Thermal Cycler Temperature Verification System

For GeneAmp® PCR Systems 2400, 9600, 9700

User's Manual

Applied Biosystems

For Research Use Only. Not for use in diagnostic procedures.

Contents

Temperature Verification System Overview 1

Introduction

Tests Performed

Use the Temperature Verification System to perform the following tests on your GeneAmp PCR Systems:

- Temperature Calibration Verification Test
- Temperature Uniformity Test

Parts Included

Note Carefully unpack the kit and check the parts included against the following list. If any part is damaged or missing, contact the shipping carrier and Applied Biosystems immediately.

- Digital Thermometer with 9V battery installed
- RTD Probe Assembly
- Cotton swabs (P/N 401–0066)
- Light Mineral Oil (P/N 186–2302)
- Probe Trays:
  - 2400 (P/N N8031076)
  - 9600 (P/N N801–3837)
  - 9700 (P/N N8050305)

IMPORTANT Your RTD probe and digital thermometer have been calibrated together at the factory. Therefore, do not use the probe or digital thermometer with any other probe or digital thermometer; they are not interchangeable.

Using the Digital Thermometer

Overview The handheld digital thermometer (Figure 1) is used with a platinum resistance temperature device (RTD) probe to verify that your thermal cycler is still within calibration tolerance. It is also used to test the uniformity of your instrument's sample block. The digital thermometer has a temperature range of -10 to 110 °C and is accurate to within +/-0.3 °C.

IMPORTANT Do not try to recalibrate or perform any service on the digital thermometer. The only user-serviceable component in the unit is the battery.

Instructions Observe the following when using the digital thermometer:

- Make sure the RTD probe is connected to the input connector located at the top of the digital thermometer.
- To turn on the digital thermometer, move the on/off range switch to the 200 position.

IMPORTANT Do not use the 630 range.

- The temperature measured by the RTD probe appears on the digital thermometer display in degrees Celsius.
- When you complete the test, move the ON–OFF/RANGE switch to the OFF position.

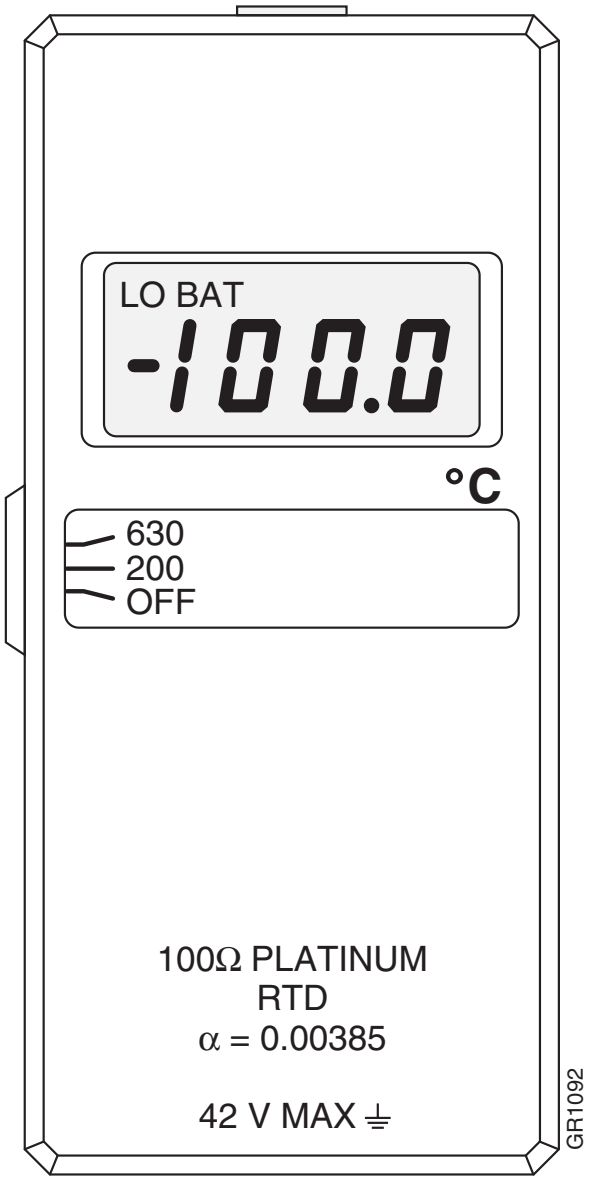

Figure 1 Front view of digital thermometer

Replacing the Battery

The digital thermometer is powered by a 9V battery, which has been installed at the factory. If the battery is low, the “LO BAT" indicator will appear on the display. Install a new battery immediately if the “LO BAT" indicator appears.

To replace the battery:

Step	Action
1	Remove the three 7/16-in. long screws securing the back cover of the digital thermometer (Figure 2).
2	Install the 9 V battery as shown in Figure 2. Make sure the battery terminals are contacting the battery clips.
3	Replace the back cover and secure it with the three screws removed in step 1.

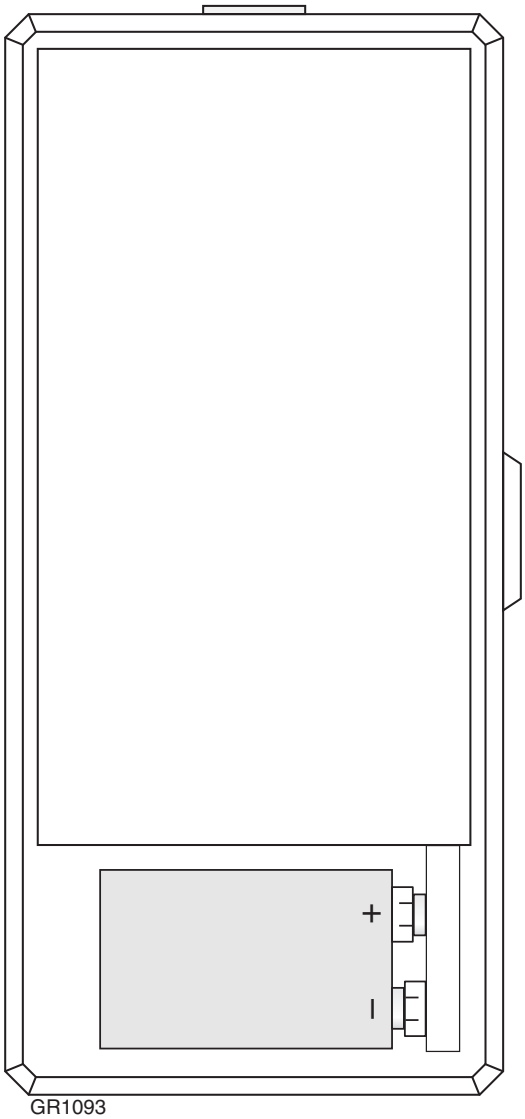

Figure 2 Digital thermometer shown with back cover removed

Temperature Display Differences

The sample temperature display on the instrument will be different than the display on the digital thermometer during heating or cooling transitions. This is because the digital thermometer measures block temperature while the thermal cycler measures sample temperature. The instrument sample temperature display is a function of the tube type and the reaction volume.

Using the Probe While Running Programs

We recommend that you use the RTD probe and digital thermometer only for the temperature calibration verification test and the temperature uniformity test, which are described in these instructions on page 1-2. If you use the RTD probe and digital thermometer while running programs other than those used in the two tests, be aware that the accuracy of the RTD probe will decrease due to the effect of the heated sample block cover.

The heated cover normally operates at 105 °C. The effect of the heated cover on the RTD probe decreases as the temperature of the sample block approaches 99.9 °C.

Getting the Temperature Verification System Recalibrated

Overview

IMPORTANT Do not try to recalibrate or perform any service on the digital thermometer. The only user-serviceable component in the unit is the battery.

We recommend that your Temperature Verification System be recalibrated once a year.

In the United States and Canada

If you are in the United States or Canada, get your Temperature Verification System serviced by calling the Applied Biosystems Service Repair Center at (800) 831-6844. The Service Repair Center will give you a Return Authorization Number and an address to which you can send your Temperature Verification System.

Outside the United States and Canada

If you are outside the United States, call your local Applied Biosystems Sales/Service office. See page 1-10 for a listing of Sales offices, and their telephone and Fax numbers.

IMPORTANT When shipping your Temperature Verification System, ship the meter and probe in the black case provided, and enclose a Decontamination Certificate.

Decontaminating The Temperature Probe

Decontaminate the temperature probe by gently swabbing the probe cone with a cotton swab, using a 20% Clorox/80% water mixture. Observe the following precautions when decontaminating your probe:

- Do not submerge the probe in Clorox.
- Do not disassemble the the probe assembly.
- Do not separate the cones.

CERTIFICATE OF DECONTAMINATION

Return Authorization:

1. This document must be completed in full, signed by the customer/operator of all Applied Biosystems equipment, and handled as follows:

- Attach to all Applied Biosystems products prior to returning to either a Applied Biosystems manufacturing facility, Field Office, or Service Repair Center for the purpose of repair, refurbishing, trade in, or replacement.
- Provide to all Applied Biosystems Representatives before they perform service on any Applied Biosystems equipment which has been located within a biological or radioactive materials laboratory.

Instrument/model/type:

_____

Instrument serial number:

_____

Reason for return/service:

_____

RMA or RAN number:

_____

Decontamination Information and Procedure:

1. Has this product been exposed to any infectious agents assigned to biosafety levels 2, 3, or 4 ?

_____

If so, please indicate agent and biosafety hazard class:

_____

2. Has this product been exposed to toxic, carcinogenic, or radioactive substances?

_____

If so, please indicate types and quantities used:

_____

3. Indicate how the equipment was cleaned or disinfected by checking one or more of the decontamination procedures described below:

Contact with infectious agents:

- The equipment was thoroughly decontaminated by spraying it or wiping it down with a 1:10 dilution of Clorox brand bleach, or equivalent, with water.
- The disinfectant remained on the unit for a minimum of 10 minutes before flushing with water.

Contact with radioactive material:

- The equipment was thoroughly decontaminated by spraying it or wiping it down with a commercially available decontaminate (i.e., Radiacwash) or equivalent.
- The instrument must be surveyed with a Geiger meter and swipe tested with a scintillation counter. Results must be attached to this form.

Chemical contamination:

- All chemical bottles have been removed from the instrument.
- The equipment was thoroughly decontaminated by rinsing areas associated with chemicals with a solvent such as alcohol or water.

