO user properties screen

Right	Function
System configuration	Provides access to several system settings like date and time, screen saver, beeper, network settings, display brightness, touch screen calibration and factory settings
Edit user	Allows to modify the user name, user initials, password, language setting and user rights
Create user	Allows to create new user accounts. A user with generic rights can only create generic or limited users and is not allowed to modify the rights settings. An administrator is allowed to create all user types and to modify the rights settings for each user.
Delete user	Allows to delete existing user accounts
Write/delete all programs	The user can edit/copy and delete all programs of all users
Write/delete own programs	The user can edit/copy and delete own programs but not of other users
Read all programs	If inactivated the access is limited to own programs. Programs of others cannot be viewed (if a program cannot be viewed it also cannot be edited or started by the user).
Start/stop programs	The user can start or stop programs

Note: The settings for the administrator “Admin” cannot be changed. The administrator “Admin” always has all rights since at least one user needs to have all rights.

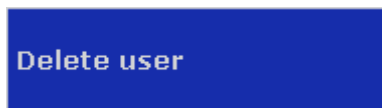
Press



to go back to the create user screen.

12.2.3 Delete user

Note: User accounts can only be deleted by users with the right “Delete User” activated (see chapter 12.2.2).



To delete a user select the user account in the user management screen (see chapter 12.2) and press the button



Note: The administrator “Admin” cannot be deleted since at least one user must be administrator of the Biometra TRIO Thermal Cycler.

Confirm the security query to delete the selected user.



Figure 58 Biometra TRIO security query

The user and all of his programs will be deleted.

12.3 Documentation

The Biometra TRIO software offers the following documentation tools:

Run-Logfile	Documentation of PCR runs	See chapter 12.3.1
Power On Logfile	Results of the latest initialisation test	See chapter 12.3.2
Self-Test Logfile	Results of extended self-tests	See chapter 12.3.3
Error Logfile	Lists all errors detected by the instrument	See chapter 12.3.4

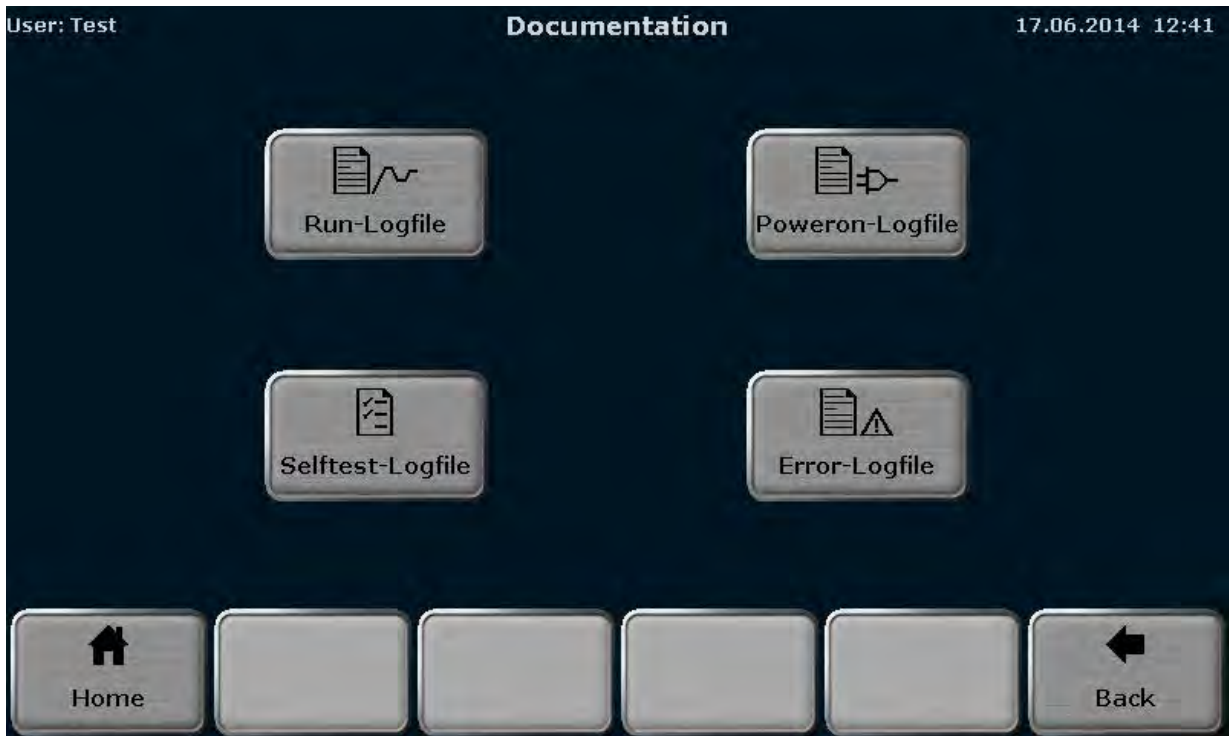


Figure 59 Biometra TRIO documentation screen

Press the corresponding button to access the desired function.

12.3.1 Run-Logfile

The Biometra TRIO Thermal Cycler lists up to 6 Run-Logfiles for the latest started programs.



Figure 60 Biometra TRIO Run-Logfile overview screen

The Run-Logfiles are sorted by date and time. The latest started program is shown in position 1 in the table. Additionally the user that started the program, the sample block number used to run the program, the program name, directory and number are shown.

To view the collected data select a Run-Logfile from the list and press button:

View Run-Logfile 

In the Run-Logfile screen the following information is listed:

- Run-Logfile name
- Log time and date
- Start and end time of the program run
- Date of the program run
- Name of the vendor
- Software version
- User initials
- Cyclor type
- Serial number Cyclor
- Block type
- Block serial number
- Block number
- Messages

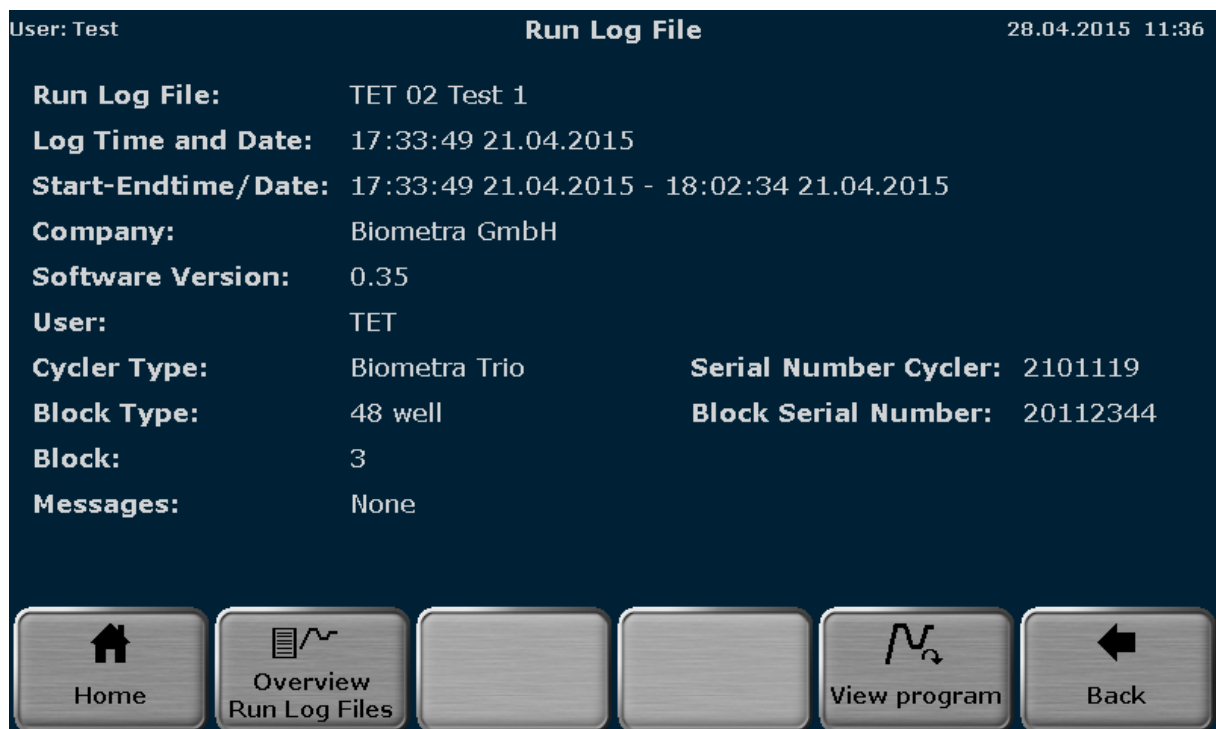


Figure 61 Biometra TRIO Run-Logfile screen

To view the corresponding program press button:

View Program 

The Biometra TRIO offers the option to show the program in graphical form or as spreadsheet (see chapter 8.1). To toggle between graphical and spreadsheet view use buttons:

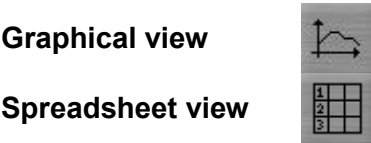
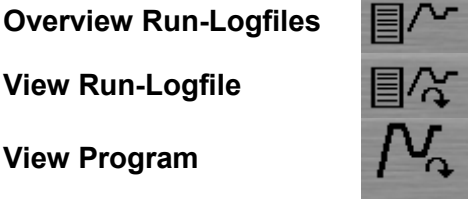


Figure 62 Biometra TRIO View program screen

Use the buttons



to toggle between the overview screen, view Run-Logfile screen and view program screen.

