

ACCQPrep Preventative Maintenance Kit

Overview

The ACCQPrep system preventive maintenance kit (ISCO part #605234815) contains the following items:

Table 1: Part List

Item	Part Number	Quantity
Lamp Assembly	605238159	1
Pump Seal Kit	880227TL	2
Injection valve rotor (coated)	209009925	1
Injection valve rotor	209009917	1
Pressure test tubing	605234817	1
Syringe	292003602	1

The table below lists scheduled maintenance tasks required for the ACCQPrep. Following the table are instructions and additional information for performing each of the tasks.

Table 2: Scheduled Maintenance Tasks

Component	ISCO Part Number	Task	Interval
Pump Seal Kit	605237101	Replacement	Annually
Lamp	605238159	Replacement	Annually
Flow cell	N/A	Cleaning (Done during the seal break in and the pressure test)	Monthly
Lamp Fan	N/A	Cleaning	Annually
Injection Valve Rotor	209009916 or 209009925	Replacement	Annually

Required Tools:

- #2 Phillips screw driver
- 1/2" open end wrench
- 7/16" open end wrench
- 5/16" open end wrench
- 1/4" open wrench
- 9/64" hex key
- 3/16" hex key
- Small needle nose pliers

Use and Disclosure of Data

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Additional Items and Solvents Required

- 100 mL ACS grade Acetone
- 3.5 L ACS grade Methanol
- 3.5 L Deionized (or better) Water
- Waste bottle
- 250 mL graduated cylinder

Time Required

- Typical time required to perform the preventative maintenance is approximately 6 hours.

Pump Seal Replacement

The following instructions are for the removal and replacement of the pump piston seals for the ACCQPrep system.

Gaining Access to the Pump

1. Turn the power OFF.
2. Access the pump compartment by grabbing the edges of front panel and pulling forward on both sides (Figure 1).



Figure 1: Front panel of the ACCQPrep

3. The solvent supply lines to the pumps have fluid in them that will be drained during this procedure. If it is acceptable to drain this fluid back into the supply bottles, follow step 'a' below, if not, go to step 'b'.
 - a. Place the solvent supply bottles below the instrument. Next, loosen the solvent supply fitting that enters the 'Y' fitting below each pair of pump heads. This will allow the fluid to drain back into the supply vessel.
 - b. Remove the solvent supply lines from the solvent bottles. Place a tray or absorbent pad beneath the 'Y' fitting below each pair of pump heads. Loosen the fitting entering the

'Y' fitting to allow the solvent to drain from the supply lines.

4. Remove the two screws securing the pump assembly in place shown in Figure 2 using the included $\frac{3}{16}$ " hex key.



Pump assembly mounting screws

Figure 2: Location of the pump assembly mounting screws

5. Remove the two stainless steel outlet lines connected to the bulkhead fittings using a $\frac{1}{4}$ " wrench. Remove the two bottom inlet lines going to the bottom 'Y' fittings and disconnect the pump power connector (Figure 3).
6. Remove the pump communication connection located behind the right side arrow pointing to the outlet lines (Figure 3).

