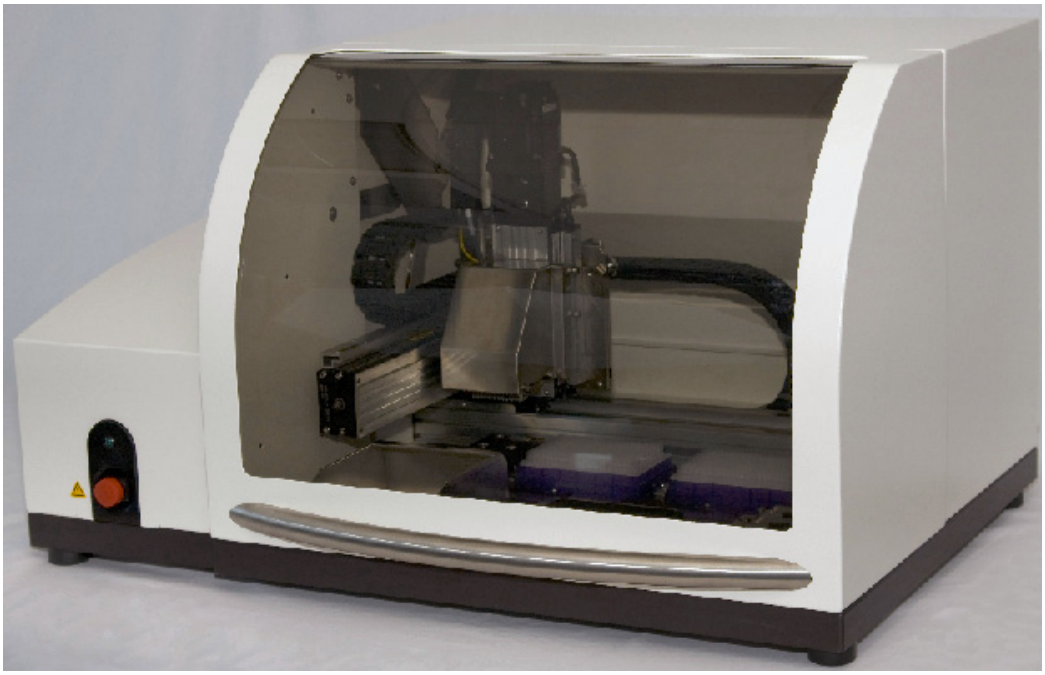
applied biosystems
by life technologies

OpenArray® AccuFill™ System

User Guide



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Contents

About This Guide

Purpose

The OpenArray® AccuFill™ System User Guide provides reference information for the OpenArray AccuFill system and describes how to prepare, maintain, and troubleshoot the system.

Safety information

Note: For general safety information, see this section and Appendix D, "Safety" on page 65. When a hazard symbol and hazard type appear by a chemical name or instrument hazard, see the "Safety" Appendix for the complete alert on the chemical or instrument.

Safety alert words

Four safety alert words appear in Applied Biosystems user documentation at points in the document where you need to be aware of relevant hazards. Each alert word—**IMPORTANT, CAUTION, WARNING, DANGER**—implies a particular level of observation or action, as defined below:

IMPORTANT! – Indicates information that is necessary for proper instrument operation, accurate chemistry kit use, or safe use of a chemical.

CAUTION! – Indicates a potentially hazardous situation that, if not avoided, may result in minor or moderate injury. It may also be used to alert against unsafe practices.

WARNING! – Indicates a potentially hazardous situation that, if not avoided, could result in death or serious injury.

DANGER! – Indicates an imminently hazardous situation that, if not avoided, will result in death or serious injury. This signal word is to be limited to the most extreme situations.

Except for IMPORTANTs, each safety alert word in an Applied Biosystems document appears with an open triangle figure that contains a hazard symbol. *These hazard symbols are identical to the hazard symbols that are affixed to Applied Biosystems instruments* (see "Safety symbols" on page 66).

SDSs

The Safety Data Sheets (SDSs) for any chemicals supplied by Applied Biosystems or Ambion are available to you free 24 hours a day. For instructions on obtaining SDSs, see "SDSs" on page 73.

IMPORTANT! For the SDSs of chemicals not distributed by Applied Biosystems or Ambion contact the chemical manufacturer.

Safety labels on instruments

The following CAUTION, WARNING, and DANGER statements may be displayed on Applied Biosystems instruments in combination with the safety symbols described in the preceding section.

Hazard symbol	English	Français
	**CAUTION!** Hazardous chemicals. Read the Safety Data Sheets (SDSs) before handling.	**ATTENTION!** Produits chimiques dangereux. Lire fiche technique associée au produit avant toute manipulation.
	CAUTION! Hazardous waste. Refer to SDS(s) and local regulations for handling and disposal.	**ATTENTION!** Déchets dangereux. Lire fiche technique associée et prendre connaissance de la régulation locale associées à la manipulation et l'élimination des déchets.
	**DANGER!** High voltage.	**DANGER!** Haute tension.
	WARNING! To reduce the chance of electrical shock, do not remove covers that require tool access. No user-serviceable parts are inside. Refer servicing to Applied Biosystems qualified service personnel.	**AVERTISSEMENT!** Pour éviter les risques d'électrocution, ne pas retirer les capots dont l'ouverture nécessite l'utilisation d'outils. L'instrument ne contient aucune pièce réparable par l'utilisateur. Toute intervention doit être effectuée par le personnel de service qualifié venant de chez Applied Biosystems.

1 Sample Preparation

This chapter covers:

Prepare sample plate for a SYBR® OpenArray® Real-Time PCR run

This section provides instructions on preparing samples for a SYBR® OpenArray® Real-Time PCR run.

Materials and equipment required

See "Ordering Information" on page 55 for a list of equipment, kits, and consumables necessary for this procedure.

Prepare the reagents for use in the master mix

To prepare the reagents for use in the master mix:

1. Dilute the concentrated stock of SYBR® Green I dye to 240X shown:

Reagent	Volume in µL
SYBR Green I dye	5
Water	495
Total volume	500

2. Check the stock concentration:
   a. Dilute a 10-µL aliquot of diluted SYBR Green I dye into 90 µL water.
   b. Measure the optical density at 494 nm.
   c. Adjust the OD to 0.5 to 0.6 units by adding SYBR Green I dye or water to the 240X dilution.
   d. Store at –20 °C in 20-µL aliquots.

3. Prepare 10 mg/mL BSA working solution as shown. Store at –20 °C in 500 µL aliquots.

Reagent	Combine
BSA	100 mg
Water	10 mL
Total volume	10 mL at 10 mg/mL

4. Prepare 15% glycerol working solution as shown. Store at room temperature.

Reagent	Combine
Glycerol	7.5 mL
Water	42.5 mL
Total volume	50.0 mL at 15%

Sample plate preparation

To prepare a sample plate for a SYBR OpenArray Real-Time PCR run:

1. Take the vacuum-sealed OpenArray plates out of the freezer and allow them to thaw for approximately 15 minutes before opening the packages.

2. Prepare Master Mix using the contents of the LightCycler® FastStart DNA Master kit. The LightCycler® kit comes with four vials labeled 1a, 1b, 2, and 3.
   a. Pipette 10 µL of LightCycler FastStart Enzyme from vial 1a to vial 1b.
   b. Mix thoroughly by pipetting up and down. *Do not* use a vortex mixer.
   c. Re-label vial 1b with the label "vial 1: Master Mix" provided in the kit or peel off the label to identify the mix containing the enzyme.

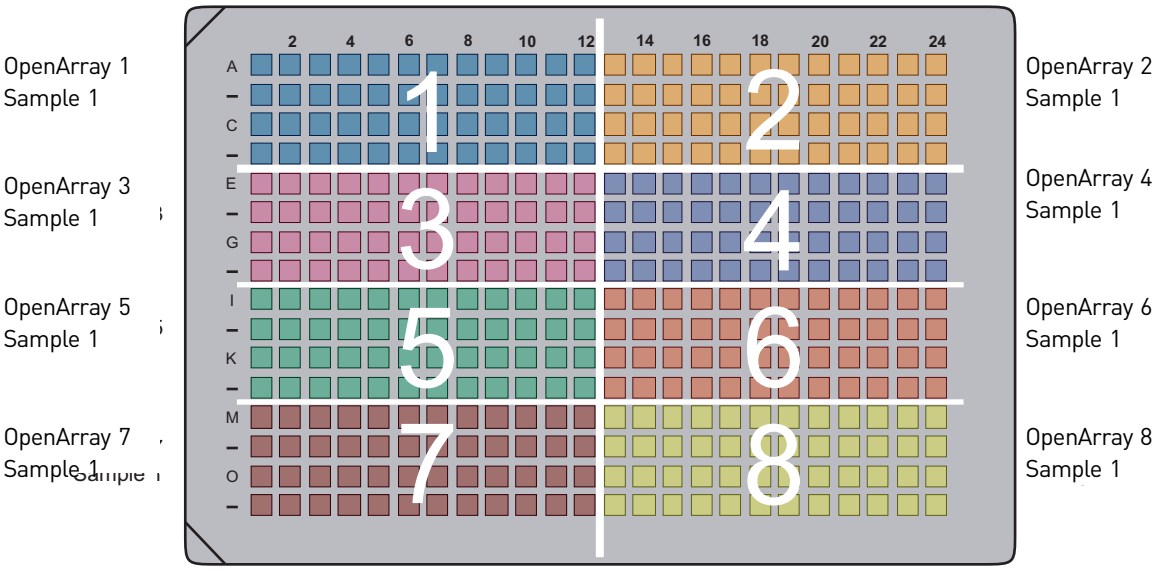
      Protect the vial from light.

3. Prepare the OpenArray PCR Master Mix as described in the following table.

Reagent	Volume (µL)	Stock Conc.	Final Conc.	Conc. Units
LightCycler® DNA Master SYBR® Green I (prepared in step 2 above)	26.4	10	1.0	X
SYBR® Green	1.1	240	1.0	X
Glycerol	8.8	15	0.5	%
Pluronic® F-68	5.3	10	0.2	X
BSA	26.4	10	1.0	mg/mL
MgCl2 (Use a 25-mM solution, i.e., vial 2 with a blue cap in the LightCycler Kit.)	26.4	25	2.5	mM
Formamide	21.1	100	8.0	X
Water	70.9	—	—	—
Final volume without sample	186.4			

Note: The volumes in this procedure are for one OpenArray plate. If you are loading more than one OpenArray plate, adjust the total volume.

4. Plan which sections of the sample plate you will load with samples. Each 4- x 12-well rectangular area corresponds to one OpenArray plate. You may find it helpful to mark these areas with a fine point silver marker, as shown in the following illustration.



5. Pipette 3.5 µL OpenArray PCR Master Mix into each well of the 48-well section of the 384-well sample plate designated OpenArray 1.

6. Pipette 1.5 µL of the template into each well.

7. Pipette gently up and down to mix. *Do not* use a vortex mixer.

8. Cover the sample plate with sealing tape. Centrifuge the plate for 1 minute at 1000 rpm.

9. Place the plate on ice to keep the samples cold.

Prepare sample plate for a TaqMan® OpenArray® Real-Time PCR run

This section provides instructions on preparing a sample plate for a TaqMan OpenArray Real-Time PCR run.

Materials and equipment required

See "Ordering Information" on page 55 for a list of equipment, kits, and consumables necessary for this procedure.

Sample plate preparation

To prepare a sample plate for a TaqMan OpenArray Real-Time PCR run, follow these steps:

1. Take the vacuum-sealed OpenArray plates out of the freezer and allow them to thaw for approximately 15 minutes before opening the packages.

2. Prepare the OpenArray PCR Master Mix as described in the tables below.

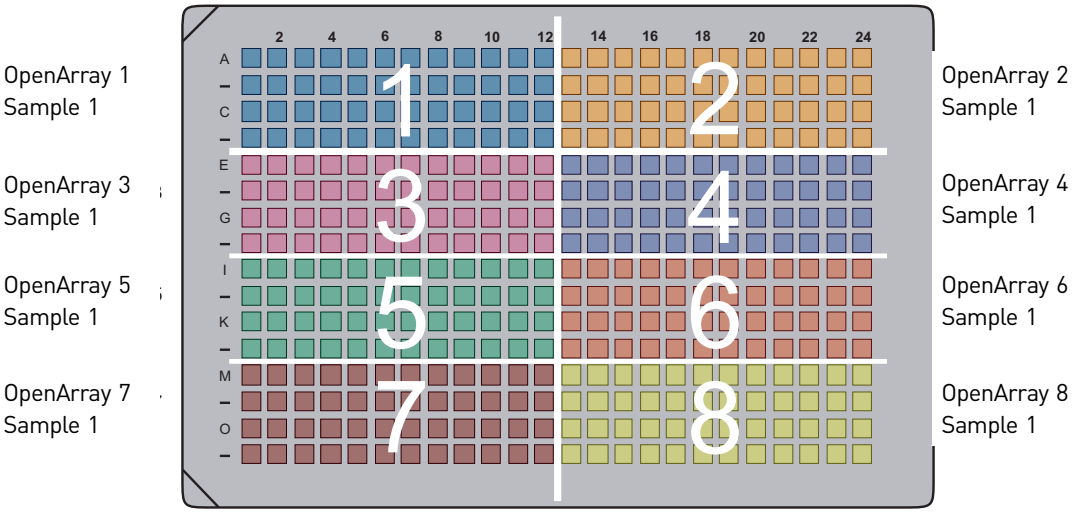
   IMPORTANT! Master Mix components will vary depending on the source of the template you are using. Use only *one* of the tables below, depending on whether your experiment uses cDNA or other DNA.

Reagent for cDNA samples	Volume (µL)	Stock Conc.	Final Conc.	Conc. Units
GeneAmp® Fast PCR Master Mix	32.0	2	1	X
5X REmix (supplied with the TaqMan OpenArray kit)	52.8	5	1	X
Water	15.8	—	—	—
Final volume without sample	200.6			

Reagents for gDNA samples	Volume (µL)	Stock Conc.	Final Conc.	Conc. Units
GeneAmp Fast PCR Master Mix	132.0	2	1.0	X
5X REmix (supplied with the TaqMan OpenArray kit)	52.8	5	1.0	X
BSA	1.3	10	0.05	mg/mL
Water	14.5	—	—	—
Final volume without sample	200.6			

Note: The volumes in this procedure are for one OpenArray plate. If you are loading more than one OpenArray plate, adjust the total volume.

3. Plan which sections of the 384-well sample plate you will load with samples. Each 4- x 12-well, rectangular area corresponds to one OpenArray plate. You may find it helpful to mark these with a fine point silver marker, as shown in the following illustration.



