

The PTC-0150 MiniCycler Operations Manual

Version 4.0

**MJ RESEARCH, Inc.
Waltham, MA**

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Documentation Conventions

Typographic Conventions

The names of keyboard keys are in sans serif type and placed within double angle brackets:

Example: «Proceed»

Items in programming menus are italicized:

Example: Select *Edit* from the Main Menu.

Graphic Conventions

The programming screens displayed in the LCD window are represented by a box containing two lines of text:

Example:

_RUN	Enter
Program	Program

Terminology

A programming option is termed “selected” when the cursor is positioned in front of it. Use the «Select» keys (see fig. 2-3) to move the cursor. In some screens selected items are also displayed in all-capital letters.

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Introduction

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Meet the PTC-150 MiniCycler

Thank you for purchasing an MJ RESEARCH PTC-150 MiniCycler. Designed by a team of molecular biologists and engineers, the MiniCycler will meet your needs for an easy-to-use, reliable, and compact programmable thermal cycler:

- Easy-to-read programming interface for quick and painless programming
- Choice of block or probe control
- Space-saving design for easy setup and transportation
- Instant Incubate feature for continuous-temperature incubations
- Customizable factory-installed protocols
- Hot Bonnet heated lid for oil-free cycling

How to Use This Manual

This manual contains all the information you need to operate your MiniCycler safely and productively:

- Chapter 2 describes the **physical characteristics** of the MiniCycler.
- Chapters 3–5 describe the basics of **installing and operating** the MiniCycler.
- Chapters 6 and 7 describe **programming** the MiniCycler.
- Chapter 8 explains the proper **maintenance** of the MiniCycler.
- Chapter 9 offers **troubleshooting** information for the MiniCycler.
- Chapter 10 describes **how to change the MiniCycler's block**.

Important Safety Information

Safe operation of the MiniCycler begins with a complete understanding of how the machine works. Please review this entire manual before attempting to operate the MiniCycler. Do not allow anyone who has not reviewed this manual to operate the machine.

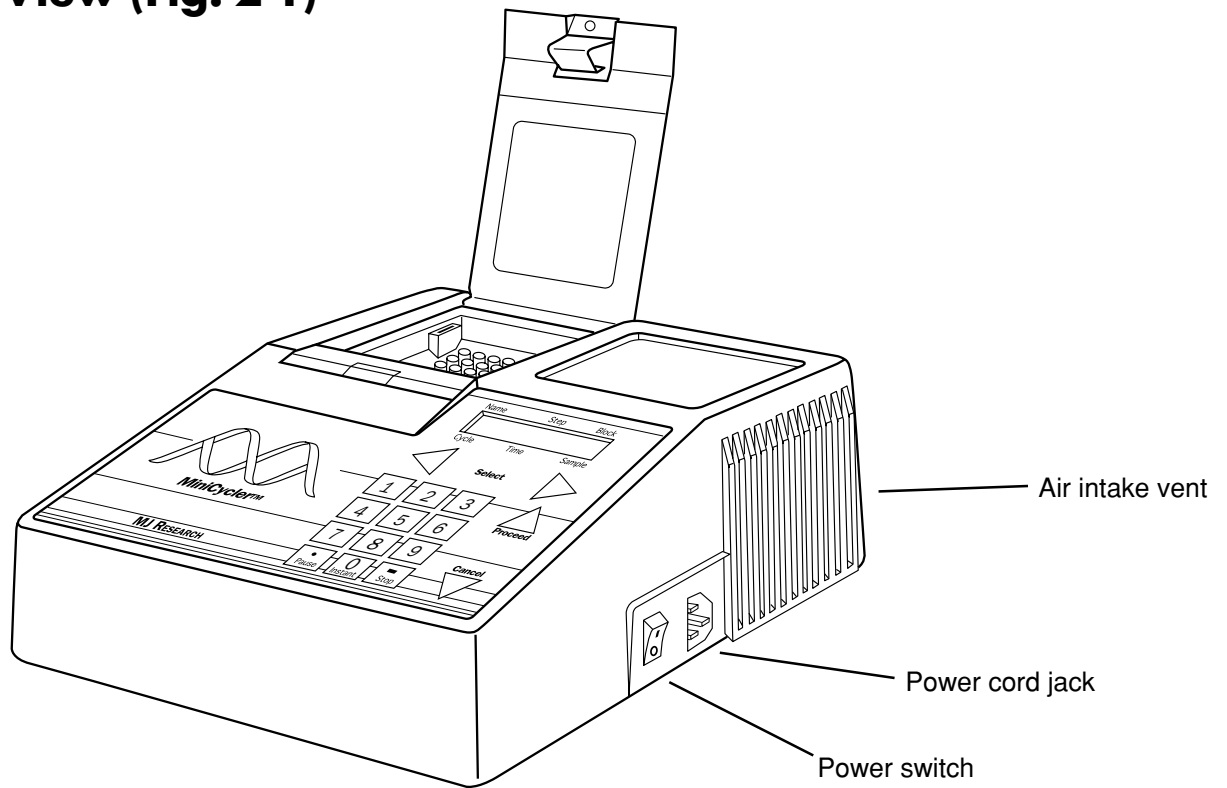
The MiniCycler can generate enough heat to inflict serious burns and can deliver strong electrical shocks if not used according to the instructions in this manual. Please read the safety warnings and guidelines in appendix B, and exercise all precautions outlined in them.

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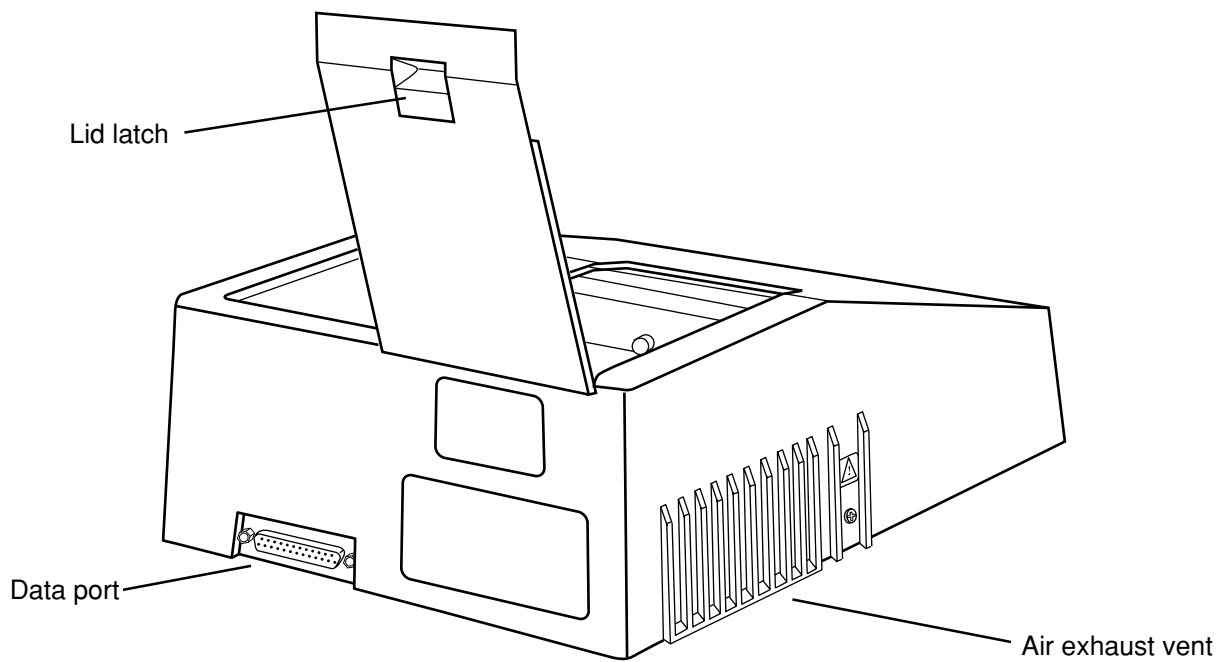
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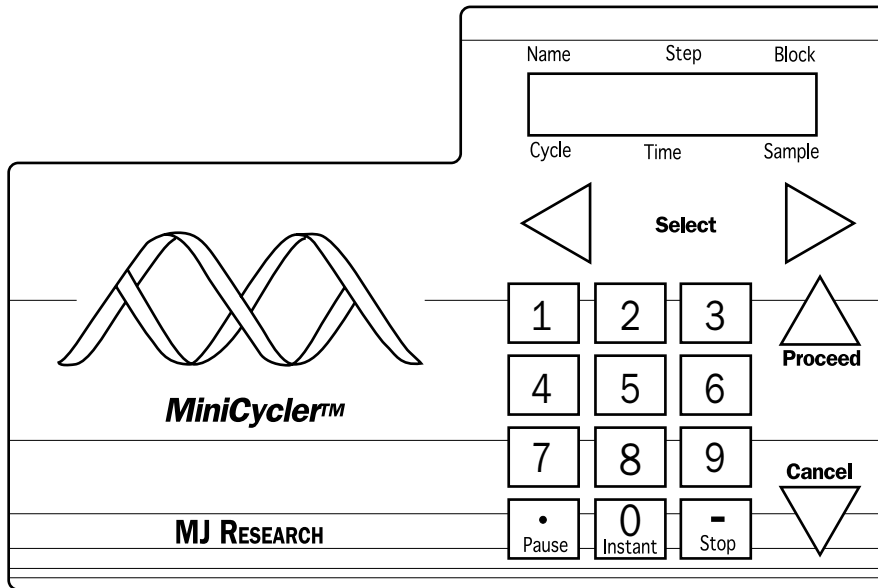
Front View (Fig. 2-1)



Back View (Fig. 2-2)



Control Panel (Fig. 2-3)



Blocks Available

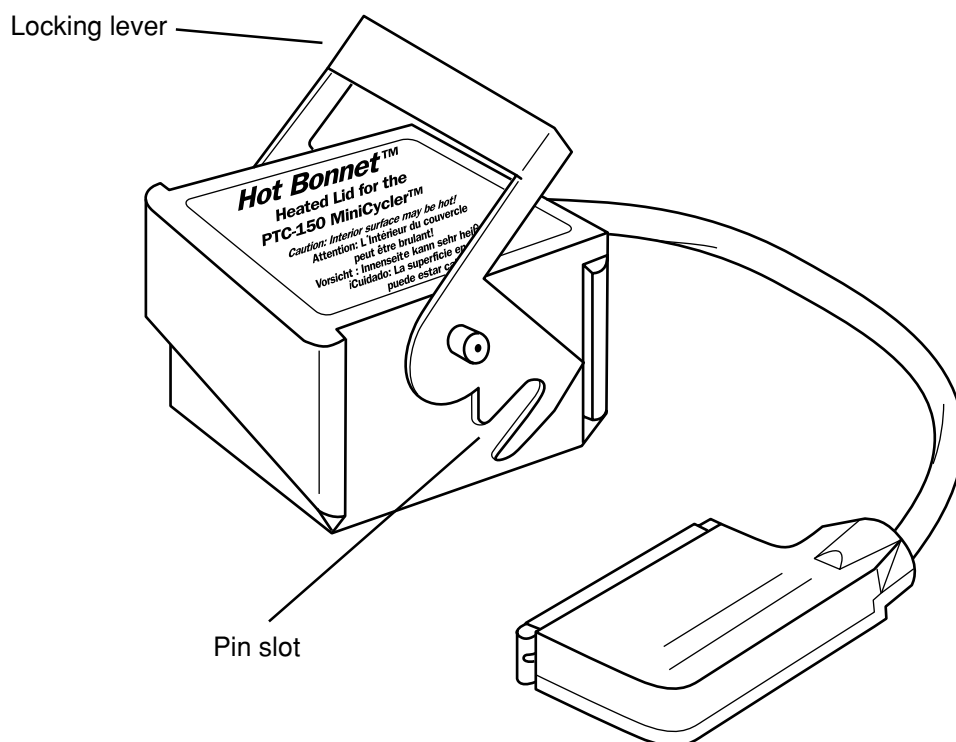
16-well block: holds 16 x 0.5mL tubes

25-well block: holds 25 x 0.2mL tubes or one 25-well V-bottom microplate

Accessories

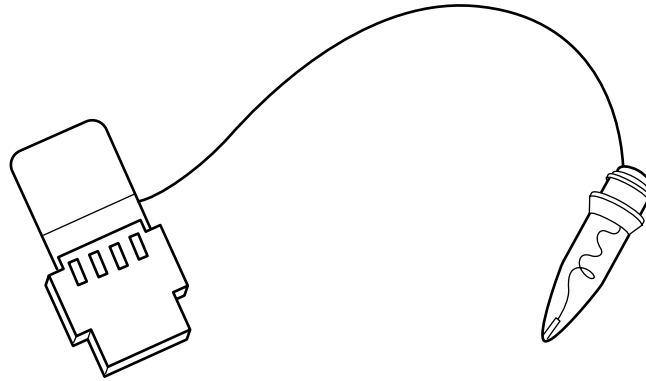
Hot Bonnet (Fig. 2-4)

Available for 16- and 25-well blocks



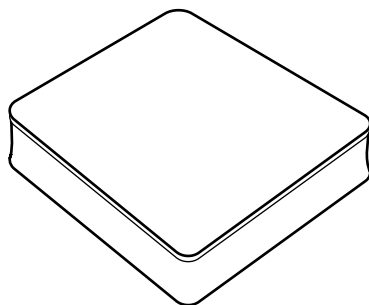
Temperature probe (Fig. 2-5)

Allows machine to control reactions according to a representative sample's temperature. Available for 16- and 25-well blocks.

