

AirSharp™

Operating Instructions

Using the AirSharp™ effectively for your analyses

Congratulations on your purchase of the AirSharp™ compressed air peak-focussing ancillary kit for GC. Below are a few simple procedures and hints on how to effectively use the AirSharp™ and how to develop a method for your particular analysis.

Method Development

As an example of how to use the AirSharp™ effectively, below is a series of chromatograms to explain the process of method development and how and when to apply the AirSharp™ to improve peak shape and signal-to-noise ratio.

Using a horse racing standard drug mixture (Figure 1) on a BPX5 column the separation of the nine

components is quite good. The concentration of this analysis is 1ppm and each compound is easily detected. This however is very useful to set up the method development for lower level analyses as will be seen later.

The four compounds that will be subjected to AirSharp™ are compounds 6, 7, 8 and 9. These are late-eluting compounds and the temperature at the point of elution of Dilantin (6) is 270 °C extending to 320 °C for Diphenoxylate (9). This gives sufficient temperature difference for the AirSharp™ to be effective (Another aspect to consider with regards to these types of late eluting compounds is that they are usually high boiling compounds that are subjected to inlet mass discrimination and so the compound response would have been reduced in any case).

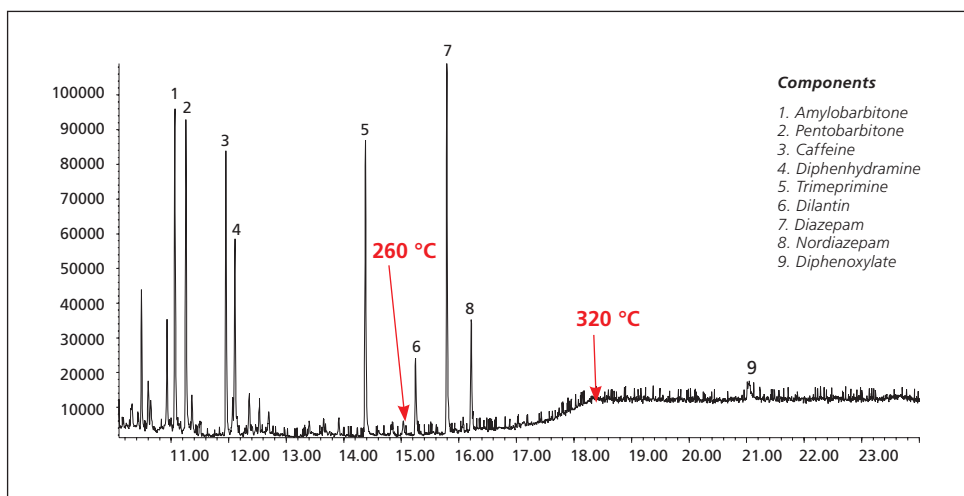


Figure 1. Chromatogram of the horse racing mixture at 1 ppm with no AirSharp™ applied.

The aim of this method development is to show how we arrive at Figure 2. The AirSharp™ can be used to increase the sensitivity of the late eluting high boiling components of the analysis as shown in Figure 2.