To export Run-Logfiles to USB press



or to export a specific Run-Logfile select a logfile from the list and press



in the Run-Logfile overview screen (see figure Figure 60). Use “Selected Run-Logfile to USB” to save single Run-Logfiles to USB or “All Run-Logfiles to USB” to save all Run-Logfiles to USB.

12.3.2 Power-On-Logfile

With each system start the Biometra TRIO Thermal Cycler performs an initial self-test and stores the results in a Power-On-Logfile.

In the Power-On-Logfile screen the following information is listed:

- Power-on by date and time
- Last Power-on by date and time
- Last Power-down by date and time
- Table with failure messages by number, date, time, error code and message

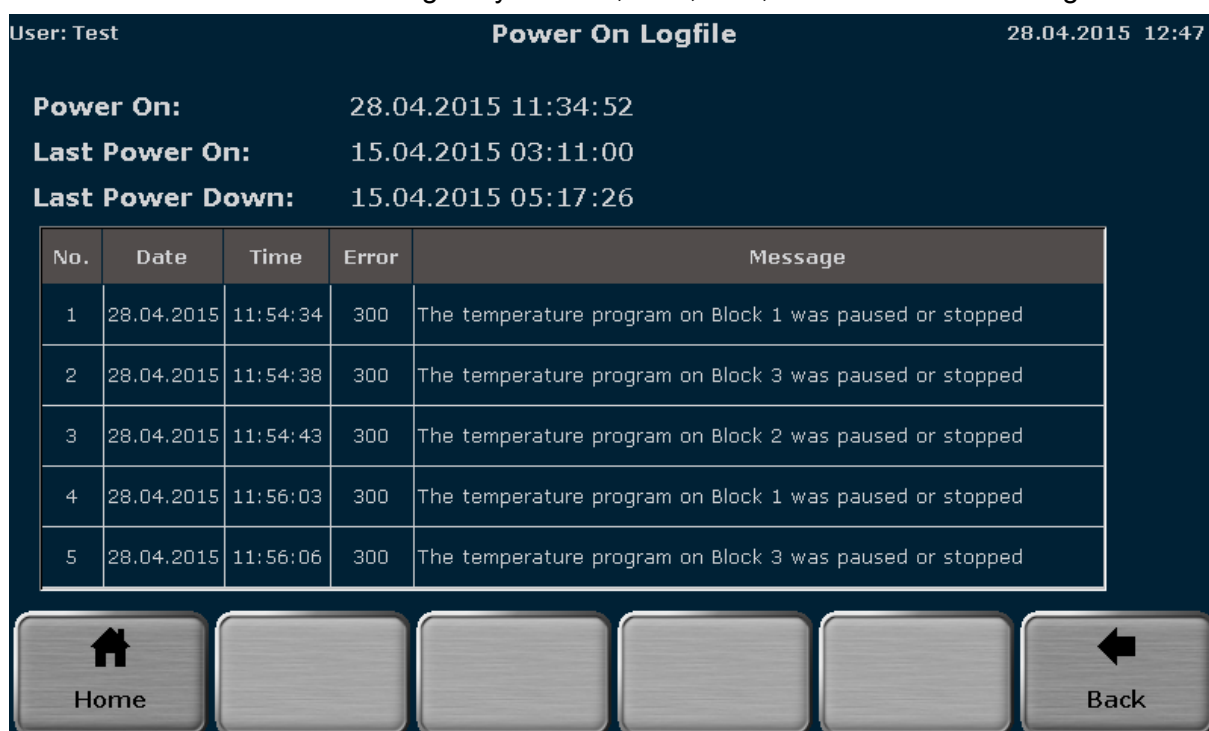


Figure 63 Biometra TRIO Power-On-Logfile screen

12.3.3 Extended Self-Test-Logfile

During the Extended Self-Test the Biometra TRIO Thermal Cycler checks several functions and components like cooler, thermal tracking, heating and cooling rate, refrigeration, heated lid and regulation (see chapter 12.4). Press on the corresponding button to see the results for a specific test summarized in tabular format.

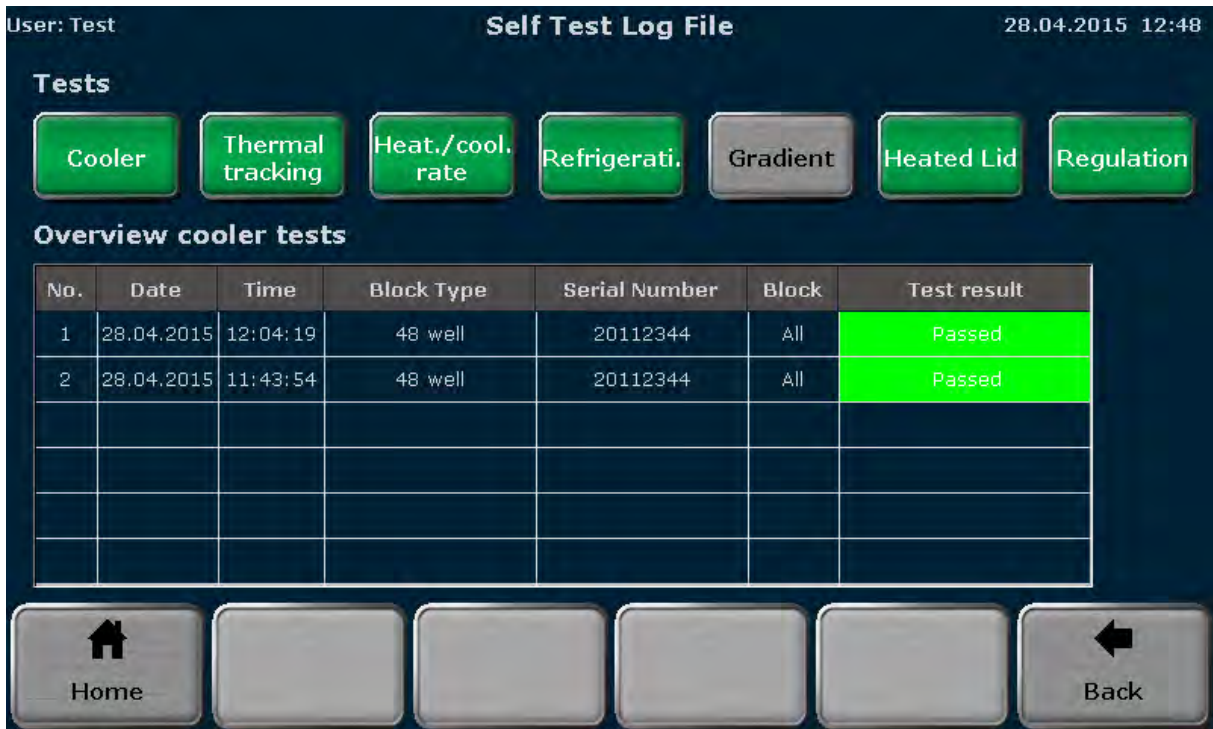


Figure 64 Biometra TRIO Extended Self-Test-Logfile screen

The test result can be “Passed” or “Failed” and is listed in the last row of each table for each test.

12.3.4 Error-Logfile

The Biometra TRIO Thermal Cycler records all errors by time and date. The latest error is shown in position in the Error Log table. Additionally the error code and error message are listed.

