

SERIES 200

PUMP



User's Manual

Series 200 Pump User's Manual

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Safety and Regulatory Information

Electromagnetic Compatibility (EMC)

United States

This product is classified as a digital device used exclusively as industrial, commercial, or medical test equipment. It is exempt from the technical standards specified in Part 15 of the FCC Rules and Regulations, based on Section 15.103 (c).

Symbols and Conventions used in this Manual

In this manual, the following graphic symbols and special text formats are used to set apart important safety information.



Warning

A warning indicates an operation that could cause *personal injury* if precautions are not followed.



Caution

A caution indicates an operation that could cause *instrument damage* if precautions are not followed.

Symbols used on the Instrument

There are two different types of warning symbols that appear on the instrument.



This symbol indicates an operation (or condition on the instrument) that could cause *personal injury* if precautions are not followed. This can be any type of hazard.

When you see this symbol, refer to the safety pages in the manual for further information.

Consulter les documents d'accompagnement.



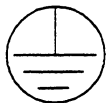
This symbol indicates the danger of electric shock, if precautions are not followed, due to the presence of high voltage.

Attention. Risque de choc électrique.

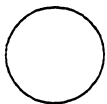
Additional graphic symbols used on the instrument are the following:



Indicates alternating current.



Indicates the primary protective grounding terminal.



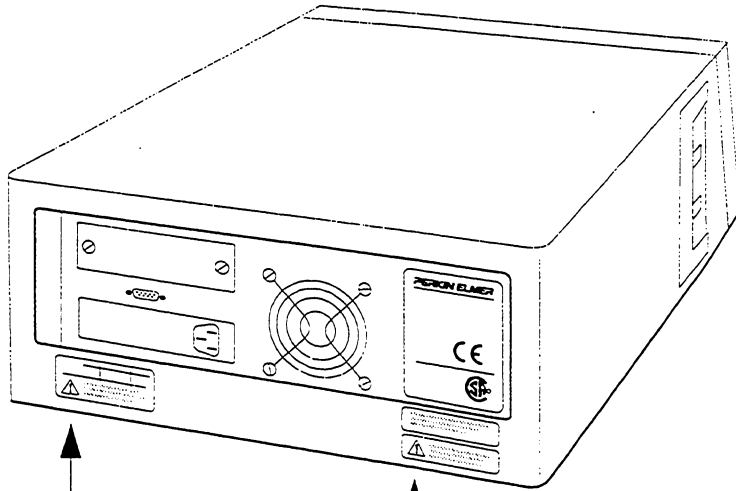
Indicates the *off* position of the main power switch.



Indicates the *on* position of the main power switch.

Warnings on the Instrument

The following warning labels are affixed to the rear panel of the instrument for all models as shown in the figure below.



**FOR PROTECTION AGAINST FIRE HAZARD,
REPLACE ONLY WITH SAME TYPE AND
RATING OF FUSE.**

**AFIN D'ASSURER LA PROTECTION CONTRE
LES RISQUES D'INCENDIE, REMPLACER
UNIQUEMENT PAR UN FUSIBLE DE MÊME
TYPE ET DE MÊME COURANT NOMINAL.**



**GROUNDING CIRCUIT CONTINUITY IS VITAL FOR
SAFE OPERATION OF EQUIPMENT. NEVER OPER-
ATE EQUIPMENT WITH GROUNDING CONDUCTOR
DISCONNECTED.**

**POUR UN FONCTIONNEMENT SANS DANGER. NE
JAMAIS UTILISER L'ÉQUIPMENT SI LE FIL DE
TERRE N'EST PAS RACCORDÉ.**

Electrical Warnings

**Warning**

Connect the instrument to an AC line power outlet that has a protective ground connection. To ensure satisfactory and safe operation of the instrument, it is essential that the protective ground conductor (the green /yellow lead) of the line power cord is connected to true electrical ground. Any interruption of the protective ground conductor, inside or outside the instrument, or disconnection of the protective ground terminal may impair the protection provided by the instrument.

**Warning**

Do not operate the instrument with any covers or parts removed.

**Warning**

Do not attempt to make adjustments, replacements or repairs to this instrument except as described in the accompanying User Documentation. Only a PerkinElmer service representative or similarly trained and authorized person should be permitted to service the instrument.

**Warning**

Use only fuses with the required current rating and of the specified type for replacement.

Quality Control/Good Laboratory Practices

Quality Control

The user should develop appropriate quality control procedures for the LC Pump (and the entire LC system) to ensure suitability for its intended use. These procedures typically consist of periodic performance verifications and routine inspections and suitability tests.

Certificate of System Conformity

Each Series 200 LC Pump is carefully built and tested in a controlled system in accordance with the requirements specified in its applicable PerkinElmer Final Assembly and Test Specification.

Each instrument is certified to meet its functional and performance specification upon release to shipment. The integrity of this quality system is routinely audited and is certified by the British Standards Institution as meeting all requirements of ISO 9001:1987 (Certification No. FM 22179).

Instrument Performance Verification (IPV)

To maintain functional performance, PerkinElmer recommends a yearly Instrument Performance Verification (IPV) of the Series 200 Pump by a PerkinElmer Service Engineer to ensure its operation within published specifications. These tests consist of measuring the most important pump characteristics such as pressure, flow rate, and composition accuracy. Certification is available for regulatory compliance. Contact your local PerkinElmer Sales and Service office.

Routine Inspection and Suitability Test

The Series 200 Pump should also be inspected weekly through the access door for signs of liquid leaks. Prior to any analysis, a system suitability test, which closely resembles the intended assay, should be performed to ensure that the LC system is operating within established criteria (e.g., peak resolution, peak asymmetry, precision, retention time, column plate count, pressure limits, signal/noise ratio, etc.)

While the Universal Test Mix (UTM) Part No. 0089-0893, can be used for a system check, we recommend that you develop a separate system suitability test and acceptance criteria for each of your assays.

Hazardous Chemicals

Before using mobile phase solvents, you should be thoroughly familiar with all hazards and safe handling practices. Observe the manufacturer's recommendations for use, storage and disposal. These recommendations are normally provided in the material safety data sheets (MSDS) supplied with the solvents.

**Warning**

Some chemicals used with this instrument may be hazardous or may become hazardous after completion of an analysis. The responsible body (e.g. lab manager) must take the necessary precautions to ensure that the surrounding workplace and that the instrument operators are not exposed to hazardous levels of toxic substances (chemical or biological) as defined in the applicable Material Safety Data Sheets (MSDS) or OSHA, ACGIH, or COSHH documents. Venting for fumes and disposal of waste must be in accordance with all national, state and local health and safety regulations and laws.

Definitions in Warning for Hazardous Chemicals

Responsible body. "Individual or group responsible for the use and maintenance of equipment, and for ensuring that operators are adequately trained." [per IEC 1010-1, Amendment 2 (draft)].

Operator. "Person operating equipment for its intended purpose." [per IEC 1010-1, Amendment 2 (draft)].

OSHA: Occupational Safety and Health Administration (United States)

ACGIH: American Conference of Governmental Industrial Hygienists

COSHH: Control of Substances Hazardous to Health (United Kingdom)

Temperature, Humidity and Environment



Caution This instrument is designed for indoor use.



Caution Do not operate in a Cold Room or a refrigerated area. The Series 200 LC Pump operates most efficiently under the following conditions:

- ambient temperature is 10 to 35°C
 - ambient relative humidity is 20 to 80% (without condensation)
 - altitude is in the range 0 to 2000 meters.
-



Warning This instrument is not designed for operation in an explosive environment.

Installation Category

This instrument is able to withstand transient overvoltage according to Installation Category II as defined in IEC 1010-1.

Pollution Degree

This equipment will operate safely in environments that contain nonconductive foreign matter up to Pollution Degree 2 in IEC 1010-1.

Storage Conditions

The Series 200 LC Pump may be stored under the following conditions:

- ambient temperature is -20 to +60°C
- ambient relative humidity is 20 to 80% (without condensation)
- altitude is in the range 0 to 12,000 meters.

Notes

Preface: Using the Manual

The PerkinElmer Series 200 LC Pump can be used with virtually every LC system or application. The Series 200 LC Pump is upgradable from a single solvent (isocratic) pump, to mixing two solvents (binary) or to mixing four solvents (quaternary). In addition, each version is available in a biocompatible version. The Series 200 LC Pump provides you with the interchangeable technology so that you do not have to buy a new pump and learn new user interfaces every time your needs change.

Using the Manual

The main function of this manual is to give you all the information you need to operate the Series 200 LC Pump as productively as possible, using all of the instrument features.



Caution

The instrument should be used according to the instructions provided in this manual. If used otherwise, the protection provided by the instrument may be impaired.

Note

The unpacking and setup procedures for the Series 200 LC Pump are included in Chapter 2 of this manual. If you have not installed the instrument, you should first refer to the instructions given in Chapter 2.

Definitions

The following definitions of key terms will help you in using this manual to operate the Series 200 LC Pump.

Method

A method is a set of conditions used to affect the column separation of samples injected by the autosampler. In an isocratic method, it consists of the solvent flow rate, the length of time the instrument pumps solvent at the specified flow rate, and the operating pressure limits.

A binary or quaternary method consists of the solvent flow rate, the length of time the instrument pumps solvent at the specified flow rate, the solvent composition, the gradient curvature (how the solvent composition changes from one step to another) and the operating pressure limits.

Isocratic, binary, and quaternary methods also include provisions for initiating timed events during the course of the method, as well as a means of limiting the amount of time the instrument continues to pump solvent after the method time expires (also referred to as the ready time).

There is a default method, method00, provided with the Pump. You can make changes to this default method and save those changes by saving them under a new method number (from 1 - 19). If you choose not to save the changes in this way, the changes will be lost when you turn the power off on the Pump.

Main Screen

The Main Screen is the top-level screen that provides you with access to all of the Pump's functions. All of the functions are displayed along the bottom of the screen. If you have one of the isocratic pump models (N291-0100 or N294-0200), the Main Isocratic Screen appears; if you have one of the binary or quaternary pump models (the N291-0101, N291-0102, N291-0201, or N291-0202) the Main Binary/Quaternary Screen appears.

MAIN ISOCRATIC SCREEN

METHOD00	STORD	SHTDN			0 PSI
DEFAULT					
	TIME	FLOW			
	10.0	2.00			
ELAPSED TIME					*0.0
PUMP	T.E.	PRES	SEQ	DIR	CNFG STRT

MAIN BINARY/QUATERNARY SCREEN

METHOD00	STORD	SHTDN			0 PSI		
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME					*0.0		
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

Current or Active Method

The current or active method is the method that is currently running on the Pump, or the method currently shown on the display. As the method progresses, certain values in the main screens change to reflect the progress of the analysis.

Sequence

A sequence is a series of stored methods that are linked together. When you start a sequence, each method in the sequence is run in the order defined during the setup of the sequence.

Conventions

The following conventions are used in this manual.

- An instrument screen display is pictured as a single-lined box:

METHOD00	STORD	SHTDN	0 PSI
DEFAULT			
TIME	FLOW		
10.0	2.00		
		ELAPSED TIME	*0.0
PUMP	T.E.	PRES	SEQ
DIR	CNFG	STRT	

- Instructions for using the softkey functions are displayed in the text as:
“press **CNFG** to ...”
- Instructions for using hard keys on the instrument keyboard are displayed in text as:
“press the **return** key ...”
- Notes emphasize significant information in a procedure or description.

Note	The pressure units are set from the Instrument Set-Up screen under the Configure menu.
-------------	---

Customer Service

Company Name and Address

PerkinElmer
710 Bridgeport Avenue
Shelton, CT 06484-4794 U.S.A.

For Sales Assistance, Technical Assistance, and Information call: 1-800-762-4000

To place an order for supplies and many replacement parts, request a free *e*-ssentials catalog or ask for information, visit our website at www.perkinelmer.com. The most up-to-date information on part numbers, product brochures, systems modules, spare parts, and application notes are in the PerkinElmer website.

Telephone:

- **U.S. only:** Call toll free **1-800-762-4000**, 8:30 a.m. to 7 p.m. EST. Your order will be shipped usually within 24 hours.
- **Worldwide:** Call your local PerkinElmer sales or service office.

Notes

Introduction

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Introduction 1

This chapter introduces you to the Series 200 LC Pump and includes the following information.

- A Description of the Pump
- The Pump's Features and Benefits
- The Main Components of the Pump
- Using the Keyboard and Display
- Instrument Specifications

A Description of the Pump

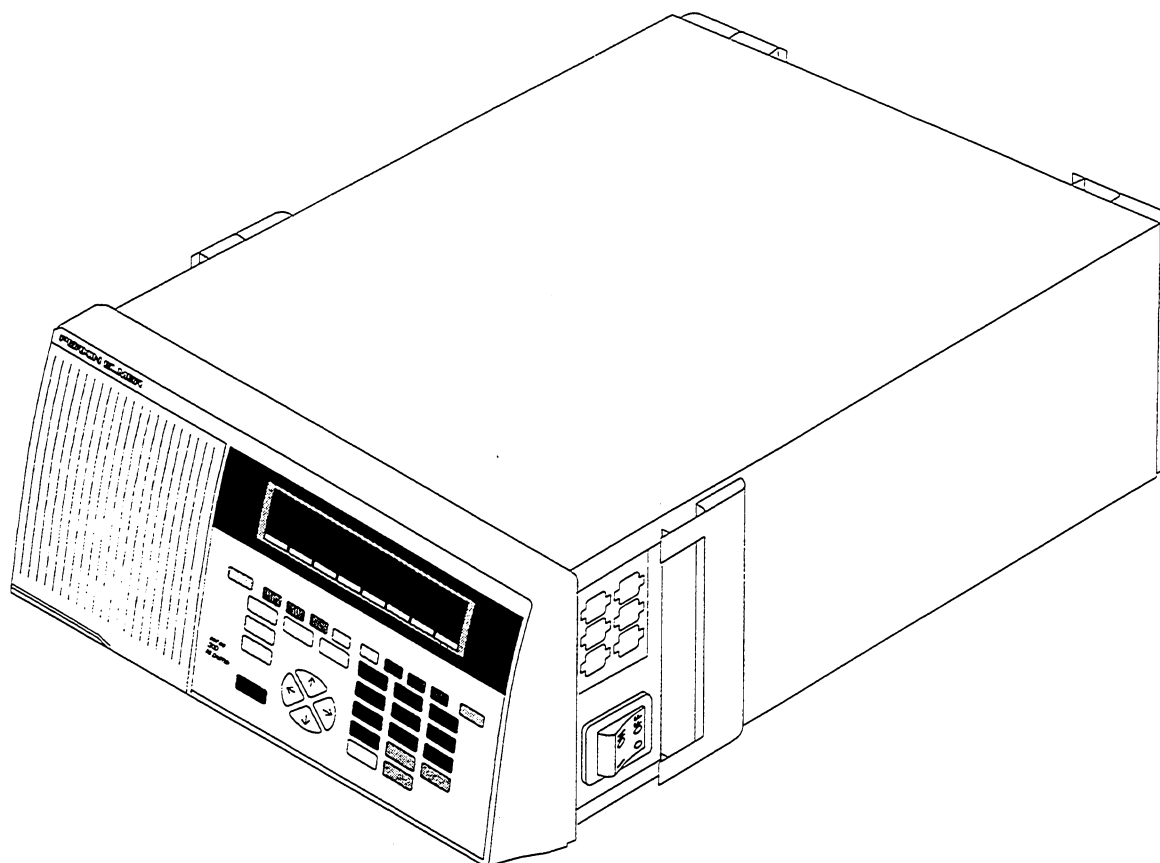


Figure 1-1. The Series 200 LC Pump.

The PerkinElmer Series 200 LC Pump can be used with virtually every LC system or application. The Series 200 LC Pump is upgradable from a single solvent (isocratic) pump, to mixing two solvents (binary) or to mixing four solvents (quaternary). In addition, each version is available in a biocompatible version. The Series 200 LC Pump provides you with the interchangeable technology so that you do not have to buy a new pump and learn new user interfaces every time your needs change.

Series 200 LC Pump Features and Benefits

- Upgradable isocratic, binary, and quaternary versions as well as biocompatible versions for maximum flexibility.
- Small, compact design (12 inches (30.5 cm) wide) saves valuable bench space.
- Directory of up to 20 methods, 20 steps each with built-in method linking increases sample throughput.
- Edit the active method or sequence in real-time with easy access function keys.
- Large backlit liquid crystal display allows clear viewing of pump method and operating parameters at all times.
- Unique function allows automatic shutdown of the pump method at final gradient method.
- Graphic display of solvent profile on-screen shows real-time status of the gradient method.
- Hold gradients or advance to the next step with a simple keystroke.
- Solvent-resistant front panel with rounded ergonomic design for easier interaction with the keypad.
- Tactile keyboard with unique “key click” so that you know your entry has been made.
- Phone-type contact closure connections and finger tight plumbing fittings make it simple to connect or disconnect your system.
- Full method documentation including modification with method name, number, date and time stamp.
- Visual access front-door panel allows quick plumbing inspection and easy servicing.
- Maintenance log coded by the serial number of the pump, including pump and seal cycles.
- Instrument log records history of operation for quick diagnosis and servicing to get you up and running quickly.
- Extensive service diagnostics.
- Pump module can be completely removed to lab bench for maintenance without having to unstack modules.

Main Components of the Series 200 LC Pump

The Series 200 LC Pump consists of the following main components. A brief description of each of the components follows.

Refer to Figure 1-2 for the location of the following components.

Power On/Off Switch

This switch is located on the right side of Pump and switches the power on or off.

Keyboard and Display

All instrument functions are accessed using the tactile keyboard.

Pump control screens, graphic displays of solvent profiles, and up to eight soft keys corresponding to the function keys on the keyboard appear in the Liquid Crystal Display.

An overview describing how to perform such basic operations as moving and entering information in the screens is provided in “**Using the Keyboard and Display**” section of this chapter.

Pump Access Door

This door provides access to the prime/purge valve, Luer-Lok™ fittings for the piston rinse (BIO version), and locking tab for the pump drawer.

Input/Output Connector Panel

The connector panel consists of six contact closure connectors for automated instrument control of devices like an LC pump, LC detector, and a data handling system.

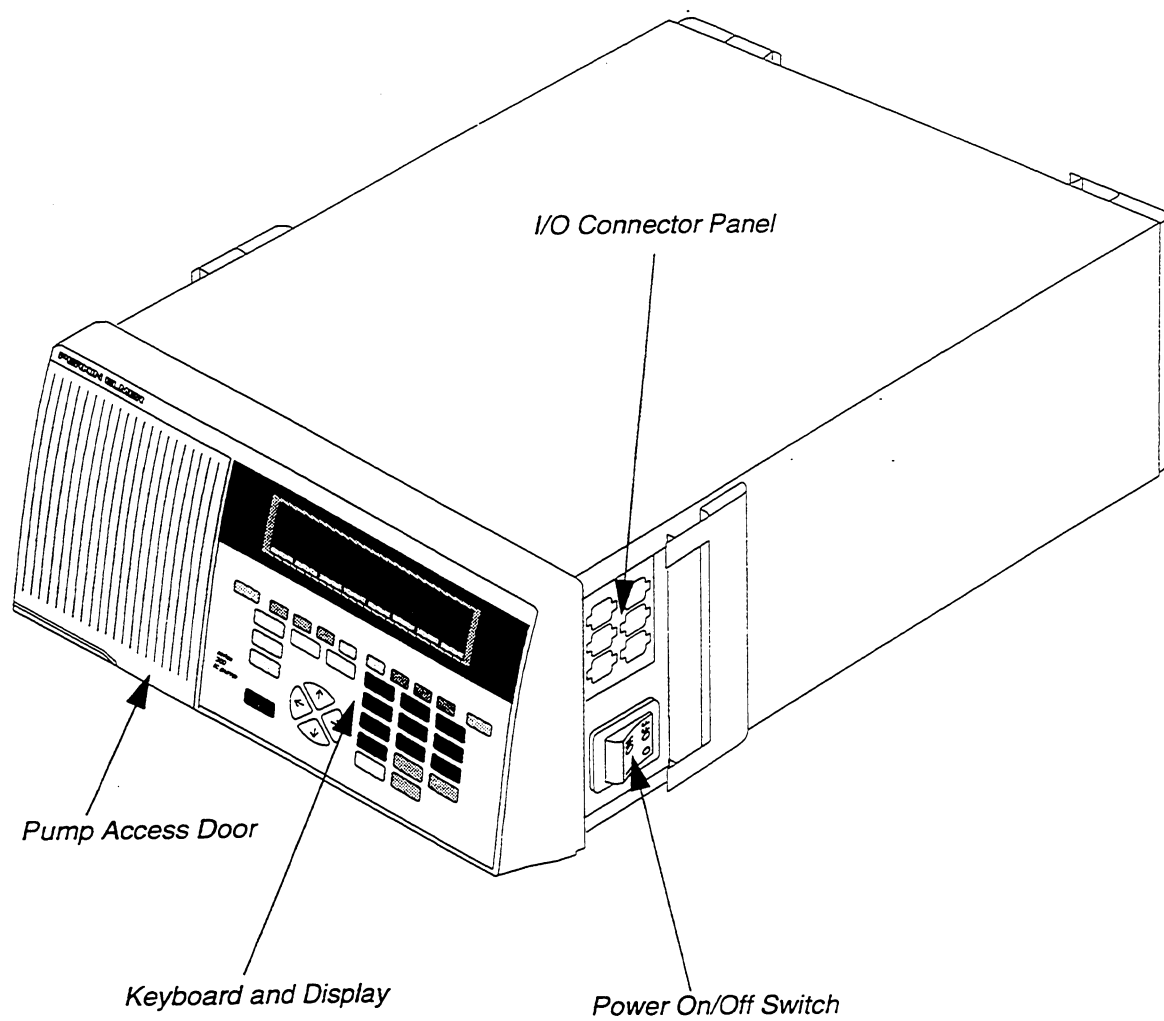


Figure 1-2. Front and Right Side View of the Pump.

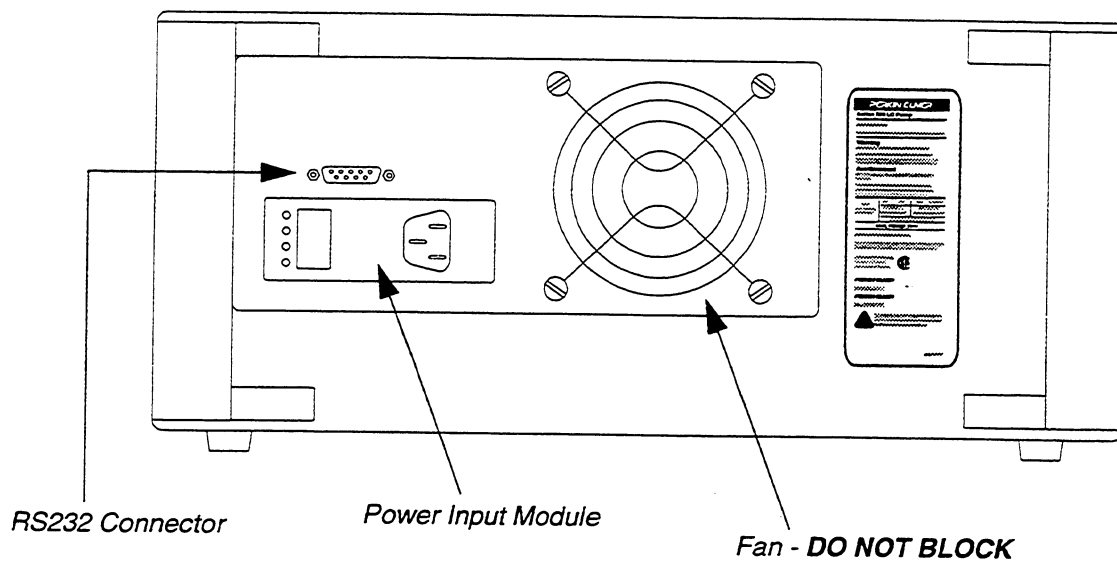


Figure 1-3. Rear Panel of the Pump.

Rear Panel

Figure 1-3 shows the location of the Pump fan and the names and locations of all the rear panel connectors. A brief description of each connector is contained in Table 1-1.

Table 1-1. Description of the Rear Panel Connectors

Connector	Description
Power Input Module	The power input module accepts the AC line cord. It also houses the system fuse(s) and the voltage selection card.
RS232 Connector	This provides connection for RS232 communications to other computers and devices.

Using the Keyboard and Display

The Series 200 LC Pump keyboard and Liquid Crystal Display (LCD) are located on the front panel of the instrument as shown in Figure 1-2. A closer view of the keyboard and display is provided here in Figure 1-4. You configure the Pump and define all of your methods by filling in information on the screens that appear in the LCD. You use the keys on the keyboard to enter the information on the screens and also to start or stop certain operations on the Pump. As you move from one screen to another, the screen-defined function keys (also referred to as soft keys) change. The functions of the various keys are summarized in Table 1-2 on the next page.

You access all of the Pump screens starting at one of the two top level main screens. The top level main screen shown in Figure 1-4, the Main Quaternary Screen, corresponds to the one provided on the binary and quaternary pump models. The other top level screen, the Main Isocratic Screen, is displayed on the isocratic pump models and is described in Chapters 3 and 4 in this manual.

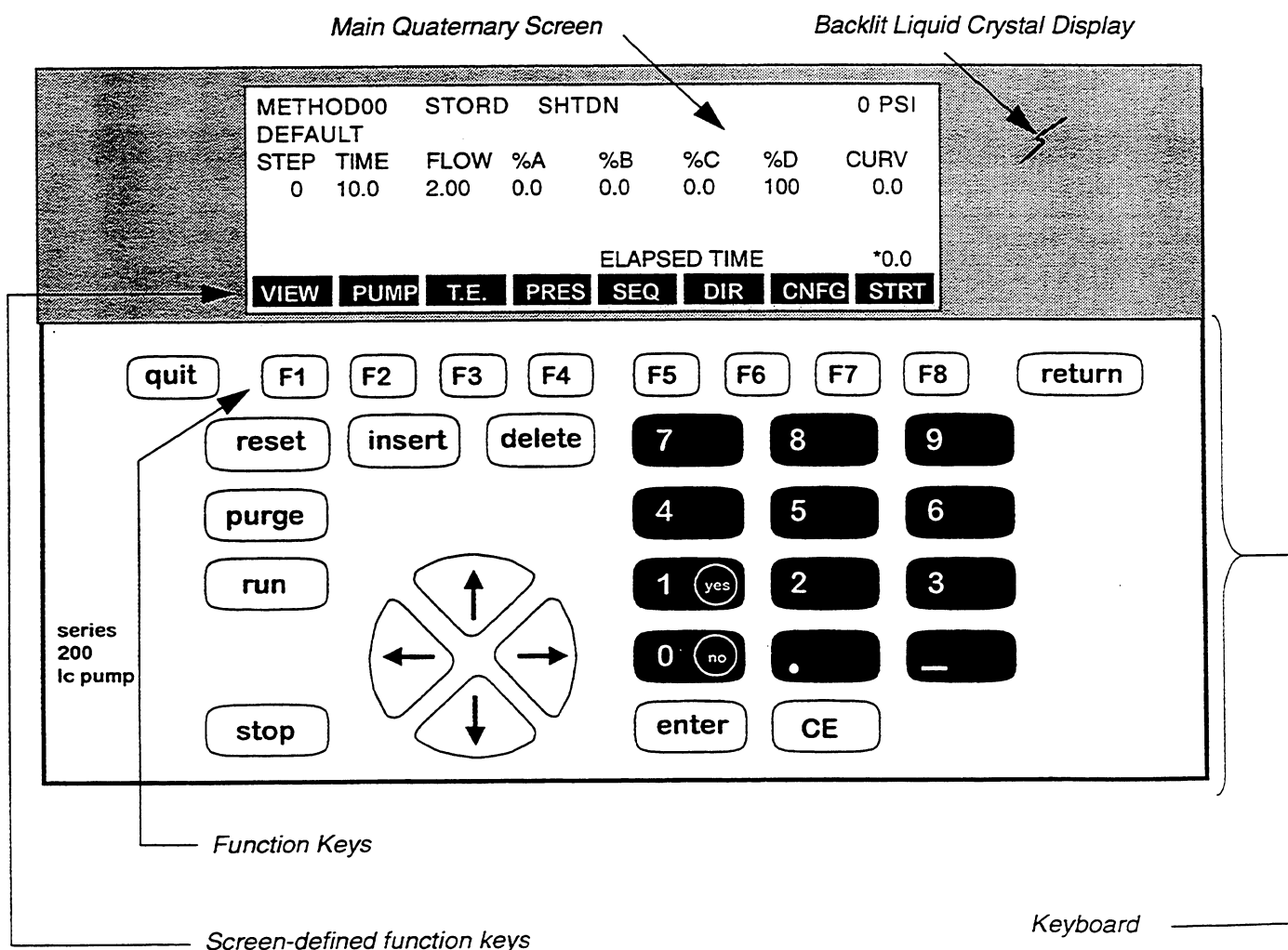









Figure 1-4. Keyboard and Display.

Summary of the Keyboard Functions

Table 1-2. Summary of Keyboard Functions.

Key	Function
 To 	Press a function key to select the corresponding screen-defined soft key label displayed above it. For example, press F7  from the Main Quaternary or Isocratic Screen to go to the Configure Screen.
reset	Press to stop a currently running method and return to the conditions in Step 0 (the equilibration step). When a Sequence is linked, the  key is only operational when the Sequence is in Hold.
purge	Press to purge the system whenever necessary (except when a Sequence is linked). You must Hold the Sequence if you want to purge.
run	Press to start running the pump method at Step 1.
stop	Press to stop the pump and reset the method to Step 0.
quit	Press quit to cancel any changes you have made to the screen and return to the previous screen.
return	Press return to save changes you have made to the current screen and return to the previous screen.
enter	Press enter to accept a typed value and move to the next position or field.
CE	Press CE to clear a typed value and redisplay the original value.
insert	Press to insert a duplicate of the current step below the current step and increment the steps following it by one (for example, when adding methods in a sequence).
delete	Press to delete the current step and decrement the steps following it by one.
Numeric Keys	Use numeric keys 1 - 9   Use numeric keys 0 and 1 (no and yes) to answer to yes/no prompts.
	Use the cursor arrow keys to accept a typed value and to move between fields within a screen or page of a screen.

Keyboard Operations

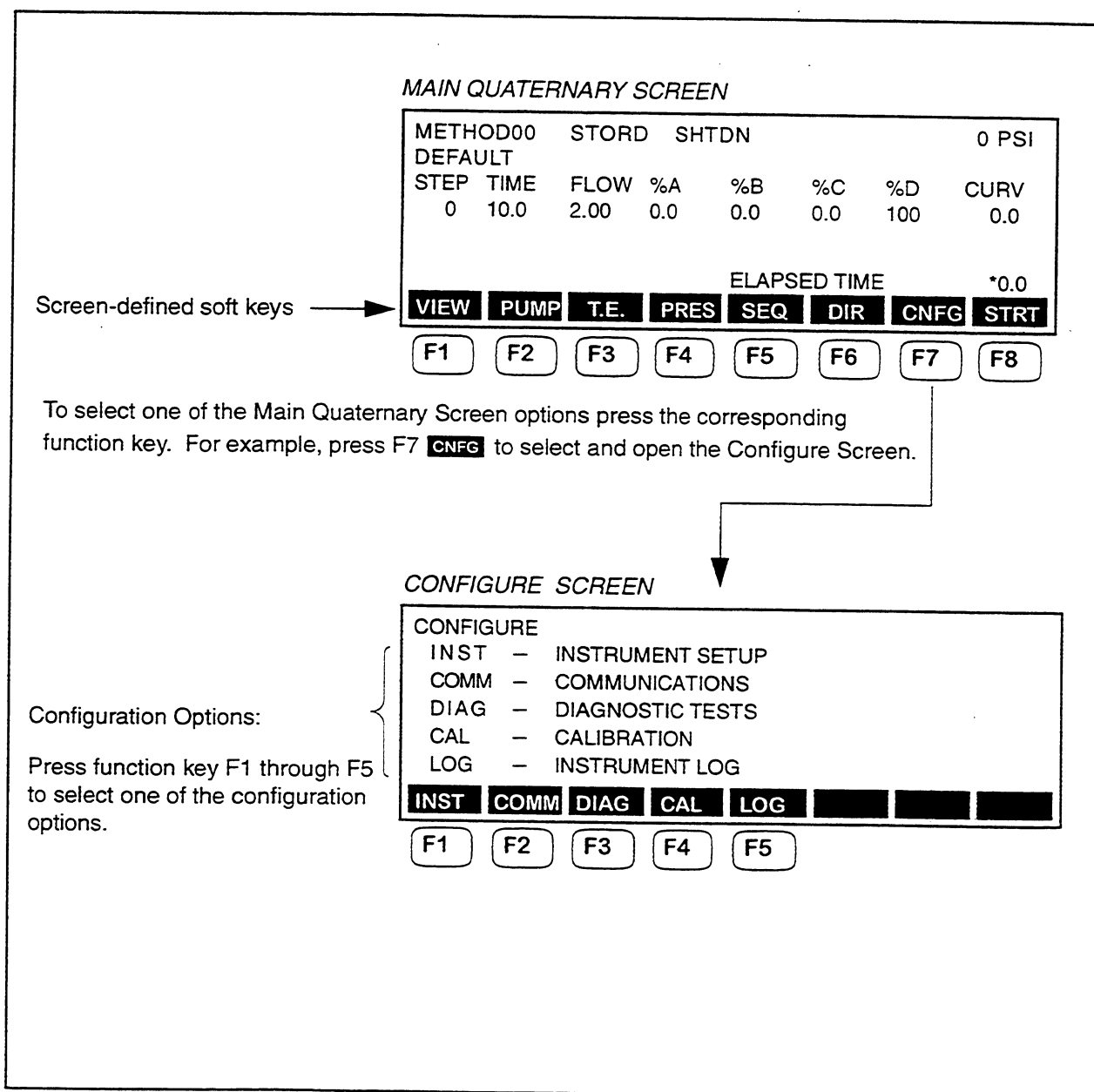
This section provides an overview of how to perform the following basic operations using the Pump keyboard:

- Selecting screen options
- Selecting data fields and entering numeric information
- Scrolling through screen lists
- Responding to screen prompts
- Saving changes to screens

Selecting Screen Options

Use the **F1** To **F8** Function Keys:

To select options displayed on a screen, either as screen-defined soft keys, or as a list of items, use the appropriate function key. Once you press the appropriate function key, the screen corresponding to the selected option appears. Typical examples include selecting options from the top level Main Quaternary Screen and the Configure Screen as shown here.



Selecting Data Fields and Entering Numeric Information

Use the **enter** key and the numeric keys

To accept the entry in a highlighted data field and move to the next field, use the **enter** key. For example, when you first enter the Pump Screen shown below, the data field for TIME is highlighted. To accept the current entry and move to and highlight the FLOW data field press the **enter** key.

To change a numeric entry use the numeric keys on the keypad.

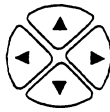
PUMP SCREEN

METHOD00	STEP	TIME	FLOW	SHTDN %A	%B	%C	%D	0 PSI CURV
	>0	10.0	2.00	0.0	0.0	0.0	100	
ELAPSED TIME								*0.0
STEP		NEW				STOR		

Use the **enter** key to accept the entry and move to the next field.

Scrolling Through Screen Lists

Use the cursor arrow keys,



If a list of items is displayed on a screen, use the cursor arrow keys to move the carrot up and down through the list of entries. The following example shows the three most common types of screen lists: the Directory Screen, Sequence Screen and the Timed Events Screen.

DIRECTORY SCREEN

carrot

List of the methods stored in the Pump's Directory.

METHOD00	STORD	SHTDN	LAST MODIFIED	
METHOD	NAME			
1	UTM		02-FEB-1994	12:00
> 2	UTM-ASSY		05-FEB-1994	07:52
3	UTM-TEST		07-MAR-1994	09:26

STORE (STOR) OR RECALL (RCL) A METHOD ?

RCL
STOR

SEQUENCE SCREEN

List of the methods, or SETs, comprising a Sequence.

SEQUENCE	SET	METHOD	FIRST	LAST	INJ
	1	2	1	1	6
>	2	3	1	1	8
	3	5	1	1	6

SET
METH
1ST
LAST
INJ
LINK
DELQ

TIMED EVENTS SCREEN

List of all the timed events for a particular method, in this case Method01.

METHOD01	SHTDN	0 PSI	
EVENT	TIME	T.E.1	T.E.2
4	11.0	YES	YES
> 5	12.0	YES	YES
6	13.0	YES	YES

EVNT
TIME
TE1
TE2
STOR
RDY

Responding to Screen Prompts

To respond to a Y/N prompt appearing in the last line of a screen, such as the one shown here prompting you to name a method, press the numeric key to select Y and the to select N.

METHOD00	STORD	SHTDN	
METHOD	NAME		LAST MODIFIED
> 2	UTM		02-FEB-1994 12:00
NAME METHOD? (Y/N) <input type="button" value="N"/>			
<input type="button" value=""/>	<input type="button" value=""/>	<input type="button" value=""/>	<input type="button" value=""/>

Saving Changes to Screens

When you finish making changes to a screen, press the key to save those changes. Changes you make to the default active method screens will remain in effect only until you turn off the power to the Pump. To permanently store changes to the default method, refer to Chapter 4 in this manual.

Instrument Specifications

Table 1-3. General Specifications.

Specification	Description
Flow Rate Range	0.01 to 10.0 mL/min (maximum pressure of 6 200 psi (420 bar) over the entire range)
Flow Rate Increments	0.01 from 0 to 0.99 mL/min and 0.1 from 1.0 to 10.0 mL/min
Flow Precision	0.3% relative standard deviation (typically <0.1%) at 1 mL/min and 1 000 psi (70) bar with water as the mobile phase
Flow Accuracy	±1% of the setting at 1 mL/min and 1 000 psi (70 bar) with water as the mobile phase
Retention Time Reproducibility	<0.3% RSD (typically <0.1% RSD)
Pressure Range	0 to 6 200 psi or 0 to 420 bar from the entire flow-rate range
Pressure Limits	High and low limit user-settable to 10 psi or 1 bar increments
Solvent Compressibility	Keyboard settable flow calibration
Composition Range	0 to 100% solvent A - D (depending on your pump configuration)
Composition Settability	Settable to 0.1%
Composition Accuracy	±1.0% absolute from 3 to 97% at 5 mL/min
Composition Precision	Typically less than 0.2% variation
Gradient Curves	Linear, exponential 1 to 9.9 (positive and negative), or step (89 profile selections)
Linearity	Within ±1% from 10 to 90% at 5 mL/min
Program Steps	Step 0 plus up to 20 solvent program steps. Battery-backed storage of up to 20 methods plus one default method.
Step Time	0 to 999 min
Step Time Increments	0.1 min, 0 to 9.9 min; 1.0 min, 10 to 99 min
Programmed Timed Events	Two built-in independent momentary (two second) contact closures
Events/Methods	Maximum of 20
Event Increments	0.1 min, 0 to 9.9 min; 1.0 min, 10 to 99 min
Dimensions (HxWxD)	14 cm x 30.5 cm x 43 cm
Weight	15.9 kg (35 lbs.)

Specification	Description
Operating Conditions:	
Environment	This instrument is designed for indoor use. It is not designed to operate in an explosive environment.
Ambient temperature	10 - 35°C
Ambient relative humidity	20 - 80% (without condensation)
Altitude	0 to 2000 meters
Installation Category	This instrument is able to withstand transient overvoltage according to Installation Category II as defined in IEC 1010-1.
Pollution Degree	This instrument will operate safely in environments that contain nonconductive foreign matter to Pollution Degree 2 in IEC 1010-1.
Storage Conditions:	
Ambient temperature	-20 to 60°C (-4 to 140°F)
Ambient relative humidity	20 - 80% (without condensation)
Altitude	0 to 12,000 meters

Table 1-4. Electrical Specifications.

Specification	Description
AC Power Requirements:	
AC power	70 Watts
AC voltage range	Grounded 100, 120, 220, 230, VAC ($\pm 10\%$) Grounded 240 VAC (+6%/-10%)
Frequency	50/60 Hz ($\pm 1\%$)
Fuse rating characteristics:	
100/120 VAC Single Fuse	1.6 A 3 AG Slow Blow, 250 V
220-240 VAC Single/Dual Fuse	0.8 A TT (5x20 mm), 250 V
Line cord	See "Installing the AC Line Cord " section in Chapter 2 of this manual.

Notes

Installation **2**

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Installation 2

This chapter describes how to install your Series 200 LC Pump. It includes the following information:

- Warnings and Precautions
- Preparing your laboratory
- Unpacking your Series 200 LC Pump
- Making the Electrical Connections
- Making the External Tubing Connections
- Making Input/Output Terminal Connections

Warnings and Precautions

Before installing the Series 200 LC Pump, read the following warnings and precautions.

