Gas Chromatography Problem Solving and Troubleshooting

Question:

Injecting a column test mixture is often recommended to diagnose a capillary column problem. What is the best type of test mixture and how should the results be interpreted?

Answer:

There are several different types of column test mixtures, but no single test mixture is best for all columns and GC systems. Column test mixtures are widely used for capillary columns, but they are rarely seen for packed columns due to the large number of packed column stationary phases. Also, most packed columns are packed in the laboratory; thus, they are not tested by the manufacturer. Nearly every capillary column is individually tested by the manufacturer, and the test chromatogram is included with the column. For this reason, the easiest column test mixture to use is the one used by the column manufacturer. In most cases, the test mixture compounds and test temperatures are unique to that specific column. Although many of the compounds in the various test mixtures are different, the column performance characteristics measured are the same. The Grob test mixture (1,2) is used by some column manufacturers. The Grob mixture is a standardized set of compounds analyzed using the same temperature program for all columns except for PLOT columns. The advantage of the Grob mixture is its universal nature; however, some of the compounds co-elute or the run times are long for some columns.

Hydrocarbons are used to measure column efficiency (number of theoretical plates, *n*) (3) and retention (partition ratio, *k*) (3). The lack of reactive functional groups makes hydrocarbons well suited for this purpose. The hydrocarbon peaks should be sharp and symmetrical without any tailing or excessive width. Alcohols are used to measure the amount of column activity. Hydroxy or amine containing compounds can interact with the inner surface of the fused-silica or metal tubing used for capillary columns. This undesirable interaction is often apparent as peak tailing. Straight chain alcohols are often used in test mixtures because they are fairly sensitive to column activity. Some mixtures contain a diol, which is a more sensitive indicator of column activity than a straight chain alcohol. Organic acids and bases are often included in test mixtures to measure column activity and pH characteristics. Substituted phenols and anilines and alkyl amines and carboxylic acids are the most commonly used compounds. The phenols and anilines are usually not as sensitive to column activity as the alcohols. The amines and carboxylic acids are very sensitive to column activity; their peaks often exhibit slight tailing even with the best columns. The heights of the acid and base peaks are often used to measure the pH of the column. The heights of these peaks are compared with the height of a hydrocarbon peak. Adsorption of the acid or base by the column is indicated by the loss of peak height relative to the hydrocarbon peak. In general, the shape of the base peak is poor for excessively acidic columns, and the shape of the acid peak is poor for excessively basic columns. Many test mixtures have a few additional compounds. Fatty acid methyl esters (FAMEs) are probably the most common. They are often used to measure retention characteristics of the stationary phase. In the Grob test mixture, they are used to determine a type of resolution measurement called trennzhal (TZ) (3).

If the column is suspected as a source of a peak shape or size problem, the injection of an appropriate test mixture often helps to diagnose possible problem sources. Split injections at a high split ratio are used to minimize injector contributions to the chromatogram. Figure 1 is a test chromatogram of the column manufacturer's test mixture. Slight tailing is evident for the alcohol peaks (peak 2 and 6). This is usually indicative of column activity or a contamination problem. Because the tailing is minor, the amount of contamination or activity is also minor. The tubing's surface or the contaminants interact with the alcohols to create the tailing peaks. It is not possible to determine from the chromatogram which problem is causing the peak tailing. If the column is highly contaminated or contains particulate matter (such as ferrule or septa particles), most or all of the peaks may exhibit tailing. Also, a poorly installed column may result in this problem. If column contamination is suspected, solvent rinsing the column (4) may eliminate the alcohol peak tailing. If proper solvent rinsing is not effective, the column is irreversibly contaminated or is suffering from activity. One of the later

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.

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eluting hydrocarbons is normally used to determine the number of theoretical plates and retention. A decrease in the number of plates is indicative of degradation of the column due to damage or age or due to a change in the average linear velocity or flow rate of the carrier gas. A change in the carrier gas also manifests itself as a change in retention time. Although a column manufacturer's test mixture does not conclusively diagnose every problem, it is useful in investigating activity, contamination, and retention problems.