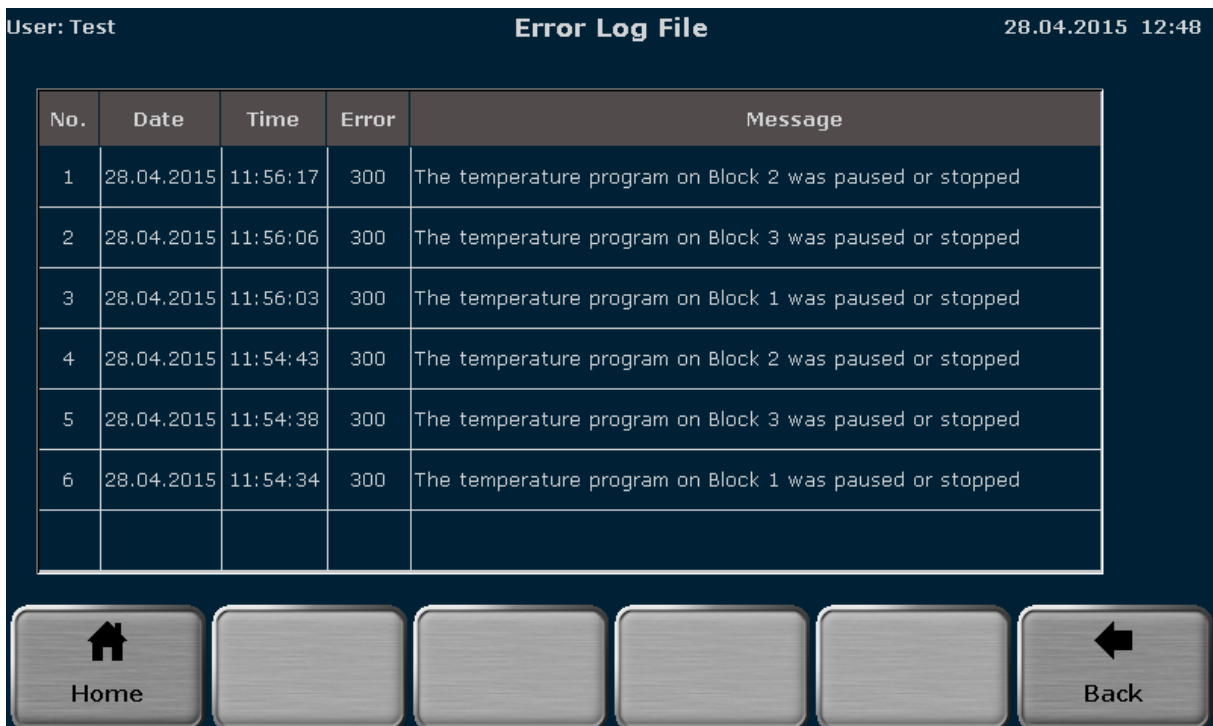


Figure 65 Biometra TRIO Error-Logfile screen

12.4 Extended Self-Test

During the Extended Self-Test the Biometra TRIO Thermal Cycler checks several functions and components:

Test	Function
Cooler	Incubates the sample block to 4°C and checks, if the sample block reaches 4°C and if the low temperature is hold for a longer time.
Thermal tracking	Checks the synchronism of the control circuits and if they work coordinated together.
Heating and cooling rate	Checks, if the specified average heating and cooling rate is reached.
Refrigeration	The cooling sink and fans are checked to work properly together.
Gradient ¹	Checks, if the sample block reaches the set gradient temperatures.
Heated lid	Tests, if the heated lid reaches the set temperature and if it holds the temperature for a longer time.
Regulation	Tests, if the sample block is controlled properly.

¹ not available for Biometra TRIO

In certain intervals the Biometra TRIO software will automatically recommend to execute the Extended Self-Test. Although it is not necessary to perform the test it is highly recommendable to follow the instructions and to let check the instrument itself.

Before starting the Extended Self-Test insert a microplate or, if applicable, two strips or a row of tubes to the first and last column of the sample block (see figure Figure 66). It is very important to load the block with plastic ware since during the Extended Self-Test the heated lid is tested and this test can only be executed properly with plastic ware inserted. Also the position of the plastic ware is important to ensure optimal pressure distribution and horizontal levelling of the heated lid.

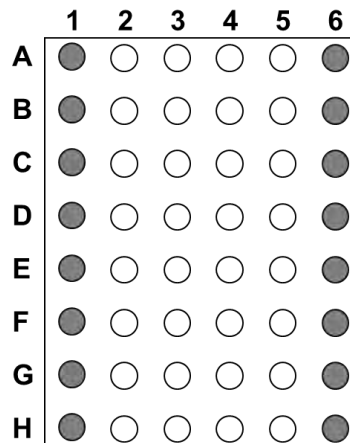


Figure 66 How to load plastic ware for the Extended Self-Test

If the plastic ware is properly inserted and the lid is closed the test can be started. To start the Extended Self-Test press

Start Extended Self-Test



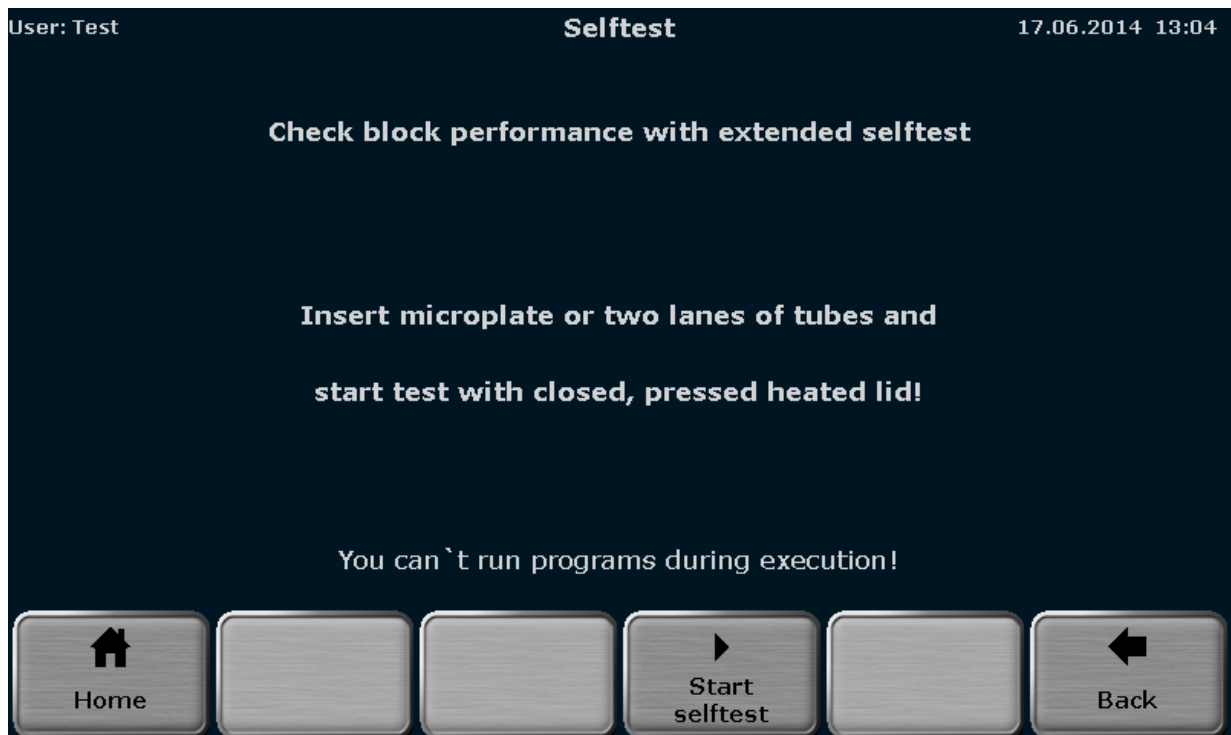


Figure 67 Biometra TRIO Extended Self-Test screen

During the test the software provides a progress overview. If a test is already passed the result “Failed” or “Passed” is displayed, for the currently performed test “Running” is shown and all other tests are not finished yet.

Note: The Extended Self-Test will take approximately 30 minutes to finish and the instrument is blocked, no other program can be started while the test is running. To stop the Extended Self-Test press

Stop Extended Self-Test



If the test is stopped, no corresponding log file will be created.

