



**Sample Sentinel**

February 1997

**MF-9069**

**INSTRUCTION MANUAL**

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Autosampler

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Bioanalytical  
Systems, Inc  
2701 Kent Avenue  
West Lafayette  
Indiana 47906

MANUFACTURER'S NOTE

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Bioanalytical Systems, Inc.  
2701 Kent Avenue  
West Lafayette, IN USA 47906-1382  
Phone: (765) 463-4527  
Fax: (765) 497-1102

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## **Preface**

### **Safety Information**

#### **INSTRUMENT CERTIFICATION**

In accordance with BAS' commitment to customer service and safety, this autosampler and its accompanying documentation have satisfied the requirements for, and have received certification from TÜV/GS (Technischer Überwachungs-Verein Rheinland), CSA (Canadian Standards Association), and the FCC (Federal Communications Commission). VDE (Verband Deutscher Elektrotechniker certification is pending. This product complies with the following CSA Standard 022.2 regulations:

- 0-M1982 - General Requirements (Canadian Electrical Code, Part II)
- 0.4-M1982 - Bonding and Grounding of Electrical Equipment (Protective Grounding)
- 151-M1986 - Laboratory Equipment

Designed and tested according to DIN 57411 Pt. 1/VDE 0411 Pt. 1, Safety Requirements for Electronic Measuring Apparatus, this autosampler has been shipped to you from our manufacturing facility in a safe condition.

#### **IDENTIFYING SAFETY INFORMATION**

This reference manual contains warnings and precautionary statements that can prevent personal injury, instrument damage, and loss of data if properly followed. All statements of this nature are called to your attention through the use of the following:

#### **CAUTION!**

#### **WARNING!**

#### **HIGH VOLTAGE!**

#### **SPECIFIC HAZARDS**

Every instrument has specific hazards, so be sure to read and comply with the following precautions. They will help ensure the safe, long-term use of your autosampler.

1. Only use fuses of the type and current rating specified. Do not use repaired fuses and do not short-circuit the fuse holder.
2. The supplied power cord must be inserted into a power outlet with a protective earth contact (ground). When using an extension cord, make sure that the cord also has an earth contact.

***Chemical Toxicity***

Although the large volume of toxic and flammable solvents used and stored in laboratories can be quite dangerous, don't ignore the potential hazards posed by your samples. Take special care to read and follow all precautions that ensure proper ventilation, storage, handling, and disposal of both solvents and samples. Become familiar with the toxicity data and potential hazards associated with all chemicals by referring to the manufacturers' Material Safety Data Sheet (MSDS).

***Sample Preparation***

Always consider the solubility of your sample in the mobile phase. Sample precipitation can plug the system by obstructing the flow through the injector and/or the column. This obstruction may result in irreparable damage to parts of the system. Particulate matter can be avoided by filtering the samples through 0.45- or 0.2-micron (or less) filters (see above).

***Solvent Requirements***

Many chemical manufacturers provide a line of high-purity or HPLC-grade reagents. Routine filtration of all solvents or eluants through a 0.45- or 0.2-micron (or less) fluorocarbon filter before placing them in the solvent reservoir will prolong the life and effectiveness of the inlet filters, check valves, seals, injectors, and columns.

Choose a mobile phase that is compatible with the sample and column you have selected for your separation. Remember that some solvents are corrosive to stainless steel. Inert/bio-compatible instrument versions are also available from BAS.

***Degas the Eluants***

Degas your LC system eluants by vacuum degassing or sparging with an inert gas. Complete information for using BAS equipment to perform either of these techniques is found in separate documentation provided with degas accessories.

***Solvent Disposal***

Make sure you have a solvent waste container or other kind of drain system available at or below the benchtop level. Most solvents have special disposal requirements and should not be disposed of directly down a drain. Follow all governmental regulations when disposing of any chemical.

### ***High-Pressure Systems and Leaks***

LC systems operate at high pressures, but since liquids are not highly compressible, they do not store much energy. Thus, little immediate danger arises from the high pressure in an LC system unless a dangerous mobile phase is used, such as THF, CH<sub>3</sub>CN, DMSO, H<sub>2</sub>SO<sub>4</sub>, etc. However, if a leak occurs, it should be corrected as soon as possible. Finally, we recommend that you always wear eye and skin protection when working on an LC system and that you always shut down the system and return it to atmospheric pressure before attempting any maintenance.

*NOTE: If, for any reason, you need to return your autosampler to BAS, flush the lines with methanol.*

## **Support Policy**

### **USER UPDATES**

To activate your warranty and receive product update information news and valuable information related to this and other BAS products, fill out and return the Warranty Enrollment Card which was shipped with the instrument.

### **DAMAGED SHIPMENTS**

Breakage of any part of this instrument during shipping should be reported immediately to BAS Customer Service. You must retain the original packing box and contents for inspection by the freight handler. BAS will replace any new instrument damaged in shipping with an identical product as soon as possible after the claim filing date. Claims not filed within 30 days after the shipping date will be invalid.

Do not return damaged goods to BAS without first contacting Customer Service for a Return Authorization Number (RA#). When a defective part is returned to BAS, the RA# immediately identifies you as the sender and describes the item being returned. Bioanalytical Systems refuses all unauthorized return shipments.

### **PRODUCT WARRANTY**

BAS products are fully warranted against defects in material and workmanship. The Sample Sentinel is unconditionally warranted for 90 days from date of shipment, except when failure is due to obvious abuse or neglect, unauthorized tampering, procedures not described in manuals, or improper connection of electronic units to other components. The rotor seal and syringes are consumables, and therefore are not covered under this warranty.

For any product expressly covered under this warranty, BAS is liable only to the extent of replacement of defective items. Bioanalytical Systems, Inc. shall not be liable for any personal injury, property damage, or consequential damages of any kind whatsoever. The foregoing warranty is in lieu of all other warranties of merchantability and fitness for a particular purpose.

**SERVICE INFORMATION**

Bioanalytical Systems provides a skilled service staff available to solve your technical problems if an equipment-oriented problem should arise. For further details, call customer service personnel (1-800-845-4246), who will route your problem to the correct individual. Following discussion of your specific difficulties, an appropriate course of action will be described and the problem resolved accordingly.

DO NOT RETURN ANY PRODUCTS FOR SERVICE UNTIL A RETURN AUTHORIZATION NUMBER (RA#) HAS BEEN OBTAINED. The RA# identifies you as the sender and describes the problem you are having in full detail. Turnaround time on service can be quoted to you at the time your RA# is issued, although we can not determine the actual amount of service required until we have received your unit and diagnosed the problem. All correspondence and shipments should be sent to:

RA # \_\_ , Service Department  
Bioanalytical Systems, Inc.  
2701 Kent Avenue  
West Lafayette, IN 47906

## Start-up Checklist

This list is a brief summary of tasks that should be completed to install your autosampler.

*NOTE: Items marked with an asterisk (\*) pertain to instruments with the sample preparation option installed. Items marked with two asterisks (\*\*) are optional advanced installation procedures that should be performed only by very experienced users with specialized applications.*

## Basic Installation

### UNPACKING

- Unpack and inspect your instrument
- Remove cardboard/foam packing insert from sample tub
- Check for parts shortages
- Read the Safety Information

### REAR PANEL

- Place the autosampler with rear panel accessible
- Install flush bottle/bracket
- Install filter/solvent line and flush bottle
- Connect the external solvent tubing to tubing extensions (sample preparation only)\*
- Connect heater/mixer to an air supply\*
- Connect the power cord
- Hardwire external events, make connection to other instruments\*\*

### FRONT PANEL

- Install solvent tray/waste container
- Connect autosampler to pump - Connect pump outlet line to port 6 on injector valve
- Connect column - Connect inlet end to injector valve port 5
- Connect column to detector

### STARTUP/PRIMING

- Turn on, check initial response to power-on
- Using the 2.5 mL syringe, flush lines with 5000  $\mu\text{L}$  from the Btl reservoir\*
- Using the 250  $\mu\text{L}$  syringe, flush lines with 500  $\mu\text{L}$

**PERFORMANCE  
VERIFICATION**

- Run a standard sample
- Date and retain run results with instrument documentation
- Customize READY participation\*\*
- Run an unretained sample to determine gradient delay time\*\*
- Optimize needle height\*\*

**List of Optional Parts, Consumables****Cables**

EW-4454	DA-5/ChromGraph
EW-4456	BAS 200/ChromGraph

**Accessories**

SS-0020	Standard 2-mL vials with PTFE-lined screw caps, 144/pkg.
MF-5270	300- $\mu$ L microvials, glass, 1000/pkg. (requires SS-0025, adaptors)
MF-5272	Aluminum caps with Teflon/silicone seals for microvials, 1000/pkg.
MF-5274	Crimper for microvials, 1/pkg.
SS-0025	Glass adaptors for 300 $\mu$ L microvials, 25/pkg. (reusable)
SS-0250	Replacement sample syringe
SS-4900	Replacement needle, side cut
A-3564-010	Replacement needle, straight cut
SS-5000	Three-piece disposable microvials and holders, 100/pkg.

**Rotor Seals**

MF-5030	Vespel
MF-5029	Tefzel

3. Do not change the external or internal grounding connections. Tampering with or disconnecting these connections could endanger you and/or damage the autosampler.

*NOTE: The instrument is properly grounded in accordance with these regulations when shipped. You do not need to make any changes to the electrical connections or to the instrument's chassis to ensure safe operation.*

4. Never run the autosampler without the top cover on. Permanent damage can occur. (The door may be opened during operation.)
5. Do not turn the instrument on if you suspect that it has incurred any kind of electrical damage. Instead, disconnect the power cord and contact a BAS Service Representative for a product evaluation. Do not attempt to use the instrument until it has been evaluated. (Electrical damage may have occurred if the autosampler shows visible signs of damage, or has been transported under severe stress.)
6. Damage can also result if the instrument is stored for prolonged periods under unfavorable conditions (e.g., subjected to heat, water, etc.).
7. Always disconnect the power cord before attempting any type of maintenance.
8. Capacitors inside the instrument may still be charged even if the instrument is turned off.
9. Never try to repair or replace any component of the instrument that is not described in this manual without the assistance of BAS.
10. The autosampler is shipped with an aqueous methanol solution in the lines. Be sure to prime the autosampler with a miscible solvent as described on page 106 prior to use.

## **GOOD LABORATORY PRACTICES**

Always follow good laboratory practices whenever you operate any high-performance liquid chromatograph.

### ***Keep Good Records***

To help identify and isolate problems with either your equipment or your chromatography, we recommend that you keep good records of all system conditions (e.g., %RSDs on retention times and peak areas, peak shape and resolution, column pressure, and autosampler reproducibility). At a minimum, keep a chromatogram of a typical sample and standard mixture, well documented with system conditions, for future reference. Careful comparison of retention times, peak shapes, column pressure, efficiency, and baseline noise can provide valuable clues to identifying and solving future problems.

## Section 1. Getting Started

This section provides you with the three basic rules you'll need for using your Sample Sentinel. It also introduces you to the instrument's command center and describes the conventions we'll use in this manual.

Before you start this section, be sure that you read the Safety Information section beginning on page iii and install your autosampler as described in Section 6.

*NOTE: Throughout our explanations, we encourage you to explore the general architecture of your instrument's menus and screens. Use the Menu Tree in Section 7 as your guide if you wish.*

### 1.1 Learning Your Way Around

#### **AS EASY AS 1-2-3!**

It's easy to learn your way around a Sample Sentinel autosampler. Just remember these three rules:

1. The arrow keys ([^], [v], [<], [>]) move the cursor in the direction printed on the key.

*HINT: Press [MENU] to jump quickly to the top of the menu structure.*

2. The shape of the cursor determines how you make a selection:

- If a triangular cursor appears, press [ENTER].
- If a blinking square cursor appears, press the [+] or [-] keys to scroll up or down through preset choices, or to increase or decrease alphanumeric entries.

3. There are four ways to accept (and automatically save) an entry. Just move the cursor out of the field by any of the following methods:

- Pressing [ENTER]
- Using the arrow keys
- Pressing [MENU]
- Pressing [STATUS]

*NOTE: You won't be able to leave a menu if errors are present or if you haven't filled in all the necessary entries.*

## VISUAL CLUES

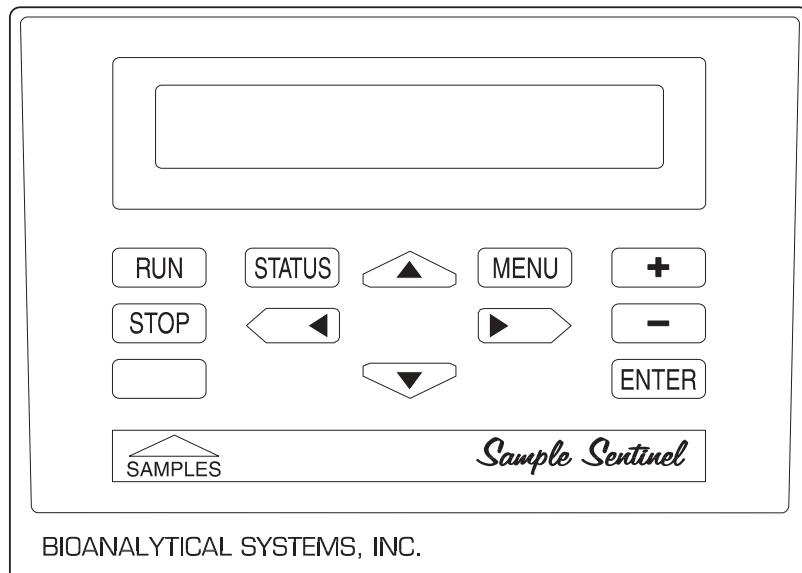
The following conventions are used on the autosampler's display:

1. Top-level menu choices are displayed in all-capital letters.
2. A field's square cursor changes to an underscore cursor when you're scrolling through preset choices or entering numerical values and characters.
3. A solid down-arrow on the right side of some displays indicates that the current menu continues on additional screens. To access additional menu lines, press the down arrow key, [v].
4. The last line of a longer menu is frequently a blank display line (without a solid down-arrow).

## 1.2 Instrument Control

Take a look at the keypad and two-line display located on the front panel (Figure 1.1). This is the command center from which you'll access menus and control the instrument's operations. A brief explanation of the keys and the main menus and screens follows.

Figure 1.1. The autosampler's front panel.



The keypad of each Sample Sentinel consists of twelve keys. Four keys directly control the instrument's operation: [RUN], [STOP], [STATUS], and a blank key called [SAMPLES]. The remaining keys either access commands ([MENU] and [ENTER]), or are used to set parameters and move around the display ([^], [v], [<], [>], [+], [-]). The function of each is explained below.

- [RUN]** Pressing [RUN] starts the run.
- The specific [RUN] operation depends on the contents of the sample queue:
1. If there are sample sets (groups of samples and associated calibration vials) in the sample queue, pressing [RUN] begins processing the first vial in the sample set.
  2. If the sample queue is empty, pressing [RUN] prompts you to press [SAMPLES] to display the Samples Menu so that you can add sample sets to the sample queue.
- [STOP]** Pressing [STOP] pauses the sample set currently running, aborts subsequent sample sets, and returns the autosampler to the initial run conditions. Refer to Section 3 for more information on the [STOP] key's function.
- [STATUS]** Pressing [STATUS] displays the Status Screen. From the Status Screen you can monitor the run in progress.
- [SAMPLES]** The [SAMPLES] key displays the Samples Menu that lets you define the resources (samples, reagents, and standards) and assign a file to each sample set. From the Samples Menu you also add sample sets to the sample queue.
- Because you manage resources and files from the Samples Menu, the [SAMPLES] key gives you real-time editing capability. That is, from the Samples Menu you can make changes to the file that is currently running. Pressing the [SAMPLES] key during a run lets you obtain information about, or edit the locations and injection volumes of, the sample and calibration vials. Sections 3 and 7 contain more information on the [SAMPLES] key.
- [MENU]** Pressing [MENU] displays the Main Menu. See below for more information.
- [ENTER]** Pressing [ENTER] accepts a selected choice or menu entry. The [ENTER] key also advances the cursor to a new field, either on the same line of the display or in the line below.
- [^], [v], [<], and [>]** Pressing any arrow key (up, down, left, or right) moves the cursor in the direction indicated on the key. The up- and down-arrow keys also move the cursor between menus and displays.
- [+] and [-]** Pressing the [+] and [-] keys scrolls you through a field's available choices or changes the value of alphanumeric entries. Holding down either key will continuously scroll the list of choices forward or backward until you release the key.

In fields that require alphabetical or numerical entries, the value of each digit is increased or decreased by one unit each time you press the [+] or [-] key. In fields that accept *either* numeric or character entries, such as the File Name field, the [+] and [-] keys scroll through the alphabet from A to Z, then through the numbers 0 to 9, and finally to a slash, hyphen, and blank space.

In other fields, the [+] key advances you through a preset list of choices while the [-] key takes you back through the list.

## MENUS AND SCREENS

Your autosampler has two kinds of displays: menus and screens. Menus require you to make selections or enter specific values. Screens display information that cannot be edited. The Menu Tree in Section 7 illustrates the structure and content of the autosampler's menus and screens.

### *Main Menu*

The Main Menu (Figure 1.2) is the top level of the menu structure. It gives you access to five other menus: FILES, COMMANDS, QUEUE, OPTIONS, and TESTS. To see the Main Menu, press the [MENU] key at any time.

---

Figure 1.2. The Main Menu.

>FILES	QUEUE	TESTS
COMMANDS	OPTIONS	

From the Files Menu you can edit, load, or delete files. From the Queue Menu you can edit or change the order of sample sets in the sample queue. The Tests Menu gives you access to the instrument's built-in diagnostics. The Commands Menu lets you run built-in instrument routines. In the Options Menu, you can set up or change your instrument's configuration. Refer to Sections 3–5 and 7–9 for more information on any of the instrument's menus.

### *Status Screen*

The Status Screen automatically appears whenever you turn on the instrument or press the [STATUS] key. The Status Screen (Figure 1.3a) displays the status of the sample queue, the current sample set, the current sample vial, and the total number of samples in the set. Pressing the down-arrow once displays the Inject Status Screen (Figure 1.3b). Pressing the down-arrow again displays the Oven/Tray Temperature Status Screen (Figure 1.3c), if these options are present. Section 3 contains more information on the Status Screen.

Figure 1.3. (top) The Queue Status Screen, (middle) the Injector Status Screen, (bottom) the Oven/Tray Temperature Status Screen.

QStatus	Set	#Samples	
IDLE	1	1/1	

Status	Vial	Vol	#Inj
READY	A01-S	1000	1/1

Oven		TrayTemp	
OFF	23	OFF	25

## MESSAGES

There are three different kinds of messages that can appear on your autosampler's display: user messages, confirmation messages, and error messages.

### *User messages*

User messages tell you about an existing instrument condition or ask for further actions. Some of these will only appear on the display for three seconds. An example of a message requiring further action is shown in Figure 1.4.

Figure 1.4. An example of a user message.

* File Protected *
No Editing Allowed

### *Confirmation messages*

Confirmation messages (Figure 1.5), indicated on the display by asterisks, appear for one second after an operation has been carried out successfully.

Figure 1.5. An example of a confirmation message.

** File Loaded **
-------------------

**Error messages**

Error messages (Figure 1.6), indicated on the display by exclamation points, are shown whenever an undesirable condition exists that prevents the instrument from carrying out an operation. Error messages remain on the display until you press a key.

Figure 1.6. An example of an error message.

```
!! Unexpected Vial in Hook !!
```

**1.3 Manual Conventions**

This manual uses several conventions. Among them are menu displays, text conventions (brackets, slashes, etc.), standard words, and several different icons.

**DISPLAYS**

Figure 1.7 shows how we will depict the two-line display. Note that, in menu illustrations, the triangular cursor location is indicated by a caret (>).

Figure 1.7. A two-line menu display.

```
>FILES          QUEUE          TESTS
          COMMANDS          OPTIONS
```

Frequently, the two lines shown on the display are only part of a longer menu. In this manual, menus having more than two lines are represented as in Figure 1.8.

Figure 1.8. A menu longer than two lines.

```
Injection Volume      1.0 (SS-2)
Injections/Sample                1
-----
Cycle Time                        2.5
Col Oven:  OFF_      Temp          18
TrayTemp:  OFF_      Temp          20
```

**COMMENTS**

Fields that appear only on some models or only when certain options are installed are documented with a comment to the right of the field. For example, in Figure 1.8, the Injection Volume field appears only on the variable-injection volume (VIV) SS-2 autosamplers.

**TEXT**

Three typographic conventions are used to differentiate between keys, menus, and fields.

***Brackets***

Brackets, [ ], indicate instrument keys. For example: Press [MENU].

***Slashes***

Slashes, / /, are used around menu choices. For example: From the Main Menu, select /FILES/.

***Capitalization***

Capitalization is used to make field and menu names appear just as they do on the display. Generally the first letters of field names are capitalized. For example: Select /FILES/, /Edit/, /Injection/.

**STANDARD WORDS**

We have also standardized the meanings of two words: "select" and "enter."

***select***

The word "select" is used when you need to choose from among available options. For example, to "select" a particular menu choice, you would move the cursor to the appropriate choice and press [ENTER]. To "select" a field entry, move the cursor to the appropriate field and use the [+] and [-] keys to scroll to the desired preset value.

***enter***

The word "enter" is used when you need to specify individual alphanumeric digits. To "enter" a particular value, move the cursor to the desired field and use the [+] and [-] keys to increment or decrement each digit in the field until the desired value or letter appears.

**1.4 What's Next?**

Now you're ready to try the practice example in Section 2, A Quick Example.



## Section 2. A Quick Example

In Section 1, you read about the three easy rules for using your autosampler's command center and some of its menus and screens. In this section you'll work through two examples. In the first example, you'll enter your run parameters from the autosampler's Samples Menu without first creating a file. In the second example, you'll create a file using the Edit Menu, load, and run that file.

Each of these examples injects a 10  $\mu\text{L}$  sample (distilled water) from the vial in position A01. In these examples you won't be changing any menu values. Instead, you'll use the default values already entered into the autosampler's menus as your run parameters. Use this practice run to make yourself comfortable with your autosampler's menus and file-entry procedures, and to become familiar with the seven steps for setting up a run.

The steps for setting up a run are as follows:

- Prepare your sample, solvents, mobile phase, vial(s), and sample tray(s)
- Enter your parameters into the Samples Menu or create an autosampler file
- Load your autosampler file
- Build your sample queue
- Enter your pump, detector, and data-output device parameters
- Install your sample tray(s)
- Start your run

The following practice examples use only a fraction of the features available on your autosampler, so we've included a more complete description of your autosampler's features and benefits first.

Before you start this section, be sure that you review the Safety Information section and complete the Start-up Checklist. This section assumes that you have installed your instrument as described in Section 6 and that your sample trays are installed.

*NOTE: Your instrument is shipped with the sample trays installed. If, for some reason your trays are not installed, refer to Section 3 for more information on installing your trays.*

## 2.1 About the Sample Sentinel Autosamplers

The Sample Sentinel autosamplers have been designed for ease of use and unsurpassed performance. Two different autosamplers are available, each with a customized set of options and capabilities to fit a wide variety of HPLC analytical applications. Sample Sentinel autosamplers may be purchased with or without tray temperature control and sample preparation options. These options are available at the time of purchase or as upgrades. Instruments purchased without the tray temperature control option hold 120 vials; those purchased with the tray temperature control option hold 105 vials.

### SS-1

The SS-1 fixed-loop autosampler functions as a stand-alone HPLC module. This instrument offers additional optimization capability: it holds four user-programmable built-in files and the sample queue can process up to 39 sample sets plus one priority sample set.

### SS-2

The SS-2 variable-injection volume (VIV) autosampler offers three injection methods: 1) full-loop, 2) traditional pull-loop where sample conservation is of primary concern, and 3) PushLoop<sup>®</sup> injection. Similar to the SS-1, it holds four user-programmable built-in files and the sample queue can process up to 39 sample sets plus one priority sample set. In addition to the built-in tray temperature control, an automated sample-preparation option with accompanying heater/mixer is also available.

## 2.2 Practice Examples

Use these examples to get acquainted with your autosampler's menus. To keep these examples simple, yet still introduce you to as many menus as possible, we'll use the instrument's default parameters.

*HINT: You may wish to refer to the Menu Tree in Section 7 as you work through this example. If at any time you lose your place, you can do one of the following:*

*Press [↶] to return to a previous screen,*

*OR*

*Press [MENU] to return to the Main Menu (the top of the menu structure) and retrace your steps.*

### PREPARE YOUR CHEMICALS

Prepare your sample, solvents, mobile phase, vial, and sample tray as follows:

1. Fill a vial from your accessory kit with distilled water. Cap it securely, and place it in position A01, in the front left corner of tray A (Figure 2.1).

*NOTE: Be sure to use either: 1) a 40–60 mil (1.0–1.5 mm) rubber or silicone septum with a 5 mil (0.1 mm) Teflon liner on one side OR 2) a single 10 mil (0.25 mm) Teflon liner by itself.*

2. Fill the flush-solvent bottle with a 70:30 MeOH:H<sub>2</sub>O solution. Refer to Section 6 for the flush-bottle installation procedure.

## ENTER YOUR PARAMETERS

There are two ways to enter your run parameters into your autosampler: from the Samples Menu and from the Edit Menu. If you have an SS-1, you will use the Samples Menu method most often. Most likely you will want to enter those parameters that are least likely to change into the Edit Menu, and those that change with each run into the Samples Menu. The Samples Menu allows you to vary individual parameters with successive runs. Should you decide to enter your parameters into the Edit Menu, there is no need to re-enter them into the Samples Menu. They are automatically copied into the Samples Menu. Refer to Section 3 for more information on entering your run parameters. For now, you may wish to familiarize yourself with both approaches for entering your parameters.

### From the Samples Menu

To enter your parameters from the Samples Menu:

1. Turn on your autosampler by pressing the on/off switch located in the lower right-hand corner of the front of the autosampler (Figure 2.1).

When the startup sequence is complete, the Status Screen appears (Figure 2.2).

2. Press the [SAMPLES] key to display the Samples Menu (Figure 2.3). When the STATUS Menu first appears, the file name field is blank. Figure 2.3 shows the how this field will appear when a file name is entered.
3. Use the [v], [<], [>], [+] and [–] keys to change the Cycle Time to 1.0.
4. Press the [v] key to move down the Samples Menu and press [ENTER] to select "Yes" to add your sample set (1) to the queue.

*NOTE: We'll tell you more about building your queue later on in this section. For now, proceed to step 5.*

5. Press [RUN] to start your run. During the run, the autosampler hook picks up the vial, carries it to the sample tower, injects the sample, returns the vial to the sample tray, and flushes the lines. When the run is complete, the instrument's status returns to "IDLE."

Figure 2.1. Front of the autosampler.

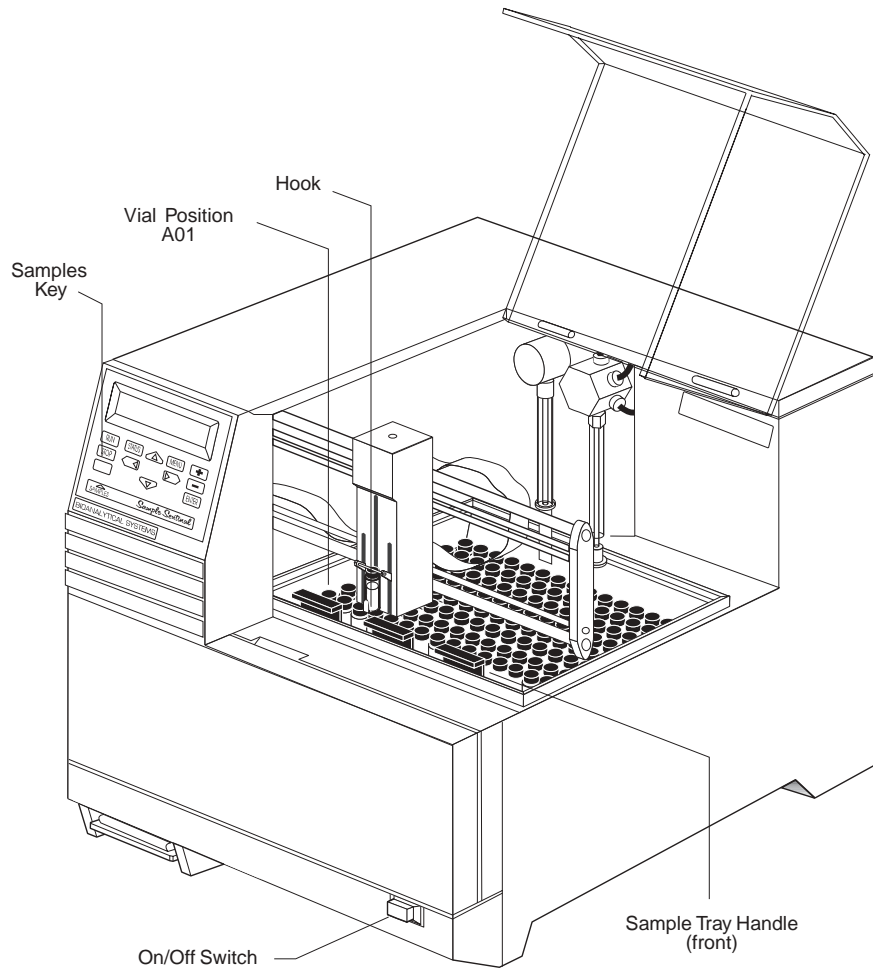


Figure 2.2. The Status Screen.

QStatus	Set	#Samples
IDLE	0	0 / 0

Figure 2.3. (top) The Samples Menu (SS-1); (bottom) the Samples Menu (SS-2).

Sample Set	1
File 1:FILENAME	
-----	
Loop Size	20
Injections/Sample	1
Cycle Time	10.0
First Sample Vial	A01
Number of Samples	1
Add to Queue?	Yes

Sample Set	1
File 1:	
-----	
Injection Volume	10
Injections/Sample	1
Cycle Time	10.0
First Sample Vial	A01
Number of Samples	1
Add to Queue?	Yes

### From the Edit Menu

In this example you'll use the Edit Menu to "create" and run the practice file. You'll use this method when you want to create more complex files and/or to create more than one file before starting a run. You will first specify a file name and then press [ENTER] in each menu field to accept its default value.

Use the following steps to create your autosampler file.

*NOTE: If you worked through the last example, the autosampler returned you to the Status Screen at the completion of the run. Go on to step 1.*

*NOTE: If you skipped the last example, turn on your autosampler and wait for the Status Screen to appear. Then proceed to step 1.*

1. Press [MENU] and select /FILES/, /Edit/ to display the Edit Menu (Figure 2.4). Notice that File 1 is already entered into the Edit File field. Press [ENTER] to edit File 1.
2. Use the [+], [-], and arrow keys to specify each character in the file name of your choice. We've chosen "PRACTICE" as our file name.

Figure 2.4. The Edit Menu.

```
Edit File                               1_
File Name: PRACTICE
-----
Injection
Calibration
Timed Events
Sample Prep  (Appears only with the Sample Preparation Option installed)
More
```

3. Move the cursor to each field in the Injection, Calibration, Timed Events, and More Menus and press [ENTER] to accept their default values. (If you were running a file other than this practice file, you would, of course, select other entries while moving through these menus.)

*NOTE: For the practice example, press the [v] key to skip the Sample Prep Menu.*

Notice that after the last field in each menu, the autosampler returns you to the next most probable choice in the menu structure. For example, after you accepted the Calibration Menu's default values, the autosampler automatically returned you to the Edit Menu with the cursor in the Timed Events field. Similarly, after you accepted the More Menu's default values, the autosampler places the cursor in the Load field of the Edit Menu. This is a pattern that occurs throughout the user interface. The instrument "anticipates" the menu choice you are mostly likely to need next. You can, of course, override that choice at any time by moving the cursor wherever you like.

## LOAD YOUR FILE

Now that you've created your practice file, use the following steps to load your practice file manually.

1. Select /Load/ from the Edit Menu.
2. Verify that File 1 is entered, and press [ENTER] to load it into the autosampler's active memory. The message "\*\*\*File Loaded\*\*\*" appears briefly. Then the Status Screen is displayed (Figure 2.5).

Loading the file manually activates any installed options (column oven or tray temperature control) that you set in the Injection Menu.

Figure 2.5. The Queue Status Screen with the queue idle.

QStatus	Set	#Samples
IDLE	0	0 / 0

## BUILD YOUR QUEUE

Build your sample queue by adding Sample Set 1 (that contains your practice sample) to the sample queue as follows:

1. Press [SAMPLES] to display the Samples Menu (Figure 2.3).
2. Press [ENTER] in each field to accept the Samples Menu's default values.

If you were injecting an actual sample instead of water, you would enter your pump, detector, and integrator/recorder or data-system parameters now. For this example, however, simply load your trays, and close the autosampler's cover.

## START YOUR RUN

To start your run, press [RUN]. If everything's running properly, the Status Screen's QStatus field will read "RUN" (Figure 2.6). The injection status field displays the time elapsed since the run was started.

Figure 2.6. The Queue Status Screen with the queue running.

QStatus	Set	#Samples
RUN	1	1 / 1

Figure 2.7. The Injector Status Screen with a run in progress.

Status	Vial	Vol	#Inj
0.05	A01-S	10	1 / 1

As you know from the first example, the autosampler injects vial A01. After the run is complete and the sample queue is empty, the vial is returned to the tray, and the instrument's queue status (QStatus) returns to "IDLE."



## Section 3. Basic Operations

This section describes the routine operation of the Sample Sentinel autosamplers. In this section you will learn the steps required for setting up and performing a run on your autosampler as follows:

- Prepare your chemicals
- Create a file that contains your autosampler run parameters
- Load your file
- Build your sample queue using the [SAMPLES] key and Samples Menu
- Start and stop a run
- Monitor the run status
- Change the parameters of a run in progress
- Insert a priority sample set into the queue

There are three options available:

- Tray temperature control
- Sample preparation (SS-2 only)
- BCD communication

The menus and fields used to control these options appear only if they are installed. This section includes the tray temperature control procedures. Section 4, *Advanced Operations*, describes the sample preparation menus (templates) and procedures. If you purchased the Sample Preparation Option, you'll first want to learn the procedures described in this section and then proceed to Section 4 to learn how to use the sample preparation templates and the heater/mixer. Contact your representative for information on the BCD option since this option requires specific installation procedures.

### Before You Start

Before beginning this section, be sure that you've completed the Start-up Checklist found in the beginning of this manual and installed your instrument according to the procedures described in Section 6.

### 3.1 Run Preparation

The Sample Sentinel autosamplers allow you to process up to 39 non-priority sample sets (groups of sample vials and associated calibration vials) within a given run. To prepare for a run, use the following steps:

- Prepare your samples, solvents, and mobile phase
- Create your file by entering the parameters for each sample set into one of the four built-in files
- Load your file
- Build your sample queue
- Start your run

Once your run has started, you can:

- Monitor the progress of the run
- Modify the currently running file (the run file) to affect subsequent vials in the active sample set
- Change the run order of, add, or delete sample sets from the sample queue
- Add a priority sample set to the queue without disrupting the calibration routine of the active sample set
- These procedures, along with the instrument's file-management features, are described in this section.

#### **PREPARE YOUR CHEMICALS**

Prepare your samples, solvents, and vials. Be sure that your samples are completely soluble in the mobile phase and that you have filtered your samples and solvents through a 0.5-micron filter. These techniques minimize sample precipitation in the lines and remove any particulate matter that could obstruct the flow through the autosampler's injector or column. Fill the solvent reservoir with flush solvent.

*NOTE: If you are using the automated Sample Preparation Option, you can use up to four solvents. Refer to Section 4 for instructions on filling your external solvent reservoirs and for setting up solvent selection in your autosampler file.*

## CREATE YOUR FILE

The group of autosampler parameters applied to a specific sample set is contained in a file. When your instrument is shipped, each of its four built-in files contains the default values. To create your own file, simply edit one of the existing files by entering your own parameters into the autosampler's Edit Menu. From the Edit Menu you can name your file and access additional menus to enter your injection, calibration, timed events, sample preparation, and other run parameters.

*NOTE: When you have entered all of your parameters in the Edit Menu, you'll have finished creating your edit file. You can't run your edit file by pressing the [RUN] key. Instead, you first have to load your edit file into the run file. For more information, see "Load your file" on page 30.*

To create your edit file, press the [MENU] key to display the Main Menu. From there, select /FILES/, /Edit/ to display the Edit Menu (Figure 3.1).

Figure 3.1. The Edit Menu.

```

Edit File           1
File Name: Vitamins
-----
Injection
Calibration
Timed Events
Sample Prep (If installed)
More

```

### Edit File

An autosampler file is identified by its number (1–4) and name, which you supply. (See "File Name," below.) The Edit File field indicates the number of the selected file. File 1 automatically appears in this field when the Edit Menu is first displayed. If you wish to edit a different file, press the [+] and [-] keys to select a different file number.

### File Name

The File Name field allows you to name your file. When you first display the Edit Menu, the File Name field contains eight blank characters. Use the [+]/[-] and [<]/[>] keys to create a file name made up of the following characters: A–Z, 0–9, \, blank, dash, or hyphen.

### Injection Menu

Selecting /Injection/ from the Edit Menu accesses the Injection Menu (Figure 3.2) from which you can enter your injection parameters. The next section describes the Injection Menu's fields.

Figure 3.2. (top) The SS-1 Injection Menu; (bottom) the SS-2 Injection Menu.

Loop Size	10.0
Injections/Sample	1
-----	
Cycle Time	10.0
TrayTemp: OFF Temp 0	<i>(If installed)</i>

Injection Volume	10.0
Injections/Sample	1
-----	
Cycle Time	10.0
TrayTemp: OFF Temp 0	<i>(If installed)</i>

**Injection Volume (Loop Size)**

Use the first field of the Injection Menu to specify the sample Loop Size (if you are using an SS-1) or the sample injection volume (if you are using an SS-2). The injection volume value depends upon the injection method and range that you entered in the More Menu's Injection Type and Injection Range fields, respectively (see page 29). To optimize injection volume, see page 169.

Full-loop injection: If you're using full-loop injection, you can use the [+] and [-] keys to select one of the following values: 5, 10, 20, 50, 100, 200, 500, and 1000  $\mu\text{L}$ .

*NOTE: Be sure that the Loop Size field value reflects the size of the sample loop installed.*

Variable-Loop Injection: You can choose either Pull- or PushLoop injection from the More Menu. These methods are described on page 29. If you use pull-loop injection, you can select a sample-injection volume from 1–1500  $\mu\text{L}$ . If you use PushLoop injection, you can specify a sample-injection volume from either 0.1–10.0  $\mu\text{L}$  or 1–200, depending upon which push type (Hi or Lo) you selected in the More Menu's Injection Type field. To achieve the highest precision, you should limit the injection volume to approximately one-half the sample loop size.

**Injections/Sample**

In this field, specify the number of injections to be made from each vial (1–99). If you have selected a sample preparation template, and want sample preparation without injections, enter 0 in this field.

*NOTE: You can't enter a 0 unless you have first selected a sample preparation template.*

### **Cycle Time**

In the Cycle Time field enter the desired time (0–655.3 minutes) between the autosampler's injections (from the injection to the end of the run). In calculating this value, be sure to allow enough time for the column to equilibrate (if you are running a gradient), all peaks to elute from the column, and for the integrator to print any post-run reports and perform any post-run calculations.

The autosampler is capable of making an injection approximately every 90 seconds, depending upon the injection and flush volumes selected. If your cycle time is controlled by a Pump Ready contact closure, you may set the Cycle Time to 0.0.

*NOTE: The default flush volume is 400  $\mu$ L, so that the total injection cycle time (including the flush cycle) is approximately two minutes.*

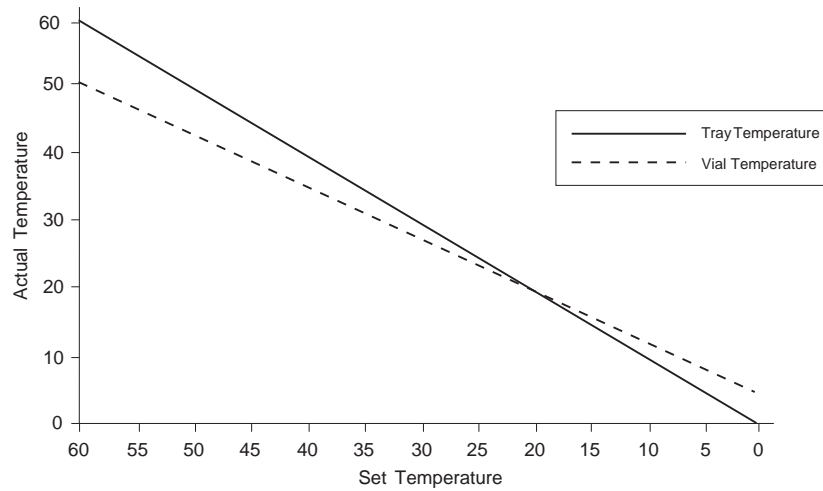
### **Tray Temperature Control**

This option allows you to heat or cool the sample tray within a range of 0 to 60 °C (in 1° increments). This field appears only if you purchased the tray temperature control option and specified Yes in the Configurations Menu Tray Temp field. If you plan to use tray temperature control, use the [+] and [-] keys to turn on the tray heater/cooler in the Tray field. Then specify the temperature in its Temp field. If set to "On" in the Injection Menu, the heater/cooler will not be activated until you load the file manually (by pressing Load from the Files Menu), or automatically when you press [RUN] to start your queue.

