

Whatman **Biometra®**



Manual

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050-801 TGradient 96

Biometra

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1 Introduction

The TGRADIENT is the new high end thermocycler in the Biometra product line. It was our goal to develop an instrument that provides fast optimization of reaction parameters as well as a high performance for daily routine analysis. Both could be realized by a completely new block architecture and the latest Peltier technology.



To increase the specificity of a reaction, the annealing temperature is one of the crucial parameters. With the new TGRADIENT different annealing temperatures can be tested within a single run. A gradient with a maximum range of 40°C can be set at any program step. We have taken strong efforts to create a gradient as linear as possible. At the same time it was very important to ensure homogenous temperature distribution within each row. The temperature data for each row of the block can be retrieved through the help menu.

The new TGRADIENT is an outstanding instrument. In addition to the advanced temperature gradient it provides high speed and reliable accuracy for routine applications. The silver block features very high heat conductivity and ensures fast ramping speeds and a fast temperature equilibration.



The new TGRADIENT will speed up your laboratory work because it saves time not only in optimizing new reactions but also in your routine work.

2 Before you start

2.1 Safety warnings

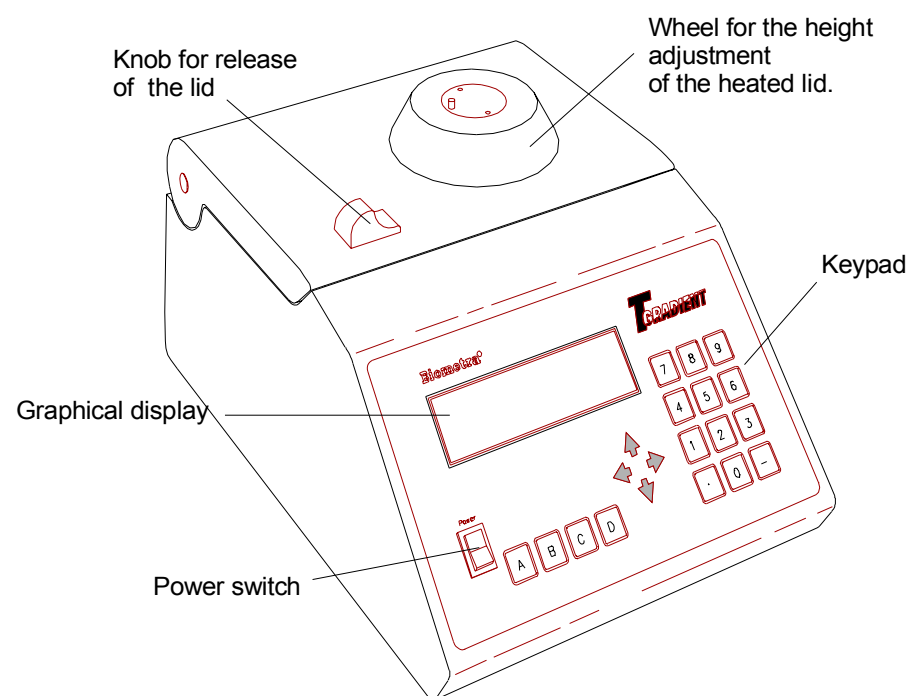
| | |
|---|---|
|  | Before connecting the unit to the power source please ensure that the voltage selector at the back side of the instrument is set to the required voltage. Danger of electric shock! Unplug the power cable before you open the TGRADIENT Thermocycler. |
|  | It is not necessary to utilize oil into the opening of the block in order to improve the heat transfer between the block and the sample tubes. If you still decide to use oil, do not use silicon oil. Mineral oil may be used. |

2.2 Warning symbols

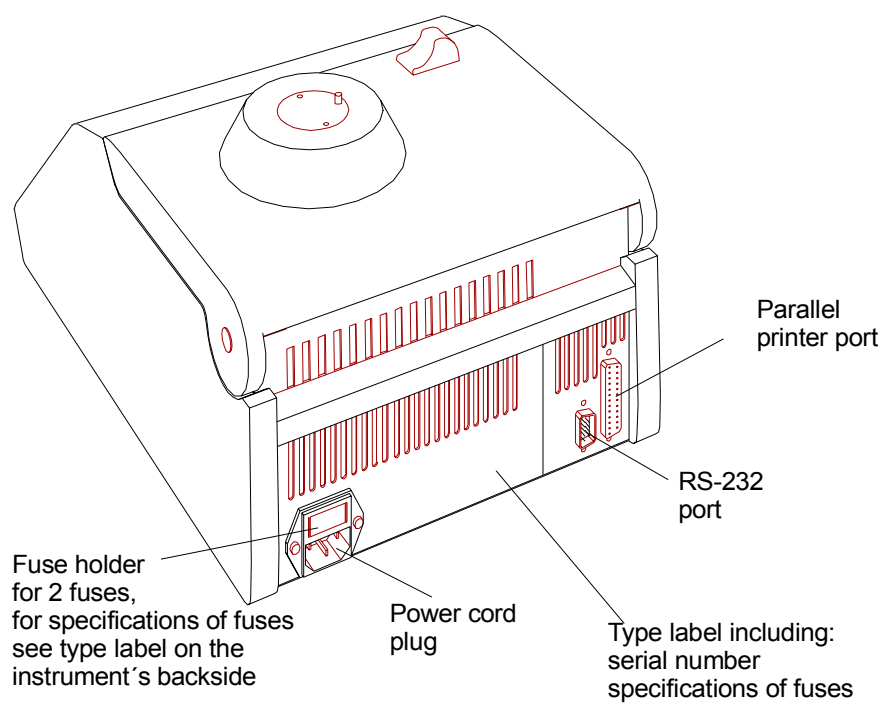
| | |
|--|---|
|  | The thermoblock and the heated lid will reach high temperatures during operation. Both thermoblock and heated lid can burn you. Rapid heating of the thermoblock can cause liquids to boil explosively. Always wear safety goggles during operation. Close the lid before starting a program. |
|  | 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. |