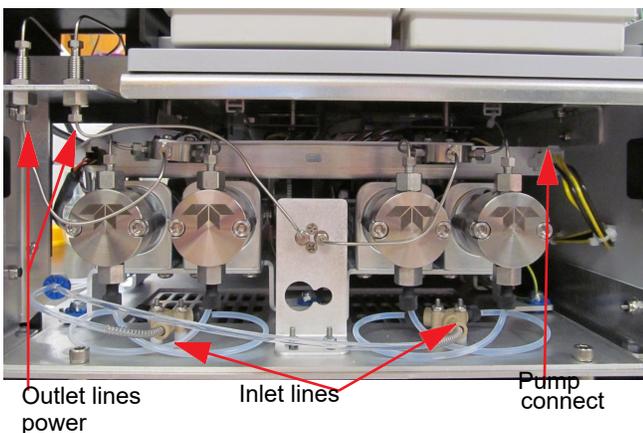


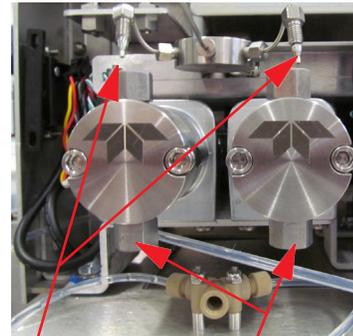
Figure 3: Pump assembly

7. Pull the pump assembly forward to the position shown in Figure 4.



Figure 4: Pump assembly

8. With the pump assembly pulled out, remove the inlet fittings connected to the bottom of the pump head. Using a $\frac{1}{4}$ " wrench, remove the stainless steel outlet lines on the top of the pump head (Figure 5).



Outlet lines removed Inlet lines removed

Figure 5: Location of fittings

Replacing the Piston Seals

1. Remove the two pump head fasteners using a $\frac{3}{16}$ " hex wrench (Figure 6).

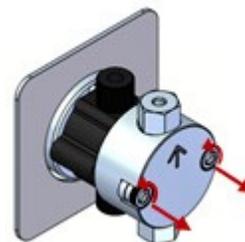


Figure 6: Removing the fasteners

2. Carefully pull the pump head forward and off the guide pins. Pull straight and slowly to prevent damage to the piston (Figure 7).

Note

Remove the washer from the piston if it did not stay in the pump head.

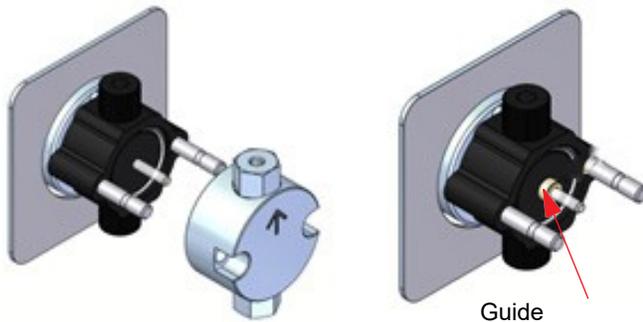


Figure 7: Removing the pump head

- Carefully pull the pump spacer block forward and off the guide pins. Pull straight and slowly to prevent damage to the piston. Also, remove the guide bushing from the piston if it did not stay in the pump spacer block (Figure 8).

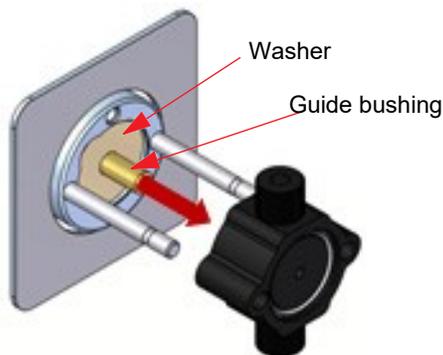


Figure 8: Removing the pump spacer block

- Insert the flanged end of the seal insertion/removal tool into the seal cavity on the pump head. Tilt it slightly so that the flange is under the seal, and pull out the seal (Figure 9).

CAUTION

Using any other tool will scratch the finish of the sealing surface and create a leak.

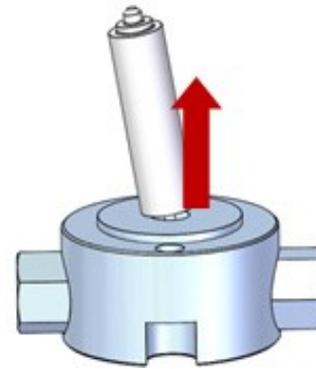


Figure 9: Removing the seals

- Use the scouring pad included in the seal replacement kit to clean the piston. Gently squeeze the piston within a folded section of the pad and rub the pad along the length of the piston. Rotate the piston frequently to assure the entire surface is scrubbed. After scouring, use a lint-free cloth, dampened with alcohol, to wipe the piston clean (Figure 10).

Note

Do not exert pressure perpendicular to the length of the piston, as this may cause the piston to break.