*HINT: The autosampler automatically delays the start of the run until the set temperatures have been reached. Once activated, if empty, the tray requires about 30 minutes to reach a temperature of 0 °C and about 10 minutes to reach a temperature of 60 °C. To save time, you may wish to load your file so that the heater/cooler can be coming to temperature while you are completing your run preparation. See page 30 for more information on loading your file. You can also save time by preheating or precooling your samples.*

The autosampler's Tray Temperature Control Option is primarily designed to maintain the temperature of the sample compartment within 2 °C. The actual temperature inside the autosampler vial is dependent upon the specified tray temperature, type of vial, sample buffer, etc. The following graph (Figure 3.3) indicates the approximate vial temperature given a specified tray set temperature.

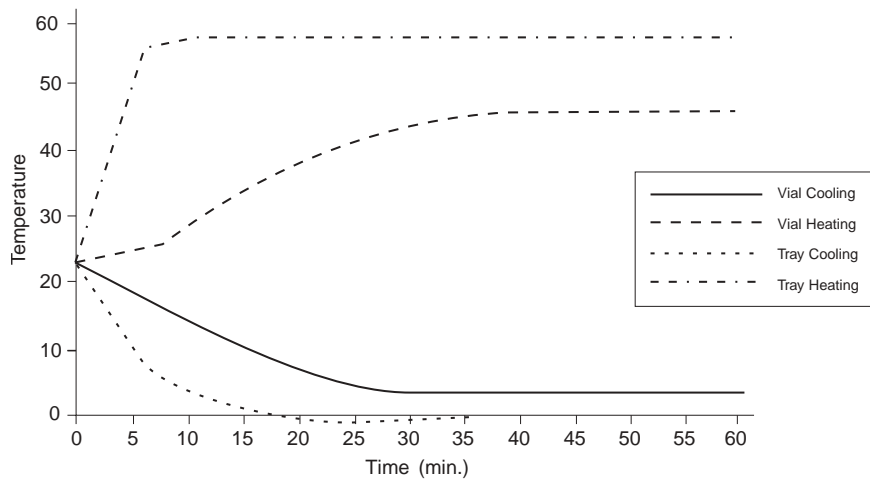
Figure 3.3. Plot showing the actual vial and tray temperature at a specified set temperature.



The Tray Temperature Control Option can also be used to heat and cool samples. The time lag in heating and cooling is due to variations in ambient temperatures, indirect contact with temperature control units, and vial type. The following graph (Figure 3.4) indicates the time required to heat (to 60 °C) or cool (to 0 °C) the tray and vials from an ambient temperature of 23 °C.

*HINT: A 5 °C change in ambient temperature results in approximately a 1 °C change in vial temperature.*

Figure 3.4. Plot of heating and cooling times of a standard glass vial and the autosampler tray. The starting ambient temperature was 23 °C.



*HINT: You can save time by preheating or cooling your samples before placing them into the autosampler.*

## Calibration Menu

By selecting Calibration from the Edit Menu, you can call up the Calibration Menu (Figure 3.5). This section tells you how to enter your calibration parameters and how each is used by other autosampler menus.

Figure 3.5. The Calibration Menu.

Number of Levels	0
Samples/Calibration	0
-----	
Injections/Level	1
Injection Volume	10
Type	Reuse same vials
Bracket samples	No

### ***Number of Levels***

Specify the number of calibration levels (0–9). The autosampler automatically copies this value to the Calib Vial 1 (of X) field in the Samples Menu, where X equals the number of levels. (Refer to page 32 for more information.) A value of zero in the Number of Levels field indicates that no calibration injections will be made for the specified sample set.

### ***Samples/Calibration***

From this field you can specify the calibration interval (the number of sample vials to be injected between each recalibration).

### ***Injections/Level***

In this field you specify the number of injections (1–99) from each calibration level (0–9).

### ***Injection Volume***

The Injection Volume field in the Calibration Menu allows you to specify an injection volume for your calibration vials that is different from the sample injection volume you specified in the Injection Menu.

Similar to the Injection Volume field in the Injection Menu, this value depends upon the injection method you select in the More Menu's Injection Type field. As with the sample injection volume, you can choose between 0.1–10.0  $\mu\text{L}$  and 1–200  $\mu\text{L}$  if you are using PushLoop injection (see page 29).

*NOTE: This field is not displayed if you have selected Full from the More Menu's Injection Type field or if you are using an SS-1.*

Using variable-volume injection, you can choose either Pull- or PushLoop injection from the More Menu. These methods are described starting on page 29. If you use pull-loop injection, you can select a calibration-vial injection volume from 1–200  $\mu\text{L}$ . If you use PushLoop injection, you can specify a calibration vial injection volume from 0.1–10.0  $\mu\text{L}$  or from 1–1500  $\mu\text{L}$ , depending upon which push type you selected in the Injection Type field. For maximum precision, you should limit the injection volume to approximately one-half the sample loop size.

If you're using full-loop injection, you can use the [+] and [–] keys to select one of the following values: 5, 10, 20, 50, 100, 200, 500, and 1000  $\mu\text{L}$ . We recommend that you enter your sample-loop volume as your calibration-vial injection volume.

*NOTE: If you want to determine optimum syringe and sample loop size, see page 169.*

### **Type**

This field allows you to specify one of two types of patterns: 1) Reuse same vials; or 2) Intersperse vials.

Reuse same vials programs the autosampler to use the same calibration vials each time a recalibration is made. Thus, each time a calibration is required, the autosampler goes to the first calibration-vial position specified in the Sample Menu's Calib Vial 1 (of X) field and injects all the calibration vials. It then injects the number of samples indicated by the Calibration Menu's Samples/calibration value.

*NOTE: If you have selected the Reuse vial calibration type, do not place calibration vials between sample vials. Instead, place them before the sample vials in the same tray or in a different location.*

*NOTE: When you reuse your calibration vials, you may place them in any positions in the sample tray(s). However, if you are doing multiple-level calibrations, your calibration vials must be placed sequentially in the tray.*

Intersperse vials programs the autosampler to use each calibration vial only once. You must manually intersperse your calibration vials between your sample vials within the sample set. The autosampler makes injections in tray-placement order. If you've specified a different injection volume for calibration vials versus sample vials, the autosampler uses the Samples/Calibration field value to determine whether the next vial is a calibration or a sample vial. It determines the location of the first calibration vial from the value entered in the Samples Menu's Calib 1 (of X) field. Figure 3.6 illustrates the vial order used by the two different calibration routines.

Figure 3.6. The different injection orders used by the Reuse and Intersperse calibration routines.

Injection Order	Reuse	Intersperse
C	A01	A01
C	A02	A02
S	A03	A03
S	A04	A04
C	A01	A05
C	A02	A06
S	A05	A07
S	A06	A08
Total vials used:	6	8

### ***Bracket Samples***

This field lets you specify whether or not you want to automatically end the sample set with a recalibration. If you select Yes, the autosampler processes the last sample and then repeats the calibration series specified for the sample set. If you select No, the autosampler simply stops and returns home after the last sample is injected.

### **Timed Events Menu**

Accessed from the Edit Menu, the Timed Events (external events) Menu (Figure 3.7) allows you to create a time program to turn on/off external devices (such as columnswitching valves and fraction collectors) at preset times during your run. Pins 9–12 of the autosampler's 12-pin terminal are used for Timed Events. In this section, we'll show you how to create a time program. Refer to Sections 6 and 9 for connecting external devices to your autosampler.

Figure 3.7. The Timed Events Menu.

Pin#	12	11	10	9
Time	TF1	TF2	TF3	TF4
0.00	Lo	Lo	Lo	Lo
-----				
0.50	Hi	Lo	Lo	Lo
2.00	Lo	Hi	Lo	Lo

The Timed Events Menu holds a maximum of nine lines, each of which sets Hi/Lo one or more timed event outputs at the time specified. For example, in Figure 3.7, Time Function 1 (TF1) is set Hi (activated) at 0.5 minutes into the run; at 2 minutes into the run, Time Function 2 (TF2) is set Hi while Time Function 1 is set Lo (deactivated). By changing the Time value in each line, you can add, delete, or insert menu lines.

### ***Adding a Line***

To add a line to the end of your program:

1. Press [v] to move the cursor to the blank line after the last displayed line.
2. Press the [+] key to copy the last existing line with a new time incremented by one minute.
3. Use the arrow keys and the [+] and [-] keys to change the time and set the time functions' states (Hi/Lo).

### ***Deleting a Line***

To remove a line:

1. Move the cursor to the Time field in the line to be deleted.
2. Hold down the [-] key until the value is zero (0.00).
3. Press the [-] key once to erase the line.

If you are deleting a line from the middle of a timed events program, the blank line remains until you move the cursor to another line.

*NOTE: If you delete both of the visible lines on the display, both lines disappear, but the cursor remains. Press [^] or [v] to move to the remaining lines in the program.*

### ***Inserting a Line***

To insert a line between two existing lines:

1. Move the cursor to the blank line at the end of the program.
2. Enter a time that falls between two existing times.
3. Move the cursor off the new line. The autosampler automatically inserts the new line.
4. Then move the cursor to the newly inserted line(s) and set the Timed Events Outputs' states as desired.

Figure 3.8 illustrates this process.

Figure 3.8. (top) The original timed events program; (middle) the original program with an added line — unsorted; (bottom) the timed events program with an added line — sorted.

Time	TF1	TF2	TF3	TF4
0.00	Lo	Lo	Lo	Lo
-----				
1.00	Lo	Lo	Hi	Lo
2.00	Lo	Hi	Lo	Lo

Time	TF1	TF2	TF3	TF4
0.00	Lo	Lo	Lo	Lo
-----				
1.00	Lo	Lo	Hi	Lo
2.00	Lo	Hi	Lo	Lo
<b>0.50</b>	<b>Lo</b>	<b>Hi</b>	<b>Lo</b>	<b>Lo</b>

Time	TF1	TF2	TF3	TF4
0.00	Lo	Lo	Lo	Lo
-----				
<b>0.50</b>	<b>Hi</b>	<b>Lo</b>	<b>Lo</b>	<b>Lo</b>
1.00	Lo	Lo	Hi	Lo
2.00	Lo	Hi	Lo	Lo

*NOTE: The Run-time clock stops and resets at the completion of the Cycle Time. Time lines longer than the Cycle Time value will never be activated.*

## Sample Prep

Once you've entered your external events parameters, the autosampler returns you to the Edit Menu with the cursor in the /Sample Prep/ field (if installed). If you did not purchase the Sample Preparation Option, this field won't appear in the Edit Menu. Instead, the autosampler will return you to the Edit Menu with the cursor in the /More/ field. See Section 4 for detailed information on the Sample Preparation Option.

**More Menu**

The More Menu (Figure 3.9) allows you to enter the remainder of your run parameters, each of which is discussed below.

Figure 3.9. (top) The SS-1 More Menu; (bottom) the SS-2 More Menu.

Equilibration Time	0.0
Gradient Delay	0.00
-----	
Viscosity	Normal
Prep Viscosity	Normal
Flush Volume	400
Needle Height	2.0

Equilibration Time	0.0
Gradient Delay	0.00
-----	
Viscosity	Normal
Prep Viscosity	Normal
Flush Volume	400
Injection Type	Push
Injection Range	.1-10.0
Needle Height	2.0

***Equilibration Time***

In this field, specify the length of time (in minutes) required for the column or LC system to equilibrate. The equilibration time allows time for the column to equilibrate prior to the first injection of a new sample set. It is only in effect before the first injection. The default value of 0.0 is a good starting value for many applications.

***Gradient Delay***

The gradient delay field is used to specify the amount of time required to allow the mobile phase to travel from the pump's proportioning valve to the injection valve. This field is used only if you are running a gradient. To determine the gradient delay value, you'll need to calculate the precolumn volume, which is best done during installation. Refer to Section 9 for a procedure for calculating your precolumn volume and gradient delay value.

***Viscosity***

Specify the sample viscosity (Normal, Medium or Viscous). The viscosity setting affects the sample syringe draw-rate after the sample loop fills. A higher viscosity setting automatically decreases the syringe draw-rate to give a more viscous sample more time to be drawn into and to travel through the sample loop.

*HINT: Use the Normal setting for running most applications.*

### **Prep Viscosity**

Specify the viscosity (Normal, Medium or Viscous) of the sample-preparation sample. The viscosity setting affects the preparation syringe (2.5 mL) draw-rate. A higher viscosity setting automatically decreases the syringe draw-rate and increases the injection delay to give a more viscous sample more time to be drawn into and to travel through the sample lines. This also reduces cavitation caused by low-boiling point solvents.

*NOTE: This field appears only if the Sample Preparation Option is installed.*

*HINT: When performing normal-phase chromatography, the viscous setting maximizes performance.*

### **Flush Volume**

Specify the flush volume to be used for flushing between injections. The acceptable range is 200–5000  $\mu\text{L}$ . The default is 400  $\mu\text{L}$ .

### **Injection Type (SS-2 only)**

With the SS-2, you can choose from four different injection methods: 1) full-loop, 2) pull-loop, 3) PushLoop injection using the high (Push Hi) injection volume range (1–200  $\mu\text{L}$ ), and 4) PushLoop injection using the low (Push Lo) injection volume range (0.1–10  $\mu\text{L}$ ). The advantages of each type are briefly described here. Refer to Section 8 for a description of how each method operates and for the excess sample volume required for injection.

The optimum injection method depends upon the amount of sample you have and the degree of precision desired.

Full: Full- or fixed-loop injection is useful when you want maximum precision and have unlimited sample. The maximum injection volume is 1000  $\mu\text{L}$  and the range from 5–1000  $\mu\text{L}$ .

*NOTE: If you select full-loop injection, the Injection Menu's Injection Volume field changes to Loop Size and the Calibration Menu's Injection Volume field does not appear.*

Pull: Pull-loop injection (traditional variable-volume injection) is useful when sample conservation is important and when large injection volumes (> 100  $\mu\text{L}$ ) are required.

Push: PushLoop injection is a variable-volume method that gives the precision of fixed-loop injection with limited amounts of sample. This method is most effective for smaller injection volumes (< 10  $\mu\text{L}$ ).

**Injection Range**

The Injection Range field displays the sample- and calibration-injection volume range, depending upon the Injection Type that you selected. For full-loop injection, the range is 5–1000  $\mu\text{L}$ . For pull-loop injection, the Injection Range field is automatically filled in with the 1–1500  $\mu\text{L}$  range. For PushLoop injection, the ranges are 1) 0.1–10.0  $\mu\text{L}$  (Push Lo) or 2) 1–200  $\mu\text{L}$  (Push Hi), depending upon your sample-loop size.

**Needle Height**

The Needle Height field enables you to control the distance between the needle tip and the bottom of the vial (0.0–20.0 mm). The default value is 2.0 mm. See Section 9 for specific instructions to optimize needle height.

*NOTE: With this value, you are actually controlling the vial's height relative to the needle tip. The vial moves; the needle is stationary.*

**SAVE YOUR FILE**

Your file is saved automatically. Once you have filled in all of the Edit Menu's parameters, you have finished creating your edit file. Exiting the Edit Menu automatically saves your edit file but does not load it. For more about file management, see page 45.

*NOTE: Pressing [RUN] after creating your file will not start your run. You must first add it to the sample queue. For more information, see "Build Your Sample Queue" below.*

**LOAD YOUR FILE**

The Load function is useful when you want to run a single sample set manually, for example to turn on your oven or tray heater/cooler.

To load your file, press /FILES/, /Load/. The Load Menu appears:

Load File 1:   FILENAME

If you want to load the last file you edited, use the [+] and [-] keys to select its file number and press [ENTER]. If, instead, you want to load a different file, select the correct file number and then press [ENTER]. The message **\*\*File Loaded\*\*** will be displayed. Once a file is loaded, it becomes your run file.

The Load function is used to run a single sample set; however, if you want to run a group of sample sets, this is done using the sample queue, by pressing [RUN]. Once [RUN] is pressed, the autosampler automatically loads each file into the queue before running it. You don't need to manually load each file. For more information on running your samples from the sample queue, see "Build Your Sample Queue" below.

## BUILD YOUR SAMPLE QUEUE

The sample queue is the list of sample sets and associated files to be run. Building your sample queue requires adding each item to the queue from the Samples Menu and then arranging those items in the desired run order from the Queue Menu. If you run a group of files from the sample queue, each file is automatically loaded before it is run. These procedures are described below.

### Adding Items to the Sample Queue

To add an item to the queue, press [SAMPLES] to access the Samples Menu. Then enter each of the values shown in the Samples Menu (Figure 3.10):

*HINT: Since the method downloads most of the values automatically, the only values that require editing are those in the File Number and Vial location fields.*

Figure 3.10. (top) The SS-1 Samples Menu; (bottom) the SS-2 Samples Menu.

Sample Set	1	
File 1:	VITAMINS	
-----		
Loop Size	100	
Injections/Sample	1	
Cycle Time	0.1	
Calib Vial 1 (of 1)	A01	<i>(if calibration is used)</i>
First Sample Vial:	A02	
Number of Samples	1	
Add to Queue?	Yes	

Sample Set	1	
File 1:	VITAMINS	
-----		
Injection Volume	100	
Injections/Sample	1	
Cycle Time	0.1	
Calib Vial 1 (of 1)	A01	<i>(if calibration is used)</i>
First Sample Vial:	A02	
Number of Samples	1	
Add to Queue?	Yes	

### Sample Set

Select the desired sample set you want to add to the queue. Each sample set can be used only one time in the sample queue. You can load a maximum of 39 sets into the sample queue, plus one priority set.

**File**

Specify the number of the file to be applied to the specified sample set. The file's parameters will determine the sequence of Sample Set prompts.

**Injection Volume, Injections/Sample, and Cycle Time**

These values are copied from the Injection Menu of the associated file.

**Calib Vial 1 (of X)**

This value is copied from the Calibration Menu of the associated file. Specify the location of the first vial in the calibration series. "X" is an indicator of the number of calibration levels specified in the file.

*NOTE: The vial position automatically wraps. For example, if the first of three calibration vials is placed in C38, the first sample vial will be A01 for instruments without tray temperature control.*

**First Sample Vial**

Specify the first sample-vial location.

**Number of Samples**

Enter the number of sample vials in the sample set.

**Add to Queue**

Select Yes and press [ENTER] to add the selected sample set to the queue. Each set can be used only one time in the sample queue.

Once you have filled in the Samples Menu for the first sample set, pressing [SAMPLES] again returns you to the top of the menu so that you can add the next sample set.

**Ordering Sets in the Sample Queue**

You add items into the queue from the Samples Menu, which you access by pressing [SAMPLES]. You can reorder them in the Queue Menu (Figure 3.11). You can reorder any sets not currently running.

Figure 3.11. The Queue Menu with no priority-vial sample sets in the queue.

Order	File Name	Set
1	1:TEST	3
-----		
2	2:SAMPLE1	4
3	3:AA1	5

### Changing the Order of Sets Within the Queue

To change the order of items in the queue, move the cursor to the line where the change in order should occur and increment/decrement to the new Order value. Refer to the example in Figure 3.12. To run sample set 5 before sample set 3, move the cursor to the order column of the sample-set 5 line, and decrement the Order value to 1 (Figure 3.12).

Figure 3.12. Changing the order of sets in the queue.

Order	File Name	Set
1	1:TEST	3
2	2:SAMPLE1	4
<b>3-&gt;1</b>	<b>3:AA1</b>	<b>5</b>

The lines rearrange themselves after you exit the field (Figure 3.13).

Figure 3.13. The Queue Menu after the Order has been changed.

Order	File Name	Set
<b>1</b>	<b>3:AA1</b>	<b>5</b>
2	1:TEST	3
3	2:SAMPLE1	4

*NOTE: Remember, Order 1 can only be changed if the sample set is not active. (When you press [Status] the QStatus column should read IDLE.)*

### Deleting Sets from the Queue

To delete an item from the sample queue, move the cursor to the line containing the item to be deleted, decrement the Order number to 0 and then blank. Moving the cursor to another line deletes the blank line from the display.

### Managing a Priority Set Within the Queue

A priority set is inserted into the active set and its samples run before other samples in the active set. It must be assigned to the active file. It appears in the Queue Menu with a P in the Set field (Figure 3.14). You can't assign or change its priority status from the Queue Menu. A priority set will automatically be deleted from the queue upon completion of the active set or when the queue (Qstatus) returns to its IDLE state (if you press [STOP]). For more information on running priority vials, see page 41.

Figure 3.14. A sample queue that contains a priority set.

Order	File Name	Set
1	1 : SAMPLE1	P
-----		
1	1 : SAMPLE1	4
2	3 : AA1	5

## ENTER YOUR OTHER LC PARAMETERS

Prepare your other LC modules for a run and enter the run parameters for your pump, detector and data-output device (recorder, integrator, or data system). Before you start your run:

- Initialize your pump and degas your solvents as required for your application.
- Be sure your detector is stabilized. Don't start your analysis until you've achieved a stable baseline.
- Set up your data-output device.

For more information on connecting your autosampler to other LC modules, see Sections 6 and 9.

## INSTALL SAMPLE TRAYS

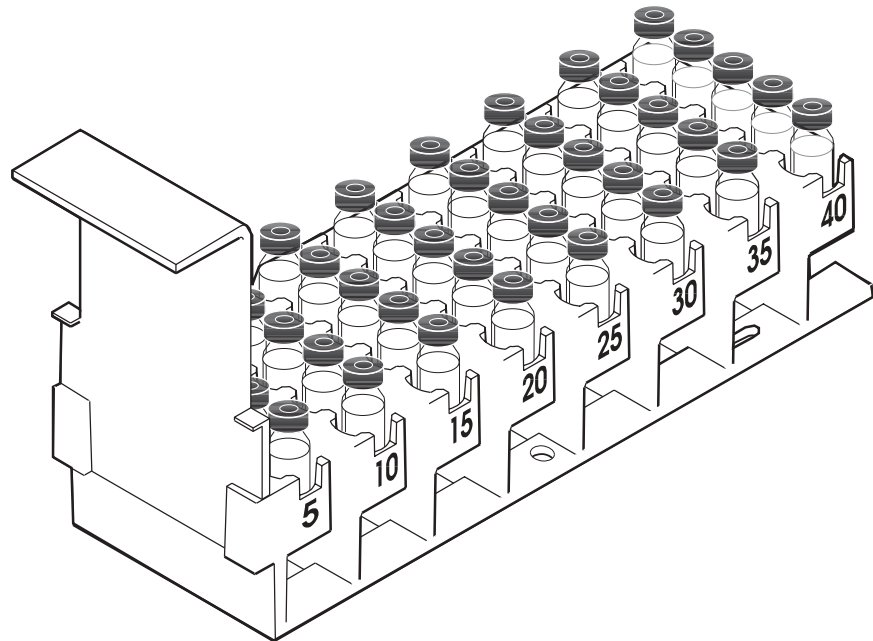
Place your calibration and sample vials into the sample trays included in your Accessory Kit. Trays A, B, and C are arranged in the autosampler from left to right. Each tray contains 40 (35, with Tray Temperature Control) vials arranged in eight (seven) rows of five (Figure 3.15). The first vial position is A01; the last is C40. The position labels are on the left of each position.

*NOTE: If you purchased the Tray Temperature Control Option, each sample tray contains 35 vials.*

Your autosampler comes with a supply of vials, silicone and Teflon septa, and screw-top vial caps. Additional vials may be ordered from BAS. Be sure to use standard size vials to ensure smooth autosampler operation.

*NOTE: Be sure to use either: 1) a 40–60 mil (1.0–1.5 mm) rubber or silicone septum with a 5 mil (0.1 mm) Teflon liner on one side or 2) a single 10 mil (0.25 mm) Teflon liner by itself.*

Figure 3.15. An autosampler tray.



Place the calibration vials and samples into the trays. After your vials are in place, install sample trays A, B, and C as follows:

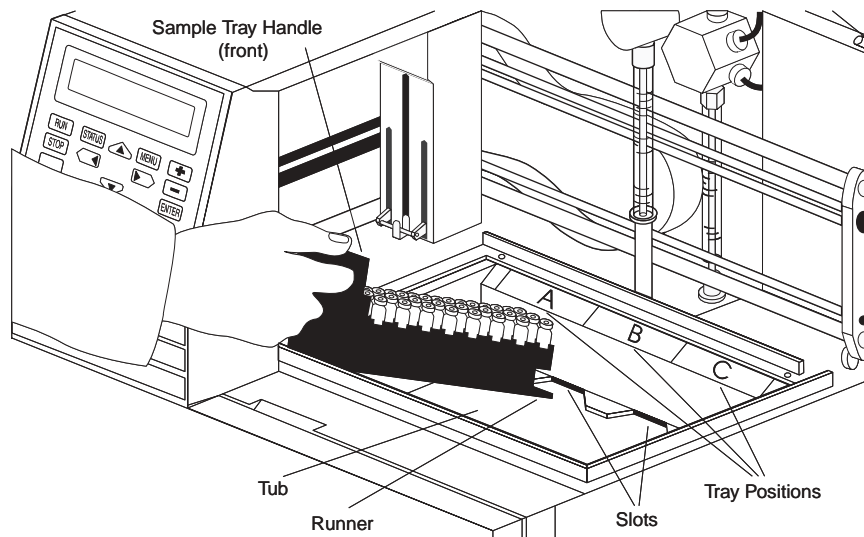
1. Hold the tray handle, tilting the back end down (Figure 3.16).
2. Insert the tray's "runners" into the slots in position A at the rear of the tub.
3. Lower the front of the tray into place and press down firmly to seat the tray.

*HINT: Pull the handle back toward you until the tray snaps into place. Try moving each tray from side to side to be sure that it's seated completely.*

4. Insert the other two sample trays into positions B and C.

**CAUTION!** Be sure that each tray is properly seated. Each tray's upper front edge should fit under the upper lip of the tub. If a tray is not completely seated, the autosampler will not be able to correctly pick up or return vials to that sample tray.

Figure 3.16. Installing a sample vial tray.



### 3.2 Starting Your Run

Once you have created and loaded your file, added your sample set(s) to the sample queue, and installed your sample tray(s), you're ready to start your run.

Press [RUN] to start your run. If there are sample sets in the sample queue, the sample queue's "RUN" Status is displayed on the Status Screen (Figure 3.17).

If the queue is empty, the autosampler prompts you to access the Samples Menu so that you can add sample sets to the queue. After you have added sets, press [RUN] to start your run.

#### MONITORING THE RUN STATUS

When [RUN] is pressed, the autosampler begins processing vials once the oven, heater/mixer and/or tray set temperatures have been reached, the equilibration delay time (entered in the More Menu) has elapsed, the door is closed, and the pump is ready. During the run, the Status Screen is displayed (Figure 3.17).

*NOTE: You can change the "Ready Requirements" from the Options, Ready Participation Menu. See "Ready Participation" in Section 9.*

Figure 3.17. The Status Screen.

QStatus	Set	#Samples	
RUN	1	1 / 1	
-----			
Status	Vial	Vol	#Inj
0.10	A01-S	1000	1 / 2
File 1:			
Oven	TrayTemp		
ON:80	ON:4		

The status screen gives you four kinds of information (described below):

- the autosampler queue status
- injection status
- oven and tray temperatures
- sample preparation status (if the Sample Preparation Option is installed)

*NOTE: The sample preparation status screen is discussed in Section 4.*

You can't edit any information on the Status screen; however, editing during a run can be done from the Samples Menu (below).

## Sample Queue Information

Information on the sample queue is presented in the first two lines of the Status screen.

The specific fields are described here.

### **QStatus**

This field displays the status of the queue (IDLE, PAUSE, RUN, EQUIL, ABORT). These conditions are described here.

IDLE indicates that no sample sets are currently being processed in the queue.

*NOTE: The IDLE status doesn't tell you whether or not there are sets loaded into the queue. To check the contents of the sample queue, press [MENU] and select QUEUE.*

PAUSE indicates that a sample set is loaded, but that processing has been suspended. If the queue is paused (the [STOP] key has been pressed once), you can either resume processing with the current or subsequent vial, or abort processing of the current sample set.

For more information, see "Stopping a Run" on page 43.

RUN indicates that a sample set in the queue is being processed.

EQUIL indicates that the autosampler is waiting for the equilibration delay (temperature zone and initial conditions) to be reached before beginning the processing of the current sample set. This status will be displayed until the oven and tray reach the set temperatures entered in the Edit Menu and/or until the Equilibration Time entered in the More Menu has elapsed.

ABORT indicates that the processing of the current sample set has been stopped (the [STOP] key has been pressed and ABORT has been selected from the Abort/Continue Menu) or that a system error has occurred. Abort is displayed until the autosampler has returned the active vial to the sample tray and flushed all the lines. For more information about Abort, Continue, and Pause, see "Stopping a Run" on page 43.

### **Set**

This field displays the number of the sample set currently being processed (1–39, P). If the queue is not running (the QStatus field reads other than RUN or PAUSE), the set field is blank.

### **#Samples**

This field displays the number of the sample currently being processed and the total number of samples in the set: current sample number/total number of samples. For example, if the autosampler is processing sample 50 in a set of 120 samples, the #Samples field will read 50 / 120.

*NOTE: The total does not indicate the total number of vials or total number of injections in the set -- it indicates the total number of sample vials in the set. Calibration vials are not included on this screen. This way, you'll have a more exact status of the run progress if you've chosen to reuse your calibration vials.*

## **Injection Status**

Pressing [v] displays the next two status screens that contain information on the injection in progress.

*NOTE: The Status Screen is updated every two seconds.*

**Status**

This field displays the progress of the current injection. A time in minutes in the Status field indicates that the queue is running (RUN) and that the current sample has been injected. This field displays the current run time (the time elapsed since the injection). If an injection has not occurred, the autosampler displays one of the following messages in the Status field:

RDY indicates that the autosampler is ready for an injection (all temperature zones and gradient conditions have been reached), but that no injection is currently in progress.

NOTRDY indicates that the autosampler is not ready for an injection. NOTRDY may indicate that the temperature zones are not stabilized, the door is open, or the Pump Ready contact closure is in the wrong state.

GRAD indicates that the autosampler is waiting for the set gradient conditions to be reached (for the gradient delay time set in the More Menu to elapse.)

HOLD indicates that an injection hold is in effect as determined by the Inject Hold input or as set in the Input Polarity Menu.

ABORT indicates that the current injection has been stopped in one of two ways: 1) [STOP] has been pressed to stop the Queue and ABORT selected from the Abort/Continue Menu, or 2) an error condition has occurred. During the ABORT sequence, the autosampler returns the current vial to the sample tray and flushes the lines.

DOOR indicates that the door is open. The autosampler automatically moves the arm to the home position to allow easy access to the sample tray. The arm automatically resumes where it left off when the door is closed.

**Vial**

This field indicates the position and type of vial (S for sample; C for calibration vial) currently being injected. If no injection is in progress (the Status is "NOTRDY"), this field is blank.

**Vol**

This field displays the injection volume for the current vial. If the current vial is a sample vial, this field indicates the Injection Volume entered in the Injection Menu. If the current vial is a calibration vial, this field displays the Injection Volume entered in the Calibration Menu.

**#Inj**

This field displays a fraction that indicates the current injection number over the total number of injections requested for the current vial: current injection number/injection per vial. For example, if the autosampler is performing the third of five injections the #Inj field will read 3 / 5.

**File #**

Pressing [v] displays the number and name of the file assigned to the sample set currently being processed.

**Temperature Status**

Pressing [v] accesses Oven/Tray Temperature Status screen which displays the actual — not the set — temperatures of the oven and tray, if the Column Oven and Tray Temperature Control Options are installed. The temperatures are displayed even if these options are turned off (not set). "OFF" indicates that an option is installed but not turned on in the current file's Injection Menu (Figure 3.18).

---

Figure 3.18. The Temperature Status screen with the oven turned off.

Oven	TrayTemp
OFF : 75	RDY : 18

The temperature field remains blank if one of these options is not present (Figure 3.19). This screen does not appear if neither option is installed.

---

Figure 3.19. The Temperature Status screen without the Tray Temperature Control Option installed.

Oven	TrayTemp
RDY : 75	

*HINT: To check the set temperature for either the oven or the tray (or other parameters), you can return to the Edit Menu and view the information for the appropriate file as long as you have not loaded that file into the Queue. Therefore, you may wish to copy your file before loading it. That way you can edit the copy if you want to check any experimental parameters after your run file is loaded.*

### 3.3 Changing the Run in Progress

Once a run has started, you may wish to change the injection volume, cycle time, calibration interval, or set assignments. You make changes to the run currently in progress (the run file) from the Samples Menu. Figure 3.20 lists the changeable parameters in bold-face type and indicates when those changes take effect. With the exception of the Cycle Time and Calib 1 (of X) values, changes made from the Samples Menu take effect on the next sample injection. Changes to the Cycle Time take effect on the current sample injection; changes to the Calib 1 (of X) field take effect on the next recalibration. Samples Menu changes remain in effect until the next sample set.

Figure 3.20. (top) The SS-1 Samples Menu; (bottom) the SS-2 Samples Menu.

Sample Set	1	
File 1: VITAMINS		
-----		
<b>Injections/Sample</b>	<b>1</b>	<i>Next sample injection</i>
<b>Cycle Time</b>	<b>0.1</b>	<i>Current recalibration</i>
<b>Calib 1 (of 3)</b>	<b>A02</b>	<i>Reuse vials only</i>
First Sample Vial	A05	<i>Cannot change</i>
<b>Number of Samples</b>	<b>1</b>	<i>Next sample injection</i>
Set	2	<i>Next sample set</i>

Sample Set	1	
File 1: VITAMINS		
-----		
<b>Injection Volume</b>	<b>5.0</b>	<i>Next sample injection</i>
<b>Injections/Sample</b>	<b>1</b>	<i>Next sample injection</i>
<b>Cycle Time</b>	<b>0.1</b>	<i>Current recalibration</i>
<b>Calib 1 (of 3)</b>	<b>A02</b>	<i>Reuse vials only</i>
First Sample Vial	A05	<i>Cannot change</i>
<b>Number of Samples</b>	<b>1</b>	<i>Next sample injection</i>
Set	2	<i>Next sample set</i>

*NOTE: Changing one vial location does not affect other vial locations.*

#### **Priority sample sets**

A priority sample set is one which can be inserted into the currently active sample set. It shares the same file and resources as specified for the currently active set. Priority sets can be added at any time.

Priority sample sets are like other sample sets with several important exceptions:

1. You assign their priority set number (P) in the Samples Menu (not in the Queue Menu).
2. Priority sets must be assigned to the currently active file.
3. You can't change the order of priority sample sets once they are loaded into the sample queue.
4. If you want to run priority sample sets, you must select Reuse same vials as your calibration routine type. (If you select Intersperse vials, the autosampler won't "know" whether a subsequent vial is a calibration vial or a sample vial and therefore won't be able to resume the calibration routine after the priority sample set is processed.)
5. Since a priority set is inserted into the currently active set, the file cannot be edited or changed between the currently active set and the priority set.

To insert a priority sample set into the sample queue, use the following steps (Figure 3.21):

1. Press [SAMPLES].
2. Use the [+] and [-] keys to change the Set field value to P.
3. Fill in the other file parameters as you would for any other sample set.
4. Add the priority set to the queue.

Figure 3.21. The Samples Menu with a priority sample set.

Sample Set	P
File 1: VITAMINS	
-----	
Injection Volume	10
Injections/Sample	1
Cycle Time	10
First Sample Vial	C23
Number of Samples	1
Add to Queue?	Yes

*NOTE: The door is equipped with a safety interlock that automatically moves the autosampler's arm to the "home" position to allow you to add samples to the tray during a run. Once the door is closed, the arm returns to its position and resumes the operation in progress before the door was opened. However, you can disable the door interlock if you wish. Press [MENU], /OPTIONS/, Ready Participation, move to Door Interlock (page 165), and*

use the [+] and [-] keys to select DISABLE. For more information on the door interlock function, refer to Section 9.

The autosampler assigns a P (priority) order to the priority set as shown in the Queue Menu (Figure 3.22):

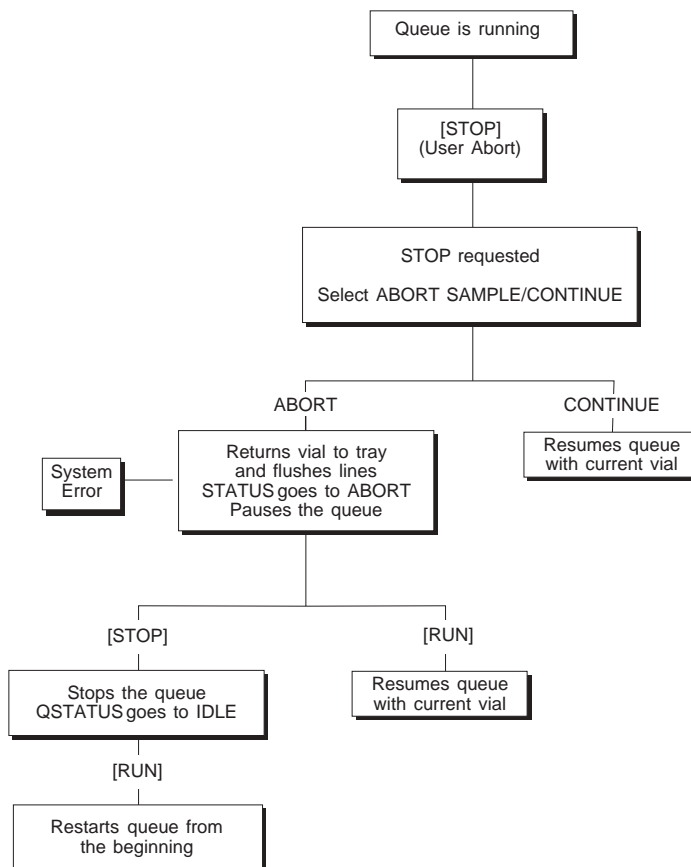
Figure 3.22. The Queue Menu with one priority sample set.

Order	File Name	Set
1	1:VITAMINS	P
-----		
1	1:VITAMINS	1
2	2:BARBITUA	2
3	3:EXPER	1 3

### STOPPING A RUN

The [STOP] key has several functions depending upon the instrument's status when the [STOP] key is pressed. Refer to Figure 3.23 as you read about the STOP/ABORT sequence.

Figure 3.23. The Queue Stop/Abort Sequence.



If the queue is in RUN when you press the [STOP] key, the autosampler suspends its current operation and waits for you to make a selection to ABORT (Figure 3.24a) or CONTINUE (Figure 3.24b):

Figure 3.24. (top) The Abort prompt; (bottom) the Continue prompt.

Stop requested. Select	ABORT Sample
Stop requested. Select	CONTINUE

If you select Continue (by pressing the [+] key), the autosampler returns the queue to the ACTIVE state and resumes the motor activity (as if you had not pressed the [STOP] key).

If you select Abort or if a system error occurs, the autosampler:

- Aborts the processing of the current sample
- Returns the active vial to the vial tray
- Flushes the lines
- Puts the queue in the PAUSE state (Figure 3.25)

Figure 3.25. The Queue Status line with the autosampler in the PAUSE state.

QStatus	Set	#Samples
PAUSE	1	1 / 1

When the queue is PAUSED, you can either:

1. Press [RUN] to resume processing of the current vial, or
2. Press [STOP] to stop the queue (the QStatus goes to IDLE) and then press [RUN] to restart processing from the beginning of the queue.

This [STOP]/Abort sequence allows you several opportunities to stop and resume or restart the run as needed.

The ABORT/CONTINUE Menu and PAUSE states let you remedy problems that could adversely influence the processing or data received from the current or subsequent vials (depleted solvent supply, etc) and resume the run without loss of previous data.

The ABORT, [STOP], [RUN] choice allows you to stop the current run and remedy problems that could have influenced the processing or data received from all prior injections (incorrect file assigned to the set, incorrect solvent concentration, etc.). This second option minimizes loss of resources (and time!).

