**Number 3**

# *The Effect of Air Bubbles on Injection Precision of Variable Autosamplers*

Air bubbles in the syringe are a common problem with most syringe based autosamplers. These bubbles can be a cause of poor injection reproducibility however, if the injection reproducibility is not affected, there is no need for concern. There are many sources for air bubbles. Finding the source of the air and eliminating it can reduce a lot of operator frustration.

One source is dissolved air in undegassed priming solution. This can be prevented by either degassing the priming solution before use, or even better, incorporating either a continuous Helium or a continuous vacuum degassing system. Some bubbles are the result of air permeating the tubing of the syringe drive system. Alcott Chromatography has minimized this source of air by using PEEK tubing in all the plumbing associated with the syringe drive. In some applications however, the PEEK tubing must be replaced with PTFE tubing for solvent compatibility reasons. PTFE tubing is permeable to air.

Finally, some bubbles are the result of cavitation by the syringe plunger. This can occur if the sample is too viscous or if the plunger tip is worn. If viscous samples are the cause, reduction the sample fill rate can minimize this effect. If the plunge tip is worn, it can be replaced with a new one.

Once the source of the air has been found and eliminated, the syringe must be reprimed. The two syringe priming techniques for the Model 718 Autosampler are described below.

## *Syringe Priming--Automatic*

Automatic syringe priming utilizes the Front Panel ***RINSE*** function to affect purging of the syringe, priming valve, needle, needle port, and all associated plumbing. The complete mechanical description is given in the introduction to ***Chapter 2, General Description,*** in the Model 718 Users Manual.

Before using the automatic priming feature, obtain a small container such as an 250 mL Erlenmeyer flask and fill it with the solvent solution to be used for priming. The chromatographic Mobile Phase for the analysis to be used with the Autosampler is the ideal priming solution. Place the priming solution dipper line into this container. (The priming solution dipper line is the PEEK tubing with the PTFE filtering frit attached.) Automatic syringe prime is now ready to be used by pressing the [RINSE] button ([FUNCTION] [9]) and following the given menu commands. As mentioned in the previous section, the 250 µL syringe usually requires only one or two rinse strokes to rid the tubing and syringe of air bubbles. The 50 µL syringe may require as many as twenty rinse strokes to remove air bubbles.

## *Syringe Priming--Manual*

Manual syringe priming may be necessary in cases where priming the syringe using the Front Panel ***RINSE*** function does not remove the air bubble from the syringe. This is more likely to occur when the 50 µL syringe is installed. Before beginning the procedure, park the needle by using ***Park Needle*** in the ***SYSTEM*** function menu. Place a small beaker or other small container under the Needle to catch the

Rinse Solution overflow. Make sure the Rinse Solvent reservoir is full and the Rinse Solvent dipper line is well submerged and primed. Have an adequate supply of laboratory towels to wipe up possible solvent spills. Once these conditions have been met, use the following procedure to manually prime the syringe:

1. Turn the power off.

2. Remove the Syringe and Priming Valve assembly as described in the previous section, but do not remove the syringe from the priming valve.

3. Turn the Syringe/Valve assembly until the "T" handle in the priming valve is facing you.

4. Rotate this valve 90° ***counter-clockwise***. The "T" handle should now be parallel to the syringe barrel.

5. Remove the plunger from the syringe barrel.

***Note: At this point, solvent should flow out of the syringe barrel. If free flow is sluggish, or does not occur, raise the Rinse Solvent Reservoir to increase the head pressure to the solvent. A Pasture pipette bulb attached to the plunger end of the syringe barrel can be used to facilitate solvent flow. The solvent should produce a steady drip at this point. If not, check and make sure the dipper line to the Rinse Solvent is well primed and filled with solvent. If the dipper line is filled with solvent and no flow occurs, the "T" handle on the priming valve has been turned in the wrong direction and is 180° out of phase. Rotate the "T" handle 180° and repeat the instructions in this note.***

6. Once a steady drip of priming solvent has been established, insert the plunger but ***do not*** push it all the way in.

7. Rotate the "T" handle 90° ***clockwise***. The "T" handle should now be perpendicular to the syringe barrel.

8. Push the plunger completely in.

***Note: Solvent should squirt out of the needle as the plunger is pushed all the way in. If solvent does not squirt out of the needle, or if it is difficult to push the plunger in, the "T" handle on the priming valve has been turned in the wrong direction and is 180° out of phase. Rotate the "T" handle 180° and push the plunger in.***

9. Check the syringe for air by repeating step 3 and retracting the plunger but not removing it from the syringe. If no air bubble is present, repeat Steps 6 and 7 and reinstall the Syringe/Priming Valve assembly. If a bubble is present, repeat Steps 4, 5, 6, and 7 until all the air is gone.

***Note: When reinstalling the Syringe/Priming Valve Assembly, the "T" handle should be perpendicular to the syringe barrel.***

10. Return power to the Autosampler and use the Front Panel ***RINSE*** function to check for bubbles.