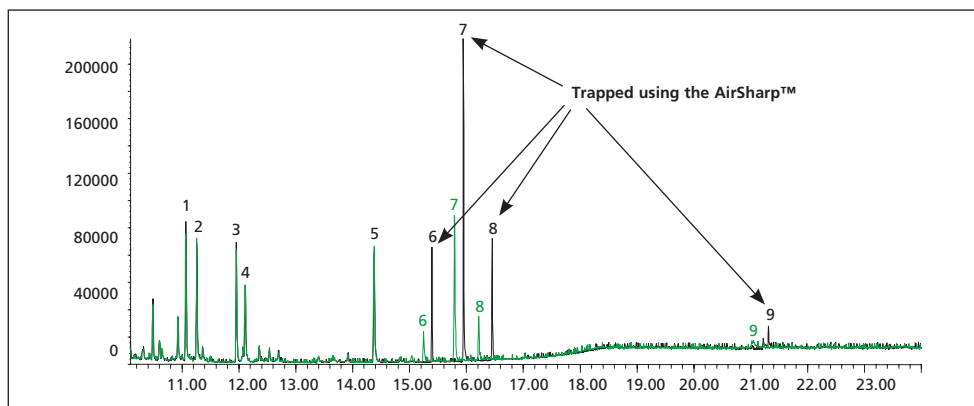


Figure 2. Overlaid chromatograms of the standard chromatogram of the horse racing mixture (in green) and the AirSharp™ focused chromatogram (in black) at 1 ppm. Note the improved signal to noise ratio of the peaks subjected to AirSharp™ focusing.

Step 1.

The first step when developing a method with the AirSharp™, after identifying which compounds you wish to sharpen and enhance, is to determine when the front of the first peak you are interested in begins eluting from the column. In our example this is Dilantin (6) (Figure 1). The AirSharp™ is controlled through external events of the GC software (Figure 3), this can be set by the user to turn the Air Sharp

on and off. Figure 3 is an example of the Agilent software that controls the external events for instruments such as the AirSharp™. As can be seen from this picture, the computer software allows the user to designate the times when the AirSharp™ is turned on and off.

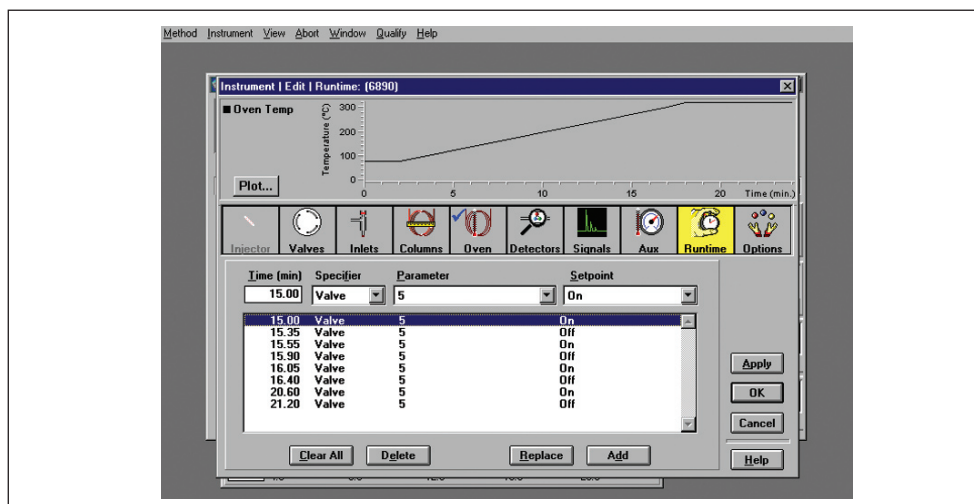


Figure 3. Screen capture of the external events software (Agilent ChemStation software) that controls the on/off functioning of the AirSharp.

In this example the front edge of Dilantin begins to elute at approximately 15.20 -15.25 minutes. The AirSharp™ is turned on at 15.00 minutes (that is 12-15 seconds prior to the normal elution time

of the compound) and should be turned off just after the compound would normally have eluted at 15.35 minutes (Figure 4).