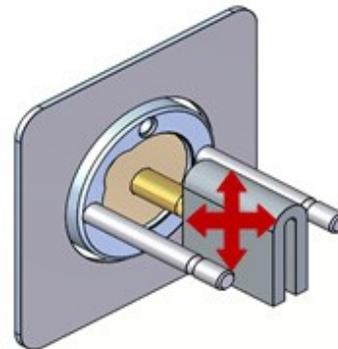


Figure 10: Cleaning the piston

- Place a replacement seal on the rod-shaped end of the seal insertion/removal tool so that the spring (energizer) is visible when the seal is fully seated on the tool. Insert the tool into the pump head (Figure 11).

Note

Be careful to line up the seal with the cavity while inserting.

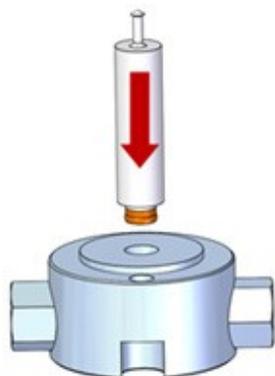


Figure 11: Replacing the seal

- Carefully replace the pump spacer block, making sure the new O-ring is properly installed.

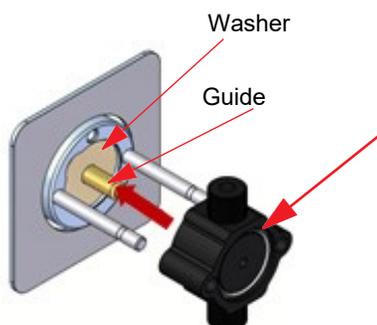


Figure 12: Reinstalling the pump spacer block

- Place the seal back-up washer on the piston. Replace the pump head. Make sure that the inlet check valves are on the bottom and the outlet check valves are on top (Figure 13).

✓ Note

Push onto guide pins straight and slowly to prevent damage to the piston. Do not force the pump spacer block or pump head into place.

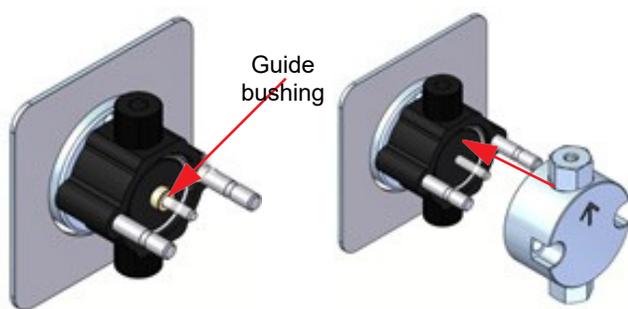


Figure 13: Reinstalling the pump head

- Reinstall fasteners. As you tighten, alternate side-to-side until snug. Turn 1 flat past snug using a $\frac{3}{16}$ " hex key (Figure 14).

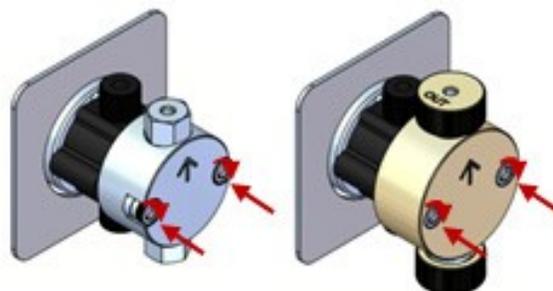


Figure 14: Reinstalling the fasteners

- Reconnect the 'Y' fittings removed in step 3a and reconnect the inlet and outlet lines. Reassemble the instrument in the reverse order of disassembly.
- Place the solvent supply lines into the proper connectors and reprime the system.

Seal Conditioning

New seals should be conditioned prior to use. Conditioning is the process of running the seals wet under controlled conditions to allow surfaces to seat and break-in for proper function of the seal. If the seals are not conditioned, the flow rates during the first few hours of operation could be up to 10% low resulting in incorrect gradient composition.

For conditioning, a premixed 50:50 solution of Methanol and water or Isopropanol and water are recommended. A premixed solution is suggested since in normal use the ACCQPrep pumps are exposed to either A solvent or B solvent, but not both.