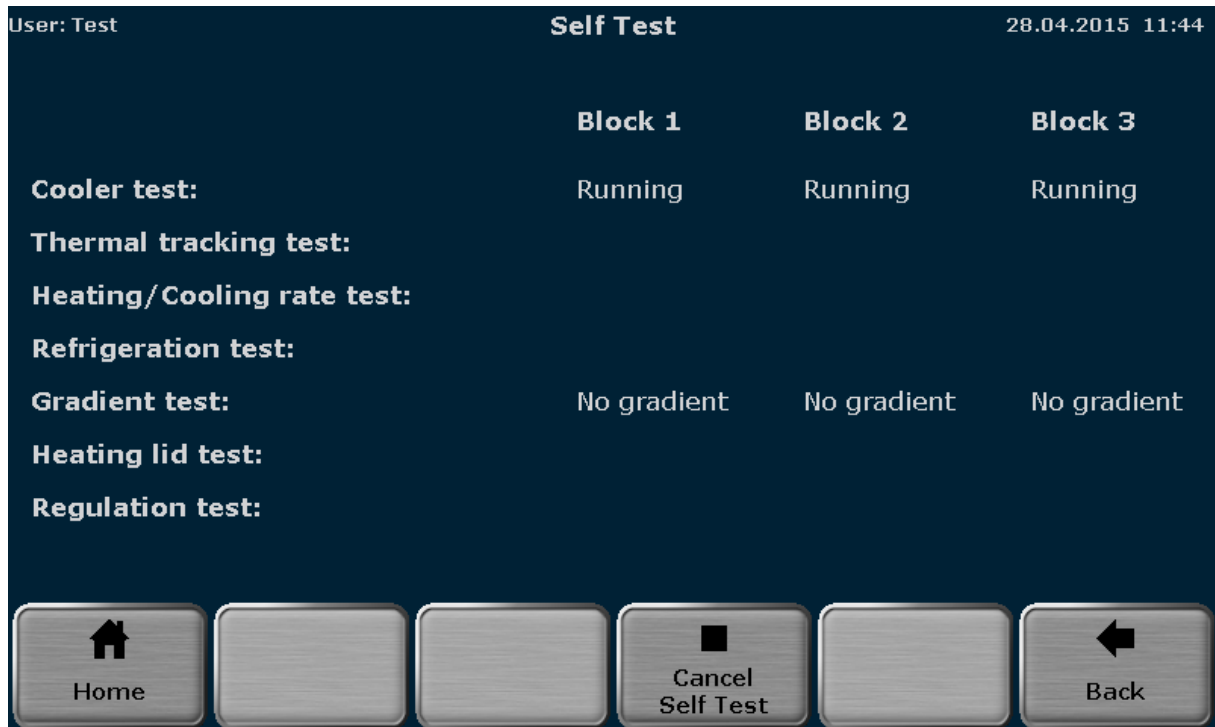


Figure 68 Biometra TRIO Extended Self-Test progress overview screen

The results of the Extended Self-Test are summarized and stored by the instrument and can be viewed by the user (see chapter 12.3.3).

12.5 Service Info File (SINF)

The Service Info File is a helpful tool for remote failure diagnosis by the Biometra service department.

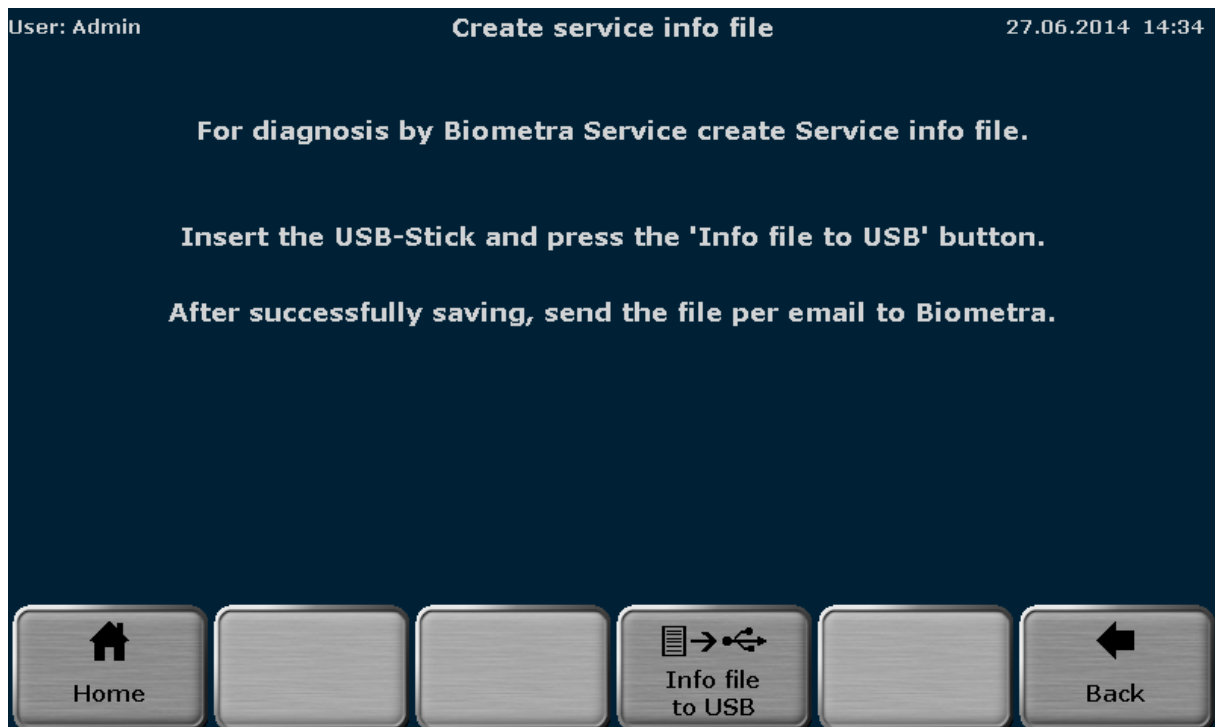


Figure 69 Biometra TRIO create Service Info File screen

To create a Service Info File connect a USB stick to the Biometra TRIO Thermal Cycler and press

Info file to USB 

The Service Info File can be send by e-mail to the Biometra service department. For contact details see chapter 12.8.

12.6 Backup

The backup function is used to synchronize the memory contents of Biometra TRIO instruments. By the backup function all folders, programs, users and user settings can be stored to USB and transferred to another Biometra TRIO instrument. Connect a USB drive to the instrument and press

Save backup file 

to create a backup file. To load a backup file select a backup from the list

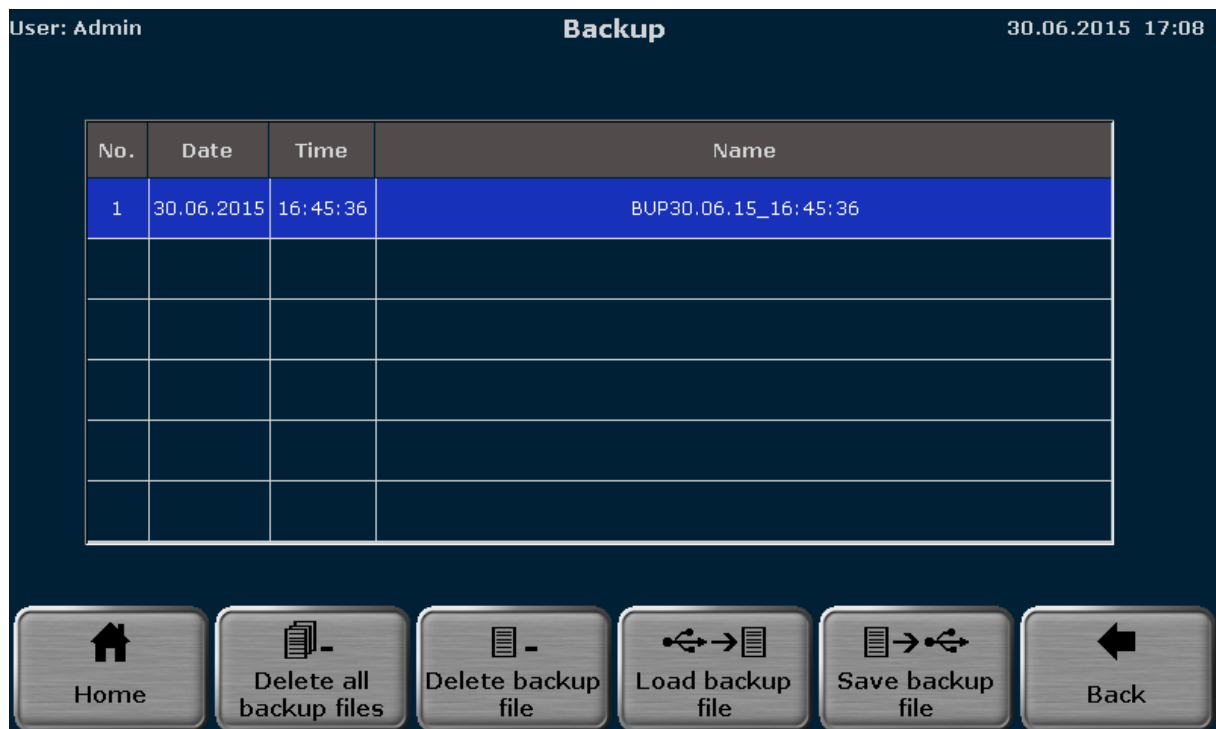


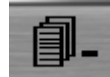
Figure 70 Biometra TRIO backup file screen

and press

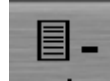
Load backup file 

Backup files that are no longer used can be deleted by using the functions

Delete all Backup Files



Delete Backup File



12.7 Cyclor Info

The Cyclor Info Tool presents several general information about the instrument like:

- Cyclor type
- Company
- Serial number Cyclor
- Block type
- Block serial number
- Software version
- Protocol version
- Revision number power board
- Software version power board
- Software version power board-logic
- Revision number processor board
- Software version processor board
- Revision number IO
- Software version IO

The information is summarized in two screens. Press

System/Cyclor Info



to toggle between the two screens.