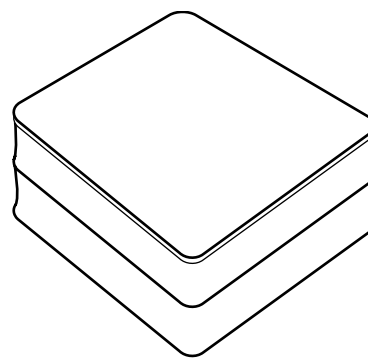


Magnetic sponge rubber pads (Fig. 2-6)

Used to help press tubes or a plate into the block. Available for use with 16- and 25-well blocks.



Thin (1cm) pad
(for 16-well block)



Thick (2cm) pad
(for 25-well block)

Specifications

Thermal range:	-9° to 105°C (no more than 35°C below ambient temperature)
Accuracy:	±0.3°C of programmed target at 90°C, NIST traceable
Thermal homogeneity:	±0.4°C well-to-well within 12 seconds of arrival at 90°C ±0.3°C well-to-well within 40 seconds of arrival at 90°C
Sample capacity:	16-well block: 16 x 0.5mL tubes 25-well block: 25 x 0.2mL tubes or one 25-well microplate
Line voltage:	100–240VAC rms (no adjustment needed among voltages within these ranges)
Frequency:	50–60Hz single phase
Power:	250W maximum
Fuses:	Two T2.5A/250V
Displays:	One 2 x 16 LCD alphanumeric display
Ports:	One 25-pin 8-bit parallel interface printer port (optional)
Memory:	80 typical programs, in nonvolatile memory; each program may have up to 100 steps; maximum of 400 steps among all programs
Weight:	3.0kg
Size:	22 x 28 x 11cm high

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Installation

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Packing List

After unpacking the MiniCycler, check to see that you have received the following:

- One MiniCycler, with the sample block of your choice
- One power cord
- Two fuses
- A 1cm–thick magnetic sponge-rubber pad (16-well block), or a 2cm–thick pad (25-well block).
- One red metal adapter plate (25–well block with heated lid only)
- The *PTC-150 MiniCycler Operations Manual* (this document)
- Warranty registration card and extended warranty application (US customers only)

If any of these components are missing or damaged, contact MJ RESEARCH or the authorized distributor from whom you purchased the MiniCycler to obtain a replacement. Please save the original packing materials in case you need to return the MiniCycler for service. See appendix D for shipping instructions.

Setting Up the MiniCycler

Insert the power cord plug into its jack (see fig. 2-1), then plug the cord into an electrical outlet (see “Power Supply Requirements,” below). Situate the machine according to the instructions below.

Environmental Requirements

Ensure that the area where the MiniCycler is installed meets the following conditions, for reasons of safety and performance:*

- Indoor, nonexplosive environment
- Ambient temperature 4–32°C
- Relative humidity between 10% and 90%
- Protection from excessive heat (e.g., radiators) and accidental spills

**Note: operation at altitudes significantly above sea-level may affect operation, particularly heat dissipation.*

Power Supply Requirements

The MiniCycler requires 100–240VAC, 50–60Hz, and a grounded outlet. The machine can use current in the specified range without adjustment, so there is no voltage-setting switch.

Air Supply Requirements

Air is taken in from a vent on the right side of the machine and exhausted from a vent on the left side of the machine (see figs. 2-1 and 2-2). If the machine does not get enough air or gets air that is too warm, it can overheat. Overheating can cause the machine to display a warning message, “H[eat] S[ink] Overheating, Check Air Flow,” or even to shut itself off.

To prevent overheating, position the machine at least 10cm from other thermal cyclers or walls. This is particularly important if two or more MiniCyclers are used in one installation: one machine can blow all of its exhaust air directly into the intake vent of its neighbor if they are placed closely side by side. If the ambient air temperature exceeds 32°C, use fans or adjust the air conditioning to cool the air that the machine receives. Keep the air intake vent clean and free of obstructions (e.g., loose sheets of paper).

4

Operation

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Turning the MiniCycler On

Move the power switch to “1” (the “On” position). A self-test of the heat pump will usually begin running (see below). Its progress is tracked in a screen in the LCD window:

SELF TEST *

This screen disappears within 10 seconds. If a problem is detected, an error message will be displayed.

Note: If either the heat sink or the block is not between 17°C and 30°C, the machine will skip the self-test.

If the self-test does not detect any problems, the Run-Enter Menu is displayed:

_RUN	Enter
Program	Program

Using the Control Panel

The control panel (see fig. 2-3) includes operation keys, an LCD window for displaying programming and machine status text, and a numeric keypad for entering values into programs.

Operation keys

- **Select keys** (left and right arrows): Move the cursor one space or option to the left or right in the LCD window.
- **Proceed:** Accepts a selected menu or screen option.
- **Cancel:** Terminates a running protocol; during programming or editing, cancels the last entry.
- **Stop:** Terminates a running protocol.
- **Pause:** Pauses a running protocol.
- **Instant:** Programs an Instant Incubation (see 5–6).

Opening and Closing the Lid

To open the MiniCycler, gently push the lid latch (see fig. 2-2) forward until it disengages, then push the lid up. To close the lid, push the lid down until the latch engages with a distinct click.

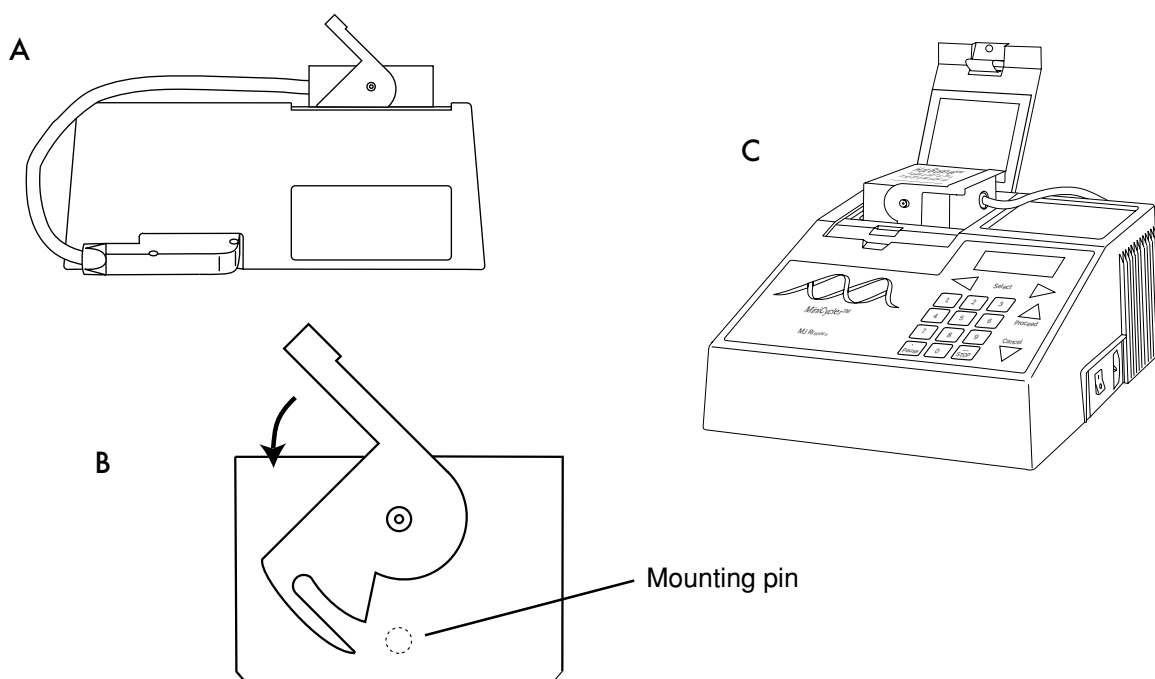
Mounting the Hot Bonnet

The Hot Bonnet mounts on two metal pins inside the retainer. When the lid's lever is lowered, the lid locks into place on the pins, and the inner lid is lowered onto the tops of loaded sample vessels (fig. 4B). (Certain models of the MiniCycler lack these pins. A retrofit to install them is available. Contact your distributor or MJ Research for further information.)

Follow this procedure to mount the Hot Bonnet:

1. Make sure the Hot Bonnet is the correct size for the block. The sticker on the side of the unit identifies it by catalogue number: HBA-1151 is for 16-well blocks; HBA-1152 is for 25-well blocks.
2. If only a few tubes are being run, make sure one tube is loaded into each corner of the block, using empty tubes if necessary.

Figure 4-1 Mounting the Hot Bonnet. A, Rear view of MiniCycler showing correct cable orientation for the Hot Bonnet. B, Hot Bonnet side view, showing mounting lever. C, Correctly mounted Hot Bonnet.



3. Plug the Hot Bonnet into its port at the back of the machine, so that the cable is to the left of the connector as you face the back of the machine (fig. 4-1A).
4. Raise the Hot Bonnet lever, and set the lid into the block with the lever pointing to the right as you face the machine's front.
5. Lower the lever as far as possible, which locks the Hot Bonnet into place and lowers the heated lid onto the loaded tubes. Figure 4-1C shows a correctly mounted Hot Bonnet.

Note: Intermittent beeping is an indication that the heated lid is not being used correctly.

If a PTC 150 is beeping, it is likely there are not four tubes of the appropriate height in each of the four corners of the sample block. Placing a tube in each of the four corners of the sample block will allow the lid to seat squarely atop the block and the beeping will stop. The Minicycler heated lid is unadjustable and designed to fit over standard sized flat-capped 0.5 mL tubes (on the 16 well cyclor) or standard sized 0.2 mL dome-capped tubes (on the 25 well cyclor). If you would like to use a non-standard tube height, or a flat-capped 0.2 mL tube, use the ADR-0153 adapter plate provided.

If the unit is programmed and plugged in correctly, carefully try to determine if the heated lid is getting hot (do not touch the inside of the lid, because if it is working, it will be extremely hot!). If the lid is cool, it may be in need of repair. Please call customer service at (888) MJCYCLE.

Selecting the Correct Sample Vessel

MJ RESEARCH offers a full range of tubes and microplates, manufactured to the specifications of each type of block to ensure a precise fit. See chapter appendix 4-A for a complete list. Keep in mind that differences in tube and plate composition and wall thickness among the many brands available can affect reaction results. Protocols may require some adjustment to ensure optimum results when using a new vessel type.

Note: certain tube and sealing options require the use of the Microseal Adapter: see page 4-7.

