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Application Note 157

Advantages of using tube-based thermal desorption for monitoring chemical warfare agents at ultra-trace levels

Summary

Chemical warfare agents (CWAs) and their respective simulants were analysed using Markes' tube-based systems. The study included the use of both 89-mm (3.5-inch) standard tubes and 114-mm (4.5-inch) depot area air monitoring systems (DAAMS) tubes, desorbed using standard- or DAAMS-configured automated thermal desorbers, respectively, connected to gas chromatography–mass spectrometry (GC–MS) or gas chromatography–mass spectrometry–flame photometric detection (GC–MS–FPD) instruments.

Two recent TD innovations were evaluated – enhanced internal standard addition and multi-tube trap stacking – to explore what advantages they offered for monitoring CWAs at ultra-trace levels.

Introduction

CWAs remain among the most toxic chemicals known to humans. Many are volatile or semi-volatile organic compounds (VOCs/SVOCs) compatible with GC analysis, for example the nerve agents VX, sarin (GB), cyclosarin (GF), soman (GD) and Russian VX (R-VX), the blister agent mustard gas and the tear gases chlorobenzylidenemalononitrile (CS) and chloropicrin (PS), which are sometimes deployed to disperse crowds during riots.

Trace-level monitoring of CW agents is required for a range of military and civilian scenarios to keep the public and government personnel safe. Applications include:

- Monitoring agent storage facilities (stockpile sites) and neighbouring locations.
- Monitoring agent destruction facilities to ensure the safety of site personnel.
- Remediation and decontamination.
- Counterterrorism (homeland defense), *e.g.* continuous monitoring of critical government buildings.
- Mobile laboratories that can be deployed in the event of a chemical incident.
- Defence research, *e.g.* testing the efficacy of equipment assigned to protect soldiers in the battlefield and checking the performance of decontamination procedures.

The extreme toxicity of the chemicals involved means that simulants are generally used during defence research to minimise the risk to scientists. The simulants selected have all been shown to behave similarly to the respective agent during GC analysis (Table 1).

Many CWAs and their respective simulants are sticky and/or reactive species that present a significant analytical challenge, especially at sub-ng and pg levels. Markes' TD technology is a global leader in terms of analytical performance and is used extensively for CW applications around the world. Relevant configurations include on-line systems (generating semi-continuous near-real-time measurement) and off-line laboratory-based installations. Both may be deployed in conventional or mobile laboratories. The most widely used sampling options include pumped monitoring onto sorbent tubes and round-the-clock in-line air monitoring and analysis.

Experimental

Standards

The simulants used in the first phase of the study were selected following the recommendations of Bartelt-Hunt *et al.*[1] and are listed in Table 1.

Simulant compound	Respective agent
Dimethyl methylphosphonate (DMMP)	Sarin (GB)
Triethyl phosphate (TEP)	Tabun (GA)
Methyl salicylate	Mustard gas (HD)
2-Chloroethyl phenyl sulfide (2-CEPS)	Mustard gas (HD)
Malathion	VX

Table 1: Simulants and their respective CWAs.

A stock solution was produced using high-purity standards of the compounds at 100 ng/µL in methanol. The stock solution was then diluted in methanol as required to produce lower-level standards as required. 1-µL aliquots of the standard solutions were then introduced onto the sampling end of pre-conditioned sorbent tubes (C2-VBXX-7011) in a flow of carrier gas using the Calibration Solution Loading Rig™ (CSLR™). The solvent was allowed to purge off the tube before the standards were sealed ready for analysis.

Markes International Ltd
T: +44 (0)1443 230935 **F:** +44 (0)1443 231531 **E:** enquiries@markes.com

Experimental conditions

TD:

Instrument:	UNITY–ULTRA-xr™ – standard- and DAAMS-configured (Markes International)
Tubes:	Inert-coated stainless steel, DAAMS 4.5" x 6 mm O.D., Quartz wool/Tenax® TA (C2-VBXX-7011) and standard 3.5-inch x 6.4 mm O.D. tubes (C1-CAXX-5003)
Flow path:	200°C
Dry purge:	1 min at 50 mL/min
Tube desorption:	300°C (8 mins) at 50 mL/min, splitless
Sample flow:	50 mL/min
Internal standard addition:	to tube
Trap purge:	1.0 min at 50 mL/min
Focusing trap:	'Chemical weapons' trap (part no. U-T10CW-2S)
Focusing trap low:	20°C
Focusing trap high:	300°C (3 min)
Trap heat rate:	MAX
Outlet split:	Splitless or 9 mL/min for re-collection tests

