

Operating manual

BiometraTRIO Triple Powered PCR thermal cycler



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1 Basic Information

1.1 User manual notes

The Biometra TRIO is intended for operation by qualified specialist personnel observing this operating manual.

For the Biometra TRIO, there are 3 different models available. In the text below these devices are collectively called Biometra TRIO. Differences are explained in the corresponding sections.

The operating manual contains information about the design and operation of the Biometra TRIO and provides personnel familiar with PCR technology with the necessary know-how for the safe handling of the equipment and its components. The operating manual further includes notes on the maintenance and service of the device and potential causes and remedies of any faults.

These operating instructions apply to all Biometra TRIO as of software version ME 1.70 - RE 1.46 (see "Cycler Info" p. 93).

Furthermore, sometimes the terms protocol and program are used synonymously, these are one or more programmed temperature steps with different or the same holding times and cycles.

Conventions Instructions for actions which occur in chronological order are numbered and combined in action units.

Safety instructions are indicated by pictographs and signal words. The type and source of the danger are stated together with notes on preventing the danger.

The elements of the control and analysis program are indicated as follows:

- Program terms are identified with SMALL CAPS (e.g., Menu FILE).
- Buttons are shown by square brackets (e.g., [OK] button)
- Menu items are separated by arrows (e.g., FILE > OPEN).

Symbols and signal words

The operating manual uses the following symbols and signal words to indicate hazards or instructions. The safety instructions are always placed before an action.



WARNING

Indicates a potentially hazardous situation which might cause fatal or very serious injuries (deformities).



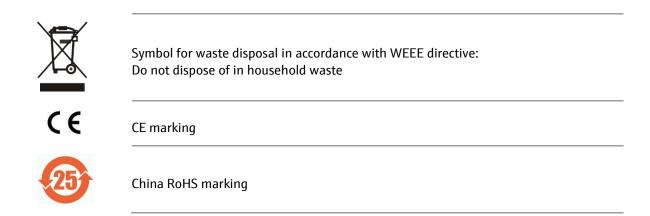
CAUTION

Indicates a potentially hazardous situation which might cause light or minor injuries.



NOTICE

Provides indications of potential material and environmental damage.



1.2 Intended use

The Biometra TRIO is an end-point thermal cycler. It is designed to amplify nucleic acids by repeated cycles of heating and cooling using DNA polymerases in a PCR reaction.

The thermal cycler is developed for Research Use Only (RUO).

The Biometra TRIO combines modern design with user-friendly software. The user interface consists of a touchscreen with a graphical or spreadsheet display that shows the time, the status and the temperature program for each run. Using the touchscreen keyboard, information and program parameters can be entered directly on the screen. Due to the high ramp rates, the Biometra TRIO thermal cycler is suitable for fast PCR applications, thereby helping to shorten program run times.

1.3 Warranty and liability

The warranty duration and liability comply with the legal requirements and the provisions in the general terms and conditions of Analytik Jena.

Deviations from the intended use described in this manual result in limitations of warranty and liability in the event of damage.

Warranty and liability claims are excluded for personal injury and property damage due to one or several of the following causes:

- use of the Biometra TRIO other than intended
- improper commissioning, operation and servicing of the device
- modifications of the Biometra TRIO without prior consultation with Analytik Jena/Biometra GmbH
- unauthorized intervention in the Biometra TRIO
- operation of the Biometra TRIO with faulty safety equipment
- use of other than original spare parts, wearing parts or consumables
- improper repairs
- improper transport and storage of the Biometra TRIO
- faults due to the non-observance of this manual

2 Safety instructions

2.1 General notes

For your own safety and to ensure error-free and safe operation of the Biometra TRIO, please read this section carefully before commissioning.

Besides the safety instructions in this user manual and the local safety regulations that apply to the operation of the device, the general applicable regulations regarding accident prevention, occupational health and safety and environmental protection have to be observed and complied with.

References to potential dangers do not replace the work protection regulations which must be observed.

2.2 Safety markings on the Biometra TRIO

Damaged or missing safety symbols can cause incorrect actions leading to personal injury or material damage. Do not remove safety symbols! Replace damaged safety symbols immediately!

The following safety symbols are attached to the inside of the heated lid and the rear of the device:



CAUTION! Hazard area!

Proceed with due care when handling samples and working with the Biometra TRIO.



CAUTION! Risk of burns!

There is a risk of burning at the heated lid, the thermal block, the samples, and the rear of the device.

The following sign is attached to the underside of the Biometra TRIO:



ATTENTION! Keep ventilation slits clear!

Keep the ventilation slits on the underside and at the rear of the device clear. Make sure that there are no objects underneath the device (e.g., paper) that may be sucked against the ventilation slot and thus interfere with ventilation.

2.3 Technical condition

The Biometra TRIO has been built and certified according to safety standard EN 61010-1. Do not modify the device in any way. Any modification of the device will lead to a loss of warranty and the EN 61010-1 certificate and poses a potential risk.

The following has to be observed:

- The operator must only operate the device in a sound and operationally safe condition. The technical condition must always comply with the legal requirements and regulations.
- Prior to every use the device must be checked for damage and sound condition.
- Any changes in the device affecting its safety must be reported by the operating personnel to the operator without delay.

2.4 Requirements for the operating personnel

The Biometra TRIO must only be operated by qualified specialist personnel instructed in the use of the device. The instructions also include imparting the contents of this manual.

In addition to the safety at work instructions in this operating manual the generally applicable safety and accident prevention regulations of the respective country of operation must be observed and adhered to. The operator must ascertain the latest version of these regulations.

The operating manual must be accessible to the operating and service personnel at all times.

2.5 Safety instructions: transport and installation

The following has to be observed:

- Drain the sample block before transporting the Biometra TRIO. Make sure there are no sample tubes in the block.
- Only ship the Biometra TRIO in its original packaging.
- Please verify that the delivery is complete on receipt of the Biometra TRIO and check for possible transport damage. In case of claims please contact Biometra GmbH.

2.6 Safety instructions – operation

2.6.1 General

The operator of the Biometra TRIO must make sure before each commissioning that the condition of the device including the safety equipment is sound.

The following has to be observed:

- Free access to the power switch on the back of the enclosure has to be ensured during operation.
- The ventilation fittings at the rear of the device must be unobstructed and operational. Covered ventilation grilles or slits etc. may cause the device to break down or may cause damage to it.

- The use of oil between the samples and the sample block is not necessary to achieve an improved heat exchange. However, if you still want to use oil, you should use mineral oil. Do not use silicone oil.
- Be careful to avoid crushing or pinching injuries when closing the device.

The Biometra TRIO may cause burns. The following has to be observed:

- The thermal block, the samples and the heated lid reach high temperatures. There is a risk of burns during contact.
- Ensure that the lid is securely closed before starting the program! Do not touch the heated lid!
- The rapid heating of the thermal block can cause liquids to boil explosively. Always wear safety goggles during operation!
- Do not touch hot sample tubes or plates and do not open them or boiling liquid may escape!
- Only use plates and tubes that are suitable for high temperatures (up to 100 °C), fit well into the thermal block (no shaking) and whose lids seal tightly!

2.6.2 Safety instructions: protection against explosion and fire

The Biometra TRIO must not be operated in an explosive environment. The Biometra TRIO must not be operated with flammable, explosive or volatile substances.

2.6.3 Safety instructions: electrical equipment

Work on the electrical components of the Biometra TRIO may only be performed by a qualified electrical technician in accordance with the applicable electrical engineering rules. Life-threatening electrical voltages may occur in the interior of the Biometra TRIO!

The following has to be observed:

- Any work on the interior of the device may only be carried out by the customer service of Biometra GmbH and specially authorized technicians.
- The electrical components must be checked regularly by a qualified electrician. Any defects, such as loose connections, faulty or damaged cables, must be repaired without delay.
- Before opening the device it must be switched off at the power switch and the mains plug must be disconnected from the mains outlet!
- The Biometra TRIO must be switched off immediately using the power switch (on the equipment back plate) and the mains plug disconnected from the power supply if any faults occur in the electric components.
- Do not operate the device near sources of strong electromagnetic radiation (for example, an unshielded, deliberately operated high frequency source), as these may affect proper operation of the device.

2.6.4 Handling of samples, auxiliary and operating materials

The operator is responsible for the selection of substances used in the process as well as for their safe handling. This is particularly important for radioactive, pathogenic, infectious, poisonous, corrosive or otherwise dangerous substances. For details contact the safety officer responsible for your location. When handling dangerous substances local safety codes and guidelines must be observed. The following general notes do not replace the specific local regulations or the regulations in the safety data sheets of the manufacturers for the auxiliary and operating materials.

The following has to be observed:

- Protective goggles and rubber gloves have to be worn when handing reagents.
- If only a few samples are treated, an (empty) tube of the same height must additionally be placed at each corner position of the block. If the number of samples in the block is too low, there is a risk of the tubes being damaged and sample liquid leaking out.
- For your own safety, please observe the potential infectious qualities of the examined biological material.

2.7 Safety instructions – maintenance and repair

The Biometra TRIO is usually repaired by the service department of Biometra GmbH or its authorized and trained specialist personnel. Unauthorized repairs can damage the device. Therefore, the operator may generally only carry out the tasks listed in section "Maintenance and Repair" p. 102.

The following has to be observed:

- The exterior of the device may only be cleaned with a damp, not dripping, cloth after the device has been switched off.
- Do not use alcohol (e.g., methanol or ethanol), organic solvents or abrasives to clean the device.
- Any maintenance on the device may usually only be carried out in the switched-off condition (unless stated otherwise).

2.8 Behavior during emergencies

In case of danger or accidents, immediately switch off the Biometra TRIO using the main switch on the equipment back plate. Disconnect the mains plug from the power supply!

3 Function and setup

3.1 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.

3.2 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 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.

3.3 Special features

3.3.1 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.

3.3.2 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.

3.3.3 Display

The Biometra TRIO thermal cycler features a 7" state-of-the-art color touchscreen user interface. The touchscreen is built-in at a flat angle to ensure reflection-free viewing and ergonomic programming. For programming PCR protocols the software incorporates Biometra's proven spreadsheet philosophy and alternatively offers graphical programming. One touch of a button leads from the spreadsheet to the alternative

graphical programming mode. This makes the creation of new programs or editing existing programs fast and easy.

3.3.4 User-specific quick start function

Many other thermal cyclers offer a so-called "List of latest programs used". In the list of latest programs used 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 creates a user-specific list and only the latest programs started by the currently logged-in user are offered for quick start. Even if a lab member does not use the device 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 additionally features a program preview. Before the start of a program the protocol steps are summarized in a clearly arranged table by the program preview tool. The preview therefore provides a comprehensive overview of the protocol structure without the need to access programming screens. The program preview is also available before the quick start of programs.

3.3.5 Easy programming

Creating new PCR programs takes much time if for each single step all parameters are 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.