3 First steps with the TGRADIENT

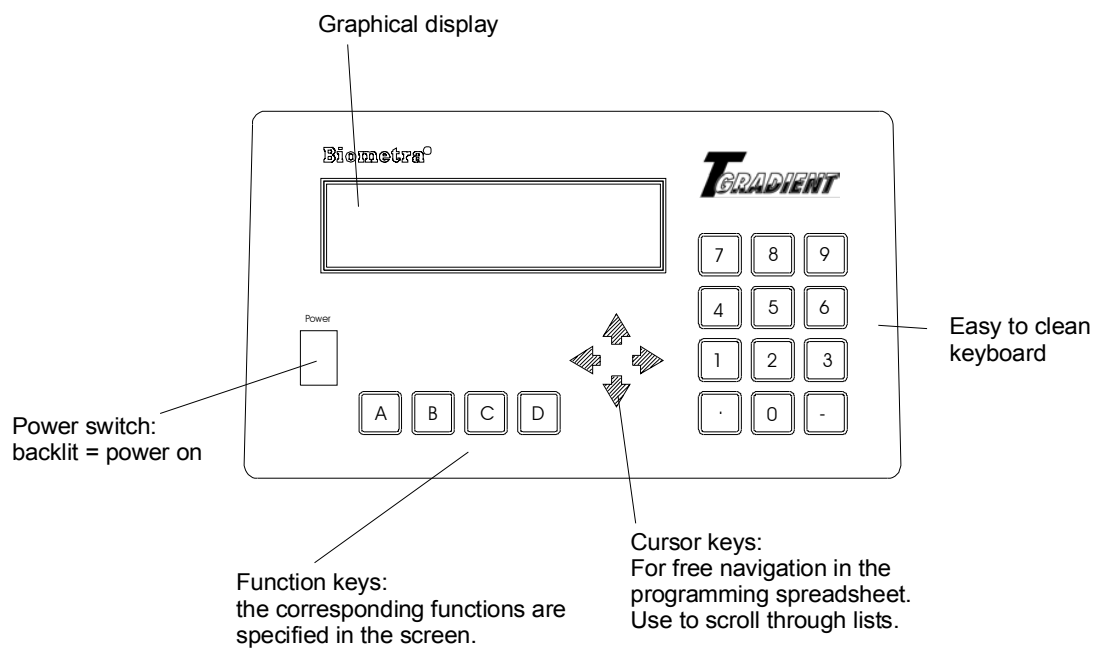
3.1 The TGRADIENT Thermocycler front View



3.2 The TGRADIENT rear view



3.3 The TGradient control panel



3.4 Initial self test

After switching on the TGRADIENT the serial number of the instrument and the software version is displayed.

```
TGradient
Serial No 1234567
Vers. 4.00 op
```

The TGRADIENT checks all programs in all directories (so called RAM check).

3.5 The TGradient display

The TGRADIENT display provides information about the instrument and about the status of the active program.

```
prog
step
temp [°C]    25.2°C
time
lid          25.4°C
A ?         B start/stop  C programs  D +
```

The individual function of the special keys **A**, **B**, **C** and **D** are specified in the display and depend on the context.

3.6 Handling of the adjustable heated Lid

To achieve optimum pressure on the tubes the TGRADIENT is equipped with an 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.

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

Open the heated lid:

First: Release pressure by turning the wheel counter clockwise. As soon as there is no more resistance the pressure has been released. **Then:** now you can open the lid with the knob.

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

3.7 Releasing wheel in case of blocked lid

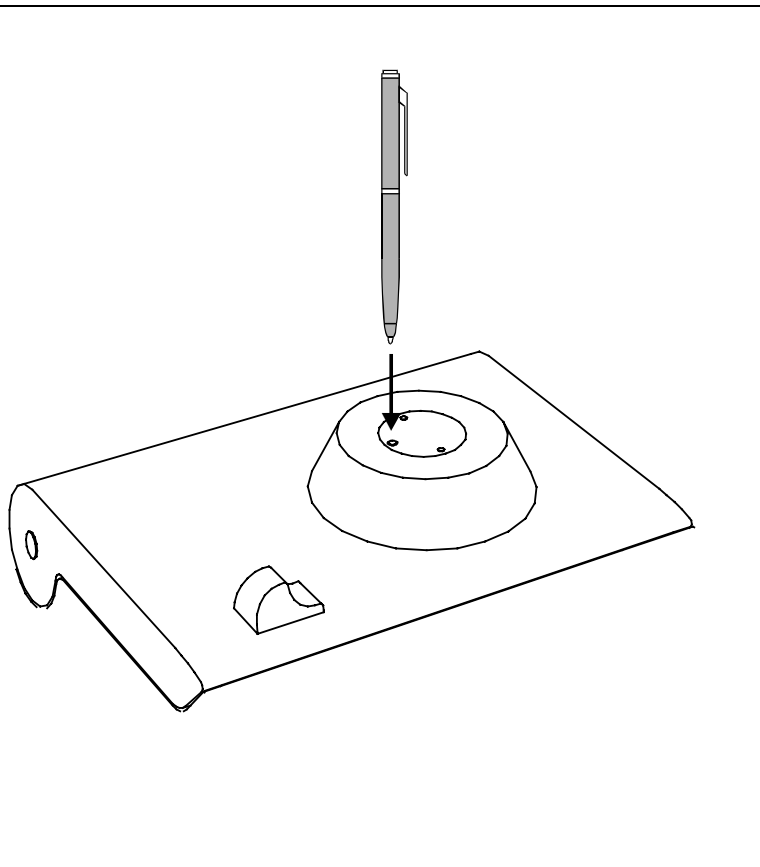
Note: When the lid is in the extreme up or down position, it may happen that the wheel is uncoupled. In this situation the clutch mechanism is active in both directions (clicking noise in either direction). To unlock wheel, press metal pin with a ballpen and turn wheel carefully. This pin overcomes the automatic clutch mechanism. Thus, care must be taken not to apply excessive pressure.