Chemical contamination to valve blocks:

The valve block was flushed and dried with appropriate gas (argon, helium, or nitrogen).

In the event that a valve block cannot be flushed properly, label it as a HAZARDOUS WASTE SOLID and dispose of it in accordance with all local, state, federal and environmental health regulation and laws.

Other:

Please describe how equipment was cleaned or disinfected. Attach additional paper, if necessary.

_____

_____

_____

By accepting authorization to return this product, I assume all responsibility and liability for biological, chemical, and radiological decontamination and cleaning. I understand that Applied Biosystems has no obligation to repair or service any product unless adequate information is provided to ensure the safety of all personnel handling the exposed product.

Print
Name:_____Title:_____

Company/Institution: _____ Phone Number:_____

Signature: _____Date: _____
Fax Number_____

Technical Support

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You can contact Applied Biosystems for technical support by telephone or fax, by e-mail, or through the Internet. You can order Applied Biosystems user documents, MSDSs, certificates of analysis, and other related documents 24 hours a day. In addition, you can download documents in PDF format from the Applied Biosystems Web site (please see the section "To Obtain Documents on Demand" following the telephone information below).

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Contact technical support by e-mail for help in the following product areas:

Product Area	E-mail address
Genetic Analysis (DNA Sequencing)	galab@appliedbiosystems.com
Sequence Detection Systems and PCR	pcrlab@appliedbiosystems.com
Protein Sequencing, Peptide and DNA Synthesis	corelab@appliedbiosystems.com
Biochromatography, PerSeptive DNA, PNA and Peptide Synthesis systems, CytoFluor®, FMAT™, Voyager™, and Mariner™ Mass Spectrometers	tsupport@appliedbiosystems.com
LC/MS (Applied Biosystems/MDS Sciex)	apisupport@sciex.com or api3-support@sciex.com
Chemiluminescence (Tropix)	tropix@appliedbiosystems.com

Hours for Telephone Technical Support

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Product	Hours
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All Other Products	5:30 a.m. to 5:00 p.m. Pacific Time

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To contact Applied Biosystems Technical Support, use the telephone or fax numbers given below. (To open a service call for other support needs, or in case of an emergency, dial **1-800-831-6844** and press **1**.)

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ABI PRISM® 3700 DNA Analyzer	**1-800-831-6844**, then press **8**	**1-650-638-5981**
DNA Synthesis	**1-800-831-6844**, then press **21**	**1-650-638-5981**
Fluorescent DNA Sequencing	**1-800-831-6844**, then press **22**	**1-650-638-5981**
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Product or Product Area	Telephone Dial...	Fax Dial...
Integrated Thermal Cyclers (ABI PRISM® 877 and Catalyst 800 instruments)	**1-800-831-6844**, then press **24**	**1-650-638-5981**
ABI PRISM® 3100 Genetic Analyzer	**1-800-831-6844**, then press **26**	**1-650-638-5981**
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Peptide Synthesis (433 and 43X Systems)	**1-800-831-6844**, then press **31**	**1-650-638-5981**
Protein Sequencing (Procise® Protein Sequencing Systems)	**1-800-831-6844**, then press **32**	**1-650-638-5981**
PCR and Sequence Detection	**1-800-762-4001**, then press **1** for PCR, **2** for the 7700 or 5700, **6** for the 6700 or dial **1-800-831-6844**, then press **5**	**1-240-453-4613**
Voyager™ MALDI-TOF Biospectrometry and Mariner™ ESI-TOF Mass Spectrometry Workstations	**1-800-899-5858**, then press **13**	**1-508-383-7855**
Biochromatography (BioCAD® Workstations and Poros® Perfusion Chromatography Products)	**1-800-899-5858,** then press **14**	**1-508-383-7855**
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PNA Custom and Synthesis	**1-800-899-5858**, then press **15**	**1-508-383-7855**
FMAT™ 8100 HTS System and Cytofluor® 4000 Fluorescence Plate Reader	**1-800-899-5858**, then press **16**	**1-508-383-7855**
Chemiluminescence (Tropix)	**1-800-542-2369** (U.S. only), or **1-781-271-0045**	**1-781-275-8581**
Applied Biosystems/MDS Sciex	**1-800-952-4716**	**1-650-638-6223**

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Australia (Scoresby, Victoria)	**61 3 9730 8600**	**61 3 9730 8799**
China (Beijing)	**86 10 64106608**	**86 10 64106617**
Hong Kong	**852 2756 6928**	**852 2756 6968**
Korea (Seoul)	**82 2 593 6470/6471**	**82 2 593 6472**
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Japan (Hacchobori, Chuo-Ku, Tokyo)	**81 3 5566 6230**	**81 3 5566 6507**
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http://www.appliedbiosystems.com/techsupp

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3	Enter the requested information and your question in the displayed form, then click **Ask Us RIGHT NOW** (blue button with yellow text).
4	Enter the required information in the next form (if you have not already done so), then click **Ask Us RIGHT NOW**. You will receive an e-mail reply to your question from one of our technical experts within 24 to 48 hours.

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Temperature Verification Tests for the 2400 2

Temperature Verification Tests

Types of Tests

- Calibration Verification tests the sample block against temperature accuracy specifications.
- Temperature Non-Uniformity tests the temperature uniformity in the sample block.

System Contents

Equipment Required

The Temperature Verification System is required to perform these tests. The Temperature Verification System should include the following:

- Digital thermometer with 9V battery installed
- RTD probe
- Cotton swabs
- Light mineral oil
- 2400 probe tray

IMPORTANT Refer to the instructions included with your Temperature Verification System for a detailed description of digital thermometer operation.

Preparing the RTD Probe Assembly for Tests

Description The RTD Probe Assembly consists of two cones, one of which measures the temperature of the sample well. The wire is attached to the cone that does not measure the temperature of the sample well. This cone is a dummy probe.

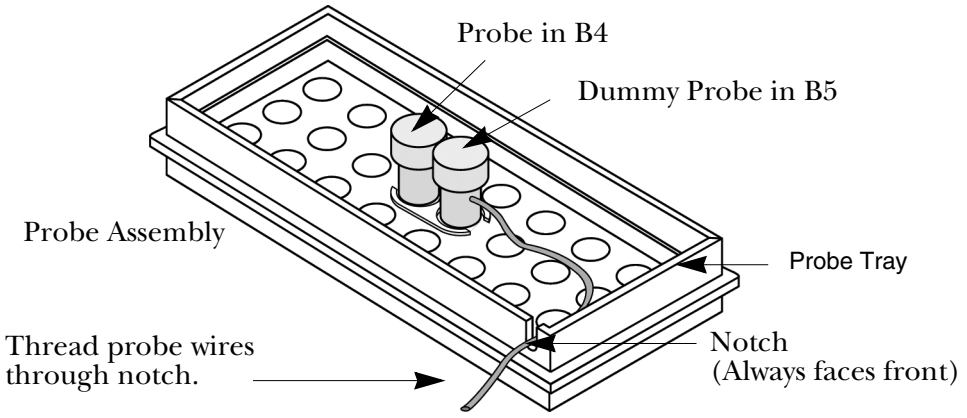
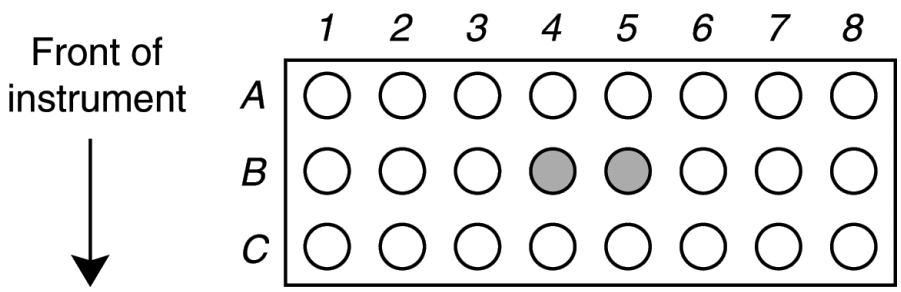


Figure 2-1 RTD Probe Assembly

Preparation **! WARNING ! BURN HAZARD! The sample block is hot to the touch and can cause burns.**

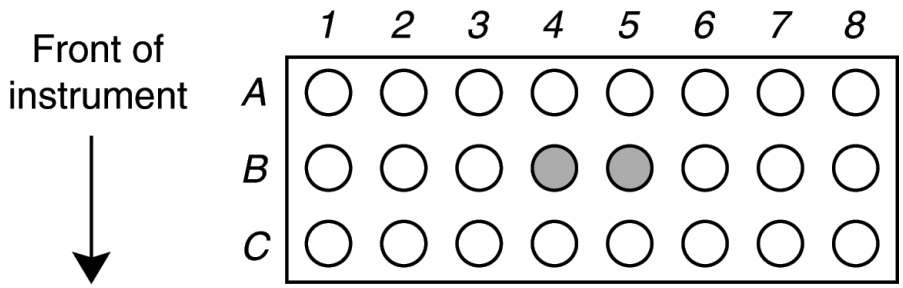
To prepare the RTD Probe Assembly for the Calibration Verification Test:

Step	Action
1	If the System 2400 heated cover is in the forward position, lift the lever, then slide the cover back.
2	Using a cotton swab, coat wells B4 and B5 with mineral oil. 
3	Place the probe tray on the sample block so that the notch faces the front of the instrument.
4	Place the Probe Assembly into wells B4 and B5 so that the dummy probe sits in B5.
5	Thread the probe wire through the notch in the probe tray.
6	Connect the probe to the digital thermometer.
7	Slide the heated cover forward and pull the lever down.

To prepare the RTD Probe Assembly for the Calibration Verification Test:

Step	Action
8	Turn on the digital thermometer by moving the ON-OFF/RANGE switch to the 200 position

To prepare the RTD Probe Assembly for the Temperature Non-Uniformity Test

Step	Action
1	If the System 2400 heated cover is in the forward position, lift the lever, then slide the cover back.
2	Using a cotton swab, coat wells A2 and A3 with mineral oil. 
3	Place the probe tray on the sample block so that the notch faces the front of the instrument.
4	Place the Probe Assembly into wells A2 and A3 so that the dummy probe sits in A3.
5	Thread the probe wire through the notch in the Probe tray (Figure 2-1).
6	Connect the probe to the digital thermometer.
7	Slide the heated cover forward and pull the lever down.
8	Turn on the digital thermometer by moving the ON-OFF/RANGE switch to the 200 position.

