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Do you lay awake at night thinking about trying to minimize the amount of dead volume in your LC system? Oh, you do? Well then, I am glad that you have found this article because we have a product for you! Extra dead-volume can be the thing of nightmares for your chromatography if not properly minimized in your LC system. So, let us face our fears together and defeat dead-volume with Phenomenex new finger tight connection solution—SecurityLINKs!



Often when using the standard HPLC column dimensions of 150 x 4.6 mm with a 5 μ m particle you don't notice all the dreadful extra dead-volume you are letting live in the system. But when you begin to lower the internal diameter of the column as well as the particle size, this hiding dead volume rears its ugly head in the form of poor asymmetry. One of the easiest places where extra dead volume can be minimized is in column connections. With traditional stainless steel tubing, once the ferrule has been swedged or bound to the tubing it cannot be adjusted and is permanent (Figure 1). The study below shows how an improperly swedged ferrule can cause major issues with regards to your chromatography compared to a zero dead volume connection.

SecurityLINK UHPLC Fitting System

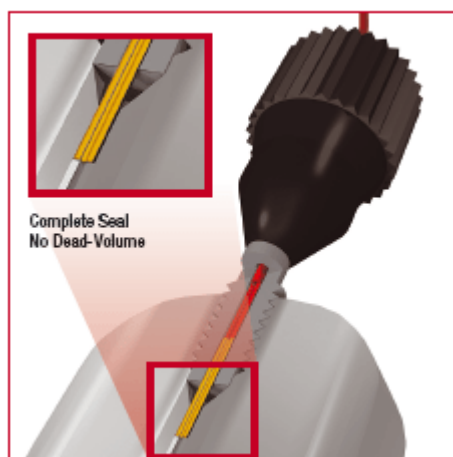


Figure 1: Connection made with SecurityLINKs tubing flush with column inlet

Poorly Connected Conventional Fittings (Nut and Ferrule)

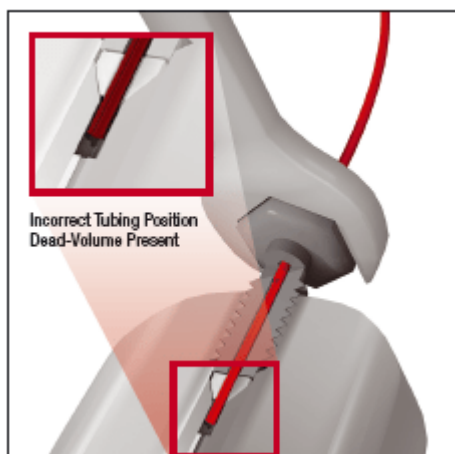
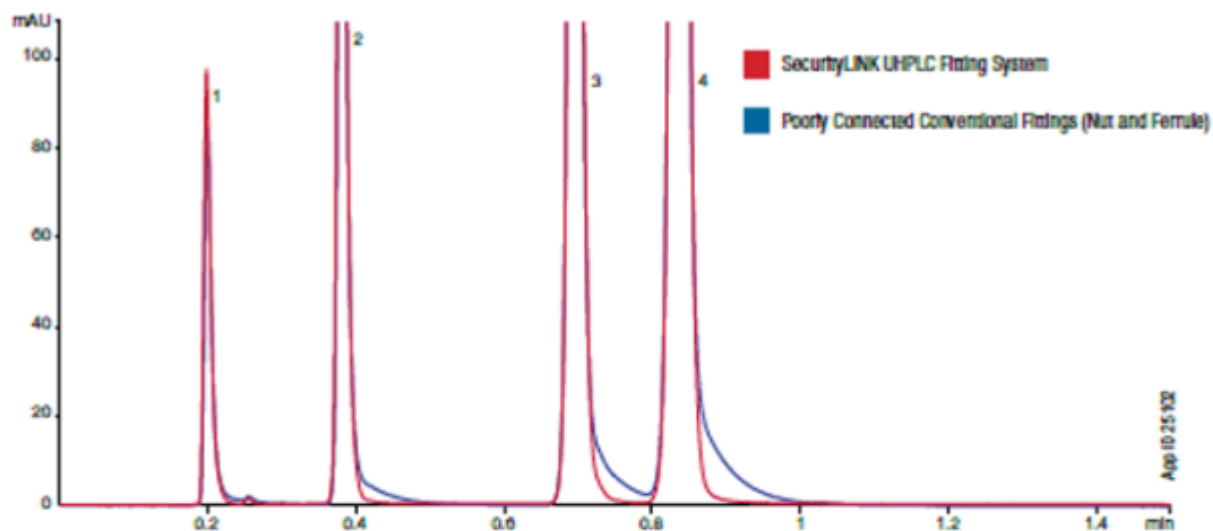


