User Notice
Users are advised that the use of this SpotArray instrument for certain applications in fabricating microarrays may require a license from one or more third parties that have patents in this area. At the time of publication of this notice, PerkinElmer Life Sciences is aware of the following third parties that have patents in this area:

Affymetrix, Inc.
Incyte Genomics
Oxford Gene Technology, Ltd
Hyseq, Inc.

There may be other third parties that have patent rights in this area. PerkinElmer is not encouraging or suggesting that the users of this equipment employ this equipment in a way that would infringe on the patent rights of any third parties. PerkinElmer suggests that users consult their own legal advisors for counsel on non-infringing or licensed uses of this machine in fabricating microarrays.
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Preface Summary

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Conventions Used in this Manual

The SpotArray uses the Windows® 2000 Operating System. We assume that the operator is acquainted with the general use of the operating system, and therefore provide only an overview of using Windows programs.

The names of the buttons and their locations are bold. The name of windows and dialog boxes are in italics. For example:

- In the SpotArray Main Window, click **Start** to start a printing procedure.
All user defined file names, for example, array content files, must be Windows compatible (i.e. they cannot contain a slash, a backslash, a dot or any other Windows reserved character).
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1.1 Overview

Welcome to the SpotArray™ family of microarray printing systems from PerkinElmer Life Sciences. There are two SpotArray printing systems:

• SpotArray 24 (24 slides), designed for research use
• SpotArray 72 (72 slides), designed for higher throughput applications

These systems are easy-to-use robotic instruments for creating microarrays. Simply load your plates and substrates, choose a printing protocol and start — the SpotArray does the rest.

Figure 1-1 shows the SpotArray 24 system; Figure 1-2 shows the SpotArray 72 system.

Figure 1–1  The SpotArray™ 24 Microarray Printing System (Instrument Module and Monitor)
1.1.1 The SpotArray System

The SpotArray system includes an instrument module, a utility module, a reservoir module, an optional humidity module, and optional cart, described in this section.

1.1.1.1 Instrument Module

The instrument module includes: substrate holder for up to 20 substrates and 4 blotting slides for the SpotArray 24, up to 68 substrates and 4 blotting slides for the SpotArray 72; plate holder for up to 4 plates; printhead for up to 48 pins; pin washer/dryer; humidity and temperature monitoring hardware; central processing unit (CPU) for running the SpotArray software. Options include humidity control, a barcode reader, lid lifting and pin lifting.

The monitor, keyboard and mouse connect to the instrument for using the SpotArray software. The SpotArray software user interface displays on the monitor and lets you start a printing protocol, or use other commands.

Figure 1–2  The SpotArray™ 72 Microarray Printing System (Instrument Module and Monitor)
An optional cart, shown in Figure 1-3 with the SpotArray 72, provides a bench-top for the instrument and monitor, and an enclosure for the utility module and humidity module.

![Optional Cart for the SpotArray Microarray Printing System](image)

**Figure 1–3** Optional Cart for the SpotArray Microarray Printing System

### 1.1.1.2 Utility Module

The **utility module** holds the power supply, pin-drying and humidity pumps, and air and water filters.

![Utility Module with On/Off switch](image)

**Figure 1–4** The SpotArray Utility Module (front)
1.1.1.3 Reservoir and Humidity Modules

The reservoir module holds the wash water supply and waste water.

The humidity control module (optional) holds the water supply for humidity generation.

![Image of reservoir and humidity modules]

*Figure 1–5  The Reservoir Module and Humidity Module*
1.1.2 The User Interface

The SpotArray software runs on a computer processor (CPU) inside the instrument module. The monitor, keyboard and mouse connect directly to the instrument module, as shown in Figure 1-6, without the need for a separate workstation (personal computer).

The User Interface, the part of the software that you see displayed on the monitor screen, runs under the Windows 2000 operating system. The user interface lets you enter information or commands, and receive status information.

![The SpotArray User Interface](image)

*Figure 1–6 Using the SpotArray Software User Interface*

The SpotArray main window is described on the following page. To learn more about the SpotArray software, see Chapter 2, “Using the SpotArray Software.”

**Note:** It is also possible to connect a workstation (personal computer) to the instrument via an Ethernet/TCP/IP connection. In that case, the SpotArray user interface software is installed and used on the workstation.
1.1.3 The SpotArray Main Window

When you start the SpotArray program, the user interface displays the **Main Window** on the monitor screen, as shown in Figure 1-7 below. This is your starting point for sending commands, entering information, or receiving status information.

**Configure Buttons**
1) Click **Plates** to add plate information to your plate library.
2) Click **Plate Sets** to select plates from the library for your printing protocol.
3) Click **Printing Protocols** to create a new printing protocol or modify an existing protocol.

**Run Buttons**
1) Click **Start** to start printing your microarray.
2) Click **Pause** to stop a printing procedure temporarily.
3) Click **Cancel** to cancel the printing procedure.

**Sensor Indicators**
Display the status of hardware sensors.

**Click the Pin Washer button to get status details.**

![Figure 1–7 The SpotArray User Interface, Main Window](image-url)
1.2 Printing a Microarray

Start the SpotArray (turn on power on the utility module, and start the SpotArray software).

Define the Plates

This lets you create plates and a description of their contents for the plate library.

For more information, see “Defining Plates” on page 3-1.

Define the Plate Set for your experiment

This lets you create a plate set (a subset of plates from the plate library) for your experiment.

For more information, see “Defining Plate Sets” on page 3-9.

Select a Printing Protocol

A printing protocol specifies how the microarray is printed. Default protocols are provided for 96-well or 384-well plates.

For information on protocols, see “Creating a Protocol” on page 3-13.

Install or verify the Pins.
Load the Substrates and Plates.

Install or verify the number and location of pins required for your protocol; load plates and substrates.

For more information, see “Preparing for Printing” in Chapter 4.

Start Your Printing Procedure

Press the Start button and select a printing protocol.

For more information, see Chapter 2 “Using the SpotArray Software.”
1.2.1 Before Running a Printing Procedure

Before you run a printing procedure, verify that the installed pins, number of plates, and plate types are correct for your printing protocol, and make a quick check of the hoses to ensure they are connected and do not have kinks that might impede the flow of air or water. Also, make sure your wash water supply is nearly full and the waste tank is nearly empty. See Chapter 4, “Preparing for Printing,” for more information.

1.2.2 Creating an Array Content File

An Array Content File is an ordered list of spots in a microarray that details the location of each spot and the source, identity, and name of the gene used to create the spot. The file may contain selected parameters for the printing protocol, depending on the file format.

The location of spots on the microarray substrate are precisely controlled so that the downstream scanning and analysis process can “know” exactly where the spots are and identify them. This control is provided by the high-precision motion control of the printhead. During “downstream” processing — after the microarrays have been printed — the array content file is referenced for the identification of each spot. You may create an array content file automatically after a printing procedure runs, or at any time using the Create Array Content File button on the Main Window.

To create an array content file

1. On the SpotArray Main Window, click Create Array Content File. The Create an Array Content File dialog box opens.

2. Click the button under the Printing protocol label to select a protocol for which you want to create a file. After you select one of the available printing protocols from the list that opens, the button fills in with the name of the protocol you select (“Demo low density 96-well plate” in the above example).

3. Click the button under the Format of array content file label; the button expands to display supported file formats. Select a format that is compatible with your microarray scanning and/or quantitation software. The following file formats are supported:
• .gal (Gene Array List) - a .gal file is used at quantitation to define the quantitation template and to correlate fluorescence data to the name of the spotted sample.
• .txt (QuantArray text file) - QuantArray uses the file to correlate fluorescence data to the name of the spotted sample.

4. Enter a path and file name, and click OK. You may want to save this file to a network-accessible drive so that it can be accessed easily by other software applications. The file extension defaults to the type of file (.gal, or .txt). By default, the SpotArray creates QuantArray text files.

Note: Downstream applications that use the .gal or .txt file often use this filename to identify the individual microarrays in the batch; therefore, we suggest that you provide a filename that describes the printing batch.

See Appendix D for a description of file formats for the array content files.

1.3 Running a Printing Procedure

To run a printing procedure, the plate contents, a plate set, and a printing protocol must already be defined. See Chapter 3 for information.

Note: Never attempt to open the door when a printing procedure is underway; the door is locked. See Pausing or Cancelling a Printing Procedure on page 1-19.

To power up the instrument and workstation

1. Power up the instrument by pressing the On/Off switch on the front of the utility module.
2. Log in to Windows® 2000 if prompted. The instrument software moves the printhead into its “home” position and runs diagnostic self-tests if the door is closed. This takes approximately one minute.
3. On the Windows desktop on the monitor screen, double-click the SpotArray application icon to start the SpotArray user interface if it doesn’t start automatically.

The diagnostic test results display on the Diagnostics tab on the Main Window. If any diagnostic self-tests fail, the Start button becomes dimmed and unavailable; you cannot start the printing procedure until the failing condition has been fixed. For more information on sensors and diagnostics, see Appendix B in this guide.
When the instrument is ready, you can start a printing procedure. The LCD text display on the instrument displays a “Ready” message, and the Start button on the Main Window becomes available to select.

To start the procedure

1. In the SpotArray Main Window, click Start in the Run box.

![Start Button](image)

The Start a Printing Procedure dialog box opens.

![Printing Protocol Dialog](image)

2. Click the button under the Printing Protocol label to open the Select a Printing Protocol dialog box. The list in the window includes the SpotArray test protocols and demonstration protocols, as well as any protocols you may have already defined.
3. Select a protocol from the list, and click **OK**.

The dialog box closes, returns you to the *Start a Printing Procedure* window, shown on the previous page, and fills the button with the name of the protocol you select.

**Note:** If you are printing a microarray for the first time and a protocol has not been created yet, you will need to create a printing protocol. See “Creating a Printing Protocol” in Chapter 3.

4. Check the box to **Create an array content file upon completion of printing**; the fields for selecting a file type and specifying a name become available. If the checkbox is not checked, these fields are dimmed and cannot be used. See Section 1.2.2 earlier in this chapter for more information on selecting and naming an array content file.
5. Click **Run Printing Procedure**. The **Getting Ready for Printing** window opens; review the checklist and confirm each checkpoint.

![Getting Ready for Printing](image)

**Caution:** Make sure that the pin configuration in the printhead exactly matches that shown in the diagram on this window.

6. When the plates and slides are loaded and the instrument door is closed, click **OK**.

The SpotArray system confirms that the door is closed, locks the door, verifies that the instrument configuration matches the protocol, and uses the optical sensor to count and locate plates and substrates. This takes approximately one minute.

The **Configuration Verified** window, with a Summary, Substrates, Plate Changes, and Finished Microarray tab, displays.

7. Check each tab to confirm the instrument configuration and description of the printing procedure. Click **OK** to start the printing procedure.

**Note:** If a plate change is required, the SpotArray pauses the procedure when it’s time to change plates, displays a prompt, and waits for you to acknowledge the prompt when the plates have been changed.
1.3.1 Configuration Verification

The *Configuration Verified* window has four tabs, described below.

1.3.1.1 Summary Tab

The *Summary* tab provides the following information:

- The name of the printing procedure.
- Name of the plate set, number of plates and number of wells in each plate.
- An alert message if pre-print (blotting) slides must be changed during the printing procedure, including the plate changes that require changing the pre-print slides.
- Number of microarrays to be printed and the number of spots per microarray.
- The number of plate changes, and amount of time between plate changes.
- The array content file name, if one was specified.
1.3.1.2 Substrates Tab

The Substrates tab, shown below, provides a list of the loaded substrates, and their barcode ID, if available.
1.3.1.3 Plate Changes Tab

The Plate Changes tab, shown below, provides the following information:

- Plate IDs and the order the plates must be loaded.
- Indicates how many plate changes are required.
- Indicates when a pre-print slide change is required in addition to a plate change.

![Configuration Verified](image)

1.3.1.4 Finished Microarray Tab

The Finished Microarray tab, shown on the next page, provides the following information:

- A graphical representation of the microarray and of the selected subarray.
- The number of the pin creating the subarray.
- The number of times the sample in each well plate will be printed.
- The sample plate ID, name, and description.
1.3.1.5 **Configuration Verification Warning**

If the configuration detected by the SpotArray does not match the protocol requirements, but the procedure can begin, a warning message describes the discrepancy. In the illustration below, the number of plates detected does not match the number of plates specified by the protocol, and may cause pin damage.
1.3.1.6 Configuration Error

If the printing procedure cannot begin because of a configuration or materials problem, a message similar to the following displays. In this case, the problem has to be corrected before the printing procedure can begin.

![Printing Cancelled]

1.3.2 What the Instrument Does During a Printing Procedure

The SpotArray system automatically performs the printing cycles using parameters in the printing protocol, and continually monitors status of the instrument sensors. For information on sensor operation, refer to Appendix B, “SpotArray Sensors.”

- If the environmental control option is installed, the SpotArray checks the environmental humidity and temperature levels. If levels are outside of the acceptable range, the SpotArray pauses, displays the status and waits until the environmental conditions are acceptable before proceeding.

- The SpotArray system turns on the pumps and washes and dries the pins for the number of cycles specified in the protocol. In the event that the water pressure is not sufficient or there is insufficient vacuum for drying, the spotting procedure will not start, or will be paused if already underway.

- If the lid-lifting option is installed and the Plates have lids checkbox for this protocol is checked, the SpotArray removes the lid from the first plate. As each plate is processed, the SpotArray replaces the lid, and removes the lid from the next plate.

- If the printing protocol requires additional plates, you are prompted to change plates. Change the plates, close the instrument door, and resume the process by clicking Resume on the Main Window.

- When all plates specified in the protocol have been processed, the SpotArray stops the pumps, opens the substrate holder, and unlocks the door. Remove all plates, pre-print slides and finished microarrays from the instrument and store them appropriately.
1.4 Monitoring a Printing Procedure

During the printing procedure, samples are loaded from each plate in succession, beginning with the plate in location P1. Samples are loaded beginning at well A1 and continuing bottom to top and left to right for each plate. For example, in a single-pin configuration and 96-well plate, sample is loaded from well A1, then A2, A3, followed by B1, B2 etc., to H12.

Wells that are marked “Full” or “Control” are dipped and printed. Wells that are marked “Empty” or “Bad” may or may not be dipped and printed, depending on the contents of neighboring wells and the pin configuration.

While a printing procedure is being executed, the following progress information displays in the *Main Window*:

- the percentage completed for processing the current batch of plates and the time remaining until the batch is complete.

- the percentage completed for the entire printing procedure and the time remaining until the printing procedure is complete.

1.4.1 Printing Tab - a Status of the Procedure

A graphic display of the completed microarray layout can be viewed on the *Printing* tab on the *Main Window* and a status bar at the bottom of the window displays the progress of the printing procedure. An illustration of the *Printing* tab is shown on the following page.
To access the Printing tab

1. On the SpotArray Main Window, click the Printing tab. The printing tab, as shown below, displays graphic status information for the procedure.

![Printing tab](image)

1.4.2 The SpotArray Instrument Sensors

During a procedure, if sensors detect any error conditions, a message will display on the Main Window. The SpotArray sensors are described in Appendix B, “SpotArray Sensors.”

1.5 Pausing or Cancelling a Printing Procedure

A printing procedure can be cancelled by the user, or it can be paused by either the user or instrument software during a procedure. You may need to stop or pause a procedure if the plates are in the wrong order or if an incorrect plate has been loaded.

---

**Note:** In order to resume the procedure, power to the instrument must be maintained, and the SpotArray software must be kept running. If the lid-lifting option is being used, do not remove or replace any plate lids manually. The SpotArray tracks the last known location of the lids.

You can also pause the procedure for the night, storing the plates in a refrigerator and leaving the substrates in the instrument to resume the procedure in the morning.
1.5.1 Pausing a Printing Procedure

If you pause a procedure while it is printing, the instrument finishes printing the current batch of plates, or cycle, moves the printhead to the home position, unlocks the door, and goes into a paused state.

1.5.1.1 User Initiated Pause

You can pause a printing procedure from the SpotArray Main Window.