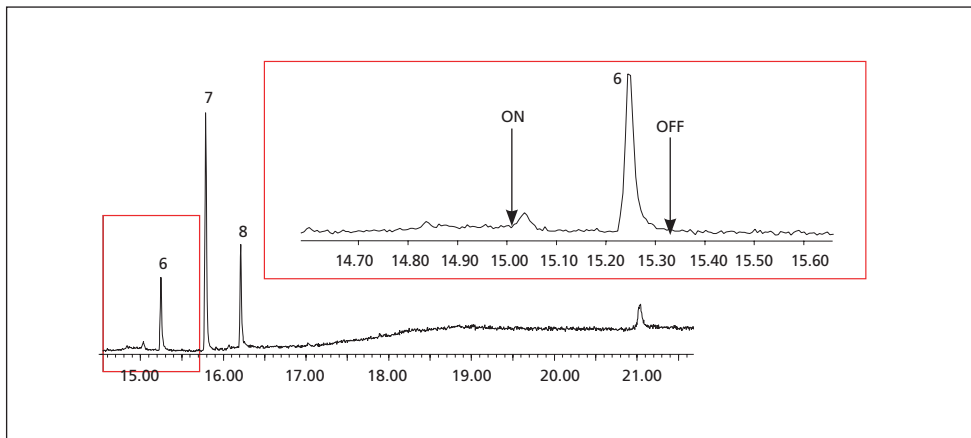


Figure 4. Enlarged section of the horse racing mixture chromatogram. The AirSharp™ should be applied at least 12-15 seconds prior to the peak first eluting from the column. In this case, this corresponds to the AirSharp™ being applied at 15.00 minutes and the compound being released from the cold trap at 15.30-15.35 minutes.

The result of this application of the AirSharp™ to Dilantin (6) at 1 ppm is shown in Figure 5. The signal-to-noise ratio has dramatically improved to the point that the peak height is almost that of Diazepam (7). The peak shape is much sharper.

Also note that the retention time of the peak has moved from 15.25 to 15.40 minutes. This indicates that once the AirSharp™ is turned off at 15.35 minutes the Dilantin is easily released from the cooled zone and carried onto the detector.

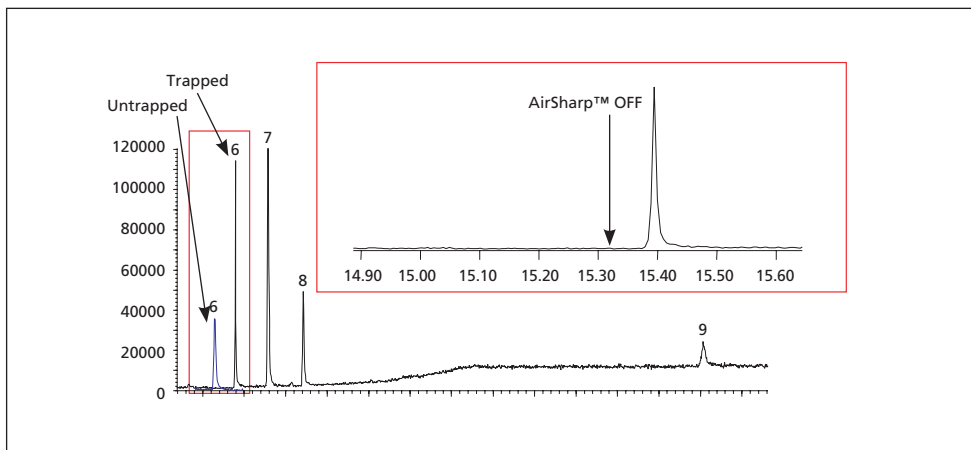


Figure 5. Result of AirSharp™ being turned off. Note the improved signal to noise ratio of Dilantin (6) from the previous chromatogram (Figure 1). This improvement along with improved peak shape is clearly shown by the overlaid peak of Dilantin (6) in blue. Note also that the AirSharp™ is turned off at approximately 15.30-15.35 minutes and the Dilantin peak appears now at about 15.40 minutes.