Figure 2: Incorrectly Swedged for Column showing inlet depth

Figure 3. Chromatogram of Poor Connection and Good Connection Overlay



UHPLC Conditions

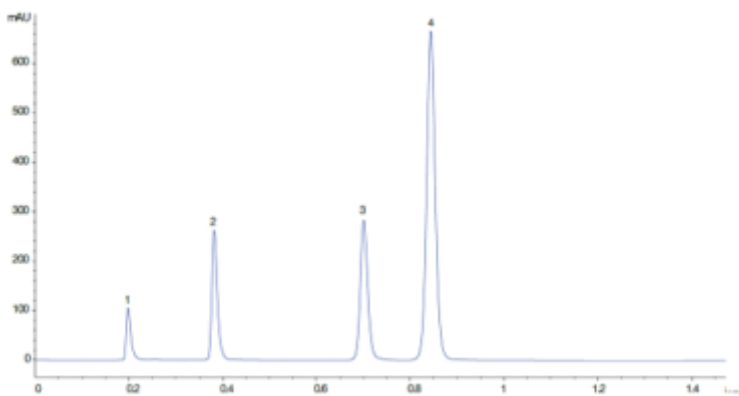
Column: Luna® Omega 1.6 µm C18
 Dimensions: 50 x 2.1 mm
 Part No.: 00B-4742-AN
 SecurityLINK Part No.: AJ1-1441
 Mobile Phase: Water/Acetonitrile (35:65)
 Injection: 0.2 µL
 Flow Rate: 0.5 mL/min
 Temperature: 25 °C
 Detection: UV @ 254 nm
 Sample:

1. Uracil
2. Acetophenone
3. Toluene
4. Naphthalene

Let's talk column hardware. It's a known issue that not all column inlet depths are created equal. Literally, different columns from different vendors have different inlet depths. You can see how this would become an issue if you were attempting to minimize your dead-volume to improve your chromatography but employ columns with varying column inlet

depths for your many methods. It would be crazy to think that you would keep a set of stainless steel tubes, one for each vendors column, to ensure the best results. With SecurityLINK systems, no collection is needed. SecurityLINK connections can adjust to the length of the tubing to fit any columns inlet. The study below shows the same SecurityLINK employed on the same system only altering what column was run. Another vendor's zero dead-volume connection system is used for performance comparison.

Figure 4. Phenomenex Luna® Omega 1.6um C18 50 x 2.1mm



UHPLC Conditions for all Columns:

Column: Luna Omega 1.6 µm C18
Agilent ZORBAX Eclipse Plus 1.8 µm C18 RRHD
Waters ACQUITY UPLC 1.7µm BEH C18
Dimensions: 50 x 2.1 mm
SecurityLINK Part No.: AJ1-1421, AJ1-1441
Mobile Phase: Water/Acetonitrile (35:65)
Flow Rate: 0.5 mL/min
Temperature: 25°C
Injection Volume: 0.2 µL
Sample:

1. Uracil
2. Acetophenone
3. Toluene
4. Naphthalene

Figure 5. Agilent® ZORBAX® Eclipse Plus C18 RRHD 1.8 µm 50 x 2.1 mm

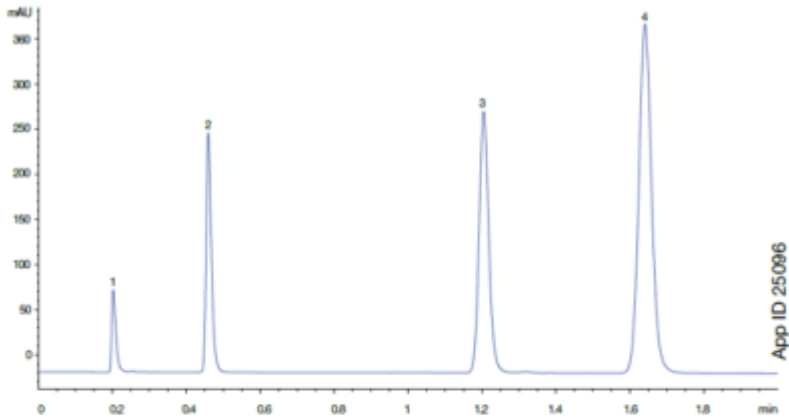
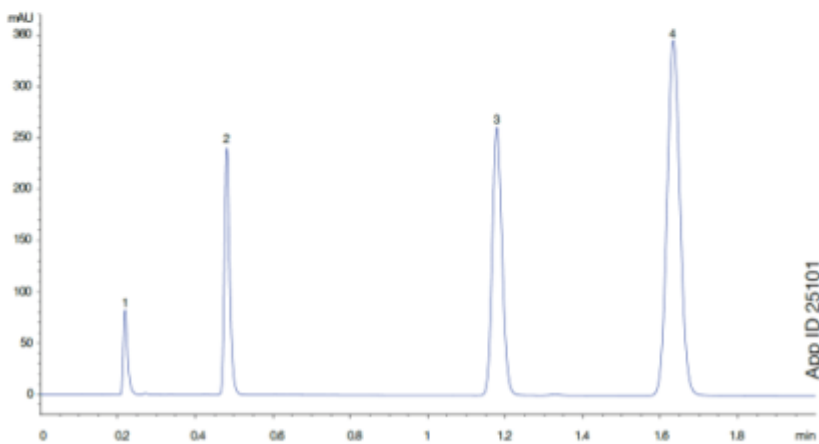