To pause a procedure

1. In the SpotArray Main Window, click the Pause button. A status message displays.

   ![Instrument Pausing dialog box]

   The instrument will finish printing the current batch of plates. Depending on the progress in the batch, this may take quite a while. After the cycle is complete, the SpotArray unlocks the instrument door.

2. Click OK to close the Instrument Pausing dialog box.

3. When you are ready, click Resume on the SpotArray Main Window. (The Pause button will have changed to Resume.)

1.5.1.2 Instrument Initiated Pause

The instrument initiates a pause whenever:

- it detects a fluid level error.
- a pre-print slide change or a plate change and pre-print slide change is required.

When the instrument initiates a pause, it unlocks the door, displays the reason for the pause on the monitor, updates the status on the instrument LCD, and goes into a paused state.
1.5.2 Changing Plates or Pre-Print Slides

When it is time to change the sample plates or change both the plates and pre-printing slides, the SpotArray pauses the printing procedure, unlocks the door, and displays the following window:

![Plate Change Window]

To change the plates and pre-print slides

**Warning:** Wear powder-free nitrile gloves when handling the plates and pre-print slides. Refer to Chapter 4 for information on handling plates, and loading the plates and substrates. See *Loading Substrates and Plates* on page 4-8.

1. Remove the plates that have been printed by lifting on the front to release the plate from the holding clips.

2. If it is time to change the pre-print slides as well, remove the used pre-print slides and dispose of them properly.

3. Load new pre-print slides if required.

4. Load the next set of plates by first positioning each plate against the left and rear pins, then pressing down in the right and front. Be sure to install the plates in the exact order specified in the Plate Changes window, and to load them with well A-1 positioned at the lower left.

5. Remove the lids from the plates if the lid-lifting option is not installed or if the protocol does not call for lid-lifting.

6. When the new plates and/or pre-prints slides are loaded and the instrument door is closed, click **Resume** on the Plate Change message.
1.5.3 Resuming a Paused Printing Procedure

A paused printing procedure, whether initiated by you or by the SpotArray, can be resumed by clicking Resume on the Main Window.

![Note: If the lid-lifting option is being used, do not remove or replace any plate lids. The SpotArray tracks the last known location of the lids.]

The instrument door must be closed for the instrument to resume the printing procedure. Once the door is closed, the SpotArray locks the door, closes the substrate holder (if open), and verifies the instrument configuration if the door was opened during the pause. If verification is successful, the SpotArray resumes the paused printing procedure.

![Note: In order to resume the procedure, the power must be maintained to the instrument and the SpotArray software must remain running.]

1.5.4 Stopping (Cancelling) a Printing Procedure

A printing procedure can be cancelled at any time before the procedure completes.

To cancel a procedure

1. Click Cancel on the Main Window.

1.5.4.1 Cancelling while the instrument is verifying configuration

If the procedure is cancelled while the instrument is verifying configuration, the SpotArray stops verification immediately and unlocks the door, opens the substrate holder, and updates the status on the instrument LCD and on the Main Window.

1.5.4.2 Cancelling while the printing procedure is being performed

If the printing procedure is cancelled while it is running, the SpotArray completes the current cycle and replaces the lid on the last plate used if the lid-lifting option is installed, and the Plates have lids checkbox for this protocol is checked. The SpotArray washes and dries the pins, unlocks the door, opens the substrate holder, and updates the status on the instrument LCD and on the Main Window. If an Array Content File had been requested, the software generates the file up to and including the point when the procedure was cancelled.

1.5.5 After the Printing Procedure...

When the printing procedure is complete, the SpotArray system stops the pumps, unlocks the door, and opens the substrate holder. The system displays a Printing Completed message. Remove all plates and substrates (slides) from the instrument. We suggest leaving the pins in the printhead between print runs whenever possible.
Using the SpotArray Software

Chapter 2

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Overview 2-1
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The SpotArray Main Window 2-2
Using Windows 2000 Applications 2-3
Exiting the SpotArray 2-6
Recording the SpotArray System Activity 2-6
Getting Help 2-9

2.1 Overview

The SpotArray software — the program that you use to create a microarray — runs under
the Microsoft Windows 2000 operating system. Programs that run under the Windows
operating system have common elements, including a User Interface (called a Graphical
User Interface, or “GUI”) that provides a visual representation of program commands and
actions.

If you are new to using Microsoft® Windows, you can find basic information in this
chapter; you will also find an overview of the SpotArray software user interface and how
to use the SpotArray.

On Windows systems, the screen on the monitor is called the desktop and each program
opens in a window on the desktop. You can move the windows around or change their
size, and you can reduce a window to a button on the task bar at the bottom of your
monitor screen.

2.2 Starting the SpotArray

To turn on the SpotArray, turn on the On/Off switch on the utility module. If the
SpotArray instrument is connected to a separate workstation, turn on the computer as
described below.

Note: Before turning off your computer or instrument when you are finished, you must
first “shut-down” — an orderly method of closing programs and windows. See Exiting
the SpotArray on page 2-6.

To start the SpotArray software

1. Turn on your computer.
2. At the Log on to Windows dialog box, enter your user name and password, and click OK. The default user name is “spotter” and the default password is “spotter20” on a new SpotArray system. See your IT department for help in setting up new user accounts and setting passwords.

3. Double-click the SpotArray program application icon on the desktop.

2.3 The SpotArray Main Window

When you start an application program it opens into a main window – or starting point – for the application. Figure 2-1 shows the SpotArray Main Window.

From this window, you can start any SpotArray action or open other windows to set up the parameters.
2.4 Using Windows 2000 Applications

Figure 2-1 calls out some of the common elements in a Windows 2000 application. This section, including the following tables, describes some of the common elements of Windows programs. For more detailed information, refer to the Windows online help. (See section 2.7.1 in this chapter.)

Table 2-1: Window Elements

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Buttons</td>
<td>You can click a button to:</td>
</tr>
<tr>
<td></td>
<td>- start an action - for example, the <strong>Start</strong> button</td>
</tr>
<tr>
<td></td>
<td>- open another window - for example, the <strong>Plates</strong> button, which opens a list of plates.</td>
</tr>
<tr>
<td></td>
<td>A SpotArray button changes color or displays a border when you hold the mouse over it, indicating that it is a valid button to click.</td>
</tr>
<tr>
<td></td>
<td>Some buttons, for example those that control optional features, may be “dimmed” and inactive. This means that the option is not installed, and therefore the button for the option cannot be used.</td>
</tr>
<tr>
<td></td>
<td>When you hold the mouse over a dimmed button, it remains gray.</td>
</tr>
<tr>
<td>Tabs</td>
<td>Window Tabs resemble file folder tabs that are lined up one behind the other. You can click a tab and the program brings the Tab contents to the front so you can see them; the other tab names remain visible.</td>
</tr>
<tr>
<td></td>
<td>The SpotArray tabs display log information, diagnostic results, printing procedure progress, or online help.</td>
</tr>
<tr>
<td>Scroll bar</td>
<td>When a list box isn’t large enough to display all of its contents at once, a scroll bar lets you move up and down to see all of the text. Click the mouse on the scroll bar and drag it up or down to move through the window contents, or click on the arrow button.</td>
</tr>
<tr>
<td>Window title</td>
<td>You can click the title bar and drag the window to another location.</td>
</tr>
</tbody>
</table>

Table 2-2: Basic Mouse Techniques

<table>
<thead>
<tr>
<th>Action</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Click</td>
<td>To quickly press and release the left mouse button.</td>
</tr>
<tr>
<td>Double-click</td>
<td>To click the left mouse button twice in rapid succession.</td>
</tr>
<tr>
<td>Drag</td>
<td>To hold down the left mouse button while moving the mouse.</td>
</tr>
<tr>
<td>Point</td>
<td>To move the mouse until the mouse pointer on the screen rests on the item of choice.</td>
</tr>
</tbody>
</table>
Table 2-3: Opening, Closing, Minimizing and Maximizing Program Windows

<table>
<thead>
<tr>
<th>Click...</th>
<th>to...</th>
</tr>
</thead>
<tbody>
<tr>
<td>×</td>
<td>Close the window (exit the program).</td>
</tr>
<tr>
<td>-</td>
<td>Reduce the window to a button on the taskbar (minimize).</td>
</tr>
<tr>
<td>Window</td>
<td>Enlarge the application to fill the screen (maximize).</td>
</tr>
<tr>
<td>Taskbar</td>
<td>Restore the full screen application to its previous size. (The maximize button changed to a Restore button.)</td>
</tr>
</tbody>
</table>

2.4.1 Choosing and Selecting Items

In Microsoft Windows, the words *choose* and *select* have different meanings. Selecting an item usually means marking it with the mouse cursor. Selecting generally highlights or changes the color of an item, but does not initiate an action. Choosing an item, however, does initiate an action.

In the SpotArray application, as you move the mouse cursor over an item that can be selected or chosen, the item changes color to yellow. When you select a SpotArray button, it becomes highlighted.

2.4.2 Using Text Boxes to Enter Information

Text boxes and edit fields (single-line text boxes) let you enter information. The example below shows the *Plate Contents* window with edit fields and text boxes.
2.4.3 Using Option Buttons, Checkboxes and the Protocol Wizard

Option buttons let you make a choice from a number of options. When you select a button, it fills with a black dot. Option buttons are mutually exclusive -- you can select only one at a time. Options that are not available on your system appear dimmed, and are inactive.

Checkboxes also let you make choices, but with checkboxes you can choose as many as are applicable, as shown in the following example. When you click a checkbox, it fills with a check mark; clicking again removes the check mark.

The Protocol Wizard guides you through the process of creating a printing protocol; you can use the step numbers to progress through the protocol windows, or use the Next, Previous, or Finish buttons to move to the previous or next windows.
2.5 Exiting the SpotArray

You can exit the SpotArray user interface and the system continues to run the current printing procedure until completion.

To exit the SpotArray user interface

1. Click the X in the top right-hand corner of the SpotArray Main Window. The following message displays:

   ![Exiting SpotArray]

   Exit the program?
   Yes  No

2. Click Yes to exit or No if you do not want to exit.

3. You may leave the computer or instrument running, or shut down the computer or instrument, after exiting the SpotArray.

To shut down the computer

1. Click the Start menu on the task bar, then click Shutdown.

2. When a message displays on the monitor screen that it is okay to turn off the computer, you can turn off the power.

2.6 Recording the SpotArray System Activity

A system log or event log, keeps a record of all SpotArray activity in a log file. In the file, the SpotArray system logs the date and time of the activity, the current user (as logged onto Windows 2000), and any message. This log may be helpful in tracking operations or for use by Service personnel to diagnose any problems.

The system log tracks the following user requests to:

- start, pause, and cancel printing procedures
- verify instrument configuration
- clean pins
- run diagnostic tests
- add, update, and delete parameters
- modify system configuration
- start-up and shut down the SpotArray application and/or instrument software

and tracks the following information:

- door open, closed, locked, unlocked
- instrument sensor status upon start-up
- instrument sensor status changes
• progress updates during a printing procedure, including start/completion of printing from each plate and start/completion of the printing procedure
• diagnostic test results
• instrument configuration verification results
• changes in the status of client and instrument software communication
• software and hardware failures

You can specify how much disk space you want to dedicate to the system log file, or leave the default of 1 MB. When the size of the log reaches the size of the specified maximum, the SpotArray overwrites the oldest entries, maintaining the same file size.

To specify event logging

1. On the SpotArray Main Window, click **Settings** in the Configure box.

2. The **Settings** dialog box appears. Click **System Settings**.
3. The System Settings window opens.

4. Fill in the information as described in the following table. Click OK to save the settings and close the window.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>System Log</strong></td>
<td></td>
</tr>
<tr>
<td>Size of System Log (MB)</td>
<td>Enter a size in megabytes for the system log file, from 10 MB to 9999 MB. The system default is 1 MB. Once set, the log file is a fixed size; if the file becomes full, new activity is written to the beginning of the file, overwriting the earlier information.</td>
</tr>
<tr>
<td><strong>Connection to instrument</strong></td>
<td></td>
</tr>
<tr>
<td>Instrument Network Name</td>
<td>The instrument network name enables the SpotArray software components to communicate with each other. The information can be entered as an IP address, or as the computer server name. For more information, refer to the SpotArray Installation Manual.</td>
</tr>
</tbody>
</table>

### 2.6.1 Viewing the System Log

You can view and print the system log file by clicking the Log tab on the SpotArray Main Window.

**To view the SpotArray event log**

1. On the SpotArray Main Window, click the Log tab.
2. Use the scroll bar to move through the display as needed.
2.7 Getting Help

2.7.1 Windows Help

The Windows 2000 online help provides information on learning how to use Windows 2000.

To open Windows 2000 help

1. Click the Start button on the task bar.
2. On the Start menu, click Help.
Chapter 3

Defining Plates, Plate Sets, and Creating Protocols

Chapter Summary
Overview 3-1
Defining Plates 3-1
Defining Plate Sets 3-9
Creating a Printing Protocol 3-13
The Printing Protocol Wizard 3-15

3.1 Overview

The SpotArray software lets you add new plates to your library, select certain plates as a set for a printing protocol, or create a printing protocol for a new configuration of microarray.

From the SpotArray Main Window, you can define the following:

- **Plates**: for each plate in your library, a description of the type of plate used and the contents of each well.
- **Plate Sets**: a subset of plates in the plate library that is an ordered list of the plates to be used to print microarrays.
- **Printing Protocol**: a list of instructions for printing microarrays.

3.2 Defining Plates

You must define each plate that will be used to print microarrays. To create an accurate array content file, you must also define the contents of each plate well. The array content file lists the location of each spot in a microarray, its plate and well source location, gene ID and gene name, using the information that is entered when defining plates.

The SpotArray is designed to know the precise location of wells in several types of plates and supports the plates listed in Table 3-1.

---

Note: If you want to use a type of plate not listed here, contact PerkinElmer Technical Support help:

telephone 888-323-9681
- or -
pbtsupport@perkinelmer.com
Table 3-1: Plates supported by the SpotArray

<table>
<thead>
<tr>
<th>Manufacturer</th>
<th>Model Code</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corning Costar</td>
<td>3364</td>
<td>96-well polypropylene</td>
</tr>
<tr>
<td>Corning Costar</td>
<td>3365</td>
<td>96-well polypropylene</td>
</tr>
<tr>
<td>Corning Costar</td>
<td>3897</td>
<td>96-well conical polystyrene</td>
</tr>
<tr>
<td>Costar</td>
<td>3795</td>
<td>96-well round polystyrene</td>
</tr>
<tr>
<td>Costar</td>
<td>3632</td>
<td>96-well polystyrene white plate (clear bottom)</td>
</tr>
<tr>
<td>Costar</td>
<td>3693</td>
<td>96-well polystyrene white 1/2 area</td>
</tr>
<tr>
<td>Costar</td>
<td>3912</td>
<td>96-well polystyrene white</td>
</tr>
<tr>
<td>Costar</td>
<td>3368</td>
<td>96-well polystyrene easy wash</td>
</tr>
<tr>
<td>Costar</td>
<td>3631</td>
<td>96-well polystyrene black plate (clear bottom)</td>
</tr>
<tr>
<td>Costar</td>
<td>3694</td>
<td>96-well polystyrene black 1/2 area</td>
</tr>
<tr>
<td>Costar</td>
<td>3915</td>
<td>96-well polystyrene black</td>
</tr>
<tr>
<td>Costar</td>
<td>3695</td>
<td>96-well polystyrene 1/2 area</td>
</tr>
<tr>
<td>Costar</td>
<td>3635</td>
<td>96-well polystyrene</td>
</tr>
<tr>
<td>Costar</td>
<td>9017</td>
<td>96-well polystyrene</td>
</tr>
<tr>
<td>Costar</td>
<td>2615</td>
<td>96-well black plate (clear bottom)</td>
</tr>
<tr>
<td>Generic 96</td>
<td>Generic 96</td>
<td>96-well plate</td>
</tr>
<tr>
<td>Greiner</td>
<td>655201</td>
<td>96-well polypropylene</td>
</tr>
<tr>
<td>Greiner</td>
<td>651101</td>
<td>96-well conical polystyrene</td>
</tr>
<tr>
<td>Greiner</td>
<td>651201</td>
<td>96-well conical polystyrene</td>
</tr>
<tr>
<td>Marsh</td>
<td>N29074</td>
<td>96-well conical polypropylene</td>
</tr>
<tr>
<td>Nunc</td>
<td>267245</td>
<td>96-well round polystyrene</td>
</tr>
<tr>
<td>Nunc</td>
<td>260844</td>
<td>96-well polystyrene</td>
</tr>
<tr>
<td>Nunc</td>
<td>245128</td>
<td>96-well conical polystyrene</td>
</tr>
<tr>
<td>Nunc</td>
<td>249944</td>
<td>96-well conical polypropylene</td>
</tr>
<tr>
<td>Nunc</td>
<td>249945</td>
<td>96-well conical polypropylene</td>
</tr>
<tr>
<td>Nunc</td>
<td>249949</td>
<td>96-well conical polypropylene</td>
</tr>
<tr>
<td>Packard</td>
<td>6005190</td>
<td>96-well polystyrene</td>
</tr>
<tr>
<td>Whatman Polyfiltronics</td>
<td>7701-5250</td>
<td>96-well conical polypropylene</td>
</tr>
</tbody>
</table>
You can add, change, copy or delete plates. You can also view which protocols use each plate and sort the list of plates by ID, name, or description.