Temperature Calibration Verification Test

Overview

The RTD probe and the digital thermometer are used to take temperature readings of one sample well at three different setpoint temperatures:

- 92 °C
- 56 °C
- 20 °C

Description of the Beginning Test Functions

When the Calibration Verification Test begins, the block and heated cover each move toward the first setpoint of 92 °C. The block temperature will increase and the cover temperature decrease. After the heated cover reaches 92 °C and the block reaches the temperature of the heated cover ± 10 °C, the instrument pauses three minutes to stabilize the temperature.

! WARNING ! BURN HAZARD! The sample block is hot to the touch and can cause burns.

The Stabilizing at Setpoint screen appears. This is the start of the 3 minute stabilization period.

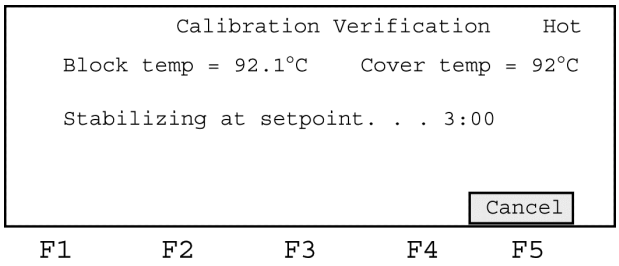


Figure 2-2 Stabilizing at Setpoint Screen

When three minutes have elapsed, the Setpoint screen will be displayed (Figure 2-3). The digital thermometer reading of the sample well temperature is entered into the Enter Actual Temp field. This process is repeated at each of the three setpoints.

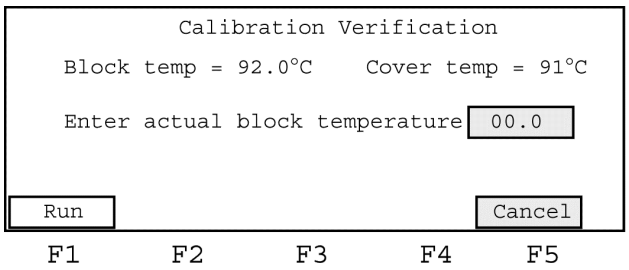
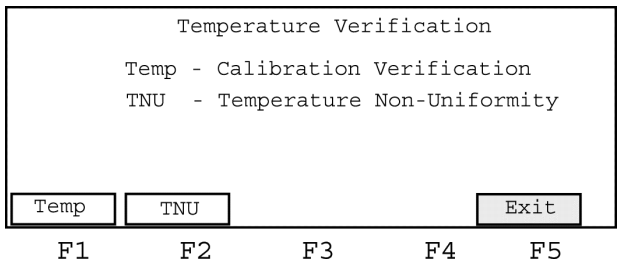
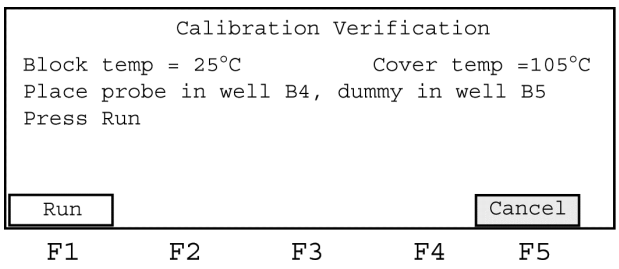
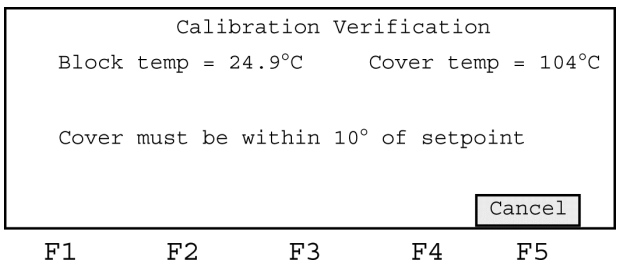


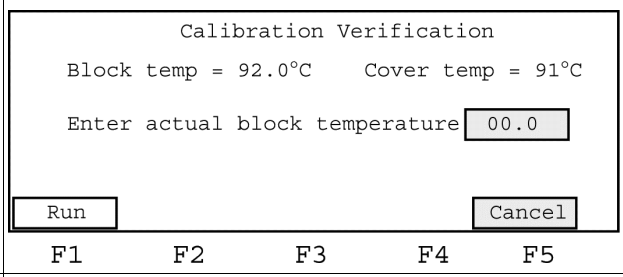
Figure 2-3 Setpoint screen for entry of digital thermometer readings

Performing the Temperature Calibration Verification Test

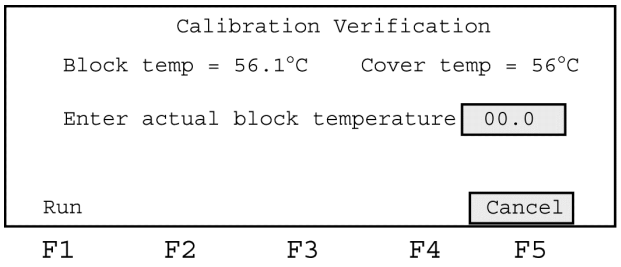
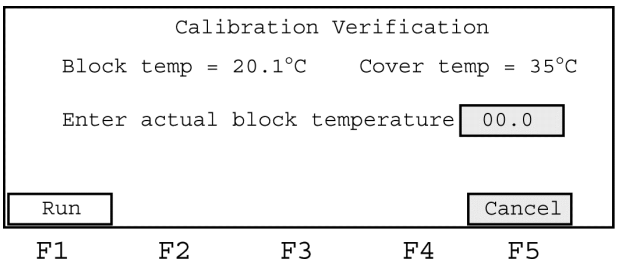
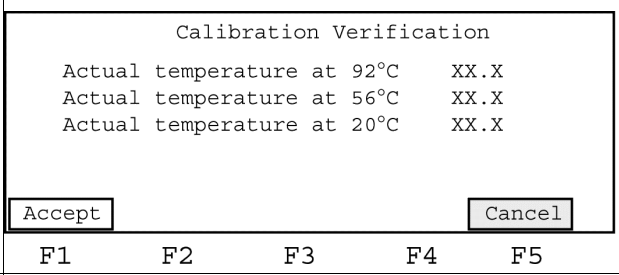
To perform the first segment of the calibration test:

Step	Action
1	From the Diagnostics screen, press F3-TmpVer. This displays the Temperature Verification screen. 
2	Press F1-Temp. This displays the Calibration Setup screen.  This screen instructs you to set up the RTD probe assembly.
3	After installing the probe assembly, slide the heated cover forward and pull the lever down.
4	Press F1-Run to initiate the Calibration Verification Test. This automatically verifies the temperature of the sample block, and displays the Setpoint Values screen.  **Note** To exit the test at any time, press F5-Cancel

To perform the first segment of the calibration test: *(continued)*

Step	Action
5	When the temperature stabilizes at 92 °C and the counter decrements to zero, read the digital thermometer. 
6	Use the numeric keys to type the value displayed on the digital thermometer into the highlighted Enter actual block temperature field.

To perform the second segment of the calibration test:

Step	Action
1	Press Enter. This will initiate the second segment of the temperature calibration process. The heated cover and sample block now approach the second setpoint temperature, 56 °C. 
2	When the temperature stabilizes at 56 °C and the counter decrements to zero, read the digital thermometer.
3	Use the numeric keys and type the value displayed on the digital thermometer in the highlighted Enter actual block temperature field.
4	Press Enter. This will initiate the third segment of the temperature calibration process. The heated cover and sample block will approach the third setpoint temperature, 20 °C. 
5	When the temperature stabilizes at 20 °C and the counter decrements to zero, read the digital thermometer.
6	Use the numeric keys and type the value displayed on the digital thermometer in the highlighted Enter actual block temperature field.
7	Press Enter. This is the last temperature entry. The GeneAmp 2400 displays the digital thermometer measurements in the right hand column of the Calibration Verification screen. 
8	Press F1-Accept to accept the calibration or F5-Cancel to cancel the calibration.
9	Turn off the digital thermometer, remove the probe assembly and clean the oil from the sample block.

Calibration Verification Specification

The digital thermometer reading at each setpoint must be +0.30 °C of the instrument setpoint temperature.

Values that are out of specification cannot be saved by pressing FI-Accept.

Temperature Non-Uniformity Test

Overview The Temperature Non-Uniformity test uses the RTD probe assembly and a digital thermometer to test the temperature uniformity of seven different wells (see the list below and Figure 2-4) in the sample block.

- A2
- A7
- B1
- B4
- B8
- C2
- C7

These wells are tested at two different setpoint temperatures:

- 94 °C
- 37 °C

Schematic A schematic representation of the Temperature Non-Uniformity Test is shown below.

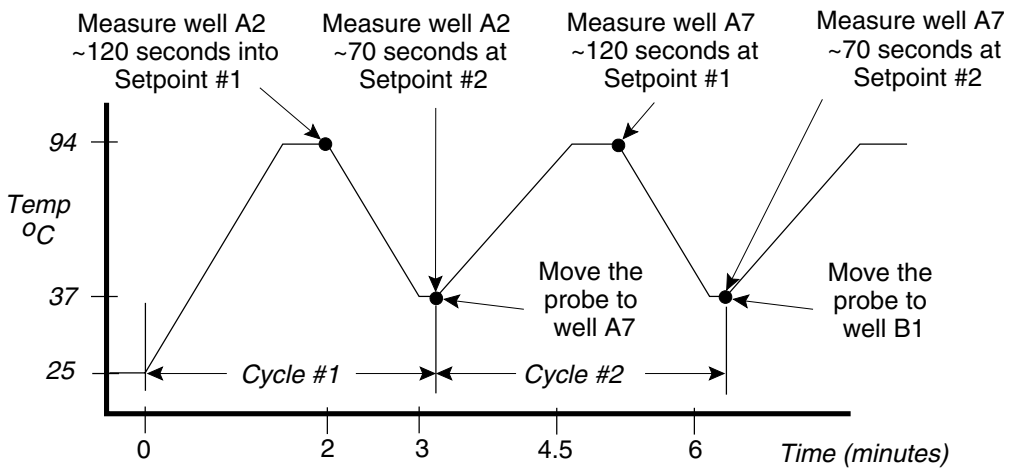


Figure 2-4 The Temperature Non-Uniformity test

Description of Beginning Test Functions

About 90 seconds after the test begins, the Stabilizing at Setpoint screen is displayed (Figure 2-5).