0.5mL tubes

The 16-well block accepts either thick- or thin-walled tubes. Make sure thick-walled 0.5mL tubes fit the wells snugly. Since these tubes were originally designed for centrifuges, some brands may not fit tightly in thermal cyclor wells. Thin-walled 0.5mL tubes were specifically designed for

thermal cycling, and the higher quality brands provide a good and consistent fit. MJ RESEARCH provides thin- and thick-walled 0.5mL tubes designed for precise block fit.

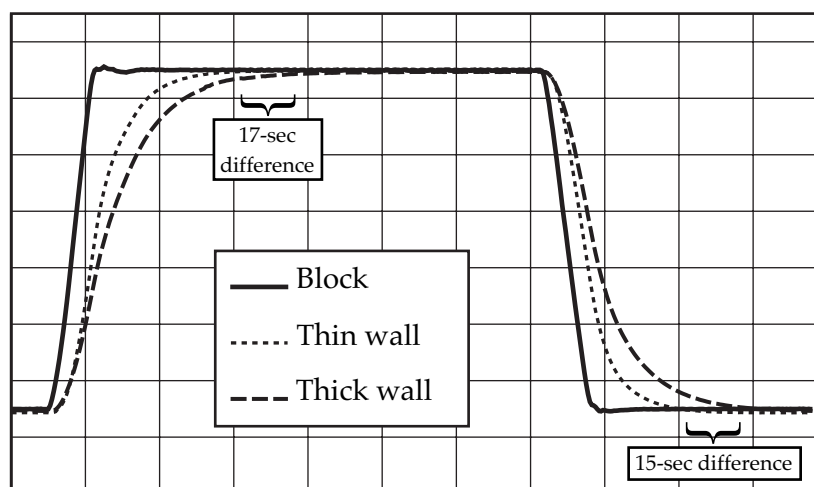
0.2mL tubes

All types of thin-walled 0.2mL tubes may be used in the 25-well block. MJ RESEARCH sells high-quality 0.2mL tubes in a number of styles, including individual tubes and strips.

Microplates

V-bottom polycarbonate or polypropylene microplates may be used in the 25-well block as long as they fit the wells snugly. Polypropylene microplates are usually preferred because they exhibit very low protein binding and, unlike polycarbonate microplates, do not lose water vapor through the vessel walls. This allows smaller sample volumes to be used—as little as 5–10 μ L. Polypropylene microplates and compatible Microseal 'A' film, mats, or strip caps for sealing are available from MJ RESEARCH. (See "Sealing with the Hot Bonnet and Caps or Film Sheets," p. 4–7, for a description of Microseal 'A'.)

Figure 4-2 Ramp rate enhancement with thin-walled tubes. To compare ramp rates of sample temperature, 50 μ L samples of water were cycled in 0.5mL thin- and thick-walled tubes in a PTC-200 thermal cycler with a 60-well block. Thermal profile: 94°C, 120 sec; 62°C, 120 sec. Data are shown for the third cycle. The cycler was run under Block Control, and the Hot Bonnet was heated to 105°C and tightened to a half-turn past the touch point. The samples in thin-walled tubes reached the target temperature more quickly. In this example, over 30 seconds could be saved per cycle by using thin-walled tubes. A comparable ramp rate enhancement can be expected when using thin-walled tubes in the PTC-150 MiniCycler.



Thin- vs. thick-walled tubes

The thickness of sample tubes directly affects the speed of sample heating and thus the amount of time required for incubations (see fig. 4-2). Thick-walled tubes delay sample heating since heat transfers more slowly through the tubes' walls. For the earliest types of thermal cyclers this delay mattered little. These machines' ramp rates were so slow (below 1°C/sec) that there was plenty of time for heat to transfer through the tube wall to the sample during a given incubation.

Modern thermal cyclers have much faster ramp rates, so the faster heat transfer provided by thin-walled tubes allows protocols to be significantly shortened. For example, in the reaction illustrated in figure 4-2, over 30 seconds can be saved per cycle by using thin-walled tubes, for an overall savings of 15 minutes in a 30-cycle run.

Sealing Sample Vessels

Water can evaporate from reaction mixtures during thermal cycling, changing the concentration of reagents. A layer of oil or wax will completely prevent evaporation from tubes and microplates, but Microseal film, mats, or caps, in conjunction with the Hot Bonnet Heated lid, are also effective and far less messy and inconvenient.

Sealing with oil or wax

Mineral oil, silicone oil, paraffin wax, or Chill-out™ liquid wax may be used to seal tubes or plate wells. Use only a small amount of oil or wax; 1-3 drops (15-50µL) are usually sufficient. **Use the same amount of oil or wax in all sample vessels to ensure a uniform thermal profile.**

Some paraffin waxes solidify at room temperature. The wax can then be pierced with a micropipette and the samples drawn off. Silicone oil and mineral oil can be poured off or aspirated from tubes if the samples are first frozen (-15° to -20°C). The samples are usually pure enough for analysis without an extraction.

Chill-out™ liquid wax (available from MJ RESEARCH) is an easy-to-use alternative to oil. This purified paraffinic oil solidifies at 14°C and is liquid at room temperature. By programming a hold step at low temperature, the wax can be solidified at the end of a run. A pipette can then be used to pierce the wax in the tubes and remove the samples. The wax is dyed red to assist in monitoring its use (clear Chill-out wax is also available). The dye has no adverse effects on fluorescent gel analysis of reaction products.

Sealing with the Hot Bonnet and caps or film sheets

The inner lid of the Hot Bonnet heats to 108°C during a 99-second pre-heat. When heated, the lid maintains a higher temperature in the upper part of the reaction vessels, which prevents condensation, refluxing, and changes in reaction concentrations. The lid also exerts pressure on the tops of vessels loaded into the sample block, helping to maintain a vapor-tight seal and to firmly seat the vessels in the block.

Caps, film, or mats must be used along with the Hot Bonnet to prevent evaporative losses. Tight-fitting caps are recommended for minimal vapor loss (and for long-term storage of reaction products). For optimal sealing, use the Easy Cap Tool (ECT-1000) to firmly seat the caps in the tubes. Microseal 'A' film may be used when a pristine sealing surface is required for each run. When a reusable sealer is appropriate, the 96-well Microseal 'M' rubber sealing mats are easy to use and effective. Microseal 'A' and 'M' sealers are sized for an array of 96 0.2mL tubes and must be cut to fit the 25-well block. Follow the manufacturer's instructions when applying caps, films, and mats.

Note: After a hold at below-ambient temperatures, a ring of condensation may form in tubes above the liquid level but below the top of the sample block. This is not a cause for concern since it occurs only at the final cool-down step, when thermal cycling is finished.

The Microseal Adapter

Certain tube and sealing options require the use of the Microseal Adapter (ADR-0153). This device is to be used in conjunction with flat-cap tubes, an array of capless 2mL tubes, or a 25-well Multiplate sealed with Microseal A. The adapter is essentially a heat-conducting metal spacer: it raises the clearance of lower-profile sample tubes that would otherwise not engage the MiniCycler lid's safety interlock, and it provides the lid-pressure the Microseal film requires for proper sealing.

Loading Sample Vessels

Loading tubes and plates

If you will be using the Hot Bonnet with a small number of tubes, load at least one empty tube in each corner of the block (see "Mounting the Hot Bonnet," p. 4-3).

To ensure uniform heating and cooling of samples, tubes and plates must make good thermal contact with the block. Adequate contact is ensured

by always following these recommendations:

- Check that the block is clean before loading samples (see chapter 8 for instructions on cleaning block wells).
- Firmly press the tubes or microplate into the wells, and use the heated lid **or** the sponge rubber pad to ensure that tubes are firmly pressed into the block. **(Do not use the sponge rubber pad with heated block.)**

Using the sponge rubber pad to improve tube contact with the block

When using the MiniCycler without the Hot Bonnet, a magnetic sponge-rubber pad may be attached to the lid to help press loaded tubes into the block wells. Two sizes of pad are available:

- A 1cm pad for use with 0.5mL tubes in the 16-well block
- A 2cm pad for use with 0.2mL tubes or 25-well microplates

Attach the magnetic backing of the pad to the metal plate under the lid. When the lid is closed, the pad will firmly seat tubes or microplates.

Using the Probe

An in-sample temperature probe is available for MiniCyclers with the 16- or 25-well block. The probe consists of a precision thermistor mounted in a thin-walled plastic tube (see fig. 2-5). A thin wire, encased in a small plastic tube, runs from the thermistor to the probe's plug, which is inserted into a jack at the side of the block. A specific amount of oil is added to the probe tube (see "Adding the oil," below), to serve as the representative sample. The tube is loaded into the block, where it can serve as the control reference for any programmed target temperature between 0 and 100°C.

When a probe-control protocol is run, the MiniCycler controls the block's temperature to keep the probe at the programmed temperature, using feedback information from the thermistor. Protocols must be tailored to fit this control mechanism (see chapter 6).

Customizing the probe vessel

For the most precise control of sample temperatures, install the probe's wire and thermistor in the same type of tube that the samples will be placed in.

Follow these steps to customize the probe vessel:

1. Cut the hinge to the probe tube's lid. Gently remove the thermistor from the probe tube; the lid will come off with it.

2. Remove the lid from the new probe tube. Add the amount of oil specified under “Adding the Oil.”
3. Gently place the thermistor into the new tube, and snap the lid closed. Make sure that the lid from the original probe tube (attached to the thermistor) fits on the new tube and that the tube is long enough to accommodate the probe wire. The sensor should rest on the tube’s bottom.

! **Caution:** The thermistor is extremely fragile. Handle it with great care.

Adding the oil

Viscous oils (not water!) are the best choice for the probe tube’s representative sample. They closely mimic the thermal characteristics of buffer solution, which changes temperature sluggishly due to the high specific heat of water.

Light and heavy mineral oil and silicone oil may be used (table 4-1). MJ RESEARCH recommends using heavy mineral oil because the formula for determining the correct volume of oil to use is easy to remember, and it is widely available and inexpensive. But whichever type of oil you use, **be careful to add the correct amount of oil.**

Loading and connecting the probe

Seat the probe tube in the center of the block.

Plug the probe into its jack (see fig. 4-3). One of the pin holes in the probe jack has been blocked off so that the probe cannot be plugged in incorrectly.

Running a protocol under probe control

To run a protocol under probe control, load and connect the probe as described above. Select a protocol that has been designed for probe control and press «Proceed». The following screen will be displayed:

```
In-sample Probe
Control? _NO Yes
```

Select *Yes* and press «Proceed». The protocol will begin running under probe control.

Figure 4-3 Correctly installed probe.

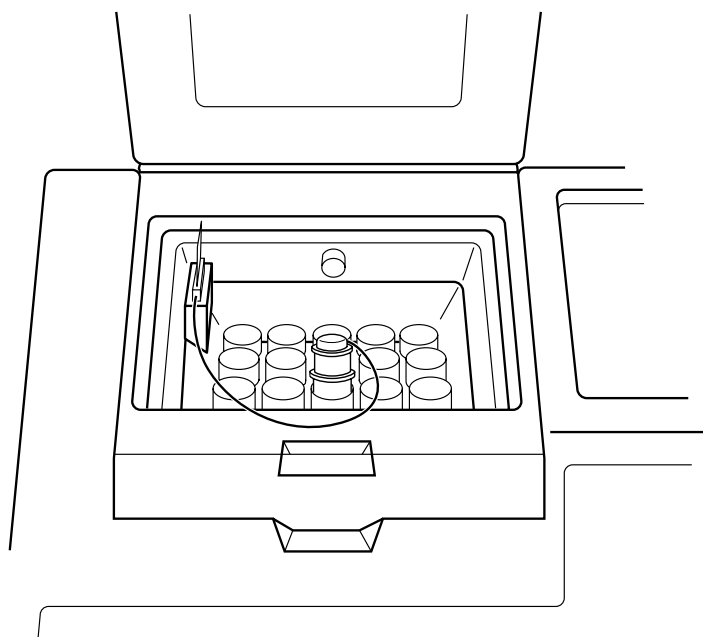


Table 4-1 Calculating the correct amount of oil

Type of oil	Sigma number	Amount to use
Heavy mineral oil	400-5	1 x volume of buffer in individual sample tube + 1 x volume of oil overlay
Light mineral oil	M5904	1.4 x volume of buffer in individual sample tube + 1 x volume of oil overlay
Silicone oil	DMPS-5X	1.7 x volume of buffer in individual sample tube + 1 x volume of oil overlay
Silicone oil	DMPS-V	2.7 x volume of buffer in individual sample tube + 1 x volume of oil overlay

Tube, Microplate, and Sealing System Selection Chart

Key

- Reaction vessel or sealer fits block without modification.
- Reaction vessel or sealer must be cut to fit.

