

# AS Autosampler Operator's Manual

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# **F** • Reordering Information

# 1 • Introduction

## 1.1 Overview

The Dionex AS Autosampler precisely delivers from 1 to  $8,000 \ \mu L$  of sample to an injection valve. The injection volume range delivered by a particular AS depends on the size of the sample syringe and the sampling mode.

#### Modes

Several sampling modes are available:

- In normal mode, sample is delivered to a single system and a sample loop is installed on the injection valve.
- In concentrate mode, the AS delivers sample to a Dionex ultra-low pressure concentrator column.
- In simultaneous mode, the AS can deliver sample to two independent ion chromatography (IC) systems. Sample is delivered equally to the two IC systems (50% of the volume to each). This mode requires the simultaneous injection option.
- In sequential mode, a diverter valve allows sample to be delivered to two independent channels or IC systems through sequential injections. This mode requires the sequential injection option.
- In sequential concentrate mode, a diverter valve allows sample to be delivered to two independent channels or IC systems on which Dionex ultra-low pressure concentrator columns are installed.

#### **Sample Temperature Control**

The sample temperature control option provides uniform heating and cooling of the vial tray.

#### **Sample Preparation**

The sample preparation option is used for automated dilutions and pipet functions.

#### **Injection Valve Options**

The AS can be configured either without an injection valve, or with one or two injection valves installed in the autosampler compartment.

#### Vials, Well Plates, and Sample Trays

The AS can sample from either vials or well plates. Several types of sample trays are available to accommodate the various vial and well plate sizes. Insulated trays are available for use with the sample temperature control option.

During sampling, the tray remains stationary and a sampling needle arm moves from position to position. Up to 99 injections can be taken from each vial or well.

#### Control

The AS can be controlled locally, from the front panel keypad and display, or remotely, with a personal computer running Windows® 2000 or Windows XP and Chromeleon® Chromatography Data Management System software (Release 6.7 or later), or Chromeleon Xpress (Release 1.0 or later). Limited remote control is also available, using TTL and relay signals.

For communication between the AS and the software, the AS is connected to a USB (Universal Serial Bus) port on the computer, USB hub, or other Dionex module equipped with a USB hub.

## 1.2 About This Manual

Chapter 1 Introduction	Introduces the AS; explains the conventions used in this manual, including safety-related information.
Chapter 2 Description	Describes the AS operating features and system components; provides the theory of operation.
Chapter 3 Operation and Maintenance	Provides operating instructions for the AS and describes routine preventive maintenance procedures.
Chapter 4 Troubleshooting	Lists problems and presents step-by-step procedures for how to isolate and eliminate the cause of each problem.
Chapter 5 Service	Provides step-by-step instructions for routine service and parts replacement procedures that the user can perform.
Appendix A Specifications	Lists the AS specifications and installation site requirements.
Appendix B Installation	Describes how to install the AS.
Appendix C Display Screens	Describes the front panel operational and diagnostic display screens.
Appendix D Front Panel Control	Provides step-by-step instructions for operating the AS from the front panel.
Appendix E TTL and Relay Control	Describes the AS TTL and relay control features.
Appendix F Reordering Information	Lists spare parts for the AS.

NOTE For details about using Chromeleon or Chromeleon Xpress to operate the AS, refer to the Chromeleon or Chromeleon Xpress Help. If you are using Chromeleon Xpress, also refer to the Chromeleon Xpress Quick Reference Guide (P/N 065070), provided on the Dionex Reference Library CD-ROM (P/N 053891).

### 1.2.1 Safety Messages and Notes

This manual contains warnings and precautionary statements that can prevent personal injury and/or damage to the AS when properly followed. Safety messages appear in bold type and are accompanied by icons, as shown below.



Indicates an imminently hazardous situation which, if not avoided, will result in death or serious injury.



Indicates a potentially hazardous situation which, if not avoided, could result in death or serious injury.



Indicates a potentially hazardous situation which, if not avoided, may result in minor or moderate injury. Also used to identify a situation or practice that may seriously damage the instrument, but will not cause injury.

IMPORTANT

Indicates that the function or process of the instrument may be impaired. Operation does not constitute a hazard.

Messages d'avertissement en français



Signale une situation de danger immédiat qui, si elle n'est pas évitée, entraînera des blessures graves à mortelles.



Signale une situation de danger potentiel qui, si elle n'est pas évitée, pourrait entraîner des blessures graves à mortelles.



Signale une situation de danger potentiel qui, si elle n'est pas évitée, pourrait entraîner des blessures mineures à modérées. Également utilisé pour signaler une situation ou une pratique qui pourrait gravement endommager l'instrument mais qui n'entraînera pas de blessures.

#### Warnhinweise in Deutsch



Bedeutet unmittelbare Gefahr. Mißachtung kann zum Tod oder schwerwiegenden Verletzungen führen.



Bedeutet eine mögliche Gefährdung. Mißachtung kann zum Tod oder schwerwiegenden Verletzungen führen.



Bedeutet eine mögliche Gefährdung. Mißachtung kann zu kleineren oder mittelschweren Verletzungen führen. Wird auch verwendet, wenn eine Situation zu schweren Schäden am Gerät führen kann, jedoch keine Verletzungsgefahr besteht.

#### Notes

Informational messages also appear throughout this manual. These are labeled NOTE and are in bold type:

**NOTE NOTES** call attention to certain information. They alert you to an unexpected result of an action, suggest how to optimize instrument performance, etc.

## 1.3 Safety and Regulatory Information

The AS is designed for IC (ion chromatography) and HPLC (high-performance liquid chromatography) applications and should not be used for any other purpose. Operation of an AS in a manner not specified by Dionex may result in personal injury.

### 1.3.1 Safety Labels

The TUV GS and cTUVus Mark safety labels and the CE Mark label on the AS indicate that the AS is in compliance with the following standards: EN 61010-1:2001 (safety), CAN/CSA-C22.2 No. 1010.1-92 + A2:97 (safety), UL 61010C-1:2002 R8.02 (safety), and EN 61326:1997, including A1:1998 and A2:2001 (EMC susceptibility and immunity).

The symbols below appear on the AS or on AS labels.

$\sim$	Alternating current
	Protective conductor terminal (earth ground)
	Power supply is on
$\bigcirc$	Power supply is off
•	Indicates a potential bazard. Refer to the operator's i

Indicates a potential hazard. Refer to the operator's manual for an explanation of the hazard and how to proceed.

# 2 • Description

## 2.1 Overview of Features

Figure 2-1 illustrates the main features of the AS Autosampler.



Figure 2-1. AS Operating Features

#### Front Panel

The front panel contains a liquid crystal display (LCD) and membrane keypad (see <u>Section 2.2</u>). The front panel permits manual control of AS functions.

#### **Power Switch**

The power switch is in the lower left corner of the AS. The switch functions only when the door is fully closed.

#### Autosampler Compartment

The autosampler compartment contains a stationary sample tray and an XYZ sampling needle arm that moves from vial to vial during operation (see <u>Section 2.3</u>). The compartment also contains an inject port, flush port, and waste port. Valves for sample injection or for diverting sample flow to different systems can optionally be included.

#### Syringe and Reservoir Organizer

The organizer on top of the autosampler holds the sample syringe, the flush liquid reservoir, and the optional prep syringe and reagent reservoirs. See <u>Section 2.3.1</u> for details.

#### Options

- **Sample Preparation**: When this option is installed, the AS can perform sample preparation functions such as diluting standards or dispensing reagents. See <u>Section 2.3.1</u> for details.
- **Sample Temperature Control** (not pictured): When this option is installed, the AS provides uniform heating and cooling of the sample tray. See <u>Section 2.3.7</u> for details.
- **Simultaneous Injection** (not pictured): When this option is installed, the AS can deliver sample to two independent ion chromatography (IC) systems simultaneously. See <u>Section 3.6.9</u> for details.
- **Sequential Injection** (not pictured): When this option is installed, the AS can deliver sample to two independent ion chromatography (IC) systems alternately.
- **Concentrator Injection** (not pictured): When this option is installed, the AS can load sample to a Dionex ultra-low pressure concentrator column. See <u>Section 2.3.10</u> for details.

#### Valve Options

One or two injection valves can be installed in the AS autosampler compartment. Or, the AS can be configured without an injection valve and instead be connected to one or two valves installed in other chromatography system modules. Two injection valves are required for simultaneous injections.

For the sequential injection option, a diverter valve is installed in the AS.

# 2.2 Front Panel Features



Figure 2-2. AS Front Panel

Information is displayed on the LCD, or *screen*. To adjust the screen contrast, use the knurled knob in the recess below the keypad (see Figure 2-2).

From the keypad, you can:

- Directly control AS operation
- Create and modify programmed series of operating steps, called *methods*
- Create and modify programmed series of injections, called *schedules*

Refer to Section 2.2.2 for a summary of how to select screens and edit parameters.

Button	Function		
Stop	<ul> <li>Stops a schedule if the AS is in Local mode, Schedule control (see <u>Section 2.5.1</u>). A confirmation message appears; press <b>Stop</b> again to abort the schedule or <b>Hold/Run</b> to resume.</li> <li>If the AS is in Direct Control (see <u>Section 2.5.1</u>), pressing <b>Stop</b> stops the current syringe and/or sampling arm movement.</li> <li><b>Note:</b> Pressing <b>Stop</b> while the AS is in Direct Control, Locked Remote mode (software control) (see <u>Section 2.5.2</u>) is not recommended. Use the controls on the software Control panel (see <u>Section 2.5.2</u>) to stop a sequence.</li> </ul>		
Home	Sends the needle arm to its home position. This button functions only when the AS is in Local mode. If a schedule is in progress, it must be on hold. If you press <b>Home</b> when a schedule is on hold, and then resume the schedule, the needle arm returns to the location it was in when you pressed <b>Home</b> .		
Insert	Inserts a new step into a method or schedule. Move the cursor to the leftmost field on the <b>SAMPLE PREP</b> , <b>TIMED EVENTS</b> , or <b>SCHEDULE</b> screen and press <b>Insert</b> . The new step is added above the cursor position. See <u>Appendix D</u> for details about front panel operation.		
	<b>Delete</b> performs several functions:		
Delete	• Cancels an entry that is in progress and restores the previous value.		
	• Returns a field to its default value (if an entry is not in progress).		
	• Deletes a line in a method or schedule. To do this, position the cursor in the leftmost field of the line that you want to delete and press <b>Delete</b> twice.		
	• Deletes an entire method or schedule. To do this, position the cursor in the method or schedule edit field, or in the time field of the <b>INIT</b> step, and press <b>Delete</b> twice.		
Hold/Run) (Hold)	At power-up, the AS is on hold (the left LED is illuminated). After you select a schedule to run, pressing <b>Hold/Run</b> starts the schedule (the right LED is illuminated). When the schedule is complete, the AS returns to hold. When running, press <b>Hold/Run</b> to pause the schedule. This button functions only when the AS is in Local mode.		
(Run)	<b>Note:</b> If the wait operation is enabled (see <u>Section 2.7.1</u> ), the AS pauses when it reaches the wait step in the method. Press <b>Hold/Run</b> to		

continue. You can disable the wait (see Section 2.7.4).

## 2.2.1 Keypad Button Functions



### 2.2.2 Display Screens

The LCD displays status information and allows access to all AS operations.

When the power is turned on, the AS performs an initialization sequence (see Section 3.1.1) and then displays the MAIN STATUS screen. Figure 2-3 is an example of the MAIN STATUS screen during AS operation.



Figure 2-3. Main Status Screen

NOTE TRAY temperature is displayed only when the sample temperature control option is installed.

To access other AS screens:

Press the Menu button. The MENU of SCREENS appears (see Figure 2-4).

MENU of SCREENS					
1	MAIN STATUS	6	FLUSH/PRIME		
2	DETAIL STATUS	7	TIME FUNCTION IN		
3	METHOD MENU	8	DIAGNOSTIC MENU		
4	SCHEDULE	9	TIME FUNCTION OUT		
5	MODULE SETUP MENU				
Help Prompt					

Figure 2-4. Menu of Screens

#### To select a screen on the menu, use one of the following methods:

- Press the keypad numeric button corresponding to the screen's number on the menu.
- Move the cursor to highlight the screen number and press Enter.

#### To display a brief description of each screen:

Press the **Help** button. See <u>Appendix C</u> for details about each screen.

#### To edit a field on the screen:

- 1. Press a cursor arrow button to position the cursor in the field to be edited. Fields that are in reverse video (blue letters on white background) can be edited. Other fields display information only.
- 2. If the field accepts numeric values, press the desired numeric buttons to enter the value.

If the field has predetermined options, press the **Select**  $\Delta$  or **Select**  $\nabla$  button to choose the desired option.

3. To confirm the selected value, press **Enter** or move the cursor out of the field by pressing an arrow button.

## 2.3 Autosampler Compartment



Figure 2-5. Autosampler Compartment Features

### 2.3.1 Syringe and Reservoir Organizer

The organizer on top of the autosampler holds the sample syringe, 1 L flush reservoir, and the optional prep syringe and reagent reservoirs (see Figure 2-5).

#### **Sample Syringe Functions**

The sample syringe performs these operations:

- Pipettes liquid from one vial to another or to waste.
- Mixes a vial by repeatedly drawing and expelling the vial contents. If the AS is equipped with the sample preparation option, mixing is done with the sample prep syringe instead of the sample syringe.
- Delivers sample to the injection valve.

The following sample syringe volumes are available:

Sample Syringe Volume	Part Number
100 µL	055064
250 µL (standard)	053916
500 μL	055065
1,000 µL	055066
5 mL*	053915
10 mL*	055068

\*A 5 or 10 mL syringe is required for simultaneous injections and optional for all other sampling modes. If a 5 or 10 mL syringe is installed, an 8.5 mL sampling needle assembly (P/N 061267) is required.

#### **Sample Preparation Option Functions**

The AS can be equipped with a sample preparation option that includes a sample prep syringe, a sample prep valve, and one 250 mL reagent reservoir. Up to three additional reagent reservoirs can be configured.

The sample prep syringe performs these operations:

- Dispenses reagents from external reservoirs to any vial
- Mixes a vial by repeatedly drawing and expelling the vial contents
- Performs dilutions, allowing preparation of multilevel calibration standards
- Performs liquid-liquid extractions by adjusting the height of the needle in the vial

Prep Syringe Volume	Part Number
250 μL	053916
500 μL	055065
1,000 µL	055066
2.5 mL	055067
5 mL (standard)	053915
10 mL	055068

The following sample prep syringe volumes are available:

The reagent reservoir connects to port A on the sample prep valve. Additional reservoirs can be connected to ports B, C, and D. Commands from the AS front panel screens or from Chromeleon or Chromeleon Xpress let you select the reservoirs.

- NOTE The 5 mL and 10 mL syringes are intended for most sample prep functions. Use the smaller syringes only for applications that require low flow rates.
- NOTE The 2.5 mL syringe (P/N 055067) can only be used with the sample preparation option. It cannot be used as a sample syringe.

### 2.3.2 Sampling Needle Arm

#### IMPORTANT

The autosampler door must remain closed during operation. If you open the door during operation, the sampling arm stops immediately. If a schedule is running, it is ended. To safely open the door during a schedule, press the Door button. See <u>Section 3.9</u> for details.

During operation, the sampling needle arm moves the sample needle along X-, Y-, and Z-axes. The sample tray remains stationary throughout the sampling process.

To sample from a vial or well plate, the arm moves to the specified position in the tray and lowers the needle into the vial or well. The sampling needle pierces the vial cap or well plate cover to allow fluid to be drawn or delivered. When sampling is complete, the arm raises the needle out of the vial or well. When running a schedule of injections, vials or wells can be sampled in any order.

The needle height (the distance from the tip of the needle to the bottom of the vial or well) can be adjusted, allowing operations such as liquid-liquid extraction. You can set the needle height in a Chromeleon or Chromeleon Xpress program (see Section 3.2.2) or in a front panel program (see page 292).

NOTE Changing the needle height is not recommended when sampling from a micro-well well plate, because the well depth is shallow. You can adjust the needle height in a deep-well well plate as required, provided the needle remains submerged in sample. This ensures accurate sample aspiration.

During a flush operation, the sampling needle arm delivers flush fluid to the flush port (see Figure 2-7).

### 2.3.3 Sampling Needle Assembly

The sampling needle assembly consists of a PEEK<sup>TM</sup> sampling needle and a line, which connects the sample syringe and the sampling needle (see Figure 2-6).



Figure 2-6. Sample Syringe with Sampling Needle Assembly

Two assemblies are available (see the table below).

Sampling Needle Assembly	Used With	Part Number
PEEK Needle with 1.2 mL line	Sample syringe volumes 1 mL or less	054271
PEEK Needle with 8.5 mL line <sup>a</sup>	Sample syringe volumes greater than 1 mL	061267

a. The 8.5 mL sampling needle assembly is standard when the AS is configured with the simultaneous injection option.

### 2.3.4 Flush, Waste, and Inject Ports

The flush port flushes the outside of the sampling needle with fluid from the flush reservoir. Excess fluid flows to the waste port. The waste port accepts waste fluid from the sampling needle and the flush port, and directs it out the drain line. The inject port (see Figure 2-7) accepts the sample from the sampling needle and delivers it to the injection valve.



Figure 2-7. Flush, Waste, and Inject Ports

### 2.3.5 Sample Vials and Well Plates

#### Vial Trays and Vials

The following vial trays are available. See the following section for well plate information.

Tray Material	Vial Size Held	Tray Capacity	Part Number
Plastic	10 mL	49	062374
Aluminum	0.3 mL, 1.5 mL	100	062481
Insulated Aluminum <sup>a</sup>	0.3 mL, 1.5 mL	100	062377

a. Required for the sample temperature control option. Includes tray covers for empty tray rows.

The following vials are available:

Vial Size and Material	Quantity	Part Number
0.3 mL polymer	Package of 100	055428
1.5 mL glass	Package of 100	055427
10 mL PolyVial™	Package of 100	055058
1.5 mL plastic	Package of 100	061696

#### IMPORTANT

Dionex does not recommend using the 10 mL plastic tray with the sample temperature control option because the plastic tray is inefficient at heating and cooling the vials. In addition, the tray is not designed for temperatures above 40 °C (104 °F) and may deform at temperatures above 40 °C.

#### Well Plates and Trays

Dionex recommends using well plates in the AS if you have limited amounts of sample or if you require more than 100 sample positions. Follow these guidelines when using well plates:

- 2 mL deep-well plates are suitable for ion chromatography (IC) applications, where concentration of ions is above 1 ppm.
- 0.5 mL and 0.45 mL micro-well plates are not recommended for IC applications.
- Where concentration of ions is below 1 ppm, Dionex recommends using 10 mL PolyVials (P/N 055058, package of 100).

Well Plate Tray Type	Includes	Part Number
Micro-well plate tray	One micro-well plate tray, two micro-well plates with 0.5 mL round (U-shaped) wells, and two covers <i>(not suitable for IC applications)</i>	066324
Deep-well plate tray	One deep-well plate tray, two deep-well plates with 2 mL wells, and two covers <i>(suitable for</i> <i>IC applications, where concentration of ions is</i> <i>above 1 ppm)</i>	066325

The following well plate tray packages are available:

Additional well plates and covers can be ordered separately. See Appendix F for reordering information.

#### Well Plate Tray Notes

- All well plate trays can be used with the sample temperature control option.
- Each well plate tray also holds ten, 1.5 mL vials.
- The micro-well plate tray can hold either type of micro-well plate (V-shaped wells or U-shaped wells).

• The AS automatically detects the type of tray installed.

### 2.3.6 Injection Valve Options

The AS can optionally have one or two injection valves installed inside the autosampler compartment. The injection valves are six-port, electrically-activated Rheodyne valves. The valves have two operating positions: Load and Inject (see <u>Figure 2-8</u>).



Figure 2-8. Injection Valve Flow Schematics

Eluent flows through either the Load or Inject path, depending on the valve position:

- In the Load position, sample flows from the AS inject port line, through the valve, and into the sample loop, where it is held until injection. Eluent flows from the pump, through the valve, and to the column, bypassing the sample loop. Excess sample flows out to waste.
- In the Inject position, sample is swept to the column for analysis. Eluent flows from the pump, through the sample loop, and on to the column, carrying the contents of the sample loop with it.

## 2.3.7 Sample Temperature Control Option

The sample temperature control option provides heating and cooling of the sample tray. You can program the temperature set point from 4 °C to 60 °C. Samples in the tray are uniformly heated or cooled to the programmed set point (to a minimum of 20 °C below ambient and a maximum of 40 °C above ambient). The absolute range is 4 °C to 60 °C.

The sample temperature control option sits on the workbench below the autosampler compartment (see Figure 2-9).



Figure 2-9. AS with Sample Temperature Control Option

**NOTE** The temperature inside the autosampler compartment may exceed ambient by up to 2 °C. The sample temperature control option is recommended for samples that are temperature sensitive.

### 2.3.8 Simultaneous Injection Option

When the AS simultaneous injection option is installed, a single AS can deliver sample to two independent chromatography systems. The sample is injected simultaneously to both systems (two injection valves are required). Dual analyses can be performed with only one sample, thus increasing sample throughput and eliminating the need to label, fill, and track two sample vials.

In Chromeleon or Chromeleon Xpress, the two chromatography systems and the AS are configured into a single timebase and each system is assigned a unique device name and channel. This lets you monitor and control both systems from one Control panel and run all samples in one sequence.

The simultaneous injection option requires a 5 or 10 mL sample syringe and an 8.5 mL sampling needle assembly (see <u>Section 2.3.3</u>). Full-loop injections are required with this option.

See Section 3.6.9 for details about simultaneous injections.

#### 2.3.9 Sequential Injection Option

When the simultaneous option is installed, a single AS can deliver sample to two independent chromatography systems. With the sequential option, sample is delivered to the first system, flow is rerouted (diverted), and sample is then delivered to the second system. Sequential injection allows different samples to be injected into each system.

The sequential injection option requires two injection valves and a diverter valve (for switching sample flow between the two systems). The diverter valve is installed in the AS autosampler compartment. The injection valves must be installed in the IC system's chromatography compartment.

See <u>Section 3.6.11</u> for details about sequential injections.

NOTE The diverter valve can only be controlled by Chromeleon or Chromeleon Xpress. It is not controllable from the AS front panel.

## 2.3.10 Concentrator Injection Option

The AS concentrator injection option lets an AS deliver sample and reagent to a Dionex ultra-low pressure concentrator column. With this option, a concentrator column is installed on the injection valve instead of a sample loop. A 5 or 10 mL sample syringe can be installed to allow larger sample loading volumes. If the 5 or 10 mL sample syringe is installed, an 8.5 mL sampling needle assembly must also be installed (see Section 2.3.3).

The AS is capable of delivering at a maximum pressure of 690 kPa (100 psi). When setting up concentrator injections, make sure to select syringe speeds that will keep the backpressure below 690 kPa (100 psi), taking into account the pressure from the installed tubing and the concentrator column. A syringe speed of 1 or 2 is required. See <u>Section 3.8</u> for details.

See Section 3.6.10 for details about concentrator injections.
## 2.4 Rear Panel Features



Figure 2-10. AS Rear Panel

#### 2.4.1 USB Receptacle

The USB receptacle provides a connection to the Chromeleon or Chromeleon Xpress computer. To enable computer control of the autosampler, connect a USB cable between the USB receptacle on the AS rear panel and a USB port on the computer or USB hub. See <u>Section B.3.8</u> for USB connection instructions.

## 2.4.2 TTL/Relay Connectors

The eight TTL and relay connectors interface with Dionex and non-Dionex modules. The input connectors allow for relay and TTL control of the autosampler by a connected module(s). The output connectors allow the AS to control other module(s). <u>Appendix E</u> describes the relay and TTL functions and the connections between the AS and other modules.

#### 2.4.3 Fuse Holder and Power Receptacle

The fuse holder contains two 3.15 amp fast-blow fuses (P/N 954745). For instructions on how to change the fuses, see Section 5.18.

The power cord plugs into the IEC 320 three-prong receptacle.



The power supply cord is used as the main disconnect device. Make sure the socket-outlet is located near the AS and is easily accessible.



Le cordon d'alimentation principal est utilisé comme dispositif principal de débranchement. Veillez à ce que la prise de base soit située/installée près du module et facilement accessible.

Das Netzkabel ist das wichtigste Mittel zur Stromunterbrechung. Stellen Sie sicher, daß sich die Steckdose nahe am Gerät befindet und leicht zugänglich ist.

## 2.5 Autosampler Control

The AS can be controlled locally, from the front panel keypad and display (see <u>Section 2.5.1</u>), or remotely, with a personal computer on which Chromeleon or Chromeleon Xpress software is installed (see <u>Section 2.5.2</u>). TTL input signals from an integrator or other remote controller can also be used to control the AS. See <u>Appendix E</u> for details about TTL control.

#### 2.5.1 Local Control

In Local mode, the AS receives commands from the front panel buttons and screens. When the AS is powered up, it is always in Local mode.

When the AS is in Local mode, two types of control are available: Direct and Schedule.

- In Direct control, you enter control commands and parameters on the AS screens, and the AS executes the commands immediately.
- In Schedule control, you define a list of injections (a schedule) to be run in sequence. The schedule can be defined, saved, and then run at a later time.

The table below summarizes the AS Local control configurations:

<b>Operating/Control Mode</b>	Autosampler Operation
Local/Direct Control	Commands are entered from the AS front panel and executed immediately after being entered.
Local/Schedule Control	Commands are entered from the AS front panel and executed by running a schedule programmed from the front panel.

#### Local/Direct Control Mode

You can directly control the following AS functions from the front panel:

- TTL and relay outputs
- Injection valve position
- Second injection valve position or diverter valve (if installed)
- Tray set point (if installed)

DETAIL	IDLE			
VIAL#:	TIME:	min	TTL1	1
INJ#:	VOL:	uL	TTL2	0
METHOD:	TRAY: 2	mL	RLY1	0
<b>TRAY: 25</b>	20 °C↑ LOOP:10	0 uL	RLY2	0
	NORMAL	FULL	INJ VLV	
LOCAL	DIRECT CON	TROL		
Help Prompt				

Set these functions from the **DETAIL STATUS** screen (see Figure 2-11).

Figure 2-11. Detail Status Screen Example

#### Local/Schedule Control Mode

The AS runs all injections according to a programmed schedule. The schedule defines a series of vials to be sampled and the operating parameters to run on each vial. A schedule consists of the following parameters:

- The vials to sample
- The number of injections to draw from each vial
- The volume of each injection
- The method to run on each injection
- The action to take when a scheduled vial is missing

You program, save, and edit schedules from the **SCHEDULE** screen (see <u>Figure 2-12</u>). The AS can store up to nine schedules in memory. The stored schedules are retained in memory when the AS is turned off. <u>Section D.6</u> describes how to create schedules.

SCHEDULE	EDIT	SAVE TO	RUN	
MISSING VIAI	ACTION:	STOP		
VIA	L# I	NJ/ INJ		
LINE STAR	Γ- <u>END</u> \	IAL VOL	uL) METHOD	
1 1	- 1	1 25.	0 1	
Help Prompt				



Each injection in a schedule is assigned a *method* (a series of operating instructions that tells the AS how to perform a single injection). A method consists of the following three phases:

Method Phase	Used To	Examples
Sample prep	Prepare the sample for injection	Pipetting, mixing, diluting
Method setup	Set parameters that remain constant throughout the method	Sample needle height, sample temperature
Timed events	Perform functions at a specific time in the method	Loading the sample loop, injecting

When the method runs, the AS performs the sample prep steps first, followed by the method setup, and then the timed events steps. Section D.5 describes how to create methods.

#### Notes

- See <u>Appendix D</u> for details about front panel operation.
- Some autosampler operations are not available from the AS front panel and can only be accessed from Chromeleon or Chromeleon Xpress. These include setting different aspirate and dispense syringe speeds and sampling from well plates.

### 2.5.2 Computer Control (Locked Remote Mode)

In Locked Remote mode, Chromeleon or Chromeleon Xpress software sends commands from the host computer via the USB interface. Connecting the AS to a Control panel (see Figure 2-13) immediately selects the Locked Remote mode. In this mode, you cannot change any operating parameters from the front panel keyboard. To return the AS to Local mode, clear the **Connect** check box on the AS Control panel.

😵 Chromeleon - [Panel Tabset1]				
	🔚 File Edit View Workspace Qualification Control Batch Window Help 💶 🗗 🛪			
E	》 ☆ ۞ ◙ ☷☶扇團屬屬幽二 ◎ 第 ■ ■ ■ ♀ ◈ ♦ = ■ ● ₹ 4 ♥ ◙ ☞ & ◎ ∰ ■ ■ ↓ ℃ (			
	ICS-3000_2 😒 👭			
	Home   Sequence Control   Status Autosamplet   Gradient Pumps   Detector Compartment   EC Detector   3D Amp Plot			
	System Log (Audit Trail)			
	2:08:10 PM 0.011 (Sampler) Start preparing next sample Sample 2 (position = 2, volume = 25.0).			
	2.08:40 PM 0.500 (Pump_1) Log Pressure: 0.0 (psi)     10.2.08:40 PM 0.500 (Pump_1) Log Pressure: 0.0 (psi)			
	▶ 2.000 Pump_2_Pressure Acq0ff			
	Status			
	Connect Status: Idle			
	Exclusive Access: ICS-3000_2			
	Mode: Normal			
	Injection Type: Full Inject Valve: LoadPosition (43) (44) (45) (46) (47) (48) (49)			
	Loop Size: 20.0 µl			
	Temperature Control Wellness (29 (30 (31 (32 (33 (34 (35)			
	Tray Set Temp 10°C - Needle Cycles 1738			
	T 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1			
	Syringe			
	Prime Flush			
	Prime Syringe: Sample 🛨 Flush Syringe: Sample			
	Prime Volume: 150 µl 🛨 Flush Volume: 100 µl 🛨			
	Prime speed: aspirate 3 - Flush speed: aspirate 3 -			
	dispense 3 🗸 dispense 3 🗸			
	Prime reservoir: Reservoir_A 🚍 More Syringe Commands			

Figure 2-13. Example AS Control Panel

There are two modes for controlling the AS using Chromeleon or Chromeleon Xpress:

• In Direct mode, you select operating parameters and commands directly from the Control panel. The AS executes the commands immediately.

• In Automatic mode, you create a list of samples (a sequence) to be processed automatically. The sequence defines a series of vials to be sampled and the AS operating parameters to run on each vial. The sequence is similar to a front panel schedule (see <u>Section 2.5.1</u>),

For instructions on how to connect the AS to Chromeleon or Chromeleon Xpress, see <u>Section 3.2.1</u>. For an overview of how to operate the autosampler using Chromeleon or Chromeleon Xpress, see <u>Section 3.2.2</u>. For detailed instructions, refer to the Chromeleon Help or Chromeleon Xpress.

## 2.6 System Wellness and Predictive Performance

The Chromeleon System Wellness panels provide easy access to the autosampler's built-in diagnostic and calibration features. You can use the Wellness panels to perform the following tasks:

- Calibrate the inject port tubing volume
- Align the sampling needle in the inject port
- Calibrate and test the leak sensor
- Monitor usage of replaceable parts (see the description of Predictive Performance below)

Wellness panels for calibrating the inject port tubing, aligning the sampling needle, and monitoring usage of parts are available on the Control panel (see Figure 2-13). A separate panel is available for calibrating and testing the leak sensor (see Section 5.11).

# NOTE You can also perform System Wellness tasks from the front panel screens. See <u>Appendix D</u> for details.

The Chromeleon Predictive Performance feature provides functions for monitoring the lifetime of replaceable parts and for planning and recording service procedures. You can use the Chromeleon Predictive Performance feature to perform the following tasks:

• Monitor how many times a part has been used since it was last replaced. Chromeleon monitors usage of the following AS parts: injection valve, diverter valve, XYZ sampling needle arm, sampling needle assembly, needle seal assembly, sample syringe, and prep syringe.

- Display a warning message in the Chromeleon Audit Trail when a monitored part is approaching its usage limit and when the limit is reached.
- Set the usage limits for monitored parts to suit your needs.
- Record when a part is serviced and reset the usage counter.

All Predictive Performance commands and parameters are available in the Chromeleon Commands dialog box (press the **F8** key or select **Command** on the **Control** menu to open the Commands dialog box).

You can view and reset the usage counters for the various replaceable parts on either the AS Wellness panel (see <u>Figure 2-14</u>) or the **USAGE LOG SCREEN** (see <u>Section C.9.6</u>). The remaining Predictive Performance functions, described above, are available only in Chromeleon.

🚟 AS Wellness Panel			
Needle Cycles 4,200	Reset		
XYZ Arm Cycles 10,400	Reset		
AS Inject Valve Cycles 2,100	Reset		
Diverter Valve Cycles1,000	Reset		
Sample Syringe Cycles 6,200	Reset		
Prep Syringe Cycles 6,200	Reset		
=> Warning. Consider performing maintenance soon			
=> Limit reached. Perform maintenance			

Figure 2-14. Example AS Wellness Panel (Default Warning Values for All Parameters Shown)

## 2.7 Theory of Operation

#### 2.7.1 Understanding the Wait Operation

NOTE This section pertains to AS operation in Local/Schedule Control mode (see <u>Section 2.5.1</u>) only.

The wait operation is a step in the sample prep phase of a method. After performing all other sample prep steps, the AS waits for a continue command before performing the method's timed events. The continue command can be sent via the front panel **Hold/Run** button or a TTL input signal.

The wait's position in the sample prep steps determines whether samples are overlapped or run sequentially:

- When the wait operation is the last step in the sample prep phase (the default), samples are overlapped (see Section 2.7.2).
- When the wait operation is at the beginning of the sample prep phase, samples are run sequentially (see <u>Section 2.7.3</u>).

NOTE The wait operation can be disabled (see <u>Section 2.7.4</u>).

#### 2.7.2 Overlapping Sample Preparation

When the wait operation is at the end of the sample prep phase (the default), the AS overlaps sample preparation. This means that while data collection is occurring for one injection, the AS performs the sample prep steps for the next injection (see Figure 2-15). For a detailed description of the schedule run flow of operations, see Section 2.7.6.



Figure 2-15. Sample Prep Overlap: Wait Operation at End of Sample Prep Phase (Default)

#### Sample Overlap with Chromeleon or Chromeleon Xpress

If Chromeleon or Chromeleon Xpress is controlling the AS, front panel settings for the wait operation are ignored. Instead, the **Sample Overlap** option determines whether sample preparation is overlapped. Sample overlap, which is selected in the Chromeleon Server Configuration program, is enabled by default. For instructions on how to configure the AS in Chromeleon, see <u>Section B.3.12</u>.

## 2.7.3 Running Samples Without Overlap

Placing the wait operation at the start of the sample prep steps allows samples to be run sequentially. The AS completes the sample prep and timed events for an injection before starting the next injection (see Figure 2-16).



Figure 2-16. No Sample Prep Overlap: Wait Operation at Beginning of Sample Prep Phase

# 2.7.4 Operating the AS as the System Master (Disabling the Wait Operation)

When the wait operation is disabled, a single run command (for example, pressing the **Hold/Run** button) starts the schedule. Thereafter, the AS performs the schedule lines without requiring additional commands. This allows the AS to act as the *system master* (the AS controls the system by sending TTL signals to start the pump and detector methods).

To disable the wait operation, press **Menu** and **5** to go to the **MODULE SETUP MENU** and press **5** to go to the **SYSTEM PARAMETERS** screen. Set the **WAIT** field to **DISABLED** (see Figure 2-17).

SYSTEM PARAMETERS		
WAIT:	DISABLED	
METHOD SYRINGE SPEED:	4	
CUT SEGMENT VOLUME:	10 uL	
SAMPLE MODE:	NORMAL	
INJECT PORT VOLUME:	43 uL	
FLUSH SYRINGE SPEED:	4	
Help Prompt		

Figure 2-17. System Parameters Screen

For details about the order of events during a schedule when the wait operation is disabled, see <u>Figure 2-20</u>.

## 2.7.5 Understanding the Cycle Time

Cycle time is an optional feature that can be used to control the time between injections. When a method is assigned a cycle time, the AS delays sample injection until the specified time has elapsed since the previous injection. Cycle time is measured from the start of the previous method's timed events (see Figure 2-18). Assuming injection occurs at time zero for each method (the default), specifying a cycle time allows a consistent time interval between injections, regardless of differences in sample prep and timed events among methods.



Figure 2-18. Cycle Time Example

A cycle's duration is never shorter than the specified time. However, cycle time will be ignored, thus delaying injection, if any of the following events occurs:

- WAIT FOR TEMP STABLE is enabled in the METHOD SETUP screen (see <u>Section C.6</u>) and the temperature did not stabilize by the end of the cycle time.
- The sample prep wait operation is enabled and a run command did not occur by the end of the cycle time.
- The previous method's timed events extend beyond the cycle time.

If the cycle time expires before the injection occurs, a warning message is displayed and the message is logged on the **MESSAGE LOG** screen (see <u>Section C.9.6</u>).

### 2.7.6 Operating Events During a Schedule

- <u>Figure 2-19</u> shows the order of events during a schedule when the wait operation is enabled.
- <u>Figure 2-20</u> shows the order of events during a schedule when the wait operation is disabled.

See <u>Section 2.7.1</u> for details about the wait operation.



Figure 2-19. Order of Events During a Schedule Run (Wait Enabled)



Figure 2-20. Order of Events During a Schedule Run (Wait Disabled)

### 2.7.7 Understanding the Status Display During a Run

During runs, the **MAIN STATUS** and **DETAIL STATUS** screens display the operations currently being performed. Because of sample prep overlap, information on multiple injections may be displayed simultaneously at certain points in the schedule. For example, in <u>Figure 2-21</u>, the AS is injecting the sample and performing the timed events for the first injection from vial 1 while preparing for the second injection.



Figure 2-21. Detail Status Screen Example

#### Notes

- The INJ# field is updated when the AS loads the loop for the next injection.
- The **TIME** field and the **METHOD** timed events clock are reset to zero at injection.
- The **TIME** field counts up from the injection until the next injection occurs.
- The **METHOD** timed events clock counts up until all timed events have been performed for an injection. The clock then stops and is no longer displayed.

#### 2.7.8 Operating Events During Flushing and Priming

The AS automatically flushes the inject port and washes the outside of the needle at the following times:

- At the beginning and end of a Chromeleon or Chromeleon Xpress sequence or a front panel schedule
- Before each injection, while the schedule or sequence is running

Flushing can also be initiated manually when the AS is idle, or as a step in the sample prep phase of a Chromeleon or Chromeleon Xpress program or front panel method.

Manual priming of the flush and reagent lines must be initiated after refilling a reservoir, or after the AS has been idle for a period of time. See <u>Section B.3.13</u> for instructions on priming the liquid lines.

#### Flushing

The AS performs the following operations during flushing:

- The sampling needle arm moves to the waste port and expels any fluid from the needle.
- The sampling needle arm moves to the inject port and delivers a volume of fluid for flushing the inject port.
- The sampling needle arm moves to the flush port and delivers a volume of flush fluid. This washes the outside of the needle.

The flush reservoir and sample syringe are always used to perform the flush.

The needle is washed with a factory-set volume. Select the volume of liquid for flushing or priming on the Chromeleon or Chromeleon Xpress Control panel (see Figure 2-13) or on the FLUSH/PRIME screen (see Section C.7). If you specify a flush volume that is greater than the sample syringe volume, the AS performs multiple syringe strokes until the volume is reached.

If you are using Chromeleon or Chromeleon Xpress to control the autosampler, you can specify separate syringe speeds for aspirating and dispensing liquid during flushing or priming. Specify the speeds on the Control panel or in the program (see Section 3.7).

#### Priming

The AS performs the following operations during priming:

- The sampling needle arm moves to the waste port and expels any fluid from the needle.
- The sampling needle arm moves to the flush port and delivers a volume of flush fluid. This primes the lines from the flush reservoir to the syringe and through the sampling needle assembly (see Figure 2-22).

When the sample preparation option is installed, additional priming is required to prime the lines from the reagent reservoir(s) to the sample syringe (see Figure 2-24). See Section B.3.13 for instructions on priming the liquid lines.

## 2.7.9 Sample and Prep Syringe Fluid Schematics

Figure 2-22 shows the fluid connections to the sample syringe and flush reservoir when no sample prep option is installed.



Figure 2-22. Fluid Schematic for Sample Syringe

Figure 2-23 shows the flow paths through the sample syringe valve ports for the two valve positions:

- In the needle position, flow is between ports A–B, C–D, and E–F.
- In the flush position, flow is between ports A–F, B–C, and D–E.



Figure 2-23. Sample Syringe Valve Port Flow Path Schematics

Figure 2-24 shows the fluid connections to the sample syringe, flush reservoir, sample prep syringe, and reagent reservoirs when the sample prep option is installed.



Figure 2-24. Fluid Schematic for Sample Syringe with Optional Prep Syringe

The prep syringe valve has two rows of port fittings. <u>Figure 2-25</u> shows a detailed view of the front and back port connections.



Figure 2-25. Prep Syringe Valve Port Connections

## 3.1 Getting Ready to Run

#### 3.1.1 Turn On the Power

Press the power switch in the lower left corner of the front door to turn on the AS power. When the power is turned on, a series of screens are displayed and the autosampler performs a power-up sequence.

- The **COPYRIGHT NOTICE** screen is displayed briefly, and then the **POWER-UP** screen appears.
- While the **POWER-UP** screen is displayed, the AS performs initialization procedures and a series of diagnostic tests. If one of the tests fails, testing stops and the **DIAGNOSTIC TEST** screen is displayed. See <u>Section C.9.2</u> if this occurs.
- When the initialization procedures are complete and the AS has passed the diagnostics, the **INSTALLED OPTIONS** screen appears. After a few seconds (or if you press a button), the **MAIN** screen is displayed. The power-up sequence is then complete.

