SFC Prep 350 User Guide

Preparative Supercritical Fluid Chromatography

PUBLISHED BY
Thar Instruments, Inc.
130 Executive Drive
Suite 2A
Newark, DE 19702
Phone 302.266.8201
Fax 302.266.8204
Email info@tharsfc.com

Visit our website at http://www.tharsfc.com

Printed in the USA
Document Number: MN56
7/2008, Initial release

SFC Prep 350 User Guide

Overview

This user guide supplied with the SFC Prep 350 Supercritical Fluid Chromatography system provides instructions for basic operation of the system. Safety information is provided in Chapter 1. An overview of the hardware and installation requirements is provided in Chapter 2. Chapters 3 and 4 contain details about the electrical and plumbing connections, respectively. Instructions for operating the SFC Prep 350 are provided in Chapter 5. An overview of the method setup for the SFC Prep 350 system is provided in Chapter 6. Chapter 7 provides maintenance procedures.

This manual should be read in its entirety before operating the SFC Prep 350 system.

Intended Use

The SFC Prep 350 system is intended for use by qualified personnel to perform purification of organic compounds. It is designed for continuous use. The system uses combinations of supercritical or liquid carbon dioxide (CO_2) and liquid organic modifiers to perform normal phase chromatographic separations at the semi-preparative and preparative levels. Separated components are directed to one of multiple collection vessels. During the collection process, gaseous CO_2 and organic vapor are separated from residual liquid modifier with dissolved compounds. Only the liquid phase is collected. The system is optimized to perform multiple overlapped or “stacked” injections with coordinated collections rather than single large volume injections.

WARNING

The user should be made aware that if the equipment is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.

Customer Support

For customer support and field service in North America please contact:

Customer Support
Thar Instruments, Inc.
575 Epsilon Drive, Suite 100
Pittsburgh, PA 15238 USA

Telephone 412 967 5665, Option # 4
Fax 412 967 9446
Email service@tharsfc.com
Internet http://www.tharsfc.com

Outside of North America, please contact your local service representative.

Using This Manual

This manual contains instructions for the safe operation of the Thar Instruments SFC Prep 350 supercritical fluid chromatography system. The following icon definitions are used on the instrument and throughout the manual.

Icon Definitions

WARNING

Calls attention to a condition or possible situation that could cause injury to the user.

VOLTAGE

Calls attention to an area of high voltage that could cause injury to the user.

CAUTION

Calls attention to a condition or possible situation that could damage or destroy either the product or the user's work.

NOTE

Calls attention to information, which may be useful for optimized instrument operation.

Table of Contents

Overview iii
Intended Use iii
Customer Support iv
Using This Manual iv
Icon Definitions iv
List of Figures viii
List of Tables x

Chapter 1 Safety

Introduction 1-1
Declaration of Conformity 1-1
Shipping and Storage 1-2
Safety Requirements 1-3
Safety Features 1-3
P-350/200 Pumps 1-3
Safety Sensors/Alarms 1-4
Additional Warnings 1-5
Flammability Warning 1-5
Electrical Considerations 1-6
Operating Considerations 1-7
Chemical Considerations 1-8
Circulating Bath 1-11
Collection Container Requirements 1-12

Chapter 2 Hardware Overview

Introduction 2-1
SFC Prep 350 System Components 2-1
CO_2 Pump 2-3
Cooling Heat Exchanger and Circulating Bath 2-4
Mass Flow Meter 2-4
Co-Solvent Pump 2-4
Static Mixer 2-4
Injection Module 2-4
UV Detector 2-4
Fraction Collection Module 2-4
Installation Requirements 2-5
Work Space Specifications 2-5
Customer Supplied Materials 2-7
Electrical Requirements 2-8
Grounding 2-8
Power Supply Requirements 2-8
Chemical Requirements 2-10
Carbon Dioxide 2-10

Co-Solvents 2-10
Plumbing Requirements 2-11
Venting Requirements 2-11

Chapter 3 Electrical Connections

Introduction 3-1
Communication 3-1
Communication Cable Connections 3-1
Power Connections 3-2

Chapter 4 Plumbing Connections

Introduction 4-1
Pump Cooling Tubing 4-1
External CO_2 Connections 4-2

Chapter 5 Operating the SFC Prep 350

Introduction 5-1
System Startup 5-1
Power on the System 5-2
Verify Water Bath Temperature 5-2
Inspect Collection Bottles and Vessels 5-2
Collection Bottles 5-2
Open SuperChrom Software 5-3
UV Detector Setup 5-5
Prime the Co-Solvent (Modifier) Pump 5-10
Install/Replace Column 5-14
Depressurize the System 5-15
Install the Column 5-17
Turn on the CO_2 Supply 5-18
Injection Module Settings 5-19
Sample Connection Size 5-20
Injection Module Priming- (mixed phase injections only) 5-20
Injection Module Priming- (modifier stream injections only) 5-21
Set the Manual BPR 5-22
High Pressure MBPR Instructions 5-22
Collection Vessels 5-23
Set Level Controller Drain Levels 5-26
Shutdown Instructions 5-27
Short Term Shutdown 5-27
Long Term Shutdown 5-28

Chapter 6 Method Setup Overview

Introduction 6-1

Method Settings 6-1
Data File Settings 6-7
Other Options 6-8
Method Startup 6-9
MBPR Instructions 6-11
Test Injections 6-12
Fraction Collection 6-13
Valve Delay Settings 6-13
Manual Collection 6-14
Collection Settings 6-17
Post Collection 6-27
Sequences 6-28
Auto Sequencing 6-29
Manually Create a Sequence 6-32
Stacked Injection Sequences 6-35
Building a Stacked Injection Sequence 6-35
Stacked Injection Collection Tips 6-44

Chapter 7 SFC Prep 350 System Maintenance

Introduction 7-1
Maintenance Schedule 7-1
Pressurizing and Leak Checking 7-3
Cleaning Instructions 7-4
Collection Vessel Cleaning 7-4
Injection Module Cleaning 7-6

Appendices

Appendix I SFC Prep 350 System Technical Specifications II
Appendix II SFC Prep 350 System Settings V
Pump Settings and Operational Ranges V
ABPR Settings X
Appendix III Waste Electrical and Electronic Equipment (WEEE) Directive XII
Appendix IV Consumables and Spare Parts XIII
Appendix IV Declaration of Conformity XIV

Index

List of Figures

Figure 2-1 SFC Prep 350 System 2-2
Figure 2-2 CO_2 Pump 2-3
Figure 2-3 SFC350 System Layout 2-6
Figure 2-4 NA and European Style Plugs 2-9
Figure 4-1 CO_2 Tank Fitting 4-2
Figure 5-1 SuperChrom Login Window 5-3
Figure 5-2 SuperChrom Main Screen 5-4
Figure 5-3 Select Mode 5-5
Figure 5-4 Select Setup 5-5
Figure 5-5 Set Channel 1 5-6
Figure 5-6 UV Lamp Alarms 5-6
Figure 5-7 Set Autorange Multiplier 5-7
Figure 5-8 Set Concentration Factor 5-7
Figure 5-9 Detector Settings 5-8
Figure 5-10 UV Detector Settings 5-8
Figure 5-11 Injection/Collection Marks 5-9
Figure 5-12 MV1 5-10
Figure 5-13 Co-Solvent Pump Display 5-11
Figure 5-14 Co-Solvent Pump Settings 5-12
Figure 5-15 Start Pump 5-13
Figure 5-16 ABPR Settings 5-16
Figure 5-17 Pressure Regulator Settings 5-16
Figure 5-18 MV2 5-17
Figure 5-19 Thar Injection Module 5-19
Figure 5-20 MBPR valve 5-22
Figure 5-21 Cyclone Needle Valves 5-23
Figure 5-22 Manual Collection Switches 5-24
Figure 5-23 Manual Cyclone Drain Switches 5-25
Figure 5-24 1L Cyclone Needle Valve 5-25
Figure 6-1 Click Method Settings 6-1
Figure 6-2 System Run Mode 6-2
Figure 6-3 Pressures 6-3
Figure 6-4 Pumping 6-4
Figure 6-5 Collection 6-5
Figure 6-6 Temperature 6-6
Figure 6-7 Data File Log Settings 6-7
Figure 6-8 Chromatography Log Settings 6-8
Figure 6-9 Start Method 6-9
Figure 6-10 Zero the Detector 6-10
Figure 6-11 Manual Back Pressure Regulator 6-11
Figure 6-12 Thar Injection Module 6-12
Figure 6-13 Valve Delay Settings 6-13
Figure 6-14 Collection Control Parameters 6-14
Figure 6-15 Collection Valve 6-15
Figure 6-16 Collection Settings 6-16
Figure 6-17 UV Input Collection Settings 6-17
Figure 6-18 Select Collection Type 6-18

Figure 6-19 mV Threshold Collection 6-19
Figure 6-20 mV Threshold Time Windows 6-20
Figure 6-21 Slope Threshold Collection 6-21
Figure 6-22 Select Slope 6-22
Figure 6-23 Timed Collection 6-24
Figure 6-24 Collection Times 6-26
Figure 6-25 Create Sequence 6-29
Figure 6-26 Select Auto Sequencing 6-29
Figure 6-27 Sequence Generator 6-30
Figure 6-28 Injection Sequence Finished Options 6-31
Figure 6-29 Create Sequence 6-32
Figure 6-30 Injection Sequence 6-32
Figure 6-31 Add Injection 6-33
Figure 6-32 Injection Sequence Finished Options 6-34
Figure 6-33 Select Stacked Inj. Builder 6-35
Figure 6-34 Stacked Prep Builder 6-36
Figure 6-35 Stacked Injection Instructions 6-37
Figure 6-36 Stacked Injection Cursor Instructions 6-38
Figure 6-37 Stacked Injection Cycle Time Determination 6-39
Figure 6-38 Total Elution Time Determination 6-40
Figure 6-39 Stacked Sequence Parameter Example 6-41
Figure 6-40 Injection Sequence Finished Options 6-41
Figure 6-41 System Settings 6-43
Figure 7-1 Select Collection Valve 7-4
Figure 7-2 Turn on Collection 7-5

List of Tables

Table 1-1	Recommended Co-Solvents	1-9
Table 1-2	Acceptable Additives	1-10
Table 1-3	Fluids not Recommended	1-11
Table 2-1	Work Space Requirements	2-5
Table 2-2	Customer Supplied Materials	2-7
Table 2-3	Power Supply Requirements	2-8
Table 2-4	CO_2 Requirements	2-10
Table 3-1	Serial Connections	3-2
Table 6-1	Collection Settings	6-18
Table 6-2	Slope Threshold Collection Settings	6-21
Table 6-3	Timed Collection Settings	6-24
Table 6-4	Sequence Parameters	6-30
Table 6-5	Add Injection Parameters	6-33
Table 6-6	Stacked Prep Builder Parameters	6-36
Table 7-1	SFC Prep 350 Maintenance Schedule	7-2

Chapter 1
Safety

Introduction

This chapter provides important safety information about the Thar Instruments SFC Prep 350 supercritical fluid chromatography system. The information and warnings contained in this chapter must be followed to properly operate the SFC Prep 350 system in a safe and successful manner. The SFC Prep 350 shipping and storage guidelines, safety features, electrical considerations, and operating considerations are provided.

WARNING	**The user should be made aware that if the equipment is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.**

Declaration of Conformity

The Declaration of Conformity for each module is located in Appendix V.

WARNING	**Thar Instruments will not accept any liability for damages directly or indirectly caused by connecting this instrument to devices which do not meet relevant safety standards.**

Shipping and Storage

The SFC Prep 350 supercritical fluid chromatography system has been designed for indoor use. It may be shipped and stored for brief intervals at temperatures between 0 and 55°C (32 – 130°F) without degradation of its safe operation. However, it is recommended that the unit stay within 15 and 35°C (60 - 95°F) during shipping and storage for optimal performance once installed. Also for optimal performance, relative humidity levels should stay at or below 60%.

Whenever it is likely that the protective safety mechanisms of the SFC Prep 350 system have been impaired due to inappropriate packaging, shipping or misuse, the SFC Prep 350 shall be made inoperable and shall be secured against further operation. Protective safety mechanisms are likely to be impaired if, for example, the SFC Prep 350 system:

- Shows visible damage.
- Has been subjected to prolonged storage under unfavorable conditions.
- Has been subjected to severe transport stresses.
- Exhibits mechanical failure by not performing an intended function.

Safety Requirements

Observe all warning labels on the system. Never remove warning labels. Do not operate the system with damaged or leaking equipment.

Safety Features

P-350/200 Pumps

The pressure rating of the system is adjusted for operation at various pressure and temperature conditions. Normally, the rated operating pressure of the tubing is 6000psi and rupture disc is 4800psi at 23°C (72°F).

Maximum Pressure Alarm

The P-350 pump contains a pressure transducer which senses the pressure at the pump outlet. If the pressure exceeds the alarm setpoint, the pump will stop and an audible beep will sound to alert the operator. When the pressure drops below the alarm setpoint, the audible alarm will stop. The pump will not restart automatically.

Rupture Disc

The P-350 and P-200 pumps are equipped with a rupture disc which is designed to open, burst, if the pressure exceeds the disc limit.

Pressure rating P-350 320bar

Pressure rating P-200 380bar

Threaded vessel enclosures 420 bar

Temperature alarm maximum Heat Exchangers 60C

Safety Sensors/Alarms

Thar recommends the following external sensors/alarms for your SFC system:

- Carbon dioxide (CO_2) sensor and alarm to provide warning for large leaks and possible dewar/cylinder malfunctions.
- Oxygen sensor/alarm to cover carbon dioxide leaks that cause the oxygen level to fall below safe levels. This will also cover nitrogen (N_2) leaks from other equipment and N_2 leaks from drying product containers after cleaning.
- LEL (Lower Explosive Limit) sensor/alarm for accidental spills of co-solvent and/or leaks that may contain solvent in an aerosol-like state.

Before proceeding with any monitoring safety configuration, check with your Environmental Health and Safety (EHS) Manager regarding applicable local, federal and international safety regulations and requirements.

Additional Warnings

WARNING	**(If applicable) Prior to opening the column oven door, turn off the oven temperature zone. Co-solvent fluids such as methanol are moderately flammable. Leaks, when confined to an enclosed space, may create a fire hazard.**

WARNING	**(If applicable) When checking fittings for leaks inside the column oven to ensure no leakage of flammable fluids from the fittings inside the column oven:** • **Turn off the oven heater.** • **Use 100% CO_2 mobile phase**

Flammability Warning

The SFC Prep 350 system has an optional column oven to help maintain the temperature stability of the chromatographic separation. Leaks of flammable mobile phase into this oven could cause flammable or explosive mixtures to form. The oven components can achieve temperatures > 180℃. These temperature le vels exceed the flash point of many flammable vapors. Avoid using any solvent with ignition temperatures in this range. Always check for leaks prior to operating the system in an unattended manner.

WARNING	**Thar recommends that you DO NOT use a highly flammable solvent as any part of the mobile phase.**

Electrical Considerations

The operating voltage of your SFC Prep 350 system was configured at the factory to operate on the standard voltage for your location. If the system is moved to an area using a different line voltage, the system must be reconfigured for the correct voltage. Contact Thar, or your dealer, for information on how to change the operating voltage.

Before powering on the SFC Prep 350 components, ensure that all power cords are connected to an appropriate source of power. The power cord should only be inserted into a socket outlet provided with a protective earth (ground) contact. The protective action of this connection must not be negated by the use of an extension cord without a ground conductor. Any interruption of the protective grounding conductor or disconnection of the protective earth ground could cause a fault condition that may result in personal injury. Thar Instruments assumes no responsibility for improperly grounded receptacles or power cords provided by the end user. All wiring to the supply outlet must meet local electrical codes. Additional details about electrical requirements and electrical connections can be found in Chapters 2 and 3, respectively.

The components in the SFC350 were designed to operate on both 50 and 60 Hz. There are no changes necessary to switch from 60 to 50 Hz. The waterbath that comes with the system is frequency specific and should not be used with a different Hz rating until contacting Thar Service.

 **WARNING** **VOLTAGE**	**Improperly grounded receptacles pose a risk of electrical shock.**

 **WARNING** **VOLTAGE**	**Do not operate the system with damaged line cords.**

Operating Considerations

Observe good laboratory practice when using the SFC Prep 350 system. Always keep in mind the following safety practices:

- Familiarize yourself with proper handling, storage, and disposal of all chemicals used with your SFC system. Refer to the Material Safety Data Sheet (MSDS) for each solvent you use, and know its chemical properties.
- Wear the appropriate personal protective equipment when working with any type of hazardous chemical.
- Wear eye protection while near the instrument to protect eyes from possible failure of column seals or fittings.
- Ensure that the system is depressurized before loosening any fittings in the CO_2 flow path.
- Exercise caution when accessing components. Internal and external surfaces of the components can become very hot or very cold during normal operation and direct contact could cause injury. The internal oven compartment is capable of reaching temperatures high enough to cause burns and the internal surfaces do not cool instantly upon opening the door. Internal surfaces such as heat sinks, valves, tubing, and fittings may become very hot or extremely cold, as low as -40°C.

WARNING	**Internal and external surfaces of the components can become extremely hot or cold during normal operation. Use caution when accessing any component.**

WARNING	**The SFC Prep350 System poses a potential rapid decompression hazard. Exposure to rapidly expanding fluids can cause injuries, including frostbite.** • **Exercise caution when opening fittings to prevent the possibility of being sprayed by expanding fluid or solvents.** • **Always depressurize the system before attempting to change or work on the bulk fluid supply.** • **Use only supply lines constructed of the appropriate material.**

Chemical Considerations

Always practice good chemical hygiene. Familiarize yourself with proper handling, storage, and disposal of all chemicals used with your SFC system. Refer to the Material Safety Data Sheet (MSDS) for each solvent you use, and know its chemical properties.

Solvents used in the mobile phase should be compatible with the materials in the SFC Prep 350 flow path including stainless steel, PEEK, and Teflon®.

CAUTION	**Thar assumes no responsibility for the use of untested or undefined fluids.**

Bulk Fluids

A bulk fluid is a fluid that makes up more than 50% of the mobile phase. Carbon dioxide is the **only** fluid recommended for use as a bulk fluid in the SFC Prep 350 system.

WARNING	**Carbon dioxide exhibits three primary hazards for humans:** • **Toxicity at levels above 10,000 ppm (PEL 5000 ppm).** • **Frostbite from uncontrolled release of pressurized CO_2 to atmosphere or contact with accumulated dry ice at a leak site.** • **Asphyxiation due to displacement of oxygen.**

As noted previously (see External CO_2 Monitoring, p. 1-4), Thar recommends installation of a CO_2 Ambient Air Sensor/Alarm unit (P/N 16001267) to ensure compliance with OSHA PEL for CO_2 in locations where CO_2 is used or stored. This is instead of, or in addition to, an oxygen monitor. Monitors purchased from an alternate source should be capable of detecting CO_2 levels to 10,000 ppm and provide an alarm at 5000 ppm.

Co-Solvents

Packed column SFC is a form of normal phase chromatography where the easiest-to-elute solutes are the most nonpolar. Pure fluids like carbon dioxide are poor solvents for polar solutes. To increase the range of solutes amenable to SFC, a more polar co-solvent or modifier is added to the mobile phase. Most organic solvents are viable co-solvents for packed column SFC. Few organic solvents are corrosive or detrimental to the components of the SFC Prep 350 system. However, several relatively nonpolar solvents corrode stainless steel, which is the recommended material for the majority of tubing in the SFC Prep 350 system. Methylene chloride (dichloromethane) is one such corrosive nonpolar solvent that is *NOT recommended* for use in the SFC Prep 350 system.

The most commonly used co-solvent in the SFC Prep 350 system is methanol. Methanol is a more polar solvent that is completely miscible with carbon dioxide.

CAUTION

Always check the compatibility of the organic modifier with the column phase in use. Some column phases will be destroyed with improper selection of co-solvent.

Recommended Co-Solvents

Co-solvents recommended for use in the SFC Prep 350 system are listed in Table 1-1 (below).

Table 1-1 Recommended Co-Solvents

Acetonitrile	Hexane
Chloroform	Isopropyl alcohol
Ethanol	Methanol

In infrequent cases, the co-solvent composition may exceed 50% of the mobile phase composition. Moving significantly above this composition can rapidly decrease the advantage of CO_2 based separations. Whenever possible, users should attempt to stay below 50% co-solvent composition.

WARNING

The recommended modifiers are Class 1B flammable materials.

Additives

The peaks of very polar solutes often tail. If for process reasons it's necessary to improve peak shape, a third component, an *additive* can be added to the mobile phase. Additives are usually too polar for direct addition to carbon dioxide alone. The additive is generally added to the co-solvent at a concentration of less than 1% of the co-solvent composition. The co-solvent and additive are then pumped together and mixed with the bulk fluid.

CAUTION

Some additives can be corrosive to columns and the column manufacturer's guidelines must be followed.

WARNING

Most additives are strong acids or strong bases.

- **Wear appropriate personal protective equipment (PPE) when handling them.**
- **Ensure that proper safety equipment is available including an eyewash station and a safety shower.**

Acceptable Additives

Table 1-2 lists acceptable additives for use in the SFC Prep350 system.

Table 1-2 Acceptable Additives

Acetic acid	Triethylamine
Trichloroacetic acid	Dimethyl ethyl amine
Trifluroacetic acid	Propylamine
Citric acid	Isopropylamine
Dimethylamine	Ammonium acetate
Trimethylamine	

Fluids Not Recommended

While this list is not complete, the fluids listed in Table 1-3 are neither recommended nor supported as bulk fluids or co-solvents in the SFC Prep 350 system.

Table 1-3 Fluids not Recommended

Ammonia	Dimethyl ether
Benzene	Dimethyl sulfoxide
Brominated solvents	Ethane
Butane	Ethylene
Carbon disulfide	Methylamine
Chlorinated solvents	Methylene chloride
Chloroform	Methyl t-butyl ether
Diethyl ether	Nitrous oxide
Di-isopropyl ether	Propane
Dimethyl acetamide	
Aqua regia	Bromine
Chlorine anhydrous	Copper chloride
Ferric chloride	Ferrous chloride
Freon 12	Guanidine
Hydrobromic acid	Hydrochloric acid
Hydrofluoric acid	Hydrofluorsilicic acid
Hydrogen peroxide	Iodine
Mercuric chloride	

Circulating Bath

Never operate the circulating bath without coolant in the bath. The minimum requirement is a 50/50 mixture of ethylene glycol and water as coolant

Collection Container Requirements

Thar supplies pressure-rated 2 liter bottles with specially designed caps for collection and waste. The bottles are rated for 0.4 bar and the caps are designed to allow CO_2 venting to avoid a pressure buildup in the bottle.

Use of different size collection containers requires prior consultation with your Thar representative.

WARNING	**Always inspect glassware for any visible damage or defects prior to use.**

WARNING	**The CO_2 supply cylinder should be secured with safety strapping prior to connecting to the system.**

Chapter 2
Hardware Overview

Introduction

This chapter provides an overview of the SFC Prep 350 hardware and installation site requirements. Detailed information about communication, electrical and plumbing connections is provided in later chapters.

SFC Prep 350 System Components

The SFC Prep 350 System contains the following components:

- System Overview (Pre-collection):
  - High pressure CO_2 pump
  - Cooling heat exchanger and circulating bath
  - Mass flow meter
  - High pressure co-solvent pump
  - High pressure static mixer
  - Pre-column heat exchanger
  - High pressure valve
  - Injection module
  - Automated back pressure regulator (ABPR)
  - Trailing heat exchanger
- System Overview (Post Collection)
  - High pressure fraction collection module
  - Fraction Collectors
  - Manual back pressure regulator (MBPR)
  - Fraction Collection Bottles with custom designed venting caps
- Control system and software:
  - PC with keyboard, mouse and multi-port Rocketport or RS-232 card
  - LCD monitor
  - Pre-loaded and configured SuperChrom software package

Figure 2-1 SFC Prep 350 System

CO_2 Pump

The CO_2 high pressure pump provides efficient pumping of CO_2 by incorporating low dead volume head and self priming check valves.

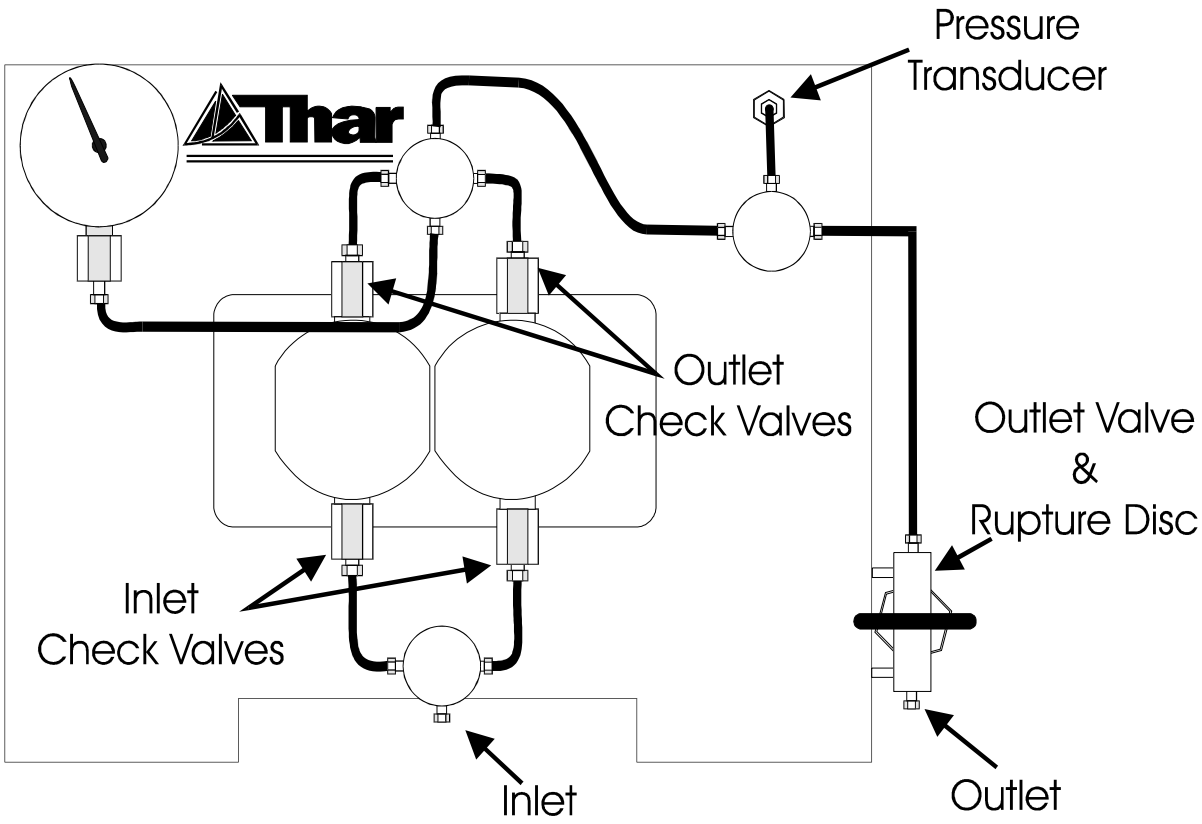


Figure 2-2 CO_2 Pump

Cooling Heat Exchanger and Circulating Bath

The heat exchanger is used to cool and liquefy CO_2 before it enters the CO_2 pump. The cooling bath circulates coolant through the pump heads and cooling heat exchanger (HE1).