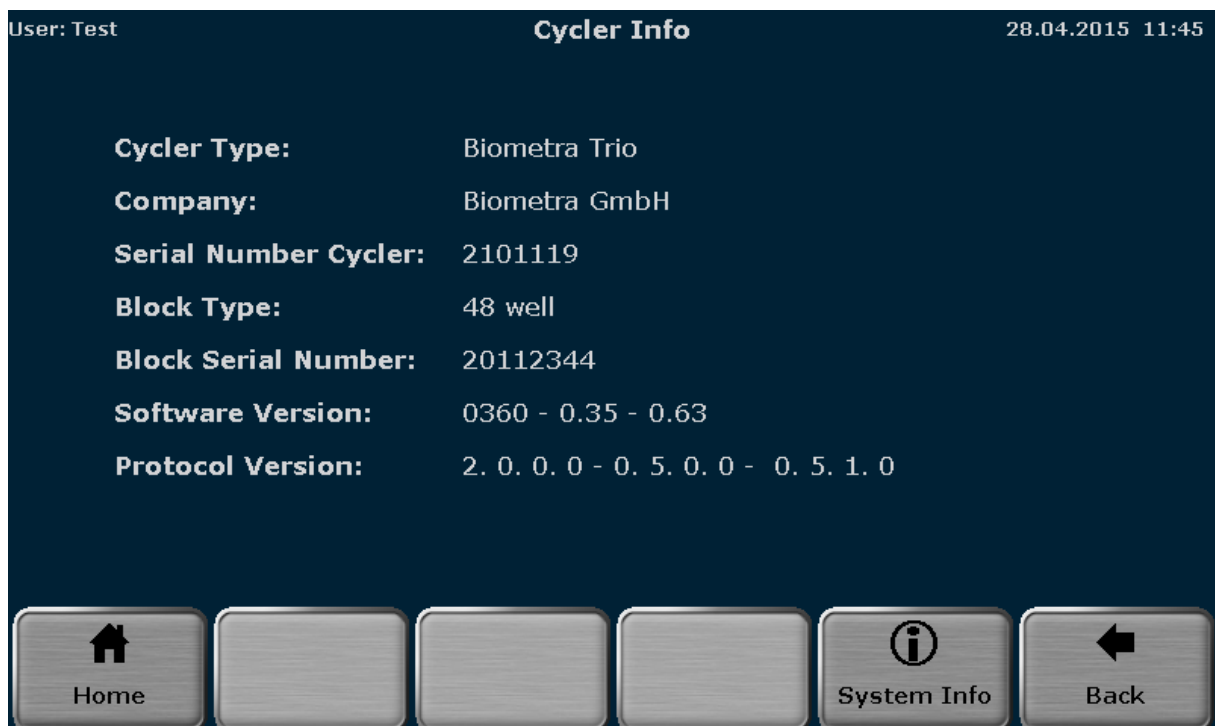


Figure 71 Biometra TRIO cyclor info screen

12.8 Contact

Shows contact details for the Biometra service department.



Figure 72 Biometra TRIO contact details screen

13 Adaptation of programs

If programs are exchanged between different thermal cycler models or of the sample block is exchanged programs might need to be adapted prior to start or during editing. The following table provides an overview for possible adaptations:

Cause	Example	Adjustment
Maximum gradient range is exceeded.	A program for the 96 well gradient silver block with a programmed temperature range > 30°C is to be started on a 96 well gradient aluminum block and therefore exceeds the block's maximum gradient range.	The temperature range of the gradient will be reduced based on the average temperature of the maximum permissible value. If the average temperature of the programmed gradient is, for example, 60°C and the temperature range is $\pm 20^\circ\text{C}$, it will be reduced to $60^\circ\text{C} \pm 15^\circ\text{C}$.
A program including a gradient step is transferred to a non-gradient instrument.	The program is including a gradient step started on a block without gradient function.	The gradient will be deleted and the average temperature/annealing temperature will be used for this step. If the average temperature/annealing temperature of the programmed gradient is for example 60°C, this value will be used for the step.
Gradient out of limit	A program for the 96 well silver block with a programmed gradient temperature below 20°C is to be started on a 96 well aluminum block and falls below minimum permitted temperature.	The lower temperature is increased to the minimum permitted value. If for example a gradient is programmed from 5°C to 25°C the gradient is changed to 20°C to 25°C.
A program including a temperature optimization step (TOS) is transferred to an instrument that does not offer this function	A TOS program is transferred from the Biometra TRIO to another thermal cycler	The mean value of the programmed temperatures in the TOS step for the left and right sample block is used.

The ramping rate exceeds the limit	A program for the 96 well gradient silver block is started on a 96 well aluminum block.	The ramp rate will be adjusted to the maximum possible ramp rate for the installed block type. If the programmed ramp rate is for example 8°C/s it will be reduced to 6°C/s.
The ramping rate is lower than the maximum	A program for the 96 well aluminum block is started on a 96 well silver block.	The ramp rate will be adjusted to the maximum possible ramp rate for the installed block type. If the programmed ramp rate is for example 6°C/s it will be increased to 8°C/s.
User defined ramping rate	The ramping rate for a program step is lower than maximum value	The user defined value is maintained and does not become adjusted

Note: If programs need to be adapted to the installed sample block prior to the start or editing a message will be displayed. The user can confirm or can deny the adaptations. If during program start the necessary adaptation is denied the program cannot be started by the instrument.

14 Short Manual

Safety Warnings



Before initial operation check the voltage selector configuration at the bottom to ensure that the instrument has the correct voltage setting. Do not open the instrument unless you are authorised to do so.



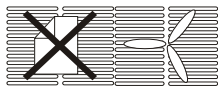
The thermoblock and the heated lid will reach high temperatures during operation. Both thermoblock and heated lid can burn you.



Rapid heating of the thermoblock can cause liquids to boil explosively. Always wear safety goggles during operation. Close the lid before starting a program.



It is not necessary to apply oil into the opening of the block in order to improve the heat transfer between the block and the sample tubes. If you still decide to use oil, do not use silicon oil. Mineral oil may be used.



Ensure that both the rear and bottom ventilation slits at the rear and bottom of the instrument are unobstructed. Insufficient ventilation can cause overheating of the instrument. **Tip:** At regularly intervals clean the bottom's ventilation slits from dust to ensure maximum venting capability.

Log in

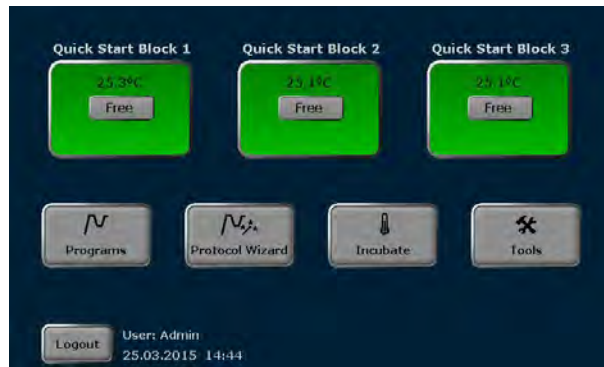
After the Biometra TRIO has passed the power-on self-test the login screen is displayed:



- Press [**Login**] to log in as existing user
- Press [**Quick start**] for the non-user specific quick start of programs
- Press [**EN/DE**] to set the login screen language

Home Screen

After log in the Biometra TRIO home screen opens:

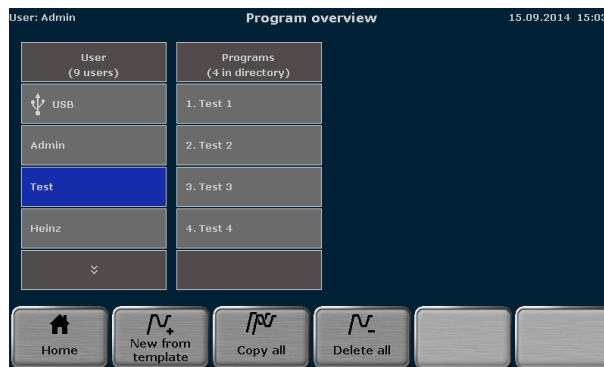


- Press **[Quick start]** for the user specific quick start of programs
- Press **[Programs]** to edit, save, copy or run programs
- Press **[Incubate]** to incubate the sample block at a constant temperature
- Press **[Tools]** to access the system settings, user management, documentation, backup tool or self-test functions
- Press **[Logout]** to log out

Edit, save and copy programs

Important note: It depends on your rights settings if you are allowed to create, delete, save, copy, start and stop programs. If you do not have a specific right you will not have access to the corresponding tools. Please ask your administrator or check your rights settings if you are missing a button or function.