After power-up, you can display the **POWER-UP**, **DIAGNOSTIC TEST**, and **INSTALLED OPTIONS** screens by selecting these options on the **MENU of SCREENS**. See (see Section C).

#### 3.1.2 Fill the Vials or Wells and Load the Sample Tray

#### **Filling Vials**

 Fill the vials to the vial shoulder or lower (see <u>Figure 3-1</u>). Do not fill to the top.

> During sampling, make sure the needle's vent is not immersed in liquid.

For 10 mL vials only: If necessary, raise the needle height inside the vial to position the vent above the liquid. You can set the needle height in a Chromeleon or Chromeleon Xpress program (see <u>Section 3.2.2</u>) or in a front panel program (see <u>page 292</u>).



Figure 3-1. Maximum Level to Fill Vials

2. Screw a cap onto each vial and tighten. To prevent septa from falling into the vials when they are pierced by the sampling needle, make sure the cap septa are pushed fully into the caps and the caps are securely tightened.

# NOTE For 0.3 mL and 1.5 ml vials (any type), Dionex recommends using vial caps with slit septa only (see Section 2.3.5).

- 3. Load the vials in the tray in the order required for the Chromeleon or Chromeleon Xpress sequence (see <u>Section 3.2.2</u>) or for the front panel schedule (see <u>Section D.6</u>).
- 4. If the sample temperature control option is installed, position tray covers (P/N 061010) over any empty positions on the tray (see



Figure 3-2). This ensures temperature stability and minimizes condensation, especially at low temperature set points.

Figure 3-2. Installing Tray Covers (Sample Temperature Control Option Only)

- 5. Install the tray in the autosampler compartment.
- 6. Close the autosampler compartment door. Make sure the door remains closed during operation.

#### IMPORTANT

If the door is opened during operation, the sampling arm stops immediately. If a schedule is running, it is aborted. To safely open the door during operation, press the Door button (see Section 3.9 for details).

#### **Filling Well Plates**

- 1. Put on powder-free latex gloves before handling well plates and covers to minimize sample contamination.
- 2. Rinse the well plates and covers thoroughly with deionized water before use to remove any contaminants.
- 3. Dry the well plates completely to prevent any changes in sample concentration. You can use 690 kPa (100 psi) nitrogen gas to assist in drying.
- 4. Use a pipet to fill the wells in the well plate. In general, pipet the amount of sample to be aspirated plus 70  $\mu$ L. See Section 3.6.4 for details about the sample volumes aspirated for each type of injection.

The table below lists the maximum fluid volumes for each well size and the maximum sample volumes available when the well is filled:

Well Plate Type	Maximum Volume per well (µL)	Sample Available (µL) <sup>a</sup>
0.45 mL micro-well with conical (V-shaped) wells	320	250
0.5 mL micro-well with round (U-shaped) wells	320	250
2.0 mL deep-well	1800	1700

a. Assumes the well contains the maximum volume and the needle height is 2 mm (the default).

- NOTE Changing the needle height is not recommended when sampling from a micro-well well plate, because the well depth is shallow. You can adjust the needle height in a deep-well well plate as required, provided the needle remains submerged in sample. This ensures accurate sample aspiration.
- 5. Place a cover over the well plate and firmly press the indentations on the cover into each well. This creates a tight seal and prevents sample evaporation.
  - NOTE Replace covers after each use to prevent contamination and sample evaporation. Well plates can be reused.

- 6. Orient the well plate with well A1 at the front-left corner and set the well plate in the well plate tray (see Figure 3-3).
- 7. Fill another well plate and install it on the right side of the tray. The two plates must be of the same type. You can install either two deepwell plates or two micro-well plates (0.50 mL or 0.45 mL).
- 8. Position the clamp in the well plate tray with the thumbscrews in the center of the tray and the arms across both well plates.

#### IMPORTANT

Always install two well plates in the tray to ensure even pressure from the clamp. Make sure that the arms on the clamp are positioned over the small "lip" on the front and back sides of both well plates. This ensures that the clamp presses down on each well plate evenly and the well plate doesn't move during sampling.



9. Tighten the thumbscrews on the clamp snugly.

Figure 3-3. Well Plate Tray with Two Well Plates Installed

10. Install the tray in the autosampler compartment.

11. Close the autosampler compartment door. Make sure the door remains closed during operation.

#### IMPORTANT

If you open the door during operation, the sampling arm stops immediately. If a schedule is running, it is ended. To safely open the door during operation, press the Door button (see <u>Section 3.9</u> for details).

#### Guidelines for improving precision when sampling from well plates:

- To avoid contaminants, rinse well plates and covers thoroughly with deionized water before use.
- Dry well plates completely to prevent any changes in sample concentration. You can use 690 kPa (100 psi) nitrogen gas to assist in drying.
- Follow the sample volume guidelines (see Step 4) when pipetting sample into the wells.
- Check the orientation of the cover to make sure the notches in the corners of the cover and well plate are aligned.
- Press the well plate cover tightly into each well.
- To avoid contaminants and evaporation, do not reuse covers. The small hole created when the sampling needle pierces the cover allows slight, but noticeable, evaporation of sample. This is especially true when samples must wait for several hours before being run.
- If sample evaporation does occur, try reducing the tray temperature.

## 3.2 Selecting Computer or Front Panel (Local) Control

When the AS is powered up, it is always in front panel (Local) control. The **MAIN** and **DETAIL** screens indicate this by displaying **LOCAL** in the operating mode field. See the example in Figure 3-4.



Figure 3-4. Main Screen—Local Control

To select computer control, follow the steps in <u>Section 3.2.1</u> to connect the AS to a Chromeleon or Chromeleon Xpress Control panel.

When the AS is under computer control, the front panel **MAIN** and **DETAIL** screens indicate this by displaying **LOCKED RMT** and **DIRECT CONTROL** in the operating and control mode fields. See the example in Figure 3-5.



Figure 3-5. Main Screen—Computer Control

To return the AS to Local mode, clear the **Connect** check box on the Chromeleon or Chromeleon Xpress Control panel (see Figure 3-6).

#### 3.2.1 Connecting to Chromeleon or Chromeleon Xpress

- 1. Start the Chromeleon Server, if it is not already running:
  - If the Chromeleon Server icon on the taskbar is crossed out in red **Server** is not running. Start the Server by right-clicking the icon and selecting **Start Server**. When the server is running

(and data acquisition is not occurring), the icon is gray 🗱.

- If the Server Monitor icon is not on the taskbar, click **Start** on the taskbar and select **All Programs** (or **Programs**, depending on the operating system) > **Chromeleon** > **Server Monitor**. Click **Start** to start the server.
- 2. Start the Chromeleon or Chromeleon Xpress client:
  - a. Click Start on the Windows taskbar and select All Programs (or Programs) > Chromeleon > Chromeleon to start the Chromeleon client.
  - b. If Chromeleon is installed, select View > Default Panel Tabset

or click the corresponding toolbar button 🛅 to display the panel tabset.

# NOTE If Chromeleon Xpress is installed, starting the client automatically displays the panel tabset.

c. To display the AS Control panel, select the **Autosampler** tab on the panel tabset (see Figure 3-6).

d. Verify that the **Connect** check box is selected. If it is not, click the box to connect the AS to Chromeleon or Chromeleon Xpress.

ų	📽 Chromeleon - [Panel Tabset1]				
	File Edit View Workspace Qualification Control Batch Window Help _ 🖬	<sup>1</sup> ×			
]	) © 0 1⊞ ⊞ ⊊ 8 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2	A			
	ICS-3000_2 🔀 👭				
	Home Sequence Control Status Autosampler Gradient Pumps Detector Compartment EC Detector 3D Amp Plot				
	System Log (Audit Trail)				
	© 208:10 PM 0.011 (Sampler) Start preparing next sample Sample 2 (position = 2, volume = 25.0).				
	1 2:08:40 PM 0.500 (Pump_1) Log Pressure: 0.0 (psi)				
	▶_:_:_ 2.000 Pump_2 Pressure AcqOff				
	Status				
	Connect Status: Idle				
	Exclusive Access: ICS-3000_2				
	Mode: Normal				
	Injection Type: Full Inject Valve: LoadPosition				
	Loop Size: 20.0 µl				
	Temperature Control Wellness				
	Tray Set Temp: 10 °C 🕂 Needle Cycles 1738 22 23 29 29 29 29 29 29 29 29 29 29 29 29 29				
	Trau Astual Targer 10.00 °C Wellness panel Needle align Calibrate IPTV 65 (8) (7) (8) (9) (2)				
	Syringe				
	Prime Flush				
	Prime Syringe: Sample 🛨 Flush Syringe: Sample				
	Prime Volume: 150 µl 🚊 Flush Volume: 100 µl 🚔				
	Prime speed: aspirate 3 - Flush speed: aspirate 3 -				
	dispense 3 - dispense 3 -				
	Prime reservoir. Reservoir_A 🔄 More Syringe Commands				

Figure 3-6. AS Control Panel

#### 3.2.2 Running a Sequence of Injections in Chromeleon or Chromeleon Xpress

This section provides a brief description of the steps required to set up and run the autosampler with Chromeleon or Chromeleon Xpress. For detailed instructions, refer to the Chromeleon or Chromeleon Xpress Help.

1. Create a program that specifies the functions the autosampler (and other system instruments) should perform at a specific time. Programs can be created automatically (with the help of a software wizard) or manually (by editing an existing program).

In addition to commands for loading and injecting the sample, a program for the AS can include commands for setting up the autosampler (syringe speed, sample needle height, etc.) and for preparing the sample before injection (pipetting, mixing, flushing, etc.).

- 2. Create a *sequence* (a list of injections). For each injection, specify the vial or well position (see Section 3.4), the injection volume, the program to run, and other parameters. Sequences can be created automatically (with the help of a software wizard) or manually (by editing an existing sequence).
- 3. Load the sequence into a batch.
- 4. Start the batch.

## 3.3 Default Operating Parameters

The <u>Table 3-1</u> lists the factory-set values for AS operating parameters:

Operating Screen	Parameter	Default Value
Method Sample Prep	Step	1 wait step only
Method Setup	Cycle Time Sample Needle Height Sample Temperature Wait for Temp Stable	Off 2 mm 20 °C No
Method Timed Events	Time=Init Inject Valve TTL/Relays	Load (for simultaneous or sequential injection, both valves at Load position) All off (0)
	Time=0.00 Inject Valve TTL/Relays	Inject (for simultaneous or sequential injection, both valves at Inject position) All off (0)
Schedule	Missing Vial Action Vial # Start/End Injections per Vial Injection Volume Method Number	Stop 1/1 1 25.0 μL; 1 mL for simultaneous injection 1
System Parameters	Wait Syringe Speeds Cut Volume Sample Mode Inject Port Volume Air Bubble Volume (not user changeable)	Enabled Refer to <u>Section 3.7</u> for Chromeleon control syringe speeds. Refer to <u>Section D.7</u> for front panel control syringe speeds. 10 μL (see <u>Section 3.6.3</u> for details) Normal varies (each inject port is individually calibrated) 8 μL if the sample syringe is <5 mL; 60 μL if a 5 or 10 mL syringe is installed
Flush/Prime	Flush Volume Flush Reservoir Flush Syringe Prime Volume Prime Reservoir Prime Syringe	250 μL Flush (always) Sample (always) 400 μL Flush Sample

Table 3-1. Default	Operating	Parameters
--------------------	-----------	------------

## 3.4 Specifying Sample Positions

When creating a schedule or sequence of injections, use the following guidelines to specify sample positions in the vial or well plate sample tray.

### 3.4.1 Vial Tray Sample Positions

For the 49-position vial tray, vial position 1 is in the lower-left corner of the tray and vial position 49 is in the upper-right corner (see Figure 3-7).



Figure 3-7. 49-Position Vial Tray

For the 100-position vial tray, vial position 1 is in the lower-left corner of the tray and vial position 100 is in the upper-right corner (see Figure 3-8).



Figure 3-8. 100-Position Vial Tray

### 3.4.2 Well Plate Tray Sample Positions

# NOTE Chromeleon or Chromeleon Xpress is required to sample from a well plate.

Sample positions in the well plates are specified with a two letter and one number identifier (for example, LB8):

- The first letter (either L or R), specifies the left or right well plate (see <u>Figure 3-9</u>).
- The second letter (A through H) and the number (1 through 12) specify the well position in the well plate. Well letters and numbers are printed on well plates and well plate covers.

For example, well plate position LA1 identifies the well located at position A1, in the well plate installed on the left side of the AS well plate tray. When the well plate is installed in the tray, position LA1 is in the lower-left corner of the tray (see Figure 3-9).



Figure 3-9. Well Plate Tray: Well Plate and Vial Positions

NOTE To specify the position of a 1.5 mL vial installed in the well plate tray, precede the vial position number with a V (V1, V2,...V10) (see Figure 3-9).

## 3.5 Specifying Injection Volumes

The range of injection volumes the AS can deliver to the injection valve depends on the size of the sample syringe installed on the AS and on the sampling mode selected on the **SYSTEM PARAMETERS** screen (see <u>Section 3.6.2</u>).

Syringe Size(s)	Sampling Mode	Injection Volume
100–1,000 μL	Normal and Sequential	1.0–1,000 μL
	Concentrate and Sequential Concentrate	100–1,000 μL
5 mL	Normal and Sequential	1.0–4,700 μL
	Concentrate and Sequential Concentrate	100–4,700 μL
	Simultaneous	1,000–4,700 μL
10 mL	Normal and Sequential	1.0–8,000 μL
	Concentrate and Sequential Concentrate	100–8,000 μL
	Simultaneous	1,000–8,000 µL

Table 3-3 lists the volume range for each syringe size and sampling mode.

#### Table 3-2. Summary of Injection Volume Ranges

You can enter the injection volume in the Chromeleon or Chromeleon Xpress sequence (see Figure 3-10 and Figure 3-11) or in the front panel schedule (see Section D.3). For volumes less than 100  $\mu$ L, specify the value in 0.1  $\mu$ L

increments.	For volumes of	of 100 µL or	greater,	specify	the value i	in 1 μL
increments.						

Chromeleon Sequence Wizard - Step 1 of 4: Unknown Samples					
Unknown Samples Generate samples from a template or import	them via clipboard or I	from another sequ	uence		
Use template	Sequence Preview	Rack Preview			
Template for Sample Name: Sample #n  Number of Vials:  Injections per Vial:  1 [19]	Name P Sample 1 P Sample 2 P Sample 3	Pos. 1 2 3	Vol. 25.0 25.0 25.0		
Start Position: 1 T [149]					
< Back	Next >	Cancel	Help		

Figure 3-10. Sequence Wizard: Specifying the Injection Volume

Sequence								
No F	<sup>2</sup> o Inj. Vol.	Program	Status	Inj. Date/Time	Name	Туре	Method	Sequence
1	35.0	Test	Finishe	3/14/2007 4:02:0	Sample 1	Unknow	default	ICS-3000 2VASTest
22	25.0	Test	Finishe	3/14/2007 4:04:1	Sample 2	Unknow	default	ICS-3000 2VASTest
2								5
						J		<u> </u>

Figure 3-11. Sequence Control Panel: Specifying the Injection Volume

## 3.6 Sample Injection Types

#### 3.6.1 Overview

The type of injection the AS performs for a particular sample depends on several factors, including the number of systems the AS is connected to and whether a sample loop or concentrator column is installed on the injection valve. Table 3-3 summarizes all of the available injection types.

Injection Type	Description		
Full	Full-loop injection to one system		
Partial	Partial-loop injection to one system		
Partial LS	Partial-loop, limited-sample injection to one system (no extra sample is aspirated)		
Large Loop	Large-loop injection to one system		
Simultaneous	Full-loop injections to two systems simultaneously		
Concentrate	Concentrate injection to one system		
Sequential Full	Full-loop injections to two systems alternately		
Sequential Partial	Partial-loop injections to two systems alternately		
Sequential Partial LS	Partial-loop, limited-sample injections to two systems alternately		
Sequential Large Loop	Large-loop injection to two systems alternately		
Sequential Concentrate	Concentrate injections to two systems alternately		
Table 3-3. St	ummary of Available Injection Types		
During an injection, the injection type is displayed on the Chromeleon or Chromeleon Xpress Control panel (see <u>Figure 3-12</u>) and on the **DETAIL STATUS** screen (see <u>Figure 3-13</u>).

		ICS-3000_2 🔀 👫
Home Sequence Control Status A	utosampler   Gradient Pumps   Dete	ctor Compartment   EC Detector   3D Amp Plot
System Log (Audit Trail)		
11:56:47 AM 0.012 (Sampler) S	Start preparing next sample San	nple 2 (position = 2, volume = 25.0).
11:57:16 AM 0.500 (EDet1) Log	g Background: 366.0 (nA)	
11:57:16 AM 0.500 {Pump_1} L	Log Pressure: 0.0 (psi) Log Pressure: 0.0 (psi)	
:: 2.000 Pump_2_Pres	ssure.AcqOff	<u> </u>
Status		
Connect Status:	ldle	
Exclusive Access: ICS	-3000_2	
	Mode:	Normal
Injection Wpe: Full	Inject Valve:	LoadPosition
Loop Size: 20.0 µl		

Figure 3-12. Injection Type Shown on Control Panel

DETAIL	INJECTING	
<b>VIAL#: 1</b>	TIME: 0.0 min TTL	1 0
INJ#: 1/3	S VOL: 25 uL TTL	2 1
METHOD: 01	TRAY: P10mL RLY	1 0
<b>TRAY: 25</b>	20 °C 1 LOOP: 25,0 uL RLY	2 0
	NORMAL FULL INJ VLV	/ L
LOCAL	SCH 1 LINE 1	
Help prompt		

Figure 3-13. Injection Type Shown on Detail Status Screen

## 3.6.2 Determining the Injection Type

To determine the injection type performed by the AS, first select the sample mode on the **SYSTEM PARAMETERS** screen (see Figure 3-14). Press **Menu**, **5**, and **5** to go to the **SYSTEM PARAMETERS** screen.



Figure 3-14. System Parameters Screen

The sample mode corresponds to the AS configuration. The following sample modes are available:

- Normal—Sample is delivered to a single system; a sample loop is installed on the injection valve.
- **Concentrate**—Sample is delivered to a single system; a concentrator column is installed on the injection valve.
- **Simultaneous**—Sample is delivered to two systems simultaneously; a sample loop is installed on both injection valves.
- **Sequential**—Sample is delivered to two systems alternately; a sample loop is installed on both injection valves.
- Sequential concentrate—Sample is delivered to two systems alternately; a concentrator column is installed on both injection valves.

## NOTE The SAMPLE MODE must be selected on the SYSTEM PARAMETERS screen. It cannot be selected from Chromeleon or Chromeleon Xpress.

If normal or sequential mode is selected, you must also enter additional parameters to specify the type of sample loop injection performed (see <u>Section 3.6.3</u>).

## 3.6.3 Determining the Sample Loop Injection Type

When the AS is in normal or sequential mode, it can perform the following types of sample loop injections:

- Full-loop—A full loop volume is injected.
- Large-loop—A full (large) loop volume is injected. Large loops are 200.0 to 7000.0 μL in volume.
- Partial-loop—A partial loop volume is injected.
- **Partial-loop, limited-sample**—A partial loop volume is injected; no extra sample is aspirated from the vial or well.

## NOTE When the AS is in simultaneous mode, it performs only full-loop injections.

The following three parameters are used to determine the type of sample loop injection the AS performs: cut volume, injection volume, and loop size (see <u>Table 3-4</u>).

Criteria for Determining the Sample Loop Injection Type	Type of Sample Loop Injection Performed
Injection volume $\geq$ loop size (loop size = 2.5 to 199.9 µL)	Full-loop
Injection volume $\geq$ loop size (loop size = 200.0 to 7000.0 µL)	Large-loop
Injection volume < loop size (cut volume > 0)	Partial-loop
Injection volume < loop size (cut volume = 0)	Partial-loop, limited-sample

### Table 3-4. Determining the Sample Loop Injection Type

The *cut volume*, which is used only for partial-loop and partial-loop, limited-sample injections, specifies a volume of sample to be "cut" (or omitted) from each side of the sample during the injection process. Using a smaller cut volume minimizes the amount of total sample aspirated from the vial or well. However, better precision is achieved with a larger cut volume. If you enter a cut volume of 0, the AS draws only the injection volume from the vial or well.

1. Enter the size of the installed loop on the PLUMBING CONFIGURATION screen (see Figure 3-15). Press Menu, 5, and 3 to go to the PLUMBING CONFIGURATION screen.



Figure 3-15. Plumbing Configuration Screen

 Enter the injection volume in the Chromeleon or Chromeleon Xpress sequence (see <u>Figure 3-16</u>) or in the front panel schedule (see <u>Section D.3</u>).

Unknown Samples Generate samples from a template or import	them via clipboard or I	from another sec	juence	
Use template	Sequence Preview	Rack Preview		
	Name	Pos.	Vol.	
Template for Sample Name:	🗑 Sample 1	1	25.0	
Sample #n 💌 🕨	👩 Sample 2	2	25.0	
Number of Vials: 3 [1x]	🗑 Sample 3	3	25.0	
Injections per Vial: 1 [199]				
Start Position: 1 🗧 [149]				
Injection Volume: 25.0 [1.01000.0 µl]				
Apply >>				
< Back	Next>	Cancel	Help	

Figure 3-16. Sequence Wizard: Specifying the Injection Volume

- If you enter an injection volume equal to or larger than the installed loop size, the AS performs a full-loop (or large-loop) injection.
- If you enter an injection volume smaller than the loop size, the AS performs a partial-loop or partial-loop, limited-sample injection.
- 3. Enter the cut volume in the Chromeleon or Chromeleon Xpress program (see Figure 3-17) or on the SYSTEM PARAMETERS screen (see Section C.6.5).
  - NOTE The cut volume is used only for partial-loop or partial-loop, limited-sample injections. For full-loop or large-loop injections, the cut volume is ignored. See <u>page 63</u> for details about cut volumes.

Program Wizard: Sampler (	Options			×
Column Temperature Column Temperature: Tray Temperature: Wait for stable temperat		Diverter Valve F     O Position     O Position	Position 1 2	
Cycle Time: Syringe Speed: Sample Needle Height: Cut Volume: Flush Volume:	0 σ [0240 min] 3 2 σ [046 mm] 0 σ [030 μ] 100 π [1009999 μ]	)		
	< Back	Next >	Cancel	Help

Figure 3-17. Program Wizard: Specifying the Cut Volume

# 3.6.4 Guidelines for Sample Volumes Used, Loop Sizes, and Injection Volumes

## Sample Volumes Used for Each Type of Sample Loop Injection

<u>Table 3-5</u> shows the volume of sample the AS aspirates from the sample vial or well plate well and the volume of sample injected onto the column.

Injection Type	Sample Volume Aspirated from Vial or Well	Sample Volume Injected
Full-loop (2.5–16.9 μL loop)	Injection volume or 2.5 × loop size + 25 $\mu$ L (whichever is greater)	Loop size
Full-loop (17.0–199.9 μL loop)	Injection volume or $4 \times \text{loop size}$ (whichever is greater)	Loop size
Large-loop (200.0–7000.0 μL loop)	Injection volume or $1 \times \text{loop size} + 500 \mu\text{L}$ (whichever is greater)	Loop size
Partial-loop	Injection volume + $2 \times cut$ volume	Injection volume
Partial-loop, limited- sample	Injection volume	Injection volume

### Table 3-5. Sample Volumes Used for Each Injection Type

See <u>Section 3.6.3</u> for descriptions of sample loop injection types and the criteria used to determine which type of injection is performed.

The following tables give several examples for full-loop, large-loop, and partial-loop injections. The examples show the amount of sample used and injected for various loop sizes and injection volumes.

## Full-Loop Sample Volume Examples

For full-loop injections, if the injection volume is more than 4 times the loop size, the full injection volume is aspirated. The excess aspirated sample is flushed through the loop, but the amount injected is equal to the loop size.

Installed Loop Size (μL)	Injection Volume from Chromeleon Sequence (μL)	Sample Aspirated from Vial or Well (μL) <sup>a</sup>	Sample Volume Injected onto Column (μL)
10	10	40	10
10	35	40	10
10	60	60	10
25	25	100	25
25	75	100	25
25	150	150	25

#### Table 3-6. Full-Loop Sample Volume Examples

a. Excess aspirated sample is used to flush the loop before and after injection.

### Large-Loop Sample Volume Examples

For large-loop injections, if the injection volume exceeds the loop size + 500 µl, the full injection volume is aspirated. The excess sample aspirated is flushed through the loop, but the amount actually injected is equal to the loop size.

Installed Loop Size (μL)	Injection Volume from Chromeleon Sequence (µL)	Sample Aspirated from Vial or Well (μL) <sup>a</sup>	Sample Volume Injected onto Column (μL)
500	500	1,000	500
500	700	1,000	500
500	1,100	1,100	500

### Table 3-7. Large-Loop Sample Volume Examples

a. Excess aspirated sample is used to flush the loop before and after injection.

### Partial-Loop and Partial-Loop, Limited-Sample Volume Examples

For partial-loop injections, the amount of sample aspirated is equal to the injection volume + 2 times the cut volume. The amount injected is equal to the injection volume.

For partial-loop, limited-sample injections, the amount of sample aspirated and injected is equal to the injection volume.

Installed Loop Size (μL)	Injection Volume from Chromeleon Sequence (μL)	Cut Volume (µL) <sup>a</sup>	Sample Aspirated from Vial or Well (μL)	Sample Volume Injected onto Column (μL)
25	10	5	20	10
25	20	10	40	20
25	10	0	10	10
25	20	0	20	20

 Table 3-8. Partial-Loop and Partial-Loop, Limited-Sample

 Sample Volume Examples

a. The cut volume specifies a volume of sample to be "cut" (or omitted) from each side of the sample during the injection process. See Section 3.6.3 for details.

### Maximum Sample Loop Sizes and Injection Volumes

The maximum loop size allowed depends on which sample syringe and sampling needle assembly are installed. If the sample loop is too large, the sample volume drawn exceeds the syringe/sampling needle assembly volume, causing sample to enter the flush bottle. To prevent sample from contaminating the flush bottle, the sample loop size must not exceed the values shown in Table 3-9.

<u>Table 3-9</u> also lists the maximum injection volume that can be specified in the Chromeleon or Chromeleon Xpress sequence or in the front panel schedule.

Sample Syringe	Sampling Needle Assembly	Maximum Sample Loop Size	Maximum Injection Volume
≤1,000 µL	1.2 mL	500 μL <sup>a</sup>	1,100 µL
5 mL	8.5 mL	4,000 μL <sup>b</sup>	4,700 μL
10 mL	8.5 mL	7,000 μL <sup>b</sup>	8,000 μL

Table 3-9. Maximum Sample Loop Sizes and Injection Volumes

- a. Requires a 1.5 mL vial or a 2 mL deep well plate.
- b. Requires a 10 mL vial.

### Notes

- To minimize the backpressure applied to the sample syringe pump, use the largest ID tubing possible to make the injection loop.
- If a 1.5 mL vial is used, the maximum loop size is 500  $\mu$ L and the maximum injection volume is 1100  $\mu$ L, regardless of the syringe size.
- For syringes that are 1,000 µL or smaller, the maximum loop size and injection volume requires the syringe to aspirate from the sample vial multiple times. This can result in reduced precision. For maximum precision, use a loop size and injection volume that require only a single sample aspiration.

## 3.6.5 Normal, Full-Loop Injections

For a normal, full-loop injection, the AS draws a volume of sample from the sample vial or well plate well. The volume of sample drawn depends on the loop size and the specified injection volume (see Section 3.6.4 for details). A portion of the sample is flushed through the loop, the loop is filled, and an additional portion remains before the loop. The sample in the loop is then injected onto the column (see Figure 3-18).



Figure 3-18. Full-Loop Injection Sequence

## Setting Up a Full-Loop Injection

- 1. On the SYSTEM PARAMETERS screen (see <u>Section C.6.5</u>), select NORMAL in the SAMPLE MODE field.
- 2. In the Chromeleon or Chromeleon Xpress sequence (see Figure 3-19) or in the front panel schedule (see Section D.3), enter an injection volume equal to or greater than the volume of the loop installed on the injection valve.

Chromeleon Sequence Wizard - Step 1 of 4: U	omeleon Sequence Wizard - Step 1 of 4: Unknown Samples					
Unknown Samples Generate samples from a template or import	them via clipboard or fro Sequence Preview	m another sequ	uence			
	Name	Pos	Vol	-		
Template for Sample Name:	Sample 1	1	25.0			
Sample #n 🔹 🕨	🗿 Sample 2	2	25.0			
Number of Vials: 3 [1x] Injections per Vial: 1 [199] Start Position: 1 ➡ [149] Injection Volume: 25.0 [1.01000.0 µ]	🖗 Sample 3	3	25.0			
Apply >>						

Figure 3-19. Sequence Wizard: Specifying the Injection Volume

### **Full-Loop Injection Notes**

- Entering an injection volume equal to or greater than the sample loop size sets up the AS for a full-loop injection. The actual volume of sample injected equals the volume of the sample loop installed.
- If the entered injection volume is more than four times the loop size, the entire injection volume is delivered during loading of the loop. For example, if a 100  $\mu$ L loop is installed and the injection volume is 600  $\mu$ L, then 350  $\mu$ L is flushed through the loop, 100  $\mu$ L is loaded into the loop, and 150  $\mu$ L remains before the loop.

## **Example Program for Normal Mode**

The following example Chromeleon or Chromeleon Xpress program shows the AS commands for sample loading and injecting when the autosampler is in normal mode (full-loop, partial-loop, or partial-loop, limited sample).

0.000	Load		;Switch the inject valve to Load and load sample onto the loop
	Wait	CycleTimeState	;Wait for the end of the specified Cycle Time
	Inject		;Switch the inject valve to Inject and sweep the sample onto the column
	Wait	InjectState	;Wait for the injection to finish

## 3.6.6 Normal, Large-Loop Injections

The injection sequence for a normal, large-loop injection is the same as described for a normal, full-loop injection (see Section 3.6.5). However, when a large loop (200.0 to 7000.0  $\mu$ L) is installed, the AS draws a set amount (500  $\mu$ L) of extra sample from the vial for flushing the loop, instead of drawing a factor of the loop volume (see Section 3.6.4 for details).

## 3.6.7 Normal, Partial-Loop Injections

For a partial-loop injection, the AS draws the volume of sample to be injected from the sample vial plus two times the *cut volume*. The cut volume specifies a volume of sample to be "cut" (or omitted) from each side of the sample during the injection process. See <u>Section 3.6.3</u> for details. The middle portion of the sample is positioned in the loop and injected (see <u>Figure 3-20</u>).



Figure 3-20. Partial-Loop Injection Sequence

## Setting Up a Partial-Loop Injection

- 1. On the SYSTEM PARAMETERS screen (see <u>Section C.6.5</u>), select NORMAL in the SAMPLE MODE field.
- 2. In the Chromeleon or Chromeleon Xpress sequence (see Figure 3-19) or in the front panel schedule (see Section D.3), enter an injection volume that is less than the volume of the sample loop installed on the injection valve.
- 3. In the Chromeleon or Chromeleon Xpress program (see Figure 3-21), or on the SYSTEM PARAMETERS screen, enter a cut volume greater than zero.

Program Wizard: Sampler (	Dptions
Temperature Column Temperature: Tray Temperature: Wait for stable tempera	
Cycle Time: Syringe Speed: Sample Needle Height:	0 (0240 min) 3 (15) 2 (046 mm)
Cut Volume: Flush Volume:	20 ∓ [030 µ]
nan dan dina sadi sadi dan sa san sa	

Figure 3-21. Program Wizard: Specifying the Cut Volume

**NOTE** For the best accuracy when performing partial-loop injections, specify an injection volume equal to half the loop size or less.

#### **Example Partial-Loop Injection Parameters:**

Sample loop size:  $100 \ \mu L$ Injection volume:  $50 \ \mu L$ Cut volume:  $20 \ \mu L$ 

To perform the injection, the AS aspirates 90  $\mu$ L from the sample vial (the inject volume plus double the cut volume) and delivers it to the injection valve. Then, enough flush fluid is drawn into the syringe to position the sample. One cut volume (20  $\mu$ L) flows to waste and the next 50  $\mu$ L is loaded into the loop. At injection, 50  $\mu$ L is injected. After injection, the remaining cut volume (20  $\mu$ L) flows to waste.

## 3.6.8 Normal, Partial-Loop, Limited-Sample Injections

For a partial-loop, limited-sample injection, the AS draws only the volume to be injected from the sample vial. The AS positions the sample in the loop and injects it onto the column (see Figure 3-22).



Figure 3-22. Partial-Loop, Limited-Sample Injection Sequence

## Setting Up a Partial-Loop, Limited-Sample Injection

- 1. On the SYSTEM PARAMETERS screen (see <u>Section C.6.5</u>), select NORMAL in the SAMPLE MODE field.
- 2. In the Chromeleon or Chromeleon Xpress sequence (see Figure 3-19) or in the front panel schedule (see Section D.3), enter an injection volume that is less than the volume of the sample loop installed on the injection valve.
- 3. In the Chromeleon or Chromeleon Xpress program (see Figure 3-21), or on the SYSTEM PARAMETERS screen, enter a cut volume of zero.

## Example Partial-Loop, Limited-Sample Injection Parameters:

Sample loop size: 100  $\mu$ L Injection volume: 50  $\mu$ L Cut volume: 0  $\mu$ L

To perform the injection, the AS aspirates 50  $\mu$ L from the sample vial (the inject volume) and then enough flush fluid is drawn into the syringe to position the sample. The 50  $\mu$ L of sample is positioned just before the loop and is then loaded onto the loop and injected.

## 3.6.9 Simultaneous Injections

The AS must be in simultaneous mode to perform a simultaneous injection (see <u>Section 3.6.2</u>). In simultaneous mode, the AS delivers equal volumes of sample to two injection valves (see <u>Figure 3-23</u>). Each injection is a full-loop size.



Figure 3-23. Simultaneous Injection Sequence

## Setting Up a Simultaneous Injection

The AS must be equipped with the simultaneous injection option in order to perform simultaneous injections. The following setup steps are required:

- Follow the instructions in <u>Section B.3.5</u> to connect the AS to two injection valves (either in the autosampler compartment or two IC systems). In this configuration, the inject port tubing is connected to a Y-connector and the legs of the Y are connected to the inject valves in both systems (see <u>Figure 3-23</u>). The tubing from the Y to each valve must be the same length.
- Install a sample syringe of 5,000 or 10,000 μL.
- Enter the sample syringe size of 5,000 or 10,000 μL on the PLUMBING CONFIGURATION screen (see Section C.6.3).
- On the SYSTEM PARAMETERS screen (see <u>Section C.6.5</u>), select SIMULTANEOUS in the SAMPLE MODE field.
- In the Chromeleon or Chromeleon Xpress sequence (see Figure 3-19) or in the front panel schedule (see Section D.3), enter an injection volume that is large enough to allow flushing the inject port, flushing the tubing from the inject port to each valve (25 µL per inch of tubing), plus enough to flush and fill both injection loops (this is about 5 times the combined loop sizes).

### **Example Simultaneous Injection Parameters:**

45  $\mu$ L = inject port volume

500  $\mu$ L = 20 inches of connector tubing (combined from both systems) x 25  $\mu$ L

 $625 \ \mu L = 5 \ x \ (100 \ \mu L \ loop \#1 + 25 \ \mu L \ loop \#2)$ 

total = 1170  $\mu$ L

The specified injection volume is loaded into the sample syringe and then delivered equally to the two sample loops.

## **Example Program for Simultaneous Mode**

The following example Chromeleon or Chromeleon Xpress programs show the AS commands for sample loading and injecting when the autosampler is in simultaneous mode. Commands differ slightly, depending on where the valves are installed.

## **Example Simultaneous Injection Program: Two Valves Installed in the AS**

0.000	Sampler	_InjectValve.LoadPosition	;Switch the first inject valve to Load
	Sampler	_InjectValve_2.LoadPosition	;Switch the second inject valve to Load
	Load		;Load sample onto the loop
	Wait	CycleTimeState	;Wait for the end of the specified Cycle Time
	Sampler	_InjectValve.InjectPosition	;Switch the first inject valve to Inject
	Sampler	_InjectValve_2.InjectPosition	;Switch the second inject valve to Inject
	Inject		;Sweep the sample onto the column
	Wait	InjectState	;Wait for the injection to finish
	Example an ICS-3	e Simultaneous Injection Program: Tw 6000 Detector/Chromatography Modu	o Valves Installed in le
0.000	InjectV	alve_1.LoadPosition	;Switch the first inject valve to Load
	InjectV	alve_2.LoadPosition	;Switch the second inject valve to Load
	Load		;Load sample onto the loop
	Wait	CycleTimeState	;Wait for the end of the

specified Cycle Time

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<pre>InjectValve_1.InjectPosition</pre>		;Switch the first inject valve to Inject
InjectValve_2.InjectPosition		;Switch the second inject valve to Inject
Inject		;Sweep the sample onto the column
Wait	InjectState	;Wait for the injection to finish

## Example Simultaneous Injection Program: Two Valves Installed in an ICS-1000/1500/2000

0.000	Pump_Inj	ectValve.LoadPosition	;Switch the first inject valve to Load
	Pump_Inj	ectValve_2.LoadPosition	;Switch the second inject valve to Load
	Load		;Load sample onto the loop
	Wait	CycleTimeState	;Wait for the end of the specified Cycle Time
	Pump_Inj	ectValve.InjectPosition	;Switch the first inject valve to Inject
	Pump_Inj	ectValve_2.InjectPosition	;Switch the second inject valve to Inject
	Inject		;Sweep the sample onto the column
	Wait	InjectState	;Wait for the injection to finish

## 3.6.10 Concentrate Injections

The AS must be in concentrate mode to perform a concentrate injection (see Section 3.6.2). In concentrate mode, the AS draws the volume to be injected from the sample vial and delivers it to a concentrator column (see Figure 3-24). The example in Figure 3-24 also shows the reagent prime and reagent flush steps for matrix elimination.



Figure 3-24. Concentrate Injection Sequence (Example for Matrix Elimination)

## Setting Up a Concentrate Mode Injection

- 1. Install a concentrator column on the injection valve.
- 2. If the sample syringe volume is greater than 1 mL, install an 8.5 mL sampling needle assembly (P/N 061267). See Section 5.4 for installation instructions.
- 3. On the SYSTEM PARAMETERS screen, select CONCENTRATE in the SAMPLE MODE field.
- 4. For front panel control, select **METHOD SYRINGE** and **FLUSH SYRINGE** speeds on the **SYSTEM PARAMETERS** screen.
- 5. For Chromeleon or Chromeleon Xpress control, enter the syringe speed in the Program Wizard. 1 is the slowest speed and 5 is the fastest. You can enter separate aspirate and dispense speeds for the Concentrate, Reagent Flush, and Reagent Prime functions. In general, use a faster speed for aspirating and a slower speed for dispensing fluid to the concentrator. See <u>Section 3.7</u> for details about selecting syringe speeds.

## NOTE If a 5 or 10 mL syringe is installed, the dispense speed is limited to speed 1 or 2 only.

Figure 3-25 shows the Program Wizard Sampler Options tab page with the Concentrate function selected.

Program Wizard: Sampler Options	×
E Function Parameters Eunction:	<u>E</u> nter
LoadPosition Aspirate = 3 Dispense = 1     Concentrate	
⊻alve Position LoadPosition	, <u>D</u> elete
Associate Speed	
3 <b>1</b> 5	]
Dispense Speed	1
< <u>B</u> ack <u>Next</u> Car	cel Help

Figure 3-25. Program Wizard Sampler Options for Concentrate

- NOTE Be sure to select syringe speeds that will keep the backpressure below 690 kPa (100 psi), taking into account the pressure from the tubing and the concentrator column. See <u>Section 3.8</u> for details.
- 6. In the Chromeleon or Chromeleon Xpress sequence (see Figure 3-19) or in the front panel schedule (see Section D.3), enter an injection volume equal to the amount of sample to be loaded onto the concentrator.

#### Example Program for Concentrate Mode (Matrix Elimination)

The following example Chromeleon or Chromeleon Xpress program shows the AS commands when the autosampler is in concentrate mode. Commands similar to these could be used for a matrix elimination application.

```
Flush
                               Volume = 100
                                                      ;AS options commands
        Wait
                               FlushState
        NeedleHeight =
                                2
        CutSegmentVolume =
                               0
        SyringeSpeed =
                                3
        CycleTime =
                                0
        Pipet Volume = 20.0, SourceVial =
                                                      ;AS sample prep
        CurrentVial, DestinationVial =
                                                      commands
        CurrentVial
        Mix SourceVial = CurrentVial,
        NumberOfTimes = 5, Volume = 250.0
        FlushSP Volume = 250.0
        DelaySP Time = 0.0
        Dispense Volume = 1000.0,
        SourceReservoir = Reservoir A,
        DestinationVial = CurrentVial
        WaitForTemperature = False
        Concentrate ValvePosition =
                                                      ;Switch the inject valve
        LoadPosition, AspirateSpeed = 3,
                                                      to Load and load sample
        DispenseSpeed = 1
                                                      onto the concentrator
        ReagentPrime Volume = 10000.0,
                                                      ;Prime sampling needle
        SourceVial = Reservoir A, ValvePosition
                                                      assembly with reagent
        = NoChange
        ReagentFlush Volume = 5000.0, SourceVial
                                                      ;Flush concentrator with
        = Reservoir A, ValvePosition = NoChange
                                                      reagent; this is the
                                                      matrix elimination step
        Wait
                  SampleReady
0.000
        Wait
                  CycleTimeState
```

Sampler InjectValve.InjectPosition

;Switch the inject valve to Inject and sweep the sample onto the column

5.000 BeginOverlap

;Start preparing the next sample in the sequence (see note below)

30.000 End

NOTE If the AS is in concentrate mode, sample preparation overlap is available only if the autosampler is controlled by Chromeleon or Chromeleon Xpress. If the AS is under front panel control, sample overlap is not available.

