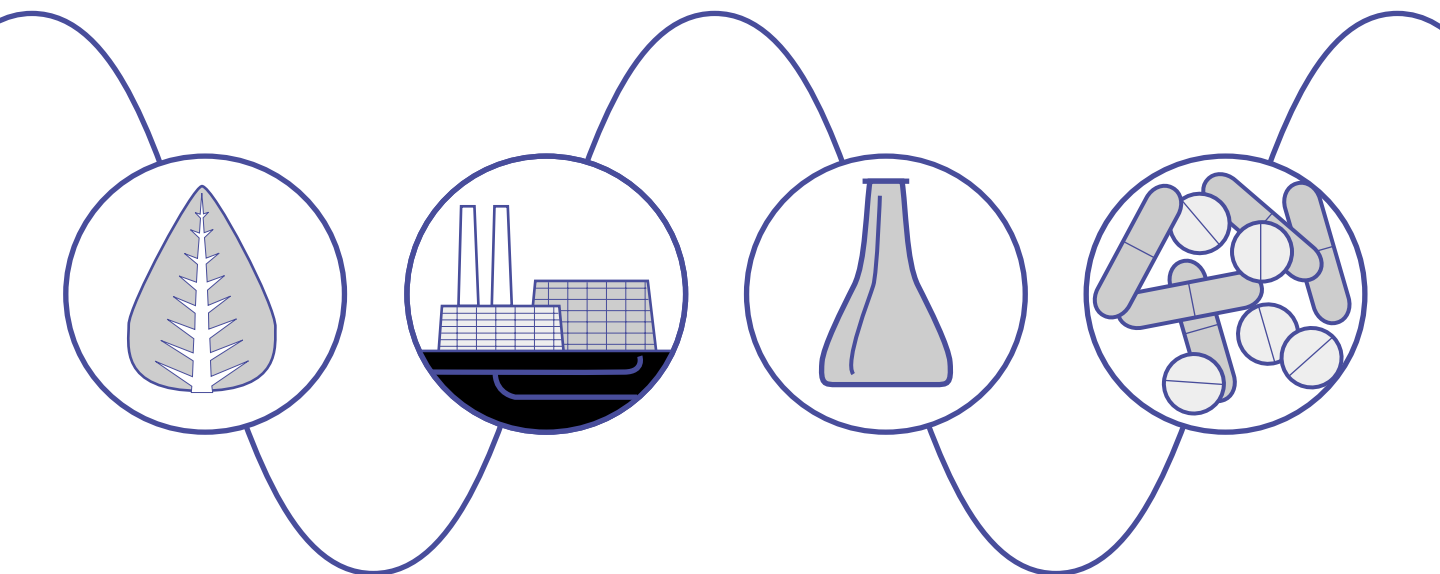


Waters 2695 Separations Module

Quick Start Guide



Waters

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71500269503, Revision A

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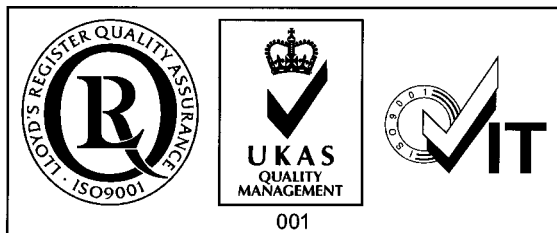
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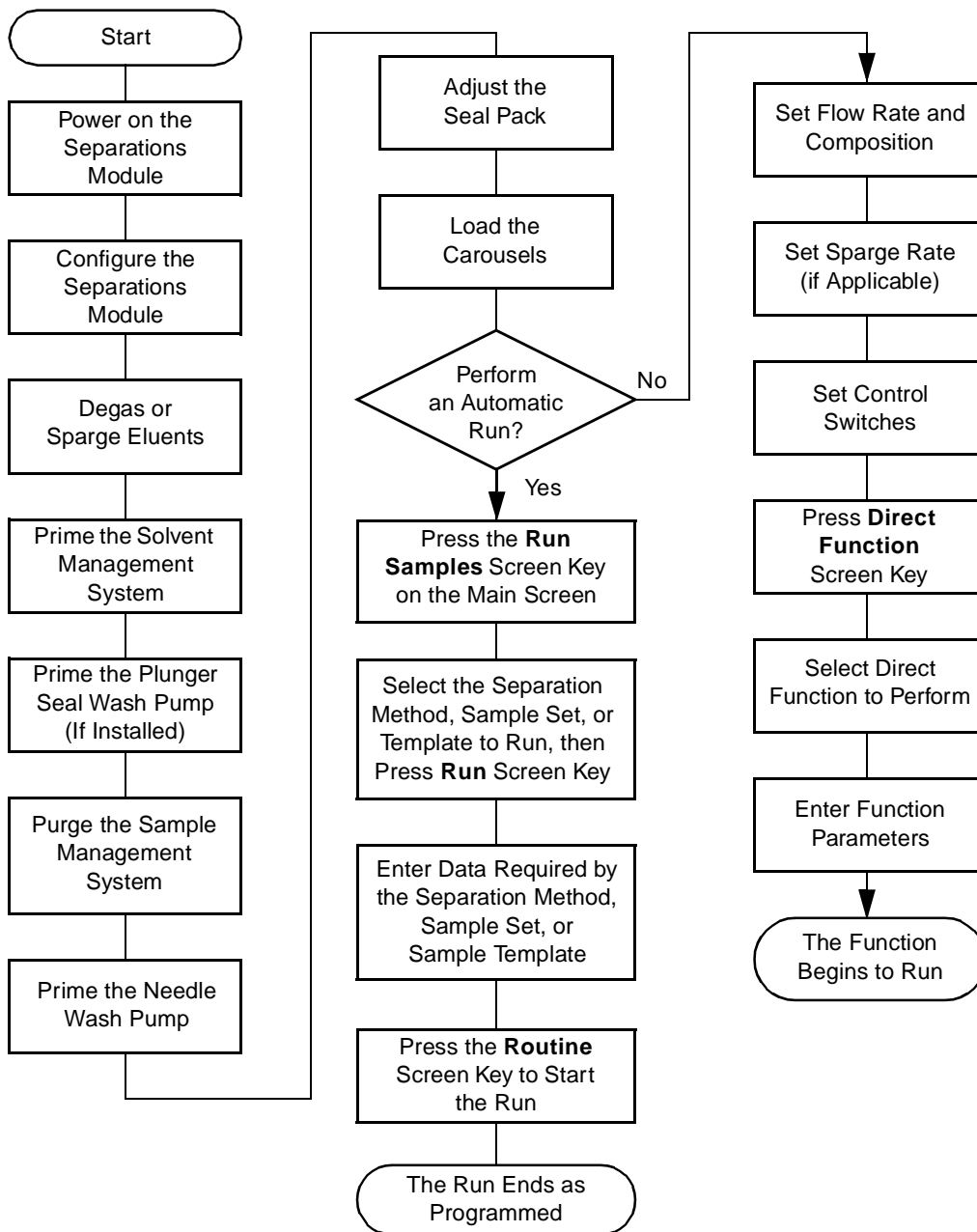
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Quick Summary

This flowchart summarizes the information contained in this Quick Start Guide. Use it as a reference after you have read the entire document.





Conventions Used in This Guide

This Quick Start Guide uses the following conventions:

- Bold text indicates user input or action. For example, press **Enter**.
- When you are instructed to “press the **X** key”, press the indicated keypad key.
- When you are instructed to “press the **X** screen key,” press the keypad key directly below the key name displayed on the screen.

Notes, Attentions, and Cautions

This guide uses the following Note, Attention, and Caution conventions:

- Notes call out information that is important to the operator. For example:

Note: *Record your result before you proceed to the next step.*

- Attentions provide information about preventing possible damage to the system or equipment. For example:



Attention: *To avoid damaging the detector flow cell, do not touch the flow cell window.*

- Cautions provide information essential to the safety of the operator. For example:



Caution: *To avoid chemical or electrical hazards, always observe safe laboratory practices when you operate your system.*



Caution: *To avoid electrical shock and possible injury, remove the power cord from the rear panel of the instrument before you perform the procedures in this section.*

Chapter 1

Introduction

The *Waters 2695 Separations Module Quick Start Guide* introduces you to the basic features of the Waters® 2695 Separations Module and describes how to make a run.

Who Should Use This Guide?

This guide is intended for both novice and experienced chromatographers who need to operate the Waters 2695 Separations Module.

What Is in This Guide?

The *Waters 2695 Separations Module Quick Start Guide* contains basic procedural information to help you set up the Waters 2695 Separations Module and make a run. In addition, this guide contains detailed flowcharts that provide Waters-recommended steps to prime, equilibrate, and purge both degasser and sparge-based 2695 Separations Modules. Refer to [Chapter 6, Recommended Preparation Procedures](#).

Note: *This guide is not designed to teach you chemistry and does not contain background or reference information.*

Refer to the *Waters 2695 Separations Module Operator's Guide* for additional information, or if you want to learn how to modify any of the procedures in this guide.

Note: *Additional information concerning regulatory matters may also be found in the front section of the Waters 2695 Separations Module Operator's Guide.*

Note: *With the purchase of the 2695 Separations Module, Waters supplies both this guide and the Waters 2695 Separations Module Operator's Guide in electronic format on a CD-ROM. To install and view these guides on your computer, refer to the Readme file on the CD-ROM.*

Before You Begin

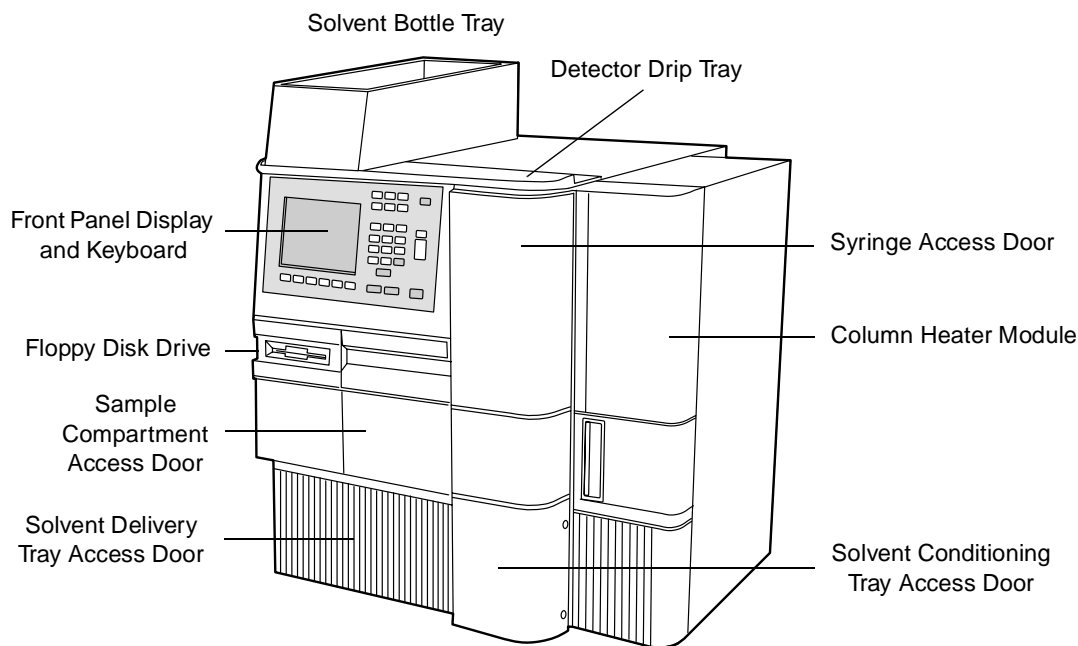
This guide assumes that:

- Your system is properly installed (refer to the *Waters 2695 Separations Module Operator's Guide*, Chapter 2, Installing the 2695 Separations Module).

- Your separation methods, sample sets, and/or sample templates are already created and stored (refer to the *Waters 2695 Separations Module Operator's Guide*, Chapter 6, Creating Methods, Sample Sets, and Sample Templates).

System Description

The Waters 2695 Separations Module ([Figure 1-1](#)) is an integrated solvent and sample management platform. This integration of two traditional high-performance liquid chromatography (HPLC) components streamlines all critical separation functions.



TP01354

Figure 1-1 Waters 2695 XE Separations Module (Front View)

The sample management system in the Waters 2695 Separations Module uses five carousels with a total capacity of 120 vials. A carrier rotates the carousels to the injection station in the sample compartment.

The Waters 2695 Separations Module is available from Waters in a number of configurations, differing from each other by the options that are included.

The two primary configurations are designated:

- **2695** – Provides quaternary solvent, high-performance solvent delivery, integral helium sparge for solvent conditioning, integral plunger seal-wash system, 120-vial

capacity sample management system, liquid-crystal display and keyboard user interface, and floppy disk drive.

- **2695 XE** – Provides the capabilities of the 2695 base product, replaces the helium sparge system with a four-channel inline vacuum degasser, and adds a column heater and a sample heater/cooler.