Mass Flow Meter

The mass flow meter measures the liquefied carbon dioxide mass output from the CO_2 pump. The feedback from the Mass Flow Meter helps control the pump and maintain an accurate and consistent CO_2 flow rate.

Co-Solvent Pump

The co-solvent or modifier pump is calibrated to deliver a consistent amount of liquid in the specified operating range of the SFC Prep350

Static Mixer

The high pressure static mixer blends the CO_2 and co-solvent flow streams.

Injection Module

The injection module introduces the sample to the injection valve. Two injection configurations are available (1) modifier stream injection and (2) combined stream injection. Plumbing connections for each configuration are provided in Chapter 4.

UV Detector

The UV detector is a variable wavelength detector. The peak detector uses the output from the UV detector to trigger fraction collection.

Fraction Collection Module

The fraction collection module uses two high pressure rotary valves to allow collection of up to 12 compound fractions.

Installation Requirements

Work space requirements are provided in Table 2-1. The SFC Prep 350 system should installed at a comfortable operating height. Space on the front and sides of the system should be available to access components.

The water bath should be placed on the floor next to the SFC Prep 350 system. The water bath requires ventilation for proper operation. Position the water bath to not impede air intake and discharge.

Work Space Specifications

Work space requirements are summarized in Table 2-1 and Figure 2-1 (next page).

Table 2-1 Work Space Requirements

Factor		Requirement	
Workspace	**System**	Width:	107 cm (42 inch)
		Depth:	92 cm (36 inch)
		Height:	135 cm (53 inch)
		Weight:	123 kg (270 lb)
	Water Bath (Floor)	Width:	40 cm (16 in)
		Depth:	43 cm (17 in)
		Height:	63 cm (25 in)
		Weight:	65 kg (145 lb)
	Computer	Width:	16.8 cm (6.6 in)
		Depth:	45.0 cm (17.7 in)
		Height:	45.6 cm (17.9 in)
		Weight:	16 kg (35 lb)
	Computer Monitor (with stand)	Width:	44.6 cm (17.6 in)
		Depth:	18.7 cm (7.3 in)
		Height:	37.0 cm (14.6 in)
		Weight:	5 kg (11 lb)
Clearance*	**System**	Front:	
		Rear:	15 cm (6 in)
		Right side:	
		Left side:	
		Top:	
	Water Bath (Floor)	Front:	10 cm (4 in)
		Rear:	10 cm (4 in)
		Right side:	10 cm (4 in)
		Left side:	10 cm (4 in)
		Top:	10 cm (4 in)

*Additional clearance of 60 cm (24 in) to the left of the unit is recommended for installation purposes. This additional space is not required once the system is installed.

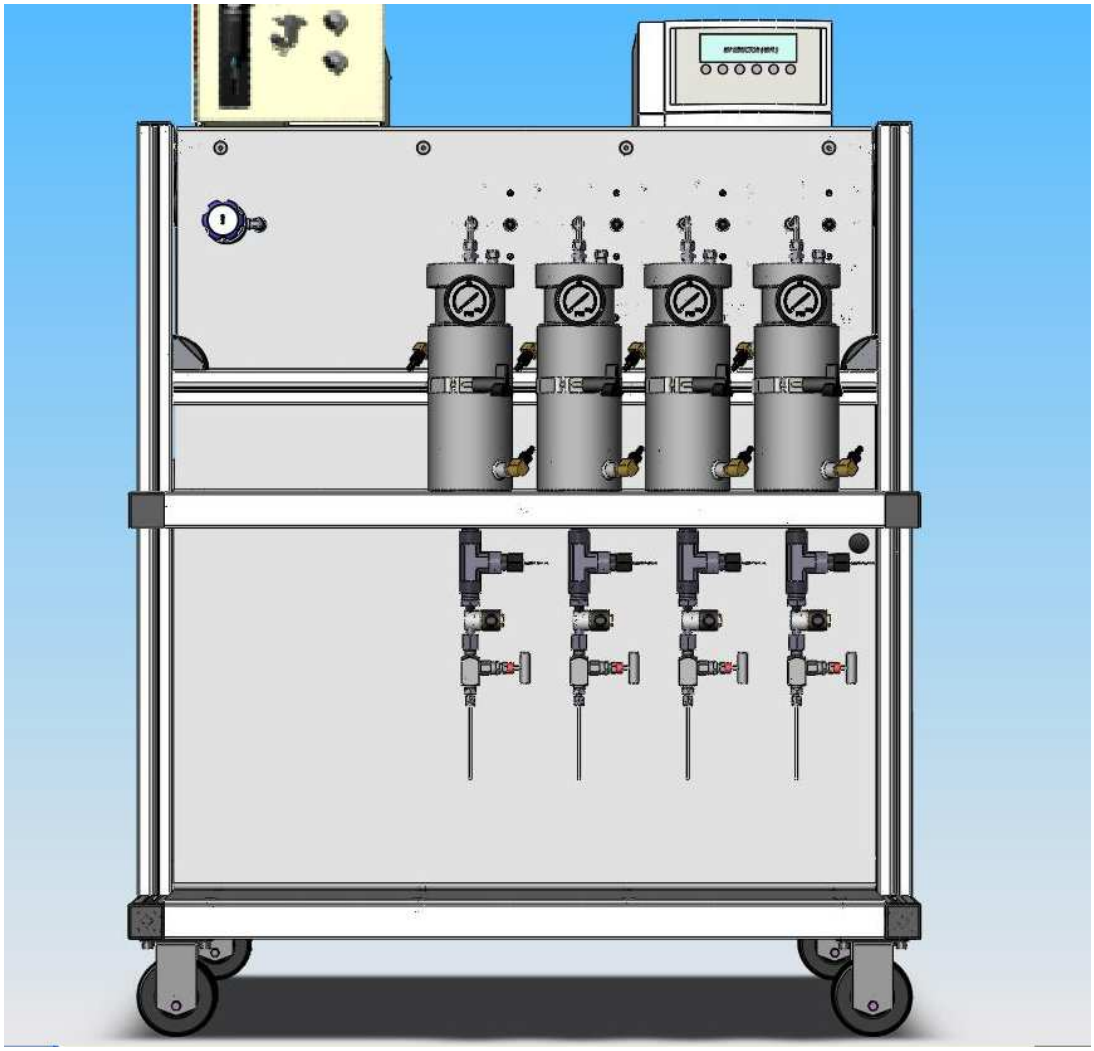


Figure 2-3 SFC350 System Layout

Customer Supplied Materials

To perform an installation of the SFC Prep 350 system the user must provide the materials listed in Table 2-2.

Table 2-2 Customer Supplied Materials

Material	Comments
3 cylinders of CO_2 with dip tube	Minimum required for system checkout
13-liter supply of 50% ethylene glycol and water	Required for the cooling bath operation
8-liter supply of co-solvent	Minimum required for system checkout
Sample in sufficient quantity for testing	Minimum required for system checkout
Ventilation tubing	1/4“ compression fitting
Column min 3 or 5 cm diameter	For testing
Completed site prep checklist	See SFC350 Site Preparation Instruction and Installation Procedures (MN50)

Electrical Requirements

Grounding and power supply requirements for the SFC Prep 350 are provided below.

Grounding

When plugged into a properly wired receptacle, the three-conductor power line provides both power to and grounding of the instrument. Proper receptacle grounding should be periodically verified. Any interruption of the protective grounding conductor, or disconnection of the protective earth ground, could cause a fault condition that may result in personal injury. Thar assumes no responsibility for improperly grounded receptacles or power cords provided by the end user.

WARNING

VOLTAGE

Improperly grounded receptacles pose a risk of electrical shock.

Power Supply Requirements

The SFC Prep 350 system is configured at the factory for the power ordered. Power supply requirements are summarized in Table 2-3. Refer to “Electrical Considerations” in Chapter 1 for additional details.

Table 2-3 Power Supply Requirements

System	System Current/Plugs	Water Bath* Current/Plugs	Computer Current/Plugs	Monitor Current/Plugs
120 VAC	28 Amp (total)/ 2 ea L5-20 plug	14 Amp/ 1 ea 5-15 plug	1.1 Amp/ 1 ea 5-15 plug	.9 Amp/ 1 ea 5-15 plug
220 VAC	18 Amp (total)/ 2 ea CEE-7 plug	7 Amp/ 1 ea CEE-7 plug	1.1 Amp/ 1 ea CEE-7 plug	.9 Amp/ 1 ea CEE-7 plug

*The water bath should be on its own circuit.

Required power cords are supplied with the system. Plugs for North American (NA) and European installations are shown in Figure 2-4.

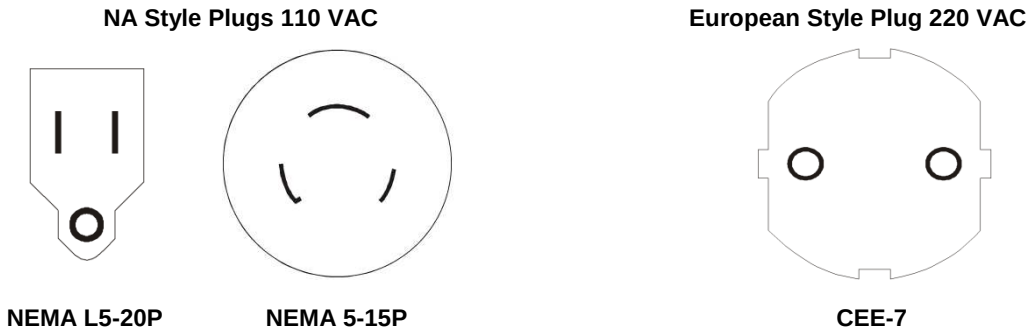
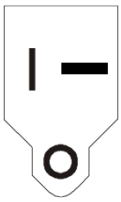


Figure 2-4 NA and European Style Plugs

NA Style Plugs 220 VAC



Chemical Requirements

Solvents used in the mobile phase should be compatible with the materials in the SFC Prep 350 flow path including stainless steel, PEEK, and Teflon.

Carbon Dioxide

Carbon dioxide is the only bulk fluid Thar Instruments recommends for use in the SFC Prep 350 system. CO_2 requirements are summarized in Table 2-4. CO_2 purity should be industrial grade or better depending on your application requirements. The CO_2 supply tank must have a dip tube to pull liquid CO_2 from the bottom of the tank. Use of low pressure tanks such as dewars, is not acceptable. Tank must be capable of delivering a minimum of 50 bar. Tanks should not be stored in cold locations.

Table 2-4 CO_2 Requirements

Purity	Industrial grade or better
Supply cylinder	Dip tube cylinder
Pressure	Minimum 50 bar
Storage temperature	Ambient

Co-Solvents

Co-solvents or modifiers are organic solvents used as a component of the SFC mobile phase. They are required for proper SFC fraction collection. Co-solvents should be HPLC grade or reagent grade.

In most cases, preparative SFC uses UV-Vis detectors; consider this when selecting an appropriate co-solvent. Refer to Chapter 1, “Safety” for recommended co-solvents and solvents to avoid.

Plumbing Requirements

CAUTION	**Particulate matter should not be introduced to the pump or damage to the check valve sealing surface may result. A filter should be placed directly upstream of the pump to capture any solids that may have formed.**

Venting Requirements

Ventilation is required for the SFC Prep 350. The SFC Prep 350 provides provisions for venting to a fume hood to the outside through a 1/4" compression fitting. Tubing connected to the fitting should be of sufficient strength to withstand the pressures. If the run is going to be greater than 5 feet or contain multiple bends larger tubing should be used to prevent restriction of the escaping gasses

WARNING	**Venting of the SFC Prep 350 is the sole responsibility of the end user.**

Chapter 3
Electrical Connections

Introduction

There are two types of electrical connections required to configure your SFC Prep 350 system: communication (Rocketport or RS-232 serial) and power cords.

Communication

Communication between modules and the PC is via Rocketport (or RS-232) serial and USB connections. All necessary cables are provided with the system.

Communication Cable Connections

Directions for connecting the communication cables for your SFC Prep 350 system are provided below.

CAUTION	**Failure to make the proper connections will result in a loss of communication with the device.**

1. Turn on the computer and allow it to complete initialization.

2. Connect the USB cable from the Peak Detector (Analog to Digital converter) to the computer USB port labeled Peak Detector. Connect the Rocketport to the computer. If the USB ports are not labeled, plug the cable into the first port. If you receive a message "Found New Hardware", unplug the cable and cancel the hardware installation.

CAUTION	**Do NOT allow Windows to install the USB/RS-232 converter or the Peak Detector. The required software was installed at the factory.**

3. Connect the USB cable from the analog interface card to the computer USB port labeled Peak Detector. If the ports are not labeled, plug the cable into the first port. If you receive a message "Found New Hardware", unplug the cable and cancel the hardware installation. Repeat the process in the next USB port.

4. All of the individual component connections between the converter and the unit will be already made. These connections listed below for reference only:

Table 3-1 Serial Connections

RS-232 Hub	To System
COM 1	P1 (Pump 1)
COM 2	ABPR1
COM 3	P2 (Pump 2)
COM 4	CN6
COM 5	UV detector
COM 6	Level Controller
COM 7	Injection module
COM 8	Collection module

Power Connections

Each module contains its own internal power supply. The system power connections should be connected to an appropriate power source. If applicable the power switches are located on the rear of each module. For additional information about power requirements refer to Chapter 2.

Chapter 4
Plumbing Connections

Introduction

This chapter provides

All required tubing is supplied with the system.

Pump Cooling Tubing

Thar CO_2 pumps require external cooling of the CO_2 for proper operation. Improper cooling of the pump heads and CO_2 will cause inefficient pump operation. The connection between the outlet of the pump heads cooling tubes and the inlet to the cooling heat exchanger will already be made.

1. Locate the “Outlet” port on the bath and slide the 5/16” ID Silastic Tubing (02321) from the HE1 inlet over the end of the fitting. The entire barbed section of the fitting must be covered or the tube may come off.
2. Locate the “Inlet” port on the water bath and slide the 5/16” ID Silastic Tubing (02321) from the CO_2 pump heads over the end of the fitting. The entire barbed section of the fitting must be covered or the tube may come off.
3. Fill the bath reservoir with a 50/50 mixture of antifreeze and water. Follow the antifreeze manufacturer’s recommendations to allow for a temperature of –20℃.
4. Turn on the bath and set the control temperature to 3℃.

External CO_2 Connections

1. Secure the CO2 supply cylinder with safety strapping.

WARNING	**Secure the CO_2 cylinder with safety strapping.**

2. Remove the cap and protective wrap from the tank valve.

3. Point the valve outlet away from all personnel and “burp” the cylinder for 2-3 seconds until liquid escapes. To “burp” the cylinder, open and close the valve in a fast, smooth motion. Wipe the valve clean and dry with a lint-free cloth.

4. Attach the inlet tubing assembly (05132) to the nozzle (ensure the Teflon disc seal is inserted into the CGA fitting prior to attaching); use a 1 1/8" open-end wrench to tighten.

NOTE	**The system is shipped with a CGA-320 fitting to connect to the CO_2 tank. Contact your gas supplier to determine if this is the standard connection for your area.**

5. Uncoil the tubing toward the rear of the system. Locate the CO_2 inlet fitting on the bottom shelf of the system and using a 9/16” backup wrench and 3/8” wrench connect the tubing to the fitting.

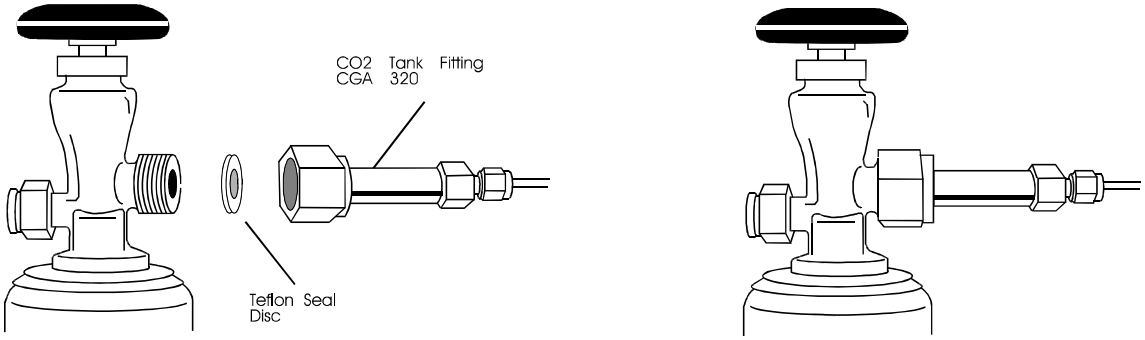


Figure 4-1 CO_2 Tank Fitting

Chapter 5
Operating the SFC Prep 350

Introduction

This chapter provides basic instructions for operating your SFC Prep 350. For information on method setup refer to Chapter 6.

WARNING	**The user should be made aware that if the equipment is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.**

System Startup

A summary of the recommended startup procedure for the SFC Prep 350 system is provided below.

1. Power on the system
2. Verify water bath temperature
3. Inspect collection bottles and vessels
4. Start SuperChrom
5. Setup UV detector*
6. Prime the co-solvent pump
7. Install/replace column
8. Turn on the CO_2 supply
9. Prime the injection module
10. Set the manual BPR pressure
11. Setup the Level Controller

*UV detector setup only required for initial system startup.

Power on the System

1. Ensure the computer and system are connected up to the correct power supply. Power on all of the components, including the PC and monitor. Note: Power switches are located on the rear of each SFC Prep 350 module.

2. Verify that all of the units are powered on.

3. Perform a normal Windows OS startup on the computer.

Verify Water Bath Temperature

Confirm the water bath temperature is set to 3℃.

Inspect Collection Bottles and Vessels

Inspect the collection bottles and vessels.

Collection Bottles

Fractions must be collected into suitable containers. Your system was shipped with 2L collection bottles for each cyclone including the waste cyclone. The bottles are rated for 0.4 bar of pressure and have custom designed caps that allow CO_2 to vent while avoiding a pressure buildup in the bottle.

Use of different size bottles requires prior notification to Thar. Depending of the version of caps that were delivered with your system there may be some modifications necessary to allow use of smaller bottles and maintain the large safety factor that Thar requires.

1. Inspect the vessels for defects.

2. Ensure the collection bottles are empty and clean before use. Inspect the vessels for buildup of solid material.

3. Replace any defective or unclean container.

4. Ensure the caps are tight on each container.

Open SuperChrom Software

1. Double click the SuperChrom icon on the desktop.

2. The User Login window displays. Click in the Login field; enter your Login and Password. Then click the **Login** button.

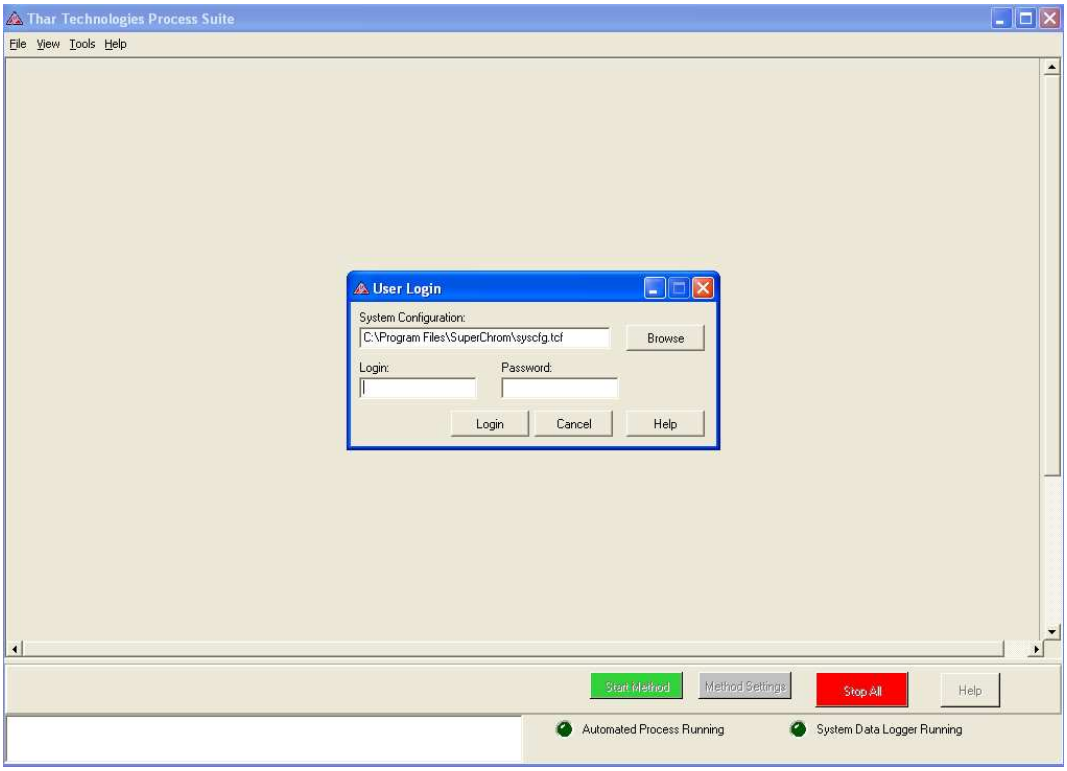


Figure 5-1 SuperChrom Login Window

NOTE	**The Login and Password fields are case sensitive.**

3. The SuperChrom software should open after (at most) a minute of communication with the system.

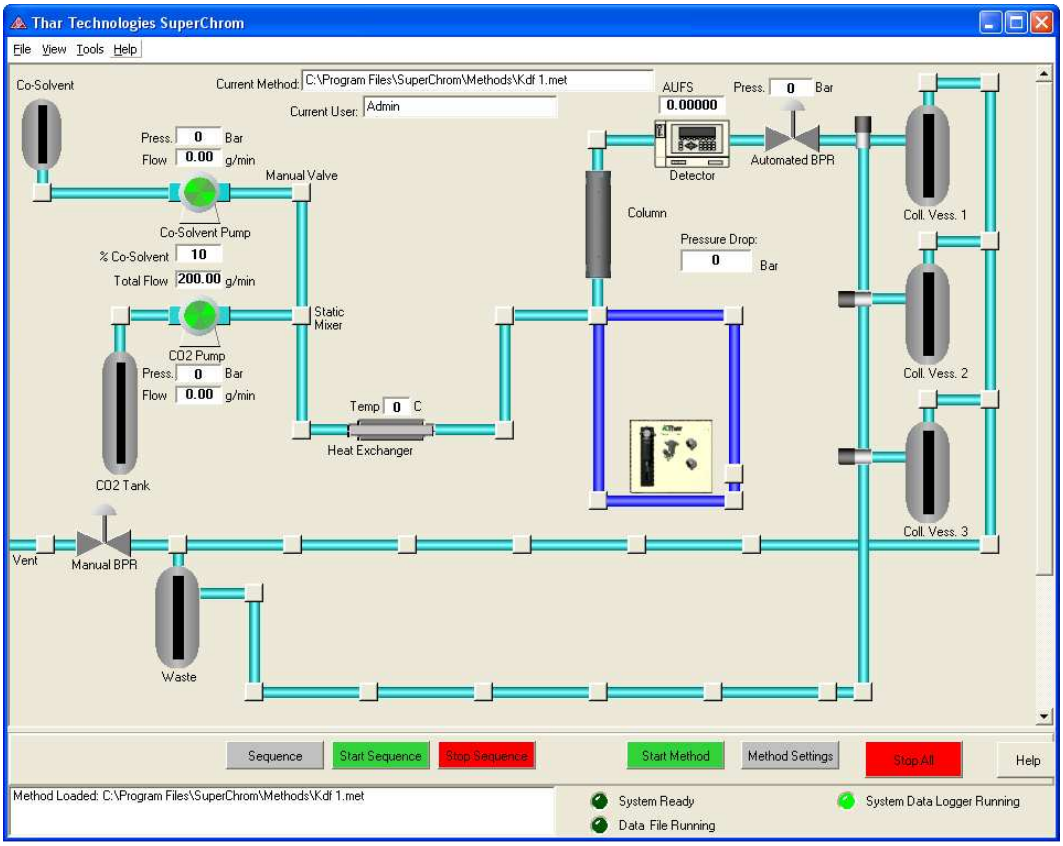


Figure 5-2 SuperChrom Main Screen

UV Detector Setup

1. Turn on the UV lamp and allow the detector to complete the wavelength validation process.

2. Press **Mode** on the UV detector touch screen display.

Figure 5-3 Select Mode

3. Then press **Setup**.

Figure 5-4 Select Setup

4. Confirm the Channel 1 signal is set to 100 mVFS.

Figure 5-5 Set Channel 1

5. Press **Next**. Ensure both lamp alarms are set to 0.

Figure 5-6 UV Lamp Alarms

6. Press **Next**. Confirm the Autorange multiplier is set to 0.2 and all others are zero

Figure 5-7 Set Autorange Multiplier

7. Press **Next**. Confirm the Concentration factor is set to 1.000.

Figure 5-8 Set Concentration Factor

8. Press the **ESC** (Escape) button.

9. In the SuperChrom main window, right-click on the detector and select **Detector Settings**.

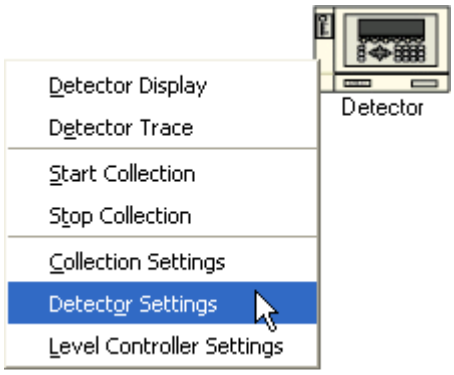


Figure 5-9 Detector Settings

NOTE **Alternatively, the UV Detector settings can be accessed by selecting View > UV Detector Settings.**