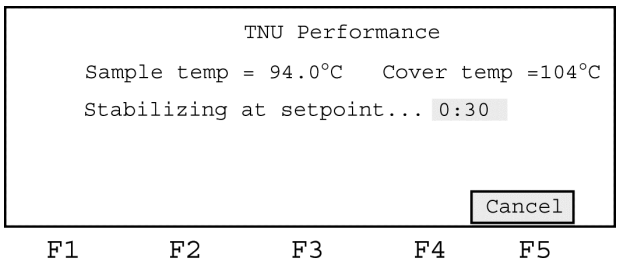


Figure 2-5 The Setpoint Screen

The instrument pauses 30 seconds to allow the temperature to stabilize. A digital thermometer reading is taken and entered using the numeric keypad.

About 40 seconds after the digital thermometer reading is entered, the heated cover and sample block approach the second setpoint value of 37 °C. Again, the instrument pauses 30 seconds to allow the temperature to stabilize. A digital thermometer reading is taken and entered using the numeric keypad.

This process is repeated in seven different sample block wells.

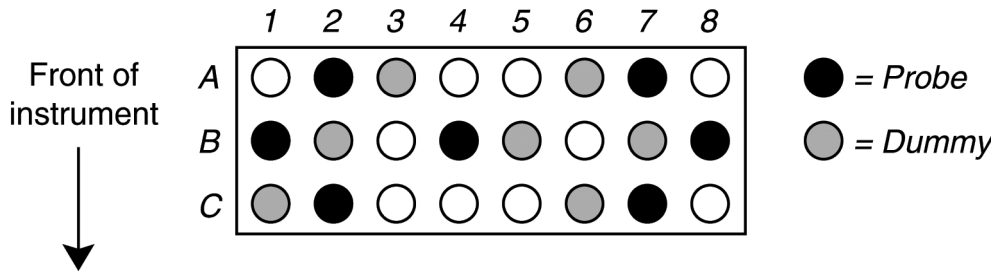
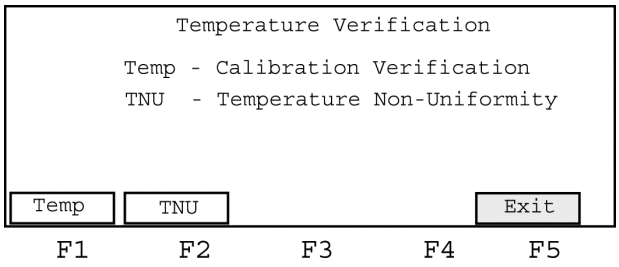
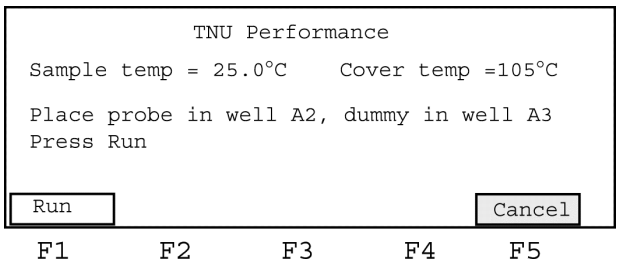
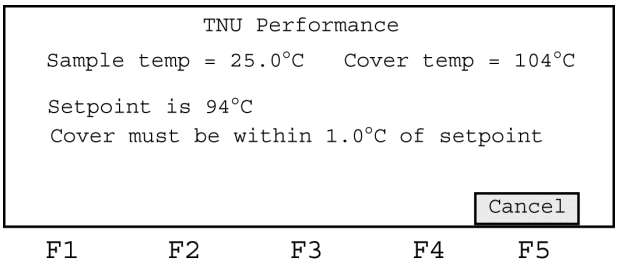


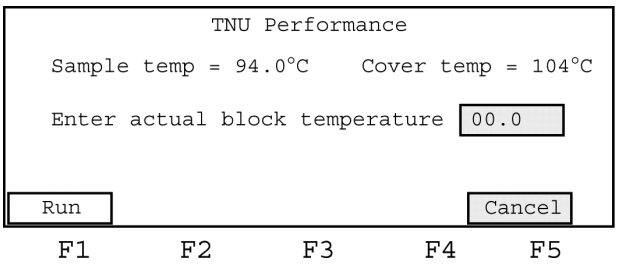
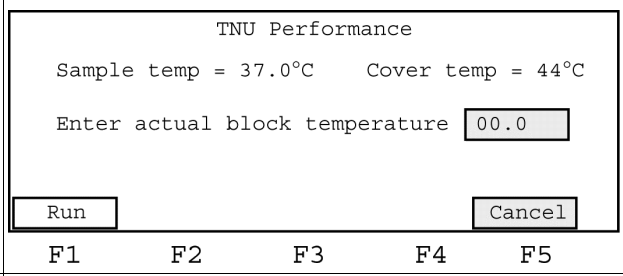
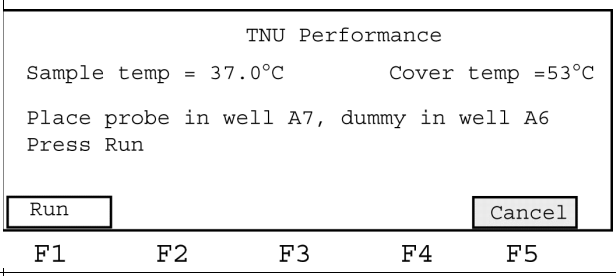
Figure 2-6 Location of wells used in Temperature Non-Uniformity test

Procedure

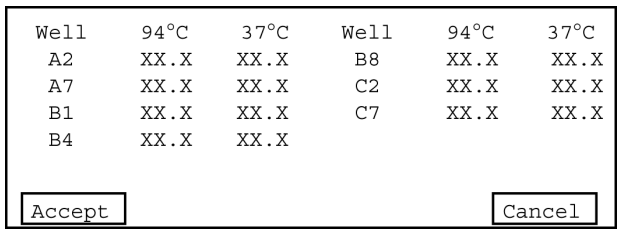
To run the Temperature Non-Uniformity Test:

Step	Action
1	From the Diagnostics screen, press F3-TmpVer. This displays the Temperature Verification screen. 
2	Press F2-TNU to display the TNU Setup screen.  This screen instructs you to set up the RTD probe assembly. **Note** Both the probe and the dummy probe are moved to seven different locations in the block during the Temperature Non-Uniformity test.
3	After installing the probe assembly, slide the heated cover forward and pull the lever down.
4	Press F1-Run to initiate the Temperature Non-Uniformity Test. This displays the TNU Setpoint Values screen. 

To run the Temperature Non-Uniformity Test:

Step	Action
5	When the temperature stabilizes at 94 °C and the counter decrements to zero, read the digital thermometer. 
6	Use the numeric keys to type the temperature displayed on the digital thermometer in the highlighted Enter actual block temperature field.
7	Press Enter. The sample block and heated cover now approach 37 °C. 
8	Read the digital thermometer.
9	Use the numeric keys to type the temperature displayed on the digital thermometer in the highlighted Enter actual block temperature field.
10	Press Enter.
11	Slide the heated cover back and move the probe assembly to wells A7 and A6, with the dummy probe in A6, as indicated by the screen shown below. 
12	Repeat steps 5 through 10 on wells A7, B1, B4, B8, C2, and C7. Make sure you place the measuring cone of the probe assembly into these wells and the dummy probe into adjacent wells.

To run the Temperature Non-Uniformity Test:

Step	Action
13	When the GeneAmp 2400 completes the Temperature Non-Uniformity Test, it displays the actual values gathered during testing. 
14	Press F1-Accept or F5-Cancel.
15	Turn off the digital thermometer, remove the probe assembly from the sample block and clean the oil from the sample block.

Temperature Non-Uniformity Specifications

The maximum temperature measured in any well at the 94 °C setpoint minus the minimum temperature measured in any well at the 94 °C setpoint must be equal to or less than 1.0 °C. This can be expressed as:

Max Temp 94 °C – Min Temp 94 °C < 1.0 °C

The maximum temperature measured in any well at the 37 °C setpoint minus the minimum temperature measured in any well at the 37 °C setpoint must be equal to or less than 1.0 °C. This can be expressed as:

Max Temp 37 °C – Min Temp 37 °C < 1.0 °C

Temperature Verification Tests for the 9600 3

Temperature Verification Tests

Types of Tests

- Calibration Verification tests the sample block against temperature accuracy specifications.
- Temperature Non-Uniformity tests the temperature uniformity in the sample block.

System Contents

Equipment Required

The Temperature Verification System is required to perform these tests. The Temperature Verification System should includethe following:

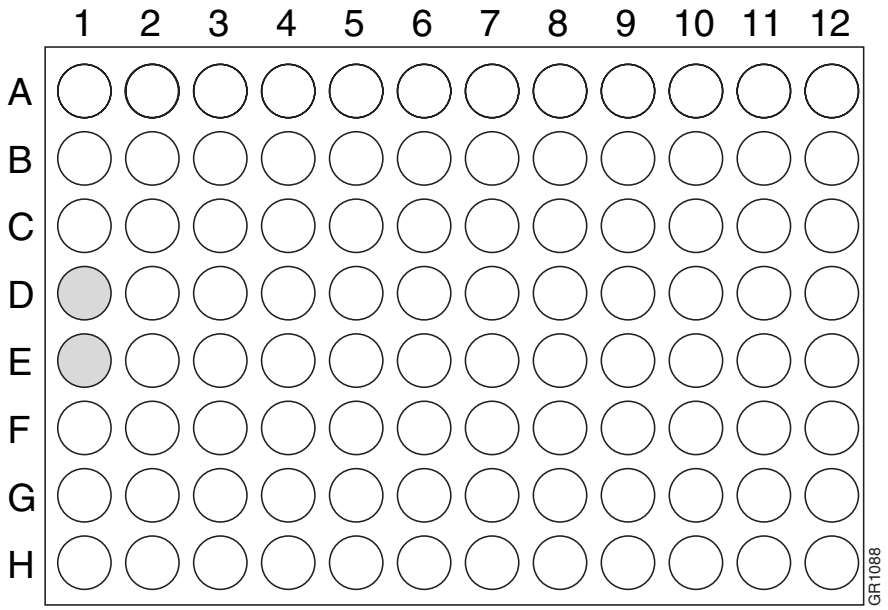
- Digital thermometer with 9V battery installed
- RTD probe
- Cotton swabs
- Light mineral oil
- 2400 probe tray

IMPORTANT Refer to the instructions included with your Temperature Verification System for a detailed description of digital thermometer operation.