MJ RESEARCH Thermal Cycler Blocks		Reaction Vessels		Sealing Options for Oil-Free Cycling			
16 (0.5ml)	25 (0.2ml)	Description	MJ RESEARCH Catalog #	Microseal 'A' film MSA-5001	Microseal 'M' mat MSM-1001	8-Strip caps TCS-0801	12-Strip caps TCS-1201
●		0.5ml tubes w/ caps, thin wall	TBI-0501				
●		0.5ml tubes w/ caps, thick wall	TBI-0601				
	●	0.2ml tubes w/ flat caps	TFI-0201				
	●	0.2ml tubes w/ domed caps	TWI-0201				
	●	0.2ml tubes, no caps	TBI-0201	○	○	○	○
	○	8-strip 0.2-ml tubes	TBS-0201	○	○	○	○
	○	12-strip 0.2-ml tubes	TBS-1201	○	○	○	○
	●	Multiplate 25-well microplates	MLP-2501	○	○	○	○
	○	Multiplate 24-well microplates	MLP-2401	○	○	○	○
	○	Multiplate 48-well microplates	MLP-4801	○	○	○	○
	○	Multiplate 96-well microplates	MLP-9601	○	○	○	○

Note: All tubes and Multiplate microplates are made from polypropylene plastic. Microseal 'A' sealing film and Microseal 'M' mats are supplied in a 96-well format and must be cut to fit the Minicycler.

Safety Warning Regarding Use of ^{35}S Nucleotides

Some researchers have experienced a problem with **radioactive contamination** when using ^{35}S in thermal cyclers. This problem has occurred with all types of reaction vessels.

The Problem

When ^{35}S nucleotides are thermally cycled, a volatile chemical breakdown product forms, probably SO_2 . This product can escape the vessel and contaminate the sample block of a thermal cycler, and possibly the air in the laboratory. Contamination has been reported with microassay plates, 0.2-mL tubes, and 0.5-mL tubes.

Polycarbonate microplates

These microplates present the largest risk of contamination. Polycarbonate is somewhat permeable both to water and the ^{35}S breakdown product. This problem is exacerbated when polycarbonate plates are held at high temperatures for long periods of time, or when the plates are sealed for oil-free thermal cycling.

0.2mL Polypropylene tubes and 25-well polypropylene microplates

These tubes are manufactured with very thin walls to enhance thermal transfer. The thin walls are somewhat fragile and can develop small cracks when subject to mechanical stress. Undamaged thin polypropylene tubes may also be somewhat permeable to the ^{35}S breakdown product. Either way, there have been reports of ^{35}S passing through the walls of microplates and 0.2-mL tubes of several different brands during thermal cycling.

0.5-mL Polypropylene tubes

Contamination problems are rarer with this type of tube, but instances have been reported.

The Solution

1. Substitute the low-energy beta emitter ^{33}P in cycle sequencing. ^{33}P nucleotides are not subject to the same kind of chemical breakdown as ^{35}S nucleotides, and they have not been associated with volatile breakdown products.
2. If ^{35}S must be used, three things will help control contamination: an oil overlay inside the tubes, mineral oil in the thermal cycler outside the tubes, and use of thick-walled 0.5-ml tubes. Always run ^{35}S thermal cycling reactions in a fume hood, and be aware that vessels may be contaminated on the outside after thermal cycling. Please be certain that you are using the

appropriate detection methods and cleaning procedures for this isotope. Consult your radiation safety officer for his or her recommendations.

If mild cleaning agents do not remove radioactivity, harsher cleaners may be used. Users have suggested the detergent PCC-54 (Pierce Chemical Co., Rockford, Illinois; Pierce Eurochemie B.V., Holland), Micro Cleaning Solution (Cole-Parmer, Niles, Illinois), and Dow Bathroom Cleaner (available in supermarkets).

Caution: Harsh cleaning agents are corrosive to aluminum and must never be used on bare aluminum blocks. MJ RESEARCH blocks are anodized, so they have a protective coating of aluminum oxide. Still, harsh agents (such as those above) must be *thoroughly* rinsed away within a few minutes of application, or the anodization will degrade.

5

Running Protocols

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Running a Protocol

Either a custom-designed protocol or one of the factory-installed protocols may be run. See appendix F for descriptions of the factory-installed protocols, which may be edited to fit your needs.

To run a protocol, select *Run Program* from the Run-Enter Menu, then press «Proceed».

If protocols have been stored in the machine, they will be displayed.

Press one of the «Select» keys until the name of the protocol you wish to run is displayed. Press «Proceed» again. The protocol will begin running.

```
Run
QUIKSTEP  ?
```

Running a protocol with the Hot Bonnet installed

Select a protocol to run as described above and press «Proceed». The following screen will be displayed:

```
Use Heated Lid?
_YES No
```

Select *Yes* and press «Proceed». The Hot Bonnet will begin its 99-second preheat to 108°C. A screen showing a timer that counts down the seconds of this preheat period will be displayed:

```
QUIKSTEP
Preheat time: 89
```

When the lid has fully preheated, the protocol will begin to run.

If the Hot Bonnet is removed when it is preheating, if tubes are not present in all corners of the block, or if the Hot Bonnet cable is disconnected from the machine, the MiniCycler will begin beeping, and the following message will be displayed:

```
Check Bonnet
Placement...
```

When the Hot Bonnet is replaced in the well, or tubes are placed in all corners of the block, or the cable is connected to the machine, the Hot Bonnet will stop beeping and re-start its 99-second preheat.

Note: If the Hot Bonnet is removed when a programmed protocol is running, the MiniCycler will begin beeping **but no message will be displayed.**

Running a protocol under probe control

To run a protocol under probe control, load and connect the probe (see p. 4–12). Select a protocol that has been designed for probe control. Do not use probe control to run a protocol designed for block control. Press «Proceed». The following screen will be displayed:

In-sample Probe Control? -NO Yes

Select *Yes* and press «Proceed». The protocol will begin running under probe control.

If probe control is not desired, select *No* and press «Proceed». The protocol will begin running under block control.

Reading the Runtime Screen

During a protocol run, a runtime screen will be displayed:

QUIKSTEP 1 65.4
1

This screen lists the program name (Quikstep in the example above), the protocol step that is running (1), the block temperature (65.4°C), and the cycle number (1). If the probe is installed, the temperature of the representative sample inside the probe is displayed (62.7°C):

CUSTOM 1 65.4
1 62.7

The runtime screen will show each step of the protocol as it executes. When the target temperature for a given step is reached, a timer in the middle of the second line will begin running:

```
QUIKSTEP 2 92.0  
1      00:10
```

The timer shows the length of time the samples have been held at the displayed temperature. When another step begins, the timer disappears until the new step's target temperature is reached. At this point the timer begins running for the new step.

Reading the Protocol Completion Screen

When the protocol ends, a message is displayed:

```
QUIKSTEP 5 75.0  
Complete
```

The number of the last step in the protocol (5 in the example above) and the temperature of the block at the completion of the program (75°) are displayed in the upper right-hand corner of the screen. The temperature reading will change as the block cools to ambient temperature.

Press «Proceed» to remove this screen. The Run-Enter Menu will be displayed, and another protocol may be run.

Manually Stepping Through a Protocol

A running protocol can be manually advanced through its steps. As soon as a step has reached its target temperature (i.e., when the timer begins running for the step), press «Proceed» to progress the protocol to its next programmed step. The next step will immediately be displayed.

Note: Pressing «Proceed» while a step is ramping will have no effect. A step must have reached its target temperature before the protocol can be manually advanced to the next step.

Protocols can be programmed to require manual stepping (see p. 6.10).

Pausing a Running Protocol

Press «Pause» to temporarily stop a running protocol. If «Pause» is pressed during temperature ramping, the protocol will pause as soon as the target temperature is reached, and samples will be held at the displayed temperature. If «Pause» is pressed after a step has reached its target temperature, the protocol will immediately begin holding samples at the current incubation temperature. The timer will stop running, and the word “Pause” will be displayed:

QUIKSTEP 2	92.0
9	PAUSE

To resume the protocol, press «Pause» again. The timer will begin running again, and the protocol will complete the step that was paused and continue on to the next step.

Stopping a Running Protocol

Press «Stop» or «Cancel» to stop a running protocol. The program will stop running, and the Run-Enter Menu will be displayed again.

Note: Turning off the machine will not stop a running protocol. Instead, the MiniCycler will assume that a power outage has occurred and will resume running the protocol when the machine is turned on again (see below).

Resuming a Protocol after a Power Outage

If a power failure occurs when a protocol is running, the MiniCycler will hold the protocol in memory for at least 24 hours and sometimes up to 10 days. (The exact duration depends on environmental conditions.)

When power is restored, the protocol will begin running again at the point at which it was stopped. When the protocol ends, the protocol completion screen will be displayed, with a special notice on its last line:

```
QUIKSTEPCOMPLETE
Press Proceed
```

Press «Proceed» as instructed. A screen about the power outage will be displayed. The screen will identify the step and the cycle that were running when the power failure occurred:

```
AC POWER FAILED
Cyc 3      Step 2
```

Press «Proceed» to remove this screen. The Run-Enter Menu will be displayed again.

Running an Instant Incubation

The MiniCycler may be used as a constant-temperature incubator by pressing «Instant» (the zero key) while the Run-Enter Menu is displayed. A screen allowing entry of the incubation temperature will be displayed:

```
TEMP: _
```

Type any incubation temperature from -9.0 to 105.0°C , then press «Proceed». The MiniCycler will incubate the sample at the specified temperature.

When the sample block reaches the incubation temperature, a timer will begin running in the lower right-hand corner of the screen. To stop and start the timer, press «Pause». To stop an instant incubation, press «Cancel» or «Stop» (see “Stopping a Running Protocol”).

6

Programming

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MiniCycler programs consist of a series of steps encoding a protocol. These steps are run using one of two temperature control methods: block control or probe control.

Programs may contain six types of steps. Two of the step types are mandatory, and four are optional:

1. **Temperature step (mandatory):** Sets a temperature for the block and the length of time it is held at that temperature. The MiniCycler brings the block to this temperature at its maximum rate of heating or cooling (up to 2.4°C/sec) unless modifying instructions are added to the program.
2. **GoTo step (optional):** Causes the program to cycle back to an earlier step for a specified number of times (up to 9,999 times).
3. **Increment step (optional):** Allows a progressive increase or decrease of temperature (–6.0° to 6.0°C/cycle) each time a step is executed in a GoTo cycle (useful in “touchdown” programs).
4. **Extend step (optional):** Allows a progressive lengthening or shortening of a temperature step hold (by 1–60 sec/cycle) each time a step is executed in a cycle (useful for accommodating an enzyme with diminishing activity).
5. **Slope step (optional):** Allow a slower-than-maximum rate of heating or cooling.
6. **End step (mandatory):** Instructs the MiniCycler to shut down its heat pump because the last line of the program has run.

Designing a New Program

Using the GoTo step to write short programs

The GoTo step allows programs of many repetitious steps to be shortened to just a few lines. When the program encounters a GoTo step, it returns to the specified step, repeats that step, and repeats all steps that follow, back to the GoTo step. When the program has returned, or cycled, back to the step the specified number of times, the program moves on to the step that **follows** the GoTo step.