Chapter 2

Preparing the Separations Module for Operation

Before you can prepare the Separations Module for operation, you need to power on the 2695 Separations Module by moving the power switch (located at the top of the left side panel) to the **I** (On) position. The startup diagnostics routine begins.

2.1 Powering On

Startup Diagnostics

The startup diagnostics routine performs the following functions and tests:

- CPU board
- Memory (RAM and ROM)
- Keypad
- Display
- External communication
- Digital signal processor (DSP)
- Floppy disk drive

Once the electronic part of the diagnostics test is complete, the front-panel screen displays the results of the tests, as shown in [Figure 2-1](#).

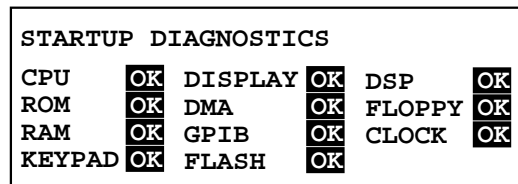


Figure 2-1 Diagnostic Test Results Screen

The Main Screen

When the initial part of the startup diagnostics routine is successful, the Main screen appears in the front panel display (Figure 2-2). The startup diagnostics routine continues by initializing the:

- Needle, syringe, and valves
- Carousel carrier
- Solvent management system

The statuses of these mechanical diagnostics tests appear in the banner area of the Main screen (Figure 2-2) while the diagnostics are running. If the startup diagnostics routine is not successful, refer to the *Waters 2695 Separations Module Operator's Guide*, Section 8.5, Troubleshooting. When the startup diagnostics routine is complete, the Separations Module enters the Idle mode.

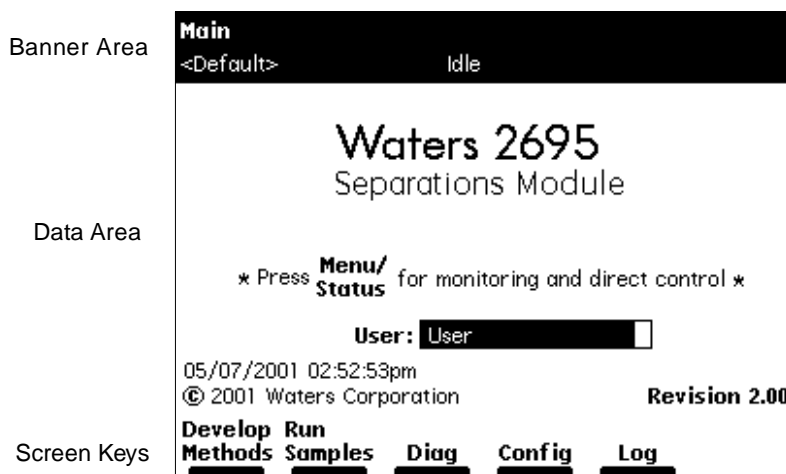


Figure 2-2 Main Screen

2.2 Configuring the Separations Module

Before you can operate the Separations Module, you need to configure the Separations Module for stand-alone or remote control and for a variety of other operating parameters.

No Interaction Mode

In this stand-alone mode, the Separations Module is not connected to the IEEE-488 interface bus. The Separations Module controls other non-IEEE devices in the HPLC system using the I/O connections on the rear panel. Refer to the *Waters 2695 Separations*

Module Operator's Guide, Section 2.7.1, I/O Signal Connections, for the procedure to make I/O connections. Use this mode if you want to suspend communication with a connected Millennium®³² Chromatography Manager workstation and operate the Separations Module and the other HPLC system components from their front panels.

To set the Separations Module to the No Interaction mode:

1. Press the **Config** screen key in the Main screen. The Configuration screen appears ([Figure 2-3](#)).
2. Select the **System** field, then press **Enter** to display a list of choices.
3. Select **No interaction**, then press **Enter**.
4. Press **Exit**.

System Controller Mode

In this stand-alone mode, the Separations Module controls up to three detector channels on the IEEE-488 bus. This can include two UV detector channels (Waters 2487 and/or 486 Tunable Absorbance Detectors) and one RI detector channel (Waters 2410 or 410 Differential Refractometer). You can also control other HPLC modules using the I/O connections.

To set the Separations Module to the System Controller mode:

1. Press the **Config** screen key in the Main screen. The Configuration screen appears ([Figure 2-3](#)).

The screenshot shows a 'Configuration' screen with a title bar containing '<Default>' and 'Idle'. The screen is divided into several sections:

- Syringe Size:** 250 μL
- Loop Size:** 100 μL
- Verify vial presence
- Verify carousel placement
- System:** Controlled by Millennium 32 [0]
- IEEE 488 Address:** 2 [0]
- Printer:** None [0]
- Baud Rate:** 1200
- Date Format:** MDY [0]
- Alarm Sound:** Continuous [0]
- Pressure Units:** psi [0]

At the bottom, there are several menu items: Reports, Comp Check, Status Layout, Auto-Shutdn, Events In, and More [0].

Figure 2-3 Configuration Screen

2. Select the **System** field, then press **Enter** to display a list of choices.
3. Select **System Controller**, then press **Enter**.

4. Press the **Detectors** screen key. A list of active devices (with their IEEE-488 interface bus addresses) appears.
 - a. Press the **Scan** screen key to update the list.
 - b. Press the **OK** screen key to return to the Configuration screen.
5. Press **Exit**. The Separations Module is ready to control Waters detectors using separation methods, sample sets, or sample templates.

Controlled by Millennium³² Mode

In this remote-control mode, a Millennium³² Chromatography Manager workstation running Millennium³² software controls the operation of your HPLC system using the IEEE-488 interface bus.

To set the Separations Module to the Controlled by Millennium³² mode:

1. Select the **System** field in the Configuration screen, then press **Enter** to display a list of choices.
2. Select **Controlled by Millennium32**, then press **Enter**. The IEEE 488 Address field is highlighted.
3. Press **Enter** to display a list of addresses.
4. Select an address that is unused by other chromatographic components connected to the Millennium³² Chromatography Manager workstation, then press **Enter**.
5. Press **Exit**. The Separations Module is ready to be controlled from the Millennium³² Chromatography Manager workstation.

Note: Millennium workstations with firmware version 3.20 or earlier recognize a 2695 Separations Module with firmware version 2.00 as a 2690 Separations Module. Future software versions may distinguish between the 2690 and 2695 Separations Modules.

Controlled by MassLynx Mode

In this remote-control mode, Micromass[®] MassLynx[™] software (version 3.5 or higher) controls the 2695 Separations Module. MassLynx software is used with Micromass mass spectrometry (MS) detectors, using the IEEE-488 interface between the Micromass computer and the Waters 2695 Separations Module.

Note: MassLynx software version 3.5 or earlier recognizes a 2695 Separation Module with firmware version 2.00 as a 2690 Separations Module. Future software versions may distinguish between the 2690 and 2695 Separations Modules.

To set the Separations Module to the Controlled by MassLynx mode:

1. Select the **System** field in the Configuration screen, then press **Enter** to display a list of choices.

2. Select **Controlled by MassLynx**, then press **Enter**. The IEEE 488 Address field is highlighted.
3. Press **Enter** to display a list of addresses.
4. Select an address that is unused by other chromatographic components connected to the Micromass computer, then press **Enter**.
5. Press **Exit**. The Separations Module is ready to be controlled from the MassLynx computer.

Controlled by Millennium 2.xx Mode

In this remote-control mode, a Millennium Chromatography Manager workstation running Millennium 2.xx software controls the operation of your HPLC system using the IEEE-488 interface bus.

To set the Separations Module to the Controlled by Millennium 2.xx mode:

1. Select the **System** field in the Configuration screen, then press **Enter** to display a list of choices.
2. Select **Controlled by Millennium 2.xx**, then press **Enter**. The IEEE 488 Address field is highlighted.
3. Press **Enter** to display a list of address pairs.
4. Select an address pair that is unused by other chromatographic components connected to the Millennium Chromatography Manager, then press **Enter**.
5. Press **Exit**. The Separations Module is ready to be controlled from the Millennium Chromatography Manager workstation.

The Separations Module appears as two devices in the Millennium System view:

- **Solvent and sample management systems** – With an even address
- **Optional sample heater/cooler** – With an odd address

Refer to the *Waters 2695 Separations Module Operator's Guide*, Section 5.2, Making Automatic Runs Under Millennium Control, for details on making runs under Millennium control.

Controlled by Operate Gradient by Event In Mode

In the Operate Gradient by Event In mode, an external autosampler (a Waters 2700 Sample Manager, for example) signals the Solvent Management System of the Separations Module to begin a gradient. In this mode, the Separations Module has no control of IEEE-488 devices, however, the Separations Module *can* control non-IEEE devices using the I/O connections on the rear panel.

To set the Separations Module to the Operate Gradient by Event In mode:

1. Select the **System** field in the Configuration screen, then press **Enter** to display a list of choices.
 2. Select **Operate Gradient by Event In**, then press **Enter**.
 3. Press the **Events In** screen key to display the Events In dialog box. If you need more detailed steps on defining Events In conditions, refer to the *Waters 2695 Separations Module Operator's Guide*, "Events In" discussion, Section 3.2.1, Setting Configuration Parameters.
 4. Set the Stop Flow field to the appropriate condition (**Ignore**, **High**, or **Low**), then press **Enter**.
 - Choose **Ignore** if the Stop Flow I/O terminals of the Separations Module are not used.
 - Choose **High** if the output connection from the external autosampler to the Stop Flow I/O terminals of the Separations Module changes to a high (more positive) TTL-level.
 - Choose **Low** if the output connection from the external autosampler to the Stop Flow I/O terminals of the Separations Module changes to a low (more negative) TTL-level.
 5. Set the Hold 1 field to the appropriate condition (**High** or **Low**), then press **Enter**.
 - Choose **High** if the output connection from the external autosampler to the Hold 1 Inject terminals of the Separations Module changes to a high (more positive) TTL-level to initiate a chromatographic run (and prevent an injection by the Separations Module).
 - Choose **Low** if the output connection from the external autosampler to the Hold 1 Inject terminals of the Separations Module changes to a low (more negative) TTL-level to initiate a chromatographic run (and prevent an injection by the Separations Module).
- Note:** *If you are configuring the Separations Module with a Waters 2700 Sample Manager, choose **Low**.*
6. Set the Logic field to **Or**, then press **Enter**.
 7. Set the Hold 2 field to **Ignore**, then press **Enter**.
 8. Press the **OK** screen key to save your selections and exit the Events In dialog box.

Controlled via RS-232 (ASCII) or (Binary) Mode

In these remote-control modes, a non-Waters data system (a mass spectrometry system, for example) controls the Separations Module using RS-232 ASCII or binary communications. The Separations Module disconnects from the IEEE-488 interface bus

when either of these modes is selected. The Separations Module can control other non-IEEE devices in the HPLC system using the I/O connections on the rear panel.

To set the Separations Module to the Controlled via RS-232 (ASCII) or (Binary) mode:

1. Select the **System** field in the Configuration screen, then press **Enter** to display a list of choices.
2. Select **Controlled via RS232 (ASCII)** or **Controlled via RS232 (Binary)**, then press **Enter**.
3. Press **Exit**. The Separations Module is ready to be controlled by a remote data system using RS-232 communications.

Setting Report Options

Use the Report Options dialog box to define the information that is sent to the printer, integrator, and/or floppy disk. (Before you print a report, select the printer in the Printer section of the Configuration screen. You select the destination of the report in the print dialog box that appears when you press the Print screen key.)

To define the information sent to a printer, integrator, and/or floppy disk:

1. Press the **Reports** screen key on the Configuration screen to display the Report Options dialog box ([Figure 2-4](#)).

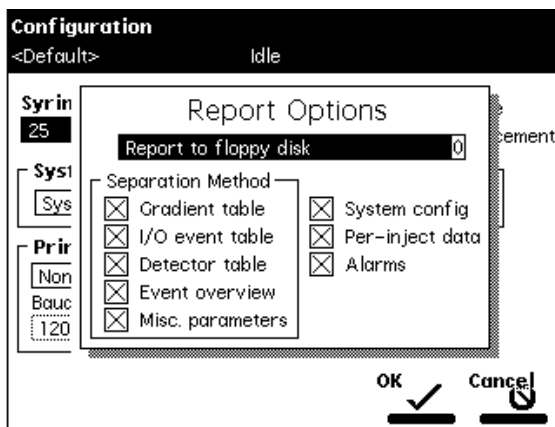


Figure 2-4 Report Options Dialog Box

2. Select the destination for the report in the drop-down list.
3. Select any option and press any numeric key to enable or disable the option, then press **Enter**. An "X" in the box means the option is enabled. The options include:
 - **Gradient table** – Generates the gradient table

- **I/O event table** – Generates the I/O events table
 - **Detector table** – Generates the detector table
 - **Event overview** – Generates an overview of all merged tables
 - **Misc. parameters** – Generates a list of all parameters not included in tables
 - **System config** – Generates a list of the instrument configuration parameters
 - **Per-inject data** – Generates a list of the minimum, maximum, and average values for temperature, pressure, the time and date of each injection, and so on
 - **Alarms** – Generates a list of the error conditions that occurred during the run
4. Press the **OK** screen key to exit the Report Options dialog box.
 5. Press **Exit** to return to the Main screen.