Lid arrested in upper position:

carefully turn wheel with pressed pin 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).

Lid arrested in down position:

carefully turn wheel with pressed pin 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), NEVER USE PIN TO FURTHER INCREASE PRESSURE. THIS WILL LEAD TO DAMAGE OF TUBES AND INSTRUMENT!

4 Creating a Program

4.1 Select a directory

In the TGRADIENT programs can be stored both in the main directory and in individual subdirectories. For easy identification of the selected storage place, the subdirectories can be named (see section 4.3).

Start with the main screen.

```
Prog
step
temp [°C]    25.2°C
time
lid          25.4°C
A ?         B start/stop  C programs  D +
```

Press [C programs] to enter the edit mode.

```

Main direct.--- -3. subdirect.:
                  -4. subdirect.:
                  -5. Subdirect.:
                  -6. subdirect.:
                  -7. subdirect.:
A ?             B          C quit   D enter
```

You are now in the main directory. The structure of the subdirectories is displayed.

4.2 Select a program store

Programs can be stored in the main directory or in subdirectories.

To create a program in the main directory press [D enter].

To access a subdirectory move right with the cursor key \Rightarrow and select a subdirectory with the cursor keys $\uparrow\downarrow$. The selected directory will be highlighted.

```

Main direct.--- -3. subdirect.:
                  -4. subdirect.:
                  -5. Subdirect.:
                  -6. subdirect.:
                  -7. subdirect.:
A ?             B          C quit   D enter
```

4.3 Enter name for subdirectory

The process for naming a subdirectory and a program is the same. For information about naming programs see section 4.4.

Press [D enter] to enter the subdirectory

```

directory:          5
program no.         Name:

edit
A list      B          C quit      D enter

```

Press [A list] to view a list of all existing files and empty memory in this directory.

Note: If neither program name nor the word “empty” is displayed, this place may be occupied by a program without name. For the programming of new protocols choose an empty memory. Existing programs that are not needed any more can be deleted to provide new memory.

```

0 empty
1 empty
2 empty
3 empty
4 empty
A list      B forward      C quit      D enter

```

Scroll through the list with cursor keys $\uparrow\downarrow$ and confirm memory with [D enter].

4.4 Enter a program name

Each program is specified by a program number and a subdirectory number. To make retrieval of a program easier, you can enter a name for each program existing of letters, numbers and symbols.

```

Directory:          3
program no.         0      Name: 
                        ^
                        ABCDEFGHIJKLMNOPQRST
                        UVWXYZ-()#°C/,⟨⟩&+.%!
A ABC      B files      C name OK      D enter

```

To enter a name for your program press [A ABC].

```

Directory:          3
program no.         0      Name: 
                        ^
                        ABCDEFGHIJKLMNOPQRST
                        UVWXYZ-()#°C/,⟨⟩&+.%!
A name      B blank      C quit      D enter

```

Choose a letter from the alphabet with cursor keys $\leftarrow\rightarrow\uparrow\downarrow$

Confirm each letter with [D enter].

```
Directory:      3
program no.    0      Name:   T
                  ^
                ABCDEFGHIJKLMNOPQRST
                UVWXYZ-()#°C/,<>&+.%!
[A name] [B blank] [C name OK] [D enter]
```

The selected letter will appear in the display and the cursor will move one step to the right. Select the next letter and confirm with [D enter].

Note: What to do if a wrong letter has been accepted?

To alter the position of the cursor within the name press [A name]. Now you can move the cursor with cursor keys ⇐⇒ through the name. This is helpful if you have confirmed an wrong letter or if you want to change the name of an existing program.

Once the name is complete, confirm name with [C name OK].

```
Directory:      3
program no.    0      Name:   TEST 1
lid temp: [ ] °C      preheating: on
edit
[A ?] [B files] [C pgm OK] [D enter]
```

4.5 Enter the lid temperature

You can now enter a temperature for the heated lid. **Note:** In general, the lid temperature should be about 10°C above the highest temperature in the protocol. Example: If your denaturation step is at 95°C, enter 105°C for the lid.

```
Directory:      3
program no.    0      Name:   TEST 1
lid temp:   105 °C      preheating: on
edit
[A ?] [B files] [C pgm OK] [D enter]
```

4.6 Select / deselect pre-heating of the lid

You can choose whether the lid is pre-heated before the program starts. This is to avoid condensation during the initial heating phase. **Note:** During the preheating of the lid, the block is held constant at 25°C.

```

Directory:      3
program no.    0      Name:  TEST 1
lid temp:     105 °C   preheating: on
edit
A on/off  B files      C pgm OK  D enter

```

Note: The default setting, which is recommended for most applications, is preheating: on.

After all pre-settings have been made, press [D enter] to open the programming spreadsheet.

4.7 Enter temperature and time settings

The programming spreadsheet:

```

Temp [°C]  time  ←  #  gradient [°C]  opt →
1
2:
3:
4:
A ?      B insert/delete  C pgm OK  D enter

```

Note: In this spreadsheet you can enter all parameters for your cycling protocol. Furthermore, you can set special parameters like touch down, ramping rates or time increments. For further details on these special parameters, see section 5.1.

Note: You can navigate in the spreadsheet without limitations with the four cursor keys
 ⇐⇒⇑⇓.

Note: Each setting is confirmed with [D enter]. The cursor moves automatically to the next field. Alternatively, you can confirm a value by moving forward with the cursor keys.