3.3.6 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 ± 47.5 °C.

3.3.7 GLP compliance

The software can manage up to 90 user accounts. By default three different userlevels 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.

3.3.8 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 Biometra TRIO 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 3.3.2) 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".

3.4 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

Abbrevations:

x = 2 for 230 V, 4 for 115 V, 5 for 100 V

4 Installation and commissioning

4.1 Content of delivery

- Thermal Cycler
- Mains connector
- Manual
- 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 with form blocks for a safe transport of this electronic device.

4.2 Unpack and Check

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



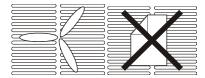
WARNING

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.

4.3 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 18 p. 107).
- Let equilibrate the Biometra TRIO Thermal Cycler to room temperature before starting operation (1 to 6 h).
- 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 Figure 3 p. 21). 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).
- In case of several cyclers side by side running simultaneously we would recommend to keep a distance of at least 10 cm between every thermal cycler.
- 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 "Operation Voltage" p. 20).
- 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.



WARNING

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.

4.4 Operation Voltage



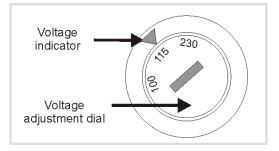
WARNING

Danger of electric shock! 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.



5 Control elements

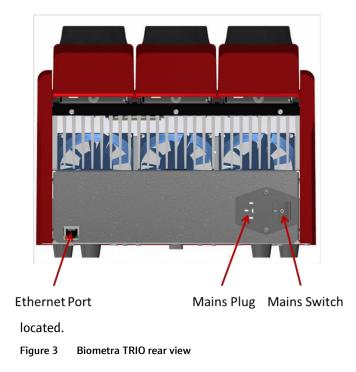
5.1 The Biometra TRIO front view





5.2 The Biometra TRIO rear view

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



5.3 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.

5.4 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.



CAUTION

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 4.
- 2. Release the pressure on the button when the lid becomes unlocked

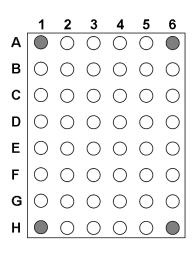
The lid opens automatically and stays open without assistance

- 3. Load the samples.
- 4. To close the lid push it down until it becomes arrested by the locking mechanism.



Figure 4 Biometra TRIO lid open button

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.



5.5 The Biometra TRIO Software

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

5.5.1 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
Тав	Tab
Shift	仓

DELETE SINGLE CHARACTER	$\langle \mathbf{X} $
DELETE ALL CHARACTERS	Del
Васк	←
Forward	→

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

5.5.2 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

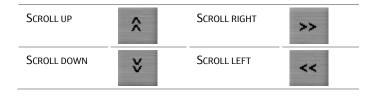
The keyboard offers to enter digits, dot and minus and infinite. Additionally the following special keys are available:

The keypad can be used to enter digits, dots, minus signs and infinite. In addition the following special keys are available:

DELETE SINGLE CHARACTER	$\langle \mathbf{X} $
DELETE ALL CHARACTERS	Del
Тав	Tab
Infinite	∞

5.5.3 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.



5.5.4 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.



5.5.5 List of used commands and icons

The Biometra TRIO thermal cycler software uses the following icons:

Command		lcon
Номе		Ħ
Васк		+
Select sample block 1		 1
SELECT SAMPLE BLOCK 2		 2
SELECT SAMPLE BLOCK 3		 з
	Rіght	 >>
	Left	
	Up	 *
	Down	*

Command			lcon
		Confirm	×
		Cancel	×
Login			0
		User management	1
		New user	1.
		User management ON/OFF	11
		Delete USER	1_
		Edit user	20
PROTOCOL WIZARD			M***
	New Polymerase		_ ● → +
	Edit Polymerase		
	Delete Polymerase		_ ● →
		Temperature Optimisation Step (TOS)	тоѕ
		Edit primer Concentration	
		Edit primer	ACGT
		CALCULATE PRIMER	
		Save annealing temperature Ta	Ta√
		PROTOCOL WIZARD INFO	
INCUBATION			l
	START INCUBATION		•

Command			lcon
Programs			\sim
	NEW PROGRAM		\\.
	OPEN TEMPLATE		M
	COPY PROGRAM/COPY ALL PROGRAMS		Tp:r
	DELETE PROGRAM		\sim
	Edit program		N
	START PROGRAM		N,
	CHOOSE BLOCK START PROGRAM		アコ
	CHOOSE BLOCK START PROGRAM		
	STOP PROGRAM		
	Done		\checkmark
	SAVE PROGRAM		
	Save as		
	Pause program		II
	CONTINUE PROGRAM		•
	SKIP STEP		M
		GRAPHICAL VIEW	Þ
		Spreadsheet view	1
		Edit step	5

Command			lcon
		INSERT STEP	Γ.
		DELETE STEP	Γ-
		Temperature	⊿Τ
		TIME INCREMENT	∆t
		HEATING RATE	∆R
		PREHEATING OFF	Off
		PREHEATING ON	On
Tools			*
	Settings		*
		DATE AND TIME	
		AUTOMATIC USER LOGOUT	₽
		Beeper	-
		Network	_~_
		DISPLAY BRIGHTNESS	×
		Display darker	÷¢÷
		DISPLAY BRIGHTER	
		Touchscreen calibration	·¢·
		Factory settings	444
	DOCUMENTATION		

Command			lcon
		Run log file	
		RUN LOG FILE OVERVIEW	
		SAVE RUN LOG FILE TO USB	□→ ↔
		SAVE ALL RUN LOG FILES TO USB	₽→⊷
		VIEW RUN LOG FILE	≣/≎
		VIEW PROGRAM	N_{a}
		Power-on-Logfile	Power On Log File
		LOG FILE SELF-TEST	
		ERROR LOG FILE	
E	EXTENDED SELF-TEST		
		START EXTENDED SELF- TEST	•
		STOP EXTENDED SELF- TEST	
9	SERVICE INFO FILE		Service info file
E	Заскир		Backup
E	BACKUP FILE	SAVE BACKUP FILE	□→ ↔
		LOAD BACKUP FILE	∎→•द• •द•→
		DELETE BACKUP FILE	- 1
		DELETE ALL BACKUP FILES] -
(CYCLER INFORMATION		(i)
(Contact		

6 Operating

6.1 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 "Power-On-Logfile" p. 87).

6.2 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 6.3
Вьоск	Shows the current block status. If a block is free a touch on the button leads to a list of previously used pro- grams for quick start for the selected sample block (not user specific before log in).	See chapter 6.5 and 6.6
EN/DE	Changes the language setting for the log in screen.	For user-specific language settings see chapter 11.2.1

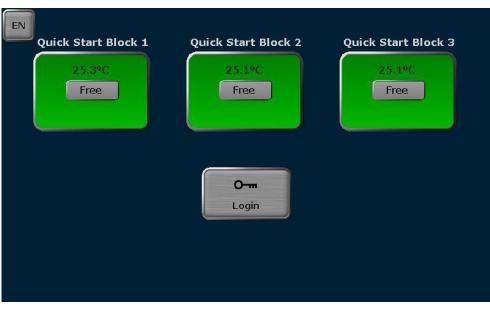


Figure 8 Biometra TRIO log in screen

6.3 Log in

After starting the Biometra TRIO Thermal Cycler the log in screen is displayed (see chapter "Log in Screen" p. 30). To log in as existing user press LOGIN.



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

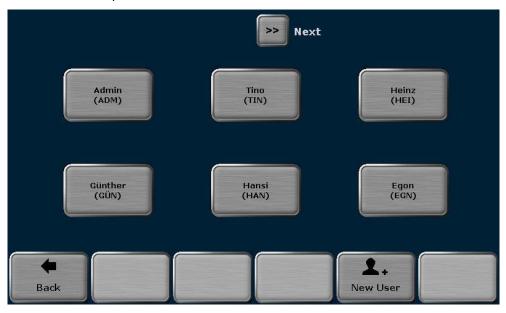


Figure 9 Biometra TRIO select user screen

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 "List of used commands and icons" p. 25).

Touch the input box PASSWORD and enter the password using the Biometra TRIO keyboard (see chapter "Keyboard" p. 23). 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.

6.4 Home Screen

After user login the Biometra TRIO software shows the home screen. The home screen offers the following functions:

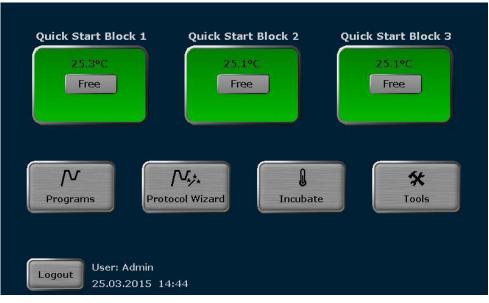


Figure 11 Biometra TRIO home screen

Command	Function	Chapter
Programs	Viewing, running and editing programs	See chapter 7
Protocol Wizard	Automatic creation of PCR programs	See chapter 10
Вьоск	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 6.5 and chapter 6.6
INCUBATE	For running a sample block at a constant temperature	See chapter 6.7
Tools	Settings, documentation, extended self-test, cycler info and diagnosis mode	See chapter 11
Log out	User log out	

6.5 Block status

In the log in screen (see chapter "Log in Screen" p. 30) and the home screen (see chapter "Home Screen" p. 32) 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 Figure 12). 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.

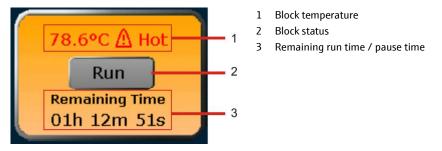
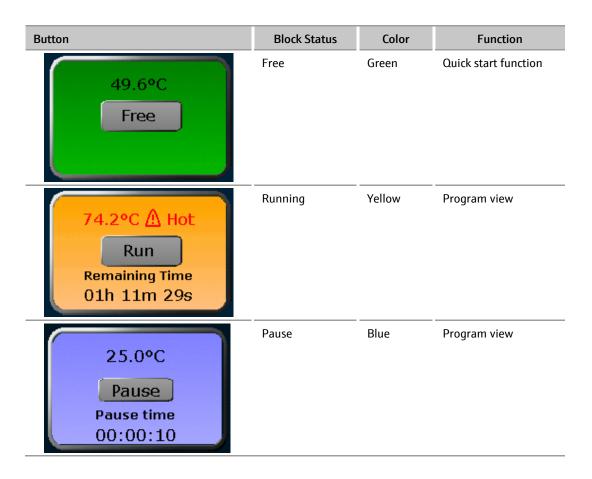


Figure 12 Biometra TRIO block status/quick start button

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 "Display during run" p. 58). If the status is FREE, the program quick start function is activated (see chapter "Quick start function" p. 34).



6.6 Quick start function

Note: Programs can only be started by users with the right START/STOP PROGRAMS activated (see chapter "Edit user" p. 81).



