Optimize Your Gas Chromatography Analysis Through the Use of a Retention Gap and Integrated Guard Column

Authors

Michel Pampaloni

PerkinElmer, Inc.
France

Kyle Oberndorfer

PerkinElmer, Inc.
Shelton, CT

Guy Gaspar

PerkinElmer, Inc.
Consultant
France

Introduction

Optimizing your gas chromatography (GC) is crucial in improving laboratory efficiencies and analytical performance. One way to optimize your GC analysis is by using retention gaps and guard columns. Although similar, they can serve different purposes. A guard column/retention gap is a short (1-5 m) piece of uncoated deactivated fused silica tubing which is placed in-line between the GC injection port and the capillary column. Guard columns are used to protect the analytical column and extend its lifetime. Retention gaps also act as guard columns, but their primary goal is to improve peak shape by focusing the sample components whenever introducing a large liquid sample onto the column.

This article illustrates the benefits of using retention gaps, the various possibilities of connecting a retention gap to the analytical column, and the use of integrated guard columns.

The Problem of Analyzing Low Concentration Analytes and How a Retention Gap Solves It

Analyzing samples with analytes at low concentrations in capillary GC is not an easy task. Of all the GC injection techniques the SPLIT injection is very common and quite easy, but is not always the best option. Often, we have to use more sophisticated techniques like Splitless and On-Column injection, which can be complicated and problematic. The common characteristic of these techniques is the presence of the sample in liquid phase.

Before going into the details, let's have a look at an example, see Figure 1.

The figure labeled "b) 0.5 µL without retention gap" in Figure 1 shows the separation of these hydrocarbons after injecting 0.5 µl of sample. The separation appears correct. However, if the concentration of solutes was 8 times lower, we would need to inject 8 times more sample. Performing a 4 µL injection to achieve that goal results in a very different chromatogram, as shown in the chromatogram labeled "a) 4 µl without retention gap". Note how the peaks are poorly shaped and very broad. Finally, the chromatogram labeled "c) 4 µL with retention gap" looks much better and compared favorably to the 0.5 µl result. The only difference between these two analyses is the addition of a deactivated capillary tube 1.5 m long and 0.32 mm inner diameter placed between the injector and the analytical column. This is known as a "Retention Gap".
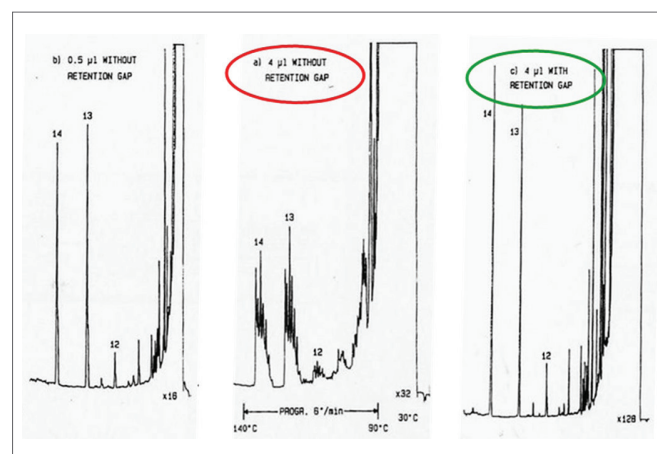

Figure 1. Alkane separation C12, C13 and C14 on a 8 m x 0,32 mm x 0,1 µm dimethyl-siloxane column, solvent acetone, on-Column injection. (from K. GROB book: On-Column Injection in Capillary Gas Chromatography) *Please note that on many of Dr. Grob's chromatograms the time goes from the right to the left!*
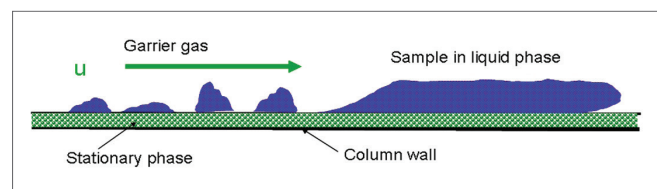

Figure 2. The spreading and the fragmentation of the liquid sample into the capillary column.