Suggested Conditioning Method

- To provide backpressure, install the pressure tubing in the column position.
- Premix the conditioning solvents and place both the A and B inlet line in the conditioning solvent.
- Prime the system by placing the prime knob in the prime position and following the prompts.
- Program a separation 60 minutes long with a constant 50%B and 0 equilibration volume. Set the flow rate to a low 10 mL/min. Select "Collect None" so no fraction collection tubes are required.
- Start a separation.
- Increase the flow rate until the system pressure is ~1000 psi. The system pressure can be found in the status bar in the upper right corner of the screen.

7. As the seals condition, the pressure will rise slightly. Once the pressure has stabilized, increase the flow rate until the pressure is ~1500 psi and operate until the pressure has stabilized
8. Stop the system.
9. Place the supply lines in the proper solvent supply containers and prime the system.

If the solvent used to condition the seals aren't typical solvents used with the attached column, use the manual control screen to flush the column with typical solvents in a suitable storage composition, such as 50% B.

Flow Cell Cleaning

Pump clean solvent through the flow cell. This will be done during the pressure test and the seal break in.

Lamp

Follow the instructions that are included with the lamp.

Lamp Fan

With the unit in standby, wipe off the lamp fan blades.

Injection Valve Rotor

Follow the instructions included with the replacement rotors.

Note

The injection valve rotor is a common wear part and is therefore not covered under warranty. Since it is a sealing surface it can be damaged by particulates in the sample. To prolong the life of the rotor, ensure your sample is properly filtered (<20 µm) to prevent premature failure. Failure is evident by leakage from an unexpected port or location such as the sample load port after injection.

Pressure Gradient Test

The pressure gradient test consists of a capillary tubing used to create backpressure and a UV absorbing compound (acetone) mixed into solvent A. During the test run the UV trace follows the gradient change verifying gradient accuracy. The backpressure allows the check valves to function properly and verifies there are no leaks at higher pressure.

Note

Any commonly used Chromatography solvent may be used as the A or B solvent. Typically, water will be used as the A solvent and Methanol will be used as the B solvent.

Preparing the Solvent A Acetone Mixture

To prepare the mixture:

1. Measure out 200 mL of solvent A into a clean 250 mL graduated cylinder.
2. Using a syringe, add 20 mL of ACS grade acetone to the graduated cylinder.
3. Place the Solvent A filter assembly (sinker) into the graduated cylinder.

4. Gently move the filter assembly up and down several times to mix the Acetone and solvent A.

Note

The Solvent B line should be inserted into the solvent B bottle.

Installing the Pressure Test Tubing in the Place of the Column

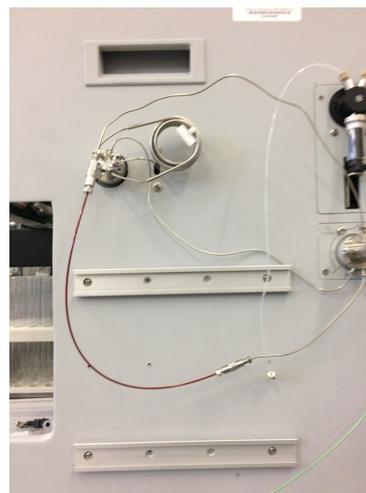
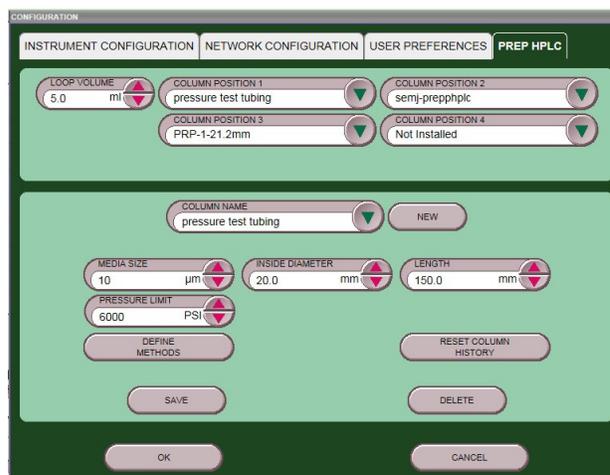


Figure 18: Side view of the ACCQPrep

Configuration Changes

To configure the changes:

1. Turn off the level sensing.
2. Configure a new column TOOLS | CONFIGURATION and select the Prep HPLC tab. and press NEW.
3. Set the column name to PRESSURE TEST TUBING, media size to 10, pressure, inside



diameter 20, and length 150.