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 "Log in Screen" p. 30) or the home screen (see chapter "Home Screen" p. 32).

Note: The quick start function can only be activated if the current block status is FREE (see chapter "Quick start function" p. 34). Is 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.

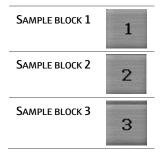
LOG IN SCREEN	Latest program list
Home screen	User specific latest program list

The latest used programs are summarized in a table.

User: Sa	ndra		Latest programs used			10.09.2018 14:5	
I	Block 3						
	No.	User	Program No.	Name	Block Type	Program Type	
	1	SK	08	Touchdown	48 well		
	2	SK	07	Vill	48 well		
2	з	SK	02	Villin	48 well		
3	4	SK	03	Two-step	48 well		
		SK	01	Villin	48 well		
		•			·		
1			Ne	w from	Choose b	lock Start	
Ho	me			emplate Edit	start prog		

Figure 13 Biometra TRIO quick start screen

To switch between the sample blocks choose the number of the sample block.



Select and touch a program in the table for quick start. Press the [START] button to start the selected program.



To start a program on multiple sample blocks press the [CHOOSE BLOCK START PROGRAM] button.



By activating the checkboxes select the sample blocks to start.



Figure 14 Biometra TRIO multiblock selection query

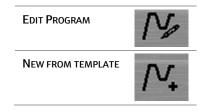
If necessary, the program can be edited before or a new program can be created using a template (see chapter "Create a new program/Use program template" p. 40).

Note: For programs including a TOS step (see chapter "Set Temperature Optimisation Step (TOS)" p. 47) 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 "Edit user" p. 81).



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



6.7 Incubation Mode

Note: Incubations can only be started or stopped by users with the right START/STOP PROGRAMS activated (see chapter "Edit user" p. 81).



The incubation mode allows to incubate samples at a constant temperature for a defined time. Press the [INCUBATE] button.

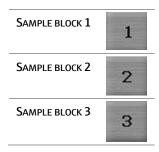


In the next screen set the block temperature, hold time, heated lid on or off, heated lid temperature and set preheating mode on or off using the virtual keypad and the corresponding buttons (see Figure 6).

Note: To set the time to endless use the following button:

ENDLESS TIME	and the second sec	_
	00	
		_

To select a sample block press:



To start the incubation press the [START INCUBATION] button.

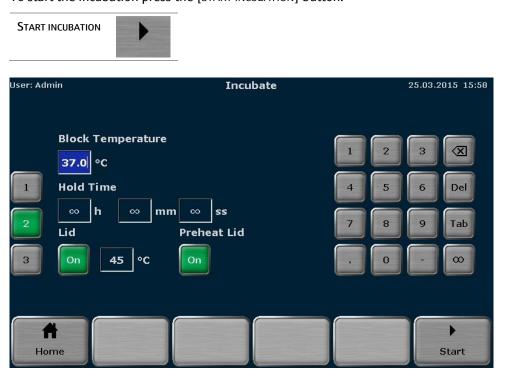
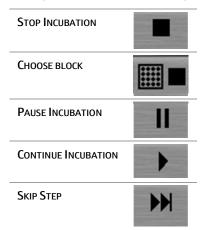


Figure 15 Biometra TRIO incubation mode screen

During incubation the following commands can be used:



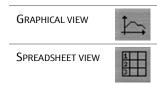
The button [CHOOSE BLOCK STOP PROGRAM] for multiblock selection is only available if more than one sample block is running.

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.

7 Creating, editing and saving programs

7.1 Programming modes

The Biometra TRIO software offers spreadsheet programming or graphical programming. Use the following buttons to toggle between the two modes (see Figure 16 and Figure 17).





The following screenshots show the spreadsheet and graphical programming mode.

Figure 16 Biometra TRIO spreadsheet programming

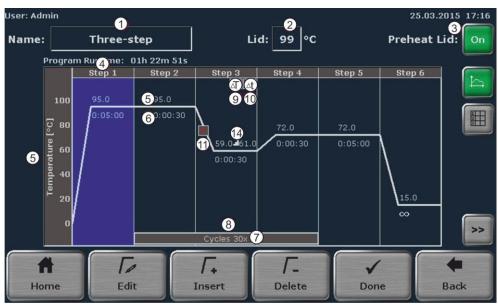


Figure 17 Biometra TRIO graphical programming screen

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 7.3
Heated lid temperature [°C]	2	2	See chapter 7.4
Preheat lid mode	3	3	See chapter 7.5
Step number	(4)	4	See chapter 7.6
Temperature [°C]	5	5	See chapter 7.6
Holding time [hh:mm:ss]	6	6	See chapter 7.6
Goto	7,12	$\overline{\mathcal{O}}$	See chapter 7.6.1
Cycles	8, 13	8	See chapter 7.6.1
Temperature increment [ΔT (°C)]	9	9	See chapter 7.6.2
Time increment [Δt (s)]	10	10	See chapter 7.6.3
Ramping rate [ΔR (°C/s)]	11		See chapter 7.6.4
TOS step	14	14)	See chapter 7.6.5

If a program has more than 6 steps, use the cursor keys to navigate in the spreadsheet or the graphical display.

7.2 Create 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 "Edit user" p. 81). Programs of other users can only be modified by users with the right WRITE/DELETE ALL PROGRAMS activated.

ENABLE USER TO EDIT OR DELETE OWN	Write/delete	
PROGRAMS	own programs	

- 1. To create a new program or to open a program template, first log in as existing user.
- 2. After log in press the [PROGRAMS] button in the home screen.



The program overview screen opens:

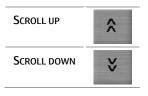
Us	er: Admin	verview	1	5.09.2014 15:03	
	User (9 users)	Programs (4 in directory)			
	🖞 usв	1. Test 1			
	Admin	2. Test 2			
	Test	3. Test 3			
	Heinz	4. Test 4			
	×				
	Home	om Copy all	Delete all		

Figure 18 Biometra TRIO program overview screen

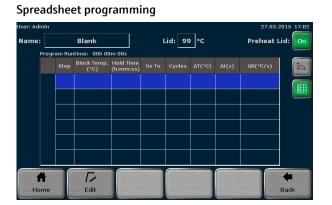
1. In the program overview screen press the following button:



2. The program template screen opens (see Figure 18). Search for a program template using the cursor keys 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 19).



Graphical programming



Figure 19 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.

3. Press the [OPEN TEMPLATE] button to open a program template.



		Γ					
Prograi	ns				Preview		
			Progran	n informatior	1		
Blank				hree-step			
				pe: 48 well			
	Three-step			'C Preheat Lic			
Three-step			Program Runtime: 01h 12m 26s				
			Step	°C	h:mm:ss	Go To	
			1	95.0	0:05:00		
Fast-1s-60c			2	95.0	0:00:30		
			3	60.0	0:00:30		
			4	72.0	0:00:30	30x Step 2	
Two-step			5	72.0	0:05:00		
			6	15.0	00		
*							
		L L					
							-
		- N		the second se			

Figure 20 Biometra TRIO program template screen

7.3 Assign program names

Each template has a specific name which is superimposed in the programming screen. To enter a new name touch the input box NAME and enter a new name using the Biometra TRIO virtual keyboard (see chapter Keyboard'''p.23).

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.

7.4 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 21) 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.

User: PCR			10.09.2018 11:	21
Name:	Osa QCAssay	Lid	Preheat Lid: On	
GoTo AT At AR		0n 99 ℃ 1 2 3 🕅 4 5 6 Del 7 8 9 Tab	Step 5 Step 6 72.0 72.0 0:00:20 0:02:00	
		Insert Delete	vcles 40x >> Done Back	

Figure 21 Biometra TRIO lid temperature setting screen



NOTICE

The heated lid automatically switches off at a temperature difference of >75 $^{\circ}$ C between the block and the heated lid. At these low block temperatures, sample condensation on the tube lid is no longer to be expected. In addition, it supports a longer service life of the Peltier elements in the block, since no unnecessary cooling against the heating cover is required.

7.5 Set preheating mode

The heated lid can be set to preheating ON or OFF using the corresponding buttons:



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.

7.6 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 22). 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 "Programming modes") and enter the desired values or select a program step and press the [EDIT STEP] button.



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 7.6
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 7.6
Gото	Defines the step number the program jumps back to.	See chapter 7.6.1
Cycles	Defines the number of repeats. The maximum number of repeats is 999.	See chapter 7.6.1
ΔT	Temperature increment to increase or decrease the set temperature with each cycle by the defined value. The increment can be set to $\pm 20^{\circ}$ C.	See chapter 7.6.2
∆t	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 Os to 240s.	See chapter 7.6.3
∆R	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 7.6.4
TOS	Temperature Optimisation Step (TOS). For optimisation of new PCR pro- tocols very often gradient enabled Thermocyclers are used. The Biomet- ra TRIO offers no gradient function but by the temperature optimisation step (TOS) function pograms making use of the three blocks and deliver- ing different annealing temperatures at the defined step can be created. It is possible that in some models this display may show a slightly higher	See chapter 7.6.5
	maximum ramping rate than is achieved according to the technical spec- ifications of the device. This is technically justified and achieves opti- mum control of the maximum value. In fact, the maximum heating and cooling rate is achieved as specified in the technical specifications.	

	Step	°C	h:mm:ss	Go To	
					V
	I		<u> </u>		
		_		1	2 3 🕅
Temperature:		_•c		4	5 6 Del
Hold Time:		h	mm ;	ss 7	8 9 Tab
Go To:		Cycl	es:		0 - ∞
	ΔΤ		AR TOS		

Figure 22 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 cursor buttons to navigate between the steps..



The currently activated step is high-lighted in the program overview table (see Figure 22).

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.

7.6.1 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 22). To define the number of repeats, enter a number in the input box CYCLES.

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.

Loops	Step	Block Temp. (°C)	Hold Time (h:mm:ss)	Go To	Cycles
	1	95.0	0:05:00		
	2	95.0	0:00:30		
35x	з	60.0	0:00:30		
	4	72.0	0:00:30	2	35
	5	72.0	0:05:00		
	6	16.0	œ		

Figure 23 PCR program with loop

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.

7.6.2 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 the following button (see Figure 22) and fill in a value between $+20^{\circ}$ 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.

7.6.3 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 the following button (see Figure 22) 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.

7.6.4 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 the [RAMPE RATE] button.



Then enter a value between 0.1°C/s and max. in tenths of degree Celsius.

It is also possible that the entered ramp rate is applied to all steps in the PCR program. For this the following checkbox to the right of the input field for the ramp rate must be activated.

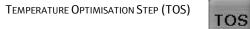


Note: The maximum average rate depends on the sample block installed in the Biometra TRIO Thermal Cycler. If the ramp rate has been adjusted, the small symbol will be shown at the corresponding step in the graphical programming mode.

7.6.5 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 the following button.



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 23).

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.