### 3.4 File Management

You've learned how to edit and load files from the Files Menu. Now you'll learn to copy, delete, protect files from editing, and lock files to prevent changes to the run file during a run.

#### SAVING FILES

As you know, the autosampler automatically saves your file when you exit the Edit Menu. If a power failure occurs while you are editing a file, all current changes will be lost. However, if a power failure occurs after you exit the Edit Menu, your changes will be saved.

#### COPYING FILES

To copy a file, press [MENU], and select /FILES/, /Copy/ to display the Copy Menu (Figure 3.26).

Figure 3.26. The Copy Menu.

```
Copy File 1:  
To File 2:
```

Enter the number of the file to be copied in the Copy File field. Enter the number of the file you wish to copy to in the To field. When you press [ENTER], the message **\*\*File Copied\*\*** appears briefly and you are returned to the Files Menu.

#### DELETING FILES

Deleting a file restores all parameters to their default values. To delete a file, press [MENU], select /FILES/, and /Delete/. The Delete Menu appears (Figure 3.27).

Figure 3.27. The Delete Menu.

```
Delete File 1:
```

Enter the number of the file you wish to delete and press [ENTER]. The message **\*\*File Deleted\*\*** appears briefly and you are returned to the Files Menu.

## PROTECTING FILES

You can protect a file from being edited or deleted by using the file protection feature in the OPTIONS Menu. Because this feature enables you to control the changes to an original run file, you can easily incorporate your autosampler files into your standard good-laboratory-practice (GLP) protocols. To display the Configurations Menu (Figure 3.28), press [MENU], select /OPTIONS/, /Configuration/.

Figure 3.28. The Configurations Menu.

Sample Syringe	250
Prep Installed	Yes
-----	
Oven Installed	No
TrayTemp Installed	Yes
<b>Key Repeat Rate</b>	<b>Medium</b>
File Name	Protect
<b>1: VITAMINS</b>	<b>On</b>
2: BARBITUA	Off
3: EXPER 3	Off
4: STAT	Off
Mode	Stand Alone
Solv Viscosity	Normal

To protect a file, move the cursor down to the Protect field in the line that contains the file to be secured and press the [+]/[-] keys to turn ON the protect function. For example, File 1 VITAMINS in Figure 3.28 cannot be edited until the protect function is turned off. Attempting to edit a protected file displays the following message:

Figure 3.29. The confirmation message to prevent editing of a protected file.

```

*File Protected*
No Editing Allowed
```

## Changes to real-time editing

If a file is protected, you will not be able to change the injection volume, number of injections per vial, or cycle time for the run in progress. These lines will not appear on the Samples Menu (Figure 3.30).

Figure 3.30. The Samples Menu for a protected file.

Sample Set	1
File 1:	VITAMINS
First Sample Vial	A01
Number of Samples	10
Set Already in Queue	

## Section 4. Advanced Operations

This section tells you how to use the optional sample preparation system to automate sample treatment, heating, and mixing. In this section you will learn:

- The terminology used in the sample preparation templates
- How the sample preparation system works
- How to use the built-in templates
- How to create your own customized file
- How to use the Edit and Samples Menus and Status Screen with the Sample Preparation Option installed
- How to run an autosampler file that uses automated sample preparation

Maintenance and troubleshooting procedures for the sample preparation system are included in Section 5, Maintenance, and Section 8, Troubleshooting, respectively.

### BEFORE YOU START

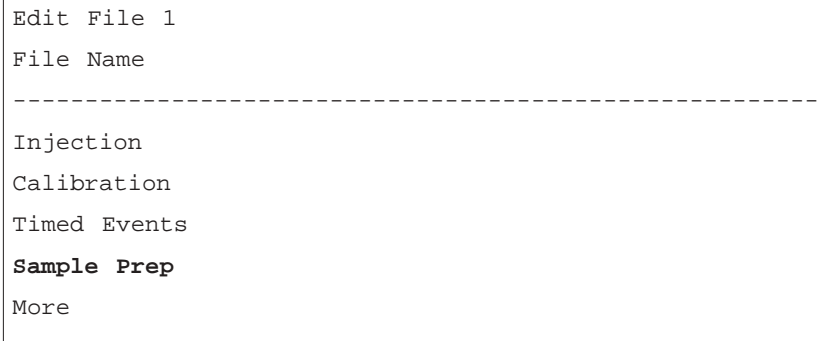
In presenting the material in this section, we assume that you have installed your sample preparation bottles and lines as described in Section 6, that you have read and used the procedures described in Section 3, and that you are comfortable with the routine operations of your autosampler.

#### 4.1 Overview

Preparing to run a sample preparation file is similar to preparing to run any other autosampler file. First, you prepare your chemicals and fill the solvent bottle(s). Then you enter your file information into the Injection, Calibration, Timed Events, and More Menus as always. Next you enter your sample preparation parameters into one of the four built-in sample preparation menus (templates) accessible from the Edit Menu's Sample Prep field (Figure 4.1). Your sample preparation parameters are stored in the same file as your other run parameters. You load your file, enter your sample set information (build your sample queue) and start your run as we discussed in Section 3.

The Overlap Enable field at the end of each template allows you to program the autosampler to begin preparing the next sample vial while the current injection vial is still eluting.

Figure 4.1. The Edit Menu.



## 4.2 Terminology

### TEMPLATES

Each of the four sample preparation menus is a built-in procedure format called a template. Each template contains a different number of steps arranged in a preset and unchangeable order. Each step carries out a specific autosampler operation or group of operations. Some steps transfer liquid between vials or dispense solvents from any of the four external solvents. Others control the operation of the heater/mixer (included in the Sample Preparation Option) or the position of the hook to adjust the distance between the needle tip and the bottom of the vial.

By using the template default values, you can run one of the four built-in procedures:

- 1) Simple dilution
- 2) DABS-CL derivatization
- 3) Rainbow dilution
- 4) Linear dilution

Or, you can enter new values and create your own customized procedures. The autosampler automatically prepares your sample(s) according to the steps specified in the template.

## DILUTION CYCLES

Within each template, the numbered steps are grouped into dilution cycles. For example, steps 1–6 in Figure 4.2 comprise the dilution cycle for Template 1.

Dilution cycles differ across templates in the number of steps and types of operations they perform. You can program the autosampler to repeat dilution cycles up to four times within a template to carry out more complex procedures. We'll see how this works in the section titled "Template Descriptions" on page 57. But first we'll discuss what happens in the simple dilution cycle used in Template 1 because its steps are used in all four templates. Once you understand how these steps control the autosampler's operation, you'll be able to apply the same principles to Templates 2–4.

Figure 4.2. Template 1.

```
Dilution Cycles 1
1- Load 500µL Solv S-1
2- Pickup 100µL Sample
3- Pickup 0µL Sample
4- PickLG 0µL Sample
5- Add 500µL to Sam+1
6- Mix for 0.5 minute
Overlap Enable: No
This Procedure requires
that 1 Extra Vial be
placed immediately after
each Sample Vial.
```

Your autosampler's simple dilution cycle is easily understood by comparing it to a manual dilution procedure. In a manual dilution, you draw a specific amount of sample into a pipet and expel a measured volume into a volumetric flask. Next you dilute to final volume with a desired amount of solvent and mix your sample/solvent mixture for a specific amount of time.

In the manual dilution we've just discussed, solvent and sample are added to the target container one at a time. In contrast, the autosampler first draws all of the resources for a given dilution into its solvent-holding loop and sample/needle lines. An air bubble is drawn before and after each resource to isolate it in the loop. Once all of the resources have been drawn into the lines, the autosampler delivers them to the target vial simultaneously. The vortex mixer then mixes the target vial for the specified amount of time.

All of the templates use this dilution routine. The next section discusses how the individual template steps carry out this general dilution procedure.

### 4.3 How Dilution Cycles Work

Delivery lines transport liquid and air through the sample preparation system as programmed in the template steps. Each step influences the operation of one or more of the six major components in the sample preparation system:

- 1) Prep syringe
- 2) Sample syringe with 6-port flush valve
- 3) Solvent holding loop
- 4) Six-port solvent selection valve with your four solvent bottles
- 5) Heater/vortex mixer
- 6) Needle

Refer to Figure 4.3 and keep these components in mind as you learn the functions of each step in a simple dilution cycle.

#### THE SIMPLE DILUTION CYCLE

Before the dilution cycle begins, the autosampler's lines contain flush solvent. The simple dilution cycle is made of five different steps: Load, Pickup, PickLG, Add, and Mix. Each of these steps is described below.

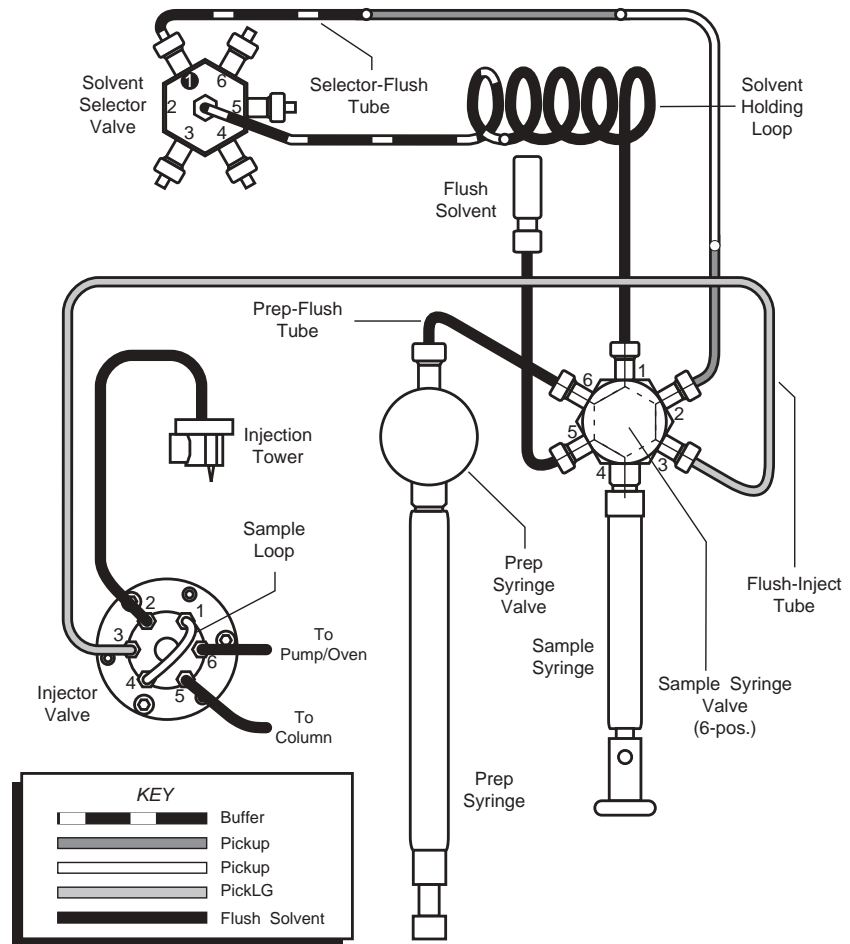
*NOTE: Some of these steps are repeated or skipped in the default methods. We'll see how this works in the sections that describe each template's dilution cycle.*

#### Load

The Load step flushes the lines with the selected solvent in a six-step process:

- 1) The 6-port solvent-selection valve rotates to the air position to draw an air bubble into the solvent holding loop.
- 2) The solvent selection valve then rotates to the selected solvent position (S-1, S-2, S-3, or S-4).
- 3) The preparation (prep) syringe draws the specified amount of solvent (0–1500  $\mu\text{L}$ ) plus a constant excess amount into the solvent holding loop. The excess solvent will be used in step 6 to flush the dead volume from the solvent-selection valve to the needle tip. The air bubble drawn into the solvent-holding loop in step 1 isolates the selected solvent (this step) from the flush solvent already present in the lines.
- 4) The solvent-selection valve moves back to the air position and the prep syringe draws another air bubble.

Figure 4.3. The sample preparation dilution process.



- 5) The solvent-selection valve rotates to the sample/needle line.
- 6) The prep syringe moves forward and expels solvent, thus flushing the dead volume with the selected solvent and expelling the second air bubble, leaving only the selected solvent behind.

## Pickup

The Pickup step causes the sample syringe to withdraw the specified volume of liquid from the source vial. The source can be the vial in the Sample (active) position, the vials in the Reagent A, B, or C locations (anywhere in the trays), or vials in the three positions adjacent to the Sample vial: Sam+1, Sam+2, or Sam+3. For example, if the Sample vial position is A01, Sam+1 would be A02, Sam+2 would be A03, etc. The following operations occur during Pickup:

- 1) The hook moves to the selected vial position and picks up the vial.
- 2) The autosampler's arm brings the vial to the sample tower and pushes the funnel out of the way.
- 3) The sample syringe draws an air bubble to isolate the sample or reagent from the selected solvent already present in the solvent holding loop (from the Load step).
- 4) The vial is raised in the sample tower. Sample is then drawn into the sample/needle line.
- 5) The vial is lowered and the sample syringe draws an air bubble to isolate the sample slug and to prevent sample loss.
- 6) The hook returns the vial to the sample tray.

This process is repeated if a second Pickup step is used.

*NOTE: Each pickup step allows you to pick up a maximum of 100  $\mu\text{L}$ . If you want to pick up more than 100  $\mu\text{L}$ , use either the second pickup step or the PickLg step.*

## PickLG

The PickLG (Pickup Large) is the same as Pickup except that it uses the solvent syringe to transport larger volumes (0–1000  $\mu\text{L}$ ).

## Add

During the Add step two events happen:

- 1) The hook moves to the designated "target" vial position (Sample, Sam+1, Sam+2, or Sam+3) and transports the vial into the sample tower.
- 2) The prep syringe expels the desired volume (0–1000  $\mu\text{L}$ ) of the liquids drawn into the solvent-holding loop and sample/needle lines, during the Load, Pickup, and PickLG steps, into the "target" vial.

*NOTE: Because the Add step uses the prep syringe, the maximum volume that can be added is the sum of the Load volume and the PickLG volume. That is, you can't add (deliver to the target vial) more than you've withdrawn into the large (prep) syringe in the Load and PickLG steps.*

**Mix**

The final step in the simple dilution cycle is the Mix step. During this step the target vial is carried to the mixer and vortex mixed for the specified length of time (0.0 to 9.9 minutes). In templates 1 and 2, the vial is returned to the sample tray after the Mix step. In templates 3 and 4, you have the option of doing an extraction operation before returning the vial to the tray.

After each dilution cycle, the autosampler automatically flushes its lines and syringes with flush solvent. This operation (not shown in the templates) returns the instrument to a "known" state. Any residual sample, reagents, or solvents from a previous cycle are flushed from the lines so that the instrument is ready for the next cycle or injection.

**CAUTION!** If you are using immiscible solvents, be sure to flush the preparation syringe (prep flush) between solvents to minimize contact between the immiscible solvents in the autosampler's lines.

**4.4 Template Programming Rules**

Just as there were rules for creating regular files, there are rules that you need to remember when creating sample preparation procedures.

*NOTE: You may wish to read through these guidelines briefly and then refer back to them as you read the section titled "Template Descriptions."*

1. Extra vials are placed in the three vial positions subsequent to the Sample vial position (Sam+1, Sam+2, Sam+3). For example, if the Sample position is A02, Sam+1 is A03, Sam+2 is A04, etc.
2. It is impossible to add, delete, or change the order of steps within a template, but you can skip any step (except an Add step) by setting its volume parameter to 0  $\mu\text{L}$ .
3. Specify one dilution cycle for each delivery of chemicals to, or for each operation carried out on, the target vial.
4. The desired final volume is specified in the Add step.
5. To perform an operation on, but not make a delivery to, the target vial, set the Add step's volume parameter to 0. This approach is useful, for example, when you want to mix or heat the contents of a vial.
6. The volume specified in the Add step in a given dilution cycle must be less than or equal to the sum of the PickLG and the LOAD volumes.
7. The Add step makes deliveries to the target vial on a last-in-first-out basis.

8. Leave the Overlap Enable field function turned on (Yes) if sample preparation for the next vial to be injected can begin while the current vial is still running.
9. The autosampler injects only from the last vial referenced in the method template's last Add step.

## 4.5 Sample Preparation Menu Differences

Before we describe each template in detail, we'll discuss how to do a sample preparation run.

Performing a sample preparation run is essentially the same as doing a run without sample preparation. You create and load your file, build your sample queue, and start your run just as you did in Section 3. However, with the Sample Preparation Option installed, the Edit and Samples Menus and Status Screens are expanded to allow you to enter and monitor your sample preparation parameters. The "new" lines that are dedicated to sample preparation in each of these menus and screens are discussed below.

### EDIT MENU

With the Sample Preparation Option installed, the Edit Menu has the Sample Prep field that you saw in Figure 4.1.

### SAMPLE PREPARATION MENU

Selecting /Sample Prep/ from the Edit Menu accesses the Sample Prep Menu from which you can use the [+] and [-] keys to select one of the four sample preparation templates (Figure 4.4):

Figure 4.4. The Sample Prep Menu.

Use Template	1
Description	Dilution
<i>(The Templates start here)</i>	

### SAMPLES MENU

The Samples Menu (Figure 4.5) has several additional lines with the Sample Preparation Option installed. These new lines reflect the number of reagent and extra vials used by the method.

#### ***Reagent Vials (Reag-A, B, and C)***

If reagents (A, B, and C) are used in the sample preparation method, a line is added on the Samples Menu for each reagent used by the method to allow you to edit its vial location. If one reagent is used, only the Reag-A appears; if two reagents are used, the Reag-B line appears. Figure 4.5 shows the Samples Menu when all three reagents are used.

*NOTE: Reagent vials can be placed in any vial position in any of the vial trays. They don't have to be in proximity to the sample vial.*

Figure 4.5. The Samples Menu with reagents, calibration, and extra vials used in the sample preparation method.

Sample Set	1
File 1:	
Injection Volume	10
Injections/Sample	1 <i>(If injection is used)</i>
Cycle Time	0.1
<b>Reag-A</b>	<b>C23</b> <i>(If reagent vials are used)</i>
<b>Reag-B</b>	<b>C24</b>
<b>Reag-C</b>	<b>C25</b>
<b>Calib Vial 1 (of X)</b>	<b>A01</b> <i>(If calibration is used)</i>
First Sample Vial	A04
Number of Samples	1
<b>File requires n extra vials after each sample</b>	
Add to queue?	Yes

#### ***File requires n extra vials after each sample***

If extra target vials are used after each sample vial, the two lines indicating the number of extra vials required are copied from the sample preparation template into the last two lines of the Samples Menu.

The "n" represents the number of extra (empty) vials placed after the Sample vial (the Sam+1, Sam+2, and Sam+3 vials). An extra vial (one in which sample is mixed with solvent, where a reaction takes place), or a pickup (PickLG) is made. "n" is selected in the Add steps of each template. Figure 4.5 illustrates the Samples Menu for a file that uses calibration with the Sample Preparation Option installed.

*NOTE: The screen actually displays a number (1, 2, or 3) in the "n" field. We've represented this value as "n" because it changes depending on whether you've chosen Sam+1, Sam+2, or Sam+3 during your run.*

#### **Changes to real-time editing**

As described in Section 3, you can make changes to the run file for the run in progress from the Samples Menu. Table 4.1 summarizes when Samples Menu changes to the run in progress take effect.

Table 4.1. When changes affect the run in progress.

File #	Can't edit
Injection Volume	Next injection*
Injections/Sample	Next injection
Cycle Time	Current injection
Reagent (A,B,C)	Next sample
Reagent A–C	Next sample
Calib Vial 1 (of x)	Next recalibration
First Sample Vial	Can't edit
Number of Samples	Next injection
*Changes to this field affect sample vials only, not calibration vials.	

### Protecting files from real-time editing

As described in Section 3, a file can be protected from real-time editing by turning on the Protect function in the Configurations Menu. File Protect prevents you from editing the injection volume, injections per sample and cycle time and eliminates some lines from the Samples Menu for a protected file that uses sample preparation just as it did for a regular file. Figure 4.6 shows the Samples Menu for a protected file that uses sample preparation and calibration.

Figure 4.6. The Samples Menu for a Protected File that uses sample preparation and calibration.

Sample Set	1
File 1: VITAMINS	
Calib Vial 1 (of 3)	A01
First Sample Vial	A04
Number of Samples	10
Already in Queue	

### STATUS SCREEN

With the Sample Preparation Option installed, an additional Status Screen is included to give you information on the current template step, the heater/mixer status and the resource locations for the active file. Figure 4.7 shows the Status Screen for a file that contains a sample preparation procedure and that uses several reagents and calibration vials.

Figure 4.7. The Status Screen for a file with sample preparation.

QStatus		Set	#Sample
RUN		1	5 / 120
-----			
Status	Vial	Vol	#Inj
NRDY	A01	1000	1 / 2
File 1: VITAMINS			
Oven		TrayTemp	
RDY:80		RDY:4	
<b>Prep</b>	<b>Vial</b>	<b>Htr/Mix</b>	<i>(only with sample preparation)</i>
<b>PickLg</b>	<b>A01</b>	<b>RDY:35</b>	

**Prep**

This line tells you which template step is in progress (Load, PickUp, PickLG, Add, Mix, Wait, or Remove). For example, Figure 4.7 indicates that even though the sample queue is running (RUN), since a Mix step is in progress, the autosampler is not ready (NRDY) to inject.

**Vial**

This field indicates the position of the vial currently undergoing sample preparation.

**Htr/Mix**

This field indicates the status (OFF, RDY, and NRDY) of the heater/mixer and its actual temperature, if turned on and set in the Heater field in templates 2 and 4. If not in use, this field reads "OFF" followed by the actual temperature.

*NOTE: The temperature is always displayed whether or not the heater/mixer is in use.*

## 4.6 Template Descriptions

You are now familiar with the basic template steps and the additional menu selections you have to make when using automated sample preparation. In this next section we'll describe the specifics of each template.

All four templates are based on the liquid transport and delivery process described in the section titled "The Simple Dilution Cycle" above. In fact, Template 1 (simple dilution) contains only the steps described above (Figure 4.2). If your protocol has more than two dilutions, simply specify the number of dilution cycles you need in the line before step 1. The autosampler will automatically replicate the template's dilution cycle a total of four times.

Each template will be discussed in two ways: with a boxed figure showing how the display looks, and with a line-by-line description of each step. You can use the line-by-line description to assist you in creating a customized method file for your own applications. Each example uses the template's default values.

**TEMPLATE 1: DILUTION**

Template 1 (Figure 4.8) is a dilution program that you can use to perform a 1:5 v/v dilution.

Figure 4.8. Template 1: Dilution.

```

Use Template 1
Description Dilution
# of Dilution Cycles 1
1- Load 500µL Solv S-1
2- Pickup 100µL Sample
3- Pickup 0µL Sample
4- PickLG 0µL Sample
5- Add 500µL to Sam+1
6- Mix for 0.5 minute
Overlap Enable:      Yes
This procedure requires
that 1 Extra Vial(s) be
placed immediately
after each Sample Vial.

```

**Line-by-Line Description of Template 1**

Use Template 1	Template 1 is selected.
Description Dilution	Template 1 is the Dilution Template.
# of Dilution Cycles 1	One dilution cycle requested for delivery to Sample vial.
1- Load 500 µL Solv S-1	The prep syringe draws 500 µL Solvent 1 (buffer) into solvent holding loop.
2- Pickup 100 µL Sample	The sample syringe draws 100 µL sample into the sample lines.
3- Pickup 0 µL Sample	Skip this step; the sample syringe is not needed.
4- PickLG 0 µL Sample	Skip this step; the prep syringe not needed.
5- Add 500 µL to Sam+1	Dilute 100 µL sample to 500 µL and deliver to Sam+1.
6- Mix for 0.5 minute	Mix sample and buffer for 0.5 minute.
Overlap Enable Yes	Turn on the Overlap Enable function to begin preparation after the last injection and flush of the previous vial (sample or calibration).

This procedure requires that 1 Extra Vial(s) be placed immediately after each Sample Vial.

One Extra (empty or extra) vial is required since mixing takes place in the Sam+1 vial position.

### HOW THE SIMPLE DILUTION STEPS WORK TOGETHER

Before continuing to the other three templates, it is important to understand how the Load, Pickup, PickLg, and Add steps affect the contents of the Add step target vial. Remember that the Add step makes deliveries to the target vial on a last-in/first-out basis. Consider the following examples to see how this works.

Figure 4.9. Template 1: Dilution Cycle with no Pickup or PickLg.

```
Use Template 1
Description Dilution
# of Dilution Cycles 1
1- Load 1000µL Solv S-1
2- Pickup 0µL Sample
3- Pickup 0µL Sample
4- PickLG 0µL Sample
5- Add 1000µL to Sam+1
6- Mix for 0.5 minute
Overlap Enable:      Yes
This procedure requires
that 1 Extra Vial(s) be
placed immediately
after each Sample Vial.
```

In Figure 4.9, 1000 µL of solvent is loaded into the holding loop and delivered into the target vial since no other reagents are drawn into the holding loop in the Pickup or PickLG steps.

In contrast, in Figure 4.10, 1000 µL of solvent and 100 µL sample are loaded into the holding loop. The Add step delivers the last 1000 µL of liquid drawn: 100 µL sample from the first Pickup step but only 900 µL solvent from the Load step. The Add step designates the desired final target-vial volume.

Figure 4.10. Template 1: Dilution Cycle showing a 1:10 dilution.

```
Use Template 1
Description Dilution
# of Dilution Cycles 1
1- Load 1000µL Solv S-1
2- Pickup 100µL ReagA
3- Pickup 0µL Sample
4- PickLG 0µL Sample
5- Add 1000µL to Sam+1
6- Mix for 0.5 minute
Overlap Enable:      Yes
This procedure requires
that 1 Extra Vial(s) be
placed immediately
after each Sample Vial.
```

**TEMPLATE 2: DABS-CL**

Template 2 (Figure 4.11) is a program to perform various chemical reactions. The default method is specifically designed to perform a (Dimethylamino)azobenzenesulfonyl chloride (DABS-Cl) derivatization procedure. This template contains the same steps as Template 1, plus three new steps described here.

**Heater Temp**

This line allows you to turn the heater on and off and set the temperature from 30–100 °C.

**Heat**

In this field you can specify a heating time from 0.0–99.9 minutes.

**Mix**

This field allows you to remix the reagents for a specified amount of time (0.0–9.9 minutes) after heating. Figure 4.11 shows the first Template 2 dilution cycle.

Figure 4.11. The first dilution cycle of Template 2.

```

Use Template 2
Description DABS-CL
# of Dilution Cycles 2
Heater: On           Temp: 65 C
1- Load 200µL Solv S-1
2- Pickup 100µL Reag-A
3- Pickup 40µL Sample
4- PickLG 0µL Sample
5- Add 140µL to Sam+1
6- Mix for 0.3 minute
7- Heat for 4.5 minute
8- Mix for 0.3 minute
Overlap Enable:      Yes

```

The default method's values are those used to perform a simple DABS-Cl derivatization of amino acids.

### Line-by-Line Description of Template 2

Use Template 2	Template 2 is selected.
Description DABS-CL	Template 2 is DABS-Cl derivatization.
# of Dilution Cycles 2	Two dilution cycles is default value: one dilution cycle per delivery to Extra vial (Sam +1).
Heater: On Temp: 65	Turn on heater; set the temperature.
1- Load 200 µL Solv S-1	Draw buffer into the solvent-holding loop.
2- Pickup 100 µL Reag-A	Sample syringe draws Reag-A into sample/needle line (e.g., DABS-Cl).
3- Pickup 40 µL Sample	Sample syringe draws 40 µL Sample into sample/needle line (e.g., Amino Acids).
4- PickLG 0 µL Sample	Skip step.
5- Add 140 µL to Sam+1	Deliver Reag-A and Sample into first extra vial (e.g.DABS-Cl).
6- Mix for 0.3 minute	Mix contents of the first Extra vial for 0.3 minutes to bring vial to temperature.
7- Heat for 4.5 minute	Heat vial for 4.5 minutes.
8- Mix for 0.3 minute	Mix reaction vial for 0.3 minute.
Repeat Dilution Steps	

9- Load 1000 $\mu$ L Solv S-1	Draw buffer into the solvent-holding loop.
10- Pickup 0 $\mu$ L Sample	Skip this step.
11- Pickup 0 $\mu$ L Sample	Skip this step.
12- PickLG 0 $\mu$ L Sample	Skip this step.
13- Add 860 $\mu$ L to Sam+1	Deliver 860 $\mu$ L Solv S-1 into reaction vial (e.g. acetate buffer).
14- Mix for 0.3 minute	Mix reaction vial for 0.3 minutes.
15- Heat for 0.0 minute	Skip this step.
16- Mix for 0.0 minute	Skip this step.
Overlap Enable Yes	Turn on Overlap Enable function.

This procedure requires that 1 Extra Vial(s) be placed immediately after each Sample Vial.

The protocol requires one reaction vial for vortex mixing Reag-A, sample, and Solv S-1.

### TEMPLATE 3: RAINBOW

Template 3 (Rainbow) allows you to do a more complex dilution process or an extraction process and is the same as Template 1 (dilution) with three additional steps. Wait and Remove steps are added after the Mix step at the end of each dilution cycle. The FlushP step has been added after the last dilution cycle before the Overlap Enable step. These new steps are described here.

*NOTE: The default values in Template 3 are designed to illustrate a dilution procedure that creates a calibration series with two components (Reagents A and B) and four calibration levels. For this illustration, the resource locations are as follows: ReagA contains your first standard, ReagB contains your second standard, and Sample, Sam+1–3 are empty.*

#### Wait

The Wait step programs a reaction time for the vial contents or to allow sufficient time for the liquid layer in an extraction to separate. Wait pauses the autosampler for the specified amount of time (0.0–99.9 minutes).

#### Remove

The Remove step positions the bottom of the vial at the desired distance (0.0–20.0 mm) from the needle tip, withdraws a desired volume of solvent (0–1500  $\mu$ L), and delivers it to waste.

## FlushP

As you know, the autosampler automatically flushes the lines and syringes with flush solvent at the end of each dilution cycle. (This step is not shown on your templates.) The FlushP step allows you to fill the autosampler's lines with any of the four solvents (or buffers) in preparation for the sample injection. You can select any of the four external solvents (S-1, S-2, S-3, S-4), or you can flush the lines with flush solvent by selecting Solv Btl. After flushing the lines, FlushP returns the prep syringe to its initial position in preparation for the next injection. Figure 4.12 illustrates the last dilution cycle for Template 3 in order to include the FlushP step.

Figure 4.12. The last dilution cycle of Template 3.

```
Repeat Dilution Steps
25- Load 1000µL Solv S-1
26- Pickup 0µL Sample
27- Pickup 30µL Reagent A
28- PickLG 0µL Sample
29- Add 1000µL to Sam+3
30- Mix for 0.5 minute
31- Wait for 0.0 minute
32- Remove 0µL to
waste at 2.0 mm height
FlushP 0µL Solv S-1
Overlap Enable:      Yes
This procedure requires
that 3 Extra Vial(s) be
placed immediately
after each Sample Vial.
```

## Line-by-Line Description of Template 3

Use Template 3	Template 3 selected.
Description Rainbow	Template 3 is Rainbow Dilution example.
# of Dilution Cycles 4	Specify a dilution cycle for the reagent(s)/solvent delivery to each of the four calibration vials.
1- Load 1000 µL Solv S-1	Draw solvent into the solvent holding loop.
2- Pickup 30 µL Reag-B	Sample syringe draws Reagent B into the sample/needle line.
3- Pickup 0 µL Reag-A	Skip step (no Reagent A is used in the first calibration vial).
4- PickLG 0 µL Sample	Skip this step.

5- Add 1000 $\mu\text{L}$ to Sample	Dilute 30 $\mu\text{L}$ Reagent B to 1000 $\mu\text{L}$ with solvent S-1 and deliver to sample vial.
6- Mix for 0.5 minute	Mix contents of sample vial for 0.5 minutes (first calibration level).
7- Wait for 0.0 minute	Skip this step.
8- Remove 0 $\mu\text{L}$ to waste at 2.0mm height	Skip this step.
Repeat Dilution Steps	
9- Load 1000 $\mu\text{L}$ S-1	Flush lines and solvent loop with the desired volume of solvent (S-1).
10- Pickup 20 $\mu\text{L}$ Reag-B	Sample syringe withdraw 20 $\mu\text{L}$ Reagent B into sample/needle line.
11- Pickup 10 $\mu\text{L}$ Reag-A	Sample syringe withdraw 10 $\mu\text{L}$ Reagent A into sample/needle line.
12- PickLG 0 $\mu\text{L}$ Sample	Skip this step.
13- Add 1000 $\mu\text{L}$ to Sam+1	Dilute 20 $\mu\text{L}$ Reagent B and 10 $\mu\text{L}$ Reagent A to 1 mL with solvent S-1 and deliver to first Extra vial (second calibration level).
14- Mix for 0.5 minute	Mix contents of first extra for 0.5 minutes.
15- Wait for 0.0 minute	Skip this step.
16- Remove 0 $\mu\text{L}$ to waste at 2.0mm height	Skip this step.
17- Load 1000 $\mu\text{L}$ Solv S-1	Draw solvent into the solvent holding loop.
18- Pickup 10 $\mu\text{L}$ Reag-B	Sample syringe draw 10 $\mu\text{L}$ Reagent B into the sample/needle line.
19- Pickup 20 $\mu\text{L}$ Reag-A	Sample syringe draw 20 $\mu\text{L}$ Reagent A into the sample/needle line.
20- PickLG 0 $\mu\text{L}$ Sample	Skip this step.
21- Add 1000 $\mu\text{L}$ to Sam+2	Dilute 10 $\mu\text{L}$ Reagent B and 20 $\mu\text{L}$ Reagent A to 1 mL with solvent S-1 and deliver to second Extra vial (third calibration level).
22- Mix for 0.5 minute	Mix contents of second Extra for 0.5 minutes.
23- Wait for 0.0 minute	Skip this step.
24- Remove 0 $\mu\text{L}$ to waste at 2.0mm height	Skip this step.
Repeat Dilution Steps	
25- Load 1000 $\mu\text{L}$ S-1	Flush lines and solvent loop with the desired volume of solvent (S-1).

---

26- Pickup 0 $\mu$ L Reag-B	Skip this step (no Reagent B needed in the last calibration level).
27- Pickup 30 $\mu$ L Reag-A	Sample syringe withdraw 30 $\mu$ L Reagent A into sample/needle line.
28- PickLG 0 $\mu$ L Sample	Skip this step.
29- Add 1000 $\mu$ L to Sam+3	Dilute 30 $\mu$ L Reagent A to 1 mL with solvent S-1 and deliver to third Extra vial (fourth calibration level).
30- Mix for 0.5 minute	Mix contents of third extra for 0.5 minutes.
31- Wait for 0.0 minute	Skip this step.
32- Remove 0 $\mu$ L to waste at 2.0mm height	Skip this step.
FlushP 0 $\mu$ L Solv S-1	Skip this step.
Overlap Enable No	Turn off the Overlap Enable function.
This procedure requires that 3 Extra Vial(s) be placed immediately after each Sample Vial	One extra vial is required for each standard level. The sample vial contains the first level; levels 2–4 are in positions Sam+1, 2, and 3, respectively.

**TEMPLATE 4: LINEAR**

Template 4 is also named for its default method; it allows you to do linear dilutions. This template (Figure 4.13) contains the heating and mixing steps added to Template 2 and the Wait, Remove and FlushP steps used in Template 3. Figure 4.13 shows the last dilution cycle of Template 4.

Figure 4.13. The last dilution cycle of Template 4: Linear.

```
Use Template 4
Description Linear
# of Dilution Cycles 3
Heater: On Temp: 30 C
21- Load 1000µL Solv S-1
22- Pickup 5µL Sample
23- Pickup 0µL Sample
24- PickLG 0µL Sample
25- Add 1000µL to Sam+1
26- Mix for 0.3 minute
27- Heat for 0.0 minute
28- Mix for 0.0 minute
29- Wait for 0.0 minute
30- Remove 0µL to
waste at 2.0 mm height
FlushP 0µL Solv S-1
Overlap Enable: No
This procedure requires
that 3 Extra Vial(s) be
placed immediately
after each Sample Vial.
```

This template can be used to perform any function or application performed by any of the other templates. The default values can be used to perform a linear dilution procedure, for example, to prepare calibration vials. If you use the default values, your diluent is Solvent S-1, Sam+1 will hold your Level 1 standard, Sam+2 will hold your Level 2 standard, and Sam+3 will hold your Level 3 standard.

---

**Line-by-Line Description of Template 4**

Use Template 4	Template 4 is selected.
Description Linear	Template 4 does linear dilution.
Dilution Cycles 3	One cycle per delivery to a target vial.
Heater: On Temp: 30 C	Turn on heat; set temperature.
1- Load 1000 $\mu$ L Solv S-1	Load 1000 $\mu$ L buffer into solvent holding loop.
2- Pickup 5 $\mu$ L Sample	Sample syringe draw sample (for the Level 1 standard) into the sample/needle line.
3- Pickup 0 $\mu$ L Sample	Skip this step.
4- PickLG 0 $\mu$ L Sample	Skip step.
5- Add 1000 $\mu$ L to Sam+1	Deliver buffer and sample to the first Extra vial (Level 1 standard).
6- Mix for 0.3 minute	Mix the first Extra vial for 0.3 minutes.
7- Heat for 0.0 minute	Skip step.
8- Mix for 0.0 minute	Skip step.
9- Wait for 0.0 minute	Skip step.
10- Remove 0 mL to waste at 2.0mm height	Skip step.
Repeat Dilution Steps	Begin dilution cycle 2.
11- Load 1000 $\mu$ L Solv S-1	Draw 1000 $\mu$ L buffer into the solvent holding loop.
12- Pickup 50 $\mu$ L Sample	Sample syringe draw sample (for the Level 2 standard) into sample/needle line.
13- Pickup 0 $\mu$ L Sample	Skip step.
14- PickLG 0 $\mu$ L Sample	Skip step.
15- Add 1000 $\mu$ L to Sam+2	Add buffer and sample to second Extra vial (Level 2 calibration vial).
16- Mix for 0.3 minute	Mix the Level 2 calibration vial.
17- Heat for 0.0 minute	Skip step.
18- Mix for 0.0 minute	Skip step.
19- Wait for 0.0 minute	Skip step.

20- Remove 0 $\mu\text{L}$ to waste at 2.0mm height	Skip step.
Repeat Dilution Steps	Begin dilution cycle 3.
21- Load 1000 $\mu\text{L}$ Solv S-1	Load 1000 $\mu\text{L}$ buffer into the solvent holding loop.
22- Pickup 100 $\mu\text{L}$ Sample	Sample syringe draws 100 $\mu\text{L}$ sample into the sample/needle line.
23- Pickup 0 $\mu\text{L}$ Sample	Skip step.
24- PickLG 0 $\mu\text{L}$ Sample	Skip step.
25- Add 1000 $\mu\text{L}$ to Sam+3	Deliver sample and buffer for the Level 3 calibration vial into the third Extra vial.
26- Mix for 0.3 minute	Mix Level 3 calibration vial for 0.3 minute.
27- Heat for 0.0 minute	Skip step.
28- Mix for 0.0 minute	Skip step.
29- Wait for 0.0 minute	Skip step.
30- Remove 0 $\mu\text{L}$ to waste at 2.0mm height	Skip step.
FlushP 0 $\mu\text{L}$ Solv S-1	Flush lines with selected solvent.
Overlap Enable No	Do not begin sample preparation on next set until current set has completed injection.
This procedure requires that 3 Extra Vial(s) be placed immediately after each Sample Vial	You need an extra vial for <u>each</u> calibration level.

#### 4.7 Starting a Sample Preparation Run

Once you have filled in all of the parameters in one of the four sample preparation templates, you're ready to enter the rest of your parameters into the Edit Menu. Before you start your run, you need to:

- Enter your Timed Events Menu parameters
- Enter your More Menu parameters
- Load your file
- Press [RUN]

## 4.8 Preparing And Running Your Samples Automatically

### A SIMPLE DILUTION RUN

If you are doing a simple dilution, you can put your concentrated sample, diluent, reagent (if used) and extra vial in one sample set. The autosampler will automatically inject the last vial used during sample preparation, in this case, the contents of your extra vial. Figure 4.14 illustrates this example. For this example, vial A01 is a reagent (ReagA), vial A02 contains the sample concentrate, and vial A03 is the extra vial.

Figure 4.14. An example of a simple dilution run using sample preparation and injection in the same sample set.