- Press **[Programs]** in the Biometra TRIO home screen. The program overview screen opens:



- To edit a program template press **[New from template]**, select a template and press **[Open template]**.
- To edit a stored program select a user directory, select a program and press **[Edit]**.

The Biometra TRIO programming screen opens. Use the following buttons to switch between the different programming modes:

Graphical mode



Spreadsheet mode



Spreadsheet programming screen

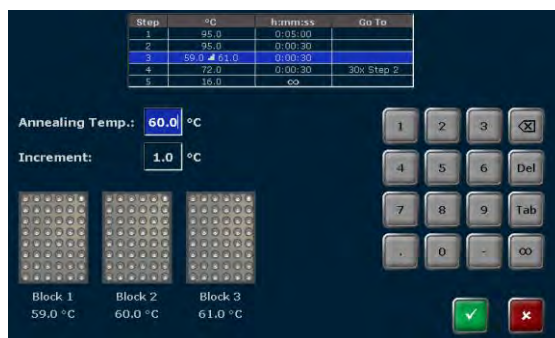
Graphical programming screen



- Define the program name. Touch the input box “Name” and enter a program name using the Biometra TRIO keyboard.
- Define the heated lid status and temperature. Touch the input box “Lid” and switch the heated lid on or off. If the heated lid is switched on enter a temperature between 30 °C and 110 °C.
- To switch heated lid preheating mode on or off press the corresponding button.
- Select a program step and press **[Edit]**. The edit step screen opens:



- Use the cursor keys at the top of the screen to switch between program steps. The number of the currently activated step is displayed.
- Touch the input box “Temperature” and enter a temperature [°C]
- Touch the input box “Holdtime” and enter the time [h:mm:ss]. To program a pause step press the button ∞.
- To program loop touch the input box “Goto” and enter the step number the program shall go back to. Then touch the input box “Cycles” and enter how often the loop will be repeated.
- To reduce or increase the annealing temperature from step to step within a loop press the button ΔT and set a negative or positive temperature increment.
- To increase or reduce the incubation time from step to step in a loop press the button Δt and set a time increment.
- To adjust the ramping rate press the button ΔR and enter any value between 0.1°C and max.
- To define a Temperature Optimisation Step press button **TOS**. Enter the primer annealing temperature and define the temperature increment. The temperature for each sample block is displayed. Press the button ✓ to confirm the settings.

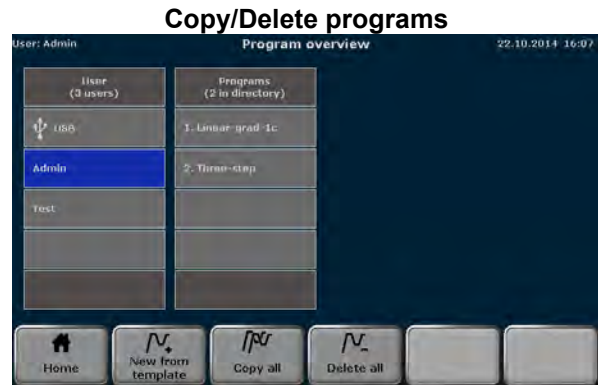
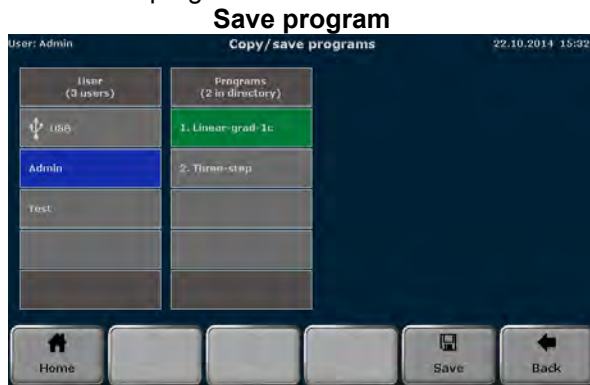


- To confirm all settings press the button ✓ in the edit step screen. The software leaves the edit step screen and shows programming screen in graphical or spreadsheet view.

- To insert a step select a program step in the programming screen and press **[Insert]**. The edit step screen opens. Enter the desired parameters and confirm the settings by pressing the button **✓**.
- To delete a step select a program step in the programming screen and press **[Delete]**. In the security query confirm to delete the step by pressing the button **✓**.

Tip: Storing samples at 15°C rather than 4°C at the end of a run will increase the instruments life time.

- When the parameters for all steps are entered confirm the settings by pressing the button **[Done]** in the programming screen. The software opens the screen to copy and save programs. Please note that the program is not saved yet.
- To save the program select a user directory and a storage place, then press the button **[Save]**. Now the program is saved.



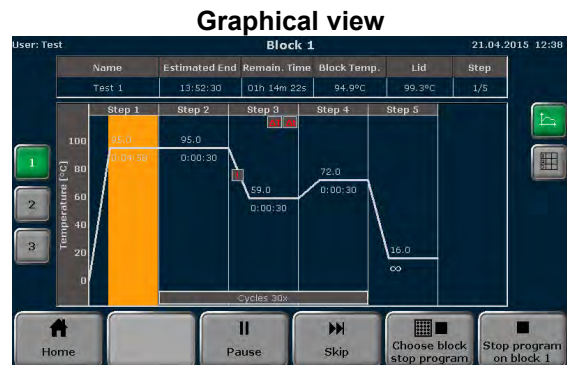
- To copy programs press the button **[Programs]** in the Biometra TRIO home screen.
- To copy all programs located in a user directory select a user directory and press the button **[Copy all]**. Select the user directory the programs should be copied to and press the button **[Save]**.
- To copy a single program select a user directory and a program and press the button **[Copy]**. Select a user directory, a storage place and press the button **[Save]**.
- To delete all programs located in a user directory select a user directory, press the button **[Delete all]** and confirm the security query.
- To delete a single program select a user directory and a program and press the button **[Delete]**. Confirm the security query to delete the selected program.

Start, pause and stop programs

- The Biometra TRIO thermal cycler stores the latest 5 edited or started programs for each user. For quick start of a program press the **[Quick start]** button in the Biometra TRIO home screen. Select a program from the list and press the **[Start]** button.
- To start a program from a user directory press the button **[Programs]** in the Biometra TRIO home screen. In the next screen select a user directory and a program and press the **[Start]** button.
- After start the running program is shown in spreadsheet or graphical view. Use the same buttons as for the programming screen to switch between the different modes.
- The current step is highlighted in yellow.

Spreadsheet view

Name	Estimated End	Remain. Time	Block Temp.	Lid	Step		
Test 1	13:52:30	01h 14m 16s	95.0°C	99.4°C	1/5		
Step	Block Temp. (°C)	Hold Time (minutes)	Go To	Cycles	ΔT(°C)	Δt(s)	ΔR(°C/s)
1	95.0	0:04:51	--	--	--	--	6.0
2	95.0	0:00:30	--	--	--	--	6.0
3	59.0	0:00:30	--	--	1.0	1	1.0
4	72.0	0:00:30	2	30	--	--	6.0
5	16.0	∞	--	--	--	--	6.0



- To pause the active program press the button **[Pause]**. The color of the highlighted step changes to blue and in the field “Step” the message **Pause** is shown.
- To continue a paused program press the button **[Continue]**.
- To skip a step press the button **[Skip]**. The program will continue with the next step.
- To stop the active program press the button **[Stop program]** button and confirm the security query. Programs with a pause in the last step have to be stopped manually as well.

Switch user management on or off (only for administrator)

- In the Biometra TRIO home screen press the button **[Tools]**, in the next screen the button **[User-Management]** and then use the checkboxes to switch the user management on or off.