Figure 19: Prep HPLC Configuration tab

4. Save changes.

5. If the unit has a column selection valve, select the column position where the pressure test tubing was installed (Figure 19).
6. Select the INSTRUMENT CONFIGURATION tab because the A line is in a narrow graduated cylinder, the solvent level sensing system should be turned OFF (Figure 20).
7. Select the USER PREFERENCES tab to turn On the graph system pressure option.

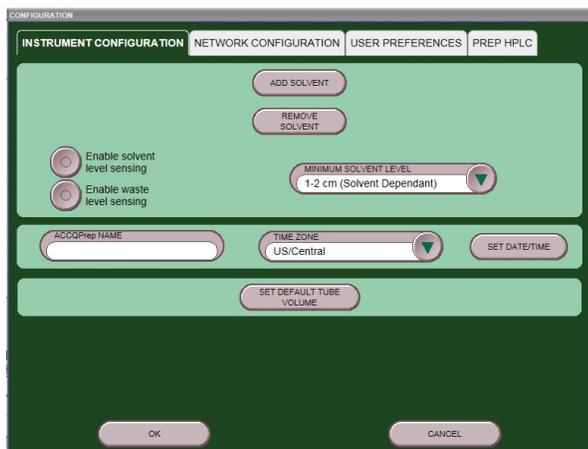


Figure 20: Instrument Configuration tab

8. Leave the CONFIGURATION screen.

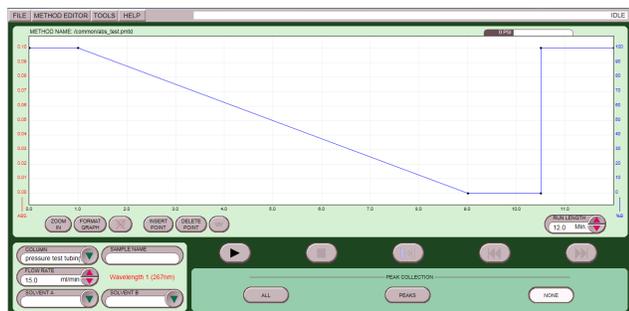


Figure 21: Main screen loaded

9. On the MAIN menu, in the COLUMN TYPE drop down menu select PRESSURE TEST TUBING.
10. Starting with the standard 20x180 mm method, create the following method:
 - o Wavelength set to 267 nm.
 - o Collection set to none.
 - o Gradient (Table below and in Figure 22).
 - o Flow Rate: 15 mL/min.

Length (min)	%B
0	100%B
1	100%B
8	0%B
1.5	0%B
0	100%B
1.5	100%B

11. SAVE the method.



Figure 22: Method to create gradient

Priming/Flushing the System

To ensure the system has the desired solvents in the flow cell and entire flow path, prime and purge the system.

1. With the A and B solvent lines in the correct solvent, turn the prime purge knob to PRIME and follow the on-screen instructions.
2. Go into the TOOLS manual control tab.
3. In the column drop down select PRESSURE TEST TUBING.
4. In the flow path, select through loop and column.