Note: At any time you can call up the help function with [A ?]

Note: In an existing protocol program steps can be deleted and inserted. For further information on deleting and inserting program steps see section 5.1.

Now enter temperature for the first step in the protocol:

```

Temp [°C]  time  ←  #  gradient [°C]  opt →
1  95,0
2:
3:
4:
A ?      B insert/delete  C pgm OK  D enter

```

Confirm temperature with [D enter] or just move on with the cursor key. If you move to the next row, the setting will be stored in the temporary memory.

| Temp [°C] | time | ← | # | gradient [°C] | opt | → |
|-----------|-------------------|---|---|---------------|-----|-----------|
| 1: 95.0 | | | | | | |
| 2: | | | | | | |
| 3: | | | | | | |
| 4: | | | | | | |
| [A] ? | [B] insert/delete | | | [C] pgm OK | | [D] enter |

Enter a time for step one.

Note: There is a convention on how time settings are entered in all BIOMETRA cyclers:
hours • minutes • seconds

If you enter a number without “point” this value will be interpreted as seconds (“300” => 5 minutes). To program minutes enter a “.” after the number of minutes. To enter hours enter “.” after the number. You can also enter any combination of hours, minutes and seconds. Example: for 1 hour, 30 minutes, 20 seconds enter 1•30•20.

The time values will be displayed in the following format: 0h 00 m 00s

Accept time setting with [D enter] or move to the next field with cursor key.

4.8 Set temperature gradient

Note: Each temperature gradient is defined by two settings: **first:** the temperature in the middle of the block, **second:** the range of the gradient. For example if you want to program a temperature gradient from 50 to 60°C, enter 55°C as temperature and 10°C for the gradient.

| Temp [°C] | time | ← | # | gradient [°C] | opt | → |
|-----------|-------------------|---|---|---------------|-----|-----------|
| 1: 95.0 | 0h 5m 0s | | | | | |
| 2: 95.0 | 0h 0m30s | | | | | |
| 3: 55.0 | 0h 0m30s | | | 10°C | | |
| 4: | | | | | | |
| [A] ? | [B] insert/delete | | | [C] pgm OK | | [D] enter |

Note: The maximum temperature gradient is 40°C.

Note: You can program as many steps with temperature gradient as you want.

Note: You can even set an inverse temperature gradient. An inverse gradient has the higher temperature on the left side of the block and the lower temperature on the right side respectively. Inverse gradient are set by adding the algebraic sign “-“ to the gradient value.

4.9 View different temperatures of the gradient in the block

To view the temperatures in the 12 different block rows press [A ?]

| Temp [°C] | time | ← | # | gradient [°C] | opt | → |
|-----------|---|---|---|----------------|-----|---|
| T1 - 6: | 50.0 50.2 50.9 | | | 52.0 53.2 54.4 | °C | |
| T7 -12: | 55.6 56.8 58.0 | | | 59.1 59.8 60.0 | °C | |
| 3: | 55.0 0h 0m30s | | | 10°C | | |
| 4: | Temperature difference left side / right side | | | | | |

In the first highlighted lane the temperatures from **row 1 to 6** are displayed. In the second highlighted lane the temperatures **from row 7 to 12** are displayed.

Note: While the TGRADIENT is cycling you can view the current gradient temperatures. The temperatures can be displayed online during each gradient step (see section 7.4). As the program moves to the next step the last gradient temperatures are stored. Thus you can check the temperatures of the last gradient in consecutive steps without gradient. For further information on the current status of the cycler see section 7.

4.10 Set cycle number

Note: In general, loops are defined by selecting the target for the back loop and the number of backloops.

| Temp [°C] | time | ← | # | gradient [°C] | opt | → |
|-----------|-----------------|----------|---------|---------------|-----|---|
| 1: | 95.0 0h 5m 0s | | | | | |
| 2: | 95.0 0h 0m30s | | | | | |
| 3: | 55.0 0h 0m30s | | | | | |
| 4: | 72.0 0h 0m30s | | 2 | | | |
| A ? | B insert/delete | C pgm OK | D enter | | | |

In the field labeled with ← you can select the target step for a backloop.

| Temp [°C] | time | ← | # | gradient [°C] | opt | → |
|-----------|-----------------|----------|---------|---------------|-----|---|
| 1: | 95.0 0h 5m 0s | | | | | |
| 2: | 95.0 0h 0m30s | | | | | |
| 3: | 55.0 0h 0m30s | | | | | |
| 4: | 72.0 0h 0m30s | 2 | 29 | | | |
| A ? | B insert/delete | C pgm OK | D enter | | | |

Enter the number of backloops. **Note:** total cycler number = (n backloops) + 1, e.g. enter 29 for a total number of 30 cycles.

4.11 Cool below ambient temperature

| Temp [°C] | time | ← | # | gradient [°C] | opt | → |
|-----------|-----------------|----------|---------|---------------|-----|---|
| 2: | 95.0 0h 0m30s | | | | | |
| 3: | 55.0 0h 0m30s | | | | | |
| 4: | 72.0 0h 0m30s | 2 | 29 | | | |
| 5: | 4.0 | pause | | | | |
| A ? | B insert/delete | C pgm OK | D enter | | | |

To hold the temperature for an indefinite time enter “0”. After pressing [D enter] “pause” is displayed.

Note: The minimum temperature you can set is -3°C .

4.12 Save program

Save program with [C] pgm OK]. The program data are stored in the permanent memory.

| | | | |
|------------------|----------|-------|--------|
| Directory: | 3 | | |
| program no. | 0 | Name: | TEST 1 |
| number of steps: | 5 | | |
| runtime: | 1h10m23s | | |
| edit | | | |
| [A] | [B] | [C] | [D] |

The runtime is calculated and displayed.