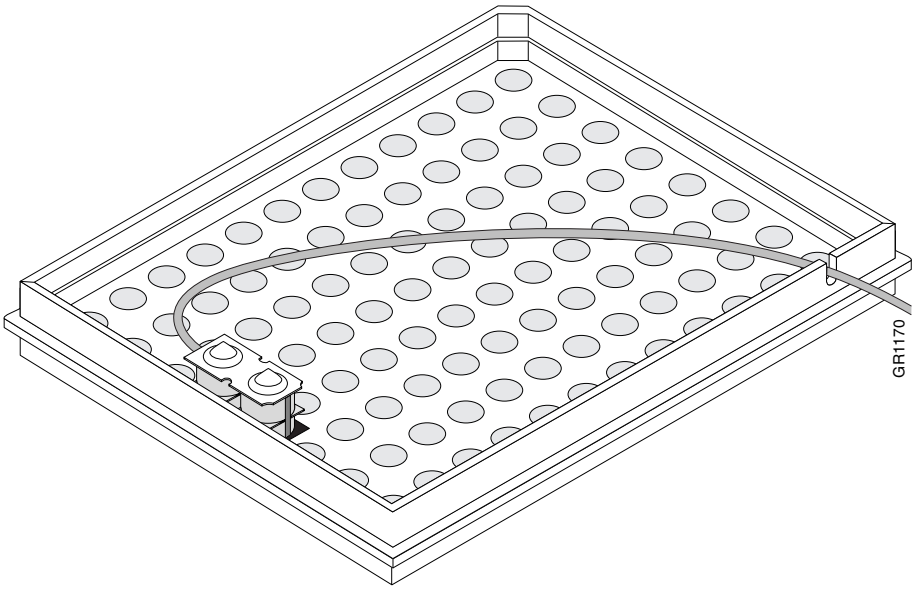
Preparation of the RTD Probe Assembly

! WARNING ! BURN HAZARD! The sample block is hot to the touch and can cause burns.

To prepare the RTD Probe Assembly for the Calibration Verification Test:

Step	Action
1	If the the sample block heated cover is in the forward position, turn the knob completely counterclockwise, then slide the cover back.
2	Coat wells D1 and E1 with mineral oil using a cotton swab. The illustration below shows the location of the wells. 
3	Place the probe tray on the sample block so that the probe tray notch faces the front of the instrument. **Note** The RTD probe assembly consists of two cones, one of which measures the temperature of the sample well. The wire is attached to the cone that does not measure the temperature of the sample well; this cone is a dummy probe.

To prepare the RTD Probe Assembly for the Calibration Verification Test: *(continued)*

Step	Action
4	Place the probe assembly into wells D1 and E1 so that the dummy probe sits in D1. Carefully thread the probe wire through the notch in the probe tray. Make sure the probe is connected to the digital thermometer. 
5	Slide the heated cover forward, then turn the cover knob clockwise until the white mark on the knob is aligned with the white mark on the cover.

The Temperature Calibration Verification Test Procedure

To perform the first phase of the calibration test:

Step	Action
1	Turn on the digital thermometer by moving the ON–OFF/RANGE switch to the 200 position.
2	Turn on the GeneAmp PCR System 9600. The main menu appears:
3	Press the OPTION key three times to move the cursor to UTIL, then press ENTER. The utilities menu appears:
4	Press the OPTION key twice to move the cursor to DIAG, then press ENTER. The following display appears:

5	Run the Verify Calibration Diagnostic Test (Test #5) by pressing 5 then ENTER.
	
	Note To ensure maximum accuracy, the temperatures of the heated cover and the sample block are the same in this test. This prevents the heated cover from affecting the accuracy of the RTD probe.
	
	The temperature of the sample block and heated cover will go to 40 °C, and the following display will appear:
	
	`Going to 40°C...`
	`Cvr= xxC  Blk = xx.xC`
	
	This display shows the current temperature of the block cover (Cvr= xxC) and sample block (Blk = xx.xC).
	
	When the temperature of the block cover is within ten degrees of the sample block temperature, the following display appears:
	
	`Wait 3 minutes`
	`Time=MM:SS Blk=95.0C`
	
	This display shows the current sample block temperature (“Blk=40.0C") and a clock, which counts up from zero in minutes and seconds (“Time=MM:SS").
	
	When the clock reaches three minutes, the following display appears:
	
	`Record Temperature`
	`Time=MM:SS Blk=95.0C`

6	Measure the temperature of well E1 using the digital thermometer. Record this temperature as T(40). See Figure 5 on the next page.

To perform the first phase of the calibration test: *(continued)*

7	Press ENTER.
	
	The temperature of the sample block and heated cover will go to 95 °C, and the following display will appear:
	
	`Going to 95°C...`
	`Cvr= xxC  Blk = xx.xC`
	
	This display shows the current temperature of the block cover (Cvr= xxC) and sample block (Blk = xx.xC).
	
	When the temperature of the block cover is within ten degrees of the sample block temperature, the following display appears:
	
	`Wait 3 minutes`
	`Time=MM:SS Blk=95.0C`
	
	This displays shows the current sample block temperature (“Blk=95.0C") and a clock, which counts up from zero in minutes and seconds (“Time=MM:SS").
	
	When the clock reaches three minutes, the following display appears:
	
	`Record Temperature`
	`Time=MM:SS Blk=95.0C`

8	Measure the temperature of well E1 using the digital thermometer. Record this temperature as T(95).

To perform the second phase of the calibration test:

Step	Action
1	Turn on the digital thermometer by moving the ON–OFF/RANGE switch to the 200 position.
2	Turn on the GeneAmp PCR System 9600. The main menu appears:
3	Press the OPTION key three times to move the cursor to UTIL, then press ENTER. The utilities menu appears:
4	Press the OPTION key twice to move the cursor to DIAG, then press ENTER. The following display appears:

5	Run the Verify Calibration Diagnostic Test (Test #5) by pressing 5 then ENTER.
	
	Note To ensure maximum accuracy, the temperatures of the heated cover and the sample block are the same in this test. This prevents the heated cover from affecting the accuracy of the RTD probe.
	
	The temperature of the sample block and heated cover will go to 40 °C, and the following display will appear:
	
	`Going to 40°C...`
	`Cvr= xxC  Blk = xx.xC`
	
	This display shows the current temperature of the block cover (Cvr= xxC) and sample block (Blk = xx.xC).
	
	When the temperature of the block cover is within ten degrees of the sample block temperature, the following display appears:
	
	`Wait 3 minutes`
	`Time=MM:SS Blk=95.0C`
	
	This display shows the current sample block temperature (“Blk=40.0C") and a clock, which counts up from zero in minutes and seconds (“Time=MM:SS").
	
	When the clock reaches three minutes, the following display appears:
	
	`Record Temperature`
	`Time=MM:SS Blk=95.0C`

6	Measure the temperature of well E1 using the digital thermometer. Record this temperature as T(40). See Figure 5 on the next page.

To perform the second phase of the calibration test: *(continued)*

7	Press ENTER.
	
	The temperature of the sample block and heated cover will go to 95 °C, and the following display will appear:
	
	`Going to 95°C...`
	`Cvr= xxC  Blk = xx.xC`
	
	This display shows the current temperature of the block cover (Cvr= xxC) and sample block (Blk = xx.xC).
	
	When the temperature of the block cover is within ten degrees of the sample block temperature, the following display appears:
	
	`Wait 3 minutes`
	`Time=MM:SS Blk=95.0C`
	
	This displays shows the current sample block temperature ("Blk=95.0C") and a clock, which counts up from zero in minutes and seconds ("Time=MM:SS").
	
	When the clock reaches three minutes, the following display appears:
	
	`Record Temperature`
	`Time=MM:SS Blk=95.0C`

8	Measure the temperature of well E1 using the digital thermometer. Record this temperature as T(95).

Temperature Measurements

Figure 3-1 illustrates temperatures during the Calibration Verifciation Test.

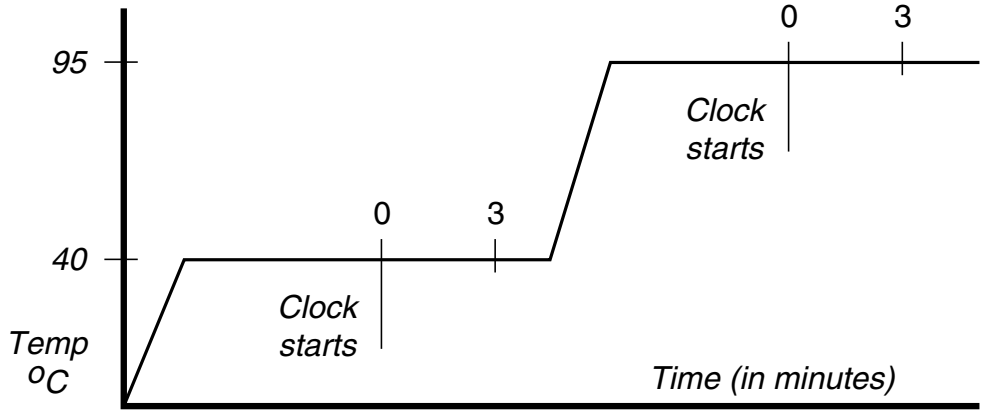


Figure 3-1 Temperature measurements in the temperature calibration verification test

IMPORTANT To exit the test at any time, press the STOP key. This will return you to the "Review History File" display. Press 5 and ENTER to return to the Verify Calibration Diagnostic test.

Calculating Test Results

Calculating the Test Results

Perform the following procedure to calculate the results of the test. You will need to refer to the calibration label in your Users Manual for the High and Low Offset values.

Note If you have more than one GeneAmp PCR System 9600 in your laboratory, make sure that the serial number on the calibration label matches the serial number on the instrument you are testing.

Calculating the Average Block Temperature at the 95 °C Hold

Use the following formula to calculate the average block temperature at 95 °C = T(95) - High Offset hold:

Block Average at 95 °C = T(95) - High Offset

If the block average is more than 0.75 °C above or below 95 °C, your GeneAmp PCR System 9600 must be recalibrated.