Air Bubbles

To prevent air from entering the system, and to ensure that pressure fluctuation does not occur, observe the following precautions:

- Ensure that the pump's solvent inlet filter is below the solvent level in the solvent reservoir.
- If the pump has not been used for a period of time, prime the pump by removing all air bubbles. Prime the pump by connecting a priming syringe to the drain valve, opening the drain valve, then purging the pump. After priming the pump, close the drain valve completely.

Solvents

- Always use clean solvents. Solvents which have been distilled in glass (HPLC Grade) are recommended.
- Filter the solvents and buffers through a 0.5-micron medium as an additional precaution.
- Degas all aqueous and most organic solvents prior to use.
- Store flammable solvents or solvents which may form hazardous by-products when the instrument is shut down, by following the recommended shutdown procedure.
- Check compatibility of solvent(s) with the type of column(s) being used.
- Know the relative polarity and miscibility of the solvents being used.

Solvents with Low Boiling Points

Liquids that have a boiling point less than or equal to 30°C should be used under pressurization in a solvent chamber to reduce the incidence of bubble formation in the pump inlet.



Warning

Do not use carbon disulfide or other solvents which have an auto-ignition temperature below 110°C.

Buffers

Exercise care when using buffers in conjunction with organic solvents. **NEVER LEAVE BUFFERS IN THE SYSTEM OVERNIGHT.** Buffers left in the system can form salt crystals which may cause premature pump seal failure, interfere with the proper check valve operation, and plug the connecting tubing or flowcell. To remove buffers, flush the system with water followed by methanol or isopropanol. Remember to change the mobile phase from methanol or isopropanol to water before using buffers.

Corrosion

The following corrosion precautions apply to the standard (stainless steel) pump and the bio-compatible (titanium) pump.

Standard Pump

All parts of the pump that contact mobile phase are made of stainless steel, Teflon, sapphire, and glass. Some of these materials are extremely sensitive to acid chlorides. If you have questions about your mobile phase or flush solvent, contact a PerkinElmer representative. Refer to Table 2-1.

Table 2-1. Solvents Which May Corrode the Series 200 LC Pump.

Aqua Regia (80% HCl, 20% HNO ₃)	Bromine
Hydrogen Peroxide	Freon 12 (wet)
Anhydrous Chloride	Chlorinated solvents
Dichloromethane	Hydrofluorosilicic Acid (20%)
Sulfuric Acid (Conc.)	Hydrobromic Acid (20%)
Hydrochloric Acid (20% and 37%)	Ferric Chloride
Hydrofluoric Acid (20%, 50%, and 75%)	Ferrous Chloride
Copper Chloride	Mercuric Chloride (Dilute)

Biocompatible Pump

All parts of the Series 200 BIO LC Pump that contact the mobile phase are made of titanium, fluoropolymers, glass, or sapphire. This makes the Series 200 BIO LC Pump inert to the most common mobile phases that are used for biological assays. If you have questions about your mobile phase, contact a PerkinElmer representative. Refer to Table 2-2.



Caution Aqua Regia and sulfuric acid are known to dissolve titanium.

Table 2-2. Solvents Which May Corrode the Series 200 Bio LC Pump.

Aqua Regia (80% HCl, 20% HNO ₃) Hydrogen Peroxide Dichloromethane	Chlorinated solvents Sulfuric Acid (Conc.)
---	---

Solvents with Auto-Ignition Temperature Below 110°C

Certain solvents have a temperature at which they are combustible upon contact with a heated surface, even in the absence of a spark or other source of ignition. A list of common solvents with their auto-ignition temperatures are in Table 2-3.



Warning Do not use carbon disulfide or other solvents which have an auto-ignition temperature below 110°C.

Table 2-3. Auto-Ignition Temperatures of Common LC Solvents.

Solvent	Auto-Ignition Temp°C
carbon disulfide	100
diethyl ether	180
cyclohexane	260
hexane	261
petroleum ether (naphtha)	288
pentane	309
THF	321

Solvent	Auto-Ignition Temp °C
dioxane	366
propanol	404
iso-octane	418
ethanol	423
isopropyl ether	443
dimethylformamide	445
methanol	446
isopropanol	455
methylethylketone	474
acetonitrile	524
ethyl acetate	524
toluene	536
acetone	538
benzene	562

Pressure Buildup

Over time, you may observe a gradual increase in the system operating pressure. If you observe pressure readings greater than 3.45 MPa (500 psi) above the normal operating pressure of your analysis, the following items may need attention:

- If you are injecting "clean" samples and there is no other apparent cause for the pressure build-up, remove, disassemble, and clean the injector.
- If you are injecting "dirty" samples, the injector may be clogged or the column packing material may have retained contaminated particulates. Remove and clean the injector, and replace the column.
- The column end fitting or column frit may be plugged. Replace the end fitting or frit to relieve excess pressure.
- In-line filters may be plugged. Replace the filter element.
- If excessive pressure persists after the injector and column are removed from the system and the pump is connected directly to the detector input, the problem could be a plugged flowcell. Flush, reverse-flush, or rebuild the flowcell.

Preparing Your Laboratory

Before installing your Series 200 LC Pump, prepare your laboratory according to the following guidelines:

Required Air Quality

To minimize contamination problems in your laboratory, provide a relatively dust-free environment. Make sure that the following gases or vapors are not present at levels exceeding federal, state, and local ordinances for continuous human exposure:

- Flammable
- Explosive
- Toxic
- Caustic
- Corrosive

Make sure that your laboratory environment consists of the following temperature and humidity levels:

- Ambient temperature between 10°C and 35°C
- Constant relative humidity between 20% and 80%, without condensation.

Use care when working with hazardous solvents or solvents that produce hazardous by-products.



Warning

Solvent vapor levels that are high enough to interfere with the detector performance should be considered hazardous to someone who is continuously exposed to the same vapors.

Adequate Bench Space

Provide bench or table space to accommodate the dimensions of the pump listed in the specification section. The weight of the Series 200 LC Pump is 15.9 kg (35 lbs.). The bench must also support the weight of the detector and other instruments in the system. Provide space at the rear of the pump for air circulation, and provide access space on both sides of the pump.

Waste/Solvent Disposal

Provide a properly labeled chemical waste container in a safe and vented place. Make sure that it is within all of the specified safety requirements for your location. Wear gloves, eye protection, and a laboratory coat when handling or disposing of chemical waste.

Unpacking Your Series 200 LC Pump

Removing the Instrument from the Shipping Container

1. The weight of the pump is 15.9 kg (35 lbs.).
2. Grasp the pump and carefully lift it so that it clears the packing foam.
3. Check for obvious signs of damage that may have occurred during shipment. Immediately report damaged or missing items to the shipping carrier and PerkinElmer.
4. Save the original packing in case you need to return the instrument.

Items Included

When you open the shipping container, the first box you will see is the Startup Kit. Depending on your application, you will receive either the Start-Up Kit for the Series 200 LC Pump, Part No. N291-0340, or the Start-Up Kit for the Series 200 BIO Compatible LC Pump, Part No. N291-0341. For N291-0340, refer to the Start-Up Kit Packing List, Part No. 0993-8973; for N291-0341, refer to the Start-Up Kit Packing List, Part No. 0993-8974. The part numbers in these lists should help you to identify items in the kits, but do not use these part numbers to order replacements. To order replacement parts, refer to the PerkinElmer HPLC and CE Supplies Catalog or to a PerkinElmer Product Description List.

Removing the Shipping Bracket

1. Open the Access door on the front panel of the Series 200 LC Pump and locate the tab as shown in Figure 2-1.
2. Press the tab and pull the pump drawer forward. Figure 2-2 shows the pump with the pump drawer pulled out.

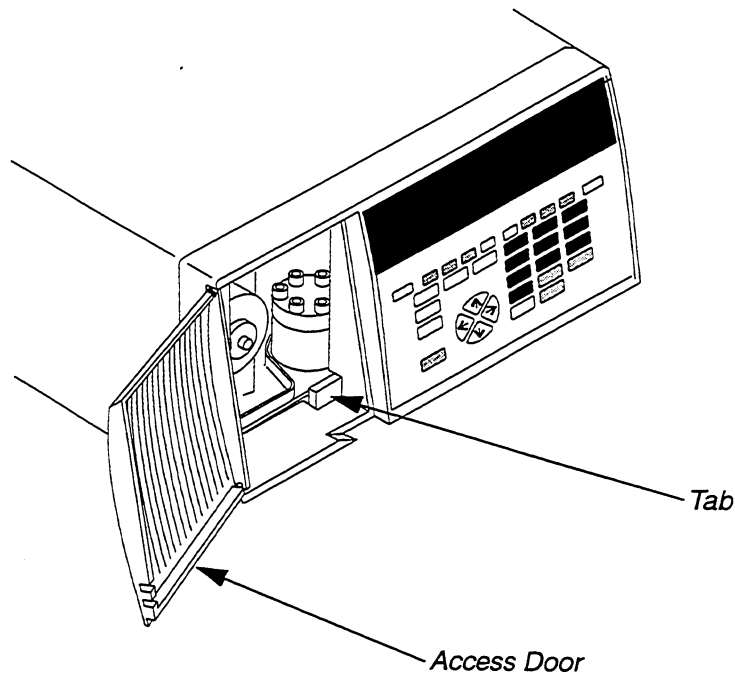


Figure 2-1. Opening the Access Door and Locating the Tab.

3. Locate the shipping bracket (Figure 2-2).
4. Cut and remove the tie wrap, then remove the shipping bracket.

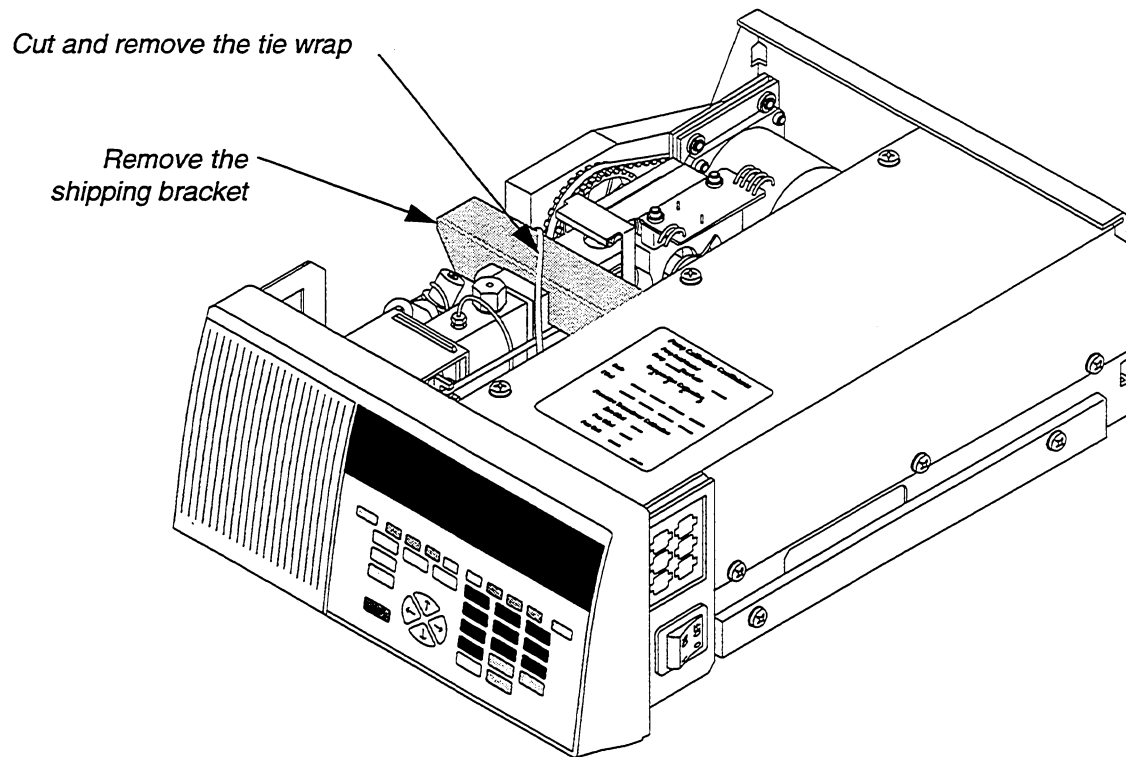


Figure 2-2. Removing the Shipping Bracket.

Making the Electrical Connections



Caution

The Series 200 LC Pump is shipped from the factory with:

- The operating voltage set to 120 VAC.
- The main AC power switch set to the OFF position.
- No fuses installed.

You need to select the appropriate voltage setting and install the proper fuse(s) before installing the line cord and applying AC power.

1

Checking Electrical Power Requirements

The Series 200 LC Pump requires a grounded, nominal 100-, 120-, 220-, 230-, or 240 VAC source. The Series 200 LC Pump uses 70 Watts maximum power.

Data processing equipment and other accessories require separate outlets. Refer to the appropriate instrument manual for the power requirements.

The AC line voltage must remain within $\pm 10\%$ of the nominal specified voltage ($+6\%/-10\%$ for 240 VAC). If the electrical supply voltage produces large AC line voltage fluctuations, install a voltage regulator between the AC line voltage outlet and the Series 200 LC Pump.

Instrument grounding is required and is accomplished by a third wire in the line voltage box. If your electrical outlet does not contain a ground, then contact your local electrical contractor.

2

Setting the Operating Voltage

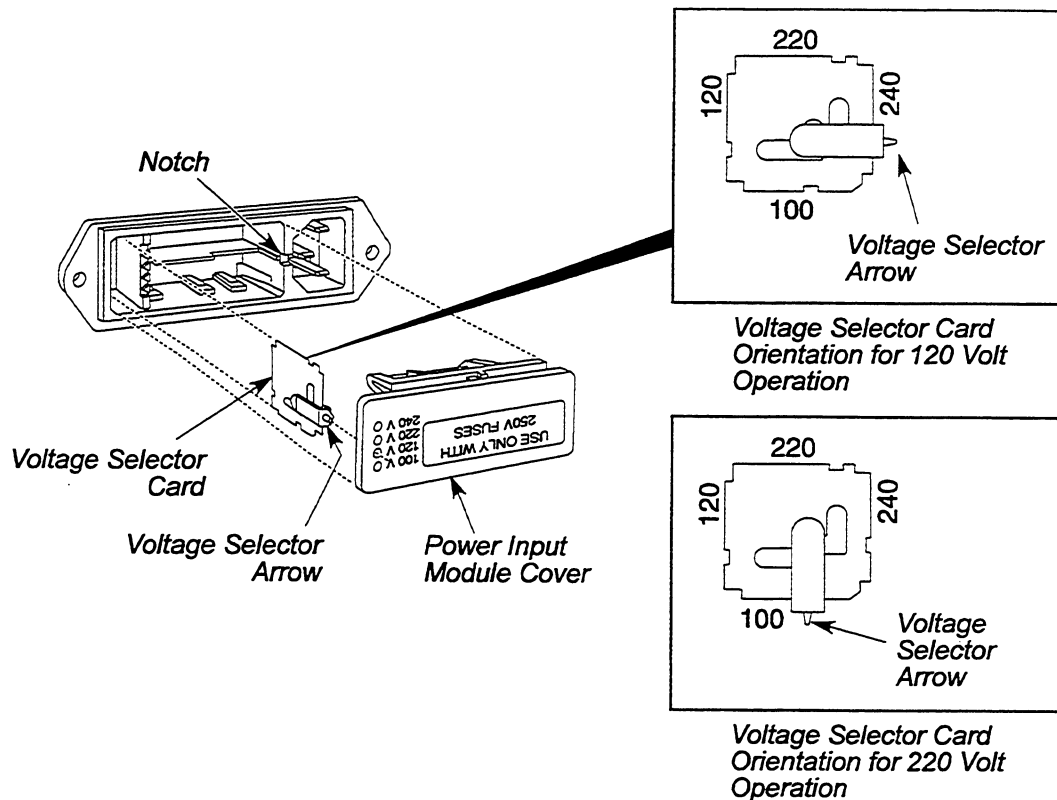
The power input module (located on the rear panel) contains an outlet for the AC line cord, the voltage selector card, and the system fuse(s). Inside the power input module is a small voltage selector card that you need to set to the local line voltage. The selected voltage is indicated by a white dot in the power input module cover next to the displayed voltage. Table 2-4 shows the setting to use for various line voltages.

Table 2-4. Voltage Selector Settings.

AC Voltage Range	AC Voltage Tap	Set Voltage Selector To:
90-110 VAC	100	100
108-132	120	120
198-242	220	220
207-253	230	240
216-264	240	240

To set the Series 200 LC Pump operating voltage:

1. Make sure the main power switch is in the OFF position.
2. Refer to Figure 2-3. Remove the power input module cover by inserting a small flat blade screwdriver into the notch and carefully pry off the cover.
3. Remove the voltage selector card from the power input module.

**Figure 2-3. Power Input Module (Exploded View).**

4. Position the number (100, 120, 220, 240) on the side of the voltage selector card so that it is **opposite** the voltage selector arrow. **THIS NUMBER MUST CORRESPOND TO THE VOLTAGE FOR YOUR LOCATION.** Figure 2-3 shows the voltage selector card settings for both the 120 V and the 220 V position.
5. Reinstall the voltage selector card into the power input module.

3 Installing the Primary Fuse(s)

After setting the voltage range, select and install the proper fuse(s). The fuse block is located on the power input module cover. See Figure 2-4 for the location of the fuse block.

Single Fuse Configuration

1. Select the required fuse from Table 2-5 below.

Table 2-5. Single Fuse Configuration Table (Use only 3AG 1/4 x 1-1/4" 250 V Fuses).

Voltage Selector	Insert the following fuse:	P/N
100	1.6 amp Slow-Blow	0998-1613
120	1.6 amp Slow-Blow	0998-1613
220	0.8 amp Slow-Blow	0999-1671
240	0.8 amp Slow-Blow	0999-1671

2. Slide the fuse into the fuse holder. See Figure 2-3.
3. Reinsert power input module cover back into power input module.

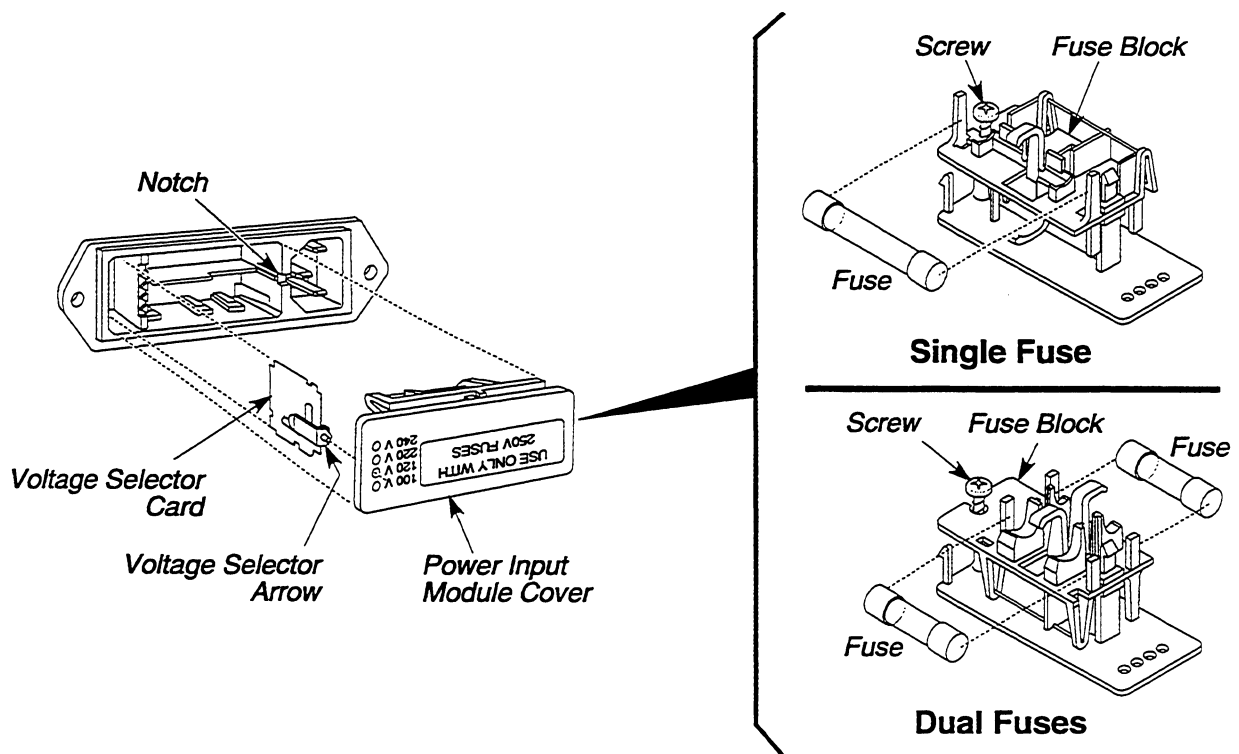


Figure 2-4. Fuse Location.

Dual Fuse Configuration

1. Select the required fuse from Table 2-6 below.

Table 2-6. Dual Fuse Configuration Table (Use only 5 x 20 mm 250V Rated Fuses).

Voltage Selector	Insert the following fuses:	P/N
220	0.8 A Type TT	0999-1670
240	0.8 A Type TT	0999-1670

2. Loosen the screw on the fuse block and remove the fuse block from the power input module cover. See Figure 2-4.
3. Invert the fuse block and reinstall it on the power input module cover. Tighten the screw.
4. Slide two fuses into the fuse block.
5. Reinsert the power module cover back into the power input module.

4

Installing the AC Line Cord

**Warning**

Ground circuit continuity is vital for safe operation of equipment.

Never operate equipment with grounding conductor disconnected. Do not remove or alter the ground pin on a three-prong line cord.

The line cord should have the electrical rating and plug appropriate for your location. See Figure 2-5 for the correct cord.

1. Select the proper line cord for your location.
2. Insure that the power switch is off and insert the line cord plug into the AC supply.


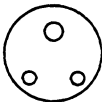
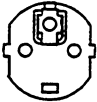
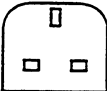


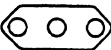
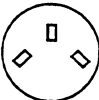


 <p><i>North America NEMA-5-15 0999-1420</i></p>	 <p><i>Old British Standard BS 546 India 0999-1423</i></p>
 <p><i>Europe CEE 7 "Schuko" 0999-1415</i></p>	 <p><i>British Standard BS 1363 United Kingdom 0999-1414</i></p>
 <p><i>Europe Switzerland 0999-1413</i></p>	 <p><i>Australia ETSA S/86 0999-1417</i></p>
 <p><i>Europe Italy 0999-1422</i></p>	 <p><i>Israel 0999-1424</i></p>
 <p><i>Denmark 0999-1416</i></p>	 <p><i>Japan 0999-1425</i></p>

Figure 2-5. AC Line Cord Selections.

Making the Tubing Connections

All PerkinElmer LC systems utilize 1/8-inch outside diameter (O. D.) tubing and fittings between the reservoirs and the pump, and 1/16-inch O.D/ 0.030-inch I. D. tubing and fittings after the pump. The 1/16-inch O.D/0.030-inch I.D. tubing is made of either stainless steel or Tefzel. Connections made with stainless steel nuts, ferrules, and tubing are rated to 6 000 psi, whereas connections made with Tefzel nuts, gripper ferrules, and tubing are rated to 1 000 psi.

1

Preparing the Tubing and Making Connections

1. Measure a piece of tubing to your required length.
2. Scribe the tubing with a triangular or knife-edge file.
3. Firmly hold the tubing on each side of the scribe mark with smooth jaw pliers, then bend the tubing back and forth to break (see Figure 2-6).
4. File any burrs off the end of the tubing.
5. First slide the nut, and then the ferrule over the end of the tubing.
6. Insert the end of the tubing into the part you wish to connect until the tubing bottoms.
7. Hold the tubing firmly in the part. Slide the nut and ferrule to the part, then turn the nut clockwise until it is fingertight.
8. Using a wrench, "set" the ferrule by tightening the nut until the ferrule "bites" into the tube (approximately 1/4 turn past fingertight), to provide a leak-free seal. **Do not over-tighten the nut!**
9. Now remove the end of the tubing from the part and insert the end in a beaker.
10. Start the pump and wait until it achieves its normal operating pressure.

If a leak occurs at the connection, tighten the fitting an additional 1/8 turn. If the leak persists, disconnect the leaking fitting and inspect it. The threads should be in good condition, the ferrule should be uniformly deformed, and the ferrule should grasp the tubing firmly. If the nut and ferrule appear to be in good condition, reconnect the fitting. If the leak persists, replace the nut and ferrule and repeat the procedure until you achieve leak-free operation.

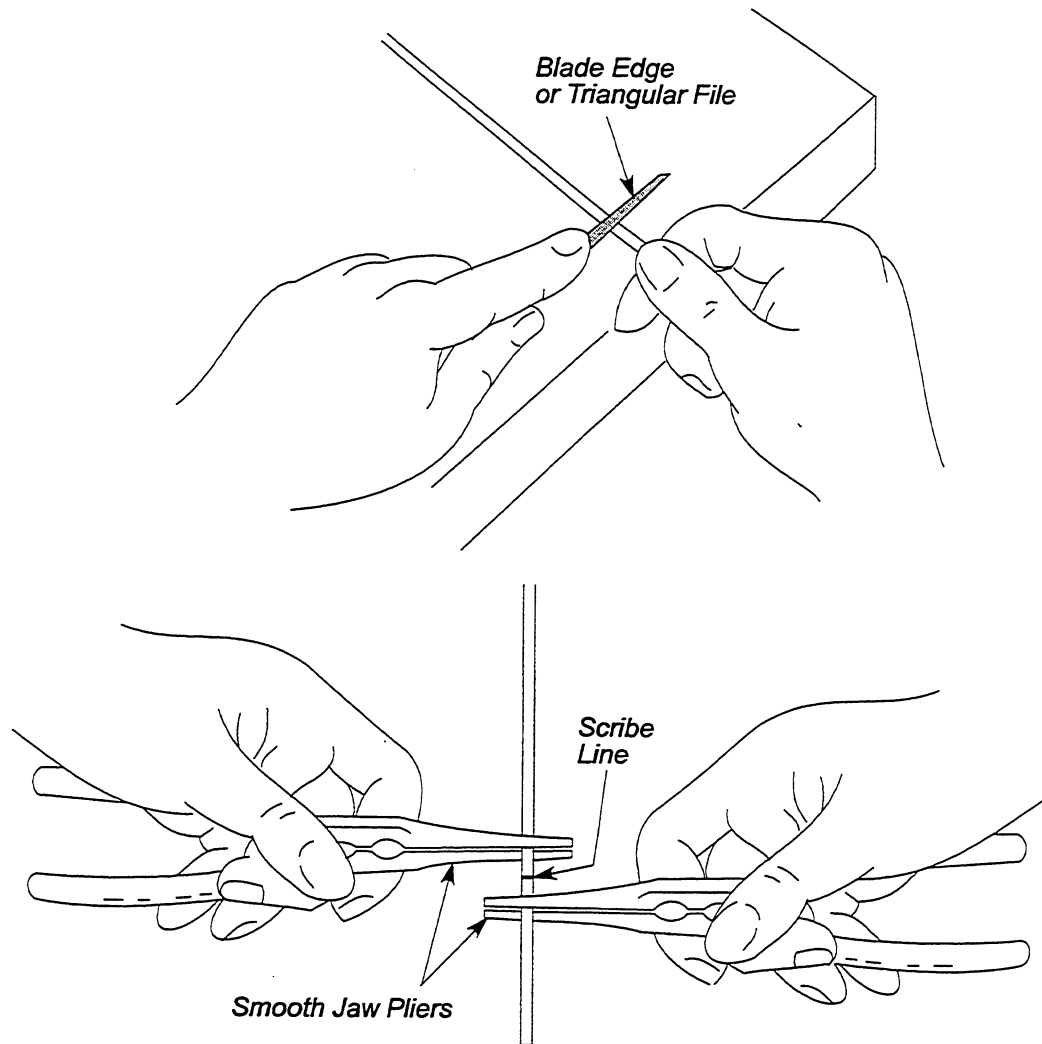


Figure 2-6. Cutting the Tubing.

2**Connecting the Pump Outlet Tubing**

You can connect the pump output tubing to exit the left or right side of the pump cabinet. The following top view of the Series 200 LC Pump shows you where to install the pump output tube. To connect the pump output tubing:

1. Refer to Figure 2-7. Open the pump access door. Locate and press the tab as you pull the pump drawer out.

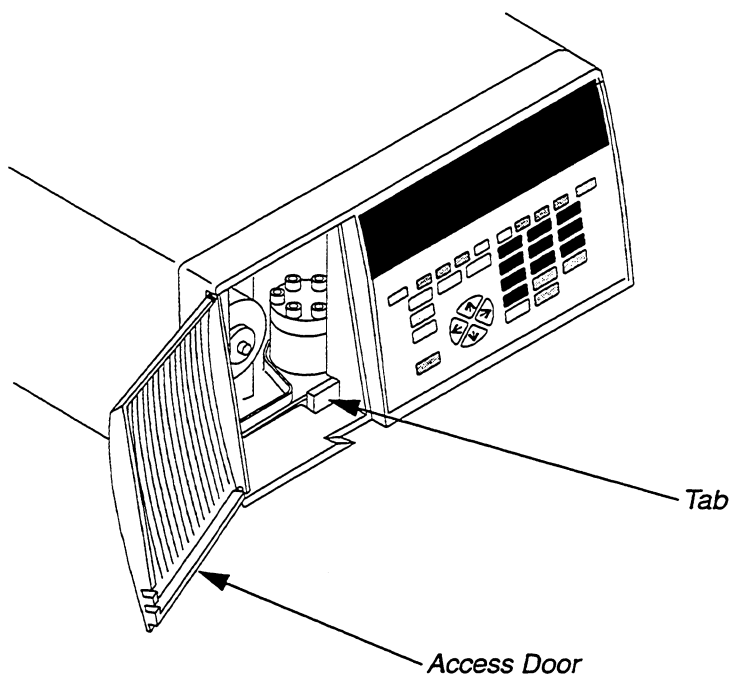


Figure 2-7. Series 200 Pump Access Door and Tab.

2. Refer to Figure 2-8. Measure and cut a piece of stainless steel tubing (P/N 0087-2036) or titanium tubing (P/N N260-1188) that you will connect between the prime/purge valve and the input port (port #2) on the injector.
3. Using the supplied nut and ferrule, connect one end of the tubing to the port on the left side of the prime/purge valve.
4. Bend the tubing to the proper shape. Then using a nut and ferrule, connect the free end to port #2 on the injector.

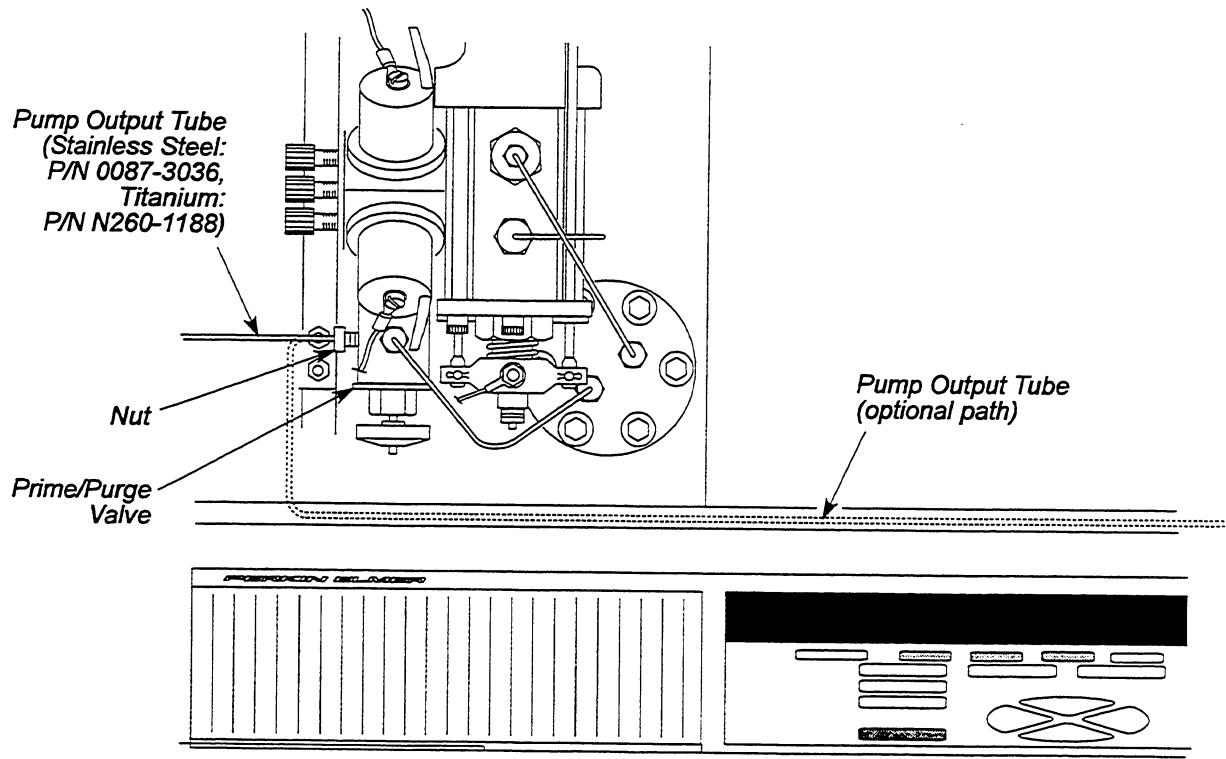


Figure 2-8. Connecting the Pump Outlet Tubing (Top View of the Pump).

3 Connecting the Series 200 LC Pump in a Typical System

Figure 2-9 shows the tubing connections that are typically made between the Series 200 LC Pump and other instruments that make up an LC system.

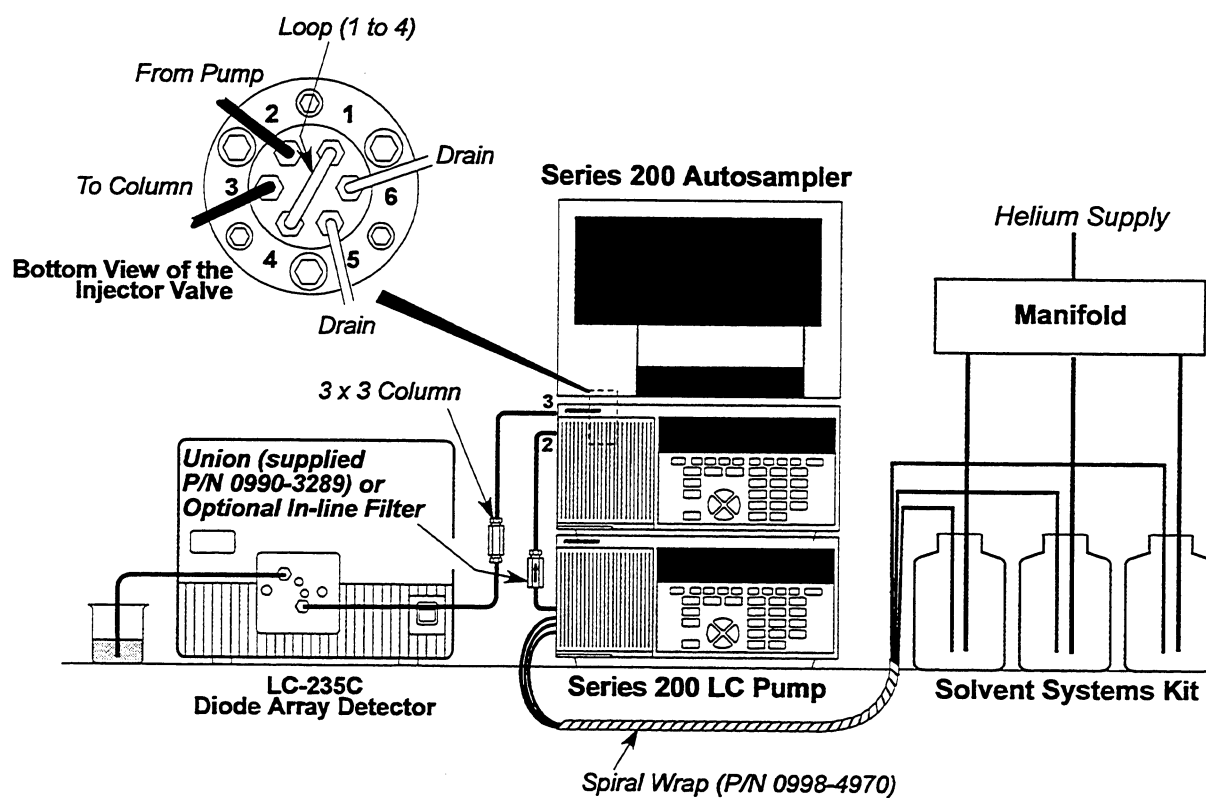


Figure 2-9. Typical System Tubing Connections.

Making Input/Output Terminal Connections

The input/output terminal on the Series 200 LC Pump uses phone jack connectors for quick contact closure connections between instruments.

The Input/Output Terminal

Figure 2-10 shows the input/output terminal connectors on the right-side panel of the Series 200 LC Pump.

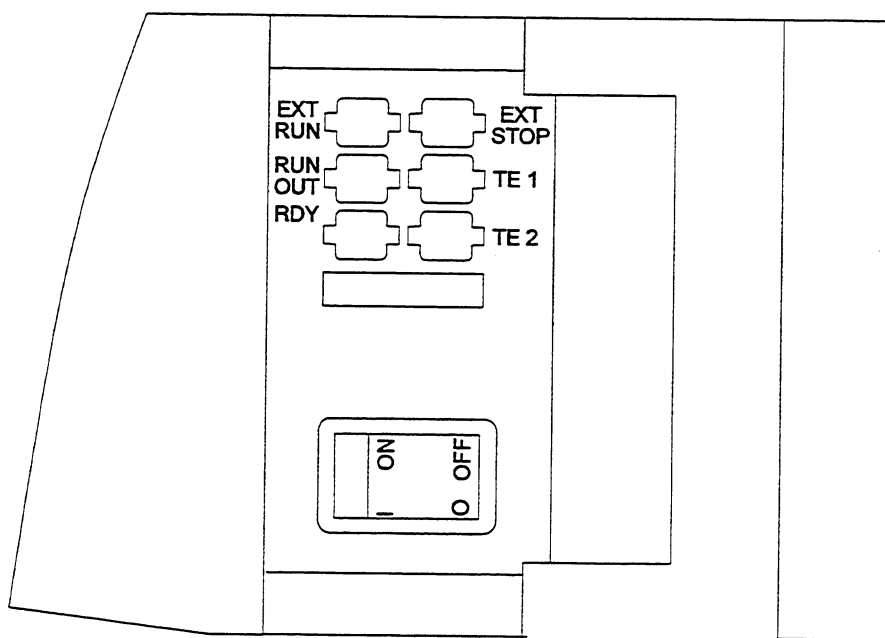


Figure 2-10. The Series 200 LC Pump Terminal Connectors.


EXT RUN (Input)

Connecting a cable to this input terminal from an external contact closure (for example, an autosampler) is identical to pressing the **run** key on the front panel. It starts the method that is currently displayed on the Series 200 LC Pump from another instrument in the LC system.

EXT STOP (Input)

Connecting an external contact closure to the external stop input is identical to pressing the **stop** key on the front panel. It stops the pump and the method that is currently running from another instrument in the LC system.

RUN OUT (Output)

This contact closure activates the moment the  key is pressed (or the external run is activated) to start the detector.

RDY (Output)

This contact closure holds closed from the moment the Method 0 time elapses and READY displays on the method screen until the run is activated. This indicates that the method has completed its equilibration and is ready to run an analysis.

TE1 and TE2 (Output)

These contact closures activate for one second when the timed event method time reaches the time you set for timed-event 1 or timed-event 2 in the timed event section of the method.

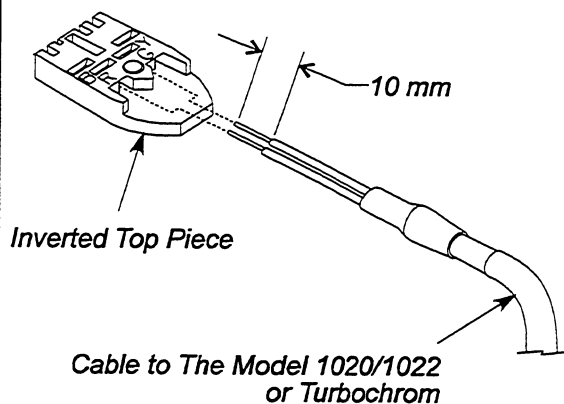
Making a Connection Between the Modular Cable and Twisted Pair Wires

You will need to make this connection when you are connecting a Series 200 Autosampler (or Model 1020/1022) to the Series 200 LC Pump. The proper way to connect the modular cable and the two wires from the Model 1020/1022 cable assembly or Series 200 Autosampler cable assembly is to use the modular connector (P/N 0999-6604) that is supplied in the Start Up Kit. To make a connection, refer to Figure 2-11 and follow these steps.