Note: To view the current remaining runtime at any time of the run press [A] info] in the main menu.

5 View/edit program data

You can check any existing program by scrolling through the editing mode with cursor keys $\leftarrow \rightarrow \uparrow \downarrow$. Select the desired program as described in section 4.2.

Note: During operation of the cycler the active program can be viewed but not modified. If you want to change settings of the active program for further experiments you have to save a copy of this program to another memory. For further information about copying programs see section 5.2.

5.1 Delete / insert program steps

The programming spreadsheet:

| Temp [°C] | time | ← | # | gradient [°C] | opt → |
|------------|------------------------|-----------------|----------------|---------------|-------|
| 2: 95.0 | 0h 0m30s | | | | |
| 3: 55.0 | 0h 0m30s | | | | |
| 4: 72.0 | 0h 0m30s | 2 | 29 | | |
| 5: | | | | | |
| A ? | B insert/delete | C pgm OK | D enter | | |

To delete or insert a program step press [**B** insert/delete]

| | | | | | |
|-----------------------|-----------------------|---------------|----------------|--|--|
| insert / delete step: | | | | | |
| 2: 95.0 | 0h 0m30s | | | | |
| 3: 55.0 | 0h 0m30s | | | | |
| 4: 72.0 | 0h 0m30s | 2 | 29 | | |
| 5: | | | | | |
| A ? | B delete input | C quit | D enter | | |

Enter the number of the step that you want to insert / delete.

Note: Additional steps will be inserted before the selected step number. The following steps will be shifted by one position.

| | | | | | |
|-------------------------|-----------------------|---------------|----------------|--|--|
| insert / delete step: 3 | | | | | |
| 2: 95.0 | 0h 0m30s | | | | |
| 3: 55.0 | 0h 0m30s | | | | |
| 4: 72.0 | 0h 0m30s | 2 | 29 | | |
| 5: | | | | | |
| A ? | B delete input | C quit | D enter | | |

Note: If you have entered a wrong number, the field can be cleared with [**B** delete input].

Confirm the step number with [**D** enter].


```

insert / delete step: 3
2: 95.0 0h 0m30s
3: 55.0 0h 0m30s
4: 72.0 0h 0m30s 2 29
5:
A insert B delete C quit D enter

```

Press **[A insert]** to insert one step at the selected position.

Note: Additional steps will be inserted before the selected step. The following steps will be shifted by one position.

Press **[B delete]** to delete the selected step.

Note: By deleting a step the following steps will be shifted by one position.

Note: The insertion and deletion of steps may have an influence on existing cycles. Be sure that all settings are updated to maintain a correct cycle.

5.2 Copy program

Select the program you want to copy as described in section 4.1 and 4.2

```

Directory:      3
program no.    0      Name:  TEST 1
lid temp:     105°C   preheating: on

A ? B files C pgm OK D enter

```

Press **[B files]**.

```

Directory:      3
program no.    0      Name:  TEST 1
lid temp:     °C     preheating: on

edit
A copy B delete pgm C quit D enter

```

To copy this program to another memory press **[A copy]**

```

copy to:      -3. subdirect.:
              -4. subdirect.:
Main direct.--- -5. Subdirect.:
              -6. subdirect.:
              -7. subdirect.:
A ? B C quit D enter

```

Select the desired subdirectory with the cursor keys $\Rightarrow \Uparrow \Downarrow$. The selected subdirectory will highlight.

```

copy to:      -3. subdirect.:
              -4. subdirect.:
Main direct.---5. Subdirect.:
              -6. subdirect.:
              -7. subdirect.:
[A] ?        [B]          [C] quit      [D] enter

```

Confirm your selection with [D enter].

```

copy from directory 3 program 0
      to   directory 6 program 
[A] list      [B]          [C] quit      [D] enter

```

Choose an empty memory from the list with [A list].

```

copy from directory 3 program 0
      to   directory 6 program 6
                        enter = OK

target program contains 0 steps
[A] list      [B]          [C] quit      [D] enter

```

Note: Before you confirm with [D enter] the target memory is checked. If there is no program in the target, memory the message “target program contains 0 steps” is displayed. If the memory already contains a program the number of steps is displayed.

Note: If you copy a program to a memory that already contains a program, the former program will be deleted!

Save a copy of the program in the new directory with [D enter].

5.3 Delete program

Select program as described in section 4.1 and 4.2

```

Directory:      3
program no.     0      Name:  TEST 1
lid temp:       °C      preheating: on

edit
[A] copy      [B] delete pgm  [C] quit      [D] enter

```

To delete this program press [B delete pgm].

```
Directory:      3
program no.    0      Name:  TEST 1
lid temp:     °C      preheating: on

delete program 0?      enter = yes
A copy      B delete pgm  C quit      D enter
```

Confirm your selection with [D enter].

Note: Once you have deleted a program it cannot be restored!

6 Further programming options

| Temp [°C] | time | ← | # | gradient [°C] | opt → |
|-----------|----------|---|---|---------------|---------|
| 1: 95.0 | 0h 5m 0s | | | | |
| 2: 95.0 | 0h 0m30s | | | | |
| 3: 55.0 | 0h 0m30s | | | | |
| 4: 72.0 | 0h 0m30s | | 2 | | |
| A ? | B header | | | C pgm OK | D enter |

Note: To program special parameters you can scroll the display to the right.

Move the cursor to the right side of the display that is labeled with opt →.