4. Pipette 3.8 µL of Master Mix into each well of the 48-well section of the 384-well sample plate designated OpenArray plate 1.

5. Pipette 1.2 µL of the sample into each well.

6. Cover the sample plate with sealing tape. Centrifuge the plate for 1 minute at 1,000 rpm. Place the plate on ice to keep the samples cold.

Prepare sample plate for a TaqMan® OpenArray® Genotyping run

This section provides instructions on preparing a sample plate for a TaqMan OpenArray Genotyping run.

Sample plate preparation

A single OpenArray plate can accept one to three loads from a sample plate, depending on how many samples per subarray are required.

To prepare a sample plate for a TaqMan Genotyping run, do the following steps.

Note: For chemical hazard alerts, see Appendix D, Safety.

1. Be sure that the OpenArray 384-well sample plate, OpenArray AccuFill Loader Tips, and Plate Holder are completely clean and dry:
   a. Soak the plate guide, loader tips, and Plate Holder in 10% bleach for at least 10 minutes.
   b. Rinse with water, then rinse with ethanol.
   c. Let the parts completely air-dry. If they are immediately needed, wipe dry with paper towels and spray with compressed nitrogen gas.

   IMPORTANT! Residual water prevents correct loading of the samples into the TaqMan OpenArray plates.

2. Fill the appropriate number of TaqMan OpenArray Genotyping Cases with TaqMan OpenArray Immersion Fluid:
   a. Using scissors, open a container of immersion fluid.
   b. Place the case in the case rack and fill the case approximately two-thirds with immersion fluid.

      IMPORTANT! **Within one hour after opening the container of immersion fluid**, fill the case with immersion fluid, insert a loaded TaqMan OpenArray plate into the case, then seal the case.

3. Remove the TaqMan OpenArray plates from the freezer, but do not open the packaging. Allow the TaqMan OpenArray plates to thaw at room temperature (approximately 5 minutes).

   Note: Unopened TaqMan OpenArray plates can remain at room temperature for up to 24 hours.

   IMPORTANT! Thaw only the TaqMan OpenArray plates you will need for the current loading session.

Prepare the DNA samples

Quality of DNA

Be sure that the DNA you use for Genotyping experiments:

- Is extracted from the raw material you are testing with an optimized protocol.
- Does not contain PCR inhibitors.
- Has an $A_{260/230}$ ratio between 1.7 and 1.9.
- Has an $A_{260/280}$ ratio between 1.7 and 1.9.
- Is intact as visualized by gel electrophoresis.
- Has not been heated above 60 °C; temperatures above 60 °C can cause degradation.

Quantity of DNA

Applied Biosystems recommends that you quantify the amount of genomic DNA in your samples. Note that:

- The TaqMan OpenArray plate requires 250 copies of haploid genome for each individual through-hole reaction.
- For optimal cluster plot results, it is important to normalize all genomic DNA samples in an experiment so that each through-hole receives the same input quantity of sample.

For an example quantification procedure, see the following topic,"Quantification procedure for human DNA samples".

Quantification procedure for human DNA samples

The recommended amount of template for each through-hole reaction in a TaqMan OpenArray plate is 250 copies of the haploid genome, equivalent to 0.84 ng for human DNA samples. Quantify human DNA samples using the TaqMan RNase P Detection Reagents Kit and the TaqMan DNA Template Reagents Kit.

Note: The recommended starting concentration for human DNA samples is 50 ng/mL. See the DNA Calculator in the Applied Biosystems *TaqMan OpenArray Genotyping System User Guide* (PN4377476).

Quantify human DNA samples

Generate a standard curve using the DNA template standards provided in the TaqMan DNA Template Reagents Kit and the RNase P gene primers and probe provided in the TaqMan RNase P Detection Reagents Kit.

Note: Refer to the appropriate instrument user guide for detailed instructions on performing and analyzing runs.

Note: For chemical hazards alerts, see Appendix D, Safety.

To quantify human genomic DNA:

1. Create and set up a sequence detector plate document.
2. Prepare the reaction plate using the following components:
   - 2× TaqMan Universal PCR Master Mix, No AmpErase® UNG
   - 20× Primer and TaqMan Probe (FAM™ dye) mix
   - DNA template standard or genomic DNA sample
   - DNase-free, sterile-filtered water

   Use at least three replicates of each standard or sample, and use all five DNA standards to ensure an accurate standard curve is generated. The range of known copy number should bracket anticipated copy numbers of the unknown samples on the same reaction plate.
3. Run the reaction plate on an Applied Biosystems Real-Time PCR System using the following thermal cycling conditions:

	AmpliTaq Gold® enzyme activation	**PCR**	
	HOLD	CYCLE (40 cycles)	
		Denature	Anneal/extend
Time	10 min.	15 sec	1 min.
Temp	95 °C	92 °C	60 °C

4. Generate a standard curve to quantify the amount of DNA in each sample.

The recommended starting concentration for human DNA samples is 50 nanograms per milliliter. See the DNA Calculator in the Applied Biosystems *TaqMan OpenArray Genotyping System User Guide* (PN4377476).

Perform multi-sample loading

The OpenArray® AccuFill™ system enables you to load more than one sample per subarray. Before performing multi-sample loading, set up the sample plates to accommodate the number of samples you will use per subarray for Genotyping applications.

The following table shows sample plate volumes, in microliters, for two-sample and three-sample loads:

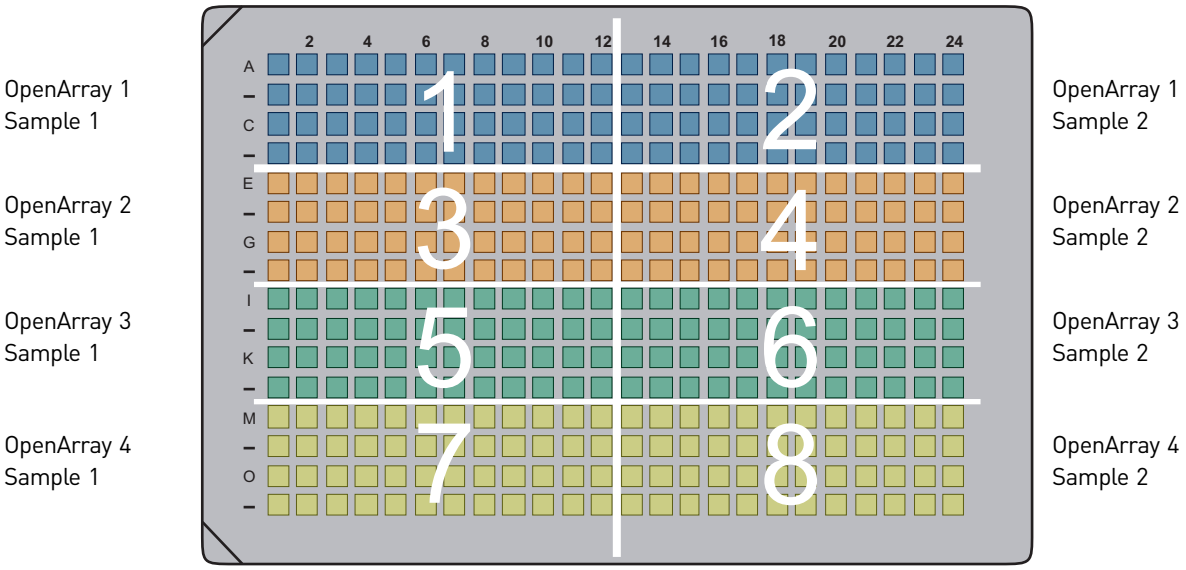
Sample PlateVolumes (µL)	
Two samples	**Three samples**
4	3

Note: Multi-sample loading is available only for Genotyping applications.

Set up the sample plate for two samples

These instructions describe how to set up a sample plate with two separate DNA samples.

1. Label the two sections of the sample plate so that you can keep track of what is loaded in each section. An example is shown in the following illustration.

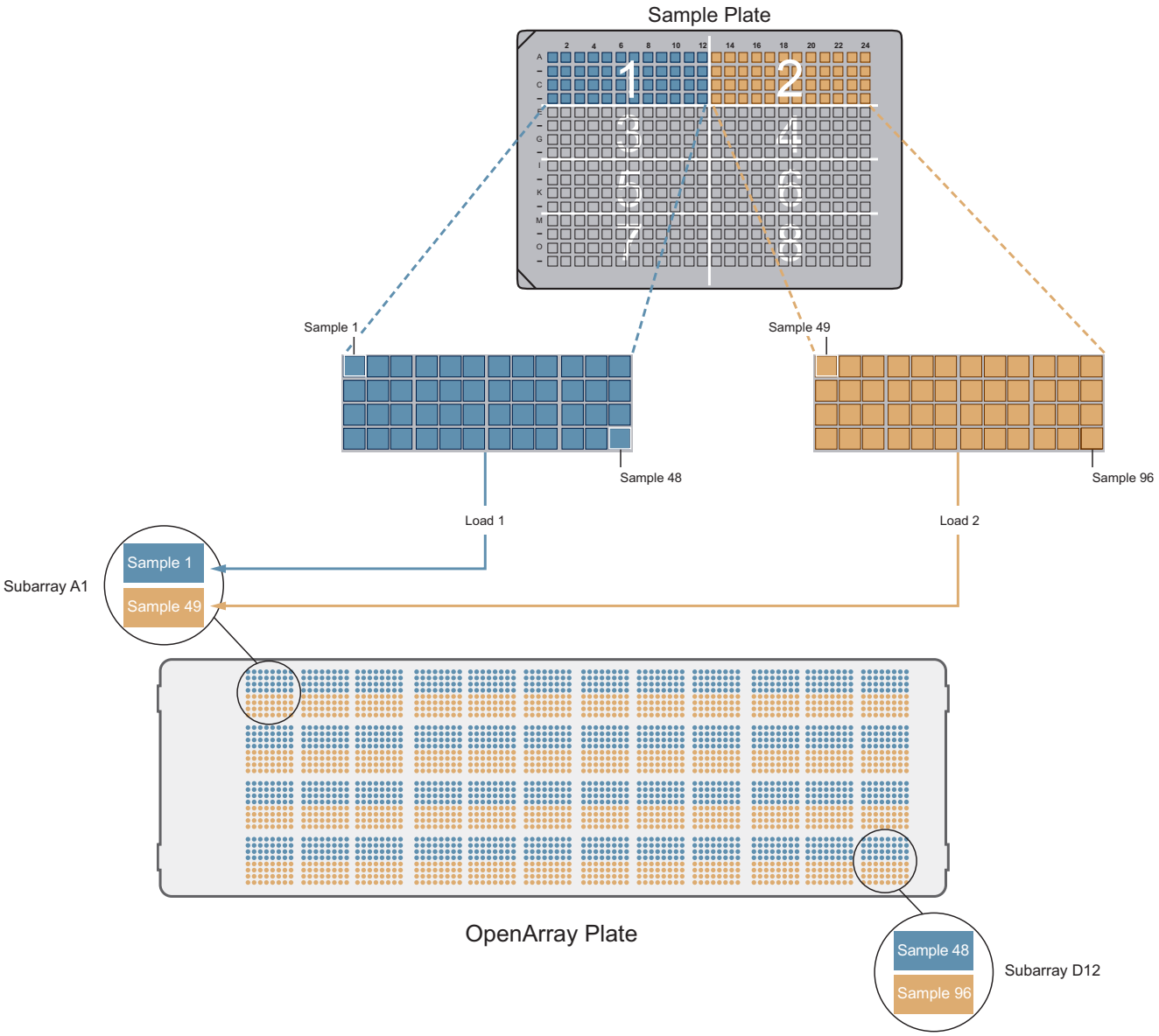


> **IMPORTANT!** Be sure to track where the samples are in the sample plate. For each sample you use, Applied Biosystems recommends creating a sample information file in comma-separated values (CSV) format. This CSV file can be used in sample integration. See "Use Sample Integration" on page 39 for more information.

2. Use the following loading arrangement. Refer to the example on page 20 for the locations of the sample plate sections.
   - OpenArray 1 - OpenArray plate for the first run is in Plate Holder position 1.
     - The first sample for the subarray comes from section 1 of the sample plate, which holds samples 1 to 48.
     - The second sample for the subarray comes from section 2 of the sample plate, which holds samples 49 to 96.
   - OpenArray 2 - OpenArray plate for the second run is in Plate Holder position 2.
     - The first sample for the subarray comes from section 3 of the sample plate, which holds samples 1 to 48.
     - The second sample for the subarray comes from section 4 of the sample plate, which holds samples 49 to 96.

- OpenArray 3 - OpenArray plate for the third run is in Plate Holder position 3.
  - The first sample for the subarray comes from section 5 of the sample plate, which holds samples 1 to 48.
  - The second sample for the subarray comes from section 6 of the sample plate, which holds samples 49 to 96.
- OpenArray 4 - OpenArray plate for the fourth run is in Plate Holder position 4.
  - The first sample for the subarray comes from section 7 of the sample plate, which holds samples 1 to 48.
  - The second sample for the subarray comes from section 8 of the sample plate, which holds samples 49 to 96.

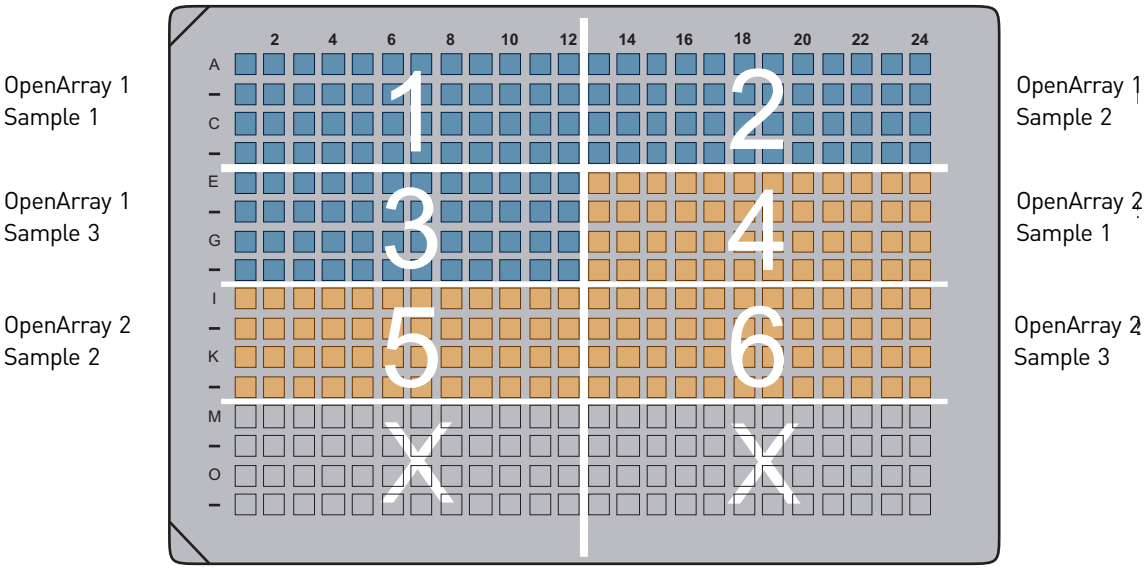
An example of two samples per subarray is shown on page 20.