**To define plates**

1. Click **Plates** in the Configure box on the **Main Window**.

![Click here](image)
The *List of Plates* dialog box opens, as shown:

2. Click **Close** if you want to close the window without making a selection
   -OR-
   Click one of the following:
   - **Add** to add a new plate definition.
   - **Change**, after selecting a plate, to change an existing definition.
   - **Duplicate**, after selecting a plate, to copy an existing definition. You can save the definition under a new name and modify it to make a new plate description.
   - **Delete**, after selecting a plate, to delete the plate definition. You cannot delete the definition for a plate that is assigned to a plate set; you must first delete it from any plate set to which it is assigned.
Clicking Add, Change, or Modify opens the Plate Contents window.

3. Enter the information as described in the following table.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plate Identification</td>
<td>The name and barcode combination must be unique.</td>
</tr>
<tr>
<td>Name</td>
<td>Enter a name for the plate; you can use alphanumeric characters. The name and identity combination must be unique.</td>
</tr>
<tr>
<td>Barcode</td>
<td>Enter the value of the barcode on the plate, if any. SpotArray systems that are equipped with a barcode reader can verify that the plates with barcodes are installed in the correct order for a selected protocol.</td>
</tr>
<tr>
<td>Description</td>
<td>Enter a description of the plate; you may want to enter information such as where the plate was obtained or the contents of the plate. This field may be left blank if the barcode field is filled in.</td>
</tr>
<tr>
<td>Plate Type</td>
<td>The plate type is the plate’s manufacturer, part number and capacity. Click the Plate Type button and select a plate type from the window that opens, then click OK. This button fills in with the plate description; the plate capacity is listed below the button.</td>
</tr>
<tr>
<td>View Contents as</td>
<td>Select to view the contents as a List or as a Map (a graphical representation of the sample wells.)</td>
</tr>
</tbody>
</table>
4. You may obtain the plate content information from a text file by clicking **Import from File** at the bottom of the *Plate Contents* window, or you may enter the following information for each well:

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Well</td>
<td>The well numbers display automatically:</td>
</tr>
<tr>
<td></td>
<td>A01 to H12 for 96-well plates</td>
</tr>
<tr>
<td></td>
<td>A01 to P24 for 384-well plates</td>
</tr>
<tr>
<td>Accession No.#</td>
<td>Enter the gene's accession number.</td>
</tr>
<tr>
<td>Gene Name</td>
<td>Enter the gene name.</td>
</tr>
<tr>
<td></td>
<td>You may download the Gene accession number and Gene Name in a text file from the GenBank internet web site.</td>
</tr>
<tr>
<td>Status</td>
<td>The default status is <strong>Full</strong>. For any well that you want to change, click the Status button, and select from:</td>
</tr>
<tr>
<td></td>
<td>Empty</td>
</tr>
<tr>
<td></td>
<td>Full</td>
</tr>
<tr>
<td></td>
<td>Bad</td>
</tr>
<tr>
<td></td>
<td>Control</td>
</tr>
<tr>
<td>Import from File</td>
<td>Click to import an existing plate contents description file. See <strong>Importing Plate Contents Information</strong> on page 3-7.</td>
</tr>
</tbody>
</table>

**Note:** Before a procedure begins, the SpotArray checks the plate contents information for the plates to be printed. Any well that has a status of “Empty” or “Bad” is not dipped, unless the pin configuration is such that pins dip into the wells marked Empty or Bad at the time that other pins dip into wells marked “Full”.

5. If you prefer to view the information as a graphic representation of the plate wells, select **Map**. The **Plate Contents** window displays as shown below. Place the cursor over a well number. You can view or enter the well information in the Well contents box by clicking the appropriate well.

6. Click **OK** to save the information and close the window, or click **Cancel** if you do not want to save the information.

### 3.2.1 Importing Plate Contents Information

You can import text files that describe the well contents of a plate. In some instances, you may receive ready-to-print plates from vendors or other users that already have a file describing the contents, or you may download a text file from a content provider’s internet web site (for example, GenBank).

#### To import a plate contents file

1. Click **Plates** in the Configure box on the SpotArray **Main Window**. The **List of Plates** window opens.

2. Click **Add**, or select a plate and click **Update**. The **Plate Contents** window opens.
3. At the bottom of the Plate Contents window (shown on page 3-7), click **Import from File**. The Open dialog box appears.

4. Specify a file: you can type the filename into the *File name* field; click the down-arrow in the “Look in” list box; or click the folder icon to browse the directories until you see the file you want to import. The contents file must be a text file with a file extension of “.txt” or “.csv”. See Appendix E for a description of the required format for Plate Contents files.

   The data from the imported file becomes part of the SpotArray database when you save the Plate definition.

### 3.2.2 Deleting a Plate

To “delete” a plate means to remove its definition from the plate library; do this only if you won’t want to use the plate again. You cannot delete a plate that is specified in a *plate set* (the plates to be used for a printing protocol). To delete a plate, you must first remove it from any plate sets that use it. (See **Removing a Plate from a Set** on page 3-12.)

**To delete a plate**

1. On the *Main Window*, click **Plates** to open the *List of Plates* window.

2. In the list of plates, highlight the plate you want to delete and click **Delete**. A confirmation window asks if you want to delete the plate contents.
3. Click **Yes** to delete the plate, or **No** if you do not want to delete the plate.

Since you cannot delete a plate that belongs to a plate set, if you attempt to delete a plate that is being used, the following message displays:

![Delete Plate Contents dialog](image)

4. Click **OK** to close the dialog box and return to the *List of Plates* window.

To see which plate sets are using the plate, click **View Usage**. If you want to delete this plate, you can edit each of the plate sets listed and remove the plate from the plate sets. See *Removing a Plate from a Set* on page 3-12.

### 3.3 Defining Plate Sets

A *Plate Set* describes all plates to be used for a printing protocol, and lists the order in which they must be loaded into the SpotArray instrument.

To **define a plate set**

1. Click **Plate Sets** in the Configure box on the *Main Window*. The *List of Plate Sets* window opens.

![List of Plate Sets](image)
2. Click **Close** if you want to close the window without making a selection - OR-
click one of the following:

- **Add** to add a new plate set.
- **Change**, after selecting a plate set, to modify an existing plate set.
- **Duplicate**, after selecting a plate set, to copy an existing plate set. You can save
  the copy with a new name and make changes.
- **Delete**, after selecting a plate set, to delete the description of an existing plate
  set. You cannot delete a plate set that is assigned to a printing protocol; you
  must first delete it from any printing protocol to which it is assigned.

The Plate Set window opens.

![Plate Set Window](image)

3. Enter a Name and Description and select the capacity of the plates in the plate set.
   Although all of the plates in a plate set do not need to be of the same type, they must
   all be of the same capacity (96-well or 384-well).

4. In the “Plates not in set” box (which lists all of the plates in the library not yet
   selected), select the plate you want to include in the set and click **Add selected
   plates to set**. The “Plates not in set” box displays a list of all available plates of the
   type that you selected (96-well or 384-well).

   **Note:** To select more than one plate, hold the Ctrl key down and click on each plate that
   you want to select, then click **Add selected plates to set**.

5. In the “Plates in Set” box, you can re-order the plates. Select a plate and click any of
   the buttons until the plates are in the order that you want them to be in:
- **To Top** to move the highlighted plate to the top of the list.
- **Up** to move the highlighted plate up one position.
- **Down** to move the highlighted plate down one position.
- **To Bottom** to move the highlighted plate to the bottom of the list.

6. To remove plates from the set before saving the Plate Set definition, highlight the plate or plates in the “Plates in set” box and click **Remove selected plates from set**.

7. Click **OK** to save the Plate Set definition, or click **Cancel** if you do not want to save your selections or changes.

---

**Note:** You can use a plate only once in a plate set.
3.3.1 Removing a Plate from a Set

Removing a plate from a plate set leaves the plate in your library, and it is still available for use by other plate sets.

To remove a plate from a plate set

1. Open the Plate Set window for the selected plate set, as described in the preceding section.

2. In the “Plates in set” box, highlight the plate or plates that you want to remove, and click Remove selected plates from set.

3. Click OK to save the modified plate set definition.

3.3.2 Deleting a Plate Set

Deleting a plate set does not delete the plates from the plate library; the plates will still be available to select for other plate sets.

To delete a plate set

1. On the Main Window, click Plate Sets. The List of Plate Sets window opens.

2. Select the plate set that you want to delete.

3. To see which printing protocols are using the plate set, click View Usage.

4. Click Delete to remove the plate set definition. At the confirmation prompt, click Yes to delete, or click No if you do not want to delete the plate set.

If a printing procedure using the plate set is currently underway, the procedure will continue as originally defined.
3.4 Creating a Printing Protocol

If this is the first time you are printing a new configuration of microarray, you need to create a printing protocol, as described in this section.

A printing protocol defines how the SpotArray system executes a printing procedure. The final results of the microarray depend on the settings in the protocol: for example, the number and position of pins, the number of plates, and the number of replicate spots.

3.4.1 Working with Printing Protocols

You can select a printing protocol from the list of available protocols to run, duplicate, or modify, or you can create a new protocol. See The Printing Protocol Wizard on page 3-15.

To select a protocol

1. On the SpotArray Main Window, click Printing Protocols in the Configure box.

2. The List of Printing Protocols window opens, as shown on the next page.
3. Sort the list by **Name** or **Description** (optional) by clicking the button at the top of either column.

4. Click one of the following to open the *Printing Protocol* wizard:

   - **Add** to create a new protocol.
   - **Change**, after selecting a protocol, to modify an existing protocol.
     If you change and save a printing protocol, the SpotArray uses the updated protocol the next time a printing procedure calls for that protocol. If a printing procedure is already under way using the protocol you just changed, the procedure finishes printing using the original protocol.
   - **Duplicate**, after selecting a protocol, to copy the highlighted protocol. You can create a new, modified protocol by clicking this button and saving the protocol with a new name.

Or, click:

   - **Delete**, after selecting a printing protocol, to delete a printing protocol.

The *Printing Protocol* wizard opens.
3.5 The Printing Protocol Wizard

The Printing Protocol wizard opens with the Basic Information window displayed, as shown below. From this window, using the wizard, you can display and set all of the parameters required for a printing procedure, including parameters for any optional features that may be installed on the SpotArray system.

**Note:** The “Barcodes” selection does not display if a barcode reader is not installed; instead, “Pins” will be number 2, and each subsequent selection moves up one number.

You can move through the windows by clicking the numbers on the left, or by clicking the Next and Back buttons at the bottom of each window to move to the previous or next window.

### 3.5.1 Basic Information

In the Printing Protocol - Basic Information window, provide a name and description for the protocol, and select the set of plates to be used to create the microarrays.
To name a printing protocol and select plates

1. Fill in the information as listed in the following table.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Name</td>
<td>Enter a name for the printing protocol. You must enter a name in order to proceed.</td>
</tr>
<tr>
<td>Description</td>
<td>Enter a description for the printing protocol.</td>
</tr>
<tr>
<td>Plate Set</td>
<td>Click the “Select from List” Plate Set button to open a list of available plate sets. Select a plate set and click OK.</td>
</tr>
<tr>
<td>Description</td>
<td>The description field fills in automatically with the description that was entered when the selected plate set was created.</td>
</tr>
<tr>
<td>Plate type</td>
<td>The plate type field fills in automatically with information that was entered when the plate set was created.</td>
</tr>
<tr>
<td>Number of plates per plate change</td>
<td>This is the number of plates to be loaded into the instrument at one time. Enter a number, from 1 to 4, of plates per plate change; this will be the number of plates that you load into the instrument at a time. To minimize plate changes, select 4 (maximum); to minimize the time that sample plates are left in the instrument, select 1.</td>
</tr>
<tr>
<td>Plates have lids</td>
<td>Check this box if the plates have lids and the lid-lifting option is installed. The SpotArray will automatically remove and replace the sample plate lids during the printing procedure.</td>
</tr>
</tbody>
</table>

2. Click Next or click “Barcodes” to go to the next window, or click Finish to save the printing protocol as is, without checking any other parameters. Click Cancel if you do not want to save the information you just entered.

Note: If a barcode reader is not installed, the Barcode button, and the associated window, are not available. In that case, clicking Next opens the Pins window.

3.5.2 Barcodes

With an optional barcode reader installed, the SpotArray can read barcodes on either or both the sample plates and substrates, as specified in the printing protocol.

3.5.2.1 Barcodes on Sample Plates

The SpotArray reads the barcodes on sample plates, beginning with Plate 1, during verification before starting the procedure, and after each plate change. The barcodes on the present sample plates are compared to the barcodes in the Plate Set information. Incorrect barcodes, or missing barcodes for protocols that require them, are reported as errors; the user can cancel the printing procedure or override the error and continue. If the lid-lifting option is installed, and the Plates have lids checkbox in the Basic Information window is checked, the SpotArray removes the lid from the first plate, reads the barcode, replaces the lid, and moves to the next plate.
3.5.2.2 Barcodes on Substrates

The SpotArray reads up to two barcodes per substrate during verification before starting the procedure. The barcode data is saved to the Array Content File (.GAL file). Missing barcodes for protocols that require them are reported as errors; the user can cancel the printing procedure or override the error and continue.

**To specify barcode information**

1. Click **Next** from the **Basic Information** window, or click “Barcodes” from the **Printing Protocol** wizard.

![Print Protocol - Barcodes](image)

2. Fill in the information as listed in the following table.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Read barcodes on plates</td>
<td>Check this box to have the SpotArray read the barcode on each plate. The SpotArray verifies that the correct plates are loaded and in the correct order.</td>
</tr>
<tr>
<td></td>
<td>Checking this box enables the subordinate checkbox.</td>
</tr>
<tr>
<td>Barcodes are required on all plates</td>
<td>Check this box to require barcodes on all plates. If the SpotArray encounters a sample plate without a barcode, the SpotArray reports an error during verification before starting a printing procedure.</td>
</tr>
<tr>
<td>Read barcodes on substrates</td>
<td>Check this box to have the SpotArray read the barcode(s) on each substrate. If a substrate has two barcodes, the SpotArray stores them in the Array Content file as a single string.</td>
</tr>
<tr>
<td></td>
<td>Checking this box enables the subordinate checkboxes.</td>
</tr>
</tbody>
</table>
3. Click **Next** or click “Pins” to go to the next window, or click **Finish** to save the printing protocol. Click **Cancel** if you do not want to save the information you just entered.

### 3.5.2.3 Supported Barcodes

Using barcodes as unique identifiers on individual plates and substrates provides a valuable way for tracking samples and workflow, and automating analysis of microarray experiments. Using barcodes allows a gene sample to be tracked through the entire microarray process including printing, hybridization, scanning, quantitation, and visualization.

