Gas Chromatography Problem Solving and Troubleshooting

Question:

The amount of baseline noise in my chromatogram suddenly increased. The GC parameters were not changed, and the sample is the same as before. What caused the sudden increase in the baseline noise?

Answer:

Whenever a GC problem occurs, the first step should be to check all of the obvious and simple items. A surprisingly large number of problems or errors originate in these areas. Do not assume that nothing has changed, even if the GC was functioning only days or hours before. Sometimes, GC parameters or samples change without anyone's knowledge. Changes are not always obvious and can be very difficult to find if assumptions and unverified conclusions are made. When one assumes nothing has changed with the sample, investigating the wrong areas of GC or peripheral devices or generating faulty conclusions may lead to many wasted hours. Carefully check the GC parameters to make sure values such as temperature, pressure, and flow rate are correct. Check other method parameters such as the split ratio, purge activation time, detector range, data system settings, and the autosampler function for potential changes or malfunctions. Finally, check non-GC areas such as gas cylinders, impurity traps, syringes, and the sample. Virtually every area of a GC is suspect, especially when a problem affects the entire chromatogram, such as increases in baseline noise or changes in all peak sizes (as opposed to a problem with a single peak).

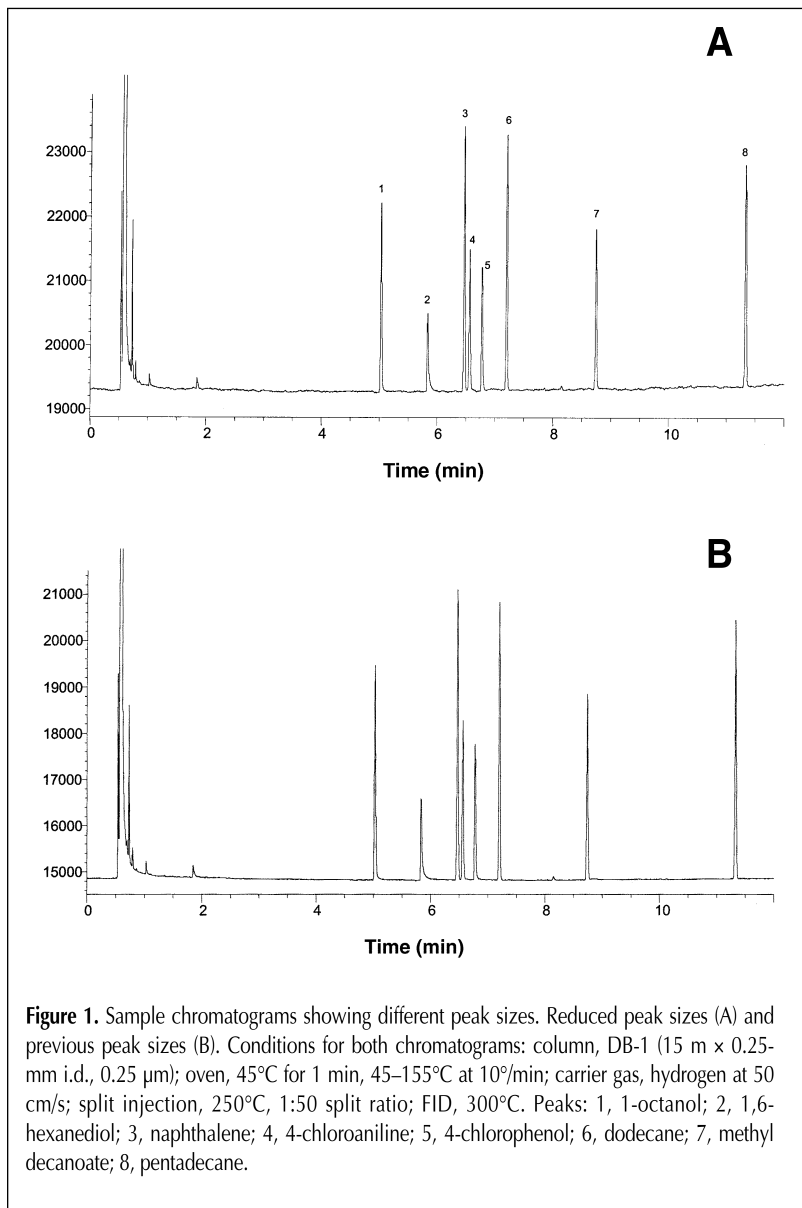

Figure 1. Sample chromatograms showing different peak sizes. Reduced peak sizes (A) and previous peak sizes (B). Conditions for both chromatograms: column, DB-1 (15 m × 0.25-mm i.d., 0.25 µm); oven, 45°C for 1 min, 45–155°C at 10°/min; carrier gas, hydrogen at 50 cm/s; split injection, 250°C, 1:50 split ratio; FID, 300°C. Peaks: 1, 1-octanol; 2, 1,6-hexanediol; 3, naphthalene; 4, 4-chloroaniline; 5, 4-chlorophenol; 6, dodecane; 7, methyl decanoate; 8, pentadecane.