1. Cut off about 10 mm of plastic sleeve from each of the two wires. Insert the two wires into the two center positions (labeled "R" and "G") in the inverted top piece.
2. Combine the top and bottom pieces. Secure them with the Phillips screw.
3. Plug the modular connector into the I/O panel connector on the Series 200 LC Pump. Figure 2-11 shows a typical connection between the Series 200 Autosampler and the Series 200 Pump.

Preparation of Interconnecting Cable ends using Modular Connector 0999-6604**STEP 1:**

Cut off 10 mm of plastic sleeve and insert the two wires into the two center positions labeled "R" and "G".

**STEP 2:**

Combine the top piece with the bottom piece and secure them with the Phillips Screw.

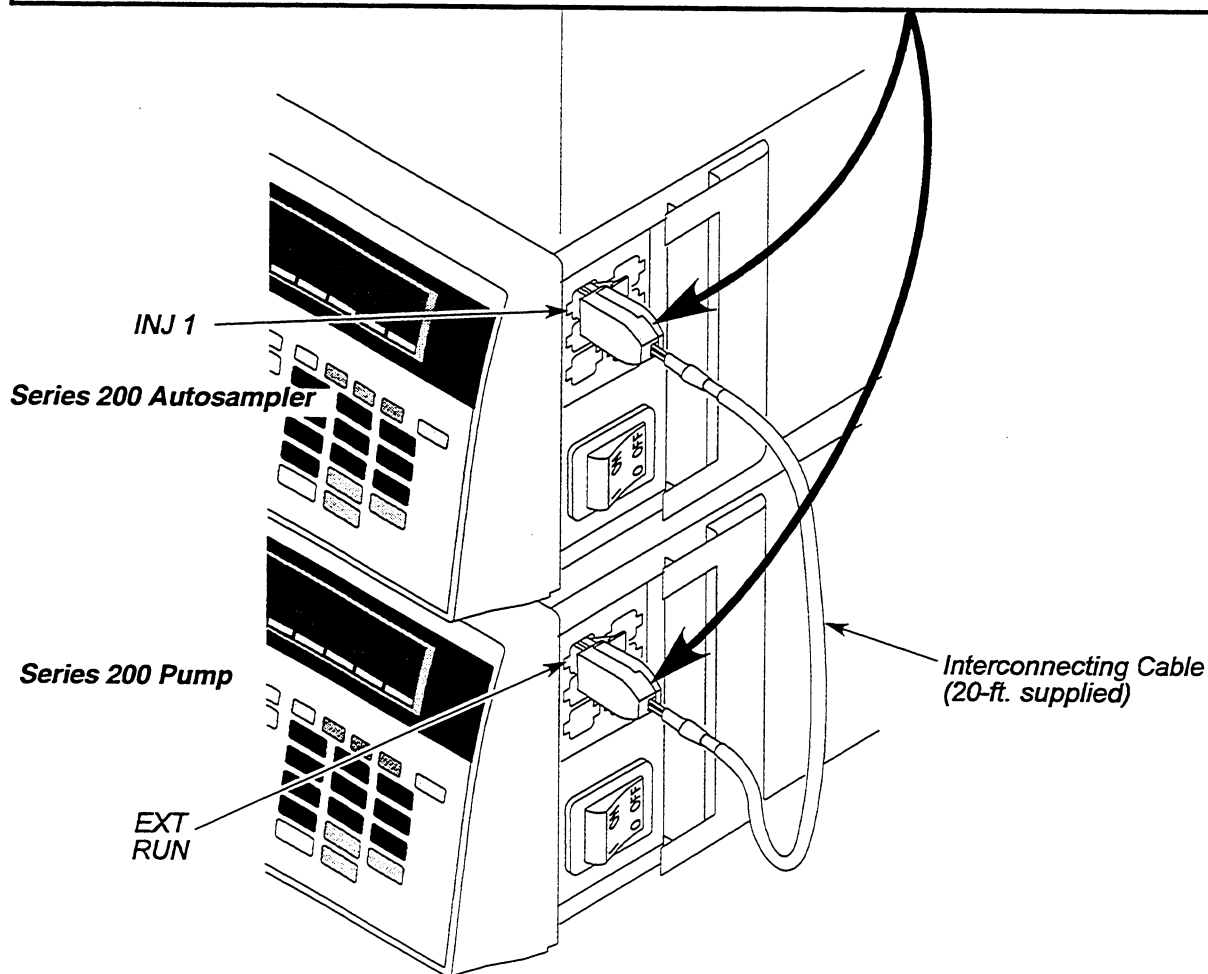
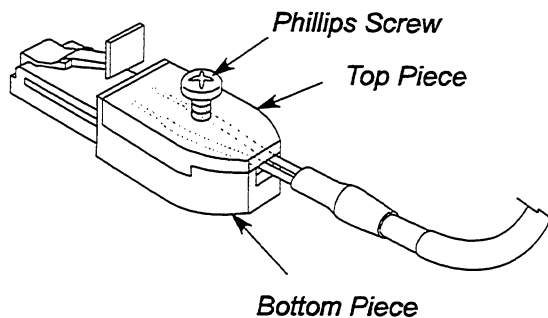


Figure 2-11. Connecting a Wire to the Modular Connector.

Making Typical System Connections

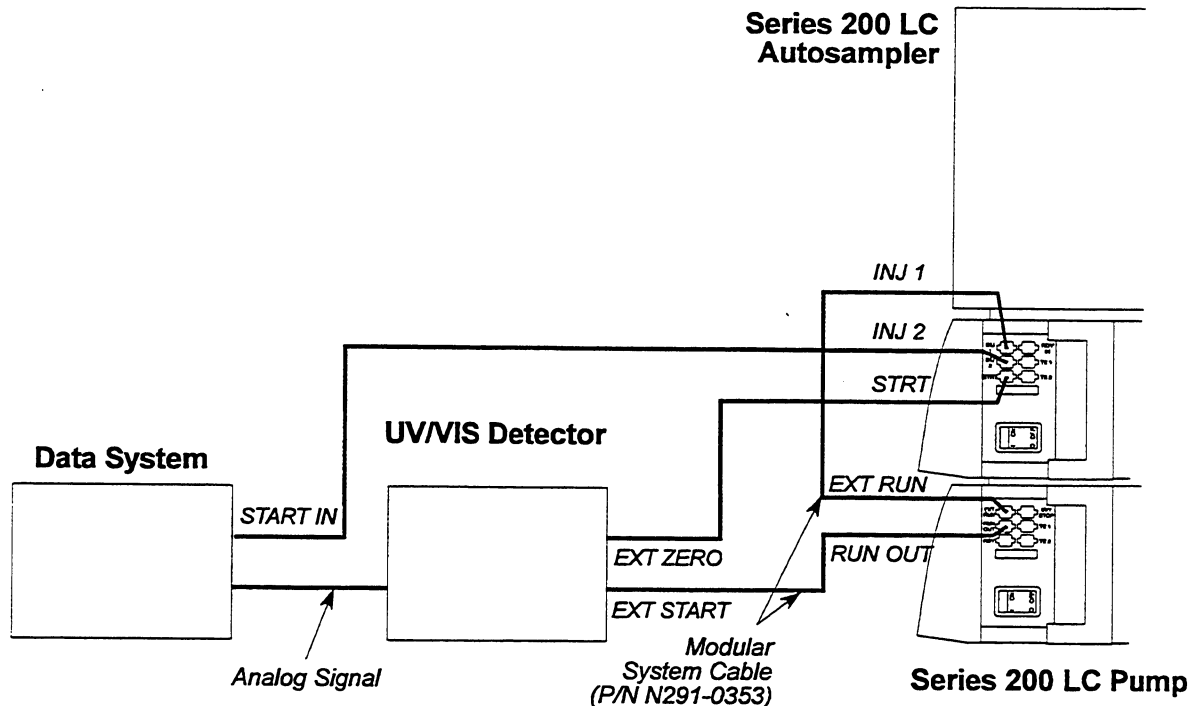


Figure 2-12. System Connection Diagram: Series 200 LC Autosampler, Series 200 LC Pump, LC UV/VIS Detector, and a Data System.

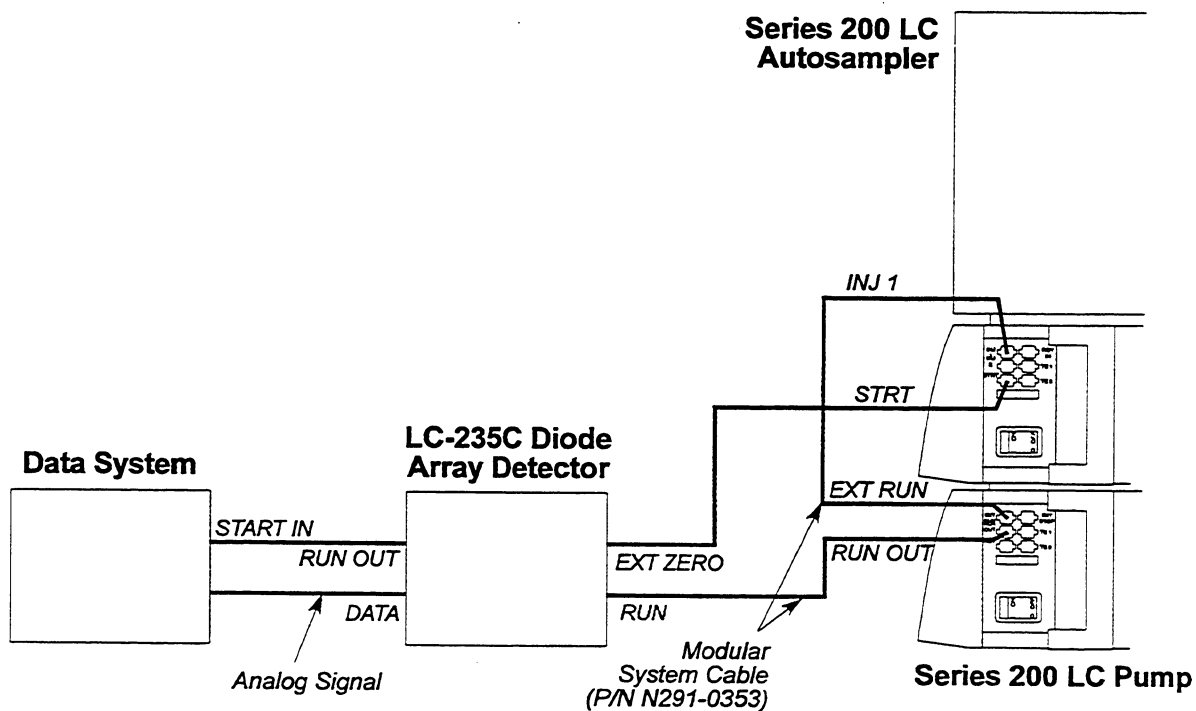


Figure 2-13. System Connection Diagram: Series 200 LC Autosampler, Series 200 LC Pump, LC-235C Diode Array Detector, and a Data System.

Configuring the Pump **3**

Powering Up the Pump ..	3-2
Defining System Parameters ..	3-3
Defining the Pressure Units ..	3-4
Adjusting the Screen Contrast ..	3-4
Setting the System Clock ..	3-5
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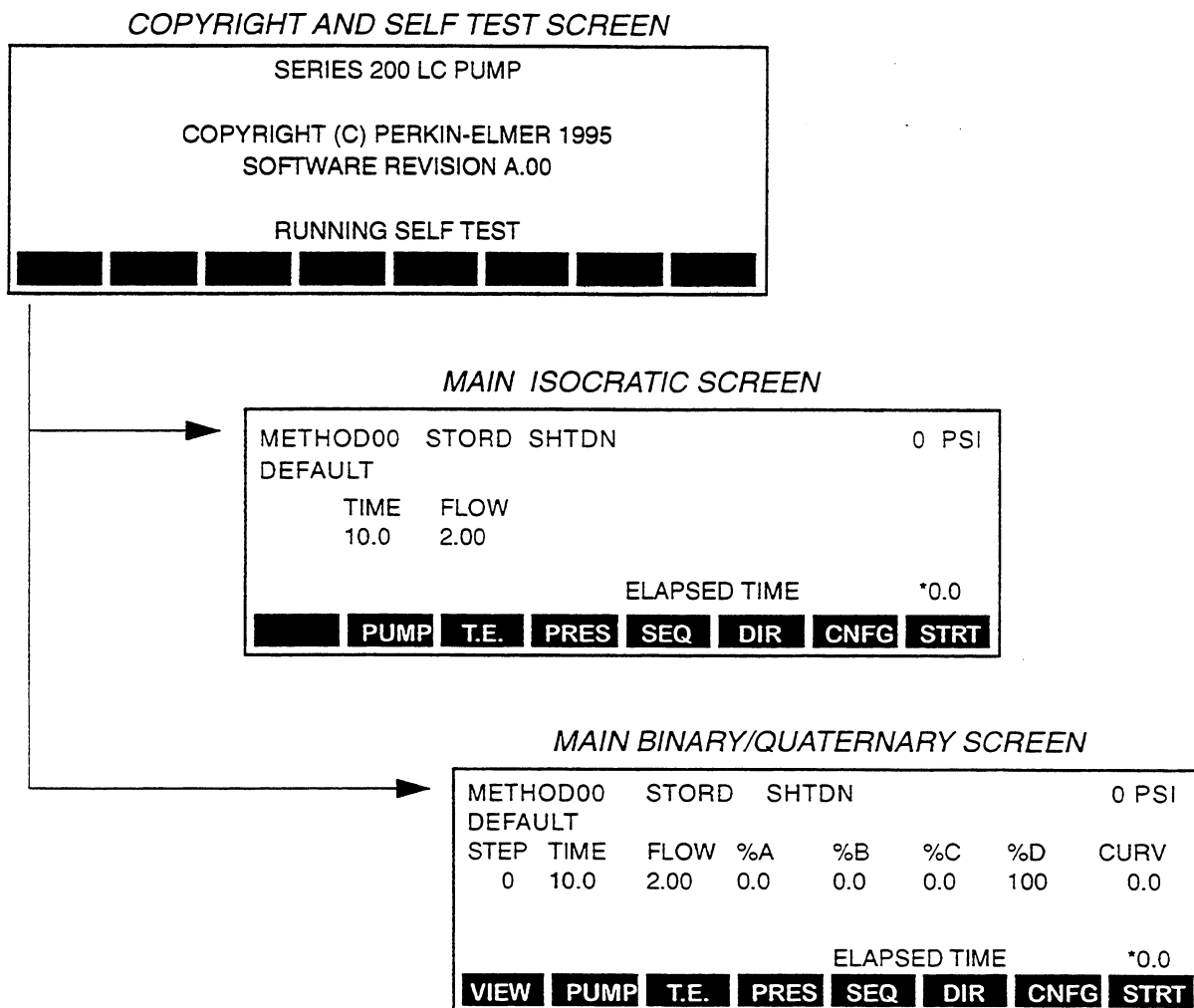
Configuring the Pump 3

This chapter describes how to configure the Series 200 LC Pump. It includes the following information:

- Powering up the Pump
- Defining System Parameters
- Selecting the Communications Option
- Accessing the Instrument Log

Powering Up the Pump

Press the power switch located on the lower right side of the Pump to the ON position. The Copyright and Self Test screen first appears on the Liquid Crystal Display. The display is located directly above the keyboard on the front panel. As soon the self test is completed, the Main Isocratic or Binary/Quaternary Screen appears. If you have one of the isocratic pump models (N291-0100 or N294-0200), the Main Isocratic Screen appears; if you have one of the binary or quaternary pump models (the N291-0101, N291-0102, N291-0201, or N291-0202), the Main Binary/Quaternary Screen appears.



The Main Screen provides access to all of the Pump's functions. The soft keys displayed along the bottom of the Main Screen corresponds to the functions that are available when you first power up the Pump. You will note that the two Main Screens are identical except for the VIEW option which is only available with the binary/quaternary models.

Defining System Parameters

This section describes how to make changes to the Pump's system parameters and includes:

- Defining the pressure units
- Adjusting the screen contrast
- Setting the system clock

You perform all of these operations from the Instrument Set-Up Screen. To access the Instrument Set-Up Screen:

METHOD00	STORD	SHTDN	0	PSI
DEFAULT				
TIME	FLOW			
10.0	2.00			
ELAPSED TIME				*0.0
PUMP	T.E.	PRES	SEQ	DIR
CNFG	STRT			

1. Press **CNFG** from the Main Screen to display the Configure Screen:

CONFIGURE	
INST	— INSTRUMENT SETUP
COMM	— COMMUNICATIONS
DIAG	— DIAGNOSTIC TESTS
CAL	— CALIBRATION
LOG	— INSTRUMENT LOG
INST	COMM
DIAG	CAL
LOG	



Caution

Do not press **DIAG. This is for service diagnostics. Do not press **CAL**. Pump calibration is set at the factory.**

2. Press **INST** to display the Instrument Set-Up Screen:

INSTRUMENT SET-UP	
UNIT	— PRESSURE UNITS
CONT	— SCREEN CONTRAST
CLK	— SYSTEM CLOCK
UNIT	CONT
	CLK

Defining the Pressure Units

INSTRUMENT SET-UP							
UNIT — PRESSURE UNITS							
CONT — SCREEN CONTRAST							
CLK — SYSTEM CLOCK							
UNIT	CONT		CLK				

1. Press **UNIT** from the Instrument Set-Up Screen to display the Pressure Units Screen:

PRESSURE UNITS							
> PSI							
PSI	BAR	MPA					

2. The current pressure unit is displayed. To change the pressure units, press **BAR** or **MPA**.
3. Press **(return)** to save the change and go back to the Instrument Set-Up Screen. To go back to the Main Screen, continue to press **(return)** until it redisplay.

Adjusting the Screen Contrast

1. To adjust the screen contrast, press **CONT** from the Instrument Set-Up Screen. The following screen appears:

ABCDEFGHIJKLMNOPQRSTUVWXYZ1234567890							
ABCDEFGHIJKLMNOPQRSTUVWXYZ1234567890							
ABCDEFGHIJKLMNOPQRSTUVWXYZ1234567890							
ABCDEFGHIJKLMNOPQRSTUVWXYZ1234567890							
CONTRAST 10							
			INC		DEC		

2. Press **INC** to increase the screen brightness (contrast) or press **DEC** to decrease the screen brightness. The contrast range is from 1 to 22, with 22 being the brightest.

3. Press **return** to go back to the Instrument Set-Up Screen. To go back to the Main Screen, continue to press **return** until it redisplay.

Setting the System Clock

1. To view and modify the system clock, press **CLK** from the Instrument Set-Up Screen. The following screen appears:

SYSTEM CLOCK			
DATE	(DA- MO-YR)	12-15-93	
TIME	(24 HR CLK)	15:06	
DAY	MNTH	YEAR	
		HOUR	MIN

2. Press the soft key that corresponds to the parameter that you want to change. Type the new value, then press **enter**.
3. After making any changes, press **return** to save the changes and go back to the Instrument Set-Up Screen. To go back to the Main Screen, continue to press **return** until it redisplay.

Selecting the Communications Option

METHOD00	STORD	SHTDN					0 PSI
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **CNFG** from the Main Screen to display the Configure Screen:

CONFIGURE							
INST	—	INSTRUMENT SETUP					
COMM	—	COMMUNICATIONS					
DIAG	—	DIAGNOSTIC TESTS					
CAL	—	CALIBRATION					
LOG	—	INSTRUMENT LOG					
INST	COMM	DIAG	CAL	LOG			

2. Press **COMM** to display the Communication Screen:

COMMUNICATION							
NONE							
PRINTER							
1020							
NONE	PRNT	1020					

3. Select the type of instrument that you are connecting to the Series 200 LC Pump by pressing the appropriate soft key.

Note Selecting the 1020 option also configures the pump for the LC 1022 and Turbochrom.

4. Press the **(return)** key to accept your selection and go back to the Configure Screen. To go back to the Main Screen, press **(return)** once more.

Accessing the Instrument Log

METHOD00	STORD	SHTDN	0 PSI
DEFAULT			
TIME	FLOW		
10.0	2.00		
ELAPSED TIME			*0.0
PUMP	T.E.	PRES	SEQ
DIR	CNFG	STRT	

1. Press **CNFG** from the Main Screen to display the Configure Screen:

CONFIGURE	
INST	- INSTRUMENT SETUP
COMM	- COMMUNICATIONS
DIAG	- DIAGNOSTIC TESTS
CAL	- CALIBRATION
LOG	- INSTRUMENT LOG
INST	COMM
DIAG	CAL
LOG	

2. Press **LOG** to display the Instrument Log Screen. This screen displays current information about your pump.

LOG	S/N XXXX	S/W	VX.XX
	PUMP CYCLES	XXXXXX	
	SEAL CYCLES	XXXXXX	
	DISPLAY HOURS	XXXXXX	
SEAL	- SET SEAL CYCLES TO ZERO		
ERR	- DISPLAY ERROR LOG		
SEAL		S/N	DISP
			ERR

3. If necessary, use one of the following soft keys to set or reset the displayed information.
 - Press **SEAL** to reset the Seal Cycles value to 0.
 - Press **S/N** to type the serial number of your pump.
 - Press **DISP** to reset the Display Hours to 0.
 - Press **ERR** to display the Error Log.
4. Press the **(return)** key to accept your changes and go back to the Configure Screen. To go back to the Main Screen, press **(return)** once more.

Notes

Creating Methods and Sequences

4

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 - Description of the Main Isocratic Screen .. **4-2**
 - Setting the Flow Rate .. **4-4**
 - Setting Timed Events and a Ready Time .. **4-5**
 - Setting the Pressure Limits .. **4-9**
- Creating a Method - Binary and Quaternary Pump Models .. **4-11**
 - Description of the Quaternary Method Screen .. **4-12**
 - Setting the Flow Rate and Solvent Composition for an Isocratic Method .. **4-16**
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Creating Methods and Sequences 4

This chapter describes how to create Methods and Sequences on your Series 200 LC Pump. It includes the following information:

- Creating a Method - Isocratic Pump Models
- Creating a Method - Binary and Quaternary Pump Models
- Modifying a Method
- Storing a Method
- Recalling a Method
- Deleting a Method
- Creating a Sequence
- Printing a Method

Note **The procedures in this chapter create methods using the values needed to run an analysis of the Universal Test Mix (P/N 0089-0893).**

Creating a Method - Isocratic Pump Models

A method is a set of conditions used to affect the column separation of samples injected by the autosampler. In an isocratic method, it consists of the solvent flow rate, the length of time the instrument pumps solvent at the specified flow rate, and the operating pressure limits. It may also include provisions for initiating timed events during the course of the method, as well as a means of limiting the amount of time the instrument continues to pump solvent after the method time expires (also referred to as the ready time).

For isocratic methods, the solvent composition remains constant throughout the separation.

This section describes how to create an isocratic method on the following Series 200 LC Pumps:

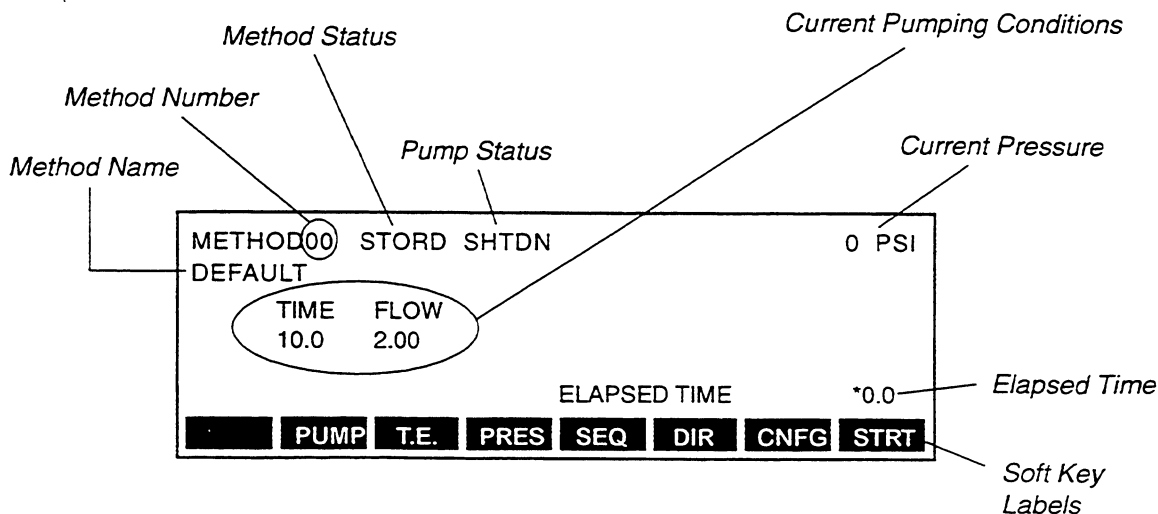
- Series 200 LC Pump Isocratic Version (50/60 Hz), Part Number N291-0100
- Series 200 Bio LC Pump Isocratic Version (50/60 Hz), Part Number N291-0200

You define an isocratic method using the following soft keys which are accessed from the Main Isocratic Screen.

1. **PUMP** key sets the flow rate
2. **T.E.** key sets timed events and a ready time
3. **PRES** key sets pressure limits

The main components of the Main Isocratic Screen are described next, followed by procedures for using the **PUMP**, **T.E.**, and **PRES** soft keys to define a pump method.

Description of the Main Isocratic Screen



Refer to Table 4-1 for a description of the Main Isocratic Screen fields.

Table 4-1. Description of the Main Isocratic Screen.

Item	Description
Method Name	Type up to eight characters to identify your method.
Method Number	Type a method number so that you can identify the method for storage and linking together method in an automated sequence. You can assign up to 20 different methods.
Method Status	<p>STORD displays when the method is stored in memory.</p> <p>CHNGD displays when you change a method value. CHNGD remains displayed until you store the method.</p> <p>This field is blank when you are creating a new method.</p>
Pump Status	<p>SHTDN displays when the pump is not running.</p> <p>START displays when you start the pump and it is pumping to achieve the minimum pressure value that is set. When START is displayed, the keyboard is locked.</p> <p>READY displays when the time in Step 0 expires.</p>
Pump Pressure	Displays the current pressure value and units.
Elapsed Time	Total time in minutes that has elapsed since pressing the STRT pump key.
Current Pumping Conditions	<p>The current values showing what the pump is doing at this exact moment.</p> <p>After pressing the PUMP key, the method parameters are displayed.</p>
Soft Key Labels	Displays the options available to you on this particular screen.

1

Setting the Flow Rate

The following procedure describes how to set flow rate using the **PUMP** soft key on the Main Isocratic Screen.

METHOD00		STORD SHTDN		0 PSI	
DEFAULT					
TIME	FLOW				
10.0	2.00				
				ELAPSED TIME	*0.0
PUMP	T.E.	PRES	SEQ	DIR	CNFG STRT

1. Press **PUMP** from the Main Isocratic Screen. The data field for Time becomes highlighted.

METHOD00		SHTDN		0 PSI	
DEFAULT					
TIME	FLOW				
10.0	2.00				
				ELAPSED TIME	*0.0
HALT				STOR	

2. Using the numeric keys, type a time value, then press **enter**. The Flow data field becomes highlighted.

METHOD00		SHTDN		0 PSI	
DEFAULT					
TIME	FLOW				
10.0	2.00				
				ELAPSED TIME	*0.0
HALT				STOR	

Press **HALT** if you want the pump to stop after the time expires. The pump will not run for the set Ready Time.

- Using the numeric keys, type a flow rate value; then press the **return** key to accept the time and flow rate values and return to the Main Isocratic Screen:

METHOD00	STORD	SHTDN	0 PSI
DEFAULT			
TIME	FLOW		
10.0	2.00		
ELAPSED TIME			*0.0
PUMP	T.E.	PRES	SEQ
DIR	CNFG	STRT	

2

Setting Timed Events and a Ready Time

Timed events control the relay contact closures which, in turn, control auxiliary instruments (for example, a column switching valve, fraction collector, autosampler, etc.). When actuated, the relays close for two seconds, then reopen. You can actuate two timed events (TE1 and TE2) a maximum of 20 times while running the method.

The ready time (READY) value is the number of minutes the pump runs after the method time expires. A ready time of 999 indicates that the pump will run indefinitely with the displayed parameters. If you want to automatically stop the pump when no sample has been injected or the run key has not been pressed, enter a ready time value of less than 999. You can use this as a way to conserve solvent after an automated sequence has ended. For example, a ready time of 10 minutes stops the pump 10 minutes after the ready time has begun.

Note You must stop the pump before you can change the ready time.

The following procedure describes how to set timed events.

METHOD00	STORD	SHTDN	0 PSI
DEFAULT			
TIME	FLOW		
10.0	2.00		
ELAPSED TIME			*0.0
PUMP	T.E.	PRES	SEQ
DIR	CNFG	STRT	

- Press **T.E.** from the Main Isocratic Screen. The Timed Events Screen appears and the data field for Time is highlighted.

METHOD00		SHTDN	EVENTS	0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	0.0			READY 999
ELAPSED TIME				*0.0
EVNT	TIME	TE1	TE2	STOR RDY

2. Using the numeric keys, type a time value (in this example, 8.0 is used) and press **enter**. The cursor moves to the T.E.1 data field.

METHOD00		SHTDN	EVENTS	0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	8.0			READY 999
				*0.0
EVNT	TIME	TE1	TE2	STOR RDY

3. Press **1 (yes)** to set T.E.1 to activate at this time or press **enter** to move the cursor to the T.E.2 data field. The following screen shows that T.E.1 is set to activate at 8 minutes.

METHOD00		SHTDN	EVENTS	0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	8.0	YES		READY 999
				*0.0
EVNT	TIME	TE1	TE2	STOR RDY

4. Press **enter** to set this entry. The cursor moves to T.E.2.

METHOD00		SHTDN	EVENTS	0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	8.0	YES		READY 999
				*0.0
EVNT	TIME	TE1	TE2	STOR RDY

5. Press **1** **yes** to set T.E.2 to activate at this time or press **enter** if you do not want to activate T.E.2. The cursor moves to the Time value for Event 2.
6. Repeat the above procedure to set timed events for Event 2 or set a READY time value.
7. Press **RDY** from the Timed Events Screen. The cursor moves to the Ready data field.

METHOD00		SHTDN	EVENTS	0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	8.0	YES		READY
				999
				*0.0
EVNT	TIME	TE1	TE2	RDY

8. Determine the number of minutes that you want the pump to run after the Step 0 time elapses. For example, if you type 10, the pump will run for 10 minutes then stop. If you type 999, the pump will run until you stop it.
9. Press the **return** key to save these changes and return to the Main Isocratic Screen.

Modifying Timed Events

If you need to make changes to the timed events, follow these steps.

METHOD00	STORD	SHTDN	0 PSI
DEFAULT			
TIME	FLOW		
10.0	2.00		
		ELAPSED TIME	*0.0
PUMP	T.E.	PRES	SEQ
DIR	CNFG	STRT	

1. Press **T.E.** from the Main Isocratic Screen to go to the Timed Events Screen.

METHOD00		SHTDN	0 PSI
EVENT	TIME	T.E.1	T.E.2
> 1	8.0	YES	
			READY
			999
EVNT	TIME	TE1	TE2
STOR	RDY		

2. Press **EVNT** on the Timed Events Screen. The following prompt appears on the bottom line of the screen as shown next.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	8.0	YES		READY 999
ENTER EVENT NUMBER 1				
EVNT		TIME	TE1	TE2
STOR				RDY

3. Using the numeric keys, type the number of the Event that you wish to modify. You may also use the cursor arrow keys to scroll through the list of Events. The following screen shows that you selected Event 1.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	8.0	YES		READY 999
EVNT		TIME	TE1	TE2
STOR				RDY

4. If you want to create Event 2, press the **insert** key on the front panel of the pump. A duplicate line of Event 1 is inserted below Event 1 and the cursor is over the time value.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
1	8.0	YES		READY
> 2	8.1	YES		999
EVNT		TIME	TE1	TE2
STOR				RDY

5. Enter a time value and press the **enter** key to set that value. Then press the soft key or press the arrow keys to select the next entry that you wish to modify.
6. When you have finished making changes to the timed events, you can store the method by pressing **STOR**. The following prompt appears on the bottom line of the Timed Events Screen.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	8.0	YES		READY 999
STORE METHOD		01		
EVNT		TIME	TE1	TE2
		STOR		RDY

To store a method, refer to the procedure under "Storing a Method" in this chapter.

3 Setting the Pressure Limits

The following procedure describes how to set the operating pressure limits. You set a minimum pressure value to detect leaks in the system. If the pressure goes below the minimum pressure value, the pump shuts down. You set a maximum pressure value to protect your column. If the pressure goes above the maximum pressure value, the pump shuts down.

METHOD00	STORD	SHTDN		0 PSI
DEFAULT				
TIME	FLOW			
10.0	2.00			
ELAPSED TIME				*0.0
	PUMP	T.E.	PRES	SEQ
		DIR	CNFG	STRT

1. Press **PRES** from the Main Isocratic Screen. The following screen appears.

METHOD00	SHTDN		0 PSI
MINIMUM PRESSURE 0 PSI			
MAXIMUM PRESSURE 6100 PSI			
* IMPORTANT - CONSULT COLUMN INFO SHEET *			
ELAPSED TIME			*0.0
	MIN	MAX	STOR

2. Press **MIN** to highlight the minimum pressure limit value. Type the minimum pressure limit value and press the **enter** key. If the operating pressure falls below this value, the pump stops. The following screen shows the minimum pressure limit value set to 0 psi.

Note The pressure units are set from the Instrument Set-Up screen under the Configure menu.

METHOD00	SHTDN	0 PSI
MINIMUM PRESSURE 0 PSI		
MAXIMUM PRESSURE 6100 PSI		
* IMPORTANT - CONSULT COLUMN INFO SHEET *		
ELAPSED TIME		*0.0
	MIN	MAX
		STOR

3. Press **MAX** to highlight the maximum pressure limit value. Type the maximum pressure limit value and press the **enter** key. If the pumping pressure exceeds this value, the pump stops. The following screen shows the maximum pressure limit value set to 6100 psi.

METHOD00	SHTDN	0 PSI
MINIMUM PRESSURE 0 PSI		
MAXIMUM PRESSURE 6100 PSI		
* IMPORTANT - CONSULT COLUMN INFO SHEET *		
ELAPSED TIME		*0.0
	MIN	MAX
		STOR

4. Press the **return** key to save all entries and return to the Main Isocratic Screen or press **STOR** to store this method. If you press **STOR**, the following prompt appears on the bottom of the screen.

METHOD00	SHTDN	0 PSI
MINIMUM PRESSURE 0 PSI		
MAXIMUM PRESSURE 6100 PSI		
* IMPORTANT - CONSULT COLUMN INFO SHEET *		
STORE METHOD	01	
	MIN	MAX
		STOR

To store a method, refer to the procedure under "Storing a Method" in this chapter.

Creating a Method - Binary and Quaternary Pump Models

A method is a set of conditions used to affect the column separation of samples injected by the autosampler. It consists of the solvent flow rate, the length of time the instrument pumps solvent at the specified flow rate, the solvent composition, the gradient curvature (how the solvent composition changes from one step to another) and the operating pressure limits. It may also include provisions for initiating timed events during the chromatography portion of the method, as well as a means of limiting the amount of time the instrument continues to pump solvent after the method time expires (also referred to as the ready time).

This section describes how to create an isocratic method and a gradient method on the following Series 200 LC Binary and Quaternary Pumps.

- 200 LC Pump Binary Version (50/60 Hz), N291-0101
- Series 200 LC Pump Quaternary Version (50/60 Hz), N291-0102
- Series 200 BIO LC Pump Binary Version (50/60 Hz), N291-0201
- Series 200 BIO LC Pump Quaternary Version (50/60 Hz), N291-0202

In an isocratic method, the Series 200 LC Pump mixes the solvents at a fixed composition throughout the separation. In a gradient method the Series 200 LC Pump mixes and changes the solvent composition throughout the separation.

Both the isocratic and gradient methods defined in this section assume the methanol and water solvents are in the following pump reservoirs.

Reservoir	Solvent
A	Methanol
B	Not used
C	Not used
D	Water

The components of the Main Quaternary Screen are described next. This is followed by an overview of the isocratic and gradient methods that will be set up in the procedures in this section. Remember, the values used in these methods are the ones needed to run an analysis of the Universal Test Mix (P/N 0089-0893).

Lastly, the procedures themselves are described. You will use the **PUMP**, **T.E.**, and **PRES** soft keys which are accessed from the Main Quaternary Screen to define the methods.

Note that the main difference between setting up the isocratic method and the gradient method (on a binary or quaternary pump) involves defining the solvent composition changes for the gradient method. The procedures for setting up the timed events, defining a ready time, and setting the pressure limits are the same for both methods.

Description of the Quaternary Method Screen

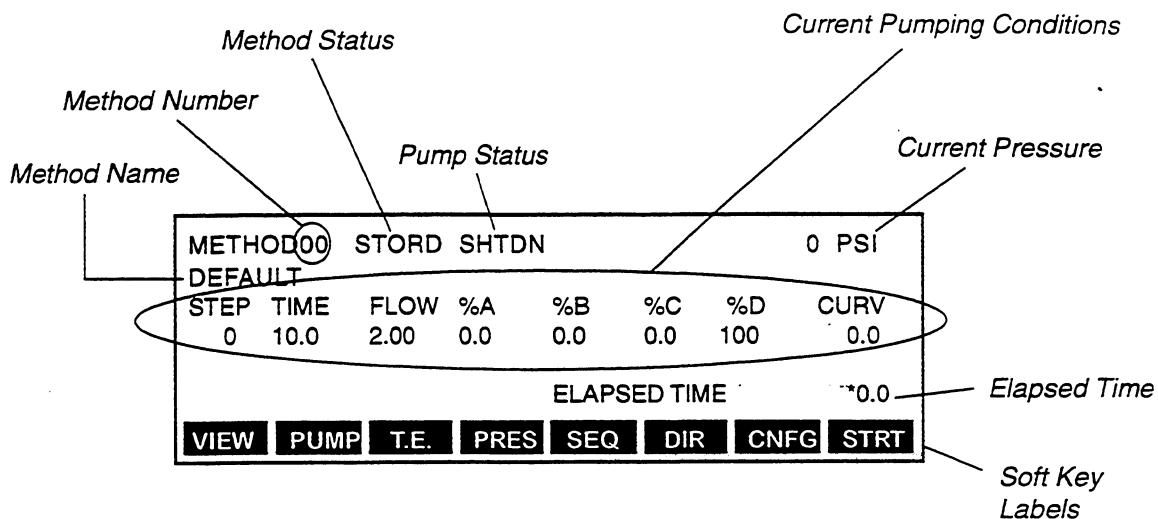


Table 4-2. Description of the Quaternary Method Screen.

Item	Description
Method Name	Type up to eight characters to identify your method.
Method Number	Type a method number so that you can identify the method for storage and linking together method in an automated sequence. You can assign up to 20 different methods.
Method Status	<p>STORD displays when the method is stored in memory.</p> <p>CHNGD displays when you change a method value. CHNGD remains displayed until you store the method.</p> <p>This field is blank when you are creating a new method.</p>
Current Pumping Conditions	<p>The current values showing what the pump is doing at this exact moment.</p> <p>After pressing the PUMP key, the method parameters are displayed.</p>

Item	Description
Pump Status	<p>SHTDN displays when the pump is not running.</p> <p>START displays when you start the pump and it is pumping to achieve the minimum pressure value that is set. When START is displayed, the keyboard is locked.</p> <p>EQUIL displays while Step 0 is running.</p> <p>READY displays when the time in Step 0 expires.</p> <p>RUN01-RUN19 displays when the pump is running. The number (01-19) corresponds to the step number.</p> <p>HOLD01-HOLD19 displays when the hold key is pressed. The number (01-19) corresponds to the step number.</p>
Pump Pressure	Displays the current pressure value and units.
Elapsed Time	Total time in minutes that has elapsed since pressing the STRT pump key.
Soft Key Labels	Displays the options available to you on this particular screen.

Isocratic Method:

For the isocratic method, the solvent composition is 70% A and 30% D. The method consists of a 10 minute equilibration step (step 0), an indefinite ready time during which the pump will run at equilibration conditions until the **run** key is pressed, and a 5 minute chromatography run (step 1). The time line for the isocratic method is shown graphically in Figure 4-1.

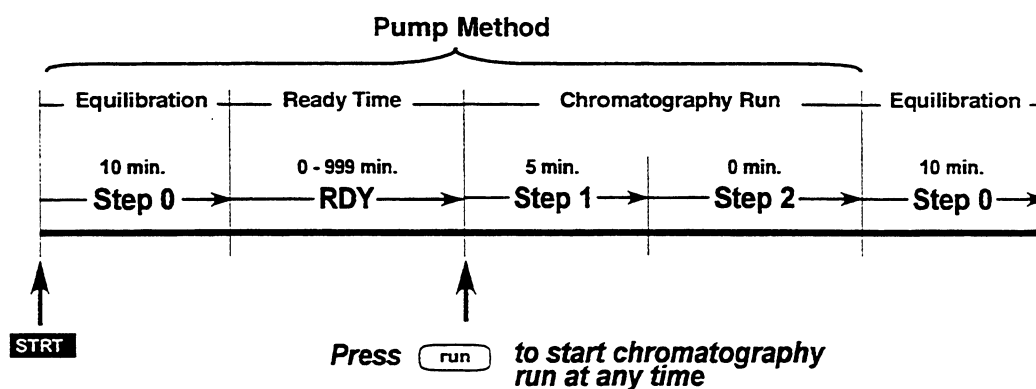


Figure 4-1. Time line representation of an isocratic analysis.