	Step	°C	h:mm:ss	Go To			
	1	95.0	0:05:00				
	2	95.0	0:00:30				
	3	59.0 🛋 61.0	0:00:30				
	4	72.0	0:00:30	30x Step 2			
	5	72.0	0:05:00				
Annealing Tei	mp.: <mark>60.</mark>	o °c		1	2	3	$\overline{\mathbf{x}}$
Increment:	1.0	°C		4	5	6	Del
		000000		7	8	9	Tab
					0		ω
Block 1	Block 2	Block 3			ſ		
59.0 °C	60.0 °C	61.0 °C				\checkmark	×

Figure 24 Biometra TRIO Temperature Optimisation Step screen

7.7 Insert step

To insert a new step, select a program step in the Biometra TRIO programming screen (see Figure 16 and Figure 17) and press the [INSERT STEP] button.



By using the [INSERT STEP] button the EDIT STEP function (see chapter "Edit parameter or step" p. 44) 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.

7.8 Delete step

To delete a step, select a program step in the Biometra TRIO programming screen (see Figure 16 and Figure 17) 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.

7.9 Save program

To save a program, press the [DONE] button.



On the Biometra TRIO spreadsheet or graphical programming screen. In the next program overview screen (see Figure 25), touch the corresponding buttons to select a user directory and a program storage location.

If the number of user directories or programs is higher than the maximum number that can be displayed in the list, the buttons above and below the list can be used to scroll up and down.

Note: The user directory of the logged in user is automatically preselected.

Note: If a new program is to be saved, the first free storage location in the directory of the currently logged in user is automatically selected. However, the storage location can be changed using the cursor keys above and below the lists. If the selected storage location is already in use, the stored program will not be overwritten; instead this program and all other following programs will be moved one position higher. The storage location number for these programs will increase by +1 and the new program is saved at the selected location.

U٩	ser: Test	programs	2	1.04.2015 12:23	
	User (3 users)	Programs (4 in directory)			
	🖞 иѕв	1. Test 1			
	Test	2. Test 4			
	Admin	3. Test 2			
		4. Test 3			
		×			
ſ					
	Home			Save	Back

Figure 25 Program overview screen

There is also the option of selecting a user directory or a program location using the quick select function.

Touch the icon above the user or program view (see Figure 25).



An overview screen opens in which a maximum of 50 users or 50 user programs are displayed. If more than 50 users are saved, scroll through the user list using the cursor keys. This is also true for more than 50 user programs (see Figure 26).

Rіght	>>
Left	<<

To select a user or program, press the corresponding button with the user or program name. The user or the program will be selected and the program overview screen (see Figure 25) appears again.

		>> N	ext	
1.Three-step	11.Fast-1s-60c	21.Linear-grad-1c	31.Three-step	41.Linear-grad-1c
2.Fast-1s-60c	12.Linear-grad-1c	22.Three-step	32.Linear-grad-1c	42.Three-step
3.Three-step	13.Three-step	23.Fast-1s-60c	33.Three-step	43.Fast-1s-60c
4.Fast-1s-60c	14.Fast-1s-60c	24.Linear-grad-1c	34.Fast-1s-60c	44.Three-step
5.Linear-grad-1c	15.Linear-grad-1c	25.Three-step	35.Linear-grad-1c	45.Linear-grad-1c
6.Fast-1s-60c	16.Three-step	26.Fast-1s-60c	36.Three-step	46.Fast-1s-60c
7.Three-step	17.Fast-1s-60c	27.Linear-grad-1c	37.Fast-1s-60c	47.Linear-grad-1c
8.Fast-1s-60c	18.Linear-grad-1c	28.Linear-grad-1c	38.Linear-grad-1c	48.Three-step
9.Linear-grad-1c	19.Fast-1s-60c	29.Three-step	39.Three-step	49.Fast-1s-60c
10.Three-step	20.Three-step	30.Fast-1s-60c	40.Fast-1s-60c	50.Linear-grad-1c

Figure 26 Quick selection screen

Before saving the program there is still the possibility of changing the program name.

To do this, press the [SAVE AS] button and enter the new program name using the Biometra TRIO keyboard (see section "Assign program names" p. 42).

```
SAVE AS
```



After selecting the user directory and the storage location and possibly changing the program name, press the [SAVE] button on the Biometra TRIO program overview screen (see Figure 25) to save the program.

SAVE



8 Starting, Copying and Deleting programs

8.1 Start program

Note: Programs can only be started by users with the right START/STOP PROGRAMS activated (see chapter "Edit user" p. 81). 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.

ENABLE A USER TO START OR STOP PROGRAMS	Start/stop programs	
ENABLE A USER TO READ ALL PROGRAMS OF OTHER USERS	Read all programs	

The Biometra TRIO software offers a quick start function for previously used programs. The quick start function is described in chapter 6.6. This chapter deals with the start of stored programs by selecting a program from a user account. To select a program for start, press the programs button in the Biometra TRIO home screen (see chapter "Home Screen" p. 32).



The program overview screen opens with the user directory of the logged in user preselected (see Figure 27).

U٩	ser: Test			Program o	verview	2	1.04.2015 10:51
	User (3 users)		(Programs 4 in directory)			
🜵 USB			1. Te	st 1			
	Test		2. Te	st 2			
	Admin		3. Те	st 3			
		4. Te	st 4				
ſ			and the second se	nal l	Į∿_		
L		mpla		Copy all	Delete all		

Figure 27 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 28). The program preview can be used to check the program prior to start. To start the selected program press:





U	ser: Test	Program	ov	erviev	v	2	1.04.2015 10:54
	User (3 users)	Programs (4 in directory)			F	review	
	🖞 ИЅВ	1. Test 1		Name:	m information Test 1 ype: 48 well Pro	gram Type: 1	-os
	Test	2. Test 2			°C Preheat Lid 1 Runtime: 01h (
			Step	°C	h:mm:ss	Go To	
			1	95.0	0:05:00		
	Admin	3. Test 3		2	95.0	0:00:30	
				3	59.0 🚽 61.0	0:00:30	
				4	72.0	0:00:30	30x Step 2
		4. Test 4			16.0		
	Home	CODY		JV. Delet	States and States of	Edit	Start

Figure 28 Biometra TRIO program overview screen with program preview

In the multiblock selection query, select the sample block to start and confirm your selection.

Start progran	า	
Please select block to start:		
✓ Block 1 ✓ Block 2 ✓ Block 3		×

Figure 29 Biometra TRIO multiblock selection query

The program will be started on the selected sample blocks.

Note For programs including a TOS step (see chapter "Set Temperature Optimisation Step (TOS)" p. 47) all blocks are started simultaneously and therefore the block selection option is not offered.

8.2 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 "Edit user" p. 81).

ENABLE A USER TO EDIT, COPY AND DELETE OWN PROGRAMS	Write/delete own programs	
ENABLE A USER TO EDIT, COPY AND DELETE ALL PROGRAMS OF OTHER USERS	Write/delete all programs	

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 18). 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 the [COPY] button.



Touch the corresponding buttons to select a user directory and a storage place in the Biometra TRIO program overview screen (see Figure 30). 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 the [SAVE] button.



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.

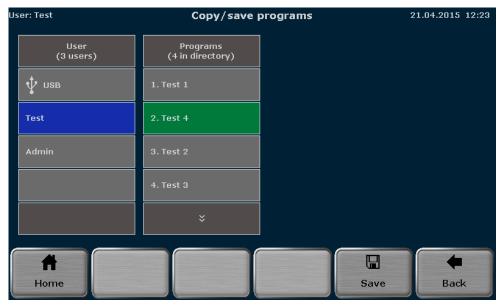


Figure 30 Biometra TRIO program overview screen

8.3 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 "Edit user" p. 81).

ENABLE A USER TO EDIT, COPY AND DELETE OWN PROGRAMS	Write/delete own programs	
ENABLE A USER TO EDIT, COPY AND DELETE ALL PROGRAMS OF OTHER USERS	Write/delete all programs	

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 30). 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 the [COPY ALL] button.



Touch the corresponding button to select a user directory in the Biometra TRIO program overview screen. 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 the [SAVE] button.



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

User: Test	Copy/save	programs	2	1.04.2015 12:27
User (3 users)				
🖞 иѕв				
Test				
Admin				
Home			Save	Back

Figure 31 Biometra TRIO program overview screen

8.4 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 "Edit user" p. 81).



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 30). 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 the [DELETE] button.



Confirm the security query to delete the program.

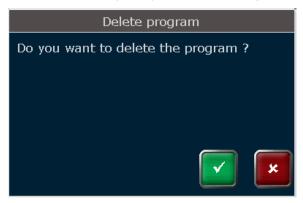


Figure 32 Biometra TRIO security query

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.

8.5 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 "Edit user" p. 81).



ENABLE A USER TO EDIT, COPY AND DELETE ALL PROGRAMS OF OTHER USERS

Write/delete all programs

To delete all programs, touch the corresponding button to select a user directory in the Biometra TRIO program overview screen (see Figure 30). 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 the [DELETE ALL] button.



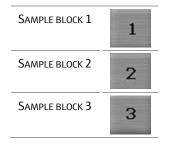
Confirm the security query to delete all programs.

Delete all programs
Do you want to delete all programs from the chosen user?

Figure 33 Biometra TRIO security query

9 Running, pausing, continuing and stopping programs

To switch between the view for the sample blocks during run select the number of the sample block in the corresponding screen.



9.1 Display during run

After a program is started (see chapter "Display during run" p. 58), the Biometra TRIO software can display the running program in spreadsheet or graphical view. Use the following buttons to toggle between both modes.

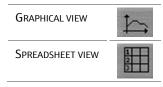




Figure 34 Biometra TRIO run screen in graphical view

User: Tes	st						21.04.2015 12:38								
		Name		Name		Estin	nated End	Remain. [.]	Гime	Bloc	ck Temp.	Lid		Step	
		Test	1	13	3:52:30	01h 14m	16s	ç	95.0°C	99.4%	c	1/5			
		Step	Block T (°C		Hold Time (h:mm:ss)		Су	cles	ΔT(°C)	∆t(s)	۵	R(°C/s)			
		2	95.	0	0:00:30		-					6.0			
2	30x	3	59.	0	0:00:30		-		1.0	1		1.0			
3		4	72.	0	0:00:30	2	3	80				6.0			
		5	16.	0	œ		-					6.0			
Но	H me					l l use) Sk	and the second second	Choose stop pr			p program n block 1		

Figure 35 Biometra TRIO run screen in 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 "Set preheating mode" p. 43), 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 Δ T, Δ t or ! are displayed (see Figure 34).

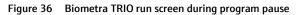
9.2 Pause Program

To pause a program press the [PAUSE] button in the Biometra TRIO run screen (see Figure 34 and Figure 35).