10. The UV Detector Settings screen displays.

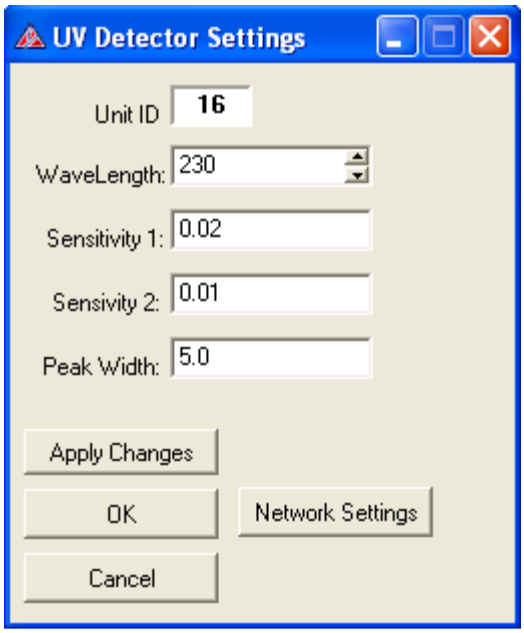


Figure 5-10 UV Detector Settings

11. Set the following:

UV wavelength	230 nm
Sensitivity 1 & 2	0.01 AUFS
Peak width	5.0 sec

12. Click **Apply Changes**.

13. Click **OK** to close the UV Dectector Settings screen and return to the main SuperChrom screen.

14. Double-click the Detector icon to open the UV Input screen.

15. Confirm the “Show Injection/Collection Marks” box is checked.

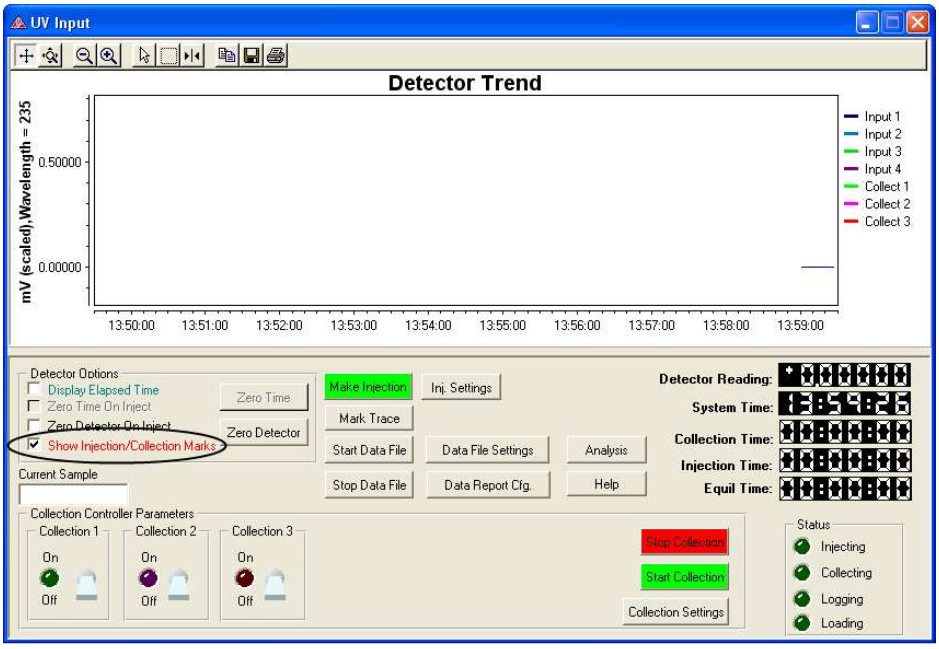


Figure 5-11 Injection/Collection Marks

16. Optional settings (not recommended for Stacked Injections). Checking the box activates the option:

    a. Display Elapsed Time - Displays the elapsed time instead of the actual time.

    b. Zero Time On Inject – Zeroes the time upon injection.

    c. Zero Detector On Inject - Zeroes the detector upon injection.

Prime the Co-Solvent (Modifier) Pump

The co-solvent pump lines must be primed for proper operation. The co-solvent reservoir should be covered and placed at the same level or slightly higher than the pump.

CAUTION	**Do NOT pump the co-solvent up from floor level.**

Follow the steps below to prime the solvent lines.

1. Place the filtered inlet line into the co-solvent reservoir. The entire filter must be immersed in the co-solvent.

2. Place a waste collection container at the end of the priming line to collect the solvent waste.

3. Close the MV1 valve which leads to the system and open the MV1 valve that leads to the priming line.

WARNING	**One side of MV1 MUST always be left open when the pump is running. Closing both sides of MV1 while the pump is running will cause the pump rupture disc to burst.**

WARNING	**Open the priming valve slowly. If there is any pressure in the system, it will bleed out through the valve.**

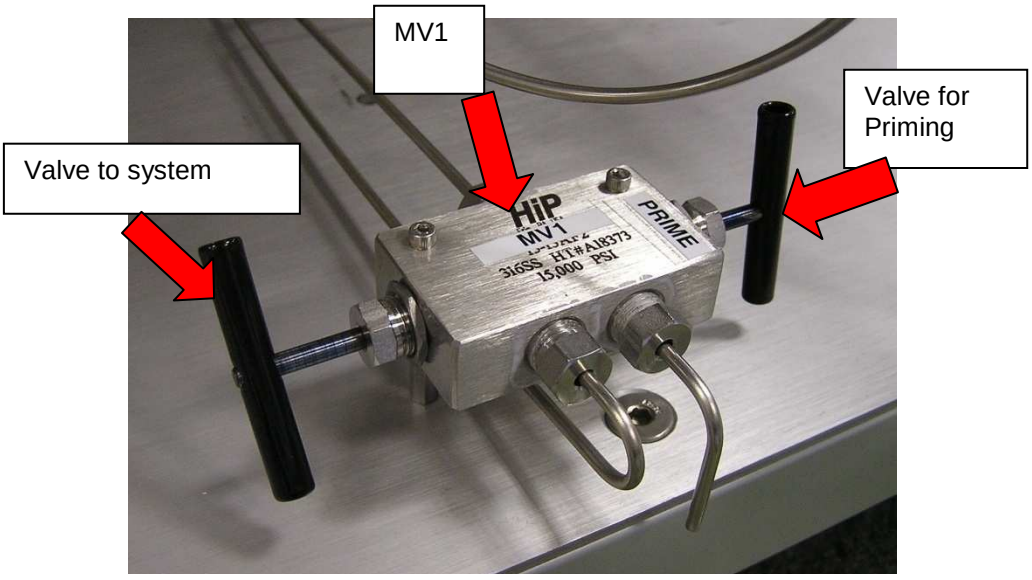


Figure 5-12 MV1

4. In the SuperChrom main screen, right-click on the Co-Solvent pump icon and select **Co-Solvent Pump Display**.

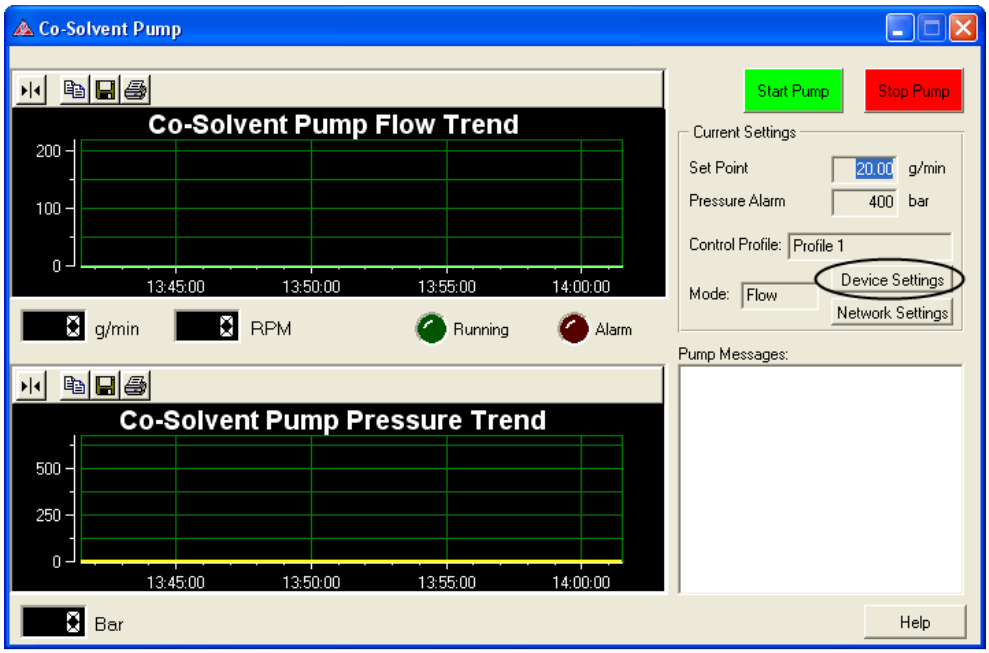


Figure 5-13 Co-Solvent Pump Display

5. In the Co-Solvent Pump display window click **Device Settings**.

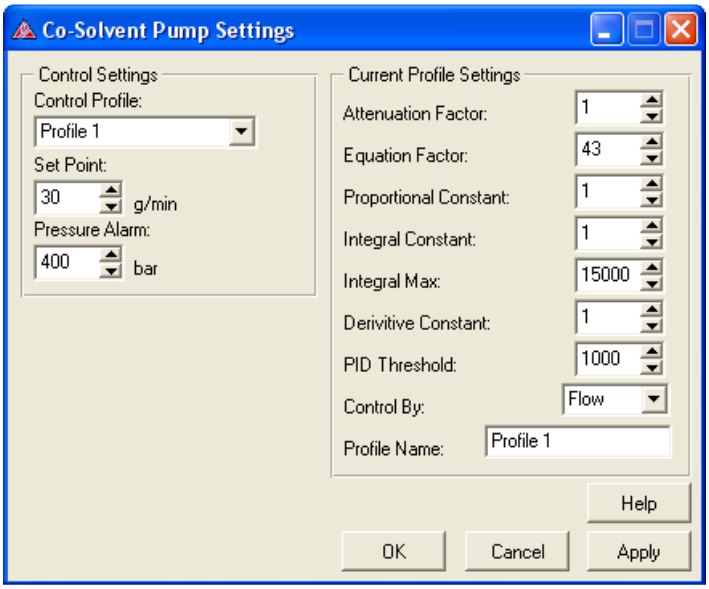


Figure 5-14 Co-Solvent Pump Settings

6. Enter a Flow Rate of 30 g/min.

7. Click **Apply**. Then click **OK** to close the pump settings window and return to the co-solvent pump display window.

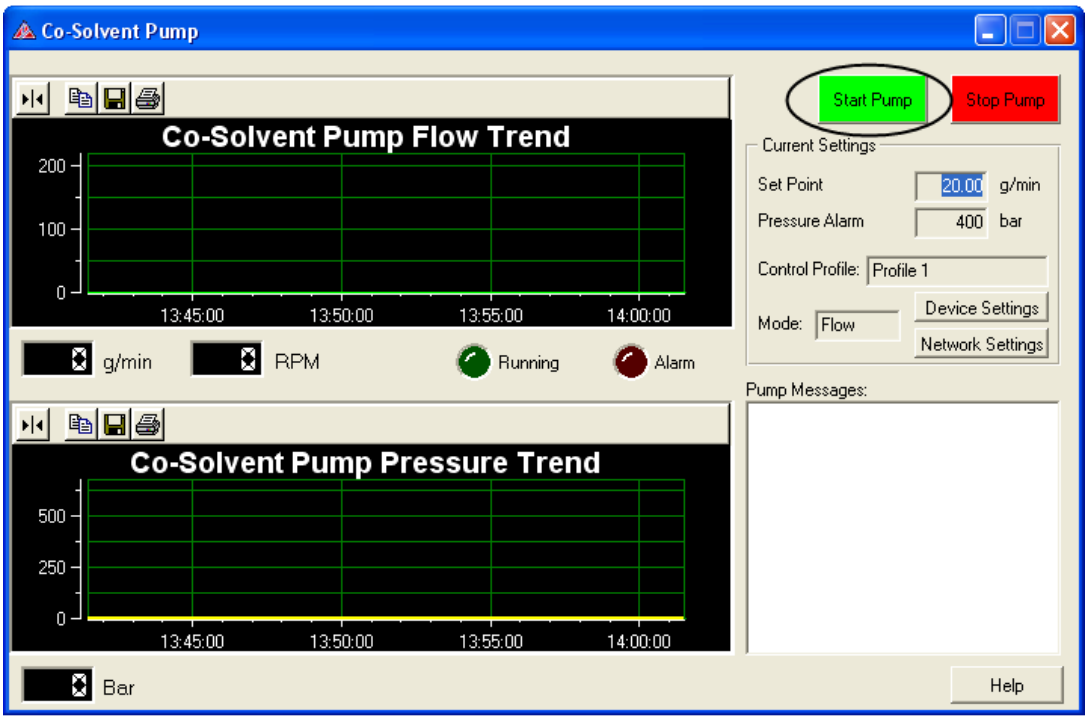


Figure 5-15 Start Pump

8. Click the **Start Pump** button.
9. Allow flow to continue until you observe no visible air bubbles in the outlet line.
10. Then click the **Stop Pump** button.
11. After the pump stops, close the Priming side of MV1 and open the System side of MV1.

Install/Replace Column

The SFC Prep 350 is equipped with 1/16” fitting that will fit standard chromatography columns. Before installing a column confirm that the column is rated for SFC conditions and/or the system operational conditions.

Columns should be installed or replaced with the instrument depressurized, no flow through the system, and (if applicable) the oven powered off.

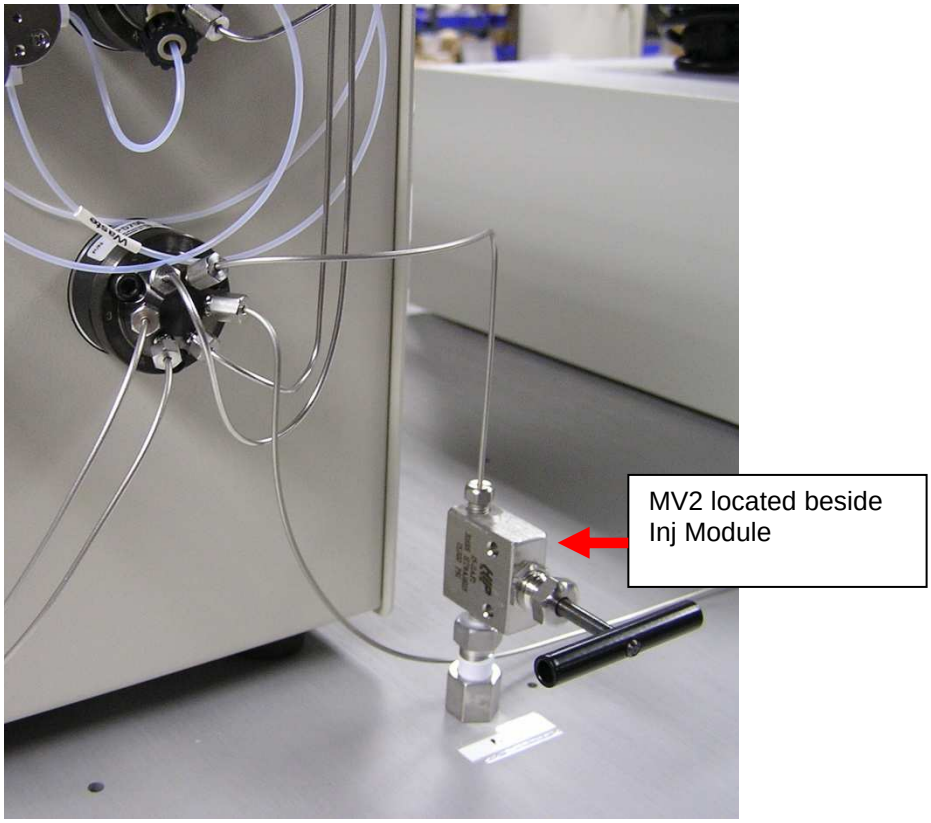
CAUTION	**Never crack the column inlet with liquid CO_2 in the system. This could destroy the column.**

Depressurize the System

If there IS pressure on the system:

1. Close MV2.

NOTE: newer systems may have the MV2 in a different location



2. Right-click on the Automated BPR (ABPR) icon and select **ABPR Settings**.

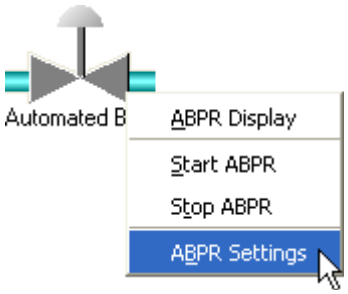


Figure 5-16 ABPR Settings

3. Change the ABPR Pressure Set Point to 0 bar.

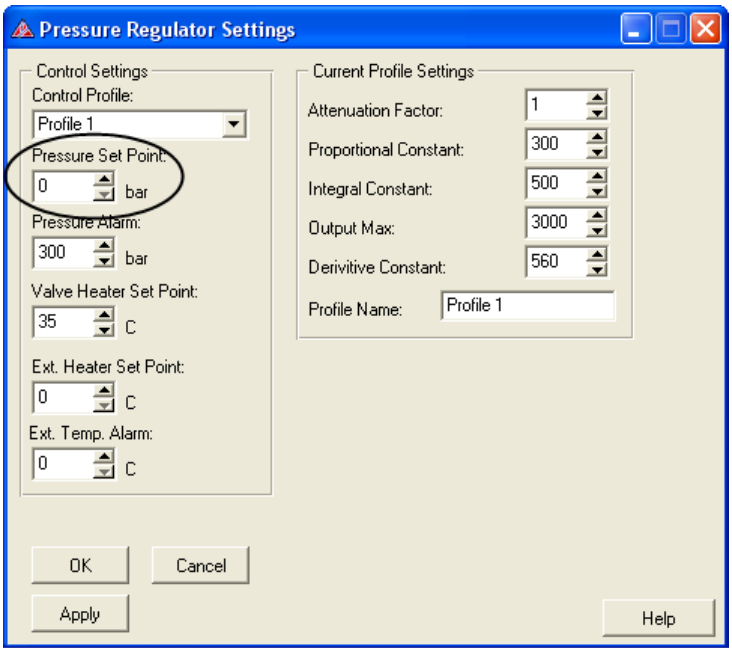


Figure 5-17 Pressure Regulator Settings

4. Click **Apply** and then click **OK**. Wait for the system to depressurize.

5. Slowly loosen the lines from the 1/16”union or the column. Allow these lines to vent fully.

Install the Column

When the system is depressurized:

1. Disconnect the lines from the 1/16” union or column.
2. Connect the line coming up from the Injection Module to the inlet of the column.
3. Connect the line going to the UV detector to the outlet of the column.
4. Open MV2 and check for leaks at the column fittings.

NOTE: newer systems may have the MV2 in a different location

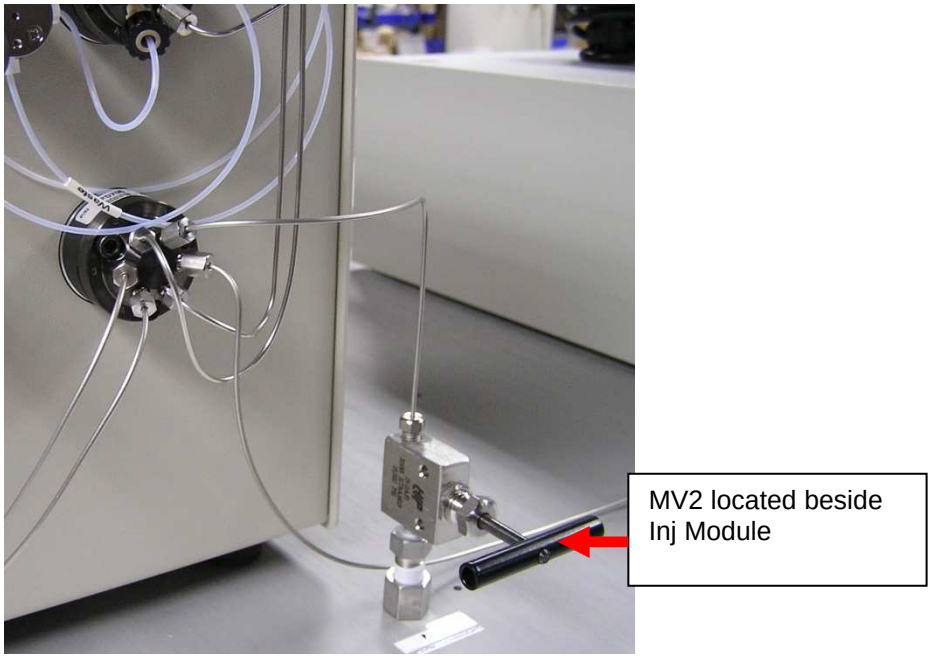


Figure 5-18 MV2

Turn on the CO_2 Supply

Use a soap solution or a designated leak check solution to check for leaks once the CO_2 has been turned on near:

1. The column fittings
2. The injection module fittings
3. Check any fitting that has frost around it-which is indicative of a leak

Injection Module Settings

1. Double-click the Injection Module icon on the main SuperChrom screen to open the Thar Injection Module window.

NOTE

Alternatively, the Thar Injection Module window can be accessed by either:

- **Selecting View > Thar Injection Module.**
- **Or by double clicking the UV detector icon and then clicking the Inj. Settings button on the UV Input screen.**

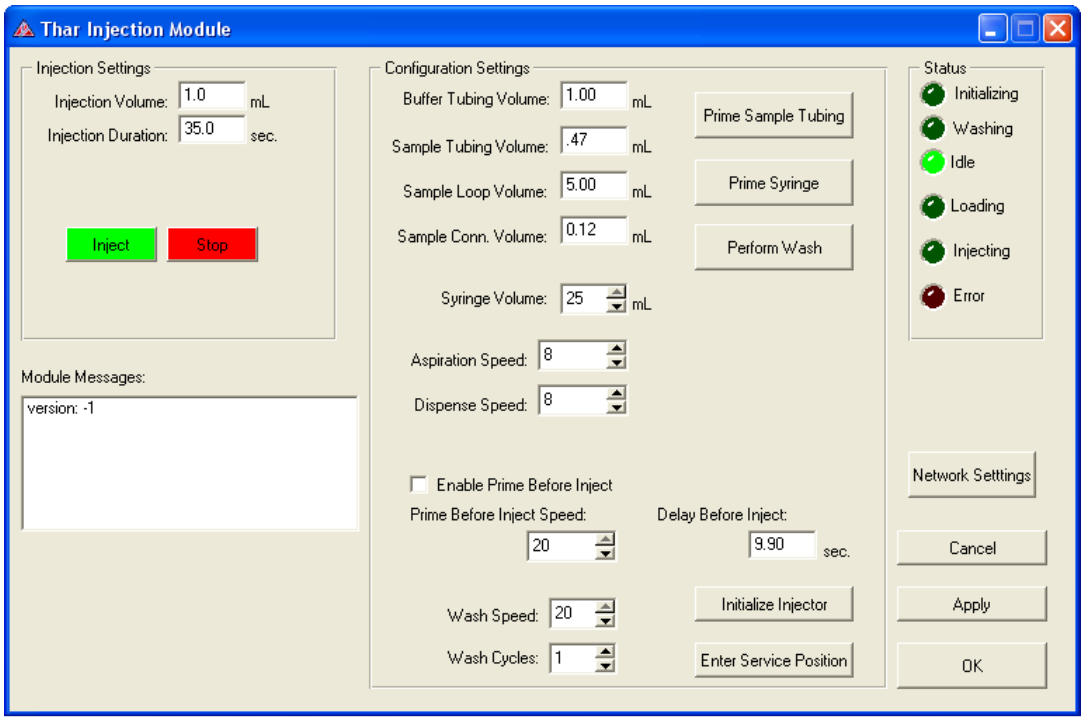


Figure 5-19 Thar Injection Module

2. Verify the sample loop size installed (5 mL loop is standard) matches the entry for "Sample Loop Size."

Sample Connection Size

The sample connection size (i.e. Sample Conn. Size) will vary slightly with each sample and sample solvent. The actual volume of the tubes and valves that make up the sample connection size is 95.7 µL. A safe setting would be between 0.10 mL and 0.15 mL. With some samples and solvents such as acetonitrile, the settings have been as high as 0.22 mL.