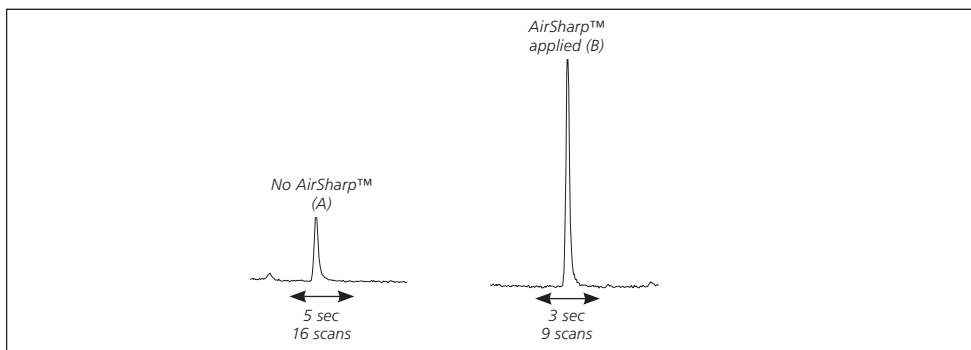


Figure 6. By using the AirSharp™ the signal to noise ratio is improved and the peak shape is narrowed. Peak A has no AirSharp™ applied and takes 5 seconds to elute. Peak B is narrower and takes 3 seconds to elute. At 3.14 scans/sec this gives approximately 16 and 9 scans respectively. If the scan rate is too low this can be a problem with regards to poor peak shape and quantitation

Figure 6 shows a comparison of the peak shape and width of the Dilantin (6) peak from Figure 5. As can be seen the peak A with no AirSharp™ applied takes 5 seconds to elute from the column. Peak B after AirSharp™ is applied takes only 3 seconds. The scan rate for these analyses is 3.14 scans/sec giving approximately 16 and 9 scans for each peak. In this instance there is probably enough scans to give a reasonably good peak shape and the peak shape does not look triangular but this could be a problem if the peak were narrower. This could result in poor peak shape or very triangular peak shape due to not enough points being scanned over the course of the peak eluting from the column.

Step 2

The next peak to be subjected to AirSharp™ is Diazepam (7). A similar procedure is used to determine when to turn the AirSharp™ on and off. The only difference being that you have to use the chromatogram that you have just run (Figure 5) to determine the retention time and not the original chromatogram (Figure 1). When you apply AirSharp™ to any chromatogram you change the

retention times of most components to a small degree. In this instance, Diazepam (7), the AirSharp™ would need to be applied at 15.55 minutes, approximately 12-15 seconds before Diazepam begins to elute and turned off at 15.85-15.90 minutes just after the Diazepam would normally have finished eluting from the column (Figure 7)

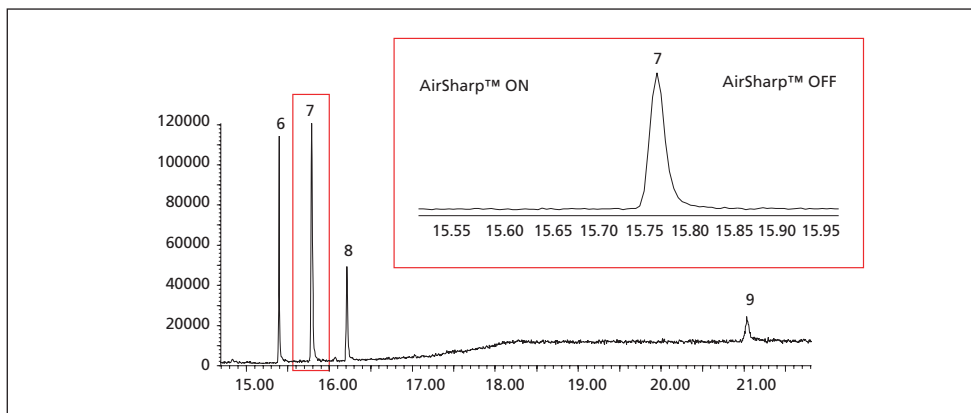


Figure 7. Chromatogram showing when to turn the AirSharp™ instrument on and off to sharpen up the Diazepam (7) peak.

Step 3

The result of this application of the AirSharp™ to both Dilantin (6) and Diazepam (7) is shown in Figure 8. As can be seen from this chromatogram

the size of the Diazepam peak has increased considerably from 120000 to 220000 in abundance.