We should mention that this performance anomaly exists only with certain solvents. The more commonly encountered "bad guys" (but not the only ones) are -

- Acetone
- Dichloromethane
- Benzene
- Methanol

These solvents effect the stationary phase and result in an increased speed of the liquid sample along the stationary phase in the column. In cases where one is injecting a relatively large sample volume (> 0.5µl) the result is that the sample quickly spreads along initial length of the column while leaving behind fragments of different sizes and shapes as show in Figure 2. In the larger and thicker fragments, the solutes are partially trapped and their evaporation is delayed as compared to solutes that are dissolved within the smaller and thinner fragments. The result is a differentiation (an offset of the molecules in time and space) which explains the broader peaks. This phenomenon, according to K.Grob, is called the "flooding effect".

After adding an empty capillary tube between the injector and column (a Retention Gap), the scenario changed (see Figure 3). Evaporation in the empty tube is carried along at the same speed as the carrier gas. Upon reaching the stationary phase the forward speed is slowed. For example, for a solute with a k'=9, the chromatographic zone advances 10 times slower. Therefore, as the leading edge reaches the stationary phase and slows – the tail continues at the greater speed and closes the gap. This is similar to the effect we can see with automobiles on highways - where a toll booth results in slowing the flow of traffic. Another analogy is passengers boarding an airplane. The first passenger moves slowly, one small step at a time, at the entrance to the plane, while simultaneously the last passengers are still making faster strides several meters at a time down the ramp to the plane.

The inventor of this mechanism, K. Grob named the length of pre-deactivated column as a "retention gap".
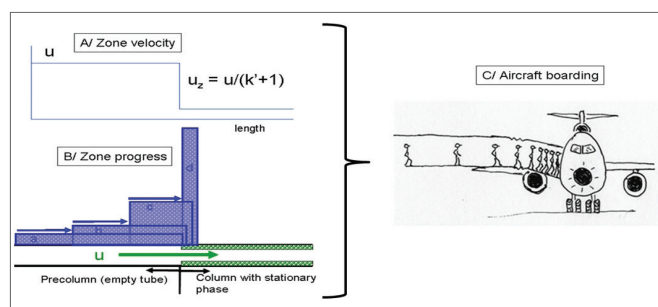

Figure 3. The sample evaporates in the empty tube and it is driven by the carrier gas; its speed is identical to the carrier gas velocity (a governed by formula shown in Fig 3 A/Zone Velocity). The front reaches the end of the tube in a distribution represented by the first rectangle from the left (Fig 3/B Zone Progress). Upon reaching the stationary phase the velocity of the initial rectangle slows – as the trailing portion catches up. This phenomenon is similar and relatable to passengers boarding a plane as show in (Fig 3/C, Aircraft boarding).[1]
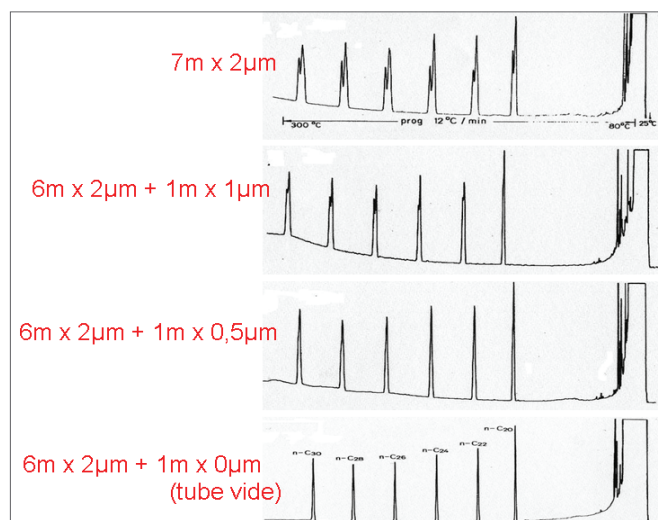