For example, consider a basic cycle sequencing protocol consisting of 30 repeats of the denaturation and annealing/extension steps. Rather than listing all 60 steps, use a GoTo step to design a short, easy-to-enter program:

Raw program:

1. 92° for 30 sec
2. 60° for 3 min
3. 92° for 30 sec
4. 60° for 3 min
5. 92° for 30 sec
6. 60° for 3 min
7. 92° for 30 sec

[continues for total of 60 lines]

Shortened program:

1. 92° for 30 sec
2. 60° for 3 min
3. GoTo step 1, 29 times (i.e., cycle back to step 1 and repeat steps 1, and 2, 29 more times)
4. End

Choosing a temperature control method

The MiniCycler can control block temperature in two ways, each of which has different implications for the speed and accuracy of sample heating:

- **Block control:** The MiniCycler adjusts the block's temperature to maintain the block at programmed temperatures, independent of sample temperature.
- **Probe control:** The MiniCycler adjusts the block's temperature to maintain an in-sample probe at programmed temperatures.

Block control

Under block control, the temperature of samples always lags behind the temperature of the block. The duration of this time lag depends on type of sample vessel and sample volume, but typically is between 10 and 30 seconds.

Probe control

Probe control is available for MiniCyclers equipped with the 16- or 25-well block. Special care must be taken to fill the probe with the correct amount of oil and to seat the probe and the samples correctly. Otherwise, actual sample temperatures can vary widely from the probe's temperature. Probe control cannot be used with microplates or slides.

Under probe control, the machine will slightly overshoot its heat targets to account for the time required for heat to transfer to the sensor within the probe tube. Therefore, probe control protocols typically require incubations that are 15–20 seconds shorter than incubations for block control protocols. For example, a denaturation step in a block-control protocol calling for 92°C for 30 sec would be reduced to 92°C for 10 sec in a probe-control protocol.

Entering a New Program

Programming the MiniCycler occurs in three stages:

1. Initiating the program
2. Naming the program
3. Entering the program's steps, including the End step

Each stage involves typing values from the keyboard or making selections from a menu. Programs may be edited as they are being entered or after they have been stored. Programs are automatically saved when the End step is entered.

Initiating the program

To initiate a new program, select *Enter Program* from the Run-Enter Menu, then press «Proceed». The Main Menu will be displayed:

_NEW	List
Edit	Delete

Select *New*, then press «Proceed». The naming screen will be displayed:

Name	A
------	---

Naming the program

Program names may be up to eight characters long and may consist of any combination of letters (Roman and Greek), numbers, punctuation marks, or Japanese *Katakana*.

Press the right «Select» key to scroll forward and the left «Select» key to scroll backward through the alphabets and characters available, which are presented in this order: Roman alphabet, selected Greek letters, punctuation marks, numbers. To access the Japanese *Katakana* syllabary, press the «.» key. A second press of «.» returns the machine to Western characters.

When the character needed is displayed next to *Name*, press «Proceed».

The character will be accepted, and the cursor will move one space to the right. Numbers and dashes may also be inserted by pressing the corresponding keys on the keypad.

When the name is complete, press «Proceed» once to accept the last character and again to accept the whole name. If the name is already in use for a program, a screen saying “Name In Use” will be displayed. If this happens, press «Proceed», then enter a different name.

Entering the program steps

When the name has been entered (CUSTOM1 is used in the following examples), the Enter Menu will be displayed:

```
Step 1    _TEMP
Goto Option End
```

Use this menu to enter each step of the program:

- *Temp* enters a temperature step.
- *GoTo* enters a GoTo step.
- *Option* enters an increment, extend, or slope step.
- *End* enters the End step.

Entering a temperature step

To enter a temperature step, select *Temp* from the Enter Menu, then press «Proceed». The first *Temp* screen will be displayed:

```
Step 1
Temperature _
```

The first line of this screen shows the number of the step being programmed (1 is used in the example above). The second line of the screen allows a target temperature (in degrees Celsius) to be entered for the step. Type any number between -9.0 and 105.0 as the target temperature (92.5 is used in the example below):

```
Step 1
Temperature 92.5
```


Press «Proceed». The temperature will be accepted, and a screen allowing entry of an incubation time will be displayed:

Step 1	Hrs	_
Min	Sec	

A value must be entered for *Hrs* (hours), *Min* (minutes), and *Sec* (seconds). Use the «Select» keys to move the cursor to each option, and do one of the following: type a number, or press «Proceed» to automatically enter a value of 0.

For example, to enter an incubation time of 30 seconds, press «Proceed» twice. The value of 0 will automatically be entered for *Hrs* and *Min*, and the cursor will be positioned at *Sec*. Use the keyboard to type "30." The screen will look like this:

Step 1	Hrs	0
Min	Sec	30_

Press «Proceed». The times will be accepted, and the Enter Menu will be displayed again. Use the Enter Menu to add another step to the program.

Entering a GoTo step

To enter a GoTo step, select *GoTo* from the Enter Menu and press «Proceed». The first *GoTo* screen will be displayed:

Step 2
Go to step _

The first line of this screen shows the number of the step being programmed (2 is used in the example above). The second line of the screen allows entry of the number of the step the program should cycle back to.

Type the number of the step the program should cycle back to (1 is used in the example below):

Step 2
Go to step 1

Press «Proceed». The number will be accepted, and a screen allowing

entry of an additional number of cycles will be displayed:

```
Step 2  Go to 1
_      more times
```

Type the additional number of times the program should cycle back to the step (24 is used in the example below):

```
Step 2  Go to 1
24     more times
```

Press «Proceed». The number will be accepted, and the Enter Menu will be displayed again. Use the Enter Menu to add another step to the program.

Entering an increment step

To enter an increment step, select *Option* from the Enter Menu and press «Proceed». The Options Menu will be displayed:

```
Step 3  _EXTEND
Increment Slope
```

Select *Increment* and press «Proceed». A screen allowing entry of the initial incubation temperature for the step will be displayed:

```
Step 3
Temperature _
```

Type a temperature and press «Proceed». The number will be accepted, and a screen allowing entry of an incubation time will be displayed:

```
Step 3
Min _  Sec
```

Type a number for *Min* and *Sec*. Press «Proceed» to automatically enter a value of 0.

When the numbers have been typed, press «Proceed». The numbers will be accepted, and a screen allowing entry of a temperature increment or decrement will be displayed:

Step 3 Add _ °/cycle

Type a temperature from -6.0 to 6.0°C (the maximum programmable range) as the number of degrees by which the incubation temperature will increase or decrease each time the step is executed in a cycle. Use a negative number to program a progressive decrease of temperature, and a positive number to program a progressive increase.

When the number has been typed, press «Proceed». The number will be accepted, and the Enter Menu will be displayed again. Use the Enter Menu to add another step to the program.

Entering an extend step

To enter an extend step, select *Option* from the Enter Menu and press «Proceed». The Options Menu (see above) will be displayed. Select *Extend* and press «Proceed». Type the initial incubation temperature and incubation time as for an increment step (see above), and press «Proceed». The temperature and time will be accepted, and a screen allowing entry of a time increment or decrement will be displayed:

Step 4 Extend _ S/cyc

Type a number from -60 to 60 (the maximum programmable range) as the number of seconds by which the incubation time will increase or decrease each time the step is executed in a cycle. Use a negative number to program a progressive decrease of incubation time, and a positive number to program a progressive increase.

When the number has been typed, press «Proceed». The number will be accepted, and the Enter Menu will be displayed again. Use the Enter Menu to add another step to the program.

Entering a slope step

To enter a slope step, two values must be specified:

- The desired temperature change for the step: This is the difference between the temperature of the block at the beginning of the slope step and the desired block temperature at the end of the slope step. For example, if the final temperature of the preceding step is 92°C

and the desired final temperature of the slope step is 65°C, the desired temperature change is 27°C.

- The slope rate for the step: This is a ratio of degrees per unit of time (usually degrees per second). Use the smallest convenient time increment to achieve the smoothest temperature curve for the step. For example, 0.1°C per second will give a smoother curve than 1°C per 10 seconds.

To enter a slope step, select *Option* from the Enter Menu and press «Proceed». The Options Menu (see above) will be displayed. Select *Slope* and press «Proceed». A screen allowing entry of the magnitude of the temperature change for the step will be displayed:

Step 4
Slope _ °

Type a number for the desired temperature change and press «Proceed». The number will be accepted, and a screen allowing entry of the rate of heating or cooling will be displayed:

_ °per Hrs
Min Sec

Type a number from 0.1 to 1.5 (the maximum programmable temperature range) for the numerator of the slope rate, and press «Proceed». The temperature will be accepted, and the cursor will move to *Hrs*. Type a time value for the denominator of the slope rate. A number must be entered for *Hrs*, *Min*, and *Sec*. Press «Proceed» to move the cursor to the next time element.

When the denominator has been typed, press «Proceed». The numbers will be accepted into the program, and the Enter Menu will be displayed again. Use the Enter Menu to add another step to the program.

Entering the End Step

To enter the End step, select *End* from the Enter Menu, then press «Proceed». A confirmation screen will be displayed:

Step 6
End

Press «Proceed». The End step will be entered into the program. The program will be stored, and the Run-Enter Menu will be displayed.

Programming an Indefinite Hold

Programs can be designed to end in an indefinite hold at a specified temperature. This allows completed reaction products to be held at a stable refrigeration temperature at the end of a run, until they can be conveniently removed.

To program an indefinite hold, program a temperature step immediately before the protocol's End step. Type the desired temperature for the indefinite hold, then give the step an incubation time of 0 hours, 0 minutes, and 0 seconds. When this temperature step is reached, the block will heat or cool the samples to the desired temperature and hold them there until you press «Proceed», or manually end the program by pressing «Cancel» or «Stop».

Creating a Program That Requires Manual Stepping

To create a program that must be manually progressed to each step, program an indefinite hold for each temperature step in the protocol (see "Programming an Indefinite Hold," above). When the protocol runs, you may then advance it through its steps at the time desired by pressing «Proceed».

Editing While Entering a Program

Changing the last value entered or last menu option chosen

To change the last value entered or last menu option chosen, press «Cancel». The choice just made will be cancelled, and another value may be entered or another menu option chosen. Press «Proceed» after changing a value, so that the program will accept it.

Changing all the values in the step being entered

To change all the values in the step currently being entered, repeatedly press «Cancel». Each time you press the key, the cursor will move backward through the values for the step, deleting them as it moves. When all values for the step have been deleted, enter new values, or press «Cancel» one more time to display the Enter Menu for the step. At this point you can re-enter the step.

Changing values in earlier steps of a program being entered

To change one or more values in earlier steps of the program you are entering, repeatedly press «Cancel» until the Enter Menu for the step you are working on is displayed. Press «Cancel» one more time to display the last-entered step. The step will be displayed in a special format:

CUSTOM1	3	94.0
	00:00:10	

In the example above, the last-entered step is a temperature step. The step number is 3, and the temperature for the step is 94.0°C. The incubation time is displayed in the form of 00:00:00. The first two digits represent hours, the second two digits represent minutes, and the last two digits represent seconds. The step in the example is programmed for a 10-second incubation.

At this point you may press the arrow keys to scroll through the program's steps. To change temperature, time, and cycling values see chapter 7.

Note: You cannot change the program name, step number, or step type. To change the program's name or the order or type of its steps, delete the program (see below) and then re-enter it.

After you have made all necessary changes, press the right «Select» key until the Enter menu is displayed for the next step in the program. Continue entering the new program.

Deleting an Incomplete Program

To delete a program that you are in the process of entering, enter the End step and press «Proceed», which will store the program. Then delete the program from memory (see below).

Deleting a Program from Memory

Any program in memory may be deleted. To delete a program, select *Enter Program* from the Run-Enter Menu and press «Proceed». The Main Menu will be displayed. Select *Delete* and press «Proceed». The deletion screen will be displayed for the first program in memory:

Delete CUSTOM1?

Press the «Select» keys until the name of the program to be deleted is displayed. When the correct name is displayed, press «Proceed». The program will be deleted, and the Run-Enter Menu will be displayed.

Listing a Program

Use the *List* option on the Main Menu to display all of a program's steps in the LCD window. To enter List mode, select *Enter Program* from the Run-Enter Menu and press «Proceed». The Main Menu will be displayed.
□ Select *List* and press «Proceed».

Press the «Select» keys until the name of the program to be listed is displayed, then press «Proceed». The first step in the program will be displayed in the LCD window. Press the right «Select» key to scroll forward and the left «Select» key to scroll backward through the list of steps.