Gradient Method:

The gradient method consists of a 10 minute equilibration step (step 0), an indefinite ready time during which the pump will run at equilibration conditions until the **run** key is pressed, and a three-step chromatography run. The steps in the chromatography run include:

- Step 1 which runs for 5 minutes at a solvent composition of 10% A and 90% D.
- Step 2 which runs for 5 minutes and during which time the solvent composition changes from 10% A and 90%D to 70%A and 30%D.
- Step 3 runs for 5 minutes at a solvent composition of 70%A and 30%D.

The manner in which the solvent composition changes in going from one step to another (in this method, from step 1 to step 2) is determined by the solvent composition curve for the gradient method at each step.

Solvent Composition Curves

When the time expires in the current step of a gradient method, the solvent composition can:

- change instantaneously to the solvent composition of the next step, or,
- change gradually to the solvent composition of the next step by a linear or proportional relationship over the specified time.

You select the method of change during the method setup by entering an appropriate value in the CURV data field. For example, a value of "0" represents an instantaneous change; a value of "1" represents a linear change. Figure 4-2 shows the relationship between curve number and the rate of solvent composition change. You can select up to 176 different curves (by selecting curve values ± 0.1 to 9.9).

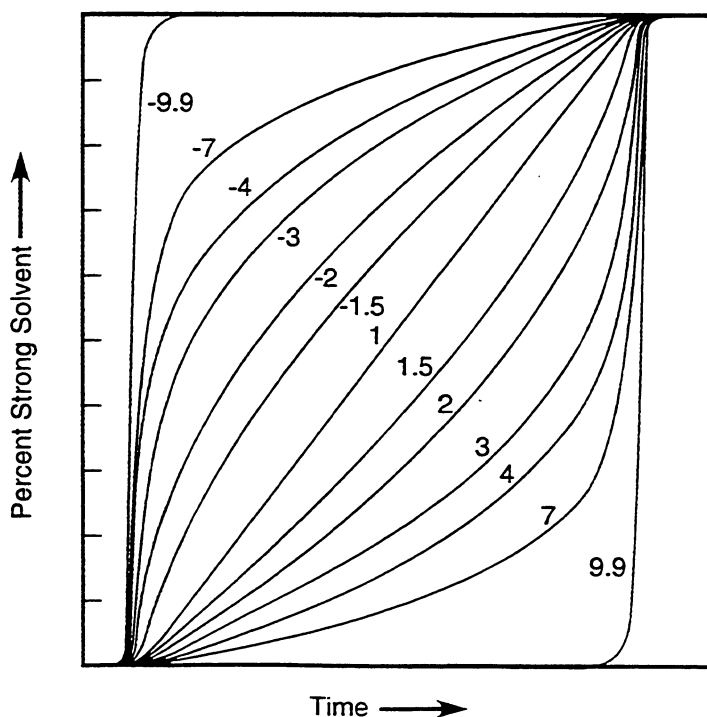


Figure 4-2. Graphic representation of selected solvent composition curves.

The time line for the gradient method is shown graphically in Figure 4-3.

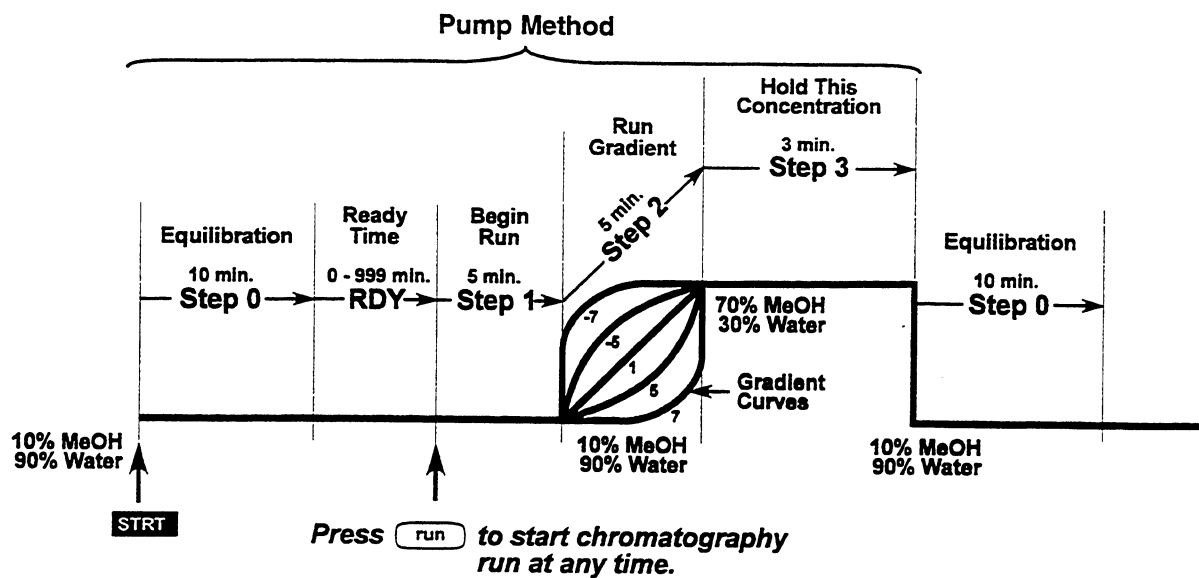


Figure 4-3. Time line representation of this gradient analysis.

METHOD00	SHTDN						0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
>0	10.0	2.00	0.0	0.0	0.0	100	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

3. Type the flow rate value and press the **enter** key. In this example we will leave the flow rate at 2.00 mL/minute.

METHOD00	SHTDN						0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
>0	10.0	2.00	0.0	0.0	0.0	100	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

4. Type the 70 for the % of Reservoir A (methanol) and press the **enter** key. Notice that the %D automatically changes to 30%.

METHOD00	SHTDN						0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
>0	10.0	2.00	70.0	0.0	0.0	30	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

5. Since we are not going to make any changes to this solvent concentration for Step 0, you can:

Press the **enter** key three times to move the cursor to the time value in Step 1.

or

Press the **insert** key to make Step1 a duplicate of Step 0. Then move the cursor to the time value by pressing the left arrow key three times.

METHOD00		SHTDN					0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30	
> 1	10.0	2.00	70.0	0.0	0.0	30	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

6. Type a time value for Step 1 and press the key. In this example, we will type a time of 5 minutes.

METHOD00		SHTDN					0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30	
> 1	5.0	2.00	70.0	0.0	0.0	30	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

7. Since we are not creating Step 2, we can assign a method number and method name by pressing and answering the prompts. For a detailed procedure, refer to **"Storing a Method"** in this chapter. The following screen shows that we named the method UTM and stored the method as number 1.

METHOD01		STORD	SHTDN				0 PSI
UTM							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1B Setting the Flow Rate and Solvent Composition for a Gradient Method

To set the flow rate and solvent composition for a gradient method, follow these steps.

METHOD00		STORD		SHTDN		0 PSI	
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **PUMP** from the Main Quaternary Screen. The data field for Time becomes highlighted.

METHOD00		SHTDN					0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
>0	10.0	2.00	0.0	0.0	0.0	100	
			* ELAPSED TIME				*0.0
STEP		NEW			STOR		

- Using the numeric keys, type a time value and press the **enter** key. In this example we will leave the time at 10.0 minutes.

Note:

Pressing **STEP** allows you to select a step number. However, since this is the Default method, it only has Step 0. We will show you how to use this later when you modify a method.

Pressing **NEW** will cancel all of the changes you have made so far to screen and allow you to create a new method.

Pressing **STOR** allows you to store the current method under a new method number and name. For more information, refer to "Storing a Method" in this chapter.

METHOD00	SHTDN		0 PSI				
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
>0	10.0	2.00	0.0	0.0	0.0	100	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

3. Type the flow rate value and press the **enter** key. In this example we will leave the flow rate at 2.00 mL/minute.

METHOD00	SHTDN		0 PSI				
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
>0	10.0	2.00	0.0	0.0	0.0	100	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

4. Type 10 for the % of Reservoir A (methanol) and press the **enter** key. Notice that the %D automatically changes to 90%.

METHOD00	SHTDN		0 PSI				
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
>0	10.0	2.00	10.0	0.0	0.0	90	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

5. Since we are not going to make any changes to this solvent concentration for Step 0, you can:

Press the **enter** key three times to move the cursor to the time value in Step 1.

or

Press the **insert** key to make Step 1 a duplicate of Step 0. Then move the cursor to the time value by pressing the left arrow key three times.

METHOD00			SHTDN				0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	10.0	0.0	0.0	90	
> 1	10.0	2.00	10.0	0.0	0.0	90	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

6. Type a time value for Step 1 and press the **enter** key. In this example, we will type a time of 5 minutes.

METHOD00			SHTDN				0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	10.0	0.0	0.0	90	
> 1	5.0	2.00	10.0	0.0	0.0	90	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

7. Since we are not going to make any changes to this solvent concentration for Step 1, you can:

Press the **enter** key eight times to move the cursor to the %A value in Step 2.

or

Press the **insert** key to make Step 2 a duplicate of Step 1 as shown in the next screen.

METHOD00			SHTDN				0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
1	5.0	2.00	10.0	0.0	0.0	90	
> 2	5.0	2.00	10.0	0.0	0.0	90	
ELAPSED TIME							*0.0
STEP	HALT	NEW			STOR		

8. Move the cursor to the %A value by pressing the right arrow key once. Type 70 for the % of Reservoir A (methanol) and press the **enter** key. Notice that the %D automatically changes to 30% and a value of 0.0 appears under CURV. Press the right arrow key three times to highlight the CURV value and type 1.

Also notice that **HALT** appears. Press **HALT** to make this step a HALT step. HALT stops the pump after the time in the previous step expires. You can remove the HALT step by highlighting HALT and pressing the **delete** key.

METHOD00	SHTDN						0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
1	5.0	2.00	10.0	0.0	0.0	90	
> 2	5.0	2.00	70.0	0.0	0.0	30	1.0
ELAPSED TIME							*0.0
STEP	HALT	NEW			STOR		

The solvent composition will change linearly from 10% to 70% A over a time a five minutes, then Step 2 will run at 70% MeOH/Water for five minutes.

9. Press the **enter** key to move the cursor to the time value in Step 3.

or

Press the **insert** key to make Step 3 a duplicate of Step 2. Then move the cursor to the time value by pressing the left arrow key four times as shown in the next screen.

METHOD00	SHTDN						0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
2	5.0	2.00	70.0	0.0	0.0	30	1.0
> 3	5.0	2.00	70.0	0.0	0.0	30	
ELAPSED TIME							*0.0
STEP	HALT	NEW			STOR		

10. Type a time value of three minutes and press the **enter** key to hold at the top of the gradient.

METHOD00	SHTDN						0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
2	5.0	2.00	70.0	0.0	0.0	30	1.0
> 3	3.0	2.00	70.0	0.0	0.0	30	
ELAPSED TIME							*0.0
STEP	HALT	NEW			STOR		

11. Since we are not creating Step 4, we can assign a method number and method name by pressing **STOR** and answering the prompts. For a detailed procedure, refer to "Storing a Method" in this chapter. The following screen shows that we named the method UTM-GRAD and stored the method as number 1:

METHOD01	STORD	SHTDN					0 PSI
UTM-GRAD							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	10.0	0.0	0.0	90.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

2 Setting Timed Events and a Ready Time

The steps in this section are the same for either an isocratic method or a gradient method.

Timed events control the relay contact closures which, in turn, control auxiliary instruments (for example, a column switching valve, fraction collector, autosampler, etc.). When actuated, the relays close for two seconds, then reopen. You can actuate two timed events (TE1 and TE2) a maximum of 20 times while running the method.

The ready time (READY) value is the number of minutes the pump runs after the time in Step 0 ends. A time of 999 indicates that the pump will run indefinitely with the displayed parameters. If you want to automatically stop the pump when no sample has been injected or the run key has not been pressed, enter a ready time value of less than 999. You can use this as a way to conserve solvent after an automated sequence has ended. For example, a ready time of 10 minutes stops the pump 10 minutes after the ready time has begun.

The following procedure describes how to set timed events.

METHOD00	STORD	SHTDN					0 PSI
UTM							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	10.0	0.0	0.0	5.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **T.E.** from the Main Quaternary Screen. The Timed Events Screen appears and the data field for Time is highlighted.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	0.0			READY 999
EVNT		TIME	TE1	TE2
		STOR		RDY

2. Type a time value and press . The cursor moves to the T.E.1 data field.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	11.0			READY 999
EVNT		TIME	TE1	TE2
		STOR		RDY

3. Press to set T.E.1 to activate at this time or press to move the cursor to the T.E.2 data field. The following screen shows that T.E.1 is set to activate at 11 minutes.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	11.0	YES		READY 999
EVNT		TIME	TE1	TE2
		STOR		RDY

4. Press to set this entry. The cursor moves to T.E.2.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	11.0	YES		READY 999

EVNT TIME TE1 TE2 STOR RDY

5. Press **1** **(yes)** to set T.E.2 to activate at this time or press **(enter)** if you do not want to activate T.E.2. The cursor moves to the Time value for Event 2.
6. Repeat the above procedure to set timed events for Event 2 or set a READY time value.
7. Press **RDY**. The cursor moves to the time value under READY.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	11.0	YES		READY 999

EVNT TIME TE1 TE2 STOR RDY

8. Determine the number of minutes that you want the pump to run after the Step 0 time elapses. For example, if you type 10, the pump will run for 10 minutes then stop. If you type 999, the pump will run until you press **(stop)**
9. Press the **(return)** key to redisplay the current method.

Modifying Timed Events

If you need to make changes to the timed events, follow these steps.

METHOD00	STORD	SHTDN					0 PSI
UTM							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **T.E.** from the Main Quaternary Screen to go to the Timed Events Screen.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
				READY
> 1	11.0	YES		999
EVNT		TIME	TE1	TE2
		STOR		RDY

2. Press **EVNT** on the Timed Events Screen. The following prompt appears on the bottom line of the screen as shown next.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
				READY
> 1	11.0	YES		999
ENTER EVENT NUMBER <input type="text" value="1"/>				
EVNT		TIME	TE1	TE2
		STOR		RDY

3. Using the numeric keys, type the number of the Event that you wish to modify. You may also use the cursor arrow keys to scroll through the list of Events. The following screen shows that you selected Event 1.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	11.0	YES		READY 999
EVNT		TIME	TE1	TE2
		STOR		RDY

4. If you want to create Event 2, press the **insert** key on the front panel of the pump. A duplicate line of Event 1 is inserted below Event 1 and the cursor is over the time value.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
1	11.0	YES		READY
> 2	11.0	YES		999
EVNT		TIME	TE1	TE2
		STOR		RDY

5. Enter a time value and press the **enter** key to set that value. Then press the soft key or press the arrow keys to select the next entry that you wish to modify.
6. When you have finished making changes to the timed events, you can store the method by pressing **STOR**. The following prompt appears on the bottom line of the Timed Events Screen.

METHOD00		SHTDN		0 PSI
EVENT	TIME	T.E.1	T.E.2	
> 1	11.0			READY 999
STORE METHOD 01				
EVNT		TIME	TE1	TE2
		STOR		RDY

To store a method, refer to the procedure under **"Storing a Method"** in this chapter.

3 Setting the Pressure Limits

The following procedure describes how to set the operating pressure limits. You set a minimum pressure value to detect leaks in the system. If the pressure goes below the minimum pressure value, the pump shuts down. You set a maximum pressure value to protect your column. If the pressure goes above the maximum pressure value, the pump shuts down.

METHOD00	STORD	SHTDN						0 PSI
UTM								
STEP	TIME	FLOW	%A	%B	%C	%D	CURV	
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0	
ELAPSED TIME								*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT	

1. Press **PRES** from the Main Quaternary Screen. The following screen appears.

METHOD00	SHTDN						0 PSI
MINIMUM PRESSURE 0 PSI							
MAXIMUM PRESSURE 6100 PSI							
* IMPORTANT - CONSULT COLUMN INFO SHEET *							
ELAPSED TIME							*0.0
		MIN	MAX		STOR		

2. Press **MIN** to highlight the minimum pressure limit value. Type the minimum pressure limit value and press the **enter** key. If the operating pressure falls below this value, the pump stops. The following screen shows the minimum pressure limit value set to 0 psi.

Note The pressure units are set from the Instrument Set-Up screen under the Configure menu.

METHOD00	SHTDN						0 PSI
MINIMUM PRESSURE 0 PSI							
MAXIMUM PRESSURE 6100 PSI							
* IMPORTANT - CONSULT COLUMN INFO SHEET *							
ELAPSED TIME							*0.0
		MIN	MAX		STOR		

3. Press **MAX** to highlight the maximum pressure limit value. Type the maximum pressure limit value and press the **enter** key. If the pumping pressure exceeds this value, the pump stops. The following screen shows the maximum pressure limit value set to 6100 psi.

METHOD00	SHTDN	0 PSI
MINIMUM PRESSURE	0 PSI	
MAXIMUM PRESSURE	6100 PSI	
* IMPORTANT - CONSULT COLUMN INFO SHEET *		
ELAPSED TIME		*0.0
MIN	MAX	STOR

4. Press the **return** key to save all entries and return to the Main Quaternary Screen or press **STOR** to store this method. If you press **STOR**, the following prompt appears on the bottom of the screen.

METHOD00	SHTDN	0 PSI
MINIMUM PRESSURE	0 PSI	
MAXIMUM PRESSURE	6100 PSI	
* IMPORTANT - CONSULT COLUMN INFO SHEET *		
STORE METHOD	01	
MIN	MAX	STOR

To store a method, refer to the procedure under "Storing a Method" in this chapter.

Modifying a Method

You can modify any data field parameter in the method screen at any time - even while that parameter is currently running. This provides you with total control and flexibility over your method and analysis. The example in this section uses a gradient method. To modify the displayed method appearing in the Main Quaternary Screen, follow these steps.

METHOD00	STORD	SHTDN					0 PSI
UTM							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **PUMP** from the Main Quaternary Screen. The data field for the first parameter, Time, is highlighted.

METHOD00			SHTDN				0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
> 0	10.0	2.00	10.0	0.0	0.0	100	
1	5.0	2.00	10.0	0.0	0.0	90.0	
ELAPSED TIME							*0.0
STEP		NEW			STOR		

The carrot (>) indicates the current STEP.

2. Press the arrow keys to move the cursor to the value that you wish to modify. If you know which STEP you want to modify, press **STEP**.

METHOD00			SHTDN				0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
> 0	10.0	2.00	10.0	0.0	0.0	100	
1	5.0	2.00	10.0	0.0	0.0	90.0	
ENTER STEP NUMBER							0
STEP		NEW			STOR		

3. Type the step number, and press the **enter** key to display that step. In the following example step 3 is selected.

METHOD00			SHTDN				0 PSI
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
2	5.0	2.00	70.0	0.0	0.0	30.0	1.0
> 3	3.0	2.00	10.0	0.0	0.0	90.0	1.0
ELAPSED TIME							*0.0
STEP		NEW			STOR		

If you want to add another STEP as a duplicate of the current STEP, press the **insert** key.

If you want to delete the current STEP, press the **delete** key.

- After modifying the displayed parameters, press the **return** key to save all changes and return to the top level Main Quaternary Screen as shown here.

METHOD00		CHNGD	SHTDN				0 PSI
UTM							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

Notice that CHNGD is displayed on the top line to indicate that the method has been changed.

Storing a Method

To store the current method, follow these steps. The example in this section uses a gradient method.

METHOD00	STORD	SHTDN					0 PSI
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **DIR** from the Main Quaternary Screen. The following screen appears with a prompt in the last line.

METHOD00	STORD	SHTDN	
METHOD	NAME		LAST MODIFIED
> 2	UTM		02-FEB-1994 12:00
STORE (STOR) OR RECALL (RCL) A METHOD ?			
		RCL	STOR

2. Press **STOR**. The Store Method prompt appears in the last line of the screen with the first available number highlighted.

METHOD00	STORD	SHTDN	
METHOD	NAME		LAST MODIFIED
> 2	UTM		02-FEB-1994 12:00
STORE METHOD 02			

3. Type a number (between 1 and 19) that you wish to assign to the method. Then press **enter** (in this example, we use method number 19).

Note: If the method number already exists, the following prompt appears in the last line of the screen:

OVERWRITE EXISTING METHOD? (Y/N) N

4. Respond to the prompt by pressing the **1** yes or **0** no key. Pressing the **1** yes key returns you to the Main Quaternary Screen (which displays the current method).

Pressing the **0** no key prompts you to name the method.

If the method number does not exist, the following prompt displays on the bottom line of the screen:

NAME METHOD? (Y/N) N

5. Respond to the prompt by pressing the **1** yes or **0** no key. Pressing the **0** no key displays the method with the method number that you just assigned. Pressing the **1** yes key prompts you to name the method using up to eight characters:

METHOD19	STORD	0 PSI
ABCDEFGHIJ	USE CURSOR AND SEL TO	
KLMNOPQRST	SELECT, BKSP TO BACKSPACE	
UVWXYZ-	PRESS ENTER WHEN COMPLETE	
0123456789		
BKSP	SEL	

6. Press the arrow keys to highlight a letter or number, then press the **SEL** key to select and enter that choice. Press the **BKSP** key to erase a selected letter or number. The following screen shows that we named Method 19 UTM-ASSY.

METHOD19	STORD	0 PSI
ABCDEFGHIJ	USE CURSOR AND SEL TO	
KLMNOPQRST	SELECT, BKSP TO BACKSPACE	
UVWXYZ-	PRESS ENTER WHEN COMPLETE	
0123456789	UTM-ASSAY	
BKSP	SEL	

7. Press the **enter** key to accept the name. The Main Quaternary Screen reappears with the information displayed for the stored method.

METHOD19	STORD	SHTDN	0 PSI
UTM-ASSAY			
STEP	TIME	FLOW	%A %B %C %D CURV
0	10.0	2.00	0.0 0.0 0.0 100 0.0
ELAPSED TIME *0.0			
VIEW	PUMP	T.E.	PRES SEQ DIR CNFG STRT

Deleting a Method

To delete a stored method, follow these steps.

METHOD00	STORD	SHTDN					0 PSI
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **DIR** from the Main Quaternary Screen. The following screen appears.

METHOD00	STORD	SHTDN		
METHOD	NAME		LAST MODIFIED	
> 2	UTM		02-FEB-1994	12:00
19	UTM-ASSY		05-FEB-1994	07:52
STORE (STOR) OR RECALL (RCL) A METHOD ?				
		RCL	STOR	

2. Select the method you wish to delete by pressing the up and down arrow keys until the desired method appears to the right of the carrot (>).
3. Press the **delete** key. The following prompt appears in the last line of the screen:

DELETE METHOD? (Y/N)

Respond to the prompt by pressing the **1 (yes)** or **0 (no)** key. Pressing the **0 (no)** key redisplay the prompt:

STORE (STOR) OR RECALL (RCL) A METHOD ?

Pressing the **1 (yes)** deletes the method and redisplay the prompt:

STORE (STOR) OR RECALL (RCL) A METHOD ?

4. Press the **return** key to return to the Main Quaternary Screen which displays information about the current method.

Creating a Sequence

A Sequence is a series of stored methods that are linked together. The procedures in this section describe how to create a Sequence on the Series 200 LC Pump in a manner that allows you to use the injection information from the Sequence on the Series 200 LC Autosampler to ensure that the Pump Sequence produces the exact chromatographic conditions required to affect the separation of the samples in the Autosampler Sequence.

Rules for Creating a Sequence

1. If you have not already done so, you need to create and store in the Pump's Directory all of the methods that you intend to include in the Sequence.
2. Methods used in a Sequence must have a STEP 1 (or more steps) in the method.
3. The maximum number of methods comprising a sequence is 10.
4. If you have previously defined a sequence, you will be able to overwrite (but not delete) methods that you have stored and assigned to the sequence, as long as you have not linked the sequence. Once you link the sequence, you cannot overwrite or delete any of the methods in the sequence.
5. A Sequence can be linked at any time, even while the system is currently running a method.
6. You can only modify a Sequence when it is not linked.

Identifying the Methods to Include in a Sequence

Before defining the sequence, you need to view the methods stored in the Pump's Directory so that you can identify the numbers and names of the methods you want to include in the Sequence.

The example used in this section assumes you have a binary or quaternary pump model so the top level screen is the Main Quaternary Screen.

METHOD00	STORD	SHTDN					0 PSI
UTM							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0
							ELAPSED TIME
							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **DIR** from the Main Quaternary Screen and the Directory Screen appears.

DIRECTORY SCREEN

METHOD00	STORD	SHTDN	LAST MODIFIED	
METHOD	NAME			
2	UTM		02-FEB-1994	12:00
> 19	UTM-ASSY		05-FEB-1994	07:52
STORE (STOR) OR RECALL (RCL) A METHOD ?				
			RCL	STOR

- Press the up and down arrow keys to view the stored methods. Write down the method numbers and names that you intend to use in the Sequence.
- Press **return** to go back to the Main Quaternary Screen.

METHOD00	STORD	SHTDN						0 PSI
UTM								
STEP	TIME	FLOW	%A	%B	%C	%D	CURV	
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0	
ELAPSED TIME								*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT	

Defining the Sequence

Press **SEQ** from the Main Quaternary Screen to display the Sequence Screen:

SEQUENCE					
SET	METHOD	FIRST	LAST	INJ	
> 1	0	1	1	0	
SET	METH	1ST	LAST	INJ	LINK
					DELQ

Refer to Table 4-3 for a description of the Sequence Screen data field parameters.

Table 4-3. Description of the Sequence Screen Data Field Parameters.

Parameter	Description
SET	The identification of a line in the Sequence. You can have from 1 to 10 sets.
METHOD	The number of the stored method that controls the set. The method MUST be stored and it must contain one or more STEPS.
FIRST	Set this to 1 for every set in the Pump Sequence.
LAST	Set this to 1 for every set in the Pump Sequence.
INJ	Set this number equal to the number in the Injections data field for each set in the Autosampler Sequence. The Injections for each set in the Autosampler Sequence corresponds to the total number of injections performed for the method in that set on the Autosampler for samples and/or calibrants.
DELQ	Pressing this soft key deletes the entire sequence so that you can generate a new sequence.

1. Type the number of the stored method that you wish to use in this set and press **enter**. The cursor moves to the FIRST vial position. The following example shows that Pump Method 2 was selected:

SEQUENCE						
SET	METHOD	FIRST	LAST	INJ		
> 1	2	1	1	0		
SET	METH	1ST	LAST	INJ	LINK	DELQ

2. Set each first and last vial number for each set in the Pump Sequence to 1. After the First entry, press **enter**; then press **enter** again after the Last entry. The data field for Inj will then be highlighted as shown in the following Sequence Screen.

SEQUENCE						
SET	METHOD	FIRST	LAST	INJ		
> 1	2	1	1	0		
SET	METH	1ST	LAST	INJ	LINK	DELQ

3. For each set in the Pump Sequence, enter the number that appears in the Injection field of the Autosampler Sequence for the corresponding set.

The Injections field on the Autosampler Sequence represents the total number of injections performed for the method in that set.

The correspondence between the Pump Sequence and Autosampler Sequence screens is shown next.

PUMP SEQUENCE SCREEN

SEQUENCE						
SET	METHOD	FIRST	LAST	INJ		
> 1	2	1	1	0		
SET	METH	1ST	LAST	INJ	LINK	DELQ

AUTOSAMPLER SEQUENCE SCREEN

SEQUENCE			INJECTIONS
SET	METHOD		
> 1	3 UTM		6
2	5 UTM-ASSAY		3
ENTER METHOD NUMBER (0 - 19)			
SET		LINK	DELS

To create another SET, go to step 4. If you are finished at this point, go to step 5.

4. To create another set (SET 2) in the Pump Sequence, press **enter** or **insert**. A second set, identical to SET 1 (except for the Inj field which is zero), is added as shown next.

SEQUENCE						
SET	METHOD	FIRST	LAST	INJ		
1	2	1	1	6		
> 2	2	1	1	0		
SET	METH	1ST	LAST	INJ	LINK	DELQ

Edit each of the fields as described in steps 1 to 3 (remember to set First and Last to 1). Press **enter** or **insert** to add up to a total of 10 sets.

Note: if you want to remove any of the sets, press the **delete** key.

5. To link the Sequence to the method in SET 1, press **LINK**.

To complete the Sequence, press **return**. This saves the entries in the Sequence Screen and returns you to the Main Quaternary Screen which displays the information about the current method.

Printing a Method

To obtain a copy of the method, you can print the current method to an external printer.

1. Connect the printer cable to the RS-232 port on the rear of the Series 200 LC Pump.

METHOD00		STORD		SHTDN		0 PSI	
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

2. Press **CNFG** to display the Configure Screen.

```

CONFIGURE
INST  - INSTRUMENT SETUP
COMM  - COMMUNICATIONS
DIAG  - DIAGNOSTIC TESTS
CAL   - CALIBRATION
LOG   - INSTRUMENT LOG

```

3. Press **COMM** to display the Communication Screen.

COMMUNICATION
NONE
PRINTER
1020

Press **PRNT** to select PRINTER. Set your printer to the following settings:

Baud	Parity	Data Bits	Stop Bits
2400	None	8	1

4. Press **return** to display the Configure Screen.

```

CONFIGURE
INST  - INSTRUMENT SETUP
COMM  - COMMUNICATIONS
DIAG  - DIAGNOSTIC TESTS
CAL   - CALIBRATION
LOG   - INSTRUMENT LOG
INST COMM DIAG CAL LOG

```

5. Press **return** to display the Main Quaternary Screen.

```

METHOD01  STORD  SHTDN      Q01.10.00  0 PSI
UTM
STEP  TIME  FLOW  %A    %B    %C    %D    CURV
  0   10.0   2.00  70.0   0.0   0.0   30.0   0.0

                                ELAPSED TIME      *0.0
VIEW PUMP T.E. PRES SEQ DIR CNFG STRT

```

Select and display the method that you want to print.

6. Press **PUMP**. The Pump Screen appears.

```

METHOD00      SHTDN      0 PSI
STEP  TIME  FLOW  %A    %B    %C    %D    CURV
>0   10.0   2.00  0.0    0.0    0.0   100

                                ELAPSED TIME      *0.0
STEP NEW STOR PRNT

```

Notice that the **PRNT** key appears. This key only appears if you have selected PRINTER in the Communication Screen.

7. Press **PRNT** to print a copy of your displayed method.

Notes

Running Methods and Sequences

5

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Running Methods and Sequences 5

This chapter describes how to run Methods and Sequences on isocratic, binary and quaternary models of the Series 200 LC Pump for the analysis of the Universal Test Mix (P/N 0089-0893). It includes the following topics:

- Before You Begin (Operating Summary and Preparing Your Mobile Phase)
- Running the Isocratic Pump Models
- Running the Binary or Quaternary Pump Models
- Running a Sequence
- Shutting Down the Pump (Short-term and Long-term)

Before You Begin

Before you run the pump, read the information contained in this section.

Operating Overview

The Series 200 LC Pump delivers mobile phase at the flow rate and composition displayed on the top level screen, the Main Isocratic Screen (for isocratic pump models) or the Main Quaternary Screen (for binary and quaternary pump models).

These two main screens, discussed in detail in Chapter 4, are repeated here for your convenience.

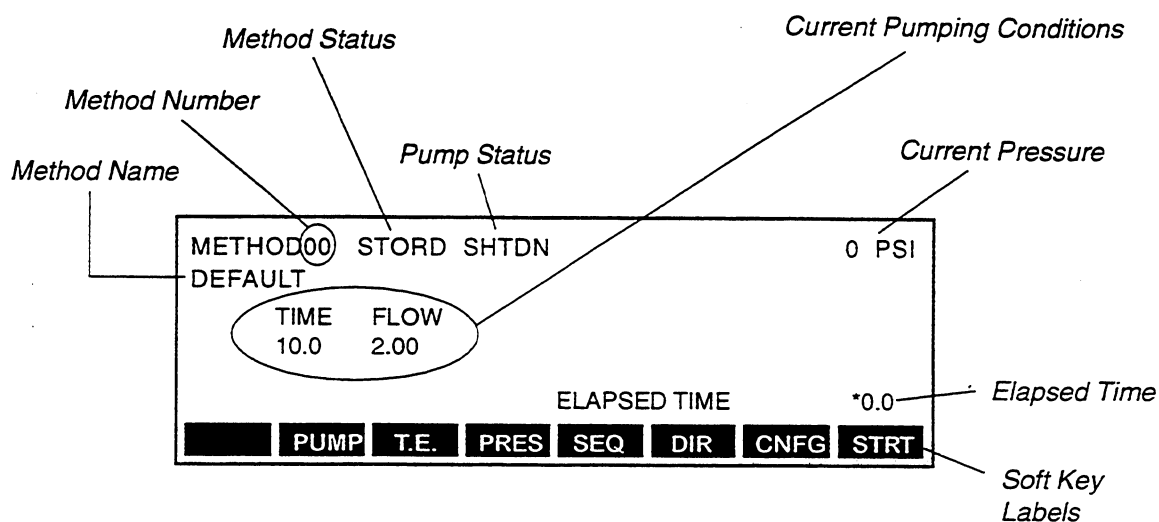


Figure 5-1. Main Isocratic Screen.

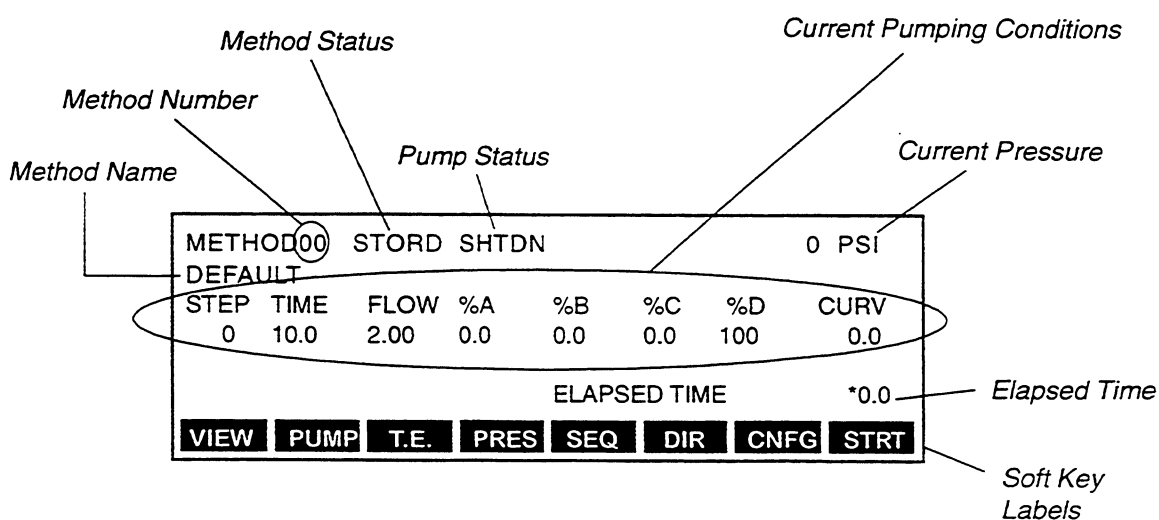


Figure 5-2. Main Quaternary Screen.

You start the pump by pressing the **STRT** soft key. The pump runs at the Step 0 conditions (binary or quaternary models). When the Step 0 time elapses, READY displays on the top line of the screen to indicate that the pump is ready to run the method. To start running a method, press the gray **run** key or to stop the pump, press the red **stop** key.

Operating Summary

This summary provides a brief overview of how to use the pump.

1. Select and fill the solvent reservoir with the proper degassed mobile phase for your analysis.
2. Switch on the Series 200 LC Pump. The power on/off switch is on the right side panel.
3. Properly configure the Series 200 LC Pump for your application as described in Chapter 3 of this manual.
 - Set the pressure units.
 - Set the type of communication (to a printer or integrator).
4. Create a method (or sequence) as described in Chapter 4 of this manual.
 - Ensure that all sections are complete (pump control, timed events, and pressure limits).
5. Verify that the tubing to your injector, column, and detector is properly connected. Also verify that the electrical connections to relays and timed events are properly connected for automated operations.
6. Prime the pump.
7. Display the method and start the pump to equilibrate the system.
8. Run the method (analysis).
9. Analyze the results.

Preparing Mobile Phase for Your Analysis

The following steps apply to all pump models unless otherwise noted. To prepare the mobile phase for your analysis:

1. Select the solvents for your analysis.

HPLC solvents are recommended to ensure purity for accurate separations. As an additional precaution, filter the solvents through a 0.5 μ porosity medium, such as the Kontes All-Glass Filter Apparatus (P/N N930-1063).

**Caution**

Even with an in-line filter, minute particulates can accumulate. This can increase the system pressure and change the analysis results.

2. Carefully pour the solvents into the solvent reservoirs.
3. Degas the solvents to remove dissolved air (which causes detection spikes or baseline drift). You can degas solvents by any of the following methods.
 - Bubble a fine stream of helium through the solvent.
Helium has the unique ability to dissolve other gases out of solvent without having itself absorbed in the solvent.
 - Place the solvent reservoir in an ultrasonic bath or insert an ultrasonic probe in the solvent.
 - Apply a vacuum to the solvent while mixing it with a magnetic mixer. (This is the least effective method.)

**Warning**

Use only heavy-walled flasks. Even a simple aspirator can produce sufficient vacuum to collapse an ordinary flask.

4. For isocratic models, insert the solvent inlet tubing into the reservoir. For binary or quaternary models:
 - Since solvent composition is defined as %A in the method, reservoir A should contain the strong solvent.
 - If you are using a single premixed solvent, put it in reservoir A. Then set Reservoir A to 100%.

Running the Isocratic Pump Models

The following procedure describes how to run the standard and biocompatible isocratic pumps. The isocratic pump does not have a proportioning valve; therefore, you will use a premixed mobile phase at the same concentration throughout the entire analysis. The procedures in this section apply to the following pumps:

Description	Part Number
Series 200 LC Pump Isocratic Version (50/60 Hz)	N291-0100
Series 200 BIO LC Pump Isocratic Version (50/60 Hz)	N291-0200

Getting the Pump Ready - Standard Model N291-0100

After selecting and preparing your mobile phase, you are ready to prepare the pump for your analysis.

Priming/Purging the Pump

Before starting an analysis, it's necessary to purge the pump of the residual mobile phase and fill it with the mobile phase for your analysis.

1. Press the **purge** key on the pump keyboard to display the following purge screen:

METHOD00		STORD		SHTDN		0 PSI	
FLOW							
0.00							
		FLOW					

2. Confirm that the solvent line is submerged in the mobile phase.
3. Refer to Figure 5-3. Open the access door on the front of the pump and connect the 30 mL syringe (P/N 0990-4849), that is supplied in the Start-Up Kit, to the fitting on the prime/purge valve.
4. Open the prime/purge valve by turning the knob one turn counterclockwise.
5. Press **FLOW** and type a flow rate of 5 mL/min. Press **enter** to enter the flow rate and start the pump.

6. Collect about 30 mL of mobile phase or until you stop seeing bubbles.
7. Stop the pump and close the prime/purge valve.