Injection Module Priming- **(mixed phase injections only)**

3. Place the solvent inlet line into a vessel containing rinse solvent.
4. Place both waste lines in a waste container.
5. Place the sample line into a vessel containing rinse solvent.
6. Select View > Thar Injection Module to open the injection module window.
7. When the syringe and the rest of the system have been properly primed, place the sample inlet line in the vessel containing rinse solvent.
8. If the Injection Module was not cleaned after its last use, remove the sample line from the rinse solvent, and place it in a separate waste container.
9. Click "Perform Wash" of sample line - remember the wash solvent comes through the sample line.

Injection Module Startup-(mixed phase only)

1. Make sure the Solvent line to the syringe is submerged in methanol.
2. Click **Prime syringe**
3. Place the sample line into a separate methanol container at the same height as the actual sample container.
4. Click **Prime Sample Tubing**
5. (while the system is running) Perform two 5 mL methanol injections at the same settings as the actual injections will be (30 sec duration, pushback value, speed, etc.)
6. When the two injections are complete, place the sample line into the desired sample solution.
7. Click **Prime Sample Tubing** (there may be a small amount of sample loss here).

CAUTION	**Do not perform test injections of less than 1 mL if you are doing a campaign of injections that are greater than 1 mL. Going from 1 mL to 5 mL injection on a 5 mL loop is acceptable.**

NOTE	**Doing a 0.25 mL injection and then going to a 5 mL injection volume will allow *some* sample into the buffer line for the 1st injection after the 0.25 mL test injection. After the first 5 mL injection, the injection module will work correctly again.**

Injection Module Priming- (modifier stream injections only)

8. Make sure the Solvent line to the syringe is submerged in methanol.
9. Click **Prime syringe**
10. Click **Prime Sample Tubing**
11. Make sure the **Enable Prime Before Inject** box is checked

Set the Manual BPR

The Manual BPR sets the pressure on the Fraction Collection Vessels. If the fraction collection vessels do not have pressure, the product will be lost. Follow the directions to set the MBPR installed on your system.

You must have 55 bar back pressure on the 1000 mL cyclones during collection.

High Pressure MBPR Instructions

Set the MBPR after 5 minutes of equilibration time at running conditions.

CAUTION	**For optimum performance there must be at least 55 bar of manual back pressure on the 1000 mL cyclones during collection.**

Figure 5-20 MBPR valve

This MBPR must be set to 55 bar to achieve optimum conditions. This will allow for a constant minimum 55 bar back pressure on the 1000 mL cyclones while in operation to achieve optimum collection conditions.

The most important pressure regarding the MBPR is the pressure on the cyclones while collecting, **trying to use 0bar on the cyclones will lead to product loss.**

Collection Vessels

When the system is at pressure, check the needle valves of each cyclone

Manually close all the needle valves on each cyclone

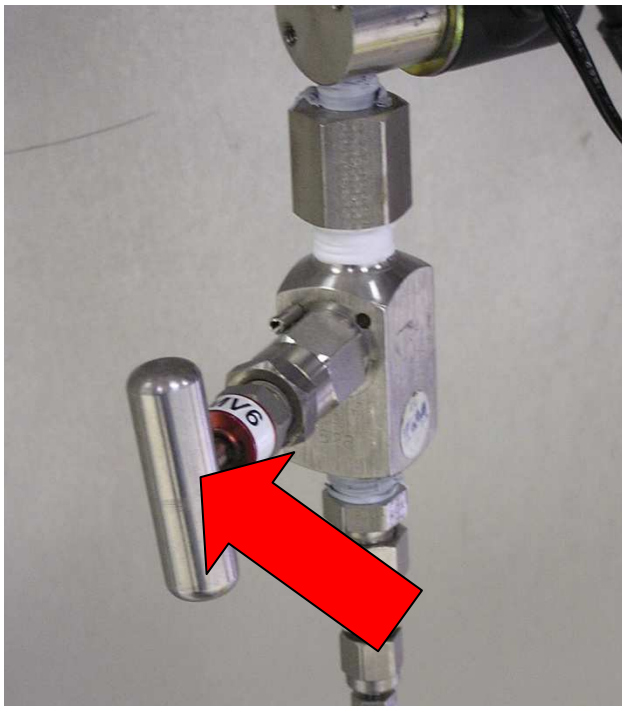


Figure 5-21 Cyclone Needle Valves

Turn On each Fraction Collection cyclone valve from the “UV Input” Screen to pressure check each one.

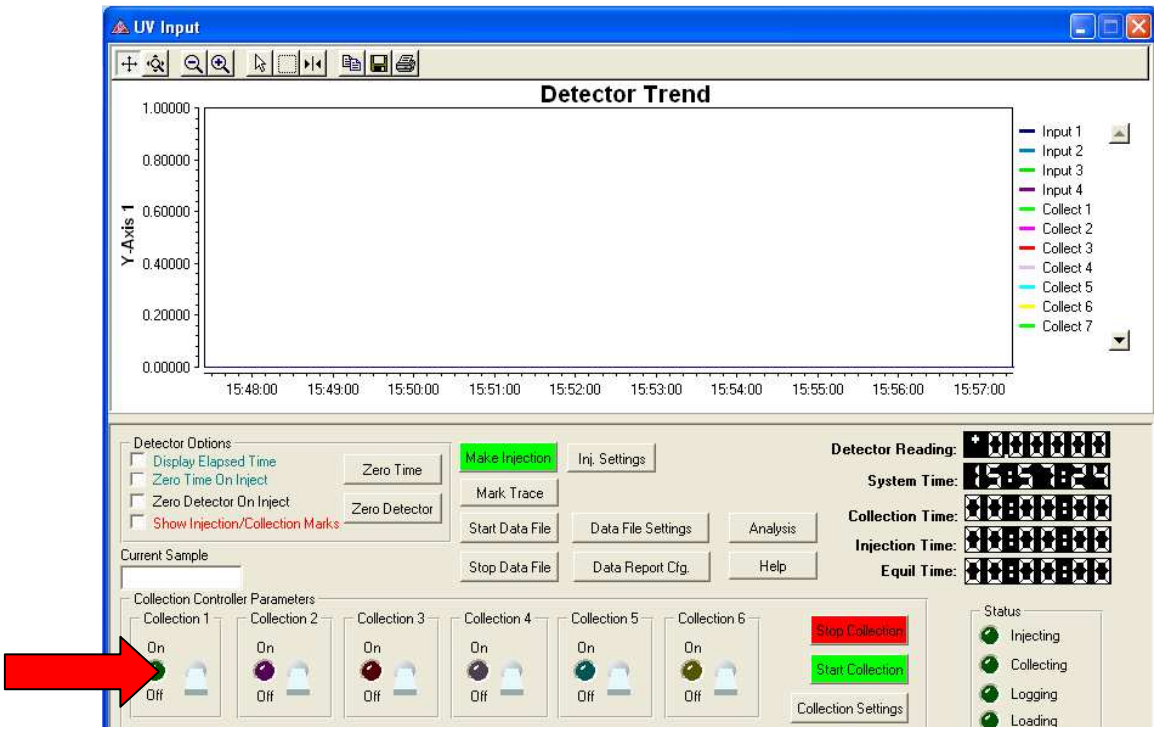


Figure 5-22 Manual Collection Switches

As each Collector is selected, make sure the Pressure is increasing in the correct fraction collector (FC)

Allow each Cyclone Collector to reach 55-60bar

On the main Superchrom screen Click the “STOP ALL” before continuing to the next step

For each cyclone
open each “Cyclone Drain” solenoid valve manually (green switches)

Figure 5-23 Manual Cyclone Drain Switches

Very slowly start opening each needle valve into each designated fraction collection container until CO_2 is observed venting into each bottle.

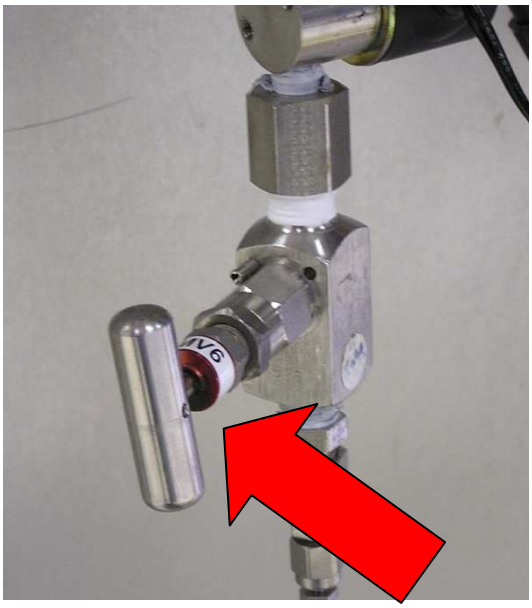
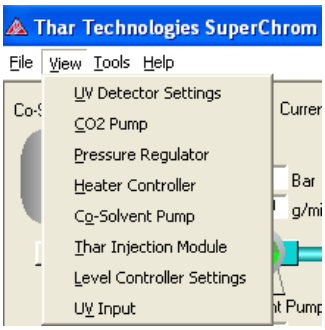


Figure 5-24 1L Cyclone Needle Valve

When needle valve is set, turn off each “Cyclone Drain” solenoid valve

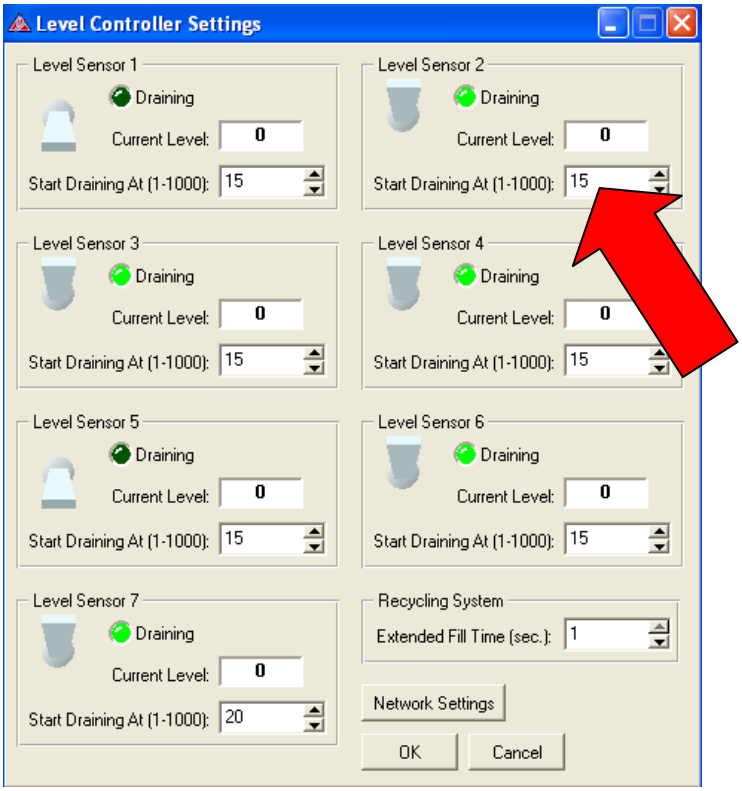
When each needle is set Start the Method and again allow to equilibrate for 2-5 minutes after the desired pressure is reached.

Click on “View” then Click on “Level Controller” to use the software’s manual drain function.



Set Level Controller Drain Levels

Set all Levels to drain between 15-20mL



Shutdown Instructions

Shutdown instructions for short term (e.g. weekend) and long term are provided below.

Short Term Shutdown

Short term shutdown is appropriate for intervals of a few days, e.g. a weekend.

1. Clean the system of residual product.
2. Perform a wash on the injection module with methanol.
3. Turn off the UV detector.
4. Turn off the CO_2 supply.
5. Leave system pressurized.
6. Cleaning solvents listed in the acceptable solvents list may remain in the cyclone. Solvents listed in the unacceptable solvents list and any other material not on the acceptable list should not be left in the cyclone.

   Acceptable Solvents

   - Methanol (MeOH)
   - Ethanol
   - Heptane
   - Hexane
   - Isopropyl alcohol (IPA)

   Unacceptable Solvents

   - Acetonitrile
   - DMSO
   - Methylene chloride
   - Tetrahydrofuran (THF)
   - Triethanolamine (TEA)
   - Trifluoroacetic acid (TFA)

7. Turn off the water baths for pumps.

Long Term Shutdown

1. Clean the system of residual product.
    a. Perform a wash on the Injection Module with methanol.
    b. If each cyclone has not been cleaned with an acceptable solvent (see previous page), execute the “Cleaning Instructions” with either methanol (MeOH) or isopropyl alcohol (IPA).
2. Make sure each cyclone is drained, including the waste cyclone.
3. Pump pure CO_2 through the system for 10 - 15 minutes.
4. Turn off the CO_2.
5. Depressurize the system.
6. Close each cyclone fully.
7. Log out of SuperChrom.
8. Shut down computer.
9. Power down each component. (Including the water bath)
10. Disconnect power to the system.

Chapter 6
Method Setup Overview

Introduction

This chapter provides basic instructions for method setup for the SFC Prep 350 system.

Method Settings

1. In the main SuperChrom screen, click the **Method Settings** button.

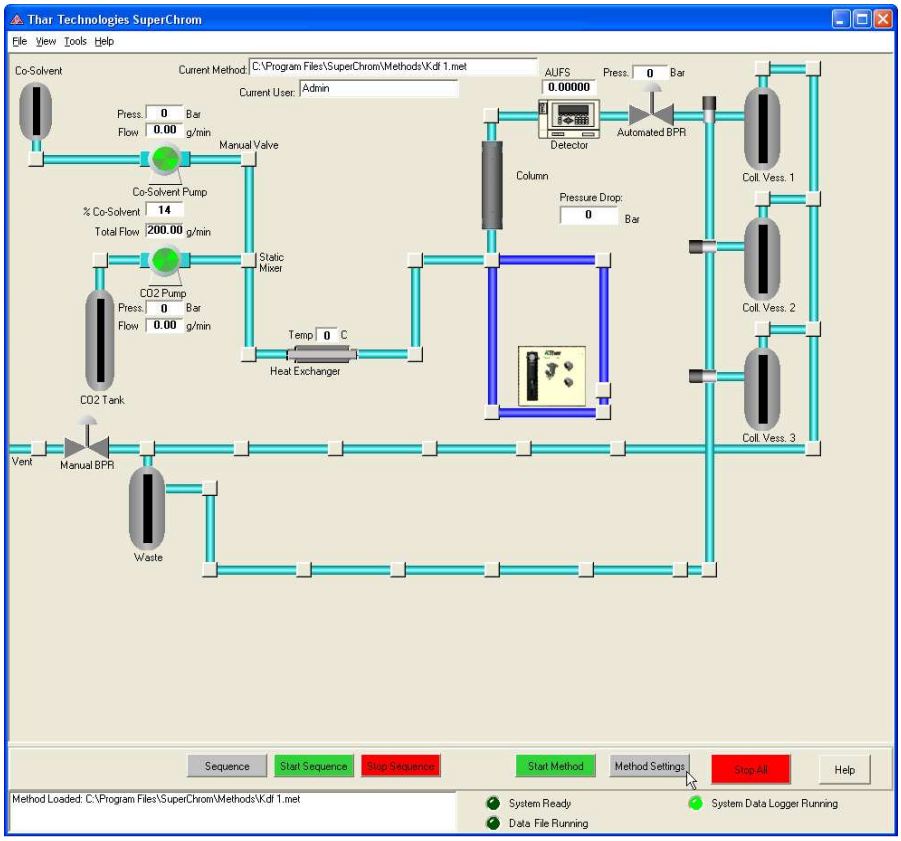


Figure 6-1 Click Method Settings

2. The System Settings window displays.

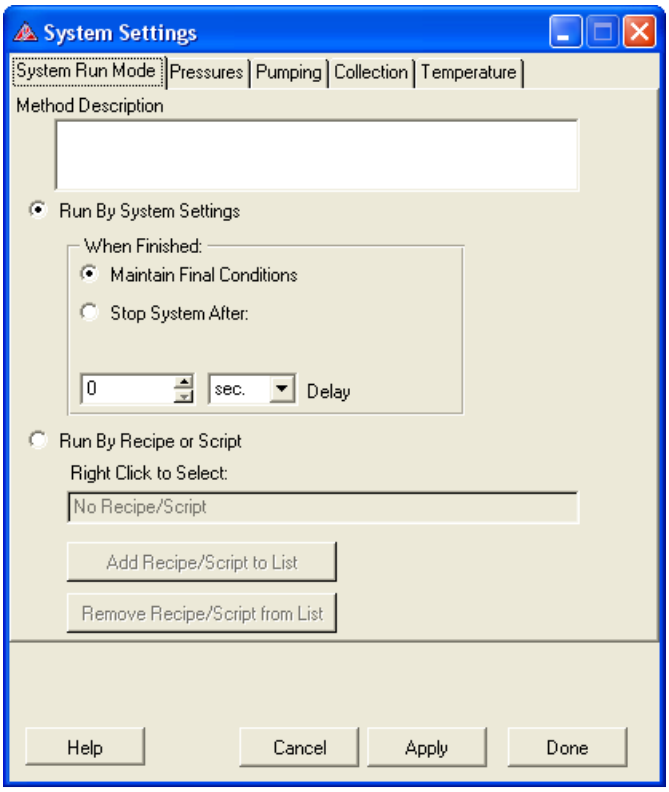


Figure 6-2 System Run Mode

3. Select the **System Run Mode** tab.

4. Enable “Run By System Settings”.

5. Select the **Pressures** tab.

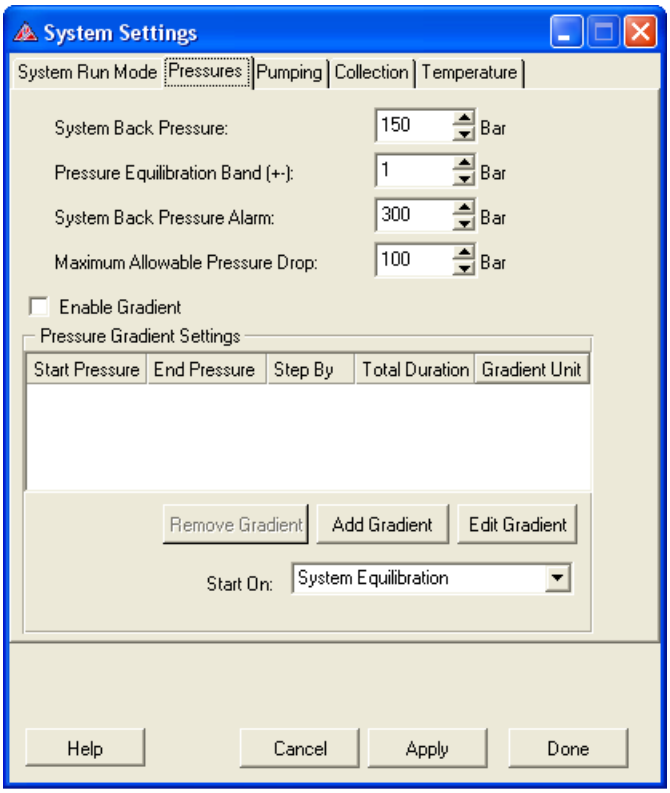


Figure 6-3 Pressures

6. Enter a value between 100 and 300 bar for the System Back Pressure.

NOTE	**The system back pressure can be set as high as 300 bar.**

WARNING	**Ensure the column hardware and/or packing material installed on the system can handle the "System Back Pressure" set point plus an additional 80 bar due to potential back pressure due to the column packing.**

7. Select the **Pumping** tab.

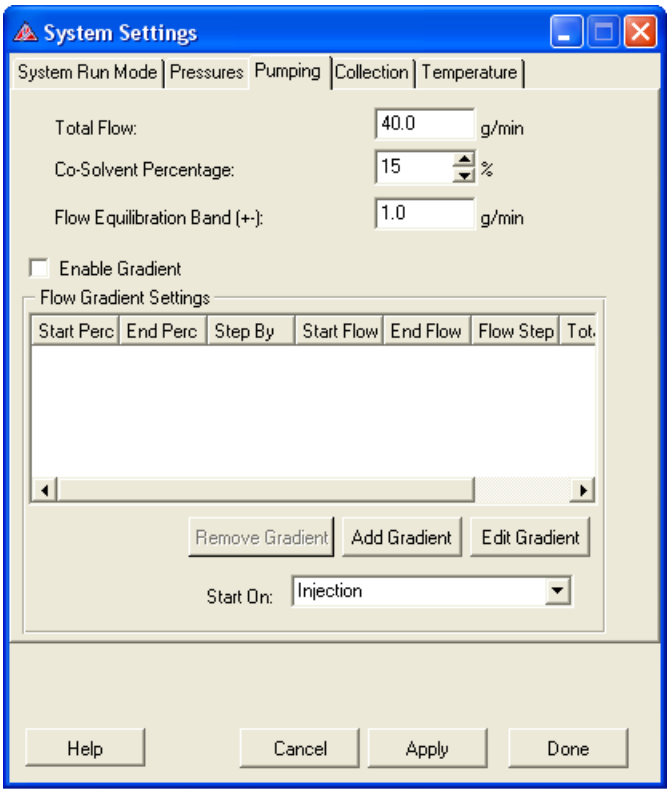


Figure 6-4 Pumping

8. Enter 40.0 g/min for the Total Flow.

9. Enter 15% for the Co-solvent (modifier) Percentage.

10. Select the **Collection** tab.

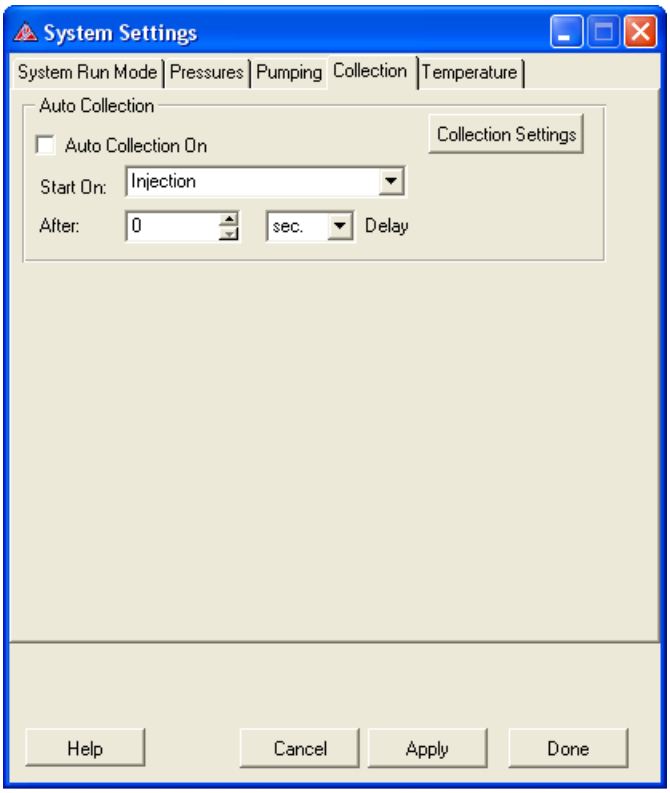


Figure 6-5 Collection

11. For test injections do not check “Auto Collection On”. If you want to collect test injection fractions, use the manual collection feature or (if the sample has minimal impurities) mV signal for collection.

12. Select the **Temperature** tab.

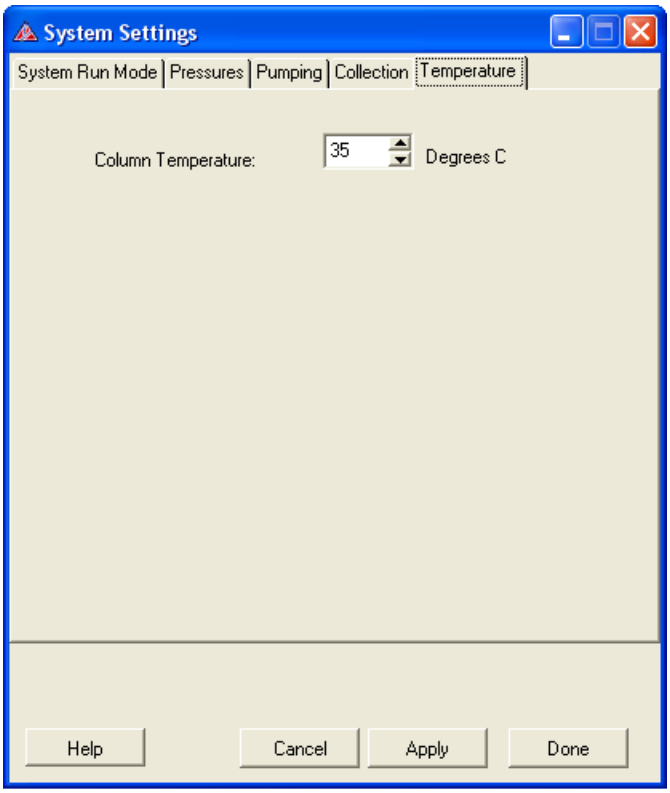


Figure 6-6 Temperature

13. Set the Column Temperature to 35°C.

14. Click **Done** to close this window.

Data File Settings

There are several options for data file storage. A data file can be created

- For each injection (recommended),
- For an entire sequence, or
- When collection operations start.

Directions for selecting data file options are provided below.