The word PAUSE is shown and in the field remaining run time the pause time is counted.

er: Te	at .				Plo	ck 1			21.04	.3:37 User: T	oct		Block 1			21.041	2015 13:3
		Nam Test		stimated End 14:56:24	Remain 01h 18r	. Time	Block Temp 95.0°C	. Lid	Step	0.07 0361.1	Name Test 5	Estimated End		Block Temp. 95.0°C	Lid 99.2°C	Step	1010 10.0
		Step	Block Ter (°C)	np. Hold Time (h:mm:ss	: Go To	Cycle	s ∆T(°C)	∆t(s)	∆R(°C/s)	h	Step 1	Step 2	Step 3 S	tep 4 St	ep 5	Step 6	
											100 95.0 0:01:34	95.0					
		2	95.0	0:00:30							2 80 80				2.0		
2	30×		60.0	0:00:30			1.0	1	1.0	2	an 60 40		0:00:30		105.00		
		4	72.0	0:00:30	2	30			6.0	3	du 40				1		
		5	72.0	0:05:00					6.0		20					15.0 00	
		6	15.0	- 00					6.0		0		Cycles 30x				
								-									
	me		Continu	e						ram +	forme Con	tinue				Stop	



Note: To press the pause button requires manual interaction. The Biometra TRIO software also allows to program a pause step (see chapter "Edit parameter or step" p. 44), then the pause step is automatically started by the instrument.

Note: For programs including a TOS step 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 "Stop Program" p. 61) before switching off the instrument.

Continue Program 9.3

To continue a paused program press the [CONTINUE] button in the Biometra TRIO run screen during program pause (see Figure 36).



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 "Set Temperature Optimisation Step (TOS)" p. 47) the program is continued on all blocks simultaneously.

9.4 Skip Step

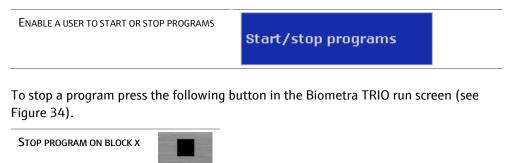
To skip a step program press the [SKIP] button in the Biometra TRIO run screen (see Figure 34).



The Biometra TRIO Thermal Cycler will immediately continue with the next program step and skips the current step.

9.5 Stop Program

Note: Programs can only be stopped by users with the right START/STOP PROGRAMS activated (see chapter "Edit user" p. 81).



If two or more sample blocks are running the Biometra TRIO thermal cycler offers the option to stop multiple sample blocks simultaneously. Press the following button in the Biometra TRIO run screen (see Figure 34).

CHOOSE BLOCK STOP PROGRAM	
---------------------------	--

Activate the corresponding check boxes to select the sample blocks to stop.

The [CHOOSE BLOCK STOP PROGRAM] button for multiblock selection is only available if more than one sample block is running.

Stop progra	m	
Please select block to stop	:	
 ✓ Block 1 ✓ Block 2 ✓ Block 3 		×

Figure 37 Biometra TRIO query for multiblock selection

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





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

10 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 the protocol wizard button in the Biometra TRIO home screen (see chapter "Home Screen" p. 32).



10.1 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.

User: Test		Se	lect/Ed	it Poly	merase ter	mpla	te	23	3.04.2015 13:09
	(Polymerase 2 in directory)			P	reviev	v		
	Innu	Taq		Program information Name: TaqPolymerase Block Type: 48 well Lid: 99 °C Preheat Lid: On					
	TaqP	olymerase					°C	h:mm:s	c .
				Initial I	Denaturation		98.0	0:05:00	
					naturation		95.0	0:01:00	
					nnealing			0:00:10)
					ongation		2.0		
					Elongation		2.0	0:05:00)
			_	Fina	al Storage		10.0	00	
		_ →	-	-					-
Home		New Polymerase	Dele Polym		Edit Polymeras	se	Ba	ck	Next

Figure 39 Biometra TRIO polymerase selection screen

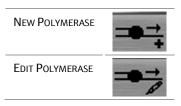
Pre-installed in the software is the program template for Analytik Jena Innupure polymerases. To create a PCR program select a polymerase from the list and press:



In the next screen (see chapter "Enter New Polymerase or edit Polymerase" p. 62) enter values for the requested parameters to continue with the automatic program creation.

10.2 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:



In the next screen (see Figure 40) 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:

- 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 "Protocol Wizard parameter screen" p. 65).
- The holdtime for the elongation step becomes calculated by the expected product length (see chapter "Primer Annealing Temperature Calculator" p. 66).

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

User: Admin	l de la companya de l	Edit polym	ierase			22.12.2	2014 12:56
Name:	InnuTaq						
		Temperature [°C]	Holdtime (h:mm:ss)				
	Initial denaturation	95.0	0:05:00		2		
	Denaturation	95.0	0:01:00		2	3	
	Annealing		0:00:10	4	5	6	Del
	Elongation	72.0			8	9	Tab
	Final elongation	72.0	0:05:00				
	Final storage	10.0	00		0	-	00
			1				
Home			Done	Ba	ck		

Figure 40 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:



In the next screen (see Figure 41) 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.

User: Admin		Select pol	ymerase		22.12.2014 13:25
	Polymerase (8 in directory)				
	InnuTaq				
	Super Taq				
	InnuTaq				
	InnuTaq				
	×				
A				-	
Home			Save	Back	

Figure 41 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:

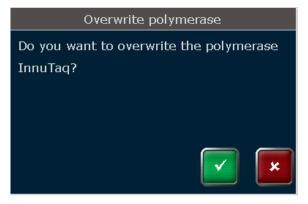


Figure 42 Biometra TRIO security query

10.3 Delete Polymerase

To delete a polymerase press:



10.4 Protocol Wizard parameter screen

For automatic calculation of PCR programs in the protocol wizard parameter screen (see Figure 43) some parameters have to be set by the user:

 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:

Elongation time [s] = product length [bases]/1000 [bases]*60s 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 "Primer Annealing Temperature Calculator" p. 66).

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.

User: Test	Protoco	l Wizard			23.04.2	2015 12:52
Polymerase Template:	InnuTaq					
PCR Method:	Two step	🗹 Three st	ер			
Product length:	500 bp		1	2	3	$\overline{\mathbf{X}}$
Primer Annealing Temp. (Ta = Tm - 5°C)	: 55.0 °C	Calc Ta	4	5	6	Del
Amplification Cycles:	30 x		7	8	9	Tab
			·	0		8
Home			В	¢ ack		➡ Next

Figure 43 Biometra TRIO protocol wizard parameter screen

After setting all program parameters press:



Test		View parameter	r	23	.04.201
Po	olyı	nerase Template: InnuTaq			
			Temperature [°C]	Hold Time (h:mm:ss)	
		Initial Denaturation	95.0	0:05:00	
		Denaturation	95.0	0:01:00	
з	30×	Annealing	55.0	0:00:10	
		Elongation	72.0	0:00:30	
		Final Elongation	72.0	0:05:00	
		Final Storage	10.0	00	

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

Figure 44 Biometra TRIO protocol wizard view parameter screen

Press the [CREATE PROGRAM] button to enter the Biometra TRIO programming screen (see chapter "Creating, editing and saving programs" p. 39).

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 7.9.

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 45 Biometra TRIO security query

10.5 Primer Annealing Temperature Calculator

Based on the primer sequences the Biometra TRIO protocol wizard can calculate the theoretical primer annealing temperature (T_a) . Press the [CALC TA] button in the Bi-

ometra TRIO protocol wizard parameter screen (see Figure 43) to access the primer annealing temperature calculator screen (see Figure 44).



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.

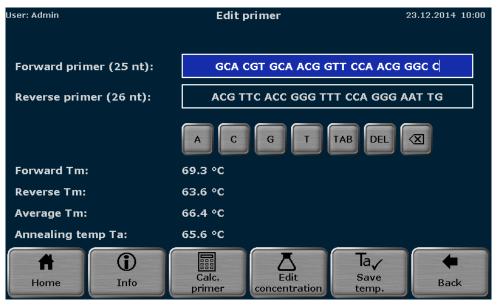


Figure 46 Biometra TRIO primer annealing temperature calculator screen

To adjust the primer or salt concentration press:



 $\overline{\Delta}$

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

User: Admin		Edit concentr	ation			23.12.2	014 10:01
Primer conce	entration	200.0 nM					
Salt concent	ration:	50.0 mM		1	2	3	$\overline{\mathbf{X}}$
Magnesium o	oncentration:	mM		4	5	6	Del
Forward Tm:		69.3 °C	ſ	7	8	9	Tab
Reverse Tm:		63.6 °C					
Average Tm:		66.4 °C			0	-	00
Annealing te	mp Ta:	65.6 °C	C.				
	i		ACGÍ	Ta	~		+
Home	Info	Calc. primer	Edit primer	Sa ten			Back

Figure 47 Biometra TRIO screen for primer concentration adjustment

Press the [EDIT PRIMER] button to toggle to the primer annealing temperature calculator screen (see Figure 47).



Or press the [CALC. PRIMER] button 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 the [SAVE TEMP.] button to transfer the calculated primer annealing temperature to the protocol wizard (see Figure 46).



The calculated T_a value will be displayed in the input box for the primer annealing temperature.

Press the [INFO] button 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

11 Tools

The Biometra TRIO software provides the following tools:

Settings	Define basic instrument settings	See chapter 11.1
User Management	Tool for the user management	See chapter 11.2
Documentation	Access to stored log files	See chapter 11.3
EXTENDED SELF-TEST	Execute extended self-test	See chapter 11.4
SERVICE INFO FILE (SINF)	Create Service Info File	See chapter 11.5
Васкир	Saves all folders, programs, users and user settings to USB	See chapter 11.6
Cycler Info	Provides general information on the instrument	See chapter 11.7
Солтаст	Biometra contact data	See chapter 11.8



Figure 48 Biometra TRIO tools screen

Press the corresponding button to access the desired function.

11.1 Settings

Note: System settings can only be modified by users with the right SYSTEM CONFIGURATION activated (see chapter "Edit user" p. 81). 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 11.1.1
AUTOMATIC USER	Switch automatic user logout on or off	See chapter 11.1.2
LOGOUT	and time settings	
Beeper	Switch beeper on or off	See chapter 11.1.3
Network	Define network settings or list net-	See chapter 11.1.4
	work users	
DISPLAY BRIGHTNESS	Adjust display contrast	See chapter 11.1.5
TOUCH SCREEN	Calibrate touch screen	See chapter 11.1.6
CALIBRATION		
FACTORY SETTINGS	Reset the settings	See chapter 11.1.7

User: Admin	Set	tings	22.10.2014 15:13
() Date and time	Automatic user logout	H Beeper	Network
Display brightness	F∳ Touch screen calibration	Factory settings	
Home			F Back

Figure 49 Biometra TRIO settings screen

Press the corresponding button to access the desired function.

11.1.1 Set Date and Time

Note: Date and time can only be edited by users with the right "SYSTEM CONFIGURATION" activated (see chapter "Edit user" p. 81).