Sample Set	1
File 1: EXAMPLE	
Injection Volume	10
Injections/Sample	1
Cycle Time	0.1
Reag-A	A01
First Sample Vial	A02
Number of Samples	
<b>File requires 1 extra vial(s) after each sample</b>	
Add to Queue?	Yes

*NOTE: In Figure 4.14, the vial in the First Sample Vial position (A01) is your first preparation vial, not your first injection vial.*

## PREPARING CALIBRATION VIALS AND INJECTING THEM

If you want to prepare several calibration vials and then inject them, it's easiest to set up two sample sets. The first set will be your sample preparation set without injections; the second set will be for your actual run with injections.

For the first set, you set Injections/Sample to 0 in the Edit and Samples Menus. For the second set, you set Injections/Sample to the desired number of sample injections in both menus. Figure 4.15 illustrates the Samples Menu for the preparation set; Figure 4.16 illustrates the Samples Menu for the injection set.

Figure 4.15. The Samples Menu for the Prep Set.

Sample Set	1
File 1: PREP	
Injection Volume	10
Injections/Sample	0
Cycle Time	10.0
First Sample Vial	A01
Number of Samples	10
Add to Queue?	Yes

*NOTE: You can decrement the Injections/Sample field to 0 only if you have first selected a sample preparation template.*

Figure 4.16. The Samples Menu for the Injection Set.

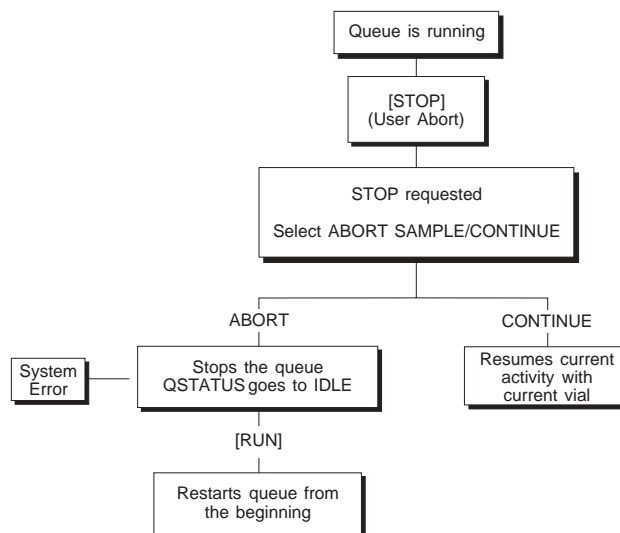
Sample Set	2
File 1: INJECT	
Injection Volume	10
Injections/Sample	5
Cycle Time	10.0
First Sample Vial	A01
Number of Samples	10
Add to Queue?	Yes

*NOTE: If you enter your sample preparation and other run parameters, load your file and then start your run, the autosampler will prepare your samples as you would expect, but inject only the vial used by the last Add step in the sample preparation template.*

## 4.9 Stopping A Run

Similar to operation without sample preparation, the [STOP] key has several functions depending upon the instrument's status when the [STOP] key is pressed. However, the [STOP] sequence discussed in Section 3 is slightly different when sample preparation is used. Refer to Figure 4.17 as you read about the STOP/ABORT sequence for sample preparation.

Figure 4.17. The Queue Stop/Abort Sequence.



If the queue is in RUN when you press the [STOP] key, the autosampler suspends its current operation and waits for you to make a selection from the ABORT/Continue Menu as described in Section 3.

If you press [+] to select Continue, the autosampler returns the queue to the ACTIVE state and resumes the motor activity (as if you had not pressed the [STOP] key), just as it did without sample preparation.

If you select ABORT or if a system error occurs, the autosampler:

- Aborts the processing of the current sample
- Returns the active vial to the vial tray
- Flushes the lines (both syringes)
- Puts the queue in the IDLE state (not PAUSE)

You can then press [RUN] to restart processing from the beginning of the queue.

This [STOP] sequence allows you to resume the run from the ABORT/CONTINUE menu. But if you press [STOP] and select ABORT, you can only restart (not resume) the queue from the beginning.

## 4.10 Customizing the Templates

### GUIDELINES

In our examples so far, the contents of the vials in the template positions have matched the autosampler's names for those positions. For example, we have placed the sample in the Sample position and stock standards in the Reagent A, B, and C positions. We have also used the vial positions in a sequential order. However, the autosampler does not limit you to this approach. In customizing the templates to your own applications, the following more general rules may help:

1. In each dilution cycle, you have seven vial positions to work with: three source-vial positions (Reagents A, B, C) and four vial positions that may be either source or target vials (Sample, Sample + 1, Sample + 2, and Sample + 3).
2. You can arrange your Pickups and Add vials in any order you wish within a dilution cycle as long as you remember that the autosampler locates the extra vials (Sample + 1–3) in relationship to the vial you've assigned to the Sample position. For example, you could Pickup from the Sam + 3 vial and Add to the Sample vial. (But, you couldn't Add to the ReagentA vial since, according to rule 1, it cannot be a target vial.)
3. You can use the vial positions in any order you wish within a dilution cycle. For example, even though the autosampler cannot skip Sample vial positions, you can program it to prepare interspersed calibration vials by Adding to the Sam + 2 position, without using the Sam + 1 position in any of the steps in a given dilution cycle.

*HINT: In loading your tray, if you want your first calibration vial to be A01, then you can use rule 3 and the autosampler's "wrap" feature by assigning the First Sample Vial as C39 (Sam + 2 will then be A01). However, you will still need to create two sample sets: one to prepare and the other to run your calibration and sample vials.*

You'll discover other shortcuts as you integrate your autosampler into your laboratory procedures.

### BLANK FORMS

After completing this section, you are now ready to begin developing your own sample preparation procedures. To assist you in documenting your sample preparation protocols, we have included a blank form (without the default values) for each of the four templates. We have provided four dilution cycles (the maximum available) for each template. We hope these forms will be helpful to you as you incorporate your autosampler into your regular laboratory procedures.

**TEMPLATE 1: DILUTION**

# of Dilution Cycles \_\_\_\_

1- Load \_\_\_\_ $\mu$ L Solv \_\_\_\_

2- Pickup \_\_\_\_ $\mu$ L \_\_\_\_

3- Pickup \_\_\_\_ $\mu$ L \_\_\_\_

4- PickLG \_\_\_\_ $\mu$ L \_\_\_\_

5- Add \_\_\_\_ $\mu$ L to \_\_\_\_

6- Mix for \_\_\_\_ minute

Repeat Dilution Steps

7- Load \_\_\_\_ $\mu$ L Solv \_\_\_\_

8- Pickup \_\_\_\_ $\mu$ L \_\_\_\_

9- Pickup \_\_\_\_ $\mu$ L \_\_\_\_

10- PickLG \_\_\_\_ $\mu$ L \_\_\_\_

11- Add \_\_\_\_ $\mu$ L to \_\_\_\_

12- Mix for \_\_\_\_ minute

Repeat Dilution Steps

13- Load \_\_\_\_ $\mu$ L Solv \_\_\_\_

14- Pickup \_\_\_\_ $\mu$ L \_\_\_\_

15- Pickup \_\_\_\_ $\mu$ L \_\_\_\_

16- PickLG \_\_\_\_ $\mu$ L \_\_\_\_

17- Add \_\_\_\_ $\mu$ L to \_\_\_\_

18- Mix for \_\_\_\_ minute

Repeat Dilution Steps

19- Load \_\_\_\_ $\mu$ L Solv \_\_\_\_

20- Pickup \_\_\_\_ $\mu$ L \_\_\_\_

21- Pickup \_\_\_\_ $\mu$ L \_\_\_\_

22- PickLG \_\_\_\_ $\mu$ L \_\_\_\_

23- Add \_\_\_\_ $\mu$ L to \_\_\_\_

24- Mix for \_\_\_\_ minute

Overlap Enable                      Yes/No

This procedure requires  
that \_\_\_\_ Extra Vial(s) be  
placed immediately  
after each Sample Vial.

**TEMPLATE 2: DABS-CL**

```
# of Dilution Cycles  __
Heater:  __  Temp:  __
1- Load  __µL Solv  __
2- Pickup  __µL  __
3- Pickup  __µL  __
4- PickLG  __µL  __
5- Add  __µL to  __
6- Mix for  __ minute
7- Heat for  __ minute
8- Mix for  __ minute
Repeat Dilution Steps
9- Load  __µL Solv  __
10- Pickup  __µL  __
11- Pickup  __µL  __
12- PickLG  __µL  __
13- Add  __µL to  __
14- Mix for  __ minute
15- Heat for  __ minute
16- Mix for  __ minute
Repeat Dilution Steps
17- Load  __µL Solv  __
18- Pickup  __µL  __
19- Pickup  __µL  __
20- PickLG  __µL  __
21- Add  __µL to  __
22- Mix for  __ minute
23- Heat for  __ minute
24- Mix for  __ minute
Repeat Dilution Steps
25- Load  __µL Solv  __
26- Pickup  __µL  __
27- Pickup  __µL  __
28- PickLG  __µL  __
29- Add  __µL to  __
30- Mix for  __ minute
31- Heat for  __ minute
32- Mix for  __ minute
Overlap Enable          Yes/No
This procedure requires
that  _ Extra Vial(s) be
placed immediately
after each Sample Vial.
```

**TEMPLATE 3:  
EXTRACTION (RAINBOW)**

```

# of Dilution Cycles  __
1- Load  ___µL Solv  __
2- Pickup  ___µL  _____
3- Pickup  ___µL  _____
4- PickLG  ___µL  _____
5- Add  ___µL to  _____
6- Mix for  __ minute
7- Wait for  __ minute
8- Remove  ___µL to
waste at  ___ mm height
Repeat Dilution Steps
9- Load  ___µL Solv  __
10- Pickup  ___µL  _____
11- Pickup  ___µL  _____
12- PickLG  ___µL  _____
13- Add  ___µL to  _____
14- Mix for  __ minute
15- Wait for  __ minute
16- Remove  ___µL to
waste at  ___ mm height
Repeat Dilution Steps
17- Load  ___µL Solv  __
18- Pickup  ___µL  _____
19- Pickup  ___µL  _____
20- PickLG  ___µL  _____
21- Add  ___µL to  _____
22- Mix for  __ minute
23- Wait for  __ minute
24- Remove  ___µL to
waste at  ___ mm height
Repeat Dilution Steps
25- Load  ___µL Solv  __
26- Pickup  ___µL  _____
27- Pickup  ___µL  _____
28- PickLG  ___µL  _____
29- Add  ___µL to  _____
30- Mix for  __ minute
31- Wait for  __ minute
32- Remove  ___µL to
waste at  ___ mm height
FlushP  ___µL Solv  __
Overlap Enable          Yes/No
This procedure requires
that  _ Extra Vial(s) be
placed immediately
after each Sample Vial.

```

**TEMPLATE 4: LINEAR**

```
# of Dilution Cycles  __
Heater:  __  Temp:  __  C
1- Load  __µL Solv  __
2- Pickup  __µL  __
3- Pickup  __µL  __
4- PickLG  __µL  __
5- Add  __µL to  __
6- Mix for  __ minute
7- Heat for  __ minute
8- Mix for  __ minute
9- Wait for  __ minute
10- Remove  __µL to
waste at  __ mm height
Repeat Dilution Steps
11- Load  __µL Solv  __
12- Pickup  __µL  __
13- Pickup  __µL  __
14- PickLG  __µL  __
15- Add  __µL to  __
16- Mix for  __ minute
17- Heat for  __ minute
18- Mix for  __ minute
19- Wait for  __ minute
20- Remove  __µL to
waste at  __ mm height
Repeat Dilution Steps
21- Load  __µL Solv  __
22- Pickup  __µL  __
23- Pickup  __µL  __
24- PickLG  __µL  __
25- Add  __µL to  __
26- Mix for  __ minute
27- Heat for  __ minute
28- Mix for  __ minute
29- Wait for  __ minute
30- Remove  __µL to
waste at  __ mm height
Repeat Dilution Steps
31- Load  __µL Solv  __
32- Pickup  __µL  __
33- Pickup  __µL  __
34- PickLG  __µL  __
35- Add  __µL to  __
36- Mix for  __ minute
37- Heat for  __ minute
38- Mix for  __ minute
39- Wait for  __ minute
40- Remove  __µL to
waste at  __ mm height
FlushP  __µL Solv  __
Overlap Enable  Yes/No
This procedure requires
that  _ Extra Vial(s) be
placed immediately
after each Sample Vial.
```

## **Section 5. Required Maintenance**

This section contains the maintenance schedule and procedures for your autosampler.

### **SPECIFIC HAZARDS**

For your own safety, please familiarize yourself with and follow these safety precautions when using this instrument:

**WARNING – High Voltage!** To avoid electrical shock, do not remove the top cover or remove parts beyond the descriptions in the operation portions of this manual or in the maintenance section.

**WARNING!** Disconnect the power cord before replacing fuses.

**WARNING!** When replacing fuses, use only fuses of the type and current rating specified. Do not use repaired fuses and do not short-circuit the fuse holder.

**WARNING!** Failure to heed these or any other cautions or warnings described in this manual may invalidate your warranty.

### **5.1 Maintenance Schedule**

Your autosampler requires only a few simple maintenance procedures to keep it in optimal working condition. Table 5.1 contains the Maintenance Schedule for the autosampler. The yearly items should be performed only by a BAS service representative or other qualified person.

Table 5.1. Maintenance Schedule.

Frequency	Procedure	Performed By
Daily	Empty flush-solvent tray.	User
Monthly	Inspect the sample needle module. Check the solvent tubing and connections for leaks. Run a standard sample. Clean the drop catch.	User User User User
Semiannually	Replace the flush-solvent inlet filter.	User
Annually	Replace the injector valve's rotor seal. Clean and lubricate the syringe drive mechanism. Clean and lubricate the XYZ arm. Verify the XYZ arm alignment.	User Service Service Service

*NOTES: Maintenance of the autosampler is the responsibility of the user. Routine maintenance is not provided under warranty.*

*This section contains recommended maintenance intervals based upon operation of the autosampler under typical conditions. Operation of the autosampler under certain harsh conditions (corrosive solvents, extreme temperatures, etc.) may require more frequent maintenance than indicated in this section.*

## 5.2 Procedures

### EMPTYING THE FLUSH-SOLVENT TRAY

For normal operation, you may need to empty the tray at least once or twice a day. To empty the tray:

1. Be sure the autosampler is in idle status.
2. Carefully slide the tray out without spilling the collected solvent.
3. Empty the tray, rinse it off with water, and slide it back into place under the black el-bowed, barbed piece of tubing.

**WARNING!** Dispose of hazardous waste according to local, state, and federal regulations.

**CAUTION!** Be sure that the level of the liquid in the waste container is below the level of the instrument or waste will collect in the lines and could backflush into the instrument.

## REPLACING THE ROTOR SEAL

Replacing the rotor seal includes the following procedures:

- Removing the injector valve
- Disassembling the valve
- Cleaning the stator
- Reassembling and replacing the valve

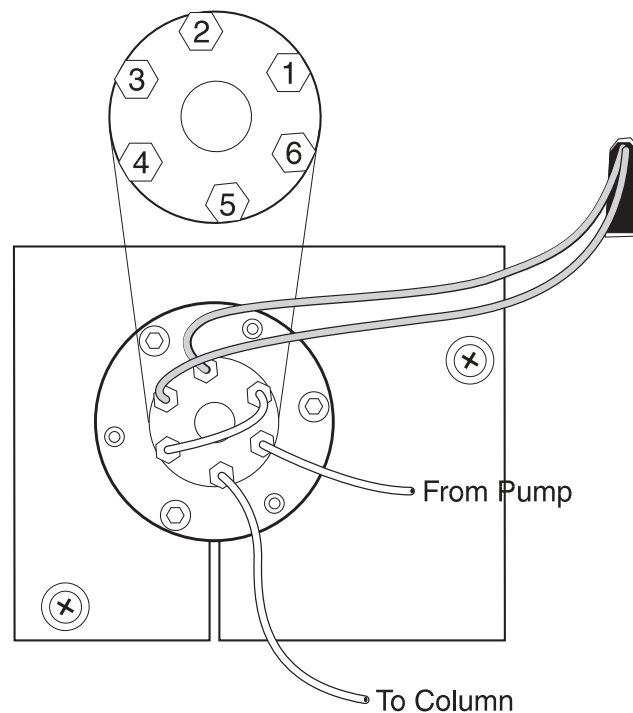
*NOTE: If you are only replacing the rotor seal, you don't have to remove the injector valve from the instrument; however, it's easier. If you wish to replace the rotor seal with the valve installed, proceed to the section entitled, "Disassembling the Injector Valve" on page 80.*

## Removing and Reinstalling the Injector Valve

To disassemble the injector valve (Figure 5.1):

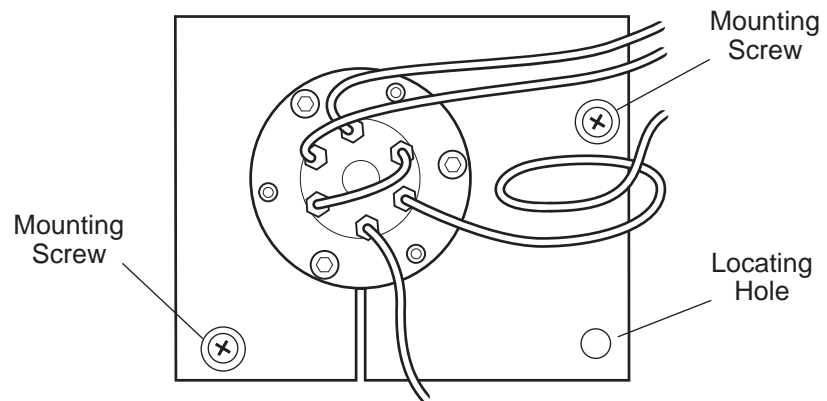
1. Using a 1/4" open-end wrench, disconnect tubing from ports 2, 3, 5, and 6.
2. Remove the two Phillips screws that secure the valve mounting plate.
3. Carefully pull the valve out of the instrument.
4. Reverse these steps to reinstall the valve.

Figure 5.1. The injector valve.



**CAUTION!** When reinstalling the bracket/valve assembly, be sure to orient the bracket with the "locating-hole" corner in the lower right as shown in Figure 5.2. You should be able to see the pin through the locating hole.

Figure 5.2. The correct injector valve bracket orientation.



### Disassembling the Injector Valve

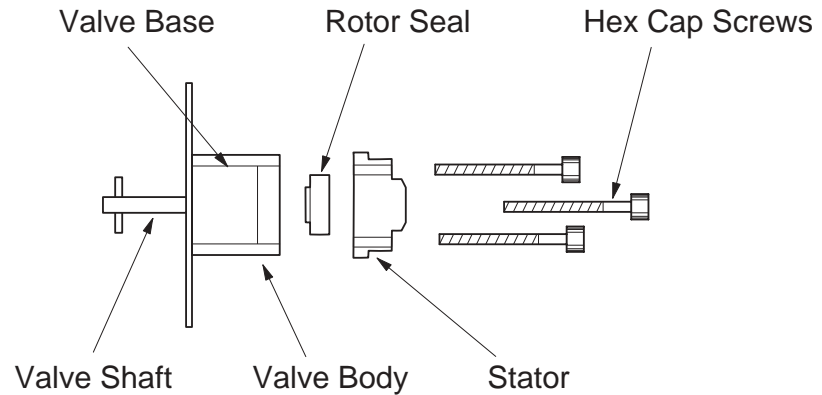
*NOTE: This repair must be done in a clean environment. Component cleanliness will affect the useful life of your injector valve.*

**CAUTION!** Do not activate the valve mechanism with the injector valve removed from the instrument or valve mechanism damage could occur.

To disassemble the injector valve:

1. Follow the procedures above for removing the injector valve.
2. Using an Allen wrench, remove the 9/64" hex cap screws from the top of the valve (Figure 5.3).
3. Carefully lift the stator from the valve body and set it onto a clean surface.

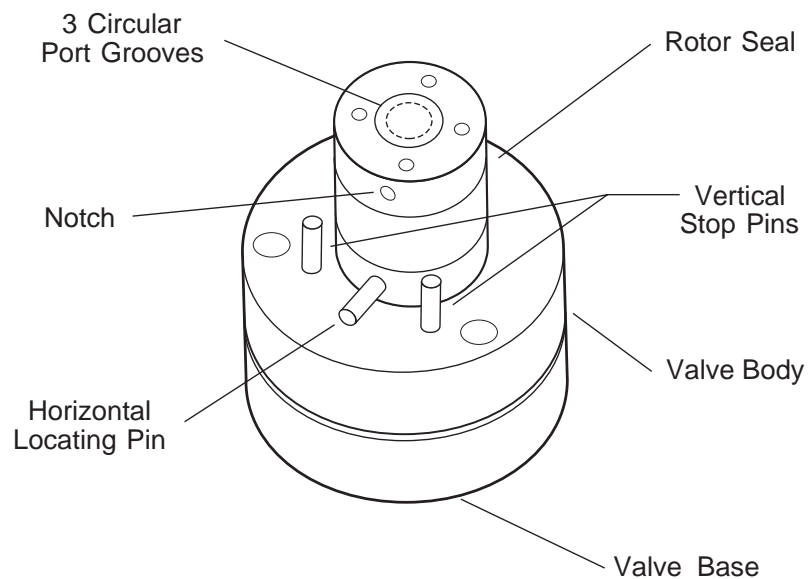
Figure 5.3. The injector valve's components.



*NOTE: Be careful not to drop the two rotation stop pins that protrude from the valve base (Figure 5.4).*

4. Remove the valve body.
5. Before removing the worn rotor seal, note the following on the rotor seal (Figure 5.4):
  - the three circular port grooves on one side of the rotor (the other side is smooth)
  - the small locating notch on the rotor's rim (*not* the V stamped on the seal)

Figure 5.4. The injector valve with valve body removed and rotor seal exposed. (*NOTE: The mounting bracket is not shown in this picture.*)



6. Remove the old rotor seal. A thin, flat-bladed tool may be needed.

### Cleaning the Stator

Clean the stator as follows:

1. Inspect the stator to determine if it requires cleaning.
2. If it is dirty or greasy, swab it with HPLC-grade methanol. If more stringent cleaning is required, use a sonicator.
3. Inspect the remaining valve components for contamination. Clean as necessary.

**CAUTION!** If the stator is scratched, replace it. Scratches can damage the rotor seal and cause valve leaks.

### Installing the Rotor Seal

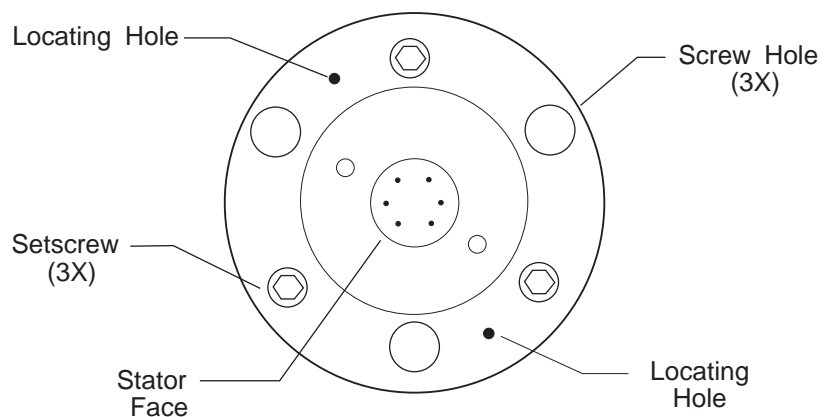
To install the rotor seal:

1. Install the new rotor seal in exactly the same orientation as the old one. Be sure that the rim notch is in line with the horizontal locating pin and that the circular port grooves are visible.
2. Install the valve body.
3. Install the stator onto the valve body carefully, observing that the two locating pins in the valve body (Figure 5.4) line up with the two holes on the bottom side of the stator assembly (Figure 5.5).

**CAUTION:** Be sure to hold the valve shaft so that the horizontal locating pin touches the left stop pin as you reassemble the valve. This puts the valve into the INJECT mode (the same mode as when removed from the instrument). If the pins are not in the correct orientation, the valve shaft may not be in the proper orientation for reinsertion into the instrument.

---

Figure 5.5. The bottom of the stator assembly.



*NOTE: These locating pins orient the stator assembly in one of two positions, but only one position aligns the mounting holes in the valve body with the screw holes in the stator.*

4. Insert the three cap screws so that each one just contacts the stator.

*NOTE: There should be a uniform gap between the stator and the valve body to maintain the necessary pressure for proper valve operation and to avoid valve binding. Do not over-tighten the valve.*

5. Tighten cap screws evenly until all setscrews are in equal contact with the valve body.

## REPLACING THE FLUSH SOLVENT INLET FILTER

Properly filtering your flush solvent extends the life of your injector valve and column. Replace the inlet filter as follows:

1. Be sure the autosampler is in the IDLE mode.
2. Remove the flush-solvent bottle cap and lift out the inlet tubing.

**CAUTION!** Be sure to follow all of the safety precautions for each solvent as prescribed by the manufacturer's MSDS.

3. Put on a new filter cartridge and put the line back into the bottle. Tighten the solvent cap.
4. Flush the inlet filter and lines with an appropriate volume of solvent (~5  $\mu\text{L}$ ).

## SAMPLE NEEDLE MODULE

The sample-needle module resides in the sample tower and is responsible for piercing the vial septum and removing sample from the vial. It easily snaps out of the needle mount and is very easy to maintain.

### **Removal**

Use the following steps to remove the sample needle module:

1. Remove the sample needle fitting from port 2 of the injection valve using a 1/4" open-end wrench.
2. Carefully (to avoid damage to the sample tubing) release the sample needle module from the tower by grasping the module edges and pulling towards the rear of the autosampler.
3. Pass the needle fitting through the autosampler's front bulkhead and remove the needle module from the autosampler.

*NOTE: If you are replacing (not inspecting) the needle, you may wish to cut the used needle to make it easier to pass the fitting through the bulkhead.*

### ***Cleaning***

To clean the sample needle module:

1. Inspect the needle module.
2. Rinse in water or sonicate if contamination is present.
3. If internal plugging is suspected, pass a cleaning wire through the needle and flush the needle with high flow from an LC pump.

**CAUTION!** The sample needle module is glued during manufacture and cannot be disassembled. Attempts to do so will damage the module.

### ***Installation***

Replace the sample needle module every six months. The needle module is easy to replace because it is designed to snap into place.

1. Remove the new module from the replacement kit.
2. Pass the valve end of the needle through the hole on the underside of the column compartment.
3. Snap the needle assembly into the mounting brackets on the backside of the sample tower (Figure 5.6).
4. Place the tube fitting and ferrule onto the needle and install it into port 2 of the injection valve (Figure 5.1).

### **Drop Catch**

The drop catch (Figure 5.6) removes the last drop of sample from the end of the needle after injection. It must be flushed monthly as follows:

1. Remove the needle module assembly as described above, but leave the drop catch installed.
2. Flush the drop catch with water, an organic solvent, an acid or base (as appropriate for the sample), and once more with water. Then dry it with a clean tissue.

**SYRINGE  
REPLACEMENT**

This section describes how to replace the sample (250  $\mu\text{L}$ ) syringe and the large (2.5 mL) sample preparation syringe.

**250  $\mu\text{L}$  Sample Syringe**

You can replace the 250  $\mu\text{L}$  sample syringe without removing the syringe drive assembly, but you must move the syringe drive about two inches out by interrupting a flush cycle. Replace the sample syringe as follows:

1. From the Main Menu, choose /Commands/, and /Flush Sample Syringe/.
2. When the plunger is lowered, turn off the power.
3. Hold the silver base of the plunger to prevent it from turning, and loosen the knurled syringe set screw under the syringe's plunger.

*HINT: You may need to insert a screwdriver or coin into the slot in the set screw to get the screw started and then lower the plunger.*

*NOTE: The syringe set screw is "captive" and won't fall down into the instrument's chassis.*

4. To loosen the top of the syringe, hold the metal tip on the top of the syringe barrel, unscrew the used syringe barrel and lift it out of the instrument.

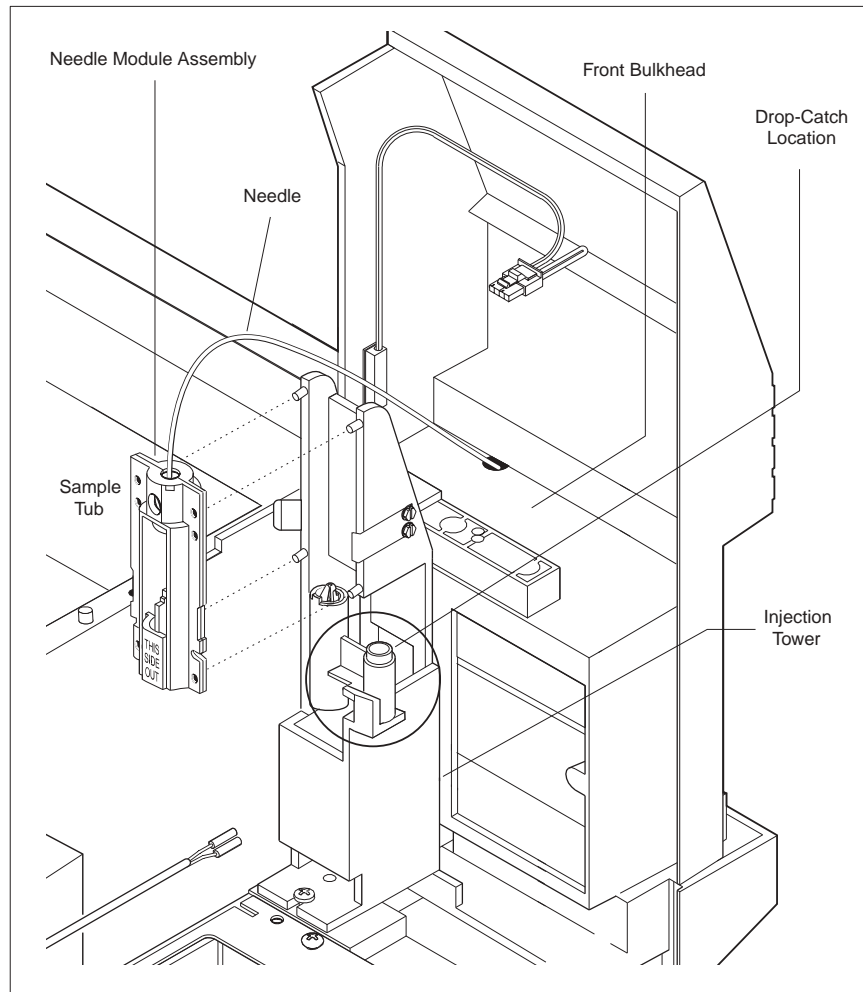
**CAUTION!** Don't try to loosen the syringe by holding the barrel. The syringe could break between the glass barrel and the metal tip.

*HINT: Push up on the syringe plunger while lifting it out of place.*

*HINT: Install the syringe in its fully-expelled position to prevent an air bubble in the syringe barrel.*

5. Reverse steps 4 and 5 to install your new syringe.

Figure 5.6. Replacing the sample needle module.



### 2.5 mL Preparation (Prep) Syringe

If you have the Sample Preparation Option, you can position the large (2.5 mL) syringe's plunger in the down position by interrupting a flush cycle, or you can remove the prep syringe without using the display and with the plunger in the normal operating position. The two procedures are detailed here.

### ***Interrupting a FlushPrep Cycle***

1. Be sure the autosampler is in the IDLE mode.
2. Move the XYZ arm out of your way by pressing [MENU], Commands, /Arm/, /Align Arm/, and selecting position A01. Then press [ENTER].
3. To start the flush cycle, from the Main Menu, choose Commands, and Flush Prep Syringe.
4. When the plunger has lowered, turn off the power.
5. Hold the plunger to prevent it from turning, and loosen the knurled syringe set screw under the syringe's plunger.

*HINT:* You may need to insert a screwdriver or coin into the slot in the set screw to get the screw started.

*NOTE:* The syringe set screw is "captive" and won't fall down into the instrument's chassis.

6. To loosen the top of the syringe, hold the metal tip on the top of the syringe barrel, unscrew the used syringe barrel and lift it out of the instrument.

**CAUTION!** Don't try to loosen the syringe by holding the barrel. The syringe could break between the glass barrel and the metal tip.

*HINT:* Push up on the syringe plunger while lifting it out of place.

7. Reverse steps 5 and 6 to install your new syringe.

*HINT:* When reinserting the bottom set screw, you may need to pull the plunger down slightly so that the end of the screw will reach the bottom of the plunger.

### ***Removing Syringe with Plunger in Operating Position***

Use the following procedure to remove the prep syringe without accessing the instrument's display:

1. Be sure the autosampler is in the IDLE mode or turned off.
2. Move the arm and hook to position A01 so that you can easily access the syringe assembly.

*HINT:* You can move the arm out of the way manually, or press [MENU], /Commands/, /Arm/, /Align Arm/, select position A01, and press [ENTER].

**CAUTION!** Before moving the arm manually, be sure the pod (the triangular assembly that moves along the XYZ arm) is pushed all the way to the left. If you try to move the arm with the sample tower pushed toward the right side of the sample tub, the arm may bind and not move easily.

3. Hold the plunger to prevent it from turning, and loosen the knurled syringe set screw under the syringe's plunger.

*NOTE: The syringe set screw is "captive" and won't fall down into the instrument's chassis.*

4. Holding the metal tip on the top of the syringe barrel, unscrew the used syringe barrel and lift it out of the instrument.

*HINT: Push up on the syringe plunger while lifting it out of place.*

*Install the syringe in its fully expelled position to prevent an air bubble in the syringe barrel.*

5. Reverse steps 3 and 4 to install your new syringe.

*HINT: When reinserting the bottom setscrew, you may need to pull the plunger down slightly so that the end of the screw will reach the bottom of the plunger.*

## **SAMPLE LOOP**

Different size sample loops are available for the Sample Sentinel autosamplers. Call BAS for sizes, part numbers, and prices. To change the sample loop:

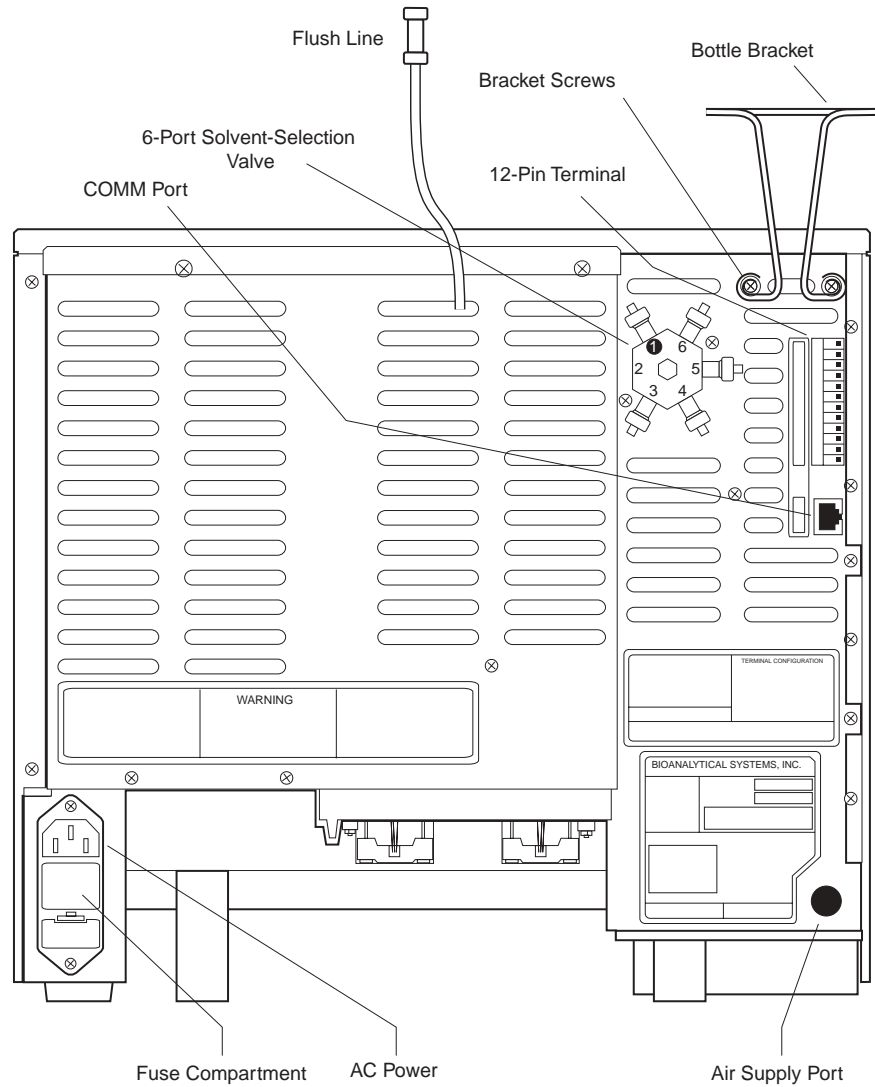
1. Be sure that the autosampler is in the IDLE mode.
2. Using an open-end wrench, loosen the fittings at ports 1 and 4, and pull out the two ends of the sample loop.
3. Insert the ends of the new sample loop in the same orientation as the old one and tighten the fittings.

*NOTE: If the Sample Preparation Option is installed, to ensure maximum reproducibility, do not perform small-volume injections with the large (preparation) syringe.*

**FUSES**

Instrument power is supplied by two 5.0-amp SB fuses housed in the fuse compartment on the lower left corner as you are facing the instrument's rear panel (Figure 5.7).

Figure 5.7. The autosampler's rear panel.



To replace the fuses:

1. Use a flat blade to press down on the plastic tab at the top edge of the fuse compartment and pull out the fuse holder.
2. Pull both fuses out of the holder and discard. Place the new fuses into the holder with the metal end visible.
3. Snap the fuse holder back into place.

### 5.3 Maintenance Options

#### SHUTDOWN

If the autosampler is not used for a period of time (after completing an overnight or over-the-weekend run), you can set it to turn off automatically. You enter the shutdown parameters in the OPTIONS Shutdown Menu (Figures 5.8 and 5.9).

Figure 5.8. The Options Menu.

Configurations
Ready Participation
Input Polarity
Output Polarity
Shutdown
Maintenance Log
Communications

Figure 5.9 illustrates the autosampler's Shutdown Menu.

Figure 5.9. The Shutdown Menu.

Delay Time	Off
TrayTemp Shutoff	No <i>(Only if tray temperature control installed)</i>
Heater Shutoff	No <i>(Only if sample prep installed)</i>
Pump Shutoff	Yes
Flush Volume	400
Prep Flush Volume	1000 <i>(Only if sample prep installed)</i>

#### ***Delay Time***

In this field you specify the desired amount of time that the autosampler should wait before initiating the shutdown procedure (Off, 5, 10, 20, 30, 45, 60, 90, 120, 240, 480 minutes).

**Oven Shutoff**

Use the [+] and [-] keys to select Yes or No. Selecting Yes causes the autosampler to automatically turn off the oven after the Shutoff Delay time has elapsed.

*NOTE: If you are using the Shutdown function with methods that call for heating and cooling, be sure to allow a long enough delay time (not less than 10 minutes) for thermal equilibrium to occur.*

**TrayTemp Shutoff**

Use the [+] and [-] keys to select Yes or No. Selecting Yes causes the autosampler to automatically turn off the Tray Temperature Control Option after the Shutoff Delay time has elapsed. You may or may not wish to turn off tray temperature control depending upon your sample requirements.

**Heater Shutoff**

Use the [+] and [-] keys to select Yes or No. Selecting Yes causes the autosampler to automatically turn off the heater after the Delay Time has elapsed. This field appears only if the Sample Preparation Option is installed.

**Pump Shutoff**

Use the [+] and [-] keys to select Yes or No. Selecting Yes causes the autosampler to automatically activate the Pump Stop output after the Delay Time has elapsed. Turning off the pump retains your solvent supply and prevents air from being pumped into the lines should the solvent reservoirs become depleted.

**Flush Volume**

This field allows you to flush the lines using the 250  $\mu$ L sample syringe. Specify the flush volume in microliters (200–5000). The default value is 400  $\mu$ L.

**Prep Flush Volume**

This field allows you flush the lines using the 2.5 mL sample preparation syringe. Specify the flush volume in microliters (0–6500). This field appears only if the Sample Preparation Option is installed.

**MAINTENANCE LOG**

The Maintenance Log Menu (Figure 5.10), accessible from the OPTIONS Menu, monitors the instrument's maintenance status and counts and displays the number of activations for each of the autosampler's routine maintenance parts listed in the menu.

Figure 5.10. The Maintenance Log Menu.

Total System Cycles
Injector Valve
Needle Wear
Sample Syringe
Prep Syringe

### ***Instrument Maintenance Status***

The Maintenance Log Menu provides a convenient way to keep track of autosampler maintenance. When you first access the Maintenance Log Menu from the OPTIONS Menu, the "Maintenance Required" confirmation message is displayed if instrument maintenance is required. This message is displayed if any of the #Cycles field values in any of the Maintenance Log menus is greater than the number of cycles entered in the Due fields. See below for more information on these fields.