Figure 4. On-Column injection, acetone solvent, methyl-siloxane (phase immobilized) 7 m x 031 mm x 2 µm. The sample is even-numbered n-alkanes C20 to C30, injected in 1,5 µl volume. Starting with the second chromatogram from the top, the first meter is replaced in succession by reduced film thicknesses. Adapted from K. Grob's book: On-Column Injection in capillary Gas Chromatography.

Retention Gap Features and Benefits

1. Serves the role as a guard column
In On-Column injection the sample is directly injected inside the capillary column and dirt from the sample and matrix are also introduced. Prior to the use of a retention gap, this would result in degradation of the column and poor chromatography. Chromatographers used to cut the first 20 to 30 cm off the analytical column to remove the contamination and restore some level close to initial performance. Naturally, this solution has limits! However, by using a retention gap in this sacrificial role, this can be done without affecting the columns performance.

2. Creates higher efficiency
With the focusing effect, the initial band of the chromatographic zone is narrower, so are the peaks. K. Grob, in his book, showed a very educational figure (see Figure 4). The evolution of the chromatograms from top to bottom is remarkable.

3. Allows large volume injection
In the same book, K. Grob shows a spectacular chromatogram by injecting 80 µl of sample with particularly sharp peaks.

4. Quicker injection
The splitless techniques and On-columns require slow injections, especially at high volumes, which is sometimes tricky. The retention gap allows for better performance.

5. Protection of the stationary phase
When performing the injections noted above, it's possible that samples in liquid phase may attack the stationary phase. Although non-bonded phases are rare today, they do still exist (chiral phases among others). With the properly sized retention gap the sample is already vaporized when it enters in the analytical column.

6. On columns injection into "narrow-bore" (<0.25 mm ID) columns
There is no needle capable of entering such narrow tubes/IDs. Therefore, a larger diameter pre-column/ Retention Gap is needed.

Connecting a Retention Gap to the Analytical Column and Implementation

There are different ways to connect a retention gap to the analytical column. But how do you choose the right length and deactivation type?

There are two more popular options:

1. Connection with Swagelok™ type connectors (see Table 1) with double ferrule inside. The assembly is not always straightforward and does require some experience.

2. Using universal connectors (see Tables 1 and 2.)

Table 1. Swagelok™ fittings.

Product		Qty.	Size	Brass Part No.	Stainless Steel Part No.
Bulkhead Adapter		1	1/4 in. to 1/4 in. tube		N9301267
Back Ferrule		pkg. 5	1/16 in.	N9300040	N9300042
			1/8 in.	N9300036	N9300038
			1/4 in.	N9300030	N9300032
Cross Union		pkg. 1	1/8 in.	N9301259	
Front Ferrule		pkg. 5	1/16 in.	N9300041	N9300043
			1/8 in.	N9300037	N9300039
			1/4 in.	N9300031	N9300033
Male Adapter Tube to Pipe		pkg. 1	1/4 in. tube to 1/8 in. NPT	N9301266	
Male Connector		pkg. 1	1/8 in. to 1/8 in. NPT	N9301253	
			1/8 in. to 1/4 in. NPT	N9301254	
			1/4 in. to 1/8 in. NPT	N9301255	
			1/4 in. to 1/4 in. NPT	N9301269	
Nut		pkg. 5	1/16 in.	N9300058	N9300059
			1/8 in.	N9300056	N9300057
			1/4 in.	N9300054	N9300055
Plug		pkg. 1	1/16 in.		N9300053
			1/8 in.	N9300060	N9300061
			1/4 in.	N9301268	N9301233
Union		pkg. 2	1/16 in.	N9300048	N9300049
			1/8 in.	N9300046	N9300047
			1/4 in.	N9300044	
Union Tee		pkg. 1	1/16 in.	N9301221	
			1/8 in.	N9301222	
			1/4 in.	N9301223	
Reducing Union		pkg. 1	1/8 in. to 1/16 in.	N9300051	N9301225
			1/4 in. to 1/8 in.	N9300050	N9301226
			1/4 in. to 1/16 in.	N9301227	

Universal Connectors

Table 2. Universal connectors.