ENABLE A USER TO CONFIGURE THE SYSTEM	
CONFIGURATION	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.

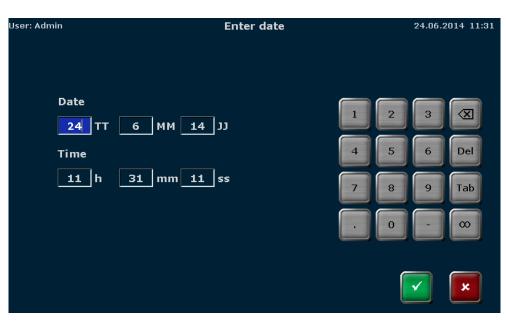
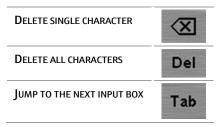
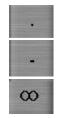


Figure 50 Biometra TRIO date and time screen

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:



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



11.1.2 Automatic user logout

Note: The automatic user logout can only be configured by users with the right "SYSTEM CONFIGURATION" activated (see chapter "Edit user" p. 81).



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 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.

User: Admin Autor	natic user logout sett	tings		22.10.2	2014 15:12
Automatic user logout	30 (30-60min)	1	2	3	$\overline{\boxtimes}$
		4	5	6	Del
		7	8	9	Tab
			0		
				~	×

Figure 51 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:



11.1.3 Configure Beeper

Note: The beeper can only be configured by users with the right SYSTEM CONFIGURATION activated (see chapter "Edit user" p. 81).

ENABLE A USER TO CONFIGURE THE SYSTEM		
CONFIGURATION	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 52 Biometra TRIO beeper configuration screen

11.1.4 Network

The NETWORK menu item can be used to configure basic settings or show a list of network users. Press the corresponding button:

	NETWORK SETTINGS
Network settings	Note: The network settings can only be configured by users with the right SYSTEM CONFIGURATION activated (see section "Edit user" p. 81).
	ENABLE A USER TO CONFIGURE THE SYSTEM CONFIGURATION System configuration
	The Biometra TRIO can handle static and dynamic IP addresses. Activate the corre- sponding checkbox to choose between dynamic (DHCP) and static IP address man- agement.
	If DHCP (see Figure 54) is activated, the device automatically receives the network configuration settings. If the static checkbox is activated, you can enter the IP address and the subnet mask using the keypad on the right side of the screen. The port number can entered manually in both setting types (static and DHCP).
	The following buttons may be helpful when completing the fields:
	DELETE SINGLE CHARACTER
	Delete all characters Del
	GO TO THE NEXT INPUT FIELD Tab

User: Admin	Network settings			25.05	.2016 14:45
Cycler name:	Cycler 2				
IP settings:	Assign IP address automatica	illy (DH	CP)		
	Static	1	2	3	
IP Address:	192 . 168 . 1 . 2	4	5	6	Del
Subnet Mask:	255 . 255 . 255 . 0	7	8	9	Tab
Port:	55555		0		
				Image: A start of the start	×

Figure 53 Network settings screen, IP settings via static

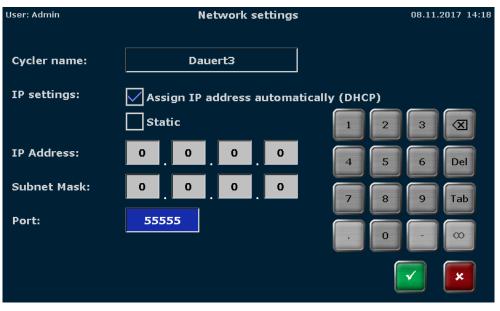


Figure 54 Network settings screen, IP settings via DHCP

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

Network usersThe software stores the users who access the Biometra TRIO via the network and lists
them in chronological order in a table. The function makes it possible to check if only
authorized persons have network access to the device.

PCR control App The app is available for iOS (Apple App Store) and Android (Google Play Store) driven systems.

The PCR Control App offers the following functions:

- Live monitoring of PCR runs on network-integrated thermal cyclers- Starting and stopping PCR protocols are stored on the thermal cycler
- Notification system for monitoring status changes on selected blocks of a thermal cycler
- Store PCR protocols on the terminal. This can be stored on a PC using a USB stick

- Copy PCR protocols from the terminal to a selected thermal cycler
- Read device-specific parameters
- Read out thermal cycler error messages

11.1.5 Display brightness

Note: The display brightness can only be edited by users with the right "SYSTEM CONFIGURATION" activated (see chapter "Edit user" p. 81).



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



Or use the horizontal slider.

User: Test		Set display brightness		17.06.2014 12:39
	Dark	_	Light	

Figure 55 Biometra TRIO display brightness settings screen

11.1.6 Touch screen calibration

Note: The touch screen can only be calibrated by users with the right SYSTEM CONFIGURATION activated (see chapter "Edit user" p. 81).

ENABLE A USER TO CONFIGURE THE SYSTEM	
CONFIGURATION	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.

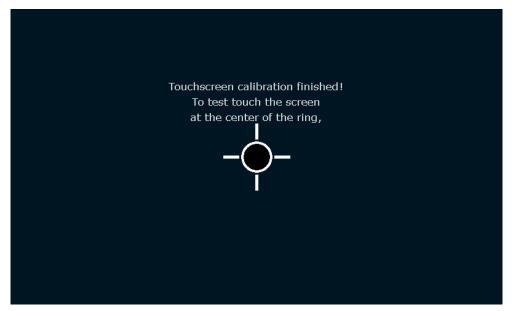


Figure 56 Biometra TRIO touch screen calibration screen

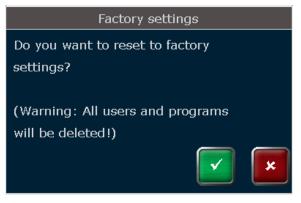
11.1.7 Factory settings

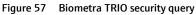
Note: The factory settings tool can only be executed by users with the right SYSTEM CONFIGURATION activated (see chapter "Edit user" p. 81).

ENABLE A USER TO CONFIGURE THE SYSTEM
CONFIGURATION
Sys

System configuration

Press the corresponding button and confirm the following security query:





By using the factory settings tool all user accounts and programs will be deleted! **Note**: Use a backup file (see chapter "Backup" p. 92) to restore the system.

11.2 User Management

The Biometra TRIO thermal cycler manages up to 90 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 the [USER MANAGEMENT] button in the Biometra TRIO user management screen (see Figure 58).



Note: The button 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 login.

User: Mike		l	Jser Man	agement 05.05.2017 13:45
	No.	Username	Initials	Group
	1	Mike	мі	Administrator
	2	Heiner	HE	Administrator
	з	Admin	ADM	Administrator
Hom	е	Edit New	V User	Remove

Figure 58 Biometra TRIO user management screen

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

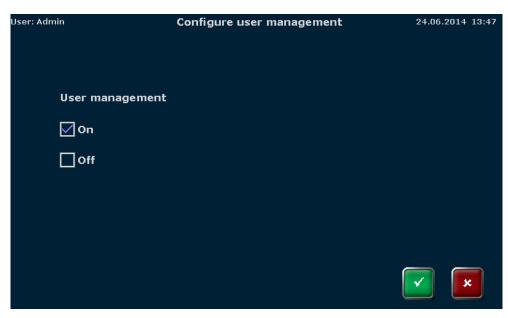


Figure 59 Biometra TRIO configure user management screen

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 "Edit user" p. 81) by the administrator and will be stored by the Biometra TRIO software.

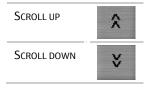
Note: If the user management is switched off, user accounts and programs are not protected and can be modified or deleted by others.

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

Administrator

- User with generic rights
- User with limited rights
- User defined

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



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

11.2.1 Create user

Note: New users can only be created by users with the right CREATE USER activated (see chapter "Edit user" p. 81).

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:



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

Use the Biometra TRIO keyboard (see chapter "Keyboard" p. 23) and set the language by activating the corresponding check box.

>>	User properties	
]

Fiaure 60	Biometra TRIC) create user screen

Password (optional):

Username:

Language:

Initials:

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.

English German Chinese

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.

11.2.2 Edit user

Note: User rights can only be edited by users with the right EDIT USER activated.



To edit the user properties select a user account in the user management screen and press the button:

EDIT USER ACCOUNT



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

	User properties	
Username:	Admin	
Initials:	ADM	
Password (optional)	****	
Verify password:	****	
Language:	🗹 English	
	German	
	Chinese	

Figure 61 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:

- Administrator
- User with generic rights
- User with limited rights

For each user group templates defining the user rights are available. However, the predefined 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 "User Management" p. 78).



Figure 62 Biometra TRIO user properties screen

Right	Function				
System configuration	GURATION Provides access to several system settings like date and time, screen saver, beeper, networ 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 lim- ited users and is not allowed to modify the rights settings. An administrator is allowed to cre- ate all user types and to modify the rights settings for each user.				
Delete user	Allows to delete existing user accounts				
WRITE/DELETE PROGRAMS OF OTHER USERS	Allows to edit/copy and delete all programs of all users				
READ ALL PROGRAMS OF OTHER USERS	If inactive 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	Allows to start or stop programs				
WRITE/DELETE OWN PROGRAMS	Allows to edit/copy and delete own programs but not those of other users.				
SHOW YOUR OWN PROGRAMS TO OTHER USERS	Allows users to hide their own programs from other users.				

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 the [USER DATA] button to go back to the create user screen.



11.2.3 Delete user

Note: User accounts can only be deleted by users with the right DELETE USER activated (see chapter "Edit user" p. 81).



To delete a user select the user account in the user management screen (see chapter "User Management" p. 78) 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 63 Biometra TRIO security query

The user and all of his programs will be deleted.

11.3 Documentation

The Biometra TRIO software offers the following documentation tools:

Run-Logfile	Documentation of PCR runs	See chapter 11.3.1
Power On Logfile	Results of the latest initialisation test	See chapter 11.3.2
Self-Test Logfile	Results of extended self-tests	See chapter 11.3.3
Error Logfile	Lists all errors detected by the instrument	See chapter 11.3.4



Figure 64 Biometra TRIO documentation screen

Press the corresponding button to access the desired function.