Sample Plate
1
2
Sample 1
Sample 48
Sample 49
Sample 96
Load 1
Load 2
Subarray A1
Sample 1
Sample 49
OpenArray Plate
Sample 48
Sample 96
Subarray D12

Set up the sample plate for three samples

These instructions describe how to set up a sample plate with three separate DNA samples.

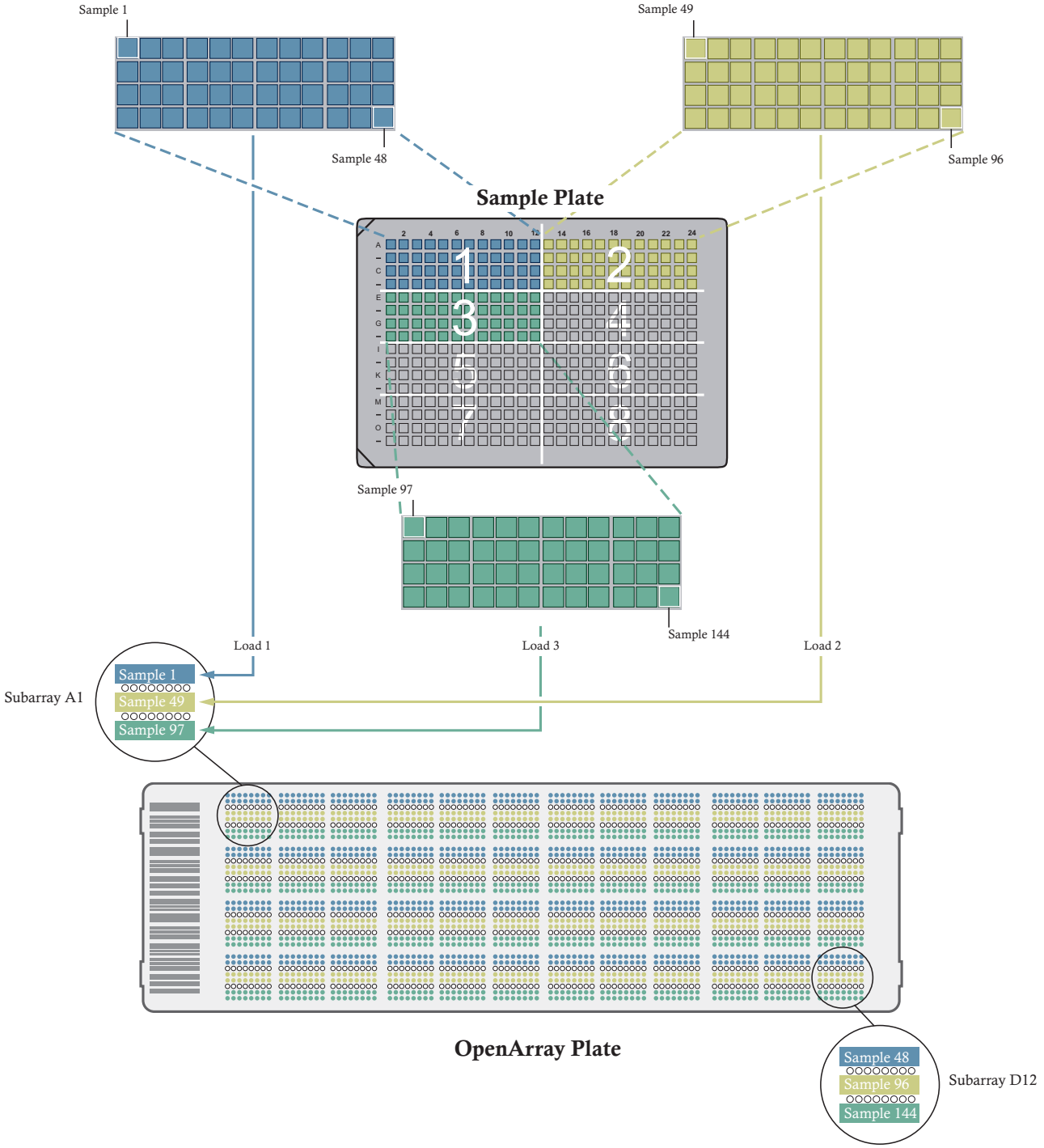
1. Label the three sections of the sample plate so that you can keep track of what is loaded in each section. An example is shown in the following illustration.



> **IMPORTANT!** Be sure to track where the samples are in the sample plate. For each sample you use, Applied Biosystems recommends creating a sample information file in CSV format. This CSV file can be used in sample integration. See "Use Sample Integration" on page 39 for more information.

2. Use the following loading arrangement.
   - OpenArray 1 - OpenArray plate for the first run is in Plate Holder position 1.
     - First sample for the subarray comes from section 1 of the sample plate, which holds samples 1 to 48.
     - Second sample for the subarray comes from section 2 of the sample plate, which holds samples 49 to 96.
     - Third sample for the subarray comes from section 3 of the sample plate, which holds samples 97 to 144.
   - OpenArray 2 - OpenArray plate for the second run is in Plate Holder position 2.
     - First sample for the subarray comes from section 3 of the sample plate, which holds samples 1 to 48.
     - Second sample for the subarray comes from section 4 of the sample plate, which holds samples 49 to 96.
     - Third sample for the subarray comes from section 6 of the sample plate, which holds samples 97 to 144.

   An example of three samples per subarray is shown on page 22.


Sample 1
Sample 48
Sample 49
Sample 96
Sample Plate
1
2
3
4
Sample 97
Sample 144
Load 1
Load 3
Load 2
Subarray A1
Sample 1
Sample 49
Sample 97
OpenArray Plate
Sample 48
Sample 96
Sample 144
Subarray D12

2 Sample Loading

This chapter covers:

Materials and equipment required

See "Ordering Information" on page 55 for a list of equipment, kits, and consumables necessary for this procedure.

Workflow

The workflow for preparing the OpenArray® AccuFill™ instrument and OpenArray cases summarizes the simplest loading scenario, which involves one sample and one OpenArray.

Workflow

"Initialize the system" (page 25)

↓

"Prepare the cases" (page 29)

↓

"Prepare for loading" (page 29)

↓

"Load an OpenArray plate" (page 32)

Important guidelines for handling the plate

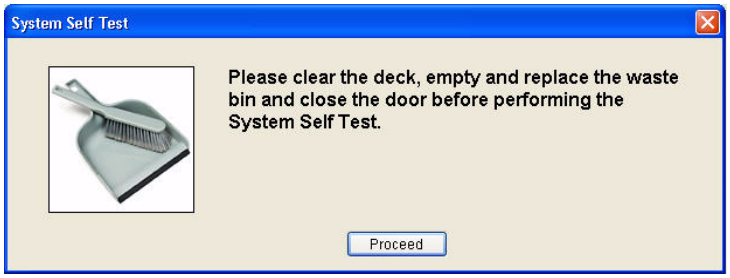
- Wear gloves that are one size smaller than the size you typically wear, to help prevent excess glove material from contacting the OpenArray plate while loading.
- Hold the OpenArray plate by the edges, at the end opposite from the barcode. Do not touch the through-holes.
- *Within one hour after opening the plate packaging*, load the TaqMan OpenArray plate with sample, place the loaded plate in a OpenArray Genotyping Case, then seal the case.
- If you drop a loaded OpenArray plate, discard it in the appropriate waste container.

Initialize the system

Initialize the system to prepare for the loading operation:

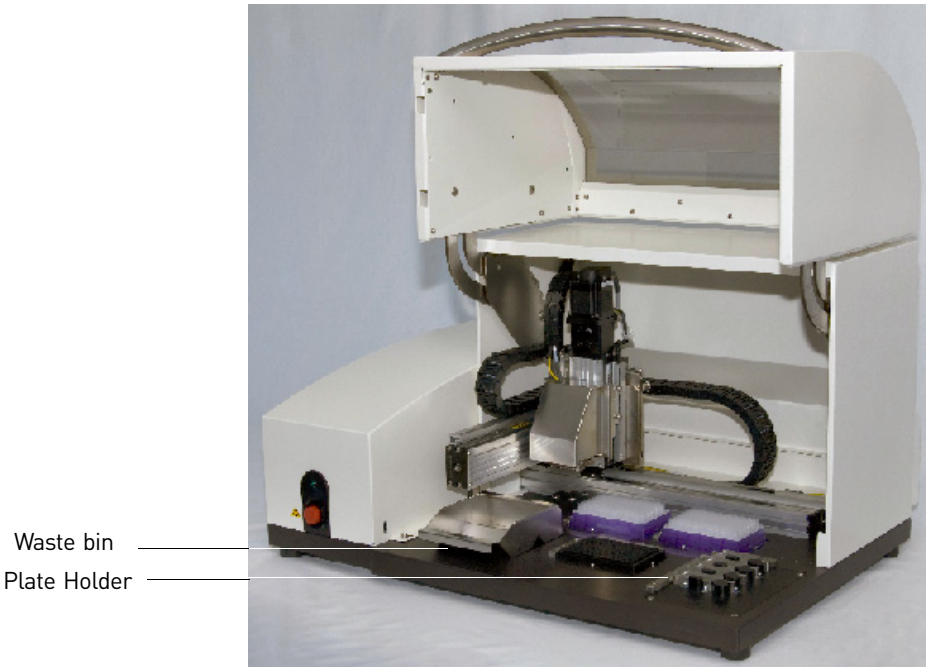
1. Make sure that the enclosure door is closed. Double-click the OpenArray® AccuFill™ icon to launch the software. The software checks the computer and connections as the system starts.

After the application has been launched, it displays a message requesting you to clear the deck and empty the waste bin of used tips.

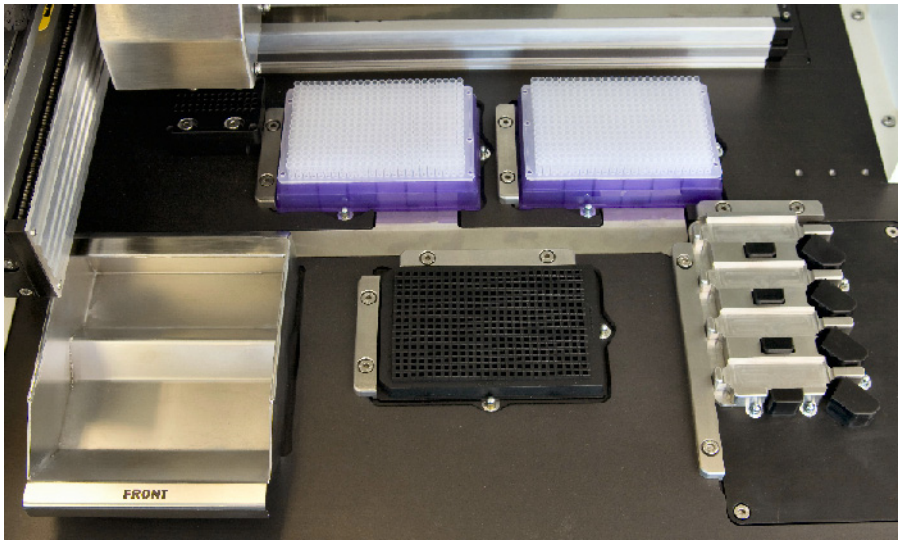


   a. Open the instrument by grasping the enclosure door handle and gently, but firmly, pulling the enclosure door up. The opened instrument is shown below.

Note: To safely operate the instrument, it is important to keep the deck clear and have enough room in the waste bin to eject the used pipette tips.



b. Empty the waste bin and place it back on the deck.



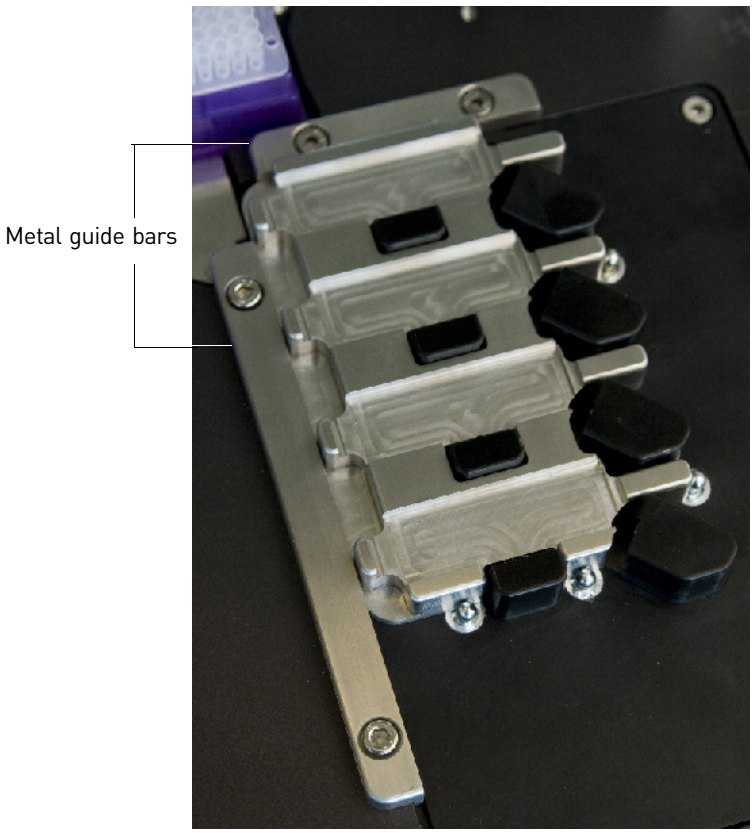
2. If there is a Plate Holder on the deck, check if there are any OpenArray plates on it. If necessary, remove them.

3. If there is not a Plate Holder on the deck, place one on the deck:

   Orient the Plate Holder so that the text etched on it is readable, with the barcode on the left.

IMPORTANT! The locating bars and Plate Holder must be clean and free of any foreign material. which can cause a shift in location.

To insert the Plate Holder into its place on the deck, align the top and left edges with the metal guide bars on the deck and firmly press down. You will hear it snap into place.