Programming for Auto Shutdown

You can set up the Separations Module to shut down automatically after a specified period of inactivity. Inactivity is defined as:

- No keyboard use
- No injections performed
- No changes sent to the Separations Module from a remotely connected Millennium³² Chromatography Manager workstation, MassLynx computer, or external autosampler
- An error condition that suspends the operation of the Separations Module

You can leave the Separations Module in the shutdown state indefinitely. The separation method you specify in the Auto Shutdown dialog box defines the initial conditions that are applied after the specified period of inactivity. Use the auto shutdown function:

- When there is a long delay between injections
- To minimize solvent flow after an unattended or long run
- To minimize sparge gas consumption
- To disable the vacuum degasser
- To disable temperature controls
- To turn off detector lamps

To enable the auto shutdown function:

1. Press the **Config** screen key in the Main screen, then press the **More** screen key once.
2. Press the **Auto Shutdown** screen key. The Auto-Shutdown dialog box appears ([Figure 2-5](#)).

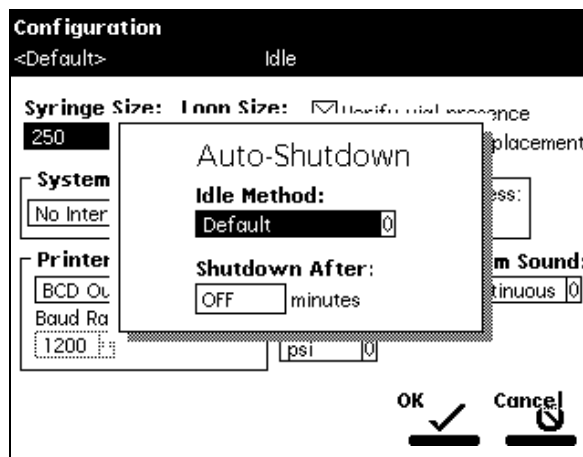


Figure 2-5 Auto-Shutdown Dialog Box

3. Select the separation method to be used while the Separations Module is shut down. Only the *initial* conditions in the method you select are used.
4. Enter a time period (in minutes) after which you want the Separations Module to shut down (or press **Clear** to disable the Auto Shutdown function).
5. Press **OK**. The Separations Module shuts down when there has been no activity for the specified period.
6. Press **Exit** to return to the Main screen.

Note: If you want to use Auto Shutdown to turn off the lamp of a 2487 or 486 Detector, program a Lamp Off event and specify a time of INIT in the I/O Events Timed Table screen (see Section 6.2.5, Setting I/O Parameter Values, in the Waters 2695 Separations Module Operator's Guide).

2.3 Preparing the Solvent Management System

For optimal performance of your Separations Module, perform the steps identified in [Figure 2-6](#).

Note: For details on how to prepare both degasser and sparge-based 2695 Separations Modules for operation, refer to [Chapter 6, Recommended Preparation Procedures](#).

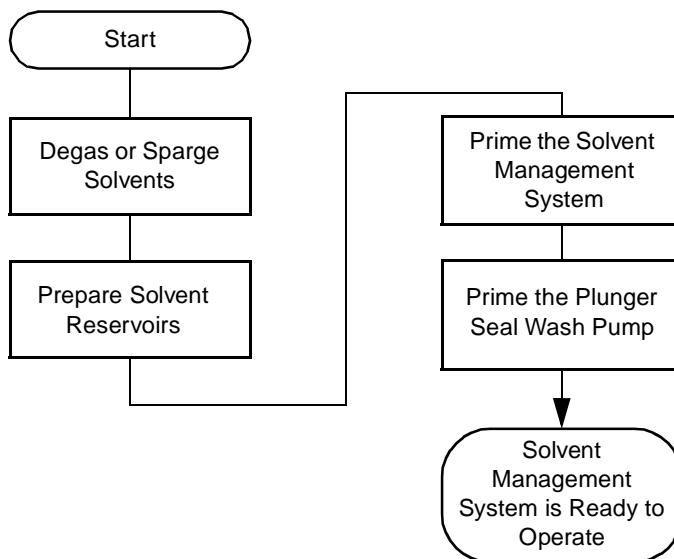


Figure 2-6 Preparing the Solvent Management System for Operation



Caution: Observe safe laboratory practices when you handle solvents. Refer to the Material Safety Data Sheet for each solvent you use.

2.3.1 AutoStartPLUS Startup Routine

AutoStartPLUS is a new startup procedure introduced with version 2.00 firmware for 2695 Separations Modules.

To standardize your daily system startup routine, Waters developed a unique sample set and methods (protocols) to prepare your 2695 Separations Modules for chromatography. A sample set with unique separation methods called AutoStartPLUS was created to purge and equilibrate the system automatically. The 2695 Separations Module has been factory-programmed with the AutoStartPLUS protocols. You can copy and save AutoStartPLUS under a different sample file name, and then edit it to suit your specific needs.

For more information about AutoStartPLUS, refer to Section 3.5.1, AutoStartPLUS Startup Procedure, in the *Waters 2695 Separations Module Operator's Guide*.

2.3.2 Degassing Solvents

Inline Vacuum Degassing

Inline vacuum degassing reduces the total dissolved gas in the mobile phase. The system uses the PerformancePLUS™ Inline Degasser, comprising a variable-speed, continuously operating vacuum pump and low volume degasser chambers that enable rapid instrument priming and equilibration. This allows you to become operational quickly from an idle state or following a solvent change.

For more information on vacuum degassing, refer to Section 1.5, the “Degasser Considerations” discussion in the *Waters 2695 Separations Module Operator's Guide*.

Note: For proper operation of the inline vacuum degasser, you must fill each vacuum chamber with solvent. Use the dry prime direct function to prime all solvent lines. See [Section 2.3.3, Priming the Solvent Management System](#), for the procedure to dry prime the solvent lines.

To control the operation of the inline vacuum degasser from the Status screen:

1. Press the **Menu/Status** key. The Status screen appears, as shown in [Figure 2-7](#). The content and layout of the Status screen varies with the options installed in the Separations Module and with the mode of operation.

The screenshot shows the 'Status (1)' screen with the following fields and controls:

- Method:** <direct>
- Flow:** 0.000 mL/min
- System:** -1 psi
- Composition:** A: 100.0%, B: 0.0%, C: 0.0%, D: 0.0%
- Sample:** Set: OFF, Current: 23.8 °C
- S1 S2 S3 S4:** All OFF
- Degasser:** Mode: Off, Pressure: * psi
- V1 V2 V3 V4:** All OFF
- Column:** Set: OFF °C, Current: 24.6 °C, Selection: 1: Position 1
- Buttons:** Sample Queue, Next Page, Direct Function

Figure 2-7 Status Screen

2. Press the **Next Page** screen key (if necessary) to display the Degasser fields.
3. Select the **Degasser** mode field, then press **Enter** to display a list of degasser modes. The modes are:

- **On** – The degasser is always on.
 - **Off** – The degasser is always off.
4. Select the desired operating mode, then press **Enter**.
- The Vac pump field displays the current status of the vacuum degasser pump.
- The Pressure field displays the current vacuum level in psia, bar, or kPa.

Sparging

In systems outfitted with the Sparge option, you can set the initial and maintenance sparge cycles from the Sparge screen. Also, you can set different sparge cycles for each of the four solvent reservoirs.

To program the sparge cycles from the Direct Function menu:

1. Press the **Direct Function** screen key in the Status screen. The Direct Functions menu appears ([Figure 2-8](#)).

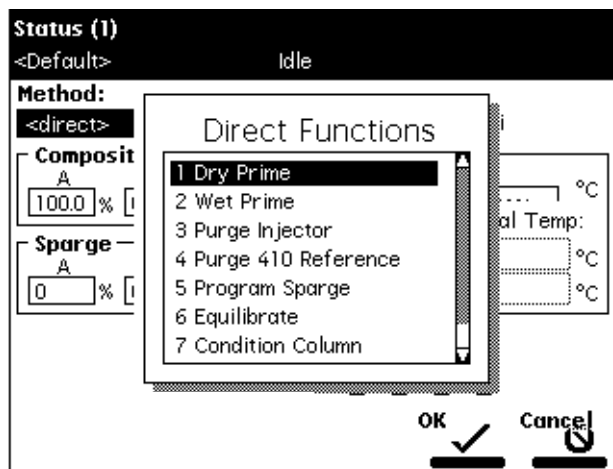


Figure 2-8 Direct Functions Menu for the Program Sparge Option

2. Select the **Program Sparge** option, then press **Enter**. The Programmed Sparge dialog box appears ([Figure 2-9](#)).

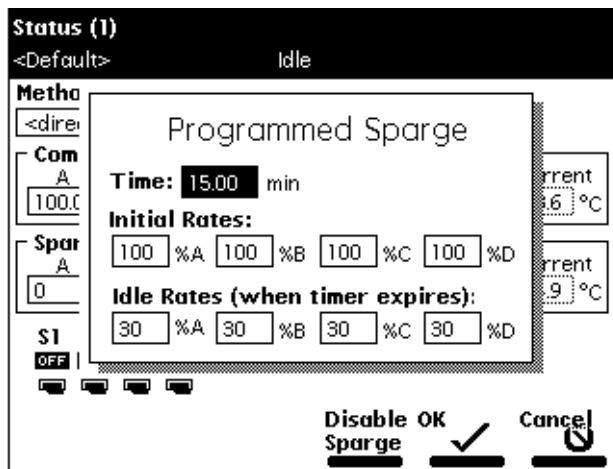


Figure 2-9 Programmed Sparge Dialog Box

3. In the Time field, enter the time to run the initial sparge cycle cycle, then enter values for each solvent reservoir for the following parameters:
 - Initial sparge cycle rates
 - Idle sparge cycle rates

At a sparge cycle rate of 100%, the sparge valve is on at all times; at a sparge cycle rate of less than 100%, the sparge valve turns on and off periodically.

Press the **Disable Sparge** screen key if you do not want to sparge the reservoirs.

Press the **OK** screen key. The timer starts counting down, and the sparge gas flows at the initial sparge cycle rates. The idle sparge cycle rate begins when the timer expires.

2.3.3 Priming the Solvent Management System

Dry Prime

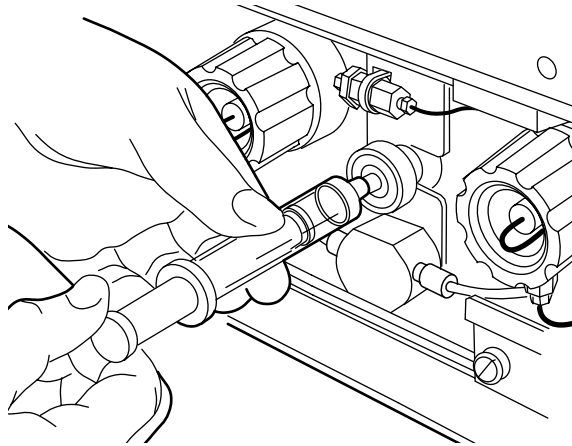
Use the dry prime option to prime the system when the fluidic path in the solvent management system is dry.

To perform a dry prime:

1. Set up the reservoirs as described in the *Waters 2695 Separations Module Operator's Guide*, Section 2.6, Making Fluidic Connections.
2. Insert the solvent tubing into the appropriate reservoir(s). Be sure the detector waste line and the sample loop waste line drain into an appropriate container.
3. Gently shake the filters in the reservoirs to remove any bubbles that may be trapped.

4. Attach an empty syringe to the prime/vent valve, as shown in [Figure 2-10](#), then open the valve by turning it counterclockwise 1/2-turn.

Note: The syringe does not lock onto the prime/vent valve; hold it in place while you pull on the plunger.



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Figure 2-10 Prime/Vent Valve with Syringe

5. Press the **Direct Function** screen key in the Status screen. The Direct Functions menu appears ([Figure 3-4](#)).
6. Select **Dry Prime**, then press **Enter**. The Dry Prime dialog box appears ([Figure 2-11](#)).