## 3.6.11 Sequential Injections

The AS must be in sequential mode to perform sequential injections (see <u>Section 3.6.2</u>). In sequential mode, the AS delivers sample to two separate chromatography systems. Each system is operated independently and can have a different sample and sample volume injected.

- **NOTE** Precision for partial-loop injections may be slightly affected by the longer tubing length from the inject port to the injection valve.
- NOTE Figure 3-26 shows an injection to inject valve #1. When the diverter valve is switched to position 2, the injection sequence is identical, except that flow is diverted to inject valve #2.



Figure 3-26. Sequential, Full-Loop Injection Sequence

NOTE Partial-loop and partial-loop, limited-sample injections can also be performed in sequential mode. The sequences of events for these injection types are the same as for normal mode, except for selection of the diverter valve position.

## Setting Up a Sequential Mode Injection

The AS must be equipped with the sequential injection option (P/N 063294) in order to perform sequential injections. The following setup steps are required:

- 1. Follow the instructions in <u>Section B.3.6</u> to install the sequential injection diverter valve, connect the AS inject port tubing to the diverter valve, and connect the diverter valve to the injection valves on two systems.
- 2. Calibrate the inject port volume of each system. This is the volume from the inject port through the diverter valve to the injection valve (see Section 5.9).
- 3. On the SYSTEM PARAMETERS screen, select SEQUENTIAL in the SAMPLE MODE field (see Figure 3-27).



Figure 3-27. System Parameters Screen: Sequential Mode

4. On the **PLUMBING CONFIGURATION** screen, enter the sample loop size installed on each valve (see Figure 3-28).

PLUMBING CONFIGURATION		
LOOP SIZE V1:	25 uL	
LOOP SIZE V2:	100 uL	
SAMPLE SYRINGE VOLUME:	250 uL	
SAMPLE SYRINGE CONFIG:	READY	
PREP SYRINGE VOLUME:	5 mL	
PREP SYRINGE CONFIG:	READY	
Help Prompt		

Figure 3-28. Plumbing Configuration Screen: Sequential Mode

5. Specify the injection volume and the cut volume required for the type of injection to be performed. The cut volume specifies a volume of sample to be "cut" (or omitted) from each side of the sample during the injection process. To set up a full-loop injection, see <u>Section 3.6.5</u>. For a partial-loop injection, see <u>Section 3.6.7</u>, and for a partial-loop limited-sample injection, see Section 3.6.8.

## **Example Program for Sequential Mode**

The following example Chromeleon or Chromeleon Xpress program shows the AS commands for sample loading and injecting when the autosampler is in sequential mode.

	Sampler	AcquireExclusiveAccess	;The timebase acquires exclusive control of the AS, to prevent another timebase from attempting to control the AS
	Diverte	erValve.Position_1	;Divert flow to system #1
0.000	Sampler	_InjectValve.LoadPosition	;Switch the inject valve on system #1 to Load
	Load		;Load sample onto the loop
	Wait	CycleTimeState	;Wait for the end of the specified Cycle Time
	Sampler	_InjectValve.InjectPosition	;Switch the inject valve on system #1 to Inject
	Inject		;Sweep the sample onto the column
	Wait	InjectState	;Wait for the injection to finish
5.00	BeginOv	verlap	;Begin preparing the next sample
	Sampler	ReleaseExclusiveAccess	;Release control; another timebase can now acquire exclusive control of the AS
30.00	End		

## 3.6.12 Sequential Concentrate Injections

In sequential concentrate mode, the AS delivers sample to two separate systems. A concentrator column replaces the sample loop on each injection valve. Each system is operated independently and can have a different sample and sample volume injected. Figure 3-29 shows a sequential concentrate injection to system #1 with reagent prime and reagent flush steps for matrix elimination.



Figure 3-29. Sequential Concentrate Injection Sequence (Example for Matrix Elimination)

## Setting Up a Sequential Concentrate Mode Injection

The AS must be equipped with the sequential injection option (P/N 063294) in order to perform sequential injections. In general, the following setup steps are required:

- 1. Follow the instructions in <u>Section B.3.6</u> to install the sequential injection diverter valve, connect the AS inject port tubing to the diverter valve, and connect the diverter valve to the injection valves on two systems.
- 2. Install a concentrator column on each injection valve.
- 3. Calibrate the inject port volume of each system. This is the volume from the inject port through the diverter valve to the injection valve (see Section 5.9).
- Follow the instructions in <u>Section 3.6.10</u> to set up a concentrate injection, except select SEQUENTIAL CONCENTRATE in the SAMPLE MODE field on the SYSTEM PARAMETERS screen (see Figure 3-30).



Figure 3-30. System Parameters Screen: Sequential Concentrate Mode

## 3.7 Selecting Syringe Speed Settings

The speed at which the sample syringe and prep syringe move can vary between a speed of 1 (the slowest) to 5 (the fastest). You can select syringe speeds on the Chromeleon or Chromeleon Xpress Control panel (see Section 3.7.1) and in the Program Wizard or Program Editor (see Section 3.7.2).

## **Controlling the Backpressure**

The AS is capable of delivering at a maximum pressure of 690 kPa (100 psi). The syringe speed affects the backpressure produced at the inject port and needle seal; a faster syringe speed produces more backpressure than a slower speed.

Other factors that affect the backpressure:

- The syringe size.
- The tubing IDs and lengths.
- The sample viscosity. Select a slower speed (1 or 2) for viscous samples.
- The presence of a concentrator column. If a concentrator column is installed, a speed of 1 or 2 is required.

Refer to <u>Section 3.8</u> to select the appropriate syringe speed, syringe size, and tubing IDs to keep the total backpressure below 690 kPa (100 psi)

## Selecting Separate Aspirate and Dispense Speeds

If you are using Chromeleon or Chromeleon Xpress (Release 6.8 SP2, or later) to control the autosampler, you can specify separate syringe speeds for aspirating and dispensing liquid during the following functions: Flush, Prime, Load, Reagent Flush, Reagent Prime, and Concentrate. For example, if the AS is loading sample, you can select a faster syringe speed for aspirating sample from the vial and a slower speed for dispensing sample to the loop or concentrator column.

## Notes

- If the AS is running under Local (front panel) control and is *never* connected to Chromeleon or Chromeleon Xpress Release 6.8 SP2 (or later), the aspirate and dispense speeds are always the same. See <u>Section D.7</u> for details about selecting syringe speeds for front panel operation.
- If the AS is connected to Chromeleon or Chromeleon Xpress Release 6.8 SP1 (or earlier) or if Moduleware Version 2.01 (or earlier) is installed, different aspirate and dispense speeds are not supported.

## 3.7.1 Selecting Syringe Speeds on the Control Panel

1. Go to the Autosampler Control panel (see Figure 3-31). Under **Syringe**, select the desired speeds for aspirating and dispensing liquids during priming and flushing. 1 is the slowest speed and 5 is the fastest.

🔞 File Edit View Workspace Qualification Control Batch Wi	ndow Help _ B ×
D 🚅 🖬 📲 🗃 🎒 X 🖻 📾 😽 🔈 🖣 💽	************************************
	ICS-3000_2 😫 👭
Home   Sequence Control   Status Autosampler   Gradient Pumps	s   Detector Compartment   EC Detector   3D Amp Plot
System Log (Audit Trail)	
2:08:10 PM 0.011 (Sampler) Start preparing next sample 2:08:40 PM 0.500 (EDet1) Log Background: 372.6 [nA]	e Sample 2 (position = 2, volume = 25.0).
2:08:40 PM 0.500 (Pump_1) Log Pressure: 0.0 [psi]	
2.06.40 PM 0.500 (Famp_2) Edg Pressure. AcqOff	
Status	
Connect Status: Idle	▼ 1
Exclusive Access: ICS-3000_2	
Mode:	Normal
Injection Type: Full Inject V	alve: LoadPosition (43) (44) (45) (46) (47) (48) (49)
Loop Size: 20.0 µl	3 7 3 9 4 4 2
Temperature Control Wellness	29 30 31 32 33 34 35
Tray Set Temp: 10 °C 🕂 Needle Cy	rcles 1738 22 23 24 25 29 29 29
Trav Actual Temp: 10.00 °C Wellness p	anel Needle align Calibrate IPTV
Syringe	
Prime	Flush
Prime Syringe: Sample 🛨	Flush Syringe: Sample
Prime Volume: 150 µl 🛨	Flush Volume: 100 µl 🗄
Prime speed: aspirate 3 🗸	Flush speed: aspirate 3
dispense 3 -	dispense 3 -
Prime reservoir: Reservoir_A	More Syringe Commands

Figure 3-31. Setting Syringe Speeds on the Control Panel

- 2. To select aspirate and dispense speeds for loading the loop, click the **More Syringe Commands** button. A second window appears.
- 3. In the **Load speed** fields, select the **aspirate** and **dispense** speeds (see Figure 3-32).



Figure 3-32. Setting Syringe Speeds for Loading the Loop

### Notes

- The aspirate and dispense speeds selected for priming are also used for flushing, and vice versa.
- If the AS is in concentrate mode, select a speed of 4 or less for dispensing during flushing and 2 or less for dispensing during loading.

## 3.7.2 Entering Syringe Speeds in the Program

NOTE If you do not include aspirate and dispense syringe speed commands in a program, the autosampler uses the speeds selected in the previous program or on the Control panel (whichever are the most recent). See <u>Section 3.7.1</u> for how to select syringe speeds on the Control panel.

To enter syringe speeds for loading the sample loop in the program:

1. Open the program in the Program Editor (in the panel tabset, click the **Sequence Control** tab and click the **Edit Program** button).

- 2. Place the cursor in the list of autosampler setup commands (for example, after the **Wait FlushState** command) and press the **Enter** key twice to create two empty lines.
- 3. Press the **F8** key to open the Commands dialog box.
- 4. Under **Sampler**, enter a value for the **LoadDispenseSpeed** (see Figure 3-33) and click **OK**.

Commands
Image: Sampler       Betention Time:       [min]         Image: Sampler_Relay_1       LoadDispenseSpeed:       3         Image: Sampler_Relay_2       LoadDispenseSpeed:       3         Image: Sampler_TTL_1       Image: Sampler_TTL_2       LoadDispenseSpeed:       3         Image: Sampler_Diverter/Valve       Image: Sampler_Calibration       Image: Sampler_DispenseSpeed       Image: Sampler_DispenseSpeed         Image: Sampler_DispenseSpeed       Image: Sampler_DispenseSpeed       Image: Sampler_Speed       Image: Sampler_Speed         Image: Sampler_DispenseSpeed       Image: Sampler_DispenseSpeed       Image: Sampler_Speed       Image: Sampler_Speed         Image: Sampler_DispenseSpeed       Image: Sampler_Speed       Image: Sampler_Speed       Image: Sampler_Speed
HushPrimeDispenseSpeed       BillshPrimeAspirateSpeed       SyringeSpeed       ✓   Comment:
Command: LoadDispense Speed = 3
OK         OK & Prev         OK & Next         Help

Figure 3-33. Commands Dialog Box

5. Press F8 again, enter a value for the LoadAspirateSpeed, and click OK.



Two new commands are added to the program.

Figure 3-34. Syringe Speed Commands in the Program

- NOTE If the AS is in concentrate mode, you can also enter separate aspirate and dispense syringe speeds for the Concentrate, Reagent Prime, and Reagent Flush functions. See <u>Section 3.6.10</u> for details about concentrate mode.
- NOTE The SyringeSpeed command shown in the program in Figure 3-34, controls the speed for various minor autosampler operations. For best results, use the default setting of 3 for this command.
## 3.8 Syringe Speed and Tubing Size Guidelines

The AS is capable of delivering at a **maximum pressure of 690 kPa (100 psi)**. Several factors affect the system backpressure, including the syringe speed, syringe size, and the IDs and lengths of the sample loop and the tubing used to connect the inject port and the injection valve. Use the following formula to determine the backpressure produced by these factors:

*backpressure = cm of tubing x kPa (psi)/mL/min/cm* (from <u>Table 3-10</u>) x mL/min (from <u>Table 3-11</u> or <u>Table 3-12</u>)

**Example:** 30 cm of 0.254-mm ID tubing x 1.6 kPa/mL/min/cm x 2.40 mL/min (5 mL syringe at speed 2) = 115 kPa

Tubing ID	Backpressure in kPa (psi) per mL/min per cm of tubing	Volume in $\mu$ L per cm of tubing
0.127 mm (0.005 in)	25 kPa (3.6 psi)	0.13
0.178 mm (0.007 in)	6.5 kPa (0.94 psi)	0.25
0.254 mm (0.010 in)	1.6 kPa (0.23 psi)	0.51
0.330 mm (0.013 in)	0.55 kPa (0.079 psi)	0.86
0.508 mm (0.020 in)	0.10 kPa (0.014 psi)	2.03

Sample Syringe Size	Sample Syringe Speed	Flow Rate (mL/min)
$100^{a}$ , 250, 500, and 1,000 $\mu L$	1	0.06
	2	0.12
	3	0.24
	4	0.60
	5	1.20
5 and 10 mL <sup>b</sup>	1	1.80
	2	2.40
	3	4.80
	4	7.20
	5	15.00

Table 3-10. Tubing Volume and Backpressure

### Table 3-11. Flow Rates Provided by the Sample Syringe

a. The maximum flow rate for a 100  $\mu$ L syringe is 0.60 mL/min.

b. In general, if you are using a 5 or 10 mL syringe, select a syringe speed of 1 or 2.

Prep Syringe Size	Prep Syringe Speed	Flow Rate (mL/min)
250, 500, and 1,000 µL	1	0.03
	2	0.09
	3	0.24
	4	0.60
	5	1.20
2.5, 5, and 10 mL <sup>a</sup>	1	1.80
	2	2.40
	3	4.80
	4	7.20
	5	15.00

### Table 3-12. Flow Rates Provided by the Prep Syringe

a. In general, if you are using a 5 or 10 mL syringe, select a syringe speed of 1 or 2.

### Example Backpressure Calculations for a Dual ICS-3000 System

The example tubing IDs and lengths for a dual ICS-3000 system shown in Figure 3-35 will be used to calculate the total backpressure produced for each system. See the following tables for details.



Figure 3-35. Example Tubing IDs and Lengths for a Dual ICS-3000 System

### System #1 Backpressure Calculations:

Tubing Tubing Flow Rate Backpressure Backpressure Produced Length from tubing (mL/min) (cm) (kPa/mL/min/cm) (kPa) Needle Seal Line 46 1.60 2.40 177 kPa 0.254 mm (0.010 in) Diverter Line #1 0.55 2.40 50 kPa 38 0.330 mm (0.013 in) 12 1.60 2.40 46 kPa 5 µL Loop 0.254 mm (0.010 in)

A 5 mL syringe and a syringe speed of 2 is assumed for this example.

Total backpressure for system #1 273 kPa

**Conclusion for system #1:** The total backpressure of 273 kPa is less than 690 kPa. Therefore, the selected tubing IDs, tubing lengths, and syringe speed can be used.

### System #2 Backpressure Calculations:

A 5 mL syringe and a syringe speed of 2 is assumed for this example.

Tubing	Tubing Length (cm)	Backpressure from tubing (kPa/mL/min/cm)	Flow Rate (mL/min)	Backpressure Produced (kPa)
Needle Seal Line 0.254 mm (0.010 in)	46	1.60	2.40	177 kPa
Diverter Line #1 0.330 mm (0.013 in)	64	0.55	2.40	85 kPa
25 μL Loop 0.508 mm (0.020 in)	15	0.10	2.40	3.6 kPa
		<b>T</b> 11 1 0		0((1))

Total backpressure for system #2 266 kPa

**Conclusion for system #2:** The total backpressure of 266 kPa is less than 690 kPa. Therefore, the selected tubing IDs, tubing lengths, and syringe speed can be used.

## 3.9 Opening the Autosampler Door During Operation

Normally, the autosampler door must remain closed while the AS is running either a sequence in Chromeleon or Chromeleon Xpress or a schedule from the front panel. If the door is opened inadvertently, the sampling arm stops immediately and the sequence or schedule is ended.

To safely open the door during a sequence or schedule, first press the **Door** button on the front panel, and then follow the on-screen instructions.

If it is not safe to open the door at this time, one of the following messages appears. The message depends on whether a Chromeleon or Chromeleon Xpress sequence or a front panel schedule is running:

STATUS:	INJECTING
	EN DOOR SAFETY SCREEN
not safe to	open door. Please wait for
status abo 10 seconds	ve to say - IDLE - for at least s during RUN (Timed Events).
NOTE: any key	press will exit this mode

Figure 3-36. Open Door Safety Screen: A Chromeleon or Chromeleon Xpress Sequence Is Running

### STATUS: INJECTING

OPEN DOOR SAFETY SCREEN WARNING: Autosampler arm motion possible, not safe to open door. Please wait for status above to say - WAITING FOR CYCLE TIME TO EXPIRE (Timed Events). NOTE: any key press will exit this mode

Figure 3-37. Open Door Safety Screen: A Front Panel Schedule Is Running

This message remains displayed until the sampling arm is in the home position and it is safe to open the door. The screen then displays the message that it is safe to open the door (see Figure 3-38).



Figure 3-38. Open Door Safety Screen: Safe to Open Door

The **Time** field counts down the amount of time remaining in which it is safe to open the door. If you open the door, the sequence or schedule pauses. When you close the door, the sequence or schedule resumes.

The timer continues counting down while the door is open. If the door is still open when the timer reaches zero, the sequence is ended.

To cancel the **OPEN DOOR SAFETY** screen, press any front panel button. If the door is open, a message cautions you that pressing a button again will end the schedule.

### Notes

- When the AS is operating under computer control, the door can be opened safely only when the sampling arm is in the home position and the autosampler is idle. This can occur at various times in a program (for example, after injection is complete, but before the next injection starts).
- When the AS is operating under front panel control, the door can be opened safely only when the sampling arm is in the home position and the status displays **WAITING FOR CYCLE TIME TO EXPIRE**. The cycle time controls the time between injections (see Section 2.7.5 and Section D.5.4). For most runs, selecting a cycle time greater than 4 minutes provides an adequate wait period between each injection for opening the door. If a method has a long sample prep phase, however, a longer cycle time may be required.

- If the AS never reaches a status that allows opening the door, check for one of the following:
  - If the AS is operating under Chromeleon or Chromeleon Xpress control, check the run length specified for the program. The run length may need to be increased to allow a longer wait period between injections.
  - If the AS is operating under front panel control, verify that a cycle time was specified for each method in the schedule and that the cycle time is long enough to provide a wait period between injections.

## 3.10 Routine Maintenance

This section describes routine maintenance procedures that the user can perform. Any other maintenance procedures must be performed by Dionex personnel.

### 3.10.1 Daily

- Check for air bubbles in the syringe(s) and tubing and remove them if they appear (see Section B.3.14).
- Check the volume of liquid in the flush bottle (and any reagent bottles that are installed) and refill when needed. After refilling the flush reservoir, manually flush the inject port (see Section 3.10.4).
- Check the volume of liquid in the waste container and empty when needed.



Neutralize acidic and caustic wastes before disposal. Dispose of all wastes in accordance with local regulations.



Neutralisez les déchets acides ou caustiques avant de les jeter. Jetez les déchets aux règlements locaux.



Neutralisieren Sie säurehaltige und ätzende Abfälle vor ihrer Entsorgung. Entsorgen Sie alle Abfälle entsprechend den lokalen Bestimmungen.

IMPORTANT

For correct drainage, make sure the end of the waste line is not submerged in waste liquid.

### 3.10.2 Periodically

- Check the alignment of the sampling arm needle. If the needle is not centered in the inject port opening when it enters the port, realign the needle using the Needle Alignment Wellness panel in Chromeleon or Chromeleon Xpress (see Section 5.8).
- Remove the drip tray (see <u>Section 5.5</u>), rinse the tray and the leak sensor, dry thoroughly, and replace (see <u>Section 5.6</u>).
- Check for leaks from the inject port, the syringe(s), the syringe valve(s), and the sampling needle line. If leaks occur, see Section 4.2.
- Monitor the usage of replaceable parts. The Chromeleon Wellness panel displays the usage counters for the various parts. Replace or service parts when recommended and reset the usage counter afterwards (see Section 5.2.1).

### 3.10.3 Annually

Perform the AS preventive maintenance procedure. An AS Preventive Maintenance Kit (P/N 060581) is available for this purpose. The kit provides parts and instructions for performing the procedure.

### 3.10.4 Manually Flushing the Inject Port

Flush the inject port after refilling the flush reservoir. The AS must be idle to run a flush cycle.

To manually flush the inject port from the Chromeleon or Chromeleon Xpress Control panel:

- 1. Open a panel tabset in Chromeleon or Chromeleon Xpress and select the **Autosampler** tab.
- 2. Enter the desired Flush Volume and Flush Speed (aspirate and dispense).
- 3. Click the **Flush** button.

To manually flush the inject port from the front panel:

- 1. Set the AS to LOCAL mode and DIRECT CONTROL.
- 2. Go to the FLUSH/PRIME screen (press Menu and 6) and enter the desired FLUSH VOLUME, or keep the default value.
- 3. Move the cursor to the **ACTION** field and select **FLUSH** (see Figure 3-39).



Figure 3-39. Initiating a Manual Flush Cycle

4. Press **Enter** to start the flush.

### 3.11 System Shutdown

No special system shutdown procedure is required. If the AS is under Chromeleon or Chromeleon Xpress control, you can load a shutdown program at the end of the schedule to automatically shut down the system. The shutdown program turns off the pump flow, the suppressor current, the sample temperature control option (if installed), and other system devices.

If you are running Chromeleon, you can use the SmartShutdown Wizard to assist you in creating and running a shutdown program. Refer to the Chromeleon Help for details. This chapter is a guide to troubleshooting minor problems that may occur while operating the AS Autosampler.

- <u>Section 4.1</u> describes error messages and how to troubleshoot them.
- The remaining sections in this chapter describe operating problems and how to resolve them.

If you are unable to resolve a problem by following the instructions here, contact Dionex Technical Support. In the U.S., call 1-800-346-6390. Outside the U.S., call the nearest Dionex office. Please have this chapter at hand when talking with Technical Support personnel.

## 4.1 Error Messages

When an error occurs, a beep sounds and a message appears. Each message is identified by a number in brackets in the lower-right corner of the AS screen. To clear the message, press any key.

The Moduleware (the instrument control firmware installed in each AS) also reports errors to Chromeleon or Chromeleon Xpress, where they are logged in the Audit Trail.

Most error messages occur because you have pressed an incorrect key or attempted to enter an incorrect value for a particular field. These types of messages generally do not need additional explanation and they are not listed here.

### • [232] Maximum number of steps reached.

A method can have a maximum of 100 steps, including the sample prep steps, the timed events steps, and one step for the method setup parameters.

Possible Causes: The method includes 100 steps.

Actions: No action is required at this time. However, before trying to create a new step in the method, delete an existing step.

### • [237] Memory is full. Cannot save additional method or schedule.

The current method or schedule occupies all available memory.

**Possible Causes:** Too many complex methods or schedules have been saved.

Actions: To free memory for future use, delete a method or schedule that is no longer used.

### • [250] DSP does not acknowledge.

The DSP (digital signal processor) did not respond to a command from the CPU.

**Possible Causes:** Communication problem between the DSP and the CPU.

### Actions:

- Turn the power off and then back on.
- If the error message appears again, contact Dionex.

### • [331] Method does not exist.

The schedule contains a method that does not exist.

**Possible Causes:** Incorrect method number entered into the schedule.

- Enter the method number of an existing method in the schedule.
- Create a new method and save it to the method number indicated in the schedule.

# • [348] A failure has occurred during power up! The status of all tests run during power up are indicated on the Diagnostic Tests screen which follows this error message.

At power-up, the AS electronics are tested. If one or more tests fails, this message appears. On the **DIAGNOSTIC TESTS** screen, an "F" indicates that a test failed and a "P" indicates it passed.

**Possible Causes:** One or more of these tests has failed, indicating a problem with that portion of the electronics. See <u>Section C.9.2</u> for a description of each test.

### Actions:

- If the **SYRINGE COMM** test fails, make sure the sample syringe cable is installed and connected to the correct connector on the AS rear panel (see <u>Section B.3.3</u>). If the prep syringe is installed, check its cable connection, also. Rerun the test.
- If the **SYRINGE COMM** test still fails or if other tests fail, contact Dionex.
- [362] Syringe size <= 1 mL require the 1.2 mL needle assy.

Actions: See <u>Section 2.3.3</u> for a description of the sampling needle assemblies and the syringe size requirements for each.

### • [363] Syringe size > 1 mL require the 8.5 mL needle assy.

Actions: See <u>Section 2.3.3</u> for a description of the sampling needle assemblies and the syringe size requirements for each.

• [366] The computed sample injection volume exceeded the needle assembly volume. The sample injection volume has been automatically reduced. Please confirm injection volume parameter.

Actions: See <u>"Maximum Sample Loop Sizes and Injection</u> <u>Volumes</u>" on page 68 for the maximum injection volumes allowed for each sampling needle assembly and syringe size.

# • [367] The computed sample injection volume exceeded the vial volume. The sample injection volume has been automatically reduced. Please confirm injection volume parameter.

Actions: See Section 3.6.4 for the guidelines for sample volumes used, loop sizes, and injection volumes. See Section 2.3.5 for a list of the available sample vial and well plate sizes.

# • [368] The computed sample injection volume exceeded the syringe volume. The sample injection volume has been automatically reduced. Please confirm injection volume parameter.

Actions: See <u>Section 3.6.4</u> for the guidelines for sample volumes used, loop sizes, syringe sizes, and injection volumes.

# • [375] An attempt was made to use the sample prep syringe. This option is not installed.

Two of the method sample prep operations (**DISPENSE** and **DILUTE**) can be used only when the sample preparation option is installed. See <u>Section D.5.3</u> for details about defining sample prep steps from the front panel.

**Possible Causes:** A **DISPENSE** or **DILUTE** operation was specified in the sample prep steps of the current method, although the sample preparation option is not installed.

### Actions:

- Remove the step(s) requiring the sample preparation option.
- Select a method that does not require the sample preparation option.

### • [376] Time-out occurred waiting for syringe to complete an action.

Every syringe action is assigned a maximum time in which the action should be completed. The syringe did not complete its task in the allotted time, indicating either a mechanical or a communication problem.

### **Possible Causes:**

• Faulty syringe

• Faulty cable

### Actions:

- Check the syringe motion. If the syringe action is faulty, the syringe may need to be replaced (see Section 5.12).
- If the syringe does not move in response to a command, the cable may need to be replaced. Contact Dionex.
- [377] Time-out occurred waiting for syringe serial port txrdy. Possible Causes:
  - Blocked syringe lines
  - Faulty syringe
  - Faulty cable

### Actions:

- Check the lines to the syringe for crimps or other blockages. Replace the lines, if necessary (see Section 5.1).
- Check the syringe motion. If the syringe action is faulty, the syringe may need to be replaced (see Section 5.12).
- If the syringe does not move in response to a command, the cable may need to be replaced. Contact Dionex.

# • [378] Time-out occurred waiting for syringe to respond to command.

Possible Causes: Communication problem.

Actions: Turn off the AS power, check the syringe cable connections, and then turn on the power again. If the message reappears, contact Dionex.

### • [379] Response from syringe contained data errors.

Possible Causes: Communication problem.

Actions: Turn off the AS power and then turn on the power again. If the message reappears, contact Dionex.

## • [380] Response from syringe was too long. Receive buffer overflowed.

Possible Causes: Communication problem.

Actions: Turn off the AS power and then turn on the power again. If the message reappears, contact Dionex.

# • [381] An attempt was made to move the syringe to an illegal position. Most likely, the specified volume was larger than the remaining syringe capacity.

Valid positions for the syringes are programmed into the AS instrument control Moduleware. There was an internal instruction to move a syringe to a position that was not programmed.

Possible Causes: Problem with the Moduleware.

Actions: Turn off the AS power and then turn on the power again. If the message reappears, contact Dionex.

# • [382] An attempt was made to move a syringe value to an illegal position.

Valid positions for the syringe valves are programmed into the AS instrument control Moduleware. There was an internal instruction to move a syringe valve to a position that was not programmed.

Possible Causes: Problem with the Moduleware.

Actions: Contact Dionex.

# • [383] An attempt was made to set the syringe speed to an illegal value.

Valid speeds for the syringes are programmed into the AS instrument control Moduleware. There was an internal instruction to move a syringe at a speed that was not programmed.

Possible Causes: Problem with the Moduleware.

Actions: Contact Dionex.

# • [384] Cannot use sample syringe to aspirate from specified source reservoir.

**Possible Causes:** The sample syringe can aspirate only from the flush reservoir or from the sampling needle.

Actions: Select a different source for the aspirate function or select the prep syringe (if installed).

# • [386] Operation could not be completed because vial tray is not installed.

Magnets on the sample trays indicate to a magnetic sensor in the autosampler compartment when a tray is installed (and the type of tray).

**Possible Causes:** (assuming a tray is installed, but is not recognized by the sensor)

- Tray magnets are missing or incorrectly installed.
- Sensors are not working.

### Actions:

- Replace the tray.
- Replace the tray sensor card. Contact Dionex.

### • [387] An illegal vial number was specified.

The type of tray installed determines which vial numbers can be used in methods and schedules. See <u>Section 3.4</u> for details about specifying sample positions for each type of tray.

**Possible Causes:** The vial number specified in the program or method sample prep steps or the sequence or schedule steps is not available for the type of tray installed in the autosampler compartment.

- Change the vial specified in the method or schedule.
- Use a tray type that accommodates the specified vial.

### • [388] A NAK command was received from the DSP.

The CPU sent a command to the DSP which the DSP cannot execute.

Possible Causes: An installed option is not being recognized.

### Actions:

- Check the **INSTALLED OPTIONS** screen (see <u>Section C.6.1</u>) to verify that all optional features are being recognized. A check mark should appear beside each installed option.
- If an installed option is not checked, turn off the AS power and then turn on the power again and recheck the **INSTALLED OPTIONS** screen. If the problem persists, contact Dionex.

### • [389] Timed out waiting for motion complete command from DSP.

The DSP could not complete a motion in the time allowed.

### **Possible Causes:**

- Mechanical problem with the autosampler needle arm
- A valve is not being actuated

- Check for an obstruction in the autosampler compartment.
- Manually test the autosampler needle arm movement, using the **XYZ TEST** screen (see Section C.9.3).
- Manually test the sample syringe, the sample prep syringe, and the injection valve(s), using the LIQUID CONTROL screen (see <u>Section C.9.5</u>).
- If the problem persists, contact Dionex.

### • [390] Received invalid command from DSP.

**Possible Causes:** Communication between the CPU and the DSP was corrupted.

Actions: Turn off the power and then turn on the power again. If the message reappears, contact Dionex.

### • [391] Schedule error. Specified schedule does not exist.

**Possible Causes:** The schedule selected to run has not been saved.

Actions: Create the new schedule or select an existing schedule.

# • [398] Battery backed up RAM failed data validation test. Setting system parameters to defaults and initializing schedule and method database.

### **Possible Causes:**

- Temporary short on CPU board
- Dead battery on CPU board
- Malfunctioning power supply

Actions: If a temporary short is suspected, the AS can still be operated. You will need to re-enter any method and schedule information and customized system setup parameters. If the problem persists, contact Dionex.

### • [408] Recoverable motion error occurred in X-axis

**Possible Causes:** An error occurred during X-axis movement of the sampling arm. The DSP was able to recover from the error and continue the motion.

Actions: Occasional occurrences of this error can be ignored. If the problem persists, contact Dionex.

# • [409] Non-recoverable motion error occurred in X-axis. Clear any physical obstructions and then press any key to reset system.

**Possible Causes:** An error occurred during X-axis movement of the sampling arm. The DSP was not able to recover from the error.

### Actions:

- 1. Turn off the power.
- 2. Check for an obstruction in the autosampler compartment.
- 3. Check for freedom of movement in all three axes of the sampling arm.
- 4. Turn on the power again. If the problem persists, contact Dionex.

### • [414] Specified volume is larger than size of sample syringe.

During normal operation (not during diagnostics), the syringe can perform up to four multiple strokes to aspirate or dispense a volume up to four times larger than the syringe size.

### **Possible Causes:**

- During a schedule or flush/prime operation, the specified volume is more than four times the size of the sample syringe.
- On the LIQUID CONTROL diagnostics screen (see <u>Section C.9.5</u>), either the specified volume exceeds the size of the sample syringe or the cumulative volume (when performing multiple actions) exceeds the size of the syringe.

- Reduce the volume specified or, if the sample preparation option is installed, use the prep syringe (not the sample syringe) for certain operations. See <u>"Sample Preparation Option Functions"</u> on page 16 for details.
- On the **LIQUID CONTROL** screen, select a different operation or reduce the volume specified.

### • [415] Cycle time expired before INIT step was completed.

When the cycle time expires, the AS ignores the cycle time and delays injection until the **INIT** step is complete. See Section 2.7.5 for details.

Possible Causes: The selected cycle time was too short.

Actions: Select a longer cycle time (see <u>Section D.5.4</u>).

### • [418] DSP not able to home XYZ motor arm

#### **Possible Causes:**

- Physical blockage of the sampling arm
- DSP malfunction

### Actions:

- 1. Turn off the AS power.
- 2. Check for an obstruction in the autosampler compartment.
- 3. Check for freedom of movement in all three axes of the sampling arm.
- 4. Turn on the power again. If the problem persists, contact Dionex.

# • [421] Timed events step took so long to execute that at least one subsequent step was missed.

#### **Possible Causes:**

• Insufficient time allowed for loading the sample loop

- On the method's **TIMED EVENTS** screen, set the injection valve to the load position in the **INIT** step (the default).
- If the valve is not set to load in the INIT step, be sure to allow enough time between the LOAD and the INJECT parameters for the loop to be completely loaded. The time needed depends on the sample syringe volume and the selected syringe speed (see <u>Section 3.7</u>).

# • [429] Format of global database has changed. Reinitializing to default values.

### **Possible Causes:**

- Dead battery on CPU board
- Malfunctioning power supply

Actions: Contact Dionex.

• [430] Format of method/schedule database has changed. Reinitializing to default values.

### **Possible Causes:**

- Dead battery on CPU board
- Malfunctioning power supply

Actions: Contact Dionex.

• [431] Format of calibration database has changed. Reinitializing to default values.

### **Possible Causes:**

- Dead battery on CPU board
- Malfunctioning power supply

Actions: Contact Dionex.

### • [432] DSP has unexpectedly reset. Press any key to reset system

#### **Possible Causes:**

Actions: Occasional occurrences of this error can be ignored. If the problem persists, contact Dionex.

### • [438] Timed out waiting for home position to be found. Possible Causes:

- Autosampler compartment door open at power-up
- DSP malfunction
- Sampling arm malfunction

### Actions:

- Close the autosampler compartment door and then press the **Home** button.
- If closing the door does not eliminate the problem, try turning the power off and then on again. If the problem persists, contact Dionex.

# • [439] Method/Schedule database has been corrupted. Reinitializing database.

### **Possible Causes:**

- Dead battery on CPU board
- Malfunctioning power supply

Actions: Contact Dionex.

• [441] Sampler door opened.

### **Possible Causes:**

- Autosampler compartment door accidentally opened
- Door not closing properly
- Door sensor malfunction
- Door magnet missing

### Actions:

- The autosampler door must be closed to operate the AS. Close the door and restart the schedule, if necessary.
- If the door appears closed, but the message still occurs, check for an obstruction that is preventing the door from closing completely.
- Make sure the magnet on the inside lower front edge of the door is present. Also make sure the magnet on the lower edge of the autosampler compartment is present. If a magnet is missing, contact Dionex.

### • [443] Leak detected in tray/syringe area.

### **Possible Causes:**

- Excess liquid in the waste container is preventing liquid from siphoning through the waste tubing
- Blocked or crimped waste tubing
- Leaking inject port
- Miscalibrated leak sensor

- Empty the waste container.
- Make sure the waste tubing is not crimped and that it is not pushed against the bottom of the waste container.
- If the inject port is leaking, see <u>Section 4.2.3</u>.
- If the tray is dry, but the error is still appearing, recalibrate the sensor (see <u>Section 5.11</u>).

• [445] Invalid leak sensor reading. May be open- or short-circuit, or bad sensor. See Leak Sensor Calibration screen for more information.

Actions: The leak sensor card, the cables, or the sensor must be replaced. Contact Dionex.

• [446] Current leak sensor voltage is outside of legal calibration range.

Attempting to calibrate when the leak sensor voltage reading is outside the normal dry range.

Possible Causes: The sensor is not dry enough for calibration.

Actions: Clean and dry the area thoroughly and recalibrate (see <u>Section 5.11</u>).

### • [447] Leak sensor needs recalibration. Go to Leak Sensor Calibration screen to perform recalibration operation.

Actions: See <u>Section 5.11</u> for calibration instructions.

• [448] Motion command to DSP was aborted. Needle may not have homed properly.

**Possible Causes:** Loose coupling in the Z-axis. The needle does not stay up.

Actions: Try the motion again. If the problem persists, contact Dionex.

• [449] Recoverable motion error occurred in Y-axis.

**Possible Causes:** An error occurred during Y-axis movement of the sampling arm. The DSP was able to recover from the error and continue the motion.

Actions: Occasional occurrences of this error can be ignored. If the problem persists, contact Dionex.

# • [450] Non-recoverable motion error occurred in Y-axis. Clear any physical obstructions and then press any key to reset system.

### **Possible Causes:**

- Vial obstructing motion
- Misaligned sampling needle arm

### Actions:

- Remove the vial obstruction.
- Align the sampling needle arm in the inject port (see <u>Section 5.8</u>).
- Turn off the power and check for freedom of movement of all three axes of the sampling arm.

### • [451] Recoverable motion error occurred in Z-axis.

**Possible Causes:** An error occurred during Z-axis movement of the sampling arm. The DSP was able to recover from the error and continue the motion.

Actions: Occasional occurrences of this error can be ignored. If the problem persists, contact Dionex.

# • [452] Non-recoverable motion error occurred in Z-axis. Clear any physical obstructions and then press any key to reset system.

### **Possible Causes:**

- Vial obstructing motion
- Bent sampling needle
- Misaligned sampling needle arm
- Incorrect X-Y position (not positioned above a vial or well)

- Remove the vial obstruction.
- If the needle is bent, replace the sampling needle assembly (see <u>Section 5.4</u>).
- Align the sampling needle arm in the inject port (see <u>Section 5.8</u>).

- Turn off the power and check for freedom of movement of all three axes of the sampling arm.
- Contact Dionex.
- [453] Recoverable motion error occurred in inject valve.

### **Possible Causes:**

Valve in wrong position

Actions: Turn off the AS power and then turn on the power again. If the message reappears, contact Dionex.

• [454] Non-recoverable motion error occurred in inject valve. Clear any physical obstructions and then press any key to reset system.

Possible Causes: Valve in wrong position

Actions: Turn off the AS power and then turn on the power again. If the message reappears, contact Dionex.

### • [455] Recoverable motion error occurred in auxiliary valve.

Possible Causes: Valve in wrong position

Actions: Turn off the AS power and then turn on the power again. If the message reappears, contact Dionex.

### • [457]-[474] Syringe reports fatal error: Syringe reports error:

**Possible Causes:** Syringe errors can result from various mechanical problems or from a communication failure between the syringe and the AS electronics.

- Check the syringe cable connections.
- For fatal errors, press a key to reset the system.
- If the error persists, contact Dionex.

### • [475] Cannot run schedule. No DSP detected.

#### Possible Causes: Electronic malfunction

Actions: Turn off the AS power and then turn on the power again. If the message reappears, contact Dionex.

### • [476] Cannot run schedule. No sample syringe detected.

### **Possible Causes:**

- Syringe cable not plugged into the rear of the AS
- Faulty syringe cable or syringe

### Actions:

- Check the syringe cable connection.
- Replace the cable. Contact Dionex.
- [479] Non-recoverable error. Both internal and external inject valves are connected. Please, power down and disconnect one inject valve before continuing.

**Possible Causes:** An injection valve is installed in both the autosampler compartment and in another module.

Actions: Disconnect one of the valves.

# • [480] Timed out waiting for tray temperature to stabilize. Either temperature control is broken, or set temperature is too far away from ambient.

The sample temperature control option was not able to reach the temperature set point within the time allotted.

### **Possible Causes:**

- Ambient temperature too high or low
- Airflow to cool the Peltier devices is restricted
- Malfunction of the sample temperature control option

#### Actions:

Go to the TEMPERATURE STATISTICS screen (press Menu, 8, and
4) and check the following parameters under TRAY SET TEMP:

**PELTIER**: The Peltier value should be above zero. If it is not, contact Dionex.

**DUTY CYCLE**: A value of 255 means the sample temperature control option is working to capacity. The ambient temperature may be too high or too low, or the airflow to the Peltier device may be restricted.