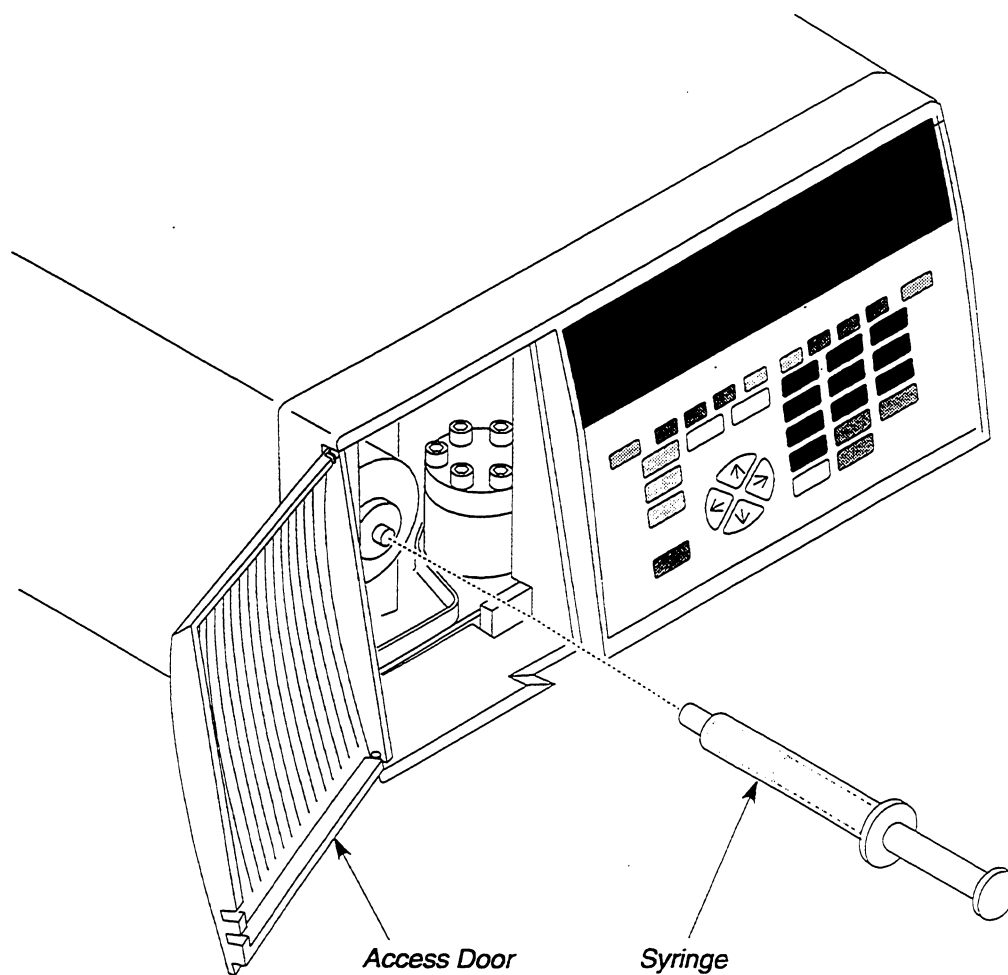


Figure 5-3. Connecting a syringe to the prime/purge valve.

Getting the Pump Ready - Biocompatible Model N291-0200 (Adding Piston Rinse Solvent)

If you have a Series 200 BIO LC Pump, you must also flush and fill the pump head with piston rinse solvent. The solvent must be compatible with your mobile phase.

1. Remove the following items from the Series 200 LC BIO Pump Start-Up Kit (P/N N291-0341).

Table 5-1. Piston Rinse Items.

Item	Part Number (P/N)
Yellow Tubing	0250-5799
Barbed Male Fitting	0990-3773
30 mL Syringe	0990-4849

2. Open the pump access door.
3. Insert the barbed end of the male fitting into one end of the yellow tubing.
4. Connect the barbed male fitting to one Luer fitting.
5. Insert the other end of the yellow tubing into a small beaker.
6. Select a rinse solvent that is compatible with your mobile phase (typically water).



Caution

Incompatible solvents may produce excessive salt precipitation that can cause damage to the pump.

7. Refer to Figure 5-4. Fill a 30 mL syringe with the rinse solvent and connect it to one Luer fitting on the prime/purge valve bracket.
8. Slowly inject the rinse solvent into the pump. The beaker should begin to fill with the rinse solvent.
9. When finished, remove the syringe and insert the plug into the Luer fitting.

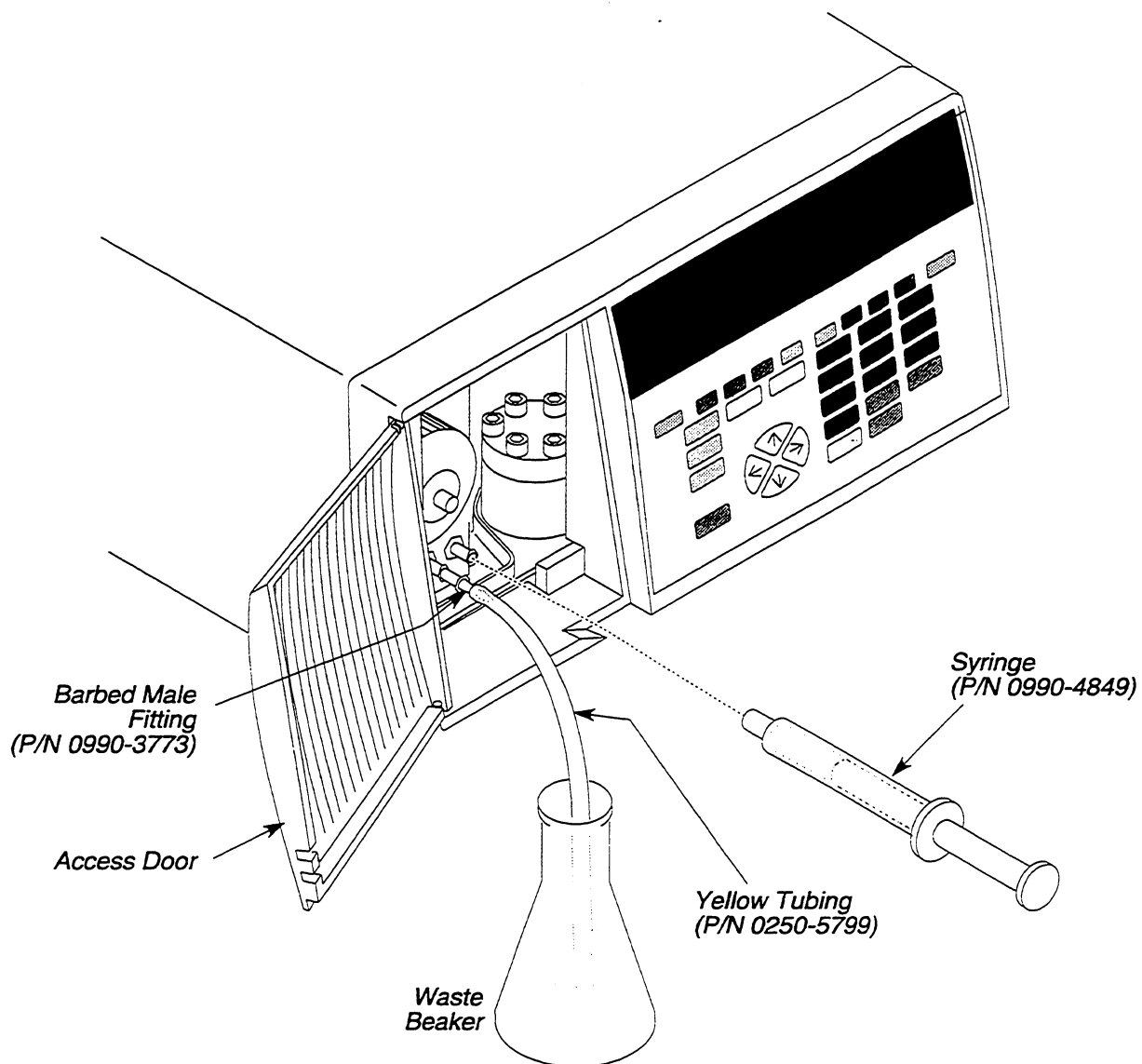


Figure 5-4. Adding piston rinse solvent to the pump head.

Running an Isocratic Method

The following procedure shows how to run an isocratic analysis of the Universal Test Mix (P/N 0089-0893) on pump models N291-0100 or N291-0200 using a PerkinElmer 3x3 C18 column. The mobile phase is typically 70% MeOH/Water.

1. Display the method you want to run as shown next in the Main Isocratic Screen.

METHOD00	STORD	SHTDN	2200 PSI	
DEFAULT				
TIME	FLOW			
10.0	2.00			
ELAPSED TIME				*0.5
PUMP	T.E.	PRES	SEQ	DIR
CNFG	STRT			

2. Press **STRT**. The pump starts running:

Note The pump allows one minute for the pressure to reach the minimum pressure value. If the minimum pressure is not reached within that time, the pump shuts down.

METHOD00	STORD	SHTDN	2200 PSI	
DEFAULT				
TIME	FLOW			
10.0	2.00			
ELAPSED TIME				*0.5
PUMP	T.E.	PRES	SEQ	DIR
CNFG				

3. Verify that the other instruments are ready.
4. Inject the Universal Test Mix (P/N 0089-0893) and verify that you obtain the chromatogram shown in Figure 5-5.

Chromatographic Conditions

Column: Pecosphere - 3 x 3C C18
(4.6 x 33mm)
Mobile Phase: 70/30 Methanol/Water
Injection Volume: 6 μ L
Flow Rate: 2.0 mL/min.
Sensitivity: 0.5 AUFS
Wavelength: 254 nm
Sample: Universal Test Mix
1. Sodium Chloride
2. Benzene
3. Toluene
4. Ethylbenzene
5. Isopropylbenzene
6. t-butylbenzene
7. Anthracene

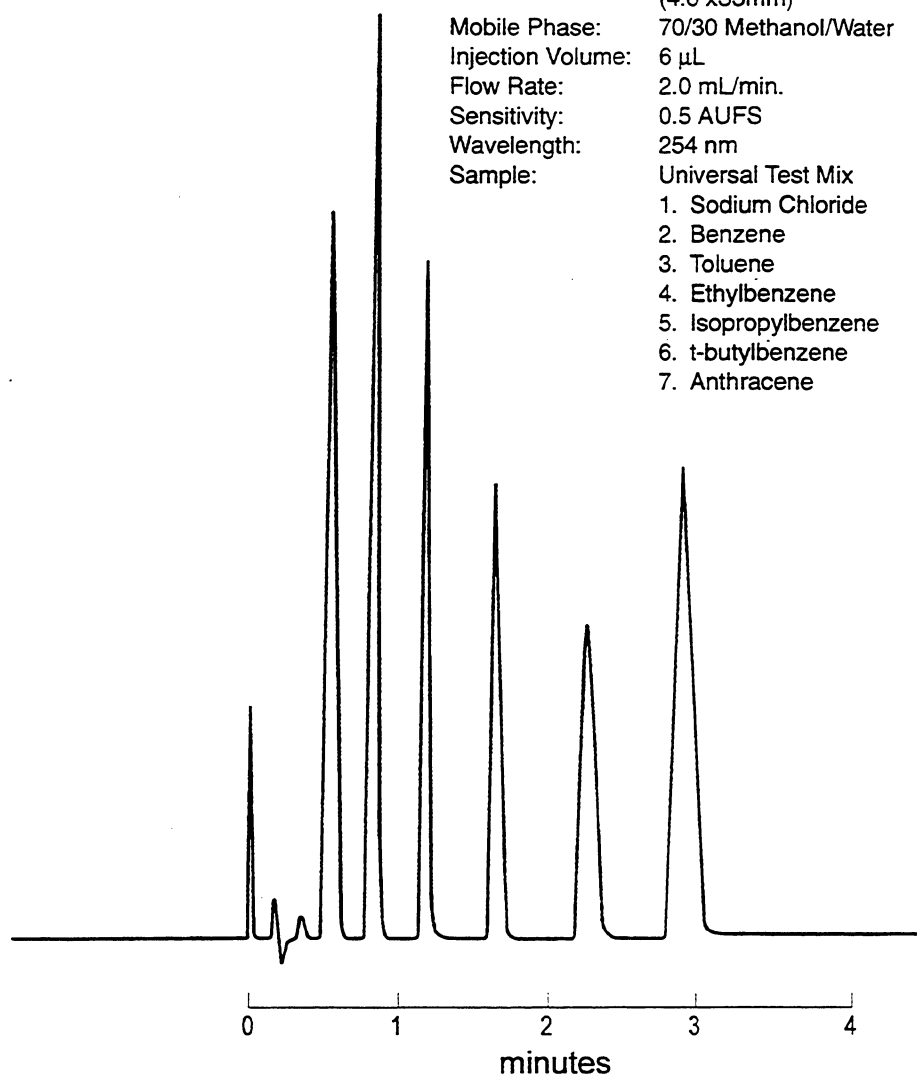


Figure 5-5. Chromatogram of the Universal Test Mix.

Running a Binary or Quaternary Pump Model (Standard and BIO Versions)

The following procedure describes how to run the standard and biocompatible binary and quaternary pumps. The procedures in this section apply to the following pumps:

Description	Part Number
200 LC Pump Binary Version (50/60 Hz)	N291-0101
Series 200 LC Pump Quaternary Version (50/60 Hz)	N291-0102
Series 200 BIO LC Pump Binary Version (50/60 Hz)	N291-0201
Series 200 BIO LC Pump Quaternary Version (50/60 Hz)	N291-0202

Getting the Pump Ready - Models N291-0101 and N291-0102

After selecting and preparing your mobile phase, you are ready to prepare the pump for your analysis.

Priming/Purging the Pump

Before starting an analysis, it's necessary to purge the pump of the residual mobile phase and fill the pump with the mobile phase for your analysis.

1. Press the **purge** key on the pump keyboard to display the following purge screen:

METHOD00	STORD	SHTDN					0 PSI
	FLOW	%A	%B	%C	%D		
	2.00	0.0	0.0	0.0	100		
	FLOW	A	B	C	D		

2. Confirm that the solvent lines are submerged in the mobile phase.
3. Refer to Figure 5-6. Open the access door on the front of the pump and connect the 30 mL syringe (P/N 0990-4849), that is supplied in the Start-Up Kit, to the fitting on the prime/purge valve.
4. Open the prime/purge valve by turning the knob one turn counterclockwise.

5. Press **FLOW** and type a flow rate of 5 mL/min. Press **enter** to enter the flow rate and start the pump.
6. Collect about 30 mL of mobile phase or until you stop seeing bubbles.
7. Stop the pump and close the prime/purge valve.

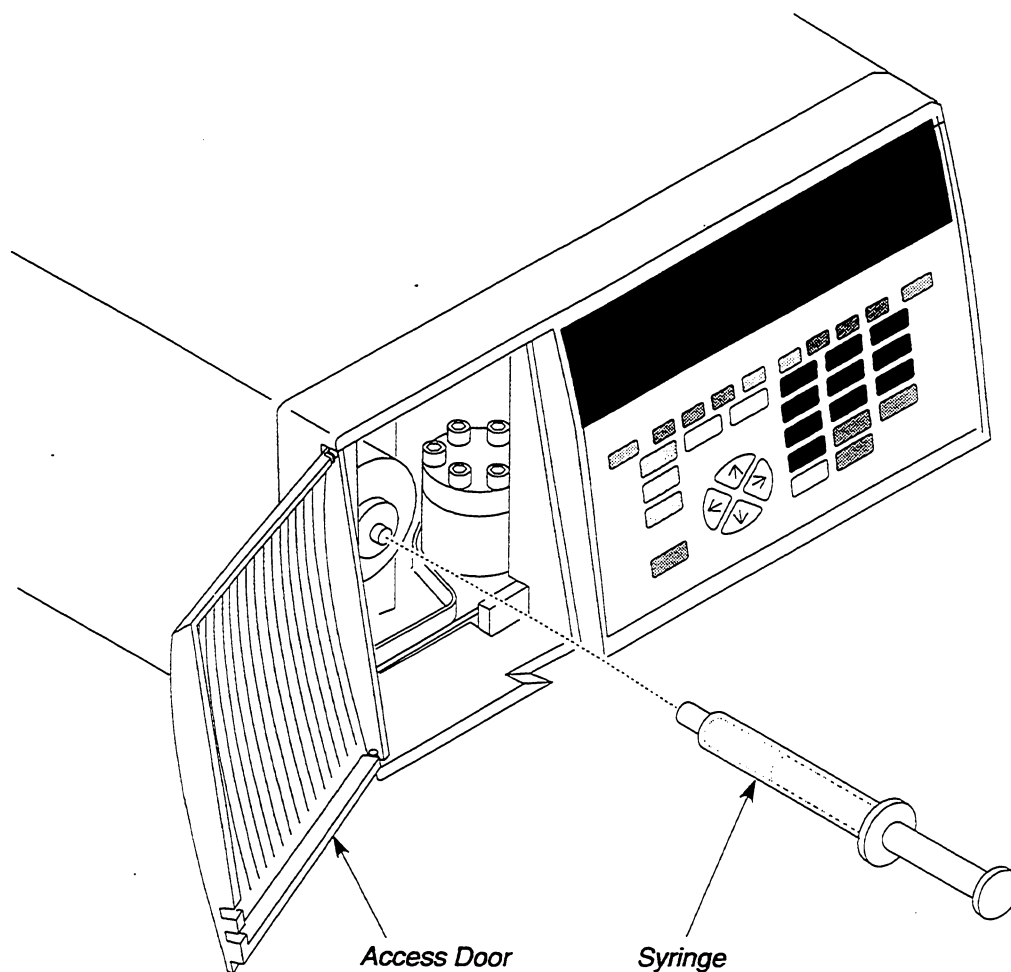


Figure 5-6. Connecting a syringe to the prime/purge valve.

Getting the Pump Ready - Biocompatible Models N291-0201 and N291-202 (Adding Piston Rinse Solvent)

If you have a Series 200 BIO LC Pump, you must also flush and fill the pump head with piston rinse solvent. The solvent must be compatible with your mobile phase.

1. Remove the following items from the Series 200 LC BIO Pump Start-Up Kit (P/N N291-0341).

Table 5-2. Piston Rinse Items.

Item	Part Number (P/N)
Yellow Tubing	0250-5799
Barbed Male Fitting	0990-3773
30 mL Syringe	0990-4849

2. Refer to Figure 5-7. Open the pump access door.
3. Insert the barbed end of the male fitting into one end of the yellow tubing.
4. Connect the barbed male fitting to one Luer fitting.
5. Insert the other end of the yellow tubing into a small beaker.
6. Select a rinse solvent that is compatible with your mobile phase (typically water).



Caution

Incompatible solvents may produce excessive salt precipitation that can cause damage to the pump.

7. Fill a 30 mL syringe with the rinse solvent and connect it to one Luer fitting on the prime/purge valve bracket (Figure 5-7).

8. Slowly inject the rinse solvent into the pump. The beaker should begin to fill with the rinse solvent.
9. When finished, remove the syringe and insert the plug into the Luer fitting.

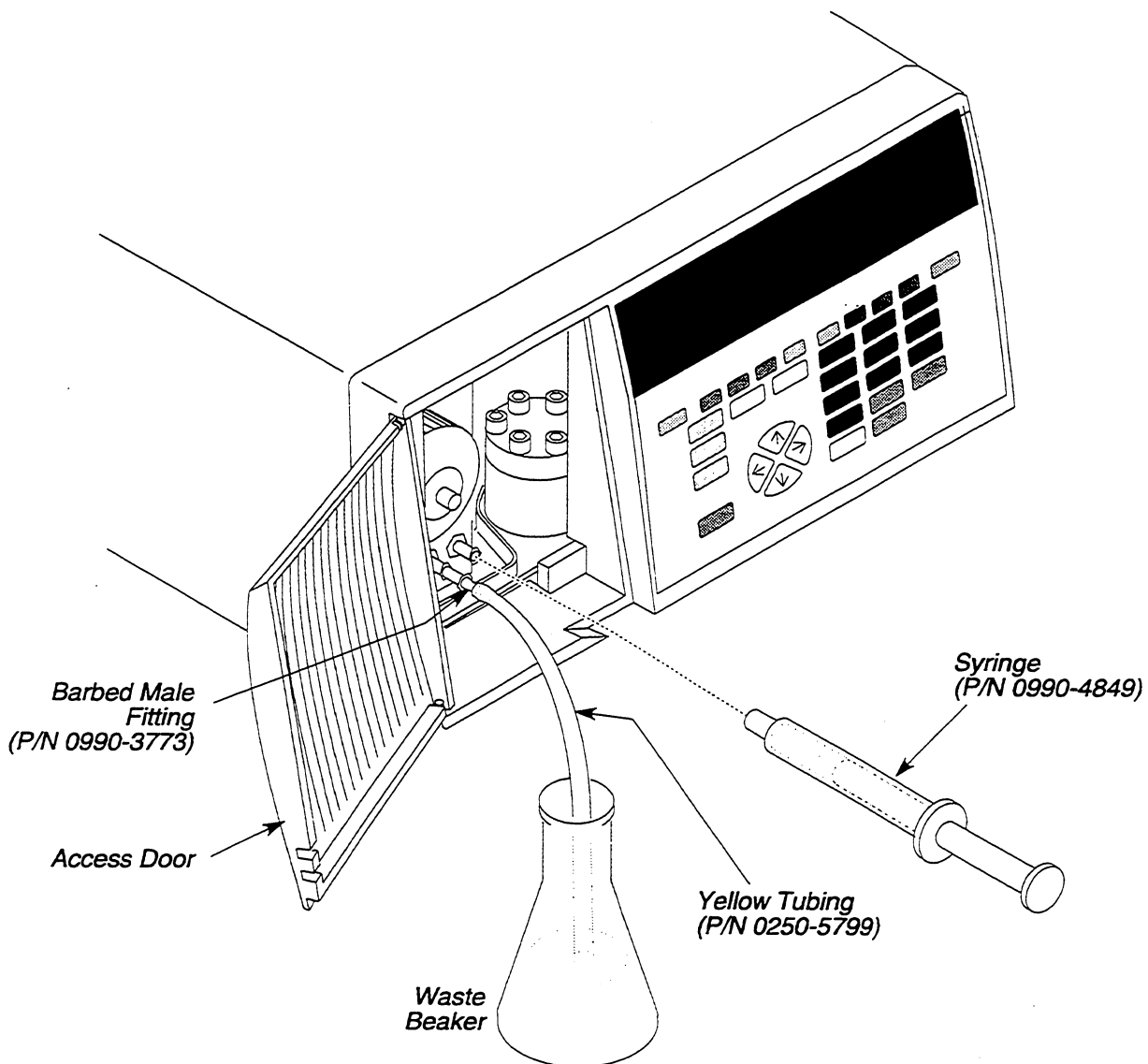


Figure 5-7. Adding piston rinse solvent to the pump head.

Running an Isocratic Method on the Binary or Quaternary Pumps

The following procedure shows how to run an isocratic analysis of the Universal Test Mix (P/N 0089-0893) on pump models N291-0101 and N291-0102 using a PerkinElmer 3x3 C18 column. The mobile phase is methanol (MeOH) and water.

1. Display the method that you want to run as shown next in the Main Quaternary Screen.

METHOD01	STORD	SHTDN						0 PSI
UTM								
STEP	TIME	FLOW	%A	%B	%C	%D	CURV	
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0	
ELAPSED TIME								*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT	

2. Verify that the other instruments are ready.
3. Press **STRT**. The pump starts running:

METHOD01	STORD	EQUIL						2200 PSI
UTM								
STEP	TIME	FLOW	%A	%B	%C	%D	CURV	
0	1.8	2.00	70.0	0.0	0.0	30.0	0.0	
ELAPSED TIME								*1.8
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG		

The pump allows one minute to achieve the minimum pressure. If the minimum pressure is not achieved within that time, the pump shuts down. After the equilibration time (the STEP 0 time) elapses, READY appears on the top line of the screen:

METHOD01	STORD	READY						2200 PSI
UTM								
STEP	TIME	FLOW	%A	%B	%C	%D	CURV	
0	1.8	2.00	70.0	0.0	0.0	30.0	0.0	
ELAPSED TIME								*11.8
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG		

4. Verify that you have a stable chromatographic baseline. Then set the chromatographic baseline on your data collection device to zero.

5. Simultaneously, press and inject the Universal Test Mix (P/N 0089-0893).
6. Verify that you obtain the chromatogram shown in Figure 5-8.

Chromatographic Conditions

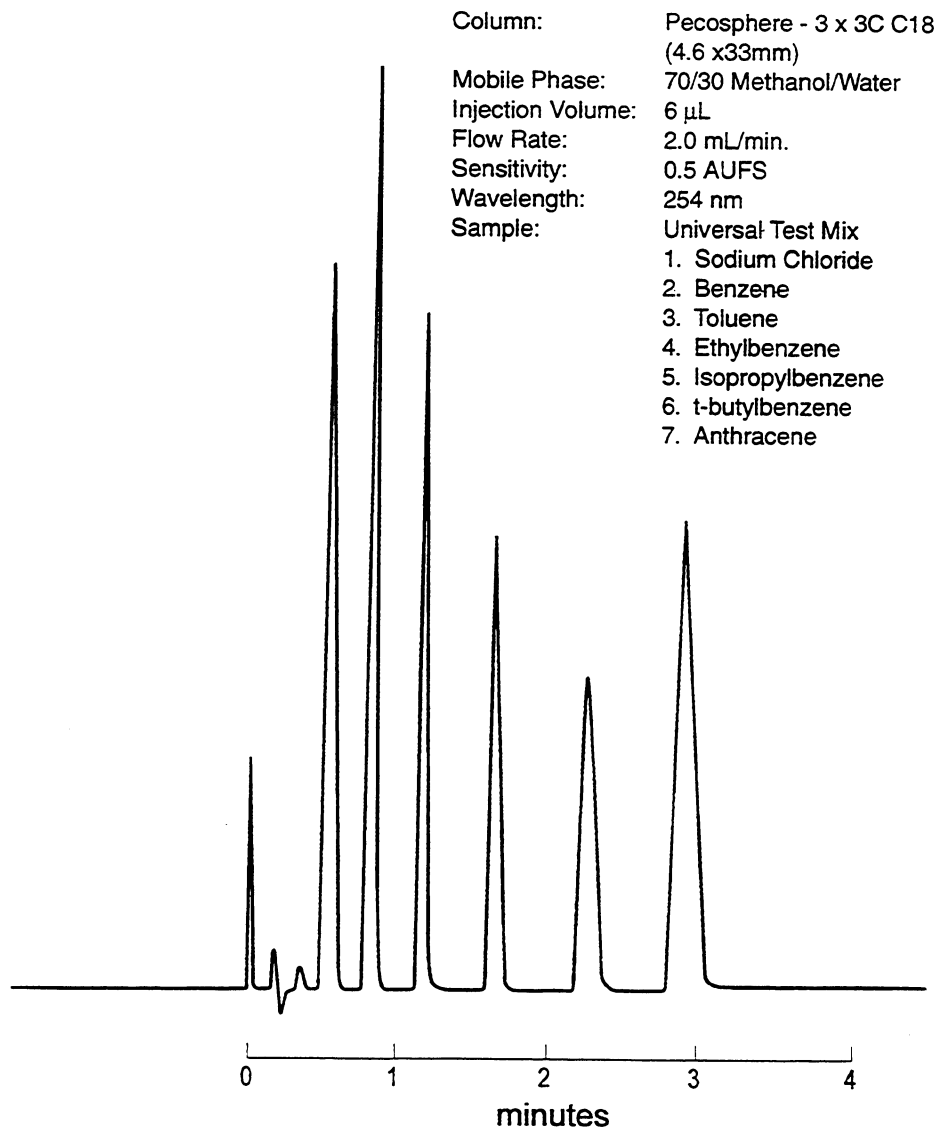


Figure 5-8. Chromatogram of the Universal Test Mix.

Running a Gradient Method

The following procedure shows how to run a gradient analysis of the Universal Test Mix (P/N 0089-0893) on pump models N291-0201 and N291-0202 using a PerkinElmer 3x3 C18 column. The mobile phase uses varying concentrations of methanol (MeOH) and water.

1. Display the method that you want to run as shown next in the Main Quaternary Screen. For example, the following method was named UTM-GRAD and is stored as METHOD 05.

METHOD05	STORD	SHTDN					0 PSI
UTM-GRAD							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	10.0	0.0	0.0	90.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

2. Verify that the other instruments are ready.

3. Press **STRT**. The pump starts running.

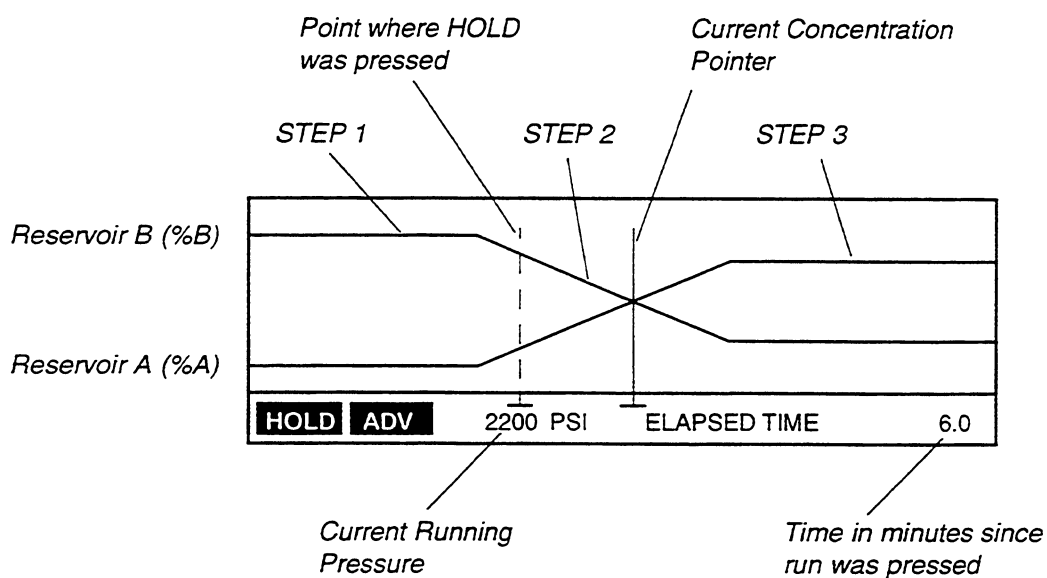
The pump allows one minute for the pressure to reach the minimum pressure value. If the minimum pressure is not reached within that time, the pump shuts down. The following screen shows that the method has been running for 1.8 minutes in Step 0.

METHOD05	STORD	EQUIL					2200 PSI
UTM-GRAD							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	1.8	2.00	10.0	0.0	0.0	90.0	0.0
ELAPSED TIME							*1.8
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	

After the equilibration time (the STEP 0 time) elapses, READY appears on the top line of the screen as shown next.

METHOD05	STORD	READY					2200 PSI
UTM-GRAD							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	11.8	2.00	10.0	0.0	0.0	90.0	0.0
ELAPSED TIME							*11.8
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	

- Verify that you have a stable chromatographic baseline. Then set the chromatographic baseline on your data collection device to zero.
- Simultaneously, press **run** and inject the Universal Test Mix (P/N 0089-0893).
- Press **VIEW** and observe the view screen. The view screen graphically represents the solvent profile in the running method. The following screen is an example of a typical view screen.

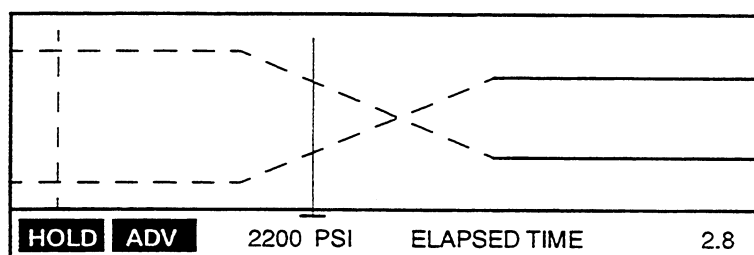


- Press **return** to display the current method screen.

METHOD05	STORD	RUN01					2200 PSI
UTM-GRAD							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
1	2.8	2.00	10.0	0.0	0.0	90.0	0.0
ELAPSED TIME							*2.8
VIEW	PUMP	T.E.	PRES	SEQ		CNFG	ADV

The above screen shows us that we are running STEP1. Notice that the TIME value and ELAPSED TIME value are identical. During a running method, you can:

- Modify solvent composition parameters that have not yet run and the VIEW screen will change to reflect these changes.
- Modify solvent composition parameters in the currently running step. The VIEW screen displays a dotted line reflecting that the step has been changed.
- Press **HOLD** to hold the method at the current conditions. When you press **CONT**, the method continues from the spot where HOLD was pressed. It is represented by a dotted line on the VIEW screen.
- Press **ADV** to advance the method to the next step. A dotted vertical line appears to mark the spot where you pressed **ADV**, and the current step and following step appear as dotted lines in the VIEW screen. The following VIEW screen shows that **ADV** was pressed while running STEP 1.

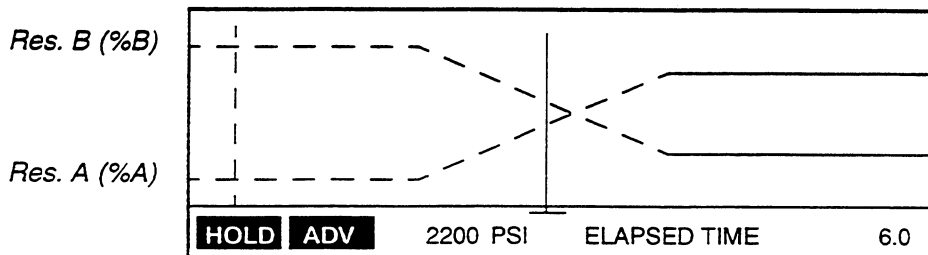


8. Press **return** to display the current method.

METHOD05	STORD	RUN02	2200 PSI				
UTM-GRAD							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
2	1.0	2.00	22.7	0.0	0.0	77.3	1.0
ELAPSED TIME							*6.0
VIEW	PUMP	T.E.	PRES	SEQ		CNFG	ADV

The method is now running STEP 2 (RUN02) and is 6 minutes into the run as shown by the ELAPSED TIME. Since the change from STEP 2 to STEP 3 is a liner gradient over 5 minutes, observe that the %A and %D values are constantly changing.

9. Press **VIEW** to display the current picture of the changing solvent composition.



Notice that the pointer shows that Reservoir A is increasing as Reservoir B is decreasing.

10. Upon completion of the method, verify that you obtain the chromatogram shown in Figure 5-9.

Chromatographic Conditions

Column: Pecosphere - 3 x 3C C18
(4.6 x33mm)

Mobile Phase: 5 min. Linear Gradient
10/90 Methanol/Water to
70/30 Methanol/Water
Hold 5 min.

Injection Volume: 6 μ L

Flow Rate: 2.0 mL/min.

Sensitivity: 0.5 AUFS

Wavelength: 254 nm

Sample: Universal Test Mix
1. Sodium Chloride
2. Benzene
3. Toluene
4. Ethylbenzene
5. Isopropylbenzene
6. t-butylbenzene
7. Anthracene

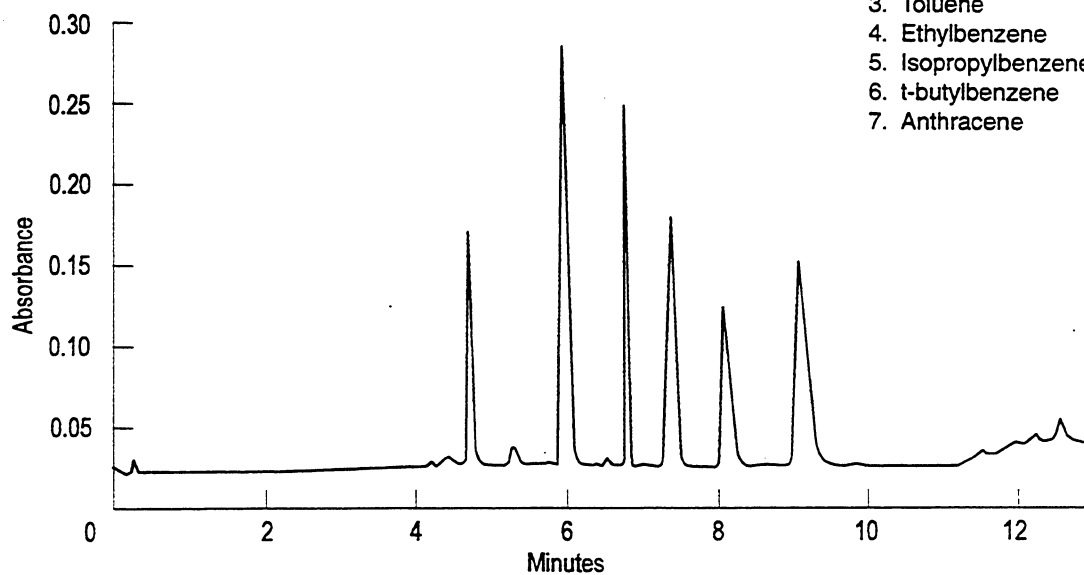


Figure 5-9. Gradient Chromatogram of the Universal Test Mix.

Running a Sequence

A sequence is a series of stored methods that are linked together. This section describes how to run a Sequence that has been created according to the directions contained in Chapter 4 of this manual.

Overview of a Sequence

The following is a summary of the main operating features of a Sequence.

- A Sequence is started by pressing **LINK** or stopped by pressing **BRK**.
- After stopping a Sequence with the **BRK** key, you can only restart the Sequence from the beginning.
- A Sequence can be interrupted by pressing **HOLD** and resumed by pressing **CONT**.

When **HOLD** is pressed, the current gradient run continues to completion, at which time all of the Sequence parameters hold at the current conditions. Now you can modify, recall, store, and even perform one or more runs with any method of your choice.

To continue the gradient and Sequence, press **CONT**. The Sequence method run continues to completion, then the Sequence parameters advance.

- Stopping the pump by pressing **stop** places the Sequence in hold. The pump assumes that the current run or method within the Sequence has completed.
- **reset** is disabled when a Sequence is linked except when the Sequence is in "Hold."
- When the Sequence ends, the pump stops.

Procedure for Running a Sequence

METHOD01	STORD	SHTDN					0 PSI
UTM							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Display the Sequence Screen by pressing **SEQ** from the Main Quaternary Screen.

SEQUENCE					
SET	METHOD	FIRST	LAST	INJ	
> 1	2	1	1	4	
SET	METH	1ST	LAST	INJ	LINK
					DELQ

2. Press **LINK** to link the Sequence with the method number in SET 1.

Observe that LINKD is displayed on the top line of the Sequence Screen.

SEQUENCE	LINKD	Q01.10.00		
SET	METHOD	FIRST	LAST	INJ
> 1	2	1	1	6

SET	METH	1ST	LAST	INJ	LINK		DELO
-----	------	-----	------	-----	------	--	------

Also note that Q01.10.00 is also displayed on the top line of the screen. This indicates the current status of the Sequence:

Q01	10	00
SET number	Vial number	Injection number

3. Refer to Table 5-3 for a summarized description of the function of the soft keys on the bottom of the Sequence Screen.

Table 5-3. Description of the Soft Keys when Running a Sequence.

Soft Key	Description
SET	Press this soft key to select a SET number and review the parameters.
BRK	Press this soft key to break a running Sequence at any time. Once a Sequence is broken (stopped), you cannot continue from the point of the break. You must restart the Sequence from the beginning.
HOLD	Press this soft key to hold a running Sequence at the current position to review and modify methods. HLD replaces LINKD and the sequence status is removed from the top line of the screen. You can continue from the point of the HOLD by pressing CONT .
CONT	Press this soft key to continue running the Sequence from the point of the HOLD.

4. Press **return** to redisplay Main Quaternary Screen which displays the current method.

METHOD01	STORD	SHTDN	Q01.10.00				0 PSI
UTM							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	70.0	0.0	0.0	30.0	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

5. To start the Sequence, press **STRT**.

If you want to hold or break a running sequence, press **SEQ** then press either **HOLD** or **BRK**.

SEQUENCE	LINKD	Q01.10.00
SET	METHOD	FIRST LAST INJ
> 1	2	1 1 4
SET		BRK HOLD

6. Press **return** to redisplay the Main Quaternary Screen.

Shutting Down the Pump

There are two types of shutdown procedures: long-term and short-term.

Short-Term Shutdown (Overnight and Weekends)

Observe all precautions pertaining to hazardous solvents, and/or those solvents which form harmful deposits or by products.

1. Remove harmful mobile phases from the pump and system.

Flush the column according to the instructions supplied with the column.

Flush buffer salts from the system with water. Evaporation leaves salt crystals which may form harmful deposits.



Caution

Damage caused by precipitating buffer salts in capillary tubing, or damage resulting from this condition, is specifically excluded from warranty.

Remove chloroform or solvents which can decompose to form hydrochloric acid from the system.

2. After removing harmful mobile phases, prepare the pump for most mobile phases by flushing it with isopropanol.



Caution

To avoid contaminating the pump, refilter or discard solvents (including water) that were exposed to the environment for more than 24 hours before use.

3. For weekend storage we recommend flushing 60%/40% MeOH/water through the pump, column, and flowcell (provided your column is compatible with MeOH/water).
4. Switch off the pump, detector, and recorder.
5. If you are using a data acquisition device (e.g., Model 1020, 1022, or Turbochrom), refer to the shutdown instructions for that device. Turning off power to the data system may not be advantageous if Methods and Data files have not been permanently stored.

Long-Term Shutdown

1. Follow Short-Term Shutdown procedure Steps 1 through 4.
2. Remove the column, and flush the pump first with water and then with isopropanol. Ensure that the pump head is filled with isopropanol.
3. Switch off the detector power switch.
4. Disconnect the pump output tube from the system and cap it.
5. Remove the solvent inlet filter from the reservoirs. Place the filter in a small polypropylene bag to prevent evaporation.
6. Store the pump in a clean, dry location.
7. Before using the pump, completely purge the pump with the correct solvent for the column before reconnecting the column and restarting the system.

Calibration and Diagnostics

6

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Calibration and Diagnostics

6



Caution

Changing pump calibration parameters voids pumping accuracy. Therefore, unauthorized resetting of calibration parameters is not covered by PerkinElmer warranty service.