No message is displayed if maintenance is not required. Pressing the down-arrow key clears the confirmation message and returns you to the Maintenance Log Menu.

### ***Total System Cycles Menu***

The Total System Cycles Menu (Figure 5.11) displays the total number of instrument cycles in the Total Cycles field. This field value is set to zero at the factory and cannot be reset. The Note field is a six-character, alphanumeric, user-programmable field. You can use the [+]/[-] keys to enter the last service date or other comments regarding the maintenance of your instrument.

Figure 5.11. The Total System Cycles Menu with the Note field used to indicate the next maintenance due date.

#Cycles	Note
35	Feb 93

### ***Injector Valve Menu***

With the exception of the Total System Cycles Menu, all of the Maintenance Log menus are identical to the Injector Valve Menu (Figure 5.12). Therefore, only the Injector Valve Menu will be described in detail in this section. Refer to Section 7 for definitions of the other Maintenance Log menus.

Figure 5.12. The Injector Valve Menu.

#Cycles	Due	Note
202	24000	021593
-----		
Reset	#Cycles	YES

**#Cycles:** This field displays the number of activations of the specific autosampler part since the last time this field (counter) was reset (0–999,999). The #Cycles value for each of the Maintenance Log menus is included in the menu descriptions below.

**Due:** This is a six-character numeric field where you enter the number of cycles before maintenance is due. If the #Cycles field value is larger than the Due field value, the "Maintenance Due" message is displayed when you access the Maintenance Log Menu from the Options Menu. The default maintenance Due field value is 12,000 (approximately once a year). The default values for the other Maintenance Log menus are included in the menu descriptions below.

*NOTE: The Due field default values for all of the Maintenance Log menus are estimates only. The actual values appropriate for your instrument will vary with the type of applications and the specific conditions being used with your LC. If the "Maintenance Required" message is displayed, parts replacement is recommended. However, you may wish to increase the Due value and experiment with extending the time before maintenance is performed. Conversely, if instrument performance indicates, you may wish to decrease the Due field value for some of the Maintenance Log menus.*

**Note:** Similar to the Total System Cycles Menu, this is a six-character, alphanumeric, user-programmable field. We recommend that you use the [+]/[–] keys to enter the last service date in this field. However, you can also use this field to document contact information or other comments regarding the maintenance of the specific autosampler part.

**Reset #Cycles:** Pressing the down-arrow key displays the Reset #Cycles field used to reset the #Cycles counter for each autosampler part. To reset the counter to 0 after maintenance is complete, press the [+]/[-] keys to select YES, and press [ENTER]. The confirmation message \*\* Counter Reset \*\* is displayed. Most likely, your service representative will reset the #Cycles field value when the instrument is serviced.

### ***Needle Wear Menu***

The #Cycles field in this menu is incremented each time the needle pierces a vial. The Due field's default value is 10,000, which corresponds to approximately every six months.

### ***Sample and Prep Syringe Menus***

The #Cycles field in both of these menus displays each full syringe stroke. However, the instrument tracks the actual distance of linear travel, thus compensating for different injection (stroke) volumes. The default Due field value for both menus is 24,000, which corresponds to approximately once a year.

## Section 6. Basic Installation and Specifications

This section describes the site requirements, basic installation procedures, and performance testing for the SS-1 and SS-2 Sample Sentinel autosamplers. Section 9 contains instructions for the advanced installation procedures and instrument configuration and initialization procedures.

*HINT: If you are setting up your autosampler for the first time, you may wish to follow the procedures in the basic installation section and skip the advanced section until you are more familiar with your instrument.*

### Basic Installation: OVERVIEW

The easiest way to install your Sample Sentinel autosampler is to unpack it and set it on the bench, complete the procedures that require access to the back of the instrument, and then complete those that require access to the front. Additional or modified procedures required for instruments with one or more of the integrated options are incorporated into this section.

*NOTE: For a list of all available options, upgrades, and accessories for your autosampler, contact your BAS sales representative. Note that all upgrades require specialized installation. Contact your BAS representative for further information.*

If you purchased the Sample Preparation Option, there are several additional steps in making the rear panel connections. These changes to the normal setup procedure are included in the rear- and front-panel sections, respectively. The sample Tray Temperature Control Option comes completely installed and requires no special user installation procedures.

### BEFORE YOU START

Before you start, review the site requirements and assemble the tools you'll need to install your instrument:

- One Phillips screwdriver
- Two 1/4" open-end wrenches
- One small, standard screwdriver

## 6.1 Site Requirements

### SPACE

The standard autosampler weighs 16 kg. With all options installed, the autosampler weighs 24 kg. The instrument requires a bench space at least 37 cm by 41 cm by 51 cm. Be sure to allow adequate ventilation away from heating or air conditioning ducts. Avoid exposure to direct sunlight.

*HINT:* You may wish to set your instrument on the bench such that you have easy access to the rear panel.

**CAUTION!** Be sure to use the proper venting apparatus if you use hazardous solvents.

### ELECTRICAL

Your autosampler automatically configures itself to operate from the local line voltage (100–260 VAC at 50 or 60 Hz). You do not need to set the line voltage. The instrument is shipped with the necessary fuses installed.

You should provide a stable, spike-free power source. This will avoid intermittent failures and unexplained occurrences (lockup, loss of memory, incorrect operation) that are directly related to poor power. We also recommend that you use an uninterruptible power supply (UPS) for maximum protection from power-related failures.

## 6.2 Unpacking

Your autosampler was shipped in a specially designed shipping container to protect it from damage during transit. Upon arrival, inspect all containers and immediately report any damage in transit to the transportation company. Have the carrier note the shipping crate's condition on both the delivery receipt and the freight bill. The carrier is responsible for all damage incurred in shipment.

All Sample Sentinels are shipped with the sample trays and syringe(s) installed. A cardboard shipping insert on top of the sample trays restrains the XYZ arm and holds the sample trays in place during shipment. Remove the shipping insert. Retain both the shipping insert and shipping crate should future shipment become necessary.

After unpacking, inspect your autosampler and its accessories for missing parts and/or physical damage. If damage is found, notify both the carrier and your BAS representative. Please do not return goods without prior authorization (a Return Authorization number) from BAS.

**ACCESSORY KIT**

The contents of your accessory kit will vary with the model and options you ordered (refer to Table 6.1). Notify your BAS representative of any shortages or damage.

Table 6.1. Accessory Kit Contents

<b>STANDARD ACCESSORY KIT</b>	
Qty	Description
1	250 mL glass bottle, screw top
3	Extra long Rheodyne fitting/bushing
3	Rheodyne ferrule
1	6.3A fuse, 250V, SB
1	Tube, stainless steel, .060DX.01 id 12 inches
1	Tube, .06X.01X4.0
1	Vial kit (100)
1	Flush waste container
1	Bracket, flush solvent bottle
1	Cable assembly (4-conductor)
1	Solvent line preparation kit (cap and lines)
1	Label set
2	Screws for bottle bracket
<b>SAMPLE PREPARATION ACCESSORY KIT</b>	
4	Solvent filter
4	.125 tubing assembly with Cheminert™ fittings

## 6.3 Installation

### REAR PANEL

Installation is easiest if you make the rear panel connections first. The rear panel installation procedures include plumbing (flush bottle), system communication, and AC power.

### Plumbing

Your accessory kit includes a 250 mL flush-solvent bottle and cap and a bottle bracket. The bracket attaches to the upper-right corner of the rear panel (as you are facing the rear of the instrument) and holds the flush bottle in the correct position and elevation for optimal flow through the lines.

*NOTE: If you wish to use your own solvent bottle or other container, be sure that it is clean and chemically inert. We recommend using glass bottles.*

#### **Bottle Bracket**

The bottle bracket fastens to the rear panel with two Phillips-head screws included in your accessory kit. To install the flush-bottle bracket:

1. Align the bracket's mounting holes with the corresponding screw holes on the autosampler's rear panel (Figure 6.1). Position the bracket so it forms a guard rail for the bottle, which sits on top of the autosampler.
2. Insert the screws into the holes and tighten them in place. Be sure that the bracket is fastened securely onto the back of the instrument.

#### **Flush Bottle**

The flush bottle rests in the bracket and connects to the factory-installed flush line (Figure 6.1) via a Cheminert® union. The cap assembly comes with the 1/8" od × 1/16" id Teflon inlet line and high-molecular-weight polyethylene filter attached.

Use the following steps to install the flush bottle:

1. Rinse the bottle with LC-grade solvent to remove any dust. Then fill with appropriate LC-grade solvent.
2. Insert the filter into the solvent bottle and screw on the cap. Make sure that the inlet filter rests on the bottom of the solvent bottle.
3. Run the vent line to an appropriate exhaust apparatus, particularly when solvents with harmful vapors are used.

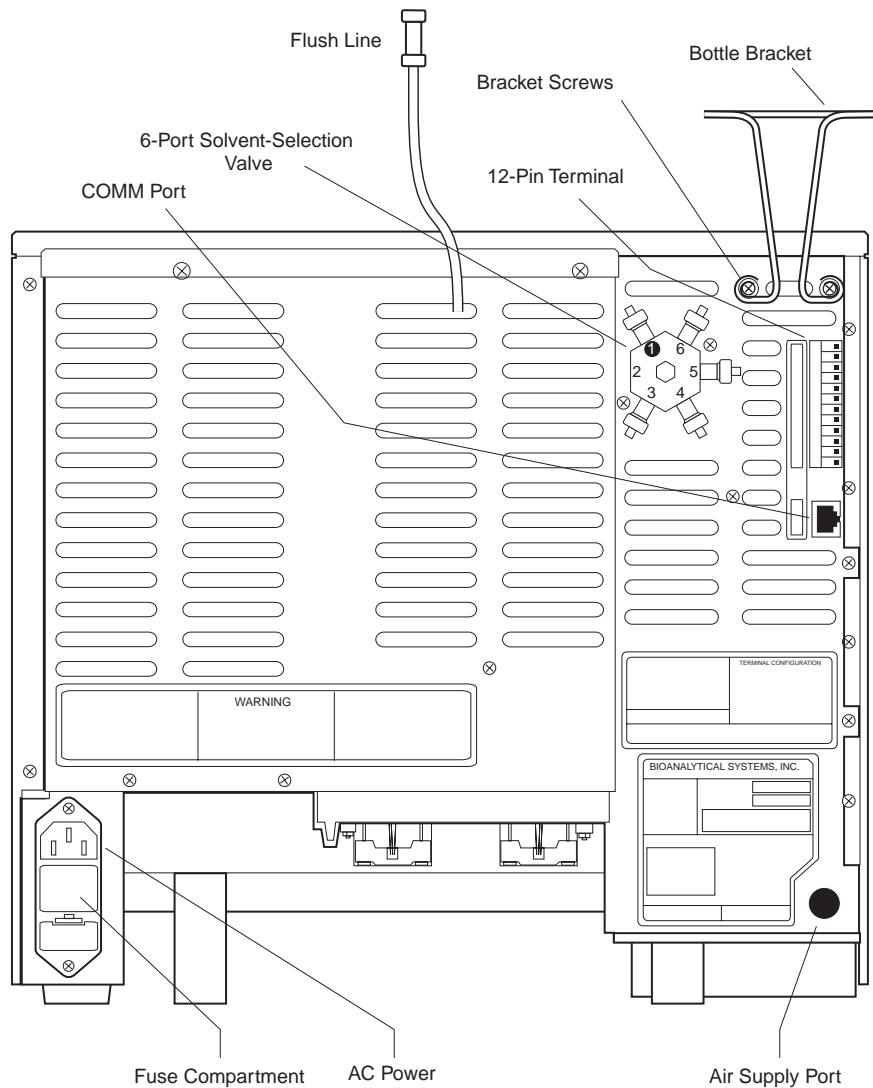
**WARNING – Chemical Hazard!** Many LC solvents are hazardous and must be handled properly. Always dispose of waste solvents in an environmentally correct manner.

- Place the flush bottle in the bracket.

*NOTE: If you did not purchase the Sample Preparation Option, proceed to "System Communication" on page 101.*

If you purchased the Sample Preparation Option, you will also need to install your external solvent bottles and lines and connect the heater/mixer to an air source.

Figure 6.1. The autosampler's rear panel connections (with the Sample Preparation Option installed).



### ***External Solvents (S1–S4) (Sample Preparation Option)***

If you purchased the Sample Preparation Option, the internal solvent-line plumbing is already installed. You only need to set up your external solvent bottles and cap assemblies (not supplied), and connect the inlets and filters to the solvent-line extensions on the rear panel (Figure 6.1). Your accessory kit contains four solvent lines and four filter assemblies.

Each solvent line has a flared (flanged) Cheminert nut on one end; the other end is flareless. To connect a solvent line, you simply connect the Cheminert nut to the Cheminert union on the appropriate solvent line extension and attach the filter assembly to the other (flareless) end.

Use the following procedure to connect each external solvent line to its extension.

1. Designate four bottles as your solvent bottles (S1–S4), and plan the required space near your autosampler.
2. Remove the solvent lines and numbered labels from your accessory kit.
3. Clip the tie-wrap that coils the solvent-line tubing during shipment.
4. Remove the red shipping cap from the end of the appropriate solvent-line extension.

*NOTE: You don't have to use all four solvent bottles. Leave the red shipping cap on the end of any solvent line you're not planning to use.*

5. Connect the Cheminert union to the Cheminert nut on the end of the appropriate solvent-line extension.

*NOTE: If you are going to use bottle caps, thread the other (flareless) end through the bottle cap.*

6. Remove a filter assembly from the accessory kit.
7. Slide the filter over the tubing with the threaded portion toward the end of the tubing.
8. Repeat steps 3–7 for the remaining solvent bottles and lines.

### **Heater/Mixer Air Supply**

The Sample Preparation Option also requires that you connect the heater/mixer to an air supply if you plan to use the vortex mixer. The heater/mixer requires a clean, filtered, regulated air supply (40–100 psi) to prevent particulates from obstructing the narrow tubing in the heater.

*NOTE: You can also use dry, filtered house air.*

Connect the autosampler's air input line to your air supply. The air inlet line is located in the lower right-hand corner of the rear panel (Figure 6.1). It is flared and has a Cheminert nut on the end.

Use the following steps as guidelines and adapt as necessary when connecting your autosampler to your air supply.

1. Remove the red cap from the end of the air input line.
2. Attach the flared end of the air inlet line to your air supply.

*NOTE: Depending upon your regulator, you may need to cut off the Cheminert nut and flared end of the air supply line and use a standard 1/4" brass fitting in order to make the connection.*

3. Connect the brass connector to your regulator and use a wrench to tighten it in place.

### **System Communication**

Your autosampler can be connected to other modules via the rear panel's 12-pin terminal. Refer to Section 9 to connect your autosampler to other LC modules.

### **AC Power**

Locate the power cord in the shipping container (not in your accessory kit). Insert one end into the AC power receptacle on the rear panel and the other end to a grounded power outlet. You don't have to worry about the fuses. Your autosampler is shipped with the correct fuses installed.

## FRONT PANEL

After you've completed all of the rear-panel connections, you can move to the front panel.

The front panel connections include:

- Solvent tray/waste container installation
- Pump-autosampler connection
- Column installation
- Autosampler-detector connection

The 250  $\mu\text{L}$  syringe, 40-vial sample trays, sample needle, and a 100  $\mu\text{L}$  sample loop are already installed.

*NOTE: The SS-1 comes with the 20  $\mu\text{L}$  sample loop installed.*

If you purchased the Sample Preparation Option, the large (2.5 mL syringe) is also factory installed. If you purchased the Tray Temperature Control Option, your trays have only 35 vial positions (to accommodate the tub insulation).

## Solvent Tray/Waste Collection Container

A solvent-collection tray is provided for small-volume applications. For larger-injection applications, or for those requiring frequent flushing, you may wish to install a carboy or other large container for waste collection. Install a waste-collection container using one of the following procedures.

### **Solvent Tray**

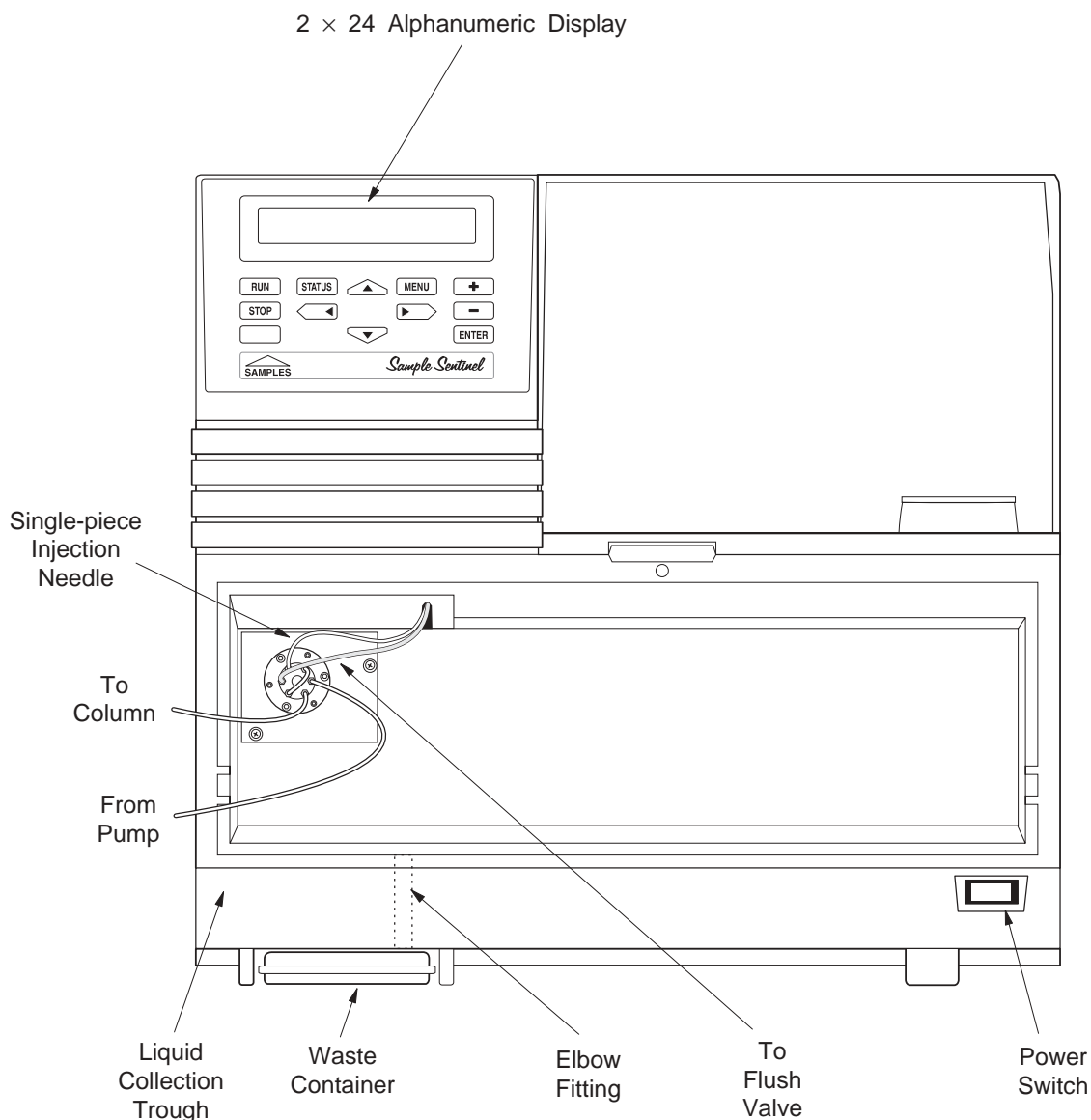
The solvent tray (the flat, white plastic container included in your accessory kit) can hold 100 mL of waste. To install it, simply slide it into the compartment under the front-left side of the instrument.

*NOTE: The back of the compartment prevents the tray from sliding too far under the instrument.*

Orient the tray so that the black-plastic, barbed elbow fitting is directed through the hole in the liquid-collection trough (across the front of the instrument) (Figure 6.2).

*NOTE: Sometimes the barbed fitting may become dislodged during shipping. Be sure that the end of the barbed fitting is directed through the hole (not into the trough).*

Figure 6.2. The autosampler's front panel connections (the optional column oven installed).



### ***Other Waste-Collection Container***

If you elect not to use the autosampler's solvent tray, you'll need to provide some other waste-collection device.

To hook up your waste container, locate the Tygon tubing included in your accessory kit and connect it to the end of the black, barbed fitting. To prevent backflushing into the lines, place the container lower than the instrument. Direct the Tygon tube into your container.

### Pump-Autosampler Connection

Your standard accessory kit contains two pieces of 0.010 id stainless-steel tubing (one 4-inch piece and one 12-inch piece). The 12-inch piece is included to make the column-detector connection, but may be used instead to make the pump-autosampler connection.

The instrument is shipped with the needle and flush lines installed. To connect your pump to the autosampler's injection valve, use the following steps:

1. Remove the column compartment cover (if you have not already done so) and set it aside.

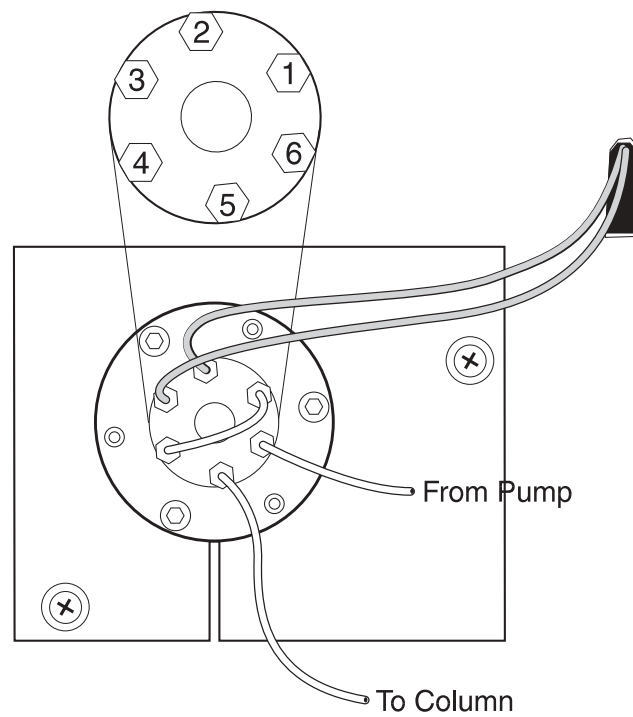
*NOTE: The cover fits snugly. To remove, place your fingers or thumb in the indentation on the top edge of the column door and pull back toward you firmly.*

2. Route the pump outlet line through one of the two notches on the left side of the autosampler.
3. Connect the line to port 6 (Figure 6.3) on the injector valve. Finger-tighten, then use a 3/16" wrench to tighten the fitting an extra 1/8 turn.

*NOTE: Do not under- or over-tighten the screws, or damage to the connections could occur. Undertightening causes leaks; overtightening may cause leaks or damage the connections. We recommend fingertight PEEK fittings (MF-4165), since they are easily replaced and do not permanently affix to the stainless steel tubing.*

---

Figure 6.3. The autosampler's injector valve.



**Sample Loop Installation**

The SS-2 autosampler comes with a 100  $\mu$ L sample loop installed; the SS-1 autosampler comes with a 20  $\mu$ L loop. If you wish to change sample loops, refer to Section 5, page 88.

**Column Installation**

*NOTE: Flush/purge the pump and autosampler lines before connecting the column. This will ensure that no air gets into the column.*

Mount a column on your autosampler as follows:

1. Locate the 4-inch length of stainless steel tubing (0.010) and fittings included in your accessory kit.
2. Connect the 4-inch tubing to the inlet end of the column.
3. Connect the other end of the 4-inch tubing to port 5 on the injector valve (Figure 6.3).

*NOTE: BAS columns are connected such that liquids flow from left to right as you read the label. If you're not using a BAS column, refer to your column documentation to orient your column in the correct direction of flow.*

4. Use a 1/4–3/16" wrench to tighten the fittings. If you're installing the column onto the autosampler for the first time, tighten the fitting 1/8-turn past finger-tight. Otherwise, tighten the fittings 1/16-turn past finger-tight.

**CAUTION!** Do not overtighten the fittings. You could damage, or cause leaks to form around, the column connections. We recommend fingertight PEEK fittings (MF-4165), since they are easily replaced and do not permanently affix to the stainless steel tubing.

**Autosampler-Detector Connection**

The 12-inch piece of 0.010 id tubing is provided in your accessory kit to connect your autosampler to your detector. Simply connect the outlet end of the column to your detector.

*NOTE: If you are using a BAS detector, you won't need the 12-inch tubing because the detector's inlet line is long enough to reach to the outlet end of your column.*

You have completed the front-panel connections. Replace the column oven cover. It snaps firmly into place.

*HINT: To replace the cover, place the bottom edge into place and then snap the top closed.*

**6.4 Instrument Startup and Priming Procedures**

After you have completed the rear and front panel connections, you need to start up and prime your autosampler to wet all of the lines.

## STARTUP

Turn on your instrument by pressing the on/off button located in the lower right-hand corner just below the front panel (Figure 6.2).

During startup, the instrument's self-diagnostic sequence

- Completes a series of ROM checks
- Completes a series of RAM checks
- Activates the injection valve
- Verifies the proper connections on all installed options
- Verifies the proper operation of all sensors
- "Homes" (returns to starting position) all syringes and motors

## PRIMING

To prime your instrument, you'll need to flush all syringes and lines with an appropriate solvent.

**CAUTION!** The autosampler is shipped with methanol in the lines. Be sure to use a miscible solvent.

*HINT: If you purchased the Sample Preparation Option, proceed to the "2.5 mL" section to beginning priming the lines using the prep syringe. It will require less time. Then return to the "250  $\mu$ L Syringe" section.*

## 250 $\mu$ L Syringe

To prime the autosampler:

1. Fill your flush solvent bottle, if you have not already done so.

*NOTE: Where possible, avoid using salts in your flush solvent, as dried salt deposits tend to corrode and abrade the system. Water, or water with a small amount of organic solvent (never more than the amount in your mobile phase), is a good choice. These liquids should be LC-grade, freshly prepared, and vacuum-filtered each day. Flush solvent will be injected onto the column when injection volume is less than the loop size!*

2. Press [MENU] and select /COMMANDS/, (Figure 6.4).

Figure 6.4. (top) The Commands Menu; (bottom) the SS-2 Commands Menu with the Sample Preparation Option installed.

```
Flush Sample Syringe
Initialize Hardware
Injector
Arm
```

```
Flush Sample Syringe
Flush Prep Syringe
Initialize Hardware
Injector
Arm
```

3. Select /Flush Sample Syringe/ to access the menu shown in Figure 6.5.
4. Use the [+] and [-] keys to select 5000  $\mu\text{L}$ . Then press [ENTER] to initiate the flush operation.

Figure 6.5. The Flush Sample Syringe Menu.

```
Flush 5000  $\mu\text{L}$ 
```

The autosampler automatically flushes the sample syringe and flush lines with the specified volume of flush solvent and returns the syringe to its starting position. The recommended 5000  $\mu\text{L}$  flush takes approximately ten minutes. During the flush sequence, a confirmation message is displayed.

*NOTE: You cannot exit from the flush sequence. If you must terminate, turn off the main power.*

## 2.5 mL Syringe

If you purchased the Sample Preparation Option, you must also flush the large (2.5 mL) sample preparation syringe and external solvent lines as follows:

1. Fill each solvent bottle with an appropriate solvent.
2. Press [MENU] and select /COMMANDS/.
3. Select /Flush Prep Syringe/ (Figure 6.4).
4. Use the [+] and [-] keys to select 5000  $\mu\text{L}$ . Then move the cursor to the Solv field and select /Btl/. Press [ENTER] to initiate the flush sequence.

Figure 6.6. The Flush Prep Menu.

Flush 5000  $\mu$ L Solv Btl

**CAUTION!** If there is residual air in the solvent-holding loop, flushing may expel solvent out the open port. To eliminate residual air that may be present during shipping, be sure to flush the lines with Btl selected before flushing with S-1 through S-4 selected.

5. Repeat this step with 1000  $\mu$ L from whichever solvent bottles (S-1 through S-4) are installed. Use the [+] and [-] keys to switch from (Btl) to S-1 through S-4.

The autosampler automatically flushes the selected solvent line with the specified volume of flush solvent and then returns the syringe to its starting position.

**CAUTION!** If you are replacing syringe(s), be sure to install the 2.5 mL syringe on the left. Injecting small sample volumes with the large syringe can adversely affect reproducibility.

## 6.5 Specifications

### Vial capacity

- 120 vials in three removable trays
- 105 vials with tray temperature control

### Variable-Volume

#### *Injection Precision*

- < 0.5% RSD @ 10  $\mu\text{L}$  or greater

#### *Injection Volume*

- 0.1–100  $\mu\text{L}$  injection standard (250  $\mu\text{L}$  syringe)
- up to 1000  $\mu\text{L}$  injections with larger loops and syringes

### Fixed-Loop

#### *Injection Precision*

- < 0.5% RSD @ 10  $\mu\text{L}$  or greater

#### *Injection Volume*

- 20  $\mu\text{L}$  standard (250  $\mu\text{L}$  syringe)
- 1000  $\mu\text{L}$  injections with larger loops and syringes

### Sample Carryover

- < 0.01% at 400  $\mu\text{L}$  flush volume
- 5 mL flush volumes can be set

### Minimum Sample Volume

- 1  $\mu\text{L}$  can be injected from 5  $\mu\text{L}$  with an end-port needle

### Needle Height

- Programmable in 0.1 mm increments

### Tray Temperature Control (optional)

- Settable in 1  $^{\circ}\text{C}$  increments from 0–60  $^{\circ}\text{C}$
- Temperature stability  $\pm 1$   $^{\circ}\text{C}$
- Typical vial temperature from 4–50  $^{\circ}\text{C}$

Sample Preparation (optional)

*Reagent Vials*

Three vials can be specified per file

*Dilution Solvents*

Four external solvents can be selected

*Heating/Mixing*

Single vial, vortex heater/mixer

Settable in 1 °C increments from 30–100 °C

*Dilution Precision*

Typically < 1.0% RSD for a 1/100 dilution (combined dilution and injection)

*Dispensing Precision*

Typically < 0.2% RSD for 1 mL

Method Files

Protected in non-volatile memory

Communications

*Remote Inputs*

Pump Ready, Inject Hold

*Outputs*

Pump Stop

Autosampler Ready

Inject

Gradient Start

Four timed-event outputs

BCD (optional)

Ambient Environment

10–40 °C, 5–95% relative humidity

Dimensions

37 cm × 41 cm × 51 cm (H × W × D)

Weight

16 kg standard; 24 kg with all options installed

Power requirements

100/120/220/240 VAC nominal; 450 VA

50/60 Hz

Product Certification

CSA, TÜV, FCC, CE Mark

## **Section 7. Menu Reference**

This section provides you with your autosampler's menu tree and menu reference. It is not necessary to read this section in order to learn how to use your autosampler. It is included in the manual as a quick reference and aid to using your instrument.

The Menu Trees represent the overall SS-1 and SS-2 menu structures. They show the location and interrelation of all the menus for your autosampler. The menu trees will also help if you become "lost" while moving through the autosampler's menus.

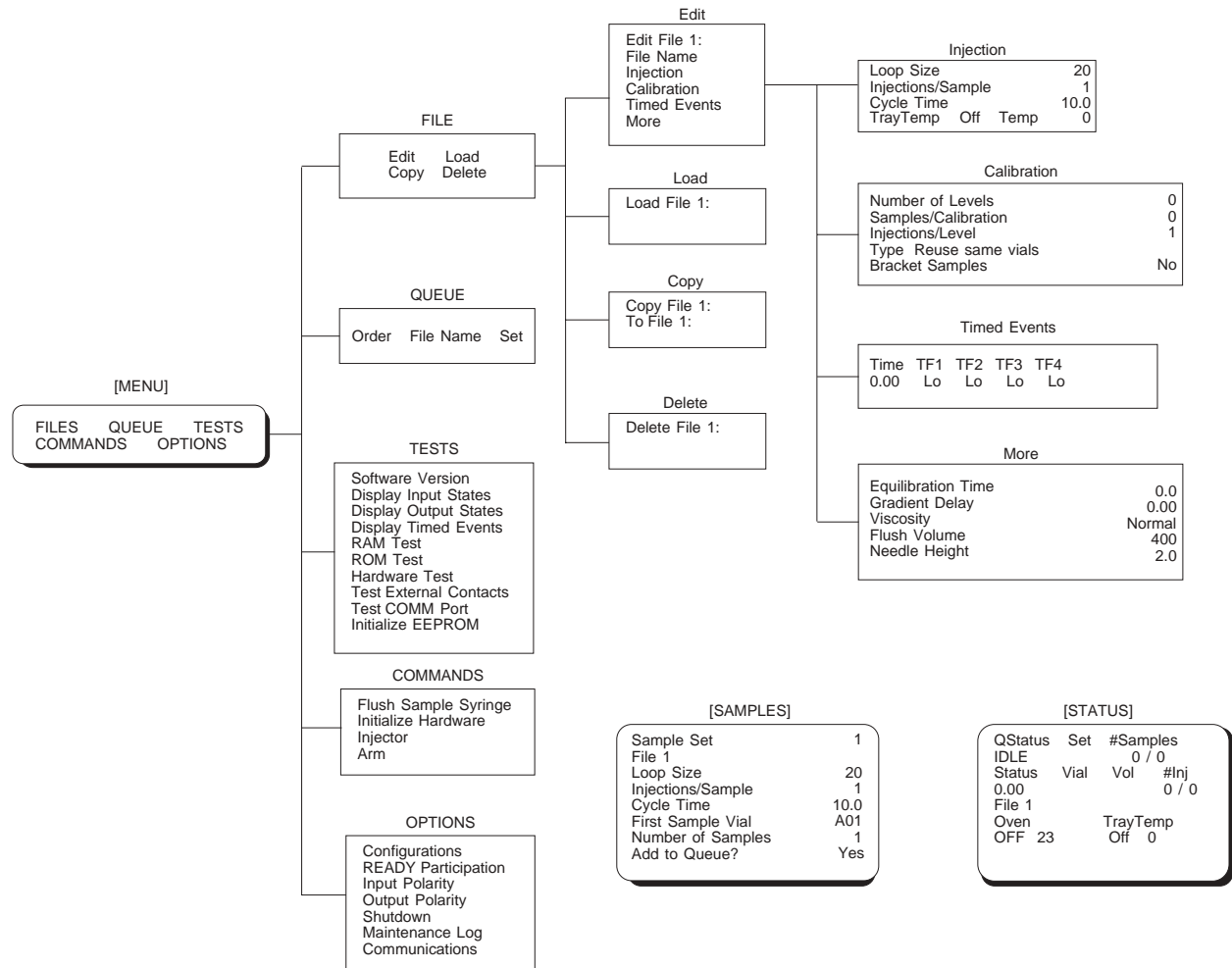
The Menu Reference is an alphabetical listing of each menu field, its definition, and, where appropriate, its allowable and default values.

### **7.1 Menu Trees**

The Menu Tree for your particular instrument is a useful reference guide to learning your way around your autosampler. You may wish to keep it open while you learn where each display is located in the overall menu structure and as you work through the operating instructions in Sections 3 and 4.