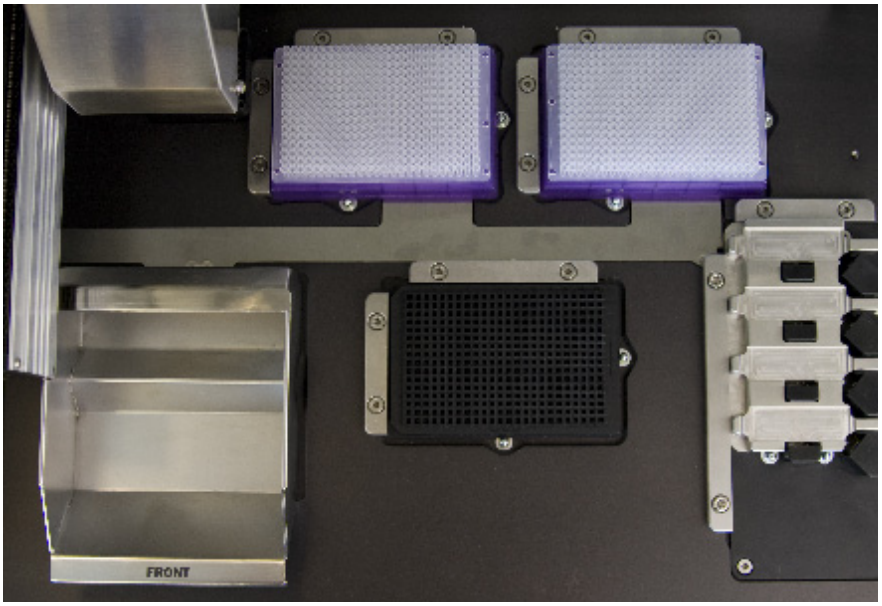


4. If necessary, replace the tip boxes.

   Note: Tip boxes contain 384 tips, divided into 8 sections. When you click Load, the AccuFill instrument loads as though a new, full box of tips is on the deck. System software prompts you to verify that tips are in the locations shown in the Setup Deck screen. Clicking a section in the Setup Deck window confirms that tips are in that section of the tip box. We recommend using a full tip box.

   a. Place tip boxes into the assigned locations.

b. Place tip boxes on the deck in the two side-by-side recessed rectangular platforms. See the illustration for the location of the tip boxes (purple and white).



c. Remove the cover before using the tips for loading.

5. Close the door on the instrument.

6. Begin the System Self Test.

Note: System Self Test runs only at start up. The test does not run again unless the system is restarted or a self test is intentionally run. The System Self Test utility is in the Instrument drop-down menu in the OpenArray® AccuFill™ application.

Click **Proceed**. The application performs a number of self tests and is then ready for you to continue.

Prepare the cases

Cases for OpenArray plates are included with the OpenArray Case Sealing Station. Fill the cases – one OpenArray plate per case – with immersion fluid:

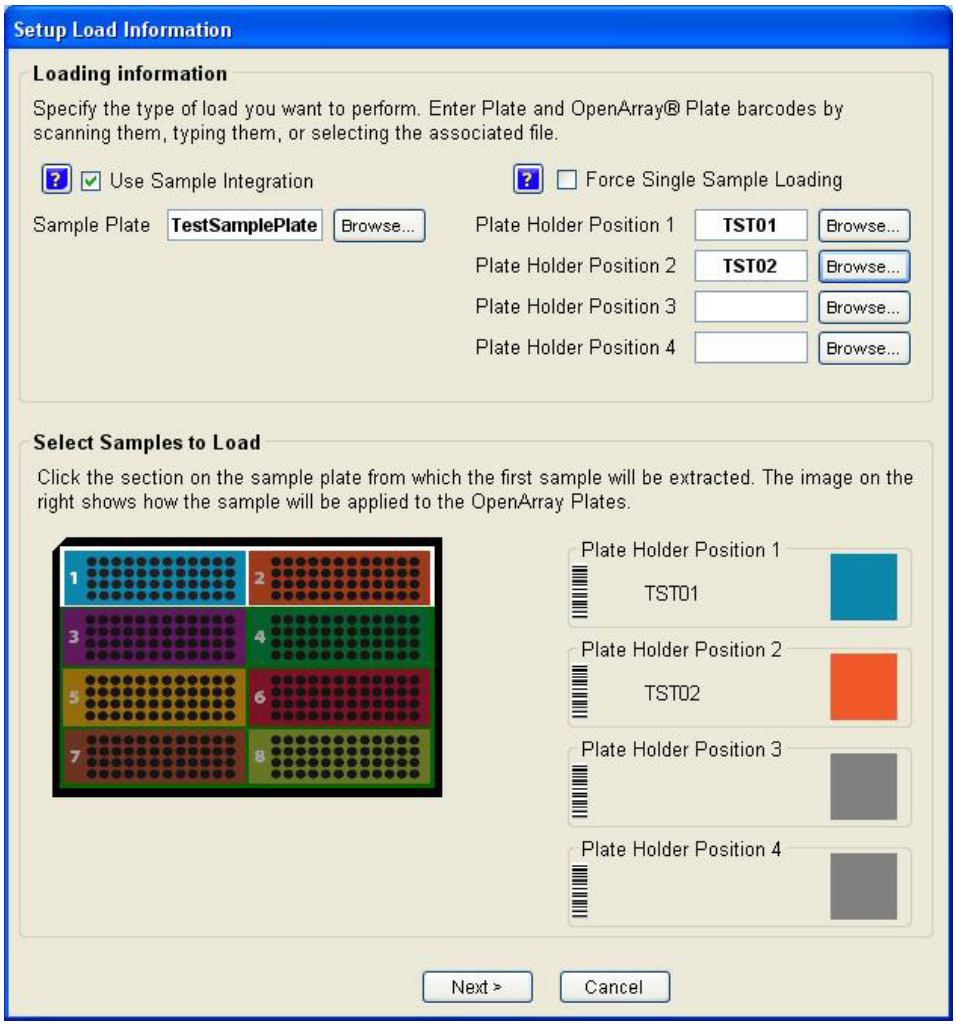
1. Use scissors that are clean and not rusty to open a container of immersion fluid.
2. Place a case in the case rack and fill it approximately 3/4 of the way with immersion fluid.

   IMPORTANT! Immersion fluid must not be exposed to air for more than 60 minutes. Make sure that the immersion fluid is degassed. Bubbles in a case negatively affect data quality.

Prepare for loading

To set up the software:

1. Click **Setup & Load**. The Setup Load Information window opens.



2. If the enclosure door on the AccuFill instrument is not open, open it by grasping the door handle and lifting the door up.

3. Type the name of the sample plate in the Sample Plate field.

   The sample plate name can also be scanned with a barcode reader, which is not supplied with the AccuFill™ system.

4. Insert the 384-well sample plate with the foil cover still in place. Press on the plate until you hear it snap into place.

   Note: Do not remove the foil from the 384-well sample plate at this stage.

   If you are using sample integration, see "Use Sample Integration" on page 39.

   If you are performing multi-sample loading, see "Perform multi-sample loading" on page 17.

5. Select **1** from the Samples Per Subarray drop-down list.

6. In the Plate Holder Position 1 text field, enter the unique serial number of the OpenArray plate that is loaded in the first position of the Plate Holder. The procedure to enter the serial number varies, depending on whether you use Sample Integration:

   If you are *not* using Sample Integration, enter the plate serial number by either:

   - Typing the serial number, or
   - Using a barcode reader to scan the serial number from the plate itself.

   If you **are** using Sample Integration, enter the plate serial number using any one of the following three methods:

   - Type the serial number. There must be a file by the same name in the default directory.
   - Use a barcode reader to scan the serial number from the OpenArray plate itself. There must be a file by the same name in the default directory.
   - Click the Browse button and locate the file for the OpenArray plate.

   As you enter the serial number, it is reflected in the representation of the OpenArray plates in the lower section of the window.

   Note: If you enter the name of an OpenArray plate that you have already used, the software warns you that the name has already been used, but will not prevent you from continuing.

7. Place the OpenArray plate into the Plate Holder. When handling OpenArray plates:

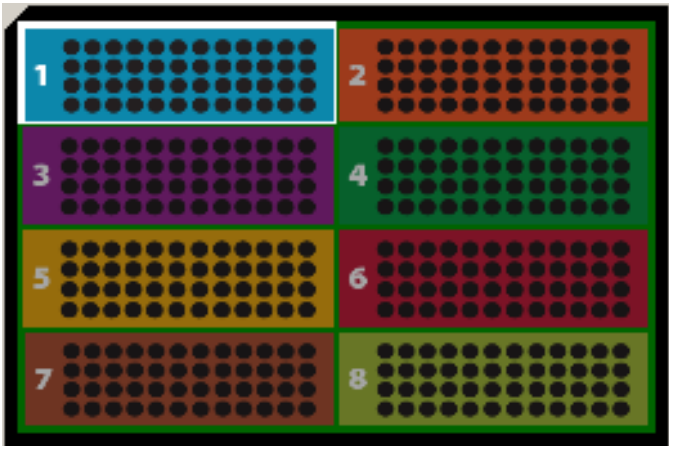
   - Always hold OpenArray plates by the edges.
   - If you inadvertently drop a loaded OpenArray plate, discard it in the sharps waste container.
   - Be sure to load OpenArray plates within an hour after you open them.

   Hold the OpenArray plate by the edges and place it into the Plate Holder with the barcode face up and to the left. You can place one to four plates for one loading run.

   If you are using more than one OpenArray plate, repeat *step 6* and *step 7* as necessary.

8. By default, the AccuFill™ software identifies the next available section of the sample plate (section 1 in the following illustration). When you choose a section of the sample plate, you will see the corresponding color on the illustration of the OpenArray plates at the right. You can choose only the location of the first sample. The application automatically chooses the next available section of the sample plate in the order shown.

   For multi-sample or multi-plate loading, you can choose sections in any order by repeating the setup procedures between plates. The setup procedure is described in"Perform multi-sample loading" on page 17.



   Note: A warning appears if you choose a sample section that you have recently used for another loading run.

9. Click **Next**.

Load an OpenArray plate

Workflow

Load a single OpenArray plate

Remove the foil from the sample plate.

↓

Load the OpenArray plate.

↓

Remove the OpenArray plate.

↓

Insert the OpenArray plate into a case.

Load multiple OpenArray plates

Remove the foil from a sample plate.

↓

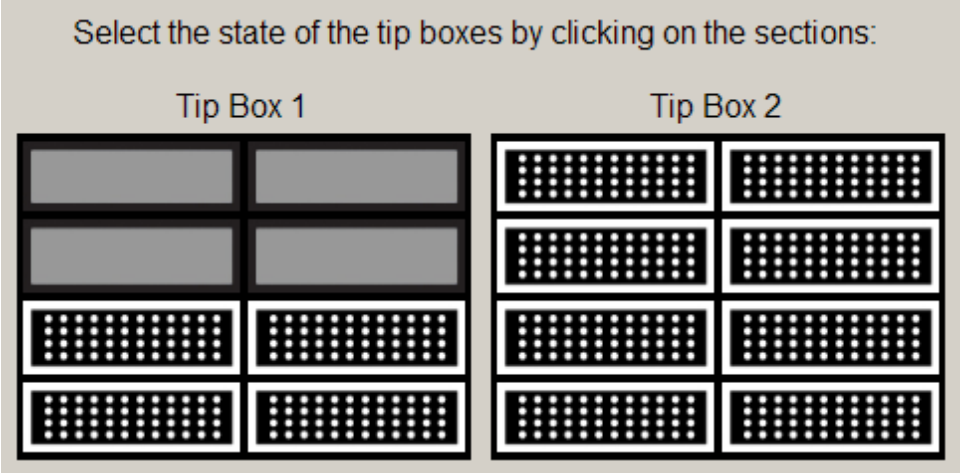
Repeat loading, removing, and encasing steps for each OpenArray plate.

Load an OpenArray plate

To load an OpenArray plate:

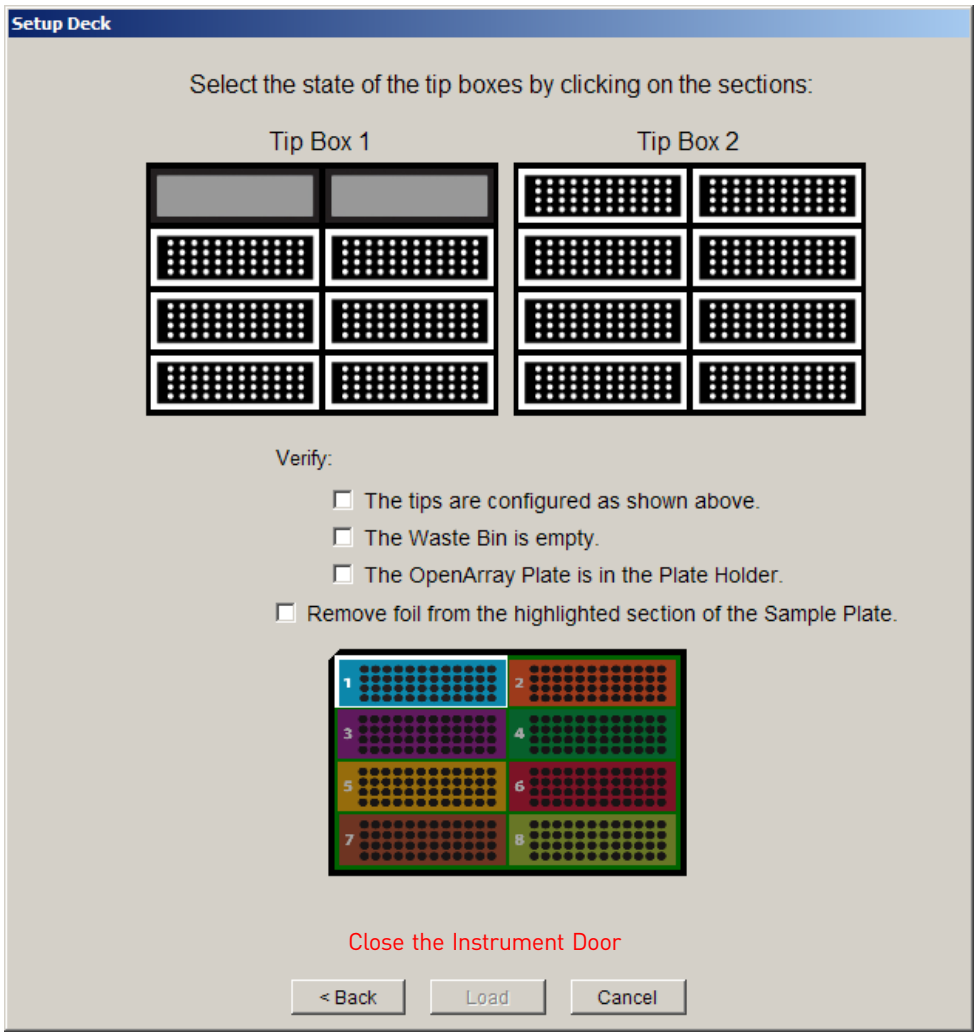
1. Visually verify that the Tip Status window in the software matches the state of the tips on the deck. Make sure that:
   - Gray areas in the Tip Status window indicate that no tips are present.
   - White areas indicate that tips are present.

