Ghost Peaks in Gas Chromatography Part 3:

Sample Contamination and Ghost Peaks Formed by The Stationary Phase Itself

Jaap de Zeeuw, Restek Corporation, Middelburg, The Netherlands.

In the last issue we discussed possible sources for ghost peaks related to the carrier gas and the gas lines. The injection port septa, O-rings can also be a source for ghost peaks. Here we look at the sample itself and the different "contaminations" that can result in unwanted "extra" peaks. Also the stationary phase in the column can play an interesting role in ghost-peak formation.

Sample Vials and Septa

Sample vials that are used can be a big source for extra peaks. The sample vial itself is usually sealed well in clean packaging. The challenge is the seal that is used. Different seals are available. There are caps with integrated seals and there are also "snap-caps" with separate seals. The seal-material is a big challenge as it can produce a lot of ghost peaks. It depends not only on the material used but also on the type of solvent, age and number of injections through the seal.

If a fresh sample is made today and analysed, a stable baseline with known peaks is observed. If the sample is left for two weeks and is analysed again, there will be more peaks showing up. Such peaks are extracted components (phthalates, siloxanes) from the seal material. The polymer that is used for sealing caps contains all kind of impurities, Figure 1 shows a sample that was stored in a vial with a 50 uL insert. A series of 20 injections was performed from the same vial. Figure 1A shows the first analysis and B shows the resulsts after 20 injections. With every injection, the outside of the needle was contaminated by the sealing material, contaminating the sample. Also more degradation of endrin was observed: the endrin-ketone

Figure 1

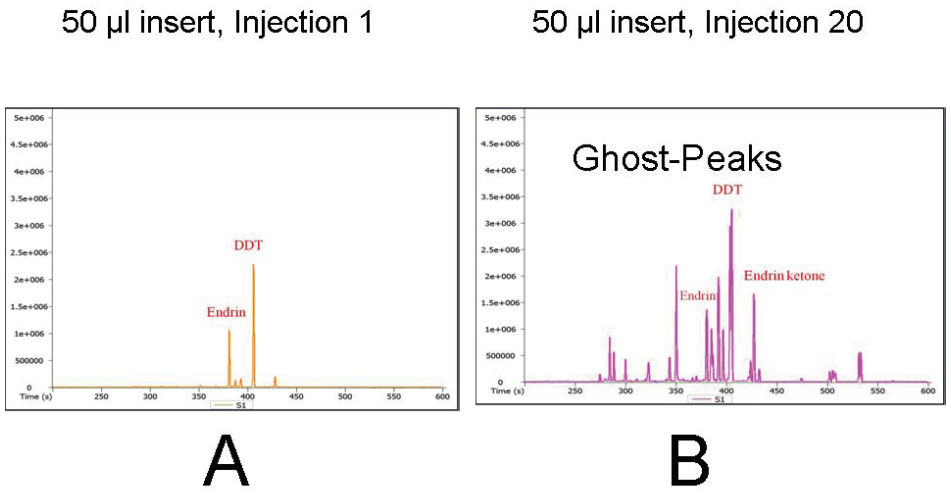

Figure 1: Contamination of sample after repeated injection using the same vial. Because the volume is small contamination increases very fast (see ref. 1 for details of the experiment).

Figure 2

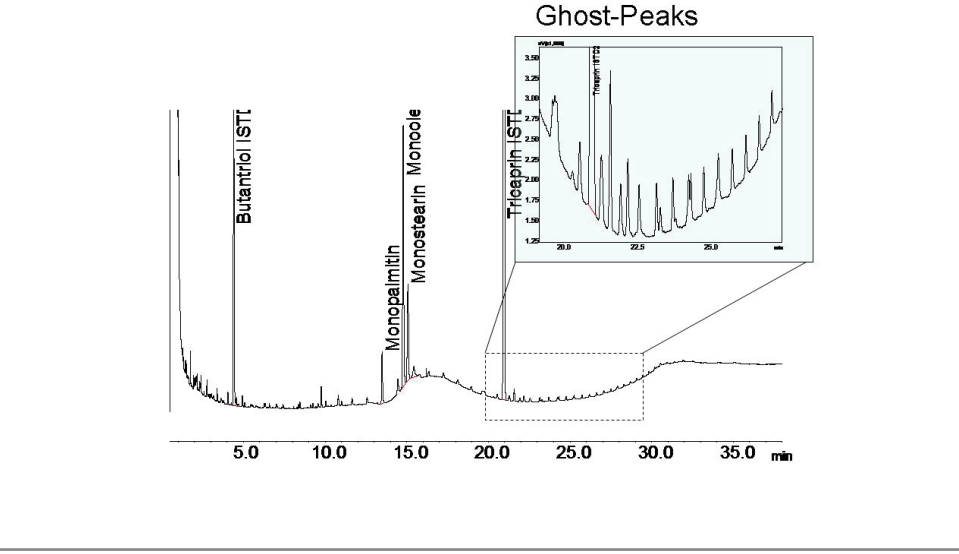

Figure 2: Ghost peaks of bleed products generated by injecting through the septum of a vial Courtesy: Franz Kramp, Shimadzu Deutschland.