To exit List mode, scroll to the last step of the program, then press the right «Select» key once. The Run-Enter Menu will be displayed.

Note: No program values can be changed in List mode. To change program values, see chapter 7 .

Printing a Program

If your MiniCycler has the optional 8-bit, 25-pin parallel printer port, it can print all the programs stored in the machine

To do this, follow the instructions under “Listing a Program” to enter List mode. If a printer is connected to the printer port, the list program screen will give you the option to print as well as list programs in the LCD window. Select *Print Programs* and press «Proceed». Each program stored in the machine will be printed.

The MiniCycler parallel printer port is compatible with Epson® LX-810 dot matrix printers and many other IBM compatibles. Only printers that support the Epson® extended graphics set will be able to print the degree character (°). Other printers will print an *x* instead of the degree character.

7

Editing Programs

Initiating Editing 7-2

Editing the Program 7-2

Editing temperatures and cycling values 7-2

Editing a time value 7-3

Editing a slope step 7-4

Initiating Editing

To edit an existing program the machine must be in Edit mode. To enter Edit mode, select *Enter Program* from the Run-Enter Menu and press «Proceed». The Main Menu will be displayed. Select *Edit* from the Main Menu and press «Proceed». A screen similar to the following will be displayed:

```

Edit
CUSTOM1?
    
```

The machine is now in Edit mode. Use the «Select» keys to select the program you wish to edit. When the name of the desired program is displayed, press «Proceed». The first step in the program will be displayed:

```

CUSTOM1  1  94.0
          00:00:20
    
```

The programmed temperature for this step is 94.0°C. The incubation time is displayed in the form of 00:00:00. The first two digits represent hours, the second two digits represent minutes, and the last two digits represent seconds. The step in the example is programmed for a 20-second incubation.

Editing the Program

Press the left «Select» key to scroll backward through the program steps, and the right «Select» key to scroll forward. As the left or right «Select» key is pressed, the cursor will progressively move backward or forward through each value in a step, and then back to the preceding or forward to the following step.

Editing temperatures and cycling values

To change a temperature or a cycling value, position the cursor on it and type the new value. Press «Proceed». The new value will be accepted, and the cursor will move forward to the next value in the step.

To cancel a change, press «Cancel». The just-typed value will disappear, and a blank line will be displayed in its place. Type a new value and press «Proceed».

Note: Be careful when editing program values. Once a value is changed or deleted, you cannot make it reappear.

Editing a time value

To edit any part of a time value, you must retype the number for each element in the time value: hours, minutes, and seconds. Press «Proceed» after typing each time element. The number just typed will be accepted, and the cursor will move to the next time element. To enter a value of 00 for hours, type a zero, then press «Proceed». The second zero will automatically be entered. To enter a value of 00 for minutes or seconds, simply press «Proceed». Both zeros will be entered at once.

For example, to change the time value for the example above from 00:00:10 to 00:00:30, press the right arrow key to move the cursor to the first digit of the time value:

```
CUSTOM1 1 94.0
  00:00:30
```

Type a zero. The zero will become the first digit of the hours element, and the values for minutes and seconds will disappear from the screen:

```
CUSTOM1 1 94.0
  0 : :
```

Press «Proceed». A second zero will automatically be entered for the hours element, and the cursor will move to the minutes element:

```
CUSTOM1 1 94.0
  00:_ :
```

Press «Proceed». Two zeros will automatically be entered for the minutes element, and the cursor will move to the seconds element:

```
CUSTOM1 1 94.0
  00:00:_
```

Type the number 30, then press «Proceed». The number will be entered

for the seconds element, and the cursor will move back to the first digit for the hours:

CUSTOM1	1	94.0
	00:00:30	

Editing a slope step

To edit a slope step you must retype both a sign (plus or minus) and a number. Press «.» (the period key) to enter a plus sign and «-» (the hyphen) to enter a negative sign. Press «Proceed» after entering the new slope step value.

8

Maintenance

Cleaning the Chassis and Block..... 8-2

Cleaning the Air Vents 8-2

**Cleaning Radioactive or Biohazardous
Materials Out of the Block 8-2**

Changing the Fuses 8-3

Cleaning the Chassis and Block

Clean the outside of the MiniCycler with a damp, soft cloth whenever something has been spilled on it or the chassis is dusty. A mild soap solution may be used if needed.

Clean the block's wells whenever anything is spilled into them or when the oil in them is discolored or contains particulate matter. It is particularly important to prevent the buildup of old, dirty oil, which will interfere with vessel seating and diminish thermal coupling of sample vessels to the block.

Clean the wells with a swab moistened with water, 95% ethanol, or, if a thorough cleaning is needed, a 1:100 dilution in water of 5.25% (0.7M) sodium hypochlorite (household bleach). If using sodium hypochlorite, swab the wells with water afterward to remove all traces of it. Do not clean the block with caustic or strongly alkaline solutions (e.g., strong soaps, ammonia, sodium hypochlorite at a higher concentration than specified above). These can damage the block's protective anodized coating.

Cleaning the Air Vents

Clean the air intake and exhaust vents with a soft-bristle brush, a damp cloth, or a vacuum cleaner whenever dust is visible in them. If these vents become clogged with dust and debris, airflow to the MiniCycler's heat sink is hampered, eventually causing overheating and shutdowns (see "Air Supply Requirements," chapter 3).

Cleaning Radioactive or Biohazardous Materials Out of the Block

When cleaning machines that have been running radioactive or biohazardous reactions, consult your institution's radiation safety officer or biosafety officer regarding methods, monitoring, and disposing of contaminated materials.

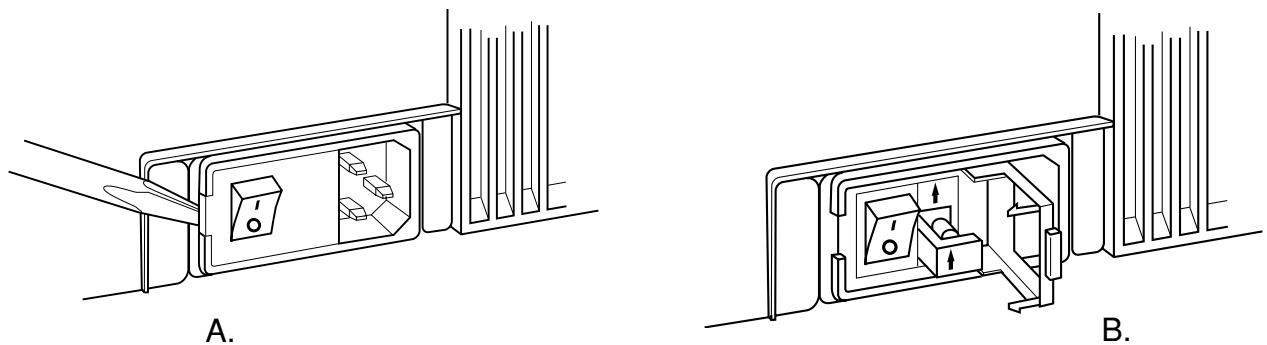
If mild cleaning agents do not remove radioactivity, harsher chemicals may be used. Users have suggested the detergent PCC-54 (Pierce Chemical Co., Rockford, Illinois; Pierce Eurochemie B.V., Holland), Micro Cleaning Solution (Cole-Parmer, Niles, Illinois), and Dow Bathroom Cleaner (available in supermarkets).

Caution: Harsh cleaning agents are corrosive to aluminum and must never be used on bare aluminum blocks. MJ RESEARCH blocks are anodized, so they have a protective coating of aluminum oxide. Still, harsh agents (such as those above) must be *thoroughly* rinsed away within a few minutes of application, or the anodization will degrade.

Changing the Fuses

The circuits in the MiniCycler are protected by two fuses (2.5TA 250V Slo-blo). When a fuse blows, the MiniCycler immediately shuts down and cannot be turned back on. The machine records the event as a power loss, so if a protocol is running when a fuse blows, the machine will resume the protocol run when the fuse is replaced and power restored (see “Resuming a Protocol after a Power Outage,” chapter 5).

⚡ Warning: The MiniCycler incorporates neutral fusing, which means that live power may still be available inside the unit even when a fuse has blown or been removed. Unplug the MiniCycler from the electrical outlet before opening its case. Never connect a power cord to the machine when the case is open.



Follow this procedure to change the fuse:

1. Move the power switch to the "0" (off) position. Disconnect the power cord from the side of the machine.
2. Insert one corner of a small flat-head screwdriver into the slot at the left side of the fuse block (fig. 8-1A). Gently pry the block loose, and pull it down and out. The fuses will be visible (fig. 8-1B).
3. Remove the fuses and examine them. A flat-head screwdriver may be used to pry the fuses loose, if necessary. A broken central wire or darkened glass indicates that a fuse has blown and should be replaced.
4. Gently press the fuse block back in place, and reconnect the power cord.



Troubleshooting

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Error Messages

Error Message	Cause	Action
Check ADC	Analog/digital converter not giving proper output.	Contact MJ RESEARCH or your distributor.
Check Block Sensor	Block's temperature sensor or sensor circuitry is malfunctioning.	Block may need servicing. Contact MJ RESEARCH or your distributor.
Check EEPROM	User Memory failing to store data.	Turn machine off, then on again. If error message appears again, contact MJ Research or your distributor.
Heat Pump Not Functioning	Heat pump is not working, so machine has shut down.	Cycler needs servicing. Contact MJ RESEARCH or your local distributor.
HS Overheating, Check Air Flow	Machine is not getting enough air, or air being taken in is not cool enough.	Ensure machine is at least 10cm away from walls and other equipment. Make sure air being taken in is cool (see chap. 3). If message persists, cycler may need servicing; contact MJ RESEARCH or your distributor.

Problems Related to Protocols

Problem	Cause	Action
Reaction is working but broad low molecular weight band is seen in gels.	Primer-dimer material often produces a broad band in the <100bp region of gels.	If obtaining appropriate reaction product/s, no need to change anything.
		Minimize primer-dimer production by designing primers with no 3' self-complementarity. Reoptimize magnesium concentration and annealing temperature to maximize desired product and minimize primer-dimers.
Reaction working but unexpected extra products or smear is seen.	Nonspecific hybridization occurring during setup.	Program a hot start into the protocol.
	Reaction component concentration too high or too low.	Check concentrations of components. May need to reoptimize magnesium concentration.
	Annealing temperature too low.	Reoptimize annealing temperature. It should be within a few degrees of the temperature for the primers using the nearest-neighbor method.
	Template not of sufficient purity.	Check extraction and purification protocols. Add additional purification steps if necessary.
	Multiple templates or host DNA in sequencing reactions.	Check nucleic acid preparations by gel electrophoresis.

(Continued)

Problem	Cause	Action
No reaction products obtained.	Protocol contains a wrong value.	Use List to check protocol's temperature control method, temperatures, and times.
	Reaction component omitted from mixture.	Check reaction assembly protocol, ensuring that mixture contains appropriate components in correct concentrations.
	Denaturation temperature too low.	Use 92° C for denaturation.
	Annealing temperature too high for primers.	Check for appropriate annealing temperatures of primers, using available computer programs or empirical testing.
	Probe failed, causing machine to run protocol under block control.	Check screen for probe failure error message. Probe may need servicing or replacing. Call MJ RESEARCH or your local distributor.
	Probe not filled with correct amount of oil.	Fill probe tube with correct amount of oil (see p. 4-11).
	Reaction mix contains an inhibitor (e.g., heme from blood).	"Spike" a complete reaction mix with a control template and primer set.
	Reaction vessels not making good thermal contact with sample block.	Use only high-quality tubes/plates that fit block snugly. Ensure that wells are free of foreign materials that would interfere with tube/plate seating.