The following screen will appear:

| Main funct. | dT [°C] | dt [s] | ↗ [°/s] |
|-------------|----------|----------|---------|
| 1: ← | | | 5.00 |
| 2: ← | | | 5.00 |
| 3: ← | | | 5.00 |
| 4: ← | | | 5.00 |
| A ? | B header | C pgm OK | D enter |

Note: To get back to the screen with the basic parameters move the cursor to the left side of the display labeled with “Main funct.” Alternatively you can move the cursor to the extreme right.

| Temp [°C] | time | ← | # | gradient [°C] | opt → |
|-----------|----------|---|---|---------------|---------|
| 1: 95.0 | 0h 5m 0s | | | | |
| 2: 95.0 | 0h 0m30s | | | | |
| 3: 55.0 | 0h 0m30s | | | + | |
| 4: 72.0 | 0h 0m30s | | 2 | | |
| A ? | B header | | | C pgm OK | D enter |

Note: In the main screen you will find an information that a special parameter has been modified. This is indicated by a + in the row labeled with opt →.

6.1 Program time increments

To compensate for loss in enzyme activity each step within a loop can be extended from cycle to cycle. Enter the desired time increment [seconds] in the row labeled with dt[s]. This value will be added to the time value from cycle to cycle.

| Main funct. | dT [°C] | dt [s] | ↗ [°/s] |
|-------------|----------|----------|---------|
| 2: ← | | | 5.00 |
| 3: ← | | | 5.00 |
| 4: ← | | 10 | 5.00 |
| 5: ← | | | 5.00 |
| A ? | B header | C pgm OK | D enter |

Note: The time increment will have an impact on the total runtime depending on the numbers of cycles and the size of the increment. A program with many cycles and large time increments will take a significantly longer time than a standard protocol.

6.2 Program touch down

For some applications it is useful to start with a higher annealing temperature and to decrease the temperature from cycle to cycle. This subsequent lowering of the annealing temperature is called touch down.

To decrease the annealing temperature from cycle to cycle enter a negative temperature increment in the row labeled with dT[s].

| Main funct. | dT [°C] | dt [s] | ↗ [°/s] |
|-------------|----------|----------|---------|
| 2: ← | | | 5.00 |
| 3: ← | | | 5.00 |
| 4: ← | -0.20 | | 5.00 |
| 5: ← | | | 5.00 |
| A ? | B header | C pgm OK | D enter |

Note: Be sure that the temperature decrease is set in a step that lies within a loop. Otherwise there will be no iterative temperature decrease.

6.3 Adjust heating and cooling ramps

Since the TGRADIENT is a extreme fast thermocycler it may be necessary to reduce the heating and cooling ramp for some applications. For example this is helpful if cycling protocols from other cyclers shall be used.

The heating or cooling ramp respectively can be set in the row labeled with ↗[°/s].

| Main funct. | dT [°C] | dt [s] | ↗ [°/s] |
|-------------|----------|----------|---------|
| 2: ← | | | 5.00 |
| 3: ← | | | 5.00 |
| 4: ← | | | 5.00 |
| 5: ← | | | 5.00 |
| A ? | B header | C pgm OK | D enter |

Note: The default is [5.00 °/s] which means maximum ramping speed.

7 Run Program

7.1 Select and start program

Main display:

```

Prog
step
temp [°C]    25.2°C
time
lid          25.4°C
[A] ?       [B] start/stop [C] programs [D] +

```

To select a program press [B] start/stop]

```

start          | -3. subdirect.:
Main direct.--- | -4. subdirect.:
               | -5. Subdirect.:
               | -6. subdirect.:
               | -7. subdirect.:
[A] ?          [B]          [C] quit    [D] enter

```

Choose directory with cursor keys ⇨ ⇧ ⇩ or enter main directory with [D] enter]

Note: The path for the program that was modified most recently is automatically pre-selected

```

               | -3. subdirect.:
Main direct.--- | -4. subdirect.:
               | -5. Subdirect.:
               | -6. subdirect.:
               | -7. subdirect.:
[A] ?          [B]          [C] quit    [D] enter

```

Enter subdirectory with [D] enter]

```

Directory:  5
program no. [ ]      Name:

Edit
[A] list    [B]          [C] quit    [D] enter

```

Enter the number of the program you want to start. Alternatively, [press A] list to select a program from the list of all programs in this subdirectory.

Scroll through the list with cursor keys ⇧ ⇩.

```

0 test 1
1 empty
2 empty
3 empty
4 empty
A list      B forward      C quit      D enter

```

Confirm highlighted program with [D enter].

The number and name of both subdirectory and the selected program will be displayed.

```

Directory:      3
program no.    0      Name:  TEST 1

                        preheating lid on
start block                        signal on
A list      B                        C quit      D start

```

To start the program press [D start].

7.2 Display during operation

During preheating of the lid the following screen is displayed:

```

Prog      0 TEST 1      dir 3
Step      1
temp[°C]  25.0°C      → 95.0
time
lid      preheating
A info    B start/stop  C programs  D +

```

Note: In the default setting (preheating on) the block is held constantly at 25°C. The program starts as soon as the lid has reached the set temperature. During the preheating phase the display of the lid temperature alternates between the current lid temperature and “preheating”.

```

Prog      0 TEST 1      dir 3
Step      1
temp[°C]  25.0°C      → 95.0
time
lid      preheating
A info    B start/stop  C programs  D +

```

As soon as the block has reached the programmed hold temperature, the timer starts to count down.

```

Prog      0 TEST 1      dir 3
Step      1      cycle 1 of 30
temp[°C]  95.0°C      → 95.0
time      0h 2m15s    → 0h 5m 0s
lid      104.9°C
A info    B start/stop  C programs  D +

```

The elapsed time versus programmed hold time is displayed

```

Prog      0 TEST 1          dir 3
Step      1                cycle 1 of 30
temp[°C]  49.9 55.0 60.0 → 50.0 55.0 60.0
time      0h 0m23s        →0h 5m 0s
lid       104.9°C
[A] info  [B] start/stop  [C] programs  [D] +

```

As soon as the block enters a gradient step the three temperatures of the left, middle and right sides of the block are displayed.