- Select a temperature that is closer to ambient. If possible, adjust the ambient temperature before running the AS.
- Check the fan intake under the AS and the fan exhaust in the back for any obstructions restricting airflow. Make sure there is at least 6 cm (2.5 in) of free space behind the AS for ventilation.
- If the problem persists, contact Dionex.

# • [482] An injection was skipped because of a problem in the autosampler. Will continue with next injection.

Possible Causes: Vial not present

Actions: Insert vial in intended location.

# • [486] A DIRECT CONTROL command was received while the sampler door is open. Cannot implement command.

- To execute Direct control commands from the front panel (Local mode), temporarily disable the autosampler compartment door lock on the **DOOR INTERLOCK BYPASS** screen (see <u>Section C.6.7</u>).
- If you are running a schedule or Chromeleon or Chromeleon Xpress sequence, press the **Door** button before opening the sample door (see <u>Section 3.9</u>).

## 4.2 Liquid Leaks

### 4.2.1 Leaking Syringe or Syringe Valve Port Possible Causes:

- Cracked syringe or leaking plunger seal
- Missing or worn Teflon® washer in the leaking valve port
- Loose syringe

- Check for a crack or a leaking plunger seal in the syringe. Replace the syringe, if necessary (see Section 5.12).
- If the leak is from one of the valve ports, remove the valve and syringe (see Section 5.12). Remove the existing white Teflon washer from inside the valve port and replace it with a new washer (P/N 061315). The washers are supplied with the syringe valve and with the AS Preventive Maintenance Kit (P/N 060581). Reinstall the syringe and valve.
- Make sure the syringe is tightened. To tighten, hold the syringe at the top and bottom fittings and turn it clockwise (as viewed from the top) (see <u>Figure 4-1</u>). Tighten fingertight only; do not overtighten.



Figure 4-1. Tightening the Syringe

### 4.2.2 Leaking Drain Line Connection

- Make sure the drain line is not crimped or otherwise blocked.
- Make sure the drain line is not submerged in liquid in the waste container. Empty the container, if needed.
- Make sure the drain line is pushed tightly onto the fitting and that the line is not elevated at any point after it exits the autosampler.

### 4.2.3 Leaking Inject Port

- Check for a leaking fitting and tighten.
- Check the sampling needle; if the needle tip appears worn or damaged, replace the sampling needle assembly (see Section 5.4).
- Check the needle seal; if the seal appears worn or damaged, replace the needle seal assembly (see Section 5.7).
- If fluid appears on the top of the inject port during injection, the needle may not be reaching the correct depth in the port or may not be aligned over the inject port. Realign the inject port (see Section 5.8).
- Air in the needle seal assembly or in the tubing from the inject port to the injection valve can cause continued flow after the syringe pump has stopped. Make sure all lines are primed and free of air bubbles. See <u>Section B.3.13</u> for priming instructions.

### 4.2.4 Leaking Fitting

Locate the source of the leak. Tighten or replace the liquid line connection (see Section 5.1).

### 4.2.5 Broken Liquid Line

Replace the line and fittings (see <u>Section 5.1</u>).

## 4.3 Condensation on Well Plate Covers

Condensation can form on the well plate covers of the micro-well plates if the tray temperature set point is at 4  $^{\circ}$ C and the ambient temperature and relative humidity in the laboratory are above 25  $^{\circ}$ C and 60% humidity.

To prevent condensation, either ensure that the laboratory temperature remains below 25  $^{\circ}$ C and the humidity below 60%, or increase the tray temperature set point.

## 5 • Service

This chapter describes AS Autosampler service and repair procedures that the user can perform. All procedures not included here, including electronics-related repair procedures, must be performed by Dionex personnel. For assistance, contact Dionex Technical Support. In the U.S., call 1-800-346-6390. Outside the U.S., call the nearest Dionex office.

Before replacing any parts, refer to the troubleshooting information in <u>Chapter 4</u> to isolate the cause of the problem.



The CPU card contains a lithium battery. If the CPU card is replaced, dispose of the used battery properly. Contact your local government for disposal or recycling practices in your area.



Substituting non-Dionex parts may impair AS performance, thereby voiding the product warranty. Refer to the warranty statement in the Dionex Terms and Conditions for more information.

## 5.1 Replacement Tubing and Fittings

The following tables list the tubing, fittings, and tubing assemblies used to plumb the AS autosampler compartment.

NOTE The AS is capable of delivering at a maximum pressure of 690 kPa (100 psi). Refer to <u>Section 3.8</u> to select the appropriate syringe speed and tubing sizes to keep the total backpressure below 690 kPa (100 psi).

### 5.1.1 Syringe and Reservoir Connections

Part Number	Description	Used For
052112	PFA tubing: 1.55-mm (0.062-in) ID	Connecting the flush reservoir
048949	1/8-in flangeless ferrule fitting	to the sample syringe valve
052267	1/4-28 flangeless nut	

### 5.1.2 Assemblies, Tubing, and Fittings

Part Number	Description	Used For
057301	Needle seal assembly, 18-in, with 10-32 ferrule fitting	Connecting the AS inject port to the injection valve
061290	Needle seal assembly, 18-in, without 10-32 ferrule fitting	Connecting the AS inject port to two injection valves for simultaneous injections
062564	Needle seal assembly, 24-in, with 10-32 ferrule fitting	Connecting the inject port of a second AS to injection valve #2 in a dual-ICS-3000 system
054271	Sampling needle assembly, with a 1.2 mL volume	Connecting the sampling needle arm to the sampling valve, use with sample syringe volumes of 1 mL or less
061267	Sampling needle assembly, with an 8.5 mL volume	Connecting the sampling needle arm to the sampling valve, use with sample syringe volumes greater than 1 mL
044777	PEEK tubing: Green 0.75-mm (0.030-in) ID	Connecting the injection valve port 6 (waste) to the AS drip
043276	Double-cone ferrule fitting	tray
043275	10-32 double-cone bolt	
042857	Sample loop, 25 µL	Connecting ports 1 and 4 on the injection valves (if installed)
049714	PEEK tubing: Blue, 0.33-mm (0.013-in) ID	Connecting ports 1 and 2 on a diverter valve to injection valves (sequential injection option only)
042855	PEEK tubing: Orange, 0.50-mm (0.020-in) ID	Connecting the simultaneous injection valve tee to each injection valve
042690	PEEK tubing: Black, 0.25-mm (0.010-in) ID	Sample loop
063405	PEEK tubing: Violet, 0.18-mm (0.007-in) ID	Sample loop

# NOTE For other replacement parts (vials, well plates, syringes, etc.), refer to <u>Appendix F</u>.

## 5.2 Resetting the Usage Counters

The Chromeleon Predictive Performance feature monitors the lifetime of serviceable parts by counting the number of times a part has been used since it was installed. After replacing or servicing a monitored part, reset the usage counter, so that Chromeleon can then begin monitoring use of the new part. See <u>Section 2.6</u> for more information about Predictive Performance.

Chromeleon monitors usage of the following AS parts: injection valve, diverter valve, XYZ sampling needle arm, sampling needle assembly, needle seal assembly, sample syringe, and prep syringe.

# NOTE Reset the counter for the XYZ sampling needle arm after lubricating or replacing the arm. Reset other usage counters after replacing the part.

You can reset usage counters from either the AS Wellness panel in Chromeleon or Chromeleon Xpress (see Section 5.2.1) or on the front panel USAGE LOG SCREEN (see Section 5.2.2).

### 5.2.1 Resetting a Usage Counter on the AS Wellness Panel

- 1. Open a panel tabset in Chromeleon or Chromeleon Xpress and select the **Autosampler** tab.
- 2. Click the **Wellness panel** button to open the AS Wellness Panel (see Figure 5-1).

🚟 AS Wellness Panel			
Needle Cycles	4,200	Reset	
XYZ Arm Cycles	10,400	Reset	
AS Inject Valve Cycles	2,100	Reset	
Diverter Valve Cycles	1,000	Reset	
Sample Syringe Cycles	6,200	Reset	
Prep Syringe Cycles	6,200	Reset	
=> Warning. Consider performing maintenance soon			
=> Limit reached. Perform maintenance			

Figure 5-1. Example AS Wellness Panel (Default Warning Values for All Parameters Shown)

 Click the **Reset** button for the part that you replaced or serviced. The number is reset to zero.

### 5.2.2 Resetting a Usage Counter on the Usage Log Screen

1. On the **MENU of SCREENS**, press **8**, **6**, and **1** to go to the **USAGE LOG** screen (see Figure 5-2).



Figure 5-2. Usage Log Screen

The monitored parts do not all fit on one screen. If necessary, press the down arrow to display the usage counter to be reset (see Figure 5-3).



Figure 5-3. Usage Log Screen (Additional Parameters)

2. Position the cursor in the **RESET** field of the part that was replaced or serviced and press **Enter**.

The number is reset to zero.

## 5.3 Replacing the Sample or Prep Syringe

Replace a sample or prep syringe after about 6,200 cycles of use. When the AS is operating under Chromeleon or Chromeleon Xpress control, a message is displayed in the Audit Trail when a syringe reaches this usage limit. If you are operating the AS from the front panel, the **USAGE LOG SCREEN** (see Section C.9.6) displays the number of cycles performed since the syringe was last replaced.

Syringe Size	Part Number
100 μL <sup>a</sup>	055064
250 µL	053916
500 μL	055065
1,000 µL	055066
2.5 mL <sup>b</sup>	055067
5 mL <sup>c</sup>	053915
10 mL <sup>c</sup>	055068

The table below lists part numbers for the available syringe sizes.

a. The 100  $\mu L$  syringe is for use as a sample syringe only.

b. The 2.5 mL syringe (P/N 055067) is for use as a prep syringe only.

c. A 5 or 10 mL syringe is required for the simultaneous injection option. When a 5 or 10 mL syringe is used (either for simultaneous injection or for loading a concentrator), an 8.5 mL sampling needle assembly is required.
### 5.3.1 Removing the Existing Syringe

- 1. Using a flathead screwdriver, unscrew and remove the syringe drive connecting screw (see Figure 5-4).
- 2. Hold the syringe at the top and bottom and unscrew it from the valve.
- Remove the white Teflon washer from the syringe valve port. Note: To remove the washer, push a piece of 0.8-mm (0.031-in) OD PEEK tubing into the center of the washer and twist to dislodge the washer.
- 4. Install a new white Teflon washer (P/N 061315) into the new syringe.



Figure 5-4. Removing the Syringe

### 5.3.2 Filling the New Syringe and Removing Bubbles

#### Method A

- 1. Fill a container with isopropyl alcohol. Place the threaded end of the syringe into the container and slowly pull liquid into the syringe.
- 2. Remove the syringe from the liquid and point the threaded end up into the air. If air bubbles are present, push the syringe plunger up and pull down rapidly with short movements to dislodge the bubbles. Repeat until all bubbles are dislodged.
- 3. Place the threaded end of the syringe into the isopropyl alcohol again and draw a full syringe volume up by slowly pulling out the plunger.
- 4. Keep the syringe in the liquid and slowly push the syringe plunger until all bubbles exit the syringe. Then, fill the syringe completely.
- 5. Remove the syringe from the liquid. While holding it vertically, push a small amount of liquid out the top to make sure no air remains.

#### Method B

- 1. Fill a container with isopropyl alcohol. Place the threaded end of the syringe into the container and slowly pull liquid into the syringe.
- 2. Remove the syringe from the liquid and hold it with the threaded end up under a stream of deionized water.
- 3. Push the syringe plunger up and pull down until all bubbles are removed.

### 5.3.3 Connecting the New Syringe and Flushing

- 1. To position the new syringe under the valve, you may need to lower move the syringe pump drive. To do this, use the **LIQUID CONTROL** screen (see Section C.9.5) to manually aspirate 100 to 1,000  $\mu$ L.
- 2. Position the syringe under the valve.
- 3. Holding the syringe at the top and bottom, screw it back into the valve and tighten fingertight.
- 4. Align the syringe piston with the connecting screw mount on the syringe drive.

- 5. Insert the syringe drive connecting screw removed in <u>Section 5.3.1</u> and tighten fingertight.
- 6. Perform a flush cycle:
  - a. Press Menu, Menu, and 6 to go to the FLUSH/PRIME screen.

FLUSH/PRIME				
FLUSH		PRIME		
250	uL	<b>VOLUME:</b>	2000 uL	
<b>FLUS</b>	н	<b>RESERVOIR:</b>	RES A	
SAMP	LE	SYRINGE:	PREP	
READ	Υ	ACTION:	READY	
Help Prompt				
	FL H 250 FLUS SAMF READ	FLUSH/F H 250 uL FLUSH SAMPLE READY	FLUSH/PRIME H PRIN 250 uL VOLUME: FLUSH RESERVOIR: SAMPLE SYRINGE: READY ACTION:	

Figure 5-5. Flush/Prime Screen

- b. Move the cursor to the ACTION field and press Select  $\Delta$  or Select  $\nabla$  to select FLUSH. Press Enter.
- 7. Check the syringe for bubbles. If any exist, remove the syringe and remove the bubbles (see <u>Section 5.3.2</u>). Reconnect the syringe and flush again.

### 5.3.4 Initializing the Syringe

1. On the MENU of SCREENS, press **5** and **3** to go to the PLUMBING CONFIGURATION screen.



Figure 5-6. Plumbing Configuration Screen

- Move the cursor to the SAMPLE SYRINGE CONFIG field or the PREP SYRINGE CONFIG field (depending on which syringe is to be removed).
- 3. Press **Select**  $\triangle$  to select the **INITIALIZE** option.
- 4. Press Enter.
- 5. The syringe plunger moves down a small amount. This is its initial position (see Figure 5-7).



Figure 5-7. Initializing the Syringe

### 5.3.5 Resetting the Syringe Usage Counter

After replacing the syringe, reset the syringe usage counter from either the AS Wellness panel in Chromeleon or Chromeleon Xpress or on the front panel **USAGE LOG SCREEN** (see Section 5.2).

# 5.4 Replacing the Sampling Needle Assembly

Replace the sampling needle assembly after about 6,200 cycles of use (the cycle count increments each time the needle is inserted into the inject port). When the AS is operating under Chromeleon or Chromeleon Xpress control, a reminder message is displayed in the Audit Trail when the number of cycles reaches this usage limit. When the AS is operating under front panel control, the **USAGE LOG** 

**SCREEN** (see <u>Section C.9.6</u>) displays the number of cycles performed since the assembly was last replaced.

Two versions of the PEEK sampling needle assembly are available. The version used depends on the volume of the sample syringe installed:

- If the sample syringe is 1 mL or less, a 1.2 mL sampling needle assembly (P/N 054271) is installed (see Figure 5-8).
- If the sample syringe is 5 or 10 mL, an 8.5 mL sampling needle assembly (P/N 061267) is installed. The 8.5 mL assembly includes the same parts as the 1.2 mL assembly, plus a coupler and an additional length of tubing to increase the volume (see Figure 5-9).



Figure 5-8. 1.2 mL PEEK Sampling Needle Assembly (P/N 054271)

NOTE The PEEK sampling needle assemblies use a different bolt and ferrule than previous sampling needle assemblies. If you remove the PEEK needle from the 0.8-mm (0.032-in) ID tubing, be sure to use the correct fitting bolt (P/N 052667) and ferrule (P/N 064553) to reattach the needle to the tubing.



Figure 5-9. 8.5 mL PEEK Sampling Needle Assembly (P/N 061267)

### 5.4.1 Removing the Old Sampling Needle Assembly

- 1. Press Menu, 8, and 5 to go to the LIQUID CONTROL screen.
- 2. Move the cursor to the VIAL# field. Press Select  $\nabla$  to select FLU and press Enter.

The needle arm moves to the flush port.

3. Turn off the power and open the autosampler door.

- 4. The line that connects the sampling needle to the sample syringe is clipped to the upper-right side of the needle arm (see Figure 5-10). Using a 2.5-mm Allen wrench, remove the M3 screw that attaches the clip to the sampling arm. Remove the clip, and then remove the tubing from the clip.
  - NOTE Some needle arms have a clip that is open at the top. This type of clip does not need to be removed. To remove the tubing from the clip, pull the clip out slightly and push the tubing up and out of the clip.



Figure 5-10. Sampling Needle Line and Tubing Clip

5. Measure the distance from the yellow sleeve on the sampling needle assembly line (see Figure 5-10) to the needle. When installing the new sampling needle assembly, position the sleeve on the new line the same distance from the needle.

- The sampling needle line is coiled and held by a bracket and clamp on the upper-right side of the autosampler compartment (see <u>Figure 5-11</u>). To remove the tubing, use a flathead screwdriver to gently pry the center bracket apart. Then, lift the tubing out of the bracket and clamp.
- 7. Measure the distance from the second yellow sleeve (see <u>Figure 5-11</u>) to the needle. When installing the new sampling needle assembly, position the sleeve on the new line the same distance from the needle.



Figure 5-11. Sampling Needle Line Bracket and Clamp (Right Side of Compartment)

- If you are replacing an 8.5 mL assembly (P/N 061267), go on to <u>Step 9</u>. If you are replacing a 1.2 mL sampling needle assembly (P/N 054271), follow the steps below to disconnect the sampling needle line:
  - a. Disconnect the sampling needle assembly from port C on the sample syringe valve.
  - b. Pull the ferrule fitting off the end of the tubing and remove the bolt.

- c. Tape one end of a piece of string or tubing to the end of the sampling needle assembly line. This will help you thread the new line through the autosampler later in the procedure.
- d. From the inside of the autosampler, pull the tubing into the autosampler compartment, along with the end of the attached string or tubing. Go on to <u>Step 10</u>.
- 9. If you are replacing an 8.5 mL sampling needle assembly (P/N 061267), disconnect the sampling needle line from the coupler that connects it to the larger ID tubing (see Figure 5-11).

# NOTE The coupler may be hidden behind the metal frame supporting the needle arm assembly.

- 10. Turn the needle fitting above the needle block (see Figure 5-12) counterclockwise until it is loose.
- 11. Pull the needle assembly up and out of the needle block and cradle.
- 12. Remove the needle assembly from the autosampler.



Figure 5-12. Removing the Needle from the Cradle

### 5.4.2 Installing the New Sampling Needle Assembly

1. From the top, carefully insert the needle into the needle block (see <u>Figure 5-13</u>). Make sure the needle is vertical and centered over the opening inside the block.



Figure 5-13. Installing the Needle in the Cradle

- 2. Continue inserting the needle, making sure it is lined up with the opening in the cradle. The needle stops when the fitting on the needle reaches the bottom of the needle block.
- 3. Tighten the fitting fingertight.
- 4. Position the yellow sleeve on the tubing in the position measured in <u>Section 5.4.1</u>, <u>Step 5</u>. Orient the tubing clip up or down, depending on the natural curve of the tubing and attach the clip to the tubing where

the yellow sleeve is attached. Screw the clip back onto the needle arm again.

IMPORTANT Do not shorten the sampling needle assembly tubing. The extra length is required to prevent sample from being pulled into the syringe during operation.

IMPORTANT The tubing must curve away from the needle arm assembly when the arm is moved toward the left/front of the autosampler (as viewed from the front). Adjust the tubing length to the needle when the needle is in the left/front corner and all the way down.

- 5. Close the autosampler door and turn on the power. Press **Home** to confirm that the tubing moves correctly.
- 6. Turn off the AS power and open the door.
- Position the second yellow sleeve on the tubing in the position measured in <u>Section 5.4.1</u>, <u>Step 7</u>. Loop the tubing at the second yellow sleeve and attach it to the center bracket on the upper-right side of the autosampler compartment (see <u>Figure 5-11</u>). Use a screwdriver to push in the bracket slightly and secure the tubing.

# **NOTE** The front and back brackets on the upper-right side of the compartment are not used.

- 8. Loop the remaining tubing and hang it on the rear clamp, leaving about 45 cm (18 in) free.
- If you are replacing an 8.5 mL sampling needle assembly (P/N 061267), follow the steps below. If you are replacing a 1.2 mL sampling needle assembly, go on to <u>Step 10</u>.
  - a. Connect the sampling needle line to the coupler that connects it to the larger ID tubing (see Figure 5-11).
  - b. Carefully move the needle arm to the left front corner of the compartment make sure it is all the way down. Verify that there is enough slack in the sampling needle line to prevent the tubing from being pulled.

- 10. If you are replacing a 1.2 mL sampling needle assembly (P/N 054271), follow the steps below:
  - a. Tape the string or tubing that was pulled into the compartment in <u>Section 5.4.1</u>, <u>Step 8d</u> to the free end of the new tubing. Pull the string or tubing, along with the new tubing, out the back of the compartment.
  - b. Slide the bolt, large end first, onto the end of the tubing and then slide the ferrule, small end first, onto the tubing (see Figure 5-14). Slide the ferrule about 10 mm (3/8 in) past the end of the tubing.



Figure 5-14. Installing the Ferrule Fitting and Bolt

- c. Insert the tubing with the bolt and ferrule into port C on the sample syringe. Push the tubing snugly into the bottom of the port.
- d. While holding the tubing with one hand to avoid twisting it, screw the bolt into the port fingertight. This seats the ferrule onto the tubing.
- e. Carefully move the needle arm to the left front corner of the compartment and make sure it is all the way down. Verify that there is enough slack in the sampling needle line to prevent the tubing from being pulled.

#### IMPORTANT

If the fitting leaks when operation is resumed, tighten the bolt slightly. Do not overtighten. Overtightening can damage the inside of the valve.

### 5.4.3 Resetting the Sampling Needle Assembly Usage Counter

After replacing the sampling needle assembly, reset the **Needle Cycles** usage counter from either the AS Wellness panel in Chromeleon or Chromeleon Xpress or on the front panel **USAGE LOG SCREEN** (see <u>Section 5.2</u>).

## 5.5 Removing the Drip Tray

# **NOTE** This section is provided as a reference when performing service procedures that require removing the drip tray.

- 1. Using a 2.5-mm Allen wrench, remove the screw that secures the drip tray to the autosampler compartment and remove the injection valve waste line from the opening in the tray (see Figure 5-15).
- 2. Pull out the tray (toward the front of the autosampler) about 6 mm (1/4 in).
- 3. Tilt the bottom of the tray out slightly and push the tray down slightly until the leak sensor on the inside of the tray (see <u>Figure 5-15</u>) clears the overhang on the front of the autosampler.
- 4. Pull the tray out of the autosampler.
- 5. The leak sensor is hooked onto the tray and its cable is connected inside the autosampler. To remove the drip tray completely from the autosampler, unhook the leak sensor from the tray.



Figure 5-15. Removing the Drip Tray

# 5.6 Installing the Drip Tray

- NOTE This section is provided as a reference when performing service procedures that require removing and reinstalling the drip tray.
- 1. Hook the leak sensor on the tray as shown in <u>Figure 5-16</u>; make sure the sensor is pushed all the way down onto the tray edge.



Figure 5-16. Drip Tray with Leak Sensor Installed

- NOTE Make sure the bottom wire on the leak sensor is not touching the tray. There should be at least a paper-width gap between it and the tray.
- 2. Tilt the top of the tray in and the bottom out, and partially slide the tray back into the autosampler. Make sure the leak sensor clears the front overhang on the autosampler.
- 3. Make sure the needle seal line is in the front of the tray (see Figure 5-15).
- 4. Straighten the tray and push it back into the autosampler until it is flush with the front. Replace the screw.
  - NOTE After installing the drip tray, make sure the tray is not tilted to the left; if it is, liquid will not drain properly during operation.

- 5. Insert the injection valve waste line into the small round opening at the right front corner of the drip tray (see Figure 5-17).
- 6. Route the needle seal line out the autosampler right side slot.



Figure 5-17. Routing the Injection Valve Waste Line to the Drip Tray: Valve Installed in Another Module

To connect the waste line when the AS is connected to two injection valves:

- 1. Insert the waste line from the first injection valve into the small round opening at the right front corner of the drip tray.
- 2. Insert the waste line from the second injection valve into the larger drain line opening (see Figure 5-18).



Figure 5-18. Injection Valve Waste Line Connections for Dual Valves

# 5.7 Replacing the Needle Seal Assembly

Replace the needle seal assembly after about 4,200 cycles of use (the cycle count increments each time the needle is inserted into the inject port). When the AS is operating under Chromeleon or Chromeleon Xpress control, a reminder message is displayed in the Audit Trail when the number of cycles reaches this usage limit. When the AS is operating under front panel control, the **USAGE LOG SCREEN** (see <u>Section C.9.6</u>) displays the number of cycles performed since the assembly was last replaced.

Follow these instructions to install any of the three needle seal assembly versions:

Needle Seal Assembly	Part Number	Use For
18-in needle seal assembly, with 10-32 ferrule fitting	057301	Connecting the AS inject port to the injection valve
18-in needle seal assembly, without 10-32 ferrule fitting	061290	Connecting the AS inject port to two injection valves for simultaneous injections
24-in needle seal assembly with 10-32 ferrule fitting	062564	Use for connecting the inject port of a second AS to injection valve #2 in a dual-ICS-3000 system

- 1. Remove the drip tray (see <u>Section 5.5</u>).
- 2. Disconnect the needle seal line fitting from port S (5) on the injection valve. Using a 5/16-in wrench, turn the fitting counterclockwise to loosen it and then complete the removal with your fingers.

If the AS is equipped with the simultaneous injection option, disconnect the needle seal line from the Y-connector in the autosampler compartment (see Figure 5-19).



Figure 5-19. Simultaneous Injection Option: Disconnecting the Needle Seal Line

If the AS is equipped with the diverter valve for sequential injections, disconnect the needle seal line from port S on the diverter valve (see Figure 5-20).



Figure 5-20. Diverter Valve: Disconnecting the Needle Seal Line

3. Rotate the fitting below the inject port until the slots in the needle seal align with the projections on the top of the inject port (see Figure 5-21).



Figure 5-21. Removing the Needle Seal Assembly

- 4. Push up from underneath and slide the entire needle seal assembly up and out of the inject port.
- 5. Remove the ferrule and bolt fitting from the end of the needle seal line. Save the fitting.

#### IMPORTANT

Do not cut the needle seal line to shorten it, and do not replace it with a different line. The needle seal line is specially constructed and the autosampler will not function correctly if this line is shortened.

6. Thread the new needle seal line (see <u>Figure 5-22</u>) down through the top of the inject port.



Figure 5-22. Needle Seal Assembly

- 7. Align the slots on the needle seal with the projections on the inject port and push the needle seal into the inject port.
- Rotate the bottom fitting until the slots are perpendicular to the projections on the inject port (see <u>Figure 5-23</u>).



Figure 5-23. Inject Port

- 9. Reinstall the drip tray (see <u>Section 5.6</u>).
- Reset the Needle Cycles usage counter from either the AS Wellness panel in Chromeleon or Chromeleon Xpress or on the front panel USAGE LOG SCREEN (see Section 5.2).
- 11. Align the sampling needle in the inject port (see <u>Section 5.8</u>) and then calibrate the inject port volume (see <u>Section 5.9</u>).
- 12. Do not connect the needle seal line to the injection valve until after aligning the inject port and calibrating the inject port volume.

### 5.8 Aligning the Sampling Needle in the Inject Port

Align the sampling needle in the inject port at the following times:

- After replacing the sampling needle assembly (see <u>Section 5.4</u>) and/or the needle seal assembly (see <u>Section 5.7</u>)
- If the needle becomes misaligned

**NOTE** Before aligning the sampling needle, always inspect the needle to make sure it is not bent. If the needle is bent, replace the sampling needle assembly (see <u>Section 5.4</u>).

Two methods are available for aligning the sampling needle:

- Automatic alignment, using Chromeleon or Chromeleon Xpress (Release 6.8 SP2 or later)
- Manual alignment, using the AS front panel screens

This section describes the alignment procedure in Chromeleon or Chromeleon Xpress.

If Chromeleon or Chromeleon Xpress (Release 6.8 SP2 or later) is unavailable, refer to Section D.8 for instructions on how to use the front panel screens to align the needle.

#### To align the sampling needle in Chromeleon:

- 1. Open the Chromeleon Browser and open a panel tabset.
- 2. Select the Autosampler tab to open the AS Control panel (see Figure 5-24).

-	😵 Chromeleon - [Panel Tabset1]								
	File Edit View Workspace Qualification Control Batch Window Help 📃 🖻 🗙								
	☆ ☆ 6 9   田田田田田田田田田田田田田田田田 4 ◎ 参 4   < □ ■ ● 7 6 9   9 @ 0 3   8 @ 計  目 ☆ 1								
	Home   Sequence Lonirol   Status   Autosampler   Gradient Pumps   Detector Lompartment   EL Detector   3D Amp Plot								
	Constituent 200 (Addit That)								
	12:08:40 PM 0.500 (EDet1) Log Background: 372.6 [nA]								
	12.08.40 PM 0.500 (Pump_1) Log Pressure: 0.0 [psi]								
	Status								
	Connect Status: Idle								
	Exclusive Access: ICS-3000_2								
	Mode: Normal								
	Injection Type: Full Inject Valve: LoadPosition								
	Loop Size: 20.0 µl (38) (39) (39) (39) (40) (42)								
	Temperature Control Wellness								
	Tray Set Temp: 10 °C 🕂 Needle Cycles 1738								
	Tran Astual Terrary 10.00 °C Wellness panel Needle align Calibrate IPTV 10 10 10 10 20 20								
	Syringe								
	Prime								
	Prime Syringe: Sample 🛨 Flush Syringe: Sample								
	Prime Volume: 150 µI 🛨 Flush Volume: 100 µI 🛨								
	Prime speed: aspirate 3  Flush speed: aspirate 3								
	dispense 3 - dispense 3 -								
	Prime reservoir. Reservoir A 🗧 More Syringe Commands								

Figure 5-24. Example AS Control Panel

S Needle Alignment		
1 Press the "Inject port align" button. This will disable the AS door lock, and set the needle to the current calibrated position.	Align the X, Y positions. Press Test to drop the needle into the port.         When the needle appears to be correctly aligned, press Verify.         If the needle position is correct, press Done. Otherwise repeat.         X-position       0	
Inject port align	Y-position 0 2 Test Verify Done Lowers needle Homes needle Saves calibration then tests	
Check Dispenses a large volume. If it leaks, press "Lower". If not, press "Home Check" Lower Lowers the needle by 2. Check for leaks again	Set the syringe speed used in your typical application. Set the valve position to the side with the greatest backpressure. Adjust the z-axis height until pressing the Test button does not produce a leak. Then click Done.	
Finish Saves calibration.	0     Move     Test     Done       z-axis height     Moves to position     Dispenses 100 uL     Saves calibration	
When finished, write these calibrated values inside the AS front door       x-axis     0     y-axis     0     z-axis     0		

3. Under **Wellness**, click the **Needle align** button. The Needle Alignment panel appears (see Figure 5-25).

Figure 5-25. Chromeleon Needle Alignment Panel

4. To complete the alignment procedure, follow the instructions on the panel and refer to the following notes:

**Step 1:** For a better view of the needle and inject port during the alignment procedure, you can open the autosampler door after clicking **Inject port align**.



Be aware of unexpected sampling arm movement when operating with the door open.



Soyez conscient de la possibilité de mouvements imprévus du bras d'échantillon lorsqu'il est utilisé avec la porte ouverte.



Achten Sie beim Betrieb mit geöffneter Tür auf unerwartete Bewegungen des Probenarms.

**Step 2:** After clicking **Test**, adjust the **X-position** and/or **Y-position** values until the needle is centered in the inject port.

#### Step 3:

- Select the syringe speed and diverter valve position (if installed) that will provide the maximum backpressure delivered to the needle seal during use. This backpressure must not exceed 690 kPa (100 psi). Refer to <u>Section 3.8</u> for guidelines on selecting syringe speeds.
- After selecting the syringe speed and the diverter valve position (if installed), click **Test**. The AS dispenses 100 µL of fluid into the port.
- Monitor the top of the inject port for fluid. If fluid appears, wipe up the fluid immediately. Then, increase the needle's z-axis height and click Move to move the needle down. Note: Each digit in the z-axis height value moves the needle 0.127 mm (0.005 in).

#### IMPORTANT

Increase the z-axis height value only until no more leaks occur. If the z-axis height is too large, the needle will hit the bottom of the port during operation and can damage the needle or port.

#### Step 4:

- After clicking Check, the AS dispenses 500 μL of fluid into the port. This larger amount of fluid provides a more thorough check than the 100 μL that is dispensed by clicking Test in Step 3.
- After clicking **Finish**, the new calibration values are displayed on the panel. Write these values on the label inside the AS front door.
- 5. If you are performing this procedure after replacing the needle seal assembly, go on to calibrate the inject port volume (see <u>Section 5.9</u>).

## 5.9 Calibrating the Inject Port Volume

Calibrate the inject port volume at the following times:

- After replacing the needle seal assembly
- If reproducibility between injections is poor

Two methods are available for calibrating the inject port volume:

- Automatic calibration using Chromeleon or Chromeleon Xpress (Release 6.8 SP2 or later)
- Manual calibration using the front panel screens

This section describes the calibration in Chromeleon or Chromeleon Xpress.

If Chromeleon or Chromeleon Xpress (Release 6.8 SP2 or later) is unavailable, refer to <u>Section D.9</u> for instructions on how to use the front panel screens to calibrate the inject port volume.

#### **Note for Sequential Injections**

For sequential injections, the volume to be calibrated includes not just the needle seal line, but the path through the diverter valve and the diverter tubing to the injection valve (see Figure 5-26).



Figure 5-26. Sequential Injections: Inject Port Volume Flow Paths

#### To calibrate the inject port volume in Chromeleon or Chromeleon Xpress:

- 1. Open a panel tabset.
- 2. Select the Autosampler tab.
- 3. Under **Wellness**, click the **Calibrate IPTV** button. The Inject Port Volume Calibration panel appears. The panel and calibration procedure differs, depending on whether the AS is configured for normal or sequential mode.
  - If the AS is in normal mode, refer to the instructions on the panel (see <u>Figure 5-27</u>) and to the notes on <u>page 157</u> to complete the calibration procedure.
  - If the AS is in sequential mode, refer to the instructions on the panel (see <u>Figure 5-28</u>) and to the notes on <u>page 159</u> to complete the calibration procedure.

🔚 Inject Port Volume Calibration	
Normal	
Disconnect the needle tubing assembly from the injection valve. Click here to see a Diagram Proceed to the next step only when System is "Idle" idle	
<b>1.</b> Setup Prepares autosampler for calibration	
Press the +1 uL button every few seconds until you observe liquid at the end of the tubing. If you have gone too far, press the -5 uL button and proceed again more slowly. +1 uL -5 uL (performs multiple steps)	
First, press the "Reset" button. When you are completed, press Test. You should observe a small drop of liquid at the end of the tubing. Otherwise, repeat steps 1 and 2. Reset Test	
Press Done to save the calibration value to the AS  Done	
When finished, write this calibrated value inside the AS front do	or
Inject tubing Volume 20	

Figure 5-27. Chromeleon Inject Port Volume Calibration Panel: Normal Mode

#### Notes for calibrating the inject port volume when in normal mode:

• **Preparing:** After disconnecting the needle seal line, route the line outside the autosampler or chromatography compartment.

Make sure the sample syringe is free of bubbles (see Section B.3.14).

• Step 1: Clicking Setup first empties the needle seal line. The AS then dispenses 25 µL into the inject port. This nearly fills the needle seal line (minus a few microliters).

• Step 2: After clicking +1 uL, wait until the sound of the syringe motor stops before clicking +1 uL again.

Continue clicking +1 **u**L until a tiny drop of liquid appears at the end of the needle seal line. If you go too far and more than a single small drop of liquid appears, click -5 **u**L. The AS will empty the line and then fill the line with 5  $\mu$ L less than was cleared. You can then begin clicking +1 **u**L again.

- Step 3: Press Reset and then Test. After clicking Test, verify that a drop of liquid appears at the end of the needle seal line. If no drop appears, or if the drop is a different size than previously observed, click -5 uL and repeat the procedure.
- Step 4: After clicking Done, the new calibration value is displayed on the panel. Write this value on the label inside the AS front door.
- **Finishing:** Reconnect the needle seal line to the injection valve. Tighten the fitting fingertight and then tighten an additional one-quarter turn using the 5/16-in wrench.

	Sequential
Disconnect b	oth tubing assemblies from the injection valves. The calibration must be done for both sides. Click here to see a <b>Diagram</b> Proceed to the next step only when System is "Idle" <b>Idle</b>
1.	Diverter Valve position Position_1
2.	Setup Prepares autosampler for calibration
3.	Enter an estimate of the tubing volume between the diverter valve and inject valve 1 (~2 uL/in; ~0.8 uL/cm) plus the pink tubing volume (between the inject port and diverter valve) $20 \mu$ and press Estimate
4.	Press the +1 uL button every few seconds until you observe liquid at the end of the tubing. If you have gone too far, press the -5 uL button and proceed again more slowly. +1 uL -5 uL
	(performs multiple steps)
5.	First, press the "Reset" button.       When you are completed, press Test. You should observe a small drop of liquid at the end of the tubing. Otherwise, repeat steps 1-3.       Reset     Test
6.	Press Done to save the calibration value to the AS Done
When fi	nished, write this calibrated value inside the AS front door

Figure 5-28. Chromeleon Inject Port Volume Calibration Panel: Sequential Mode

#### Notes for calibrating the inject port volume when in sequential mode:

• **Preparing:** After disconnecting the diverter valve lines from the injection valves, route the lines outside the autosampler or chromatography compartment.

Make sure the sample syringe is free of bubbles (see Section B.3.14).

• Step 1: Select Diverter Valve Position\_1 to calibrate the inject port volume for system 1.

- Step 2: Clicking Setup empties the needle seal line and the diverter line.
- Step 3: Enter an approximate volume using the following formula: 0.8 μL/in (2 μL/in) for the diverter line plus the volume of the needle seal line (which is written on a label on the tubing and inside the AS front door). Then, click Estimate. The AS dispenses the volume entered, which nearly fills the diverter and needle seal lines (minus a few microliters).
  - NOTE The value used for approximating the diverter line volume (0.8  $\mu$ L/cm, 2  $\mu$ L/in) assumes a tubing ID of 0.330 mm (0.130 in). For volume per length values for other tubing IDs, see Section 3.8.
- Step 4: After clicking +1 uL, wait until the sound of the syringe motor stops before clicking +1 uL again.

Continue clicking +1  $\mathbf{u}$ L until a tiny drop of liquid appears at the end of the diverter line. If you go too far and more than a single small drop of liquid appears, click -5  $\mathbf{u}$ L. The AS will empty the line and then fill the line with 5  $\mu$ L less than was cleared. You can then begin clicking +1  $\mathbf{u}$ L again.

- Step 5: After clicking Test, verify that a drop of liquid appears at the end of the diverter line. If no drop appears or if the drop is a different size than previously observed, click -5 uL and repeat the procedure.
- Step 6: After clicking Done, the new calibration value is displayed on the panel. Copy this value to the label inside the AS front door.
- **Calibrate System 2:** Select **Diverter Valve Position\_2** and repeat the calibration procedure for system 2.
- **Finishing:** Reconnect the diverter lines to the injection valves. Tighten the fittings fingertight and then tighten an additional one-quarter turn using the 5/16-in wrench.

### 5.10 Replacing the Leak Sensor

- 1. Remove the drip tray from the autosampler (see Section 5.5).
- 2. Unhook the leak sensor from the drip tray and pull out the leak sensor cable until the connector is visible.
- 3. Unplug the old leak sensor and connect a new leak sensor (P/N 053669).
- 4. Attach the sensor to the drip tray, push the cable inside the autosampler compartment, and reinstall the drip tray (see <u>Section 5.6</u>).
  - NOTE Make sure the bottom wire on the leak sensor is not touching the tray. There should be at least a paper-width gap between it and the tray.
  - NOTE After installing the drip tray, make sure the tray is not tilted to the left; if it is, liquid will not drain properly during operation.
- 5. Calibrate the new sensor (see <u>Section 5.11</u>).

### 5.11 Calibrating the Leak Sensor

If you are calibrating an existing (not a new) leak sensor, complete the following steps before calibrating the leak sensor.

- 1. Remove the drip tray from the autosampler (see <u>Section 5.5</u>) and thoroughly clean and dry the tray and sensor.
- 2. Replace the sensor on the tray and reinstall the drip tray (see Section 5.6).

The leak sensor can be calibrated from the front panel or from Chromeleon.

#### To calibrate the leak sensor from the front panel:

1. Press Menu, 8, and 0 to go to the LEAK SENSOR CALIBRATION AND STATUS screen.



Figure 5-29. Leak Sensor Calibration and Status Screen

2. Press Select  $\triangle$  or Select  $\nabla$  to select CAL and then press Enter.

The current measured value becomes the new calibration value.

#### To calibrate the leak sensor from the Chromeleon Wellness panel:

- 1. Open the Chromeleon Browser and expand the Dionex Templates folder.
- 2. Expand the **Panels** folder and the **Wellness** folder.
- 3. On the right pane of the Browser window, double-click **Dionex\_autosampler\_wellness.pan**. The Wellness panel appears.

4. Click **Connect to Timebase** on the **Control** menu. Connect to the timebase in which the AS is configured. When connected, the controls on the Wellness panel are enabled (see Figure 5-30).

🚟 Autosampler / Timebase: ICS-3000_1 Computer na	ame: AMAR
System Status           Image: System	Diagnostic            • 10:31:11 AM {EGC_1}             • ID:31:11 AM {EGC_1}             • ID:31:11 AM {EGC_1}             • ID:31:11 AM {EGC_1}
AS Autosampler Details Calibration leak detector tray 2.45 V 3/25/2005 external 2.45 V 3/25/2005 inject port volume: 20 µl = 3/25/2005	
Diagnostic Tests	
leak detector	
tray Passed(Dry)	
external Passed(Dry)	

Figure 5-30. Dionex Autosampler Wellness Panel

#### 5. Under Calibration: leak detector, click tray.

The AS calibrates the sensor and then uploads the new value to Chromeleon. Chromeleon stores the new calibration value as the current value.