Problems Related to Machine Performance

Problem	Cause	Action
Beeper sounds continuously (MiniCyclers with Hot Bonnet only).	Hot Bonnet is not plugged into the machine.	Cancel protocol. Plug Hot Bonnet cable into its port at back of machine, and restart protocol.
	Tubes are not in all 4 corners of the block so lid is tilted.	Put tubes (empty if necessary) in all 4 corners of the block.
	A microplate or flat-capped tubes are being used.	Need to use red metal adapter.
Block does not heat or cool.	Heat pump or fans are malfunctioning.	Contact MJ RESEARCH or your distributor.
Machine will not power up.	Power cable is not plugged into machine, electrical outlet is dead, or fuse has blown	Make sure power cord is plugged into machine correctly. Make sure electrical outlet is operational. Check fuses.
Machine starts in middle of a protocol when turned on.	Machine was turned off before protocol run had finished.	See p. 5-5.
Strange characters appear in LCD window, or text in window freezes or scrolls repetitively.	LCD window or machine microprocessor is malfunctioning.	Reset the microprocessor by turning machine off, then back on while pressing the «Cancel» key at the same time. If problem persists, contact MJ RESEARCH or your distributor.

10

Changing the Sample Block

Opening and Closing the MiniCycler's

Case 10-2

Changing the Sample Block 10-2

! **Caution:** Avoid changing the sample block frequently. The cables and connectors are fragile and can be damaged in as few as 50–100 uses.

Opening and Closing the MiniCycler's Case

Follow this procedure to open and close the MiniCycler's case:

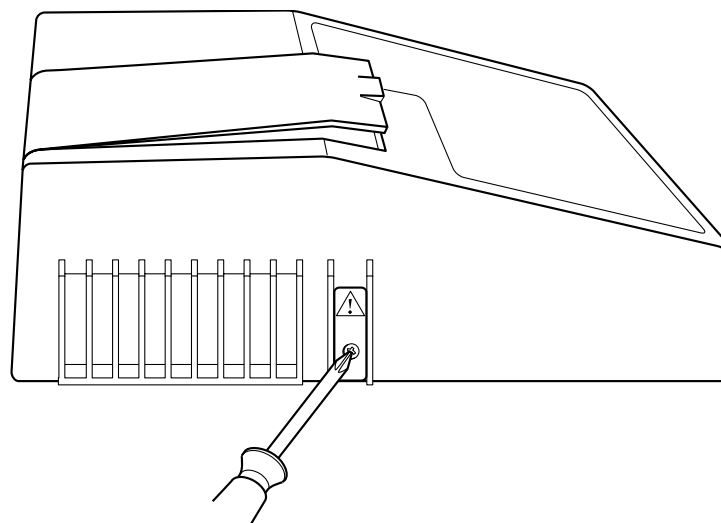
1. Unplug the power cord from the machine. Put the power switch in the "0" position.
2. Using a Philips-head screwdriver, remove the large black screw on the left side of the machine (fig. 10-1A).
3. Place the tip of a finger under the edge of the case, just beneath the screw hole, and pull outward and upward on the case. The case latch will release, and the case can then be lifted up and over to reveal the interior of the machine.

⚡ **Warning:** Do not plug the power cord into the machine while the case is open. You could receive a serious shock.

! **Caution:** The case is designed to open to a 90° angle. Do not force the case to open wider than this, to avoid damaging the case's hinge.

4. To close the case, push it down to cover the machine. Replace the screw holding the case to the machine base.

Figure 10-1 Opening the MiniCycler case.



Changing the Sample Block

Follow this procedure to change the sample block:

1. Unplug the power cord from the machine and open the case as described above.
2. Locate the block, and identify the two white ribbon cables exiting from it (fig. 10-2).
3. Grasp the ribbon cables just above their board connections and gently pull upward to disconnect them.
4. Locate the metal latch holding the block onto the circuit board (fig. 10-3). Press down on the latch, then pull the block toward the latch. The block will slide out over the latch (fig. 10-4).
- ✓ **Tip:** If you have trouble sliding the block out, insert the tip of a flat-head screwdriver between the latch and the bottom of the instrument. Use the screwdriver to push the latch down while sliding out the block.
5. Slide the new block into place; firm pressure may be required.
6. Reconnect the ribbon cables to their pins on the board. Make sure to align the pins with to the connectors correctly.
7. Close the case as described above.

Figure 10-2 Changing the sample block.

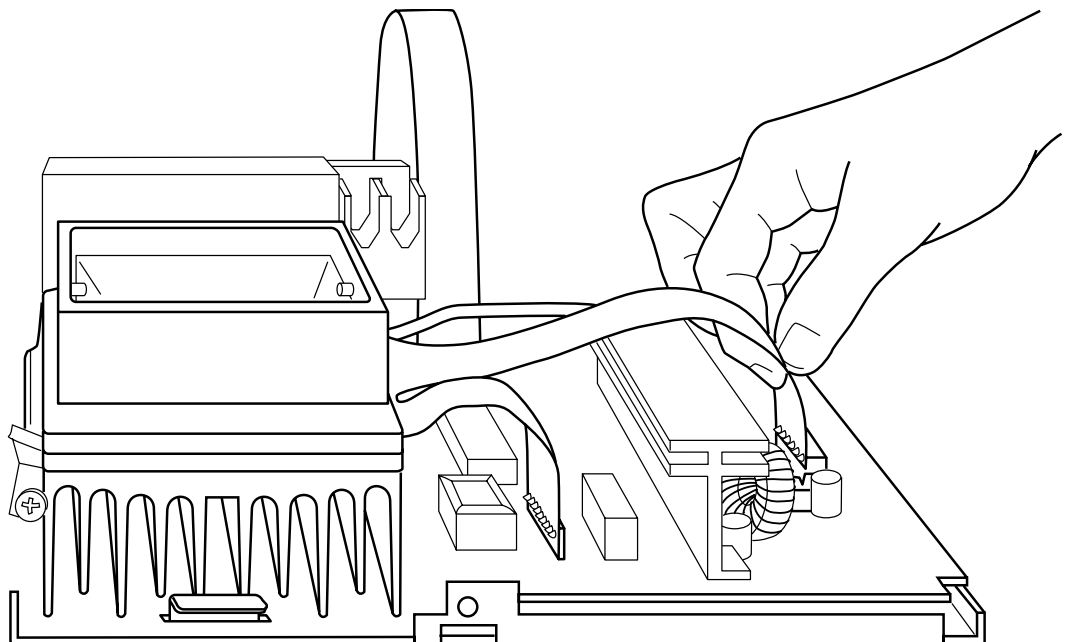


Figure 10-3 Releasing the sample block latch.

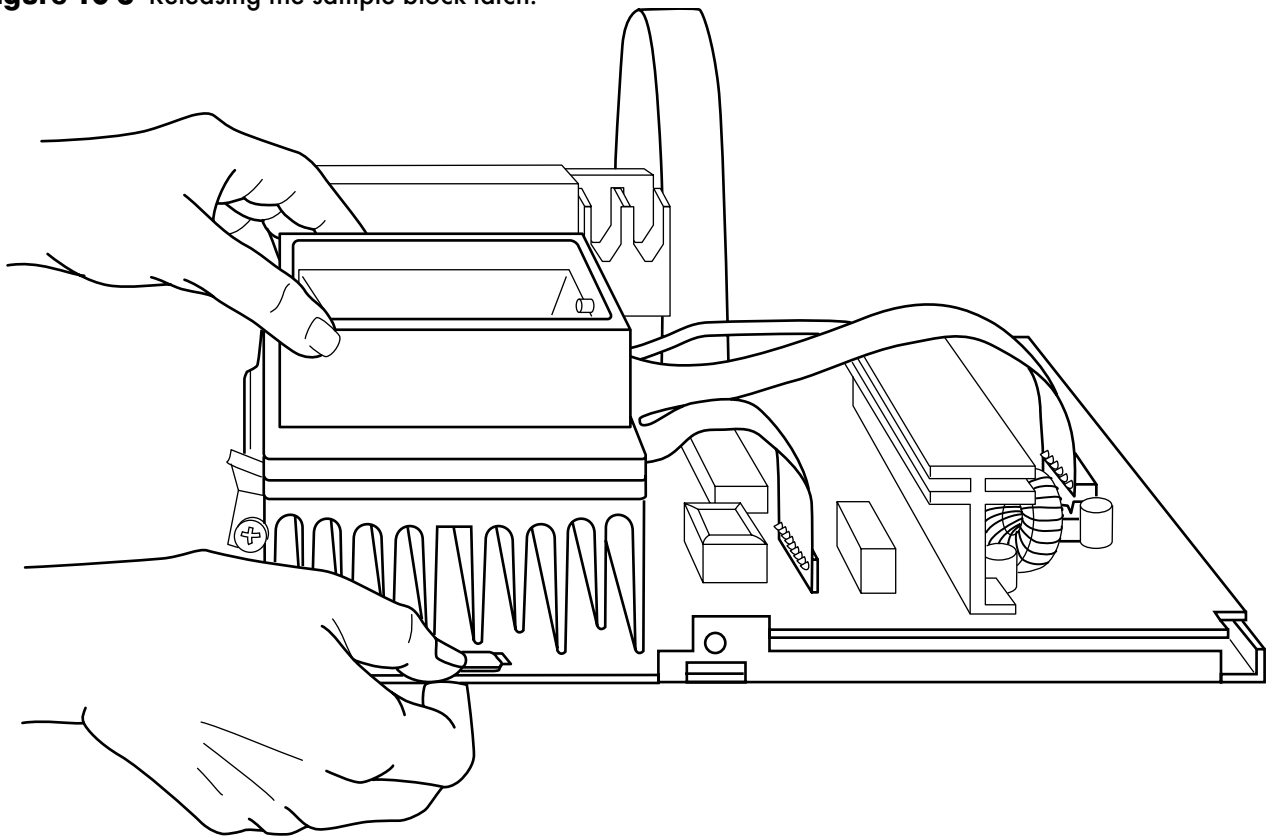
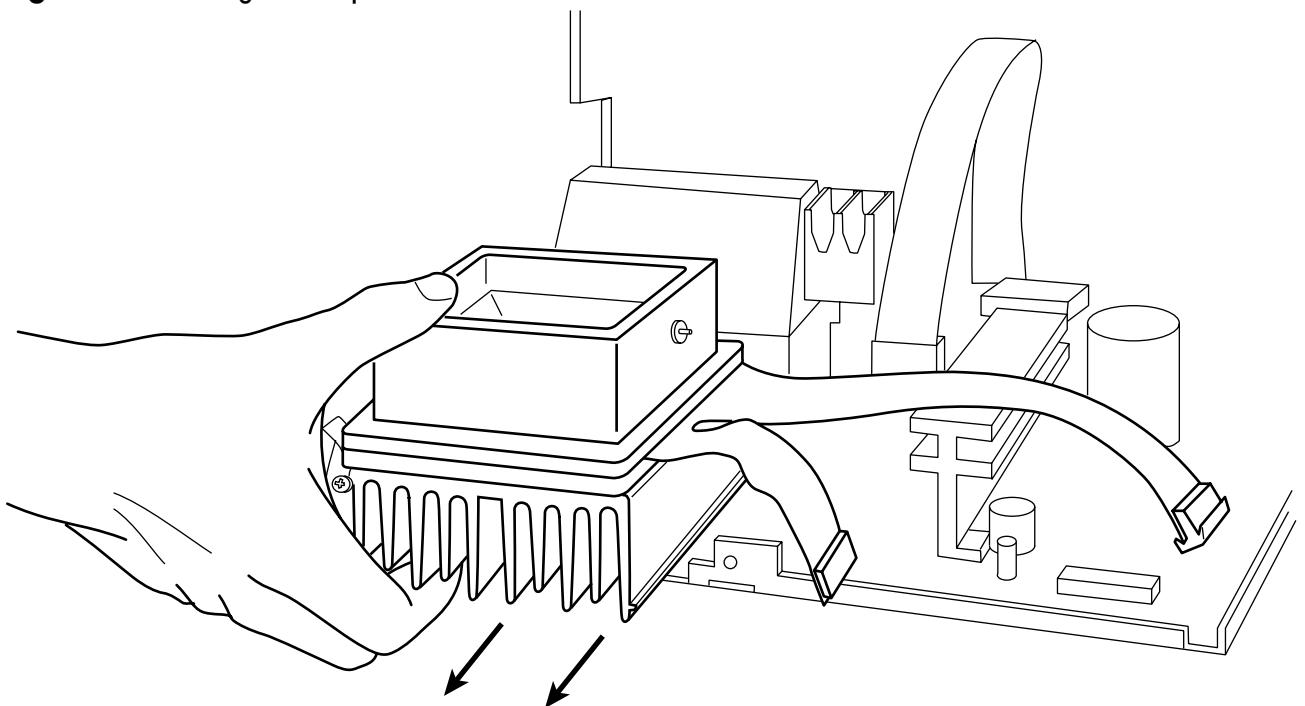


Figure 10-4 Sliding the sample block out.