GC

Column:	BPX5™ 30 m × 0.25 mm × 0.25 µm
Carrier gas:	Helium
Column flow:	2 mL/min, constant flow
Oven:	40°C, 20°C/min to 300°C (4 min)

MS

Source:	250°C
Transfer line:	310°C
Quadrupole:	200°C
Scan range:	m/z 35–350

Results and discussion

Excellent peak shape was achieved for all compounds, even for DMMP, which frequently exhibits tailing during capillary GC analysis (Figure 1).[2]

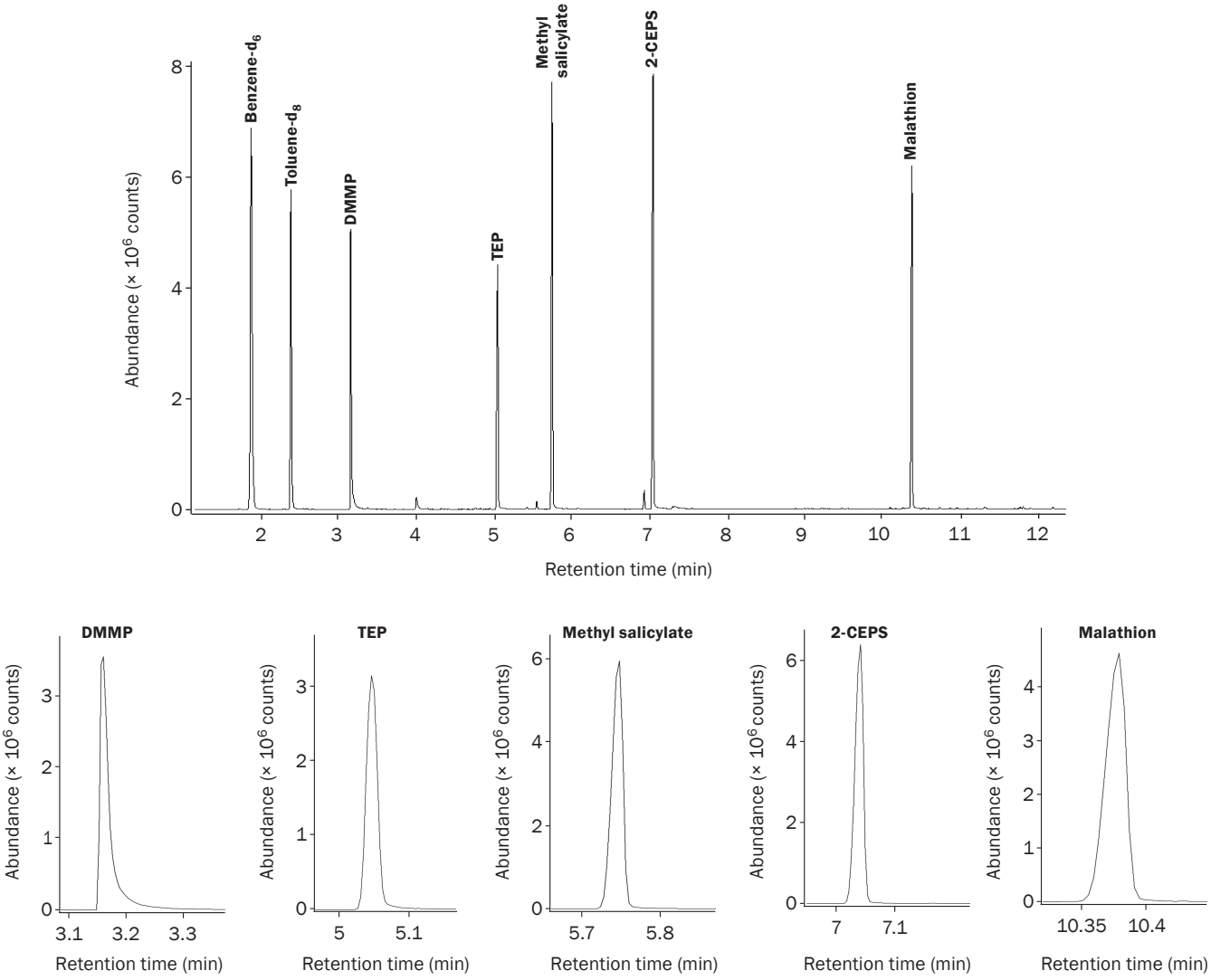

Figure 1: (Top) Merged extracted ion chromatogram (EIC) of the high (100 ng) level standard. (Bottom) A close-up view of the EIC results obtained for each target analyte at 100 ng illustrating peak shape.