   If the software and the tips on the deck do not match, click the appropriate section in the Tip Status window.



Note: Use tips within one week after opening the box. Discard any unused tips after one week.

2. Verify each of the following conditions and, when verified, select its check box:
   - Tips are configured as shown in *step 1*.
   - Waste bin is empty.
   - OpenArray plate is in the Plate Holder.



Note: The software will not continue until you select all the check boxes.

3. With forceps, peel off the foil covering the area of the sample plate containing the samples to be loaded on the OpenArray plate.

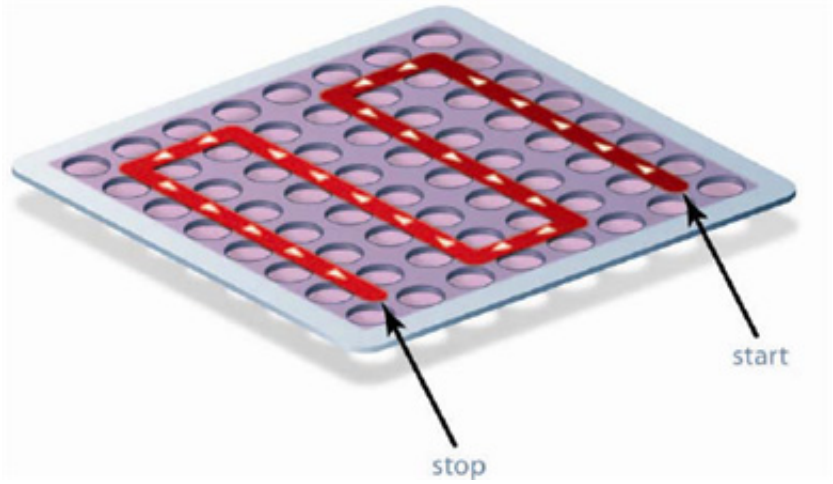
4. Select the check box **Remove foil from the highlighted section of the Sample Plate**.

5. Close the instrument door.

6. Click **Load**.

   Note: If the number of OpenArray plates in the instrument differs from the number that is entered in the Setup Load Information window, an error message instructs you to remove any extra OpenArray plates. Correct the error and continue.

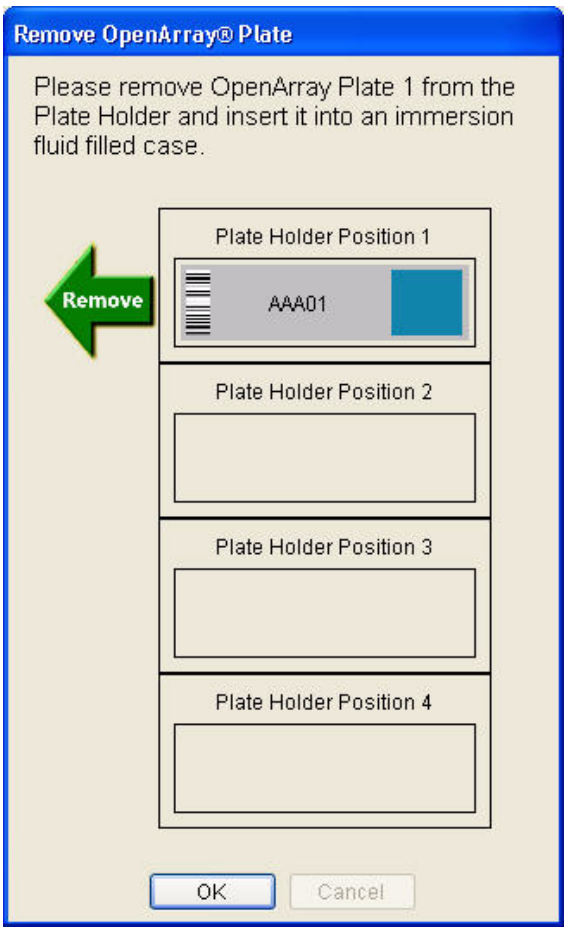
   You can follow the progress of the OpenArray plate loading on the screen. The samples in each tip are loaded in the OpenArray plate. Each tip fills the 64 through-holes in one subarray, travelling in the pattern shown (the following illustration shows the load path for only one sample):



   Sections of the sample plate are loaded in a three sample/subarray order: first, section 2, then section 1, and then section 3.

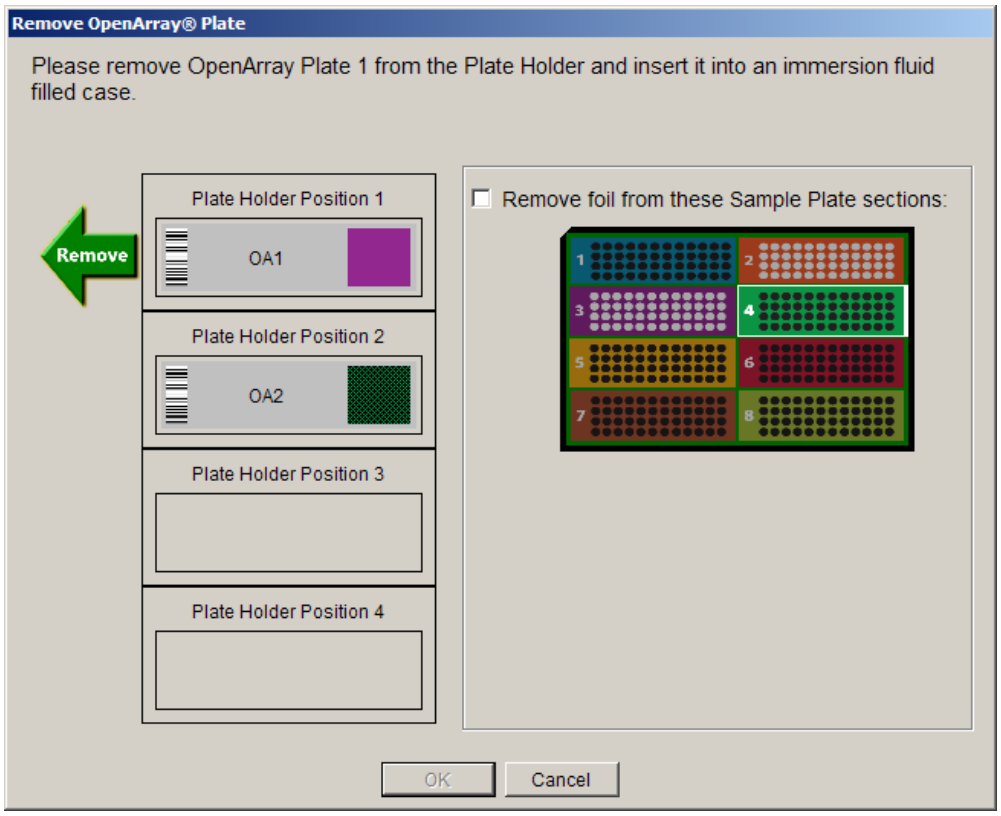
7. If you are loading only one OpenArray plate, follow *step a*.
   If you are loading more than one OpenArray plate, follow *step b* through *step g*.
   a. If you are loading only one OpenArray plate, you will see the Remove OpenArray Plate window when the load sequence is done.
      Click **OK**.

      Open the instrument, carefully remove the loaded OpenArray plate, and immediately insert it into a prepared case.
      See "Insert the loaded OpenArray® plate into a case" on page 42. Skip steps 7b through 7g and proceed to "Loading history" on page 38.



   b. If you are loading more than one OpenArray plate, you will see the Remove OpenArray Plate window on page 37 when the load sequence is done. Open the instrument, carefully remove the loaded OpenArray plate and insert it into a prepared case immediately. See "Insert the loaded OpenArray® plate into a case" on page 42.
   c. Remove the foil from the next sample plate section as indicated by the illustration in the window and close the enclosure door.
   d. Select the **Remove foil check box** and click **OK**.
   e. Close the instrument door.
   f. The instrument loads the next OpenArray plate.

g. Continue from *step b* until the last OpenArray plate has been loaded.



Note: After you have finished your experiment, clean the OpenArray AccuFill instrument.

Loading history

The software displays the loading history, which logs the following information for the completed load:

- OpenArray Plate Barcode
- Sample Plate Barcode
- Sample Plate Selected Area
- Sample Number
- Plate Holder Position
- Sample Load Time
- Load Status

Save the loading history

You can save the load information by either:

- Clicking the **Copy All** button to capture the contents of the table. You can then paste the data into an existing spreadsheet or word processing document. The contents are copied in a tab-delimited format.
- Clicking the **Save As** button to save the contents of the table. This method saves the data to a CSV file with a name and location of your choosing.

To not save any information, click **Close**.

3 Sample Integration

Use Sample Integration

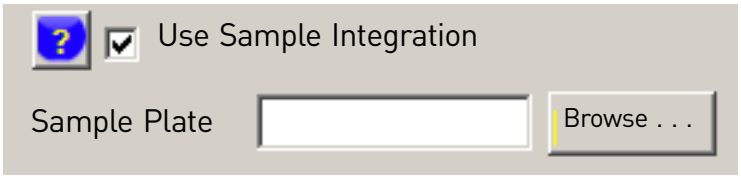
Sample Integration combines the information about your samples – stored in a comma-separated values (CSV) file – with the information about the pre-loaded assays in the OpenArray®plate. The output of Sample Integration is the assignment of all samples to their respective assays in one new updated plate file.

The following table shows the file extension used for the resulting Sample Integration file, depending on the type of chemistry used in your analysis run.

Type of chemistry kit	File type	File extension
SYBR® OpenArray Real-Time kits	transcript	.tpf
TaqMan® OpenArray Real-Time kits		
TaqMan OpenArray Genotyping Analysis kits	SNP	.spf

To use Sample Integration:

1. In the Setup Load Information window, check the **Use Sample Integration** check box. A Browse button appears next to the Sample Plate field.

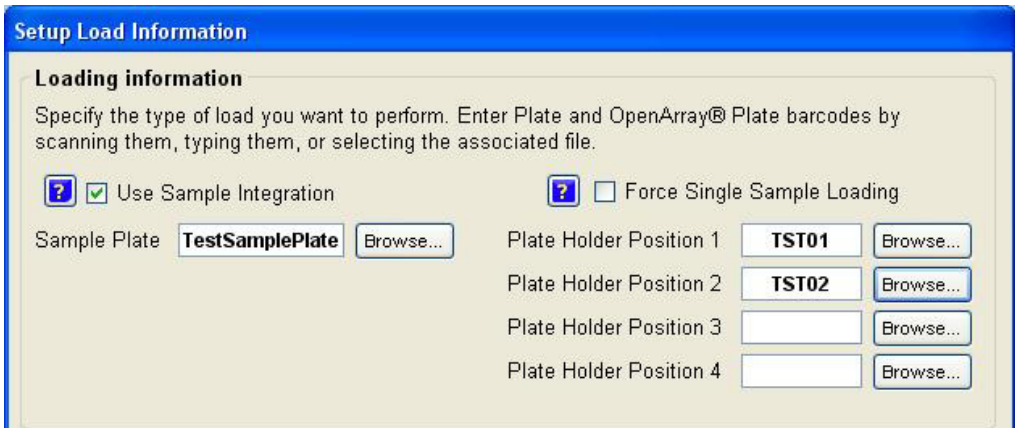


   Select **Use Sample Integration** to combine sample information with assay information. This method creates a complete plate file that allows you to further process your samples.

2. Type the name of a file in the **Sample Plate** field or click **Browse** to locate the plate file for the sample (in CSV format). By default, sample plate files are stored in the directory `C:\OpenArray\Sample Plates`.

3. Optionally select the **Force Single Sample Loading** check box to have the same sample loaded in each subarray.

   Note: When you select Force Single Sample Loading, the same sample is loaded into each of the 64 through-holes of a subarray on the OpenArray plate, even when the plate is formatted for two or three samples per subarray.



4. Select the OpenArray plate file provided by Applied Biosystems by typing the name of the file in the **Plate Holder Position** field or click **Browse** to locate the plate file for the OpenArray plate. OpenArray plate files are located in:

   `C:\OpenArray\OpenArray Plates`

   When the run is done, the integrated data is saved, with the prefix "Loaded," by default in:

   `C:\OpenArray\Loaded OpenArray Plates`

5. The newly created plate file must now be used to process the OpenArray plates in the OpenArray NT Cycler. Transfer the newly created plate file by using your local network or some form of portable media such as a CD or a USB drive.

4 Prepare the plate for the NT Cycler

This chapter covers:

Workflow

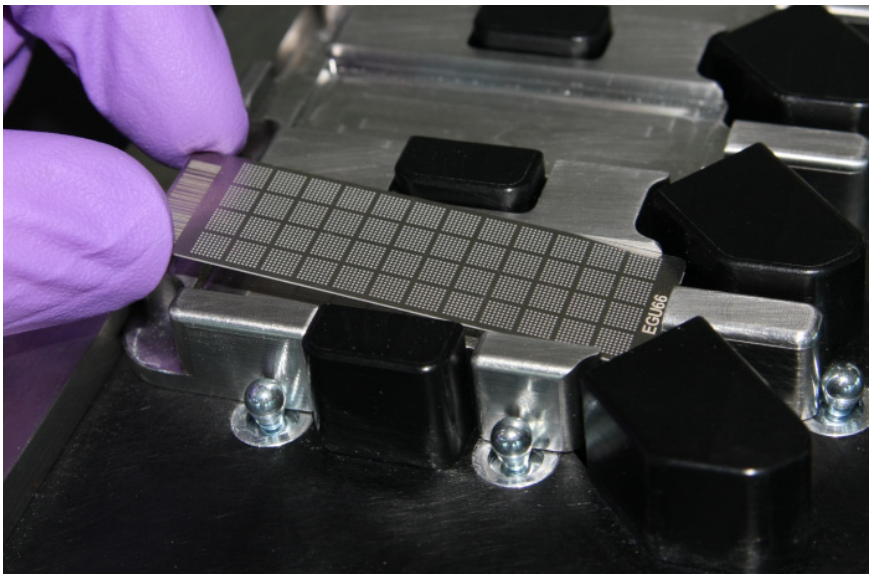
Workflow
"Insert the loaded OpenArray® plate into a case" (page 42)
↓
"Seal the OpenArray® plate" (page 43)
↓
"Frame the case" (page 45)

Insert the loaded OpenArray® plate into a case

IMPORTANT! Immersion fluid in OpenArray cases must not exposed to air for more than 60 minutes. Make sure that the immersion fluid is degassed. Bubbles in a case negatively affect data quality.