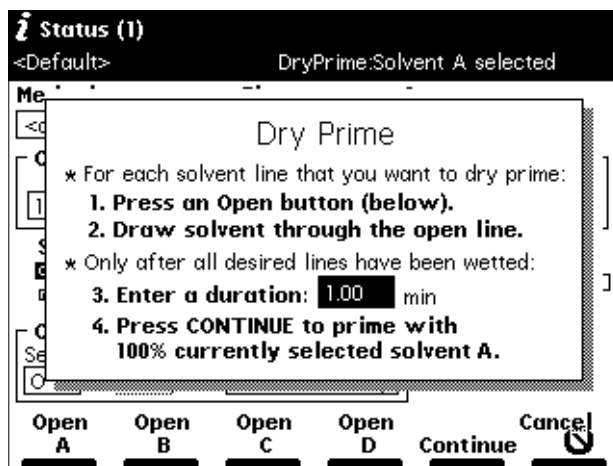


Figure 2-11 Dry Prime Dialog Box

7. Press the screen key corresponding to the solvent line you want to prime.
8. Withdraw the syringe plunger to pull solvent through the tubing. You may need to exert force to pull the air and solvent through the system. Continue until you pull all the air through the solvent line into the syringe.
9. Repeat steps 7 and 8 for each solvent line you want to prime, then close the prime/vent valve.
10. Press a screen key corresponding to the solvent line with which you want to prime the system. Waters recommends that you prime using the solvent with the lowest viscosity to help purge air from the lines, especially if the inline vacuum degasser is installed.
11. In the Enter a Duration field, enter the length of time (in minutes) to prime the solvent management system. Start with a value of 5 minutes. Press **Continue**.
The solvent management system begins to operate. At the end of the priming period, the solvent management system turns off and the Separations Module enters the Idle mode.
12. Perform a wet prime (described next).

Wet Prime



Attention: To avoid damage to the face seals, perform a wet prime only when there is solvent in the solvent management system fluidic path.

Perform a wet prime when you want to change reservoirs or solvents, or if the Separations Module has been idle for some time. The wet prime replaces solvent in the path from the

reservoirs to V3 (waste). If the solvent lines are dry, perform the dry prime procedure before continuing.



Attention: To avoid precipitating salts in the Separations Module, use an intermediate solvent such as water when you change from buffers to high-organic-content solvents. Refer to the Waters 2695 Separations Module Operator's Guide, Section C.3, Solvent Miscibility, for information on solvent miscibility.

To perform a wet prime:

1. In the Status screen Composition field, enter **100%** for the first solvent you want to use for wet priming. Waters recommends that you start the wet prime using the solvent with the lowest viscosity to help purge air from the lines, especially if the inline vacuum degasser is installed.
2. Press the **Direct Function** screen key in the Status screen. The Direct Functions menu appears (Figure 3-4).
3. Select the **Wet Prime** option, then press **Enter**. The Wet Prime dialog box appears (Figure 2-12).

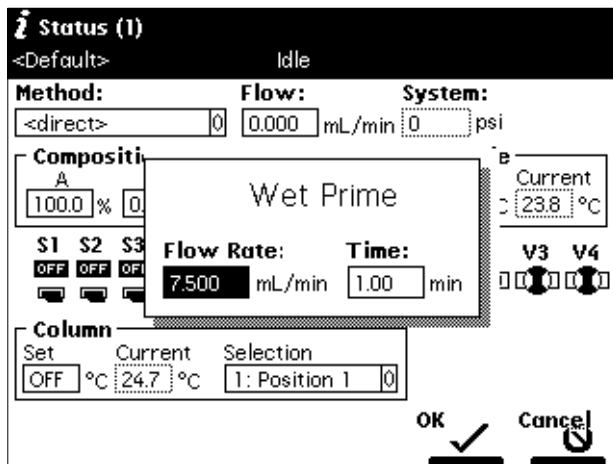


Figure 2-12 Wet Prime Dialog Box

4. Gently shake the filters in the reservoirs to remove any bubbles that may have formed.
5. Enter the flow rate and the duration of the prime (using the composition entered in the Status screen), then press **OK**. The solvent management system begins to operate.

At the end of the time period you specify, the solvent management system returns to previous conditions and the Separations Module enters the Idle mode.

6. Repeat steps 1–5 for each solvent, as appropriate.

Equilibrating Solvents in the Vacuum Degasser

Solvents are equilibrated in their respective degasser chamber by setting a zero flow rate for a short time period. The steps that follow describe this process.

To equilibrate solvents in the inline vacuum degasser after you have wet primed the system:

1. Enter the initial solvent composition for the run in the Status screen.
2. Set the Degasser Mode parameter to **On**.
3. Press the **Direct Function** screen key. The Direct Functions menu appears ([Figure 3-4](#)).
4. Select **Wet Prime**, then press **Enter**. The Wet Prime dialog box appears ([Figure 2-12](#)).
5. Enter a flow rate of **0.000** mL/min and a time of **5** min. At the end of the time period, the solvents are degassed and the Separations Module enters the Idle mode.

The solvents in the inline vacuum degasser are now equilibrated. Prime the plunger seal-wash pump as described below.

Priming the Plunger Seal-Wash Pump

For reversed-phase HPLC applications, use an aqueous plunger seal-wash solution with enough organic content to inhibit bacterial growth. For example, use an 80% water:20% methanol solution or an 80% water:20% acetonitrile solution, depending on your application. For GPC separations, use a 50% methanol:50% water seal-wash solution. If required, use an intermediate solution to prevent immiscibility or precipitate problems when you switch between GPC and reversed-phase analytical solvents.

To prime the plunger seal-wash pump:

1. Press the **Diag** screen key in the Main screen. The Diagnostics screen appears ([Figure 2-13](#)). Refer to the *Waters 2695 Separations Module Operator's Guide*, Section 8.3, Performing Main Diagnostics, for more information on the Diagnostics screen.

Diagnostics					
<Default>		Idle			
Serial Number:		<Not-Entered>			
Transducers			Lifetime Counters:		
System:	0	psi	Eluent delivered =	0.0	L
Primary:	0	psi	Injection count =	0	
Sample:	-1	psi			
C/D Ratio:		0.00			
Prime SealWsh	Comp Check	Prime NdlWsh	Adjust Seals	Service Utilities	Other Tests

Figure 2-13 Diagnostics Screen

2. Attach the syringe adapter to the syringe (both are found in the Startup Kit).
3. Remove the inlet filter from the seal-wash inlet tube.
4. Fill the syringe with the seal-wash solution, then attach the syringe adapter to the end of the seal-wash inlet tube.
5. Press the **Prime SealWsh** screen key, then press the **Start** screen key. The seal-wash procedure begins. Push on the syringe plunger to force seal-wash solvent through the system.
6. When seal-wash solvent flows out of the seal-wash waste tube, press the **Halt** screen key.
7. Reinstall the inlet filter and place the seal-wash inlet tube in the seal-wash reservoir.
8. Press **Close** to return to the Diagnostics screen.

Before you make a run, perform a Condition Column or Equilibrate direct function as necessary.

2.4 Preparing the Sample Management System

[Figure 2-14](#) shows the steps in preparing the sample management system for operation.

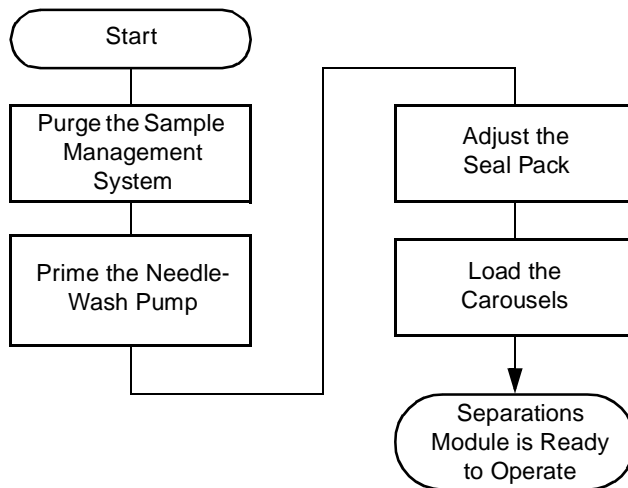


Figure 2-14 Preparing the Sample Management System for Operation

2.4.1 Purging the Sample Management System

Purge the sample management system whenever you:

- Prime the solvent management system
- Change solvents
- See bubbles in the syringe
- Start using the Separations Module at the beginning of each day



Attention: To avoid precipitating salts in the Separations Module, use an intermediate solvent such as water when you change from buffers to high-organic-content solvents.

To purge the sample management system:

1. In the Main screen, press the **Menu/Status** key to display the Status screen ([Figure 2-7](#)).
2. Enter the appropriate solvent composition in the Composition fields.
3. Press **Direct Functions** to display the Direct Functions menu ([Figure 3-4](#)).

4. Select **Purge Injector**, then press **Enter**. The Purge Injector dialog box appears ([Figure 2-15](#)).

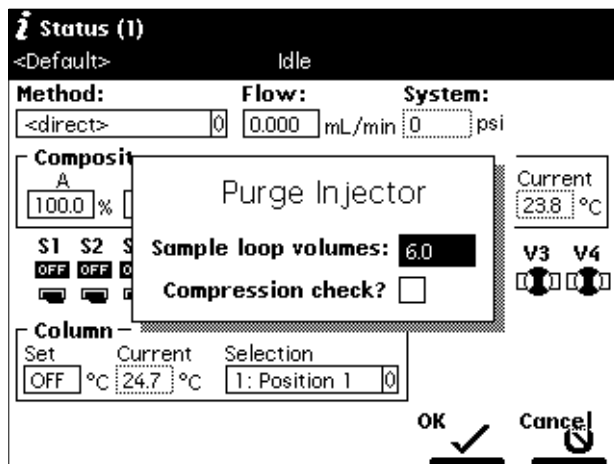


Figure 2-15 Purge Injector Dialog Box

5. Enter the number of sample loop volumes in the Sample Loop Volumes field (use the default value of 6.0 volumes), then press **Enter**. The solvent management system ramps up to a predetermined flow rate to flush the sample loop with the selected volume.
6. Leave the Compression Check check box blank the first time you start up the system to prevent a compression test from occurring. The Separations Module does not allow a compression test before the seal pack is adjusted.
7. Press **OK**. The purge cycle begins.

2.4.2 Priming the Needle-Wash Pump

The needle-wash pump flushes the needle in the sample management system, preventing carryover of sample between injections. The needle wash also extends the life of the injector seals by removing buffered mobile phase and sample from the needle.

Selecting a Needle-Wash Solvent

Use a needle-wash solvent based on the sample and mobile phase chemistries, making sure all solutions/buffers are miscible and soluble. [Table 2-1](#) lists some needle-wash solvents recommended for use with certain mobile phase conditions. High sample concentrations may require other needle-wash solvents.

Table 2-1 Suggested Needle-Wash Solvents

Chromatographic Condition	Needle-Wash Solvent
Buffered aqueous, reversed-phase	80% organic, 20% aqueous ^a
Nonaqueous, reversed-phase	100% MeOH
Normal phase	Mobile phase
GPC	Mobile phase
Ion exchange	H ₂ O

a. If the aqueous portion of the analytical solution is pH-adjusted to ensure sample solubility, match the pH of the aqueous portion of the needle-wash solution to it.

If acids are used, avoid long-term exposure of the needle-wash system to halide- containing acids.

To prime the needle-wash pump:

1. Be sure the needle-wash fluidic lines are properly installed, as described in the *Waters 2695 Separations Module Operator's Guide*, Section 2.6.6, Installing Needle Wash and Plunger Seal Wash Reservoirs.
2. Press the **Diag** screen key in the Main screen. The Diagnostics screen appears ([Figure 2-13](#)).
3. Press the **Prime NdlWsh** screen key. The 30-second needle-wash procedure begins. If solvent does not flow out of the waste tube within 30 seconds, press the **Start Again** screen key.

2.4.3 Adjusting the Seal Pack

Adjust the seal pack whenever:

- You start up the Separations Module for the first time. The Separations Module does not perform injections and compression checks until the seal pack has been adjusted.
- A “Compression Check Failed” error appears on the screen.
- A “Missing Restrictor” alarm appears during a diagnostic procedure.
- You change the seal pack.
- You rebuild the seal pack.

Before you adjust the seal pack, purge the sample management system ([Section 2.4.1, Purging the Sample Management System](#)) to ensure that there is no air in the syringe. Air

in the system may cause Alarm Seal Geometry or Alarm Missing Restrictor dialog boxes to erroneously appear.

To adjust the seal pack:

1. Press the **Diag** screen key in the Main screen to display the Diagnostics screen.
2. Press the **Adjust Seals** screen key. The adjustment procedure begins. When the test is complete, the results are shown in the dialog box that appears.

2.4.4 Loading Carousels

Loading carousels involves:

- Loading the vials into the carousel
- Loading the carousel into the sample compartment

Loading Vials

Use only 12 × 32 mm, 2-mL vials with snap-on, crimp-on, or screw-on caps.

Insert the prepared vials into the appropriate positions in the carousel(s). Be sure to place vials in the numbered slots that correspond to the sample set (if any) you are using. If you are not using a sample set, enter the sample vial positions in the Inject Samples dialog box. If you are using the bar code option, make sure you have correctly attached labels to the vials.

Loading the Carousel

[Table 2-1](#) lists the carousels and their color codes.