Linearities

Calibrations were performed over eight levels between 100 pg and 100 ng using 1-µL injections of the standard solutions. Note that while low ng and pg levels are more representative of the levels collected during air monitoring, 100-ng levels are seen in some important chamber tests.

The linearities for all five compounds were excellent across this extended dynamic range with R^2 values ≥0.9991 (Figure 2).

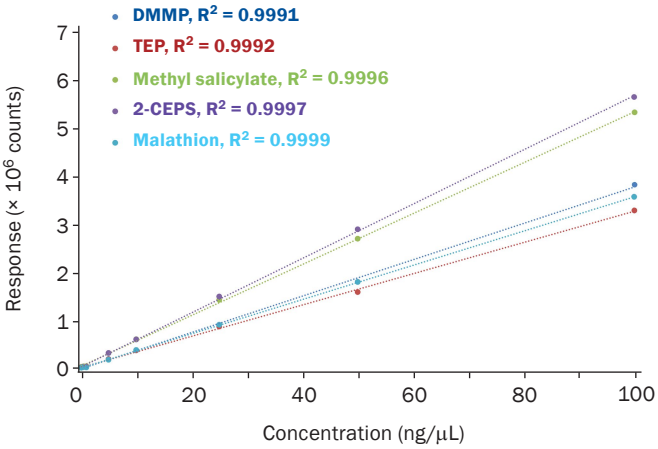

Figure 2: Excellent linearities were obtained for the target compounds for a range of 0.1–100 ng.

Carryover

Carryover, in the tube and system, was assessed by loading tubes with a 100-ng standard and desorbing them twice (Figure 3 and Table 2). Figure 3 shows a maximum carryover of 1.5% even at these extremely high levels.

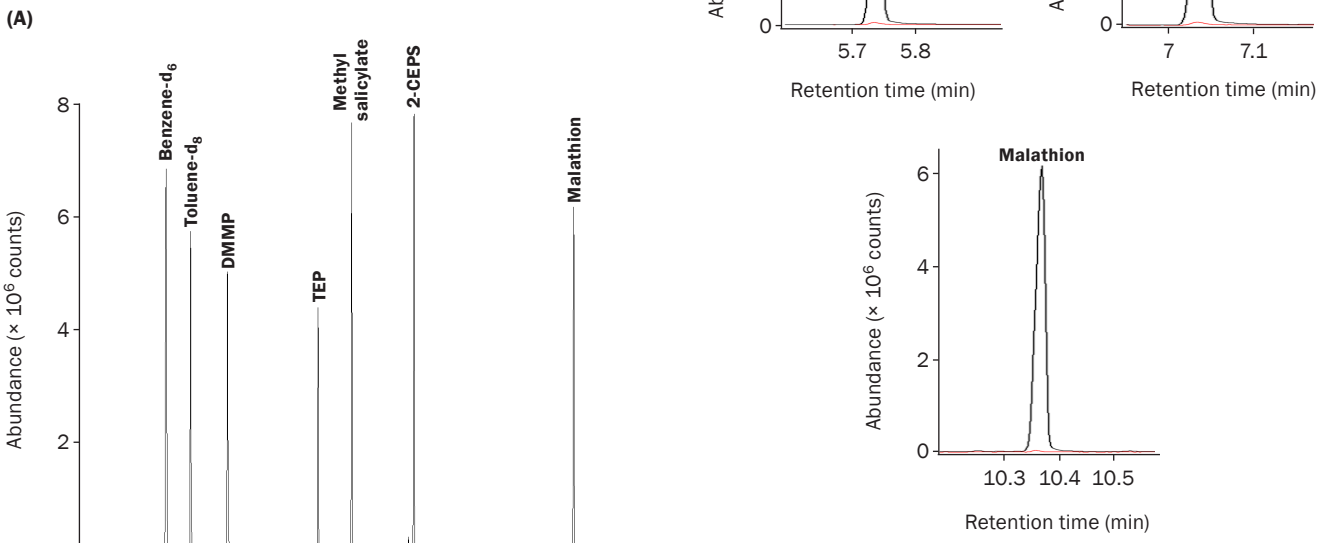

Figure 3: (A) EICs from original desorption of 100-ng/µL standards (black) overlayed with the subsequent re-desorption (red). (B) Close-ups of each overlayed peak.