For example:

If the measured temperature of well E1 was 95.2 °C, and the High Offset printed on your calibration label is -0.1, you would make the following calculation:

Block Average at 95 °C = 95.2 - (-0.1)

= 95.3 °C

In this example, since 95.3 °C does not differ by +/-0.75 °C from your programmed target temperature, your instrument would not need to be recalibrated.

Note The offset is the number of degrees Celsius that the temperature of well E1 differed from the average temperature of the block when the instrument was calibrated at the factory.

Calculating the Average Block Temperature at the 40 °C Hold

Use the following formula to calculate the average block temperature at the 40 °C hold:

Block Average at 40 °C = T(40) - Low Offset

If the block average is more than 0.75 °C above or below 40 °C, your GeneAmp PCR System 9600 must be recalibrated.

For example:

If the measured temperature of well E1 was 39.9 °C, and the Low Offset printed on your calibration label is +0.1, you would make the following calculation:

Block Average at 40C = 39.9 - (+0.1)

= 39.8 °C

In this example, since 39.8 °C does not differ by more than +/-0.75 °C of your programmed target temperature, your instrument would not need to be recalibrated.

Completing the Calibration Verification Test Procedure

To complete the test:

Step	Action
1	Remove the probe assembly from the sample block and move the digital thermometer ON–OFF/RANGE switch to the OFF position.
2	Clean the oil from wells D1 and E1 using cotton swabs.

IMPORTANT If your instrument needs to be recalibrated, contact an Applied Biosystems Service Representative

The Temperature Uniformity Test

Preparation

Note Use this procedure to test the temperature uniformity of the sample block in the GeneAmp PCR System 9600.

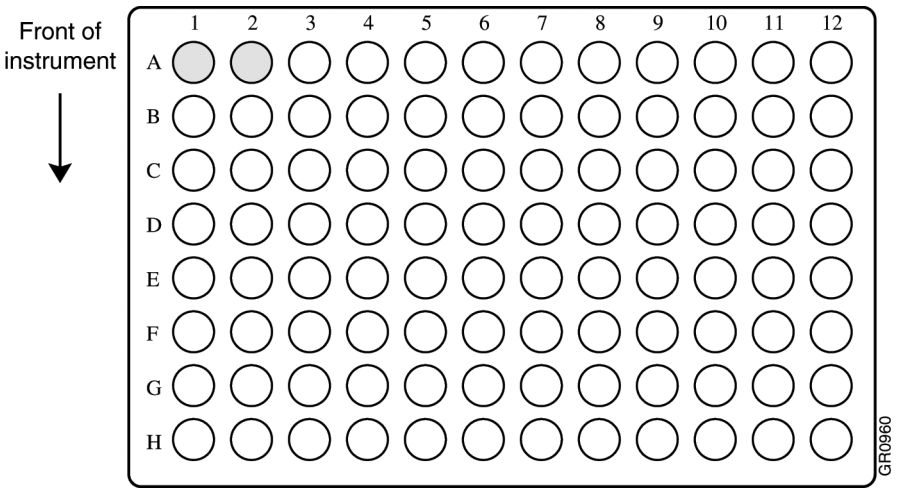
Equipment Required

- Temperature Verification System

See page 3-1 for items that are part of ths system.

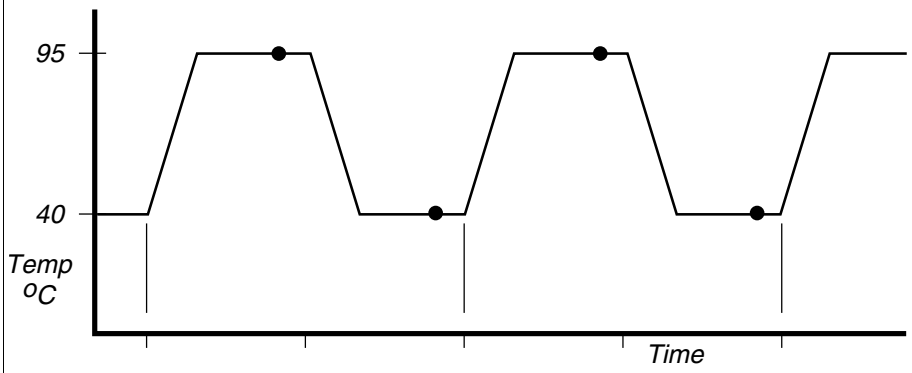
Preparation for the Temperature Uniformity Test

To set-up the thermal cycler for the TNU test:

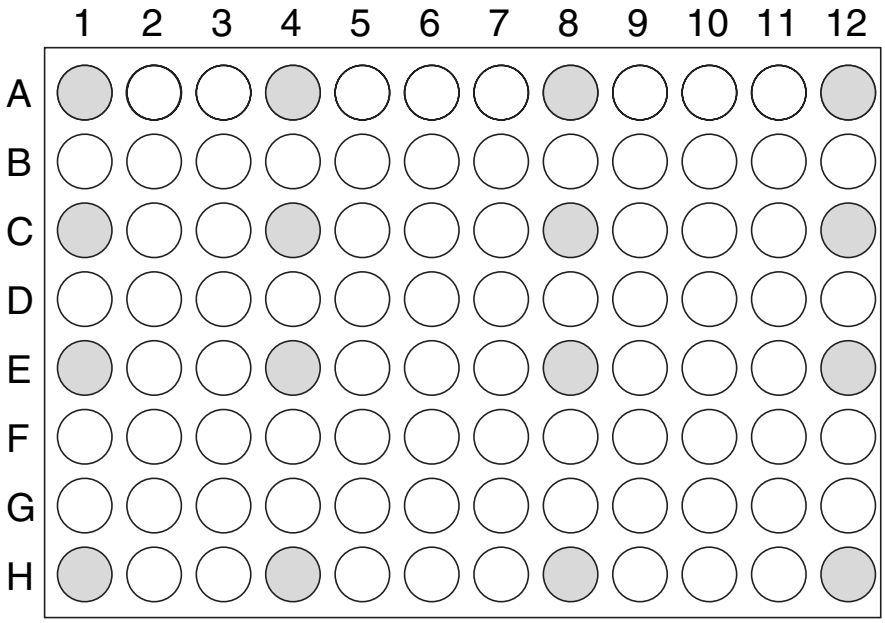
Step	Action
1	If the the sample block heated cover is in the forward position, turn the cover knob completely counterclockwise, then slide the cover back.
2	Coat all the wells in sample block rows A, C, E, and H with mineral oil using a cotton swab.
3	Place the probe tray on the sample block with the notch facing the front of the instrument. **Note** The RTD probe assembly consists of two cones, one of which measures the temperature of the sample well. The wire is attached to the cone that does not measure the temperature of the sample well; this cone is a dummy probe.
4	Place the probe assembly into wells A1 and A2 so that the dummy probe sits in A2. Carefully thread the probe wire through the notch in the probe tray. Make sure the probe is connected to the digital thermometer. 
5	Slide the heated cover forward and turn the cover knob clockwise until the white mark on the knob and the white mark on the cover are aligned.
6	Turn on the digital thermometer by moving the ON–OFF/RANGE switch to the 200 position.

Performing the Temperature Non-Uniformity Test

To perform the TNU test:

1	Turn on the GeneAmp PCR System 9600 and create a two–temperature CYCL program with the following parameters: ♦ Setpoint #1 Temperature = 95 °C ♦ Hold Time = 2:00 minutes ♦ Ramp Time = 0:00 minutes ♦ Setpoint #2 Temperature = 40 °C ♦ Hold Time = 2:00 minutes ♦ Ramp Time = 0:00 minutes ♦ Cycles = 99 **Note** Refer to your Users Manual for detailed instructions on how to set up and run a CYCL program.
2	On the third cycle, measure the temperature of well A1 90 seconds into Setpoint #1 (95 °C setpoint temperature) using the digital thermometer (see Figure 7). The time remaining clock on the run–time display will read “0:30" (30 seconds). Record this temperature.
3	Still on the third cycle, measure the temperature of well A1 90 seconds into Setpoint #2 (40 °C setpoint temperature) using the digital thermometer. The time remaining clock on the run–time display will read “0:30" (30 seconds). Record this temperature. 
4	After you measure the second temperature of well A1, turn the cover knob completely counterclockwise, then slide the heated cover back.
5	Move the probe assembly to wells A4 and A5, placing the dummy probe in A5.
6	Slide the heated cover forward, then turn the cover knob clockwise until the white mark on the knob and the white mark on the cover are aligned.

To perform the TNU test: *(continued)*

7	Repeat the measurements on wells A4, A8, A12, C1, C4, C8, C12, E1, E4, E8, E12, H1, H4, H8, and H12. Make sure you place the measuring cone of the probe assembly into these wells and the dummy probe into adjacent wells (see Figure 8). **Note** The temperature display on the digital thermometer may not match the temperature display on the System 9600. This is due to the effect of the heated sample block cover on the RTD probe. If you suspect any temperature calibration problems, perform the temperature calibration verification test described on pages 4 through 9 of these instructions. 

Post-Test and Clean-Up Procedures

Performing TNU post-test procedures and clean-up:

1	After you have completed all measurements, remove the probe assembly from the sample block and turn off the digital thermometer.
2	Clean the oil from the sample block using cotton swabs.

Test Results

Calculate the test results as follows:

- For the 16 Setpoint #1 measurements (95 °C hold), subtract the lowest measured temperature from the highest measured temperature.
- For the 16 Setpoint #2 measurements (40 °C hold), subtract the lowest measured temperature from the highest measured temperature.
  - If either result is more than 1 °C, your instrument must be serviced by an Applied Biosystems Service Representative.

Temperature Verification Tests for the 9700 4

Temperature Verification Tests

Types of Tests

- Calibration Verification tests the sample block against temperature accuracy specifications.
- Temperature Non-Uniformity tests the temperature uniformity in the sample block.