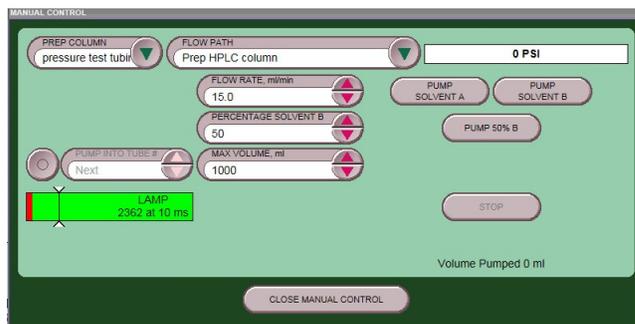


Figure 23:

5. Prime solvent A 15 mL and solvent B 30 mL.
6. Close the MANUAL CONTROL screen.
7. Remove the bottom front panel of the ACCQPrep and from the MAIN screen:
 - a. Verify the previously setup run is loaded.
 - b. Press PLAY to start the test run.
 - c. Choose MANUAL INJECTION.
 - d. Choose START EQUILIBRATION.
 - e. Watch the tubing for air going into the A and B inlet tee.

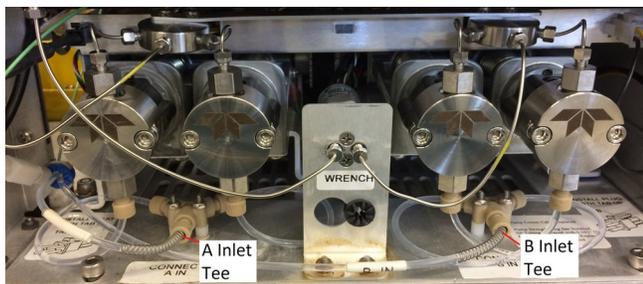


Figure 24: Location of tubing

Interpreting the Test Run

The following are ideal results:

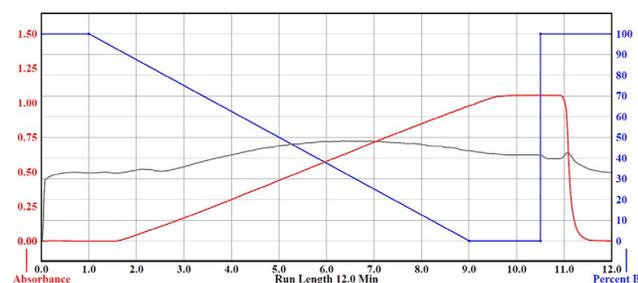


Figure 25: Ideal results

- The UV Trace when at 100% or at 0% B is flat.
- The UV trace during the gradient change is straight.
- The maximum UV is between 0.9 AU and 1.1 AU.
- The pressure is relatively stable no spikes or dropouts.
- The pressure changes gradually with gradual gradient changes and sharply with sharp gradient changes.
- If Maximum or UV absorbance does not match the ideal results:
 - Improper solvent A to Acetone ratios, the below graph shows solvent A mixed improperly. In the example below, acetone made up about 5% of the A mixture.

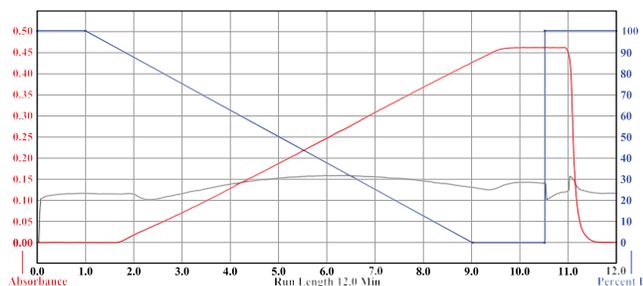


Figure 26: Non-ideal results

- Solvent is contaminated, 5 mL acetone dissolved in 100 mL solvent should have a UV spectrum as shown below. If it does not the solvent is contaminated.