Table 2-1 Carousels and Color Codes

Code	Color	Vial Numbers
A	Blue	1 to 24
B	Yellow	25 to 48
C	Red	49 to 72
D	Green	73 to 96
E	White	97 to 120

To load the carousels:

1. Open the carousel door. The Door is Open dialog box appears in the screen ([Figure 2-16](#)).

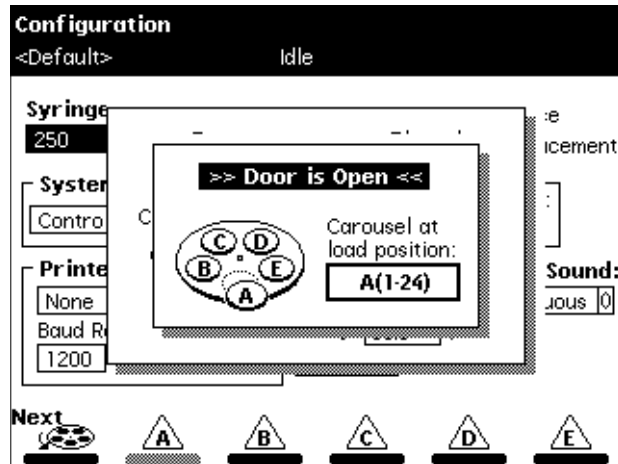


Figure 2-16 Door is Open Dialog Box

2. Press the **Next** screen key (or select the desired carousel screen key) to position the carousel turntable for loading the appropriate carousel.
3. Load the carousel into the sample compartment.
4. Repeat steps 2 and 3 until all appropriate carousels are loaded.
5. Close the carousel door. If you enable the "Verify carousel placement" check box (the default setting) in the Configuration screen, an error message appears if you attempt to make an injection from a carousel that is in the wrong position.

Chapter 3

Direct Control Runs

This chapter describes the procedures for performing a direct control (manual) run. [Figure 3-1](#) provides an overview of the procedures. To make an automatic run, refer to [Chapter 4, Automatic Runs](#).

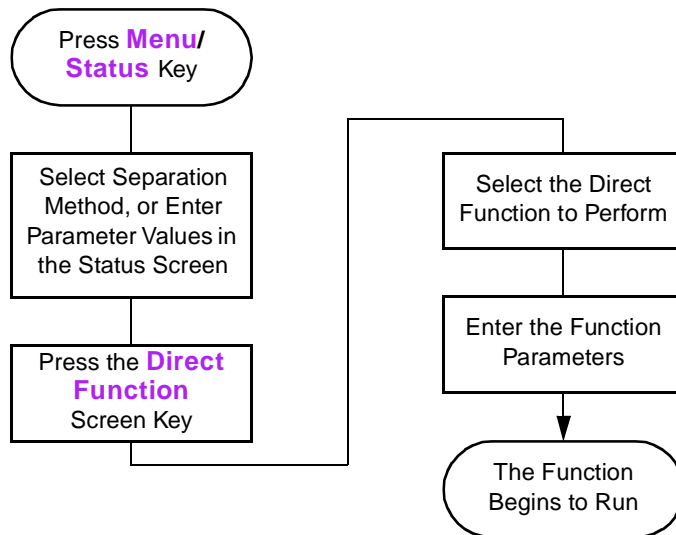


Figure 3-1 Setting Up a Direct Control Run

You perform direct control runs from the Status screen ([Figure 3-2](#) and [Figure 3-3](#)). The fields in the Status screen indicate the status or values of the current operating parameters, as described in [Table 3-1](#). The Status screen also allows you to enter new values for certain parameters.

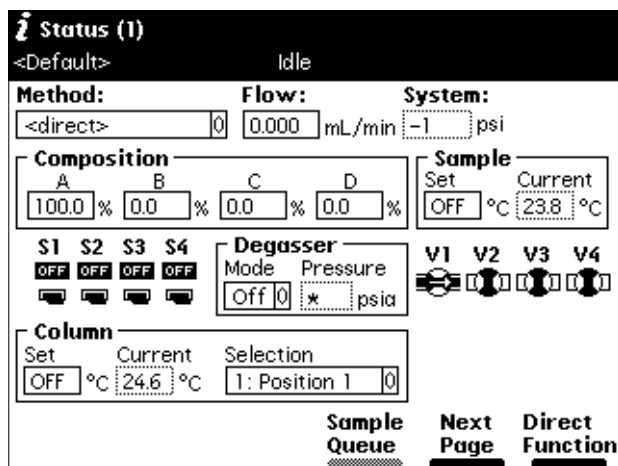


Figure 3-2 First Page of Status Screen (Default)

Press the **Next Page** screen key to display the second page of the Status screen ([Figure 3-3](#)).

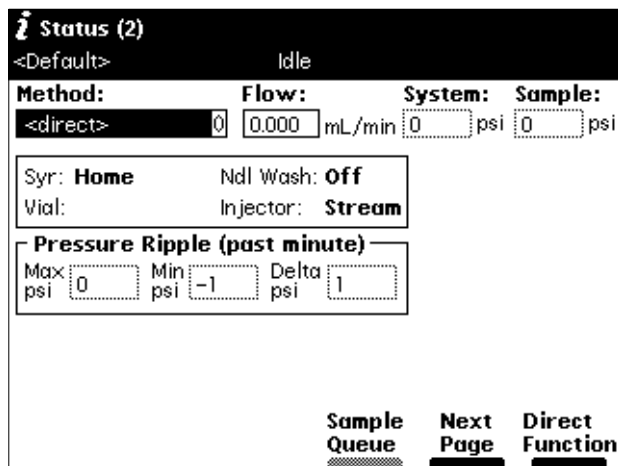


Figure 3-3 Second Page of Status Screen (Default)

Table 3-1 Current Status Screen Parameters

Parameter	Description
Method	Indicates the current separation method. When you change any parameter in the Status screen, this field changes to <direct>.
Flow	Indicates the current flow rate of the solvent management system.
System	Indicates the current system pressure in psi, bar, or kPa.
Sample: Set	Indicates the user-specified sample compartment temperature.
Sample: Current	Indicates the current actual sample compartment temperature.
Composition	Indicates the current solvent composition.
S1 – S4	Indicates the status of the event switches on the I/O connector.
Degasser <ul style="list-style-type: none"> • Mode • Vac pump • Pressure 	<p>Indicates the status of the inline vacuum degasser (if installed).</p> <p>Selects the mode of operation: On (enabled) or Off (disabled).</p> <p>Indicates whether the vacuum pump motor is active or idle.</p> <p>Current vacuum level in psi, bar, or kPa.</p>
Sparge	Indicates the sparge rate cycle for each solvent (if sparge is installed)
V1 – V4	Indicates the position (open or closed) of each of the valves in the sample management system. See the <i>Waters 2695 Separations Module Operator's Guide</i> , Section 1.3, Sample Management System Overview, for a description of the valves.
Pressure Ripple <ul style="list-style-type: none"> • Max psi • Min psi • Delta psi 	<p>Indicates pressure readings in the solvent management system for the last minute.</p> <p>Maximum pressure</p> <p>Minimum pressure</p> <p>Peak-to-peak pressure difference</p>
Syr	<p>Indicates the current position of the syringe. Valid positions are:</p> <ul style="list-style-type: none"> • Home – Default position • Empty – Highest position (to purge the syringe) • Full – Lowest position (to draw in sample)
Vial	Indicates the number of the vial that is currently under the injector.
Ndl Wash	Indicates whether the needle-wash pump is on.
Sample	Indicates the current pressure in the sample loop.

Table 3-1 Current Status Screen Parameters (Continued)

Parameter	Description
Column Temp	Indicates the desired and current temperature of the column (if the column heater is installed).
Injector	Indicates the current position of the injector. Valid positions are: <ul style="list-style-type: none"> • Stream – In the high pressure stream • Seal – In the lower seal position • Wet – In the needle wash position • Vial – In the sample vial
486 (1) and (2) <ul style="list-style-type: none"> • λ • Lamp • AUFS 	Indicates the current status of the following parameters for the 486 Detector(s) (if installed). <ul style="list-style-type: none"> • Wavelength • Lamp On or Off • Absorbance Units Full-Scale
2487 (1) and (2) <ul style="list-style-type: none"> • λ • Lamp • AUFS 	Indicates the current status of the following parameters for the 2487 Detector(s) (if installed): <ul style="list-style-type: none"> • Wavelength • Lamp On or Off • Absorbance Units Full-Scale
2410 and 410 <ul style="list-style-type: none"> • Cell • Col 1 • Col 2 	Indicates the current status of the following parameters for the 2410 or 410 Detector (if installed): <ul style="list-style-type: none"> • Cell Temperature • External Temp 1^a • External Temp 2^a

a. Enabled if the optional column heater module is configured with the Waters 2410 or 410 Detector.

To access the direct control functions:

1. Press the **Menu/Status** key (if necessary) to display the Status screen ([Figure 3-2](#)).
2. Press the **Direct Function** screen key. The Direct Functions menu appears ([Figure 3-4](#)). [Table 3-2](#) describes each of the direct functions.
3. Select the function you want to specify (press the number beside the function, or use the arrow keys), then press **Enter**. Follow the instructions for each function.

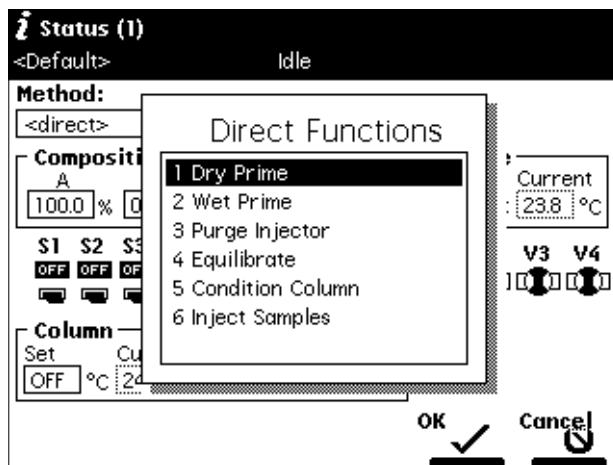


Figure 3-4 Direct Functions Menu

Table 3-2 Direct Control Functions

Direct Control Function	Description
Dry Prime	Opens the fluidic path (from the selected solvent reservoir to the prime/vent valve) to replace air with solvent, then performs a prime.
Wet Prime	Replaces solvent in the path from the reservoirs to V1 and then to waste. Use to change the solvent(s) in the system.
Purge Injector	Removes mobile phase from the sample loop and from the syringe and fills it with fresh mobile phase. As an option, performs a compression test.
Purge 2410/410 Reference (if applicable)	Purges the Waters 2410 or 410 RI Detector reference cell.
Program Sparge (if applicable)	Sets the sparge rate for each reservoir.
Equilibrate	Delivers mobile phase at the current conditions for the specified period of time. Equilibrates the temperature in the sample compartment and column heater at preset values.
Condition Column	Delivers mobile phase using the gradient table specified in the current separation method without injecting a sample or running the Events table.
Inject Samples	Injects a sample one or more times from the specified vial(s) using the selected method.

Chapter 4

Automatic Runs

This chapter describes how to set up the Separations Module to make automatic runs in:

- System Controller or No Interaction mode
- Controlled by Millennium³² Chromatography Manager modes (2.xx or Millennium³²)
- Controlled by MassLynx mode

System Controller or No Interaction Mode

If your HPLC system uses the 2695 Separations Module in a System Controller or No Interaction mode, you make an automatic run using a sample set or sample template stored in the Separations Module. Refer to [Section 4.1, Making Automatic Runs in Stand-Alone Mode](#), for details. Refer to the *Waters 2695 Separations Module Operator's Guide*, Chapter 6, Creating Separation Methods, Sample Sets, and Templates, for information on creating and editing separation methods, sample sets, and sample templates.

Controlled by Millennium³² Chromatography Manager Modes

If your HPLC system is controlled by the Millennium³² Chromatography Manager, you make an automatic run using a project, instrument method, and sample set stored in the Millennium³² Chromatography Manager. Refer to the procedures in [Section 4.2, Making Automatic Runs Under Millennium Control](#), for details on making a run using Millennium³² software.

Controlled by MassLynx Mode

If your HPLC system is controlled by Micromass MassLynx software, you make an automatic run using the sample processing list containing the LC method(s) that define the Separations Module and detector (other than the MS Detector) operating parameters. Refer to [Section 4.3, Making Automatic Runs Under MassLynx Control](#), for details on making a run using MassLynx software.

4.1 Making Automatic Runs in Stand-Alone Mode

[Figure 4-1](#) shows the steps for setting up and making an automatic run with a Separations Module in a stand-alone configuration (System Controller or No Interaction mode).