Figure 6. Waters® ACQUITY® UPLC® BEH C18 1.7 µm 50 x 2.1 mm



Comparative separations may not be representative of all applications.

Now here is the part where SecurityLINK tubing becomes the thing of dreams, well at least a chromatographer's dreams. On top of providing the correct depth every time, SecurityLINK tubing also has the added feature of clicking when you have reached the proper torque during installation. This prevents possible over tightening and allows you to free your mind from worrying if you have tightened your tubing enough to ensure no leaking

will occur when your method changes over to a new higher pressure one in the middle of the night during your overnight sequence of a bajillion important samples...oh sorry, just re-living bad memories.

Press play on the video below to see for yourself just how simple SecurityLINKs are to install:

So, if you are looking for an easy care-free way to ensure that you will have a flush seal between your tubing and any column you employ than SecurityLINK connections are the product for you!

To learn more about Phenomenex SecurityLINK products, click the following link: [CLICK HERE](#)

You can also check out our full technical application “Conventional Low Volume Stainless Steel Tubing Connection Compared to Stainless Steel Zero Dead-Volume”

TN-3006

APPLICATIONS

Conventional Low Volume Stainless Steel Tubing Connection Compared to Stainless Steel Zero Dead-Volume SecurityLINK™ Connection

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Genevieve Hodson
Technical Specialist
Genevieve loves to pick up a round of blue-ball, watching lightning storms in the distance, lazy summers in the Swiss Hill Country and drinking a good cup of coffee with a book of equal good reads.

Background

When using a UHPLC system, narrow column ID, or sub-2 μm particle column, overall system dead-volume is an important variable to minimize. A bad connection can contribute greatly to system-dead-volume and result in poor peak shape and lower overall efficiency.

Introduction

The minimization of dead-volume within a UHPLC system is an important consideration. Additional dead-volume in a system will lead to more diffusion, thus causing poor peak shape and potential failing results during method transfers or system suitability testing. When utilizing a UHPLC system, even small amounts of extra dead-volume created by poor connections can contribute greatly to diffusion, due to the overall decrease in the systems dead-volume. An increase in the dead-volume of a system, for both UHPLC or HPLC, can also cause chromatography problems when columns with smaller inner diameters or core-shell/sub-2 μm particles are used. An incorrect, or poor, connection can be defined as any connection where the base of the tubing used does not sit flushed up against the inlet of the column and subsequently contributes to the dead-volume of the system. Although most chromatographers consider the connection of the column as the largest potential location for an incorrect connection, poor connections can happen in many places throughout the system, including connections from the autosampler to the column heater.

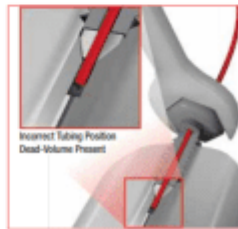
There are two main approaches to creating a zero dead-volume connection. The first involves swaging your stainless steel tubing at the exact depth of the column's inlet. While effective, this only allows for the connection to be used with one specific vendor's columns and relies on the scientist's proper measurements before the tubing is swaged together. If the distance was measured or implemented incorrectly then the result is a permanent poor connection. The second approach involves the use of a zero dead-volume connection where the connection is not permanently swaged and allows for a perfectly flush connection independent of the column's inlet depth.

This technical note investigates the effects of poor and zero dead-volume connections using a sub-2 μm particle column on a UHPLC system.

Figure 1.
Ideal Flush Connection



Figure 2.
Incorrect/Poorly Made Connection



If you found this article helpful, then you will definitely want to check out: “Tips to Maintain HPLC/UHPLC Systems and Improve Column Longevity.”

If you have any questions about the above information please reach out to our Technical Experts via Live Chat, 24/7. You might even get to chat with Genevieve herself!

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