6. To test the leak sensor, click tray under Diagnostic Tests: leak detector.

### 5.12 Removing the Sample or Prep Syringe Valve

- 1. On the MENU of SCREENS, press **5** and **3** to go to the PLUMBING CONFIGURATION screen.
- 2. Move the cursor to the **SAMPLE SYRINGE CONFIG** field or the **PREP SYRINGE CONFIG** field (depending on which syringe requires initialization).
- 3. Press **Select**  $\triangle$  to select the **INITIALIZE** option.
- 4. Press Enter.

The syringe plunger moves to the initial position and the syringe motor shuts off.

- 5. Unscrew and remove the syringe drive connecting screw and the two screws on the syringe valve (see Figure 5-31).
- 6. Pull the valve and syringe assembly straight out to remove it.
- 7. Holding the syringe at the top fitting, unscrew it from the valve.
- 8. Set the syringe aside in a safe place.



Figure 5-31. Removing the Syringe Valve

# 5.13 Replacing the Sample Syringe Valve

- 1. Disconnect the liquid lines from the existing valve.
- 2. Remove the existing valve (see <u>Section 5.12</u>).
- When the valve is removed, the syringe drive shaft is visible (see <u>Figure 5-32</u>). When the syringe is in the INITIALIZE position (the position selected when the valve was removed), the slot in the drive shaft should be in the position shown in <u>Figure 5-32</u>. The position depends on whether a prep syringe is installed.



Figure 5-32. Sample Syringe Drive Shaft Initial Positions

If the slot in the drive shaft is not in the correct position, the syringe drive may need replacing. Contact Dionex for assistance. Do not attempt to manually turn the drive shaft.

 Check the back of the new sample syringe valve (P/N 054242). The valve drive shaft should be perpendicular to valve ports A and D (see Figure 5-33).

> If it is not, rotate the valve rotor until the drive shaft is in the correct position.



Figure 5-33. Sample Syringe Valve Drive Shaft Initial Position

- 5. To verify that the syringe drive shaft is in the correct position, fill a syringe with deionized water and insert it into port D on the valve. Push liquid through the port and verify that it exits through port C.
- 6. Each valve port must contain a white Teflon washer. Check the inside of each port and insert a washer (P/N 061315), if one is not already present. The washers are supplied with the valve and with the AS Preventive Maintenance Kit (P/N 060581).
- 7. Holding the syringe at the top fitting, screw it into the bottom of the valve at port D and tighten fingertight.
- 8. With valve ports A and D at the 12 o'clock and 6 o'clock positions, respectively, push the valve onto the syringe drive shaft.
- 9. Replace the two valve thumbscrews and partially tighten.
- 10. Carefully align the syringe piston with the connecting screw mount on the syringe drive.
- 11. Insert the syringe drive connecting screw and tighten fingertight. Finish tightening the valve thumb screws.
- 12. Reconnect the valve liquid lines. Tighten the fittings fingertight.

IMPORTANT If a fitting leaks after operation is resumed, tighten it slightly. Do not overtighten; overtightening can damage the inside of the valve.

- 13. On the MENU of SCREENS, press 5 and 3 to go to the PLUMBING CONFIGURATION screen. Set the initial position (see Section 5.3.4).
- 14. On the **PLUMBING CONFIGURATION** screen, move the cursor to the **SAMPLE SYRINGE VOLUME** field. Press **Select**  $\Delta$  or **Select**  $\nabla$  to select the syringe volume. Press **Enter**.

# 5.14 Replacing the Prep Syringe Valve

- 1. Disconnect the liquid lines from the existing valve.
- 2. Remove the existing valve (see Section 5.12).
- 3. When the valve is removed, the syringe drive shaft is visible. When the syringe is in the initial position (the position selected when the valve was removed), the slot in the drive shaft should be in the 11 o'clock position (see Figure 5-34).



Figure 5-34. Prep Syringe Drive Shaft Initial Position

If the slot in the drive shaft is not in this position, the syringe drive may need replacing. Contact Dionex for assistance. Do not attempt to manually turn the drive shaft.

 Check the back of the new prep syringe valve (P/N 054243). The valve drive shaft should be perpendicular to valve ports N and D (see Figure 5-35).



Figure 5-35. Prep Syringe Valve Drive Shaft Initial Position (Back View)

If it is not, rotate the valve rotor until the drive shaft is in the correct position. To check the position, insert the end of a paper clip into valve port N. If the drive shaft is in the correct position, you will be able to insert the paper clip about 1.5 cm into the port. If the drive shaft position is incorrect, you will not be able to insert the paper clip more than about 1 cm into the port. In this case, rotate the valve 180 degrees and recheck the position.

- 5. Each valve port must contain a white Teflon washer. Check the inside of each port and insert a washer (P/N 061315), if one is not already present. The washers are supplied with the syringe valve and with the AS Preventive Maintenance Kit (P/N 060581).
- 6. Holding the syringe at the top fitting, screw it into the bottom of the valve at port S and tighten fingertight.
- 7. With valve ports L and B at the 12 o'clock position and the syringe at the 6 o'clock position, push the valve onto the syringe drive shaft.
- 8. Replace the two valve thumbscrews and partially tighten.
- 9. Carefully align the syringe piston with the connecting screw mount on the syringe drive.
- 10. Insert the syringe drive connecting screw and tighten fingertight. Finish tightening the valve thumbscrews.
- 11. Reconnect the valve liquid lines. Tighten the fittings fingertight.

IMPORTANT If a fitting leaks when operation is resumed, tighten it slightly. Do not overtighten. Overtightening can damage the inside of the valve.

- 12. On the MENU of SCREENS, press 5 and 3 to go to the PLUMBING CONFIGURATION screen. Set the initial position (see Section 5.3.4).
- 13. On the **PLUMBING CONFIGURATION** screen, move the cursor to the **PREP SYRINGE VOLUME** field. Press **Select**  $\Delta$  or **Select**  $\nabla$  to select the syringe volume. Press **Enter**.
## 5.15 Replacing a Sample or Prep Syringe Pump Assembly

The following syringe pump assemblies are available: Sample syringe (P/N 062382), dual sample syringe (P/N 062379), and sample prep syringe (P/N 062365)

- 1. Turn off the AS power.
- 2. Disconnect the liquid lines from the existing syringe valve.
- 3. Unplug the syringe cable from the rear panel connector.
- 4. Lift up the syringe pump assembly and remove it from the organizer.
- 5. Set the new syringe pump assembly into the organizer.
- 6. Connect the liquid lines.
- 7. Plug in the cable.
- 8. Turn on the AS power.

# 5.16 Rebuilding the Injection or Diverter Valve

Rebuild the injection valve after about 2,100 cycles of use. Rebuild the diverter valve after about 1,000 cycles of use. When the AS is operating under Chromeleon control, a reminder message is displayed in the Audit Trail when the number of cycles reaches this usage limit. If the AS is operating under front panel control, the **USAGE LOG SCREEN** (see Section C.9.6) displays the number of cycles performed since the valve was last rebuilt.

- 1. Turn off the liquid flow.
- 2. Disconnect each liquid line connected to the injection valve (see Figure 5-36) or diverter valve (see Figure 5-37).



Figure 5-36. Injection Valve Installed in the Autosampler Compartment



Figure 5-37. Diverter Valve Plumbing Connections

- 3. If you are rebuilding an injection valve installed inside the AS autosampler, follow the steps below to pull the valve away from the compartment slightly, for better viewing of the valve parts during the procedure.
  - a. Remove the drip tray as described in <u>Section 5.5</u>.
  - b. Using an Allen wrench, remove the screw on the bracket that attaches the injection valve to the AS compartment (see Figure 5-37).
  - c. Pull the valve forward far enough to allow you to inspect the injection valve on all sides.
- 4. Follow the instructions provided in the valve rebuild kit (P/N 057896) to replace the rotor seal, isolation seal, and stator face.

Notes on replacing the stator screws (the last step in the valve rebuild kit instructions):

- Before tightening the stator screws, make sure the stator and the stator ring are squarely aligned and the space between them is uniform all the way around the valve.
- If you have trouble gripping the stator screws in order to tighten them, use the Allen wrench held lightly between the thumb and forefinger. Tighten them until you estimate they are fingertight.
- After finger tightening each screw, tighten each an additional one-half turn.

## (IMPORTANT) Do

Do not overtighten the stator screws. Tighten them further in small increments, only if the valve leaks or does not pass qualification.

- 5. If you pulled the valve out from the autosampler compartment, slide the valve back into position in the compartment and reinstall the three bracket screws.
- 6. Reconnect all liquid lines to the valve. Refer to <u>Figure 5-36</u> for the valve flow schematic.

# 5.17 Replacing the Well Plate Tray Thermal Pads

Replace the two thermal pads (P/N 064999) on the well plate tray annually.

- 1. Remove the well plates from the tray.
- 2. Starting at a corner, peel off the existing thermal pad from the tray. The pad is made of a sticky material, but there is no adhesive attaching it to the tray.
- 3. Remove the pad from the other side of the tray.
- 4. Remove the new thermal pads from their bags.
- 5. Thermal pads are shipped with protective covering on both sides. One side is clear and the other is patterned. Peel off the clear covering from the pad.
- 6. Note the two slightly recessed rectangles on the well plate tray (see Figure 5-38).



Figure 5-38. Well Plate Tray with Thermal Pads Removed

- 7. Turn over the pad and align the edges of the pad with the edges of the recessed rectangle on the left side of the tray. Place the pad (sticky side down) in the tray (see Figure 5-39).
- 8. Peel off the protective covering from the top of the pad.
- 9. Install the remaining thermal pad on the right side of the tray.



Figure 5-39. Well Plate Tray with One Thermal Pad Installed

# 5.18 Changing the Main Power Fuses

The fuse holder is part of the main power receptacle on the rear panel of the AS autosampler compartment.

1. Turn off the main power.



HIGH VOLTAGE—Disconnect the main power cord from its source and also from the rear panel of the AS.



HAUTE TENSION—Débranchez le cordon d'alimentation principal de sa source et du panneau arrière du AS.



HOCHSPANNUNG—Ziehen Sie das Netzkabel aus der Steckdose und der Netzbuchse auf der Rückseite des AS.

 A recessed lock is located on each side of the fuse holder (see Figure 5-40). Using a small screwdriver, push each lock toward the center to release it. The fuse holder pops out approximately 0.16 cm (<sup>1</sup>/16 in) when the locks release. When both locks are released, pull the fuse holder straight out of its compartment.



Figure 5-40. Autosampler Compartment Main Power Fuse Holder

3. The holder contains two fuses. Replace both with new 3.15 amp fast-blow IEC 127 fuses (P/N 954745). Dionex recommends replacing both fuses.

- 4. Reinsert the fuse holder into its compartment. The fuse holder is keyed to fit only in its proper orientation. Apply sufficient pressure evenly against the holder to engage the two locks. The holder is flush against the panel when both locks are engaged.
- 5. Reconnect the main power cord and turn on the power.

# A • Specifications

# A.1 Autosampler

#### A.1.1 Electrical

Main Power	100 to 240 Vac, 50/60 Hz; 6 amps. The AS power supply is main
	voltage auto-sensing and requires no adjustment.

**Fuses** Two 3.15 amp fast-blow IEC 127 fuses (P/N 954745)

## A.1.2 Environmental/Physical

Ambient Operating Temperature	10 to 40 °C (50 to 104 °F)
Operating Humidity	10 to 95% relative humidity, non-condensing
Dimensions	58 cm high x 30 cm wide x 46 cm deep (23 in x 12 in x 19 in)
Weight	< 30 kg (< 65 lb)

## A.1.3 Valves (Optional)

Injection	One or two, two-position, six-port, electrically-activated Rheodyne
	valves, PEEK

**Diverter** One two-position, six-port, electrically-activated Rheodyne valve, PEEK

## A.1.4 Injection

Injections per Vial	1 to 99
Vial Sizes Available	0.3 mL polymer vials 1.5 mL glass vials 10 mL PolyVials
Well Plate Sizes Available	Deep-well plate, 96 wells, 2.0 mL Micro-well plate, 96 conical (V-shaped) wells, 0.45 mL Micro-well plate, 96 round (U-shaped) wells, 0.5 mL
Variable Volume Range	<i>If the sample syringe is 1,000</i> μ <i>L or less:</i> 1.0 to 99.9 μL in 0.1 μL increments; 100 to 1,000 μL in 1 μL increments (normal or sequential mode)
	100 to 1,000 $\mu$ L in 1 $\mu$ L increments (concentrate or sequential concentrate mode)
	<i>If the sample syringe is 5,000</i> μ <i>L:</i> 1.0 to 99.9 μL in 0.1 μL increments; 100 to 4,700 μL in 1 μL increments (normal or sequential mode)
	100 to 4,700 $\mu$ L in 1 $\mu$ L increments (concentrate or sequential concentrate mode)
	1,000 to 4,700 $\mu$ L in 1 $\mu$ L increments (simultaneous mode)
	If the sample syringe is 10,000 $\mu$ L: 1.0 to 99.9 $\mu$ L in 0.1 $\mu$ L increments; 100 to 8,000 $\mu$ L in 1 $\mu$ L increments (normal or sequential mode)
	100 to 8,000 $\mu$ L in 1 $\mu$ L increments (concentrate or sequential concentrate mode)
	1,000 to 8,000 $\mu$ L in 1 $\mu$ L increments (simultaneous mode)

<b>Injection Loop Size</b> 25 μL standard; other sizes are available		
Concentrator Column	Dionex ultra-low pressure; maximum injection pressure is 690 kPa (100 psi)	
Maximum Pressure Delivery	690 kPa (100 psi)	

## A.2 Sample Temperature Control (Optional)

Temperature Control Range	4 to 60 °C in 1 °C increments at 24 °C (75 °F) ambient, stable to $\pm 0.2$ °C
Operating Humidity	5 to 90% relative humidity, noncondensing Cooling capacity is slightly reduced at high humidity
Dimensions (Sample Temp. + Autosampler)	65 cm high x 30 cm wide x 46 cm deep (2.75 in x 12 in x 19 in)
Weight (Sample Temp. +	<38 kg (<81 lb)

**NOTE** The temperature inside the autosampler compartment may exceed ambient by up to 2 °C. If samples are temperature sensitive, Dionex recommends installing the sample temperature control option/

Autosampler)

This chapter provides installation instructions for the AS Autosampler, including initial setup instructions, optional component connections (sample preparation option, diverter valve for sequential injections, second AS kit), and USB cable connections for computer control.

If the installation instructions refer you to a manual for another Dionex instrument or chromatography component, you can find the manual on the Dionex Reference Library CD-ROM (P/N 053891), located in the AS Ship Kit (P/N 062380 or P/N 062381).

# **B.1 Facility Requirements**

- Make sure the AS meets the power and environmental specifications listed in <u>Appendix A</u>.
- Install the AS on a sturdy workbench at a height that ensures convenient viewing of the front panel display and access to the interior.
- Place the AS on the workbench to the left of the other system module(s). This allows the shortest liquid line connections between the AS and the chromatography components (injection valve, pump, column, etc.).
- If two AS Autosamplers are to be used with an ICS-3000 Detector/Chromatography module (DC), place one AS on the left side of the DC and one on the right side. Use the Second AS Kit (P/N 062578) to set up the AS on the right side (see <u>Section B.3.7</u>).

# **B.2** Unpacking



Two or more persons should lift the AS, which weighs more than 18 kg (40 lb). Lift the AS only from each side of the cabinet bottom. Lifting from the front door will damage the door hinges.



Au moins deux personnes peuvent soulever l'AS, qui pèse plus de 18 kg (40 lb). Ne soulevez l'AS que par chaque côté du fond de l'armoire. Son soulèvement par la porte du panneau avant endommagera les charnières de la porte.



Der AS wiegt über 18 kg. Daher sollte das Gerät nur von zwei oder mehr Personen angehoben werden. Greifen Sie dazu an beiden Seiten unter das Gerät. Heben Sie den Autosampler nicht an der Vordertür an, da dadurch die Türangeln beschädigt werden könnten.



<ul><li><i>Figure B-3.</i></li><li><i>AS with foam top cap installed</i></li><li>2. Remove the foam top cap.</li></ul>
<ul> <li>Figure B-4.</li> <li>AS with sleeve assembled over cradle</li> <li>Note: The AS is inserted into the bottom foam of the main shipping container. The tubing is coiled in front of the AS.</li> <li>Using the handholds in the cradle, place the AS on the bench.</li> <li>Remove the sleeve (see Figure B-5).</li> </ul>
Figure B-5. AS sitting on cradle (with the sleeve removed) Note: The rear edge of the cradle and the rear edge of the AS are aligned.

<ul> <li><i>Figure B-6.</i></li> <li><i>AS fully wrapped and taped within polyethylene bag</i></li> <li>5. Remove the polyethylene bag (see <u>Figure B-7</u>). Be careful not to damage the drain tube.</li> </ul>
<i>Figure B-7.</i> <i>AS sitting on foam spacer</i> <b>Note:</b> The rear of the AS is aligned with the rear edge of the foam.
<ul> <li>Figure B-8.</li> <li>5-cm (2-in) wide masking tape securing tubing within the recess at the rear of the AS</li> <li>6. Remove the masking tape.</li> <li>7. Remove the coiled tubing inside the recess. Be careful not to damage the tubing.</li> </ul>
<ul> <li>Figure B-9.</li> <li>Coiled inlet tubing inserted into the recess in the rear of AS</li> <li>8. Pull enough of the inlet tubing through to connect to the syringe and reservoir organizer.</li> </ul>



	Figure B-13. Fully assembled dual syringes with bottle
	<ul> <li>Figure B-14.</li> <li>Shipping bracket hooked into frame and attached to sample arm and chassis with screws</li> <li>13. Remove the sturdy tie, the two cap screws, and the bracket Figure B-15.</li> </ul>
CAUTION: REMOVE BEFORE USE	Figure B-15. Shipping bracket with M3 Allen cap screws





After unpacking, save the shipping restraints. To stabilize the sampling arm and prevent damage, always reattach these shipping restraints before moving or shipping the AS.

# **B.3** Autosampler Installation

### B.3.1 Before You Begin

- Install the chromatography system components (pump, detector, column, suppressor, etc.) before installing the autosampler.
- Before connecting the AS to a Chromeleon or Chromeleon Xpress PC, make sure Chromeleon is installed on the PC. Refer to *Installing the Chromeleon Chromatography Management System with a Dionex Ion Chromatograph* (Document No. 031883) for instructions.

## **B.3.2 Installation Checklist**

- □ Verify that all shipping restraints are removed
- $\Box$  Connect the syringe(s) and drain line
- □ Connect the injection valve
- □ (Optional) Connect the AS to the Chromeleon PC or to the Chromeleon Xpress PC
- □ (Optional) Install a diverter valve for sequential injections
- □ (Optional) Install a second AS kit
- $\Box$  Plug the power cord into a grounded power source
- $\Box$  Turn on the system power
- $\Box$  Select the module setup options
- $\Box$  Prime the liquid lines
- $\Box$  Remove bubbles from the syringe

## B.3.3 Connecting the Syringes and Drain Line

- 1. Remove the acorn nut from the fitting on the sampling needle assembly, which extends from the AS rear panel. Gently pull the line out the rear panel until there is enough free tubing (about 45 cm (18 in)) to route the line to the front of the sample syringe.
- 2. Remove the 1/4-28 plug from port C on the sample syringe valve. Make sure the white Teflon washer remains installed inside the port.
- 3. Connect the sampling needle assembly line to port C (see Figure B-19).



Figure B-19. Syringe Connections

- 4. Fill the flush reservoir (P/N 057408) with flush fluid (normally deionized water) and place it in the organizer.
- 5. Connect the line that extends from the flush reservoir cap to the line connected to port E (see Figure B-19).

- 6. If the sample temperature control option is installed, locate the drain line (P/N 055075) in the AS Ship Kit (P/N 062381) and push the line onto the fitting on the lower right front corner of the sampler temperature control unit.
- 7. Place the end of the drain line that extends from the lower right front corner of the autosampler compartment (see <u>Figure B-20</u>) or sample temperature control option into a waste container.



Figure B-20. Autosampler Compartment Drain Line

IMPORTANT

For correct drainage, make sure the tubing is not bent, pinched, or elevated at any point. Do not allow the end of the waste line to be submerged in waste liquid.



Neutralize acidic and caustic wastes before disposal. Dispose of all wastes in accordance with local regulations.



Neutralisez les déchets acides ou caustiques avant de les jeter. Jetez les déchets aux règlements locaux.



Neutralisieren Sie säurehaltige und ätzende Abfälle vor ihrer Entsorgung. Entsorgen Sie alle Abfälle entsprechend den lokalen Bestimmungen.

- 8. Connect the cable from the sample syringe to the **SAMPLE SYRINGE** connector on the rear panel (see Figure B-21).
- 9. If the sample preparation option is installed, connect the cable from the prep syringe to the **PREP SYRINGE** connector on the rear panel.



Figure B-21. Rear Panel Connections

#### **Sample Preparation Option Setup**

- 1. Fill the reagent reservoir(s) (P/N 054808) with the desired reagent(s) and place them in the organizer.
- 2. If you are installing a single reagent reservoir, connect the line that extends from port A on the prep syringe valve to the reservoir cap (see Figure B-22).
- 3. If you are installing additional reagent reservoirs, connect the lines from ports B, C, and/or D to the additional reservoirs.
  - NOTE Port letters A through D on the prep syringe valve correspond to reagent reservoirs A–D on the AS front panel screens and in software controls.



Figure B-22. Sample Preparation Option Connections

### B.3.4 Connecting a Single Injection Valve

- NOTE If your AS is equipped with the simultaneous injection option, see <u>Section B.3.5</u> for connection instructions.
- NOTE If your AS is equipped with a diverter valve for sequential injections, see <u>Section B.3.6</u> for connection instructions.
- 1. Open the AS and chromatography system module doors.
- 2. The needle seal line that extends from the inject port in the autosampler is coiled inside the AS door, on the right side. Route this line through the side slot on the AS (see Figure B-23) and then through the side slot on the chromatography system module to the injection valve.

# **IMPORTANT** Do not cut the needle seal line to shorten it, and do not replace it with a different line. The needle seal line is specially constructed and the autosampler will not function correctly if this line is cut.



Figure B-23. AS Drip Tray

- Connect the needle seal line from the AS to port S (5) on the injection valve (see <u>Figure B-24</u>).
- Locate the green waste line connected to port W (6) on the injection valve (see Figure B-24).



Figure B-24. Injection Valve

- 5. If you are connecting the AS to an ICS-3000 Detector/ Chromatography module (DC), route the waste line from the injection valve through the side slot on the DC and then through the side slot on the AS.
- 6. If you are connecting the AS to an ICS-1000/1500/2000, this green waste line (labeled **AS TRAY**) is connected with a union to a Teflon waste line (see Figure B-25), which is routed through the tubing chase to the chromatography system module rear panel.
- 7. Disconnect the green **AS TRAY** line from the union and route the line to the drip tray in the autosampler compartment.



Figure B-25. Injection Valve Waste Line Union (ICS-1000/1500/2000)

You can either leave the disconnected Teflon waste line and union in the tubing chase, or pull the line out from the front.

- 8. To ensure the green waste line stays in place in the opening in the AS drip tray, sharply bend the line about 12 mm (1/2 in) from the end, being careful not to restrict the inner diameter.
- 9. Push the bent end of the line into the small round opening on the right side of the AS tray (see Figure B-23).

IMPORTANT

Make sure the injection valve waste line is not elevated above the injection valve at any point between the valve and the AS drip tray.

B.3.5 Connecting Two Injection Valves (for Simultaneous Injections)

# Connecting to Two Valves Installed in Chromatography System Modules

This section explains how to plumb two chromatography systems in parallel.

1. Locate the Y-connector, which is installed inside the AS autosampler compartment (see Figure B-26).



Figure B-26. Simultaneous Injection Y-Connector Inside the AS

- 2. Connect one of the lines from the Y-connector to port S (5) on the injection valve of the first chromatography system module (see Figure B-27).
- 3. Connect the other line to port S (5) on the injection valve of the second chromatography system module.



Figure B-27. Injection Valve Connections for Simultaneous Injection: Valves Installed in Two Chromatography System Modules

4. Route the green waste lines connected to port W (6) on each injection valve through the side slots to the AS drip tray. Insert one line into the small round opening in the tray (see Figure B-23). Insert the other line about 12 mm (1/2 in) into the large drain opening in the tray.

#### IMPORTANT

Make sure the injection valve waste lines are not elevated above the injection valves at any point between the valves and the AS drip tray.

#### Connecting to Two Valves Installed in the AS

1. Route the outlet lines from the pump and column in both chromatography systems through the slot on the right side of the AS (see Figure B-28).



Figure B-28. Routing Pump and Column Lines to AS Injection Valves

- 2. Connect the pump outlet line from system #1 to port P (2) on the bottom injection valve (valve #1) and connect the column outlet line from system #1 to port C (3) on valve #1 (see Figure B-29).
- 3. Connect the pump outlet line from system #2 to port P (2) on the top injection valve (valve #2) and connect the column outlet line from system #2 to port C (3) on valve #2 (see Figure B-29).



Figure B-29. Injection Valve Connections for Simultaneous Injection: Valves Installed in the AS

4. Route the green waste lines connected to port W (6) on each injection valve through the side slots to the AS drip tray. Insert one line into the small round opening in the tray (see <u>Figure B-30</u>). Insert the other line about 12 mm (1/2 in) into the large drain opening in the tray.

#### IMPORTANT

Make sure the injection valve waste lines are not elevated above the injection valves at any point between the valves and the AS drip tray.



Figure B-30. Injection Valve Waste Line Connections for Dual Valves

## **B.3.6** Connecting a Diverter Valve (for Sequential Injections)

When the AS is configured for sequential injections, a high-pressure diverter valve must be installed in the second valve position inside the AS. A Diverter Valve Kit (P/N 063294) with the parts needed to install the valve is available. In addition to the kit, you will need a 5.5 mm nut driver to complete the installation.

#### Installing the Diverter Valve

1. Connect the cable from the diverter valve (see <u>Figure B-31</u>) to the matching cable that extends from the lower-right corner inside the autosampler compartment (see <u>Figure B-32</u>). The cables are keyed to fit only one way.



Figure B-31. Diverter Valve Assembly



Figure B-32. Diverter Valve Cable and Mounting Studs

- 2. Dress the cable toward the rear of the AS.
- 3. Align the keyhole openings on the valve assembly with the four studs mounted on the right side of the autosampler compartment (see Figure B-32) and push the assembly onto the studs (see Figure B-33).
- 4. Make sure the cable is not pinched and then slide the assembly back to secure it on the narrow end of the keyholes.
- 5. Using a 5.5 mm nut driver, attach the four nuts with washers to the mounting studs.



Figure B-33. Diverter Valve Assembly Installed

#### **Connecting the Plumbing**

1. Connect the pink needle seal tubing from the AS inject port to port S on the diverter valve (see Figure B-34).



Figure B-34. Diverter Valve Plumbing Connections

2. Connect the blue sample out line from port 1 on the diverter valve to port S on the injection valve of system #1 (see Figure B-35).



Figure B-35. Diverter Valve Plumbing Connections to Injection Valves

- 3. Connect the blue sample out line from port 2 on the diverter valve to port S on the injection valve of system #2.
- 4. Route the green waste lines connected to port W (6) on each injection valve through the side slots to the AS drip tray. Insert one line into the small round opening in the tray (see <u>Figure B-23</u>). Insert the other line about 12-mm (1/2-in) into the large drain opening in the tray.
- Calibrate the inject port volume (the volume of the needle seal and diverter valve line) for each system. Refer to <u>Section 5.9</u> for instructions.

### B.3.7 Connecting a Second AS to a Dual ICS-3000 System

When an ICS-3000 Detector/Chromatography module (DC) is configured with two injection valves, two AS Autosamplers can be connected to the DC. The first AS is installed on the left side of the DC and the second AS is installed on the right side.

A Second AS Kit (P/N 062578) is available for connecting the second AS to the DC. The kit consists of a 24-in needle seal assembly (P/N 062564) and a metal tubing chase (062579) and mounting screws (P/N 057058). The tubing chase routes the needle seal line across the front of the AS compartment to the DC.

- 1. Remove the existing needle seal assembly from the AS and install the 24-inch assembly (see Section 5.7).
- 2. After reinstalling the drip tray, do not fully tighten the drip tray screw.
- 3. Use the two screws to attach the metal tubing chase to the front of the AS compartment (see Figure B-36).
- 4. Route the needle seal line behind the face of the drip tray through the tubing chase to the left side of the AS (see Figure B-36).



Figure B-36. Second AS Installation: Tubing Chase Installation

 Route the needle seal line out the AS compartment through the slot on the left side (see <u>Figure B-37</u>). Verify that the tubing is not pinched when the door is closed.



Figure B-37. Second AS Installation: Needle Seal Line Routed Out the Left Side of the AS

- 6. Align the sampling needle in the inject port (see <u>Section 5.8</u>).
- 7. Calibrate the inject port volume (see <u>Section 5.9</u>).

- 8. Connect the needle seal line to port S on the second injection valve in the DC.
- 9. Locate the green waste line connected to port W (6) on the injection valve (see Figure B-24).
- 10. Route the waste line from the injection valve through the side slot on the DC and then through the side slot on the AS.
- 11. To ensure the green waste line stays in place in the opening in the AS drip tray, sharply bend the line about 12 mm (1/2 in) from the end, being careful not to restrict the inner diameter.
- 12. Push the bent end of the line into the small round opening on the right side of the AS tray (see Figure B-23).

# Make sure the injection valve waste line is not elevated above the injection valve at any point between the valve and the AS drip tray.

13. Calibrate the inject port volume. Refer to <u>Section 5.9</u> for instructions.

### B.3.8 Connecting the AS to Chromeleon or Chromeleon Xpress (Optional)

#### **Before You Begin**

Before connecting the USB cable and turning on the AS power, verify that the following tasks have been completed:

- Chromeleon or Chromeleon Xpress is installed on the PC.
- The Chromeleon or Chromeleon Xpress software license is installed.

#### NOTE Dionex strongly recommends installing Chromeleon or Chromeleon Xpress before connecting the AS to the PC. When you install the chromatography software first, the USB driver for the AS is automatically loaded and the Windows operating system detects the AS when the power is turned on.

If installation is required, refer to *Installing the Chromeleon Chromatography Management System with a Dionex Ion Chromatograph* (Document No. 031883) for instructions. The manual is available in two formats: as a printed manual (shipped with Chromeleon or Chromeleon Xpress) and as an Adobe PDF file (on the Chromeleon CD-ROM and the Dionex Reference Library CD-ROM).

#### **Connecting the USB Cable**

#### IMPORTANT

The USB standard limits the USB cable length to 5 meters (16 ft). Each USB device can be separated from the PC by no more than five hubs. Thus, each USB device can be located no more than 30 meters (32 yds) from the PC.

The AS rear panel (see <u>Figure B-21</u>) provides a USB receptacle for connecting the AS to a USB port on the Chromeleon or Chromeleon Xpress PC or on a USB hub.

Typically, the AS is connected to a USB port on a system module that has an internal hub (for example, an ICS-2000 or ICS-3000 module). The system module is then connected to the USB port on the PC.

#### NOTE If you are installing an AS with an ICS-3000 Ion Chromatography System, refer to *ICS-3000 Ion*

Chromatography System Installation Instructions (Document No. 065032). The manual contains information about USB compatibility issues with ICS-3000 systems.

- 1. Locate the USB cable (P/N 960777) in the AS Ship Kit (P/N 062380 or P/N 062381).
- 2. Plug the "B" connector of the USB cable into the USB receptacle on the rear panel of the AS.
- Plug the "A" connector of the cable into the USB port on one of the following locations: a module with a built-in USB hub (see <u>Figure B-38</u>), an external hub (see <u>Figure B-39</u>), or directly to the PC (see <u>Figure B-40</u>).



Figure B-38. Example USB Connections: Connecting the AS to an Internal Hub on Another Device


Figure B-39. Example USB Connections: Connecting the AS to an External USB Hub



Figure B-40. Example USB Connections: Connecting the AS Directly to the PC

### **B.3.9** Connecting the Power Cord

Connect the power cord (IEC 320 C13) (ordered separately) from the main power receptacle on the rear panel to a grounded power source. The AS power supply is auto-sensing, so no adjustment is required to select the line voltage.



SHOCK HAZARD—To avoid electrical shock, use a grounded receptacle. Do not operate the AS or connect it to AC power mains without an earthed ground connection.



The power supply cord is used as the main disconnect device. Make sure the socket-outlet is located near the AS and is easily accessible.



Operation at AC input levels outside of the specified operating voltage range may damage the AS.



DANGER D'ÉLECTROCUTION—Pour éviter toute électrocution, il faut utiliser une prise de courant avec prise de terre. Ne l'utilisez pas et ne le branchez pas au secteur C.A. sans utiliser de branchement mis à la terre.



Le cordon d'alimentation principal est utilisé comme dispositif principal de débranchement. Veillez à ce que la prise de base soit située/installée près du module et facilement accessible.



STROMSCHLAGGEFAHR—Zur Vermeidung von elektrischen Schlägen ist eine geerdete Steckdose zu verwenden. Das Gerät darf nicht ohne Erdung betrieben bzw. an Wechselstrom angeschlossen werden.



Das Netzkabel ist das wichtigste Mittel zur Stromunterbrechung. Stellen Sie sicher, daß sich die Steckdose nahe am Gerät befindet und leicht zugänglich ist.

### B.3.10 Turning On the Power

Press the power switch in the lower left corner of the front door to turn on the AS power. When the power is turned on, a series of screens are displayed and the autosampler performs a power-up sequence.

- The **COPYRIGHT NOTICE** screen is displayed briefly, and then the **POWER-UP** screen appears.
- While the **POWER-UP** screen is displayed, the AS performs initialization procedures and a series of diagnostic tests. If one of the tests fails, testing stops and the **DIAGNOSTIC TEST** screen is displayed. See Section C.9.2 if this occurs.
- When the initialization procedures are complete and the AS has passed the diagnostics, the **INSTALLED OPTIONS** screen appears. After a few seconds (or if you press a button), the **MAIN** screen is displayed. The power-up sequence is then complete.

### **B.3.11 Selecting Module Setup Options**

Press Menu and 5 to go to the MODULE SETUP MENU (see Figure B-41).



Figure B-41. Module Setup Menu Screen

#### Set the Time and Date

1. On the **MODULE SETUP MENU**, press **4** to go to the **TIME/DATE** screen.



Figure B-42. Time/Date Screen

- The SET TIME field uses a 24-hour time format (hour:minutes:seconds). Press numeric buttons to enter the time directly or press Select △ or Select ▽ to increase or decrease the current values.
- 3. Move the cursor to the **DATE** field.
- Press numeric buttons to enter the day and year directly, or press
   Select △ or Select ⊽ to increase or decrease the current values. Press
   Select △ or Select ⊽ to select the month.
- 5. After selecting the last option, press **Enter** before exiting the screen.

#### Select the Sample Mode

1. On the MODULE SETUP MENU, press 5 to go to the SYSTEM PARAMETERS screen (see Figure B-43).



Figure B-43. System Parameters Screen Example: Normal Mode Selected

2. Move the cursor to the **SAMPLE MODE** field and select the mode (see <u>Table B-1</u>). Refer to <u>Section 3-16</u> for details about the AS sample modes.

SAMPLE MODE	DESCRIPTION
NORMAL (default)	Injections to a single system
CONCENTRATE	Injections to a single system when the sample loop is replaced by a concentrator column
SIMULTANEOUS	Injections to two systems simultaneously
SEQUENTIAL	Injections to two systems in sequence
SEQUENTIAL CONCENTRATE	Injections to two systems in sequence when the sample loop is replaced by a concentrator column
-	

Table B-1. AS Sample Modes

#### Select the Method and Flush Syringe Speeds

Refer to the tables in <u>Section 3.8</u> to select the correct method and flush syringe speeds to ensure the backpressure generated does not exceed 690 kPa (100 psi).

- If the AS is under Chromeleon or Chromeleon Xpress control, set the syringe speeds on the Control panel or in a program. See <u>Section 3.7</u> for details.
- If the AS is *always* under front panel control (it is never connected to Chromeleon or Chromeleon Xpress), set the syringe speeds on the **SYSTEM PARAMETERS** screen. See <u>Section D.7</u> for details.

#### **Select Plumbing Configuration Options**

1. On the MODULE SETUP MENU, press **3** to go to the PLUMBING CONFIGURATION screen (see Figure B-44).



Figure B-44. Plumbing Configuration Screen Example

2. Move the cursor to the **SAMPLE SYRINGE VOLUME** field and select the volume of the sample syringe installed.

# NOTE For the simultaneous option, the sample syringe size must be either 5,000 or 10,000 µL.

- 3. If the sample preparation option is installed, move the cursor to the **PREP SYRINGE VOLUME** field and select the volume of the prep syringe.
- 4. Move the cursor to the **LOOP SIZE V1** field and enter the size of the sample loop installed on the injection valve.

- NOTE The upper limit for the loop size depends on the volume of the sample syringe installed: 500  $\mu$ L (if the syringe volume is  $\leq 1$  mL), 4,000  $\mu$ L (if the syringe volume is 5 mL), and 8,000  $\mu$ L (if the syringe volume is 10 mL).
- 5. For the simultaneous or sequential mode, move the cursor to the **LOOP SIZE V2** field and enter the size of the sample loop installed on the second injection valve.

#### Select the Wait Operation (for Front Panel Operation Only)

- NOTE Do not complete this section if the AS will be controlled by Chromeleon or Chromeleon Xpress. The software enables the wait function automatically and ignores the setting on the SYSTEM PARAMETERS screen.
- 1. Go to the **MODULE SETUP MENU** and press **5** to go the **SYSTEM PARAMETERS** screen.



Figure B-45. System Parameters Screen

- 2. Move the cursor to the **WAIT** field and press **Select**  $\triangle$  or **Select**  $\nabla$  to select either **ENABLED** or **DISABLED**.
  - If the AS will be controlled by another instrument through TTL or relay connections, set the **WAIT** field to **ENABLED**.
  - If the AS will control other modules (system master) through TTL or relay connections, set the **WAIT** field to **DISABLED**
- 3. Press Enter.

#### Notes

- When Wait is enabled, the AS waits for a continue command (either from the front panel **Hold/Run** button or from a TTL input) before performing the next line in a schedule of injections. See <u>Section 2.7.1</u> for details.
- When Wait is disabled, a schedule of injections is started by pressing the **Hold/Run** button. The AS then performs all injections in the schedule without waiting for any continue commands. See <u>Section 2.7.4</u> for details.

#### B.3.12 Configuring the AS in Chromeleon or Chromeleon Xpress

If the AS is connected to a Chromeleon or Chromeleon Xpress PC, refer to *Installing the Chromeleon Chromatography Management System with a Dionex Ion Chromatograph (IC)* (Document No. 031883) for detailed instructions on how to configure the AS in the software. In summary:

- Install the software, enter the license, and connect the USB cable before turning on the AS power.
- When you start the AS after connecting to the PC, the PC detects the new USB device.
- You are given an option to either create a new timebase or to add the AS to an existing timebase.
- An AS can be included in the following types of timebases:

Single: A single timebase consisting of one system (for example, pump, detector, eluent generator, and autosampler).

Simultaneous: A single timebase consisting of two separate systems (for example, a pump and detector used for anions and another pump and detector used for cations) and a single AS equipped with the simultaneous injection option.

Sequential: Two single system timebases in which the AS is shared between the two systems.

# NOTE If you are configuring an AS for simultaneous injections, refer to <u>Section B.5</u> for additional

# Chromeleon or Chromeleon Xpress setup instructions.

• After configuring the AS in one or two timebases, you can start Chromeleon or Chromeleon Xpress and connect to a Control panel (see <u>Figure B-46</u>). For Chromeleon Xpress, connection to a panel is automatic.

🛠 Chromeleon - [Panel Tabset1]	
File Edit View Workspace Qualification Control Batch Wi	ndow Help
D 📽 🖬 📲 🛅 🗇   X 🖻 📾 🙌 🚺 💿 🔍	义 ジ 0 0   田田臣関國國戰 2 □   ◎ 昭■■■□ 2 ◎ 参 +   < ■
	ICS-3000_2 23 **
Home Sequence Control Status Autosampler Gradient Pumps	; Detector Compartment EC Detector 3D Amp Plot
System Log (Audit Trail)	
2:08:40 PM 0.500 (EDet1) Log Background: 372.6 [nA]	
2:08:40 PM 0.500 (Pump_1) Log Pressure: 0.0 (psi)	
Image: Contract of the second seco	
Status	
Connect Status: Idle	
Exclusive Access: ICS-3000_2	
Mode:	Normal
Injection Type: Full Inject V	alve: LoadPosition (43 (44 (46 (47 (48 (49
Loop Size: 20.0 µl	38 37 38 39 40 41 42
Temperature Control Wellness	29 30 31 32 33 34 35
Tray Set Temp: 10 °C 🛨 Needle Cy	rcles 1738
Tray Actual Temp: 10.00 °C Wellness p	anel Needle align Calibrate IPTV (15) (16) (17) (18) (19) (20) (21)
Syringe	
Prime	Flush
Prime Syringe: Sample 🛨	Flush Syringe: Sample
Prime Volume: 150 µl 🛨	Flush Volume: 100 µl 🚍
Prime speed: aspirate 3 -	Hush speed: aspirate 3
dispense 3 🗸	dispense 3 💌
Prime reservoir: Reservoir_A	More Syringe Commands

Figure B-46. Example AS Control Panel

### **B.3.13 Priming the Liquid Lines**

This procedure explains how to prime the liquid lines from the Chromeleon or Chromeleon Xpress Control panel. If you prefer, you can use the **FLUSH** screen on the front panel to prime the lines (see <u>page 217</u>).