This chapter contains the procedures to view the pump calibrations and run the pump diagnostics. The only pump calibration that you are likely to change is the zero offset in the Pressure Transducer Calibration. The pump diagnostics provide a way for you to determine the cause of possible problems.

Calibrations

This section contains procedures for performing the following calibrations.

- Checking Pump Calibration
- Checking P-Count Performance
- Checking Pressure Transducer Calibration

Checking Pump Calibration



Caution

Changing pump calibration parameters voids pumping accuracy. Therefore, unauthorized resetting of calibration parameters is not covered by PerkinElmer warranty service.

The correct pump calibration parameters are on the label that is attached to the top of the electronics cover. To verify the correct pump calibration parameters:

METHOD00		STORD		SHTDN		0 PSI	
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **CNFG** from the Main Quaternary Screen to display the Configure Screen.

```

CONFIGURE
INST  — INSTRUMENT SETUP
COMM  — COMMUNICATIONS
DIAG  — DIAGNOSTIC TESTS
CAL   — CALIBRATION
LOG   — INSTRUMENT LOG

```

2. Press **CAL** to display the Pump Calibration Screen.

PUMP CALIBRATION - ENTRIES AND TESTS					
RAMP LENGTH 200	FLOW FACTOR 1100	COMPOSITION OR EXTENDED PUMP CALIBRATION			
RAMP	FLOW	TEST	COMP	EXTD	

3. Press **COMP** to view the Composition Calibration Screen.

COMPOSITION CALIBRATION					
REFILL STEPS	TOTAL	A	B	C	D
() MAJOR	1000	0	0	0	0
SCALE/1000	SCALE	1000	1000	1000	1000
HUN%	OFFSET	0	0	0	0
SCAL	OFFS	A	B	C	D

4. Press **return** to display the Pump Calibration Screen.

PUMP CALIBRATION - ENTRIES AND TESTS					
RAMP LENGTH 200	FLOW FACTOR 1100	COMPOSITION OR EXTENDED PUMP CALIBRATION			
RAMP	FLOW	TEST	COMP	EXTD	

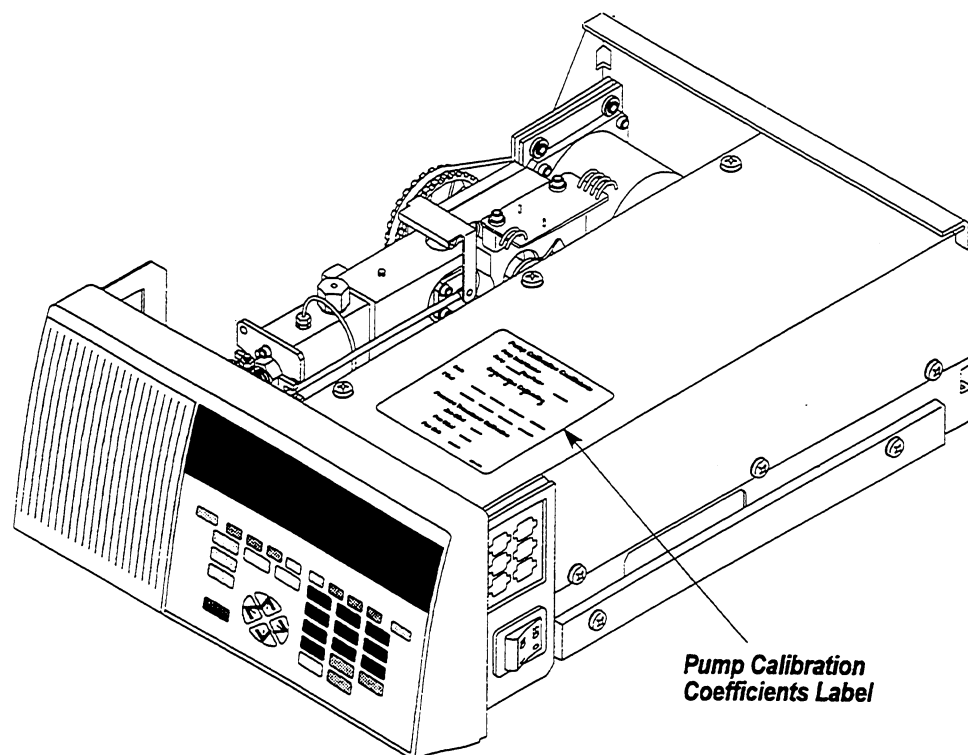


Figure 6-1. Location of the Pump Calibration Coefficients Label.

Pump Calibration Coefficients			
Pump Serial Number _____			
Ramp _____		Flow Factor _____	
Composition Calibration			
A	B	C	D
Scale	_____	_____	_____
Offset	_____	_____	_____
Pressure Transducer Calibration			
Zero Offset		_____	
Pres Gain		_____	
N291-1070			

Figure 6-2. The Pump Calibration Coefficients Label.

5. Press **EXTD** to view the Extended Pump Calibration Screen.

EXTENDED PUMP CALIBRATION - ENTRIES					
EFFECTIVE UNTIL POWER OFF					
SEGMENTS	1	2	3	4	5
ACCL	50000	250000		320000	55000
CNTS	15			199	205
RATE			5500		
	ACCL	CNTS	CNTS		

6. Press the **return** key three times to redisplay the Main Quaternary Screen.

METHOD00	STORD	SHTDN					0 PSI
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

Checking the P-Count Performance

Stability of the P-Count RANGE, while pumping, is indicative of consistent pumping performance. For example, a high RANGE value may suggest that there are bubbles in the pump head.

Note To run the P-Count Performance test you need a minimum pressure of 200 psi.

To check the P-Count Performance:

METHOD00	STORD	SHTDN					0 PSI
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **STRT** from the Main Quaternary Screen to start the pump.
2. Press **CNFG** from the Main Quaternary Screen to display the Configure Screen.

```

CONFIGURE
INST  - INSTRUMENT SETUP
COMM  - COMMUNICATIONS
DIAG  - DIAGNOSTIC TESTS
CAL   - CALIBRATION
LOG   - INSTRUMENT LOG

```

3. Press **CAL** to display the Pump Calibration Screen.

PUMP CALIBRATION - ENTRIES AND TESTS							
RAMP LENGTH 200	FLOW FACTOR 1100		COMPOSITION OR EXTENDED PUMP CALIBRATION				
RAMP		FLOW		TEST		COMP	EXTD

4. Press **TEST** to display the Calibration Tests Screen.

CALIBRATION TESTS

PCNT — P-COUNT PERFORMANCE

PCAL — PRESSURE TRANSDUCER CALIBRATION

RCAL — RAMP LENGTH CALIBRATION

PCNT

PCAL

5. Press **PCNT** to display the P-Count Performance Screen.

P-COUNT PERFORMANCE

AVE(10) P-COUNT = 1278

RANGE = 8

The RANGE should be less than 10.

Checking the Pressure Transducer Calibration

To check the pressure transducer calibration:

1. Open the prime/purge valve to drop the pressure to 0 psi.

METHOD00	STORD	SHTDN					0 PSI
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

2. Press **CNFG** from the Main Quaternary Screen to display the Configure Screen.

CONFIGURE	
INST	— INSTRUMENT SETUP
COMM	— COMMUNICATIONS
DIAG	— DIAGNOSTIC TESTS
CAL	— CALIBRATION
LOG	— INSTRUMENT LOG
INST	COMM
DIAG	CAL
LOG	

3. Press **CAL** to display the Pump Calibration Screen.

PUMP CALIBRATION - ENTRIES AND TESTS		
RAMP	FLOW	COMPOSITION OR
LENGTH	FACTOR	EXTENDED PUMP
200	1100	CALIBRATION
RAMP	FLOW	TEST
	COMP	EXTD

4. Press **TEST** to display the Calibration Tests Screen.

CALIBRATION TESTS							
PCNT - P-COUNT PERFORMANCE							
PCAL - PRESSURE TRANSDUCER CALIBRATION							
RCAL - RAMP LENGTH CALIBRATION							
PCNT						PCAL	RCAL

5. Press **PCAL** to display the Pressure Transducer Calibration Screen.

PRESSURE TRANSDUCER CALIBRATION							
ZERO OFFSET = 128							
PRES GAIN = 10000							
ELEVATED PRESSURE = 6000 PSI							
1 ML	10 ML	ZOFF	GAIN	POFF		ZERO	PRES

6. Press **ZERO** and wait until the ZERO OFFSET value is highlighted. This indicates that the test is complete. Then press

return to set the values and redisplay the Calibration Tests Screen,

or

quit to maintain the original values and redisplay the Calibration Tests Screen.

CALIBRATION TESTS							
PCNT - P-COUNT PERFORMANCE							
PCAL - PRESSURE TRANSDUCER CALIBRATION							
RCAL - RAMP LENGTH CALIBRATION							
PCNT						PCAL	RCAL

Diagnostics

The following diagnostics are used to check the condition of the Series 200 LC Pump electronic hardware.



Caution

Some tests will delete all methods, the Sequence, configuration, and calibration parameters.

To display the Diagnostics Screen:

METHOD00	STORD	SHTDN					0 PSI
DEFAULT							
STEP	TIME	FLOW	%A	%B	%C	%D	CURV
0	10.0	2.00	0.0	0.0	0.0	100	0.0
ELAPSED TIME							*0.0
VIEW	PUMP	T.E.	PRES	SEQ	DIR	CNFG	STRT

1. Press **CNFG** from the Main Quaternary Screen to display the Configure Screen.

CONFIGURE	
INST	— INSTRUMENT SETUP
COMM	— COMMUNICATIONS
DIAG	— DIAGNOSTIC TESTS
CAL	— CALIBRATION
LOG	— INSTRUMENT LOG
INST	COMM
DIAG	CAL
LOG	

2. Press **DIAG** to display the first screen:

KPD1	— KEYPAD DISPLAY TEST 1
KPD2	— KEYPAD DISPLAY TEST 2
PROM	— PROM TEST
RAM	— RAM TEST
BBUP	— BATTERY BACKUP TEST
COUNT=	01
KPD1	KPD2
PROM	RAM
BBUP	
	MORE

The COUNT value is the number of times that you want to run the test.

3. Press **MORE** to display the next diagnostics screen.

TPU	-	TPU TEST
DUA1	-	DUART TEST INTERNAL
DUA2	-	DUART TEST EXTERNAL
INP	-	REMOTE INPUTS TEST
COUNT= 01		
TPU	DUA1	DUA2
INP		
		MORE

4. Press **MORE** to display the next diagnostics screen.

OUT	-	OUTPUT RELAYS TEST
PART	-	READ PROM PART NUMBERS
CLRM	-	CLEAR ALL METHODS
CRAM	-	CLEAR BATTERY RAM
COUNT= 01		
OUT	PART	CLRM
CRAM		
		MORE

5. Press **MORE** to display the next diagnostics screen.

VALV	-	VALVE COIL CONTINUITY
COUNT= 01		
OUT	PART	CLRM
CRAM		
		MORE

6. Press **MORE** to redisplay the first diagnostics screen.

The following sections describe the function and use of each diagnostic.

Keypad Display Test 1

This test is designed to test the general functionality of the keypad. If a number is displayed next to pass, the keypad passed the test. If a number is displayed next to fail, call your PerkinElmer Service Engineer.

KPD1	—	KEYPAD DISPLAY TEST 1
KPD2	—	KEYPAD DISPLAY TEST 2
PROM	—	PROM TEST
RAM	—	RAM TEST
BBUP	—	BATTERY BACKUP TEST
COUNT	=	01
KPD1	KPD2	PROM
RAM	BBUP	
		MORE

1. Press **KPD1** to start the test.
2. A screen similar to the following appears.

KPD1	—	KEYPAD DISPLAY TEST 1
PASS = 01 FAIL = 00		

3. Press the **return** key to display the initial diagnostics screen.

Keypad Display Test 2

This test is designed to test the operation of each of the keypad keys. If a number is displayed next to pass, the keypad passed the test. If a key works, it is highlighted. If a key fails to highlight, call your PerkinElmer Service Engineer.

KPD1	—	KEYPAD DISPLAY TEST 1
KPD2	—	KEYPAD DISPLAY TEST 2
PROM	—	PROM TEST
RAM	—	RAM TEST
BBUP	—	BATTERY BACKUP TEST
COUNT=	01	
KPD1	KPD2	PROM RAM BBUP MORE

1. Press **KPD2** to start the test.
2. A screen similar to the following appears.

QUIT	F1	F2	F3	F4	F5	F6	F7	F8	F9	RET
RST		INS			DEL		7	8	9	
PRG							4	5	6	
RUN					^		1	2	3	
			<		>		0	.	-	
STOP					v		ENT	CE		

3. Press each key on the keyboard and verify that the corresponding label highlights on the screen. If a label fails to highlight, contact your PerkinElmer Service Engineer.
4. Press the **return** key to display the initial diagnostics screen.

PROM Test

This test is designed to perform a checksum on each of the PROMS. These checksums will be compared to the checksums stored in the first even/odd PROM pair. If a failure occurs, call your PerkinElmer Service Engineer.

KPD1	—	KEYPAD DISPLAY TEST 1					
KPD2	—	KEYPAD DISPLAY TEST 2					
PROM	—	PROM TEST					
RAM	—	RAM TEST					
BBUP	—	BATTERY BACKUP TEST					
COUNT	=	01					
KPD1	KPD2	PROM	RAM	BBUP			MORE

1. Press **PROM** to start the test. The following screen appears during the test.

PROM	—	PROM TEST					
PASS	=	01	FAIL	=	00		

2. If a failure occurs, a screen similar to the following appears.

PROM	—	PROM TEST					
U02	CHECKSUM IS	XXXX					
CORRECT	CHECKSUM IS	XXXX					
PASS	=	00	FAIL	=	01		

3. Press the **return** key to redisplay the initial diagnostics screen.

Ram Test

This test is designed to perform a series of tests by writing a number of patterns and then reading to verify that the pattern was correctly written. Each RAM is tested with three patterns. The first is modulo 256 of each byte address. The second is the hexadecimal value 55 and AA written into the same part of every other byte.

The system RAM is tested first, then the battery backup RAM is tested. The contents of the battery backup RAM are stored in the system during testing. The battery backup RAM will not be tested if the system RAM fails. If a failure occurs, call your PerkinElmer Service Engineer.

KPD1	—	KEYPAD DISPLAY TEST 1
KPD2	—	KEYPAD DISPLAY TEST 2
PROM	—	PROM TEST
RAM	—	RAM TEST
BBUP	—	BATTERY BACKUP TEST
COUNT=	01	
KPD1	KPD2	PROM
RAM	BBUP	
		MORE

1. Press **RAM** to start the test. If the test is successful, the following screen appears.

RAM	—	RAM TEST
PASS =	01	FAIL = 00

2. If a failure occurs, the following screen appears.

RAM	—	RAM TEST
RAM U25	FAILS TESTS AT ADDRESS XXXX	
DATA WRITTEN IS	XXXX	
DATA READ IS	XX	

3. Press the **return** key to redisplay the initial diagnostics screen.

Battery Backup Test

This test is designed to test the battery backup RAM. The contents of the battery backup RAM are stored in the system during testing. The battery backup RAM will not be tested if the system RAM fails. If a failure occurs, call your PerkinElmer Service Engineer.



Caution

This test will delete all methods, the Sequence, configuration and calibration parameters.

KPD1	—	KEYPAD DISPLAY TEST 1
KPD2	—	KEYPAD DISPLAY TEST 2
PROM	—	PROM TEST
RAM	—	RAM TEST
BBUP	—	BATTERY BACKUP TEST
COUNT=	01	
KPD1	KPD2	PROM
RAM	BBUP	
		MORE

1. Press **BBUP** to start the test. The following screen appears.

THIS TEST WILL DELETE ALL METHODS SEQUENCE, CONFIGURATION AND CALIBRATION PARAMETERS .. CONTINUE ?							
BBUP — BATTERY BACKUP TEST							
YES	NO						

2. Press **YES** to run the test, or press **NO** to redisplay page 1 of the Diagnostics Screen. If you run the test, the following screen appears.

TURN SERIES 200 OFF FOR AT LEAST 1 MIN. PRESS F8 SOFTKEY IMMEDIATELY AFTER POWER UP.							
BBUP — BATTERY BACKUP TEST							

3. Follow the instructions on the screen. If the test is successful, the following screen appears.

BBUP — BATTERY BACKUP TEST							
YES	NO						

If the test fails, the following screen appears. Call your PerkinElmer Service Engineer.

TEST PATTERN NOT DETECTED IN U306							
BBUP — BATTERY BACKUP TEST							

TPU Test

This test is designed to test the TPU. If a failure occurs, call your PerkinElmer Service Engineer.

TPU	—	TPA TEST
DUA1	—	DUART TEST INTERNAL
DUA2	—	DUART TEST EXTERNAL
INP	—	REMOTE INPUTS TEST
COUNT= 01		
TPU	DUA1	DUA2
INP		
		MORE

1. Press **TPA** to start the test. The following screen appears during the test.

TPU	—	TPU TEST
PASS = 01 FAIL = 00		

2. If the test fails, a screen similar to the following appears.

TPU	—	TPU TEST
PASS = 00 FAIL = 01		

3. Press the **return** key to redisplay the initial diagnostics screen.

DUART TEST Internal

This test is designed to test the internal DUART. The DUART is configured for the local loop-back mode. Data bytes 55 and AA are transmitted. The test verifies that bytes 55 and AA are received. If a failure occurs, call your PerkinElmer Service Engineer.

TPU	—	TPA TEST
DUA1	—	DUART TEST INTERNAL
DUA2	—	DUART TEST EXTERNAL
INP	—	REMOTE INPUTS TEST
COUNT=01		
TPU	DUA1	DUA2
INP		
		MORE

1. Press **DUA1** to start the test. The following screen appears during the test.

DUA1		—	DUART TEST INTERNAL				
PASS = 01		FAIL = 00					

2. If a failure occurs, a screen similar to the following appears.

DUA1		—	DUART TEST INTERNAL				
PASS = 00		FAIL = 01					

3. Press the **return** key to display the diagnostics screen.

DUART Test External

This test is designed to test the external DUART. If a failure occurs, call your PerkinElmer Service Engineer.

Note You cannot run this test without a special test fixture.

```

TPU      -   TPA TEST
DUA1     -   DUART TEST INTERNAL
DUA2     -   DUART TEST EXTERNAL
INP      -   REMOTE INPUTS TEST

COUNT= 01

```

TPU	DUA1	DUA2	INP				MORE
-----	------	------	-----	--	--	--	------

1. Press **DUA2** to start the test. The following screen appears.

PLACE TEST CONNECTOR ON J8 (SERIAL)
PRESS DUA2
DUA2 — DUART TEST EXTERNAL

2. Attach the test fixture and press **DE A2**.
3. If a failure occurs, a screen similar to the following appears.

DUA2 - DUART TEST EXTERNAL

U23 FAILURE

PASS = 00 FAIL = 01

--	--	--	--	--	--	--	--

4. Press the **return** key to display the diagnostics screen.

Remote Inputs Test

This test is designed to test the remote inputs. If a failure occurs, call your PerkinElmer Service Engineer.

```

TPU      -   TPA TEST
DUA1     -   DUART TEST INTERNAL
DUA2     -   DUART TEST EXTERNAL
INP      -   REMOTE INPUTS TEST

COUNT=01

```









1. Press **INP** to start the test. The following screen appears during the test.

FLAG	1
MP CONTACT	1
EXT RUN	0
EXT STOP	0

2ML

STOP

2. Press **2ML** to start the pump at 2 mL/min. The following screen appears.

FLAG	1	
MP CONTACT	1	
EXT RUN	0	
EXT STOP	0	
		
		
		
		2ML STOP

3. Press **STOP** to stop pumping.
4. Press the **return** key to display the diagnostics screen.

Output Relays Test

This test is designed to test the output relays. If a failure occurs, call your PerkinElmer Service Engineer.

OUT	-	OUTPUT RELAYS TEST
PART	-	READ PROM PART NUMBERS
CLRM	-	CLEAR ALL METHODS
CRAM	-	CLEAR BATTERY RAM
COUNT= 01		
OUT	PART	CLRM CRAM MORE

1. Press **OUT** to start the test. The following screen appears during the test.

READY	OFF
RUN OUT	OFF
TE1	OFF
TE2	OFF
NEXT TOGL	

2. Press the **TOGL** key to switch the relay on or off. Press **NEXT** to select the next relay.
3. Press the **return** key to display the diagnostics screen.

Read PROM Part Numbers

This screen displays the PROM part numbers.

OUT	-	OUTPUT RELAYS TEST
PART	-	READ PROM PART NUMBERS
CLRM	-	CLEAR ALL METHODS
CRAM	-	CLEAR BATTERY RAM
COUNT= 01		
OUT	PART	CLRM
CRAM		
		MORE

1. Press **PART** to display the part numbers. The following screen appears..

U2	N291 - XXXX

2. Press the **return** key to display the diagnostics screen.

Clear All Methods

This screen clears all stored methods.

OUT	-	OUTPUT RELAYS TEST					
PART	-	READ PROM PART NUMBERS					
CLRM	-	CLEAR ALL METHODS					
CRAM	-	CLEAR BATTERY RAM					
COUNT= 01							
OUT	PART	CLRM	CRAM				MORE

1. Press **CLRM** to start to clear all methods. The following screen appears.

CLRM - CLEAR ALL METHODS							
ARE YOU SURE?							
YES	NO						

2. Press **YES** to run the test or press **NO** to display the diagnostics screen.
3. Press the **return** key to display the diagnostics screen.

Clear Battery RAM

**Caution**

This test will delete all methods, the Sequence, configuration, and calibration parameters.

This diagnostic clears all battery backup RAM.

OUT	-	OUTPUT RELAYS TEST
PART	-	READ PROM PART NUMBERS
CLRM	-	CLEAR ALL METHODS
CRAM	-	CLEAR BATTERY RAM
COUNT= 01		
OUT	PART	CLRM
CRAM		
		MORE

1. Press **CRAM** to clear the battery backup RAM. The following screen appears.

DELETE ALL METHODS, SEQUENCE CONFIGURATION AND CALIBRATION PARAMETERS?							
CRAM - CLEAR BATTERY RAM							
YES	NO						

2. Press **YES** to run the test or press **NO** to display the diagnostics screen.
3. Press the **(return)** key to display the diagnostics screen.

Valve Coil Continuity Test

This test is designed to test the continuity of the solenoid valve.

VALV - VALVE COIL CONTINUITY							
COUNT= 01							
VALV							

1. Press **VALV** to start the test. The following screen appears.

VALV - VALVE COIL CONTINUITY							
A	YES						
B	YES						
C	YES						
D	YES						

If you have the quaternary pump, your screen should look like the above screen. "YES" displayed next to the valve designation indicates that the valve is connected and works.

If you have a binary pump, "YES" should be displayed next to two of the proportioning valve designations (A and B).

If you have an isocratic pump, you do not have a proportioning valve; therefore, "NO" is displayed next to all proportioning valve designations.

2. Press the **return** key to display the diagnostics screen.

Maintenance 7

- Guidelines for Maintaining the Pump .. 7-2
 - Viewing the Instrument Log .. 7-2
 - Before Calling Your Service Engineer .. 7-3
- Maintaining Pump Accessories .. 7-4
 - Replacing a Solvent Inlet Filter/Sparger .. 7-4
 - Replacing an In-line Solvent Filter Element .. 7-6
- Replacing a Fuse .. 7-8
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- Maintenance on Internal Pump Parts .. 7-11
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 - Replacing Piston Seals in the Stainless Steel Pump .. 7-20
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 - Inspecting and Cleaning Pump Pistons .. 7-29
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Maintenance 7

We describe how to perform routine and preventive maintenance procedures for the Series 200 LC pump in this chapter. We included the following information:

- Guidelines for maintaining the pump
- Maintaining pump accessories
- Replacing a fuse
- Maintaining internal pump parts

Important Information about Pump Seal Life when Using Specialty Mobile Phases

The Series 200 pump has been designed to handle a wide array of mobile phase chemistries. The standard high pressure seal installed in the pump (P/N 0990-7324) has been used for many years and has had an excellent record. You will, however, occasionally observe conditions where a seal life of less than 500 hours occurs.

Assuming that mobile phase chemistries have been filtered to 0.5 μ before use, the first way to try to increase seal life is to clean the piston with a mild abrasive (as described on page 7-29), and replace the seal and backup ring. This often solves the problem. If this has been tried and seal life is still less than 500 hours, the probability is that the mobile phase chemistry is causing the poor seal performance.

A number of mobile phase chemistries have been identified that can cause the problem. Chemistries with ammonium salts in combination with NH_4OH in high percentages of organic, and combinations involving these agents with chlorinated solvents are two examples. To aid in these situations, a new high pressure seal with a higher carbon fill has been found to work effectively when the seal-life problem occurs. This seal (P/N 0990-7345) and a corresponding backup ring are provided in your pump startup kit.

Guidelines for Maintaining the Pump

You can achieve long and trouble-free performance from your Series 200 LC Pump by performing the routine and preventive maintenance procedures described in this chapter.

Preventive maintenance ensures that your Series 200 LC Pump will perform consistently at an optimal level. To maintain the pump in its optimum operating condition, we recommend the following:

- Adhere to standard laboratory cleanliness practices.
- Use only high-purity solvents (preferably HPLC grade) for mobile phases. (Water should be bottled HPLC grade, or filtered and deionized.)
- Filter the solvents to avoid particulate contamination and tubing blockages.
- Use only high-purity gases when drying contact areas.
- Ensure that all new tubing is passivated and thoroughly flushed before making pump connections. (The tubing available from PerkinElmer is passivated.)
- Follow the short- and long-term shutdown procedures that are outlined in Chapter 5 in this manual.

In addition to preventive maintenance, you can perform routine maintenance on the pump, as needed, to keep your Series 200 LC Pump in proper working condition. Routine maintenance consists of replacing the normal wear items such as the high- and low-pressure piston seals, pump pistons, fuses, check valves, and columns when you observe a degradation in performance.

- Replace high- and low-pressure piston seals, backup rings, and if you have a biocompatible (BIO) pump, also replace the wash seals.
- Clean the check valves with isopropanol in an ultrasonic bath.

Viewing the Instrument Log

Periodically check the Instrument Log as described in section “Accessing the Instrument Log” in Chapter 3 of this manual. This lists the number of pump and seal cycles performed by the pump. You can use this information to help set up a maintenance schedule.

Note	Maintenance cycles for the pump piston seals and check valves are similar; therefore, to reduce downtime, check, clean or replace if necessary, the pump piston seals.
-------------	---

Before Calling Your Service Engineer

To save time and money, refer to the Troubleshooting Guide in Chapter 8 before calling your local PerkinElmer Service Engineer. If you have a problem, it may be minor and you may be able to correct it yourself by using the Troubleshooting Guide to pinpoint the problem.

**Warning****Before starting any maintenance work:**

1. **Switch off all instruments in the system.**
 2. **Disconnect the Pump from the electrical supply.**
 3. **Follow the maintenance instructions exactly as described in this chapter.**
-

Maintaining Pump Accessories

The section describes how to perform the following maintenance procedures:

- Replacing solvent inlet filters and spargers.
- Replacing in-line solvent filter elements.
- Replacing a fuse.
- Cleaning the pump.

Replacing a Solvent Inlet Filter/Sparger

Stainless Steel and titanium solvent inlet filters/spargers are identical (see Figure 7-1) in shape and porosity. The solvent inlet filters are attached to the ends of the pump solvent inlet lines (in the reservoirs) to trap particulates. Solvent inlet filters are also used as spargers when they are attached to the end of the degas lines in each reservoir.

Over time, solvent inlet filters may become partially covered with particulates, thereby restricting solvent flow to the pump. This is a cause of pump flow fluctuations. Water as a mobile phase encourages growth of micro-organisms which the solvent inlet filter traps, thereby restricting solvent flow. You can recognize this situation by observing air bubbles in the solvent inlet tubing.

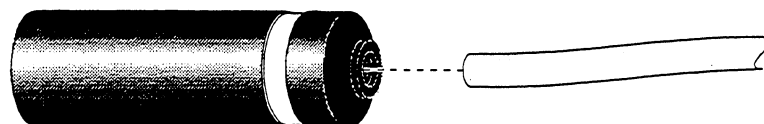
The lifetime of solvent inlet filters or spargers depends on solvent quality, usage, and laboratory environment. Replace or clean the solvent filters every 90 days. You can temporarily unplug solvent inlet filters until you obtain new replacements by removing the solvent inlet filter from the tubing and blowing dry air or helium in the reverse direction of the usual flow. (Force helium into the smaller end of the filter.)

Removing and Replacing Solvent Inlet Filters/Spargers

1. Refer to Table 7-1 for a list of replacement filter/spargers.
2. Cut the tubing as close to the filter/sparger as possible with a razor blade to remove the filter/sparger from the tubing.
3. Grip the solvent inlet filter with one hand and with a piece of sandpaper in your other, grip the Teflon tubing 1/4-inch from the end.
4. Firmly screw the solvent inlet filter/sparger onto the end of the tubing (see Figure 7-1).

Table 7-1. Solvent Inlet Filters/Spargers.

Pore Size	Material	Part No.	Use
10 μ	Stainless Steel	0990-3610	All solvent filtering and organic solvent sparging except buffers.
40 μ	Stainless Steel	0990-3615	Water sparging only.
10 μ	Titanium	N260-0070	All solvent filtering and organic solvent sparging including buffers.
40 μ	Titanium	N260-0089	Water sparging only.

**Figure 7-1. Solvent Inlet Filter/Sparger.**

Replacing an In-line Solvent Filter Element

The in-line solvent filter is typically installed between the pump output tubing and the injector input port. The in-line solvent filter (stainless steel, P/N 0990-3606 or titanium, P/N N260-0259) is available from PerkinElmer. Periodically inspect the in-line solvent filter element and replace it if necessary.

The stainless steel in-line solvent filter contains a stainless steel 0.5 m porosity filter element. Order the In-Line Solvent Filter Replacement Kit (P/N 0254-0311).

The titanium in-line solvent filter contains a titanium 0.2 m porosity filter element (P/N N260-1477).

Replacing a Stainless Steel Filter Element

1. Unscrew the filter fitting from the filter body (see Figure 7-2).
2. Unscrew and remove the filter element from the filter fitting. Discard the filter element and both seals.
3. Insert a new seal over the end of the filter element and screw the filter element into the filter fitting. Insert the O-ring seal in the filter body.
4. Screw the filter fitting into the filter body until it is fingertight.
5. To make a leak-proof seal, use two 1/2-inch wrenches and turn the filter retaining nut into the filter body until it is snug. **Do not overtighten the filter retaining nut!**

Replacing a Titanium Filter Element

1. Unscrew and remove the filter retaining nut from the filter body using a 1/2-inch wrench (see Figure 7-3).
2. Insert a wire (such as a small paper clip) into the inlet end of the filter retaining nut to dislodge the filter.
3. Insert a new filter seal (P/N N260-1262) on the new filter (P/N N260-1477).
4. Insert the new filter element into the filter retaining nut (filter seal side up). Screw the filter retaining nut into the filter body until it is fingertight.
5. To make a leak-proof seal, use two 1/2-inch wrenches and turn the filter retaining nut into the filter body until it is snug. **Do not overtighten the filter retaining nut!**

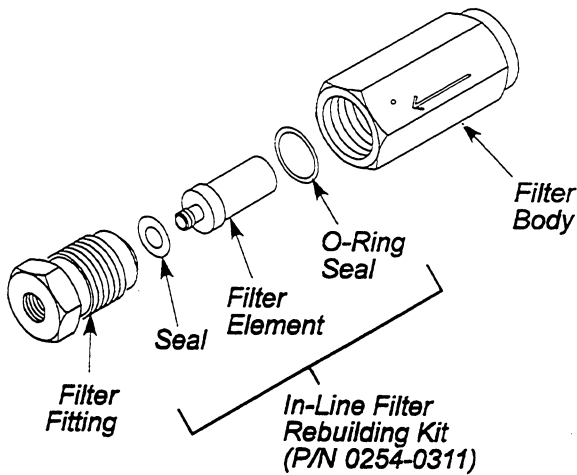


Figure 7-2. Exploded View of the Stainless Steel In-line Solvent Filter.

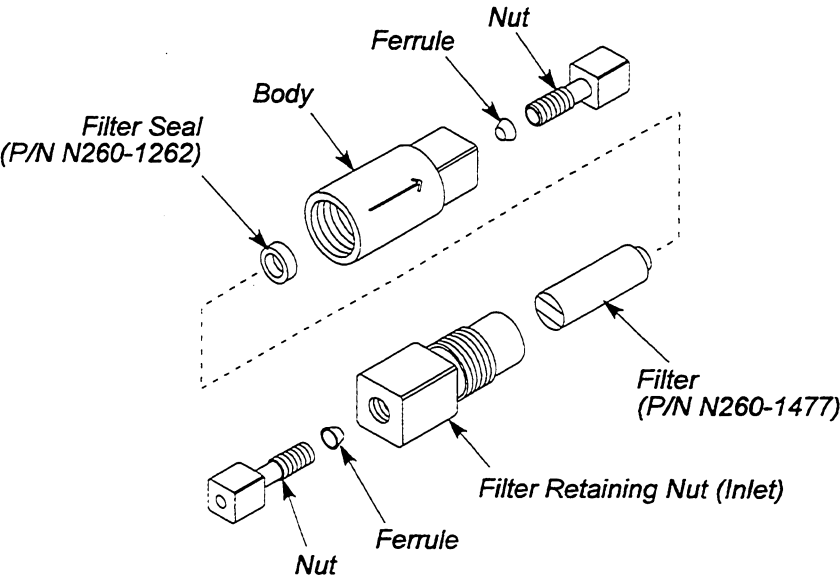


Figure 7-3. Exploded View of the Titanium In-line Solvent Filter.

Replacing a Fuse

To replace a blown fuse (refer to Figure 7-4):

1. Locate the power on/off switch on the right side panel and switch off the pump.
2. Unplug the AC line cord from the AC outlet. Then unplug the AC line cord from the power input module on the rear of the pump.
3. Remove the power input module cover by inserting a small screwdriver in the notch, then carefully prying off the cover.

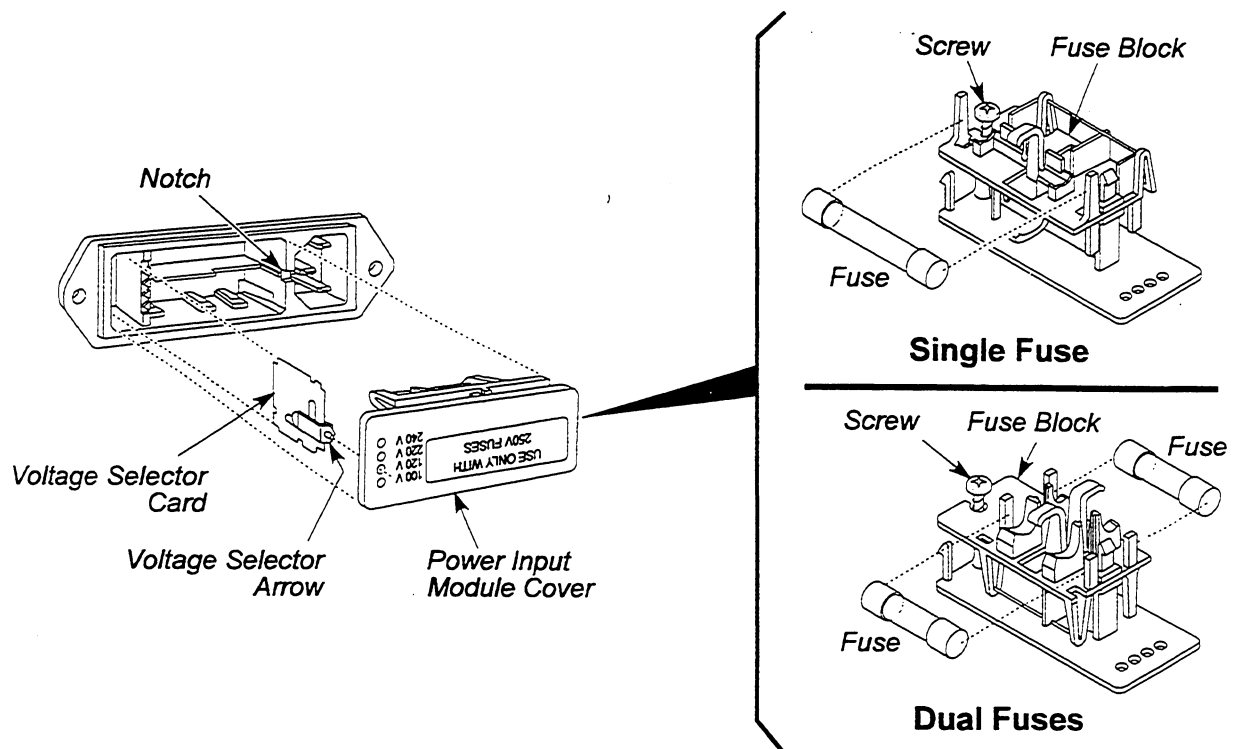


Figure 7-4. Fuse Locations on the Power Input Module.

4. Carefully remove the blown fuse from the fuse block, and replace it with a fuse of identical rating (see Table 7-2).
5. Insert the power input module cover into the power input module.
6. Make sure the power switch is off, then plug one end of the AC line cord into the power input module and the other end into the AC outlet.
7. Switch the pump power on.

**Caution**

If a replacement fuse blows, contact your local PerkinElmer Service Engineer.

Table 7-2. Voltage and Fuse Selection.

AC Voltage Range	Set Voltage	Fuse	Fuse Part No.
90-110V	100	1.6 amp Slow-Blow	0998-1613
108-132V	120	1.6 amp Slow-Blow	0998-1613
198-242V	220	0.8 amp Slow-Blow	0999-1671
216-264V	240	0.8 amp Slow-Blow	0999-1671
Dual Fusing (if required)			
198-242	220	0.8 amp TT	0999-1670
216-264	240	0.8 amp TT	0999-1670

Cleaning the Pump

This section describes how to clean stainless steel and titanium pump parts. Perform this procedure to increase the inertness of titanium parts by creating an oxide surface on the titanium.

To clean the pump:

1. Disconnect and remove the column.
2. Insert the end of the pump output tubing into a beaker.
3. Change the mobile phase in the pump reservoir to isopropanol.
4. Start the pump and collect 20 mL of mobile phase in a beaker to flush the lines of impurities.
5. Connect the pump output tubing to the flowcell inlet.
6. Set the pump flow rate to 3 mL/minute and pump the following sequence of solvents for five minutes each. (Ensure that 15 mL to 20 mL of each solvent is collected):



Warning

Follow standard laboratory procedures and use extreme care when working with strong acids and bases.

- ❶ 100% filtered, distilled water
 - ❷ 10% ammonium hydroxide/water solution
 - ❸ 100% filtered, distilled water
 - ❹ 20% nitric acid/water solution
 - ❺ 100% filtered, distilled water
7. Pump 15 mL to 20 mL of 100% isopropanol to prepare your system for:
 - Any mobile phase except buffers (with buffers flush the flowcell with water first)
 - Short-term shutdown
 - Long-term shutdown

Maintenance on Internal Pump Parts

Routine maintenance of the internal pump parts for both the stainless steel and titanium pumps includes the following:

- Replacing and cleaning check valves.
- Replacing high- and low-pressure piston seals.
- Replacing high- and low-pressure pistons.
- Rebuilding the pulse damper.

Necessary Tools and Kits

Refer to Table 7-3 for a list of the tools required but not supplied. Refer to Table 7-4 for a list and part numbers of the available maintenance and start up kits.

Table 7-3. Tools Required But Not Supplied.

Needle-nose pliers	7/64-in. hex wrench
Phillips screwdriver	5/64-in. hex wrench
Flat-blade screwdriver	5/8-in. wrench
7/16-in. hex wrench	1/4-in. wrench
9/64-in. hex wrench	Torque wrench to 100-in./lbs (5/8-in. crows foot wrench attachment)

Table 7-4. Maintenance and Startup Kits.

Kit	Part Number
Series 200 LC Pump Startup Kit (supplied)	N291-0340
Series 200 BIO LC Pump Startup Kit (supplied)	N291-0341
Series 200 LC Pump Maintenance Kit	N291-0345
Series 200 BIO LC Pump Maintenance Kit	N291-0346

Preparing the Pump for Maintenance

To replace piston seals or clean the high-pressure piston, first remove the pump head from the pump assembly.

Note	To assist you when working on the pump, fold out a complete exploded view of the stainless steel pump (Figure 7-22) or the titanium pump (Figure 7-23) at the end of this chapter.
-------------	---

To prepare the pump for maintenance:

**Warning**

Before starting any maintenance work:

1. **Switch off all instruments in the system.**
2. **Disconnect the Pump from the electrical supply.**
3. **Follow the maintenance instructions exactly as described in this chapter.**

1. To prevent mobile phase from siphoning, carefully remove the solvent inlet lines (avoid twisting them) from the mobile phase reservoirs. Insert the inlet filter portion of the lines into a polypropylene bag or beaker.
2. Remove the solvent system.
3. Open the access door on the front panel and locate the tab (see Figure 7-5).
4. Press the tab and pull the pump drawer forward.