1. To access the Data File settings, select Tools > Data File > Log Settings.

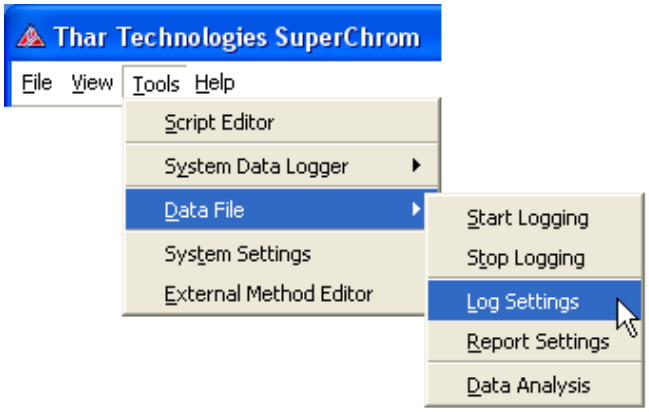


Figure 6-7 Data File Log Settings

NOTE	**Alternatively, the Data File Settings can be accessed by double-clicking the UV detector icon and then clicking the Data File Settings button on the UV Input screen.**

2. Select the General tab:

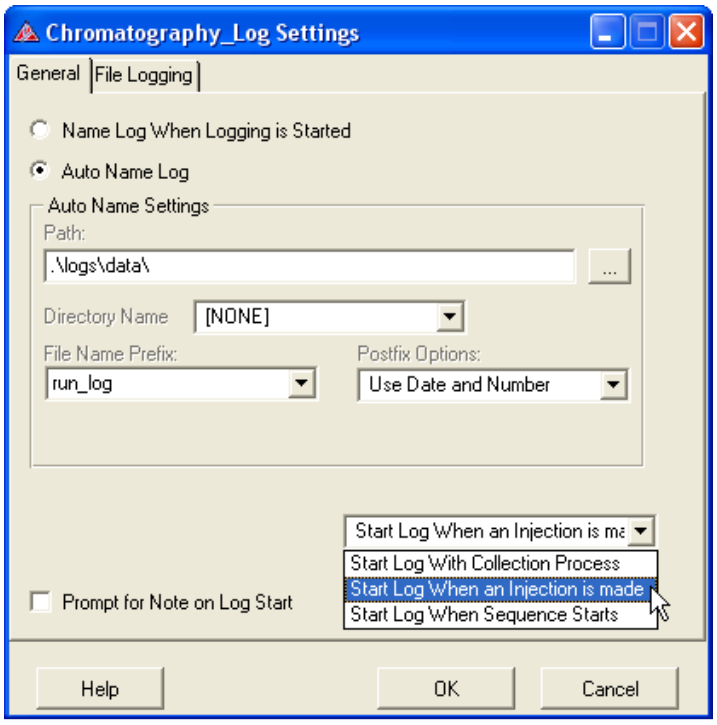


Figure 6-8 Chromatography Log Settings

3. Enable “Auto Name Log”.
4. Select “Start Log When an Injection is made” (recommended).

Other Options

You have the option to store an entire sequence as a data file or just when the collection operations start.

1. In the menu bar select Tools > Data File > Log Settings.
2. In the “General” tab check “Auto Name Log” and select “Start Log With Collection Process” or select “Start Log When Sequence Starts”

One of the disadvantages to storing entire sequences and/or collection processes as a single log is that RSD% information (especially for retention time) is more difficult or nearly impossible to compile.

Method Startup

1. Turn on the CO_2 supply.
2. Allow 2-5 minutes for flow surge to equilibrate.
3. If the co-solvent pump has not been primed review Prime Co-Solvent Pump.
4. Confirm MV2 is open (see Figure 5-23).
5. If your method is setup, click “Start Method.” If you have not set up your method, review Method Setup.

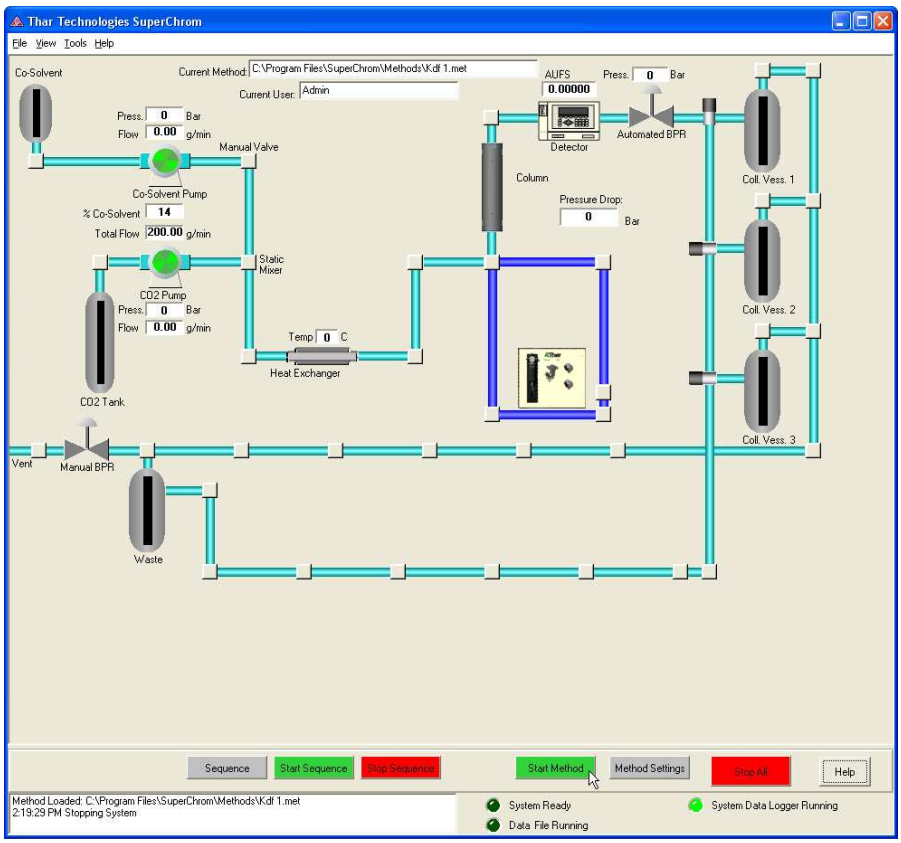


Figure 6-9 Start Method

6. Wait a 2 - 5 minutes to allow system to equilibrate to the desired pressure.

7. Double-click the UV detector icon to open the UV Input screen.

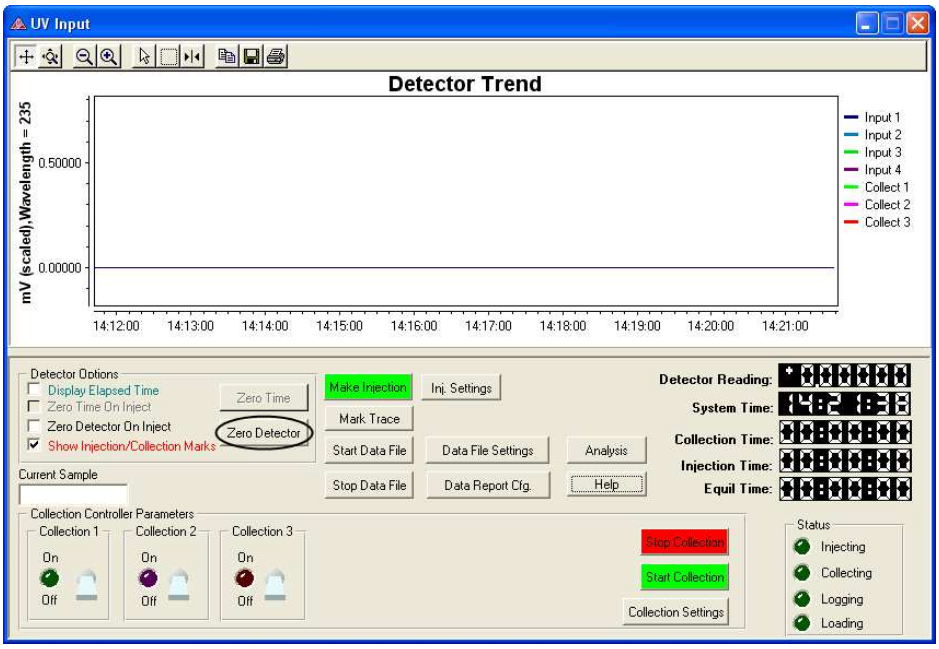


Figure 6-10 Zero the Detector

8. Zero the detector by clicking the **Zero Detector** button.

MBPR Instructions

Set the MBPR after 5 minutes of equilibration time at running conditions.

CAUTION	**For optimum performance there must be at least 55 bar of manual back pressure on the 1000 mL cyclones during collection.**

Figure 6-11 Manual Back Pressure Regulator

This MBPR must be set to 55 bar to achieve optimum conditions. This will allow for a constant minimum 55 bar back pressure on the 1000 mL cyclones while in operation to achieve optimum collection conditions.

The most important pressure regarding the MBPR is the pressure on the cyclones while collecting, **trying to use 0bar on the cyclones will lead to product loss.**

Test Injections

NOTE: while doing mixed phase injections the injections can not be closer than 2 minutes apart

1. Double-click the Injection Module icon to open the Thar Injection Module window.

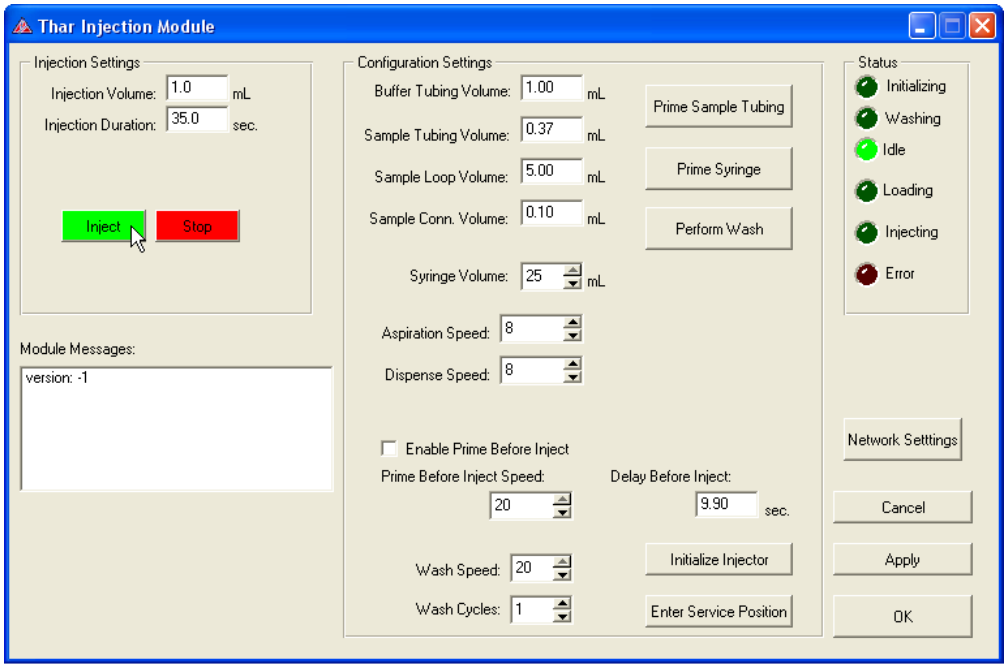


Figure 6-12 Thar Injection Module

2. Enter the “Injection Volume” and “Injection Duration”.
3. If using a custom loop size, change “Sample Loop Size”
4. Change “Sample Conn. Size” to .10
5. Start the method. When the system is equilibrated, click the **Inject** button on the Thar Injection Module screen.

NOTE	**Alternatively an injection can be made by clicking the Make Injection button on the UV Input screen.**

Fraction Collection

Valve Delay Settings

There will be “Valve Delay” settings in an Appendix of this manual for your specific unit

Enter the same valve delay setting for both Start Collecting Conditions and Stop Collecting Conditions for the desired flow rate and Co-Solvent (modifier) %

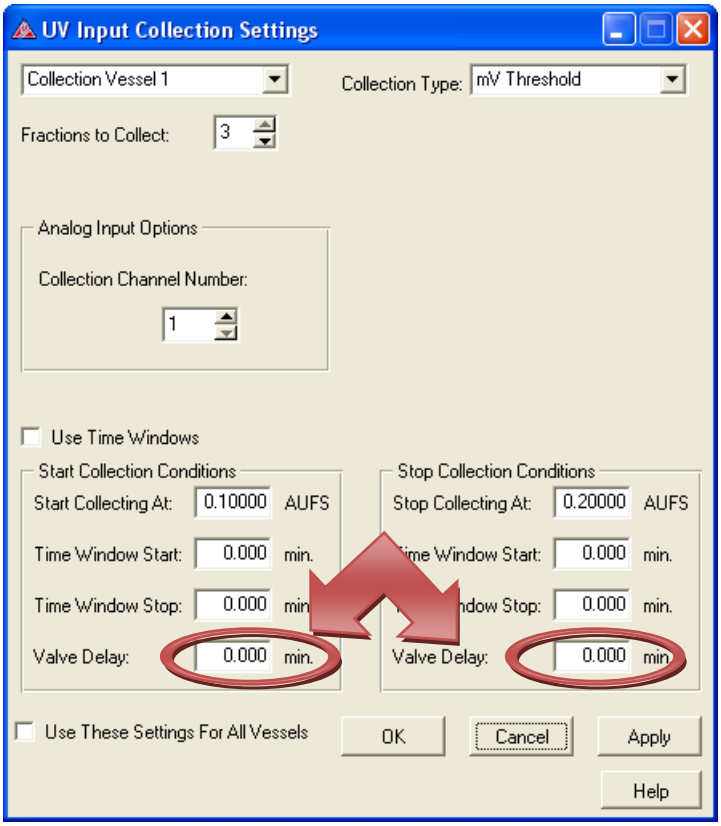


Figure 6-13 Valve Delay Settings

Manual Collection

If you want to collect fractions from a test injection, manual collection is the best option. Directions are provided below.

1. Open the UV Input screen by double-clicking on the UV detector icon on the SuperChrom main screen.

2. When the desired peak displays in the Detector Trend window, turn On each cyclone valve by clicking the Collection On/Off toggle icon.

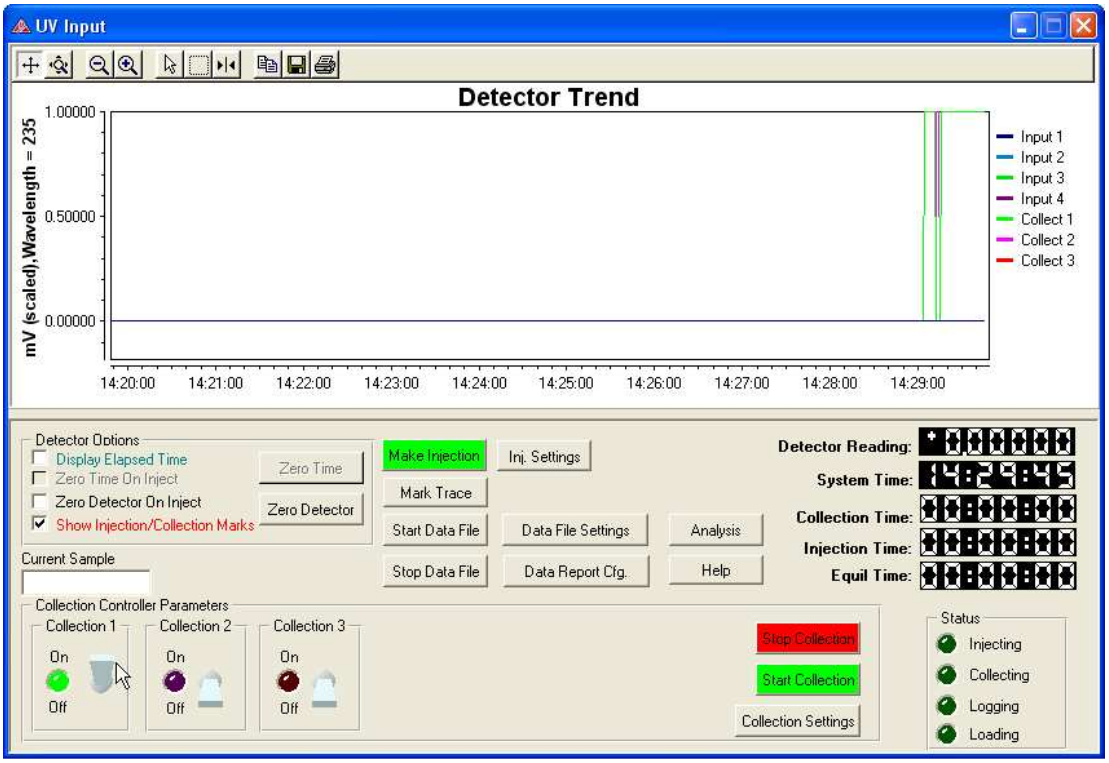


Figure 6-14 Collection Control Parameters

3. Alternatively to start collection, select each cyclone manually from the main SuperChrom screen.

4. Right-click on the collection valve and select “Start Collection.”

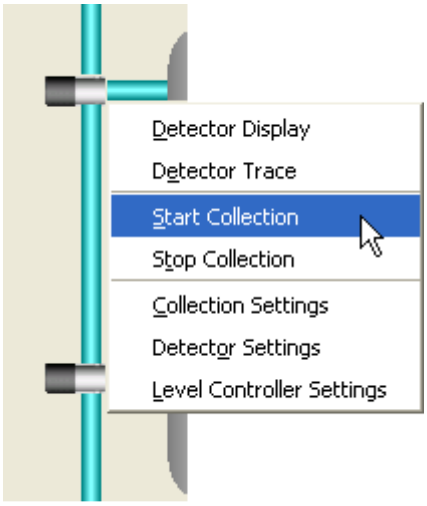


Figure 6-15 Collection Valve

5. To stop collection at the end of the peak
    a. On the UV Input screen click the **Stop Collection** button or
    b. Turn the Collection switch Off
    c. Alternatively, from the SuperChrom main screen, right-click on the collection valve and select “Stop Collection”.

6. After performing test injections review the “Collection Instructions” (next page).

Performing non-stacked injections will allow the user to easily use all the UV detector settings found in UV Detector Setup.

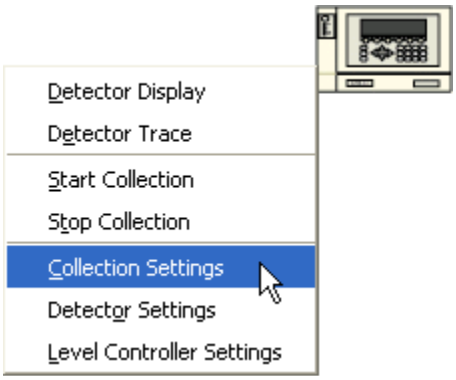


Figure 6-16 Collection Settings

There are several option to access the Collection Settings:

- Right click on the detector
- OR Right click on any of the graphical valves in the SuperChrom screen
- OR Click “Method” then click on the “Collection” tab
- Double Click on the UV detector. At the bottom right of the UV screen there is a tab for “Collection Settings”

Collection Settings

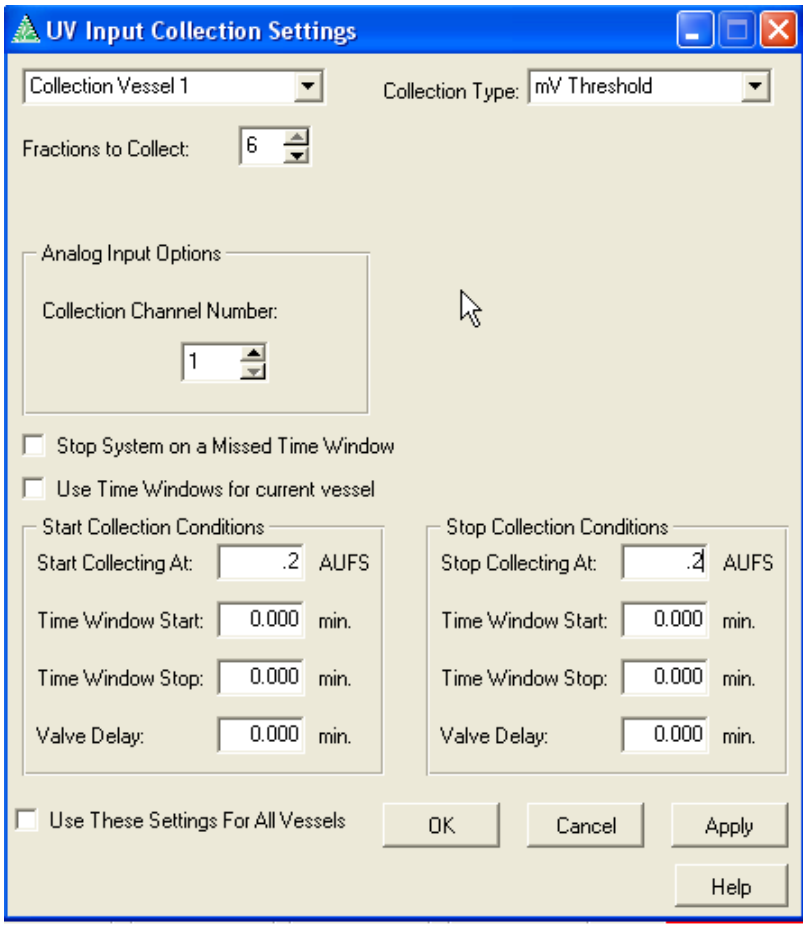


Figure 6-17 UV Input Collection Settings

Once the “UV Input Collection Settings” window is open the collection settings can be modified.

- Select “Collection Vessel X”
- Select “Fractions to Collect”

Collection Type

Select the Collection Type. There are three choices: mV Threshold, Slope Threshold or Timed Collection. The options are explained on the following pages. Note: Changing "Collection type" will change the "UV Input Collection Settings" window for each Collection Type.

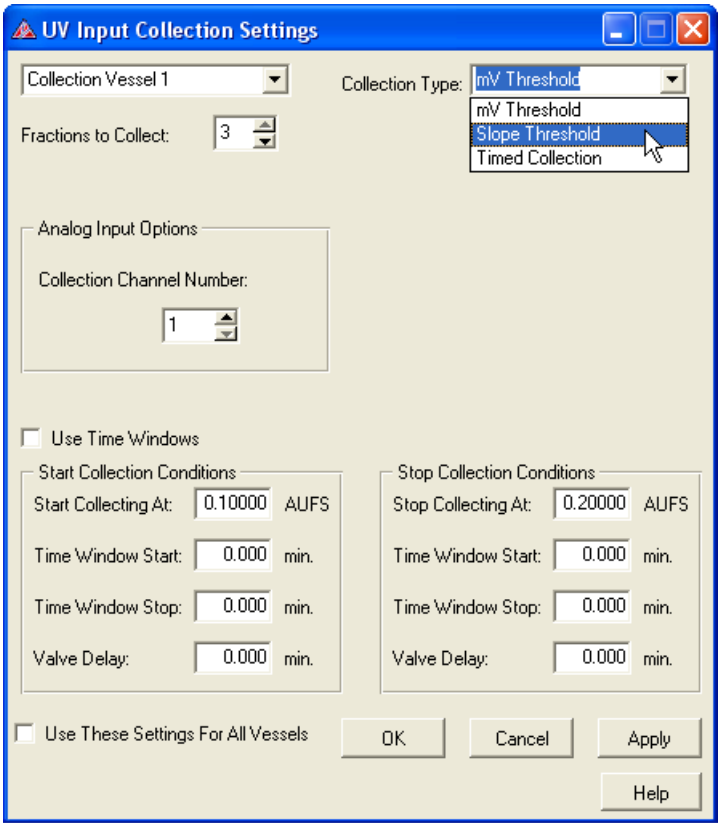


Figure 6-18 Select Collection Type

Table 6-1 Collection Settings

Start collection At	?	AUFS	Stop collection At	?	AUFS
Time Window Start	?	Min	Time Window Start	?	Min
Time Window Stop	?	Min	Time Window Stop	?	Min
Valve Delay		min	Valve Delay		min

mV Threshold

This setting collects on the signal coming from the UV detector. Available settings are listed in Table 5-1 (previous page).

A typical “Start Collection At” and “Stop Collection At” value is 0.10 - 0.20 AUFS.

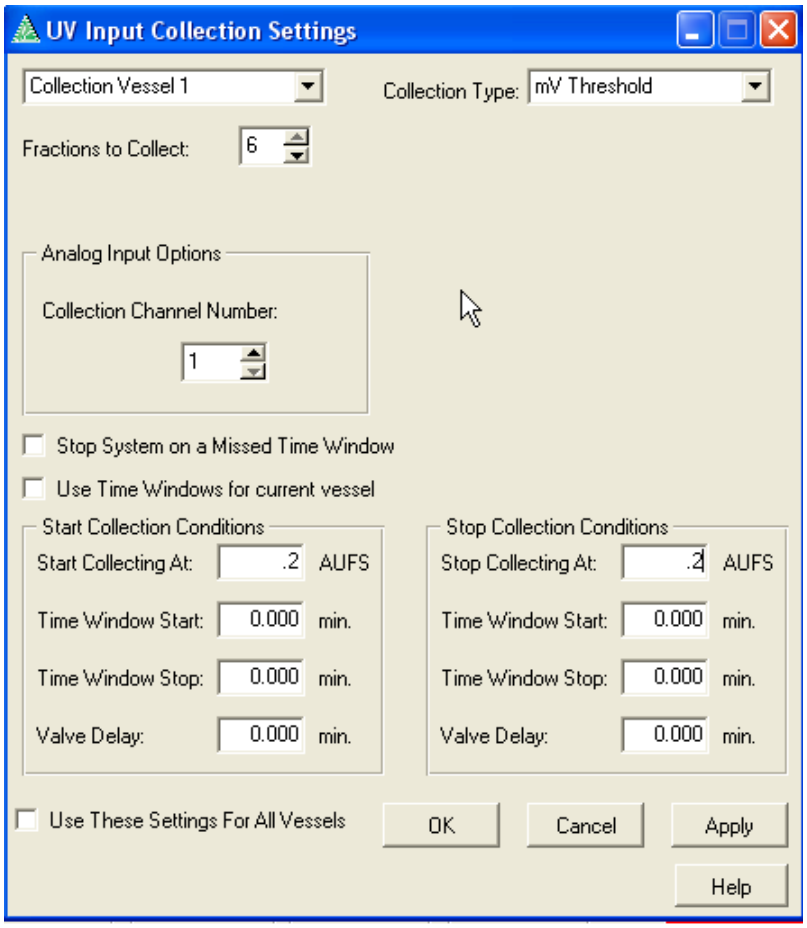


Figure 6-19 mV Threshold Collection

mV Threshold Time Windows

Time windows can be used if there are impurities that might trigger the start of collection before the desired peaks. The System can be stopped for missed time window, if desired.