Another type of test mixture is one designed specially for a particular analysis or GC system. Column manufacturer's test mixtures are usually intended for split injections using flame ionization, thermal conductivity, or mass spectrometric detection. Sometimes these test mixtures are not compatible with a particular GC system because of concentration, injection, or detection reasons. Your own test mixture may be a better measure of system performance. It should contain the compounds that are most difficult to resolve and obtain good peak shapes. This test mixture is analyzed using the same conditions as used for actual samples. Specific performance characteristics such as peak resolution or shape can be set for the test mixture. The test mixture may not indicate that there is a problem that may negatively affect the actual analysis. Conversely, the actual samples may not be as demanding of the GC system as the column test mixtures. Although a manufacturer's test mixture may indicate a slight problem or less than best performance, the failing may not affect the actual analyses. Your own test mixture can be in the concentration range of actual samples. This is beneficial because the performance of many GC systems can depend on the sample concentration levels. In general, trace or low level analyses are more demanding on a GC system. A column test mixture may require changes to the GC conditions that are not necessary for your own test mixture. When using a selective detector such as nitrogen–phosphorus or electron conductivity, the column test mixture is not suitable because many of the test compounds cannot be detected by the selective detector. In these cases, a specific test mixture is required and is rarely available from the GC or column manufacturers. With your own test mixture, column efficiency, retention, response, and peak shapes can all be easily compared.

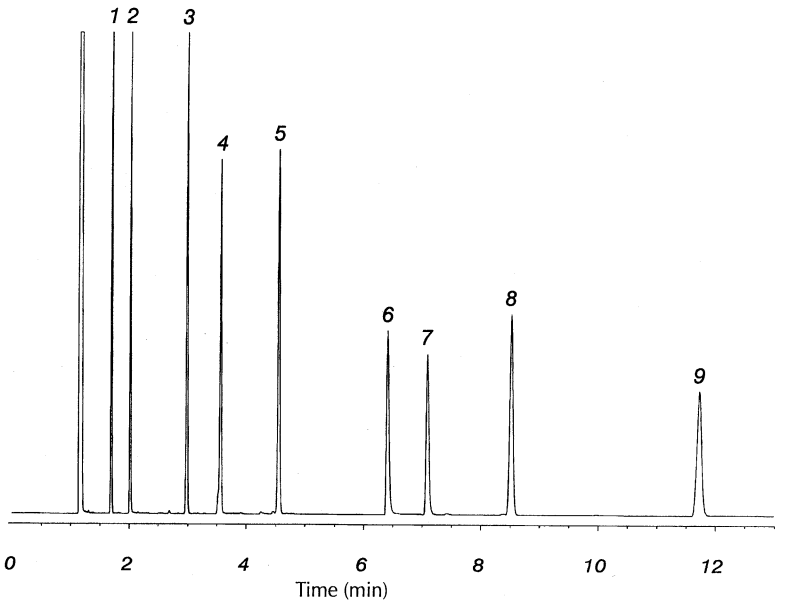

Figure 1. Chromatogram of a column test mixture. Conditions: column, DB-17, 30 m × 0.25-mm i.d., 0.15-µm film; oven, 110°C isothermal; injector, split (1:75) (250°C); detector, FID (300°C); carrier gas, hydrogen (42 cm/s). Peaks: 1, undecane; 2, 1-octanol; 3, tridecane; 4, 2,4-dimethylphenol; 5, 2,6-dimethylaniline; 6, 1, undecanol; 7, ethyl decanoate; 8, 1-methylnaphthalene; 9, hexadecane.

A practice that helps to evaluate test chromatograms is to copy a good or reference test mixture chromatogram onto an overhead transparency. If the chromatogram is copied in the actual size, it can be overlaid on a newly generated test chromatogram. This makes it easy to compare the two chromatograms especially for inexperienced chromatographers. The overhead transparency can be marked with acceptable peak size, width, and retention values. One technique is to draw boxes around the specific peaks. The peaks in the new chromatogram should not be outside of the boxes (i.e., indicating incorrect retention or excessive height or width) or shorter than a specified height. An overhead transparency chromatogram can be created for each GC system or analysis. If the test sample is analyzed before samples are run or after system maintenance or repair, this practice is very useful in detecting many GC problems before they significantly impact the laboratory.

References

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