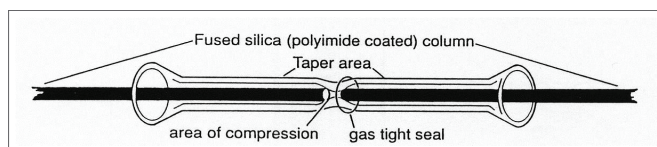| Dimension	Part No.
Universal Connector (pkg. 5)	**N9302149**
Metal Universal Connectors: 0.25 mm i.d. (pkg. 10)	**N9301167**
Universal Y Splitter (pkg. 1)	**N9303448**
Polyimide Sealing Resin (5 g)	**N9301343**
Undeactivated Presstight Column Connectors (pkg. 5)	**N9303962**

Figure 5. PerkinElmer undeactivated presstight column connector Product Number N9303962.[2]

For column "tightness" it is very important to make clean cuts at the ends of the fused silica capillary tubes before inserting them into the connector. Some users suggest wetting the ends in methyl-pyrolidone to soften the polyimide layer. Others advise putting the resin paste inside the connector to achieve the same goal.

Whatever the choice of connector (Swagelok or Universal), it will still likely remain the Achilles heel of the system – especially for GC/MS. Air intrusion in these situations is almost inevitable.

Retention Gap with Integrated Connection.
As mentioned just above, the connection is often the weakest point of the system. Currently the preferred solution for this problem is using columns with integrated guards – thus requiring no interconnection.

Capillary columns with an integrated guard offer multiple advantages over systems with a pre-column and a connector:

• No leaks for a more robust method

• No column connections for easier, faster maintenance

• No peak distortions due to connector dead volume and thermal capacity

Some chromatography literature on this subject provides complicated tables. However, based on our experience, very few simple rules will cover the vast majority of situations:

A. Required length

ID =0,32 mm :   L= 0,25 m/µl injected + 1 m

ID =0,25 mm :   L= 0,40 m/µl injected + 1 m

After selecting the appropriate ID, you can apply these equations, where the first term is capacity (proportional to the injected volume) to ensure the best performance. The addition of one extra meter serves both the role of guard column and allows for gradual cutting/trimming to remove contamination from the column.

B. Retention gap deactivation

In order to improve the focusing effect of the retention gap, it is recommended to use deactivated inner surface pre-columns. The type of deactivation is chosen as a function of the sample solvent being used:

• Non-polar deactivation for injection of none or weakly polar solvents for instance alkanes, carbon disulfide, and ethers.

• Intermediate polar deactivation for injection moderately polar solvents: methylene chloride (dichloromethane), and toluene.

• Polar deactivation for injection of strongly polar solvents such as acetonitrile, methanol, and water.

This is an important selection criterial because an incompatible solvent can result in the formation of droplets inside the tube.

Conclusion

In summary, depending on the sample matrix and injection technique that you use, you can dramatically improve your chromatographic results by simply using a retention gap. Among the various potential solutions, the use of integrated guard columns, where connections aren't required, is by far the best choice.

We offer a wide choice of Integrated Guard columns, for your GC regardless of instrument make, model or manufacturer, which eliminate the inconvenience and problems that often result in trying to make leak-free connections between a guard column and the analytical column. These innovative columns incorporate both guard and analytical columsn in a continuous length of tubing, thus eliminating the issue.