System Contents

Equipment Required

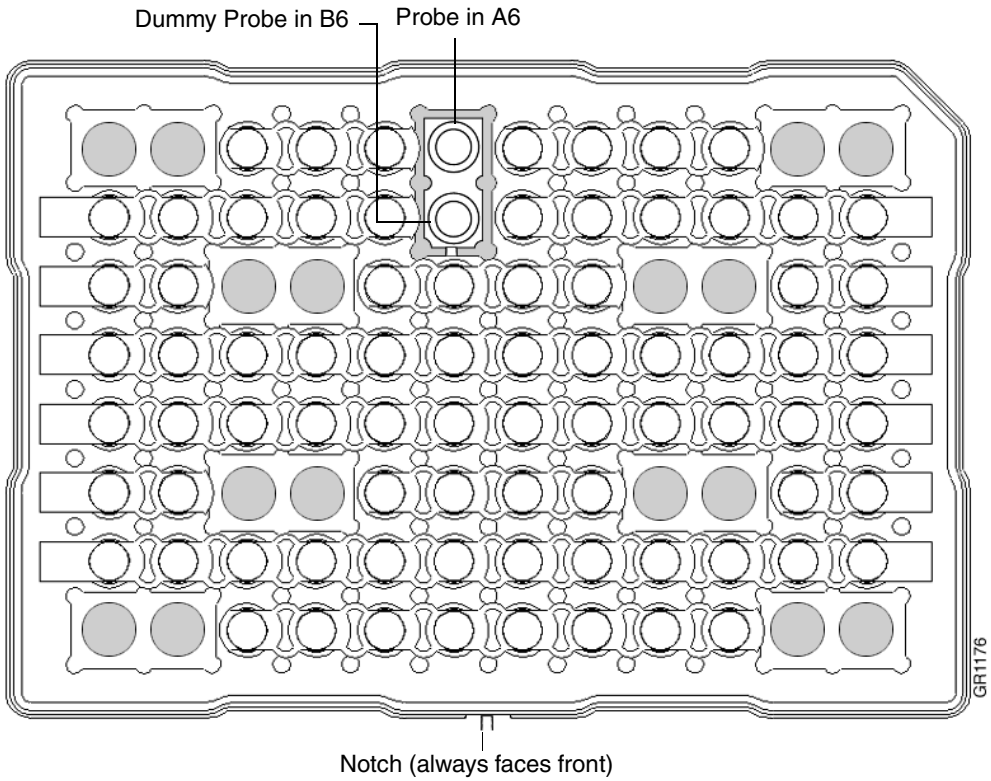
The Temperature Verification System is required to perform these tests. The Temperature Verification System should include the following:

- Digital thermometer with 9V battery installed
- RTD probe
- Cotton swabs
- Light mineral oil
- 9700 probe tray

IMPORTANT Refer to the instructions included with your Temperature Verification System for a detailed description of digital thermometer operation.

The RTD Probe Assembly

Schematic This schematic shows the probe tray from the top view. Note the locations of the probe and dummy, and also the notch.



The Tray Assembly The 9700 temperature verification system is equipped with a tray assembly that ensures accurate measurement of temperature during the calibration verification and TNU procedures. The tray consists of a 96 well plate with caps. There are nine locations where no caps are present. One location is for the calibration verification test and the other eight locations are for TNU testing.

IMPORTANT The 9700 tray must be used for accurate calibration and TNU measurements

Performing The Calibration Verification Test

Overview

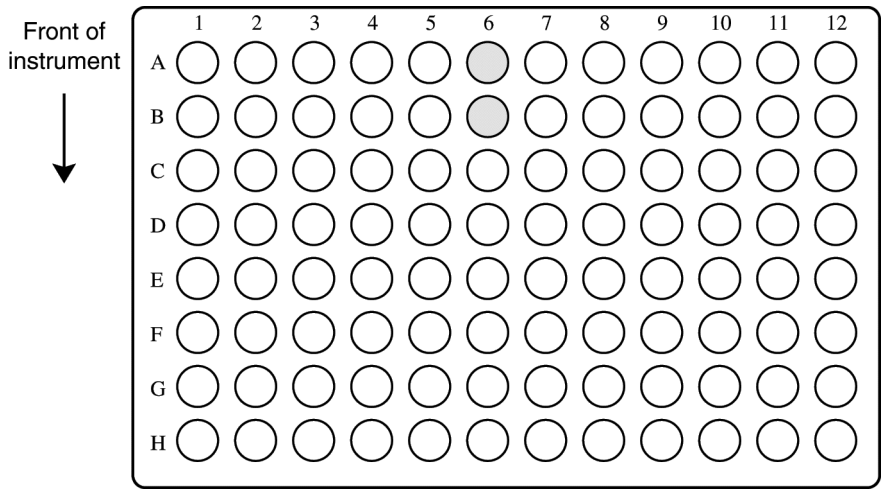
The RTD probe (P/N N8051210) and the digital thermometer (included in Temperature Verification Kit, P/N N8010435) are used to take temperature readings of one sample well at two different setpoint temperatures:

IMPORTANT A properly calibrated 9700 probe and meter must be used in determining calibration verification. The 9600 probe (now obsolete) is not compatible with the 9700, and will give inaccurate temperature readings. The 9700 Temperature Verification System probe is compatible with all 0.2 ml GeneAmp PCR thermal cycler models (2400 and 9600.)

- 85 °C
- 45 °C

Preparation

To prepare the RTD Probe assembly:

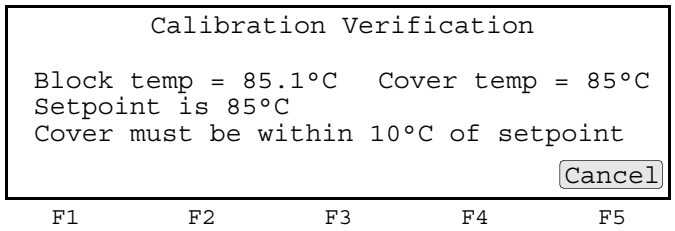
Step	Action
1	If the System 9700 heated cover is in the forward position, lift the lever, then slide the cover back.
2	Using a cotton swab, coat wells A6 and B6 with mineral oil. 
3	Place the probe assembly into wells A6 and B6 so that the dummy probe sits in B6.
4	Place the probe tray over the probe assembly with the notch in the tray facing the front of the instrument.
5	Thread the probe wire through the notch and carefully seat the probe tray over the probe and wire being sure not to crimp the wire in any wells.
6	Connect the probe to the digital thermometer.
7	Slide the heated cover forward and pull the lever down.
8	Turn on the digital thermometer by moving the ON-Off Range switch to the 200 position.

Description of Beginning Test Functions

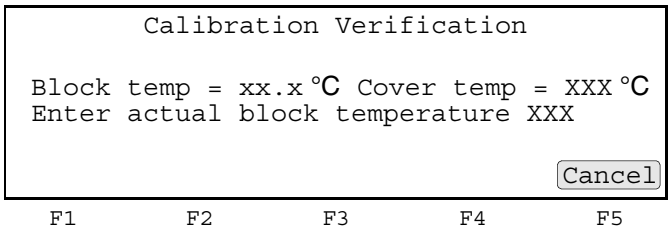
When the Calibration Verification Test begins, the block and heated cover each move toward the first setpoint of 85 °C. (The block temperature will increase and the cover temperature decrease.) After the heated cover reaches 85 °C and the block reaches the temperature of the heated cover ± 10 °C, the instrument pauses three minutes to stabilize the temperature.

! WARNING ! BURN HAZARD! The sample block is hot to the touch and can cause burns.

The Stabilizing at Setpoint screen will be displayed.

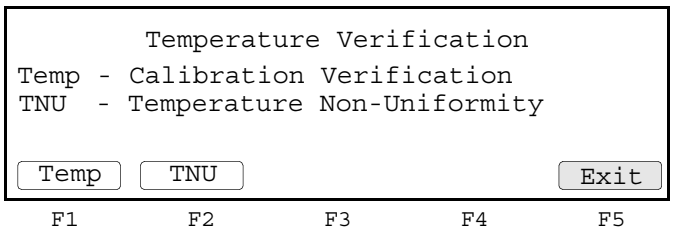
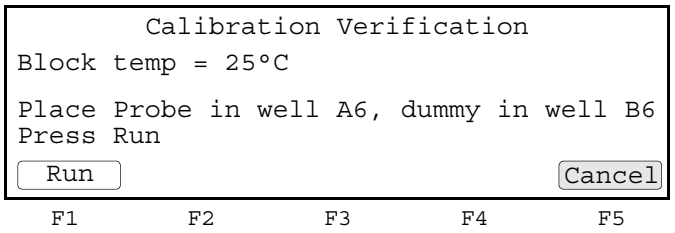
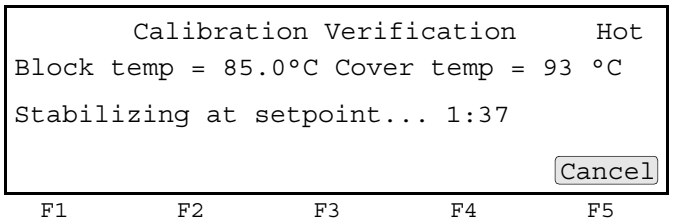


When three minutes have elapsed, the Setpoint screen will be displayed. The digital thermometer reading of the sample well temperature is entered into the Enter Actual Temp field. This process is repeated at each of the two setpoints.

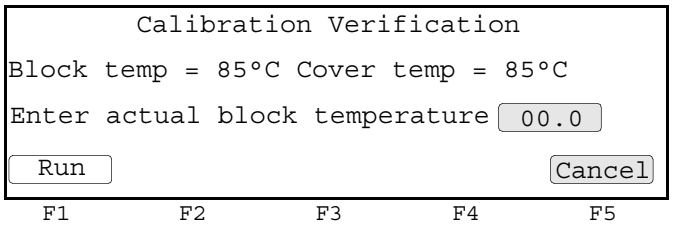
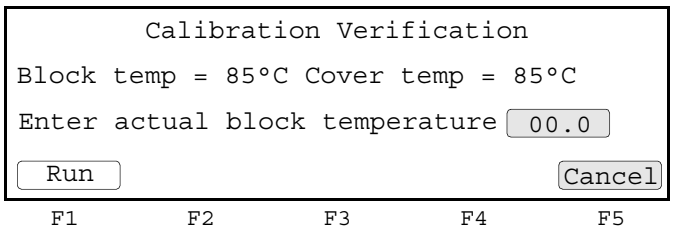
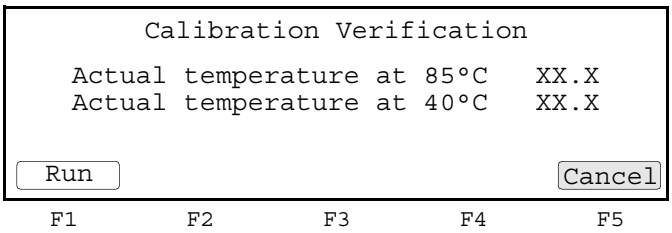


Procedure

To run the Calibration Verification Test:

Step	Action
1	From the Diagnostics screen, press TmpVer. This displays the Temperature Verification screen. 
2	Press Temp. This displays the Calibration Verification Setup screen.  **Note** This screen instructs you to set up the RTD probe assembly.
3	After installing the probe assembly, slide the heated cover forward and pull the lever down.
4	Press Run to initiate the Calibration Verification Test. This automatically verifies the temperature of the sample block, and displays the Setpoint Values screen.  **Note** To exit the test at any time, press Cancel

To run the Calibration Verification Test: *(continued)*

Step	Action
5	When the temperature stabilizes at 85 °C and the counter decrements to zero, read the digital thermometer. 
6	Use the numeric keys to type the value displayed on the digital thermometer into the highlighted Enter actual block temperature field.
7	Press Enter. This will initiate the second segment of the temperature verification process. The heated cover and sample block now approach the second setpoint temperature, 45 °C. 
8	When the temperature stabilizes at 45 °C and the counter decrements to zero, read the digital thermometer.
9	Use the numeric keys and type the value displayed on the digital thermometer in the highlighted Enter actual block temperature field.
10	Press Enter. This is the last temperature entry. The GeneAmp 9700 displays the digital thermometer measurements in the right hand column of the Calibration Verification screen. 
11	Press Accept to accept the verification values or Cancel to reject the values.