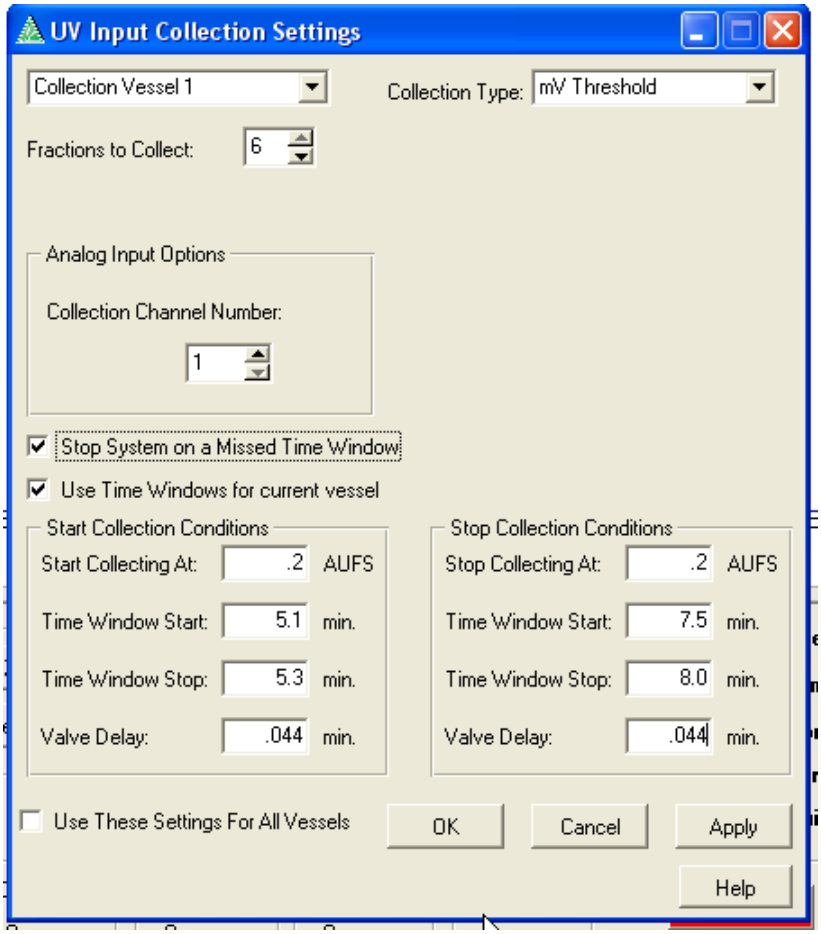


Figure 6-20 mV Threshold Time Windows

- **Time Window Start**: Describes where the computer will start looking for the signal strength to start/end collection

- **Time Window Stop**: Describes where the computer will stop looking for the desired signal strength and either shut down or stop collecting.

Slope Threshold

This setting will collect on the value of the slope of the signal coming from the UV detector. Available settings are listed in Table 6-2 (this page).

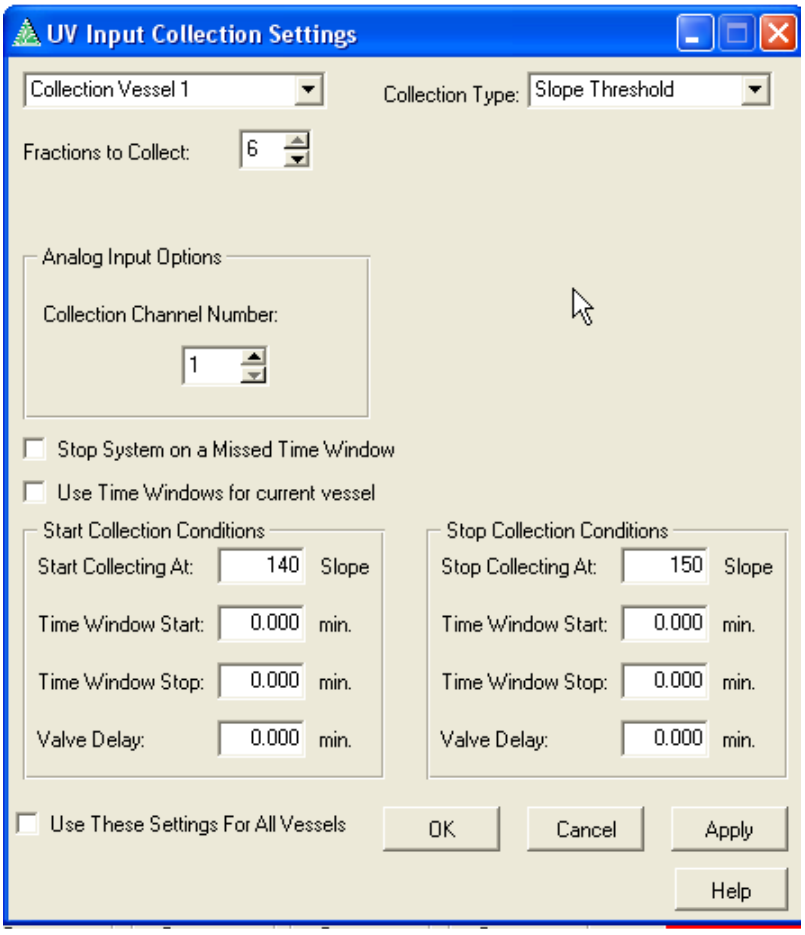


Figure 6-21 Slope Threshold Collection

Table 6-2 Slope Threshold Collection Settings

Start collection At	?	Slope	Stop collection At	?	Slope
Time Window Start	?	Min	Time Window Start	?	Min
Time Window Stop	?	Min	Time Window Stop	?	Min
Valve Delay		min	Valve Delay		min

Values for the slope are determined from an existing chromatogram. To determine the slope:

1. Double-click on the Detector to open up the “UV Input” screen
2. Find desired chromatogram, and click the cursor button. 
3. Right click on the line that is on the chromatogram.

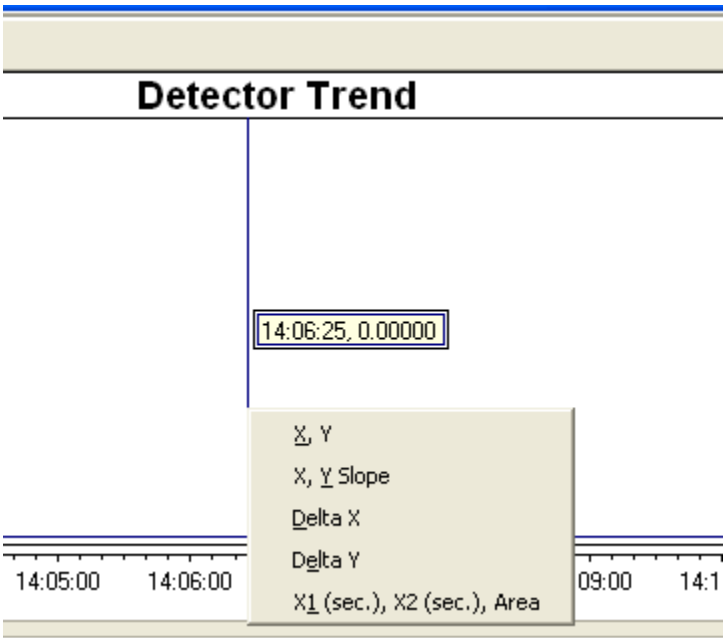
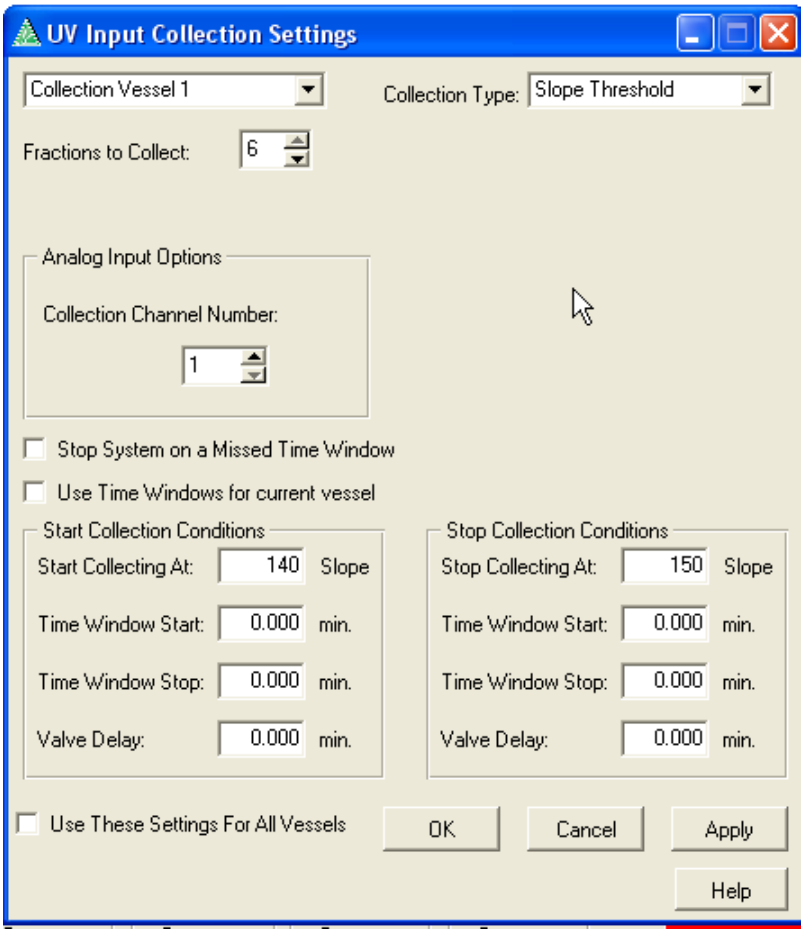


Figure 6-22 Select Slope

4. Choose “X, Y_slope”
5. Determine where the collection should start.

Slope Threshold Time Windows

Time windows can be used if there are impurities that might trigger the start of collection before the desired peaks. The System can be stopped for missed time window, if desired.



- **Time Window Start**: Describes where the computer will start looking for the Slope to start/end collection.

- **Time Window Stop**: Describes where the computer will stop looking for the desired Slope and either shut down or stop collecting.

Timed Collection

This setting collects solely on time values entered by the user.

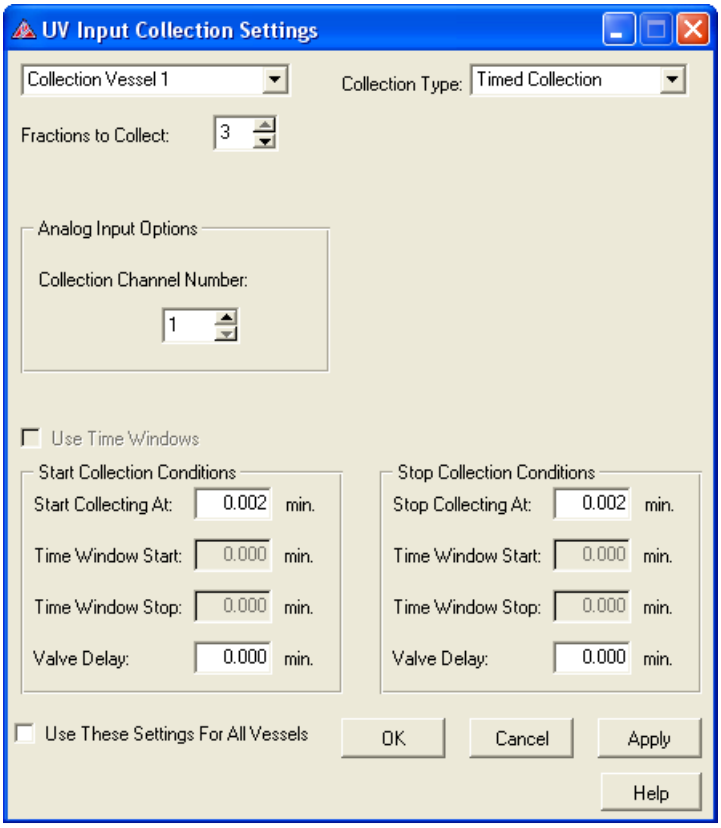


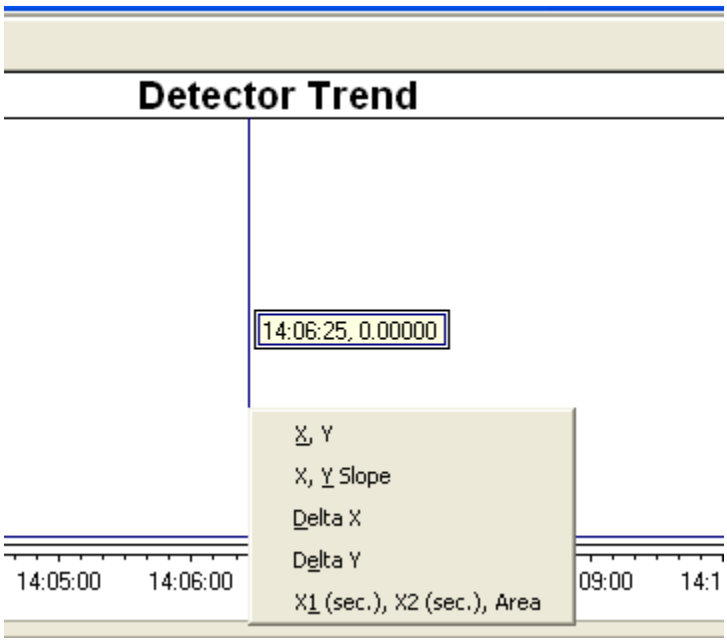
Figure 6-23 Timed Collection

Available settings for timed collection are listed below. Time Windows are not available for Timed Collection.

Table 6-3 Timed Collection Settings

Start collection At	?	min	Stop collection At	?	min
Valve Delay		min	Valve Delay		min

To determine the time settings:

1. Double click on the Detector to open up the “UV Input” screen
2. Find desired chromatogram, and click the cursor button. 
3. Right-click on the line that is on the chromatogram. (See Figure 5-36, previous page.)
4. 

5. Choose “Delta X.” Two lines will appear on the “Input Screen.”
6. Place one of the lines on the Injection Mark.
7. Place the 2nd line near each desired peak to determine at what times collection is required and total run time

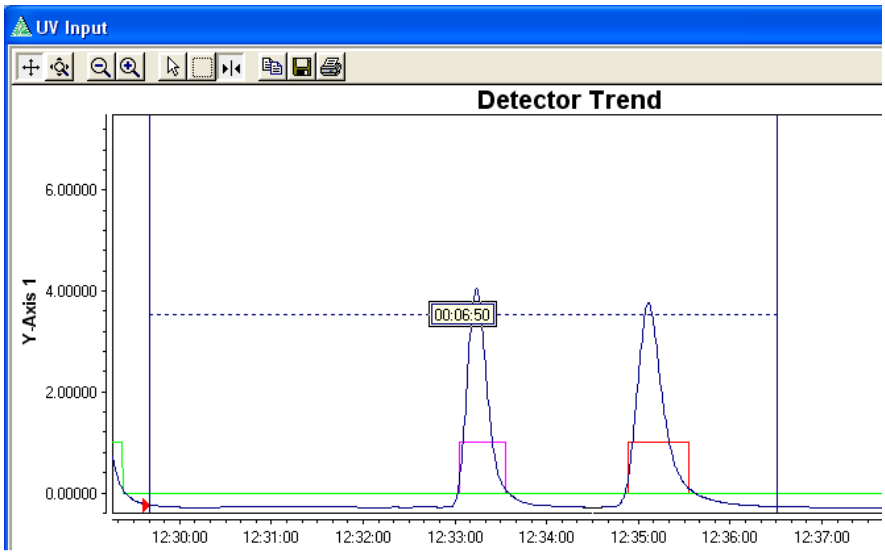


Figure 6-24 Collection Times

Post Collection

1. Check the level in the bottles/containers used for collection.
2. If the bottles are not near capacity, allow the method to continue to run.
3. Set Co-solvent to 40%.
4. Manually select collection to each cyclone (used for collection during the sequence) and rinse with 50 – 100 mL of solvent.
5. Click “Stop All” and manually drain each cyclone into the collection bottles/container.
6. Check the level from the level controller in each Fraction Collector to ensure it is completely drained.

Sequences

There are two options for setting up a sequence:

1. Auto Sequencing- performs a set number of injections for each designated method
2. Manually Add Injections

To create an injection sequence, the following parameters of an existing chromatogram must be known:

- Method
- Run duration
- Start injection option
- Injection valve duration
- Delay time for solvent equilibration
- Injection volume
- Wavelength setting for UV detector

Directions for both options are provided on the following pages.

NOTE: while doing mixed phase injections the injections can not be closer than 2 minutes apart

Auto Sequencing

1. Click the **Sequence** button on the main SuperChrom screen.

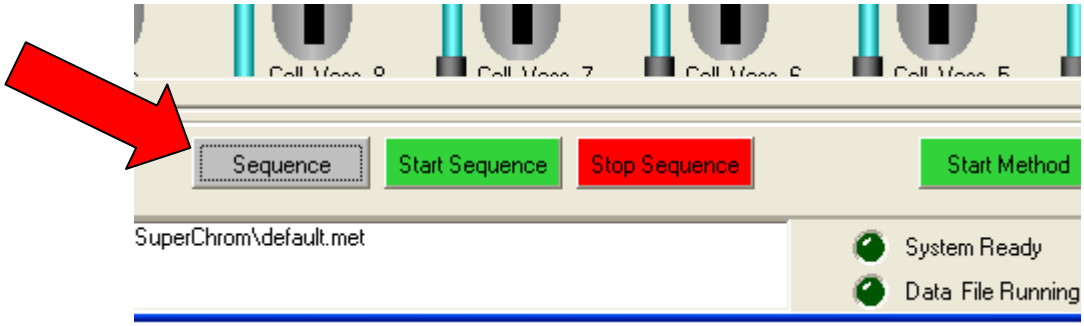


Figure 6-25 Create Sequence

2. The Injection Sequence screen displays. Click the **Auto Sequencing** button.

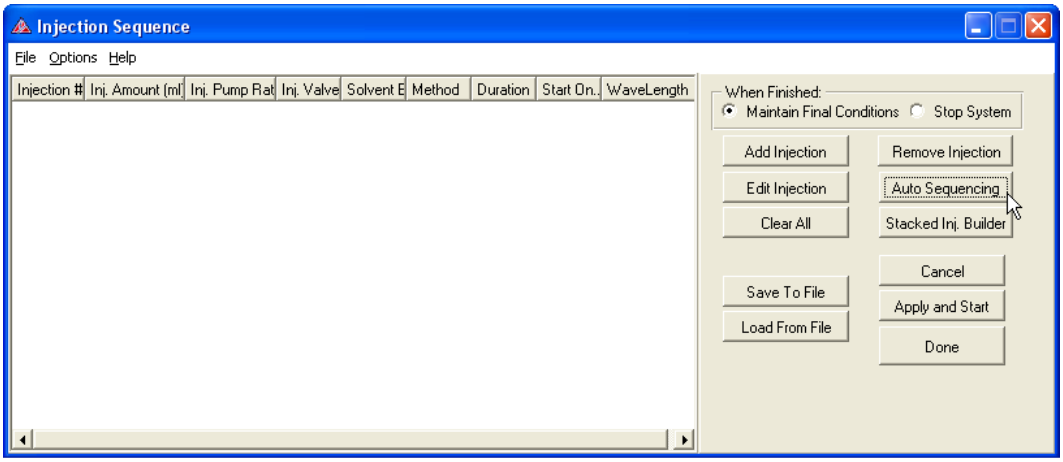


Figure 6-26 Select Auto Sequencing

3. NOTE: while doing mixed phase injections the injections can not be closer than 2 minutes apart

4. The Sequence Generator window displays.

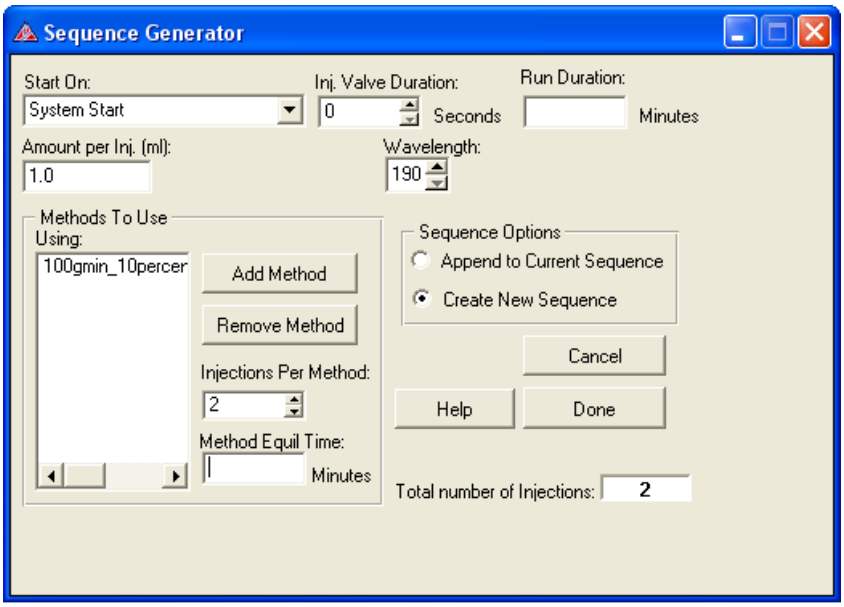


Figure 6-27 Sequence Generator

5. A brief description of each field is provided in Table 6-3.

Table 6-4 Sequence Parameters

Field	Description	
Start On	System Start	
Inj. Valve Duration	Amount of time injection valve will be in the “inject” position (seconds)	
Run Duration	Length of run after injection (minutes)	
Amount per Inj (mL)	Injection volume	
Wavelength	Sets UV wavelength	
Methods To Use	Click **Add Method** and select the desired method. More than one method can be selected.	
Injections Per Method	Number of injections for selected method	
Method Equil Time	Set if desired (minutes)	
Sequence Options	Append to Current Sequence	Adds the stacked sequence to the existing sequence in the “Injection Sequence” box.
	Create New Sequence	Creates a new sequence and erases any existing sequences in the “Injection Sequence” box

6. Complete all fields, then click **Done**. You will return to the Injection Sequence screen.

7. Select a “When Finished” option on the Injection Sequence screen:

    a. “Maintain Final Conditions”-will maintain conditions after the sequence has ended

    b. “Stop System” –will stop system when the sequence has ended

Figure 6-28 Injection Sequence Finished Options

8. Then click **Done**.

9. Review the values in the Injection Sequence screen. If the values are correct, click **Done**.

Manually Create a Sequence

1. Click the **Sequence** button on the main SuperChrom screen.

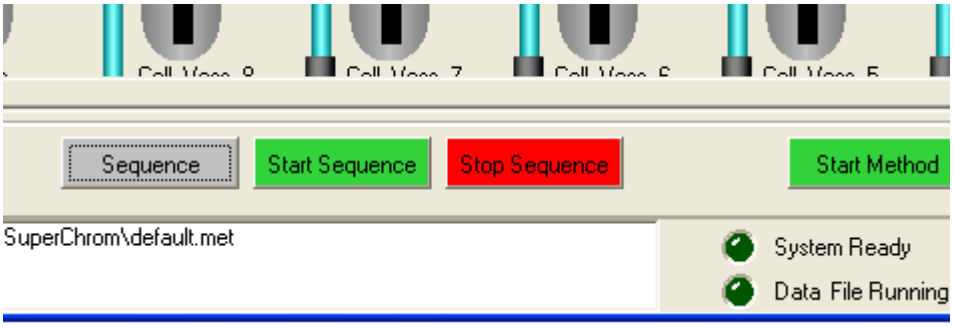


Figure 6-29 Create Sequence

2. The Injection Sequence screen displays. Click the **Add Injection** button.

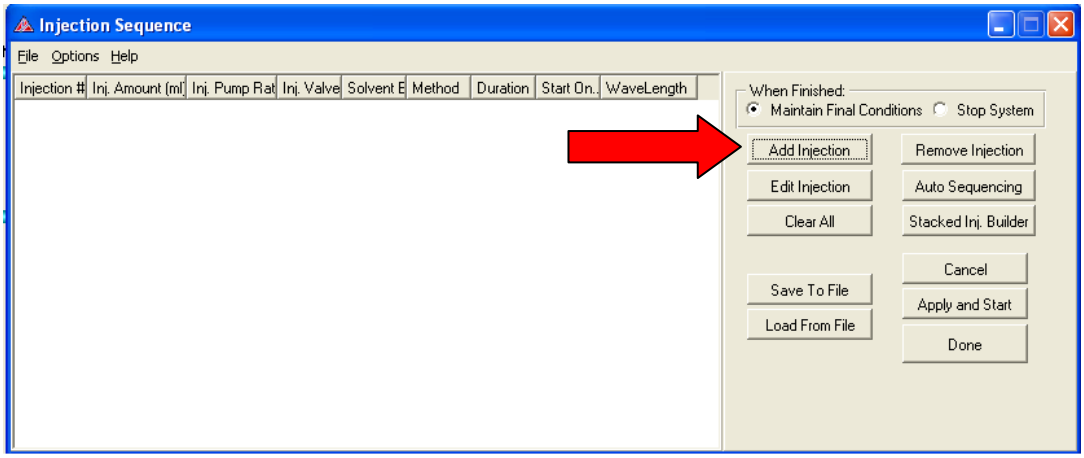


Figure 6-30 Injection Sequence

3. NOTE: while doing mixed phase injections the injections can not be closer than 2 minutes apart

4. The Add Injection screen displays.

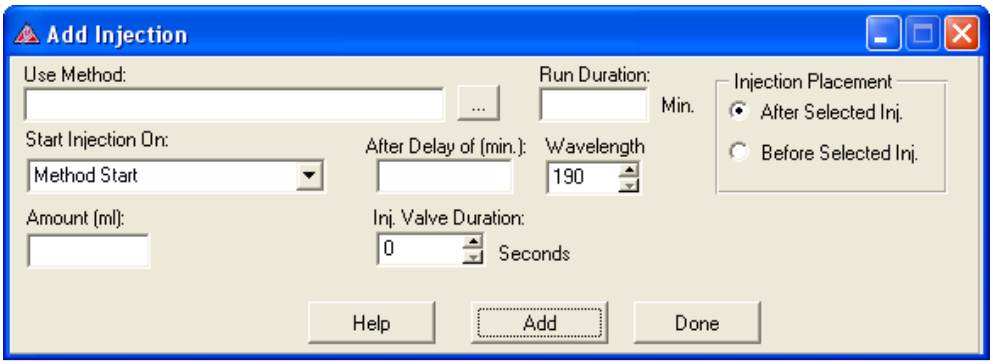


Figure 6-31 Add Injection

Table 6-5 Add Injection Parameters

Field	Description	
Use Method	Click the square box to select a method from the method files	
Run Duration	Length of the run after injection (minutes)	
Injection Placement	After Selected Inj.	Places injection after highlighted injection on the “Injection Sequence” list
	Before Selected Inj.	Places injection before highlighted injection on the “Injection Sequence” list
Start Injection On	Method Start	Starts injection process when sequence is started
	Method Equilibration	Starts injection process when the current method is equilibrated
After Delay of (min.)	Delay is typically used for solvent % changes between methods	
Wavelength	Sets UV wavelength	
Amount (mL)	Sample injection volume	
Inj. Valve Duration	Amount of time injection valve will be in the “inject” position (seconds)	

5. Complete all fields. Click **Add** until the desired injection number has been reached.

6. Then click **Done**. You will return to the Injection Sequence screen.

7. Select a “When Finished” option on the Injection Sequence screen:
    a. “Maintain Final Conditions”-will maintain conditions after the sequence has ended
    b. “Stop System” –will stop system when the sequence has ended

Figure 6-32 Injection Sequence Finished Options

8. Then click **Done**.
9. Click Done on the Injection Sequence screen to accept any new sequences, changed and/or appended sequences

Stacked Injection Sequences

Stacked injections are used to reduce the time to separate a desired amount of sample. Typically, stacked injections are used when relatively “clean” samples need to be separated. As such, use of Time Windows is normally discouraged. Also the optional UV detector settings found on page 5-12 are not recommended.