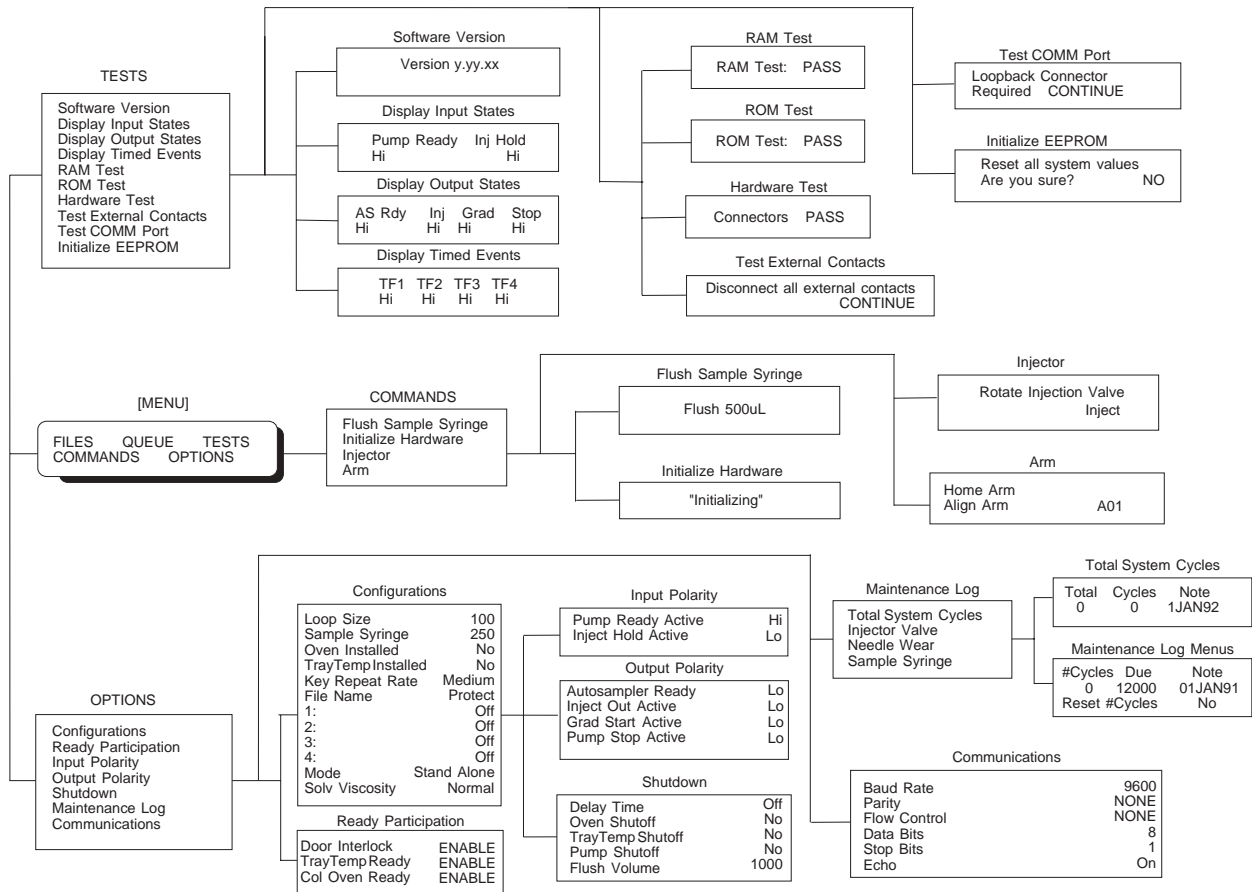
# SS-1 Menus

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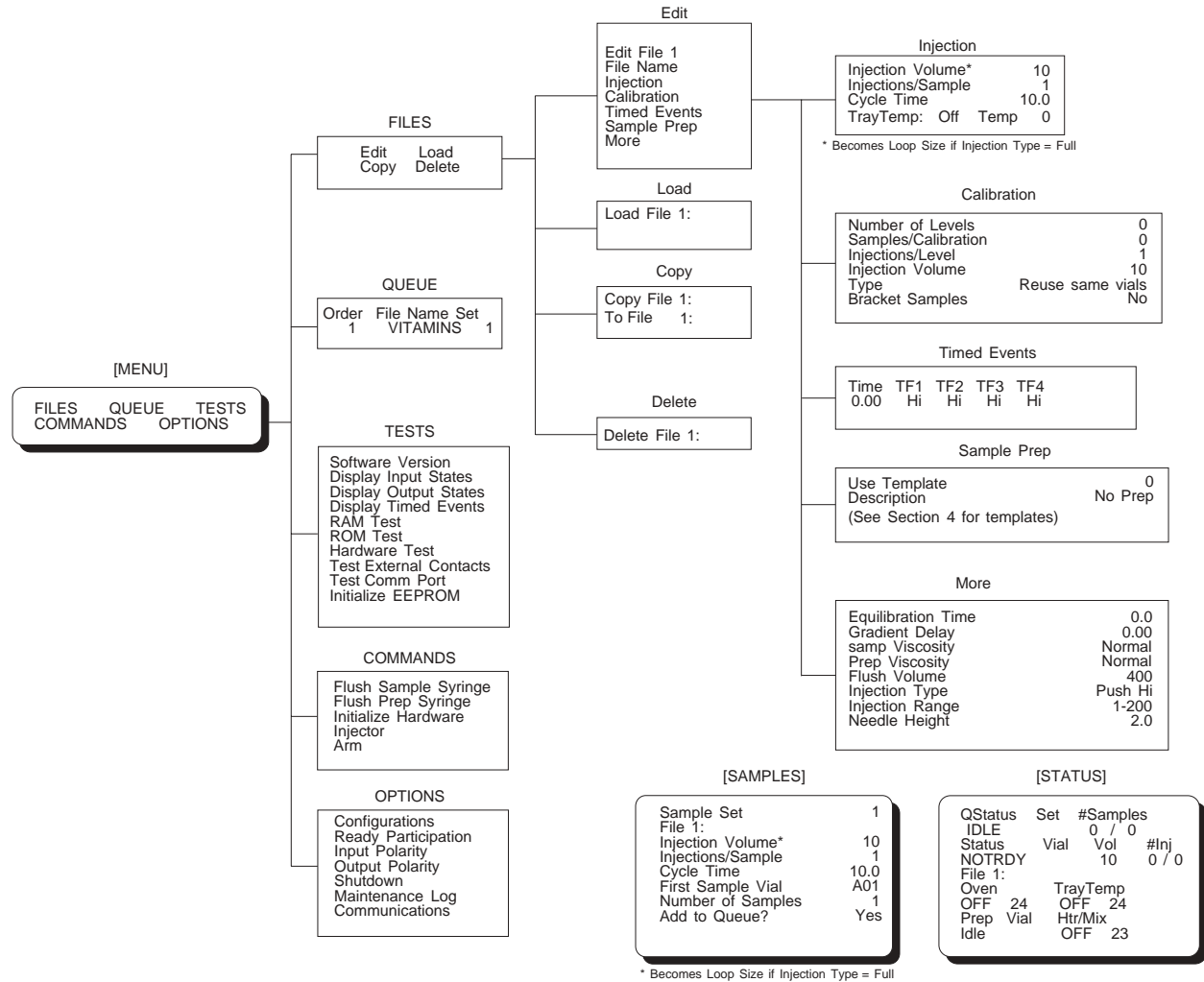
# SS-1 Menus

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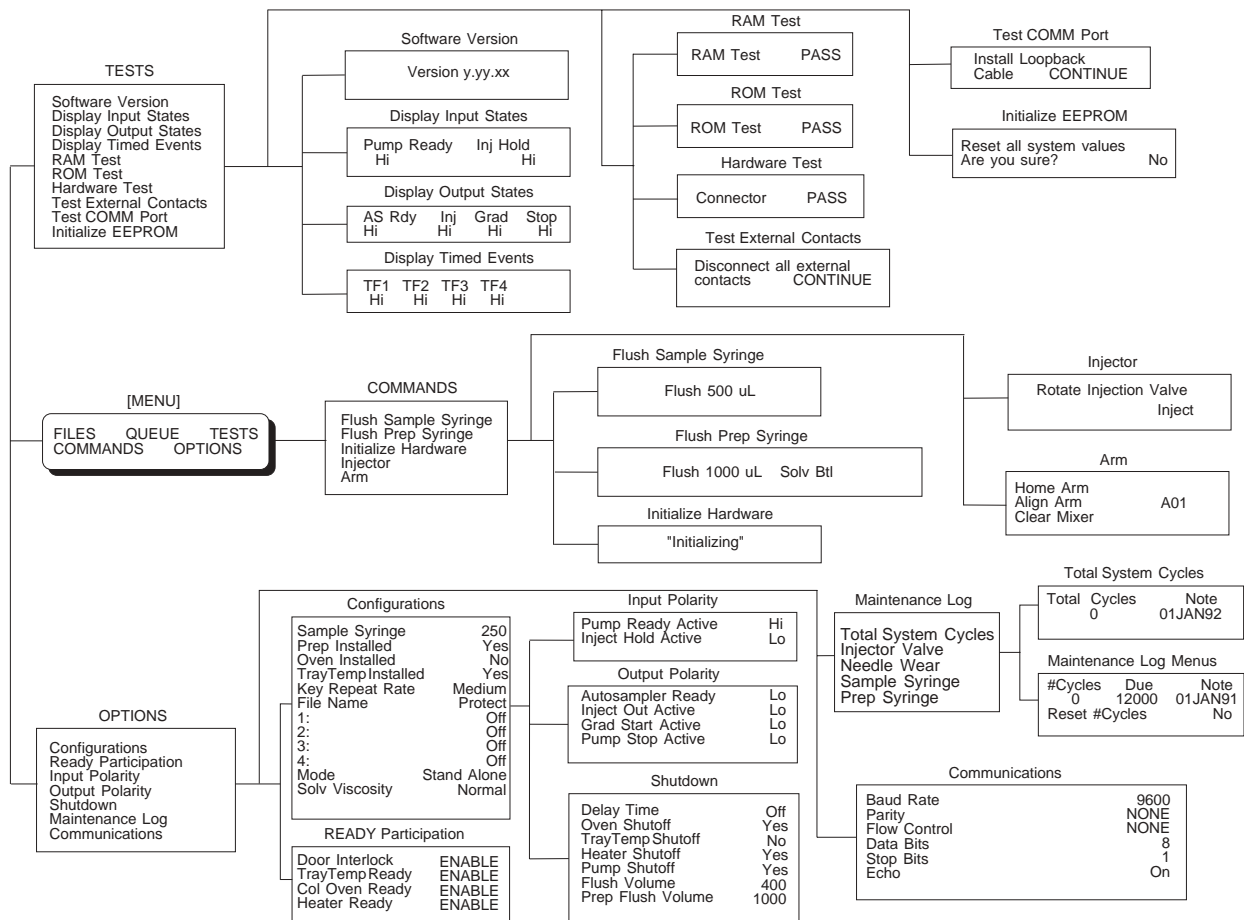
# SS-2 Menus

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# SS-2 Menus

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## 7.2 Menu Reference

For quick reference, we have included this alphabetical list of each field, including a short definition, and allowable and default values. For a more detailed explanation of the functions of your autosampler, refer to Section 3, *Basic Operations*, and Section 4, *Advanced Operations*.

Some fields appear only if a specific option is installed; we have indicated in the definition when this is the case.

### #Cycles

This field appears in all of the Maintenance Log menus and counts the number of activations (injections) for the entire autosampler or for a specific functional part. For the Total System Cycles and Injector Valve Menus, this value represents the number of injections. For the flush and solvent valves, this field is incremented each time the valve changes direction. For the sample and preparation (prep) syringes, this field is incremented each time a syringe stroke is completed.

### #Inj

In the Status Screen, this field contains the number of the current injection over the total number of injections for the vial currently being processed, expressed as a fraction: current injection number/total injections per vial.

### #Samples

In the Status Screen, this field contains the number of the sample currently being processed over the total number of samples in the set, expressed as a fraction: current sample/total samples per set.

### Add $\mu$ L to sample

A sample preparation command that delivers a specified volume from the prep syringe to the specified target vial (Sample, Sam + 1, Sam + 2, Sam + 3). The volume default value is different for each template. The target default value is the sample vial (Sam). See Section 4 for more information.

### Add to Queue

This Sample Menu field allows you to add (Yes/No) a specified sample set to the sample queue. The default value is Yes.

### Align Arm

This Commands Menu routine moves the arm to the vial position specified (A01–C40). It is primarily for service use only. For more information, refer to Section 5.

### Arm

From the Commands Menu, this choice accesses the built-in commands used to manually align or move the arm to its home position.

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<b>AS Rdy</b>	In the Display Outputs Menu, this indicates the level of the Autosampler Ready output signal (Hi/Lo). The default setting is Lo. You can't change the signal level from this menu; however, you can change its polarity from the Output Polarity Menu.
<b>Autosampler Ready</b>	In the Output Polarity Menu under OPTIONS, this field lets you specify the active level (polarity) of the Autosampler Ready output (Hi/Lo).
<b>Bracket Samples</b>	In the Calibration Menu, this prompt asks (Yes/No) if you want to reinject the calibration vials (recalibrate) after all of the sample vials in the sample set have been injected. The default value is No.
<b>Calib Vial 1 (of X)</b>	In the Calibration Menu, this field specifies the position (A01–C40) of the first calibration vial within the sample queue. X (1–9) is the number of calibration levels in the run. The vial position default value is A01; the calibration levels (X) default value is 1.
<b>Calibration</b>	This Edit Menu choice accesses the menu used to enter the calibration parameters.
<b>Col Oven</b>	In the Injection Menu, this field sets the column oven to turn on or off once the file is loaded. The default value is Off. This field also indicates the column temperature (15–80 °C). The default setting is Off.
<b>Col Oven Ready</b>	When this function is enabled in the Ready Participation Menu, the autosampler will not indicate a "ready" status until the column oven has reached its set temperature. Pressing the [+] and [-] keys lets you enable or disable this function.
<b>Commands</b>	The Main Menu choice that displays the Commands Menu used to access the autosampler's built-in procedures that let you manually flush the syringe(s), initialize the hardware, control the injector valve and XYZ arm, and reset the software.
<b>Configurations</b>	Displays the menu used to set or change the instrument's configuration, key repeat rate (cursor speed), and to protect files from being edited.
<b>Connector Test</b>	This Hardware Test diagnostic verifies all of the cable connections and instrument sensors. See Section 9 for more information on running this test.
<b>Copy</b>	Accesses the menu that lets you copy files.
<b>Copy File #</b>	Specifies the number of the file (1–4) to copy from.

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<b>Cycle Time</b>	Specifies the time (0–655 minutes) between injections (run time). The default setting is 10.0 minutes.
<b>Delay Time</b>	The time entered into the Shutdown Menu that the autosampler should wait before initiating the shutdown sequence. The default setting is off.
<b>Delete</b>	Accesses the menu used to delete files.
<b>Delete File #</b>	Specifies the number of the file to be deleted (1–4). Pressing [ENTER] restores the current file parameters to their default settings and erases the user-specified file name.
<b>Description</b>	Specifies the name of the sample preparation template in use (No Prep, Dilution, DABS-CL, Rainbow, and Linear).  <i>NOTE: This field appears only if the Sample Preparation Option is installed.</i>
<b>Door Interlock</b>	When enabled, prevents the autosampler from indicating a "ready" status until the door is closed. Pressing the [+] and [–] keys lets you enable or disable this function. The default setting is ENABLE.
<b>Due</b>	This is a 6-character numeric field that appears in each Maintenance Log menu (with the exception of the Total System Cycles Menu) where you enter the number of cycles indicating that maintenance is due. This number varies for each autosampler component. Use the [+] and [–] keys to enter or edit the Due field value. Refer to Section 5 "Maintenance Log" for the default values for this field.
<b>Edit</b>	Displays the menu used to edit/change a file that is not yet been added to the sample queue.
<b>Equilibration Time</b>	This More Menu field lets you specify the time (0–99.9 minutes) required for the system to re-equilibrate after initializing the new HPLC method. The default setting is 0.0.
<b>File #</b>	Lets you select the number of a file to edit.
<b>File Name</b>	A name that you designate for the file that contains your run parameters. File names can be up to eight characters long and may contain only the following characters: A–Z, 0–9, /, -, or blank. Once entered into the Edit Menu, the autosampler automatically copies it to the Samples and Queue Menus, and Status Screen.

**File requires Z Extra vials after each Sample vial**

Is supplied automatically by the sample preparation software and indicates the required number of empty vials (1–3) after each sample vial in a set. If Sam is used as a target vial,  $Z = 0$ ; if Sam and Sam + 1 are used as target vials,  $Z = 1$ ; if Sam, Sam + 1, and Sam + 2 are used as target vials,  $Z = 2$ ; if Sam, Sam + 1, Sam + 2, and Sam + 3 are used as target vials,  $Z = 3$ . The default value differs with each template.

**FILES**

This Main Menu choice provides access to the menus used for editing, loading, copying, and deleting files.

**Fill**

Indicates that the injector valve is in the Fill position. Pressing [+] and [–] and [ENTER] switches the valve between the Fill and Inject positions.

**Flush  $\mu\text{L}$** 

Located in the Commands Menu, this field specifies the volume of a manual flush cycle (100–5000  $\mu\text{L}$ ) for the sample syringe. The default value is 500  $\mu\text{L}$ . Pressing [ENTER] in this field manually flushes the lines, sample loop and syringe(s).

**Flush Prep Syringe**

This Commands Menu choice lets you manually flush the lines and the 2.5 mL prep syringe with a specified volume (1000–5000  $\mu\text{L}$ ) of one of the four external solvents (S-1 to S-4) or your flush solvent (Btl). 1000  $\mu\text{L}$  is the default setting.

**Flush Sample Syringe**

This Commands Menu choice lets you manually flush the lines and sample syringe with a specified volume (0–5000  $\mu\text{L}$ ) using the 250  $\mu\text{L}$  (sample) syringe. 500  $\mu\text{L}$  is the default setting.

**Flush Volume**

*In the More Menu*, this field specifies the desired volume of flush solvent (100–5000  $\mu\text{L}$ ) for the automatic flush cycle that occurs after each injection. The default value is 400  $\mu\text{L}$ .

*In the Shutdown Menu*, this field specifies the desired volume of flush solvent (0–5000  $\mu\text{L}$ ) for the sample-syringe flush that occurs as a part of the shutdown sequence. The default value is 400  $\mu\text{L}$ .

**Grad**

This Display Output States Menu field indicates the level of the Gradient Delay output signal. You can't change the signal level from this menu. The signal must be changed from the Options, Output Polarity, Gradient Start Active field.

**Grad Start Active**

This Output Polarity Menu choice lets you specify the active level (Hi/Lo) of the gradient start output.

<b>Gradient Delay</b>	This More Menu choice lets you Specify the time (0.0–99.99 minutes) required for the mobile phase to travel from the proportion valve to the head of the column. The default is 0.00 minutes (no delay).
<b>Hardware Test</b>	Accesses the Connector Test Menu used to verify that instrument and sensor connections are made. (See also Connector Test.)
<b>Heat</b>	Used in sample preparation to specify a desired heating time (0–99 minutes).
<b>Heater</b>	This sample preparation template field turns the heater/mixer on/off. The default is Off. TEMP, in the same line, specifies the heater temperature (30–100 °C).
<b>Heater Ready</b>	When enabled, this Ready Participation Menu field prevents the autosampler from indicating a "READY" status until the heater/mixer has reached its set temperature. Pressing the [+] and [-] keys lets you enable or disable this function. This field appears only with the Sample Preparation Option installed.
<b>Heater Shutoff</b>	The field in the Shutdown Menu that lets you program the autosampler to automatically turn off the heater/mixer during the Shutdown sequence.
<b>Home Arm</b>	Returns the XYZ arm to its "home" position alongside the rear-left corner of the sample tub.
<b>Htr/Mix</b>	In the sample preparation status line (Prep), this field displays the current state (on/off) of the heater/mixer and its actual (not set) temperature. The temperature is always displayed whether or not the heater/mixer is in use.
<b>Initialize EEPROM</b>	This Tests Menu choice erases the current EEPROM values, restores all file parameters to their default values, and erases all hardware configuration information. (Any installed options must be reset to "YES" in the Configurations Menu.)
<b>Initialize Hardware</b>	Lets you manually reset the autosampler's syringe(s), solvent selector, flush, and injector valves to their initial pre-injection positions.
<b>Inj Hold</b>	This Input Polarity Menu choice indicates the current level of the Inject Hold input signal (Hi/Lo) from your pump.

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<b>Inject</b>	<p><i>In the Display Outputs Menu</i>, Inject indicates the level of the Inject Hold output signal (Hi/Lo).</p> <p><i>In the Injector Menu</i>, Inject indicates that the injector valve is in the Inject position. Pressing [+] and [-] switches the valve between the Fill and Inject positions.</p>
<b>Inject Hold Active</b>	This Input Polarity Menu field lets you specify the active level (polarity) of the Inject Hold input signal (Hi/Lo).
<b>Inject Out Active</b>	This Output Polarity Menu choice lets you specify the active level (polarity) (Hi/Lo) of the Inject Out output signal.
<b>Injection Range</b>	This Injection Menu field indicates the allowable injection volume range (in microliters) as determined by the injection method (Injection Type) selected. The allowable ranges for each injection type are as follows: Push Hi (1–200), Push Lo (0.1–10.0); Pull (1–1500); Full (5–1000). This field does not appear on the SS-1 instrument.
<b>Injection Type</b>	This Injection Menu choice specifies the type of injection method: Push (PushLoop), Pull (pull-loop), and Full (full-loop). The default is Push. The Injection Type value determines the Injection Range. (See also Injection Range.) If this field entry is Full, the Injection Menu's Injection Volume field becomes Loop Size. This field does not appear on the SS-1 instrument.
<b>Injection Volume</b>	Selects the total sample- or calibration-vial injection volume (1–1500 $\mu\text{L}$ ). 10 $\mu\text{L}$ is the default value.
<b>Injections/Level</b>	The Calibration Menu choice allows for multiple injections at a given calibration level. Specifies the number of injections (0–99) for each calibration level. The default value is 1.
<b>Injections/Sample</b>	This Injection Menu choice specifies the number of injections for each sample vial (1–99). A value greater than 1 allows for multiple injections. The default value is 1. A value of 0.0 allows for sample preparation without injection.
<b>Injector</b>	This Commands Menu choice accesses the menu that allows you to manually control the injector's position.
<b>Injector Valve</b>	This Maintenance Log menu tracks the #cycles (activations) of the injector valve since #Cycles field value was last set. An activation occurs each time the valve changes direction. For example, an activation takes place when the valve changes from the Inject to the Fill po-

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sition. When the #Cycles value exceeds the Due field value, the "Maintenance Required" message is displayed when the Maintenance Log Menu is accessed. The default maintenance Due field value is 12,000 (approximately once a year).

**Input Polarity**

This Options Menu choice lets you specify the active signal level (Hi/Lo) for a specific autosampler input. For more information, refer to Section 9.

**Intersperse Cals**

This Calibration Menu field lets you specify a calibration sequence in which calibration vials are placed at regular intervals between sample vials. (See also TYPE.)

**Key Repeat Rate**

This Configuration Menu field is used to adjust the cursor scrolling speed: Fast [8 pulses per second (pps)], Medium (6 pps), and Slow (4 pps). Default: Fast.

**Load**

This Files Menu field accesses the Load Menu for activating (loading) files.

**Load  $\mu$ L Solv S-1 (2,3,4)**

A command used in sample preparation that initializes the solvent syringes, flushes the sample lines with the specified solvent (S1–S4), and loads the holding loop with the specified volume (0–1500  $\mu$ L). The volume default is different for each template; the solvent default is S1.

**Load File**

Copies the edit file into the run file, which activates any options (oven, tray temperature control, heater) set in the Edit Menu.

**Loop Size**

A field in the Injection Menu where you can specify your sample loop size fixed-loop injection. Allowable values are: 5, 10, 20, 50, 100, 200, 500, and 1000  $\mu$ L. For an SS-2, if the More Menu's Injection Type is set to Full, this field appears on the Injection Menu in place of Injection Volume. For an SS-1, this field appears in the Injection Menu in place of the Injection Volume field.

**Maintenance Log**

This Options Menu item is used to access autosampler maintenance status, use, and service information.

**Mix**

This sample preparation template command is used to vortex-mix, heat, and then remix for a specified period of time (in minutes).

**More**

This Edit Menu choice accesses the menu used to set the equilibration time, gradient delay, sample viscosity information, flush volume, injection method, and needle height.

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<b>Needle Wear</b>	This Maintenance Log menu tracks the #cycles (vial pierces) of the sample needle since #Cycles field value was last set. When the #Cycles value exceeds the Due field value, the "Maintenance Required" message is displayed when the Maintenance Log Menu is accessed. The default maintenance Due field value is 10,000 vial pierces (approximately six months).
<b>Normal</b>	This More Menu Viscosity choice indicates a standard syringe speed appropriate for most sample viscosities.
<b>Note</b>	This 6-character alphanumeric field appears in all Maintenance Log Menus and is programmable using the [+] / [-] keys. It is designed to hold the date of the last instrument maintenance, but may also be used to hold any service-related information on the autosampler or one of its parts.
<b>Number of Levels</b>	This Calibration Menu field specifies the number of calibration (concentration) levels (0–9) in the calibration series for a given sample set. A value of 0 indicates that there are no calibration vials in the sample set. The default is 0.
<b>Number of Samples</b>	This Samples Menu field specifies the total number of sample vials (1–120) in the sample set. The default is 1.  <i>NOTE: With the Tray Temperature Control Option installed, the total number of vials is 105.</i>
<b>OPTIONS</b>	This Main Menu field accesses the Options Menu for setting/changing the instrument configuration, for performing error recovery and instrument shut-down, and for monitoring the maintenance log.
<b>Order</b>	This Queue Menu field is used to edit the processing order of sample sets within the sample queue. You can reorder any sets that are not currently running. The default order value is 1 (first, except when a priority set is added to the queue.)
<b>Output Polarity</b>	This Options Menu choice lets you specify the active signal level (Hi/Lo) for a specific autosampler output. For more information, refer to Section 9.
<b>Overlap Enabled</b>	This last line in each sample preparation template allows sample preparation for the next sample to begin while the current sample is still running. The default is Yes. No delays sample preparation of the subsequent sample until the current sample's run is complete.

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<b>P</b>	This Queue Menu Order field entry indicates that the set has been assigned a priority status (from in the Sample Menu). See Section 3 for more information on managing priority sample sets.
<b>PickLG <math>\mu</math>L Sample</b>	This sample preparation command causes the prep syringe to withdraw the specified volume from the specified source vial (Sample, Sam + 1, Sam + 2, Sam + 3, Reag-A, Reag-B, or Reag-C). Volume and source-vial default values are different for each template.
<b>Pickup <math>\mu</math>L Sample</b>	This sample preparation command causes the sample syringe to withdraw the specified volume from the specified source vial (Sample, Sam + 1, Sam + 2, Sam + 3, Reag-A, Reag-B, or Reag-C). The volume and source vial default values are different for each template.
<b>Prep</b>	This Status Screen field displays the current sample preparation template step in progress.
<b>Prep Flush Volume</b>	<p>This Shutdown Menu field specifies the desired volume of flush solvent (0–5000 <math>\mu</math>L) for the prep-syringe flush that occurs as a part of the shutdown sequence. The default value is 1000 <math>\mu</math>L.</p> <p><i>In the Maintenance Log Menu, this field tracks the #cycles (prep syringe strokes) since #Cycles field value was last set. When the #Cycles value exceeds the Due field value, the "Maintenance Required" message is displayed when the Maintenance Log Menu is accessed. The default Maintenance Due field value is 24,000 strokes (approximately once a year).</i></p>
<b>Prep Installed</b>	This Configurations Menu field indicates whether or not (Yes/No) the Sample Preparation Option is installed. The default setting is No.
<b>Prep Viscosity</b>	This More Menu field indicates the viscosity of the sample preparation resource (sample/reagent) to be picked up in the PickLg step(s) by the 2.5 mL prep syringe. Allowable values are Normal, Medium, and Viscous. This value affects the overall syringe rate.
<b>Protect</b>	This Configurations field allows you to restrict editing of selected files as named in the Files Menu. If Protect is turned on (Yes) for a particular file, you will be unable to edit or view a particular file until Protect is turned off (No). The default is No.
<b>Pump Ready Active</b>	This Options, Input Polarity Menu field indicates the active level (polarity) of the pump ready input signal (Hi/Lo).

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<b>Pump Shutoff</b>	This Shutdown Menu field lets you program the autosampler to automatically turn off the pump during the Shutdown sequence.
<b>Pump Stop Active</b>	The Output Polarity Menu field lets you specify the active level (polarity) (Hi/Lo) of the Pump Stop output signal.
<b>Push Hi</b>	One of the PushLoop injection types in the Injection or Calibration Menus. This choice corresponds to an injection range of 1–200 $\mu\text{L}$ .
<b>Push Lo</b>	One of the PushLoop injection types in the Injection or Calibration Menus. This choice corresponds to an injection range of 0.1–10 $\mu\text{L}$ .
<b>QStatus</b>	This first status screen field lists the current state of the sample queue (RUN, IDLE, EQUIL, etc.). Refer to Section 3 for a description of the various autosampler states.
<b>QUEUE</b>	This Main Menu entry displays and allows you to edit the items in the sample queue. (See also Order.)
<b>RAM Test</b>	This Tests Menu choice initiates the autosampler's RAM test to verify proper operation.
<b>READY Participation</b>	This Options Menu choice lets you define the "Autosampler Ready" requirements and set the input- and output-signal polarities.
<b>Reagent A (B,C)</b>	These sample preparation template fields specify the reagent vial position(s) (A01–C40, without tray temperature control; A01–C35, with tray temperature control) required by the sample preparation method.
<b>Remove <u>  </u><math>\mu\text{L}</math> To Waste At <u>  </u>mm Height</b>	This sample preparation command withdraws a specified volume (0–1500 $\mu\text{L}$ ) from a specified source vial (Sample, Sam + 1, Sam + 2, Sam + 3) at a specified distance (height 0–20 mm) between the needle tip and the bottom of the source vial and delivers it to waste. This function enables you to select a given layer of sample for extraction. Volume and source defaults are different between Templates 3 and 4. (See Section 4.)
<b>Reset #Cycles</b>	This Maintenance Log Menu field lets you reset the #Cycles counter for each autosampler part. To reset the counter to 0 after maintenance is complete, press the [+]/[-] keys to select YES, and press [ENTER]. The confirmation message ** Counter Reset ** is displayed.

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<b>Reuse same vials</b>	This Calibration Menu choice lets you select the calibration sequence type that programs the autosampler to use the same set(s) of calibration vials throughout the run. The autosampler will automatically (Yes) reinject from the same calibration vials according to the calibration interval specified. (See also Type and Samples/Calibration.)
<b>ROM Test</b>	This Tests Menu choice initiates the autosampler's ROM test to verify proper operation.
<b>Rotate Injection Valve</b>	This Commands, Injector Menu choice lets you manually (outside of a run) rotate the injection valve between the Inject and Fill positions.
<b>S-1 (2,3,4)</b>	Indicates one of the four external solvents used in sample preparation.
<b>Sample Prep</b>	This Files, Edit Menu choice accesses the sample preparation templates (menus) and appears only if the Sample Preparation Option is installed.
<b>Sample Set</b>	This Samples Menu field indicates the sample set number (1–39) in the sample queue. A set is a group of sample vials and associated calibration vials.
<b>Sample Syringe</b>	This Maintenance Log Menu tracks the #cycles (sample syringe strokes) since the #Cycles field value was last set. When the #Cycles value exceeds the Due field value, the "Maintenance Required" message is displayed when the Maintenance Log Menu is accessed. The default Maintenance Due field value is 24,000 strokes (approximately once a year).
<b>Sample Syringe Size</b>	This Configurations Menu field specifies the size of the sample (small) syringe (250, 500, 1000, 2500, and 5000 $\mu\text{L}$ ). The default is 250 $\mu\text{L}$ .
<b>SAMPLES</b>	Is the autosampler's "special" key that displays the Samples Menu, which lets you define the resources and file for each sample set and add sample sets to the sample queue. For more information, see Section 1.
<b>Samples/Calibration</b>	This Calibration Menu field specifies the number of sample vials (0–99) between calibration vials (the calibration interval). A value of 0 indicates that there are no calibration vials in the set. The default setting is 0.
<b>Set</b>	<p><i>In the Queue Menu</i>, Set indicates the sample set assigned to the file in the same line.</p> <p><i>In the Status Menu</i>, Set indicates the current sample set being processed.</p>

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<b>Shutdown</b>	This Options Menu choice lets you specify the power-down sequence that turns off the temperature zones, zeroes the detector, turns off the pump, and flushes the autosampler's lines before putting the autosampler in the Idle mode.
<b>Software Version</b>	This Tests Menu field displays the current software version installed in the instrument.
<b>Solv Viscosity</b>	This Configurations Menu field allows you to adjust the prep syringe speed (draw rate) to accommodate the viscosity of your solvent. Range: Normal, Medium, Viscous. Default value: Normal.
<b>Status</b>	This key accesses the Status Menu that displays the queue, injection, temperature zones, and sample preparation status information.
<b>Temp</b>	This Injection Menu field lets you specify the column oven temperature (15–80 °C) and the tray temperature (0–60 °C), if installed.
<b>Test COMM Port</b>	This Tests Menu choice lets you perform a read/write test to verify proper operation of the communications (COMM) port. A loop-back cable is required for this test.
<b>Test External Contacts</b>	This Tests Menu choice activates all of the external contacts and tests them for proper activation. This menu displays the current levels (Hi/Lo) of the autosampler's input and output signals.
<b>TESTS</b>	This Main Menu choice accesses the autosampler's built-in diagnostic routines.
<b>TF1 (2,3,4)</b>	<p><i>In the Timed Events Menu</i>, this field indicates one of the four open-collector outputs capable of handling 500 mA at 28 V. The default state is Lo ("open"). This output can be set to Hi or Lo to activate an external device such as a fraction collector or column-switching device.</p> <p><i>In the Display Timed Events States</i>, these fields display the current levels (Hi/Lo) of the autosampler's time-function output signals.</p>
<b>Time</b>	In the Timed Event Menu, this field is used to specify a relay activation time (0.01–655.0 minutes).
<b>Timed Events</b>	This Edit Menu choice accesses the menu used to program external outputs. [See also TF1(2,3,4).]

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<b>To File #</b>	Located in the Copy Menu, this field lets you specify the destination file number (1–4) for copying a file. The default is 1.
<b>Total Cycles</b>	This unprogrammable Maintenance Log Menu field displays the number of injections for the total life of the autosampler. This value is set at the factory and cannot be reset.
<b>Total System Cycles</b>	This unprogrammable Maintenance Log Menu field contains instrument usage information.
<b>TrayTemp</b>	<p><i>In the Edit Menu</i>, this field lets you turn on/off the sample tray heater/cooler. The default setting is No. This field also indicates the tray temperature (0–60 °C).</p> <p><i>In the Status Screen</i>, this field displays the sample tray heater/cooler's current state (On/Off) and its actual (not set) temperature. The actual temperature is displayed even if the tray heater/cooler is turned off. This field appears only if the Tray Temperature Control Option is installed.</p> <p><i>NOTE: This field appears only if the Tray Temperature Control Option is installed.</i></p>
<b>TrayTemp Installed</b>	Indicates whether or not the Tray Temperature Control Option is installed (Yes/No). The default setting is No.
<b>TrayTemp Shutoff</b>	The field in the Shutdown Menu that lets you program the autosampler to automatically turn off the heater/cooler during the Shutdown sequence. This field appears only if the Tray Temperature Control Option is installed. The default is Yes.
<b>Type</b>	This Calibration Menu field lets you specify the type of calibration sequence: Reuse same vials or intersperse calibration vials (Intersperse Cals).
<b>Use Template</b>	Used to select the number of a desired sample preparation template file: 0:No Prep, 1:Dilution, 2:DABS-CL, 3:Rainbow, and 4:Linear. The default value is 0: No Prep.
<b>Vial</b>	<p><i>In the Injection Status Screen (Status)</i>, the vial field specifies the injection-vial position and vial type (A01-S). The vial position falls in the A01–C40 range without the Tray Temperature Control Option installed and in the A01–C35 range with the Tray Temperature Control Option installed. The vial type is either S (sample) or C (calibration vial).</p> <p><i>In the sample preparation Status Screen (Prep)</i>, this field displays the vial currently undergoing sample preparation.</p>

<b>Viscosity</b>	This Edit, More Menu choice is used to specify the sample viscosity (Normal, Medium, or Viscous), which affects the syringe drawrate.
<b>Vol</b>	This Status Screen field indicates the injection volume for the active vial.
<b>Wait for _ minutes</b>	This sample preparation command returns the vial to the sample tray and pauses all sample preparation activity for the specified period of time (0–99.9 min). The default value is 0.0 minutes.



## **Section 8. Troubleshooting**

This section provides information to help you troubleshoot possible autosampler problems. We have divided it into five sections:

- a brief theory of operation
- a troubleshooting guide to general chromatographic problems
- an autosampler-specific troubleshooting table that lists symptoms, possible problems, remedies
- a list of possible error messages, and
- a description of the autosampler's diagnostic tests.

If, after reviewing the troubleshooting table and diagnostics included in this section, you would like additional assistance in troubleshooting a particular problem, don't hesitate to contact BAS Service (1-800-845-4246).

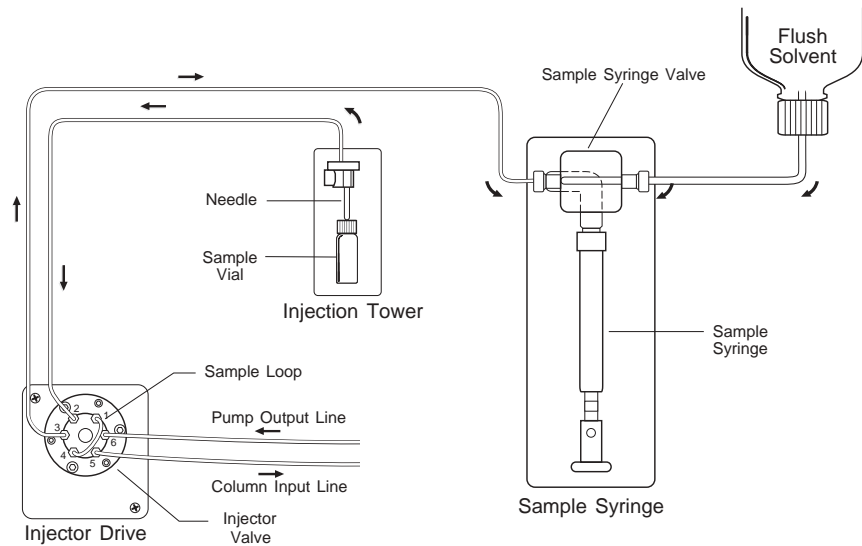
### **8.1 Theory of Operation**

#### **BASIC AUTOSAMPLER**

The autosampler consists of a number of subsystems. It is important for you to be familiar with how these subsystems contribute to the instrument's operation in order for you to be able to trace a problem or symptom to its probable cause.

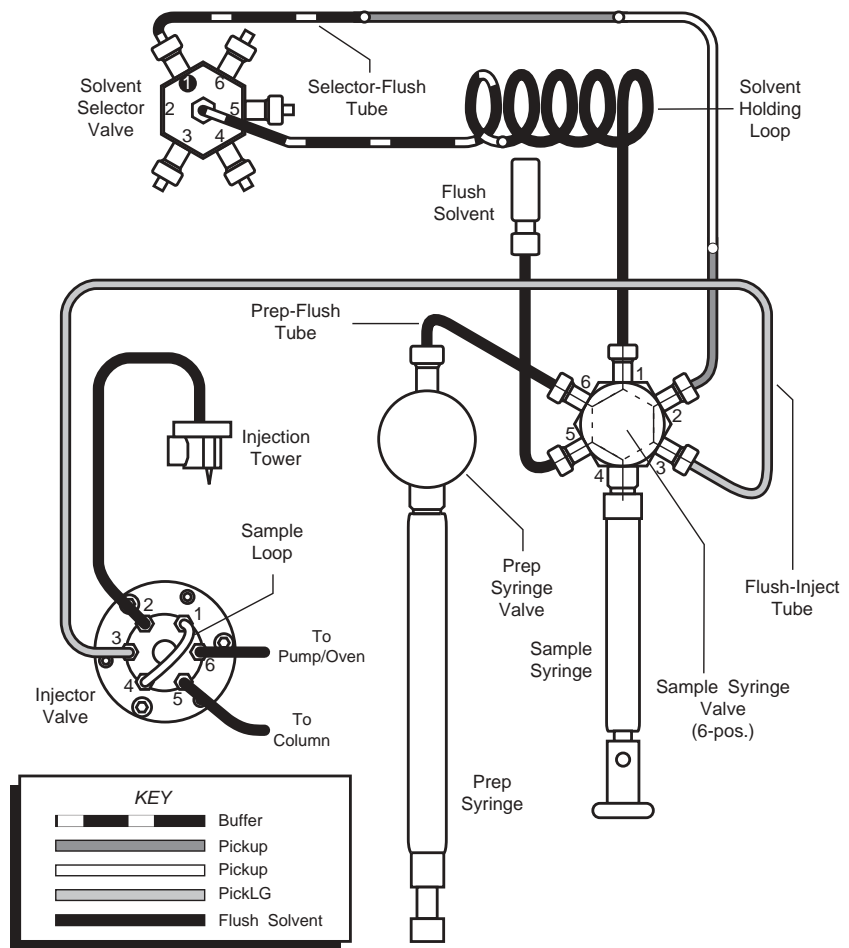
The basic autosampler consists of the following components: 1) flush valve, 2) small (250  $\mu$ L) syringe, 3) injector valve, 4) sample loop, 5) needle and hook, 6) flush funnel, 7) pump output line, 8) column input line, and 9) sample vial. Figure 8.1 illustrates the autosampler's major components without the Sample Preparation Option. Figure 8.2 illustrates the major components with sample preparation installed.

Figure 8.1. The autosampler's major components without the Sample Preparation Option.



Vials filled with samples to be analyzed are loaded into three trays that hold up to 40 vials each (35 with the Tray Temperature Control Option installed). During operation, a mechanical arm (the XYZ arm) locates a sample vial. The hook on the end of the arm moves under the selected vial and lifts it from the sample tray. The XYZ arm transports the vial to the sample tower and the sample needle for sample injection. The injection valve switches between the Inject and Fill positions (Figure 8.3) as necessary to draw sample or solvent into the autosampler lines and to inject sample onto the column. All of the Sample Sentinel autosamplers perform full-loop injection. The SS-2 offers both pull-loop and PushLoop injection. These injection routines are described here.

Figure 8.2. The autosampler's major components with the Sample Preparation Option installed.



**Full-loop injection**

In full-loop injection, an air bubble is drawn into the needle to isolate the incoming sample slug from the flush solvent already present in the lines (Figure 8.4). With the injector valve in the FILL position, the syringe retracts to pull the sample from the vial into the sample loop. The injector valve rotates to the INJECT position, and the pump pushes the sample from the sample loop onto the column.

**Pull-loop injection**

The pull-loop injection routine is useful when you have a limited amount of sample. Only the desired sample amount is pulled from the vial into the sample loop. Specifically, an air bubble is drawn into the needle to isolate the incoming sample slug from the flush solvent already present in the lines (Figure 8.5). With the injector valve in the FILL position, sample is drawn into the needle. The sample vial is lowered from the end of the needle and sufficient air drawn into the needle to pull the sample slug into the sample loop. The injector valve then rotates to the INJECT position and sample is expelled onto the column.

Figure 8.3. The injector valve's inject and fill positions.

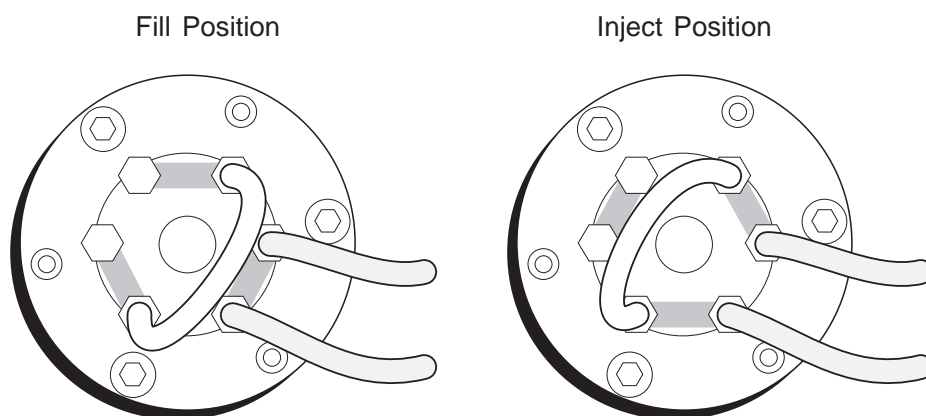


Figure 8.4. Full-loop injection just prior to sample injection.