Figure 3

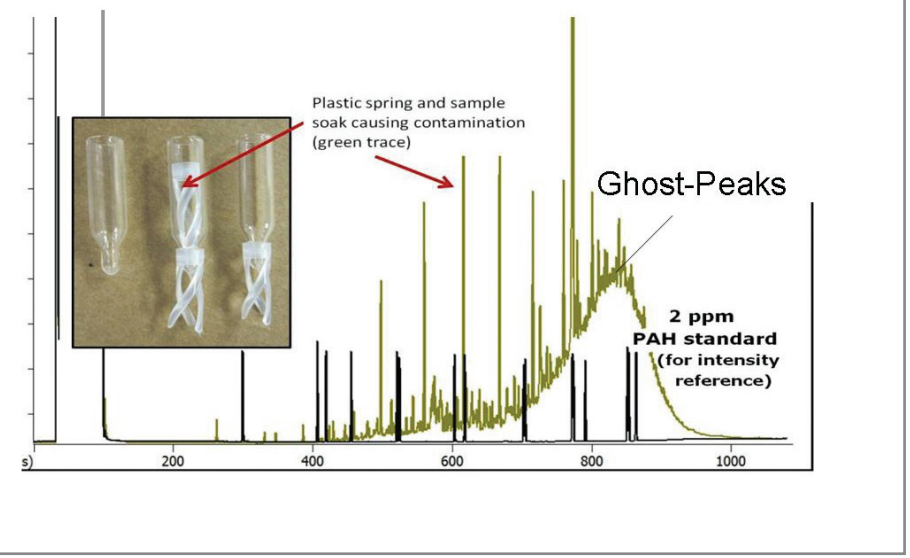

Figure 3: Ghost peaks generated by extraction of a "spring" that was left inside an insert. See ref. 2 for details.

response increased significantly after the 20 injections.

This problem causes the biggest issues in trace analysis. Another example is shown in Figure 2. The samples are injected on-column to look for late eluting triglycerides in biodiesel. If the septum of a vial is penetrated once the solvent vapour gets into contact with the seal. Also here softeners in the septum are transported in the liquid sample by the solvent vapour. This effect is accelerated with every septum penetration by the syringe needle. The effect can already be observed after three injections from one vial. As FID is used all impurities are detected and complicate the measurement of triglycerides.

Accidental Contaminations

Mistakes can always happen and here is one that even happened to an experienced person in the lab. There are vials that use a kind of polymeric "spring" whose function is to push the insert upwards, so the syringe needle will reach the bottom of the insert. The analysis was done using FID and suddenly a huge forest of ghost peaks was observed (see Figure 3). As can be seen, by accident a" spring" made it inside one of the inserts. The analysis shows an extract of this "spring", which explained the huge contamination.

Just be aware that this can happen to anyone.