Appendix A

Safety Warnings

- ! **Warning:** Operating the MiniCycler before reading this manual can constitute a personal injury hazard. Only qualified laboratory personnel trained in the safe use of electrical equipment should operate this machine.

- ⚡ **Warning:** Always disconnect the power cord before opening the base of the PTC-150 MiniCycler, to avoid receiving an electrical shock.

- ⚡ **Warning:** The MiniCycler incorporates neutral fusing, which means that live power may still be available inside the unit even if a fuse has blown or been removed. Although the machine will not be operational, you could still receive a shock if you fail to disconnect the power cord from the machine before opening the base, after a fuse has blown. Never open the MiniCycler's base with the power cord connected, and never connect the power cord to the machine when the base is open, even if a fuse has blown.

- 🔥 **Warning:** The MiniCycler Hot Bonnet inner lid can become hot enough to inflict serious burns during the course of normal operation. Use extreme caution if the Hot Bonnet is removed while still hot. Do not touch the inner lid when it is hot. Do not place a hot MiniCycler Hot Bonnet on a flammable surface, to avoid starting a fire.

- 🔥 **Warning:** The MiniCycler's block can become hot enough during the course of normal operation to cause burns or cause liquids to boil explosively. Wear safety goggles or other eye protection at all times during operation.

Safe Use Guidelines

The MiniCycler is designed to be safe to operate under the following conditions:*

- Indoor use
- Ambient temperature 4–32°C
- Relative humidity 10–90%, noncondensing
- Transient overvoltage per Installation Category II, IEC 664
- Pollution degree 2, in accordance with IEC 664

*Note: operation at altitudes significantly above sea-level may affect operation, particularly heat dissipation.

Electromagnetic Interference

The MiniCycler has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 of the US FCC Rules. These limits are designed to provide a reasonable protection against harmful interference when the equipment is operated in a commercial environment. This machine generates, uses, and can radiate radiofrequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this machine in a residential area is likely to cause harmful interference, in which case the user will be required to correct the interference at his or her own expense.

In addition, the MiniCycler design has been tested and found to comply with the EMC standards for emissions and susceptibility established by the European Union at time of manufacture.

FCC Warning

Changes or modifications to the MiniCycler not expressly approved by the party responsible for compliance could void the user's authority to operate the equipment.

Appendix B

Warranties

U.S. Domestic Warranty, Standard

MJ RESEARCH, Inc. warrants NEW MJ RESEARCH BRAND THERMAL CYCLERS (MODELS PTC-100, PTC-150, PTC-200 & PTC-225) against defects in material and workmanship for a period of two years from the date of purchase. If a defect is discovered, MJ RESEARCH, Inc. will, at its option, repair, replace, or refund the purchase price of the THERMAL CYCLER at no charge to the customer, provided the product is returned to MJ RESEARCH, Inc. within the warranty period. In no event will MJ RESEARCH, Inc. be responsible for damage resulting from accident, abuse, misuses, or inadequate packaging of returned goods.

ANY implied warranties, including implied warranties of the merchantability and fitness for a particular purpose, are limited in duration to TWO years from the date of original retail purchase of this product.

The warranty and remedies set forth above are exclusive and in lieu of all others, oral or written, expressed or implied. No MJ RESEARCH dealer, agent, or employee is authorized to make any modification, addition, or extension to this warranty, except in the form of the extended warranty outlined below.

MJ RESEARCH, Inc. is not responsible for special, incidental, or consequential damages resulting from any breach of warranty, or under any other legal theory, including downtime, lost samples or experiments, lost reagents, lost profits, goodwill, damage to or replacement of equipment, property, and any costs of recovering or reproducing experimental results and data.

Exclusions: This warranty applies only to machines sold in the U.S.A. Under no circumstance will MJ RESEARCH ship a repaired or replaced machine, or grant a refund of purchase price, to a user in a nation in which there was an authorized MJ RESEARCH distributor at the time of purchase. THIS WARRANTY IS NOT TRANSFERABLE FROM THE ORIGINAL PURCHASER TO A SUBSEQUENT OWNER. FURTHERMORE, THIS WARRANTY DOES NOT APPLY TO INSTRUMENTS USED OUTSIDE THE U.S.A., EXCEPT WHEN EXPRESSLY AUTHORIZED IN WRITING BY MJ RESEARCH, INC.

U.S. Domestic Extended Warranty, Optional

MJ RESEARCH, Inc. will offer to each ORIGINAL PURCHASER of an MJ RESEARCH BRAND thermal cyclers the opportunity to purchase an extension of the warranty coverage explained above for an additional two years. The coverage must be purchased through a purchase order received by MJ RESEARCH, Inc. within 30 days of receipt of the offer of extended warranty or the offer to renew the extended warranty. These offers apply only to machines sold and used in the U.S.A.

Some states OF THE U.S.A. do not allow the exclusion or limitation of incidental or consequential damages or limitations on how long an implied warranty lasts, so the above limitation or exclusion may not apply to you. This warranty gives you specific legal rights. You may also have other rights depending upon your state of residence.

All provisions of this warranty are voided if the product is resold, repaired, or modified by anyone other than MJ RESEARCH or an authorized distributor.

Appendix C

Shipping Instructions for US Residents

Users residing in the United States should follow these instructions for shipping a machine to MJ RESEARCH for factory repair or an upgrade. Users outside of the United States should send machines to their distributor, in accordance with shipping instructions obtained from the distributor.

1. Call MJ RESEARCH to obtain a return materials authorization (RMA) number. Machines returned without an RMA number will be refused by the Receiving Department.
2. Thoroughly clean the machine, removing oil and radioactive and other biohazardous substances. To protect the health of our employees, MJ RESEARCH will not repair or upgrade any machine that is excessively oily or that emits ionizing radiation upon arrival at our factory. **PLEASE ELIMINATE ALL BIOHAZARDS!**
3. Pack the machine in its original packaging. If this has been misplaced or discarded, call MJ RESEARCH to request shipment of packaging materials. You can also request a loaner machine, which will be provided if available (a rental fee may apply). You can use the loaner's packaging to return the machine needing repair.
4. Write the RMA number on the outside of the box.
5. Ship the machine (freight prepaid) to the following address. We recommend you purchase insurance from your shipper.

Ship to: Repair Department
MJ Research, Inc.
590 Lincoln St.
Waltham, MA 02451

Appendix D

PCR Licensing table

PCR* LICENSING INFORMATION

Background: The PCR process is covered by patents owned by Hoffmann-La Roche, Inc. and F. Hoffmann-La Roche Ltd. (collectively called Roche). Roche has granted exclusive and non-exclusive licenses for various applications of PCR, and Roche and its licensees provide end-user licenses within their designated fields. These licenses have different terms depending on the field, and different rules may apply in different countries. In the U.S. and most other nations, the user should always have proper license to perform PCR.

The following table reflects MJ Research’s understanding of the requirements set by Roche and its licensees for particular classes of users to purchase PCR licenses in the United States. This table is not endorsed by Roche, and all thermal cyclers users are encouraged to contact Roche for further information.

“Authorization” of thermal cyclers: The Perkin-Elmer Corporation holds the exclusive license from Roche to grant sublicenses to perform PCR in specific fields, such as research and forensics. Since 1992, Perkin Elmer has generally issued licenses in the form of a running royalty with sale of specific licensed enzymes, which carry a label stating that reactions must be run in an “authorized” thermal cycler.

MJ Research does not sell “authorized” thermal cyclers and believes that PE’s right to charge a thermal cycler “authorization” fee is probably legally unenforceable in the U.S. (this matter is under review by the U.S. Federal District Court). However, Perkin Elmer will sell such an “authorization” for any individual thermal cycler. It may be prudent in some circumstances to “authorize” an MJ cycler after purchase.

TYPE OF USE	PCR* for Human & Animal Diagnostics†		OTHER PCR† <i>RESEARCH, FORENSICS, ETC.</i>	NON-PCR† <i>CYCLE SEQUENCING, PRINS, ETC.*</i>
	<i>WITH DIAGNOSTIC SERVICE LICENSES†</i>	<i>WITH LICENSED TEST KITS†</i>		
THERMAL CYCLER LICENSE*	Usually None (see actual terms of license)	Usually None (see actual terms of)	Thermal Cycler “authorized” for PCR	None
ENZYME LICENSE*	Usually None (see actual terms of)	Usually None (see actual terms of)	Enzyme with PCR label license	None

N. B. For each column, both the indicated thermal cycler license and the indicated enzyme license are

* This table refers to PCR licenses only; other processes, and particular types of enzyme or thermal cycler, may require separate license.

† For definitive information on where your application fits, please contact Roche.

Addresses for Contacting Hoffmann-LaRoche:

In the United States:
Kathe Kiehn
Licensing Manager
Roche Molecular Systems, Inc.
1145 Atlantic Avenue
Alameda CA 94501 USA
(510)814-2970 • Fax: (510)814-2977

In other nations:
Dr. Agnieszka Junosza-Jankowski
PCR licensing Manager
F. Hoffmann-La Roche Ltd.
Building 222/350
CH-4002 Basel, Switzerland
41 61 687 3031 • Fax: 41 61 687 2113

Note: Please do not construe the information on this page as legal advice—for more information, please contact your institution’s legal department.

Appendix E

CE Conformity

DECLARATION OF CONFORMITY

MJ RESEARCH, INC. hereby declares that the MJ RESEARCH PTC-150 MiniCycler™ thermal cycler conforms to the following directives and standards:

APPLICATION OF COUNCIL DIRECTIVES: 89/336/EEC & 73/23/EEC

STANDARDS TO WHICH CONFORMITY IS DECLARED: EN55011; EN50082-1; Canadian Standards Association (CSA) Std. 1010 (EMC for industrial, scientific, medical equipment; EMC for generic immunity; compliance to low-voltage directive respectively)

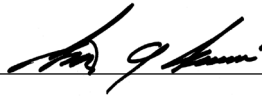
TYPE OF EQUIPMENT: Scientific Equipment (for Laboratories)

MODEL NUMBER: PTC-150

YEARS OF MANUFACTURE: 1997 and onward

MJ RESEARCH INC, as manufacturer of the equipment described above, certifies that this model has been tested and conforms to the applicable Directives and Standards of the European Union (EU) as described above. Test data to verify this conformity are available for inspection at the MJ RESEARCH, INC. at the address listed below. The instrument in question carries the “CE” mark on the nameplate.

MANUFACTURER’S OFFICIAL:



Robert A Iovanni
Senior Electrical Engineer
MJ RESEARCH, INC.
590 Lincoln Street
Waltham, Massachusetts 02451
UNITED STATES OF AMERICA
Tel: (617) 923-8000
Fax: (617) 923-8080

Date: 8/5/99

Appendix F

Factory-Installed Protocols

Note: A programmed time of 0:00:00 means “hold at this temperature forever.”

Name	Temp. Control Method	Step	Temp. & Time
ICEBUKET	Block	1	4°, 0:00:00
		2	END
LIGATION	Block	1	15°, 0:00:00
		2	END
37°-1 HR	Block	1	37°, 1:00:00
		2	4°, 0:00:00
		3	END
37°-6 HR	Block	1	37°, 6:00:00
		2	4° 0:00:00
		3	END
65°	Block	1	65°, 0:00:00
		2	END
DENATURE	Block	1	95°, 0:05:00
		2	END
BOIL	Block	1	100°, 0:05:00
		2	END
CUT&KILL	Block	1	37°, 1:00:00
		2	70°, 0:05:00
		3	END

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