7.3 View remaining run time

Display during operation:

```

Prog      0 TEST 1          dir 3
Step      1                cycle 1 of 30
temp[°C]  49.9 55.0 60.0 → 50.0 55.0 60.0
time      0h 0m23s        →0h 5m 0s
lid       104.9°C
[A] info  [B] start/stop  [C] programs  [D] +

```

Press [A info] to call up the current remaining time calculation.

```

Additional information about active pgm
Status:                               Step 2
                                       Cycle 3 of 30
Remaining time: 2h 57m
Stop or pause: key [B] in main menu
[A] gradient  [B]      [C] quit      [D]

```

The current remaining run time is displayed in line 4.

7.4 View current gradient temperatures

While the TGRADIENT is cycling you can view the current gradient temperatures. To retrieve detailed information about the temperatures in the different rows press [A info] in the main screen.

```

Additional information about active pgm
Status:                               Step 2
                                       Cycle 3 of 30
Remaining time: 2h 57m
Stop or pause: key [B] in main menu
[A] gradient  [B]      [C] quit      [D]

```

Then press [A gradient]


```

Temperatures of current gradient
T 1- 6:  49.4 49.6 50.3 51.4 52.6 53.8 °C
T 7-12:  55.0 56.2 57.3 58.4 59.1 59.4 °C
remaining time: 2h56m

A gradient      B          C quit      D

```

Note: The temperatures can be displayed online during each gradient step (see section 7.4). As the program moves to the next step the temperature data of the last gradient are stored in the temporary memory. Thus you can check the temperatures of the last gradient in consecutive steps.

Note: If there has not been yet a gradient step in the protocol yet, you will get the message: “No gradient data available”.

To leave the window press [C quit]

7.5 Pause / stop program

Main display during operation:

```

Prog          0 TEST 1   dir 3
Step          1          cycle 8 of 30
temp [°C]     72.0°C     → 72.0
time          0h 0m18s   → 0h 0m30s
lid           104.8°C
A info      B start/stop C programs D +

```

To pause or stop the active program press [B start/stop]

```

Stop/pause
directory:      3
program no:    0 test 1
A ?           B pause  C quit  D stop

```

Select [B pause] to pause program
 [D stop] to stop program
 [C quit] to return to the program display

When you select [B pause], the following screen is displayed:

```

Prog          0 TEST 1   dir 3
Step          1          cycle 8 of 30
temp [°C]     72.0°C     → 72.0
time          0h 0m18s   → 0h 0m30s
lid           104.8°C
A info      B start/stop C programs D +

```

The program pauses in the current state. The word **pause** is displayed and alternates with the elapsed time. You can proceed with the program with [B start/stop].

8 Special functions

Main screen

```
Prog
step
temp [°C]    25.2°C
time
lid          25.4°C
[A] ?       [B] start/stop [C] programs [D] +
```

To change special features of the TGRADIENT press [D+].

```
1 print
2 signal
3 language
4 standard mode
5 test mode
[A] back [B] forward [C] quit [D] enter
```

Scroll though the list with the cursor keys \uparrow \downarrow . Enter desired function with [D enter]

8.1 Print protocols

```
1 graphic protocol*
2 graphic protocols*
3 print program
4 print all programs
[A] [B] [C] quit [D] enter
```

Scroll through the list with the cursor keys \uparrow \downarrow . Confirm the highlighted function with [D enter].

8.2 Switch beep on/off

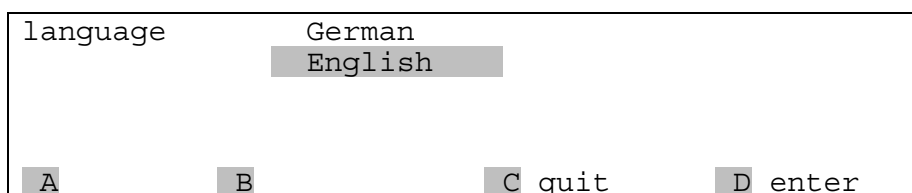
There is an option for an audible signal whenever a program enters a pause and when the program is finished. If this option is active, the beep can be switched off by pressing any key of the keyboard.

```
with signal    yes
               no
[A] [B] [C] quit [D] enter
```

In this window you can select whether there is a signal or whether there is not. Using the cursor keys you can toggle between signal yes/no. Confirm your selection with [D enter].

8.3 Select language

The display of the TGRADIENT can be switched from English to German. Select language with cursor keys \downarrow \uparrow . As you toggle between the two settings the language will change.



To confirm your selection press [**D** enter]

8.4 RS232 Control

The Tgradient can be programmed and controlled by Personal Computer. The Tcontrol iT software is available with the order number 050-820. To activate remote control, the Tgradient has to be set to RS232 control mode.

For detailed information on controlling Tgradient with PC please refer to the Tcontrol iT manual.

8.5 Block type

Display and calculation of gradient temperatures depends on the block version used. A temperature gradient can be run with either 48 well and 96 well modules. After changing block modules from one type to the other, the block type must be set in the instruments software.

Go to special function 8 [block type] and set correct block version.

9 Maintenance

The TGRADIENT was built to operate for a long time without the need for periodical maintenance. The housing may be cleaned from time to time with a smooth cotton cloth. Do not use strong detergents or organic solvents for cleaning.

10 Exchange of modules

For exchange of modules you need two hexagonal keys (number 3 and number 5). These keys are included in the shipment of the module.

Remove module

- ➡ Remove power plug
- ➡ Place instrument upside down on a soft surface
- ➡ Remove the 2 screws that close the shutter (hexagonal key number 3)
- ➡ Remove the 4 screws that hold the block (hexagonal key number 4)
- ➡ unplug connectors from the block (control and power supply)
- ➡ remove module

Install module

Follow description in reversed order.