The SpotArray barcode reader supports Code 128, 39, and Interleave 2 of 5 (ITF) symbologies. The maximum number of digits supported by each symbology depends on the minimum element width (the barcode “pitch”) in the printed barcode. Alphabetic and numeric digits can be combined, but alphabetic digits take more space than numbers, thus decreasing the maximum number of allowed digits.

Tables 3-2 and 3-3 list the maximum number of alphanumeric characters for substrate barcodes and plate barcodes.

#### Table 3-2: Substrate Barcodes

<table>
<thead>
<tr>
<th>Element Width</th>
<th>Code 128</th>
<th>Code 39</th>
<th>ITF</th>
</tr>
</thead>
<tbody>
<tr>
<td>.010&quot;</td>
<td>.254 mm</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>.0075&quot;</td>
<td>.19 mm</td>
<td>12</td>
<td>10</td>
</tr>
<tr>
<td>.005&quot;</td>
<td>.127 mm</td>
<td>22</td>
<td>16</td>
</tr>
</tbody>
</table>

#### Table 3-3: Plate Barcodes

<table>
<thead>
<tr>
<th>Element Width</th>
<th>Code 128</th>
<th>Code 39</th>
<th>ITF</th>
</tr>
</thead>
<tbody>
<tr>
<td>.010&quot;</td>
<td>.254 mm</td>
<td>26</td>
<td>18</td>
</tr>
<tr>
<td>.0075&quot;</td>
<td>.19 mm</td>
<td>32</td>
<td>24</td>
</tr>
</tbody>
</table>
Pre-printed barcode labels are commercially available, or you can print your own that conform to the guidelines in Tables 3-2 and 3-3. PerkinElmer has qualified the following blank labels:

<table>
<thead>
<tr>
<th>Manufacturer</th>
<th>Part Number</th>
<th>Size (inches)</th>
<th>Size (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Substrate</td>
<td>Intermec</td>
<td>E17512</td>
<td>0.920&quot; x 0.550&quot;</td>
</tr>
<tr>
<td>Plate</td>
<td>Intermec</td>
<td>E16809</td>
<td>2.000&quot; x 0.250&quot;</td>
</tr>
</tbody>
</table>

### 3.5.2.4 Placing the Barcode Labels

The barcode labels must be placed on the substrate or plate as shown in the following illustration.
3.5.3 Selecting Pins

Specify the number of pins to be used with this protocol.

The number of pins specified here should correspond to the actual number of pins installed in the printhead when printing with this protocol. The SpotArray makes it easy to select pins using a graphical representation of the allowable pin configurations. The maximum number of pins are:

- 4 x 12 with 4.5 mm spacing (384-well plates)
  - supporting pin configurations of 1, 4, 16, 32, and 48
- 2 x 6 with 9 mm spacing (96-well plates)
  - supporting pin configurations of 1, 4, 8, and 12

3.5.3.1 Selecting the Pin Configuration

To select pins

1. Click **Next** from the **Barcodes** window, or click “Pins”. The **Pins** window opens. The **Pins** window shows the number of allowed pins for the plate type.

**Note:** You must verify that the pins installed in the printhead match the pins illustrated in the diagram in the “Pins” window.
2. It is possible to print with just one pin -- that pin must be in position A1. Pin A1 position corresponds to the location of well A1 on the sample plate and is required as the starting point for your array.

3. Select the button for the pin configuration to be used:
   - for 96-well plates, you may select 1, 4, 8, or 12 pins;
   - for 384-well plates, you may select 1, 4, 16, 32, or 48 pins.

4. To use pin lifting, if the pin-lifting option is installed:
   - each zone must have the same pin configuration
   - zones must be adjacent, and Zone A must be used (that is, configurations of Zone B and Zone C, or Zone A and Zone C, are not valid)

Pin-lifting is enabled by checking a box on the Array Layout window. See Array Layout on page 3-23.

5. Click Next or click “Pre-Print” to go to the next window, or click Finish to save the printing protocol. Click Cancel if you do not want to save the information you just entered.
3.5.4 Pre-Printing

Pre-printing ("blotting") ensures consistent spot size by removing excess sample from the pins. When the pins are dipped into the wells of a plate, excess sample may accumulate on the tip and sides of the pins. The excess sample must be removed from the pins before quality spots can be created. Excess sample is removed by pre-printing onto blotting slides prior to printing onto the substrates.

To set Pre-printing

1. Click “Pre-Print”, or click **Next** in the *Pins* window. The *Pre-print* window opens.

2. Provide the information as described in the following table.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Pre-prints</strong></td>
<td>Specify how many times the pins must be printed onto the blotting substrate before printing on the microarray substrate.</td>
</tr>
<tr>
<td>Number of pre-prints after each sample load</td>
<td>Valid values range from 0 to 99 with a default of 5. The number of pre-prints required depends on the type of sample and type of pre-printing (blotting) substrate. The higher the number of pre-prints, the lower the amount of remaining sample available for printing.</td>
</tr>
<tr>
<td>Pre-print spot spacing, center to center (µm)</td>
<td>Specify in microns the center-to-center spacing that is needed between pre-print spots. This spacing should be large enough to accommodate the largest pre-print spot size. Valid values range from 25 to 1000 microns and can be changed in increments of 1 micron. The default value is 300. The smaller the spots, the smaller the space between the spots can be.</td>
</tr>
</tbody>
</table>
3. Click **Next** or click “Array Layout” to go to the next window, or click **Finish** to save the printing protocol. Click **Cancel** if you do not want to save the information you just entered.

### 3.5.5 Array Layout

The values specified in this window for printing determine the array pattern; that is, the number of duplicate spots and the amount of space between spots, and where the array should be positioned on the substrate.

**To specify the margins and spot spacing**

1. Click **Next** from the *Pre-Print* window, or click “Array Layout” from the *Protocol Wizard*. The *Array Layout* window opens.
2. Fill in the information as described in the following table.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Spots</strong></td>
<td>Specify the number of times each sample is to be printed in the array (replicate spots). The SpotArray prints the replicates adjacent to each other. Valid values range from 1 to 10,000 with a default of 2.</td>
</tr>
<tr>
<td>Number of times to print</td>
<td>Valid values range from 1 to 10,000 with a default of 2.</td>
</tr>
<tr>
<td>each spot within array</td>
<td></td>
</tr>
<tr>
<td>Nominal spot diameter (µm)</td>
<td>This value has no effect on printing, but is exported in .GAL format array content files to set up the quantitation grid in the quantitation software. Enter a value that represents the minimum spot diameter that you expect.</td>
</tr>
<tr>
<td>Spot spacing, center to</td>
<td>Specify in microns the center-to-center spacing of the printed spots. Typically the center-to-center spacing is specified at 1.5 to 2 times the diameter of the spots. Valid values range from 100 to 1000 microns and can be changed in increments of 1 micron. The default value is 200.</td>
</tr>
<tr>
<td>center (µm)</td>
<td></td>
</tr>
<tr>
<td><strong>Subarray Spacing</strong></td>
<td>Check this box to use pin lifting, if the pin-lifting option is installed. The advantages and disadvantages of pin lifting are described in the next section. See Pin Lifting Advantages on page 3-25.</td>
</tr>
<tr>
<td>Use pin lifting</td>
<td></td>
</tr>
<tr>
<td>**Location of array on</td>
<td>Select <strong>Top</strong>, <strong>Bottom</strong>, <strong>Center</strong> or <strong>Custom</strong>. The diagram in the window will move to reflect your selection. If you choose <strong>Custom</strong>, the edit boxes under <strong>Custom Array Location</strong> become available.</td>
</tr>
<tr>
<td>substrate</td>
<td></td>
</tr>
<tr>
<td>Leave space for a barcode/</td>
<td>Check this box to leave room at the bottom of the substrate for a barcode label. A barcode label requires 14mm space at the bottom.</td>
</tr>
<tr>
<td>label on the bottom.</td>
<td></td>
</tr>
<tr>
<td><strong>Custom array parameters</strong></td>
<td>In the edit boxes, specify in mm:</td>
</tr>
<tr>
<td>Upper left spot center</td>
<td>From left - specify in mm the amount of space from the left edge of the substrate to the beginning of the microarray.</td>
</tr>
<tr>
<td></td>
<td>From top - specify in mm the amount of space from the top edge of the substrate to the beginning of the microarray. The top margin is limited by the pin configuration; for example, pins in Zone A cannot reach the top of the slide. If you specify an invalid amount, an error message displays.</td>
</tr>
<tr>
<td>Subarray dimensions</td>
<td>Width - specify in mm the amount of space for the total width of each subarray. This amount should be greater than two times the spot space (as specified in the Printing window) and less than the pin distance.</td>
</tr>
<tr>
<td></td>
<td>Height - specify in mm the amount of space for the total height of each subarray.</td>
</tr>
<tr>
<td><strong>Redraw Array</strong></td>
<td>Click this button to refresh the screen and redraw the diagram of the array with your latest selections and parameters.</td>
</tr>
</tbody>
</table>

3. Click **Next** or “Pin Motion” to go to the next screen, or click **Finish** to save your changes and close. Click **Cancel** if you do not want to save the information you just entered.
3.5.5.1 Pin Lifting Advantages

Generally, it is desirable to create compact microarrays — with the spots printed closely together — because compact microarrays are easier to hybridize, scan, and quantitate. With a fixed, single zone printhead, you often need to choose between creating compact microarrays with a low-pin configuration, which prints more slowly, or using a high-pin configuration that prints faster, but does not create a compact microarray.

Pin lifting divides the printhead into two or three zones that independently can lift or lower pins for printing. The separate zones of the printhead are washed, dried, and loaded with sample as a single unit, but each zone operates independently during printing. This allows the creation of more compact arrays, as described in the next section, and at the same time allows using a higher pin count for faster printing.

3.5.5.2 How Pin Lifting Works

The following two examples show how a microarray might look with and without pin lifting, with all other parameters for the protocol remaining the same.

Figure 3-1 illustrates a microarray created with an 12-pin configuration and no pin lifting. Each pin creates a subarray. Because of the 9mm spacing between pins, there is unused space between the subarrays.

Figure 3–1 Microarray Resulting from Protocol without Pin Lifting
Figure 3-2 illustrates a microarray created with a 12-pin configuration with three zones for pin lifting. Notice that the space between subarrays that is unused without pin lifting can be filled when using pin lifting. The subarrays are separated by the spot spacing indicated in the protocol, rather than by the pin spacing.

Figure 3-2 The Same Protocol using Pin -Lifting

After the first zone -- Zone C -- prints, the printhead moves slightly to print Zone B just below the subarray of Zone C; then the printhead moves again slightly to print Zone A just below the subarray of Zone B.

Note: If more than half of the pin spacing is required for each subarray in a two-zone configuration, or if more than a third of the pin spacing is required for each subarray in a three- zone configuration, the empty spaces between subarrays cannot be used for pin lifting because they are not large enough. The SpotArray will not let you create a protocol using pin lifting under these circumstances.

3.5.6 Pin Motion - Setting Printhead Velocity, Dwell Time and Motion

The pin motion that you specify here, together with the surface properties of the substrate, the viscosity of the sample being printed, and the humidity level in the printing environment, affects the size of the spot. Pin motion includes the velocity — how fast the printhead moves downward and upward; the dwell time — how long the printhead remains stationary after lowering the pins; and overtravel — the distance the printhead continues to move downward after the pins touch the substrate or plate.
The default values are a good starting point for typical printing conditions; you can then adjust the default values empirically if needed.

The substrate thickness also needs to be defined here. Substrate thickness is specified by the substrate manufacturer. The substrates supported by the SpotArray can have dimensions in the following ranges:

<table>
<thead>
<tr>
<th>Dimension</th>
<th>Specification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Length</td>
<td>2.95 inches to 3.00 inches (74.9mm to 76.2 mm)</td>
</tr>
<tr>
<td>Width</td>
<td>0.97 inches to 1.02” inches (24.6 mm to 26.12mm)</td>
</tr>
<tr>
<td>Thickness</td>
<td>0.03 inches to 0.05 inches (0.76 mm to 1.27 mm)</td>
</tr>
</tbody>
</table>

At the time of this printing, Telechem slides are 0.96 mm thick and Corning slides are 1.0 mm thick.

**To set pin motion**

1. Click **Next** from the *Array Layout* window or click “Pin Motion”. The *Pin Motion* windows displays.
2. Fill in the information as described in the table below.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Printing approach velocity (mm/sec)</td>
<td>Specify the speed at which the printhead should approach the substrate after pins have been loaded with sample. The default value is 15 mm/sec.</td>
</tr>
<tr>
<td>Printing departure velocity (mm/sec)</td>
<td>Specify the speed at which the printhead should leave the substrate after touching the pins to the substrate. The default value is 10 mm/sec.</td>
</tr>
<tr>
<td>Printing overtravel (µm)</td>
<td>The overtravel distance is the amount of distance the printhead continues to travel downward once the pins touch the surface of the substrate. The default value is 500 microns. This accommodates variations in slide thickness and pin length.</td>
</tr>
<tr>
<td>Printing dwell time (mSec)</td>
<td>Enter the amount of time that the printhead motion is stationary after lowering the pins onto the substrate during printing. The default value is 25 mSec.</td>
</tr>
<tr>
<td>Sample load overtravel (µm)</td>
<td>The amount of distance the printhead continues to travel downward once the pins touch the bottom of a well while loading. The default value is 500 microns. It is okay for Stealth pins to touch the well bottoms.</td>
</tr>
<tr>
<td>Sample load dwell time (mSec)</td>
<td>Enter the amount of time that the printhead motion is stationary after lowering the pins into the sample plate during sample uptake. The default is 1000.</td>
</tr>
<tr>
<td>Substrate thickness (mm)</td>
<td>Enter the thickness, in millimeters, of the substrates on which you are printing with this protocol. This information is used to determine how far downward (the Z direction) the printhead must travel for the pins to contact the substrate. Valid values range from 0.1 to 2.0, with a default value of 1.0 millimeter.</td>
</tr>
<tr>
<td>Maximum number of printed spots per sample load</td>
<td>Specify how many spots can be printed after pre-printing (blotting). This information is used to determine if more than one load is required to print all of the specified spots. This value must be empirically derived, and is dependent on the number of pre-prints and the surface properties of your substrates. The approach velocity and dwell time while printing also affect the number of spots that can be printed because slower velocities and longer dwell times deposit more sample onto the substrate. This value depends on the type of pin used and the sample volume it is able to hold. The Telechem SMP3 pin holds approximately 0.25 µl of sample, and is able to print approximately 150 replicate spots before reloading with sample. If your printing protocol requires more, the pins will be loaded (dipped into the sample) more than once. Valid values range from 1 to 999 with a default of 150.</td>
</tr>
<tr>
<td>Speed of printhead X-Y motion</td>
<td>Select a speed for the printhead to move along the X-Y axis: slow, medium, or fast. The recommended speed is medium. The effect of this setting on cycle times is modest. Slow could improve array geometry; Fast will slightly reduce cycle time.</td>
</tr>
</tbody>
</table>
3. Click **Next** or click “Wash & Dry” to go to the next window, or click **Finish** to save the printing protocol. Click **Cancel** if you do not want to save the information you just entered.

### 3.5.7 Wash and Dry

You can specify how many times the pins should be washed, and for how long. Washing and drying the pins prevents sample carry-over from contaminating array spots and plates.

**To set the wash and dry cycles**

1. Click “Wash and Dry”, or click **Next** from the *Pre-Print & Printing* window. The *Wash & Dry* window opens.

2. Enter the information for each field, as described in the table below.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Length of wash procedure (sec)</td>
<td>Specify the amount of time, in seconds, that the wash jets will spray the pins. Valid values range from 0.5 to 10.0, with a default value of 2.0 seconds.</td>
</tr>
<tr>
<td>Number of times to wash</td>
<td>Specify the number of times to wash the pins. The default is 1.</td>
</tr>
<tr>
<td>Length of dry procedure after washing (sec)</td>
<td>Specify the amount of time, in seconds, that the dryer vacuum will be applied to the pins after washing. Valid values range from 3 to 20 with a default of 3 seconds.</td>
</tr>
</tbody>
</table>

3. Click **Next** or click “Environmental Control” to go to the next window, or click **Finish** to save the printing protocol. Click **Cancel** if you do not want to save the information you just entered.
3.5.8 Specifying Environmental Control

The environmental control module is optional but recommended. You can set the relative humidity and specify in your protocol that the printing procedure pause or stop if the humidity or temperature goes out of range.