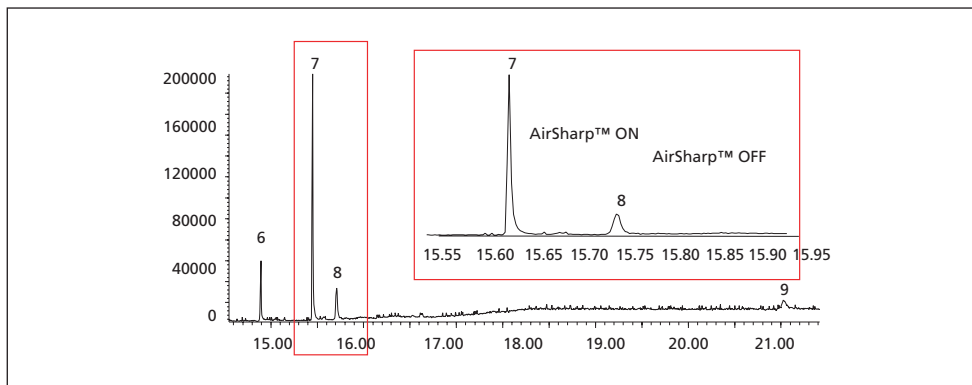


Figure 8. Chromatogram is showing the effects of applying AirSharp™ to Dilantin (6) and Diazepam (7).

Using the chromatogram in Figure 8 you can apply AirSharp™ to the Nordiazepam (8) peak in a similar manner as described in steps 1 and 2. This can be seen in the expanded section of Figure 8. As an end

result of applying the AirSharp™ to all compounds 6, 7, 8 and 9 you will obtain a chromatogram that looks like Figure 9.

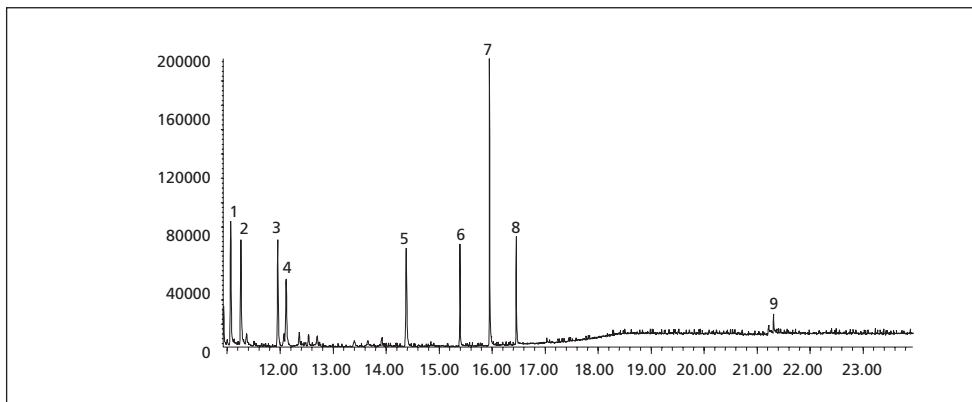


Figure 9. Chromatogram showing the effects of applying the AirSharp™ to peaks 6, 7, 8 and 9.

The true effect of the AirSharp™ on this mixture can be seen when you overlay the original chromatogram and the chromatogram that has been subjected to the AirSharp™ (Figure 2). This shows the original chromatogram (in green) and the AirSharp™ chromatogram (in black) at a level of 1 ppm. As can be seen from the early eluting peaks,

the levels injected are essentially the same but the real difference can be seen with peaks 6, 7, 8 and 9. These have a much higher signal-to-noise ratio than the corresponding peaks in the non-AirSharp™ chromatogram.