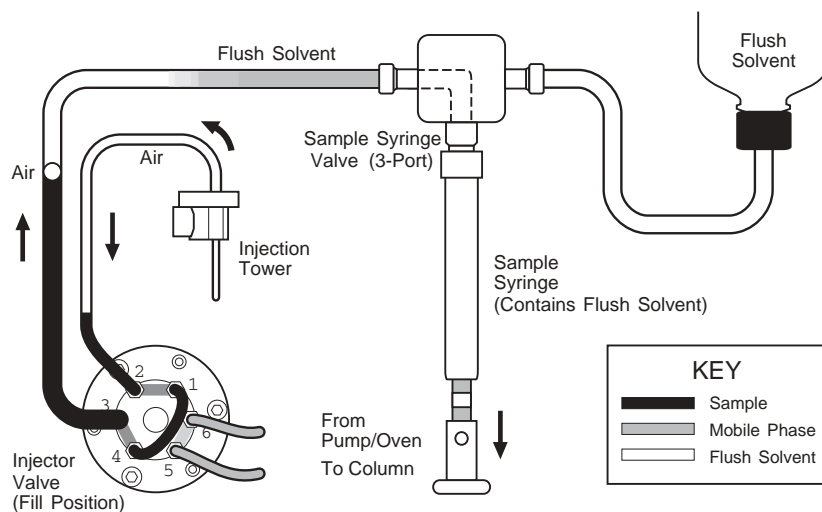
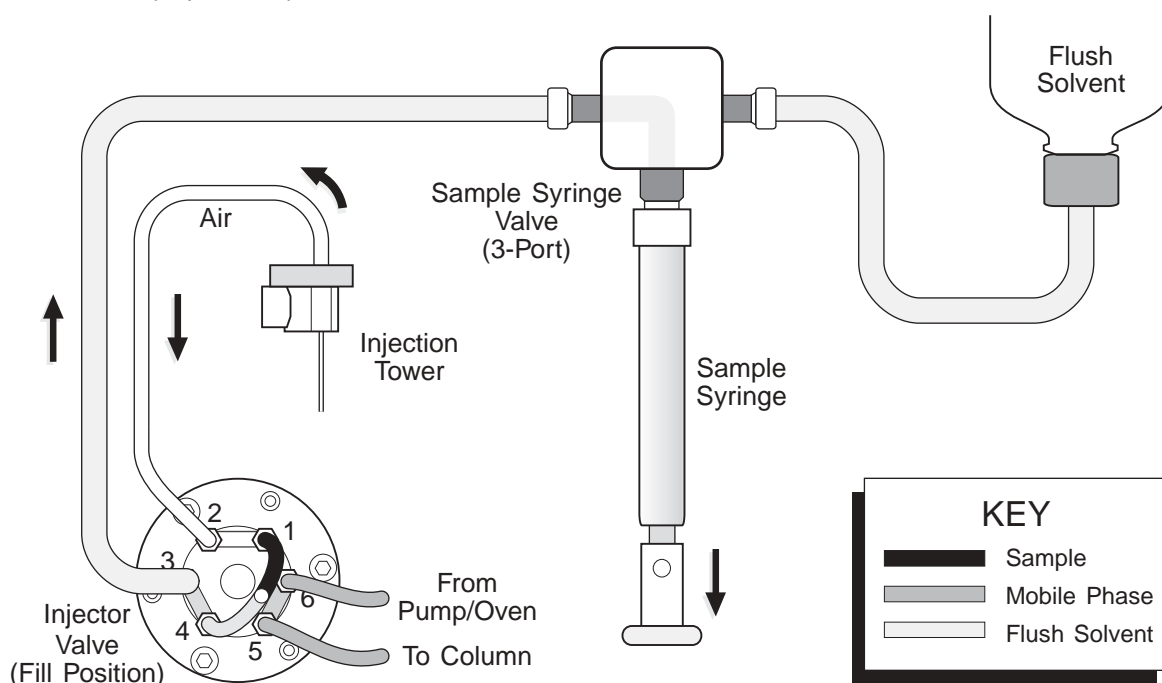


Figure 8.5. Pull-loop injection sequence.



### PushLoop Injection

The concept behind PushLoop injection is that it is more accurate to push a column of fluid than it is to pull it. Pulling liquids through a small tube can cause "cavitation" within the tubing. Cavitation is the outgassing of dissolved gases caused by reducing the pressure on a column of liquid. This forms bubbles within the column of liquid, resulting in inaccuracies and nonreproducible volumes.

With PushLoop injection, sample volume is separated from the previous flush solvent by an air bubble. With the valve in the Inject position, the sample slug is pulled up to and past the injection valve. After compensating for the syringe backlash, the valve is switched to the Fill position and the desired volume of sample is pushed into the sample loop. In PushLoop injection, sample volumes are "pushed" into the sample loop from solution directly adjacent to the loop. The sample solution is essentially at the "bottom" of the column of liquid. Any residue from the previous solvent has been minimized. The starting boundary of the delivered sample slug is well defined by the valve. There is no uncertainty about the air bubble or solvent droplets that may affect accuracy or reproducibility. Finally, the injector valve is then switched back to the Inject position, and sample is expelled from the sample loop onto the column. Figures 8.6, 8.7, and 8.8 illustrate this routine.

Figure 8.6. PushLoop injection. (Step 1: The sample slug is pulled up to and past the injection valve.)

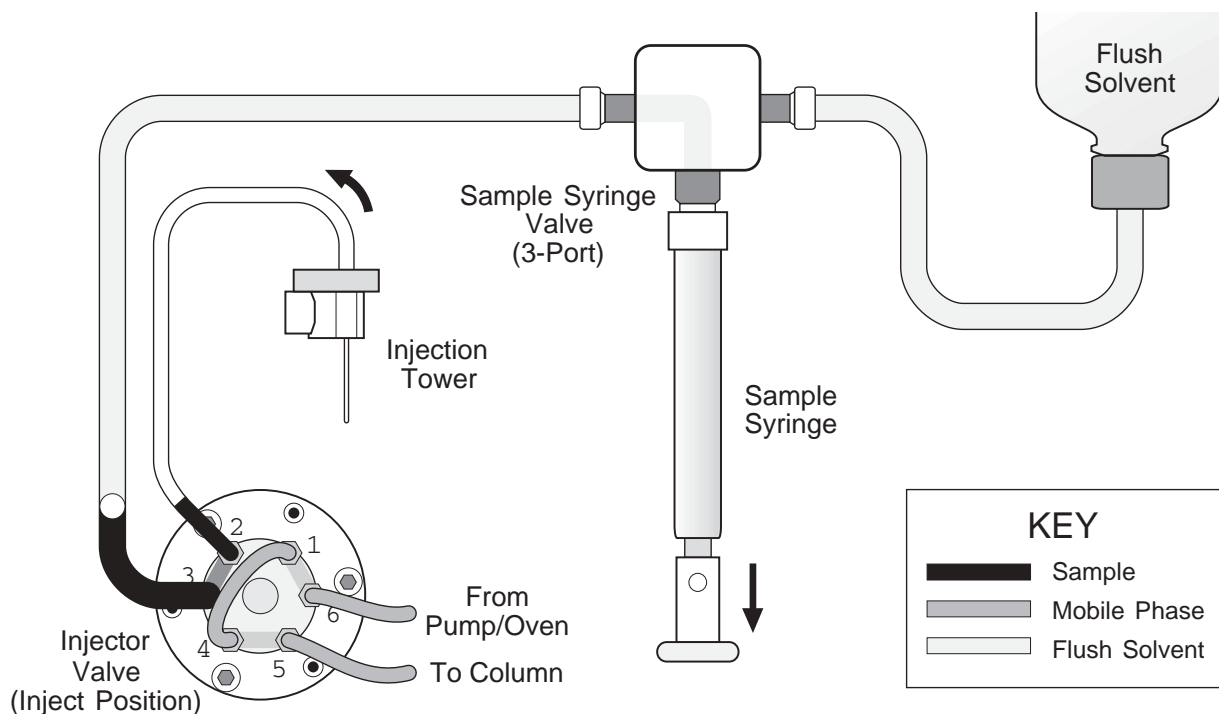
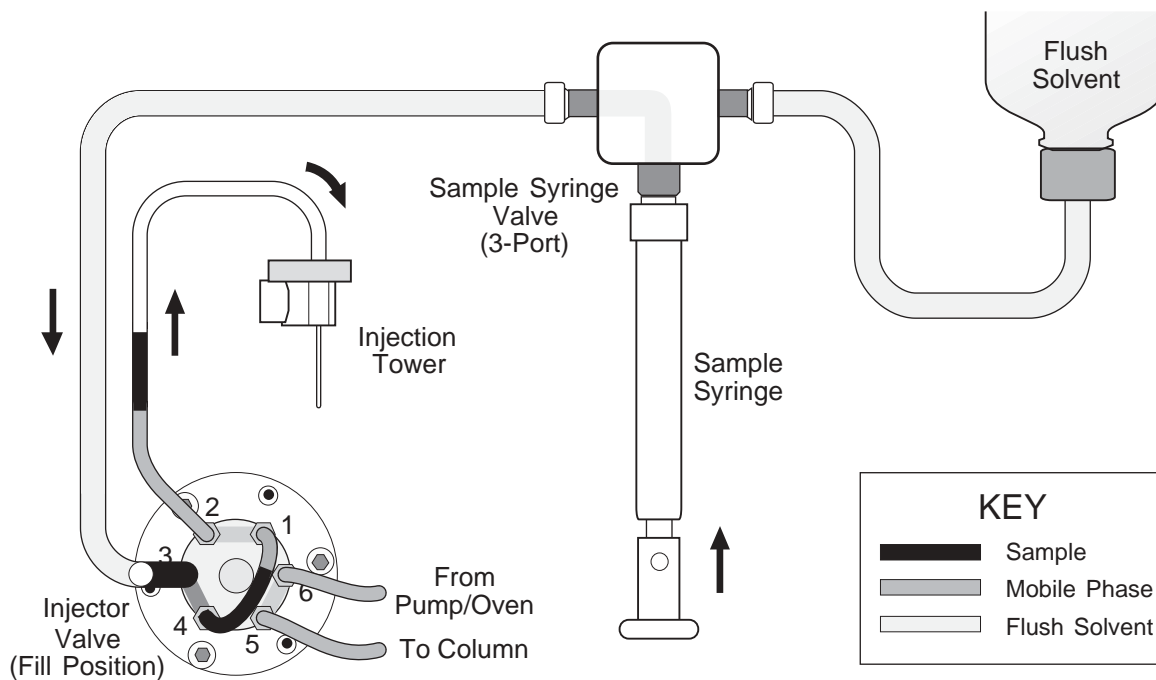


Figure 8.7. PushLoop injection. (Step 2: The injection valve is switched to the Fill position, and the desired volume of sample is pushed into the sample loop.)





## 8.2 General Chromatography Troubleshooting

This section focuses on general chromatographic problems and is designed to help you define the particular instrument or condition causing the problem. Once you have ascertained the cause, use the ordered outline to fix the problem. Each of these problems can be detected by monitoring the chromatography running conditions or by observing the chromatogram. For more detailed chromatographic troubleshooting, refer to any HPLC troubleshooting reference book or call your local sales or service representative.

<b>SYMPTOM</b>	<b>CAUSE/REMEDY</b>
1. No flow.	a) Check mobile phase connections b) Check for leaks c) Check pump troubleshooting guide
2. High backpressure.	a) Check flowrate and system/column specifications b) Check for tubing or column blockage c) Check pump troubleshooting guide
3. Unstable baseline or drift.	a) System/column not equilibrated; allow more time b) Check detector troubleshooting guide c) Check pump troubleshooting guide
4. Baseline noise.	a) Check for air bubbles in system, degas solvents b) Check for system/solvent contamination c) Check pump troubleshooting guide
5. No peaks.	a) Check detector and data system connections b) Check autosampler troubleshooting guide c) Check sample retention with chromatographic conditions
6. Contaminating/ghost peaks.	a) Clean system and column b) Check autosampler troubleshooting guide c) Check pump troubleshooting guide
7. Poor peak shape.	a) Check system for leaks b) Check fittings and tubing lengths c) Check column performance d) Check autosampler troubleshooting guide e) Check pump troubleshooting guide f) Check detector troubleshooting guide

<b>SYMPTOM</b>	<b>CAUSE/REMEDY</b>
8. Poor retention-time reproducibility.	a) Check system for leaks and bubbles b) System/column not equilibrated, allow more time c) Check column performance d) Check pump troubleshooting guide e) Check autosampler troubleshooting guide
9. Poor peak area reproducibility.	a) Check column performance b) Check autosampler troubleshooting guide
10. Non-integrated or too many peaks.	a) Check integrator or data system troubleshooting guide
11. No instrument control.	a) Check cable connections b) Check system configuration c) Check individual instrument troubleshooting guide

### 8.3 Chromatographic Problems

SYMPTOM	POSSIBLE PROBLEM	REMEDY
1. Baseline drift when the injector valve changes from INJECT to FILL.	a) Lack of thermal stability in the column, detector, or mobile phase.	a) Eliminate drafts around the column. Wait until the column has reached thermal equilibrium. If the problem persists, contact your local BAS service representative.
2. Shifting retention times.	a) Temperature variations in the lab. b) Column-heater problems.	a) Stabilize temperature. b) Call your BAS service representative.
3. Sensitivity changes.	a) Inconsistent sample injection volumes.  b) Leaks between the injector valve and the column causing inconsistent sample-volume injection. c) Air bubbles aspirated into the lines along with sample.  d) Incomplete sample mixing during sample preparation.  e) Sample adsorption to the walls of the injector valve or sample loop. f) Sample incompatible with the mobile phase. g) Sample insoluble in solvent.  h) Air in syringe/sample lines.	a) Check for a partially blocked injector valve and observe the pressure. If the problem persists, replace the injection valve (refer to Section 5). b) Tighten/remake fitting.  c) Replenish/prepare fresh sample and repeat the injection. c') Slow down syringe speed. d) Check mix time (Sample Preparation Option only). Thaw frozen samples completely prior to sample preparation to prevent sample "layering." e) Change the mobile phase concentration.  f) Change solvent, use mobile phase if possible. g) Be sure sample solvent is lower elution strength than mobile phase. h) Flush sample lines. Use degassed solvent.
4. Irregular peak shapes.	a) The volume of sample injected exceeds the column's capacity. b) The strength of the solvent used to dilute the sample exceeds the initial concentration of the mobile phase.  c) Degradation of sample component(s).	a) Decrease the volume injected or dilute the sample. b) If appropriate for your application, ensure that the flush solvent is of the same composition as the solvent used to dissolve the sample and that it never exceeds the initial composition of the mobile phase. c) Prepare fresh sample and repeat injection.
5. Band broadening.	a) Fitting problem at the injector-column connection. b) Tubing i.d. too large.	a) Inspect for damage, mismatch, incorrect assembly. b) Use 0.005-inch i.d. tubing.
6. Smaller than expected peak heights.	a) Small injection due to wrong-size syringe. b) Wrong injection. c) Air in lines. d) Misadjusted syringe-drive mechanism.	a) Check syringe size. b) Check injection volume value in file. c) Flush. d) Call your BAS service representative.

SYMPTOM	POSSIBLE PROBLEM	REMEDY
7. Peaks during a blank injection (from a previous injection).	a) Sample carryover due to residual sample in the sample loop.  b) Sample carryover due to incorrectly assembled sample-loop fittings (adding dead volume that can contain residual sample).  c) Sample too concentrated.	a) Insufficient flush volume. Increase Injection Volume in Edit Menu; add a blank vial between sample vials; or use a stronger flush solvent.  b) Check and remake fittings.  c) Dilute sample.
8. No peaks.	a) Plugged or bent needle. b) Leaking flush valve fitting. c) Plugged or leaking tubing.	a) Clean/replace needle. Tighten fittings and syringe. b) Tighten fitting. c) Replace faulty tubing.
9. Incorrect precision and/or accuracy problem on a sample of known content.	a) Air leak. b) Worn out syringe.	a) Tighten all fittings and repeat the run. b) Replace syringe.
10. Irreproducibility of peak areas.	a) Plugged or bent needle. b) Leaking flush valve fitting. c) Plugged or leaking tubing. d) Sample viscosity too low or too high.  e) Solvent viscosity too low or too high.	a) Clean/replace needle. Tighten fittings and syringe. b) Tighten fitting. c) Replace faulty tubing. d) Change viscosity setting in the FILE/Edit/More Menu. e) Change Solv Viscosity setting in the OPTIONS/Configurations Menu.

**8.4 Hardware Problems**

<b>SYMPTOM</b>	<b>POSSIBLE PROBLEM</b>	<b>REMEDY</b>
<u>Startup</u>		
1. Nothing works when instrument is turned on.	a) Power cord unplugged or faulty. b) Blown fuse. c) Defective power switch, driver, transformer.	a) Plug in/replace cord. b) Replace fuse. c) Call your local BAS service representative.
2. Fan is running but display doesn't work.	a) Display component(s) failure.	a) Call your local BAS service representative.
3. No response from keypad but display works.	a) Defective keypad.	a) Call your BAS service representative.
4. Tray temperature control won't turn on.	a) File isn't loaded.	a) Load file.
<u>Communications</u>		
5. EEPROM fails to retain memory.	a) Reinitialize the EEPROM.	a) If unsuccessful, may be a defective PCB. Call your BAS service representative.
6. External devices (timed events) don't work.	a) Wrong settings. b) Interface equipment problem.	a) Check Section 9 for proper pinouts. b) Refer to respective instrument manuals.
<u>Vials</u>		
7. Vial stuck in tower.	a) Needle bent.	a) Replace needle module (see Section 5).
8. Vial stuck in mixer.	a) Run Clear Mixer routine in the Commands Menu.	a) See page 157.
9. Instrument can't locate vial.	a) Incorrect vial position entered into Samples Menu. b) TrayTemp Option incorrectly selected in Configurations Menu. (Instruments with tray temperature control have fewer tray rows.) c) Sample incorrectly placed in sample tray. d) Arm misaligned. e) Arm movement restricted.	a) Check Samples Menu. b) Check configuration. c) Check tray arrangement. d) Home arm. See "Special Commands" on page 155. e) Remove restriction and home arm.
10. Vials don't fit in tray.	a) Incorrect vials.	a) Vial diameter should be 0.44–0.47 inches.
11. Broken vials.	a) Tray not aligned or not seated properly.	a) Seat tray completely in tray chamber and pull handle back toward you.
12. Torn septa.	a) Bent needle.	a) Replace needle.
<u>Injector</u>		
13. Injector valve won't switch.	a) Slipping injector drive.	a) Call your BAS service representative.

<b>SYMPTOM</b>	<b>POSSIBLE PROBLEM</b>	<b>REMEDY</b>
14. Injector valve continues to rotate until fault is generated.	a) Loose or faulty sensor cable.	a) Call your BAS service representative.
15. Injector valve does not rotate to the proper position.	a) Valve requires excessive torque.	a) Call your BAS service representative.
16. Motor fails to stop after syringe is fully returned.	a) Possible defective sensor.	a) Call your BAS service representative.
17. Sample loop won't fill.	a) Blocked injector valve. b) Blocked sample loop. c) Worn syringe. d) Blocked needle.	a) Follow procedures on page 158 to isolate blockage and flush, or take apart valve and sonicate. DO NOT clean valve with a cleaning wire or you could damage it. b) Backflush loop. If problem persists, replace the loop. c) Substitute a "known-good" syringe and determine if symptoms persist. d) Clean with a cleaning wire.
18. Rapid valve-seal wear.	a) Too much valve seal pressure against valve. b) Particulate matter in valve. c) Inadequately filtered sample. d) Buffer crystallization.	a) Decrease LC system pressure. b) Use an in-line filter before the injector valve. c) Filter with a 0.5- $\mu$ m filter before injection. d) Don't allow buffers to stand in system. Run a flush sequence from the Commands Menu when the system is not in use.
19. Injection from incorrect vial.	a) Incorrect vial position entered in Samples Menu. b) Sample placed in incorrect position in vial tray.	a) Check Samples Menu. b) Check tray arrangement.
20. Samples injected incorrectly in a pattern.	a) Error in file (calibration vials placement/sample preparation resource placement).	a) Check file (calibration/sample preparation template).
<u>Needle</u>		
21. Needle bent or broken.	a) Incorrect vials. b) Alignment problem. c) Arm sticking.	a) See list of recommended replacement parts in the front of the manual. b) Call BAS. c) Call BAS.
22. Needle plugging.	a) Multiple/incorrect septa.	a) Verify a single septum of type recommended.

<b>SYMPTOM</b>	<b>POSSIBLE PROBLEM</b>	<b>REMEDY</b>
<u>Syringe</u>		
23. Loud buzzing from syringe drive.	a) Defective limit switch.	a) Call your BAS service representative.
24. Flush valve won't activate.	a) Defective valve/coupler/motor.	a) Call your BAS service representative.
25. Flush valve actuated, but syringe can't draw flush solvent.	a) Plugged inlet filter in solvent reservoir.	a) Replace inlet filter (Section 5).
<u>Arm</u>		
26. Hook doesn't lift vial.	a) XYZ arm stepping motor failure.	a) Call your BAS service representative.
<u>Leaks</u>		
27. Leakage during loop filling.	a) Blocked loop or waste line or valve not deflected far enough.	a) Flush sample loop or replace; check and flush waste line. Check the sensor.
28. Cross-port leakage.	a) Worn rotor seal.	a) Replace according to instructions in Section 5.
29. Liquid on vial caps.	a) Leaky flush valve. b) Leaky injector valve.	a) Replace flush valve. b) Replace rotor seal (Section 5).
<u>Other</u>		
30. An automatic sequence is interrupted, or instrument will not start.	a) Column-oven, heater, door interlock(s) activated.	a) Check the Ready Participation Menu.
31. In Stand Alone mode, Status Screen indicates a run in progress, but the instrument is locked up.	a) Configuration is incorrectly set to SpectraNET mode.	a) In the Configurations Menu, set Mode to Stand Alone.
32. Contaminated sample.	a) Dirty vials. b) Improper septa made of silicone or other rubber-like material.	a) Replace vials. b) Use recommended septa. NOTE: BAS septa are made of Teflon-faced silicone.
33. Elevated system pressures.	a) Block between column and autosampler.	a) Crack each fitting and observe pressure. See instructions in "Testing for Tubing Restrictions" on page 158.

## 8.5 Display Messages

As we mentioned in Section 1, there are three different kinds of messages that can appear on your autosampler's display: user messages, confirmation messages, and error messages. This section describes the possible conditions that generate these messages.

### USER MESSAGES

User messages are displayed as a result of an incorrect operation on the part of the user.

Autosampler Busy

You can't issue a command from a menu or press a key while the autosampler is in the middle of a previously requested operation.

Autosampler Busy No  
Loading Allowed

You can't load a sample set while the queue is running.

Can not add Priority  
Set during final cali-  
bration

You cannot add a priority set during bracketing (after all the sample vials have been injected).

Can not add Priority  
Set at this time

You cannot add a priority set to other than set 1. You cannot add a priority set unless the file's Calibration Menu calls for interspersed calibration vials (you must select reuse calibration vials in order to add priority sets to the queue).

File in Queue Cannot  
be Copied to

You cannot copy to a file assigned to a sample set in the queue.

File in Queue Cannot  
be Deleted

You cannot delete a file assigned to a sample set in the queue.

File in Queue Cannot  
be Edited

You cannot edit a file assigned to a sample set in the queue from the Edit Menu.

*NOTE: You can edit the run-file copy of the active file from the Samples Menu.*

File Protected Cannot  
Be Copied To

You can't copy to a file that is protected (Protect is set to On) in the Configurations Menu.

Use SAMPLES key to  
Add Samples to Queue

You can't start a run by pressing the [RUN] key when the queue is empty. You must first load or add sample set(s) to the queue.

**CONFIRMATION  
MESSAGES**

	Confirmation messages inform you that the autosampler is completing a requested operation or command. These messages disappear when the operation is complete.
Aligning Arm	The Align Arm command is in progress.
Clearing Mixer	The Clear Mixer command is in progress.
File Copied	The requested file has been copied.
File Deleted	The requested file has been deleted.
File Loaded	The requested file has been loaded into the queue.
Flush Samp Syringe	The Flush Sample Syringe command is in progress.
Flush Prep Syringe	The Flush Prep Syringe command (page 155) is in progress.
Homing Arm	The Home Arm command is in progress.
Initializing	The Initialize Hardware command (page 156) is in progress.
Set Added to Queue	The requested sample set has been added to the sample queue.

**ERROR MESSAGES**

There are three kinds of errors: 1) hardware errors, 2) system errors, and 3) diagnostics errors. Hardware and system errors are discussed below. Diagnostic errors are discussed in the diagnostics section on page 149.

**Hardware Errors**

Hardware errors occur during the normal use and operation of the instrument. Most of the time you will be able to clear them. For some, you may need to call your BAS service representative. Pressing any key clears the error message. Then take the steps suggested for each message to resolve the problem.

Arm Failed to Home on X, Y, Z	The XYZ arm could not make the final X-, Y-, or Z-axis motion required return to the home position. Check for a dislodged vial, tray, or other obstruction. Run the Initialize Hardware routine. If the error persists, contact your BAS service representative.
Arm Jam on X, (Y, Z) Axis	The XYZ arm is obstructed on its left-to-right (X) axis; its front-to-back (Y) axis; or its vertical (Z) axis. Check for a dislodged vial, tray, or other obstruction. Run the Initialize Hardware routine. If the error persists, contact your BAS service representative.

---

Flush Valve Misaligned	The flush valve did not turn all the way to the required position. Perform the Initialize Hardware routine. If the error persists, run the Test Connectors routine. If the error persists, contact your BAS service representative.
Hardware Failure	A problem occurred with the valves or syringes. Run the Initialize Hardware diagnostic on page 156 and/or the Injector Test (page 156). If you are unable to clear the error, contact your BAS service representative.
Hardware Not Homed	The solvent valve was requested to move to a certain position, but the hardware was not initialized properly. Or, on powerup, the solvent valve could not home properly. Run the Initialize Hardware diagnostic on page 156. If you are unable to clear the error, contact your BAS service representative.
Hardware Timeout	A requested mechanical operation was not completed in the allotted time. The problem could be with the arm, valves, or syringe(s). Run the Initialize Hardware routine on page 156. If the problem persists, try manually flushing the syringe(s) using the Flush Sample (page 155) and Flush Prep Syringe (page 155) commands. If the problem persists, contact your BAS service representative.
Injector Valve Failure	The injector valve failed to move (between the INJECT and FILL positions, or vice versa) for the requested operation. Issue the Injector command from the Commands Menu. If the error persists, call your BAS service representative.
Inject Valve Misaligned	The injector valve is positioned between the INJECT and FILL positions. Run the Injector test (page 156), and call your BAS service representative if the error persists.
IValve Move Is Impossible	The injector valve cannot move to the requested position from its current position. Move the injector valve to another position using the Injector command, and call your BAS service representative if the error persists.
IValve Move Not Detected	The system did not detect a requested movement to the required position. Check for a loose connection to the injector. Try to manually move the injector to another position. If the error persists, call your BAS service representative.
Inject Valve Overshoot	The injector valve moved past the desired position. Run the Injector test (page 156), and call your BAS service representative if the error persists.
Injector Valve Under-shoot	The injector valve failed to move far enough to the desired position. Run the Injector test (page 156), and call your BAS service representative if the error persists.

---

Parameter Out of Range	A value too large or too small was entered. For example, an injection volume too large for the installed syringe was entered. Compare the size of the installed syringe/sample loop to the edit file parameters.
Solvent Valve Misaligned	The 6-port solvent selection valve failed to turn far enough to reach the next required port or turned past the next required port. Issue the Initialize Hardware command on page 156. If the error persists, issue the Flush Prep Syringe (page 155) using various external solvent positions (S-1, S-2, S-3, S-4). This message appears only on instruments with the Sample Preparation Option installed.
SValve Not Present	A Load or Flush Prep Syringe command was issued without the Sample Preparation Option installed. The Configurations Menu incorrectly indicates that the Sample Preparation Option is installed (Sample Prep Yes). Check the Configurations Menu under OPTIONS. If the problem persists, call your BAS service representative.
Syr 1(2) Not Enough to Expel	One of the syringe(s) is not retracted far enough to expel the requested volume in the ADD step (sample preparation syringe) or flush operation (both syringes). Run the Initialize Hardware test (page 156), and restart the run (sample preparation) or the flush cycle.
Syringe Failure	A syringe failed to moved to the appropriate position required for an operation. Run the Initialize Hardware test (page 156). If the error repeats, call your BAS service representative.
Syringe Overfill	A volume has been requested for injection or ADD that is too large for the size of the installed syringe. Check the method and syringe volume(s).
TrayTemp Failure	The Tray Temperature Control Option was unable to reach the desired set point and has turned itself off returning the instrument to a READY state. Any method loaded will be executed without the Tray Temperature Control Option. Pressing any key clears the error. This error indicates a problem with the Tray Temperature Control Option. Call your BAS service representative for assistance.
Unexpected Vial in Hook	This message appears if a vial is found in the hook where no vial is expected for the requested operation. To clear the error, remove the vial and restart or resume the requested operation.
Vial Not Found in Hook	This message appears if a vial is not found in the hook when a vial is expected. Verify that a vial exists in the requested tray location, check the file and vial assignments in the Samples Menu, and check to see if a vial is lodged in the sample tower. (If so, the needle may be bent.)

## System Errors

System errors occur very rarely and abort normal operation. Since they are not usually problems you can correct, we will list some of them, but not describe them in detail. Should you see any of the following messages or other messages not described in this section, document them, and contact your BAS service representative for assistance.

```
Invalid Current Location
Invalid Command
Not in GET/RET Sequence
Z Must be Up/No Vial
Move Z Not Allowed
Get LocNum: Bad Row, Col
Zup Only in Serv Block
Invalid Target Location
SValve Extra Interrupt
Bad Vial Number
Unexpected Syr Event
```

## 8.6 Diagnostics

The Sample Sentinel autosamplers have built-in diagnostic routines accessible from the Tests Menu (Figure 8.11). These tests enable you to access the operation of your autosampler. Before describing each of these tests, we'll describe how the diagnostic tests are run and how to interpret the error screen information, should a test fail.

All of the test screens are arranged similarly. A test is initiated by pressing [ENTER]. While a test is in progress, the name of the test is displayed along with three dots (Figure 8.9).

Figure 8.9. The In-progress Screen.

```
Test External Contacts: ...
```

### INTERPRETING THE RESULTS

When the test is complete, either a PASS or FAIL result is displayed. Should a test fail, an error screen similar to that shown in Figure 8.10 will appear that contains the following information:

- Test identification
- Fault code (diagnostic error)
- Module identification
- [RUN]/[STOP]

Figure 8.10. An example of an error screen.

Connector:	ERROR #3
Syringe1	[RUN] / [STOP]

*NOTE: The error number in this screen is just an example to show you the screen layout.*

**Test Identification**

The test identification is the name of the Identification test being run.

**Fault Code**

The fault code is the error number associated with the specific failed result. Document and report the fault code to your service representative to receive further assistance in diagnosing and solving the problem.

**Module Identification**

This field displays the specific autosampler board or assembly where the problem occurred.

**[RUN]/[STOP]**

This field prompts you to press [RUN] to repeat the test or to press [STOP] to exit the current test and return to the Tests Menu.

**TEST DESCRIPTIONS**

All diagnostics and built-in assessment routines are accessible from the Tests Menu (Figure 8.11). Each of the routines is described here.

Figure 8.11. Tests Menu.

Software Version
Display Input States
Display Output States
ROM Test
RAM Test
Hardware Test
Test External Contacts
Test Comm Port
Initialize EEPROM

**Software Version**

Selecting this field displays the current software version number, x.yy.zz.

## Display Input States

This menu (Figure 8.12) shows the current states of the TTL-input device signals (Hi or Lo). This information can be used to check the level of the Pump Ready and Injection Hold input lines from other LC system modules. You can use this information to diagnose situations where the autosampler is waiting for ready or waiting to inject. Make sure the Input Polarity is set to the proper level for each of these signals. (See Section 9 for more information on the signal levels.) Pressing the up- or down-arrow keys returns you to the Test Menu's Display Input States field should you wish to repeat the test.

*NOTE: You can change the active polarity of a specific input signal from the OPTIONS Input Polarity Menu.*

Figure 8.12. Display Inputs States Menu.

Pump Ready	Inj Hold
Hi	Hi

## Display Output States

This menu shows the current states of the TTL-output device signals (Hi or Lo). This information is used when hardwiring your autosampler to other LC system modules. (See Section 9 for more information.) Pressing the up- or down-arrow keys returns you to the Test Menu's Display Output States field should you wish to repeat the test.

*NOTE: You can change the active polarity of a specific input signal from the OPTIONS Input Polarity Menu. This menu shows the states of the TTL output device signals (Hi or Lo) as shown in Figure 8.13.*

Figure 8.13. Display Output States Menu.

Display Output States			
AS Rdy	Inject	Grad	Stop
Lo	Lo	Lo	Lo

Pressing the up- or down-arrow keys returns you to the Test Menu's Display Output States field should you wish to repeat the test.

### Display Timed Events

This menu (Figure 8.14) displays the current states of the timed events outputs. This information is used when hardwiring your autosampler to other LC system modules. Refer to Section 9 "System Communication" for more information.

Figure 8.14. Display Timed Events Menu.

TF1	TF2	TF3	TF4
Lo	Lo	Lo	Lo

### RAM Test

This test (Figure 8.15) writes to and then reads the contents of memory. If the write information is the same as the read information, the test is successful. You may wish to run this test once a month to verify proper software operation.

Figure 8.15. RAM Test Menu.

```
Short Memory Test:
```

If successful, the autosampler returns a PASS result (Figure 8.16). If unsuccessful, the autosampler returns a "FAIL" result and displays an error message. Document the test results and error message, and call your BAS service representative.

Figure 8.16. Successful RAM Test.

```
Short Memory Test:  PASS
```

Press [STOP] to return to the Tests Menu.

### ROM Test

It evaluates check-sums on the contents of the instrument's ROM and compares the results to known values.

If successful, the autosampler returns a PASS result. If unsuccessful, the autosampler displays the number of the failed chip(s) along with the regular error screen information (Figure 8.17). If the test fails, document the test results (if failed) and call your BAS service representative.

Figure 8.17. Failed ROM Test.

```
ROM Test:                               Error #: U78, U79
CPU                                       [RUN] / [STOP]
```

## Hardware Test

This is the most important autosampler diagnostic. It checks all of the cable connections, sensors, motors, circuitry, and attached components.

Before running this test, be sure that the XYZ arm is in the home position (at the left rear of the sample compartment), or false errors may be reported. You can use the Home Arm command in the Commands Menu (see "Special Commands" on page 155) if the arm is not at home.

If the Hardware Test fails, the screen displays the test identification, fault code, applicable module or connection, and the [RUN]/[STOP] fields. For example, the message "J10/J40/J45" in Figure 8.18 indicates that further troubleshooting is required. Document the screen information and report it to your BAS service representative when you call for assistance.

Figure 8.18. An example of an error screen.

Connector:	ERROR #3
J10/J40/J45	[RUN] / [STOP]

Pressing [STOP] returns you to the Tests Menu.

Most of the failed Hardware Test results require that you contact your service representative for assistance. However, there are several messages that indicate problems that you can most likely resolve without a service call. These are described here.

### ***Opt Config***

This message indicates an incorrect configuration. Check the Configurations Menu (under OPTIONS) and verify that the options field entries match your installed options.

### ***Arm,#***

This message indicates that the XYZ arm was not in its "home" position when the Hardware Test was initiated. Select /COMMANDS/, /Arm/, /Home arm/.

## Test External Contacts

This test activates all of the external contacts and tests for proper activation. It should not be run while the external contacts are wired to other equipment, or the test will report false errors. Before running this test, disconnect the 12-pin connector block from the rear of the instrument. After the test is complete, plug the connector back in to resume normal instrument operation.

Pressing [ENTER] with the cursor in the Test External Contacts field displays a warning prompt (Figure 8.19).

Figure 8.19. The External Devices Menu.

Disconnect all external contacts	CONTINUE
----------------------------------	----------

From this menu, you can use the [+] and [-] keys to select CONTINUE or ABORT. Selecting Continue starts the test.

Selecting ABORT discontinues the test and returns you to the Test External Contacts field of the Tests Menu. If ABORT is displayed and you want to resume to test, press the left- or right-arrow keys. The display will again show CONTINUE.

### Test COMM Port

A loop-back connector is required to run this test used to assess the operation of the communications port located on the instrument's rear panel. This test sends a signal to the communications port and expects a signal in return. A successful result is reported as a PASS; an unsuccessful result is indicated by the regular error screen.

*NOTE: If you wish to perform this test, contact your BAS Field Service Engineer for the procedure and necessary parts information.*

### Initialize EEPROM

The Electronically Erasable Programmable Read Only Memory (EEPROM) stores all files, user-set variable values, and system configuration information when the power is turned off. Reinitializing the EEPROM restores the default values to all files and erases the system configuration information. Pressing [ENTER] in this field displays the message shown in Figure 8.20:

Figure 8.20. The Initialize EEPROM Menu.

Reset all system values	YES
-------------------------	-----

If you want to exit this routine before running it, you can:

- Press the [+] [-] keys to select No
- Press [MENU] to call up the Main Menu
- Press [Status] to display the Status Screen
- Press the up-arrow key to return to the Tests Menu.

**CAUTION!** This routine erases any current file values and returns all field values to their default values. Initialize EEPROM also erases all configuration information. After running this routine, be sure to reset all installed options to "YES" in the Configurations Menu.

## 8.7 Special Commands

The autosampler has several built-in commands that let you to manually manipulate some of the autosampler's components (the arm, injector valve, etc.) outside of a run. These commands, useful at times during maintenance and troubleshooting procedures, are accessible from the Commands Menu (Figure 8.21).

Figure 8.21. The Commands Menu.

```
Flush Sample Syringe
Flush Prep Syringe
Initialize Hardware
Injector
Arm
```

### FLUSH SAMPLE SYRINGE

This command lets you manually flush the 250  $\mu\text{L}$  sample syringe. You used this command when you primed your autosampler. It can also be useful for maintenance and troubleshooting. To access this test:

1. Fill your flush solvent bottle, if you have not already done so.
2. Press [MENU] and select /COMMANDS/ (Figure 8.21).
3. Select /Flush Sample Syringe/ to bring up the menu shown in Figure 8.22.
4. Use the [+] and [-] keys to select the desired flush volume in microliters. Then press [ENTER] to initiate the flush operation.

Figure 8.22. Flush Syringe Menu.

```
Flush 5000  $\mu\text{L}$ 
```

The autosampler automatically flushes the sample syringe and flush lines with the specified volume of flush solvent and returns the syringe to its starting position. During the flush sequence a confirmation message is displayed.

### Flush Prep Syringe

If you purchased the Sample Preparation Option, you can also flush the large (2.5 mL) sample preparation (prep) syringe and external solvent lines manually as follows:

1. Fill each solvent bottle with an appropriate solvent.
2. Press [MENU] and select /COMMANDS/.
3. Select Flush Prep Syringe (Figure 8.23).
4. Use the [+] and [-] keys to select the desired flush volume in microliters. Then move the cursor to the Solv field and select the desired solvent (S1–S4, Btl). Press [ENTER] to initiate the flush sequence.

Figure 8.23. Flush Prep Menu.

```
Flush 5000 µL Solv Btl
```

The autosampler automatically flushes the selected solvent line with the specified volume of flush solvent and then returns the prep syringe to its starting position.

## INITIALIZE HARDWARE

The Initialize Hardware command allows you to manually control the autosampler's syringes and injector valve. This command brings the syringe(s) to the initial position(s) and removes any backlash.

## INJECTOR

The Injector command allows you to manually switch the injector valve between the inject and fill positions. Selecting /Injector/ displays the menu shown in Figure 8.24.

Figure 8.24. The Injector Menu.

```
Rotate Injection Valve INJECT
```

Pressing the [+] or [-] key and then pressing [ENTER] changes the injector valve's position.

## ARM

The Arm Menu contains two routines for returning the XYZ arm to its home position and for aligning the arm. Figure 8.25 shows the Arm Menu. The two routines are described below.

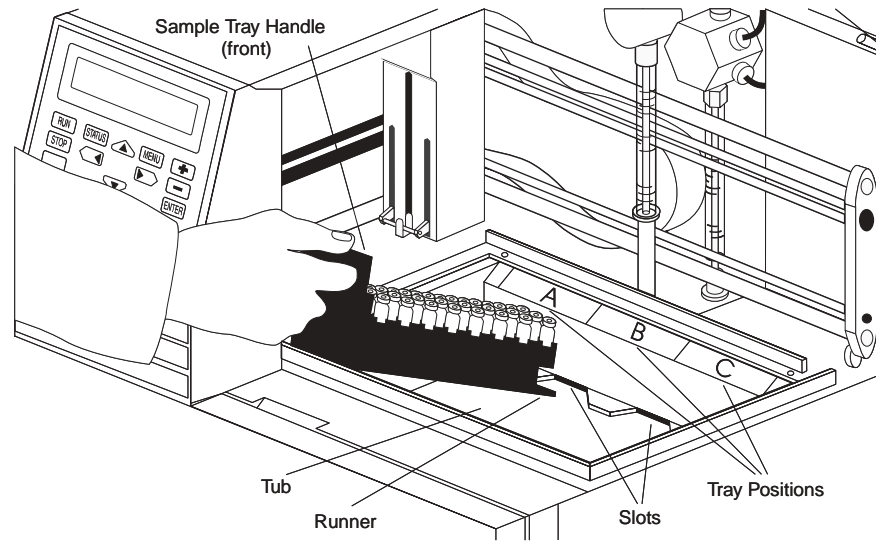
Figure 8.25. The Arm Menu.

```
Home Arm
Align Arm
Clear Mixer
```

## Home Arm

Pressing [ENTER] with the cursor in this field moves the arm from its present position to its home location to the left of the rear-left corner of the tub (Figure 8.26).