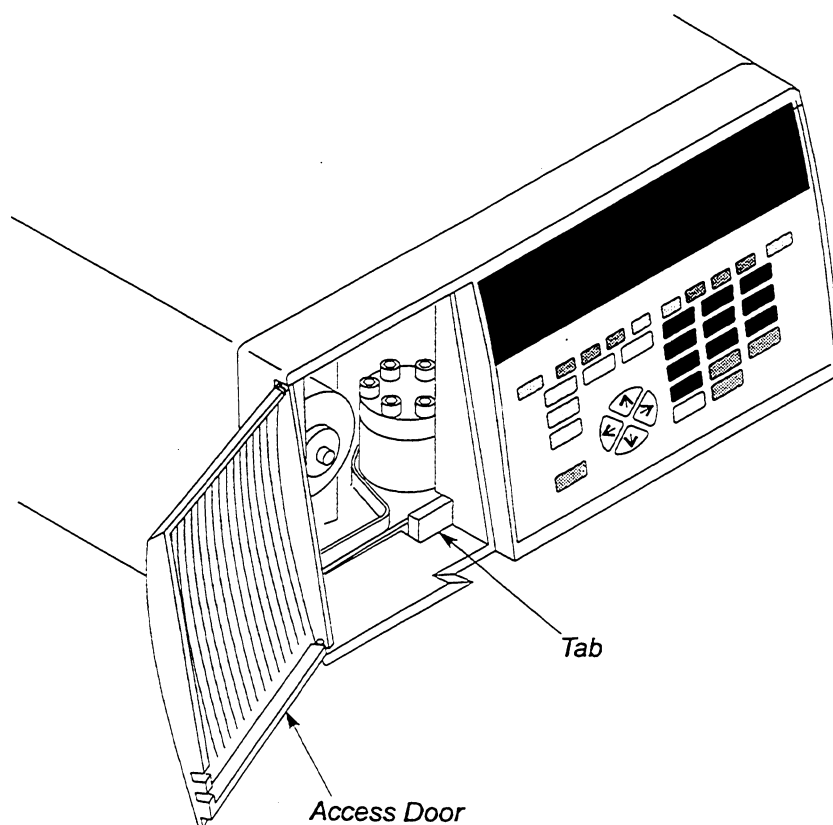


Figure 7-5. Series 200 LC Pump with the Pump Access Door Opened.

Removing the Pump Head

Before removing the pump head assembly you must first remove the low-pressure piston assembly.

Removing the Low-Pressure Piston Assembly

Note

This procedure applies to both the stainless steel pump and titanium pump.

To remove the low-pressure piston assembly:

1. Remove the low-pressure piston guard from the pump head assembly.
2. Remove the hairpin clip from the upper end of each tie rod locking pin.
3. Pull the tie rod locking pins from the low-pressure piston assembly.
4. Carefully slide the low-pressure piston assembly straight out of the pump head assembly to remove it (see Figure 7-6).

**Caution**

Sideways movement of the pump head may break the piston. The piston is made of synthetic sapphire and it is extremely fragile. When removing the piston, carefully support the piston as you slide it straight out.

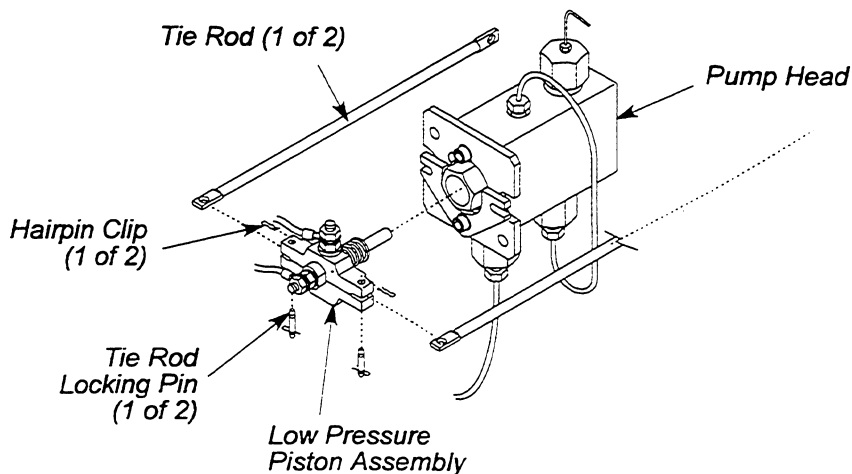


Figure 7-6. Removing the Low-pressure Piston Assembly From the Pump Head Assembly.

Removing the Pump Head Assembly

To remove the pump head assembly:

1. Using a 9/64-inch hex wrench, loosen and remove the #8 x 1/2-inch socket cap screws and #8 washers that secure the pump head assembly to the hex standoffs.
2. Using a 1/4-inch open end wrench, disconnect the stainless steel (or titanium if you have a biocompatible version) fitting from the output check valve.
3. Loosen and remove the solvent input tubing fitting from the center of the proportioning valve.



Caution

In the Biocompatible Pump: Remove the two piston rinse tubes that connect the pump head assembly to the prime/purge valve panel before removing the pump head assembly (see Figure 7-7).

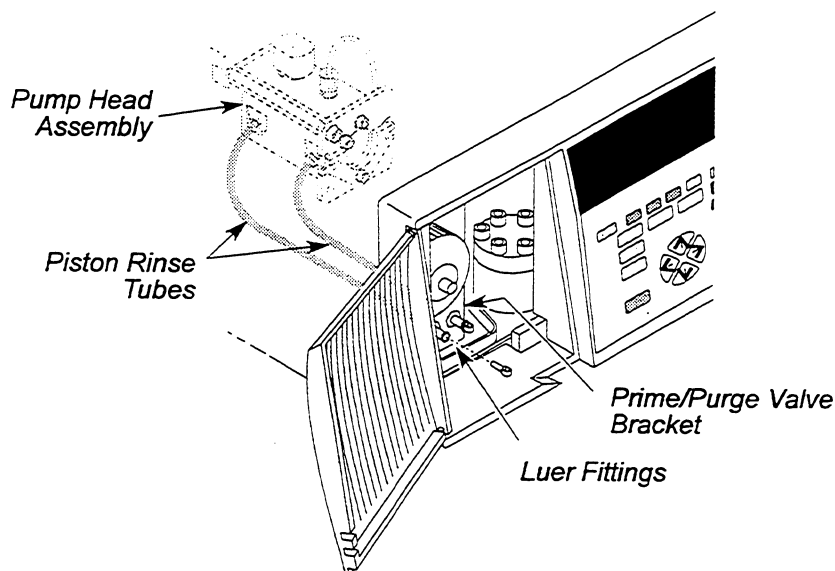


Figure 7-7. Removing the Piston Rinse Tubes.

4. Using a 1/4-inch open end wrench, loosen the fittings and remove the tubing that is connected between the pulse damper and prime/purge valve.
5. Carefully remove the pump head assembly by pulling it straight away from the pump casting (which contains the high-pressure piston).

6. Place the pump head assembly on a flat surface so that you can replace the check valves and piston seals.

**Caution**

Sideways movement of the pump head may break the piston. The piston is made of synthetic sapphire and it is extremely fragile. When removing the pump head assembly, carefully support it as you slide it straight out.

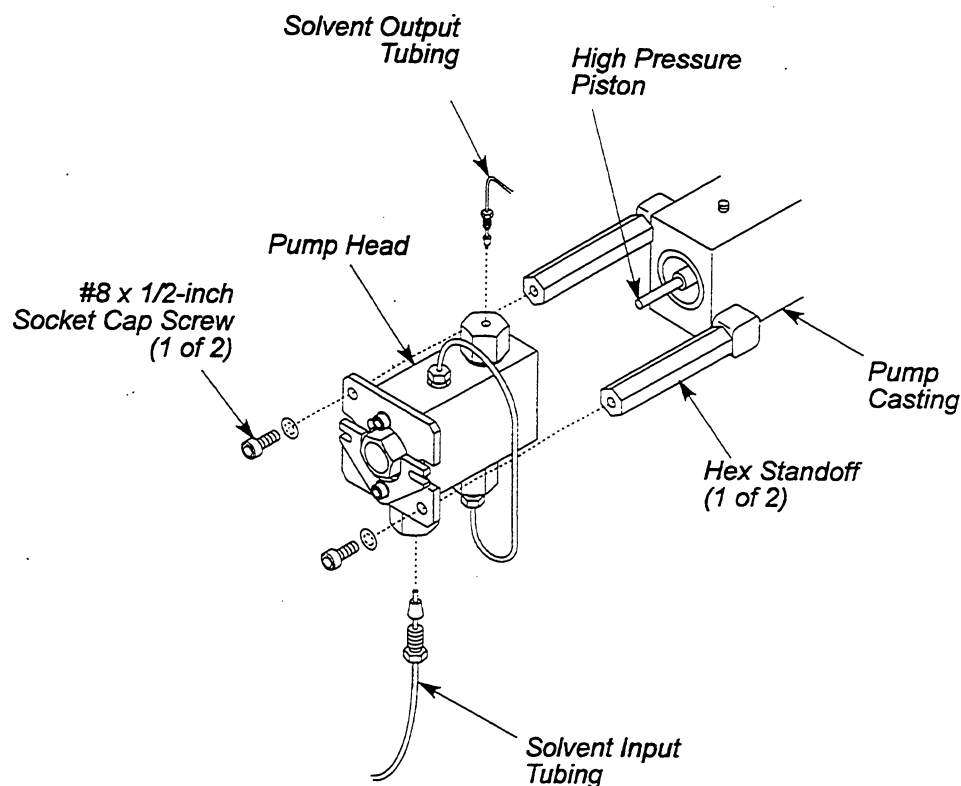


Figure 7-8. Removing the Pump Head.

Replacing Check Valves

Maintenance cycles for the piston seals and check valves are similar; therefore, to minimize downtime, perform as much maintenance as possible at one time.

- Replace high- and low-pressure piston seals, back-up rings, and if you have a biocompatible (BIO) version with titanium pump parts, also replace the piston wash seals.
- Clean the check valves with isopropanol in an ultrasonic bath.

Typical indications of check valve failure include unusual variations of pressure readings on the screen or a change/inconsistency in the peak retention times.

Stainless Steel Pump

The stainless steel pump head contains an input check valve (P/N 0254-0177), an intermediate check valve (P/N 0254-0177), and an output check valve (P/N 0254-0197).

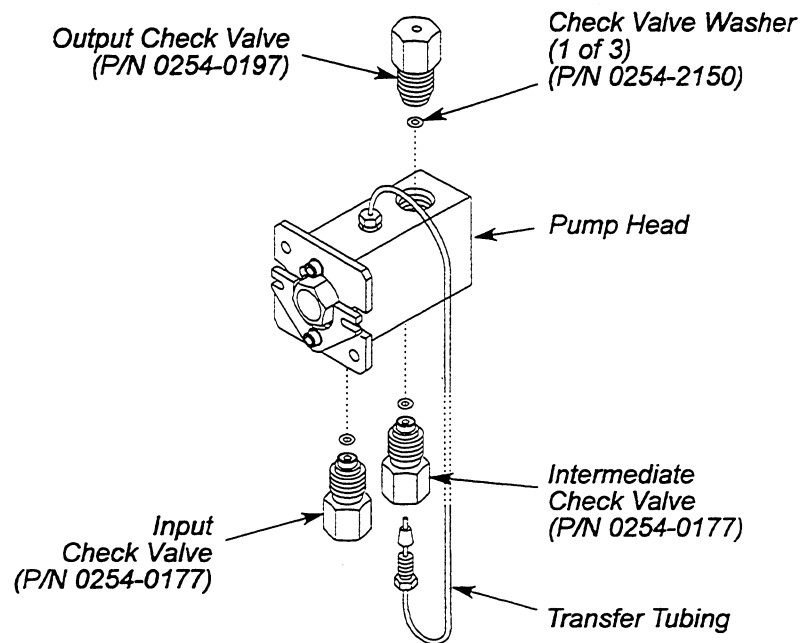


Figure 7-9. Stainless Steel Check Valves.

Titanium Pump

In the biocompatible version, the titanium pump head contains an input check valve (P/N N260-0226), an intermediate check valve (P/N N260-0226), and an output check valve (P/N N260-0192).

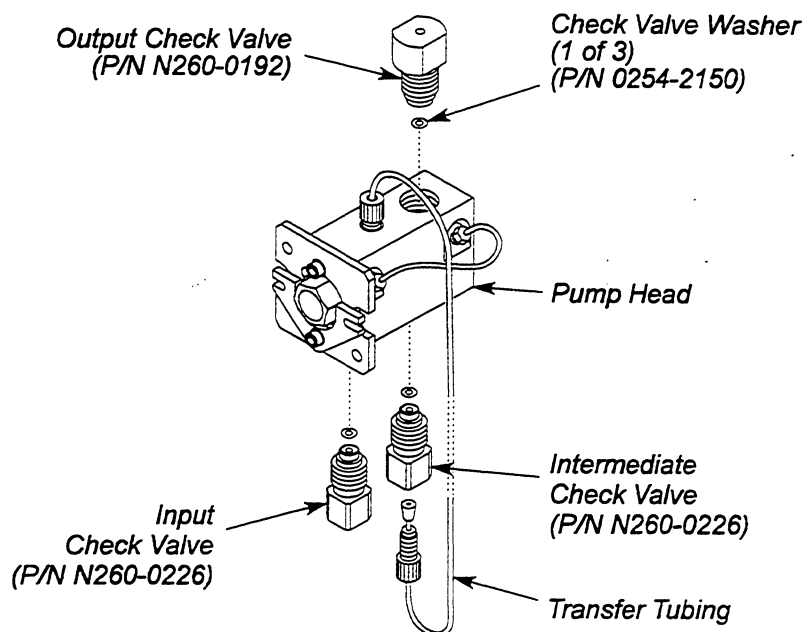


Figure 7-10. Titanium Check Valves.

Replacing Input and Intermediate Check Valves

The intermediate check valve is identical to the input check valve. Both check valves are located on the underside of the pump head. The input check valve is closest to the low-pressure piston (see Figure 7-9 or Figure 7-10).

To replace the input and intermediate check valve:

1. Using a 7/16-inch wrench, disconnect and remove the transfer solvent tubing from the intermediate check valve.
2. Using a 7/16-inch wrench, disconnect and remove the input tubing from the input check valve.

Note If the check valve loosens before the fitting on the solvent input tubing loosens, hold the check valve body with a 5/8-inch wrench while disconnecting the fitting from the check valve.

3. Remove the input check valve with a 5/8-inch wrench.

**Caution**

Use a mirror to inspect the check valve port after removing the old check valve. If the check valve washer is split, then remove all of the pieces from the pump head.

4. Inspect the check valve washer (P/N 0254-2150). Replace a damaged or missing check valve washer. A new check valve washer is supplied with each new input check valve.

Note The check valve washers must sit in the recess of the check valve.

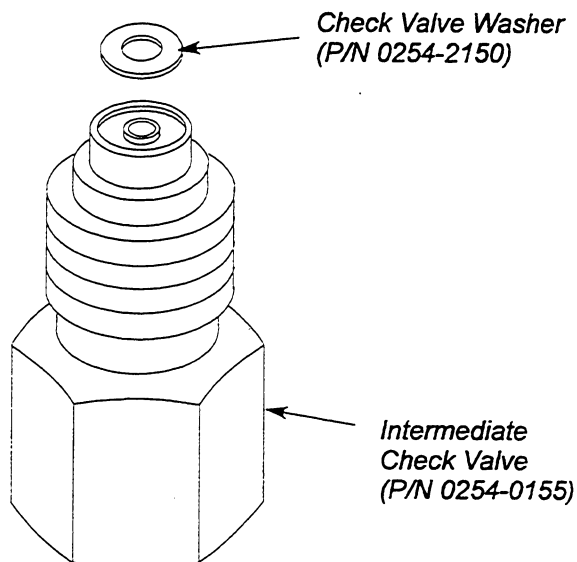


Figure 7-11. Check Valve Washer.

5. Connect the new input check valve to the pump head and tighten it until it is fingertight.

Note Do not use Teflon tape or other sealers on the check valve threads.

6. Using a 5/8-inch wrench, remove the intermediate check valve.
7. Inspect the check valve washer (P/N 0254-2150). Replace a damaged or missing check valve washer. A new check valve washer is supplied with each new input check valve.

Note The check valve washers must sit in the recess of the check valve.

8. Install the new intermediate check valve and tighten it until it is fingertight.
9. Using a torque wrench with a 5/8-inch socket or crow's foot wrench, torque the input and intermediate check valves to 75 inch-lbs (85 kg-cm). Use PerkinElmer's Torque Wrench Kit (P/N 0254-0871) or an equivalent. When torquing, be sure to follow the instructions supplied with the torque wrench.
10. Reconnect the intermediate and input solvent tubing (1/8 to 1/4 turn past a snug fit) to the proper check valves. **Do not overtighten the check valve!** (Refer to Figure 7-9 or Figure 7-10.)

Replacing the Output Check Valve

To replace the output check valve:

1. Using a 1/4-inch wrench, disconnect the high-pressure metal tubing connecting from the output check valve to the pulse damper.

Note If the check valve loosens before the fitting on the output tubing loosens, hold the check valve body with a 5/8-inch wrench while disconnecting the fitting from the check valve.

2. Using a 5/8-inch wrench, remove the old output check valve.
3. Inspect the check valve washer (P/N 0254-2150). Replace damaged or missing check valve washers. A new check valve washer is supplied with each new input check valve.

Note The check valve washers must sit in the recess of the check valve.

4. Install the new output check valve and tighten until it is fingertight.

5. Using a torque wrench with a 5/8-inch socket or crow's foot wrench, torque the output check valve to 75 in-lbs (85 kg-cm). Use PerkinElmer's Torque Wrench Kit (P/N 0254-0871) or equivalent. When torquing, always follow the instructions provided with the torque wrench.
6. Using a 1/4-inch wrench, reconnect the line from the output check valve leading to the pulse damper. **Do not overtighten the check valve!**

Replacing Piston Seals in the Stainless Steel Pump

Large fluctuations in system pressure, low system pressure, leaks in the system, or if you have a biocompatible pump, an increase of the solvent level in the beaker where the piston rinse output tubing is directed are all indications of piston seal failure.

The exploded view of the piston seals from the pump head (Figure 7-12) shows the locations of both the low- and high-pressure piston seals. Table 7-5 lists common replacement parts for stainless steel pumps with their PerkinElmer part numbers.

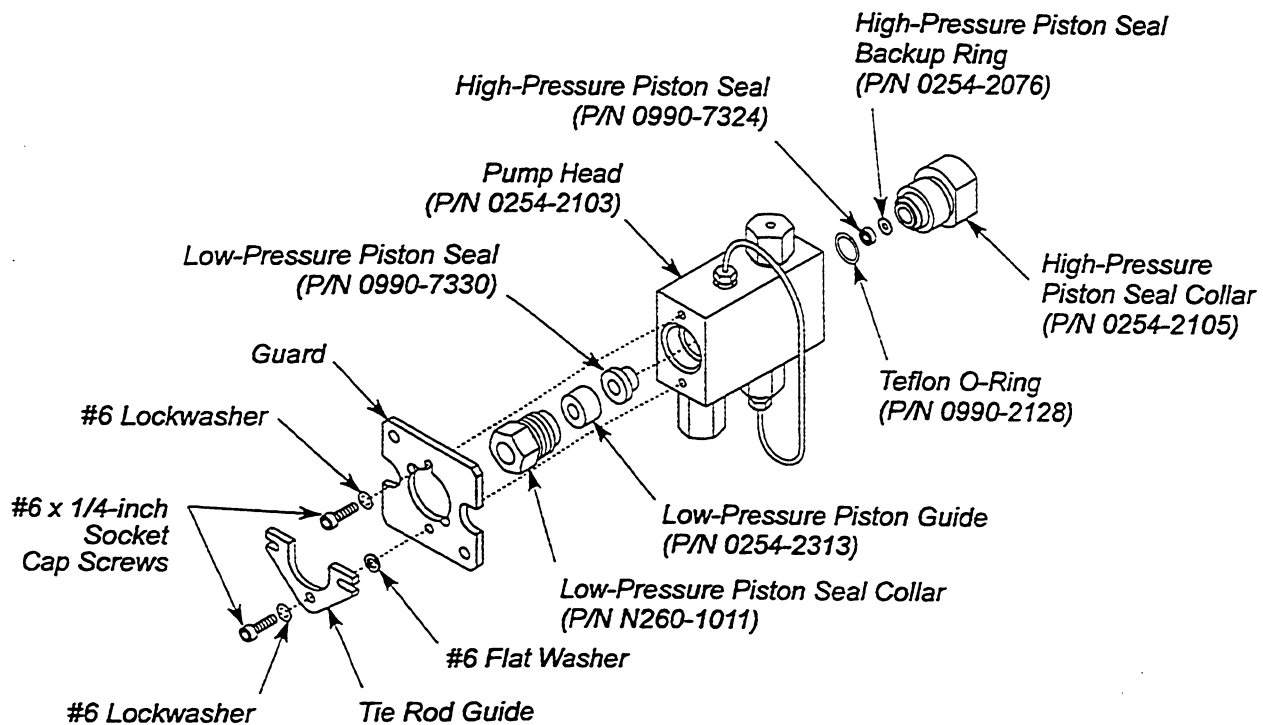


Figure 7-12. Exploded View of Piston Seals From the Pump Head.

Table 7-5. Replacement Parts for Stainless Steel Pumps

Part	Part Number
Low-Pressure Piston Seal	0990-7330
Low-Pressure Piston Guide*	0254-2313
Low-Pressure Piston	N260-0117
Low-Pressure Piston Seal Collar	N260-1011
High Pressure Piston Seal (yellow)	0990-7324
High-Pressure Piston	N260-0124
High-Pressure Piston Seal Backup Ring*	0254-2076
Stainless Steel Pump Head	0254-2103
High-Pressure Seal Collar*	0254-2105
O-Ring (High-Pressure Side)*	0990-2128
High-Pressure Piston	N260-0124

* It is required that you replace these items when you replace the associated piston seal.

Many of the above parts are included in the Series 200 LC Pump Maintenance Kit (P/N N291-0345).

Replacing the Low-Pressure Piston Seal

To replace the low-pressure piston seal:

1. Remove the pump head. (See the "Removing the Pump Head" section on page 7-13.)
2. Using a 7/64-inch hex wrench, loosen and remove the #6 x 1/4-inch socket cap screw and #6 washer that secure the tie rod guide to the pump head assembly. Then remove the tie rod guide.
3. Using a 7/64-inch hex wrench, loosen and remove the other #6 x 1/4-inch socket cap screw and #6 washer directly above it (see Figure 7-12). Then remove the guard.
4. Remove the low-pressure piston seal collar with a 3/4-inch wrench.
5. Remove the low-pressure piston seal from the pump head with a small clean tool.
6. Insert a new low-pressure piston seal (P/N 0990-7330) in the pump head.
7. Remove the low-pressure piston guide from the low-pressure piston seal collar.
8. Insert a new low-pressure piston seal guide (P/N 0254-2313) in the low-pressure piston seal collar.
9. Insert the low-pressure piston seal collar in the pump head and turn it until it is finger-tight. Make a leak-free seal by tightening the low-pressure piston seal collar an additional 1/2 turn with a 3/4-inch wrench.
10. Inspect and clean the low-pressure piston. (See the "Inspecting and Cleaning Pump Pistons" on page 7-29.)

Replacing the High-Pressure Piston Seal

To replace the high-pressure piston seal:

1. Remove the high-pressure piston seal collar with a 3/4-inch wrench.
2. Using the piston seal removal tool (P/N N260-1295), push the high-pressure piston seal and backup ring out of the high-pressure piston seal collar.

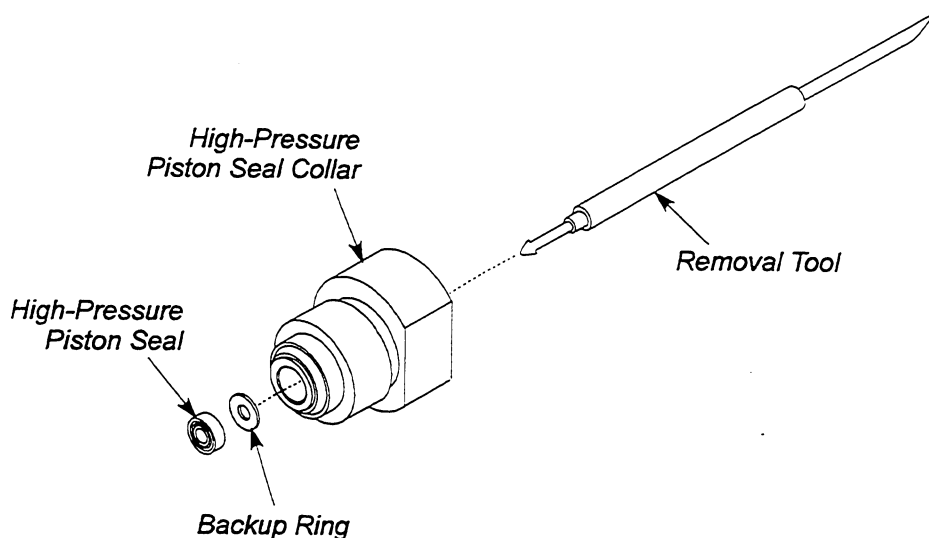


Figure 7-13. Removing the High-pressure Piston Seal and Backup Ring.

3. Place a new backup ring and high-pressure piston seal into the high-pressure piston seal collar.



Caution

Always position the piston seal so that the spring side is not facing the piston.

4. Insert the new high-pressure piston seal in place with your thumb.
5. Remove the Teflon O-ring from the pump head and discard it.
6. Insert a new Teflon O-ring (P/N 0990-2128) over the end of the high-pressure piston seal collar.
7. Insert the high-pressure piston seal collar in the pump head and turn it until it is finger-tight. Make a leak-free seal by tightening the high-pressure piston seal collar an additional 1/2 turn with a 3/4-inch wrench.
8. Inspect and clean the high-pressure piston. (See the "Inspecting and Cleaning Pump Pistons" section on page 7-29.)

Replacing Piston Seals in the Titanium Pump

Large fluctuations in system pressure, low system hydraulic pressure, leaks in the system, or an increase of the solvent level in the beaker (where the piston rinse output tubing is directed) are all indications of piston seal failure.

Table 7-6 lists common replacement parts for biocompatible (BIO) titanium pumps with their PerkinElmer part numbers. Many of the following parts are included in the Series 200 LC Pump Maintenance Kit (P/N N291-0346).

Table 7-6. Replacement Parts for Titanium Pumps

Part	Part Number
Low-Pressure Piston Seal	0990-7339
Low-Pressure Piston Seal Holder	N260-1174
O-ring (For Low-Pressure Piston Seal Cover)	0990-2018
Low-Pressure Piston	N260-0104
Low-Pressure Piston Seal Cover	N260-1175
High-Pressure Piston Seal (2 required)	0990-7338
High-Pressure Piston Seal Holder	N260-1434
Backup Ring*	0254-2076
Pump Head	N260-1433
High-Pressure Seal Cover	N260-1172
O-ring (Black, High-Pressure Side)	0990-2009
O-ring (White, High-Pressure Side)	0990-2018
High-Pressure Piston	N260-0124
1/8-inch Tefzel Nut	N260-1189
1/8-inch Ferrule	0990-3771
Piston Seal Replacement Kit	N260-0061

* It is required that you replace these items when you replace the associated piston seal.

Note Store the piston seals in a vial of isopropanol to properly break them in and extend their life.



Caution

1. Always replace both high-pressure piston seals and low-pressure piston seals whenever seal replacement is necessary.
 2. Clean both pistons or replace them if necessary.
-

Replacing the Low-Pressure Piston Seals

To replace low-pressure piston seals:

1. Remove the pump head. See the "Removing the Pump Head" section on page 7-13.

**Caution**

In the Biocompatible Pump: Remove the two piston rinse tubes that connect the pump head assembly to the prime/purge valve panel before removing the pump head assembly (see Figure 7-7).

2. Remove the low-pressure piston seal cover screws with a 7/64-inch hex wrench, then remove the low-pressure piston seal cover and the O-ring.
3. Using the barbed end of the seal removal tool (P/N N260-1295), carefully remove one piston seal from the low-pressure piston seal holder and the other piston seal from the pump head.
4. Insert a new low-pressure piston seal into the pump head, insert the other new low-pressure piston seal into the low-pressure piston seal holder, and insert the low-pressure piston seal holder into the pump head (see Figure 7-14).
5. Clean the piston. See the "Inspecting and Cleaning Pump Pistons" section on page 7-29.
6. Replace the O-ring on the low-pressure piston seal holder.
7. Replace the low-pressure piston seal cover and loosely install the cover screws.

Note **The low-pressure piston seal cover has a slight concave shape. Install the cover so that the convex side faces the low-pressure piston seals.**

8. Install the low-pressure piston through both seals.
9. With the low-pressure piston in place, equally tighten both cover screws.
10. Reinstall the pump head (see "Reinstalling the Pump Head" on page 7-29).

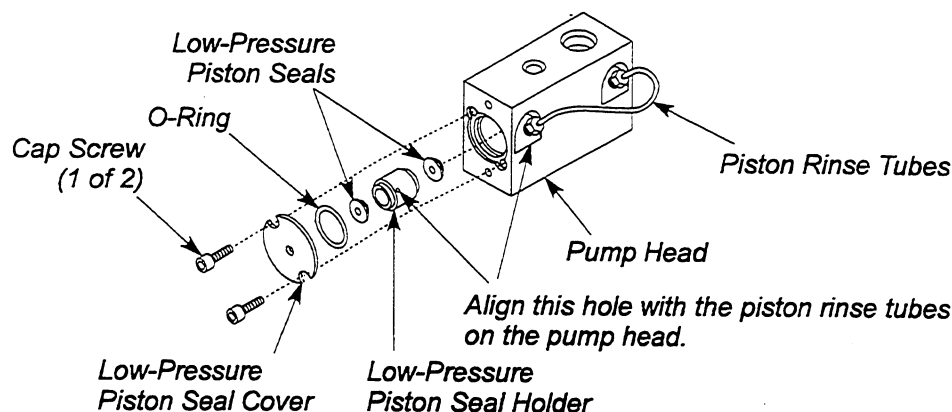


Figure 7-14. Replacing the Low-pressure Piston Seals.

Replacing the High-Pressure Piston Seals

Mobile phase leakage around the pump head is an indication of a defective high-pressure piston seal.

To replace high-pressure piston seals:

1. Remove the pump head. Refer to the "Removing the Pump Head" section on page 7-13.

Note It is not necessary to remove the high-pressure piston to replace the high-pressure seals.

There are two piston seals for the high-pressure piston. Although both seals have the same part number (P/N 0990-7338), we will refer to them by separate names:

- Wash seal - This seal is closest to the high-pressure seal cover.
 - High-pressure seal - This is the seal closest to the pump head.
2. Remove the wash seal from the high-pressure seal cover with the barbed end of the seal removal tool (Figure 7-15).
 3. Carefully remove the piston seal holder containing the high-pressure piston seal.
 4. Remove the black O-ring from the holder. Remove the white Teflon O-ring from the pump head with the sharp end of the seal removal tool.
 5. Remove the high-pressure seal and backup ring from the piston seal holder with the barbed end of the seal removal tool.
 6. Discard the used seals and O-rings.

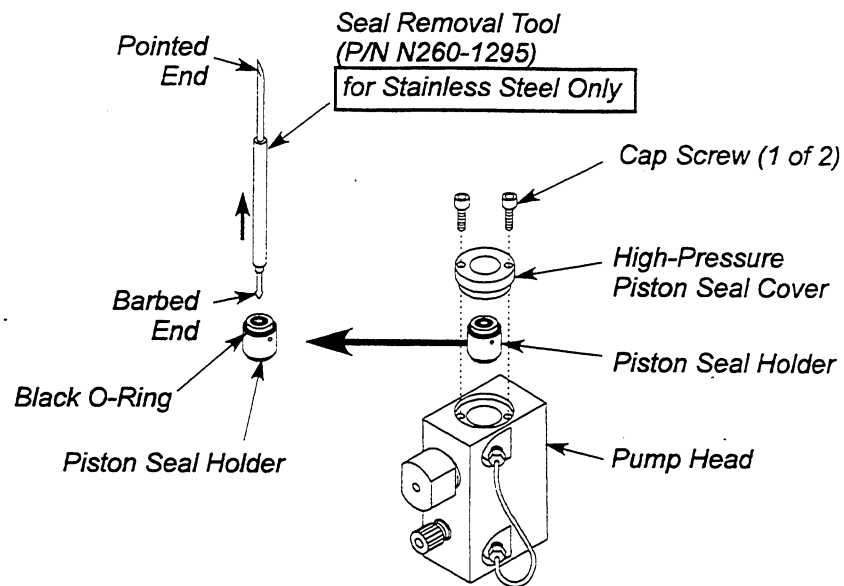


Figure 7-15. Removing the High-pressure Piston Seals.

7. Insert new seals, backup ring, and white O-ring into the piston seal holder as shown in Figure 7-16.



Caution

There is only ONE correct way to insert the seals. Refer to Figure 7-16 for the correct installation.

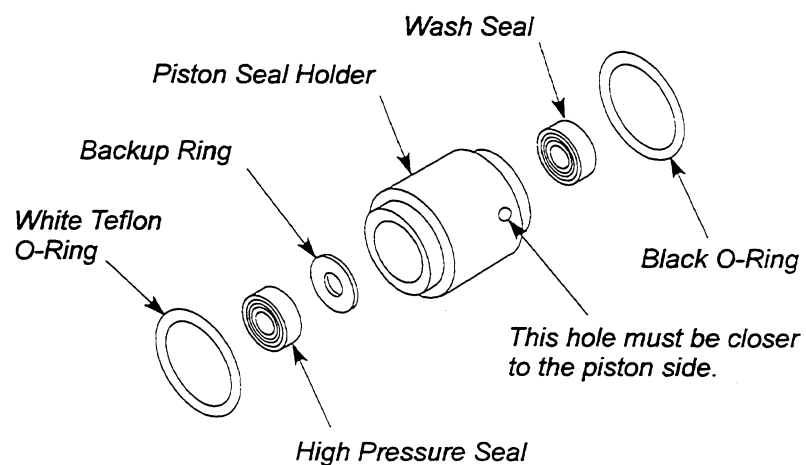


Figure 7-16. High-pressure Piston Seals and Holder (exploded view).

8. Insert the piston seal holder into the pump head. Align the holes in the holder with the piston rinse fittings on the pump head (Figure 7-17).

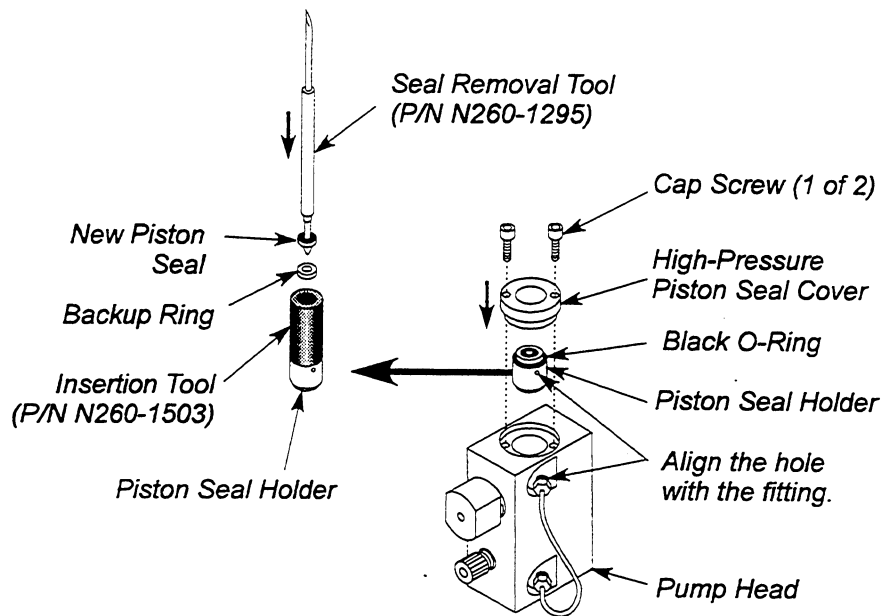


Figure 7-17. Installing New High-pressure Piston Seals.

9. Insert a new black O-ring on the top of the piston seal holder.
10. Replace the high-pressure seal cover. Tighten each cap screw with a 7/16-inch wrench, alternating 1/4 turn between each screw, until both screws are equally torqued to approximately 12 foot-pounds.
11. Inspect and clean the pump pistons.
12. Reinstall the pump head (refer to the "Reinstalling the Pump Head" section on page 7-29).



Caution

To properly break in the pump seal, pump organic solvent (methanol or isopropanol) for 15 to 30 minutes at low-pressure (less than 2 000 psi) and at a low flow rate (3 mL/min or less). Then run the pump for an additional 15 minutes at higher pressures (2 000 - 5 000 psi).

Inspecting and Cleaning Pump Pistons

It's good practice to clean the pump pistons each time you replace pump piston seals. First, visually inspect the pistons for scratches. Subsurface imperfections in the sapphire can appear to be scratches. If you are not replacing piston seals, remove the pump head (refer to the "Removing the Pump Head" section on page 7-13) to expose the piston.

Note It is not necessary to remove the high-pressure piston for cleaning.

Gently scrub the piston with "Pepsodent" toothpaste or an equivalent toothpaste that has a high diatomaceous earth content. Flush the toothpaste from the piston with distilled water, or hand wipe the piston with a Kimwipe tissue saturated with distilled water.



Caution

If you cannot remove the scratches from the piston, then replace the piston. Refer to either the section, "Removing the Low-Pressure Piston" on page 7-13 or "Replacing the High-Pressure Piston" on page 7-30.

Reinstalling the Pump Head

To reinstall the pump head:

1. Arrange the pump head with the high-pressure side facing the high-pressure piston (which is sticking out of the pump casting).
2. Carefully insert the high-pressure side of the pump head over the high-pressure piston and hold it in place.



Caution

Sideways movement of the pump head may break the piston. The piston is made of synthetic sapphire and it is extremely fragile. When installing the pump head assembly, carefully support it as you insert the pump piston into the pump head assembly.

3. Align the guard and tie rod guide with the holes in the pump head. Secure both items with the proper screws.

4. Reconnect the solvent input tubing to the proportioning valve (or if you have a Series 200 LC Isocratic Pump, reconnect the solvent input tubing to the input check valve).
5. Reconnect the solvent output tubing to the output check valve. Reconnect the tubing between the pulse damper and prime/purge valve. To make a leak-free connection, use a 1/4-inch open end wrench to tighten the fittings about 1/4 turn past fingertight.
6. Carefully insert the low-pressure piston assembly into the pump head.
7. Connect the tie rods to the low-pressure piston assembly with the retaining pins and hairpin clips.
8. Replace the low-pressure piston guard.
9. Slide the pump drawer into the cabinet until the tab locks it in place. Replace the solvent system, insert the solvent lines into the reservoirs, and plug the AC line cord into power input module and the AC outlet.

Replacing the High-Pressure Piston



Caution

The pump head must be installed so that you can relieve the spring tension by following steps 1-3.

To replace the high-pressure piston:

1. Turn the pump cam to bring the cam follower to its lowest point, to fully retract the high-pressure piston.
2. Loosen the setscrew on top of the pump casting with a 5/64-inch hex wrench (see Figure 7-18).
3. Remove the low-pressure piston assembly.



Caution

Sideways movement of the pump head may break the piston. The piston is made of synthetic sapphire and it is extremely fragile. When removing the pump head, carefully support it as you slide it straight out.