11 Trouble shooting

11.1 Slow heating and cooling

The T1 THERMOCYCLER 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. Be sure that 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). Dirt can be removed easily from the inlet with a conventional vacuum cleaner.

11.2 Adaptation of protocols from other cyclers

Since the T1 THERMOCYCLER is an extremely fast thermocycler 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 section 6.3. Alternatively, the time settings may be extended.

Note: Protocols that have been optimized with a TGradient thermocycler can be run without changes.

12 Trouble shooting

12.1 Slow heating and cooling

The TGRADIENT 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. Be sure that the inlet is not clogged by dust or other material (e.g. a sheet of paper placed under the cyclers can be attached to the inlet as the fan is in operation). Dirt can be removed easily from the inlet with a conventional vacuum cleaner.

12.2 Restart due to unrecognized power failure

High voltage fluctuation can lead to an automatic restart of the thermocycler. In this case the cyclers restarts at the step where there power failure has occurred. To avoid voltage fluctuation, do not connect the cyclers to a socket shared by a strong power consumer like a refrigerator or a centrifuge.

12.3 Adaptation of protocols from other cyclers

Since the TGRADIENT is an extremely fast thermocycler 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 section 6.3. Alternatively, the time settings may be extended.

13 Appendix

13.1 Technical specification TGradient 96

| | |
|---------------------------|---|
| Thermal range | -3°C – 100°C |
| Temperature accuracy | ± 0.1°C |
| Thermal homogeneity | ± 0.3°C within 15 sec. |
| Range of Gradient | 40°C |
| Maximum heating speed | 4°C/sec. |
| Max. cooling speed | 3°C/sec. |
| Sample capacity | 96 x 0.2ml tubes, 96 well microplates, strips |
| Line Voltage | 115 Volt or 230 Volt |
| Frequency | 50-60 Hz |
| Maximum power consumption | 310 Watt |
| Fuses | see label on the back of the instrument |
| Display | LCD graphical display, back-lit |
| Ports | serial: RS232 (bi-directional data transfer) parallel: Centronics (printer) Port for in tube-sensor |
| | |
| | |
| memory | Total capacity: 1500 steps in 10 directories with maximum 100 programs each |
| | |
| Dimensions (WxDxH) | 25 x 34 x 16,5 cm |
| Weight | 8,8 kg |
| Storage conditions | |
| Operating conditions | 5°C – 35°C |
| | |
| | |

13.2 Ordering information

| | |
|---|-----------|
| TGRADIENT Thermocycler | Order-No. |
| TGRADIENT 48 for 48 x 0.5ml tubes | 050-800 |
| TGRADIENT 96 for 96 x 0.2ml tubes, strips or 96 well microplates | 050-801 |
| | |
| | |
| TGRADIENT Modules | |
| TGRADIENT 48 Module for 48 x 0.5ml tubes | 050-810 |
| TGRADIENT 96 Module for 96 x 0.2ml tubes, strips or 96 well microplates | 050-811 |
| TGRADIENT 384 Module for 384 well microplates (non gradient) | 050-812 |
| TGRADIENT in situ Module for 4 microscopic slides (non gradient) | 050-813 |

For information about tubes, strips, microplates and other consumables please contact BIOMETRA or your local BIOMETRA distributor.

14 Instructions for return shipment

In case of faults occurring with our TGRADIENT Thermocycler please proceed as follows:

- Return only defective devices. For technical problems which are not definitively recognizable as device faults please contact the Technical Service Department at **Biometra** (Tel.: +49/551/5068640, Fax: ++49/551/5068666, e-mail: info@biometra.co.uk).
- Important: Cleanse all parts of the instrument of biologically dangerous, chemical or radioactive contaminants. If a an instrument is contaminated, **Biometra** will be forced to refuse to accept the device. The sender of the repair order will be held liable for possible losses resulting from insufficient decontamination of the device.
- Please prepare written confirmation that the device is free from biologically dangerous and radioactive contaminants. The declaration of decontamination (see 16) has to be attached to the outside of the packaging.
- Use the original packing material. If not available, please use a comparable robust box. Be sure that the instrument fits tightly into the box. When packaging, protect the device in a way to avoid
- Label the outside of the box with “CAUTION! SENSITIVE ELECTRONIC INSTRUMENT!”
- Please enclose a note which contains the following:
 - a) Sender’s name and address,
 - b) Name of a contact person for further inquiries with telephone number,
 - c) Description of the fault, which also reveals during which procedures the fault occurred, if possible

15 Warranty

This Biometra instrument has been carefully built, inspected and quality controlled before dispatch. Hereby Biometra warrants that this instrument conforms to the specifications given in this manual. This warranty covers defects in materials or workmanship for 24 months as described under the following conditions:

This warranty is valid for 12 month from date of shipment to the customer from Biometra or an authorized distributor. This warranty will not be extended to a third party without a written agreement of Biometra.

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

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

16 Equipment Decontamination Certificate

To enable us to comply with German law (i.e. §28 StrlSchV, §17 GefStoffV and §19 ChemG) and to avoid exposure to hazardous materials during handling or repair, will you please complete this form, prior to the equipment leaving your laboratory

COMPANY / INSTITUTE _____

ADDRESS _____

TEL NO _____ FAX NO _____ E-MAIL _____

| EQUIPMENT | Model | Serial No |
|-----------|-------|-----------|
| | _____ | _____ |
| | _____ | _____ |
| | _____ | _____ |
| | _____ | _____ |

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

Hazardous materials used with this equipment

Has the equipment been cleaned and decontaminated? YES / NO (delete)

Method of cleaning / Decontamination

NAME _____ POSITION _____

(HEAD OF DIV./ DEP./ INSTITUTE / COMPANY)

SIGNED _____ DATE _____

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

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

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