Figure 8.26. The arm in home position.



### Align Arm

This routine moves the arm to the vial position requested (Figure 8.27). It is used primarily by BAS service engineers to properly adjust the arm's x-y-z reference points. If you suspect that the arm's alignment is incorrect, call your BAS Service representative.

Figure 8.27. Align Arm Menu.

Align Arm	A01
-----------	-----

*NOTE: The arm "senses" its position relative to its home position. Therefore, if you request the arm to go to position A01, and it goes to position A06, the system configuration may incorrectly indicate that the Tray Temperature Control Option is installed. Check the system configuration in the OPTIONS Configurations Menu against the installed options.*

### Clear Mixer

This routine is used to remove a vial from the mixer if a power failure occurs during a mix step in a sample preparation procedure. The arm picks up the vial from the mixer and returns to the home position. You must manually return to the vial to its proper location in the sample tray.

*NOTE: This routine appears on the Arm Menu only with the Sample Preparation Option installed.*

## 8.8 Testing for Tubing Restrictions

If there is a restriction in the solvent lines, you will probably hear a loud grinding sound from the syringe-drive mechanism due to increased resistance to syringe travel.

*NOTE: Before beginning this procedure, be sure to set up a beaker or other container to catch the solvent.*

To isolate a block to a section of tubing:

1. Place the Autosampler injection valve into the FILL position (press [MENU], COMMANDS, Injector Test, and use the [+] and [-] keys to select /Fill/).
2. Disconnect the injection valve-to-flush valve tubing at the injection valve. Initiate a flush sequence (press [MENU], /COMMANDS/, /Flush Sample Syringe/, page 155 or /Flush Prep Syringe/, page 155). Solvent will flow freely on the return stroke of the plunger if there are no restrictions present. Flow restriction is unlikely at this point due to the large internal tubing diameter.
3. Reconnect the tubing.
4. Disconnect the needle and check the flow.
5. If no restriction is found, reattach all of the tubing, and flush the syringe to verify that the restriction still exists.
6. If a restriction is still present, remove and clean the sample needle as described in Section 5, page 83.

## **Section 9. Advanced Installation and Features**

This section contains advanced installation procedures as well as instructions for using some of the advanced autosampler functions. The installation procedures covered in this section are:

- System Communication
- Communication Modes
- Setting the Polarity
- Ready Participation

The advanced functions included in this section are:

- Instrument Configurations and Other Features
- Gradient Delay
- Needle Height
- Optimization (injection volume and solvent viscosity)

We recommend that you familiarize yourself with Sections 2 and 3 before trying the procedures in this section.

### **9.1 System Communication**

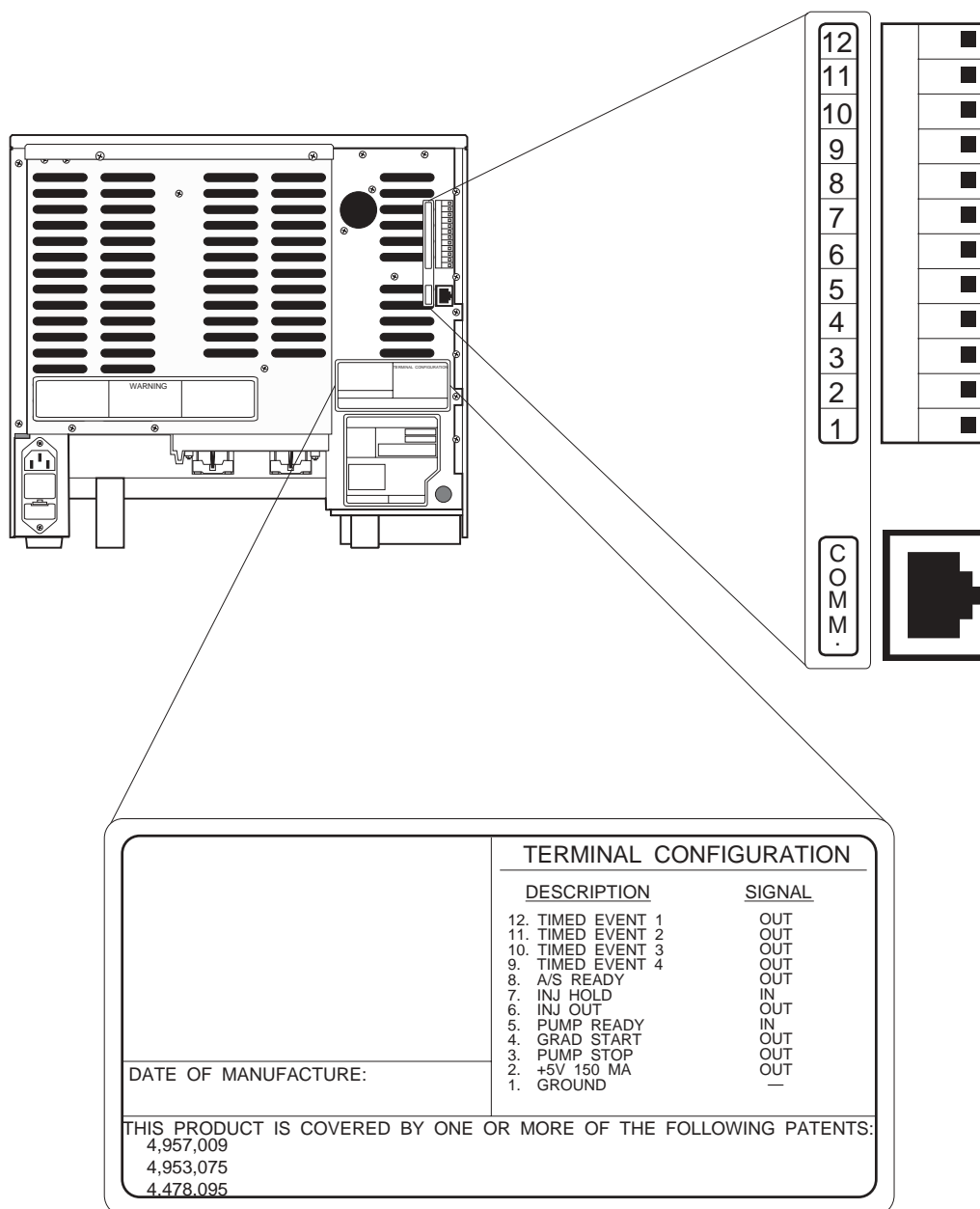
Your autosampler can operate as a separate unit or as part of an automated LC system. The 12-pin, in-line connector on the instrument's rear panel allows communications between your autosampler and other components in an LC system (Figure 9.1). This simplistic communications system, known as "hardwiring," provides injection synchronization between your autosampler and the other components of your LC system.

A mating connector (provided in the accessory kit) allows you to attach control wires from other LC devices to the 12-pin terminal by loosening the appropriate screw, inserting the wire, and then tightening the screw. Once all connections are made, you can quickly make and break connections without disturbing individual wires.

Hardwire communications allow the autosampler to start and stop a pump or detector, to begin a gradient program (gradient pumps only), or to control other features of the externally connected instruments. In addition, the terminal has four timed-function outputs that can be used to control valve-switching devices or other devices on a timed basis. (Refer to Section 3, "Timed Events Menu," page 25, to set up a timed-events program.)

This section describes the pin assignments, the recommended wiring configuration, and some optional wiring configurations.

Figure 9.1. The 12-pin terminal.



---

**PIN ASSIGNMENTS**

*NOTE: All inputs and outputs are referenced to ground.*

**Pin 1 - GROUND**

This is the ground reference used for all inputs and outputs.

**Pin 2 - +5VDC**

This regulated +5 VDC supply can be used to power pull-up resistors or external relays. The current is limited to a maximum of 300 mA to protect against external short circuits affecting the autosampler's operation.

**Pin 3 - PUMP STOP  
(output)**

This pulsed output is used to stop the pump after the last sample has been processed. It is activated when the last sample in the queue has completed its last injection, or when the shutdown file has been activated (using the special shutdown command). See Section 5 for more information on the Shutdown command.

**Pin 4 - GRADIENT START  
OUTPUT**

This signal activates the pump's gradient program. It occurs at a specified amount of time before the Inject Out line is activated. The time is specified as a Gradient Delay value in the More Menu (under Files). This momentarily active, pre-injection signal permits the gradient to start prior to sample injection to compensate for system delay volume. The result is an injection that occurs just as the gradient front reaches the injection valve. See the last section in this section for more information on the gradient delay function.

**Pin 5 - PUMP READY  
(input)**

Through this input, the pump signals the autosampler that the mobile phase flow rate is stable and that the pump is ready for an analysis to begin. (Other devices may also be connected here.) This input must be continuously active for the autosampler to proceed with injections. From the Input Polarity Menu you can select whether a high or low level on this line means "ready." The default is Hi.

**Pin 6 - INJECT OUT  
(output)**

This line sends a momentary signal each time the autosampler makes an injection. This signal allows the synchronization of external devices (pumps, detectors, integrators, etc.) to the autosampler's injection. See "Outputs" below for the specifications of this output.

**Pin 7 - INJECT HOLD  
(input)**

This signal puts the autosampler in the hold state with sample in the loop. Releasing the hold will cause an immediate injection. The signal must remain continuously active to hold injections. That is, when active, this signal will delay an imminent injection until this line goes to the inactive state. This function can be used with external equipment to precisely control the timing of the injection valve activation. The active level (Hi or Lo) of this input can be selected in the Input Polarity Menu (see Advanced Installation) at the end of this section.

**Pin 8 - A/S READY**

This line is used to signal to other devices that the autosampler is in a READY condition. See "Outputs" below for the specifications of this output.

**Pins 9 thru 12 - TF4-TF1 (timed outputs)**

These user-programmed outputs are useful for triggering column switching valves or other external hardware. These outputs are controlled by the time functions defined in the currently active file (see Section 3, "Timed Events Menu" on page 25).

**OUTPUTS**

All output signals, including Time Functions 1 through 4 (TF1-4), are open collector outputs capable of sinking (handling) currents up to 600 mA at voltages up to 24 VDC. The active state is user programmable in the Timed Events Menu (see Section 3).

**INPUTS**

All inputs accept TTL-level inputs, with a transition activating each. The two inputs are tied to +5 VDC through pull-up resistors. A "Lo" level is from 0 to 0.8 V; a "Hi" level is from 2 to 5.5 V.

**9.2 Normal Operation**

For normal stand-alone operation, you do not need to need to change any of the values in the Communications Menu. Leave the communications parameters set to the default values (Figure 9.2). Details are provided for information only.

---

Figure 9.2. The Communications Menu default values used for normal stand-alone operation.

Baud Rate	9600
Parity	NONE
Flow Control	NONE
Data Bits	8
Stop Bits	1
Echo	On

The allowable entries and default values are listed below for each of the communications parameters.

Baud Rate: Allowable entries: 1200, 2400, 4800, or 9600 Bd; default: 9600Bd.

Parity: Allowable entries: None, Even, or Odd; default: NONE.

Flow Control: Allowable entries: NONE or XON/OFF; default: NONE.

Data Bits: Allowable entries: 8 or 7; default: 8.

Stop Bits: Allowable entries: 1 or 2; default: 1.

Echo: Allowable entries: On or Off; default: On.

*NOTE: If the Communications Menu doesn't appear as one of the OPTIONS Menu choices, return to the OPTIONS Configurations Menu, and verify that Mode is set to Stand Alone.*

### 9.3 BCD Communication

The BCD Communication Option provides BCD communication between the Sample Sentinel autosamplers and BAS ChromGraph data systems. This option provides tray and/or vial location information as binary-coded decimal data. The BCD assembly must be installed by a BAS service representative if ordered after initial instrument delivery. Installation instructions are provided in the BCD Option Kit. Contact your representative for further information.

### 9.4 Setting the Polarity

From the Options Menu you can change the sense of your autosampler's input and output signals as required by your other LC system modules. For example, when shipped, the Autosampler Ready Output is set to send a positive voltage (Hi) when the instrument is ready. If you change the Autosampler Ready Output Polarity to Lo, the instrument will send a 0-volt signal (Lo) when it's ready. The procedure for changing the polarity of the input and output signals is described below.

#### INPUT POLARITY

To change the polarity of an input signal, select /Input Polarity/ from the Options Menu. Use the [+] and [-] keys to select either Hi or Lo for the desired input signal shown in Figure 9.3.

Figure 9.3. The Input Polarity Menu.

Pump Ready Active	Hi
Inj Hold Active	Lo

This menu shows the level for the active state for each input signal.

*NOTE: If you want to determine the current state of a particular signal, return to the Main Menu, and select /TESTS/Display Input States/.*

#### OUTPUT POLARITY

To change the polarity of an output signal, select /Output Polarity/ from the Options Menu. Use the [+] and [-] keys to select either Hi or Lo for the desired output signal shown in Figure 9.4.

Figure 9.4. The Output Polarity Menu.

Autosampler Ready	Lo
Inject Out Active	Lo
Grad Start Active	Lo
Pump Stop Active	Lo

This menu shows the level for the active state for each output signal.

*NOTE: If you want to determine the current state of a particular signal, return to the Main Menu, and select /TESTS/Display Output States/.*

Tables 9.1 and 9.2 show the output signal levels relative to the polarity settings and instrument status for the autosampler's input and output signals, respectively.

Table 9.1. Input Levels of Autosampler Input Signals.

		-----Polarity-----	
		Hi	Lo
Pump Ready	Ready	Hi*	Lo
	Not Ready	Lo	Hi
Inj Hold	Active	Hi	Lo*
	Inactive	Lo	Hi

Note: The asterisked (\*) values indicate default settings.

Table 9.2. Output Levels of Autosampler Output Signals.

		-----Polarity-----	
		Hi	Lo
Autosampler Ready	Ready	Hi	Lo*
	Not Ready	Lo	Hi
Inject Out	Active	Hi	Lo
	Inactive	Lo	Hi*
Gradient Start	Active	Hi	Lo
	Inactive	Lo	Hi*
Pump Stop	Active	Hi	Lo
	Inactive	Lo	Hi*

Note: The asterisked (\*) values indicate default settings.

## 9.5 Ready Participation

When the instrument is shipped, the following conditions must be satisfied in order for the Status Screen to indicate that the autosampler is "ready" for operation:

- The door must be closed.
- The heater must be at the set temperature (with the Sample Preparation Option only).
- The tray must be at the set temperature (with the Tray Temperature Control Option installed).

The Options Ready Participation Menu allows you to customize the "autosampler ready" requirements to fit your own needs. For example, you may wish to prepare your first sample set (without injection) while the heater is still warming up. In this case, you would want to disable the heater condition in order to complete the sample preparation run. Or, you may wish to run your autosampler with the door removed. In this case you would want to disable the door interlock.

To access the Ready Participation Menu (Figure 9.5), select /OPTIONS/Ready Participation/.

Figure 9.5. The Ready Participation Menu.

Door Interlock	ENABLE
Heater Ready	ENABLE

To change any of the above requirements, move the cursor to the appropriate line, and use the [+] and [-] keys to change ENABLE to DISABLE.

## 9.6 Instrument Configurations and Other Features

Your autosampler is shipped with a 100  $\mu$ L sample loop (the SS-1 series instruments come with a 20  $\mu$ L loop), a 250  $\mu$ L sample syringe, and a 2.5 mL sample preparation syringe (with the Sample Preparation Option installed). Should you wish to change your syringe sizes, you can do so from the Configurations Menu (Figure 9.6). This menu also reflects tray temperature control. If you would like to adjust the rate at which fields cycle through the available choices, you can change the Key Repeat Rate entry. Section 3 discusses the File Protect function also available from this menu.

Figure 9.6. The Configurations Menu.

Sample Syringe Size	250
Prep Syringe Size	2500
-----	
Prep Installed	Yes
Oven Installed	No
TrayTemp Installed	Yes
Key Repeat Rate	Medium
File	Protect
1:	Off
2:	Off
3:	Off
4:	Off
Solv Viscosity	Normal

## DISPLAY CONTRAST

You can vary the display's contrast to make it easier to read.

To change the display's contrast, first press [STATUS] to access the Status Screen. Then simultaneously press [>] and [+] to *increase* the contrast, or [>] and [-] to reduce the contrast.

## 9.7 Gradient Delay

Gradient delay is a user-defined value that delays the injection of a sample. This delay is the amount of time required for a change in the mobile phase to travel from the gradient proportioning valve to the injection valve. The volume of mobile phase contained between the two valves is also referred to as the precolumn volume (PV).

Do not use gradient delay in conjunction with the BAS 200 Chromatograph or the BAS PM-80 Gradient Pump. Both these instruments synchronize the start of a gradient and signal the autosampler to inject when appropriate.

Although not required for normal autosampler operation or with BAS pumps, a gradient delay is recommended when using large volume pre-filters or low flow rates (< 0.5 mL/min). The gradient delay time can be found under the FILE/Edit/More Menu (Figure 9.7). You then use the [+] or [-] keys to enter the desired time in minutes.

The following procedure is used to calculate the gradient delay time for your HPLC system.  
(See Figure 9.8.)

1. Inject an unretained sample into your LC system with a column in-line and using 100% A as the mobile phase (e.g., MeOH).
2. Change to 100% B (e.g., 0.1% Acetone in MeOH) as fast as possible.
3. Subtract the retention time ( $R_t$ ) of the unretained peak from the retention time of the start of the baseline shift ( $T_2 - T_1$ ).
4. Subtract the time required to switch mobile phases from 100% A to 100% B.
5. The resulting time is the gradient delay time.

### EXAMPLE

The following example illustrates how to apply this procedure.

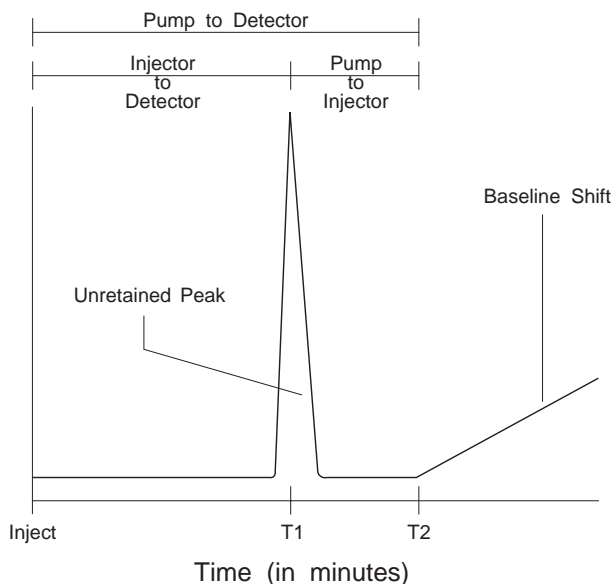
An unretained peak has an  $R_t$  of 3.0 min with a baseline shift at 4.5 min and a complete mobile-phase switch time of 0.1 min. The gradient delay is calculated as follows:

$$\begin{aligned} \text{Gradient Delay} &= (T_2 - T_1) - 0.1 \text{ min} \\ &= (4.5 - 3.0) - 0.1 \\ &= 1.4 \text{ minutes} \end{aligned}$$

Figure 9.7. The More Menu (Gradient Delay).

Equilibration Time	0.0
<b>Gradient Delay</b>	<b>0.00</b>
-----	
Viscosity	Medium
Flush Volume	250.0
Injection Type	Push
Injection Range	1-1500
Needle Height	2.0

Figure 9.8. The delay time calculation using a hypothetical chromatogram.



## 9.8 Needle Height

For some applications, such as liquid-liquid extractions and low-injection volumes, you may need to change or optimize the "needle height." On Sample Sentinel autosamplers, the needle doesn't move. Instead, the bottom of the *vial* moves relative to the needle tip. The "needle height" is the approximate distance between the needle tip and the bottom of the vial (0–20 mm).

The More Menu's default setting of 2 mm (Figure 9.9) is sufficient for most applications and accommodates a wide variety of vials. However, in cases where you have very limited sample volumes, and especially for microvials, further optimization may be required.

*NOTE: The thickness of the bottom of the vial varies considerably between vial types (1.8 mL vials, microvials, and vials with micro-inserts, etc.) and materials (glass, plastic, etc.).*

Figure 9.9. The More Menu (Needle Height).

Equilibration Time	0.0
Gradient Delay	0.00
-----	
Viscosity	Medium
Flush Volume	250.0
Injection Type	Push
Injection Range	1-1500
<b>Needle Height</b>	<b>2.0</b>

**CAUTION!** Operating the autosampler with a needle height different from the default height may bend the needle. Do not adjust the needle height without a spare needle available.

Use the following procedure to optimize the needle height for your sample vials:

1. Place 200–300  $\mu\text{L}$  water in a vial in position A01.
2. Create an autosampler file that will make two 200  $\mu\text{L}$  injections (see Section 3 for more information).
  - a. Set the Injection Menu's Injection Volume field equal to 200.
  - b. Set the Injection Menu's Injections/Sample to 2.
3. Run the file and observe the amount of fluid remaining in the vial. The fluid height indicates the position of the needle tip from the vial's bottom.
4. Adjust the More Menu's Needle Height value until the needle tip is located approximately 1–2 mm from the bottom of the vial.
5. Repeat steps 1–4 to confirm the needle height.

## 9.9 Optimization

### Injection Volume

This section describes how to select the sample loop and syringe size to obtain maximum reproducibility when performing large-volume injections.

The maximum injection volume depends upon:

- the volume of the syringe currently installed as designated under /OPTION/Configuration/Syringe Size/
- the Injection Mode selected under /FILE/More/Injection Type
- the volume of the installed sample loop

By changing the sample syringe and/or the sample loop, you can set up your autosampler to deliver injection volumes larger than can be delivered by the 250  $\mu\text{L}$  syringe shipped with the instrument. Use the following steps to prepare your instrument for large-volume injections:

1. Determine the correct sample loop size for the injection volume desired. (See Table 9.3.)
2. Use the injection volume and algorithms outlined below to determine the required syringe size. (See Table 9.4.)

The following algorithms define the amount of sample needed for your injection volume:

PushLoop	$(\text{Injection Volume} + 15 \mu\text{L})$
Pull Loop	$(\text{Injection Volume} + 1.1 \mu\text{L})$
Full Loop	$(\text{Injection Volume} \times 1.33) + 70 \mu\text{L}$

3. Change your sample loop and syringe. (Refer to Section 5 for instructions.)
4. Enter the syringe size in the /FILE/Configuration/Syringe Size field.
5. Enter the Injection Mode in the /OPTIONS/More/Injection Type/ field.
6. Enter your injection volume in the Injection Menu.

*NOTE: If the Full-loop mode is chosen, the maximum value depends only on Loop Size (assuming that the syringe size is larger than the loop size). (Loops come only in the sizes included in the Parts and Accessories list located at the front of your Reference Manual).*

*NOTE: For Pull- and PushLoop injection, to ensure maximum reproducibility, we recommend that you do not inject more than half of your loop volume.*

Table 9.3. Recommended Injection Size for Selected Sample Loop Sizes.\*

Sample Loop Size (μL)	Injection Volume (μL)		
	<u>PushLoop</u>	<u>Injection Mode</u> <u>Pull</u>	<u>Full</u>
5	2.5	2.5	5
10	5.0	5.0	10
20	10	10	20
50	25	25	50
100	50	50	100
200	100	100	200
500	200	250	500
1000	200	500	1000

\* These recommended volumes will provide the best chromatographic performance. Maximum injection sizes will be determined by the particular system being used.

The syringe size needed will be the total volume determined by these algorithms. For example, for a 500 μL full-loop injection (using a 1000 μL loop), the Total Volume (TV) needed is

$$TV = (500 \mu\text{L} \times 1.33) + 70 \mu\text{L}$$

$$TV = 735 \mu\text{L}$$

Therefore you will need a 1000 μL syringe since there are no 735 μL syringes available. (See Table 9.4.)

Table 9.4. Maximum Injection Size for Syringe Size.

Injection Type	Syringe Size (in μL)				
	<u>250</u>	<u>500</u>	<u>1000</u>	<u>2500</u>	<u>5000</u>
Push Hi	200	200	200	200	200
Pull	212	461	956	1500	1500
Full	100	200	500	1000	1000

**SOLVENT VISCOSITY**

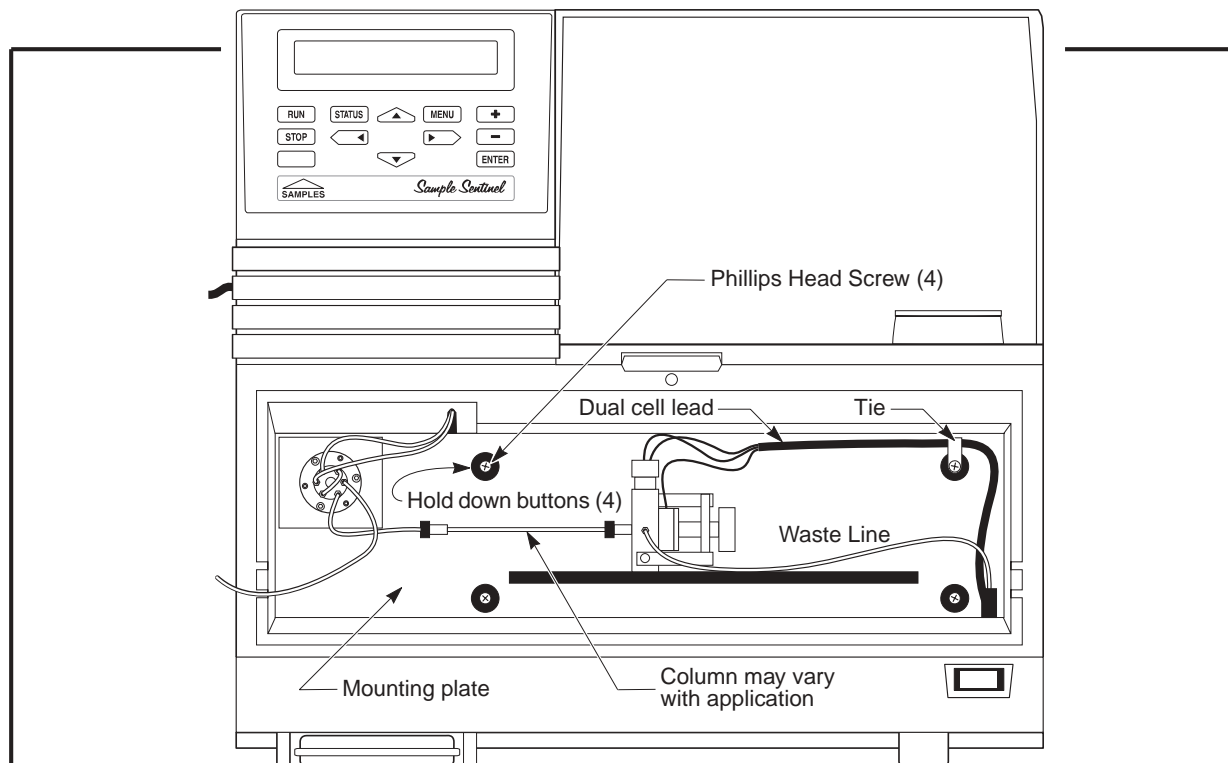
You can adjust the syringe speed (draw rate) for your solvent viscosity from the Solv Viscosity field in the Configurations Menu (Figure 9.10).

Figure 9.10. Configurations Menu -- Solv Viscosity field.

Sample Syringe Size	250
Prep Syringe Size	2500
-----	
Prep Installed	Yes
Oven Installed	No
TrayTemp Installed	Yes
Key Repeat Rate	Medium
File	Protect
1:	Off
2:	Off
3:	Off
4:	Off
<b>Solv Viscosity</b>	<b>Normal</b>

Using the [+]/[-] keys you can choose among three different syringe speeds: 1) Normal (fastest), 2) Medium, and 3) Viscous (slowest). "Normal" lets you optimize your cycle time and is optimal for most applications. With either very viscous solvents or normal-phase (low viscosity) solvents, you may wish to use the "Viscous" setting to slow down the syringe rate to prevent cavitation and unreproducible injection volumes.

## 9.10 Electrochemical Detector Accessory



### A-1822

#### Sample Sentinel Conversion Installation Instructions

1. Remove front panel. It is held in place by detents and can be removed by hand.
2. Remove all tubing and loop from the injection valve.
3. Remove (but keep) the four Phillips head screws that hold in the black tray.
4. Lift out the original black plastic tray and store for future use.
5. Install the mounting plate assembly over the mounting holes that are molded into the front of the sample sentinel.
6. Place a hold down button (included with the mounting plate assembly) over three of the holes. **Do not install a hold down button over the upper right hole yet.**
7. Using the Phillips screws that were removed earlier, screw down the three buttons and tighten.
8. Starting from the outside of the autosampler, route the the dual cell lead (EW-8122) under the cabinet. Guide it through the rectangular opening located in the lower right corner of the white conversion tray.
9. Slide the strain relief tie down over the cell lead then attach the tie, along with the fourth button, to the last mounting hole. Use the last Phillips screw and tighten.
10. Reinstall the loop and tubing on the injection valve.
11. The conversion is now complete. Connect the column and flow cell in accordance with the type of configuration ordered. **The waste line from the flow cell must be routed in a way that will prevent pinching when the outer cover is installed. The same opening used by the cell lead will also guide the waste line.**



## Section 10. Glossary

We have included a glossary to define certain technical terms used throughout the manual's text. These terms should be consistent with standard definitions used throughout the analytical industry, and are added here as a quick reference only.

### **A**

#### **active set/vial**

The sample set or vial currently being processed. The active vial refers to the vial currently being injected or currently undergoing sample preparation.

#### **Add**

1) A step used in sample preparation used to deliver all Pickup volumes and the Load volume to the target vial. 2) The process of putting a sample set into the sample queue.

### **B**

#### **backlash**

The slack in the syringe and its drive mechanism.

### **C**

#### **calibration interval**

The number of sample vials to be injected between calibration-vial injection(s).

#### **calibration level**

The number of different concentration levels used in the calibration series.

#### **calibration vial**

A vial that contains a known concentration of the compound(s) of interest; a standard.

#### **cycle time**

The time between autosampler injections.

### **D**

#### **dead volume**

The volume from the solvent-selection valve to the needle tip.

#### **decrement**

Press the [-] key to decrease the value of a parameter.

#### **diluent**

The solvent or buffer used to dilute a sample. On the autosampler, the volume of diluent is specified in the Load step of all of the sample preparation templates.

#### **dilution cycle**

A preset sequence used in the sample preparation templates that transfers solvents, sample, and reagents from the solvent bottle(s) or source vial(s) to the target vial(s).

## **E**

**extra vials** In sample preparation, an empty or extra reagent vial where samples and solvents are mixed; an empty reaction vial. It is always placed in relationship to the sample vial (Sam+1, Sam+2, Sam+3).

## **E**

**file/method** "File" and "method" are used interchangeably to describe the autosampler run parameters. However, the word "file" is often used in a more specific sense to mean the autosampler files in which the parameters are stored.

**flush solvent** The solvent used to wash the internal tubing, syringes, and sample needle after an injection. This solvent should be of sufficient strength to easily remove the small amounts of sample that can cling to the tubing walls.

**full-loop injection** A technique that injects a sample volume equal to the loop volume.

## **G**

**guard column** A small column placed just before the separation column. The guard column is used to remove contaminants that might otherwise damage the main column or interfere with the separation.

## **H**

**holding loop** The isolated section of connecting line used in sample preparation to hold measured amounts of sample, reagents, and solvents until they are delivered into a vial.

**home** The position to the left of the rear-left corner of the sample tub where the XYZ arm resides when it is not transporting a vial.

**hook** The attachment on the end of the pod that moves a vial up and down (along the Z axis). The hook lifts the vial from the sample tray and holds it during transport to the sample tower for injection.

## **I**

**increment** Press the [+] key to increase the value of a parameter.

**initialization** The process that takes place between dilution cycles that brings the autosampler to a "known" state. During this process the lines and syringes are automatically flushed with flush solvent. Any residual resources present from a previous dilution cycle are flushed out of the lines to prepare the autosampler for the next injection.

## **J - L**

**linear dilution** A dilution sequence in which resource A (buffer) is added to resource B, C, and D.

**loading a file** Loading a file copies the original edit file into the run-file location to be run. The original edit file remains.

## **M**

**method** A group of parameters applied to one or more vials.

**mobile phase** In HPLC, the liquid used to move the sample through the column.

## **N - O**

**open-collector outputs** An output, usually a transistor, that has no connection to the collector.

## **P**

**PCB** A printed circuit board.

**PickLG** A step used in the sample preparation templates that retracts the prep syringe to draw a large volume of solvent or buffer into the holding loop.

**Pickup** A step used in the sample preparation templates that retracts the sample syringe to draw a small volume of sample or reagent into the holding loop.

**pod** The autosampler component attached to the XYZ arm that contains the hook.

**precision** The degree to which results are reproducible (analysis of the same sample yields similar results).

**prep syringe** The large 2.5 mL sample preparation syringe used to transfer large volumes of liquid (usually buffer or solvents).

**prime** The process that flushes lines with flush solvent, etc. Priming is done during installation and whenever a syringe is replaced.

**pull-loop injection** Variable-loop injection.

**pull-up resistor** Resistors used to pull inputs or nonactive open-collector outputs to "Hi" states.

## **Q**

**queue** One or more vial sequences/sets subjected to one or more methods run in a series that use(s) the same column and solvents. The sample queue lists the order and file name for each sample set to be processed during a given run.

## **R**

**reagent transfer step** In a sample preparation program, a step used to deliver solvents, reagents, or samples from one location (vial or solvent bottle) to another (target or sample vial).

**real-time editing** The ability to edit the run file for a run currently in progress. On the Sample Sentinel, this function is provided by the [SAMPLES] key.

**recalibration** The reinjection of the calibration vial(s) after the injection of each series of sample vials within a sample set.

**recalibration interval** The number of sample vials injected between each calibration series injection. The value in the Samples/Calibration field.

**resource** One of the samples, calibration vials, reagents, or solvents used in an autosampler run.

**routine** The type of order in which calibration vials are arranged within a sample set. Calibration vials may be reused throughout the set (Reuse same vials) or they may be interspersed (Interperse) among the sample vials in the sample tray.

**run file** The copy of the edit file that has been loaded and the file that is currently running. From the Samples Menu, changes can be made to the run file during a run, but those changes can't be saved until after the run has completed. Also, changes can't be copied to a file that is in use (assigned to a sample set in the queue).

**S**

<b>sample needle</b>	The needle that pierces the septum of the vial cap and through which vial contents are drawn into the holding loop.
<b>sample syringe</b>	The 250 $\mu\text{L}$ syringe. In sample preparation, this syringe is used in sample preparation to transfer small volumes of liquid (usually samples or reagents).
<b>sample tower</b>	The autosampler location and component where injection takes place.
<b>sample tray</b>	One of the three racks that holds the sample vials in position in the autosampler.
<b>sample tub</b>	The autosampler compartment that holds the sample trays.
<b>sample vial</b>	The container that holds the sample in the sample tray. Sample vials must have a 12 mm diameter.
<b>sampling valve</b>	The valve through which sample is injected onto the column.
<b>set</b>	A group of samples vials and associated calibration vials run with a particular method.
<b>solvent reservoir</b>	One of the four bottles on the back of the autosampler that contains the external solvents used in sample preparation. The solvent in the first position usually contains the buffer.
<b>solvent-selection valve</b>	The 6-port valve that regulates the flow of solvents and air into the sample holding loop. This valve allows the flow of waste out of the loop.
<b>standard</b>	A vial that contains a known concentration of sample. Peaks generated from this vial are used to plot the calibration curve used to determine the concentration of the compound of interest. (See also calibration vial.)
<b>syringe refill and loop-isolation valve</b>	The valve in the sample preparation system that regulates the flow of liquid from the syringes.

**T - V**

<b>template</b>	In sample preparation, one of the four file formats that contain a unique, preset, unchangeable sequence of steps used to carry out a series of sample preparation procedures. The
-----------------	--

four types of templates include simple dilution, heating, extraction, and heating and extraction.

### **W - X**

#### **Wait**

A step used in the sample preparation templates that pauses all autosampler activity. This step may be used to allow a mixture to settle or to react.

#### **X**

In the Calibration Menu's Calib 1 (of X) field, X indicates the number of calibration levels in the calibration sequence. X is not visible on the screen; it is used for discussion purposes only.

#### **XYZ arm**

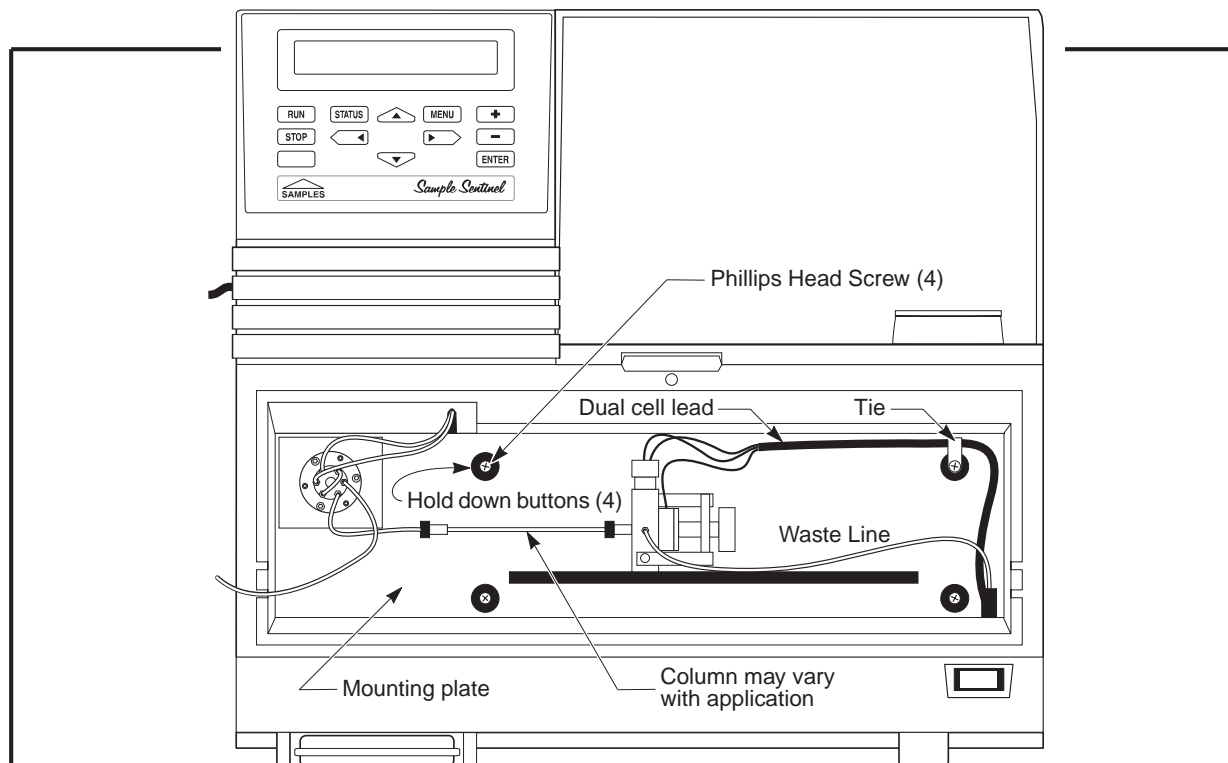
Also referred to as the "arm," this component carries the sample vials to and from the sample tray and to the various workstations (sample tower, heater mixer) in the autosampler.

### **Y - Z**

#### **Z**

In the sample preparation templates, the variable used to represent the number of clean (extra) vials subsequent to the sample vial. Z equals 1, 2, or 3.

## Appendix: Electrochemical Detector Accessory



### A-1822

#### Sample Sentinel Conversion Installation Instructions

1. Remove front panel. It is held in place by detents and can be removed by hand.
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8. Starting from the outside of the autosampler, route the the dual cell lead (EW-8122) under the cabinet. Guide it through the rectangular opening located in the lower right corner of the white conversion tray.
9. Slide the strain relief tie down over the cell lead then attach the tie, along with the fourth button, to the last mounting hole. Use the last Phillips screw and tighten.
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11. The conversion is now complete. Connect the column and flow cell in accordance with the type of configuration ordered. **The waste line from the flow cell must be routed in a way that will prevent pinching when the outer cover is installed. The same opening used by the cell lead will also guide the waste line.**

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