The recommended operating humidity is 50 percent to 65 percent with high-salt buffers to ensure high-quality printing. With the Environmental Control Module installed, the SpotArray automatically adds humid air to the printing environment when necessary and pauses printing, if directed by the printing protocol, when the humidity is out of range.

Although the SpotArray instrument does not control the temperature in the printing environment, it does monitor the temperature. When the temperature is out of range, the SpotArray pauses printing, if specified by the printing protocol.

Note: If your experiment does not depend on the ambient temperature, do not select the checkbox that tells the SpotArray to stop printing when the temperature is out of range.

To set the environmental control options

1. Click Next from the Wash & Dry window, or click “Environmental Control” from the Printing Protocol wizard. The Environmental Control window opens
2. Fill in the information as listed in the following table.

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Humidity</strong></td>
<td>Check this box to have the SpotArray system pause a printing procedure if the humidity level goes outside of the acceptable range. When the humidity level returns to the acceptable range, the SpotArray resumes the printing procedure.</td>
</tr>
<tr>
<td>Do not print when humidity is outside the acceptable range</td>
<td></td>
</tr>
<tr>
<td>Minimum acceptable humidity (% RH)</td>
<td>Enter the minimum and maximum percentage humidity at which it is acceptable to print.</td>
</tr>
<tr>
<td>Maximum acceptable humidity (% RH)</td>
<td>The recommended range is 50 percent to 65 percent.</td>
</tr>
<tr>
<td>Control humidity level during printing procedure</td>
<td>When this box is checked, the SpotArray automatically adds humid air when needed while a procedure is running.</td>
</tr>
<tr>
<td>Target humidity level (%RH)</td>
<td>The SpotArray attempts to keep the humidity at this level. The default is 60 percent. The best humidity level for your experiment may require empirical determination.</td>
</tr>
<tr>
<td><strong>Temperature</strong></td>
<td>Check this box to have the SpotArray system pause a printing procedure if the temperature goes outside the acceptable range, as set. When the temperature returns to the acceptable range, the SpotArray resumes the printing procedure. Do not check this box if your experiment does not depend on ambient temperature.</td>
</tr>
<tr>
<td>Do not print when temperature is outside of acceptable range</td>
<td></td>
</tr>
<tr>
<td>Minimum acceptable temperature (C)</td>
<td>Enter the minimum temperature and maximum temperature at which it is acceptable to print.</td>
</tr>
<tr>
<td>Maximum acceptable temperature (C)</td>
<td><em>Note:</em> the SpotArray will stop printing if the temperature is outside the specified range. To ensure uninterrupted printing, specify a temperature range wide enough to accommodate normal temperature fluctuations. The best temperatures for your experiment may require empirical determination.</td>
</tr>
</tbody>
</table>

3. Click **Next** or click “Finished Microarray” to go to the next window, or click **Finish** to save the printing protocol. Click **Cancel** if you do not want to save the information you just entered.
3.5.9 Viewing the Finished Microarray Layout

You can view the layout of the finished array that will be created using this printing protocol.

To view the layout of the finished microarray

1. Click Next, or click “Finished Microarray”. The Finished Microarray window opens. It may take a few seconds for this window to display.

This window displays the following information:

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microarray</td>
<td>A graphical representation of the microarray layout.</td>
</tr>
<tr>
<td>Selected subarray</td>
<td>A graphical representation of the subarray that you select with the mouse from the microarray layout.</td>
</tr>
<tr>
<td>Number of spots</td>
<td>The number of times each sample is printed.</td>
</tr>
<tr>
<td>Array</td>
<td>The number of the array.</td>
</tr>
<tr>
<td>Pin</td>
<td>The number (position) of the pin creating the spot.</td>
</tr>
<tr>
<td>Plate ID</td>
<td>The barcode of the plate from which the sample for the array was obtained.</td>
</tr>
<tr>
<td>Plate Name</td>
<td>The name of the sample plate.</td>
</tr>
<tr>
<td>Plate Description</td>
<td>The description of the plate as entered when the plate was defined.</td>
</tr>
</tbody>
</table>
2. Click **Finish** to save the printing protocol or click **Cancel** to close the printing protocol without saving your changes.

### 3.5.10 How Can You Change the Microarray?

If you want to change the finished microarray layout, you can go back to the *Array Layout* window and make changes before you click the **Finish** button.
Chapter 4

Preparing for Printing

Chapter Summary

Overview 4-1
 Handling the Pins and Printhead 4-1
 Maintaining the Pin Wash Water 4-6
 Checking the Connections 4-7
 Loading Substrates and Plates 4-8
 Handling the Plates 4-10

4.1 Overview

Correct handling and setup of the equipment are important for producing high-quality microarrays. Before running a printing procedure, perform the following preparations, using instructions in this chapter:

- Install pins, or check the position of installed pins
- Clean the pins if necessary
- Check the wash water level
- Check the waste level
- Check the (optional) humidity water level
- Make sure hoses and cables have been re-connected and check the hoses for kinks
- Load the required substrates and sample plates for the protocol

4.2 Handling the Pins and Printhead

The spotting pins, if handled properly, can create millions of spots before wearing out. Follow the handling instructions in this section.

4.2.1 Pins and Printhead

The SpotArray uses contact printing, meaning that when the tip of the pin touches the substrate surface, a small volume of sample coating the tip of the pin leaves a spot on the substrate. The pins are set with a precise slip-fit in the printhead, allowing them to move freely up and down in the printhead upon contact with the substrate during printing.

Note: Always wear powder-free nitrile gloves when handling the pins and printhead. Oil from your fingertips and hands will cause corrosion on the brass printhead, and can cause sample or contaminants to stick to the pins. Finger oil on the pin shafts will interfere with their free sliding in the printhead.
The pins are very fragile. To protect the pins and ensure they continue to move freely in the printhead, the pins and printhead must be handled properly and kept clean.

- Do not drag the pin tips along the printhead when locating the pinhole during installation, as this may bend the pin tips.
- The reservoir-tip pins have very small slots that are susceptible to clogging from dust, dried sample, particulates, salts or other contaminants.
- Handle the pins as little as possible; if they are printing satisfactorily, leave them in the printhead.
- Always store the pins in a protective sleeve or holder when they are not in the printhead.
- Do not allow sample to dry in the pins.

4.2.2 Pin Configurations

The maximum number of pins are:

- 4 x 12 with 4.5 mm spacing (384-well plates)
  - supporting pin configurations of 1, 4, 16, 32, and 48
- 2 x 6 with 9 mm spacing (96-well plates)
  - supporting pin configurations of 1, 4, 8, and 12

For any pin configuration, you must install a pin in position A1; position A1 in the printhead corresponds to position A1 in the plate. Make sure the plate is installed in the correct orientation (not backwards) to prevent mis-identified spots in the microarray.

If pin lifing is used, each zone of the printhead must have the same pin configuration. For more information, see “Selecting Pin” in Chapter 3.

4.2.2.1 Installing Pins

To install pins

1. Close the instrument door.

2. On the SpotArray Main Window, click Change Pins to bring the printhead into position at the front of the instrument for installing or changing pins. Open the instrument door.

3. If the pin-lifting option is installed, flip up the metal lid that protects the pins in the printhead.
4. Wearing powder-free nitrile gloves, remove a pin from its protective sleeve, and gently drop it into position A1 in the printhead. All pin configurations require a pin in position A1.

**Warning:** Be careful not to drag the pin tips along the printhead surface while locating the pinhole. Dragging will bend the pin tips.

Check the pin orientation to ensure that the pin is fully inserted into the pinhole - the pinhead is slightly rectangular, requiring it to be oriented correctly to slip completely into the printhead.

5. If you are printing with additional pins, place each additional pin in one of the defined positions for your configuration. Note that when printing from 96-well plates, you will place a pin in every other pinhole.

6. Close the instrument door.

7. Click **Resume** to return the printhead to the rear of the instrument.

### 4.2.2.2 Removing Pins

**To remove a pin**

1. On the **Main Window**, with the instrument door closed, click **Change Pins** to bring the printhead into position for removing or changing pins. Open the instrument door.
2. Wearing powder-free nitrile gloves, use a glass slide to gently push up the pin(s) you want to remove.

3. When the pin head clears the printhead, grasp the pin head and carefully pull the pin straight out to clear the printhead without dragging the pin. You may remove the pins using your fingers or a pair of tweezers.

4. To store the pin, replace it in its protective sleeve.

5. Close the instrument door and click Resume to return the printhead to the rear of the instrument.

4.2.2.3 Cleaning pins

Use the SpotArray washer/dryer to wash the pins, outside of a protocol, before and after changing pins.

⚠️ **Warning:** Wash the pins thoroughly to prevent sample carry-over. Do not allow sample to dry in the pins.

If the pins are performing satisfactorily, leave them in the printhead. If the pins or printhead require additional inspection and cleaning, refer to Chapter 5, “Periodic Maintenance,” for instructions.
To clean pins

1. On the Main Window, click **Wash & Dry Pins**.

![Wash & Dry Pins](image)

2. The **Wash & Dry Pins** window opens. Specify how many wash cycles and the length of the dry cycle.

![Wash & Dry Pins settings](image)

3. Specify how long, in seconds, for the wash jets to spray the pins. Valid values range from 0.5 to 10.0.

4. Specify how long, in seconds, for the dryer vacuum to be applied to the pins after washing. Valid values range from 3 to 20.

5. Specify the number of times to wash the pins.

6. Click **Start** to clean the pins. The SpotArray performs a wash and dry procedure. The following status message displays:

![Instrument Status](image)
4.3 Maintaining the Pin Wash Water

Rinse and fill the wash tank.

1. Disconnect the vacuum hoses and wash water hose from the reservoir.

2. Disconnect the water sensor cable.

3. Wheel the reservoir to the fill/drain area and open the drain port until all waste water is drained.

4. Rinse the tank, close the drain port, and fill the water supply tank with distilled water.

5. Wheel the reservoir back to the instrument and re-connect the labeled hoses and the water level sensors.
4.4 Checking the Connections

Visually check the hoses to ensure they have been re-connected after filling/emptying the reservoirs, don’t have water in them, or don’t have kinks in them that might impede flow. Refer to the following illustration.
4.5 Loading Substrates and Plates

You can load up to 4 pre-printing (blotting) substrates; up to 20 printing substrates for the SpotArray 24 or up to 68 printing substrates for the SpotArray 72; and up to 4 sample plates. Load any number of substrates and plates that you need, and the SpotArray detects the number and location.

**Note: Always** wear powder-free nitrile gloves when handling the substrates and plates.

The SpotArray supports glass substrates within the ISO 8037/1 microscope slide dimensional specifications, as listed below, and accommodates substrates with corner chamfers up to 2mm.

<table>
<thead>
<tr>
<th>Dimension</th>
<th>Specification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Length</td>
<td>75.0 mm to 76.2 mm (2.95” x 3.00”) excluding corner features</td>
</tr>
<tr>
<td>Width</td>
<td>24.6 mm to 25.5 mm (0.970” x 1.02”) excluding corner features</td>
</tr>
<tr>
<td>Thickness</td>
<td>0.93 mm to 1.29 mm (0.037” x 0.051”)</td>
</tr>
<tr>
<td>Corner feature</td>
<td>acceptable</td>
</tr>
<tr>
<td>Bevel</td>
<td>acceptable</td>
</tr>
<tr>
<td>Flatness Variation</td>
<td>The flatness variation will directly affect the amount of overtravel needed and effective dwell time when printing.</td>
</tr>
</tbody>
</table>

4.5.1 Loading Substrates

Refer to the figure on the next page.

**To load a pre-print slide or printing substrate**

1. Orient the slide or substrate in portrait mode (from the front of the instrument). If a barcode is affixed to the substrate or the substrate has a thumb area (frosted end), it should be face up on the end toward the front of the instrument.

2. When you begin the printing procedure, flexure spring clips close automatically to hold the slides in place.

4.5.2 Loading Plates

Plates are nominally 127.76 mm x 85.47 mm x 14.35 mm and are made of rigid polystyrene or semi-rigid polypropylene. Polypropylene is recommended because of its hydrophobic properties. For a list of supported plates, see Chapter 3.

Up to four plates can be accommodated between plate changes. Refer to the figure on the next page.
To load a plate

1. Place the plate into the plate holder in portrait orientation. Well A1 is toward the front of the instrument and Well H12 (96-well) or Well P24 (384-well) is toward the rear. Backward placement leads to mis-identified spots on the microarray.

2. Ensure that the two rear edges of the plates are located positively against dowel pins and press the front of the plate firmly down so that the flexure spring clips press against the other two edges.

Caution: Remove the lids from the plates, unless the lid-lifting option is installed and the Plates have lids checkbox is checked for the protocol.

Caution: When using lid lifting ...

1. Using lids that are tightly fitting, warped, or that stick to the surface of the plate will result in severe damage to your pins or sample materials. Make sure that the plate lids are easily lifted off the plates and are free from warping.

2. We recommend using a rigid polystyrene, low-profile universal lid such as a Corning 3098 or Nunc 240773.

3. Lids should be less than 7.6 mm in height.
4.5.3 Handling the Plates

The samples in microtiter plates are susceptible to evaporation; keep the plates covered except for the time during which they are being printed. Humidity control is recommended. If evaporation is of particular concern, you can load one plate at a time for a printing procedure, as specified in the printing protocol.

5.1 Overview

To keep SpotArray in top condition for printing high-quality microarrays, you need to perform some routine maintenance occasionally. This chapter offers guidelines; how often you need to perform some of the routine maintenance depends on the amount of usage.

In addition to maintaining the hardware, you should back up the database regularly.

5.2 Backing up the SpotArray Database

The SpotArray database is where your plate library and protocol definitions are stored. Make a copy of the SpotArray database regularly to ensure that, in the event that the SpotArray database becomes corrupted, a recent backup copy of the database will be available to restore.

5.2.1 Copying the Database

You can copy the database file to another location on your hard drive. If you have a workstation connected to the instrument, you can copy the database file to backup media, such as a ZIP disk, writable CD-ROM, or to a network location. Backing up to removable media or to another disk drive protects your information in the rare occurrence of disk failure.

To copy the database to another location

1. Exit the SpotArray user interface.
2. Click Start on the Windows task bar, point to Programs, point to Accessories, and click Windows Explorer.
3. In the left pane of the explorer window that opens, locate the database file: PackardBioChip\SpotArrayClient\V10\data\spotter.mdb
4. Hold down the Ctrl key, and drag the file to the backup location.

5.2.2 Restoring the Database

The Database can be restored as required. Locate the most recent .mdb file on your backup media.

To restore the database

5. Open the Windows Explorer and locate the backup database file: spotter.mdb

6. Drag the file to: C:\PackardBioChip\SpotArrayClient\V10\data\spotter.mdb

7. When a message displays asking if you want to replace the file with the backup file, answer Yes.

5.3 SpotArray Hardware Maintenance

Maintenance should be performed based on the amount of usage, or as needed. Regular maintenance includes the following:

- Before a procedure (see Chapter 4, Preparing for Printing)
- After a procedure
- As needed; for example, cleaning the pins and printhead
- Long-term storage

⚠️ Warning: Always use powder-free nitrile gloves when handling pins or the printhead. Do not touch the pins unless you have read “Handling the Pins and Printhead” in Chapter 4.

5.4 Inspecting and Cleaning Pins

If the normal wash and dry cycles are not sufficient to clean the pins, or if pins are clogged, they will need an ultrasonic cleaning. To do so, you must remove the pins. This section includes instructions from removing the pins, inspecting the pins, and cleaning the pins.