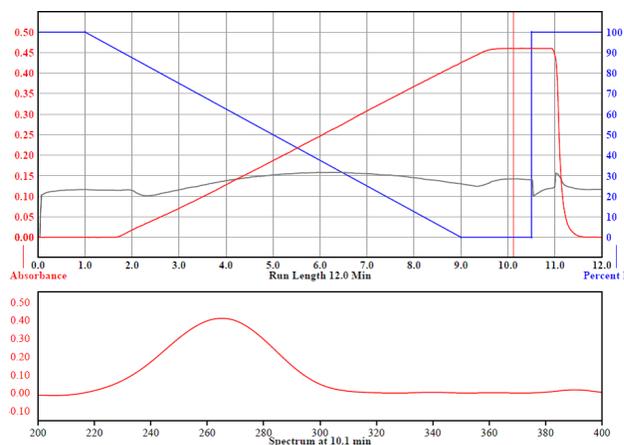


Figure 27: 5 mL acetone dissolved in 100 mL solvent

- High grade HPLC or better is used. HPLC or high grade acetone will typically produce a slightly higher maximum absorbance. To turn on the UV spectrum with the run open click on the uv button, select the chromatogram at the highest absorbance.
- If the pressure drops:
 - Typically caused by air entering the pumps.
 - The air bubble must be slightly larger than the internal diameter of the tubing for the UV trace or pressure trace to register it. Air will typically build up in the high point of the A and B inlet lines and not be pushed out until the pump flow rate reaches around 7 mL/min, because of this air bubbles if present will commonly be seen around 4.5 min during this test. The air in the below test was induced at 2 min.

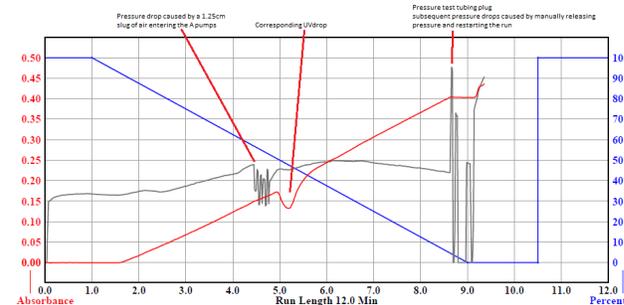


Figure 28: Results with air bubble

- Nonlinear UV Trace
 - Typically caused by failing check valves, the below graph shows at around 3 min a nonlinear UV trace caused by failing A pump check valves

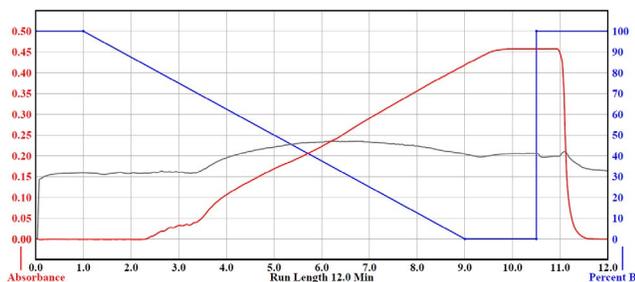


Figure 29: Results from failing A pump check valves

- If the Check valves are severely failing, as shown below, a corresponding pressure drops and spikes will be seen as shown below. No air was seen in the lines during the following graph.

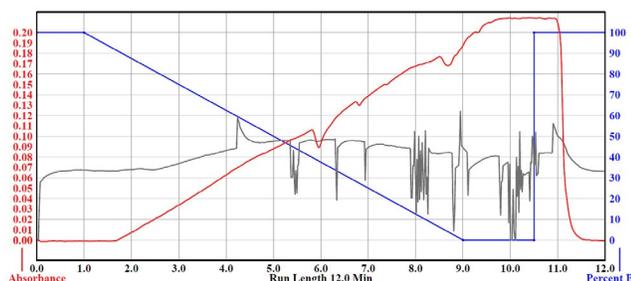


Figure 30: Results with severely failing check valves

- Following the pressure gradient test.
 - Prime the system with the desired solvents then perform a leak check.

Leak Check

The pressure test tubing can be run up to 6,000 psi. To verify the system does not leak:

1. Go to the MANUAL CONTROL screen run 50% B through sample loop and column at 10 mL/min increase the flow rate until the pressure reads approximately 5,500 psi.
2. Visually inspect for leaks.
3. Let the system run at 5,500 psi for a minimum of 10 minutes.

Teledyne ISCO

P.O. Box 82531, Lincoln, Nebraska, 68501 USA
Toll-free: (800) 775-2965 • Phone: (402) 464-0231 • Fax: (402) 465-3001

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