OpenArray cases are included with the OpenArray Case Sealing Station. The front of cases is framed in black; the back of the cases is gray. To insert the loaded OpenArray plate into the case:

1. Gently lift the OpenArray plate from the Plate Holder with the thumb and index finger of your left hand, placing your fingers on each side near the end with the barcode. Keep the side with the barcode facing you. Switch hands and now hold the OpenArray plate by the sides using your index and thumb of your right hand.



2. Hold the case at the top with the thumb and index finger of your left hand so that the black framed side is facing you. Slide the OpenArray plate into the case so that it sits in the rails along both inside edges of the case.

3. Slide the OpenArray plate all the way down into the case. The bottom of the OpenArray should touch the bottom of the case and have approximately 1 cm (0.39 in.) of space at the top.

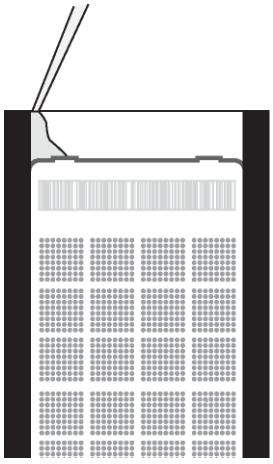
4. If necessary, adjust the fluid level so it is at the top of the OpenArray plate. Remove any excess immersion fluid with a transfer pipette.

Seal the OpenArray® plate

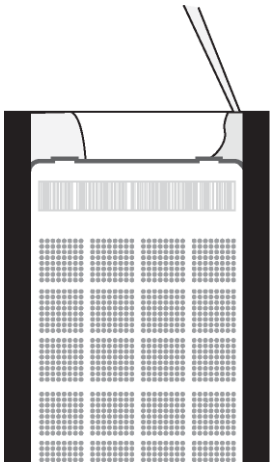
Seal the OpenArray plate in the case with ultraviolet (UV) glue:

Note: If your tube of glue has been open longer than two weeks, discard it and use a new tube. Store glue in a dark place (for example, a drawer) because ambient light can cure the glue in the tip.

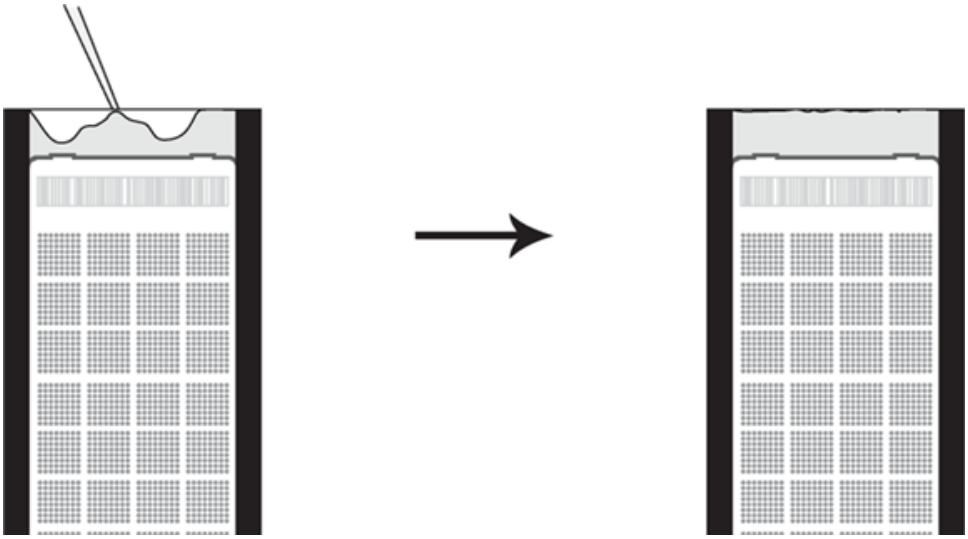
1. Place a drop of UV glue on one edge of the case opening; fill until it reaches the top of the OpenArray plate. Angle the pipette tip so that the glue reaches the inside rail, as shown:



2. Place a similar amount of glue on the other side of the case opening, as shown:



3. Add glue on each side of the case until the glue runs together in the middle. Fill the case to the top. Make sure both the left and right sides are covered with glue, as shown:



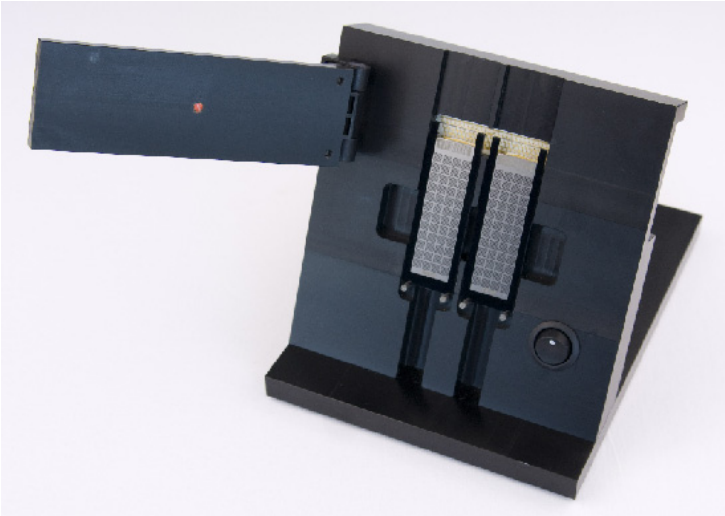
4. If there are air bubbles in the glue, carefully remove them by inserting a small transfer pipette.

5. With a laboratory wipe, wipe any excess glue from the surface of the glass.

6. To cure the glue, place the OpenArray case into the Case Sealing Station:
   a. Open the door. Place up to two cases into the device and close the door.
   b. Turn the switch to **ON**. Allow the glue to cure, then turn the switch to **OFF**.
   c. Open the door, turn the cases over so the other side faces the light. Cure the glue again. Remove the OpenArray cases.

Instrument	Product	Curing time
OpenArray NT Imager	TaqMan® OpenArray Genotyping case	2 minutes
OpenArray NT Cycler	OpenArray Real-Time PCR case	3 minutes each side

The cases with OpenArray plates in the Case Sealing Station are shown:



Note: The Case Sealing Station emits UV light as indicated on the label (not shown above). Keep at least 25 cm (10 in.) away from the light when operating the Case Sealing Station.

7. If there is glue on the side of the case, carefully remove it with a razor blade.

8. Clean the case with a laboratory wipe that has been thoroughly sprayed with ethanol. To dry the case, wipe the case downward with a clean laboratory wipe. Gently handle the case; be sure to not apply pressure on the OpenArray plate within the case.

   Note: Dust, glue, or excess sample on the case may interfere with thermal uniformity and can fluoresce. Make sure you thoroughly clean each case.

Frame the case

To ensure that the case achieves tight contact with the OpenArray NT Cycler thermal block, you must adhere a frame with an adhesive pad to the OpenArray case.

Note: Framing the case does not apply to TaqMan OpenArray Genotyping Analysis kits nor does it apply to the OpenArray NT Imager.

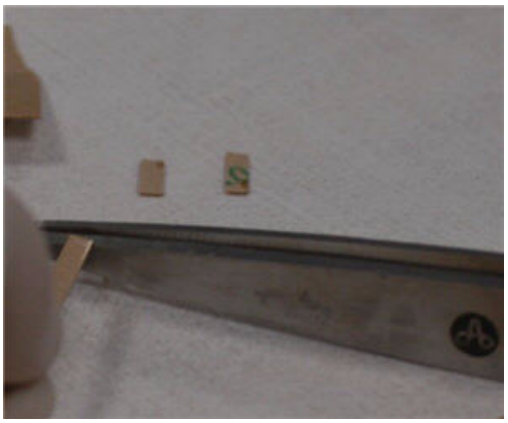
1. Position the OpenArray plate and case on a laboratory wipe, so that the black framed side faces up.

2. Make sure the frame is clean, without any residual adhesive from previous runs.

3. Use scissors to cut two rectangles from the frame adhesive.

   IMPORTANT! Use scissors that are clean and also not rusty.

Cut two rectangles, each about 3 mm in length, from the frame adhesive, as shown:



4. With forceps, remove the backing from one side of the tape of one rectangle, exposing a sticky side. Stick the tape in the middle of a long side of the frame.

5. Repeat *step 4* to place adhesive on the other, long side of the assembly.

6. With forceps, peel the backing off the pieces of tape. Align two corners of the frame with two corners of the case and bring the other corners of the frame down onto the case. Flatten the frame against the case for 10 seconds.

The OpenArray plate is now sealed, framed, and ready to insert in the OpenArray NT Cycler.

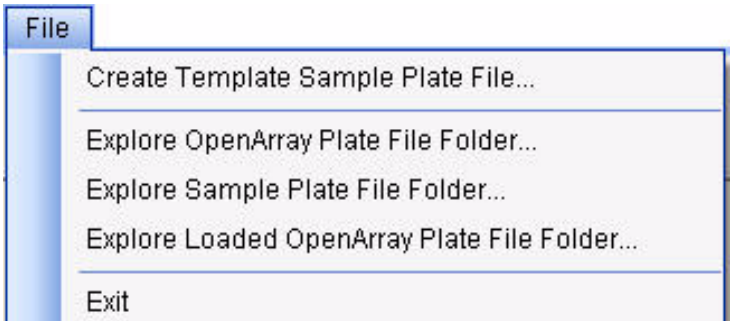
5 OpenArray® AccuFill™ Software

This chapter covers:

Overview

This chapter describes the menus available in the OpenArray® AccuFill™ software and the commands available on them.

The File menu



Create Template Sample Plate File Folder

This command creates a sample plate file in CSV format that you can open in a spreadsheet program such as Microsoft Excel® software. After you select the command, type the file name in the Save As window, then click **OK**.

Explore OpenArray Plate File Folder

This command opens the current OpenArray plate file folder in a Windows Explorer window. The default location of the plate file folder is `C:\OpenArray\OpenArray Plates`.

Explore Sample Plate File Folder

This command opens the current sample plate file folder in a Windows Explorer window. The default location of the plate file folder is `C:\OpenArray\Sample Plates`.

Explore Loaded OpenArray Plate File Folder

This command opens the current OpenArray plate file folder in a Windows Explorer window. The default location of the plate file folder is `C:\OpenArray\Loaded OpenArray Plates`.

The Instrument menu

Setup and Load OpenArray Plates

This command launches the Setup and Load function. Select this menu item when you want to start a run. It is similar to the Setup and Load button.

Cancel Current Operation

This command stops the current loading operation. The menu item is active only when you have started a run.

Show Deck

This command returns the view to the instrument deck after you have selected another view such as the Diagnostics screen.

Show Loading History

This command opens the Loading History window. From the Loading History, you can:

- View the record of past loading runs.
- Click the **Copy All** button to copy the information to an external spreadsheet program.
- Click **Save As** to save the information to a CSV file.

Show Instrument Log

This command displays the instrument log on the right side of the screen. The log contains a detailed list of each event that happens on the instrument. Its columns include:

- Number – sequential numbering of the events
- Date – the date and time the event took place
- Message – a description of this event

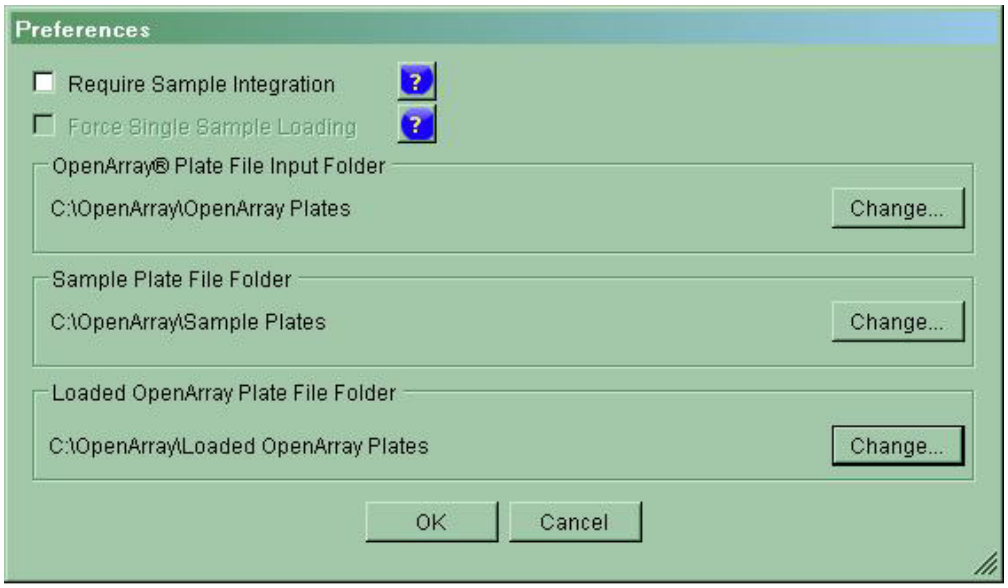
To close the instrument log, select **Hide Instrument Log** from the Instrument menu.

Run System Self Test

This command displays a message telling you to clear the deck, empty the waste bin, and close the door before moving forward with the System Self Test. After you have completed all those tasks, clicking Proceed starts the system self test. The test runs and displays its progress in a System Self Test window. When the test is done, the window closes, and you can proceed with normal operation.

Edit Preferences

This command opens the Preference window that enables you to establish settings for several software options.



The software options are:

- **Require Sample Integration** – When you select this option, Sample Integration is automatically selected in the Setup Load Information window and you cannot change that selection.
- **Force Single Sample Loading** – When you select Force Single Sample Loading, Single Sample Integration is automatically selected in the Setup Load Information window and you cannot change that selection. With Single Sample Loading, the same sample is loaded into each of the 64 through-holes that make up a subarray of the OpenArray plate, even when the OpenArray plate is formatted for 2 or 3 samples per subarray. This method is a convenient way to create technical replicates for analysis.

Help

The Help menu contains information about the OpenArray AccuFill System including a summary of the system, serial numbers, and versions of software and firmware. A link to the Software License Agreement is also provided.