11.3.1 Run-Logfile

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

User:	Test	Overview Run Log Files 25.05.2016 15:0							
	No.	Date	Time	User	Block	Program	Dir.	Program No.	
		25.05.2016	15:08:34	TES	1	Three-step	TES	01	
	2	25.05.2016	14:18:35	ADM	1	Linear-grad-1c	ADM	01	
	з	25.05.2016	14:18:00	ADM	1	Linear-grad-1c	ADM	01	
	4	25.05.2016	14:17:52	ADM	1	Linear-grad-1c	ADM	01	
	5	25.05.2016	14:16:25	ADM	1	Linear-grad-1c	ADM	01	
	6	25.05.2016	13:57:19	ADM	1	Linear-grad-1c	ADM	01	¥
	,							·	_
	Ħ				→↔	∄→ ↔	≣/☆		
H	lome				ected Lo e to USE		View tun Log F	-ile Ba	ick

Figure 65 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
- Cycler type

- Serial number Cycler
- Block type
- Block serial number
- Block number
- Messages
- Serial number Cycler
- Block type

User: Test	Run Log Fi	le	28.04.2015 11:36
Run Log File:	TET 02 Test 1		
Log Time and Date:	17:33:49 21.04.2015		
Start-Endtime/Date:	17:33:49 21.04.2015 - 1	8:02:34 21.04.2015	
Company:	Biometra GmbH		
Software Version:	0.35		
User:	TET		
Cycler Type:	Biometra Trio	Serial Number Cycler:	2101119
Block Type:	48 well	Block Serial Number:	20112344
Block:	3		
Messages:	None		
Home		المركز	H Back

Figure 66 Biometra TRIO Run-Logfile screen

To view the corresponding program press button:



The Biometra TRIO offers the option to show the program in graphical from or as spreadsheet. To toggle between graphical and spreadsheet view use buttons:

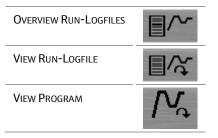
GRAPHICAL VIEW	
Spreadsheet view	1

Tools



Figure 67 Biometra TRIO View program screen

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



To export Run-Logfiles to USB press:



Select a logfile from the list and press the following button in the Run-Logfile overview screen (see Figure 66).



Use [SELECTED RUN-LOGFILE TO USB] to save single Run-Logfiles to USB. Use [ALL RUN-LOGFILES TO USB] to save all Run-Logfiles to USB.

11.3.2 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

	st			Power On Logfile 28.0	4.2015 12:47	
Powe	er On:		28.0	4.2015 11:34:52		
Last	Power O	n:	15.0	4.2015 03:11:00		
ast	Power D	own:	15.0	4.2015 05:17:26		
No.	Date	Time	Error	Message		
1	28.04.2015	11:54:34	300	The temperature program on Block 1 was paused or stopped		
2	28.04.2015	11:54:38	300	The temperature program on Block 3 was paused or stopped		
з	28.04.2015	11:54:43	300	The temperature program on Block 2 was paused or stopped		
4	28.04.2015	11:56:03	300	The temperature program on Block 1 was paused or stopped		
5	28.04.2015	11:56:06	300	The temperature program on Block 3 was paused or stopped		

Figure 68 Biometra TRIO Power-On-Logfile screen

11.3.3 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 figure below). Press on the corresponding button to see the results for a specific test summarized in tabular format.

User: Te	est		Sel	f Test Log File		28.04.2	015 12:48
	ts poler rview cod	Thermal tracking oler test	Heat./cool. rate	Refrigerati. G	iradient	Heated Lid Re	gulation
No.	Date	Time	Block Type	Serial Number	Block	Test result	
	28.04.2015	12:04:19	48 well	20112344	All	Passed	
2	28.04.2015	11:43:54	48 well	20112344	All		
Н	A ome						4 Back

Figure 69 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.

11.3.4 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.

ser: Test			est Error Log File					
No.	Date	Time	Error	Message				
	28.04.2015	11:56:17	300	The temperature program on Block 2 was paused or stopped				
2	28.04.2015	11:56:06	300	The temperature program on Block 3 was paused or stopped				
3	28.04.2015	11:56:03	300	The temperature program on Block 1 was paused or stopped	he temperature program on Block 1 was paused or stopped			
4	28.04.2015	11:54:43	300	The temperature program on Block 2 was paused or stopped				
5	28.04.2015	11:54:38	300	The temperature program on Block 3 was paused or stopped	The temperature program on Block 3 was paused or stopped			
6	28.04.2015	11:54:34	300	The temperature program on Block 1 was paused or stopped				
1 Ho	ome				F Back			

Figure 70 Biometra TRIO Error-Logfile screen

11.4 Extended Self-Test



NOTICE

The Extended Self-Test must be performed under the following conditions to achieve reproducible and meaningful results:

- Ambient temperature in the range 20 °C to 25 °C
- Mains voltage depending on the voltage type of the device: 100 V/115 V/230, ±10 % each
- The device must be cooled to the ambient temperature of 20 °C to 25 °C after the last application.

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 coordi- nated 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.

Test	Function
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 71). 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.

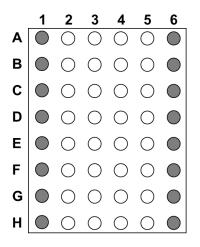


Figure 71 How to load plastic ware for Extended Self-Test

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

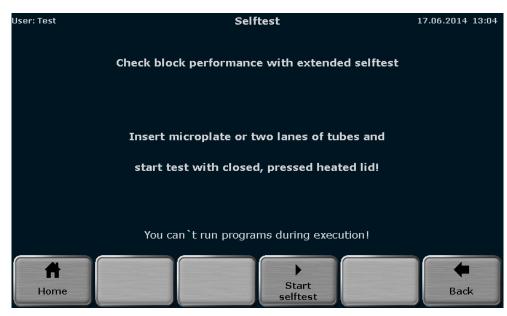


Figure 72 Biometra TRIO Extended Self-Test screen

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:



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

User: Test	Self Test		28.04.2015 11:44
	Block 1	Block 2	Block 3
Cooler test:	Running	Running	Running
Thermal tracking test:			
Heating/Cooling rate test:			
Refrigeration test:			
Gradient test:	No gradient	No gradient	No gradient
Heating lid test:			
Regulation test:			
Home	Cancel Self Test		Back

Figure 73 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 "Extended Self-Test-Logfile" p. 88).

11.5 Service Info File (SINF)

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

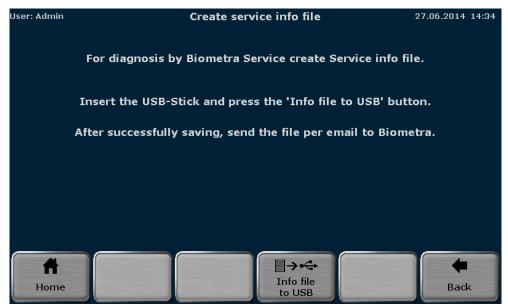


Figure 74 Biometra TRIO create Service Info File screen

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 11.8.

11.6 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 the [SAVE BACKUP FILE] button to create a backup file.

SAVE BACKUP FILE	<u></u>]→↔
------------------	-------------

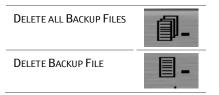
ser: Adr	min			Backup 30.06.201					
N	No.	Date	Time		Name				
	1	30.06.2015	16:45:36	BUP30.06.15_16:45:36					
	_								
1		D	elete all	Delete backup	•� → 🗐 Load backup	□→ ↔ Save backup	+		
Ho	me		ckup files		file	file	Back		

Figure 75 Biometra TRIO backup file screen

To load a backup file select a backup from the list and press:



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



11.7 Cycler Info

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

- Cycler type
- Company
- Serial number Cycler
- 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 the info button to toggle between the two screens.

System/Cycler Info

Test	Cycler Info	28.04.2015 11:45				
Cycler Type:	Biometra Trio					
Company:	Biometra GmbH					
Serial Number Cycler:	2101119					
Block Type:	48 well					
Block Serial Number:	20112344					
Software Version:	0360 - 0.35 - 0.63					
Protocol Version:	2. 0. 0. 0 - 0. 5. 0. 0 - 0. 5. 1. 0					
H	G					
Home	System	Info Back				

Figure 76 Biometra TRIO cycler info screen

11.8 Contact

Shows contact details for the Biometra service department.

User: Admin	Contact	09.11.2017 07:50
Biometra GmbH Rudolf-Wissell-Strasse 30 37079 Göttingen Germany 智 +49 (0)551 / 50686-0 昌 +49 (0)551 / 50686-66 図 info@analytik-jena.com www.analytik-jena.com	Contact	09.11.2017 07:5
Home		H Back

Figure 77 Biometra TRIO contact details screen

12 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 alumi- num block and therefore exceeds the block's maximum gradient range.	The temperature range of the gradient will be re- duced based on the average temperature of the max- imum permissible value. If the average temperature of the programmed gradient is, for example, 60° C and the temperature range is ± 20°C, it will be re- duced to 60° C ± 15°C.
A program including a gradient step is trans- ferred to a non- gradient instrument.	The program is including a gradient step started on a block without gra- dient function.	The gradient will be deleted and the average tem- perature/annealing temperature will be used for this step. If the average temperature/annealing tempera- ture 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 pro- grammed 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 func- tion	A TOS program is transferred from the Biometra TRIO to another ther- mal 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 pos- sible 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 alumi- num block is started on a 96 well silver block.	The ramp rate will be adjusted to the maximum pos- sible 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.

13 Short Manual



WARNING

Before initial operation check the correct operating voltage is set on the voltage selection switch on the underside of the device. Do not open the housing! Danger of electric shock!



CAUTION

The sample block and the heated lid can reach high temperatures during operation. Both the sample block and the heated lid can burn you. The samples are heated up quickly and may come to an explosive boil. Wear goggles when handling hot samples. Make sure that the lid is closed securely before starting the program!



NOTICE

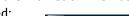
The use of oil between samples and sample block is not necessary. If you wish to use oil, never use silicone oil. Use mineral oil. Ensure that the ventilation slits are clear.

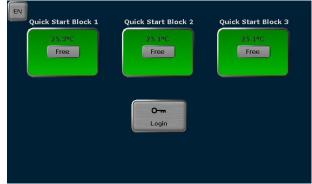
Important: Release the lid wheel before opening the lid! Adjust the lid wheel for every individual run!

Log in

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



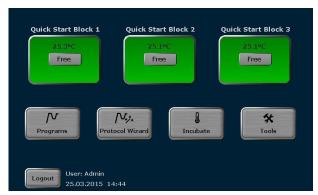




- Press [LOGIN] to log in as existing user
- Press [QUICK START] for the non-user specific quick start of programs
- Press [EN/DE/CN] to set the login screen language

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:

User: Admin	Program overview	15.09.2014 15:03
User (9 users)	Programs (4 in directory)	
🖞 иѕв	1. Test 1	
Admin	2. Test 2	
Test	3. Test 3	
Heinz	4. Test 4	
×		
Home New fi	om Convell Delete ell	

- 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

lame:			Test 1			.id: 99	• •c		Prehea	t Lid: On
	rogr	am Rur	time: 01h 17	m 50s			~			
	,		Block Temp. (°C)) <u>b</u>
ſ		2	95.0	0:00:30					6.0	
	30×	3	59.0 4 61.0	0:00:30			1.0	1	1.0	
		4	72.0	0:00:30	2	30			6.0	
		5	16.0	~~~~					6.0	
-			E.		-	Г	-			
Hor	ne		Edit	Ins	+ ert	Del	- ete	Do	ne	Back

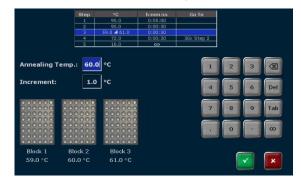
Graphical programming screen



- 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:

				Go To			
	1	95.0	0:05:00		Â		
		95.0	0:00:30				
	3	59.0 4 61.0	0:00:30				
	4	72.0	0:00:30	30x Step 2	¥		
	5	16.0	60				
Temperature: Hold Time: Go To:	95. 0 		mm 30 es:	55 7	2 5 8 0	3 6 9 -	Del Tab
Temp. incr./dec	r.:	l∘c			ſ	v	×

- 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 the 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 and press [DELETE]. In the security query confirm to delete the step by pressing the button ✓.
- 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].