15 Troubleshooting

15.1 Forgotten password

Passwords can be changed by the systems administrator(s) and by users with the activated right “**Edit User**” (see chapter 12.2.2). If the administrator Admin has forgotten the password and no one else has the right “**Edit user**” use the function “Factory settings” (see chapter 12.1.7) to reset the system and load a backup file (see chapter 12.6) to restore it.

15.2 Slow heating and cooling

The Biometra TRIO is equipped with a strong ventilator for the cooling of the heat sink. The inlet of this fan is located at the bottom side of the instrument. Make sure the inlet is not clogged by dust or other material (e.g. a sheet of paper placed under the cyclor can be attached to the inlet as the fan is in operation). Dust can be removed easily from the inlet using a conventional vacuum cleaner.

15.3 Auto restart

The Biometra TRIO Thermal Cycler features an automatic restart. If a power failure occurs during the run the instrument will continue the run when the power returns. In case of long-term power failure (longer than 30 minutes) the instrument keeps the sample block at 4°C (freeze step) and the user can decide to repeat the run with the same samples or to discard them.

Note: After power failure the user has to log in again. In addition for a limited time information can be displayed.

15.4 Auto restart due to unrecognized power failure

High voltage fluctuation can lead to an automatic restart of the Thermal Cycler. In this case the cyclor restarts at the step where there power failure has occurred. A message is displayed showing (a) that a power failure occurred during the run and (b) when and at which step the program was started again (see 15.3). To avoid voltage fluctuation, do not connect the cyclor to a socket shared by a strong power consumer like a refrigerator or a centrifuge.

Note: After power failure the user has to log in again. In addition for a limited time information can be displayed.

15.5 Adaptation of protocols from other cyclers

Since the Biometra TRIO is a fast instrument it may be necessary to reduce the heating and cooling ramps to run protocols from other cyclers. For the setting of the heating and cooling ramps see chapter 8.6.4. Alternatively, the time settings may be extended.

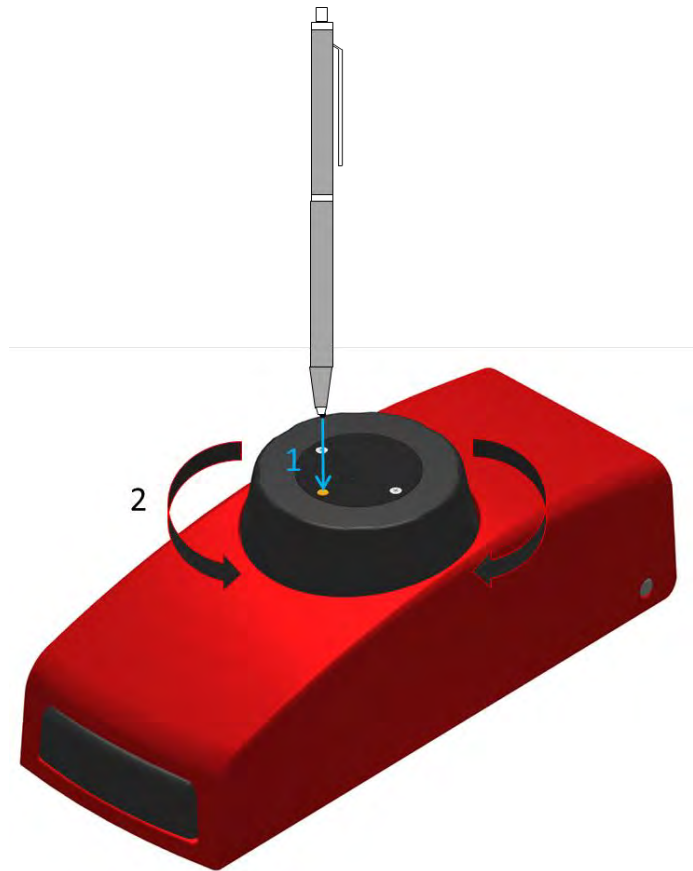
15.6 Releasing wheel in case of blocked lid

When the lid is in the highest or lowest position, it may happen that the wheel is uncoupled. In this situation the clutch mechanism is active in both directions (clicking noise in either direction).

To unlock wheel, press down metal pin with a ball pen and turn wheel carefully. This pin overrides the automatic clutch mechanism. Thus, care must be taken not to apply excessive pressure.

Release lid in upper position:

- 1) press pin
- 2) carefully turn wheel while holding the pin down **CLOCKWISE**, until you feel normal resistance (no more clicking noise, clutch is released). Release pin and turn lid down, until the clutch mechanism is activated (clicking noise, optimum pressure applied).



Release lid in down position:

- 1) press pin
- 2) carefully turn wheel while holding the pin down **COUNTER CLOCKWISE**, until you feel normal resistance (no more clicking noise, clutch is released). Release pin and turn wheel counter clockwise until pressure is completely released. Open lid.

Important: When the clutch mechanism is active (= optimum pressure is applied), do not use the pin to further increase lid pressure, as this would lead to damage of tubes and instrument!

16 Maintenance and Repair

16.1 Factory calibration

This instrument is supplied fully calibrated by the manufacturer before delivery, using equipment traceable to national standards. The Biometra Quality Management- System is certified by BSI for BS EN ISO 9001:2008 and the used measurement system is tested by an accredited testing laboratory in accordance with EN ISO 17025 at prescribed intervals and gets regularly calibrated. If the instrument is installed in an environment that meets the requested working conditions (see chapter 2.5), a temperature verification or calibration after installation is not required.

16.2 Cleaning and Maintenance

Depending upon the installation site the Biometra TRIO Thermal Cycler should be cleaned from time to time to maintain maximum performance. Occasionally cleaning of the air inlet contributes to maintain the instruments efficiency as insufficient airflow will lead to reduced heating and cooling rates (see chapter 15.1). The air inflow is located at the bottom side of the instrument. Be sure that the inlet is not clogged by dust or other material (e.g. a sheet of paper or a plastic bag). Dust can be removed easily from the inlet with a conventional vacuum cleaner. Additionally, the Thermal Cycler housing may be cleaned from time to time with a smooth cotton cloth. Do not use strong detergents, organic solvents or abrasive agents to clean the instrument and the sample block!

Important: Appropriate safety regulations must be observed when working with infectious or pathogenic material.

16.3 Servicing and Repair

The Biometra TRIO Thermal Cycler contains no user serviceable parts. Do not open the housing instrument. Service and repair may only be carried out by the Biometra service department or otherwise qualified technical personal.

Dependent on the usage the Biometra TRIO thermal cycler automatically shows a message screen recommending a preventive maintenance. Furthermore the Biometra service department offers maintenances and temperature verifications. Biometra recommends an annual maintenance and a biannual temperature check for all thermal cyclers. Please call the following phone number for detailed information: +49 551-50881-10/12.

16.4 Firmware update

For instruction for firmware upgrade, please contact the Biometra service department or your local distributor/sales representative.

16.5 Replacement of spare parts

Only original spare parts mentioned in these operating instructions are allowed.

17 Service

Should you have any problems with this unit, please contact our service department or your local Biometra dealer:

Biometra GmbH

Service Department

Rudolf-Wissell-Straße 14 - 16

D-37079 Göttingen

Phone: ++49 (0)5 51 50 68 6 - 10 or 12

Fax: ++49 (0)5 51 50 68 6 -11

e-mail: Service@biometra.com

Note: If you would like to send the unit back to us, please read the following return instructions in chapter 17.1.