Sometimes, the problem that presents itself can be misleading. In this case, an increase in baseline noise is an example of this type of situation. Careful examination and comparison of the problem chromatogram to a previously acceptable one reveals that the actual problem is more of a decrease in peak height and less of an increase in baseline noise (Figures 1A and 1B). The data system scaled the chromatogram height relative to the tallest peak (note the difference in the scale of the *y*-axis in Figures 1A and 1B). Because the peaks are smaller, the baseline is magnified, thus the apparent increase in baseline noise. Comparing peak heights or areas shows that the peaks have decreased in size. Simply visually comparing the two chromatograms easily leads to the erroneous conclusion that only the baseline noise increased. The actual problem is a significant decrease in peak size and only a small increase in baseline noise. While there is a small increase in baseline noise, all of the peaks are approximately 35% smaller in the problem chromatogram (Figure 1A) compared with the acceptable chromatogram (Figure 1B). Also, all of the peak sizes decreased by the same amount and not by variable amounts. By identifying the actual problem, finding the source becomes easier.

Because all of the peaks decreased in size by the same amount, the sample is suspect. Carefully verify that the sample has not changed. If it is freshly prepared, check for any possible preparation or

calculation errors. If the sample is older, check for any possible handling or storage problems. It is very unlikely that the sample has degraded (it is unusual for all of the compound to degrade by the same amount) or suffered from solvent evaporation (the peaks would be larger and not smaller). Preparation of a new sample is an easy method for checking all of the possibilities (unless the same preparation error is repeated). The syringe is another suspect area. If the syringe is leaking around a removable needle or past the plunger, a smaller volume than desired is injected. Using a different syringe (preferably a new one) is a quick check of the old syringe. Check the injection or syringe technique to make sure that the same sample volume is being injected. Different model syringes may have different volume markings or needle volumes. Finally, if an autosampler is being used, check its settings and proper operation.

The injector's function is to transfer the injected sample into the column, thus it is also suspect in this case. Verify that the injector temperature and split ratio are correct. Lower injector temperatures and higher split ratios usually introduce less sample into the column. Usually, the peaks are reduced in size by differing amounts, but this is not an absolute. Finally, check the injector for leaks.

A change in the flow or velocity of the carrier gas can be eliminated as the problem source. Except for extreme changes in the carrier gas velocity or flow, peak sizes for capillary columns should not be significantly affected. Because the retention times in both chromatograms are essentially the same, the carrier gas velocity or flow has not changed. Contaminants in the carrier gas may alter detector sensitivity. If a gas cylinder or supply has been recently changed, the new source of gas is suspect. Changing to a different gas supply (e.g., a change to a different cylinder) can often verify or eliminate the carrier gas as a source of the decreased detector response (or increased baseline noise). If other GCs are connected to the same gas supply, similar problems should occur with the other GCs (provided they have the same detectors). If the problem occurs with multiple GCs being fed by the same gas supply, the gas supply is a very likely problem source. Expired gas-impurity gas traps can also contaminate the gas, so check any traps for expiration.

If a column becomes active and adsorptive, active compounds (i.e., usually hydroxyl or amine-containing compounds) may be adsorbed, thus reducing peak size. Nonactive active compounds such as hydrocarbons are not susceptible to adsorption problems, and their peak sizes are unaffected by an active column. The sample being analyzed contains active and nonactive compounds. Because all of the peaks decreased by the same amount, column activity can be safely ruled out. If the column was active, only the active compounds should have been affected. (peaks 1, 2, 4, and 5 in Figure 1A).

Check detector temperatures and settings (e.g., attenuation and range) for any changes. Small changes in either area can grossly affect peak size, depending on the type of detector. Especially, check the detector gas flows for changes; the flows may have drifted over time or accidentally changed. Any variation in detector gas flows may also significantly influence peak sizes. Do not use the reading of a pressure gauge as a definitive measure of gas flows; this may be misleading. The detector gas flows for some GCs are set by adjusting a flow or pressure regulator until a specified pressure reading is obtained. It is assumed that the proper detector gas flows are obtained at this pressure.

The pressure gauges for the GC in this case were reporting the correct pressures, thus it was assumed that the detector gas flow rates were correct. After eliminating the previously mentioned possible causes and with the lack of other possibilities, a more detailed investigation was performed. One of the procedures was to measure the detector gas flow rates using a flowmeter instead of relying on the pressure gauges. It was discovered that at the recommended and previously used pressure settings, the air flow to the detector was approximately 40% lower than recommended. Upon increasing the pressure to obtain the proper air flow rate (400 mL/min compared with 250 mL/min), the peak sizes and baseline noise returned to the previously obtained values (Figure 1B). The pressure gauge then reported a pressure approximately 50% higher than the one previously used to obtain the same air flow rate. A partial blockage of the gas line in the GC or a faulty gauge or regulator is probably responsible for the reduced air flow rate at the recommended pressure setting.

By making an assumption and faithfully relying on a gauge reading, your initial assessment of the situation was incorrect. It was assumed that the detector air flow rates were correct because the pressure reading was correct. By taking the 15–20 min necessary to check all of the easy and obvious areas, your assumption error would have been quickly revealed and the problem quickly found.

The purpose of *Chromatography Problem Solving and Troubleshooting* is to have selected experts answer chromatographic questions in any of the various separation fields (GC, GC–MS, HPLC, TLC, SFC, HPTLC, open column, etc.). If you have questions or problems that you would like answered, please forward these to the *Journal* editorial office with all pertinent details: instrument operating conditions, temperatures, pressures, columns, support materials, liquid phases, carrier gas, mobile phases, detectors, example chromatograms, etc. In addition, if you would like to share your expertise or experience in the form of a particular question accompanied by the answer, please forward to JCS Associate Editor, *Chromatography Problem Solving and Troubleshooting,* P.O. Box 48312, Niles, IL 60714. All questions/answers are reviewed to ensure completeness. The *Journal* reserves the right not to publish submitted questions/answers.

Dean Rood
Associate Editor