Save program

Start, pause and stop

programs

Save program					copy belete programs				
Jser: Admin	Copy/save p	Copy/save programs 22.10.2014 15:32		User: Admin		Program	overview		22.10.2014 16:0
User (3 users)	Programs (2 in directory)			User (3 use	rs)	Programs (2 in directory)			
🖞 изв	1. Linear-grad-1c			🖞 изв	1. Li				
Admin	2. Three-step			Admin	2. TI				
				Test					
Home		and the second se	Gave Back	Home	New from template	Copy all	Delete all		

- 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]. 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.
- 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 home screen. Select a program from the list and press [START].
 - To start a program from a user directory press the button [PROGRAMS] in the 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

Graphical view



- 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].
- To stop the active program press the button [STOP PROGRAM] button and confirm the security query. Note: Programs with a pause in the last step have to be stopped manually as well.

Switch user management on or off (only aministrator)

In the home screen press [TOOLS], in the next screen the button [USER-MANAGEMENT]. Then use the checkboxes to switch the user management on or off.

Conv/Delete programs

14 Fault removal

14.1 Forgotten password

Passwords can be changed by the systems administrator(s) and by users with the activated right EDIT USER (see chapter "Edit user" p. 81). If the administrator Admin has forgotten the password and no one else has the right EDIT USER use the function FACTORY SETTINGS (see chapter "Factory settings" p. 77) to reset the system and load a backup file (see chapter "Backup" p. 92) to restore it.

14.2 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 cycler 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.

14.3 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 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.

14.4 Auto restart due to unrecognized power failure

High voltage fluctuation can lead to an automatic restart of the Thermal Cycler. In this case the cycler 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. To avoid voltage fluctuation, do not connect the cycler to a socket shared by a strong power consumer like a refrigerator or acentrifuge.

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

14.5 Mirror programs to several thermal cyclers

Using the backup function, complete thermal cyclers can be mirrored. Copy complete users (via user management) and their programs from one thermal cycler to the other without having to create each individual user or program. The prerequisite is that both cyclers have the same software version (see chapter 11.7).

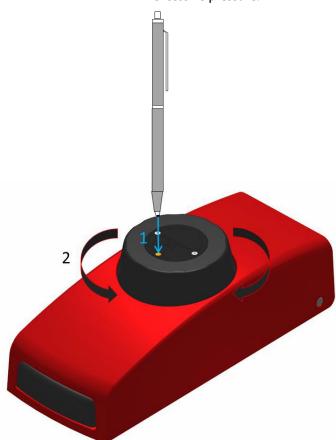
14.6 Adapt protocols from other cyclers

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 7.6.2. Alternatively, the time settings may be extended.

14.7 Release 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.
- 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.
- 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. This leads to damage of tubes and instrument!

15 Maintenance and Repair

15.1 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 9001:2015 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 "Technical specifications" p. 107), a temperature verification or calibration after installation is not required.

15.2 Servicing and Repair

The Biometra TRIO is mainly maintenance-free. The care and maintenance tasks which can be performed by the customer are limited to the cleaning and disinfection of the housing and sample block.

All maintenance work and repairs beyond this scope must only be performed by Biometra GmbH customer service personnel or authorized and trained persons. Any unauthorized intervention limits warranty entitlements. If the device exhibits any faults or defects, please contact the Biometra GmbH customer service department immediately.

In order to guarantee sound and safe operation and to ensure laboratory certification, we recommend concluding a maintenance agreement with regular device validation.

15.3 Cleaning



WARNING

Risk of electric shock! Prior to commencing any maintenance or cleaning work, switch off the device and pull out the mains plug. After cleaning, wait until the Biometra TRIO is completely dry before recommissioning it.



WARNING

If you are handling infectious and pathogenic materials observe the safety regulations. Decontaminate the device in accordance with section "Disinfecting the device" p. 103.

The professional decontamination of radioactive contaminations depends on the type of substance used. Please contact your radiation protection officer on this matter and observe the relevant safety regulations!



NOTICE

The use of alcohol, organic solvents or abrasives can damage the paintwork.

- Only wipe the housing of the Biometra TRIO with a soft clean cloth which may be slightly wetted with a commercial neutral detergent, if necessary.
- Clean the ventilation slits on the underside and at the rear of the device with a vacuum cleaner.

15.4 Disinfecting the device



WARNING

Biological hazard! Clean the Biometra TRIO with particular care after analyzing potentially infectious material. Wear suitable protective equipment, e.g., protective gloves.



NOTICE

The only suitable cleaning method for the housing is wipe disinfection.

When using spray disinfectants there is a risk that the liquid enters the sensitive electronic system through the ventilation slits. If the disinfectant has a spray nozzle, apply disinfectant to a suitable cloth before using it on the device.

- Avoid contamination by handling sample substances with care.
- Wipe spilled samples or reagents immediately with an absorbent cloth or piece of paper.
- If the Biometra TRIO is used for the analysis of infectious material, great care must be taken because the TRIO cannot be decontaminated as a whole device.
- Immediately remove visible contamination by suitable means. Do not allow solvents to enter the device.
- The sample block is also suitable for wipe and spray disinfection. The only suitable cleaning method for the housing is wipe disinfection.

Device part	Recommended disinfectants	Provider
Sample chamber	Descosept AF	Dr. Schuhmacher GmbH
	Meliseptol HBV (cloths)	B. Braun
Housing	Descosept Spezial	Dr. Schuhmacher GmbH

15.5 Firmware update

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

15.6 Spare parts

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

16 Service

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

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

16.1 Instructions for return shipment



WARNING

Risk of damage to health due to improper decontamination! Perform a professional decontamination before returning the device to Analytik Jena.



NOTICE

Biometra GmbH must refuse acceptance of contaminated devices. The sender may be liable for any damage caused by inadequate decontamination of the device.

- Please clean all device components from biologically hazardous, chemical or radioactive contamination (see also section "Disinfecting the device" p. 103).
- Download the decontamination declaration as an editable PDF document in German or English from the Internet: https://www.analytikje-

na.com/fileadmin/content/service/customer/Declaration_of_decontamination_en_01.pdf.

Complete the form and attach the signed decontamination declaration to the outside of the shipment.

- Only use the original packaging for the shipment and insert the transport lock. If the original packaging is no longer available, please contact Biometra GmbH or your local dealer.
- Please attach the warning note
 "CAUTION! SENSITIVE ELECTRONIC DEVICE!" to the packaging.
- Please include a sheet containing the following data:
 - Name and address of the sender
 - Name and telephone number of a contact for inquiries
 - A detailed description of the fault, the precise conditions and situations under which the fault occurs
 - 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!

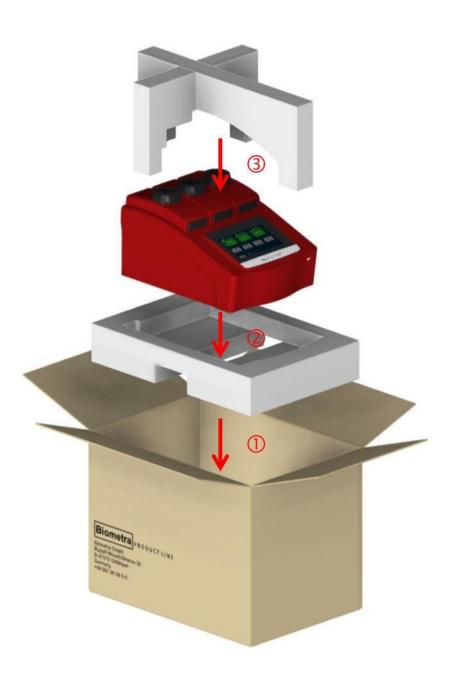
16.2 Packing of the Biometra TRIO Thermal Cycler

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



NOTICE

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.



17 Disposal

The operator of the Biometra TRIO must dispose of any waste materials that occur during measurements (sample materials) in accordance with statutory and local regulations.

At the end of its service life, the Biometra TRIO and all its electronic components must be disposed of as electronic waste in accordance with valid regulations.

18 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)		
	95°C	+/- 0.60 °C after 15 s		
Temperature Uniformity	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 90 user directories		
Language		English, German, Chinese		
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 to 60 Hz		
Noise emission		Very low		
Interfaces ¹		USB A, Ethernet		

Order Number	846-x-070-720	846-x-070-723	846-x-070-724	
Working conditions	15 °C to 35 °C, 70 % air	15 °C to 35 °C, 70 % air humidity, max 2000 m NN		
Dimensions (WxDxH)	30 cm x 41 cm x 25 cm	30 cm x 41 cm x 25 cm		
Weight	17.3 kg	17.3 kg		
* Measured inside the block				

** Capacity increases to 35×0.5 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 230 V, 4 for 115 V, 5 for 100 V

19 Declaration of Conformity

analytikjena

An Endress+Hauser Company

EU – Konformitätserklärung EC - Declaration of Conformity

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Göttingen, den 21.09.2018

Biometra erklärt als Hersteller in alleiniger Verantwortung, dass die Produkte Biometra declares as manufacturer under sole responsibility that the products

Typen / <i>types:</i>	Biometra TRIO 30	Biometra TRIO 48	Biometra TRIO Combi		
BestNr. / <i>Order No.:</i>	846-x-070-720	846-x-070-723	846-x-070-724		
0/02/100.	x = 2 für/for 230 V, 4 für/for 115 V, 5 für/for 100 V				

den folgenden Europäischen Richtlinien und angewandten harmonisierten Normen entsprechen: *conform to the following European Directives and applied harmonized standards:*

Richtlinie /	Norm /	Ausgabejahr /
<i>Directive</i>	<i>Standard</i>	<i>Year of Publication</i>
2014/35/EU	EN 61010-1	2010
Niederspannungsrichtlinie / <i>LVD</i>	EN 61010-2-10	2014
2014/30/EU EMV / <i>EMC</i>	EN 61326-1 EN 55011 EN 61000-3-2 EN 61000-3-3	2013 2009 2014 2013
2011/65/EU RoHS	EN 50581	2012

Dr. Juergen Otte

Head of Quality Management For and behalf of Biometra GmbH



Certificate number FS 68563

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