5.4.1 Removing or Changing Pins

Remove pins for cleaning only if necessary; handling pins should be kept to a minimum.

⚠️ Warning: Always wear powder-free nitrile gloves when handling the pins or printhead.
To remove a pin

1. Close the instrument door.

2. On the SpotArray Main Window, click **Change Pins** to bring the printhead into position at the front of the instrument for removing or changing pins.

3. Open the instrument door.

4. Flip up the metal lid on the printhead if the pin-lifting option is installed.

5. Wearing powder-free nitrile gloves, use a glass slide to gently push up the pin(s) you want to remove.

6. When the pin head clears the printhead, grasp the pin head with tweezers and carefully pull the pin straight out to clear the printhead without dragging the pin.

7. To store the pin, replace it in its protective sleeve.

8. Repeat for each pin to be removed.

9. Click **Resume** to return the printhead to the rear of the instrument.

To install new pins, see the following section; to inspect or clean the pins you have removed, see one of the following sections.

See **Inspecting the Pins** on page 5-4.

See **Cleaning the Pins** on page 5-6.

To install pins

1. Close the instrument door.

2. On the SpotArray Main Window, click **Change Pins** to bring the printhead into position at the front of the instrument for installing or changing pins.
3. Wearing powder-free nitrile gloves, remove a pin from its protective sleeve, and gently drop it into position A1 in the printhead. All pin configurations require a pin in position A1.

Check the pin orientation to ensure that the pin is fully inserted into the pinhole - the pinhead is slightly rectangular, requiring it to be oriented correctly to slip completely into the printhead.

4. If you are printing with additional pins, place each additional pin in one of the acceptable positions for your configuration. Note that when printing from a 96-well plate, you will place a pin in every other pinhole.

5. Close the instrument door. On the Main Window, click Resume to return the printhead to the rear of the instrument.

5.4.2 Inspecting the Pins

If a pin does not print correctly, inspect the tip under a microscope.

To inspect the pin tip

1. Remove the pin as described in section 5.4.1 above.
2. Examine the pin tip under a microscope. The slot reservoir should be clearly visible to the tip of the pin, and the pin tips should not touch.

A clogged pin, as shown below, must be cleaned:

A bent pin, as shown below, must be replaced:
5.4.3 Cleaning the Pins

If a loss of printing quality is observed, the spotting pins can be cleaned by following the procedures described below. To clean the pins, carefully remove them from the printhead, as described in the previous section.

Note: Always wear powder-free nitrile gloves during this procedure, and any time you handle the pins and printhead.

The pins can be cleaned by sonication in a bath containing a microarray cleaning solution, such as ArrayIt™ Micro Cleaning Solution from Telechem, followed by extensive rinsing with distilled water, and drying with clean forced air.

- During sonication, the pins must never come into direct contact with the sonicator. Direct contact between the pins and the sonicator can lead to permanent damage to the pins.
- The pins should never be left in microcleaning solution for longer than 10 minutes.
- The pins and the printhead should be stored and kept clean and dry between printing experiments. Prolonged storage of the pins or printhead in a wet environment can cause corrosion.

To clean the pins

1. Wearing powder-free nitrile gloves, remove the pins using instructions above. (See Cleaning the Pins on page 5-6.) Prepare a solution of 2 to 5 percent of Micro Cleaning Solution (MCS-1).

2. Suspend the pins in a sonication bath of the MCS-1 solution using the Microarray Pin Wash Station from TeleChem (see Figures 5-1 to 5-3 below).

Figure 5–1 Placing Pins Into the Pin Wash Station
3. Clean the pins in the bath for approximately 5 minutes (no longer than 10 minutes).

4. Rinse the pins in hot tap water, approximately 2 minutes, until the MCS-1 solution is completely washed away.

5. Sonicate for 3 minutes in distilled water.

**Note:** Never expose the pins to de-ionized water, which promotes corrosion. Use distilled or double-distilled water.

6. Blow dry the pins with forced air, using an oil-free compressor or a hair dryer.

**Warning:** Do not use air from a canister; the propellant will ruin the surface-tension properties of the pins.

7. Install the pins immediately into the printhead, or put the protective sleeves on the pins until you are ready to re-install them in the printhead.
5.5 Removing the Printhead

To remove the printhead

1. Close the instrument door, if it is not already closed. On the SpotArray Main Window, click Change Pins. The printhead moves to the change pins position.

2. If the pin-lifting option is installed on your system, lift the lid over the pins.

3. Wearing powder-free nitrile gloves, remove the pins using instructions in section 5.4.1 on page 5-2.

4. If the pin-lifting option is installed, remove the three “waffles” from the printhead.
5. Support the printhead with one hand. With the other hand, loosen one printhead mounting screw, and then the other.

6. When the screws are fully backed out, remove the printhead from the instrument and place it on the bench.

7. To clean the printhead, see section 5.6 on page 5-11.
5.5.0.1 Replacing the Printhead

To replace the printhead

1. Wearing powder-free nitrile gloves, hold the printhead in your left hand and check the printhead orientation -- there should be two mounting bumps away from you and one mounting bump toward you.

2. Place the printhead under the printhead bracket in the instrument.

3. Continue to hold the printhead with your left hand while you tighten the two thumbscrews with your right hand. *Tighten by hand only, do not use a tool.*
4. If the pin-lifting option is installed on your system, replace the “waffles” into the printhead. Be sure that the tabs on the waffles are facing to the left.

5. Close the instrument door. On the Main Window, click Resume to return the printhead to the rear of the instrument.

5.6 Cleaning the Printhead

The printhead is manufactured of specially polished brass and requires cleaning if it becomes contaminated by spilling or finger oil. In addition, any liquid splashed onto the printhead or pins will cause corrosion and may require immediate removal of the printhead for cleaning.

To protect the printhead and pins:

- Do not let liquids come into contact with the pin shaft or the printhead
- Never let sample or any aqueous liquid dry on the tips of the pins or the printhead

5.6.1 When Does the Printhead Need Cleaning?

The printhead should be cleaned under the following circumstances:

- If liquid has splashed the shafts of the pins, the pinholes where the pins rest in the printhead, or on the printhead.
- If the printhead has been handled without using gloves, or if pins that have finger oil on them have been installed.
- When any loss of performance is noticed, such as pins sticking in the up position.
5.6.2 How to Clean the Printhead

Occasionally, the pins do not move freely up and down in the printhead, resulting in DNA that does not print, referred to as “spot drop-out.” When this occurs, the printhead must be cleaned. In most cases only dust needs to be removed from the printhead. Less frequently, dulling of the brass printhead, caused by oxidation, must be removed. Refer to the appropriate instructions below for removing dust or oxidation.

**Warning:** Always use powder-free nitrile gloves then handling the pins and printhead.

**To remove dust from the printhead**

1. On the SpotArray Main Window, click **Change Pins** to bring the printhead into position at the front of the instrument for removing pins, and remove the pins as described in section 5.4.1 on page 5-2.

2. Remove the printhead as described in section 5.5 above.

3. Rinse the printhead thoroughly with ethanol. This operation should be done in a fume hood.

4. Air dry the printhead on a lint-free, absorbent material. If rapid drying is needed, use the warm air of a hair dryer. Do not use compressed (“clean”) air, as this will leave a hydrocarbon film on the printhead.

5. Replace the printhead.

6. On the Main Window, click **Resume** to return the printhead to the rear of the instrument.

**To clean oxidation from the printhead**

1. On the SpotArray Main Window, click **Change Pins** to bring the printhead into position at the front of the instrument for removing pins, and remove the pins as described in section 5.4.1 on page 5-2.

2. Remove the printhead as described in section 5.5 above.

3. Soak the printhead in 250 mL hot (hot as hot tap water, not boiling) de-ionized water supersaturated with oxalic acid. This operation should be done in a fume hood.

4. If brushing is required, use a soft bristle tooth brush.

5. Rinse thoroughly in de-ionized water.

6. Perform a final rinse with ethanol.

7. Air dry the printhead on a lint-free, absorbent material. If rapid drying is needed, use the warm air of a hair dryer. Do not use compressed (“clean”) air, as this will leave a hydrocarbon film on the printhead.
8. Replace the printhead.

9. On the Main Window, click Resume to return the printhead to the rear of the instrument.

### 5.6.3 Run the Test Print Protocol

Run a test of the printing capability before starting your printing procedure, using one of the test protocols for 96-well or 384-well plates provided with the SpotArray. Testing lets you check the pins, to be sure they are moving freely and printing as you intend for the microarray to be created. Load a single pre-print substrate, and load a test plate filled with buffer only in all wells; use the same buffer (for example 3x SSC) that carries the sample to be printed.

The SpotArray ships with sample plates, two test protocols and four demonstration protocols:

<table>
<thead>
<tr>
<th>Name</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Test 96-well plate</td>
<td>Test protocol for 1 96-well plate</td>
</tr>
<tr>
<td>Test 384-well plate</td>
<td>Test protocol for 1 384-well plate</td>
</tr>
<tr>
<td>Demo low density 96-well plate</td>
<td>Protocol for creating low density array from 1 96-well plate</td>
</tr>
<tr>
<td>Demo low density 384-well plate</td>
<td>Protocol for creating low density array from 1 384-well plate</td>
</tr>
<tr>
<td>Demo 96-well with 4 pins and 1 96-well plate</td>
<td>Startup protocol with 4 pins and 1 96-well plate</td>
</tr>
<tr>
<td>Demo 384-well with 4 pins and 1 384-well plate</td>
<td>Startup protocol with 4 pins and 1 384-well plate.</td>
</tr>
</tbody>
</table>

Sample plates have gene information that can be used for verifying printing and the generation of array content files, for example:

- Well A1 contains Gene A1
- Well B2 contains Gene B2
When you receive a new instrument, carefully examine the shipment as soon as possible. If any problems are noted, they must be reported to PerkinElmer and the shipping agent within ten days of receipt of the instrument.

1 Return Materials Authorization

In the unlikely event that it is necessary to return the instrument, obtain the packing list or invoice and contact PerkinElmer to obtain a Return Materials Authorization (RMA) number.

Contact information:

- Customer Service by telephone (888) 323-9681 or 978-439-8410
- Technical Support by e-mail: PBTsupport@perkinelmer.com

Please be prepared with the following when calling:

- serial number of your instrument
- version number of the application software
- nature of the problem
- steps you have taken
- access to your instrument
- software log files
- your phone number, fax number, and e-mail address

Write the RMA number on the Return Materials Authorization Form (which is shipped with the instrument) and attach the form to the outside of the box containing the returned items. This number is used to track the returned equipment and enables PerkinElmer to expedite the process.

2 Repacking for Return

Please contact your local PerkinElmer service representative. Your service representative will pack the equipment for return.
The SpotArray system includes a number of sensors that are monitored by the SpotArray software to ensure proper operation.

1 Operation of the SpotArray Sensors

Instrument Sensors

The sensor status is checked on startup, and at regular intervals during operation. The sensors are described in Table 1.

### Table B-1: SpotArray Sensors

<table>
<thead>
<tr>
<th>Sensor</th>
<th>Description</th>
<th>Sensor Behavior &amp; the SpotArray Response to Sensor Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Door</td>
<td>Identifies whether or not the instrument door is open. Status: <strong>open</strong> or <strong>closed</strong></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Main door open</td>
<td>Checks once every second.</td>
</tr>
<tr>
<td></td>
<td>Main door closed</td>
<td></td>
</tr>
<tr>
<td></td>
<td>If <strong>open</strong>, the SpotArray</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• does not perform actions that move the printhead, including diagnostic tests, printing, cleaning pins or loading pins.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>If <strong>closed</strong>, the SpotArray</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• displays the status on the instrument LCD and user interface</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• performs self-diagnostics</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• waits for a request for an operation</td>
<td></td>
</tr>
</tbody>
</table>
### Appendix B The SpotArray Sensors

**Wash Fluid Availability**

Identifies whether or not there is enough fluid in the reservoir to process 4 plates.

<table>
<thead>
<tr>
<th>Status</th>
<th>Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>OK</td>
<td>The SpotArray accepts requests to run or resume a printing procedure or clean pins.</td>
</tr>
<tr>
<td>low</td>
<td>The SpotArray does not accept requests to clean pins, or perform/resume printing procedures.</td>
</tr>
<tr>
<td>sensor not connected</td>
<td>The SpotArray takes no action on sensor status changes.</td>
</tr>
</tbody>
</table>

**Check the reservoir level and cable connection.**

**Wash Pump**

Identifies whether or not the water pump for the washer is operational.

<table>
<thead>
<tr>
<th>Status</th>
<th>Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>OK or failed</td>
<td>The SpotArray accepts requests to perform or resume a printing procedure, or wash pins.</td>
</tr>
</tbody>
</table>

If failed, the SpotArray does not accept requests to perform or resume a printing procedure, or wash pins.

If the wash pump fails during a procedure, the SpotArray displays the status on the instrument LCD and user interface. The SpotArray terminates the printing procedure.

Check the hose connections, check for kinks in the hoses or a clogged filter; check the reservoir level.
<table>
<thead>
<tr>
<th>Sensor</th>
<th>Description</th>
<th>Sensor Behavior &amp; the SpotArray Response to Sensor Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wash Fluid Pressure</td>
<td>Identifies whether or not there is sufficient pressure to operate the washer.</td>
<td>Checks during diagnostic self-tests, when verifying instrument configuration, and each time the pins are washed.</td>
</tr>
<tr>
<td></td>
<td>Status: <strong>OK</strong> or low</td>
<td>If <strong>OK</strong>, the SpotArray</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• accepts requests to perform or resume a printing procedure, or clean pins</td>
</tr>
<tr>
<td></td>
<td></td>
<td>When <strong>low</strong>, the SpotArray</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• does not accept requests to perform a printing procedure and clean pins</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• if pressure becomes low during a printing procedure, the SpotArray finishes the wash cycle, cancels the procedure, displays the status on the LCD and user interface, and unlocks the instrument door.</td>
</tr>
<tr>
<td>Waste Fluid Space Availability</td>
<td>Identifies whether or not there is enough space left in the waste fluid container to process four plates.</td>
<td>Checks every 5 seconds</td>
</tr>
<tr>
<td></td>
<td>Status: <strong>OK</strong> container too full capacity not monitored</td>
<td>The sensor must be connected, and indicate that space is available before the SpotArray can start a printing procedure, resume a paused procedure, or clean pins.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>If the capacity is not monitored, the SpotArray</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• ignores status changes</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• accepts requests to clean pins</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• starts and resume paused procedures</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A warning message displays on the user interface that the waste fluid sensor is disabled.</td>
</tr>
<tr>
<td>Dryer Vacuum</td>
<td>Identifies whether or not there is sufficient vacuum to operate the pin dryer.</td>
<td>Checks during diagnostic self-tests, when verifying the instrument configuration, and each time pins are dried.</td>
</tr>
<tr>
<td></td>
<td>Status: <strong>OK</strong> or low</td>
<td>When <strong>low</strong>, the SpotArray</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• does not accept requests to perform a printing procedure or clean pins.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• if the pin dryer vacuum fails during a printing procedure, the instrument cancels the procedure, displays a message on the instrument LCD and user interface, and unlocks the instrument door.</td>
</tr>
</tbody>
</table>
### Sensor Description

<table>
<thead>
<tr>
<th>Sensor</th>
<th>Description</th>
<th>Sensor Behavior &amp; the SpotArray Response to Sensor Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Humidity Generator Fluid Availability (Optional Feature)</td>
<td>Identifies whether or not there is enough fluid left in the container used by the humidity generator. Status: OK Fluid too low Fluid availability not monitored</td>
<td>Checks every 5 seconds. If OK, the SpotArray • accepts requests to perform or resume a printing procedure or clean pins. If low, the SpotArray • does not accept requests to start or resume a printing procedure. • turns off the humidity generator if it is on. If not monitored, the SpotArray • takes no action on sensor status change. • starts or resumes a printing procedure regardless of the sensor status. A warning message displays on the user interface that the humidity fluid level is not monitored.</td>
</tr>
<tr>
<td>Humidity Level</td>
<td>Indicates the current humidity level within the instrument enclosure. Status: Humidity level acceptable Humidity level high Humidity level low Waiting to start... humidity level low Waiting to start... humidity level high Waiting to resume... humidity level low Waiting to resume... humidity level high</td>
<td>Checks every 5 seconds. You can specify in a protocol that the SpotArray print only when the humidity is within an acceptable range. See the following section, “Using The Optional Environmental Module.”</td>
</tr>
</tbody>
</table>

Check the reservoir level and cable connection.
Using the Optional Environmental Module

The humidity and temperature sensors are part of the SpotArray base system. The ability to raise the humidity level above ambient is part of the optional environmental control module.