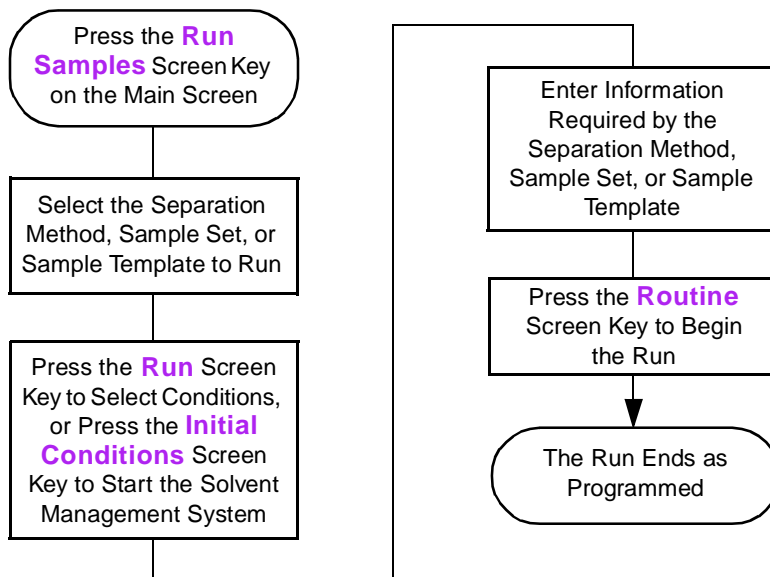


Figure 4-1 Making an Automatic Run in a Stand-Alone Configuration

Running a Sample Set

Note: If you plan on importing sample sets stored on a diskette, refer to the "Importing Methods" discussion later in this section.

To run a sample set in a stand-alone mode:

1. Press the **Run Samples** screen key in the Main screen ([Figure 2-2](#)). The Run Samples screen appears ([Figure 4-2](#)). [Table 4-1](#) describes the icons used in the Run Samples screen.

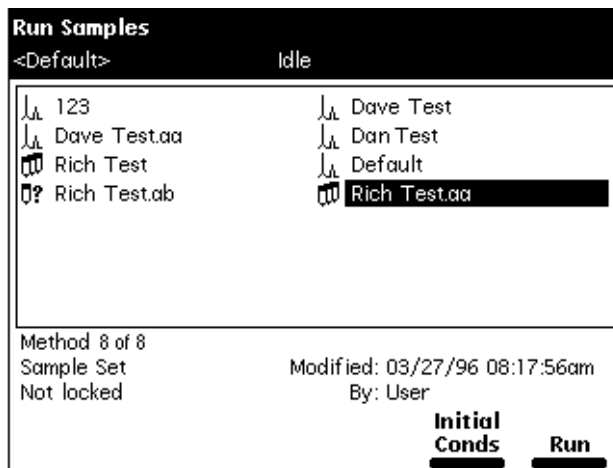


Figure 4-2 Run Samples Screen

Table 4-1 Run Sample Screen Icons

Icon	Description
	Separation Method
	Sample Set
	Sample Template

2. Select the sample set you want to run.
3. Press one of the following screen keys to perform the indicated tasks:
 - **Initial Conds** – Starts the solvent management system at the initial conditions (specified in the first line of the sample set) and begins equilibrating the temperature in the sample heater/cooler and/or column heater. When the system is equilibrated, press the **Run** screen key.
 - **Run** – Displays the selected sample set table (to view or edit), as shown in [Figure 4-3](#). Use the screen keys to modify the table as necessary.

Run Samples						
<Default>			Idle			
Job: Rich Test[002]					Functional View	
	vials	function	method	inj	μL	min
1	*	Equil	Dave Test	0 *	*	45.00
2	*	Purge	Dan Test	0 *	*	*
3	1-2	AutoStds	Dave Test	0 1	10.0	22.50
4	5-24	Sample	Dave Test.aa	0 2	15.0	22.50
5		Purge	Dan Test	0 *	*	*
6	*	Condition	Dave Test	0 *	*	30.00
7	25-30	AutoAdd	Dave Test.aa	0 1	10.0	15.00
8						

7 rows total

Insert Row **Copy Down** **Row Details** **More** **Next View** **Start**

Figure 4-3 Sample Set Screen – Functional View

4. Load the sample vials into the carousel(s) according to the locations specified in the sample set (see [Section 2.4.4, Loading Carousels](#)).
5. Press the **Start** screen key. The Run Samples screen appears with a list of the solvents required for the run ([Figure 4-4](#)). Check that you have prepared the Separations Module with the indicated solvent(s).

Run Samples						
<Default>			Idle			
Job: Rich Test[004]					Functional View	
	vials	Solvents required:				min
1	*	A: Methanol				45.00
2	*	B: Water				*
3	1	Routine schedules the samples after completion of current work				22.50
4	5-24					22.50
5	*	STAT runs the new samples as soon as possible				*
6	*					30.00
7	25-30					15.00
8						

7 rows total

Initial Conds **Cancel** **STAT** **Routine**

Figure 4-4 Run Samples Screen with Solvents Required Dialog Box

6. Press the **Routine** screen key to start a normal run. To modify a run in progress, refer to the “Editing a Separation Method” discussion that follows. To make a stat run, refer to the “Performing a Stat Run” discussion that follows.

Making a Run from a Sample Template

Note: If you plan on importing sample templates stored on a diskette, refer to the “Importing Methods” discussion later in this section.

You can perform an automated run in a stand-alone mode using a sample template stored in the Separations Module. Screen prompts give you step-by-step directions to prepare for the run. Refer to the *Waters 2695 Separations Module Operator’s Guide*, Chapter 6, Creating Methods, Sample Sets, and Sample Templates, for a description of sample templates.

To run a sample template in a stand-alone mode:

1. Press the **Run Samples** screen key in the Main screen. The Run Samples screen appears (Figure 4-2).
2. Select the sample template you want to run (see Table 4-1), then press **Enter**. The Setup dialog box appears, as shown in Figure 4-5.

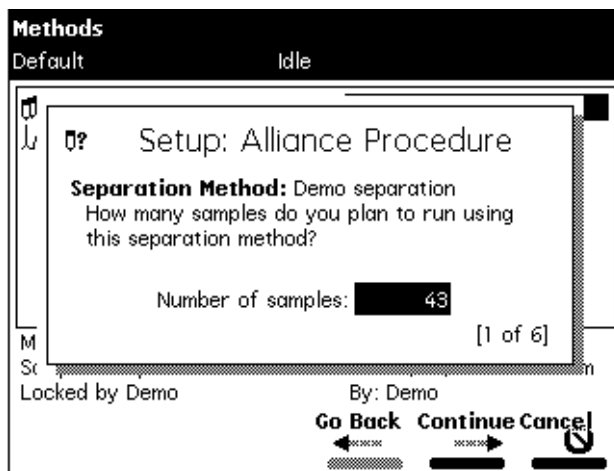


Figure 4-5 Setup Dialog Box

3. Enter the number of samples to run, then press the **Continue** screen key.
4. Enter the location of the first sample vial, then press the **Continue** screen key. You must use consecutive sample vials on one or more carousels.
5. Repeat steps 3 and 4 if the sample template includes multiple separation methods.
6. Enter the number of standards in the run, then press the **Continue** screen key.
7. Enter the location of the first standard vial, then press the **Continue** screen key. You must use consecutive standard vials.

8. Make sure that the solvents listed in the dialog box are primed and in the appropriate inlet tubes, then press the **Start** screen key.

Modifying a Sample Set During a Run

You can modify operating conditions during a run in a stand-alone mode by:

- Using the stat run function
- Editing the separation method

Performing a Stat Run

You can use the stat run function to interrupt a run in progress and perform one or more injections after the current injection is done, then continue with the scheduled run. Performing a stat run is most useful when running separation methods that use identical solvents and temperatures.

To perform a stat run *while a sample set is running*:

1. Load the sample(s) into the carousel, then place the carousel in the sample compartment.
2. Press the **Run Samples** screen key in the Main screen. The Run Samples screen appears ([Figure 4-2](#)).
3. Select the separation method or sample set you want to use for the stat run, then press the **Run** screen key.
4. Enter the carousel position of the vial(s) and other parameters in the appropriate fields in the screen.
5. Press the **Start** screen key. A screen appears with a list of the solvents required for the run, as shown in [Figure 4-4](#). Check that you have prepared the Separations Module with the indicated solvent(s).
6. Press the **Stat** screen key. The stat run starts immediately after the current injection is complete. The original sample set resumes after the stat run is complete.

Editing a Separation Method

You can modify a separation method while a sample set is running. If the separation method is being used for the current injection, the current injection is not affected by the changes. You must save the changes to the modified separation method before you can apply the method to all subsequent injections that use that separation method. Use the procedures in the *Waters 2695 Separations Module Operator's Guide*, Section 6.1, Creating and Editing Separation Methods, to modify and save the separation method.

Importing and Exporting Methods

You can use the floppy disk drive in the Separations Module to import and export separation methods, sample sets, and sample templates to 1.44-MB, standard

PC-formatted floppy disks. Also, by saving reports and tables to a floppy disk, you can use the disk drive as an alternative to a hard-copy printer.

Importing Methods

To import one or more methods from a floppy disk:

1. Insert the disk that contains the method(s) into the floppy disk drive.
2. Press the **Methods** screen key in the Main screen, then press the **More** screen key twice to access the Methods screen (Figure 4-7).
3. Press the **Import Methods** screen key. The Import Methods screen appears, displaying a list of methods stored on the disk (Figure 4-6). To update the list, press the **Refresh** screen key.

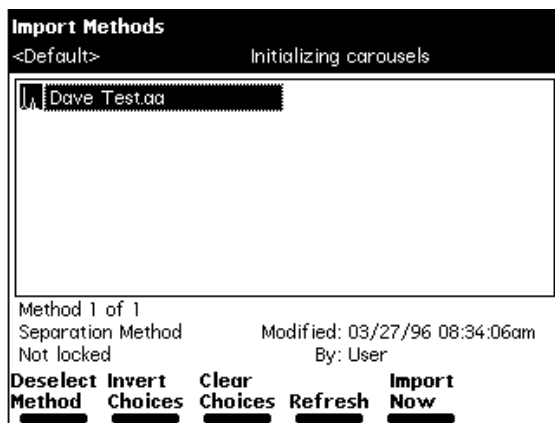


Figure 4-6 Import Methods Screen

4. Select the method you want to import into the Separations Module, then press the **Select Method** screen key. To select additional methods, select each method and press **Select Method** again.
5. After you select all the methods you want to import, press the **Import Now** screen key. The file transfer begins.



Attention: Do not remove the floppy disk from the drive before the import is complete; the Separations Module cannot detect a partially imported method. Waters recommends that you carefully review any imported method before you run the method.

An error message appears if you attempt to import a method that already exists in the Separations Module. You can choose to overwrite the existing method.

An error message appears if you attempt to transfer more than 60 methods into the Separations Module.

Exporting Methods

To export one or more separation methods, sample sets, or sample templates to a floppy disk:

1. Insert a preformatted 3.5-inch disk into the floppy disk drive in the Separations Module.
2. Press the **Methods** screen key in the Main screen, then press the **More** screen key twice to access the Methods screen ([Figure 4-7](#)).

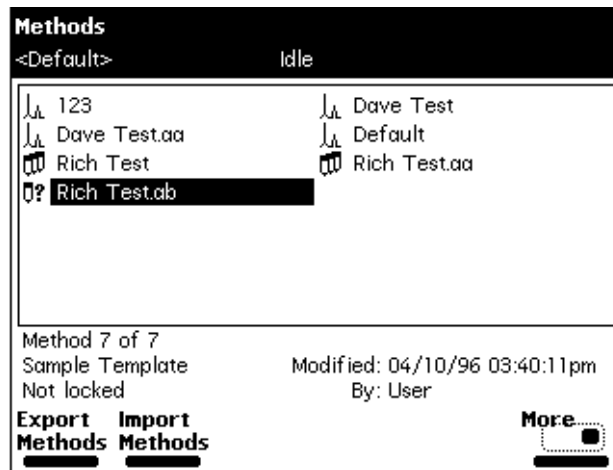


Figure 4-7 Methods Screen

3. Press the **Export Methods** screen key. The Export Methods screen appears, displaying a list of currently stored methods ([Figure 4-8](#)).

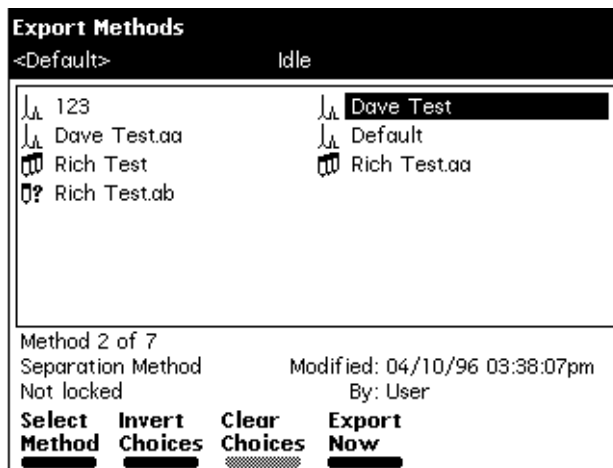


Figure 4-8 Export Methods Screen

4. Select the method you want to export to the disk, then press **Select Method**. To select an additional method, select the method desired, then press **Select Method** again.

To select all methods except the Default method:

- a. Select the **Default** method, then press the **Select Method** screen key.
 - b. Press the **Invert Choices** screen key. All methods except the Default method are selected.
5. When you have selected all the methods you want to transfer, press the **Export Now** screen key. The file transfer begins.