# NOTE To reduce bubble formation while priming, degas the flush solution.

1. Open the Chromeleon or Chromeleon Xpress Control panel (see Figure B-47).

Syringe			4				
Prime		Flush					
Prime Syringe: Sa	mple 🕂	Flush Syringe:	Sample				
Prime Volume: 20	00 µl 🕂	Flush Volume:	100 µl 🕂				
Prime speed: aspirate 5	•	Flush speed: aspirate	5 -				
dispense 4	•	dispense	4 -				
Prime reservoir:	sh_Reservoir 📫	More Syringe Con	nmands				

Figure B-47. Prime Commands on the Control Panel

- 2. Enter the **Prime Volume**. Refer to <u>Table B-2</u> or <u>Table B-3</u> for the suggested volumes.
- 3. If the sample preparation option is installed, select **Prep** in the **Prime Syringe** list and select **Flush\_Reservoir** in the **Prime reservoir** list.
- 4. Click Prime.

The flush reservoir line and sampling needle assembly are primed (see <u>Figure B-48</u>). If the sample preparation option is installed, the reagent holding loop is also primed (see <u>Figure B-49</u>).

5. Check for bubbles in the lines and repeat the priming procedure, if necessary, to eliminate the bubbles.

- 6. If the sample preparation option is installed, follow these steps to prime the lines from each installed reagent reservoir (see Figure B-49):
  - a. Select the reagent reservoir, enter 2000  $\mu$ L in the Prime Volume field, and press Prime.
  - b. Repeat Step 6 for each additional reagent reservoir.
- When priming is complete, there are often small air bubbles in the top area of the syringe. Remove the bubbles before proceeding. See <u>Section B.3.14</u> for instructions.

#### **Suggested Priming Volumes**

Sample Syringe Size	Suggested Priming Volume
≤1,000 µL	2,000 to 2,500 µL
5,000 μL	9,999 μL <sup>a</sup>
10,000 µL	19,998 μL <sup>b</sup>

# Table B-2. Suggested Priming Volumes(Sample Preparation Option Not Installed)

- a. The maximum value that can be entered.
- b. Run two priming cycles of 9,999  $\mu$ L each.

Reservoir	Prep Syringe Size	Suggested Priming Volume
Flush	250 μL	2,000 µL
	500 μL	2,000 µL
	1,000 µL	4,000 μL
	2.5 mL	5,000 µL
	5 mL	9,999 μL <sup>a</sup>
	10 mL	19,998 μL <sup>b</sup>
A, B, C, and D <sup>c</sup>	All	2,000 µL

# Table B-3. Suggested Priming Volumes(Sample Preparation Option Installed)

- a. The maximum value that can be entered.
- b. Run two priming cycles of 9,999 µL each.
- c. Repeat the priming procedure for each reagent reservoir.



Figure B-48. Sample Syringe Fluid Schematic: System without the Sample Preparation Option



Figure B-49. Sample Syringe Fluid Schematic: System Without the Sample Preparation Option

#### Priming Procedure Using the Front Panel Flush Screen

- 1. On the MAIN screen, set the AS to LOCAL mode and DIRECT CONTROL.
- 2. Press Menu and 6 to go to the FLUSH screen (see Figure B-50).
- 3. Enter the desired **PRIME VOLUME**. Refer to <u>Table B-2</u> or <u>Table B-3</u> for the suggested volumes.
- 4. If the sample preparation option is installed:
  - a. Move the cursor to the **PRIME RESERVOIR** field and select the reservoir.
  - b. Move the cursor to the **PRIME SYRINGE** field and select the **PREP** syringe.

If the sample preparation option is not installed, the **PRIME RESERVOIR** and **PRIME SYRINGE** fields default to **FLUSH** and **SAMPLE**, respectively, and cannot be edited.

- 5. Move the cursor to the **PRIME ACTION** field and select **PRIME**.
- 6. Press **Enter** to start the prime cycle.

FLUSH/PRIME						
FLUSH PRIME						
VOLUME:	400	uL	<b>VOLUME:</b>	2000 uL		
<b>RESERVOIR:</b>	FLUS	н	<b>RESERVOIR:</b>	RES A		
SYRINGE:	SAMP	LE	SYRINGE:	PREP		
ACTION:	READ	Y	ACTION:	PRIME		
Help Prompt						

Figure B-50. Initiating a Priming Cycle

- 7. Repeat the procedure as required (see the previous priming guidelines).
- When priming is complete, there are often small air bubbles in the top area of the syringe. Remove the bubbles before proceeding. See <u>Section B.3.14</u> for instructions.

### B.3.14 Removing Bubbles from the Syringe

- 1. Fill a container with isopropyl alcohol.
- 2. Using a flathead screwdriver, unscrew and remove the syringe drive connecting screw (see Figure B-51).
- 3. Hold the syringe at the top and bottom and unscrew it from the valve.
- 4. Point the threaded end up into the air. Push the syringe plunger up and pull down rapidly with short movements to dislodge the bubbles. Repeat until all bubbles are dislodged.
- 5. Place the threaded end of the syringe into the isopropyl alcohol and slowly pull out the plunger to draw a full syringe volume into the syringe.



Figure B-51. Removing the Syringe

- 6. Keeping the syringe in the liquid, slowly push the syringe plunger until all bubbles exit the syringe. Then fill the syringe.
- 7. Alternatively, fill the syringe with isopropyl alcohol and then remove the syringe from the liquid. Hold the syringe with the threaded end up under a stream of deionized water. Push the syringe plunger up and pull down until the bubble is removed.
- 8. Remove the syringe from the liquid. While holding it vertically, push a small amount of liquid out the top to make sure there is no air present.
- 9. Position the syringe under the valve.
- 10. Holding the syringe at the top and bottom, screw it back into the valve and tighten fingertight.

- 11. Align the syringe piston with the connecting screw mount on the syringe drive.
- 12. Insert the syringe drive connecting screw removed in <u>Step 2</u> and tighten fingertight.

# **B.4 Injection Valve Plumbing**

The injection valve is plumbed at the factory with all tubing and fittings for connections to the pump, inject port, and column.

A 25  $\mu$ L PEEK sample loop (P/N 042857) is installed between ports 1 and 4. Other sample loop sizes are available. Contact Dionex for information.

Figure B-52 shows the valve connections. This information is for reference, should you need to replace any tubing or fittings.



Figure B-52. Injection Valve Plumbing

### B.5 Setting Up Chromeleon or Chromeleon Xpress for Simultaneous Injections

### **B.5.1** Assigning Unique Names to Devices

NOTE If the AS will be connected to a dual ICS-3000 system, you may skip this section. The Chromeleon Server Configuration program automatically assigns unique names to each device in a dual ICS-3000.

When you add a device to a timebase, the Chromeleon Server Configuration program assigns a default name to the device. For example, **Pump** for an analytical pump, **ECD** for a conductivity detector, **Pump\_ECD** for a combined pump and detector, and **Sampler** for an autosampler. In addition, other components (relays, TTLs, injection valves, etc.) are assigned device names also.

If you assign multiple devices of the same type to a timebase, you must edit the device names to ensure that each one is unique. The Chromeleon Audit Trail displays error messages that identify duplicate device names in a timebase, for example:

▲ 11:45:59 The symbol "Pump\_ECD" already exists.

# NOTE Chromeleon automatically assigns unique names to the two injection valves in the AS.

To rename a device, open the device Properties dialog box by doubleclicking the device in the timebase. The **General** tab page displays the device name. In the **Device Name** edit box, add \_2 to the existing name. In the example below, the device name for the second ICS-2000 in a timebase is renamed **Pump\_ECD\_2**. This device will be identified as System #2 on the Control panel.

Dionex IC5-2000 System
Solvents Eluent Generator State Devices Error Levels Demo Chromatogram TTL Inputs Trend Calibration Diagnostics General Options Signals Head Type & Limits
Device Name: Pump_ECD_2
Mode <u>M</u> odule Serial No:          Moduleware version: <u>M</u> oduleware version: <u>M</u> odule version: <u>M</u> odule version: <u>M</u> odule version: <u>M</u> odule version: <u>M</u> oduleware version:
OK Cancel Apply Help

In addition to renaming the device name on the **General** tab page, you also need to rename signals and other devices associated with System #2. For example, if System #2 is an ICS-2000 (as in the previous example), select the **Signals** tab, click **Change**, and add **\_2** to the signal name. Repeat for the remaining signals. See the following example:

# NOTE You only need to rename the enabled signals (i.e., the signals with checks next to their names).

Dionex ICS-2000 System				X
Solvents Eluent Gene Demo Chromatogram TTL I General Dptions	erator   nputs   (	State Devic Trend   Ca Signals	es   alibration   Head Ty	Error Levels Diagnostics ype & Limits
Name	Unit	Factor		
✓ ECD_1_2	μS	1.00000		
ECD_Total_2	μS	1.00000		
Channel_Pressure_2	psi	1.00000		
Deactivate check boxes to remo	ve unused	channels from	Online.	
	<u>C</u> hange			
OK	C	ancel	Apply	Help

Also, select the **State Devices** tab and add \_2 to the name of each enabled device. Press **F2** to edit a device name. See the following example:

)ionex ICS-2000 System
Demo Chromatogram TTL Inputs Trend Calibration Diagnostics General Options Signals Head Type & Limits Solvents Eluent Generator State Devices Error Levels
Name
✓ Pump_ECD_Relay_1_2
✓ Pump_ECD_Relay_2_2
✓ Pump_ECD_TTL_1_2
Pump_ECD_TTL_2_2
✓ Pump_InjectValve_2
✓ Pump_ECD_Analog_Out_2
Deactivate check boxes to remove unused relays from Online.
Hit F2 to edit selection.
OK Cancel Apply Help

# B.5.2 Creating Sequences, Programs, and Quantification Methods

Follow these guidelines when creating sequences, programs (PGM file), and quantification methods (QNT file) for simultaneous injection:

- Create one sequence that includes calibration standards for both systems and the unknown samples.
- Create a PGM file for the unknowns that includes commands for controlling both systems.
- Calibrate each system separately by creating separate calibration PGM files.
- Create a single QNT file that includes the components from both systems.

#### Creating a Method File for Systems Sharing an AS

For simultaneous injection analyses, create a single method file with all components for each detector channel included in one component list. Use the **Duplicate Column** command on the Peak Table, Amount Table, and Peak Tracking pages of the method to create two extra **Ret. Time** columns; associate each column with a different channel.

# NOTE To add a column, right-click in the Ret. Time column and select the Columns>Duplicate Column command on the context menu.

For detailed instructions, refer to Chromeleon Help.

In the following example Peak Table, the **Peak Name** column includes the components from two detector channels: one for cations and one for anions. The **Ret. Time ECD\_1** column is associated with the cation detector channel and displays only the cations. The **Ret. Time ECD\_2** 

<u>2</u>	AMAR_local\System_A\Shared_CC80						_ 0	×		
No.	Peak Name	Ret.Time	Ret.Time ECD_1	Ret.Time ECD_2	Window	Standard	Int.Type	Cal.Type	Peak Type	
1	Lithium		3.267 min		0.250 AG	External	Area	Lin	Auto	
2	Fluoride			3.700 min	0.100 AG	External	Area	QOff	Auto	
3	Sodium		3.900 min		0.250 AG	External	Area	Lin	Auto	
4	Ammonium		4.400 min		0.250 AG	External	Area	QOff	Auto	
5	Chlorite			5.087 min	0.250 AG	External	Area	QOff	Auto	
6	Potassium		5.560 min		0.250 AG	External	Area	Lin	Auto	
7	Bromate			5.770 min	0.250 AG	External	Area	QOff	Auto	
8	Chloride			6.500 min	0.100 AG	External	Area	QOff	Auto	
9	Nitrite			7.300 min	0.150 AG	External	Area	QOff	Auto	
10	Bromide			8.100 min	0.200 AG	External	Area	QOff	Auto	
11	Magnesium		8.400 min		0.250 AG	External	Area	Lin	Auto	
12	Calcium		10.500 min		0.250 AG	External	Area	Lin	Auto	
13	Chlorate			10.700 min	0.250 AG	External	Area	QOff	Auto	
14	Nitrate			12.200 min	5.000 RG	External	Area	QOff	Auto	
15	Phosphate			16.400 min	0.250 AG	External	Area	QOff	Auto	
16	Sulfate			18.000 min	0.250 AG	External	Area	QOff	Auto	-
•	A Detection	n λ Peak Tab	le 🖌 Amount	Table 🖌 Pea	•					Γ

column is associated with the anion detector channel and displays only the anions.

If two anion (or two cation) detector channels were being used, the measured retention time for each anion (or cation) would appear in both **Ret. Time** columns, as in the following example:

Z	EM_CD\DEM	0\ICS-90\A9	59HC_A54A9	5C Simultane	ous\Sim	ultaneous	_AS9HC_	AS4ASC	_ 🗆	×
No.	Peak Name	Ret.Time	Ret.Time ECD_1	Ret.Time ECD_2	Window	Standard	Int.Type	Cal.Type	Peak Type	
1	Fluoride		1.000 min	3.700 min	0.100 AG	External	Area	QOff	Auto	
2	Chloride		1.600 min	6.500 min	0.100 AG	External	Area	QOff	Auto	
3	Nitrite		1.900 min	8.090 min	0.150 AG	External	Area	QOff	Auto	
4	Bromide		3.000 min	10.700 min	0.200 AG	External	Area	QOff	Auto	
5	Nitrate		3.600 min	12.800 min	0.250 AG	External	Area	QOff	Auto	
6	Chlorite			5.087 min	0.250 AG	External	Area	QOff	Auto	
7	Phosphate		5.400 min	16.400 min	0.250 AG	External	Area	QOff	Auto	
8	Bromate			5.770 min	0.250 AG	External	Area	QOff	Auto	
9	Sulfate		7.200 min	17.800 min	0.250 AG	External	Area	QOff	Auto	
10	Chlorate			11.400 min	0.250 AG	External	Area	QOff	Auto	
┓	✓ Peak Table A Amount Table Peak Table A									

This appendix illustrates and describes the screens available for display on the front panel of the AS Autosampler. Figure C-1 illustrates the hierarchy of screens.



Figure C-1. AS Menu Structure

### C.1 Menu of Screens

Pressing **Menu** displays the **MENU of SCREENS**, which provides top-level access to the menu structure.



Figure C-2. Menu of Screens

There are two ways to select a screen from a menu:

- Move the cursor to the field containing the screen number and press Enter.
- Press the numeric button on the front panel keypad that corresponds to the screen number on the menu.

To display a brief description of each screen, press Help.

- NOTE If no keypad buttons are pressed within a 2-hour period, the backlight automatically turns off. To turn the backlight on again, press any button on the keypad. Press the button a second time if you want to activate its function.
- NOTE If the simultaneous injection mode is enabled, several screens change to allow control of two injection valves. Both versions of these screens are described in this appendix.

# C.2 Main Status Screen

Use the **MAIN STATUS** screen to:

- Display the status of basic operating parameters
- Select operating and control modes



Figure C-3. Main Screen

Screen Field	Description					
Status Line (top of screen)	The top line on the screen displays the instrument's current operating status. The following status values are displayed as they occur:					
	IDLE	No method is currently running.				
	PREPARING TO RUN SCHEDULE	The AS is performing pre-schedule operations, such as flushing.				
	HOLDING	The schedule and method are on hold.				
	CHECKING FOR SAMPLE VIAL PRESENCE	At the beginning of a method, the AS checks that the specified sample vial is present.				
	PIPETTING	The AS is pipetting from one vial to another during the method's sample prep steps.				
	DISPENSING	The AS is dispensing from a reservoir during the method's sample prep steps.				
	MIXING	The AS is mixing the vial contents during the method's sample prep steps.				
	FLUSHING	The needle is being flushed (either during sample prep or between schedule lines).				
	DELAYING xx.x min	The AS is pausing for the period of time specified in the sample prep phase of the method.				

Screen Field	Description	
	DILUTE	The AS is performing a dilution as part of the sample prep phase of the method.
	REAGENT PRIME	The AS is priming the lines with reagent.
	REAGENT FLUSH	The AS is flushing the inject port with reagent.
	CONCENTRATE	The AS is loading sample onto the concentrator column.
	WAITING	The AS is waiting for a command to continue running a method. The command can be from the front panel (pressing <b>Hold/Run</b> ), from Chromeleon or Chromeleon Xpress, or from a TTL or relay input.
	WAITING FOR RUN TO FINISH	The AS has completed the overlapped sample prep steps and is waiting for the timed events of the previous method to be completed.
	EXECUTING SETUP AND INIT STEP	The AS is executing the method setup conditions and the <b>INIT</b> step of the method's timed events.
	WAITING FOR CYCLE TIME TO EXPIRE	The AS is waiting for the cycle time to expire before starting the method's timed events.
	WAITING FOR TEMP TO STABILIZE	The AS is waiting for the tray temperature to stabilize before starting the method's timed events.
	EXECUTING TIMED EVENTS	The AS is executing the method's timed events. This status message is displayed until a load or inject step is executed.
	LOADING LOOP	Sample is being loaded into the loop during the method's timed events.
	LOADING CONCENTRATOR	Sample is being loaded onto the concentrator during the method's timed events.
	INJECTING	Sample is being injected during the method's timed events.
	ABORTING, PLEASE WAIT	The AS is performing operations necessary to abort the schedule.

Screen Field	Description		
	REMOTE COMMAND IN PROGRESS	The AS is performing a command while under Chromeleon or Chromeleon Xpress control.	
	REMOTE DIAGNOSTIC IN PROGRESS	The AS is performing a diagnostic command while under Chromeleon or Chromeleon Xpress control.	
	REMOTE CALIBRATION IN PROGRESS	The AS is performing a calibration command while under Chromeleon or Chromeleon Xpress control.	
VIAL#	The number of the vial the injection volume is taken from. The AS updates the vial number after executing the <b>INIT</b> step of the method's timed events.		
INJ#	The number of injections completed using the current vial on the current schedule line followed by the total number of injections scheduled for this vial.		
TRAY °C	If the sample temperature control option is installed, this field displays the current tray temperature. A symbol to the right of this field indicates the temperature status (see below). If the sample temperature control option is not installed, the field displays "" and it is not illuminated.		
	<ul> <li>The temperature is stabilized</li> <li>↑ The temperature is increasing</li> <li>↓ The temperature is decreasing</li> <li>− The temperature control option is off</li> </ul>		
TIME	The number of minutes since the last injection. The clock counts up until the next injection begins or until the maximum time of 999.9 minutes is reached.		
VOL	The volume of sample to be injected.		
Operating Mode LOCAL or LOCKED RMT (remote)	Select <b>LOCAL</b> mode for control from the front panel. When the AS is connected to Chromeleon or Chromeleon Xpress, connecting the AS to a Control panel automatically selects <b>LOCKED RMT</b> . In Locked Remote mode, the front panel keypad is disabled to prevent any changes to operating parameters. To return the AS to Local mode, disconnect the AS from the Control panel, or turn off the AS power.		

Screen Field	Description
Control Mode SCH (schedule) or DIRECT CONTROL	Press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to select <b>SCH</b> (schedule) or <b>DIRECT</b> <b>CONTROL</b> mode. When the AS is in <b>LOCKED RMT</b> mode, Direct Control is always selected. In Schedule mode, the screen displays the number of the currently running schedule, the schedule line, and the method. Enter the schedule number to run (1 to 9) in the <b>SCH</b> field.

### C.3 Detail Status Screen

Use the **DETAIL STATUS** screen to:

- Display detailed information about the status of AS operating parameters. This screen displays all the information shown on the **MAIN STATUS** screen, plus additional parameters.
- Set several parameters for Direct Control operation.

NOTE See <u>Section C.2</u> for descriptions of the parameters common to the MAIN STATUS and the DETAIL STATUS screens.

DETAIL		NJECT	ING			
VIAL#: 1	-	TIME:	0.0	min	TTL1	0
INJ#: 1/3	•	VOL:	25	uL	TTL2	1
METHOD: 01		TRAY:	P10 m	L	RLY1	0
<b>TRAY: 25</b>	20 °C↑	LOOP:	25.0	uL	RLY2	0
		NORM/	AL FU	LL	INJ VLV	Ĺ
LOCAL	S	CH 1	LINE	1		
Help prompt						

Figure C-4. Detail Status Screen

DETAIL	INJECTING	
<b>VIAL#: 1</b>	TIME: 0.0 min TTL1	0
INJ#: 1/3	VOL: 25 uL TTL2	1
METHOD: 01	TRAY: P10 mL RLY1	0
<b>TRAY: 25</b>	20 °C↑ LOOP: 100 / 25 uL RLY2	0
	SIMULTANEOUS INJ VLV 1	L
LOCAL	SCH 1 LINE 1 INJ VLV 2	L
Help prompt		

Figure C-5. Detail Status Screen: Simultaneous Injection Mode

					_
DETAIL	INJE	CTING			
<b>VIAL#: 1</b>	TIM	E: 0.0	min	TTL1	0
INJ#: 1/3	vo vo	L: 25	uL	TTL2	1
METHOD: 01	TRA	Y: P10 m	۱L	RLY1	0
<b>TRAY: 25</b>	20°C1 CON	ICENTR/	ATOR	RLY2	0
				INJ VLV	L
LOCAL	SCH 1	LIN	E 1		
Help prompt					

Figure C-6. Detail Status Screen: Concentrate Mode

Screen Field	Description	Values	
TRAY °C	If the sample temperature control option is not installed, "" is displayed and the field is not illuminated.	4 to 60 °C (default=20)	
If the sample temperature control option is installed, this field allows you to select the tray temperature set point in °C. The actual measured temperature and a symbol indicating the temperature status are displayed to the right of this. = The temperature is stabilized ↑ The temperature is increasing ↓ The temperature is decreasing The temperature control option is off			
	<ul> <li>The temperature is stabilized</li> <li>↑ The temperature is increasing</li> <li>↓ The temperature is decreasing</li> <li>− The temperature control option is off</li> </ul>		
	Pressing <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ turns the tray temperature control on and off. The default is off. When on, the default set point is 20 °C.		

Screen Field	Description	Values
TRAY (type)	The tray type in use.	<b>NONE, 2 mL,</b> <b>P10 mL</b> (10 mL plastic), <b>WP.5mL</b> , <b>WP 2mL</b> (0.5 mL or 2 mL well plate)
LOOP	The size of the loop in microliters. <b>Note:</b> If the AS is equipped with two injection valves, two loop sizes are shown. If the AS is in concentrate mode, <b>CONCENTRATOR</b> is displayed.	
(Injection Type)	The type of injection performed. This field is updated when the initial conditions of the method's timed events are executed.	See <u>Table C-1</u>
TTL1 TTL2 RLY1 RLY2	Provides TTL and relay control of other devices. In Direct Control, select 1 (on) or 0 (off). In Schedule control, set the TTL and relay fields from the <b>TIMED EVENTS</b> screen for the method being run. <u>Appendix E</u> describes TTL and relay control.	<b>1</b> (on) <b>0</b> (off)
INJ VLV	Displays, and lets you set, the position of the injection valve.	L (load) I (inject)
INJ VLV 1 INJ VLV 2 (simultaneous injection mode only)	If the AS is equipped with two injection valves (for simultaneous injections), these fields display, and let you set, the position of each valve. Injection valve #1 is the bottom valve and injection valve #2 is the top valve. See <u>Section 3.6.9</u> for details about the simultaneous injection option.	L (load) I (inject)
Diverter	Displayed when the AS is equipped with a diverter valve for sequential injections and sequential or sequential concentrate is the selected sampling mode.	A (system 1) B (system 2)

Injection Type	Description
NORMAL FULL	Full-loop injection to one system
NORMAL PARTIAL	Partial-loop injection to one system
NORMAL LS	Limited-sample injection to one system
NORMAL LARGE	Large-loop injection to one system
CONCENTRATE	Concentrate injection to one system
SIMULTANEOUS	Full-loop injections to two systems simultaneously
SEQ FULL	Full-loop injections to two systems alternately
SEQ PARTIAL	Partial-loop injections to two systems alternately
SEQ CONC	Concentrator injections to two systems alternately
Table C	C-1. Injection Types

#### NOTE For details about injection types, see <u>Section 3.5</u>.

# C.4 Method Menu

Use the **METHOD MENU** to:

- Enter the number of the method to be created or edited
- Enter the number to save the method to
- Access the three method editing screens



Figure C-7. Method Menu

To open a method editing screen, move the cursor to the screen's number and press **Enter**. To return to this screen, press **Menu**.

NOTE A method can have a maximum of 100 steps. The total includes the sample prep steps, the method setup (which counts as one step), and the timed events steps. For example, a default method would have a total of four steps: one for the sample prep Wait step, one for the method setup, and two timed events (INIT and time 0.00).

### C.4.1 Sample Prep Screen

Use the **SAMPLE PREP** screen to specify a sequence of operations to be performed before the method's timed events.

**NOTE** See <u>Section D.5.3</u> for details about how to define sample prep steps.

	SAMPLE PREP METHOD:25				
1	PIPET	20.0	uL	<b>#SMP to</b>	S+1
2	MIX	#SMP	250	uL 10	times
3	FLUSH	250	uL		
4	WAIT				
He	lp Prompt				

Figure C-8. Sample Prep Screen

#### **Notes About Entering Vial Positions**

Several of the sample prep operations require you to specify a vial position. You can enter an absolute or a relative position:

- To specify an *absolute* vial position, press numeric buttons from 1 to 100 (depending on the number of vial positions available in the currently installed tray)
- To specify a *relative* position, press Select △ or Select ∨ to scroll through a list of positions: SMP (the current sample vial), S+1 (one position past the sample vial), or S+9 (9 positions past the sample vial). The destination vial list also includes a WST (waste) option.

Prep Operation	Descrip	tion	
DELAY	Specifies for how long the AS waits (0.0 to 999.9 minutes) before proceeding to the next step in the sample prep phase (default=0.0).		
NEEDLE	The height of the needle above the bottom of the vial, from 0 to 46 mm (default=2 mm). The needle height is used for all pipet, dispense, and mix operations until a new height is specified.		
	NOTE	For 0.3 mL and 1.5 mL vials, do not set the needle height greater than 2 mm. This ensures that the vent in the sampling needle is positioned correctly inside the vial.	
PIPET	Moves a The sam	a specified volume of sample from one vial to another. apple syringe is always used for pipetting.	
	μL: #:	Pipet volume: from 1.0 to 99.9 $\mu$ L (in 0.1 $\mu$ L increments) or 100 to 1,000 $\mu$ L (in 1 $\mu$ L increments) (default=20 $\mu$ L). Source vial position (the vial from which to	
	to #:	pick up the specified volume): (default= <b>S+1</b> ). Destination vial position (the vial to receive the volume of sample): (default= <b>SMP</b> ).	
МІХ	Mixes the vial contents by repeatedly drawing and expelling volume of sample. Mixing is done with the prep syringe, if installed; otherwise, the sample syringe is used.		
	<b>#</b> :	Mix vial position (the position of the vial to be mixed): (default = <b>SMP</b> ).	
	<b>X</b> :	Mix cycles (the number of times to repeat the mixing operation): from 1 to 99 (default=10).	
	uL	Mix volume (the volume to be drawn and expelled): from 1.0 to 9,999 $\mu$ L. The default is 500 $\mu$ L if the prep syringe is installed; otherwise, it is 250 $\mu$ L. The mix volume must be equal to or less than the syringe volume.	

Prep Operation	Descrip	ription				
FLUSH	Flushes (default= always u the syrin the volue	the needle with a specified volume, from 1 to 999 $\mu$ L =250 $\mu$ L). The sample syringe and flush reservoir are used for flushing. If the volume specified is greater than ge volume, the syringe takes multiple strokes to achieve me.				
	This ope before ea	ration is independent from the flush that takes place ach injection in a schedule (see Section $2.7.6$ ).				
<b>DISPENSE</b> (sample prep option only)	Moves a specified volume of reagent from a reservoir to a vial using the prep syringe. <b>DISPENSE</b> is available only if the sample preparation option is installed. $\mu$ L: Dispense volume: from 100 to 9,999 $\mu$ L (default=1,000 $\mu$ L). source: Source reservoir (the reservoir from which reagent is dispensed): <b>RES A, RES B, RES C</b> , or <b>RES D</b> (default= <b>RES A</b> ). <b>to #</b> : Destination vial position (the vial to receive the dispensed volume): (default= <b>SMP</b> )					
<b>DILUTE</b> (sample prep option only)	Dilutes a diluent. option is	volume of concentrate with a specified volume of <b>DILUTE</b> is available only if the sample preparation installed.				
	μ <b>L:</b> #:	Concentrate volume: from 1.0 to 99.9 $\mu$ L or 100 to 1,000 $\mu$ L (default=10 $\mu$ L). Source vial: The sample syringe picks up the concentrate from the source vial and dispenses it to the destination vial (default vial= <b>S+1</b> ).				
	μ <b>L</b> : source:	Diluent volume: from 10.0 to 99.9 $\mu$ L or 100 to 9,999 $\mu$ L (default=10 $\mu$ L) (default=990 $\mu$ L). Diluent reservoir (the reservoir from which diluent is dispensed): <b>RES A, RES B, RES C</b> , or <b>RES D</b> (default= <b>PES A</b> )				
	to # :	Destination vial position (the vial that the concentrate and diluent volumes are delivered to): (default vial= <b>SMP</b> ).				

Prep Operation	Description
WAIT	The AS waits for a continue command before proceeding with the method. The command can be from the front panel (pressing <b>Hold/Run</b> ) or a TTL input. Each method must have one wait step and no more. Sample prep operations can be added before and after the wait step. By default, the wait step is at the end of the sample prep phase. The wait step can be disabled from the <b>SYSTEM PARAMETERS</b> screen (see Section C.6.5).
<b>REAGENT FLUSH</b> (concentrate mode only)	Moves the specified volume of reagent from a reservoir or vial to the inject port. This operation is used to flush reagent onto a concentrator column.
	<ul> <li>μL: Reagent volume: The volume range and default depends on the source, the syringe size, and vial size. See <u>"Notes on Reagent Volume Ranges" on page 241</u>.</li> <li>source: Reagent source (enter either the vial from which to pick up the volume of reagent or select the flush reservoir (FLH). If the sample prep option is installed, a reagent reservoir can be selected: RES A, RES B, RES C, or RES D (default=FLH).</li> <li>valve: Load/inject valve position: L (load), (inject), or N (no change) (default=N).</li> </ul>
REAGENT PRIME (concentrate mode only)	Moves the specified volume of reagent (100 to 50,000 $\mu$ L) from a reservoir to the waste port. This operation is used to prime the sampling needle assembly with reagent when the AS is in concentrate mode. $\mu$ L: Enter the reagent volume. The volume range and default depends on the source, the syringe size, and the vial size. See the notes under <b>REAGENT FLUSH</b> . source: Enter either the vial from which to pick up the volume of reagent or select the flush reservoir ( <b>FLH</b> ). If the sample prep option is installed, a reagent reservoir can be selected: <b>RES A, RES B, RES C</b> , or <b>RES D</b> (default= <b>FLH</b> ). valve: Select the position of the load/inject valve: L (load), L(inject) or N (no change) (default=N)
### Notes on Reagent Volume Ranges

- If the source is a reservoir, the allowed range is 100 to 50,000 μL. The default is 2,000 μL if the syringe volume is less than 5,000 μL, and 10,000 μL if the syringe volume is 5,000 μL or 10,000 μL.
- If the source is a vial, the range depends on the volume of the sample syringe:

Sample Syringe Volume ( $\mu L$ )	Reagent Flush Range (μL)	Default (µL)
100 to 1,000	100 to 1,000	1,000 (any size vial)
5,000	100 to 4,700	1,000 (1.5 mL vial) 4,000 (10 mL vial)
10,000	100 to 8,000	1,000 (1.5 mL vial) 8,000 (10 mL vial)

**NOTE** The **REAGENT FLUSH** and **REAGENT PRIME** operations are performed *after* the autosampler loads the sample onto the concentrator column. See <u>Section 3.6.10</u> for details about concentrator injections.

## C.4.2 Method Setup Screen

Use the **METHOD SETUP** screen to set the initial conditions and default parameters for a method.

METHOD SETUP	METHOD: 25
CYCLE TIME: SAMPLE NEEDLE HEIGHT: SAMPLE TRAY TEMPERATURE:	123 min 46 mm 20 °C
WAIT FOR TEMP STABLE:	YES
Help Prompt	

Figure C-9.	Method Setup	Screen
-------------	--------------	--------

Screen Field	Description	Values
CYCLE TIME	The time between injections. When a method is assigned a cycle time, its timed events are not executed until the specified time has elapsed. See <u>Section 2.7.5</u> for details.	1 to 240 minutes (default=off)
	Cycle time is ignored in the following situations:	
	• If <b>WAIT FOR TEMP STABLE</b> is enabled in the <b>METHOD SETUP</b> screen (see <u>Section C.6</u> ) and the tray or column temperature has not stabilized by the end of the cycle time.	
	• If the wait step of the method's sample prep phase is enabled and a continue command has not occurred by the end of the cycle time.	
SAMPLE NEEDLE HEIGHT	Controls the height of the sample needle above the bottom of the vial.	0 to 46 mm (default=2)
	NOTE For 0.3 mL and 1.5 mL vials, do not the needle height greater than 2 mm. ensures the vent in the needle positioned inside the vial.	t set This is

Screen Field	Description	Values
SAMPLE TRAY TEMPERATURE	The sample tray temperature set point. Pressing <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ turns the tray temperature control on and off. Available only when the sample temperature control option is installed.	4 to 60 °C (default=off) (default set point=20)
WAIT FOR TEMP STABLE	Specifies whether the AS should wait until the temperature of the sample tray has stabilized before continuing.	YES/NO

## C.4.3 Timed Events Screen

Use the **TIMED EVENTS** screen to specify a sequence of events after the method's sample prep phase is completed.



Figure C-10. Timed Events Screen

	TIMED EVENTS		1	METHO	D: 25	
TIME	VALVE1	VALVE2	TTL1	TTL2	RLY1	RLY2
INIT	LOAD	LOAD	0	0	0	0
0.00	INJECT	INJECT				
Help Prompt						
L						

Figure C-11. Timed Events Screen: Simultaneous Injection Mode

When entering timed event parameters:

- Press **Select**  $\Delta$  or **Select**  $\nabla$  to select a value.
- Press **Delete** to clear a value from a field. When a field is blank, the value from the previous step remains in effect.

Screen Field	Description	Values
TIME	The elapsed time at which the method step starts. Every method begins with an <b>INIT</b> (initial) step and a <b>TIME = 0.00</b> step. You can edit the events for these steps, but cannot delete them. The initial step occurs when the timed events for the previous injection have ended.	0 to 999.99 minutes
VALVE	Controls the position of the injection valve in the chromatography system module. An AS TTL output must be connected to a TTL input on the chromatography system module. See <u>Appendix E</u> for details about TTL connections. Only one <b>LOAD</b> and one <b>INJECT</b> command can be specified in a method. Sample loading must occur before the injection. By default, <b>LOAD</b> occurs at the <b>INIT</b> step and <b>INJECT</b> occurs at <b>TIME = 0.00</b> .	LOAD/ INJECT
NOTE	If the schedule specifies an injection per vial of 0, the method's sample prep steps occurs.	en only the
VALVE1 VALVE2 (simultaneous injection mode only)	If the AS is equipped with, or connected to, two injection valves (for simultaneous injections), these fields are used to select the position of each valve. If the valves are installed in the AS, injection valve #1 is the bottom valve and injection valve #2 is the top valve. Specify one <b>LOAD</b> and one <b>INJECT</b> command for each valve. Sample loading must occur before the injection. By default, <b>LOAD</b> occurs at the <b>INIT</b> step and <b>INJECT</b> occurs at <b>TIME = 0.00</b> . See <u>Section 3.6.9</u> for details about the simultaneous injection option.	LOAD/ INJECT
TTL1, TTL2 RLY1, RLY2	Provides TTL and relay control of other devices. See Appendix E for details.	<b>1</b> (on) <b>0</b> (off)

# C.5 Schedule Screen

SAVE TO **RUN** 1 SCHEDULE EDIT 1 1 SKIP **MISSING VIAL ACTION:** VIAL# INJ/ INJ **START- END** VIAL VOL(uL) **METHOD** LINE **25.**0 3 1 1-4 4 1 -5 5 5 1 25.0 1 6 6 6 1 5.0 1 **Help Prompt** 



Screen Field	Description	Values
EDIT	The schedule number to edit. Press a numeric button to select the number or press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to scroll through the choices. Press <b>Enter</b> to display the selected schedule.	1 to 9
SAVE TO	The schedule number to save the current schedule to. It does not have to be the <b>EDIT</b> number.	1 to 9
RUN	The schedule number to run. <b>Note</b> : The schedule will not start until you press <b>Hold/Run</b> or send a TTL/RLY run command.	1 to 9
MISSING VIAL ACTION	The action the AS takes if a vial is missing from the tray. Select <b>SKIP</b> to ignore the missing vial and move to the next vial in the schedule. Select <b>STOP</b> to stop the schedule. To resume, press <b>Hold/Run</b> .	SKIP/STOP (default=STOP)
LINE	Each line contains the parameters for one vial in the schedule. Line numbers are assigned automatically when you insert steps. (A step is one horizontal line on the <b>SCHEDULE</b> screen.) If consecutive vials have identical parameter values, line numbers are shown in a range format (see the example above). A schedule can have a maximum of 203 steps, containing up to 999 lines.	1 to 999 (automatically assigned)

Screen Field	Description	Values
VIAL# START END	The first and last vial numbers for the schedule step. The vials in this range are sampled according to the parameters specified in the step. The starting vial does not have to be a smaller number than the ending vial. To sample just one vial, enter the same vial number for the start and end.	1 to 100 (1.5 mL tray) 1 to 49 (plastic 10 mL tray)
INJ/VIAL	The number of injections performed on each vial. If zero injections is specified, only the sample prep portion of the method will be performed.	0 to 99 (default=1)
INJ VOL (µL) (non- simultaneous injection mode)	The volume of sample delivered for each injection. The maximum injection volume depends on the loop size installed. The volume entered determines whether the injection is a full-loop; partial-loop; or partial-loop, limited-sample. See <u>Section 3.5</u> for details.	1.0 to 99.9 μL 100 to 1,000 μL (default=25)
<b>INJ VOL (μL)</b> (simultaneous injection mode)	If the AS is equipped with the simultaneous injection option, the injection volume is loaded into the sample syringe and then delivered equally to the two sample loops. See <u>Section 3.6.9</u> for details about the simultaneous injection option.	1,000 to 4,700 μL (with 5 mL syringe) 1,000 to 8,000 μL (with 10 mL syringe) (default= 1,000 μL)
METHOD	The method number to run on each vial in the range.	1 to 99
]	NOTE The optional diverter valve for sequential cannot be controlled from a front panel Chromeleon or Chromeleon Xpress is re automatically switch the diverter valv	injections schedule. quired to e during

sequential runs (see Section 3.6.11).

# C.6 Module Setup Menu

MODULE SETUP MENU					
1	INSTALLED OPTIONS	6	<b>INJECT PORT ALIGN</b>		
2	FRONT PANEL	7	INTERLOCK BYPASS		
3	PLUMBING CONFIG				
4	TIME/DATE				
5	5 SYSTEM PARAMETERS				
Н	elp Prompt				

Figure C-13. Module Setup Menu Screen

## C.6.1 Installed Options Screen

The **INSTALLED OPTIONS** screen lists the optional features installed in the AS. The AS automatically detects which options are installed and displays a check mark in the appropriate fields. This screen is displayed briefly at power-up.



Figure C-14. Installed Options Screen

### NOTES

- The INJECTION VALVE option is always enabled, even if a valve is not installed in the autosampler compartment. This allows all injection valve commands issued from Chromeleon or Chromeleon Xpress to control the valve in an IC system (ICS-1000, ICS-1500, ICS-2000, or ICS-3000).
- The **INJECTION VALVE 2** option is shown only when the simultaneous injection mode is enabled. Refer to <u>Section 3.6.9</u> for details about simultaneous injections.

• If the AS is in a sequential injection mode and a diverter valve is installed, the diverter valve is listed on the screen (see Figure C-15).



Figure C-15. Installed Options Screen: Diverter Valve Installed

### C.6.2 Front Panel Screen

Use the **FRONT PANEL** screen to select display options.



Figure C-16. Front Panel Screen

Screen Field	Description	Values
DISPLAY PANEL BACKLIGHT	Selects the brightness of the display panel backlight. The off option darkens the display completely. When off, press any button to turn the light on again.	LOW/MEDIUM/ HIGH/OFF (default=HIGH)
KEY ACTUATION SOUND	Turns the key sound on and off.	<b>ON/OFF</b> (default= <b>OFF</b> )
ENTRY ERROR SOUND	Turns the error sound on and off.	ON/OFF (default=OFF)
HELP LANGUAGE	Selects the language for help and error messages.	ENGLISH/ JAPANESE

## C.6.3 Plumbing Configuration Screen

Use the **PLUMBING CONFIGURATION** screen to specify the plumbing options installed in the AS.