Post-Test and Clean-Up Procedures

Performing TNU post-test procedures and clean-up:

1	After you have completed all measurements, remove the probe assembly from the sample block and turn off the digital thermometer.
2	Clean the oil from the sample block using cotton swabs.

Calibration Verification Specification

The calibration verification procedure is fully automated. After entering the calibration verification values, the instrument will display a pass or fail result.

The Temperature Non-Uniformity Test

Overview

The temperature Non-Uniformity test uses the RTD probe assembly and a digital thermometer to test the temperature uniformity of eight different wells in the sample block.

IMPORTANT Perform this test after the instrument has been off for at least two hours. The first pass of the TNU test will be performed with the heated cover at 37 °C. If the heated cover is hot when the test is started it will take a considerable amount of time for the heated cover to cool off and the test to start.

The first eight TNU measurements will be made at 37 °C with the heated cover at or near 37 °C. After completion of the 37° measurements the heated cover will reach and maintain a setpoint of 105°C. Once the setpoint is reached, the instrument will instruct you to measure TNU for the eight wells at 94 °C.

IMPORTANT The TNU procedure outlined here can only be performed on instruments with firmware version 1.63 or greater. If an instrument has an older version of firmware, then the 9700 must be upgraded for the TNU procedure to work properly (to upgrade the firmware on a 9700 see page 3-18 of the User's Manual or 2-13 of the Service Manual.)

Probe	Dummy
A1	A2
A12	A11
C4	C3
C9	C10
F4	F3
F9	F10
H1	H2
H12	H11

Setpoint Screen After starting the TNU test, the heated cover will heat to 37 °C and the block will stabilize at the 37 °C setpoint.

```
            TNU Performance
Sample temp = 25.9°C Cover temp = 31°C
Cover Setpoint is 35°C
Cover must be within 1°C of Setpoint
                                  Cancel
  F1        F2        F3        F4        F5
```

Once the heated cover has stabilized and the block has reached the 37 °C setpoint, the instrument will request placement of the probe in wells A1 and A2. After starting the test the block will begin cycling up to 94°C and then immediately back to the setpoint for the measurement. The instrument pauses 30 seconds to allow the temperature to stabilize. A digital thermometer reading is taken and entered using the numeric keypad.

This process is repeated for the remaining seven sample block wells shown in Figure 4-1:

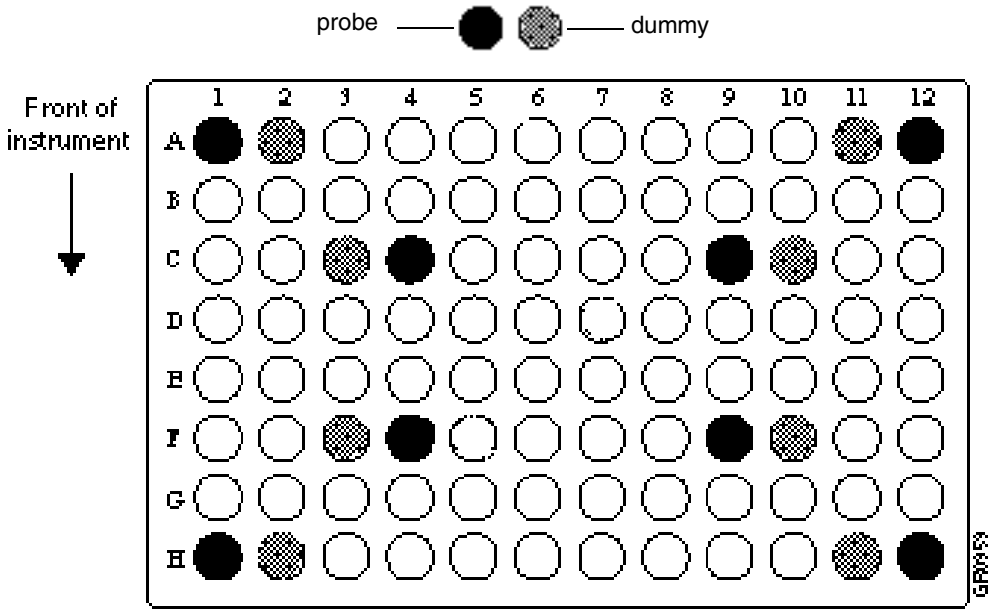
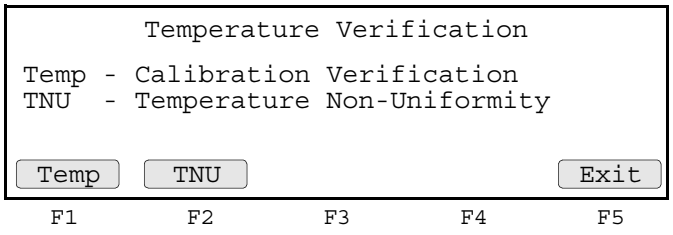
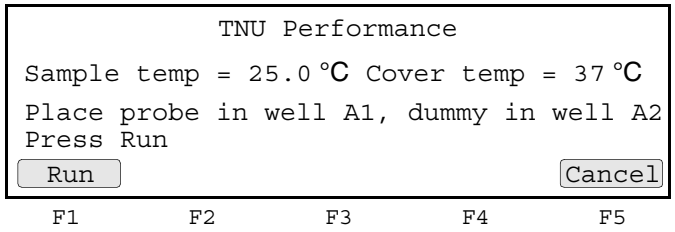
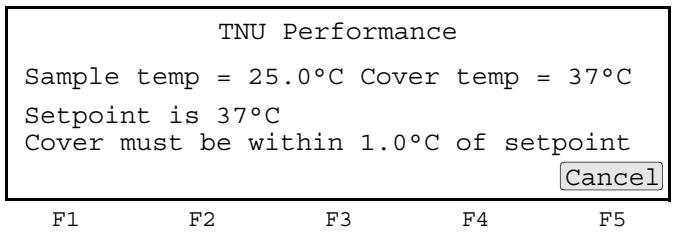


Figure 4-1 9700 Sample Block Wells for TNU Measurement

Procedure

To run the Temperature Non-Uniformity Test (TNU):

Step	Action
1	From the Diagnostics screen, press TmpVer. This displays the Temperature Verification screen. 
2	Press TNU to display the TNU Setup screen  This screen instructs you to set up the RTD probe assembly. For a detailed procedure, see page 4-4. **Note** Both the probe and the dummy probe are moved to 8 different locations in the block during the Temperature Non-Uniformity test.
3	After installing the probe assembly, slide the heated cover forward and pull the lever down.
4	Press Run to initiate the Temperature Non-Uniformity Test. This displays the TNU Setpoint Values screen. 

To run the Temperature Non-Uniformity Test (TNU): *(continued)*

Step	Action
5	When the temperature stabilizes at 37 °C and the counter decrements to zero, read the digital thermometer. TNU Performance Sample temp = 37°C Cover temp = 37°C Enter actual block temperature 00.0 Run Cancel F1 F2 F3 F4 F5
6	Use the numeric keys to type the temperature displayed on the digital thermometer in the highlighted Enter actual block temperature field.
7	Press Enter. Place the probe in well A12. Press Run. The sample block will now approach 94 °C and return to the 37 °C setpoint. TNU Performance Sample temp = 37.0°C Cover temp = 38°C Place probe in well A12,dummy in well A11 Press Run Run Cancel F1 F2 F3 F4 F5
8	After the instrument counts down 30 seconds, read the digital thermometer.
9	Use the numeric keys to type the temperature displayed on the digital thermometer in the highlighted Enter actual block temperature field. Press Enter.
10	Repeat steps 5 through 10 on wells C4, C9,F4, F9, H1, and H12. Make sure you place the measuring cone of the probe assembly into these wells and the dummy probe into adjacent wells as shown in Figure 4-1 on page 4-10. **Note** When measuring wells in row H leave a loop of wire over the sample block before securing the tray over the probe assembly.
11	After all eight wells have been measured at 37°C the heated cover will warm up to 105°C and the test will repeat measuring TNU at 94°C
12	After the heated cover reaches 105°C you will be instructed to move the probe to well A1. Repeat steps 5 through 10 for the 94°C setpoint for all eight wells. **Note** Before being instructed to move the probe to another well the sample block will cool to 37°C.
13	When the GeneAmp 9700 completes the temperature Non-Uniformity test, it displays the actual values gathered during testing.
14	Press Accept or Cancel.
15	Turn off the digital thermometer, remove the probe and tray assembly from the sample block and clean the oil from the sample block.

Temperature Non-Uniformity Specifications

The maximum temperature measured in any well at the 94 °C setpoint minus the minimum temperature measured in any well at the 94 °C setpoint must be equal to or less than 1.0 °C. This can be expressed as:

Max Temp94 °C – Min Temp94 °C < 1.0 °C

The maximum temperature measured in any well at the 37 °C setpoint minus the minimum temperature measured in any well at the 37 °C setpoint must be equal to or less than 1.0 °C. This can be expressed as:

Max Temp37 °C – Min Temp37 °C < 1.0 °C

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Printed in the USA, 09/2001
Part Number 4303200 Rev. B

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