Figure 4

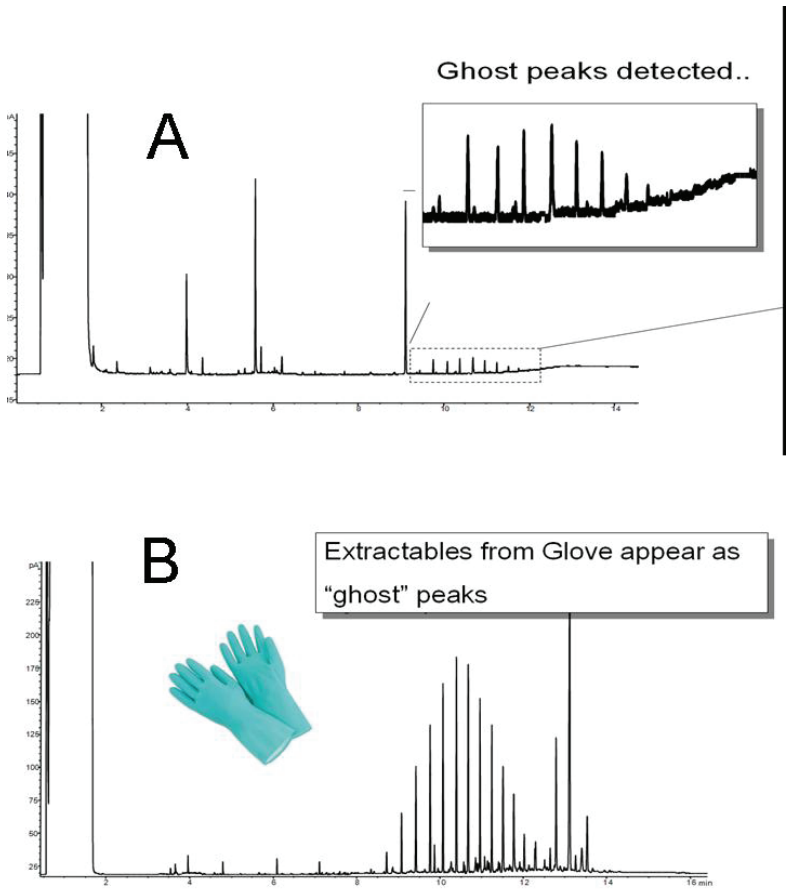

Figure 4: Ghost peaks caused by contamination of gloves. See ref. 4 for details.

Contamination by Gloves

When we work in the lab we often use the nitrile gloves to protect our hands from chemicals. The gloves, however, are also a source for contamination. In Figure 4A we were running a trace analysis and we saw this line of ghost peaks. We checked all possible sources: septa, vials, O-ring. At last we looked at the gloves we were using for protection. We took a small piece of glove and added extraction solvent. The result is shown in Figure 4B. The same peaks appeared, which gave us a good heads up that this can be a viable source for contamination. And let's be honest: If you take a new glove, you already can smell something.

Syringes

Syringes are best used for injection

Figure 5

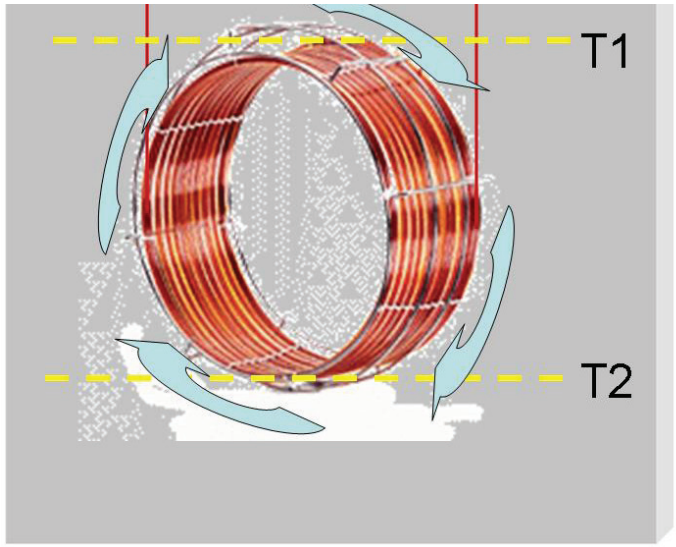

Figure 5: Schematic of column position in an oven and flows. T1 and T2 are respectively temperature at top side, and temperature at bottom side of the oven.

Figure 6

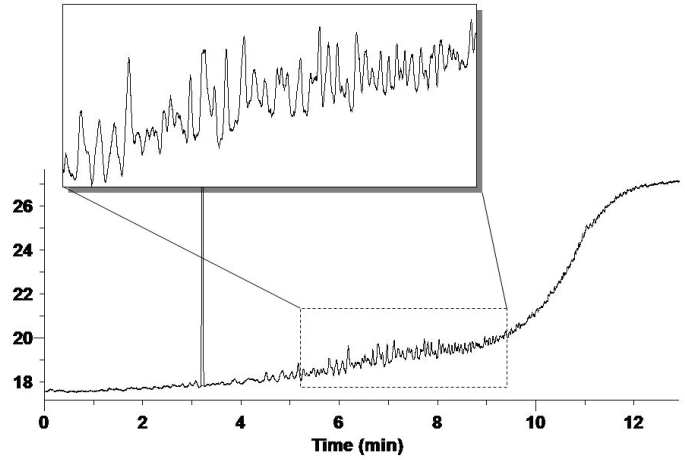

Figure 6: Baseline after fast cooling. Column : 30 m x 0.25 mm cyanopropyl/phenyl phase type 624; Carrier : H_2, 1.5 mL/min const. flow; Oven: 60 °C → 260 °C, 20 °C/min; Detector: FID.