PLUMBING CONFIGURATION			
LOOP SIZE V1:	25 uL		
LOOP SIZE V2:	uL		
SAMPLE SYRINGE VOLUME:	250 uL		
SAMPLE SYRINGE CONFIG:	READY		
PREP SYRINGE VOLUME:	5 mL		
PREP SYRINGE CONFIG:	READY		
Help Prompt			

Figure C-17. Plumbing Configuration Screen

Screen Field	Description	Values
LOOP SIZE V1	The volume of the loop installed on the first (or only) injection valve. The upper limit depends on the volume of the sample syringe installed: $500 \ \mu L$ (if the syringe volume is $\leq 1 \ mL$ ) $4,000 \ \mu L$ (if the syringe volume is $5 \ mL$ ) $8,000 \ \mu L$ (if the syringe volume is $10 \ mL$ ) This field is not available in concentrate mode.	2.0 to 99.9 μL 100 to x μL (default=25)
LOOP SIZE V2	The volume of the loop installed on the second injection valve. This field is available in simultaneous and sequential injection modes only.	See LOOP SIZE V1
	• In simultaneous mode (see <u>Section 3.6.9</u> ), the value entered for <b>LOOP SIZE V2</b> is for informational purposes only.	
	• For sequential mode (see <u>Section 3.6.11</u> ), the value of <b>LOOP</b> <b>SIZE V2</b> determines the injection type (see <u>Section 3.5</u> ). It is also used to compute the injection volume.	

Screen Field	Description	Values
SAMPLE SYRINGE VOLUME	The volume of the sample syringe installed. Press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to scroll through the choices. If the AS is equipped with the simultaneous injection option (see <u>Section 3.6.9</u> ), select either the 5,000 or 10,000 µL size. For sequential and concentrate modes, any sample syringe size (except 2.5 mL) can be installed.	100, 250, 500, 1,000, 5,000, 10,000 μL (default=250) For simultaneous injection mode: 5,000 or 10,000 μL (default=5,000)
SAMPLE SYRINGE CONFIG	The action to be performed by the sample syringe. These actions are used to configure the syringe at initial setup or after a new syringe is installed. <b>INITIALIZE</b> moves the syringe to an initial position. <b>READY</b> is displayed when syringe configuration is complete; no action occurs. See <u>Section 5.3</u> for details.	INITIALIZE READY
PREP SYRINGE VOLUME	The volume of the prep syringe installed. Press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to scroll through the choices.	250, 500, 1,000, 5,000, 10,000 μL (default=5,000)
PREP SYRINGE CONFIG	The action to be performed by the prep syringe. See the <b>SAMPLE SYRINGE CONFIG</b> section for details.	INITIALIZE READY

## C.6.4 Time/Date Screen

Use the **TIME/DATE** screen to set the time and date on the real-time clock.

TIME/DATE			
ACTUAL TIME: SET TIME: DATE:	12:35:00 12:34:56 16 JUNE 06		
Help Prompt			

Figure C-18. Time/Date Screen

Screen Field	Description
ACTUAL TIME	Displays the time on the real-time clock.
SET TIME	Sets the 24-hour real-time clock. Enter the hour first, followed by minutes, and then seconds. For example, if the time is 8:35 a.m. and 50 seconds, enter 08:35:50. If the time is 8:35 p.m. and 50 seconds, enter 20:35:50. Press numeric buttons to enter the time directly, or press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to increase or decrease the current value.
DATE	Sets the date on the real-time clock. Press numeric buttons to enter the day and year directly, or press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to increase or decrease the current values. Press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to select the month.

### C.6.5 System Parameters Screen

Use the **SYSTEM PARAMETERS** screen to set default system operating parameters. The parameters vary, depending on whether the AS is controlled exclusively from the front panel or if it is sometimes or always under Chromeleon or Chromeleon Xpress control.



Figure C-19. System Parameters Screen: AS Controlled Exclusively from Front Panel



Figure C-20. System Parameters Screen: AS Controlled from Chromeleon or Chromeleon Xpress

Screen Field	Description	Values
WAIT	Enables or disables the wait step in the sample prep phase of the method. When wait is enabled, the AS	ENABLED DISABLED
	waits for a run command from either the front nanel <b>Hold/Run</b> button or a TTL input signal	(default = <b>ENABLED</b> )
	before performing a method's timed events steps.	
	This allows another instrument to control the	
	timing of each injection in a schedule.	

Screen Field	Description	Values
	When wait is disabled, a run command is required to start a schedule. Thereafter, the AS performs the schedule lines without requiring additional run commands. Disabling wait allows the AS to act as a system master. The AS can send TTL signals to other modules in the system, thereby controlling system functions (starting the pump, starting the detector method, etc.).	
	NOTE This parameter is used for front panel operation only. If the AS is controlled by Chromeleon or Chromeleon Xpress, the software automatically enables the wait function and the setting on the SYSTEM PARAMETERS screen is ignored.	
METHOD SYRINGE SPEED	(This parameter is displayed only when the AS is controlled exclusively from the front panel.) The syringe speed to use during a front panel method run. For water, accept the default value of 3; more viscous liquids require slower speeds. For concentrate mode, select a speed of 2. Be sure to select a speed that does not produce backpressure of more than 690 kPa (100 psi). See <u>Section 3.8</u> for details. In general if you are using a 5 or 10 mL syringe, select a speed of 1 or 2.	1 (slowest) to 5 (fastest) (default=4) For concentrate mode: select 2
PIPET SYRINGE SPEED	(This parameter is displayed only when the AS is controlled by Chromeleon or Chromeleon Xpress.) The syringe speed used for various pipet actions. Accept the default value.	1 (slowest) to 5 (fastest) (default=4)
	NOTE For Chromeleon or Chromeleon Xpress operation, specify syringe speeds in the program or on the Control panel (see <u>Section 3.7</u> ).	
CUT SEGMENT VOLUME	For partial-loop injections, specifies a volume of sample to be "cut" (or omitted) from each side of the sample during the injection process. See <u>Section 3.5</u> for details about injection types.	0 to 30 μL (default=10)
SAMPLE MODE	The sample delivery mode.	See the table below.

Screen Field	Descriptio	n	Values
INJECT PORT VOLUME	The volume injection va that must b accurately determined changed un installed. If mode (see <b>9</b> port volum volume for	1 to 999 μL (default=25)	
FLUSH SYRINGE SPEED	(This parameter is displayed only when the AS is controlled exclusively from the front panel.)1 (slowest)The syringe speed to use for flush operations, including reagent flushing and priming after concentrator loading. Be sure to select a speed that does not produce backpressure of more than 690 kPa (100 psi). See Section 3.8 for details. In general if you are using a 5 or 10 mL syringe, select a speed of 1 or 2.1 (slowest)		
EMPTY SYRINGE SPEED	This param controlled I The speed t syringe em	eter is displayed only when the AS is by Chromeleon or Chromeleon Xpress. the syringe moves during various pty actions. Accept the default value.	1 (slowest) to 5 (fastest) (default=4)
SAMPLE MODE		DESCRIPTION	
NORMAL (default)		Injections to a single system (full-loop, partial-loop, limited-sample).	partial-loop, or
CONCENTRATE		Injections to a single system when the s replaced by a concentrator column.	sample loop is
SIMULTANEOUS		Injections to two systems simultaneous Requires the simultaneous injection opt	ly (full-loop). tion.
SEQUENTIAL		Injections to two systems in sequence ( loop, or partial-loop, limited-sample). If sequential injection option.	full-loop, partial- Requires the
SEQUENTIAL CONCENTRATE		Injections to two systems in sequence when the sample loop is replaced by a concentrator column. Requires the sequential injection option.	

### C.6.6 Inject Port Alignment Screen

Use the **INJECT PORT ALIGNMENT** screen to align the sampling needle in the inject port. See Section D.8 for detailed instructions.

NOTE You can also align the inject port from the Chromeleon or Chromeleon Xpress Control panel. This is an easy to use method and is recommended when Chromeleon or Chromeleon Xpress is available. See <u>Section 5.8</u> for detailed instructions.

INJECT PORT ALIGNMENT				
X ADDRESS: Y ADDRESS: Z ADDRESS:	1340 1930 230			
Z ADDRESS:	230			
Help Prompt				



Screen Field	Description	Values
X ADDRESS	The X-axis position of the needle arm.	1290 to 1390 (default=1340)
Y ADDRESS	The Y-axis position of the needle arm.	1880 to 1950 (default=1930)
Z ADDRESS	The Z-axis position of the needle arm.	180 to 280 (default=230)

### C.6.7 Door Interlock Bypass Screen

Use the **DOOR INTERLOCK BYPASS** screen to temporarily disable the autosampler compartment door lock.

NOTE The door interlock bypass can be used only for Direct control, Local mode operation. It cannot be used for Schedule control or computer control (Locked Remote).



### DOOR INTERLOCK BYPASS YES

NOTE: After setting bypass you have a 15 minute window to perform operations with the sample door open.

Help Prompt

Figure C-22. Door Interlock Bypass Screen



Be aware of unexpected sampling arm movement when operating with the door open.

Soyez conscient de la possibilité de mouvements imprévus du bras d'échantillon lorsqu'il est utilisé avec la porte ouverte.



Achten Sie beim Betrieb mit geöffneter Tür auf unerwartete Bewegungen des Probenarms.

Screen Field	Description
DOOR INTERLOCK BYPASS	Press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to select <b>YES</b> (bypass the door lock) or <b>NO</b> (do not bypass the door lock) and then press <b>Enter</b> . The default setting is <b>NO</b> .
	After selecting <b>YES</b> and pressing <b>Enter</b> , the door lock alarm is disabled for 15 minutes. If you open the autosampler compartment door during this time, the sampling arm continues its current operation. However, if a schedule is running, opening the door ends the schedule. To extend the bypass for another 15 minutes, press <b>Enter</b> again.
	When the 15 minutes is up, the <b>DOOR INTERLOCK BYPASS</b> setting automatically reverts to <b>NO</b> . When <b>NO</b> is selected, opening the autosampler compartment door stops the sampling arm operation and ends the currently running schedule.

## C.7 Flush/Prime Screen

Use the FLUSH/PRIME screen to:

- Set the parameters for and then initiate a flush cycle that flushes the inject port and washes the outside of the needle.
- Set the parameters for and then initiate a prime cycle that primes the lines to the reservoir(s) and the sampling needle.
  - NOTE To make sure the backpressure during flushing remains below 690 kPa (100 psi), select a flush syringe speed that is appropriate for the syringe size and tubing volume. See <u>Section 3.8</u> for details.

If the AS is controlled by Chromeleon or Chromeleon Xpress, set the syringe speed on the Chromeleon or Chromeleon Xpress Control panel (see <u>Section 3.7</u>). If the AS is always controlled from the front panel, select the syringe speed on the SYSTEM PARAMETERS screen (see <u>Section C.6.5</u>).

FLUSH/PRIME					
FLUSH			PRIME		
VOLUME:	250	uL	<b>VOLUME:</b>	2000 uL	
<b>RESERVOIR:</b>	<b>FLUS</b>	н	<b>RESERVOIR:</b>	RES A	
SYRINGE:	SAMP	LE	SYRINGE:	PREP	
ACTION:	READ	Y	ACTION:	READY	
Help Prompt					

Figure C-23. Flush/Prime Screen

Screen Field	Description	Values
FLUSH VOLUME	The volume of fluid used to flush the inject port. After the inject port is flushed the exterior of the needle is washed with a volume of fluid set at the factory.	100 to 9,999 μL (default=250)
FLUSH RESERVOIR	The source reservoir for the flush liquid. This is always the reservoir connected to port E on the sample syringe valve (see <u>Section B.3.3</u> ).	FLUSH
FLUSH SYRINGE	The sample syringe is always used for flushing.	SAMPLE
FLUSH ACTION	To start a flush cycle, first make sure the AS is in Direct Control mode; then press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to select <b>FLUSH</b> and press <b>Enter</b> .	READY, FLUSH
PRIME VOLUME	The volume of fluid used for priming.	100 to 5,000 μL (default=400)
PRIME RESERVOIR	The reservoir used for the prime cycle. If the sample preparation option is installed, press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to scroll through the choices. If the sample preparation option is not installed, the flush reservoir is always used and this field cannot be edited. The prep syringe is always used for <b>RES A, B, C, and D</b> .	FLUSH, RES A, RES B, RES C, RES D
PRIME SYRINGE	The syringe used for the prime cycle. If the sample preparation option is not installed, the sample syringe is always used and this field cannot be edited.	SAMPLE, PREP
PRIME ACTION	To start a prime cycle, first make sure the AS is in Direct Control mode; then, press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to select <b>PRIME</b> and press <b>Enter</b> .	READY, PRIME

# C.8 Time Function In Screen

Use the TIME FUNCTION IN screen to:

- Display the AS functions that can be controlled via TTL input from another device
- Select a TTL signal mode for each function

See <u>Appendix E</u> for details about TTL-controlled functions and connections.



Figure C-24. Time Function In Screen

Screen Field	Description	Values
MODE	Select the signal mode that corresponds to the signal type of the controlling device. <b>NORMAL EDGE</b> , the default mode, is compatible with the TTL output signals provided by Dionex modules.	NORMAL EDGE INVERTED EDGE NORMAL PULSE INVERTED PULSE
UNUSED:TTL1	Not active.	
START- CONTINUE: TTL2	A signal to TTL2 from the controlling device starts the schedule (if it is not currently running), or continues the current schedule (if the AS is at the wait step of a method).	
TRAY TEMP ON/OFF: TTL3	A signal to TTL3 from the controlling device turns the sample temperature control option on and off.	
UNUSED:TTL4	Not active.	

# C.9 Diagnostic Menu

To go to the **DIAGNOSTIC MENU**, press **Menu** and **8**.



Figure C-25. Diagnostic Menu Screen

## C.9.1 Power-Up Screen

Use the **POWER-UP** screen to display the revision levels of the Moduleware and USB BIOS code, the module's ID number (if connected), and the serial number of the AS. This is the same power-up screen that displays when you turn on the AS.



Figure C-26. Power-Up Screen

### C.9.2 Diagnostic Tests Screen

This screen lets you test the AS electronics components.

DIAGNOSTIC TESTS				
P CPU P LEAKS	P	DSP COMM XYZ HOME	P P P	SYRINGE COMM INTERLOCK INJECT VALVE
Help Prompt				

Figure C-27. Diagnostic Tests Screen

These tests are run automatically at power-up. If any test fails, this screen is displayed and indicates the status of each test:

- "--" The test was not run
- ">" The test is in progress
- "P" The test passed
- "F" The test failed

To run a test manually, position the cursor in the edit field next to the test, press **Select**  $\Delta$  or **Select**  $\nabla$  to select the asterisk (\*), and press **Enter**.

Test	Description
CPU	Checks the CPU internal configuration and the Moduleware checksum.
LEAKS	Checks whether the leak sensors detect a leak.
DSP COMM	Checks communication between the CPU and the DSP (Digital Signal Processor) hardware.
XYZ HOME	Tells the DSP to search for the XYZ home position.
SYRINGE COMM	Checks communication between the CPU and the sample syringe.
INTERLOCK	Checks that the autosampler door is closed.
INJECT VALVE	Checks that the injection valve operates correctly.
<b>DIVERTER</b> (sequential injection mode)	Checks that the diverter valve (if installed) operates correctly.

l

Test	Description
INJECT VALVE 1 INJECT VALVE 2 (simultaneous injection mode)	If the AS is equipped with two injection valves (for simultaneous injections), these tests check that each valve operates correctly.

## C.9.3 XYZ Test Screen

Use the XYZ TEST screen to test operation of the sampling needle arm.

		XYZ TEST			
			X	Υ	Ζ
ACTION:	CYCLE	ACTUAL:	1234	5678	901
X SENSOR	t: 0	1:	1234	5678	901
Y SENSOR	t: 1	2:	0	0	0
Z SENSOR	R: 0	3:	0	0	0
Z HOME	E: 1	4:	0	0	0
Help Promp	t				

Figure C-28. XYZ Test Screen

Screen Field	Description	n
ACTION	The action HOME GO TO X	the needle arm should perform: Moves all three axes to the home position. Positions the arm at the specified location (1, 2, 3, or 4).
	CYCLE	Continuously cycles the arm through the four positions.
	PATTERN	Continuously cycles through a four corners test pattern, with the needle moving about halfway into the vial.
	VIALS	Continuously cycles the arm through each position in the currently installed tray, with the needle tapping the top of each vial.
	VIALS+I	Continuously cycles the arm through each position in the currently installed tray, with the needle inserted into each vial.
ACTUAL XYZ	Displays th indicate the	e needle arm's current XYZ position. The numbers number of motor steps away from the home position.

Screen Field	Description
XYZ 1, 2, 3, 4	Allows entry of XYZ values for four needle arm positions. The numbers indicate the number of motor steps away from the home position.
X SENSOR Y SENSOR	Indicates the current state of the X and Y sensors. The X and Y sensors detect evenly spaced slots that indicate the home position and mark distance intervals.
Z SENSOR	Indicates whether a vial is present. When the needle is down and a vial is present, "1" is displayed. If no vial is present at the current location when the needle is down, "0" is displayed.
Z HOME	Indicates whether the Z axis is at the home position. An entry of "0" indicates home.

To run a test:

- 1. Move the cursor to the **ACTION** field and select an action to perform. For the **GO TO** action, enter the desired XYZ positions.
- 2. Press Enter.

### C.9.4 Temperature Statistics Screen

Use the **TEMPERATURE STATISTICS** screen to:

- Manually control the temperature of the optional tray compartment
- Monitor the status of temperature-related values

# NOTE This screen is available only when the temperature control option is installed.

			TEM	PER/	TUR	E S1	ATI	STICS	
	VIN:	0.1	Α	MAX	(: 10	.2	Α	TIME:	12:34:56
T.	RAY	SET	TEM	P: 35	°C				
	AC	TUAL	: 35	.0°	C=				
*	PEL	TIER:	10	.0	4				
L	CON	NTROL	.: 07	HE	X				
	DUT	YCYC	: 12	3					
	Help I	Prompt							

Figure C-29. Temperature Statistics Screen

MIN, MAX, TIMEDisplays the minimum and maximum values selected value, and the length of time the value monitored. An asterisk indicates the value bei select a value to monitor, move the cursor to t and press Enter.TRAY SET TEMPThe temperature to maintain in the tray.ACTUALDisplays the measured temperature. The read every 3 seconds. A symbol to the right of the	
<b>TRAY SET TEMP</b> The temperature to maintain in the tray. <b>ACTUAL</b> Displays the measured temperature. The readi every 3 seconds. A symbol to the right of the	measured for the has been ng monitored. To he desired field
ACTUAL Displays the measured temperature. The reading every 3 seconds. A symbol to the right of the	
the temperature's status:	ing is updated reading indicates
<ul> <li>The temperature is stabilized</li> <li>↑ The temperature is increasing</li> <li>↓ The temperature is decreasing</li> </ul>	
<b>PELTIER</b> Displays the measured Peltier currents.	
<b>CONTROL</b> Displays the state of the Peltier control bits:	
bit 0: Saturate (0=duty cycle, 1=saturate) bit 1: Peltier enable (1=on) bit 2: Heat/cool (0=heat, 1=cool) bits 3 through 7: Always 0	
<b>DUTY CYCLE</b> Displays the Peltier duty cycle (from 0 throug	h 255).

### C.9.5 Liquid Control Screen

Use the **LIQUID CONTROL** screen to:

- Manually control the sample and prep syringes
- Manually control the inject valve
- Manually position the needle at a specified vial

The AS must be in Local mode, Direct Control to manually control these functions.

LIQUID CONTROL				
VIAL#:	1	NEEDLE HEIGHT:	46	
ASPIRATE:	100	uL from NEEDLE	_	
DISPENSE:	100	uL SYRINGE SPEED:	4	
SYRINGE:	SAMF	PLE INJ VALVE:	LOAD	
ACTION:	ASPII	RATE		
Help Prompt				

Figure C-30. Liquid Control Screen



Figure C-31. Liquid Control Screen: Simultaneous or Sequential Mode

Screen Field	Description	Values
VIAL#	The position for the sampling needle. Press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to scroll through vial numbers and options. Use numeric buttons to enter an absolute vial number. The range of valid vial numbers depends on the type of tray currently installed. Press <b>Enter</b> to move the needle to the selected position.	Absolute vial numbers (default=1), INJ (inject port), FLU (flush port)
ASPIRATE	The volume of liquid to be drawn during an aspirate action.	1.0 to 99.9 μL or 100 to 1,000 μL (default=100 μL)
from	The source for the action. The choices depend onthe syringe in use:Sample:NEEDLE, FLUSHPrep:NEEDLE, FLUSH, RES A, RES B, RES C, RES D, AIR	
DISPENSE	The volume of liquid to dispense during a dispense action.	1.0 to 99.9 μL or 100 to 1,000 μL (default=100 μL)
SYRINGE	The syringe to use for an action.	SAMPLE, PREP
ACTION	The action to perform. Press <b>Select</b> $\triangle$ or <b>Select</b> $\nabla$ to scroll through the choices; press <b>Enter</b> to start the action.	ASPIRATE, DISPENSE, EMPTY, FILL
NEEDLE HEIGHT	The height of the needle above the bottom of the vial. If 0 is selected, a small distance is added to prevent the needle from touching the bottom of the vial.	0 to 46 mm (default=46 mm)
SYRINGE SPEED	The syringe speed to use. Use a value of 5 for water. More viscous liquids require slower speeds.	1 (slowest) to 5 (fastest) (default=4)
INJ VALVE	The injection valve position.	INJECT, LOAD
INJ VLV 1 INJ VLV 2 (simultaneous injection mode)	If the AS is equipped with two injection valves (for simultaneous injections), these fields select the position of each valve. Injection valve #1 is the bottom valve and injection valve #2 is the top valve.	INJECT, LOAD
DIVERTER VLV	If the AS is equipped with a diverter valve for sequential injections, this field selects the valve position.	<b>A</b> (system 1) <b>B</b> (system 2)

### C.9.6 Logs Menu



Figure C-32. Logs Menu Screen

### Usage Log Screen

Use the **USAGE LOG** screen to:

- Monitor for how long various AS components have been in use
- Reset counters after replacing a component

The status of each parameter is updated in real time.



Figure C-33. Usage Log Screen

NOTE Press the down arrow to see additional parameters.





Screen Field	Description
MODULE ON	The number of hours the AS has been powered-up.
BACKLIGHT	The number of hours the backlight has been on.
INSERTS INJ PORT	The number of needle insertions into the inject port since the counter was last reset to 0. Reset the counter after replacing the sampling needle assembly and the needle seal assembly.
SAMP SYRINGE USE	The number of aspirate or dispense cycles the sample syringe has performed since the counter was last reset to 0. Reset the counter after replacing the syringe.
PREP SYRINGE USE	The number of aspirate or dispense cycles the prep syringe has performed since the counter was last reset to 0. Reset the counter after replacing the syringe.
XYZ NEEDLE ASSEM	The number of XYZ sampling needle arm movements completed since the counter was last reset to 0. The count is incremented when the needle arm moves to the home position, to the vial, and to the inject port.
INJECT VALVE	The number of times the injection valve has performed a load/inject cycle since the counter was last reset to 0. Reset the count after replacing the injection valve
DIVERTER VALVE	The number of times the diverter valve has cycled the flow between system 1 and system 2 since the counter was last reset to 0.
RESET	To reset a counter to 0, move the cursor to the corresponding asterisk (*) field and press <b>Enter</b> .

### Message Log Screen

Displays a list of errors that have occurred.

		MESSAGE LOG							
1	11:22:33	<b>29 SEPTEMBER 07</b>	#208						
	Incorrect key pressed								
2	12:13:14	29 SEPTEMBER 07	#567						
	Missing vial detected								
3	15:67:02	29 SEPTEMBER 07	#331						
Method does not exist									
Help Prompt									

Figure C-35. Message Log Screen

# NOTE All messages are cleared from the log when the AS power is turned off.

Each message includes the time and date the message was recorded, the error code, and a brief description of the error.

- To see a full-screen description of an error, move the cursor to the error's entry number and press **Help**.
- Press **Select** △ or **Select** ∇ to go to the beginning or ending of the log, respectively.

### C.9.7 Leak Sensor Calibration and Status Screen

Use this screen to display the status of the leak sensors and to calibrate them.

LEAK SENSOR CALIBRA	TION	AND STATUS
	TRAY	
<b>MEASURED VALUE:</b>	2.48	
<b>CURRENT CONDITION:</b>	DRY	
CALIBRATION VALUE:	2.49	
LOW LEAK THRESHOLD:	2.29	
ENABLE LEAK DETECTION	YES	
Help Prompt		

Figure C-36. Leak Sensor Calibration and Status Screen

Screen Field	Description
MEASURED VALUE	Reports the current measured voltages for the two leak sensors. <b>TRAY</b> is the sensor installed in the autosampler drip tray. The <b>OVEN</b> fields are for support of the thermal compartment option. <b>Note:</b> The thermal compartment option is not currently available.
CURRENT CONDITION	Reports the current state of the sensors: <b>DRY</b> if the sensor reading is within the dry range, <b>WET</b> if the sensor reading is within the wet range, or <b>ERR</b> if the sensor is out of range.
	If the measured value drops 0.02 volts below the calibration value, the current condition is wet. If the measured value goes 0.06 volts above the calibration value, the sensor needs to be recalibrated.
	To calibrate a sensor, press <b>Select</b> $\Delta$ or <b>Select</b> $\nabla$ to select <b>CAL</b> and then press <b>Enter</b> . The current measured value becomes the new calibration value. See <u>Section 5.11</u> for detailed instructions.
CALIBRATION VALUE	Reports the voltage values set for the calibration.
LOW LEAK THRESHOLD	Reports the minimum voltage reading that is interpreted as a dry sensor. A reading below this voltage indicates a wet sensor.

### C.9.8 Keyboard Test Screen

Use the **KEYBOARD TEST** screen to conduct an interactive test of the front panel keypad buttons.

STOP	HOLD/RUN	UP	RIGHT	7	8	9
НОМЕ	DOOR	DOWN			5	6
INSERT	SEL UP			1	2	3
DELETE Help Prompt	SEL DOWN	HELP	MENU	0	•	E

Figure C-37. Keyboard Test Screen

To test the buttons:

- 1. Press a button on the keypad. If the button is working correctly, the button's display changes to reverse video.
- 2. Continue pressing all buttons in turn. Only the most recently pressed button shows in reverse video.
- 3. To end the test and return to the **DIAGNOSTIC MENU**, press **Menu** twice.
- 4. If any buttons do not work, contact Dionex.

### C.9.9 Code Versions Screen

Displays revision numbers for the current digital signal processor (DSP) code, syringe operating codes, moduleware, and USB BIOS code.

CODE VERSIONS									
DSP REV n.nn SAMPLE SYRINGE REV Xnnn PREP SYRINGE REV Xnnn MODULEWARE REV n.nn USB BIOS REV n.nn									

Figure C-38. Code Versions Screen

# C.10 Time Function Out Screen

Use the **TIME FUNCTION OUT** screen to enable TTL control of an injection valve installed in an ICS-1000, ICS-1500, ICS-2000, or ICS-3000.

TIME FUNCTION OUT								
	TTL	STATE						
LOAD/INJECT VALVE:	1	ENABLED						
LOAD/INJECT VALVE:	2	DISABLED						
Help Prompt								

Figure C-39. Time Function Out Screen

Screen Field	Description	Values
LOAD/INJECT VALVE: TTL1,	If the AS is not connected to Chromeleon or Chromeleon Xpress, a TTL connection is required	ENABLED/ DISABLED
TTL2	between the AS TTL output and a TTL input on the IC system. This will allow you to control the injection valve in the IC system from the AS front panel. See <u>Appendix E</u> for TTL connection instructions.	(default= TTL1 ENABLED)

## D.1 Accessing AS Screens

- Press the **Menu** button to display a menu of screens.
- To select a screen on the menu, press the numeric button corresponding to the screen's number on the menu, or move the cursor to highlight the screen number and press **Enter**.

#### **Example**:

To go to the SYSTEM PARAMETERS screen from the MAIN STATUS screen:

- 1. Press the **Menu** button to display the **MENU of SCREENS**.
- 2. Press 5 to go to the MODULE SETUP MENU.
- 3. Press 5 again to go to the SYSTEM PARAMETERS screen.

To return to the MAIN STATUS screen:

- 1. Press Menu twice.
- 2. Press Enter or 1.

## **D.2 Entering or Changing Values**

NOTE You can enter or change screen field values only when the AS is in Local mode (see <u>Section 3.2</u>). To change most fields, the AS must also be in Direct control.

- 1. Press a cursor arrow button to position the cursor in the field.
- 2. Enter the desired value, using the following buttons.
  - To select from a list of predetermined options, press **Select** △ or **Select** ⊽; repeat to scroll through the choices.
  - To enter a numeric value, press the numeric buttons.
  - To cancel an entry that is in progress and restore the previous value, press **Delete**.

• To return a field to its default value (if an entry is not in progress), press **Delete**.

IMPORTANT

To confirm the entry, press Enter or a cursor arrow button.

## D.3 Running a Schedule of Injections from the Front Panel

This section describes how to run a schedule of injections from the AS front panel. To run a schedule (sequence) of injections from the computer, refer to the Chromeleon or Chromeleon Xpress Help or user's guide.

Before you can run a schedule, you must first create a method and schedule. See <u>Section D.5</u> for details about creating methods and <u>Section D.6</u> for details about creating schedules.

#### Notes

- The AS can run a schedule while you are entering or editing any method or schedule, even the one that is currently running.
- Changes made to the currently running method or schedule will be implemented with the next injection.
- When saving changes to the currently running method or schedule, or switching to a different schedule, the clock continues running unaffected. Only those parameter changes which affect the schedule after the current time will be implemented in the current run.
- The AS can store up to nine (1 through 9) schedules in memory.
- Methods and schedules are retained in memory even after the AS power is turned off.

## D.3.1 Selecting and Starting the Schedule

- 1. Go to the MAIN STATUS or DETAIL STATUS screen.
- Position the cursor in the control mode field and press Select ∆ or Select ∇ to select SCH (schedule) control (see Figure D-1).



Figure D-1. Main Status Screen—Schedule Control Mode

- 3. Press Enter.
- 4. Press a numeric button to select the desired schedule number or press **Select**  $\Delta$  or **Select**  $\nabla$  to scroll through the numbers (1 through 9).
- 5. Press Enter.
- 6. To start the schedule, press the **Hold/Run** button.
  - NOTE You can also select the schedule from the SCHEDULE screen (see <u>Figure D-2</u>). Enter the number in the RUN field and press Enter. Press the Hold/Run button to start the schedule.

SCHED	DULE		ED	IT	1	S	AVE	ТО	1	R	UN	1
MISSING VIAL ACTION: SKIP												
VIAL#					IN	J/	IN	J				
LINE	STAR	<b>T-</b>	END	)	VI	AL	VC	)L(ul	_)	ME	ГНО	D
1-4	1	-	4			3	2	5.0	-		1	
5	5	-	5			1	2	5.0			1	
6	6	-	6			1		5.0			1	
Help Prompt												

Figure D-2. Schedule Screen

## D.3.2 Placing a Running Schedule on Hold

To place a running schedule on hold, press Hold/Run or Stop.

The AS will finish the current method step before suspending operation.

While the schedule is on hold, the time since the last injection continues to count up. If the schedule is put on hold during a method's timed events, the timed events clock is suspended until the schedule resumes. See Section 2.7.7 for details about the clock displays.

To resume running the schedule, press Hold/Run.

NOTE If wait is enabled on the SYSTEM PARAMETERS screen (see <u>Section C.6.5</u>), the schedule is automatically put on hold when it reaches the wait step in a sample prep phase. The schedule will resume running only if a TTL signal is received (see <u>Section E.2</u>) or if the Hold/Run button is pressed.

### D.3.3 Stopping a Running Schedule

- 1. To halt a schedule, press **Stop**.
- 2. When the following message appears, press **Stop** again.



Figure D-3. Stop Message Screen

When a schedule is aborted, the AS performs the following sequence of operations:

• The sampling needle arm moves to the waste port and empties any fluid from the needle.
- The needle arm moves to the flush port and delivers a volume of flush fluid that washes the outside of the needle.
- The needle arm then moves to the inject port, where a volume of flush fluid is delivered for flushing the inject port.
- The needle arm moves back to the flush port, and the outside of the needle is again flushed.
- The needle arm moves to the home position.

## D.4 Running Under Direct Control from the Front Panel

#### NOTE This section describes how to control the AS directly from the front panel (Local mode). To run under direct control from the computer (Locked Remote mode), refer to the Chromeleon or Chromeleon Xpress Help.

The AS must be in Direct Control to initiate a manual flush or priming cycle, or to perform certain maintenance or diagnostic procedures.

The following AS functions can also be controlled directly (not as part of a schedule):

- TTL and relay outputs
- Injection valve position
- Injection valve 2 position (if installed)
- Diverter valve position (if installed)
- Tray temperature set point (if installed)

#### **Selecting Direct Control Parameters**

- 1. Go to the **DETAIL STATUS** screen (press **Menu** and **2**).
- If the AS is not in Direct Control mode, position the cursor in the control mode field and press Select ∆ or Select ∇ to select DIRECT CONTROL. Press Enter.

DETAIL		IDLE			
VIAL#:		TIME:	min	TTL1	1
INJ#:		VOL:	uL	TTL2	0
METHOD:		TRAY: 2	mL	RLY1	0
<b>TRAY: 25</b>	20 °C↑	LOOP: 10	0uL	RLY2	0
		NORMAL	FULL	INJ VLV	
LOCAL	DIF	RECT CON	TROL		
Help Promp	t				

Figure D-4. Detail Status Screen Example

- 3. Position the cursor in the field for the parameter to be changed.
- 4. To set the **TRAY** temperature:
  - a. If the field displays "--", temperature control is currently turned off. Press **Select**  $\Delta$  or **Select**  $\nabla$  to turn on temperature control.
  - b. Press numeric buttons to enter the desired temperature setting.
- 5. For other parameters, press **Select**  $\triangle$  or **Select**  $\nabla$  to select the desired setting.
  - TTL and relay fields: Select 1 for on or 0 for off.
  - Inject valve(s): Select L to set the valve to load or I to set the valve to inject.
- 6. After setting a parameter, press Enter.
  - NOTE The TRAY field is available only when the sample temperature control option is installed. Tray temperature can be set to between 4 °C and 60 °C.

#### IMPORTANT

Dionex does not recommend using the 10 mL plastic tray with the sample temperature control option because the plastic tray is inefficient at heating and cooling the vials. In addition, the tray is not designed for temperatures above 40 °C (104 °F) and may become deformed over time if used above 40 °C.

## D.5 Creating Methods from the AS Front Panel

NOTE This section describes how to create methods from the front panel (Local mode). To create methods (programs) from the computer, refer to the Chromeleon or Chromeleon Xpress user's guide or Help.

A method defines a series of operating instructions that tells the AS how to perform a single injection. A method consists of three phases:

- Sample prep (Section D.5.3)
- Method setup (Section D.5.4)
- Timed events (Section D.5.5)

Parameters for each phase are defined on separate screens. A method definition always includes the parameters from all three of the screens.

#### Notes

- The method phases are run in the order listed above. See <u>"Local/Schedule</u> <u>Control Mode" on page 28</u> for details.
- A method can have a total of 100 steps, including the sample prep steps, the method setup (which counts as one step), and the timed events steps.
- You can create a new method when the AS is running, on hold, or stopped.

## D.5.1 Creating a Method

1. Press **Menu** and **3** to go to the **METHOD MENU** screen (see Figure D-5).

METHOD MENU	EDIT 25	SAVE TO 25
1 SAMPLE PREP 2 SETUP 3 TIMED EVENTS		
Help Prompt		

Figure D-5. Method Menu

- 2. In the **EDIT** field, enter the number of an unused method.
- 3. Press Enter.
- 4. Go to each method screen and enter the method parameters.
  - For details about defining sample prep steps, see Section D.5.3.
  - For details about selecting method setup parameters, see <u>Section D.5.4</u>.
  - For details about defining timed events, see <u>Section D.5.5</u>.
- 5. Save the method (see <u>Section D.5.2</u>).

## D.5.2 Saving a Method

- 1. From any of the three method screens, press the **Menu** button to return to the **METHOD MENU**.
- 2. Position the cursor in the **SAVE TO** field.
- 3. Do one of the following:
  - To save the current method, press Enter.
  - To save to a different method, enter a new number and then press **Enter**.

## D.5.3 Defining Sample Prep Steps

### Overview

Use the **SAMPLE PREP** screen (see <u>Figure D-6</u>) to specify a sequence of operations to be performed before the method's timed events. Six standard sample prep operations are available: **DELAY**, **NEEDLE**, **PIPET**, **MIX**, **FLUSH**, and **WAIT**. In addition, if the sample preparation option is installed, **DISPENSE** and **DILUTE** operations are added. If a concentrator is installed, **REAGENT FLUSH** and **REAGENT PRIME** operations are added.

Sample Prep Operation	Description
DELAY	Specifies for how long the AS waits before proceeding to the next step in the sample prep phase.
NEEDLE	The height of the needle above the bottom of the vial.
PIPET	Moves a specified volume of sample from one vial to another.
МІХ	Mixes the vial contents by repeatedly drawing and expelling a volume of sample.
FLUSH	Flushes the needle with a specified volume.
	This operation is independent from the flush that takes place before each injection in a schedule (see Section 2.7.6).
DISPENSE	(Sample prep option only) Moves a specified volume of reagent from a reservoir to a vial.
DILUTE	(Sample prep option only) Dilutes a volume of concentrate with a specified volume of diluent.
REAGENT PRIME	(Concentrate mode only) Primes the lines with reagent.
REAGENT FLUSH	(Concentrate mode only) Flushes the concentrator with reagent.
WAIT	The AS waits for a continue command before proceeding with the method. The command can be from the front panel (pressing <b>Hold/Run</b> ) or a TTL input.

- Except for **WAIT**, each operation can be performed multiple times, or not at all.
- The sample prep steps must include a single wait step. The wait operation can be disabled, however, from the **SYSTEM PARAMETERS** screen (see <u>Section C.6.5</u>).

• When multiple injections are scheduled from a single vial, the sample prep phase is performed once only (before the first injection). The method setup and timed events are performed for each injection.

## **Specifying Vial Positions**

Vial positions in prep operations can be specified by an absolute number or a relative position.

• To specify an absolute number, press the numeric button(s) for the desired position. The valid vial positions depend on the type of tray installed:

Tray Type	Valid Vial Numbers
1.5 mL, aluminum	1 to 100
10 mL, plastic	1 to 49

## NOTE To sample from a well plate, the AS must be under Chromeleon or Chromeleon Xpress control.

To enter a relative position, press Select △ or Select ∨ to scroll through the choices. SMP is the sample vial specified for the current injection in the schedule. S+1 is one vial past the sample vial. S+2 is two vials past the sample vial, and so on, up to S+9. WST is the waste port.

#### **Defining Sample Prep Steps**

1. On the METHOD MENU, move the cursor to the 1 field (SAMPLE PREP) and press Enter to display the SAMPLE PREP screen (see Figure D-6).

WAIT		
Prompt		
	<b>WAIT</b> Prompt	Prompt

Figure D-6. Sample Prep Screen (Default)

2. Press **Insert** to add a new step above the wait step. A **DELAY** step of 0 minutes is added by default (see Figure D-7).



Figure D-7. Example Sample Prep Screen

- To select a prep operation, move the cursor one field to the right and press Select △ or Select ∇ to scroll through the list of operations. As you select each type of operation, the parameters for that operation are displayed to the right.
- 4. Press Enter or a cursor arrow to select an operation.
- 5. After selecting an operation, enter its parameters. Refer to the following sections for details about each sample prep operation.

NOTE To delete a sample prep step, position the cursor in the step's leftmost column and press **Delete** twice.

#### Using the Wait Operation

The wait operation puts a method on hold until the AS receives a run command from the front panel **Hold/Run** button or a TTL input signal.

- The AS inserts a single wait operation at the end of every method's sample prep phase.
- You cannot delete the wait operation from the sample prep phase, but you can change its position or disable it.

The wait operation's position in the sample prep phase determines whether samples are run overlapped or sequentially. When the wait is the last sample prep step (the default), samples are overlapped (see <u>Section 2.7.2</u>). When the wait is the first sample prep step, samples are run sequentially (see <u>Section 2.7.3</u>).

## Changing the Position of the Wait Operation in the Sample Prep Phase

- To insert a step above the wait line, position the cursor on the wait line and press the **Insert** button.
- To insert a step below the wait line, position the cursor on the blank line below wait and press the **Insert** button.

#### **Enabling and Disabling the Wait Operation**

- 1. If you are editing a method, save the current settings (see <u>Section D.5.2</u>).
- 2. Go to the **MODULE SETUP** menu and press **5** to go to the **SYSTEM PARAMETERS** screen.
- 3. Position the cursor in the WAIT field and press Select  $\Delta$  or Select  $\nabla$  to select ENABLED or DISABLED.
- 4. Press Enter.

#### Specifying a Delay Between Prep Steps

The **DELAY** operation specifies the number of minutes the AS waits before proceeding to the next step in the sample prep phase.



## Specifying a Needle Height

The **NEEDLE** operation specifies the height of the needle above the bottom of the vial. This height is used for **PIPET**, **MIX**, and **DISPENSE** operations. When pipetting, the height applies only to the source vial. The height is measured from the tip of the needle to the bottom of the vial. Zero is closest to the bottom of the vial; 46 is closest to the top of the vial.

NOTE For 0.3 mL and 1.5 mL vials, use a maximum needle height of 2 mm, to ensure the needle's vent is positioned correctly inside the vial.



NOTE The needle height specified here does not change the default sample needle height specified on the METHOD SETUP screen (see Section D.5.4).