17.1 Instructions for return shipment

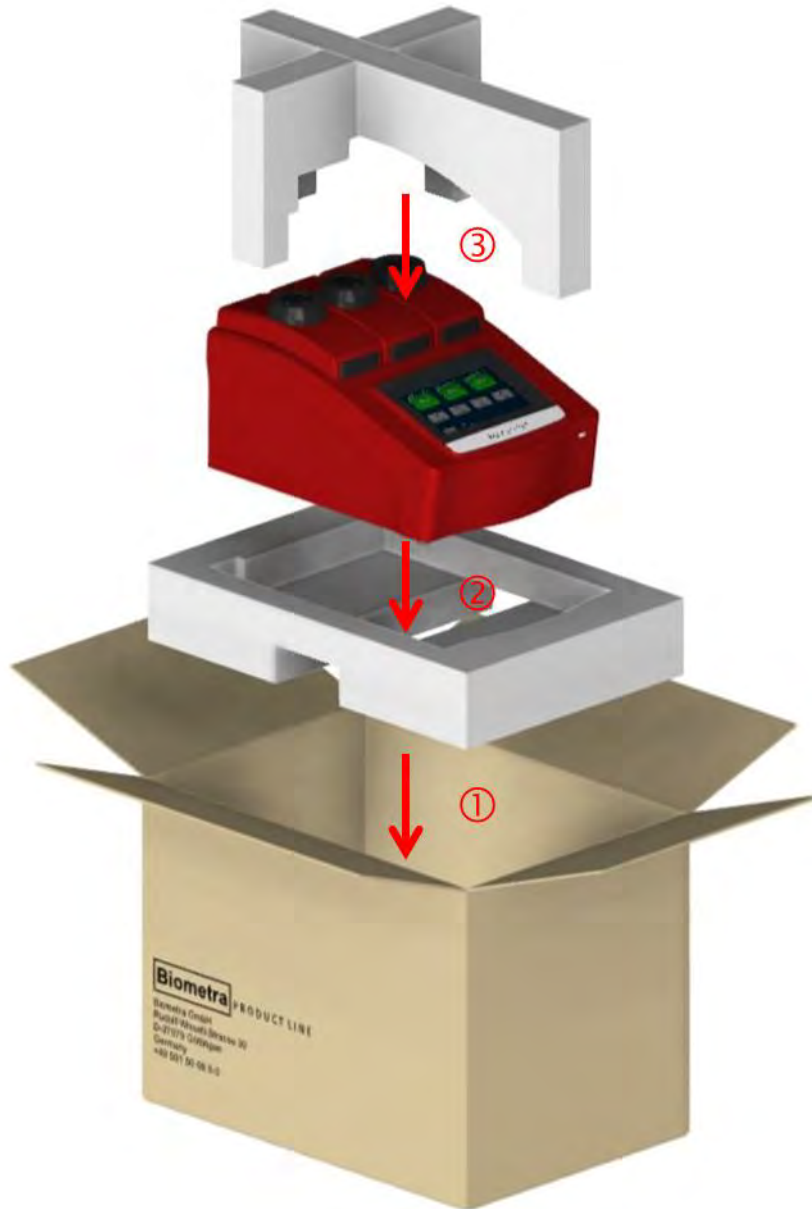
In case of an instrument failure that cannot be fixed by the procedures described in section 13 please proceed as follows:

- Return only defective devices. For technical problems which are not definitively recognisable as device faults please contact the technical service department at Biometra (Tel.: +49 551-50881-10/12, Fax: +49 551-50881-11, e-mail: service@biometra.com).
- Please contact our service department for providing a **return authorization number (RAN)**. This number has to be applied clearly visible to the outer box. **Returns without the RAN will be not be accepted!**
- **Important:** Carefully clean all parts of the instrument of biologically dangerous, chemical or radioactive contaminants. If an instrument is contaminated, Biometra will be forced to refuse to accept the device. The sender of the repair order will be held liable for possible losses resulting from insufficient decontamination of the device.
- Please prepare written confirmation that the device is free from biologically dangerous and radioactive contaminants. The declaration of decontamination (see section 18) must be attached to the outside of the packaging.
- Use the original packing material. If not available, contact Biometra or your local distributor.
- Label the outside of the box with “CAUTION! SENSITIVE ELECTRONIC INSTRUMENT!”
- Please enclose a note which contains the following:
 - a) Sender's name and address,
 - b) Name of a contact person for further inquiries with telephone number,
 - c) Description of the fault, which also reveals during which procedures the fault occurred, if possible

17.2 Packing of the Biometra TRIO Thermal Cyclers

Biometra uses an extra designed packaging system with inserts of foamed material.

Note: The Thermal Cycler is only protected from transport damage if the original transport box is used and the packing instructions are followed. Biometra is not responsible for transport damage by improper packing.



18 Equipment Decontamination Certificate

To enable us to comply with German law (i.e. §71 StrlSchV, §17 GefStoffV and §19 ChemG) and to avoid exposure to hazardous materials during handling or repair, please complete this form, prior to the equipment leaving your laboratory.

COMPANY / INSTITUTE _____

ADDRESS _____

PHONE NO _____ FAX NO _____

E-MAIL _____

EQUIPMENT	Model	Serial No
	_____	_____
	_____	_____
	_____	_____
	_____	_____

If on loan / evaluation Start Date: _____ Finish Date _____

Hazardous materials used with this equipment:

Method of cleaning / decontamination:

The equipment has been cleaned and decontaminated:

NAME _____ POSITION _____
(HEAD OF DIV./ DEP./ INSTITUTE / COMPANY)

SIGNED _____ DATE _____

PLEASE RETURN THIS FORM TO BIOMETRA GMBH OR YOUR LOCAL BIOMETRA DISTRIBUTOR TOGETHER WITH THE EQUIPMENT.

PLEASE ATTACH THIS CERTIFICATE OUTSIDE THE PACKAGING. INSTRUMENTS WITHOUT THIS CERTIFICATE ATTACHED WILL BE RETURNED TO SENDER.

General Information for Decontamination:

Please contact your responsible health & safety officer for details.


Use of radioactive substances:

Please contact your responsible person for details.

Use of genetically change organism or parts of those:

Please contact your responsible person for details.

19 Note for the disposal of electric / electronic waste.

	Note	for disposal of electric / electronic waste	
	Hinweis	für die Entsorgung von Elektroaltgeräten	
	Renseignement	du <u>traitement des déchets</u> des appareils électrique / électronique	

This symbol (the crossed-out wheelie bin) means, that this product should be brought to the return and / or separate systems available to end-users according to your country regulations, when this product has reached the end of its lifetime.

For details, please contact your local distributor!

This symbol applies only to the countries within the EEA*.

EEA = European Economic Area, comprising all EU-members plus Norway, Iceland and Liechtenstein.

Dieses Symbol (die durchgestrichene Abfalltonne) bedeutet, dass dieses Produkt von der Firma Biometra für eine kostenlose Entsorgung zurückgenommen wird. Dies gilt nur für Geräte, die innerhalb Deutschlands gekauft worden sind.

Kontaktieren Sie für die Entsorgung bitte die Biometra Service-Abteilung!

Außerhalb Deutschlands wenden Sie sich bitte an den lokalen Händler.

Dieses Symbol gilt nur in Staaten des EWR*.

***EWR = Europäischer Wirtschaftsraum, umfasst die EU-Mitgliedsstaaten sowie Norwegen, Island und Liechtenstein.**

Cet symbole (conteneur à déchets barré d'une croix) signifie que le produit, en fin de vie, doit être retourné à un des systèmes de collecte mis à la disposition des utilisateurs finaux en conséquence des réglementations par la loi de votre pays.

Pour des information additionnel nous Vous demandons de contacter votre distributeur!

Cet symbole s'applique uniquement aux pays de l'EEE*.

EEE = Espace économique européen, qui regroupe les États membres de l'UE et la Norvège, Islande et le Liechtenstein.

20 Declaration of Conformity

EU – Konformitätserklärung

EC - Declaration of Conformity

Göttingen, August 2015

im Sinne der EG-Richtlinie über elektrische Betriebsmittel zur Verwendung innerhalb bestimmter Spannungsgrenzen 2006/95/EG

following the EC directive about electrical equipment for use within certain limits of voltage 2006/95/EC

und / and

im Sinne der EG-Richtlinie für die elektromagnetische Verträglichkeit 2004/108/EG.

following the EC directive about the electromagnetic compatibility 2004/108/EC .

Hiermit erklären wir, dass folgende **Thermocycler:**

*Herewith we declare that the following **Thermocyclers:***

Typen / types: **Biometra TRIO 30, Biometra TRIO 48, Biometra combi**

Best.-Nr. / Order No.: 846-x-070-720, 846-x-070-723, 846-x-070-724

x = 2 für / for 230 V, 4 für / for 115 V, 5 für / for 110V

den grundlegenden Anforderungen der
corresponds to the basic requirements of

EG-Niederspannungsrichtlinie 2006/95/EG und der
EC low voltage directive 2006/95/EC and the

EG-Richtlinie über die elektromagnetische Verträglichkeit 2004/108/EG entsprechen.
EC directive about the electromagnetic compatibility 2004/108/EC .

Dr. Juergen Otte
Head of Quality Management

For and behalf of Biometra GmbH



Certificate number **FS 68563**

21 Warranty

This Biometra instrument has been carefully build, inspected and quality controlled before dispatch. Hereby Biometra warrants that this instrument conforms to the specifications given in this manual. This warranty covers defects in materials or workmanship as described under the following conditions:

This warranty is valid for 24 month from date of shipment to the customer from Biometra. This warranty will not be extended to a third party without a written agreement of Biometra.

This warranty covers only the instrument and all original accessories delivered with the instrument. This warranty is valid only if the instrument is operated as described in the manual.

Biometra will repair or replace each part which is returned and found to be defective. This warranty does not apply to wear from normal use, failure to follow operating instructions, negligence or to parts altered or abused.

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