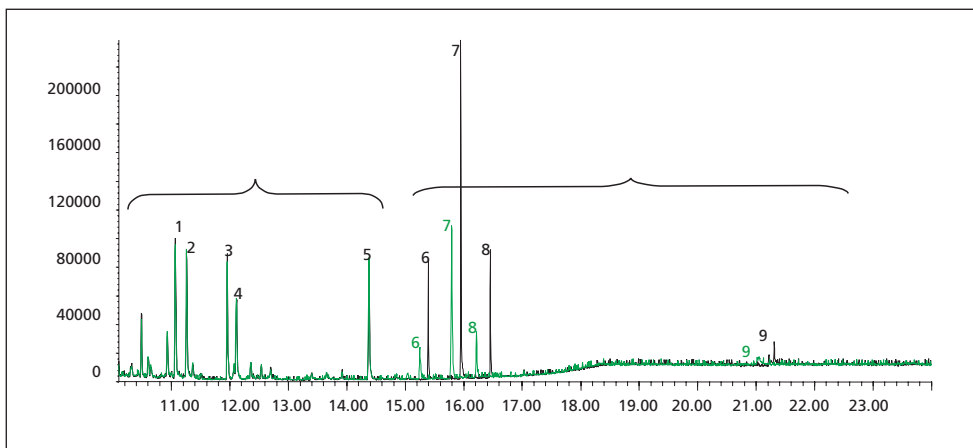


Figure 2. Overlaid chromatograms of the standard chromatogram of the horse racing mixture (in green) and the AirSharp™ focused chromatogram (in black) at 1 ppm. Note the improved signal to noise ratio of the peaks subjected to AirSharp™ focusing.

Figure 10 below is an identical chromatogram to that shown in Figure 2 except the level is 0.5 ppm on column. This shows the value of the AirSharp™ improving the signal-to-noise ratio of the peaks of

interest so they can be easily detected. The early eluting peaks are easily detected even at 0.5 ppm but the later eluting peaks were almost indistinguishable amongst the noise of the baseline.

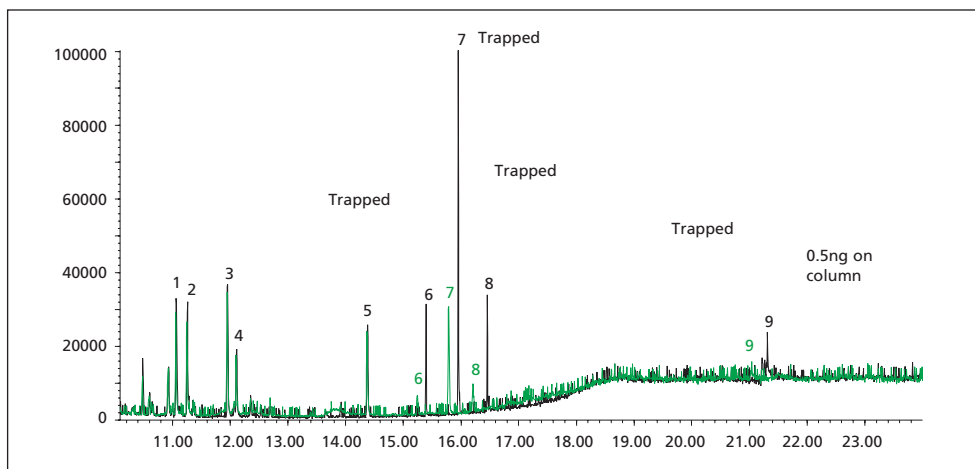


Figure 10. Overlaid chromatograms of the standard chromatogram of the horse racing mixture (in green) and the AirSharp™ focused chromatogram (in black) at 0.5 ppm. Note the improved signal to noise ratio of the peaks subjected to AirSharp™ focusing and the distinguishing the peaks of interest from that of the baseline noise.

The AirSharp™ is a compressed air peak-focussing ancillary kit for GC. Should you experience any difficulties with the installation or in the use of the AirSharp™ don't hesitate to contact SGE staff through our technical support page on our website: www.sge.com

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