You may specify in a printing protocol that the SpotArray should print only when the humidity and temperature are within the specified ranges. See “Creating a Protocol” in Chapter 3. When this feature is enabled in the printing protocol and either humidity or temperature goes out of range, the SpotArray automatically pauses the procedure, and waits for the conditions to return to within range; if the humidity becomes too low, the SpotArray automatically generates humidity if the environmental control module is installed.

<table>
<thead>
<tr>
<th>Sensor</th>
<th>Description</th>
<th>Sensor Behavior &amp; the SpotArray Response to Sensor Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temperature</td>
<td>Indicates the current temperature within the instrument enclosure.</td>
<td>Checks every 5 seconds. You can specify in a protocol that the SpotArray print only when the temperature is within an acceptable range. See the following section, “Using Optional Environmental Module.”</td>
</tr>
<tr>
<td></td>
<td>Status:</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Temperate acceptable</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Temperature high</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Temperature low</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Waiting to start... temperature low</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Waiting to resume... temperature high</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Waiting to start... temperature high</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Waiting to resume... temperature low</td>
<td></td>
</tr>
<tr>
<td>Filtered Air Supply (Optional Feature)</td>
<td>Indicates the delivery of filtered air to the instrument enclosure.</td>
<td>Checks every 5 seconds while the instrument door is closed. Note: If the air filtration system fails during a printing procedure, the SpotArray completes the current printing cycle and pauses the printing procedure. When the air filtration resumes operation, the SpotArray automatically resumes the paused procedure, unless there are other reasons for the pause.</td>
</tr>
<tr>
<td></td>
<td>Status:</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Air filtration operational</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Air filtration not operational</td>
<td></td>
</tr>
</tbody>
</table>

Check the hose connections; check for kinks and a clogged filter.
Humidity Sensor

If the humidity is too low, the SpotArray will start or resume a printing procedure, unless the printing protocol specifies to print only when the humidity is within the acceptable range. In that case, the SpotArray pauses the procedure and waits for the humidity level to rise to within the acceptable range.

If the humidity level is too high, the SpotArray will start or resume a printing procedure, unless the printing protocol specifies to print only when the humidity is within the acceptable range. In that case, the SpotArray pauses the procedure and waits for the humidity level to drop to within the acceptable range.

When a protocol specifies to print only when the humidity level is within the acceptable range, and the humidity levels goes outside the range during a printing procedure, the SpotArray:

• pauses the printing procedure
• displays the status on the instrument LCD and user interface
• waits for the humidity level to be within the acceptable range
• resumes the procedure when the humidity level is again within the acceptable range (if there are no other causes for the pause).

Temperature Sensor

If the temperature becomes too low, the SpotArray will start or resume a printing procedure, unless the printing protocol specifies to print only when the temperature is within the acceptable range. In that case, the SpotArray pauses the procedure and waits for the temperature to return to within the acceptable range.

If the printing protocol being used specifies to print only while the temperature is within range, the instrument:

• pauses the printing procedure
• displays the status on the instrument LCD and user interface.
• waits for the temperature to be within the acceptable range.

If the temperature becomes too high, the SpotArray will start or resume a printing procedure, unless the printing protocol specifies to print only when the temperature is within the acceptable range. In that case, the SpotArray pauses the procedure and waits for the temperature to drop to within the acceptable range.

If the printing protocol being used specifies to print only while the temperature is within range, the instrument:

• pauses the printing procedure
• displays the status on the instrument LCD and user interface
• waits for the temperature to be within the acceptable range.

If the temperatures goes out of range while running a printing procedure using a protocol that specifies to print only when the temperature is within range, the instrument:
• pauses the printing procedure
• displays the status on the instrument LCD and user interface.
• waits for the temperature to be within the acceptable range
• resumes the procedure when the temperature is again within the acceptable range (if there are not other causes for the pause)
2 Disabling the Fluid Level Sensors

The fluid level sensors include the wash fluid level and waste fluid level sensors for the pin washer, and the humidity fluid level sensor for the optional humidity module.

If one of these sensors detects a problem, the associated button on the Main Window turns red. In Figure B-1, the Pin Washer button indicates a problem with the pin washer. You can click the button to get more information.

The SpotArray will not begin or resume a printing procedure if one of the fluid level sensors indicate a problem. Check the fluid levels; replenish the wash fluid, empty the waste fluid, or replenish the humidity generation fluid, as required.

If the fluid levels are okay (verified by a visual check), and the sensor itself is not functioning, you can override the sensor in order to continue printing. When a sensor is disabled, the SpotArray ignores the sensor; the user interface does not reflect the current sensor reading, and the printing procedures run, regardless of the current sensor reading.

**Warning:** If you disable the pin washer sensors, you must make certain during the printing procedure that there is enough wash water, and enough room for waste water, to process the plates for your procedure. Pins may not be washed if there is not enough wash water, and could result in contaminated samples or contaminated microarrays.

If the optional environmental control module is installed, and you disable the sensor for the humidity fluid level, you must make certain that there is enough water to generate humidity as required.
To disable a pin wash fluid level sensor

1. On the Main Window, click the Pin Washer button. The Pin Washer dialog box opens.

2. Click the Wash Fluid Level button to disable the wash sensor.

![Pin Washer dialog box]

To disable the humidity fluid level sensor

1. On the Main Window, click the Humidity Fluid Level button.

2. The SpotArray displays a warning message. Click Yes to disable the sensor, click No if you do not want to disable the sensor.
To re-enable a sensor

1. On the Main Window, click the button for the sensor you want to re-enable: the Pin Washer button or the Humidity Fluid Level button.

2. When the sensor dialog box opens, click the button for the sensor you want to re-enable. At the Re-enabling Sensor prompt, click Yes to re-enable the sensor or click No to leave the sensor disabled.
Appendix Summary

This system has been tested to the Laboratory EMC standard, EN61326-1. Electrostatic caution should be used during operation.

This instrument is designed and certified to meet the following regulatory and safety standards.

1 Electrical and Mechanical Safety Standards

The product is CSA listed to the following standard:
CAN/CSA C22.2 No. 1010.1 with A1:92+A2:95
Electrical Equipment for Laboratory Use

2 Electromagnetic Emissions Standards

FCC Part 15 Class A, Radiated and Conducted (USA)
EN55011:1991 Class A, Radiated and Conducted (Europe)
ICES-003, Industry Canada, Interference-Causing Equipment Standard, Digital Apparatus, Class A

3 Electromagnetic Immunity Standards

EN50082-1, Electromagnetic Compatibility- Generic Immunity Standard, Part 1, Residential, Commercial, and Light Industry
ED61000-4-2, Electrostatic Discharge
EN61000-4-3, Radiated Electromagnetic Fields
EN61000-4-4, Electrical Fast Transient/Burst
EN61000-4-5, Surge Immunity Requirements
EN61000-4-6, Conducted Disturbances Induced by Radio-Frequency Fields
EN61000-4-11, Voltage Dips, Short Interruptions and Voltage Variation Immunity Tests
4 **FCC Label for Class A Products**

This device complies with Part 15 of the FCC Rules. Operation is subject to the following two conditions:

- This device may not cause harmful interference,
- This device must accept any interference received, including interference that may cause undesired operation.

5 **ICES-003 Label for Class A Products**

This Class A digital apparatus meets all requirements for the Canadian Interference-Causing Regulations.

Cet appareil numérique de la classe A respecte toutes les exigences du Reglement sur le matériel brouiller du Canadas.

6 **Other EU Conformance**

The product is CE marked in conformance with the following directives:


---

**Note:** Declaration of Conformity is on file and may be obtained at:

PerkinElmer Life Sciences  
40 Linnell Circle  
Billerica, MA 01821, USA
Appendix Summary

Overview D-1
QuantArray Text Format D-1
Gene Array List (.gal) Format D-5

1 Overview

The SpotArray creates array content files after a printing procedure or from any protocol as requested from the Main Window. Two formats are supported: QuantArray Text and Gene Array List (.gal). By default, the SpotArray generates QuantArray Text files.

2 QuantArray Text Format

The QuantArray Text format is written to an ASCII text file that is viewable by applications such as Microsoft® Notepad. The file contains one text line for each spot in the microarray. Each line contains tab-delimited fields in the order described in Table D-1.

2.1 Description of File Format

<table>
<thead>
<tr>
<th>Field</th>
<th>Field Description</th>
<th>Field Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Row number of the subarray</td>
<td>Numeric</td>
</tr>
<tr>
<td>2</td>
<td>Column number of the subarray</td>
<td>Numeric</td>
</tr>
<tr>
<td>3</td>
<td>Row where the spot is located within a subarray</td>
<td>Numeric</td>
</tr>
<tr>
<td>4</td>
<td>Column where the spot is located within a subarray</td>
<td>Numeric</td>
</tr>
<tr>
<td>5</td>
<td>Gene descriptor (a user assigned name or number)</td>
<td>Alphanumeric</td>
</tr>
<tr>
<td>6</td>
<td>Comment starting with the ‘#’ character</td>
<td>Alphanumeric</td>
</tr>
</tbody>
</table>
2.2 Subarrays

A subarray is the set of spots printed by one pin within a 4.5 mm x 4.5 mm square or 9 mm x 9 mm square area. Subarrays are numbered from top left to bottom right. For example, a microarray created by 4 pins would minimally result in a microarray with 4 subarrays that have the row and column numbers as shown:

Spots within a subarray are identified by the same numbering scheme. For example, the spots in a subarray with 16 spots in a 4 by 4 pattern would have the following row and column numbers:
2.3 Example QuantArray Array Text File

The following example is a QuantArray Content File that describes a microarray containing four subarrays (2 x 2), with 16 spots (4 x 4) in each subarray.

```
1 1 1 1- # -EST -91 Control (nonhuman) clone.
1 1 1 2HSCG8S08 # X54937mRNA for cannabinoid receptor.
1 1 1 3HSU14969 # U14969Ribosomal protein L28.
1 1 1 4HSU58046 # U58046p167.
1 1 2 1- # -EST-35 Control (nonhuman) clone.
1 1 2 2578986 # S78986"Id1 (Id1-a), transcription regulator helix-loop-helix protein."
1 1 2 3HUMGPIB # J03259Platelet glycoprotein Ib beta chain.
1 1 2 4HSRAFR # X03484mRNA for raf oncogene.
1 1 3 1- # -EST-34 Control (nonhuman) clone.
1 1 3 2HSPTBMR # X60648Polypyrimidine tract-binding (PTB)
mRNA for polypyrimidine tract-binding protein (pPTB).
1 1 3 3AB007877 # AB007877KIAA0417.
1 1 3 4D79986 # D79986mRNA for KIAA0164 gene.
1 1 4 1HSHMG1 # X12597mRNA for high mobility group-1 protein (HMG-1).
1 1 4 2HSVACM1 # X81882mRNA for vasopressin activated calcium mobilizing receptor-like protein.
1 1 4 3HUMN8220 # D83032"mRNA for nuclear protein, NP220."
1 1 4 4HSU06863 # U06863Follistatin-related protein precursor.
1 2 1 1- # -EST -91 Control (nonhuman) clone.
1 2 1 2HSPXF # X75535mRNA for PxF protein.
1 2 1 3HSU65093 # U65093msg1-related gene 1 (mrg1).
1 2 1 4HSU20489 # U20489Glomerular epithelial protein 1 (GLEPP1).
1 2 2 1- # -EST-35 Control (nonhuman) clone.
1 2 2 2AF047347 # AF047347Adaptor protein X11alpha.
1 2 2 3HSU65093 # U65093msg1-related gene 1 (mrg1).
1 2 2 4HSU20489 # U20489Glomerular epithelial protein 1 (GLEPP1).
1 2 3 1- # -EST-34 Control (nonhuman clone).
1 2 3 2D14041 # D14041mRNA for H-2K binding factor-2.
1 2 3 3HSU09413 # U09413Zinc finger protein ZNF135.
1 2 3 4HSPKCB2A # X07109mRNA for protein kinase C (PKC)
type beta II.
1 2 4 1AF047470 # AF047470"Malate dehydrogenase precursor (MDH) mRNA, nuclear gene encoding mitochondrial protein."
1 2 4 2HUMFKBP25A # M90820Rapamycin-binding protein (FKBP25).
1 2 4 3HUMIDNAL # M74715alpha-L-iduronidas (IDUA).
1 2 4 4HUMGCSL # L35546Gamma-glutamylcysteine synthetase light subunit.
2 1 1 1- # -EST -91 Control (nonhuman) clone.
2 1 1 2HUMASH1 # L08424Achaete scute homologous protein (ASH1).
```
2 1 1 3HSU09303 # U09303T cell leukemia LERK-2 (EPLG2).
2 1 1 4D89289 # D89289mRNA for N-Acetyl-beta-D-glucosaminide.
2 1 2 1- # -EST-35 Control (nonhuman) clone.
2 1 2 2U01244 # U01244fibulin-1D.
2 1 2 3AF026844 # AF026844Ribosomal protein L41.
2 1 2 4HUMMUPCAD # D78586CAD mRNA for multifunctional protein CAD.
2 1 3 1- # -EST-34 Control (nonhuman) clone.
2 1 3 2HS1054RNA # X57346mRNA for HS1 protein.
2 1 3 3HUMBGR1A # M81886Glutamate receptor type 1 (HBGR1).
2 1 3 4HSHK2 # Y08319mRNA for kinesin-2.
2 1 4 1AF006621 # AF006621Embryonic lung protein (HUEL).
2 1 4 2D49958 # D49958Fetus brain mRNA for membrane glycoprotein M6.
2 1 4 3HUMVBICE # D45050mRNA for ubiquitin conjugating enzyme.
2 2 1 1- # -EST-91 Control (nonhuman) clone.
2 2 1 2HSU97188 # U97188Putative RNA binding protein KOC (koc).
2 2 1 3HUMBIGFII # M35410Insulin-like growth factor binding protein 2 (IGFBP2).
2 2 2 1- # -EST-35 Control (nonhuman) clone.
2 2 2 2HSSRIP140 # X84373mRNA for nuclear factor RIP140.
2 2 2 3AF036270 # AF036270EEN-B2-L2.
2 2 2 4AF039136 # AF039136Fas binding protein (hDaxx).
2 2 3 1- # -EST-34 Control (nonhuman) clone.
2 2 3 2HSRNABS69 # X86098mRNA for BS69 protein.
2 2 3 3HSGPX4 # X71973GPx-4 mRNA for phospholipid hydroperoxide glutathione peroxidase.
2 2 3 4HSU96136 # U96136Delta-catenin.
2 2 4 1D87343 # D87343mRNA for DCRA.
2 2 4 2HSAP001435 # AF001435Clone iota unknown protein.
2 2 4 3HSU16127 # U16127Glutamate/kainate receptor subunit (EAA5).
2 2 4 4AB002334 # AB002334mRNA for KIAA0336 gene.
3 Gene Array List (.gal) Format

The gene array list format is written to an ASCII text file that is viewable by applications such as Microsoft® Notepad and Wordpad. The file contains header information and information for each spot in the microarray.

GAL files conform to the ATF format, a standard tab-delimited text file format readable by many downstream scanning and quantitation software applications. They can be created in Microsoft Excel by saving an Excel spreadsheet as Text (Tab delimited).

3.1 Description of File Format

Gene array list files contain two major sections: the header section and the spot data section.