4.2 Making Automatic Runs Under Millennium Control

If your Separations Module is configured for Millennium Chromatography Manager control (Millennium version 2.xx or Millennium³²), you make an automatic run from Run Samples (QuickSet in Millennium versions earlier than 3.20) in the Millennium software. When the Separations Module is running under Millennium control, the word “Remote” appears in the banner area.

Making a Run Using Millennium Software

To make an automatic run, perform the following steps at the Millennium workstation:

1. Create a chromatographic system that includes the Waters 2695 Separations Module:
 - For Millennium software version 2.1x, create a system that includes the Waters 2695 Solvent Management System and the Sample Management System. Refer to the Millennium documentation for information on creating a chromatographic system.
 - For Millennium³² software, create a system that includes the Waters 2695 Separations Module. Refer to the *Millennium³² Help* for information on creating a chromatographic system.
2. Create a Millennium instrument method that uses the system you created in step 1. Refer to the Millennium documentation for information on creating and/or modifying an instrument method.

Note: Any parameter values not explicitly set in the Millennium instrument method automatically take on the values in the Default separation method in the Separations Module.

You can edit the Default separation method in the Separations Module to set parameter values appropriate for your applications.

3. Create a method set that uses the instrument method you created in step 2. Refer to the Millennium documentation for information on creating and/or modifying a method set.
4. From Millennium³² software, run the samples in Run Samples (QuickSet in Millennium versions earlier than 3.20) using the method set you created in step 3. When you enter Run Samples, use the HPLC system you created in step 1. Refer to the Millennium documentation for information on using Run Samples.
5. Process and print out the data acquired from the chromatographic run. Refer to the Millennium documentation for information on developing a processing method.

Note: You can operate the Separations Module from its front panel whenever Run Samples is not running on the Millennium Chromatography Manager workstation.

4.3 Making Automatic Runs Under MassLynx Control

If your Separations Module is configured for MassLynx software control (version 3.5), you make an automatic run from the MassLynx software. When the Separations Module is running under MassLynx control, the word “Remote” appears in the banner area.

Making an Automatic Run Using MassLynx Software

To make an automatic run, perform the following steps at the Micromass MassLynx computer:

1. Configure an LC-MS system that includes the Separations Module. Refer to MassLynx Help for information on configuring an inlet system.
2. From the Inlet Editor, create an LC method for the Separations Module and detector (other than the mass spectrometer). Refer to MassLynx Help for information on using the Inlet Editor.
3. Acquire a single sample from the Inlet Editor. To acquire multiple samples, create a sample processing list from the MassLynx Top Level screen. Refer to MassLynx Help for information on sample processing lists and acquisition.
4. Process and print out the data acquired from the chromatographic run. Refer to MassLynx Help for information on processing data.

You can operate the Separations Module from its front panel whenever MassLynx is not running.

4.4 Stopping a Run

To stop a run in progress, press one of the following keys from the Status screen:

- **Stop Inject** (screen key) – Stops the current injection and displays a dialog box requesting you to abort or resume the injection
- **Hold Inject** (screen key) – Completes the current function, suspends operation of subsequent functions, and displays screen keys that allow you to abort or resume the sample set
- **Hold Gradient** (screen key) – Holds the current gradient conditions. A screen key allows you to resume the gradient
- **Stop Flow** (on the keypad) – Stops the flow of solvent, suspends operation of the current function, and displays screen keys that allow you to abort or resume the function

If any alarm condition specified in the separation method, sample set, or sample template is set to “Stop Function” or “Stop Flow” and the alarm condition occurs, a dialog box appears notifying you of the alarm condition. The following screen keys are in this dialog box:

- **Abort** – Cancels the current function
- **Resume** – Resumes the current function

Chapter 5

Powering Off the Separations Module

Before Powering Off

Before you power off the Separations Module, remove any buffered mobile phase present in the fluidic path.



Attention: To avoid damaging your column, remove the column before you perform the following procedure.

To remove buffered mobile phase from the fluidic path of the Separations Module:

1. Replace the buffered mobile phase with HPLC-quality water and prime the system for 10 minutes at 3 mL/min (see [Section 2.3.3, Priming the Solvent Management System](#), the [“Wet Prime”](#) discussion).
2. Perform three injector purge cycles to ensure that the sample loop is clean (see [Section 2.4.1, Purging the Sample Management System](#)).
3. Replace the plunger seal-wash solvent with a fresh solution of 80% water: 20% methanol or a miscible solvent, and prime the plunger seal-wash pump (see [Section 2.3.3, Priming the Solvent Management System](#), the [“Priming the Plunger Seal-Wash Pump”](#) discussion).
4. Replace the water mobile phase with a solution of 90% methanol: 10% water, and flush the system for 10 minutes at 3 mL/min.
5. Replace the needle-wash solvent with a solution of 90% methanol: 10% water or a miscible solvent, and perform a needle-wash prime (see [Section 2.4.2, Priming the Needle-Wash Pump](#)).

Powering Off

To power off the Separations Module:

1. Ensure that the system has been flushed properly (see the [“Before Powering Off”](#) discussion above).
2. Press the power switch to the 0 (Off) position. The time and date of power-off cycles are entered in the log file.

Chapter 6

Recommended Preparation Procedures

This chapter outlines the steps to prepare both degasser- and sparge-based 2695 Separations Modules for operation using Waters-recommended procedures. Flowcharts provide the sequence of steps to prime, equilibrate, and purge a Separations Module that is in one of the following states:

- A new or dry Separations Module
- A Separations Module that has been turned off or left idle with no flow
- A Separations Module that requires a solvent change, such as from a buffered solvent to an organic solvent

For step-by-step instructions on how to use Direct Functions to prime, equilibrate, and purge your Separations Module, refer to the *Waters 2695 Separations Module Operator's Guide*, Chapter 4, Section 4.4, Performing Direct Functions.

Note: All flowchart references in this sections refer to chapters or figures in the *Waters 2695 Separations Module Operator's Guide*.

[Figure 6-1](#) outlines how to prime, equilibrate, and purge a sparge-based Separations Module that is **new** or **dry**.

Note: All flowchart references refer to sections or figures in the *Waters 2695 Separations Module Operator's Guide*.

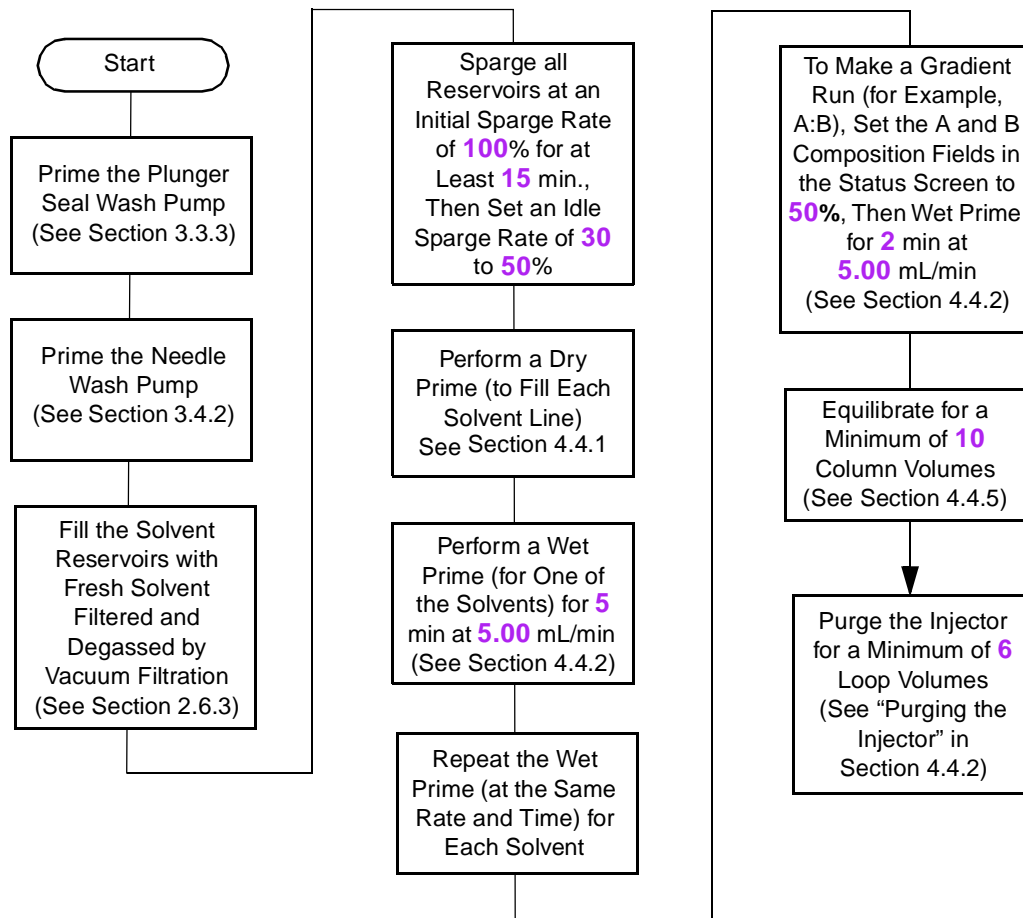
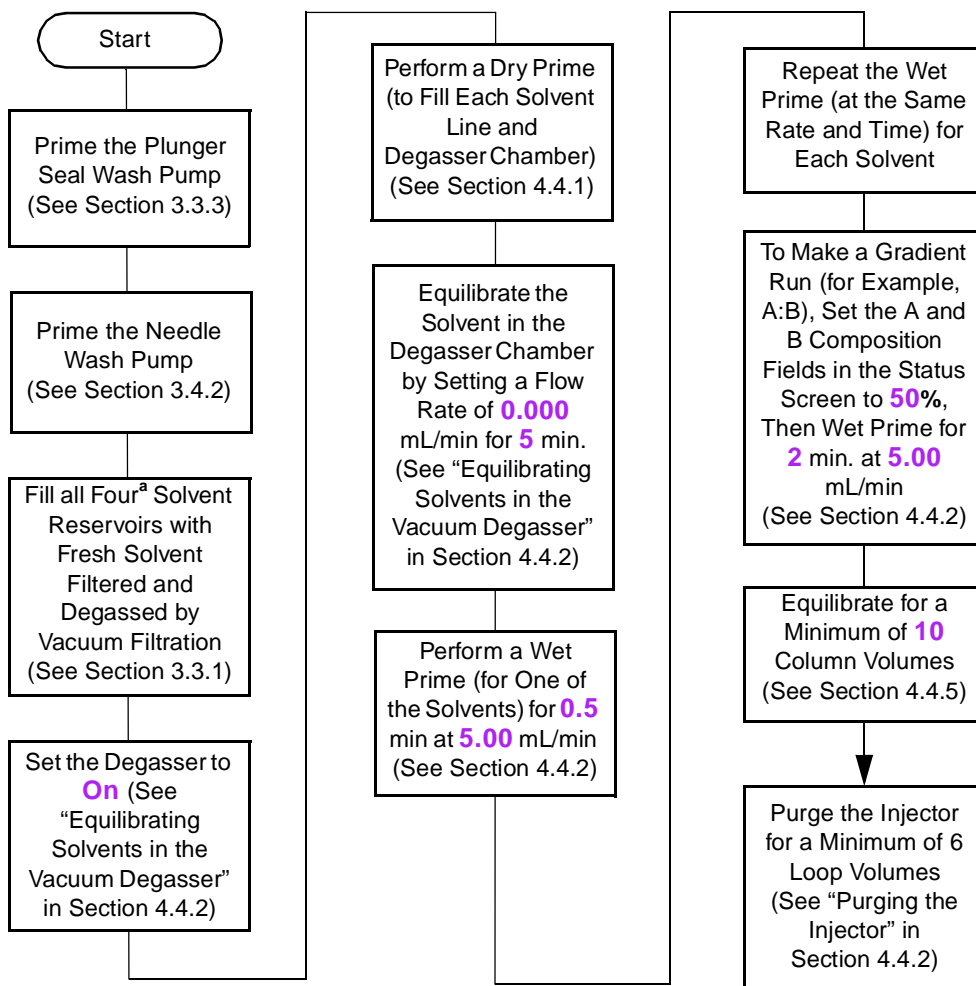


Figure 6-1 Preparing a New or Dry Separations Module for Operation (Sparge-Based Unit)

Figure 6-2 outlines how to prime, equilibrate, and purge a vacuum degasser-based Separations Module that is **new** or **dry**.

Note: All flowchart references refer to sections or figures in the Waters 2695 Separations Module Operator's Guide.