A Ordering Information

This appendix covers:

How to order from the Applied Biosystems website

To order...	Procedure
Instrument parts and accessories	1. Go to **www.appliedbiosystems.com**. 2. Click **Products ▸ Gene Expression ▸ Gene Expression Analysis Instruments ▸ OpenArray® Real-Time PCR Platform**.
Reagents	In the *OpenArray Real-Time PCR Platform* page, click **Related Products**.
Consumables	

Required equipment

The OpenArray AccuFill System (PN 4457243) includes the following components supplied by Applied Biosystems:

Item	Part Number
OpenArray® AccuFill™ Instrument	4457244
OpenArray AccuFill Software	4457245
Computer	4398961
Monitor	4404414

Required equipment and materials not included

The following items are not supplied:

Item	Source
TaqMan® OpenArray Real-Time PCR Platform	Applied Biosystems, PN 4408510
OpenArray NT Cycler System	Applied Biosystems, PN 4453106
OpenArray NT Imager	Applied Biosystems, PN 4412491
OpenArray Case Sealing Station	Applied Biosystems, PN 4409361
Uninterruptible Power Supply (UPS)	Franek PN: 5135-003 (recommended)
Electrical power strips	–
Bleach (10%)	–
DNA-ExitusPlus™	AppliChem, PN A7089
Ethanol	–
Compressed nitrogen gas	–

Optional materials

Item	Source
Barcode reader	–
Fine-point silver marker	–

Required reagents

Reagents required for an OpenArray Real-Time PCR plate run:

Item	Source
Hi-Di™ Formamide	Applied Biosystems, PN 4311320
Pluronic® F-68 (10% stock)	Invitrogen, PN 24040-032
LightCycler® FastStart DNA Master, SYBR® Green I kit	Roche Applied Science, PN 12239264001
Albumin, bovine (BSA)	Sigma, PN A7906
SYBR® Green I	Sigma, PN S9430
Glycerol, 99%	Sigma, PN G5150

Reagents required for a TaqMan OpenArray plate run:

Item	Source
5× TaqMan OpenArray® REMix Solution	Applied Biosystems, PN 4453927
GeneAmp® Fast PCR Master Mix	Applied Biosystems, PN 4362070
Albumin, bovine (BSA)	Sigma, PN A7906

Required consumables

Item	Source
OpenArray 384-Well Sample Plates	Applied Biosystems, PN 4453929
OpenArray AccuFill Loader Tips	Applied Biosystems, PN 4457246
TaqMan OpenArray Genotyping Accessories Kit – or – OpenArray Real-Time PCR Accessories Kit	Applied Biosystems, PN 4404572 – or – Applied Biosystems, PN 4453975
Pipettes	MLS
Forceps	MLS
Scissors	MLS
Sealing tape	MLS

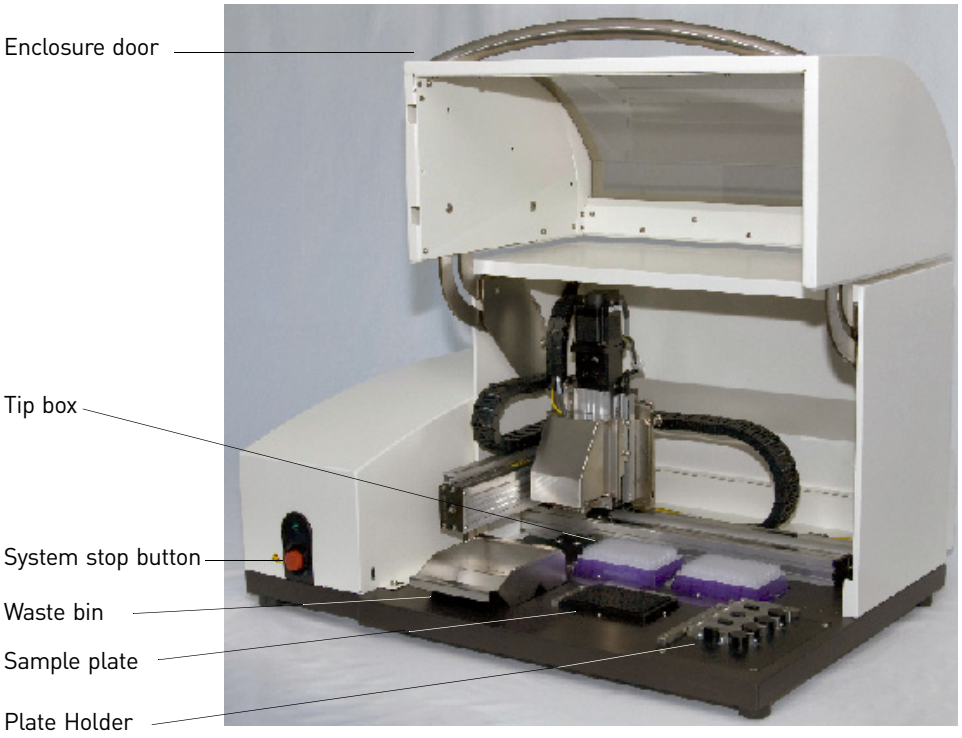
B System Overview

This appendix covers:

OpenArray® AccuFill™ System

The OpenArray® AccuFill™ System (PN 4457243) is an automated system for loading samples from 384-well plates to OpenArray plates. The OpenArray AccuFill system draws the master mix and sample across the surface of an OpenArray plate. The solution does not need to be dispensed in specific volumes because the holes are self-metering. Nor does the solution need to be forced into the holes. The same coating process that holds liquid in the through-holes allows the liquid to be drawn in by capillary action. Because there are no closed wells, air bubbles are not trapped during the dispensing process.

The OpenArray® AccuFill™ instrument and its component parts are shown:



Available run types

The loading procedure is optimized for the downstream Real-Time PCR application on the OpenArray platform. You may choose to perform qPCR using one of the following available chemistries: SYBR®, OpenArray Real-Time PCR, TaqMan® OpenArray Real-Time PCR, or TaqMan OpenArray Genotyping. Use only the chemistry that your application requires.

- For SYBR OpenArray Real-Time PCR runs (with OpenArray SYBR® Real-Time PCR Plates), see "Prepare sample plate for a SYBR® OpenArray® real-time PCR run" on page 10.
- For TaqMan OpenArray Real-Time PCR runs (with TaqMan OpenArray Real-Time PCR Plates), see "Prepare sample plate for a TaqMan® OpenArray® real-time PCR run" on page 13.
- For TaqMan OpenArray Genotyping runs (with TaqMan OpenArray Genotyping Plates), see "Prepare sample plate for a TaqMan® OpenArray® genotyping run" on page 15.

C Cleaning the OpenArray® AccuFill™ instrument

This appendix covers:

Cleaning instructions

After you have finished your experiment, clean the OpenArray® AccuFill™instrument.

Clean the outside of the instrument

Clean the outside of the OpenArray AccuFill instrument by wiping it with a clean, dry cloth. **Do not use solvents.**

Clean the inside of the instrument

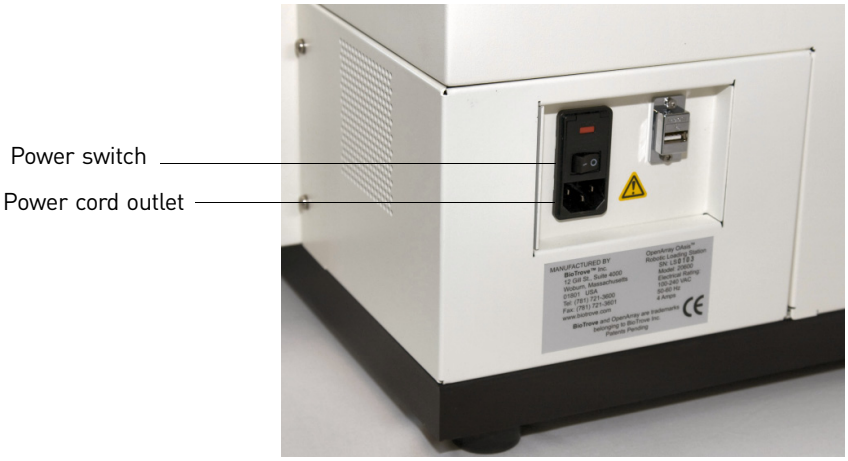
Note: For chemical hazard alerts, see Appendix D, Safety.

Inside the AccuFill instrument, the waste bin and Plate Holder are removable parts that you can clean. To clean the parts:

1. Remove the waste bin and Plate Holder from the deck of the AccuFill instrument.
2. Spray the parts with DNA-ExitusPlus™ and wait 10 minutes.
3. Rinse the parts with tap water to wash away the solution.
4. Rinse the parts with ethanol.
5. Allow the parts to dry for 5 minutes before using them again. Make sure the parts are *completely dry* before loading the next plate.

Spill accidents

If liquids or other materials spill inside the AccuFill instrument, turn off the instrument by pressing the power switch on the left side of the instrument. Unplug the power cord from the electrical outlet. Contact Applied Biosystems service personnel. For the latest service and support information, go to **www.appliedbiosystems.com**.



D

Safety

This appendix covers:

Instrumentation safety

Symbols on instruments

Electrical symbols on instruments

The following table describes the electrical symbols that may be displayed on Applied Biosystems instruments.

Symbol	Description
	Indicates the **On** position of the main power switch.
	Indicates the **Off** position of the main power switch.
	Indicates a standby switch by which the instrument is switched on to the **Standby** condition. Hazardous voltage may be present if this switch is on standby.
	Indicates the **On/Off** position of a push-push main power switch.
	Indicates a terminal that may be connected to the signal ground reference of another instrument. This is not a protected ground terminal.
	Indicates a protective grounding terminal that must be connected to earth ground before any other electrical connections are made to the instrument.
	Indicates a terminal that can receive or supply alternating current or voltage.
	Indicates a terminal that can receive or supply alternating or direct current or voltage.

Safety symbols

The following table describes the safety symbols that may be displayed on Applied Biosystems instruments. Each symbol may appear by itself or with text that explains the relevant hazard (see "Safety labels on instruments" on page 8). These safety symbols may also appear next to DANGERS, WARNINGS, and CAUTIONS that occur in the text of this and other product-support documents.

Symbol	Description
	Indicates that you should consult the manual for further information and to proceed with appropriate caution.
	Indicates the presence of an electrical shock hazard and to proceed with appropriate caution.
	Indicates the presence of a hot surface or other high-temperature hazard and to proceed with appropriate caution.

Environmental symbols on instruments

The following symbol applies to all Applied Biosystems electrical and electronic products placed on the European market after August 13, 2005.

Symbol	Description
	**Do not dispose of this product as unsorted municipal waste.** Follow local municipal waste ordinances for proper disposal provisions to reduce the environmental impact of waste electrical and electronic equipment (WEEE). **European Union customers:** Call your local Applied Biosystems Customer Service office for equipment pick-up and recycling. See **www.appliedbiosystems.com** for a list of customer service offices in the European Union.

Locations of safety labels on instruments

The caution label shown below is located on the OpenArray® AccuFill™ instrument in two places:

- Near the power switch on the left side of the instrument
- Near the power inlet

General instrument safety

WARNING! PHYSICAL INJURY HAZARD. Using the instrument in a manner not specified by Applied Biosystems may result in personal injury or damage to the instrument.

WARNING! PHYSICAL INJURY HAZARD. For safety information related to the centrifuge and thermal cycler, refer to the manufacturer's documentation.

WARNING! PHYSICAL INJURY HAZARD. Using the centrifuge or thermal cycler in a manner not specified by the manufacturer may result in personal injury or damage to the instrument.

Move and lift the instrument

CAUTION! PHYSICAL INJURY HAZARD. The instrument is to be moved and positioned only by the personnel or vendor specified in the applicable site preparation guide. If you decide to lift or move the instrument after it has been installed, do not attempt to lift or move the instrument without the assistance of others, the use of appropriate moving equipment, and proper lifting techniques. Improper lifting can cause painful and permanent back injury. Depending on the weight, moving or lifting an instrument may require two or more persons.

Move and lift stand-alone computers and monitors

WARNING! Do not attempt to lift or move the computer or the monitor without the assistance of others. Depending on the weight of the computer and/or the monitor, moving them may require two or more people.

Things to consider before lifting the computer and/or the monitor:

- Make sure that you have a secure, comfortable grip on the computer or the monitor when lifting.
- Make sure that the path from where the object is to where it is being moved is clear of obstructions.
- Do not lift an object and twist your torso at the same time.
- Keep your spine in a good neutral position while lifting with your legs.
- Participants should coordinate lift and move intentions with each other before lifting and carrying.
- Instead of lifting the object from the packing box, carefully tilt the box on its side and hold it stationary while someone slides the contents out of the box.

Operate the instrument

Ensure that anyone who operates the instrument has:

- Received instructions in both general safety practices for laboratories and specific safety practices for the instrument.
- Read and understood all applicable Safety Data Sheets (SDSs). See "About SDSs" on page 73.

Clean or decontaminate the instrument

CAUTION! Before using a cleaning or decontamination method other than those recommended by the manufacturer, verify with the manufacturer that the proposed method will not damage the equipment.

Physical hazard safety

Ultraviolet light

WARNING! ULTRAVIOLET LIGHT HAZARD. Looking directly at a UV light source can cause serious eye damage. Never look directly at a UV light source and always prevent others from UV exposure. Follow the manufacturer's recommendations for appropriate protective eyewear and clothing.

Compressed gases

WARNING! PHYSICAL HAZARD. Nonflammable compressed nitrogen gas. Contents are under pressure. Receive proper training on the handling of compressed gases before use. Exposure to rapidly expanding gas may cause frostbite. High concentrations of vapors in the immediate area can displace oxygen and cause asphyxiation. Use only in areas with adequate ventilation. Read the SDS, and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

WARNING! EXPLOSION HAZARD. Pressurized gas cylinders are potentially explosive and can cause severe injury if not handled properly. Always cap the gas cylinder when it is not in use and attach it firmly to the wall or gas cylinder cart with approved brackets or chains.

Moving parts

WARNING! PHYSICAL INJURY HAZARD. Moving parts can crush and cut. Keep hands clear of moving parts while operating the instrument. Disconnect power before servicing the instrument.

WARNING! PHYSICAL INJURY HAZARD. Do not operate the instrument without the arm shield in place. Keep hands out of the deck area when the instrument is spotting.

Solvents and pressurized fluids

WARNING! PHYSICAL INJURY HAZARD. Always wear eye protection when working with solvents or any pressurized fluids.