## Pipetting Liquid from One Vial to Another

The **PIPET** operation uses the sample syringe to transfer liquid from a source vial to a destination vial. After pipetting, mix the destination vial before sampling from it. Be sure to specify enough mix cycles to completely mix the vial contents (see "Mixing a Vial" below).



NOTE Unlike the DILUTE operation, the PIPET operation does not include an automatic mixing step. If the sample preparation option is installed, use the DILUTE operation, not PIPET, for dilutions (see <u>"Diluting" on</u> page 288).

#### **Mixing a Vial**

The AS mixes the vial by repeatedly drawing up and expelling a volume of the contents. The prep syringe is used if the sample preparation option is installed; otherwise, the sample syringe is used. When specifying the volume, enter a value that is less than or equal to the volume of the syringe. If the volume is greater than the syringe volume, an error message occurs and the syringe volume is used.



## **Flushing the Needle**

The sample syringe is used to flush the needle with the specified volume of liquid. If you specify a volume greater than the syringe volume, multiple flushes are done.



## **Dispensing from a Reagent Reservoir**

**DISPENSE** is available only if the sample preparation option is installed. During dispensing, the prep syringe moves a volume of reagent from a reservoir to a vial.



## Diluting

**DILUTE** is available only if the sample preparation option is installed. The following steps occur during a dilution:

- The sample syringe picks up a volume of concentrate from the concentrate vial and dispenses it to the destination vial.
- The prep syringe dispenses a volume of diluent to the destination vial.
- The sample syringe mixes the destination vial five times, using 80% of the volume in the vial.
- The sample syringe is flushed with a volume equal to the amount of concentrate plus one full syringe stroke.



## **Reagent Prime after Concentrator Loading**

NOTE REAGENT PRIME and REAGENT FLUSH are included in the sample prep steps. However, they are not performed until after the Load (concentrate) step in the timed events (see <u>Section D.5.5</u>).

**REAGENT PRIME** is used to prime the sampling needle assembly with reagent after sample has been loaded onto a concentrator column. See <u>Section 3.6.10</u> for details about concentrator injections. See <u>Section 2.3.3</u> for details about the sampling needle assembly.

The following steps occur during reagent prime:

- (Optional) The inject valve is switched to a specified position (load or inject). For most applications, the valve position is not changed.
- The needle arm moves to the waste port.
- The prep syringe delivers the specified volume of reagent from the reagent reservoir. This primes the sampling needle assembly with reagent.



NOTE Although it is possible to select a vial or the flush reservoir as the reagent source, REAGENT PRIME is designed to be used only when a reagent reservoir is the selected reagent source and the prep syringe is installed.

#### **Reagent Flush after Concentrator Loading or Reagent Priming**

**REAGENT FLUSH** is used to flush reagent onto the concentrator column after sample is loaded. See <u>Section 3.6.10</u> for details about concentrator injections.

The following steps occur during reagent flush:

- (Optional) The inject valve is switched to a specified position (load or inject). For most applications, the valve position is not changed.
- If the reagent source is a vial, the needle arm moves to the vial and picks up the volume of reagent (see the notes below for volume ranges and defaults).
- The needle arm moves to the inject port.
- The syringe delivers the specified volume of reagent from the reservoir to the inject port. From there it moves onto the concentrator. The sample syringe and the flush reservoir are used by default for reagent flushing. If the sample prep option is installed, the prep syringe is used and a reagent reservoir can be specified.

V	blume to Flush	Reagent Source Inje	ect Valve Position
RGNT_FLH	1000 uL	FLH VIv	Ň
	Volume	Source	Inject Valve
	100.0 to 50,000 μL (see notes)	Vial or flush reservoir. Also, RES A, B, C, or D, if sample prep is installed	L (load) I (inject) N (no change)

#### Notes on reagent prime and flush volume ranges:

• If the source is a reservoir, the allowed range is 100 to 32,600  $\mu$ L. The default is 2,000  $\mu$ L if the syringe volume is less than 5,000  $\mu$ L, and 10,000  $\mu$ L if the syringe volume is 5,000 or 10,000  $\mu$ L.

Sample Syringe Volume ( $\mu L$ )	Reagent Prime/Flush Range ( $\mu L$ )	Default (μL)
100 to 1,000	100 to 1,000	1,000 (any size vial)
5,000	100 to 4,700	1,000 (1.5 mL vial) 4,000 (10 mL vial)
10,000	100 to 8,000	1,000 (1.5 mL vial) 8,000 (10 mL vial)

• If the source is a vial, the range depends on the volume of the syringe:

• If you specify a prime or flush volume greater than the syringe volume and the source is the flush or a reagent reservoir, the syringe takes multiple strokes to complete the operation.

## D.5.4 Selecting Method Setup Parameters

To go to the **METHOD SETUP** screen:

- 1. Go to the **METHOD MENU**.
- 2. Position the cursor on the **2** field (SETUP).
- 3. Press Enter.

Figure D-8 shows the default setup parameters.



Figure D-8. Method Setup Screen (Default)

The table below shows the value ranges for each parameter. See the following section for details about each parameter.

Parameter	Value Range	Default
Cycle Time	1 to 240 minutes	Off ()
Sample Needle Height	0 to 46 mm	2 mm*
Tray Temperature (optional)	4 to 60 °C	Off () 20 °C (when on)
Wait For Temp Stable	Yes/No	No

\*Sample needle height is measured from the bottom of the vial. Zero mm is closest to the bottom; 46 mm is closest to the top of the vial.

## Controlling the Time Between Injections (Cycle Time)

The **CYCLE TIME** parameter is an optional feature that can be used to control the time between injections. See Section 2.7.5 for details.

- 1. On the METHOD SETUP screen, position the cursor in the CYCLE TIME field.
- 2. If the field displays "--", press **Select**  $\Delta$  or **Select**  $\nabla$  to turn on the cycle time.
- 3. Press numeric buttons to enter the desired time between injections.
- 4. Press Enter.

#### Controlling the Needle's Position in the Vial

The **SAMPLE NEEDLE HEIGHT** parameter positions the tip of the needle at the specified distance above the bottom of the vial. This height is always used for sample injections. It is also the default height for mixing, pipetting, and dispensing, unless a **NEEDLE** operation is specified in the sample prep steps (see <u>Section D.5.3</u>).

- 1. On the METHOD SETUP screen, position the cursor in the SAMPLE NEEDLE HEIGHT field.
- 2. Press numeric buttons to enter the height setting.
- 3. Press Enter.

## Setting the Tray Temperature

The **TRAY TEMPERATURE** parameter is available only when the sample temperature control option is installed. Otherwise, this field displays "--" and is not illuminated.

- 1. On the **METHOD SETUP** screen, position the cursor in the **TRAY TEMPERATURE** field.
- 2. If the field displays "--", tray temperature control is currently turned off. Press **Select**  $\Delta$  or **Select**  $\nabla$  to turn on tray temperature control.
- 3. Press numeric buttons to enter the desired temperature setting.
- 4. Press Enter.

## Waiting for the Temperature to Stabilize

The **WAIT FOR TEMP STABLE** parameter delays the start of the method until the temperature of the sample tray has reached the specified set point(s).

- 1. Position the cursor in the WAIT FOR TEMP STABLE field.
- 2. Press Select  $\Delta$  or Select  $\nabla$  to select either YES (wait) or NO (do not wait).
- 3. Press Enter.

## D.5.5 Defining Timed Events Steps

To go to the TIMED EVENTS screen:

- 1. Go to the **METHOD MENU**.
- 2. Position the cursor in the **3** field (TIMED EVENTS).
- 3. Press Enter.

Figure D-9 shows the default screen.

		TIMED EVENTS	1	МЕТНО	D: 1
TIME	VALVE	TTL1	TTL2	RLY1	RLY2
INIT	LOAD	0	0	0	0
0.00	INJECT				
Help Prompt					

Figure D-9. Timed Events Screen (Default)

TIMED EVENTS METHOD: 25						
TIME	VALVE1	VALVE2	TTL1	TTL2	RLY1	RLY2
INIT	LOAD	LOAD	0	0	0	0
0.00	INJECT	INJECT				
Help Prompt						

Figure D-10. Timed Events Screen (Simultaneous Injection Option)

### Understanding the INIT and Time Zero Steps

The method timed events always start with two steps. The parameters in these steps can be changed, but the steps cannot be deleted.

- The first step is the **INIT** (initial conditions) step. The initial conditions are executed immediately after the method setup.
- The second step is the time zero step, which is executed after the **INIT** step. When the time zero step is executed, the timed events clock begins counting up from zero and any additional timed events run at their specified times.
  - NOTE When a cycle time is specified, the AS delays executing the time zero step until the specified time has elapsed since the previous injection (see Section 2.7.5 for details).

## Adding Timed Events Steps

- 1. Use one of the following methods to add a step:
  - Move the cursor to the empty **TIME** field below the last step. Enter the elapsed time at which to start the new step and press **Enter** or a cursor arrow button.
  - Move the cursor to any **TIME** field and press **Insert**. This adds a new step before the cursor position. Enter the elapsed time at which to start the new step and press **Enter** or a cursor arrow button.

After you add a step, the steps are reorganized in chronological order.

2. Enter the remaining parameters for the new step (see the following sections).

To retain a value set in the previous step, leave the field blank. A blank field means there is no change from the previous step.

## **Deleting Timed Events Steps**

To delete a step, move the cursor to the step's **TIME** field and press **Delete** twice.

## **Controlling the Injection Valve**

The VALVE parameter sets the position of the injection valve to either LOAD or INJECT. A method's timed events can include only one Load and one Inject command. The Load command must precede the Inject.

By default, the valve is set to **LOAD** in the **INIT** step and to **INJECT** in the time zero step. This allows the valve to be loaded before timed events begin. Then, you do not need to include the time for loading the loop in the timed events.

You can clear the **LOAD** and **INJECT** parameters from the **INIT** and/or time zero steps and then move them to other steps if desired.

- To clear a LOAD or INJECT parameter, position the cursor in the field and press **Delete**.
- To move the LOAD or INJECT parameter to a different step, position the cursor in the VALVE field of the desired step and press Select ∆ or Select ⊽ to select LOAD or INJECT.

#### Notes

- If you move the LOAD and/or INJECT parameters to different steps, be sure to allow enough time between the LOAD and the INJECT for the loop to be completely loaded. The time needed depends on the sample syringe volume and the syringe speed (see Section 3.7).
- If the simultaneous injection option is installed, you can set the positions of both valves in the timed events.

#### Setting TTL and Relay Outputs

The AS has two TTL and two relay outputs for control of functions in other devices. For example, if you connect TTL output 1 on the AS to TTL input 1 on a pump, a signal from the AS will turn the pump motor on and off.

In the TTL and RLY fields on the TIMED EVENTS screen, select **1** (on) or **0** (off).

See <u>Appendix E</u> for details about TTL and relay control.

## D.5.6 Editing a Method

After creating a method, you can modify it by changing, adding, or deleting steps and parameters. These changes can be made at any time. If the method you are editing is currently running, changes made to steps that have not yet been completed are performed in the current run. Changes made to steps that have already been completed are performed the next time the method runs.

- 1. Go to the **METHOD MENU** screen (press **Menu** and **3**).
- 2. In the **EDIT** field, enter the number of the method to be edited.
- 3. Go to the method screen to be changed and edit the parameters as required.
- 4. Save the method (see <u>Section D.5.2</u>). The changes can be saved to the same method number or to a new (unused) method number. If you save to a new number, the original method remains unchanged.

## D.5.7 Deleting a Method

- 1. Go to the **METHOD MENU** screen (press **Menu** and **3**).
- 2. Position the cursor in the EDIT field and press Delete.

A confirmation message appears.

3. Press **Delete** again to delete the method.

## D.5.8 Copying a Method

- 1. Go to the **METHOD MENU** screen (press **Menu** and **3**).
- 2. Position the cursor in the **EDIT** field and enter the number of the existing method to be copied.
- 3. Position the cursor in the **SAVE TO** field and enter a new (unused) method number.
- 4. Press Enter.

The parameters from the original method are copied to the new method and the original method is unchanged.

## D.5.9 Example Method for Dilution

## NOTE The sample preparation option is required for this method.

This method uses the sample preparation option to dilute a sample before injection. In this method, the AS delivers 600  $\mu$ L of the concentrated sample from vial 6 and 5400  $\mu$ L of diluent from reagent reservoir A to the sample vial. Default values are used for all other method parameters. Figure D-11 shows the sample prep steps for the method.



Figure D-11. Dilution Method Example: Sample Prep Steps

## D.6 Creating Schedules from the AS Front Panel

A schedule defines injection parameters for each vial to be sampled. A schedule consists of the following parameters:

- The vials to sample
- The number of injections to draw from each vial
- The volume of each injection
- The method to run on each injection
- The action to take when a scheduled vial is missing

You can create a new schedule or edit an existing schedule when the AS is running, on hold, or stopped. If the schedule you are editing is currently running, changes to steps that have not yet been completed are performed in the current run. Changes made to steps that have already completed are performed the next time the schedule runs.

## D.6.1 Creating a New Schedule

#### 1. Go to the **SCHEDULE** screen.

If you are opening the screen for the first time after the power is turned on, the screen displays an empty (unused) schedule (see Figure D-12). Otherwise, the screen displays the last schedule that was edited.

SCHEDU	LE	EDIT	SA	VE TO	RUN
MISSING	VIAL A	ACTION	: STO	P	
	VIAL	#	INJ/	INJ	
LINE S	TART-	END	VIAL	VOL(uL)	METHOD
1	1 -	1	1	25.0	1
Help Prompt					
1 Help Pro	1 - mpt	1	1	25.0	1

Figure D-12. Schedule Screen (Default Settings)

- If a schedule is currently displayed, position the cursor in the EDIT field. Enter the number of an unused schedule, or press Select ∆ or Select ∇ to scroll through the choices (1 through 9).
- 3. Press Enter.
- 4. Select an action to take if a vial is missing (see <u>Section D.6.3</u>).
- 5. Define the schedule lines required for your sample injections (see <u>Section D.6.4</u>).

## NOTE If injections per vial is zero for a particular vial, only the sample prep phase of the method is run.

6. Save the schedule.

## D.6.2 Saving a Schedule

- 1. Position the cursor in the **SAVE TO** field.
- 2. Do one of the following:
  - To save the current schedule, press **Enter**.
  - To save to a different schedule, enter a new number and then press **Enter**.

## D.6.3 Selecting an Action if a Scheduled Vial Is Missing

Before beginning each injection, the AS checks that a vial is present in the scheduled position in the tray. If the vial is missing, the AS can either stop and wait, or skip the vial and continue. The action is specified in the schedule.

1. In the SCHEDULE screen, move the cursor to the MISSING VIAL ACTION field and select either STOP or SKIP.

**STOP**: The AS stops when a scheduled vial is missing, allowing you to place a vial in position. To resume the schedule, press **Hold/Run**.

**SKIP**: The AS ignores a missing vial and continues to the next vial.

2. Press Enter or a cursor arrow to confirm the choice.

## D.6.4 Defining Schedule Lines

Each schedule *line* contains operating parameters for one vial. Line numbers are automatically generated when you define parameters for the vials in the schedule. If the parameters are different for each vial, line numbers are shown consecutively. If you define identical parameters for a series of vials, line numbers are shown in a range format (see Figure D-13). A schedule can include up to 999 lines.

Each range of vials (or one horizontal line on the screen) is one schedule *step* (see Figure D-13). A schedule can have up to 203 steps.



Figure D-13. Schedule Screen Example

## **Inserting Steps**

Position the cursor in the LINE field below where you want the new step to appear and press **Insert**.

## Notes

- The new step is inserted above the cursor.
- The **START** and **END** vial numbers are set to one more than the previous end number. Other parameters (injections per vial, injection volume, and method number) are copied from the previous step.
- If you insert a new step at the beginning of a schedule, parameters are copied from the first step.
- To add a step at the end of a schedule, position the cursor on the blank step at the bottom of the screen and press **Insert**.

## **Deleting Steps**

Position the cursor in the step's LINE field and press Delete twice.

#### LINE

After you enter the **START** and **END** vial numbers, the **LINE** field displays the appropriate schedule line numbers. You cannot edit the **LINE** field.

### START and END

Enter the starting and ending vials for a range of vials to be sampled.

Tray Type	Tray Capacity	Valid Vial Numbers
1.5 mL, aluminum	100 vials (1.5 or 0.3 mL)	1 through 100
10 mL, plastic	49 PolyVials	1 through 49

- A range is a group of consecutively numbered vials that will be sampled using the same schedule parameter values (injections per vial, injection volume, and method number). The starting vial can be a higher number than the ending vial.
- For a single vial, enter the same number for **START** and **END**.

## NOTE To sample from a well plate, the AS must be under Chromeleon or Chromeleon Xpress control.

#### INJ/VIAL

Enter the number of injections (0 to 99) to perform on each vial.

- If you enter 0, only the sample prep phase of the method is run.
- If you enter multiple injections per vial, the sample prep phase of the method is performed once only (before the first injection). The method setup and timed events are performed for each injection.

#### INJ VOL

Enter the volume of sample to deliver for each injection. The range allowed depends on the sampling mode and the size of the sample syringe:

Sampling Mode	Sample Syringe Size	Inject Volume Ranges	Default
Normal	Any	$1.0 - 1,000 \ \mu L$	25.0 µL
Simultaneous	5 mL	1,000–4,700 μL	1,000 μL
	10 mL	1,000–8,000 μL	1,000 μL
Concentrator	100–1,000 μL	100–1,000 μL	1,000 μL
	5 mL	100–4,700 μL	4,000 μL
	10 mL	100–8,000 μL	8,000 μL

## **METHO**D

In the **METHOD** field, enter the method to run (1 through 99) on each vial in the range.

## D.6.5 Editing a Schedule

- 1. Go to the **SCHEDULE** screen.
- 2. In the **EDIT** field, enter the number of the schedule to be edited.
- 3. Press Enter.
- 4. Change the schedule parameters.
- 5. Save the schedule. The changes can be saved to the same method number or to a new (unused) method number. If you save to a new number, the original method remains unchanged.

## D.6.6 Deleting a Schedule

- 1. Go to the **SCHEDULE** screen.
- 2. Position the cursor in the **EDIT** field and press **Delete**. A confirmation message appears.
- 3. Press **Delete** again to delete the schedule.

## D.6.7 Copying a Schedule

- 1. Go to the **SCHEDULE** screen.
- 2. Position the cursor in the **EDIT** field and enter the number of the schedule to be copied.
- 3. Position the cursor in the **SAVE TO** field, enter a new (unused) schedule number and press **Enter**. The parameters from the original schedule are copied to the new schedule and the original schedule is unchanged.

## D.7 Selecting Syringe Speeds for Front Panel Operation

For front panel operation, syringe speeds are set on the **SYSTEM PARAMETERS** screen (see Figure D-14).

NOTE The syringe speed settings described in this section are used only if the AS is controlled exclusively from the front panel. When the autosampler is controlled by Chromeleon or Chromeleon Xpress (even if it is not currently connected to the software), the AS uses the syringe speeds selected in the software (see Section 3.7).

Two syringe speed parameters are available: **METHOD SYRINGE SPEED** and **FLUSH SYRINGE SPEED**.

- The **METHOD SYRINGE SPEED** is used for the following operations:
  - Aspirating and dispensing sample during loop or concentrator loading
  - Aspirating and dispensing sample during sample prep operations, except for mixing and dilution
  - Aspirating from and dispensing to the flush reservoir during sample prep operations and loop or concentrator loading
  - Aspirating an air bubble
- The FLUSH SYRINGE SPEED is used for the following operations:
  - Flushing
  - Reagent priming after concentrator loading
  - Reagent priming and flushing after concentrator loading (when a reservoir is the specified source).

## Selecting the Method or Flush Syringe Speed

1. Go to the SYSTEM PARAMETERS screen (press Menu, 5, and 5).



Figure D-14. System Parameters Screen

- 2. Position the cursor in the **METHOD SYRINGE SPEED** field.
- 3. Press **Select** ∆ or **Select** ∨ to select the desired speed or enter the speed by pressing a numeric button. 1 is the slowest speed and 5 is the fastest. The default speed is 3, which corresponds to the speed required for water. Select a slower setting for more viscous samples.

In addition, be sure to select a speed that will keep the backpressure below 690 kPa (100 psi), taking into account the pressure from the tubing and the concentrator column (if installed). See <u>Section 3.8</u> for details.

 Position the cursor in the FLUSH SYRINGE SPEED field. Press Select ∆ or Select ∇ to select the desired speed or enter the speed by pressing a numeric button (1 through 5). The default speed is 4.

## D.8 Aligning the Sampling Needle from the AS Front Panel

Two methods are available for aligning the sampling needle:

- Automatic alignment, using Chromeleon or Chromeleon Xpress (Release 6.8 SP2 or later) (recommended method) (see <u>Section 5.8</u>)
- Manual alignment, using the front panel screens

This section describes the procedure for aligning the sampling needle manually using the front panel screens. Dionex recommends manual alignment only if Chromeleon or Chromeleon Xpress (Release 6.8 SP2 or later) is unavailable.

Align the sampling needle in the inject port after replacing the sampling needle assembly and/or the needle seal assembly, or if the needle becomes misaligned at anytime.

- **NOTE** Before aligning the sampling needle, always inspect the needle to make sure it is not bent. If the needle is bent, replace the sampling needle assembly (see <u>Section 5.4</u>).
- NOTE This procedure has numerous steps. To help maintain your place in the procedure, check off each step after completing it.
- 1. Close the autosampler door and turn on the power.
- 2. (Optional) For a better view of the needle and inject port during the alignment procedure, temporarily disable the door lock to allow operation with the door open.
  - a. On the MENU of SCREENS, press 5 and then 7 to go to the DOOR INTERLOCK BYPASS screen.



Figure D-15. Door Interlock Bypass Screen

b. Press Select ∆ or Select ∨ to select YES (bypass the door lock) and press Enter. The door lock will be disabled for 15 minutes. You can now perform the alignment with the door open. To extend the bypass for another 15 minutes, select YES and press Enter again.



Be aware of unexpected sampling arm movement when operating with the door open.



Soyez conscient de la possibilité de mouvements imprévus du bras d'échantillon lorsqu'il est utilisé avec la porte ouverte.



Achten Sie beim Betrieb mit geöffneter Tür auf unerwartete Bewegungen des Probenarms.

3. Press Menu and 6 to go to the INJECT PORT ALIGNMENT screen.



Figure D-16. Inject Port Alignment Screen

- 4. Record the current values in the **X**, **Y**, and **Z ADDRESS** fields.
  - X \_\_\_\_\_
  - Υ\_\_\_\_
  - Ζ\_\_\_\_\_
- 5. Verify that the cursor is in the X **ADDRESS** field and press **Enter**.

The needle arm moves to the inject port.

Move the cursor to the Z ADDRESS field and enter 180. Press Enter.
The needle moves down 180 steps. Note: Each step is 0.127 mm (0.005 in).

- 7. Verify that the needle is centered over the inject port opening.
- 8. Adjust the alignment by changing the **X ADDRESS** and/or **Y ADDRESS** values. Change the values in two- to five-step increments.

After each value change, press **Enter**. The needle arm moves up and then adjusts to the new position.

- To move the needle to the right, increase the **X ADDRESS** value.
- To move the needle to the left, decrease the **X ADDRESS** value.
- To move the needle forward, increase the **Y ADDRESS** value.
- To move the needle back, decrease the **Y** ADDRESS value.
- 9. After each adjustment, move the cursor to the **Z ADDRESS** field (keep the value at **180**) and press **Enter**. The needle arm moves back down. Observe the alignment and adjust the position again if needed.
- 10. When the adjustments are complete, press **Home**.

The needle arm moves to the home position.

- 11. Press Enter.
- 12. Position the cursor in the X or Y ADDRESS field and press Enter.

The needle arm moves to the selected position.

- 13. Recheck the alignment and adjust the X or Y ADDRESS values, if necessary.
- 14. Position the cursor in the **Z ADDRESS** field.
- 15. Enter the following value in the **Z ADDRESS** field: **230**.
- 16. Press Enter.

The needle moves into the inject port.

- 17. Press Home.
- 18. Press Menu three times to return to the MENU of SCREENS.

19. Press 8 to go to the DIAGNOSTICS MENU. Press 5 to go to the LIQUID CONTROL screen.



Figure D-17. Liquid Control Screen

20. Select VIAL#: INJ and press Enter.

The needle moves into the inject port.

- 21. Position the cursor in the from field and select FLUSH.
- 22. Position the cursor in the **SYRINGE** field and select **SAMPLE**.
- 23. Position the cursor in the ACTION field and select FILL.
- 24. Press Enter.

The syringe fills.

- 25. Select **SYRINGE SPEED: 5**. This speed ensures a fast enough flow rate to detect a leak.
- 26. Select ACTION: EMPTY and press Enter.
- 27. Repeat the FILL and EMPTY actions until the new needle seal tubing is filled.
- 28. Once the tubing is filled and fluid reaches the needle, empty the syringe again and monitor the top of the inject port for fluid.

## If fluid appears, wipe up the leak immediately. Then, lower the needle's Z position (see the next step).

- 29. To lower the needle's Z position:
  - a. Press Menu, Menu, 5, and 6 to return to the INJECTION PORT ALIGNMENT screen.
  - b. Increase the Z ADDRESS field value two steps and press Enter.
  - c. Repeat Step 17 through Step 26, above, and check again for leaks.

- d. If a leak occurs, continue increasing the Z address in two-step increments, checking for leaks after each increase, until no more leaks occur.
  - NOTE Increase the Z address only until no more leaks occur. If the Z address is too large, the needle will hit the bottom of the port during operation and can damage the needle or port.
- 30. When all adjustments are complete, press Home.
- 31. Factory calibration settings are recorded on a label on the inside of the autosampler compartment door. If you changed the X, Y, or Z inject port coordinates during alignment, record the new values on the label.
- 32. If you are performing this procedure after replacing the needle seal assembly, calibrate the inject port volume (see Section D.9).

## D.9 Calibrating the Inject Port Volume from the AS Front Panel

Two methods are available for calibrating the inject port volume:

- Automatic calibration, using Chromeleon or Chromeleon Xpress (Release 6.8 SP2 or later) (recommended method) (see <u>Section 5.9</u>)
- Manual calibration, using the front panel screens

This section describes the procedure for calibrating the inject port volume manually using the front panel screens. Dionex recommends manual calibration only if Chromeleon or Chromeleon Xpress (Release 6.8 SP2 or later) is unavailable.

## Note for Sequential Injections

For sequential injections, the volume to be calibrated includes the needle seal line, the path through the diverter valve, and the diverter tubing to the injection valve (see Figure D-18).



Figure D-18. Sequential Injections: Inject Port Volume Flow Paths

# NOTE This procedure has numerous steps. To help maintain your place in the procedure, check off each step after completing it.

- 1. Disconnect the needle seal line fitting from port S (5) of the injection valve. For sequential mode, disconnect the diverter valve line fitting from port S (5) on each injection valve. For simultaneous mode, disconnect the needle seal line fitting from the Y-connector.
- 2. Press Menu, 8, and 5 to go to the LIQUID CONTROL screen.

LIQUID CONTROL					
VIAL#:	1	NEEDLE HEIGHT:	46		
ASPIRATE:	100	uL from NEEDLE	_		
DISPENSE:	100	uL SYRINGE SPEED:	4		
SYRINGE:	SAMF	PLE INJ VALVE:	LOAD		
ACTION:	ASPI	RATE			
Help Prompt					

Figure D-19. Liquid Control Screen

3. Select VIAL#: INJ and press Enter.

The needle arm moves to the inject port.

## **NOTE** Throughout the procedure, retain the current or default settings for parameters, unless instructed otherwise.

4. Select the following options:

from FLUSH SYRINGE SPEED: 5 SYRINGE: SAMPLE ACTION: FILL

5. Press Enter.

NOTE To initiate a selected action, the cursor must be in the ACTION field when you press Enter.

6. Select **ACTION: EMPTY** and press **Enter**.
7. Select the following options:

**ASPIRATE**: **60**  $\mu$ L for normal, simultaneous, and concentrate modes; **120**  $\mu$ L for sequential and sequential concentrate modes (to empty the needle seal line and diverter tubing) **from NEEDLE** 

SYRINGE SPEED: 3 ACTION: ASPIRATE

- 8. Press Enter.
- 9. Select VIAL#: FLU and press Enter.

The needle arm moves to the flush port.

- 10. Select ACTION: EMPTY and press Enter.
- 11. Select the following options:

from FLUSH SYRINGE SPEED: 5 ACTION: FILL

- 12. Press Enter.
- 13. Select the following options:

DISPENSE: 50  $\mu L$  SYRINGE SPEED: 5 ACTION: DISPENSE

- 14. Press Enter.
- 15. Select VIAL#: INJ and press Enter.

The needle arm moves to the inject port.

16. Select the following options:

DISPENSE: 25  $\mu$ L for normal mode; 75  $\mu$ L for sequential mode SYRINGE SPEED: 1 ACTION: DISPENSE

17. Press Enter.

The syringe dispenses 25  $\mu$ L or 75  $\mu$ L.

18. Select the following options:

DISPENSE: 1  $\mu L$ SYRINGE SPEED: 1 ACTION: DISPENSE

19. Press Enter.

The syringe dispenses 1  $\mu$ L.

- 20. Continue pressing **Enter**, counting each key press, **until a tiny drop of liquid appears at the end of the needle seal line**. After each key press, wait until the sound of the syringe motor stops before pressing **Enter** again.
- Record the volume entered (including the volume in <u>Step 17</u> and <u>Step 19</u>). Also, notice the size of the liquid drop. This will be compared to the size of the liquid drop in <u>Step 34</u>.
- 22. Select the following options:

```
ASPIRATE: 60 \muL for normal mode; 120 \muL for sequential mode (to empty the needle seal line and diverter tubing) from NEEDLE
```

SYRINGE SPEED: 3 ACTION: ASPIRATE

- 23. Press Enter.
- 24. Select VIAL#: FLU and press Enter.

The needle arm moves to the flush port.

25. Select the following options:

SYRINGE SPEED: 5 ACTION: EMPTY

- 26. Press Enter.
- 27. Select the following options:

from FLUSH SYRINGE SPEED: 5 ACTION: FILL

28. Press Enter.

29. Select the following options:

DISPENSE: 50  $\mu L$ SYRINGE SPEED: 5 ACTION: DISPENSE

- 30. Press Enter.
- 31. Select VIAL#: INJ and press Enter.

The needle arm moves to the inject port.

32. Select the following options:

DISPENSE: volume (microliters) recorded in <u>Step 21</u> SYRINGE SPEED: 3 ACTION: DISPENSE

- 33. Press Enter.
- 34. Verify that a drop of liquid appears at the end of the needle seal tubing. If no drop appears or if the drop is a different size than previously observed, repeat <u>Step 7</u> through <u>Step 34</u>.

For sequential mode, switch the diverter valve to the other position and repeat Step 22 through Step 33.

- 35. Select VIAL#: FLU and press Enter.
- 36. Select the following options:

SYRINGE SPEED: 5 ACTION: EMPTY

- 37. Press Enter.
- 38. Press **Home** to move the needle arm to the home position.
- 39. Press Menu twice to go to the MENU of SCREENS.

40. Press 5 twice to go to the SYSTEM PARAMETERS screen.



Figure D-20. System Parameters Screen

- 41. In the **INJECT PORT VOLUME** field, enter the volume (microliters) recorded in <u>Step 21</u>. For sequential mode, enter the volume recorded for system 1 in the **S1** field and the volume for system 2 in the **S2** field.
- 42. Press Enter.
- 43. Factory calibration settings are recorded on a label on the inside of the autosampler compartment door. Record the new inject port volume on the label.
- 44. If you are performing this procedure after replacing the needle seal assembly:
  - a. Slide the bolt and ferrule removed from the old needle seal line onto the end of the new needle seal line, or install a new bolt and ferrule fitting.
  - b. Connect the needle seal line fitting to the injection valve. Tighten the fitting fingertight and then tighten it an additional one-quarter turn using the 5/16-in wrench.

#### E.1 TTL and Relay Connections

A 12-pin connector strip for TTL/relay control is located on the AS rear panel. The connector provides two relay outputs, two TTL outputs, and four TTL inputs (see Figure E-1).



Figure E-1. TTL and Relay Connector on Rear Panel



Relay loads in excess of 200 mA or with included power supplies over 60 V may damage the relay drivers on the CPU.

The outputs can be used to control functions in external devices such as a Dionex IC system module. When connected to a controlling device, the inputs can be programmed to perform the following AS functions:

- Start and continue a schedule
- Turn the sample (tray) temperature control option on/off

Relay outputs 1 and 2 can be programmed to switch any low-voltage control. Switched current must be less than 200 mA and 42 V peak.

#### E.1.1 Connecting a TTL or Relay

 Locate the twisted pair of wires (P/N 043598) and the 12-position connector plug (P/N 923686) (see <u>Figure E-2</u>) in the AS Ship Kit (P/N 062380 or P/N 062381).



Figure E-2. 12-Position Connector Plug

- 2. Follow these basic steps to connect the TTL or relays.
  - a. For each relay or TTL to be used, connect an active wire (red) and a ground wire (black) to the 12-position connector plug at the appropriate pin locations. Refer to Figure E-1 or the label on the AS rear panel for the connector pin assignments.
  - b. To attach a wire to the plug, strip the end of the wire, insert it into the plug, and use a screwdriver to tighten the locking screw. If necessary, multiple ground wires can be attached to a single TTL input/output ground pin.



When attaching wires to the connector plug, be careful not to allow stray strands of wire to short to the adjoining position on the connector.

c. Plug the connector into the 12-pin connector on the AS rear panel.

- d. Connect the wires from the AS connector plug to the TTL or relay connector pins on the other module(s). Additional connector plugs are provided with other Dionex modules.
- NOTE Check the polarity of each connection. Connect signal wires to signal (+) pins and ground wires to ground (-) pins.

#### E.1.2 Selecting TTL Input Control Types

The AS TTL inputs respond to four types of signals to accommodate different controlling devices. The default control type, **Normal Edge**, is compatible with the output signals provided by Dionex modules.

If the device connected to the AS does not send a normal edge signal, select the appropriate control type. Refer to the documentation provided with the controlling device and the information below to select the correct type. Select a different control mode from the **TIME FUNCTION IN** screen (see Figure E-3).



Figure E-3. Time Function In Screen–Selecting Signal Modes

#### **TTL Input Control Types**

• Normal Edge: In normal edge operation, the negative (falling) edge of a signal turns on the function. For example, for the **START-CONTINUE** function, the negative edge starts the



schedule (if it is not currently running), or continues the current schedule if it is on hold.

The action of the positive (rising) edge depends on the function: For the **START-CONTINUE** function, the rising edge has no effect. For the **TRAY TEMP ON/OFF** function, the rising edge turns off sample temperature control.

- *Inverted Edge*: The inverted edge mode works identically to the normal edge mode except that the positive and negative edges are reversed in function.
- *Normal Pulse*: In normal pulse operation, the negative (falling) edge of the TTL signal is the active edge and the positive (rising) edge is ignored.



A pulse width of 50 ms or more is guaranteed to be detected. A pulse width of 4 ms or less is guaranteed to be ignored. The action for pulse widths that are greater than 4 ms and less than 50 ms is undefined.

• *Inverted Pulse*: The inverted pulse mode operates identically to the normal pulse mode except that the positive and negative edges are reversed in function.



#### E.2 Controlling TTL and Relay Outputs

The AS provides two TTL outputs and two relay contacts for control of functions in external devices, such as an integrator. The relay outputs can be used to switch any low-voltage control. Switched current must be less than 200 mA and 60 V peak blocking. The relay-contact closures are normally open. When the relay is closed, current flows to the connected device.



# Relay loads in excess of 200 mA or with included power supplies over 60 V may damage the relay drivers on the CPU.

The TTL outputs are normally at 5 volts. Setting a TTL output to 0 volts turns on the action in the connected device.

After connecting the TTL and Relay outputs (see Section E.1), turn on and off the output states in Direct Control from the **DETAIL** screen (see Figure E-4), or in Schedule Control during the timed events portion of the method. You can also use Chromeleon or Chromeleon Xpress to control the outputs.

- To turn on a TTL or relay output, set the corresponding output field on the **DETAIL** screen or method **TIMED EVENTS** screen to 1 (on).
- To turn off a TTL or relay output, set the corresponding output field to 0 (off).

DETAIL		IDLE			
VIAL#:		TIME:	min	TTL1	1
INJ#:		VOL:	uL	TTL2	0
METHOD:		TRAY: 2	mL	RLY1	0
<b>TRAY: 25</b>	20 °C↑	LOOP: 10	0 uL	RLY2	0
		NORMAL	FULL	INJ VLV	
LOCAL	DII	RECT CON	TROL		
Help Prompt					

Figure E-4. Detail Status Screen - Direct Control of TTL and Relays

#### E.2.1 Example TTL/Relay Connections

The following example connections can be used to connect AS relay and TTL outputs to TTL inputs on an ICS-1000/1500/2000 system. These connections allow the AS to control the following functions:

- Injection valve position
- Pump flow
- Detector autozero
  - NOTE Refer to the ICS-1000/1500/2000 operator's manual for an example of how to set up the AS and the IC system for *stand-alone operation* (operation without Chromeleon or Chromeleon Xpress software).



Figure E-5. Example TTL/Relay Connections: ICS-1000/1500/2000

# **F** • Reordering Information

Item		
Vials and Vial Trays		
Vials, 0.3 mL polymer (package of 100)		
Vials, 1.5 mL glass (package of 100)		
Vials, 1.5 mL plastic (package of 100)		
Vials, 10 mL PolyVial (package of 100)		
Tray, insulated aluminum, for 0.3 or 1.5 mL vials, 100-vial capacity, compatible with sample temperature control		
Tray, plastic, for 10 mL vials, 49-vial capacity		
Vial tray covers		
Well Plates and Well Plate Trays		
Micro-well plate tray and two well plates with 96, 0.5 mL round (U-shaped) wells, and two well plate covers <i>(not suitable for IC applications)</i>		
Deep-well plate tray and two well plates with 96, 2 mL wells, and two well plate covers <i>(suitable for IC applications, where concentration of ions is above 1 ppm)</i>		
Well plates, 96 micro-well, 0.5 mL round (U-shaped) wells (package of 10) <i>(not suitable for IC applications)</i>		
Well plates, 96 micro-well, 0.45 mL conical (V-shaped) wells (package of 20) <i>(not suitable for IC applications)</i>		
Well plates, 96 deep-well, 2 mL wells (package of 5) (suitable for IC applications, where concentration of ions is above 1 ppm)		
Well plate covers for 0.45 mL and 0.5 mL well plates (package of 10)		
Well plate covers for 2 mL well plates (package of 10)		
Well plate thermal pad		
Reservoirs		
Flush reservoir		

Part Number	Item				
062596	Reagent reservoir				
с ·					
062382	Syringes				
062370	Dual comple syringe pump assembly				
062265	Duai-sample symmetry pump assembly				
062363	Sample syringe velve				
054242	Sample syringe valve				
054243	Prep syringe valve				
061315	Tetlon washer (for sample and prep valve ports)				
055064	Syringe, 100 µL				
053916	Syringe, 250 µL (standard)				
055065	Syringe, 500 µL				
055066	Syringe, 1,000 µL				
055067	Syringe, 2.5 mL (prep syringe use only)				
053915	Syringe, 5 mL (standard for prep syringe)				
055068	Syringe, 10 mL				
Tubing and Fittings					
063405	PEEK tubing: Violet, 0.18-mm (0.007-in) ID				
042690	PEEK tubing: Black, 0.25-mm (0.010-in) ID				
049714	PEEK tubing: Blue, 0.33-mm (0.013-in) ID				
042855	PEEK tubing: Orange, 0.50-mm (0.020-in) ID				
044777	PEEK tubing: Green 0.75-mm (0.030-in) ID				
052113	PFA tubing: 0.78-mm (0.032-in) ID				
052112	PFA tubing: 1.55-mm (0.062-in) ID				
048949	Ferrule fitting, 1/8-in flangeless				
052267	Nut, 1/4-28 x 1/8-in flangeless				
052231	Ferrule fitting, 1/16-in flangeless				
052230	Nut, 1/4-28 x 1/16-in flangeless				
043276	Ferrule fitting, double-cone				
043275	Bolt, 10-32 double-cone				
039056	Union, 14-28 x 1/8-in				

Part Number	Item			
Tubing Assemblies				
042857	Sample loop, 25 µL PEEK			
057301	Needle seal assembly, 18-in, with 10-32 ferrule fitting			
061290	Needle seal assembly, 18-in, without 10-32 ferrule fitting			
062564	Needle seal assembly, 24-in, with 10-32 ferrule fitting			
054271	Sampling needle assembly, 1.2 mL volume			
061267	Sampling needle assembly, 8.5 mL volume			
052267	Nut, 1/4-28 x 1/8-in flangeless (for sampling needle fitting)			
064553	Ferrule fitting, 1/4-28 (for sampling needle fitting)			
Kits				
057896	Valve Rebuild Kit (for injection or diverter valves)			
062578	Second AS Kit (for connecting a second AS to a dual ICS-3000 system)			
063294	Diverter Valve Kit (for sequential injections)			
060581	AS Preventive Maintenance Kit			
Miscellaneous Items				
055075	Drain line			
053669	Leak sensor			
960777	USB cable			
043598	Twisted pair of wires (for TTL/relay connections)			
923686	12-position connector plug (for TTL/relay connections)			
954745	Fuse, 3.15 amp fast-blow IEC 127			

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