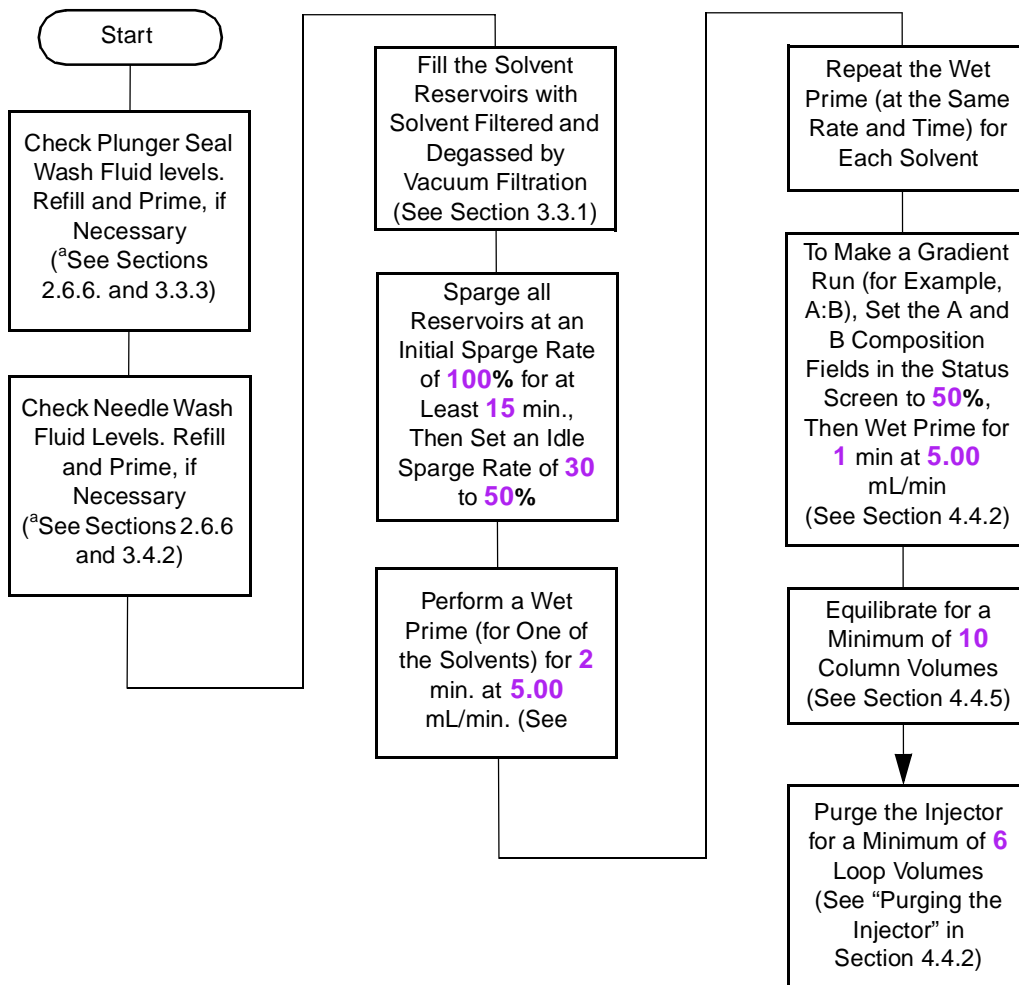


^aTo maximize degasser efficiency (and fill all four degasser chambers with solvent), Waters recommends that all four solvent reservoirs be filled (even if four solvents are not used). If you choose water as one of the "unused" solvents, change the water weekly to prevent bacterial

Figure 6-2 Preparing a New or Dry Separations Module for Operation (PerformancePLUS Degasser-Based Unit)

Figure 6-3 outlines how to prime, equilibrate, and purge a sparge-based Separations Module that is **idle** or has been **powered off** with no solvent flow.

Note: All flowchart references refer to sections or figures in the Waters 2695 Separations Module Operator's Guide.

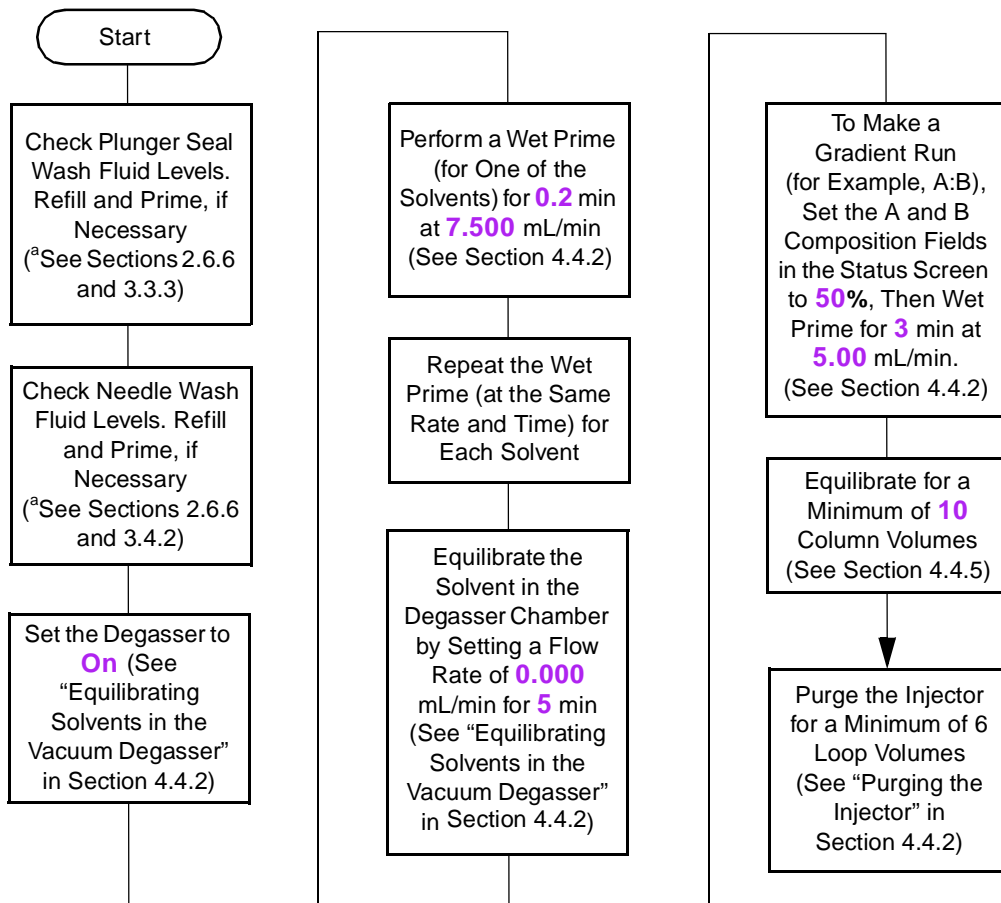


^a If you have just performed the steps from the flowchart “Changing from a Buffered Solvent to an Organic Solvent”, ensure that the Plunger Seal Wash and Needle Wash solvents are compatible with the new solvents.

Figure 6-3 Preparing an Idle or Powered-Off Separations Module for Operation (Sparge-Based Unit)

Figure 6-4 outlines how to prime, equilibrate, and purge a vacuum degasser-based Separations Module that is **idle** or has been **powered off** with no solvent flow.

Note: All flowchart references refer to sections or figures in the Waters 2695 Separations Module Operator's Guide.

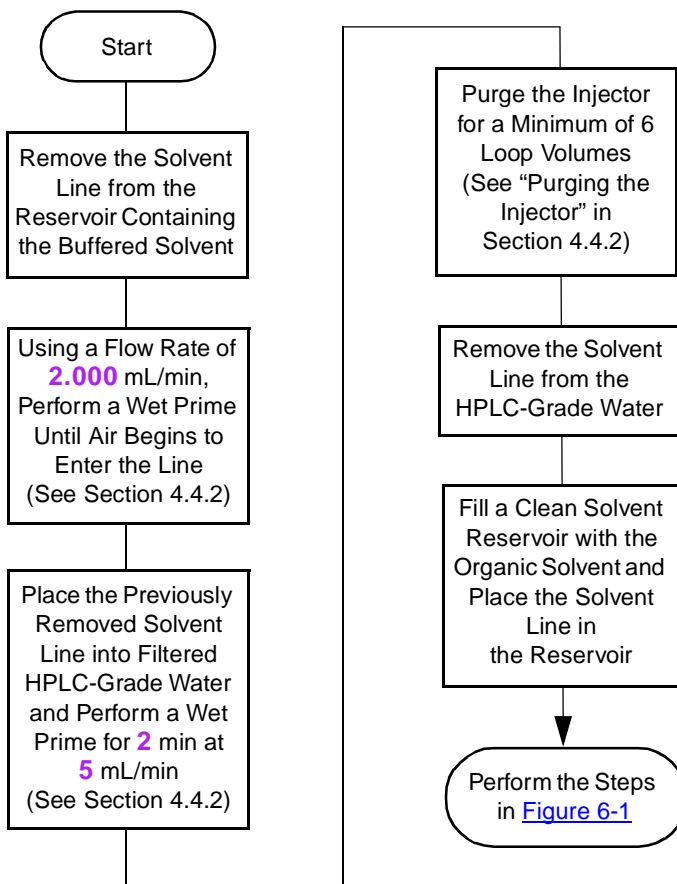


^a If you have just performed the steps from the flowchart "Changing from a Buffered Solvent to an Organic Solvent", ensure that the Plunger Seal Wash and Needle Wash solvents are compatible with the new solvents.

Figure 6-4 Preparing an Idle or Powered-Off Separations Module for Operation (PerformancePLUS Degasser-Based Unit)

[Figure 6-5](#) outlines how to change from a buffered solvent to a totally organic solvent in a sparge-based Separations Module.

Note: All flowchart references refer to sections or figures in the Waters 2695 Separations Module Operator's Guide.

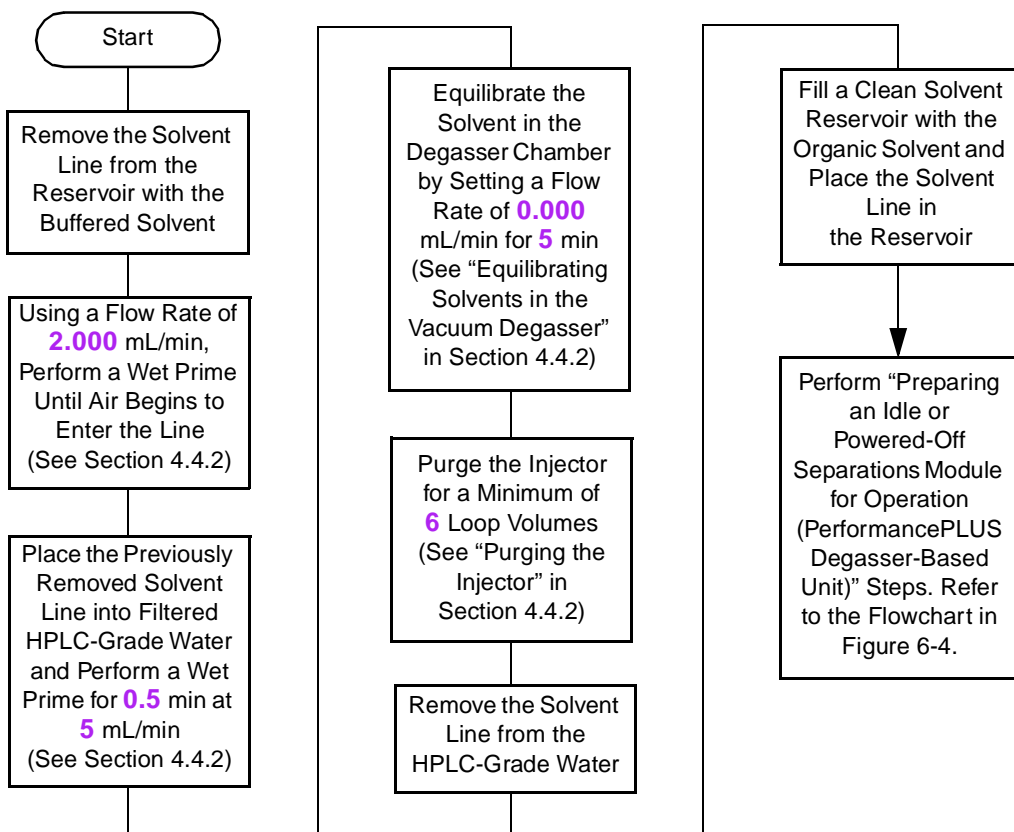


Attention: To avoid having salts precipitate in the Separations Module, use an intermediate solvent such as water when you change from buffers to high-organic-content solvents. Refer to Section C.3, Solvent Miscibility, for information on solvent miscibility.

Figure 6-5 Changing from a Buffered Solvent to an Organic Solvent (Sparge-Based Unit)

Figure 6-6 outlines how to change from a buffered solvent to a totally organic solvent in a degasser-based Separations Module.

Note: All flowchart references refer to sections or figures in the Waters 2695 Separations Module Operator's Guide.



Attention: To avoid having salts precipitate in the Separations Module, use an intermediate solvent such as water when you change from buffers to high-organic-content solvents. Refer to Section C.3, Solvent Miscibility, for information on solvent miscibility.

Figure 6-6 Changing from a Buffered Solvent to an Organic Solvent (PerformancePLUS Degasser-Based Unit)