WARNING! PHYSICAL INJURY HAZARD. To avoid hazards associated with high-pressure fluids in polymeric tubing:

- Be aware that PEEK™ tubing is a polymeric material. Use caution when working with any polymer tubing that is under pressure.
- Always wear eye protection when near pressurized polymer tubing.
- Extinguish all nearby flames if you use flammable solvents.
- Do not use PEEK tubing that has been severely stressed or kinked.
- Do not use PEEK tubing with tetrahydrofuran or nitric and sulfuric acids.
- Be aware that methylene chloride and dimethyl sulfoxide cause PEEK tubing to swell and greatly reduce the rupture pressure of the tubing.
- Be aware that high solvent flow rates (~40 mL/min) may cause a static charge to build up on the surface of the tubing. Electrical sparks may result.

Electrical safety

WARNING! ELECTRICAL SHOCK HAZARD. Severe electrical shock can result from operating the AccuFill without its instrument panels in place. Do not remove instrument panels. High-voltage contacts are exposed when instrument panels are removed from the instrument.

Fuses

WARNING! FIRE HAZARD. Improper fuses or high-voltage supply can damage the instrument wiring system and cause a fire. Before turning on the instrument, verify that the fuses are properly installed and that the instrument voltage matches the power supply in your laboratory.

WARNING! FIRE HAZARD. For continued protection against the risk of fire, replace fuses only with fuses of the type and rating specified for the instrument.

Power

WARNING! ELECTRICAL HAZARD. Grounding circuit continuity is required for the safe operation of equipment. Never operate equipment with the grounding conductor disconnected.

WARNING! ELECTRICAL HAZARD. Use properly configured and approved line cords for the voltage supply in your facility.

WARNING! ELECTRICAL HAZARD. Plug the system into a properly grounded receptacle with adequate current capacity.

Overvoltage rating

The AccuFill system has an installation (overvoltage) category of II, and is classified as portable equipment.

Workstation safety

Correct ergonomic configuration of your workstation can reduce or prevent effects such as fatigue, pain, and strain. Minimize or eliminate these effects by configuring your workstation to promote neutral or relaxed working positions.

CAUTION! MUSCULOSKELETAL AND REPETITIVE MOTION HAZARD. These hazards are caused by potential risk factors that include but are not limited to repetitive motion, awkward posture, forceful exertion, holding static unhealthy positions, contact pressure, and other workstation environmental factors.

To minimize musculoskeletal and repetitive motion risks:

- Use equipment that comfortably supports you in neutral working positions and allows adequate accessibility to the keyboard, monitor, and mouse.
- Position the keyboard, mouse, and monitor to promote relaxed body and head postures.

Safety and electromagnetic compatibility (EMC) standards

This section provides information on:

- U.S. and Canadian safety standards
- Canadian EMC standard
- European safety and EMC standards
- Australian EMC Standards

The OpenArray AccuFill instrument has been tested to, and complies, with the standards listed below.

U.S. and Canadian safety standards

UL 61010-1:2004, 2nd Edition/CSA-C22.2 No. 61010-1, "Safety Requirements for Electrical Equipment for Measurement, Control and Laboratory Use: Part 1: General Requirements."

ANSI/UL 61010-1,"Safety Requirements for Electrical Equipment for Measurement, Control and Laboratory Use -Part 1: General Requirements."

CAN/CSA-C22.2 #61010-1, "Safety Requirements for Electrical Equipment for Measurement, Control and Laboratory Use -Part 1: General Requirements."

Canadian EMC standard

The OpenArray AccuFill instrument has been tested to, and complies with, ICES-001, Issue 3: "Industrial, Scientific, and Medical Radio Frequency Generators."

European safety and EMC standards

Safety

The OpenArray AccuFill instrument meets European requirements for safety (2006/95/EC). This instrument has been tested to and complies with standards EN 61010-1:2001 (second edition), "Safety Requirements for Electrical Equipment for Measurement, Control and Laboratory Use, Part 1: General Requirements."

EMC

This instrument meets European requirements for emission and immunity (EMC Directive 2004/108/EC). This instrument has been tested to and complies with standard IEC 61326 (Group 1, Class A), "Electrical Equipment for Measurement, Control and Laboratory Use – EMC Requirements."

Australian EMC Standards

The OpenArray AccuFill instrument has been tested to and complies with standard AS/NZS 2064, "Limits and Methods Measurement of Electromagnetic Disturbance Characteristics of Industrial, Scientific, and Medical (ISM) Radio-frequency Equipment."

Chemical safety

General chemical safety

Chemical hazard warning

WARNING! CHEMICAL HAZARD. Before handling any chemicals, refer to the Safety Data Sheet (SDS) provided by the manufacturer, and observe all relevant precautions.

WARNING! CHEMICAL HAZARD. All chemicals in the instrument, including liquid in the lines, are potentially hazardous. Always determine what chemicals have been used in the instrument before changing reagents or instrument components. Wear appropriate eyewear, protective clothing, and gloves when working on the instrument.

WARNING! CHEMICAL HAZARD. Four-liter reagent and waste bottles can crack and leak. Each 4-liter bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position. Wear appropriate eyewear, clothing, and gloves when handling reagent and waste bottles.

WARNING! CHEMICAL STORAGE HAZARD. Never collect or store waste in a glass container because of the risk of breaking or shattering. Reagent and waste bottles can crack and leak. Each waste bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position. Wear appropriate eyewear, clothing, and gloves when handling reagent and waste bottles.

Chemical safety guidelines

To minimize the hazards of chemicals:

- Read and understand the Safety Data Sheets (SDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. (See "About SDSs" on page 73.)
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing). For additional safety guidelines, consult the SDS.

- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with adequate ventilation (for example, fume hood). For additional safety guidelines, consult the SDS.
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer's cleanup procedures as recommended in the SDS.
- Comply with all local, state/provincial, or national laws and regulations related to chemical storage, handling, and disposal.

SDSs

About SDSs

Chemical manufacturers supply current Safety Data Sheets (SDSs) with shipments of hazardous chemicals to new customers. They also provide SDSs with the first shipment of a hazardous chemical to a customer after an SDS has been updated. SDSs provide the safety information you need to store, handle, transport, and dispose of the chemicals safely.

Each time you receive a new SDS packaged with a hazardous chemical, be sure to replace the appropriate SDS in your files.

Obtain SDSs

The SDS for any chemical supplied by Applied Biosystems is available to you free 24 hours a day. To obtain SDSs:

1. Go to **www.appliedbiosystems.com**, click **Support**, then select **SDS**.
2. In the Keyword Search field, enter the chemical name, product name, SDS part number, or other information that appears in the SDS of interest. Select the language of your choice, then click **Search**.
3. Find the document of interest, right-click the document title, then select any of the following:
   - **Open** – To view the document
   - **Print Target** – To print the document
   - **Save Target As** – To download a PDF version of the document to a destination that you choose

Note: For the SDSs of chemicals not distributed by Applied Biosystems, contact the chemical manufacturer.

Chemical waste safety

Chemical waste hazards

CAUTION! HAZARDOUS WASTE. Refer to Safety Data Sheets and local regulations for handling and disposal.

WARNING! CHEMICAL WASTE HAZARD. Wastes produced by Applied Biosystems instruments are potentially hazardous and can cause injury, illness, or death.

WARNING! CHEMICAL STORAGE HAZARD. Never collect or store waste in a glass container because of the risk of breaking or shattering. Reagent and waste bottles can crack and leak. Each waste bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position. Wear appropriate eyewear, clothing, and gloves when handling reagent and waste bottles.

Chemical waste safety guidelines

To minimize the hazards of chemical waste:

- Read and understand the Safety Data Sheets (SDSs) provided by the manufacturers of the chemicals in the waste container before you store, handle, or dispose of chemical waste.
- Provide primary and secondary waste containers. (A primary waste container holds the immediate waste. A secondary container contains spills or leaks from the primary container. Both containers must be compatible with the waste material and meet federal, state, and local requirements for container storage.)
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing). For additional safety guidelines, consult the SDS.
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with adequate ventilation (for example, fume hood). For additional safety guidelines, consult the SDS.
- Handle chemical wastes in a fume hood.
- After emptying a waste container, seal it with the cap provided.
- Dispose of the contents of the waste tray and waste bottle in accordance with good laboratory practices and local, state/provincial, or national environmental and health regulations.

Waste disposal

If potentially hazardous waste is generated when you operate the instrument, you must:

- Characterize (by analysis if necessary) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure the health and safety of all personnel in your laboratory.
- Ensure that the instrument waste is stored, transferred, transported, and disposed of according to all local, state/provincial, and/or national regulations.

IMPORTANT! Radioactive or biohazardous materials may require special handling, and disposal limitations may apply.

Biological hazard safety

General biohazard

WARNING! BIOHAZARD. Biological samples such as tissues, body fluids, infectious agents, and blood of humans and other animals have the potential to transmit infectious diseases. Follow all applicable local, state/provincial, and/or national regulations. Wear appropriate protective equipment, which includes but is not limited to: protective eyewear, face shield, clothing/lab coat, and gloves. All work should be conducted in properly equipped facilities using the appropriate safety equipment (for example, physical containment devices). Individuals should be trained according to applicable regulatory and company/institution requirements before working with potentially infectious materials. Read and follow the applicable guidelines and/or regulatory requirements in the following:

- U.S. Department of Health and Human Services guidelines published in *Biosafety in Microbiological and Biomedical Laboratories* (**www.cdc.gov/od/ohs/biosfty/bmbl5/bmbl5toc**)
- Occupational Safety and Health Standards, Bloodborne Pathogens (29 CFR§1910.1030; **www.access.gpo.gov/ nara/cfr/waisidx_01/29cfr1910a_01.html**).
- Your company's/institution's Biosafety Program protocols for working with/handling potentially infectious materials.

Additional information about biohazard guidelines is available at:

www.cdc.gov

Safety alerts

For the definitions of the alert words **IMPORTANT**, **CAUTION**, **WARNING**, and **DANGER**, see "Safety alert words" on page 7.

General alerts for all chemicals

Avoid contact with (skin, eyes, and/or clothing). Read the Safety Data Sheets (SDSs), and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

Specific alerts for instrumentation

WARNING! PHYSICAL INJURY HAZARD. Moving parts can crush and cut. Keep hands clear of moving parts while operating the instrument.

WARNING! ULTRAVIOLET LIGHT HAZARD. Looking directly at a UV light source can cause serious eye damage. Never look directly at a UV light source. Follow the manufacturer's recommendations for the appropriate eyewear and clothing.

CAUTION! PHYSICAL INJURY HAZARD. Do not attempt to lift or move the OpenArray® AccuFill™ instrument without the assistance of others, the use of appropriate moving equipment, and proper lifting techniques. Improper lifting can cause painful and permanent back injury. Depending on the weight, moving or lifting an instrument may require two or more persons.

CAUTION! Do not tip the OpenArray® AccuFill instrument on end. Tipping damages the instrument hardware and electronics and is an unsafe practice.

Documentation and Support

Related documentation

The following related documents are shipped with the system:

Document	Part number	Description
OpenArray® AccuFill™ Site Preparation Guide	4457095	Provides information for setting up the OpenArray AccuFill System.
OpenArray AccuFill Quick Reference Card	4457450	Provides brief, step-by-step procedures to help you quickly learn to use the OpenArray AccuFill System.

The following document provides supplementary instructions:

Document	Part number	Description
TaqMan® OpenArray Genotyping System User Guide	4377476	Provides information about the TaqMan OpenArray Genotyping System, including step-by-step procedures to: • Prepare a TaqMan OpenArray Genotyping Plate, using the OpenArray Autoloader and OpenArray Case Sealing Station. • Run a TaqMan OpenArray plate on the OpenArray NT Imager, then analyze the data with the OpenArray SNP Genotyping Analysis Software. • Maintain the TaqMan OpenArray Genotyping Instrument Platform.

Obtaining support

To obtain additional documentation and the latest services and support information for all locations, go to:

www.appliedbiosystems.com

At the Applied Biosystems web site, you can:

- Access worldwide telephone and fax numbers to contact Applied Biosystems Technical Support and Sales facilities.
- Search through frequently asked questions (FAQs).
- Submit a question directly to Technical Support.
- Order Applied Biosystems user documents, SDSs, certificates of analysis, and other related documents.
- Download PDF documents.
- Obtain information about customer training.
- Download software updates and patches.

Glossary

case sealing station	Uses UV light to cross-link the UV-curable glue and seal the OpenArray® plates inside cases filled with Immersion Fluid.
CSV file	Comma-separated values file that contains sample data.
frames	Black, metal, curved supports that you affix to the OpenArray cases to ensure good contact with the thermal block on the OpenArray NT Cycler.
multi-sample loading	Refers to the process of loading two or three samples, each from different sections of the sample plate, into one subarray, broken into two or three distinct sections.
OpenArray NT Real-Time PCR instrument	Performs simultaneous thermal cycling and imaging of the OpenArray plates.
Plate Holder	Accurately positions the OpenArray plate for sample loading in the OpenArray AccuFill System. Additional Plate Holders can be ordered from Applied Biosystems.
sample DNA	The DNA from any source of interest (e.g., tissue, whole organism, cDNA library).
sample integration	Sample Integration connects the unique identifiers for each sample to their specific locations on an OpenArray plate. It does this using two files: • a CSV file that describes the 384-well Sample Plate stored in the Sample Plate File Folder • a TPF, SPF, or EPF file that describes the OpenArray plate stored in the Open Array Plate Input Folder After loading the sample, the OpenArray® AccuFill™ System generates an output file for each OpenArray plate in TPF format.
serial number	The unique alphanumeric plate identification number that is coded for each OpenArray plate. This number is visible opposite the barcode on the plate, and is tagged to the information contained within the plate file.
.spf file	A single nucleotide polymorphisms (SNP) plate file that is the output of Sample Integration. It is used to process an OpenArray plate in the OpenArray NT Cycler.
SYBR	SYBR® Green I Dye, a fluorescent dye that binds preferentially to double-stranded DNA. The optics in the OpenArray NT Cycler can detect the intensity of fluorescence emitted from bound SYBR Green I. Based on fluorescence monitoring from SYBR Green I, the OpenArray NT Cycler calculates the number of copies of target DNA.

subarray	The OpenArray plate is divided into 48 subarrays; each subarray consists of 64 through-holes in an 8x8 square configuration.
TaqMan®	A dual-labeled probe that is commonly used in laboratories for qPCR studies. The TaqMan® OpenArray Real-Time PCR Plates qPCR platform is ideally suited for TaqMan assays.
target DNA	The particular sequence of DNA in the sample in which your research is focused.
through-hole	Bottomless reaction wells in the OpenArray plate. There are 3072 through-holes in each OpenArray plate. Each through-hole contains a single assay. A self-metering loading system ensures a consistent 33-nL reaction volume within each through-hole. Proprietary plate coatings hold reagents within the through-holes.
.tpf file	A transcript plate file that is the output of Sample Integration. It is used to process an OpenArray plate in the OpenArray NT Cycler.

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