The Integrated Guard column is so cost-effective that we challenge you to compare our price against that of a conventional connection, even if you assemble it yourself. See the vast variety of PerkinElmer Integrated Guard columns in Table 3.
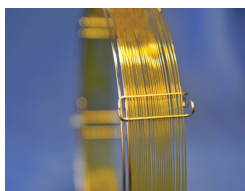
Integrated Guard Columns

Table 3. Integrated Guard Columns.

Elite-1

ID	df (mm)	Temp Limits (°C)	Integrated Guard	30 m Part No.
0.25 mm	0.25	-60 to 330/350	5 m	N9305600
0.53 mm	1.00	-60 to 330/350	5 m	N9305601
	5.00	-60 to 340/360	5 m	N9305602

Elite-5

ID	df (mm)	Temp Limits (°C)	Integrated Guard	30 m Part No.	60 m Part No.
0.25 mm	0.25	-60 to 330/350	5 m	N9305603	
	0.25	-60 to 330/350	10 m	N9305604	
	1.00	-60 to 330/350	5 m	N9305605	
0.32 mm	0.25	-60 to 330/350	5 m	N9305606	N9305607
	1.00	-60 to 330/350	5 m	N9305608	
0.53 mm	5.00	-60 to 340/360	5 m	N9305609	

Elite-5 ms II

ID	df (mm)	Temp Limits (°C)	Integrated Guard	15 m Part No.	30 m Part No.
0.25 mm	0.10	-60 to 330/350	5 m		N9305610
	0.25	-60 to 330/350	5 m	N9305611	N9305612
	0.25	-60 to 330/350	10 m		N9305613
	0.50	-60 to 330/350	5 m		N9305614
	0.50	-60 to 330/350	10 m	N9305615	N9305616
0.32 mm	0.25	-60 to 330/350	5 m		N9305617
	1.00	-60 to 330/350	5 m		N9305618

Elite-5 ms

ID	df (mm)	Temp Limits (°C)	Integrated Guard	30 m Part No.	60 m Part No.
0.25 mm	0.10	-60 to 330/350	5 m		
	0.25	-60 to 330/350	5 m		N9305619
	0.25	-60 to 330/350	10 m	N9305620	N9305621
	0.50	-60 to 330/350	5 m	N9305622	N9305623
	0.50	-60 to 330/350	10 m		N9305624
0.32 mm	0.50	-60 to 330/350	5 m		N9305625
	1.00	-60 to 330/350	5 m		N9305626

Elite-624

ID	df (mm)	Temp Limits (°C)	Integrated Guard	30 m Part No.
0.25 mm	1.40	-60 to 330/350	5 m	N9305627
0.32 mm	1.80	-60 to 330/350	5 m	N9305628
0.53 mm	3.00	-60 to 340/360	5 m	N9305629

Elite-1301

ID	df (mm)	Temp Limits (°C)	Integrated Guard	30 m Part No.
0.53 mm	3.00	-60 to 330/350	5 m	N9305630

Elite-1701

ID	df (mm)	Temp Limits (°C)	Integrated Guard	30 m Part No.
0.25 mm	0.25	-60 to 330/350	5 m	N9305631

Elite-WAX ETR

ID	df (mm)	Temp Limits (°C)	Integrated Guard	30 m Part No.
0.25 mm	0.25	-60 to 330/350	5 m	N9305632
0.32 mm	1.00	-60 to 330/350	5 m	N9305633
0.53 mm	1.00	-60 to 340/360	5 m	N9305634

References

1,2. Hüthig Buch Verlag GmbH Heidelberg GROB, Konrad:
On-column injection in capillary gas chromatography;
basic technique, retention gaps, solvent effects /
by Konrad Grog – 2.ed. – Heidelberg : Huethig, 1991
1. Page 121, Figure A42. 2. Page 374, Figure C5

For a complete listing of our global offices, visit www.perkinelmer.com/ContactUs