3.1.1 Description of the Header Section

The header section describes basic file information and provides information about each of the blocks. Each line is explained in Table D-2:

<table>
<thead>
<tr>
<th>Record</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Required Lines</strong></td>
<td></td>
</tr>
<tr>
<td>ATF 1.0</td>
<td>First line of an ATF file; the same in all GAL files: File format (ATF) and version (1.0)</td>
</tr>
<tr>
<td>8 5</td>
<td>Second line of an ATF file: 8 (number of optional header lines plus 1). 5 (number of spot data columns).</td>
</tr>
<tr>
<td>&quot;Type=GenePix Array List v1.0&quot;</td>
<td>Type of file; the same in all GAL files.</td>
</tr>
<tr>
<td><strong>Optional Lines</strong></td>
<td></td>
</tr>
<tr>
<td>&quot;BlockCount=4&quot;</td>
<td>Number of blocks described in the file, 4 in this example.</td>
</tr>
<tr>
<td>&quot;BlockSize=0&quot;</td>
<td>Type of block described, rectangular in this example. 0 = rectangular. 1 = orange-packing #1. 2 = orange-packing #2.</td>
</tr>
<tr>
<td>&quot;URL=...&quot;</td>
<td>The URL for the Go To Web command.</td>
</tr>
<tr>
<td>&quot;Supplier=PerkinElmer Life Sciences&quot;</td>
<td>The manufacturer that supplied the array or arrayer.</td>
</tr>
<tr>
<td>&quot;ArrayerSoftwareName=SpotArray&quot;</td>
<td>The name of the arrayer software.</td>
</tr>
<tr>
<td>&quot;ArrayerSoftwareVersion=2.0&quot;</td>
<td>The version number of the arrayer software.</td>
</tr>
</tbody>
</table>
3.1.2 Description of the Spot Data Section

The spot data section contains lines that describe each spot in detail. It includes the block, column, and row numbers for spots, as well as descriptive name and identifier information.

There is one text line for each spot in the microarray, containing a field for each of the descriptive columns. Each line contains tab-delimited fields in the order described in Table D-3.

### Table D-3: Spot Data

<table>
<thead>
<tr>
<th>Field</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Block</td>
<td>Block number for the spot.</td>
</tr>
<tr>
<td>Column</td>
<td>Column where the spot is located within a block.</td>
</tr>
<tr>
<td>Row</td>
<td>Row where the spot is located within a block.</td>
</tr>
<tr>
<td>Name</td>
<td>Gene name (limited to 40 characters)</td>
</tr>
<tr>
<td>ID</td>
<td>Gene identifier (limited to 40 characters)</td>
</tr>
</tbody>
</table>

Note: Positions on arrays are measured in microns with respect to the origin, which is the top left corner of the slide.
3.2 Blocks (Subarrays)

A block is a set of spots printed by one pin within a 4.5 mm x 4.5 mm square or a 9 mm x 9 mm square area. Blocks are also called subarrays. Blocks are numbered from top left to bottom right. For example:

```
Spot 1,1  Spot 1,2  Spot 1,3  Spot 1,4
Spot 2,1  Spot 2,2  Spot 2,3  Spot 2,4
Spot 3,1  Spot 3,2  Spot 3,3  Spot 3,4
Spot 4,1  Spot 4,2  Spot 4,3  Spot 4,4
```
3.3 Example GAL file

The following simple array list file describes four blocks ("BlockCount=4"), each with 24 columns and 5 rows. For simplicity, the data record information (name, ID, etc.) is included only for the first two spots:

ATF    1.0
8      5
"Type=GenePix ArrayList V1.0"
"BlockCount=4"
"BlockType=0"
"URL=http://www.perkinelmer.com/microarray/
"Supplier=PerkinElmer Life Sciences"
"ArrayerSoftwareName=SpotArray"
"ArraySoftwareVersion=1.0"
"Block1= 400, 400, 100, 24, 175, 5, 175"
"Block2= 4896, 400, 100, 24, 175, 5, 175"
"Block3= 400, 4896, 100, 24, 175, 5, 175"
"Block4= 4896, 4896, 100, 24, 175, 5, 175"
"Block" "Column" "Row" "Name" "ID"
1     1     1    VPS8    YAL002W
1     2     1    NTG1    YAL015C
Appendix E

Plate Contents Files

Appendix Summary
Overview E-1
Plate Contents File Format E-1
Creating a Plate Contents File E-3

1 Overview

The SpotArray uses the information in the *plate contents* files to create plate definitions in the SpotArray database. These plate definitions are then used by printing protocols to create array content files (.txt or .gal files) that identify each spot printed in the final microarray. The array content files are used at image quantitation after scanning.

You can import plate well data into the SpotArray from ASCII comma-delimited files (.txt or .csv files) as described in Chapter 3, section 3.2.1. You can also easily create plate contents files with applications such as Microsoft® Notepad or Microsoft® Excel by cutting and pasting gene list information into a structured file format. The format required for a plate contents file, and instructions for creating a file are described below.

2 Plate Contents File Format

A plate contents file consists of:

- two lines of header information describing the entire plate, followed by
- a line of data for each well or sample in the plate.

A description of each line in the file is given below. Generally, a comma is required at the end of each line. *Do not use any spaces or tabs after the commas within a line.*

2.1 First Line of Header Information

The first header line is as follows:

```
Product,Version,“Platecontent,”
```

Where:

- *Product* is the name of the instrument or software using this plate contents file.
- *Version* is the software version number.
- *Platecontent,* is a required label, or text string, at the end of the line.
2.2 Second Line of Header Information

The second header line is as follows:

“Plate”, Plate Name, Plate Barcode, Description, No. of Columns, No. of Rows,

Where:

- **Plate** is a required label, or text string, at the beginning of the line.
- **Plate Name** is the text name of the plate (up to 40 characters).
- **Plate Barcode** is the barcode identifier of the plate, if used. This field may be left blank.
- **Description** is the text description of the plate.
- **No. of Columns** is the number of numeric columns in the plate. (12 for 96-well plates and 24 for 384-well plates).
- **No. of Rows** is the number of alphabetic rows in the plate (8 for 96-well plates and 16 for 384-well plates).

For example,

Plate, 96-well Control Plate, 678-593, Positive/Negative controls for yeast set, 12, 8,

2.3 Sample Data - Third and Following Lines

The third and following lines, one for each well or sample in the plate, are as follows:

Row, Column, Accession No., Gene Name, Status,

Where:

- **Row** is the alphabetic row identifier of the well (A-H for 96-well plates and A-P for 384-well plates).
- **Column** is the numeric column identifier of the well (1–12 for 96-well plates and 1-24 for 384-well plates).
- **Accession No.** is the unique GenBank or other alphanumeric identifier of the sample.
- **Gene Name** is the name of the gene or sample.
• **Status** indicates the status of the sample:
  0 = empty well (no sample)
  1 = full
  2 = control sample
  3 = sample in well is bad

For example,

A,1,M21304,HUMGLP,1,

You can use an accession number, a gene name, or both to identify your samples. We recommend that at least one of these two fields be filled in for every well. The “Empty” and “Bad” designations will pass through to the array content files. Also, the SpotArray will not dip into those regions of the plate where all samples are labeled as Empty. The “Control” designation changes the color of spot location diagrams during printing, but does not affect the .gal file.

Successive lines each contain information about the sample in a specific well and there should be a line for each sample you wish to identify. When the file is imported, omitted wells - those for which a line is not included - are assumed to be empty.

3 Creating a Plate Contents File

A plate contents file can easily be created using Microsoft Excel or Microsoft Notepad. The example below shows the first few lines of a Microsoft Excel spreadsheet that will generate a generic 96-well plate contents file. Note that in a plate contents file, lines beginning with the # character are treated as comments, which are used in this example to indicate the contents of the individual entries on the line below.
This sample spreadsheet, and a similar one for creating a generic 384-well plate contents file, are installed on the SpotArray instrument in the folder \Packard Biochip\spotarray client\samples.

Using one of these files as a starting point, you can create a plate contents file.

**To create a plate contents file**

1. From your SpotArray system monitor, locate the directory \Packard Biochip\spotarray client\samples on your SpotArray instrument.

2. Open either the 96-well or 384-well sample spreadsheet in Microsoft® Excel.

3. Cut and paste in accession numbers and gene names from your own or a published gene list or website, creating a unique plate contents file for each plate in your library.

4. Assign a status to each sample, or use the default status of “1” or full.

5. When you have created and saved a Microsoft Excel file, save it as a comma delimited plate content file with the default .csv suffix:
   - Click Save As… from the File menu
   - Select CSV (Comma delimited) in the Save as type: dialog box.
   - Enter a new filename that will enable you to identify the plate import file later.

   The resulting file may be imported directly into the SpotArray software as described in Chapter 3, Section 3.2.1.

Alternatively, a file can be created directly from within any generic text editing software package, such as Microsoft Notepad, by following the file structure described in the first section of this appendix. See *Plate Contents File Format* on page E-1.

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**Note:** Be sure to separate the fields with a comma with no spaces or tabs, and end each line with a comma.
1 Overview

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

2 Physical Characteristics

2.1 Main Modules

To minimize use of bench space, the SpotArray is divided into two main modules:

Instrument module. The instrument module includes the controller, substrate holder, plate holder, washer/dryer, and printhead.

- **SpotArray 24**
  Instrument module dimensions: 26” W x 25” D x 16” H (660 x 635 x 407 mm)
  Instrument module weight: 60 lbs (27 kg)
- **SpotArray 72**
  Instrument module dimensions: 40” W x 28” D x 17” H (1,016 x 711 x 432 mm)
  Instrument module weight: 100 lbs (45 kg)

Utility module. The utility module is a separate unit which may be placed on the floor. It includes the power supplies, wash pump and vacuum pump.

- Utility module dimensions: 25” W x 28” D x 14” H (635 x 711 x 356 mm)
- Utility Module weight: 100 lbs.

Optional cart. The optional cart provides a benchtop for the instrument, a monitor stand and a cabinet below with doors to store the utility module and reservoirs.

- Cart dimensions: 58” W x 34” D x 32” H (1475 x 865 x 815 mm)
- Cart weight: 225 lbs. (102 kg)
2.2 Reservoirs

Reservoirs or tanks for wash fluid, wash waste fluid, and humidity generation are located outside the utility module.

- Wash reservoir capacity: 5 gallon (20 liter)
- Wash waste reservoir capacity: 5 gallon (20 liter)
- Humidity reservoir capacity: 2.5 gallon (10 liter); Use only 1.3 gal (5 liters)

3 Electrical and Environmental Specifications

3.1 Electrical Specifications

Supply Voltage: 100 - 130/200-240 VAC
47-63 Hz
12 Amp/6 Amp

Installation/Overvoltage Category II
Pollution Degree 2

3.2 Environmental Specifications

Operating Temperature: 15-30 degrees C (59-86 degrees F)

Operating Humidity: 30-65 % non-condensing
**Glossary**

**Array.** A collection of spots printed on a substrate, arranged in a pattern of regular rows and columns, which constitute the contents of all plates in the plate set. When the printhead contains more than one pin, the array will be made up of subarrays, one for each pin.

**Array content file.** An ordered list of spots in a microarray, which details the location of the spot and the source, identity, and name of the gene used to create the spot. The file also contains the values for the printing protocol parameters. There are two types of files: .gal, and .txt.

**Arrayer.** See Spotter.

**Barcode.** A set of letters and/or numbers encoded into a pattern of bars that can be read and interpreted by a device called a barcode reader. Barcodes are most often printed on labels, with the labels affixed to objects that need to be identified and tracked (plates and substrates, in this case).

**Blotting.** See pre-printing.

**Client (User Interface).** A software application program, running on a computer workstation or instrument CPU, that provides the GUI (user interface) and allows the user to monitor, configure, and control the instrument. The client software communicates with the instrument software to send configuration and control commands to and receive status information from the instrument.

**Dip.** See sample uptake.

**GUI.** See Graphical User Interface.

**Graphical User Interface (GUI).** The portion of a client application program that provides a visual interface between the program and the user. A GUI consists of windows, dialog boxes, icons, buttons, etc. that allow a user to control the execution of the program and view its status with a minimum of typing. Also User Interface.

**Instrument.** The hardware that comprises the entire microarray printer, including the instrument module, the utility module, and wash and waste reservoir.

**Instrument module.** The portion of the instrument comprising the printhead, motion-control subsystem, CPU, washer, dryer, substrate holders, plate holders, and enclosure. The instrument module is separate from the utility module to conserve bench space and to divide the weight of the entire instrument into two more-manageable portions.

**Margins.** See Printable area.

**Microarray.** A substrate with an array of spots printed on it; the end product of a microarray printing instrument. The microarray is the finished, physical object; an array is an ordered collection of spots on the microarray.

**Pin.** A device used by the SpotArray to take up liquid sample from a well in a plate and deposit, or print, a fraction of the taken-up liquid sample as a spot onto a substrate. A pin has a slot in its tip to take up enough sample to print many spots, up to 100 or more.

**Pin lifting.** An optional feature that divides the printhead into two or three zones that print independently, allowing the creation of more compact microarrays. All zones are washed, and dried, and loaded with sample as a single unit, but lift or lower separately for printing.
**Pin loading.** The act of placing pins into the printhead.

**Plate.** A plastic plate, often called a micro-titer plate or micro-well plate, which has 96 or 384 wells in which sample is stored. These plates are made to standard outside dimensions and standard well location and spacing. Plates from different manufacturers have different well shapes and are made from different plastics.

**Pre-printing.** The process of removing excess sample from the sides and slot reservoir in the tip of a pin after taking up sample. This excess sample comes off while printing the first few spots. Therefore, part of the print procedure includes printing these uncontrolled spots onto substrates that will not be used; this process of pre-printing is also called *blotting*.

**Print, Printing.** The act of depositing spots onto substrates from pins by touching the pins onto the substrates.

**Printable area.** The portion of the substrate upon which the microarray may print spots. The maximum size substrate that the SpotArray supports is 25.5 mm x 75 mm. The user may reduce this size to account for barcode(s) or thumb grip locations and to leave margins that may be required by the microarray scanner.

**Printhead.** The block that holds the spotting pins. The printhead holds between 1 and 48 pins.

**Printing procedure.** A set of printing cycles that result in the printing of the contents of all plates onto a batch of slides, as specified by the printing protocol.

**Printing protocol.** A set of parameters that define how the SpotArray prints microarrays.

**Printing cycle.** A series of moves that result in the printing of the contents of all pins onto a batch of slides, as specified by the printing protocol. The moves consist of one or more dips into the plate, pre-printing (or blotting), printing spots onto all of the substrates in the batch, washing, drying, and returning printhead to the plate.

**Protocol.** See *Printing protocol*.

**RH.** See *Relative humidity*.

**Relative humidity.** The amount of water vapor contained within an air sample, expressed on a scale of 0 - 100%.

**Sample.** Solution of array target reagent contained within the wells of a plate. The pins take up the sample and deposit it onto the substrate as spots.

**Sample uptake.** The act of moving the tip of the pin(s) into and then out of the sample(s) contained in the well(s) of a plate.

**ScanArray.** Trademark name of a line of microarray fluorescence scanners made by PerkinElmer Life Sciences.

**Spot.** The dried remains of sample printed onto a substrate by a pin.

**Spotter.** An instrument used to print spots of target material, typically DNA, in arrays on a substrate. A spotter may also be referred to as a microarray printing system or an arrayer.

**Spotting protocol.** See *printing protocol*.

**Subarray.** A collection of adjacent spots printed on a substrate by one pin within the printhead. Due to pin-to-pin tolerances, spots printed by adjacent pins must be spaced at least one spot width apart, thus creating separate rectangles of spots on the substrate. When the printhead contains more than one pin, the group subarrays created by the pins will comprise an array.
**Substrate.** The piece of glass upon which the array is printed. These take the form of microscope slides, which have chemically active coatings on the glass that allow the printed spots of sample to bind to the substrate.

**Target.** The unlabeled DNA printed onto the substrate.

**Utility module.** The portion of the instrument comprising the power supplies, pumps, and filters, intended to be placed under a bench. The utility module is connected to and is controlled by the instrument module.

**Well.** An individual reservoir in a plate in which sample fluid is placed.
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