Figure 7

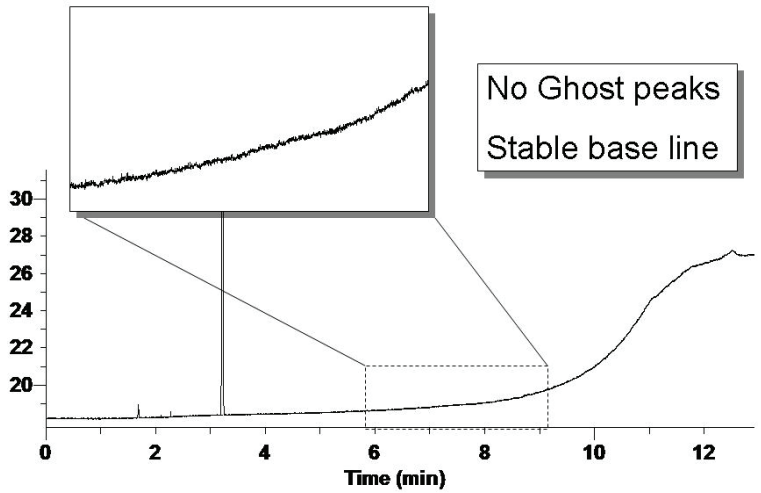

Figure 7: Baseline after cooling with a negative temperature programme of -10 °C/min. from 260 °C → 60 °C. Same column and conditions as in Figure 6. Note the improved stability of the baseline.

of the same sample. If this is not the case, cross contamination can happen very easily and components from another sample can show up. Syringes need to be rinsed with suitable solvents that will remove the previous analytes as well as the solvent. Be careful running a trace analysis and a purity analysis in one system. In purity analysis the matrix can be non-polar or polar and memory effects can be very big. Note that very often the outside of the needle is also transferring components into the "wash" vials. Such components can easily show up as "ghost" peaks in a different analysis. More syringe information can be found in reference 3.

Stationary Phase Degradation and Focusing

The stationary phase inside the column will also degrade when heated at higher temperatures. The degradation products are usually small cyclic siloxanes that are relatively volatile and sent to the detector. Such products will produce an increase of the baseline. Intensity of the background depends on the type of polymer used as stationary

phase. Such bleed products can also produce concrete peaks and cause a disturbance of the baseline. To form concrete peaks a focusing mechanism must be in place. This focusing mechanism is created at the end of a programme cycle when the oven is cooled down. At higher temperatures, bleed products are formed throughout the entire column length. As the column is wound on a cage, these bleed products will move in a circle from the top to the bottom of the oven (see schematics in Figure 5).

The moment the oven cools down, there will be a temperature-gradient in the oven. It is the intention of manufacturers to have the cool down as fast as possible. This results in a non-homogeneous distribution of the temperature, during this cooling-step. The bleed products will also follow the "chromatographic separation rules" and that side of the oven that has a lower temperature, will provide a higher retention for the bleed products. This results in a "focusing-effect" for the bleed products on one-side of the column (see Figure 5). The temperature T1 and T2 will always be a few degrees different. Depending on this difference the focusing will be more or less. As a 30 m column contains typically > 60 coils, there are > 60 focusing bands formed when the column is reaching the lower temperature. When the column is subjected to the next temperature programme (without injection), all these focused bands will start to move and elute resulting in a baseline with a lot of disturbances/ ghost peaks. Figure 6 shows a typical example of such a baseline. The problem is typical for cyanopropyl-phenyl phases like the 1701, 1301 and 624-type and is directly related to the bleed of such phases.

These phenomena can be eliminated by controlling the cool-down temperatures, using a negative temperature programme. When using this, the oven temperature will be controlled and the bleed products will have time to be "smeared" over the capillary tubing. As a result, there is no focusing and for the next analysis the baseline will be stable. Figure 7 shows the result if the same column used in Figure 6 was cooled down with a gradient of 10 °C/min.

Practically it's important to use the controlled cooling for the first minutes cooling. As soon as the signal stabilizes, a faster cooling can be applied.

This effect strongly depends on the GC type and design and the bleed of the column used. Some GCs can give very big ghost peaks based on this mechanism and others may not show any ghost peak at all. It all depends how homogeneous the GC cools down. The controlled cool down is an easy way to "fix" this. It does add extra analysis time.

New column technology with lower bleed phases, will reduce this effect further.

References

1. Jack Cochran, http://blog.restek.com/?p=3604
2. Julie Kowalski, http://blog.restek.com/?p=7114
3. Alan Sensue, http://blog.restek.com/?p=6555
4. How dirty are you? Julie Kowalski, Michelle Misselwitz and Jack Cochran, In house presentation.

Jaap de Zeeuw studied six years of chemistry and graduated in 1979. Jaap has 33 years' experience in GC capillary technology and has developed many PLOT columns as well as bonded-phase columns. He is also the originator of simple concepts for fast GC–MS using a high vacuum inside the capillary column. He has published more than 100 publications in the field of GC on column technology and application. He worked for 27 years for Chrompack/Varian and for the last six years has served as an international specialist on gas chromatography for Restek in The Netherlands.