4. Remove the pump head assembly. Refer to the "Removing the Pump Head" section on page 7-13.

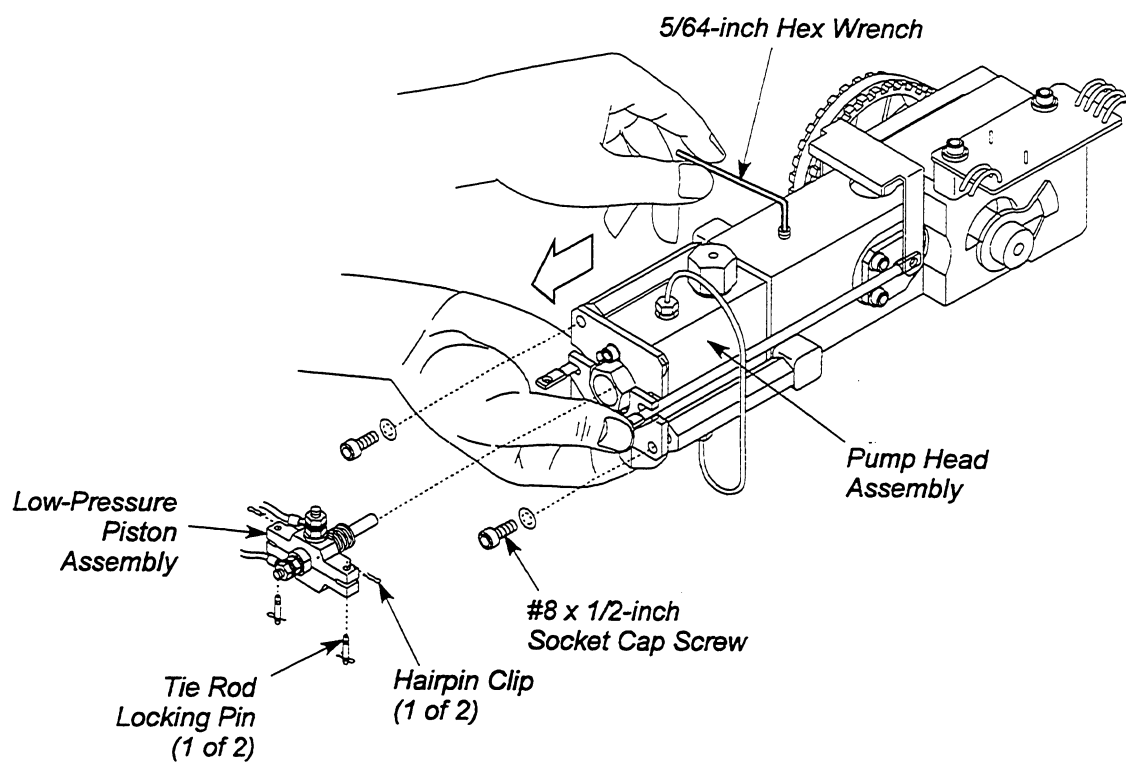


Figure 7-18. Loosening the Setscrew and Removing the Pump Head Assembly.

Removing the Old High-Pressure Piston

To remove the old high pressure piston follow these steps and refer to Figure 7-19. The steps listed here are repeated in Figure 7-19.

1. Remove the pump piston spring retainer and piston spring from the pump.
2. Remove the two tie rods.
3. Remove the tie rod retainer by pushing it down and sliding it off both ends of the cam follower pin.
4. Remove two cap screws from each retaining plate by using a 9/64-inch hex wrench.
5. Remove the two retaining plates.
6. Remove the two bushings from the cam follower pin.
7. Remove the cam follower pin by pulling it straight out with pliers.
8. Carefully pull the high-pressure piston out of the pump casting.
9. Remove the cam follower from the high-pressure piston.

Installing a New High-Pressure Piston

To install a new high-pressure piston:

1. Insert the cam follower into the fork on the new piston assembly.
2. Slide the pump piston into the pump.
3. Insert the cam follower pin through the pump casting and cam follower in the piston.

Note Lift the cam follower from the opposite side so that the pin will slide in easily.

4. Replace the two bushings on the cam follower pin. Lubricate the bushings.
5. Replace the retaining plates and cap screws on the pump casting. Tighten the cap screws with a 9/64-inch hex wrench.
6. Replace tie rods on each side of the cam follower pin.
7. Replace the tie rod retainer on the spring. Then insert the ends of the cam follower pin through the holes in each side of the tie rod retainer.

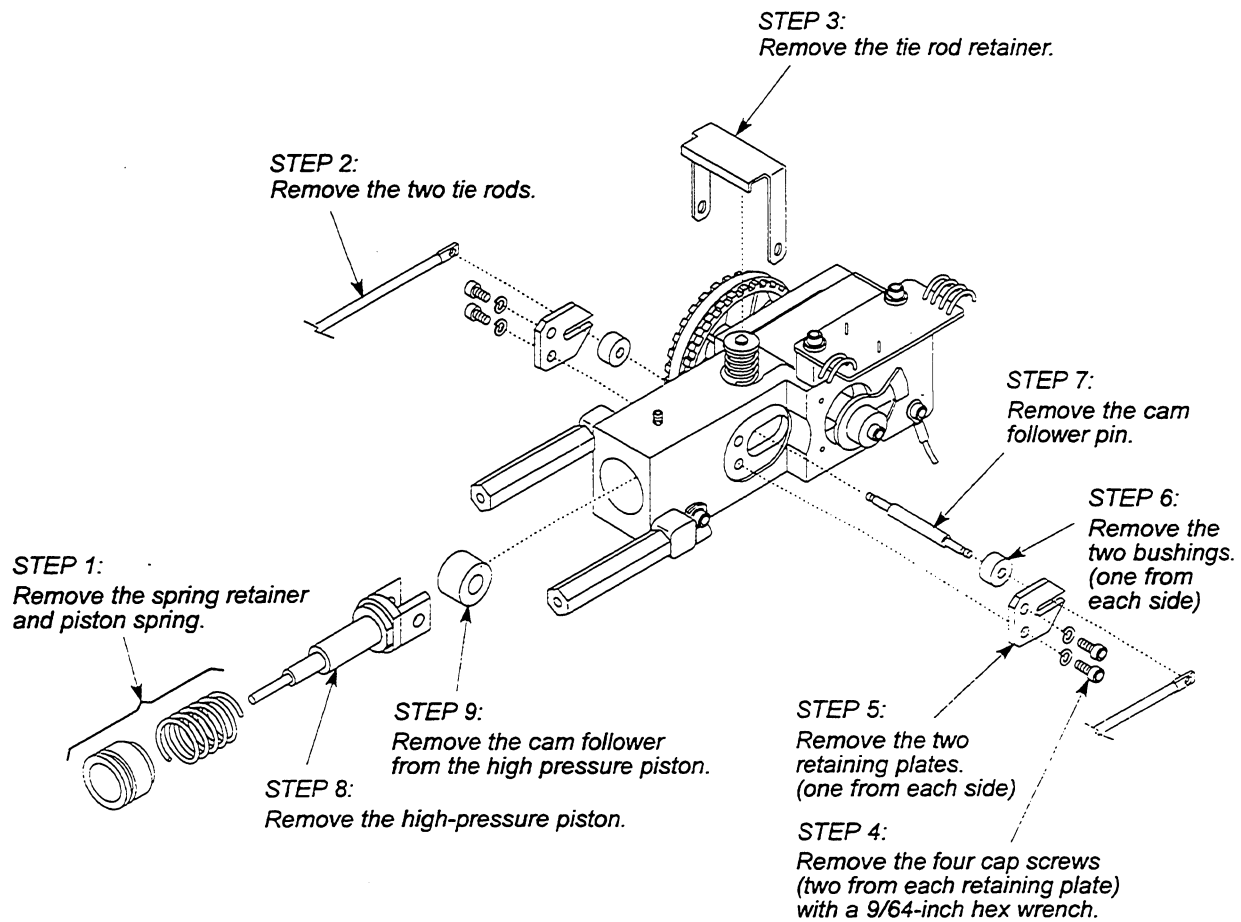


Figure 7-19. Steps to Remove a High-pressure Piston.

8. Insert the piston spring and spring retainer into the pump casting.
9. Carefully insert the high-pressure side of the pump head over the high-pressure piston and hold it in place.



Caution

Sideways movement of the pump head may break the piston. The piston is made of synthetic sapphire and it is extremely fragile. When installing the pump head, carefully support it as you slide the pump piston into it.

10. Tighten the setscrew (on top of the pump to lock the spring retainer into place) with a 9/64-inch hex wrench.

11. Reinstall the tie rod holder on the pump head.
12. Reinstall the low-pressure piston, tie rod locking pins, and clips.
13. Reconnect the solvent input tubing to the proportioning valve (or if you have a Series 200 LC Isocratic Pump, reconnect the solvent input tubing to the input check valve).
14. Reconnect the solvent output tubing to the output check valve. Reconnect the tubing between the pulse damper and prime/purge valve. To make a leak-free connection, use a 1/4-inch open end wrench to tighten the fittings about 1/4 turn past fingertight.
15. Replace the low-pressure piston guard.

Removing/Rebuilding the Pulse Damper

Excessive baseline noise or a faint tint of blue in the waste container are signs of a failed pulse damper, which must be rebuilt or replaced.

Note	When rebuilding the pulse damper, always replace the damper seal.
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Necessary Tools

Refer to Table 7-7 for a list of the tools required but not supplied.

Table 7-7. Tools Required But Not Supplied.

1/4-in. hex wrench	1/4-in. open end wrench
Blue food coloring	1/2-in. open end wrench
Flat-blade screwdriver	Torque wrench

Removing the Pulse Damper

To remove the pulse damper from the cabinet:

1. Gain access to the pulse damper (refer to the "Preparing the Pump for Maintenance" section on page 7-11).
2. Loosen and remove the tubing (using a 1/4-inch opened end wrench) connected to the top of the pulse damper.
3. Remove the six screws that secure the cover over the electronics area. Remove the cover.
4. Unplug the pressure transducer harness from the pump control board.
5. If equipped, remove the optional mixing coil.
6. Remove the two screws that secure the pulse damper to the chassis (accessible from the underside of the chassis).
7. Remove the pulse damper from the pump and put it on a flat surface.

Rebuilding the Pulse Damper

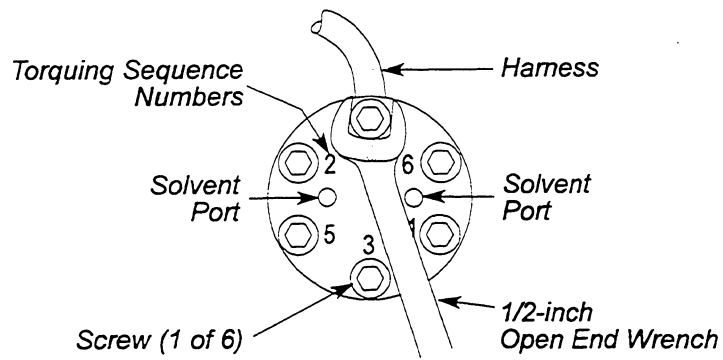
To rebuild the pulse damper, follow these steps and refer to Figure 7-20.

1. Hold the pulse damper firmly in place with a 1/2-inch open end wrench (see top view of pulse damper in Figure 7-20) or insert the pulse damper into a bench vise with brass jaws.
2. Unscrew the six large 1/4-inch hex head cap screws (using the 1/4-inch hex wrench) that secure the pulse damper cover by following the number sequence shown in Figure 7-20.
3. Remove the pulse damper cover shown in the exploded view of the pulse damper in Figure 7-20.
4. Carefully remove the diaphragm and square damper seal.
5. Remove the plug from the housing by grasping it with your fingernails and pulling it out.

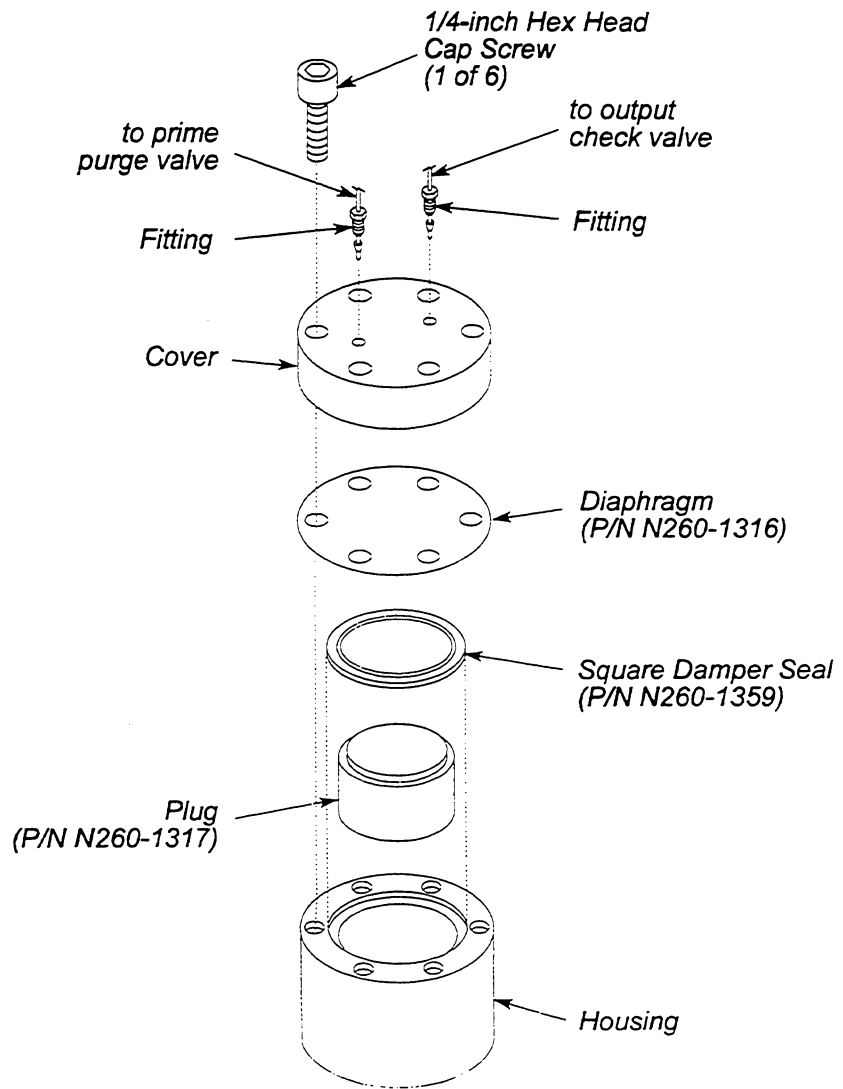
**Caution**

Never use a sharp object (for example, a screwdriver or tweezers) to remove the plug. Scratching the corner of the housing cavity may cause the pulse damper to leak.

6. Inspect the plug for damage or swelling (from contact with solvents), then clean the cavity. Replace a damaged plug (P/N N260-1317).
7. Install the plug into the housing with the concave side facing up.
8. Apply one drop of blue food coloring (about 1/16-inch diameter) in the center of the concave side of the plug. The presence of blue coloring in the waste solvent reservoir indicates a ruptured diaphragm.
9. Install a new square damper seal (P/N N260-1359) into the housing.
10. Place a new diaphragm (P/N N260-1316) on the pulse damper housing. Align the holes in the diaphragm with the holes in the housing.
11. Align the holes in the pulse damper cover with the holes in the housing and position the solvent ports perpendicular to the harness.
12. Apply Fluoro Glide FB (P/N 0990-8353) to the threads of the six 1/4-inch hex head cap screws, then screw all six screws fingertight.
13. Torque all six 1/4-inch hex head cap screws to 36 inch-lbs by following the number sequence in shown in the top view of the pulse damper in Figure 7-20. Then torque all six screws to 84 inch-lbs, and finally to 180 inch-lbs.
14. Reinstall the pulse damper in the pump (and, if equipped, reinstall the optional mixing coil).
15. Start the pump, pressurize the system to 6 000 psi, and check for leaks. Observe the pressure display on the pump to verify that the pulse damper is working properly.



Top View of Pulse Damper with Torquing Sequence



Exploded View of Pulse Damper

Figure 7-20. Rebuilding the Pulse Damper.

Lubricating the Cam Wiper Assembly

Once a year or after 2 000 hours of use, lift the cam wiper and apply approximately 1/4 oz. (6 cc or 7 cc) of Molykote Grease (P/N 0990-8511) to the white felt. See Figure 7-21 for the location of the cam wiper.

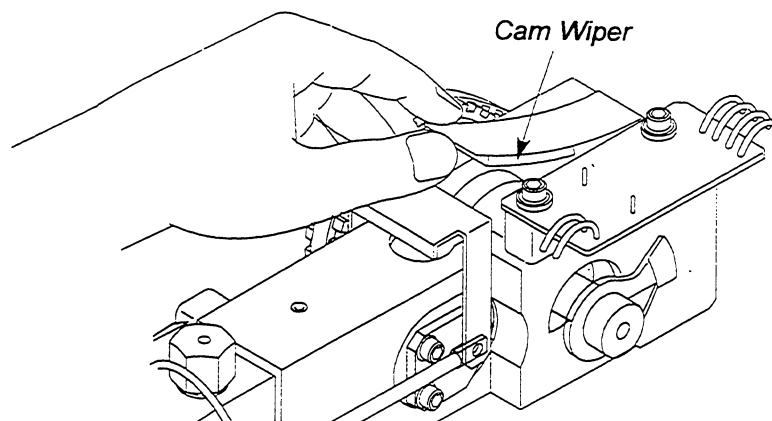


Figure 7-21. Location of the Cam Wiper.

Reassembling the Series 200 LC Pump

To reassemble the Series 200 LC Pump:

1. Slide the pump drawer into the cabinet.
2. Replace the solvent system.
3. Return all mobile phase reservoirs in the solvent system and insert the corresponding tubing.



Caution Do not overfill the solvent reservoirs.

4. Reconnect the power cord, then switch on the pump.
5. Purge the pump and check for leaks.

Troubleshooting 8

Troubleshooting Guide Table .. 8-1

Troubleshooting 8

This chapter describes how to solve some basic performance and instrument problems.

Table 8-1. Troubleshooting Guide Table.

PROBLEM	PROBABLE CAUSE	SOLUTION
1. Pump will not start.	Series 200 LC Pump line cord is not plugged into the AC outlet.	Plug Series 200 LC Pump line cord into the AC outlet.
	Power switch is off.	Turn on the power switch.
	Blown fuse.	Replace the fuse.
	Faulty low-voltage power supply board.	Call your PerkinElmer Service Engineer.
2. Pump automatically stops.	System pressure exceeded the maximum pressure of 6 100 psi.	Check for a plugged column by disconnecting the column inlet tube; then restart the pump.
		If the pump still shuts down, check for a plugged injector by disconnecting the tubing from the injector inlet, then restart the pump.
		The optional in-line filter may be plugged. Clean or replace the filter element.
	Injector valve improperly positioned or blocked.	Check the injector for proper rotation.
	Plugged flowcell.	Turn the pump off and disconnect the flowcell inlet tubing.
		Backflush the flowcell by pumping mobile phase in the reversed direction.
3. No solvent flow; pump motor is running.	A solvent leak somewhere in the system.	Locate the leak and repair it.
	Air in the pumping system.	Prime the pump to remove the air.
	Dirty pump piston.	Clean the pump piston.

PROBLEM	PROBABLE CAUSE	SOLUTION
3. No solvent flow; pump motor is running. (continued)	No mobile phase in the solvent reservoirs.	Refill the solvent reservoirs.
	Clogged solvent inlet filter.	Clean or replace the solvent inlet filter.
	Faulty check valves (most likely the input check valve).	Identify and replace the check valve.
	Defective pump motor.	Call your PerkinElmer Service Engineer.
4. Mobile phase leaking from the pump head.	Loose mobile phase inlet and /or outlet fittings.	Tighten the fittings.
	Loose check valve.	Tighten the check valve.
	Defective piston seal.	Replace the defective piston seal.
	Dirty pump piston.	Clean the pump piston.
5. Pump flow fluctuation	Mobile phase flow restriction in the solvent inlet filter.	Clean or replace the solvent inlet filter.
	Air in the solvent inlet tubing.	Prime the pump.
		Degas the mobile phase.
	Defective output check valve.	Replace the output check valve.
6. Pump piston not reciprocating.	Motor drive failure.	Call your PerkinElmer Service Engineer.
7. Flow rate too high or too low.	Pump out of calibration.	Call your PerkinElmer Service Engineer.
8. Piston seals have a short life span.	Dirty mobile phase.	Filter the mobile phase.
		Clean the pistons.
	Scratches on the pump piston.	Replace the pump piston.
9. Noisy chromatographic baseline	Air in the pumping system.	Prime the pump.
	Air bubbles passing through the detector flowcell.	Degas the mobile phase. Add a backpressure device to the detector.
	Faulty or dirty check valve	Clean or replace the check valve.
	Faulty pulse damper.	Repair or replace the pulse damper.
	Detector failure.	Check or repair the detector.
10. Spiking on the chromatographic baseline.	Air bubbles passing through the detector flowcell.	Prime the pump. Degas the mobile phase. Locate and repair solvent leaks.

PROBLEM	PROBABLE CAUSE	SOLUTION
11. Excessive pressure fluctuation.	Sticking or dirty check valve.	Clean or replace the check valve.
	Air in the pumping system.	Prime the pump. Degas the mobile phase.
	Insufficient back pressure.	Add sufficient back pressure.
	Faulty piston seals.	Replace piston seals.
12. Inconsistent sample retention times.	Changing flow rate.	Run the start up diagnostics, then call your PerkinElmer Service Engineer.
	Column activity changing.	Replace the column.
	Leak in the system.	Locate and repair the leak.
	Incorrect or contaminated mobile phase.	Replace the mobile phase.
	Injector problems.	Identify and repair.
	Gas in the mobile phase.	Degas the mobile phase.
13. Pump shuts down after one minute. It does not reach Pmin.	Pmin is set too high.	Change the Pmin to a lower value.
	Leak in the system.	Locate and repair.
	Faulty pressure transducer.	Call your PerkinElmer Service Engineer.
	Pressure transducer needs calibration.	Call your PerkinElmer Service Engineer
14. Pump shuts down. It exceeds Pmax.	Pmax is set too low.	Change the Pmax setting.
	Plugged tubing.	Locate and repair. Also, filter the mobile phase.
	Pressure transducer needs calibration.	Call your PerkinElmer Service Engineer.
15. Pump will not initialize to the active method.	Start up diagnostics failure.	Switch the pump off, then on. If the problem still persists, call your PerkinElmer Service Engineer.
16. Unable to prime the pump.	Solvent inlet filter is plugged.	Clean or replace the solvent inlet filter.
	Inlet filter is not submerged in the reservoir.	Submerge the inlet solvent filter in the reservoir (s).
	Kinked solvent inlet tubing.	Repair or replace the solvent inlet tubing.
	Proportioning valve is not working properly.	Call your PerkinElmer Service Engineer.

PROBLEM	PROBABLE CAUSE	SOLUTION
17. Cannot stop sparging the mobile phase	Sparging valve is not turned off.	Pull the valve up to stop sparging.
	Leak in the sparging lines.	Locate and correct the leak or call your PerkinElmer Service Engineer.
	No helium.	Check the source of helium. Turn on the helium.
	Control valves closed.	Open the control valves.
	High viscosity solvent.	Use the 40 μ sparger.

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About the Micro Pump System 9

The Micro Pump System and this Chapter

The Micro Pump System includes two 30- μ L based Series 200 isocratic pumps which are used in a high-pressure blending mode. The pumps are controlled by the (Macintosh) computer of the PerkinElmer Sciex mass spectrometer.

The differences between the standard 100- μ L based pump system and the 30- μ L based system, besides the way that they are controlled, include the micro pump tubing, piston, associated seals, etc. The flow-rate range of the micro pump system is from 1 μ L/min to 3 mL/min.

Since operation (setting flows, solvent conditions, etc.) of the micro pumps is accomplished by the mass spectrometer computer, this chapter does not contain operating procedures for the system. It does, however, contain a paragraph which describes the proper choice of pumps and mixer configurations. It also contains an exploded (hardware) drawing of the stainless steel micro pump (Figure 9-3), a system plumbing diagram (Figure 9-2) and an electrical connection diagram (Figure 9-1). Part number changes in the stainless steel micro pump maintenance procedures are included in Table 9-4.

NOTE: There is no 30- μ L based titanium pump.

Proper Choice of Pump and Mixer Configurations

The following table provides information about the Series 200 pump family; it provides a guide for properly choosing your pump. The choice depends on the flow rate and the gradient delay required for your column.

Table 9-1. Gradient Flow Rate, Range and Column Flows for the Series 200 Pumps

Pump	Minimum Flows for Gradient Analysis	Chromatography
Series 200 Isocratic	no gradient	Isocratic, narrow bore,
Series 200 Binary	900 $\mu\text{L}/\text{min}$	Low-pressure blending
Series 200 Quaternary	900 $\mu\text{L}/\text{min}$	Low-pressure blending
Series 200 Narrow Bore Kit	500 $\mu\text{L}/\text{min}$	Low-pressure blending
Series 200 Micro Pumps	50 $\mu\text{L}/\text{min}$	High-pressure blending

High-pressure mixing pumps require dynamic or static mixing for proper gradient formation. Generally, for best results, the internal volume of the mixing chamber should be approximately equal to the flow rate. Refer to Table 9-2 for available mixers, their gradient flow-rate range, and their type of chromatography. Refer to Table 9-3 for the mixer configurations for the Series 200 pumps.

Table 9-2. Available Mixers and their Uses

Gradient Flow Rate Range (μL/min)	Mixer Type	Chromatography
40 - 70	*ASI 50 μL (Static)	micro bore
50 (ballistic)	Lee Vistotec 10 μL (Static)	micro bore
150 - 300	Lee 250, ASI 250 (Static)	analytical
300 - 500	ASI 350 μL (Static)	analytical
500 - 1100	ASI 250, or 350 plus 500 μL	analytical

* ASI mixers are cartridges requiring holder hardware.

Table 9-3. Recommended Mixer Configurations for the Series 200 Micro Pumps

Gradient Flow Rate Range (μL/min)	Chromatography Type	Static Mixer	P-E PartNumber
40 - 70	Micro Bore Ballistic Gradient	Lee 10 μL	N260-2341
50	Micro Bore Gradient	ASI 50 μL	N300-0050
100 -250	Narrow Bore	Lee 250 or ASI 250	N260-2342 and N300-0250
275 - 500	Narrow Bore (2.4 and 2.6 to 3.9 mm Columns)	ASI 350	N300-1350
500 - 1100	Analytical	Lee or ASI 250 μL plus ASI 150 μL	N260-2306 or N300-0250 plus N300-0150

Pump System Maintenance Changes

Only the part number differences between the 30- μ L and 100- μ L pump system maintenance procedures are included in this chapter; the maintenance (and troubleshooting) procedures themselves are basically the same for both systems.

Refer to Chapter 7 of the manual and note the changes to the parts (and part numbers) that have been included in Table 9-4, below:

Table 9-4. 30- μ L System Parts and Standard 100- μ L System Parts for the Series 200 Pumps

Item	Page in Pump Manual	30-μL System Part (see Fig 9-3)	100-μL System Part (Fig 7-22)
Low-pressure piston seal	7-21 & 7-39	0992-3366	0990-7330
Low-pressure piston guide	7-21 & 7-39	N291-5006	0254-2313
Low-pressure piston	7-21	N291-0510	N260-0104
Low-pressure seal collar	7-21 & 7-39	N291-5010	N260-1011
High-pressure seal collar	7-21 & 7-39	N291-5008	0254-2105
High-pressure piston seal	7-21 & 7-39	0992-3367	0990-7324
High-pressure piston seal backup ring	7-21 & 7-39	(part of) 0992-3367	0254-2076
High-pressure piston guide bushing	7-39	N291-5009	0254-2065
Low-pressure piston assy	7-21	N291-0512	N260-0117
High-pressure piston	7-21 & 7-39	N291-0511	N260-0124
Pump Head	7-21 & 7-39	N291-5011	0254-2103

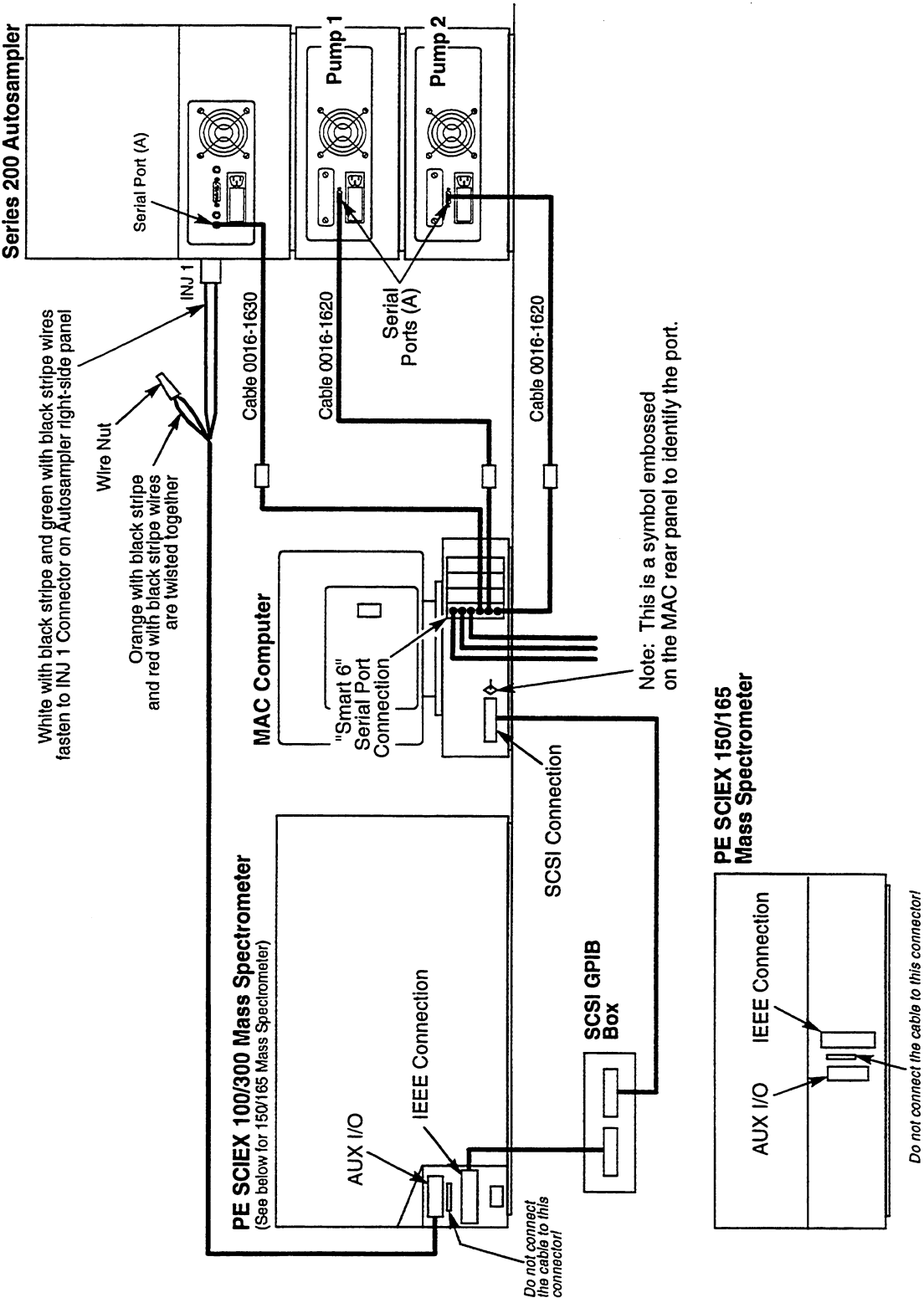


Figure 9-1. Micro Pump System Connection Diagram.

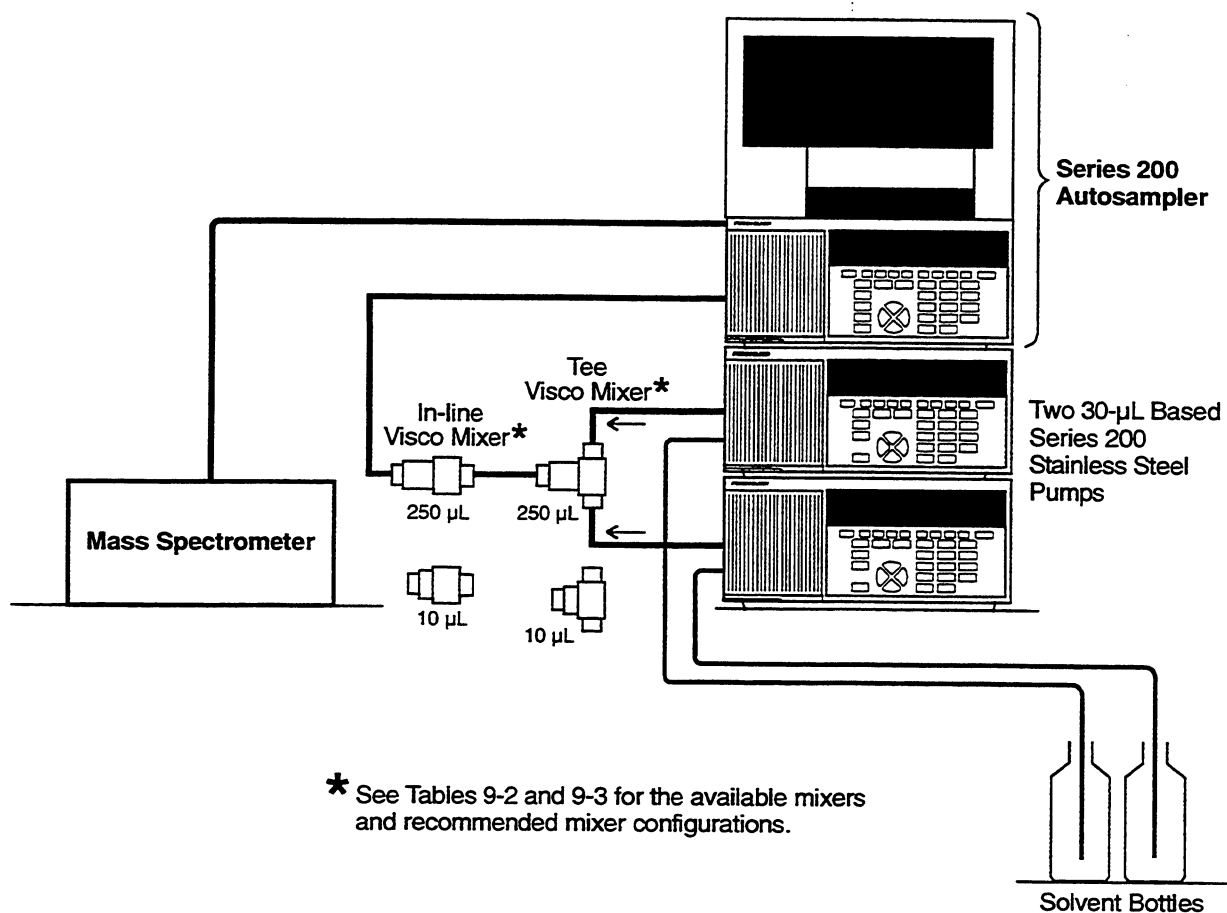


Figure 9-2. Micro Pump System Plumbing Connections.

Appendix

Appendix A

LC Solvent Polarity and Property Scale

Solvent	Adsorptive Energy	Boiling Point °C	Viscosity	Refractive index	UV Cutoff/nm
*Hexane	0.00	69	0.33	1.375	210**
*Isohexane	0.01	99	0.30	1.404	210**
Petroleum ether, skellysolveB, etc.	0.01	175-240	0.30		210
n-Decane	0.04	174	0.92	1.412	
Cyclohexane	0.04	81	1.00	1.427	210
Cyclopentane	0.05	49	0.47	1.406	210**
Carbon tetrachloride	0.18	77	0.97	1.466	265
Amyl chloride	0.26	108	0.43	1.413	225
*Butyl chloride	0.26	77	0.47	1.436	220
Xylene	0.26	138-144	0.62-0.81	1.500	290
*i-Propyl ether	0.28	69	0.37	1.368	220
Toluene	0.29	111	0.59	1.496	285
Chlorobenzene	0.30	132	0.80	1.525	330
Benzene	0.32	80	0.65	1.501	280
*Chloroform	0.40	61	0.57	1.443	245
Methyl-i-butylketone	0.43	117		1.394	330
*Tetrahydrofuran	0.45	65	0.35	1.408	220
Ethylene dichloride	0.49	84	0.79	1.445	230
Methylethylketone	0.51	80	0.30	1.381	330
Acetone	0.56	57	0.32	1.359	330
*Dioxane	0.56	101	1.54	1.422	220
Ethyl acetate	0.58	77	0.45	1.370	260
*Amyl alcohol	0.61	138	4.10	1.410	210

Solvent	Adsorptive Energy	Boiling Point °C	Viscosity	Refractive index	UV Cutoff/nm
Dimethyl sulfoxide	0.62	189	2.24		
Nitromethane	0.64	101	0.67	1.394	380
*Acetonitrile	0.65	82	0.37	1.344	210**
Pyridine	0.71	115	0.94	1.510	305
*i-propanol, n-propanol	0.82	83	2.30	1.380	210
Ethanol	0.88	79	120	1.361	210
*Methanol	0.95	65	0.65	1.329	210
Ethylene glycol	1.11	198	19.90	1.427	210
Acetic Acid	Large	118	1.26	1.372	
*Water	Larger	100	1.00	1.333	190**
Salts and buffers	Very Large	100+			
* Most commonly used with a UV detector.					
** Extremely pure grades may be used as low as 195 nm.					
The Hildebrand solvent scale is a list of common solvents used in Liquid Chromatography in order of increasing energy of adsorption on alumina. The values are different, but the order is essentially the same on silica gel as on alumina.					

Table A-1. List of Common Chromatographic Solvents in Order of Increasing Polarity with RI Values (from "Principles of Adsorption Chromatography," I. R. Snyder, Dekker.)

The starting solvent selected for a given separation can be chosen by matching the relative polarity of the solvent to that of the sample. This is done as a first approximation by selecting the solvent to match the most polar functional group on the sample molecule (for example, alcohols for OH, amines for NH₂, etc.). From this attempt, the separation can be refined by the following procedure:

1. If the sample appears at the solvent front, the solvent is too polar to allow the adsorbent to retard the sample. Change to a solvent that is higher up (lower polarity) on the scale.
2. Conversely, if the sample does not appear in a reasonable time, change to a solvent or solvent blend that is lower down (higher polarity) on the scale.

Solvent blends are most useful when they are composed of pairs which differ in the adsorption energy by no more than 100%. For example, propanol at energy 0.82 and chloroform at energy 0.40.

Solvent gradients should be generated on the same basis of no more than doubling the energy of the starting solvent. The range is more than sufficient for most, if not all, LC separations.

When changing solvents or when regenerating after a gradient, a minimum of five column volumes of the new starting solvent should flow through the system before making another sample injection.

About Solvent Miscibility

Normally, solvents are completely miscible if they are on the same half of the scale. In other words, all solvents on the upper half of the scale are completely miscible, all solvents on the bottom half are miscible with each other, and all solvents from the center half (from 1/4 to 3/4 down the table) are mutually miscible. There are several "universal" solvents, such as, tetrahydrofuran and acetonitrile which are miscible with almost all other solvents except hexane and pentane. By far, the most useful "rinsing or cleanout" solvent is isopropanol, which is miscible with all others at all concentration levels.

Ultraviolet Absorption Bands of Common Chromophores

Chromophore	System	λ_{Max}	ϵ_{Max}	λ_{Max}	ϵ_{Max}	λ_{Max}	ϵ_{Max}
Ether	-O-	185	1 000				
Thioether	-S-	194	4 600	215	1 600		
Amine	-NH ₂	195	2 800				
Thiol	-SH	195	1 400				
Disulfide	-S-S-	194	5 500	255	400		
Bromide	-Br	208	300				
Iodine	-I	260	400				
Nitrile	-C \equiv N	160	-				
Acetylide	-C \equiv C-	175-180	6 000				
Sulfone	-SO ₂	180	-				
Oxime	-NOH	190	5 000				
Azido	>C=N-	190	5 000				
Ethylene	-C=C-	190	8 000				
Ketone	>C=O	195	1 000	270-285	15-30		
Thioketone	>C=S	205	strong				
Esters	-COOR	205	50				
Aldehyde	-CHO	210	1 500				
Carboxyl	-COOH	200-210	50-70				
Sulfoxide	>S \rightarrow O	210	1 500				
Nitro	-NO ₂	210	strong				
Nitrite	-ONO-	220-230	1 000-2 000	300-400	10		
Azo	-N=N-	285-400	3-25				
Nitroso	-N=O	302	100				
Nitrate	-ONO ₂	270 (shoulder)	12				
	(acyclic)	210-230	21 000				
	-(C=C) ₃ -	260	35 000				
	-(C=C) ₄ -	300	52 000				
	-(C=C) ₅ -	330	118 000				

Chromophore	System	λ_{Max}	ϵ_{Max}	λ_{Max}	ϵ_{Max}	λ_{Max}	ϵ_{Max}
Nitrate	$-(\text{C}=\text{C})_2-$ (alicyclic)	230-260	3 000-8 000				
	$\text{C}=\text{C}-\text{C}\equiv\text{C}$	291	6 500				
	$\text{C}=\text{C}-\text{C}=\text{N}$	220	23 000				
	$\text{C}=\text{C}-\text{C}=\text{O}$	210-250	10 000-20 000			300-350	weak
	$\text{C}=\text{C}-\text{NO}_2$	229	9 500				
Benzene		184	46 700	202	6 900	255	170
Diphenyl				246	20 000		
Naphthalene		220	112 000	275	5 600	312	175
Anthracene		252	199 000	375	7 900		
Pyridine		174	80 000	195	6 000	251	1 700
Quinoline		227	37 000	270	3 600	314	2 750
Isoquinoline		218	80 000	266	4 000	317	3 500

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