NOTE: while doing mixed phase injections the injections can not be closer than 2 minutes apart

Building a Stacked Injection Sequence

1. Click on the **Sequence** button in the SuperChrom main screen to open the Injection Sequence window.

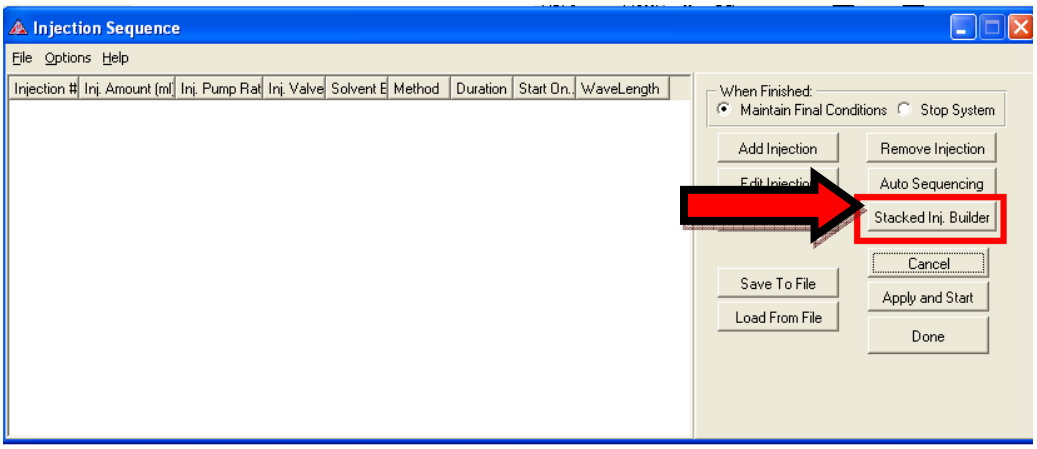


Figure 6-33 Select Stacked Inj. Builder

2. Click on the **Stacked Inj. Builder** button.
3. The Stacked Prep Builder screen displays (next page).

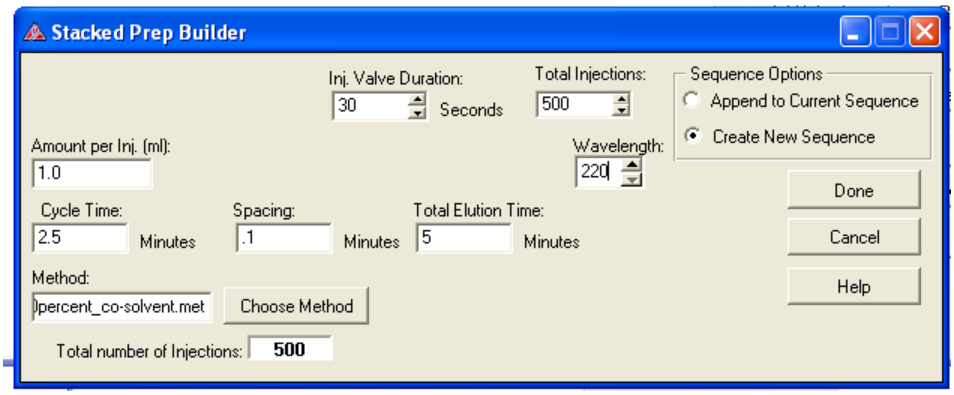


Figure 6-34 Stacked Prep Builder

4. A brief description of each field is provided in Table 6-5.

Table 6-6 Stacked Prep Builder Parameters

Field	Description	
Inj. Valve Duration	Duration sample loop is swept by the mobile phase	
Total Injections	Total number of injections	
Sequence Options	Append to Current Sequence	Adds the stacked sequence to the existing sequence in the “Injection Sequence” box.
	Create New Sequence	Creates a new sequence and erases any existing sequences in the “Injection Sequence” box
Amount per Inj (mL)	Injection volume	
Wavelength	Sets UV wavelength	
Cycle time	Time from beginning of Peak 1 to the end of Peak 2 (minutes)	
Spacing	Time between repeated injection cycles (minutes)	
Total Elution Time	Total time for one injection (minutes)	
Method	Click the **Choose Method** button and select the desired method	

5. To Create an automated stacked sequence:

6. In the “Choose Method” box select the desired method:

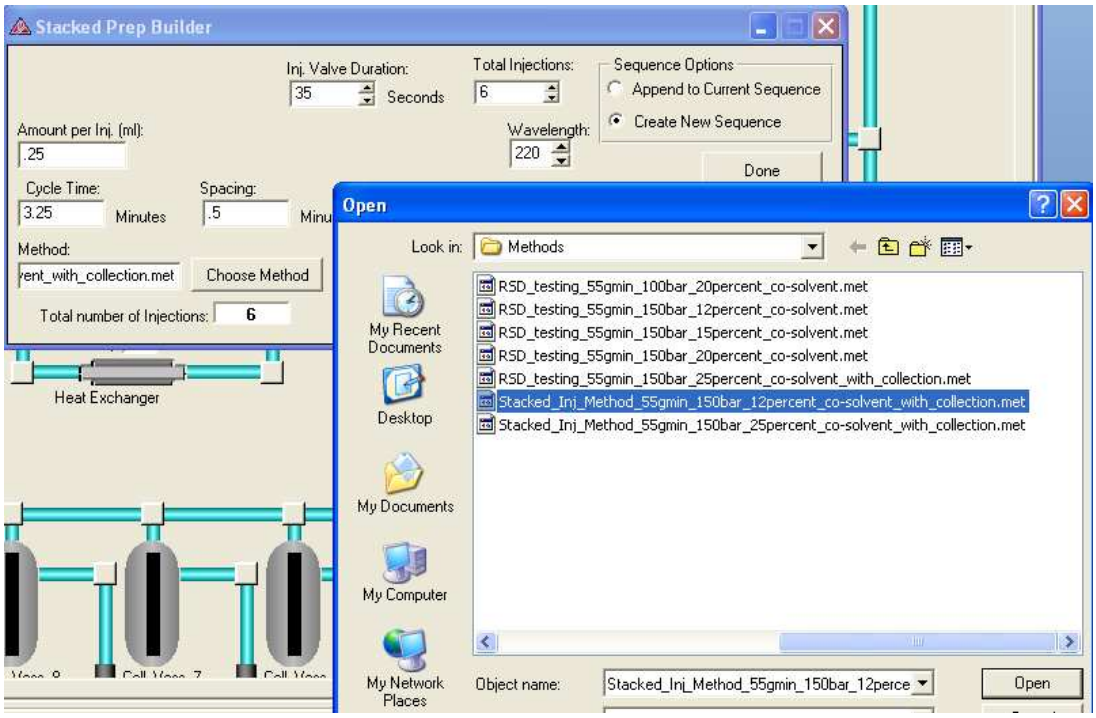
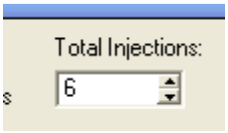
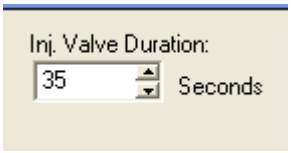


Figure 6-35 Stacked Injection Instructions

7. Click “Open”
8. Enter the desired 

Total Injections
9. Injection duration: xx sec 

10. To determine the time settings
11. Double click on the Detector to open up the “UV Input” screen
12. Find desired chromatogram
13. Click the cursor button 
14. Right click on the line that’s on the “Detector Trend” screen to see this:

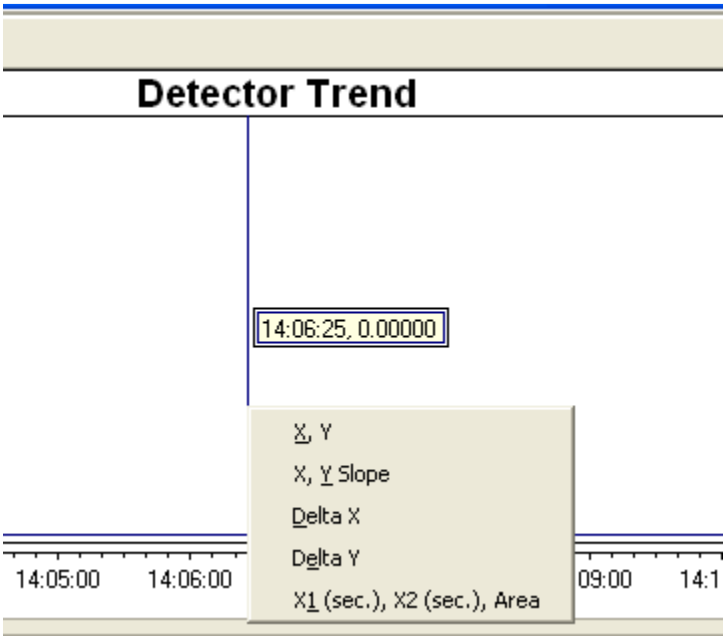


Figure 6-36 Stacked Injection Cursor Instructions

15. Choose “Delta X”

16. Two lines will appear on the “Detector Trend”
17. Place one of the lines on the desired beginning
18. Place the 2nd line near the peak to determine the desired ending
19. Enter Cycle Time 

: _____min (Time from beginning of peak 1 to end of peak 2→ See Diagram below)

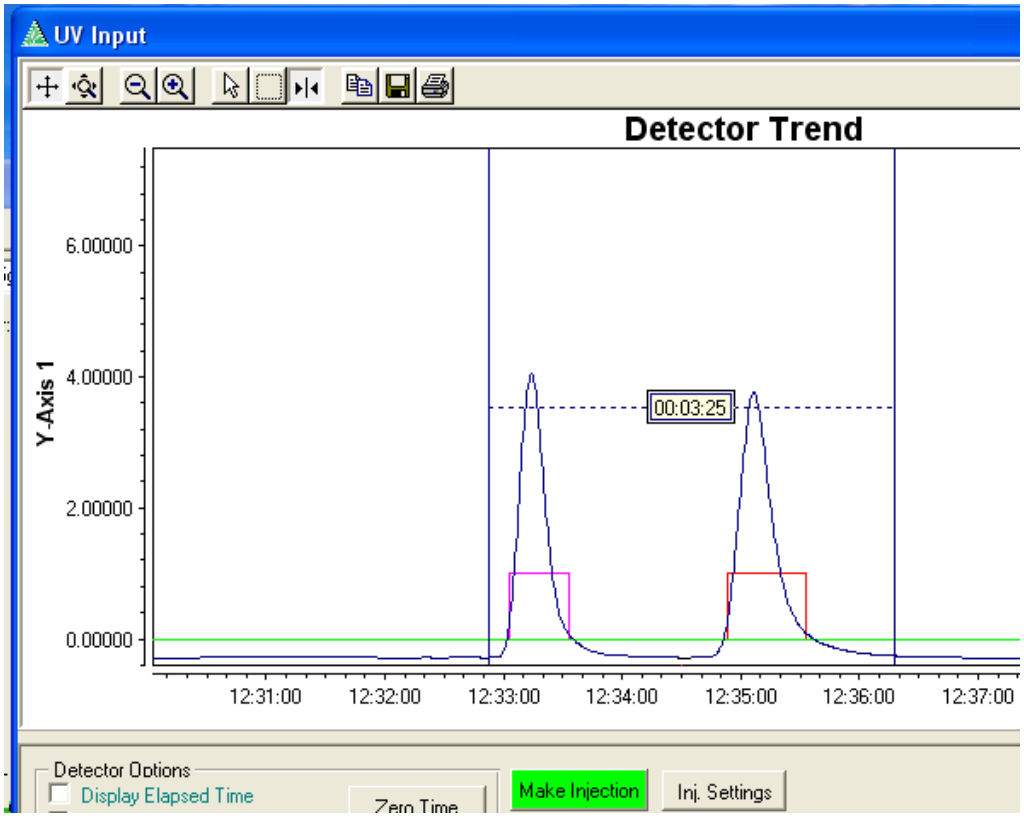
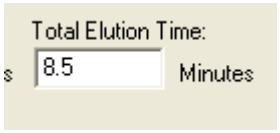


Figure 6-37 Stacked Injection Cycle Time Determination

20. Enter Spacing: XX min

(Time wanted between repeated injection cycles)

21. Enter Total Elution Time XX minutes



(Total time for one injection → See Diagram below)

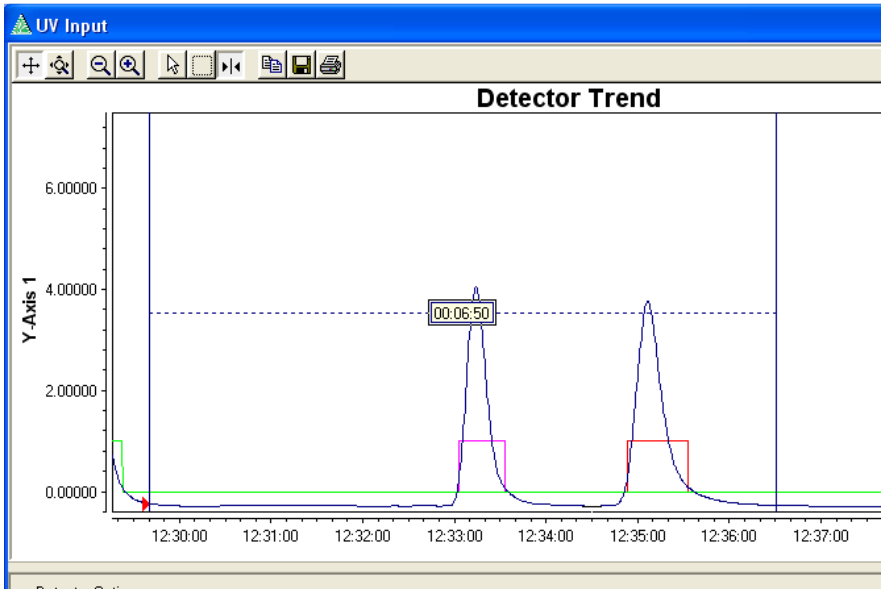
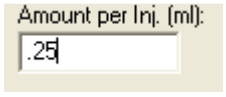


Figure 6-38 Total Elution Time Determination

22. Enter Wavelength xx nm

23. Enter XX mL for Amount per Injection



24. Click “Done”

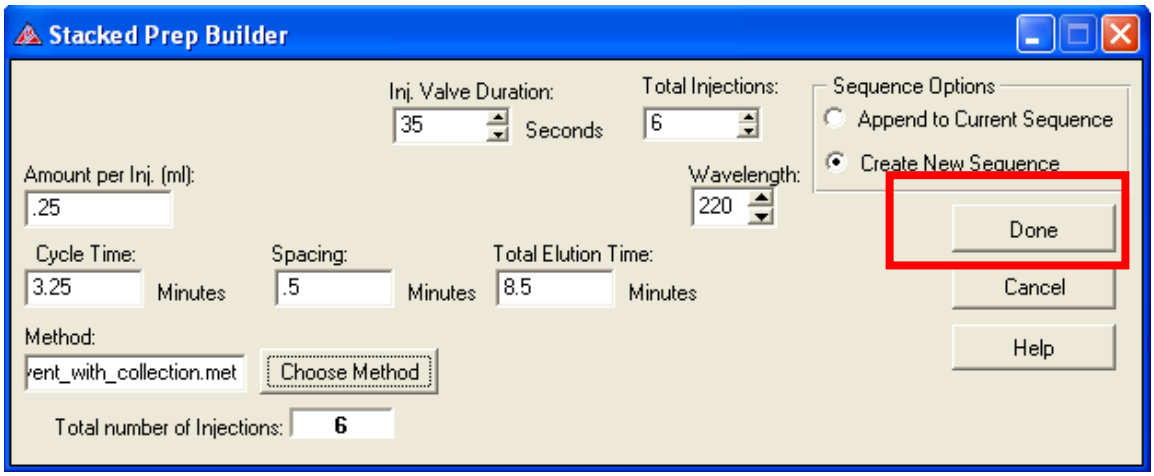


Figure 6-39 Stacked Sequence Parameter Example

25. Check to make sure the values you entered (Inj Valve Duration, Wavelength, total injections, Inj Amt (mL) are displayed properly in the list of injections.

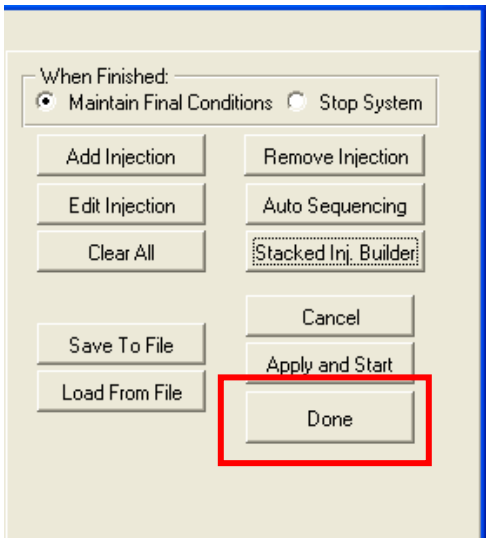
26. Select a “When Finished” option on the Injection Sequence screen:

    a. “Maintain Final Conditions”-will maintain conditions after the sequence has ended

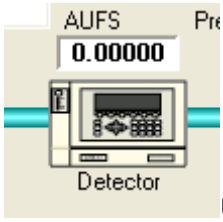
    b. “Stop System” –will stop system when the sequence has ended

Figure 6-40 Injection Sequence Finished Options

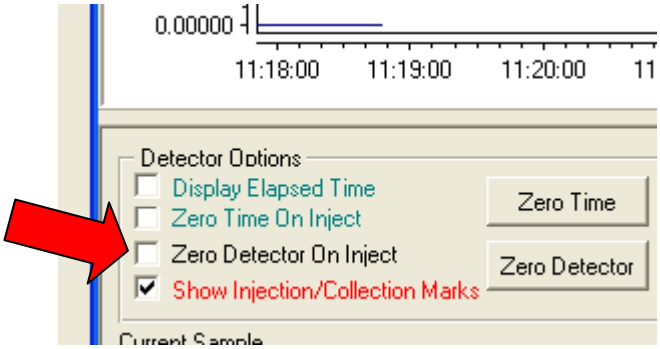
27. then click “Done”



Note: the last injection will be longer to compensate for the stacking. This assures that the last peaks are logged and collected.

28. Make sure method is still setup for automated collection… check the values in collection settings against the chromatograms generated in the test injections
29. Click “OK”
30. Double click on the 

UV detector, make sure the “Zero Detector on Inject” box is not checked



NOTE

The last injection will be longer to compensate for the stacking. This assures that the last peaks are logged and collected.

31. Make sure the method is setup for automated collection when doing stacked injections. From the main SuperChrom screen, click the **Method Settings** button.

32. Select the Collection tab. Check “Auto Collection On” and select Start On “Injection”. Then click **Done**.

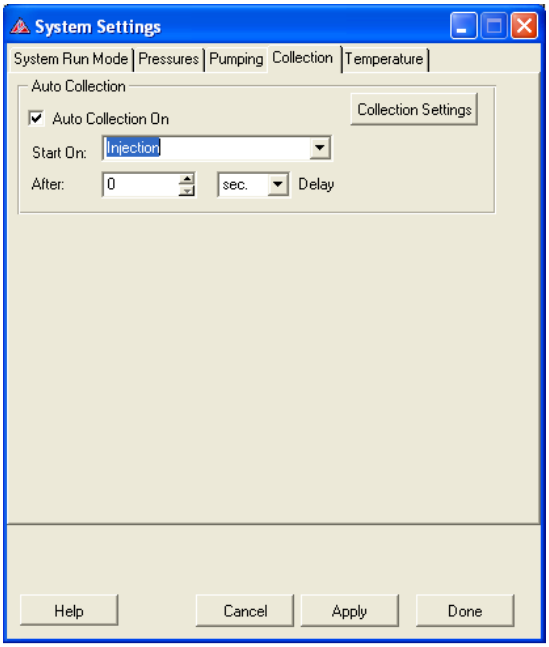


Figure 6-41 System Settings

Stacked Injection Collection Tips

- Shut off time windows unless absolutely necessary
- If a time window is necessary to avoid an impurity, pay very close attention to when the time window is set up. In some cases, the solvent from the next injection may change the retention time of the desired peak.
- Do not start a stacked injection sequence until the desired method is already running and equilibrated.

Chapter 7
SFC Prep 350 System Maintenance

Introduction

This chapter provides maintenance procedures and schedules for the SFC Prep 350 system. Periodic preventative maintenance is necessary to maximize performance of the system. Table 6-1 summarizes the procedures and recommended frequency.

Maintenance Schedule

The maintenance schedule for your SFC Prep 350 system is summarized below. Procedures for specific modules are located in the Operators Manual for the module.

CAUTION	**Many parts on the system are custom made to Thar Instruments specifications. Use of parts from other vendors may void your warranty.**

Table 7-1 SFC Prep 350 Maintenance Schedule

Maintenance Procedure		Frequency*	Comments
Pressurizing and leak checking		As needed	
Cleaning			
Replace CO_2 tank washer		Every other tank	As needed
P-350/200 Pumps			Refer to the P-350/200 High Pressure Pump Operations Manual (MN-10) for instructions
	Replace piston seals	6 months	
	Replace piston backup seals	6 months	
	Replace check valves	1 year	
	Replace rupture disc	As needed	
	Replace pump head o-ring	As needed	
	Calibration	1 year	Must be performed by qualified personnel with certified equipment
ABPR	Align the valve		Refer to the Automated Back Pressure Regulator Manual (MN-04) for instructions
	Replace seat		
	Replace needle		
	Replace packing		

*Performance maintenance at scheduled intervals or as needed.

Pressurizing and Leak Checking

Pressurizing the system and performing a leak check is required when the system is initially installed and after each component or column change.

1. Ensure the valves are in the following positions:

Valve	Position
MV1 – Prime	Closed
MV1 - System	Open
MV2	Open
MV3 thru MVX	Closed
MBPR1	Fully CW

2. Open the CO_2 supply tank and allow the system to reach tank pressure.

3. Start at the CO_2 supply cylinder and check all fittings in the CO_2 flow path for leaks. Use a small amount of leak detector per fitting and a paper towel to catch any excess.

4. If a leak is detected, remove pressure from the fitting. Then tighten the fitting a small amount at a time and recheck. Tighten just enough to stop the leak.

WARNING

Remove the pressure from the fitting before tightening. Tightening or loosening a fitting that is under pressure may cause the fitting to fail.

CAUTION

Over-tightening fittings will reduce the seating ability of the ferrule.

Cleaning Instructions

A cleaning method can be developed for any process, however, it is the responsibility of the user to either have their QC department check to make sure all the solvent coming out of the cyclones contains no residual product, or do the QC work themselves. Thar cannot develop a universal cleaning method for proprietary compounds where no material properties are publicly available.

NOTE	**Cleaning is the responsibility of the user.**

Collection Vessel Cleaning

1. Set Co-solvent to 40 - 100%.
2. Start Method.
3. Select each cyclone manually from the main SuperChrom screen. Right-click on the collection valve and select “Start Collection”.

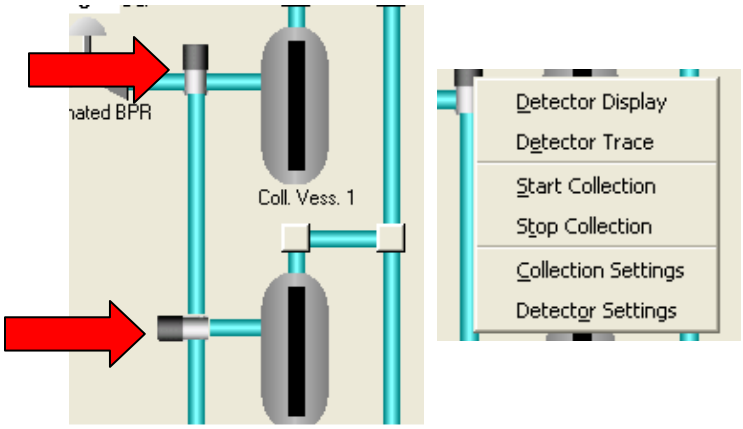


Figure 7-1 Select Collection Valve

4. Or Turn On each cyclone valve from the “UV Input” Screen.

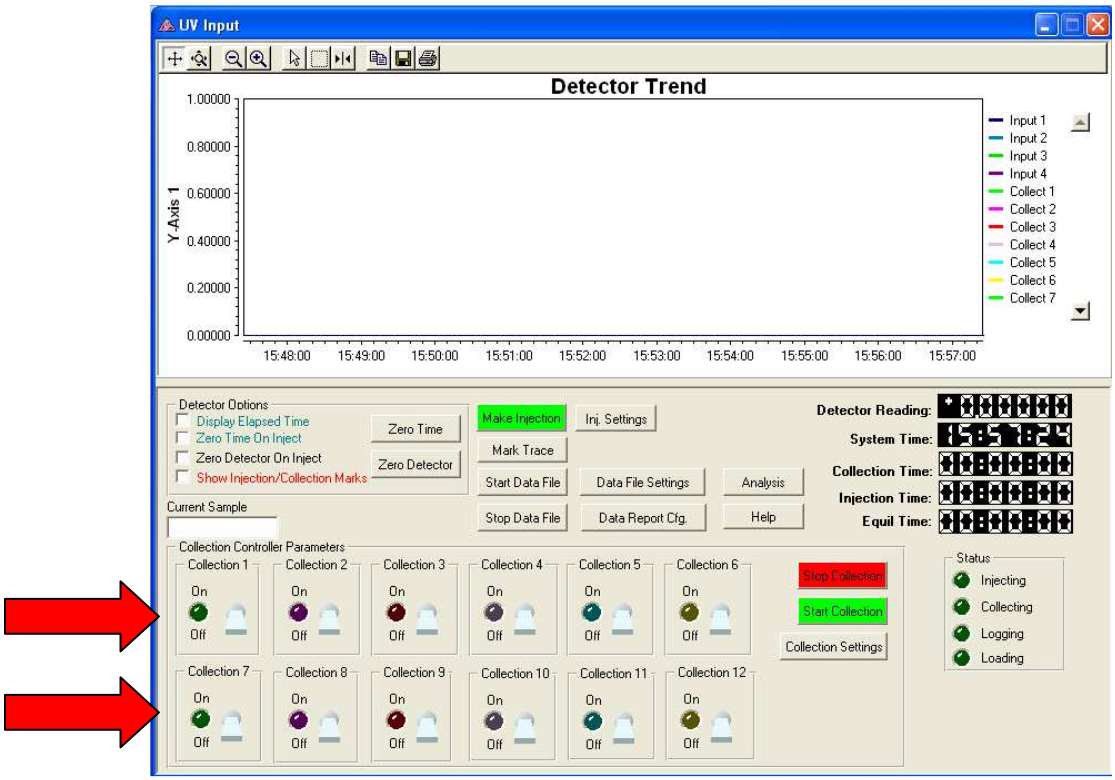


Figure 7-2 Turn on Collection

5. After 50 to 100 mL of co-solvent has run through the cyclone, click **Stop Collection** or turn the Collection switch to “Off”.

6. Click “Stop All” and manually drain each cyclone into the collection bottles/container.

7. Check the level from the level controller in each Fraction Collector to ensure it is completely drained.

Injection Module Cleaning

1. Place the “Sample tubing” on the Injection module in the waste beaker and rinse the outside of the tubing with methanol.
2. Click **Wash** in the Injection Module window. Ensure a response in the hardware and the software (a wash includes 2 full aspirations of the syringe).
3. To double check the wash
    a. Start a method
    b. Take the “Sample Tubing” out of the waste beaker and click **Inject** this will inject any solvent left in the sample tubing from the wash.
    c. View the UV data from this injection to determine if there is any residual product in the sample line.

Appendices

Appendix I SFC Prep 350 Technical Specifications

Appendix II SFC Prep 350 System Settings

Appendix III Waste Electrical and Electronic Equipment (WEEE) Directive

Appendix IV Consumables and Spare Parts

Appendix V Declaration of Conformity

Appendix I
SFC Prep 350 System Technical Specifications

Physical and Environmental

			Comments
Dimensions (width x depth x height)	System	107 x 92 x 135 cm (42 x 36 x 53 in)	
	Water bath	40 x 43 x 63 cm (16 x 17 x 25 in)	
	Computer	20 x 43 x 43 cm (8 x 17 x 17 in)	
	Computer monitor	43 x 41 x 43 cm (17 x 16 x 17 in)	
Weight	System	136 kg (300 lb)	
	Water bath	65 kg (145 lb)	
	Computer	20 kg (45 lb)	
	Computer monitor	20 kg (25 lb)	
Operating temperature			Optimal: 18 - 27°C (65 - 81°F)
Relative humidity			Optimal: 25 – 60%
Maximum oven temperature setting			80°C

Operational

Maximum pressure	400 bar
Maximum operating temperature	80°C
Maximum heat rate	~6°C/min
Temperature accuracy	± 0.5°C

General

Sound pressure level	< 70 dB
Working temperature	5 - 40°C (41 - 104°F)
Storage temperature	-20 to +60°C (-13 to +140°F)
Humidity	20 – 80% RH
IP rating	IP22
Duty rating	Continuous
Pollution degree	2
Wetted parts	SS316, SS304, GFPM

Programming

Control	User customizable PI

Electrical

Power requirements		90 – 179 VAC; 50/60 Hz System - 28amps Computer and Monitor – 2amps 180 – 255 VAC; 50/60 Hz System - 18amps Computer and Monitor – 2amps
Fuses	P-350	00467 Fuse, 10A SlowBlo (110V Units)
		00555 Fuse, 10A SlowBlo (220V Units)
	P-200	00467 Fuse, 10A SlowBlo (110V Units)
		00555 Fuse, 10A SlowBlo (220V Units)
	Six-Zone Heat Controller	04527 Fuse, 2.0Amp 3AG Fast Blow 04526 Fuse, 1.0Amp 5 X 20mm Fast Blow
	ABPR	04334 Fuse, 1.6Amp 5 X 20MM Slow Blow (220V units) 00382 Fuse, 3Amp, 1/4" X 1-1/4' GLS Slow Blow (110 V uni
	Injection Module	04334 Fuse, 1.6Amp 5 X 20MM Slow Blow (220V units) 00382 Fuse, 3Amp, 1/4" X 1-1/4' GLS Slow Blow (110 V uni
	Collection Module	04334 Fuse, 1.6Amp 5 X 20MM Slow Blow (220V units) 00382 Fuse, 3Amp, 1/4" X 1-1/4' GLS Slow Blow (110 V uni

UV detector	04334 Fuses(2), 1.6Amp 5 X 20MM Slow Blow (220V units) 04334 Fuse(1), 1.6Amp 5 X 20MM Slow Blow (110V units)
	All fuses UL- listed and CSA-certified
Grounding-system	Requires properly grounded receptacle

*Normal operating current. High inrush currents are possible at power on.

Communication

Inputs

Serial communication port	RS232C (9600 baud, 8 data bits, odd parity, 1 stop bit) Rocketport (9600 baud, 8 data bits, odd parity, 1 stop bit)
PC control	Microsoft Windows XP

Appendix II
SFC Prep 350 System Settings

Pump Settings and Operational Ranges

Operational Ranges

Operating pressure range:	55 – 400 bar
Total system flow rate:	100 - 350 g/min
CO_2 flow rate:	40 – 350 g/min
Modifier flow rate:	20 – 200 mL/min

CO_2 Pump (P350) Settings

1. Right click on the CO2 pump.
2. Click on CO2 Pump Settings.

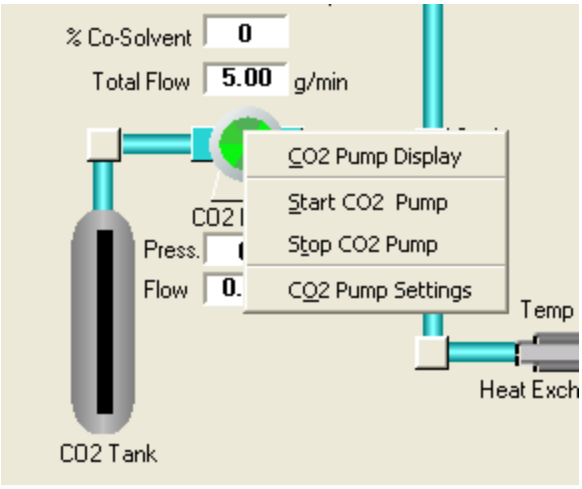


Figure I-A Select CO2 Pump Settings

3. Check the settings against the following table.

Table I-A CO_2 Pump (P350) Settings

Attenuation Factor	1
Equation Factor	10
Proportional Constant	1
Integral Constant	2
Integral Max	15000
Derivative Constant	1
PID Threshold	0
Control By	Flow

Co-Solvent Pump (P200) Settings

1. Right click on the Co-Solvent pump and select Co-Solvent Pump Settings.

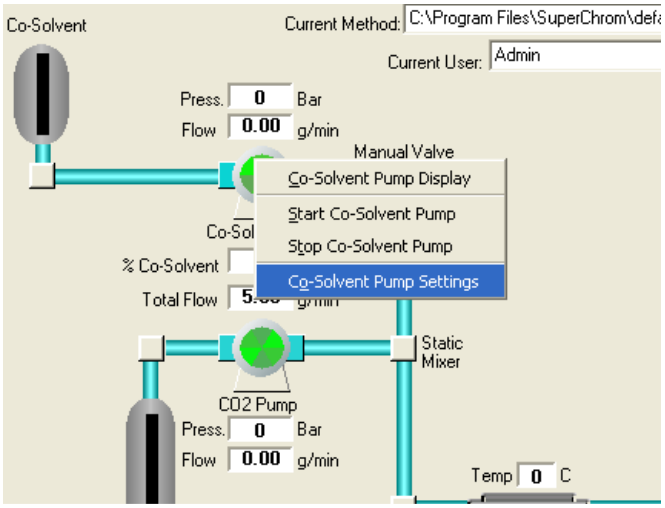


Figure I-B Select Co-Solvent Pump Settings

2. Check the settings against the following table.

Table I-B Co-Solvent Pump (P200) Settings

Attenuation Factor	1
Equation Factor	12
Proportional Constant	2
Integral Constant	1
Integral Max	8000
Derivative Constant	1
PID Threshold	1000
Control By	Flow

Heat Exchanger Settings

1. Right click on the Heat Exchanger and select Heater Controller Settings.

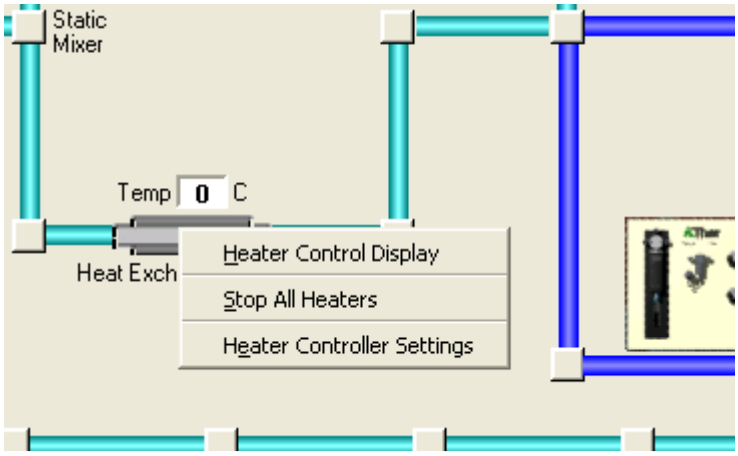


Figure I-C Select Heater Controller Settings

2. Change each zone at the top of the screen in Zone Settings>>Select Zone.

Figure I-D Zone Settings

3. Compare each zone to the following tables.

Table I-C Zone 1 Settings

Zone Name	HE 2		
Temp Set Point	35	Alarm Set Point	60
Max. Temp	60	Max. Alarm	60
Cycle Time	5	Dead Band	0
Proportional	299	Integral	349
Derivative	87	Upper Integral B	299
Lower Integral B	299	SSR Slope Min	0
SSR Slope Max	100	SSR Slope	1000
TC Type	J		

Table I-D Zone 2 Settings

Zone Name	Mobile Phase Temp		
Temp Set Point	35	Alarm Set Point	60
Max. Temp	60	Max. Alarm	60
Cycle Time	5	Dead Band	0
Proportional	299	Integral	349
Derivative	87	Upper Integral B	299
Lower Integral B	299	SSR Slope Min	0
SSR Slope Max	100	SSR Slope	1000
TC Type	J		

Table I-E Zone 3 Settings

Zone Name	HE 3		
Temp Set Point	35	Alarm Set Point	60
Max. Temp	60	Max. Alarm	60
Cycle Time	5	Dead Band	0
Proportional	425	Integral	33
Derivative	1	Upper Integral B	425
Lower Integral B	425	SSR Slope Min	0
SSR Slope Max	100	SSR Slope	1000
TC Type	J		

Table I-F Zone 4 Settings

Zone Name	NONE **OR** HE4 if system has been upgraded to modifier stream injection		
Temp Set Point	35	Alarm Set Point	60
Max. Temp	60	Max. Alarm	60
Cycle Time	5	Dead Band	0
Proportional	553	Integral	607
Derivative	151	Upper Integral B	553
Lower Integral B	553	SSR Slope Min	0
SSR Slope Max	100	SSR Slope	1000
TC Type	None **OR** J if system has been upgraded to modifier stream injection		

Table I-G Zone 5 Settings

Zone Name	NONE		
Temp Set Point	35	Alarm Set Point	60
Max. Temp	60	Max. Alarm	60
Cycle Time	5	Dead Band	0
Proportional	553	Integral	607
Derivative	151	Upper Integral B	553
Lower Integral B	553	SSR Slope Min	0
SSR Slope Max	100	SSR Slope	1000
TC Type	NONE		

Table I-H Zone 6 Settings

Zone Name	NONE		
Temp Set Point	35	Alarm Set Point	60
Max. Temp	60	Max. Alarm	60
Cycle Time	5	Dead Band	0
Proportional	553	Integral	607
Derivative	151	Upper Integral B	553
Lower Integral B	553	SSR Slope Min	0
SSR Slope Max	100	SSR Slope	1000
TC Type	NONE		

ABPR Settings

1. Right click on the ABPR and select ABPR Settings.

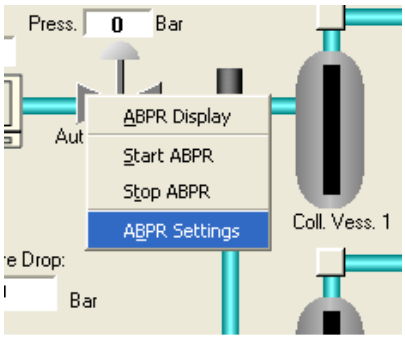


Figure I-E ABPR Settings

2. Compare the settings on the “Pressure Regulator settings” to the table below.

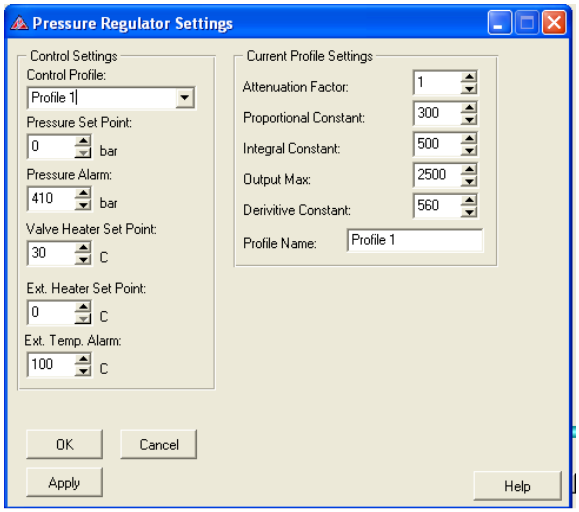


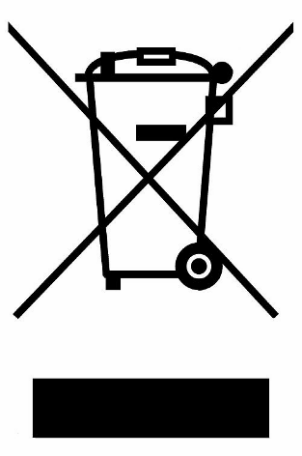
Figure I-F Pressure Regulator Settings

Table I-I Pressure Regulator Settings

Pressure Set Point	[user specified]	Attention Factor	1
Pressure Alarm	410	Proportional Constant	300
Valve Heater Set point	30	Integral Constant	500
Ext. Heater Set Point	0	Output Max	2500
Ext. Temp Alarm	100	Derivative Constant	560

Appendix III
Waste Electrical and Electronic Equipment (WEEE) Directive

In conformance with European Union Directive 2002/96/EC on Waste Electrical and Electronic Equipment (WEEE), this device may not be disposed of in domestic waste.



This disposal requirement also applies to countries outside of the EU, per their specific requirements.

Please dispose of this product in accordance with local regulations at the collecting point specified for electrical and electronic equipment.

If you have any questions, please contact the responsible authority or the distributor from which you purchased this device.

Should this device be passed on to other parties (for private or professional use), the content of this regulation must also be related.

Thank you for your contribution to environmental protection.

Appendix IV
Consumables and Spare Parts

Module	Part number	Description	Quantity
Base System	01075	Seal, 75-100 mL 1.17”VID, Polyimide	2
	06254	Disc, 1500 psi 1/4” MNPT Rupture	1
	01070	Seal, 3-50 mL, Polyimide, Med spring (EGC)	3
P-350 Pump	04093	Seal, Piston Spring	2
	04133	PEEK Seal Backup	2
	02805	Disc, 6000 psi 3/16" 72*F Rupture 3/8" OD	3
	01377	Check Valve, 3/8" Dia 10,000 Psi	4
	01794	Check Valve housing	4
	04263	Piston Assembly	2
P-200	01033	Seal, Piston Spring	
	01032	PEEK Seal Backup	
	02805	Disc, 6000 psi 3/16" 72*F Rupture 3/8" OD	
	01377	Check Valve, 3/8" Dia 10,000 Psi	
	01794	Check Valve housing	
	00754	Piston Assembly	
UV Detector	05944	Fitting, 1/16” Short Rheodyne SS	1
	05945	Fitting, 1/16" Lite-Touch Ferrule (Gilson Cell)	2
	05946	Fitting, 1/16" 10-32 Male Nut (Gilson Cell)	1
	06854	Cell Assy, SFC Prep Flow	1
	06186	Lamp, Uv - For Gilson Detector	1
	06384	Lamp, Vis - For Gilson Detector	1
ABPR-200	04497	Seat, PEEK for 04062 Micrometering	1
	06409	Kit, Rebuild for Micrometering	2
Injection Module	06828	Kit, Rebuild for Inj & Col Module 2 Pos Valve	2
	06830	Stator for Inj & Col Module 2 Pos Valve	1
	06721	Syringe, 25 ML XLP	1
	06676	Solenoid Valve Assy, 24 VDC, 250 psi, 3-way FC	1
Fraction Collection Module	06831	Kit, Rebuild for Collection Module 12 Pos Valve	1
	06832	Stator for Collection Module 12 Pos Valve	1
	06828	Kit, Rebuild for Inj & Col Module 2 Pos Valve	1
	06830	Stator for Inj & Col Module 2 Pos Valve	1

Appendix IV
Declaration of Conformity

CE

DECLARATION OF CONFORMITY

We, Thar Technologies, Inc.
575 Epsilon Drive
Suite 100
Pittsburgh, Pennsylvania 15238
USA

Declare that the product:

High Pressure Pump, model P-200A

is in conformity with the following documents:

- EEC directives 73/23 and 93/68 (low voltage directive), applied with the following standard:

  EN 61010-1: 2001 Safety requirement for laboratory equipment

Thar Technologies will not accept any liability for damages directly or indirectly caused by connecting this instrument to devices, which do not meet relevant safety standards.

- EEC directives 89/336, 92/31 and 93/68 (EMC directive), applied with the following standard:

  EN 61326: 2002 EMC requirements for electrical equipment for laboratory use

Use shielded cables and connectors for all remote connections.

May, 2007 Edward J. Bates, Operations Manager

CE

DECLARATION OF CONFORMITY

We, Thar Technologies, Inc.
575 Epsilon Drive
Suite 100
Pittsburgh, Pennsylvania 15238
USA

Declare that the product:

Automated Back Pressure Regulator, model ABPR-200

is in conformity with the following documents:

- EEC directives 73/23 and 93/68 (low voltage directive), applied with the following standard:

  EN 61010-1: 2001 Safety requirement for laboratory equipment

Thar Technologies will not accept any liability for damages directly or indirectly caused by connecting this instrument to devices, which do not meet relevant safety standards.

- EEC directives 89/336, 92/31 and 93/68 (EMC directive), applied with the following standard:

  EN 61326: 2002 EMC requirements for electrical equipment for laboratory use

Use shielded cables and connectors for all remote connections.

May, 2007 Edward J.Bates, Operations Manager

CE

DECLARATION OF CONFORMITY

We, Thar Technologies, Inc.
575 Epsilon Drive
Suite 100
Pittsburgh, Pennsylvania 15238
USA

Declare that the product:

High Pressure Pump, model P-350A

is in conformity with the following documents:

- EEC directives 73/23 and 93/68 (low voltage directive), applied with the following standard:

  EN 61010-1: 2001 Safety requirement for laboratory equipment

Thar Technologies will not accept any liability for damages directly or indirectly caused by connecting this instrument to devices, which do not meet relevant safety standards.

- EEC directives 89/336, 92/31 and 93/68 (EMC directive), applied with the following standard:

  EN 61326: 2002 EMC requirements for electrical equipment for laboratory use

Use shielded cables and connectors for all remote connections.

May, 2007 Edward J.Bates, Operations Manager

Index

A

ABPR
  Settings X
Additives 1-10
Alarms 1-4
Auto Sequencing 6-29

B

Bulk fluid 1-8

C

Circulating bath 2-4
Cleaning 7-4
CO_2 Pump 2-3
  Settings V
Collection
  Bottles 5-2
  Containers 1-12
  Instructions 6-13
  Post collection 6-27
  Settings 6-17
  Timed 6-24
  Type 6-18
  Vessels 5-23
Collection vessel
  Cleaning 7-4
Columns
  Installation 5-14
Consumables XIII
Co-Solvent 1-9
Co-Solvent Pump 2-4
  Prime 5-10
  Settings VI
Co-Solvents 2-10
Customer support iv

D

Data file settings 6-7
Declaration of conformity 1-1
Depressurize 5-15

F

Field service iv
Flammability warning 1-5
Fluids not recommended 1-11
Fraction Collection 6-13
Fraction Collection Module 2-4

G

Grounding 2-8

H

Heat exchanger 2-4
Heat Exchanger
  Settings VII

I

Icon definitions iv
Injection module
  Cleaning 7-6
  Priming-in mixed phase 5-20
  Priming-in modifier stream 5-21
  Settings 5-19
  Startup-in mixed phase 5-21
Injection Module 2-4
Installation 2-5

L

Leak checking 5-18
Level Controller Drain Settings 5-26

M

Maintenance schedule 7-1
Manual BPR 5-22
Manual Collection 6-14
Mass flow meter 2-4
MBPR Manual Back Pressure Regulator 6-11
Method
  Settings 6-1
  Startup 6-9
Mobile phase solvents 1-8, 2-10
Modifier ..... *See* Co-solvent
mV Threshold Collection 6-19

O

Operating voltage 1-6

P

Power
  Supply 2-8
Pump
  Operational ranges V

S

Safety
  Sensors 1-4
Safety features 1-3
Sample
  Connection size 5-20
Sequence
  Manual 6-32
Sequences 6-28
Shipping 1-2
Shutdown 5-27
  Long term 5-28
  Short term 5-27
Slope Threshold Collection 6-21
Spare parts XIII
Stacked injection 6-44
Stacked injection Sequences 6-35
Startup 5-1
  Method 6-9
Static mixer 2-4
Storage 1-2
SuperChrom 5-3

T

Test Injection
  Collection 6-14
Test Injections 6-12
Time Windows
  mV Collection 6-20
  Slope Threshold Collection 6-23

U

Using this manual iv
UV Detector 2-4
  Setup 5-5

V

Valve Delay
  Settings for Collection 6-13
Venting 2-11

W

Water bath 5-2

Z

Zone
  Settings VIII