Compound	Carryover (%)
Benzene-d_6	0.8
Toluene-d_8	0.4
Dimethyl methylphosphonate (DMMP)	0.7
Triethyl phosphate (TEP)	1.5
Methyl salicylate	1.1
2-Chloroethyl phenyl sulfide (2-CEPS)	1.5
Malathion	0.4

Table 2: Carryover data.

Reproducibility

Reproducibility was tested at low and high concentrations to verify method parameters. The relative standard deviation (%) can be seen in Table 3 for 5, 50 and 100 ng/µL. Values for all compounds at all levels are at or below 5%.

RSD (%)	5 ng/µL (n = 10)	50 ng/µL (n = 7)	100 ng/µL (n = 10)
Dimethyl methylphosphonate (DMMP)	3.21	4.30	2.46
Triethyl phosphate (TEP)	2.97	2.81	2.41
Methyl salicylate	2.72	1.73	1.31
2-Chloroethyl phenyl sulfide (2-CEPS)	4.04	1.82	1.76
Malathion	5.02	2.06	1.71

Table 3: The reproducibility of target simulants at three different concentration levels.

Internal standard

Internal standards reduce analytical uncertainty by identifying variability between injections that may otherwise have gone unnoticed, for example variable detector response.

Markes Instrument Control (MIC) software allows a gas-phase internal standard to be automatically introduced to the sampling end of sorbent tubes at two different stages of air monitoring operation to help improve precision:

- Pre-loading sorbent tubes prior to transport and sampling.
- Immediately before tube desorption as part of the automated analytical process.

Deuterated compounds are commonly used as internal standards, and, in this study, the sorbent tubes were pre-loaded with benzene-d_6 before introducing the simulant standards. Note that Markes' automated thermal desorbers offer a specific method template for introducing a gas-phase internal standard onto conditioned tubes before they are used for sampling or loading standards.

During real-world field studies, some sampling discrepancies and anything that compromises tube seals during storage and transport (before and after tube sampling) will impact the level of pre-loaded internal standard. This will then be observed by a deviation in internal standard levels for the affected sample tube. Likewise, if the pre-loaded internal standard value in a tube matches others in the batch while levels of all other analytes on that tube are significantly lower than other samples in the batch, this might indicate pump failure and the collection of smaller than expected air volumes during sampling.

The addition of an internal standard during automated TD serves as a quality control check of the system itself. Toluene-d_8 was used in this study. Results are plotted over time to verify the system's performance, show any outliers and to check detector response and peak shape. Reproducibility in this study was found to be 1.1% for toluene-d_8.

Note that both pre-sampling and analytical internal standards can be used in the same study provided that different internal standard compounds are selected for each.

Figure 4 shows tube replicates of the 100-ng/µL standard containing both internal standards. The peak shown at 1.9 minutes is benzene-d_6 and the peak at 2.4 minutes is toluene-d_8.

In the second data file, the benzene-d_6 peak is missing, and the early-eluting target compounds (between 2.5 and 4 minutes) are noticeably lower than expected; however, the presence of toluene-d_8 at expected levels shows that the system is working correctly.

A close-up comparison of the failed standard (red trace in Figure 5) with a successful standard (black trace in Figure 5) shows this more clearly.

The most likely cause of a result like this is breakthrough (loss of volatiles), probably caused by passing too large a volume of air through the tube during sampling or standard loading. If this had happened during field sampling without a pre-loaded internal standard, erroneously low results could have been reported for the more volatile compounds.

Improving detection limits – reaching trace levels more easily

The very high toxicity of chemical agents means detection is required at extremely low, *e.g.* picogram levels, necessitating the concentration of large volumes of air using pumped sorbent tubes before analysis.

Calculations were performed to determine the limit of quantitation (LOQ, calculated using a signal-to-noise ratio of 10) and limit of detection (LOD, calculated by using a signal-to-noise ratio of 3) from the linearity data (Table 4).

Compound	LOQ (ng)	LOD (ng)
Dimethyl methylphosphonate (DMMP)	0.207	0.062
Triethyl phosphate (TEP)	0.159	0.048
Methyl salicylate	0.066	0.020
2-Chloroethyl phenyl sulfide (2-CEPS)	0.068	0.020
Malathion	0.147	0.044

Table 4: Target compounds and their respective limits of quantitation (LOQ) and limits of detection (LOD).

Markes' TD systems now offer users the option to 'stack' the desorptions of two or more samples on the same focusing trap. This is useful, for example, where air volumes are limited by sampling time or analyte breakthrough and it requires multiple samples to be collected in parallel at the monitoring location.

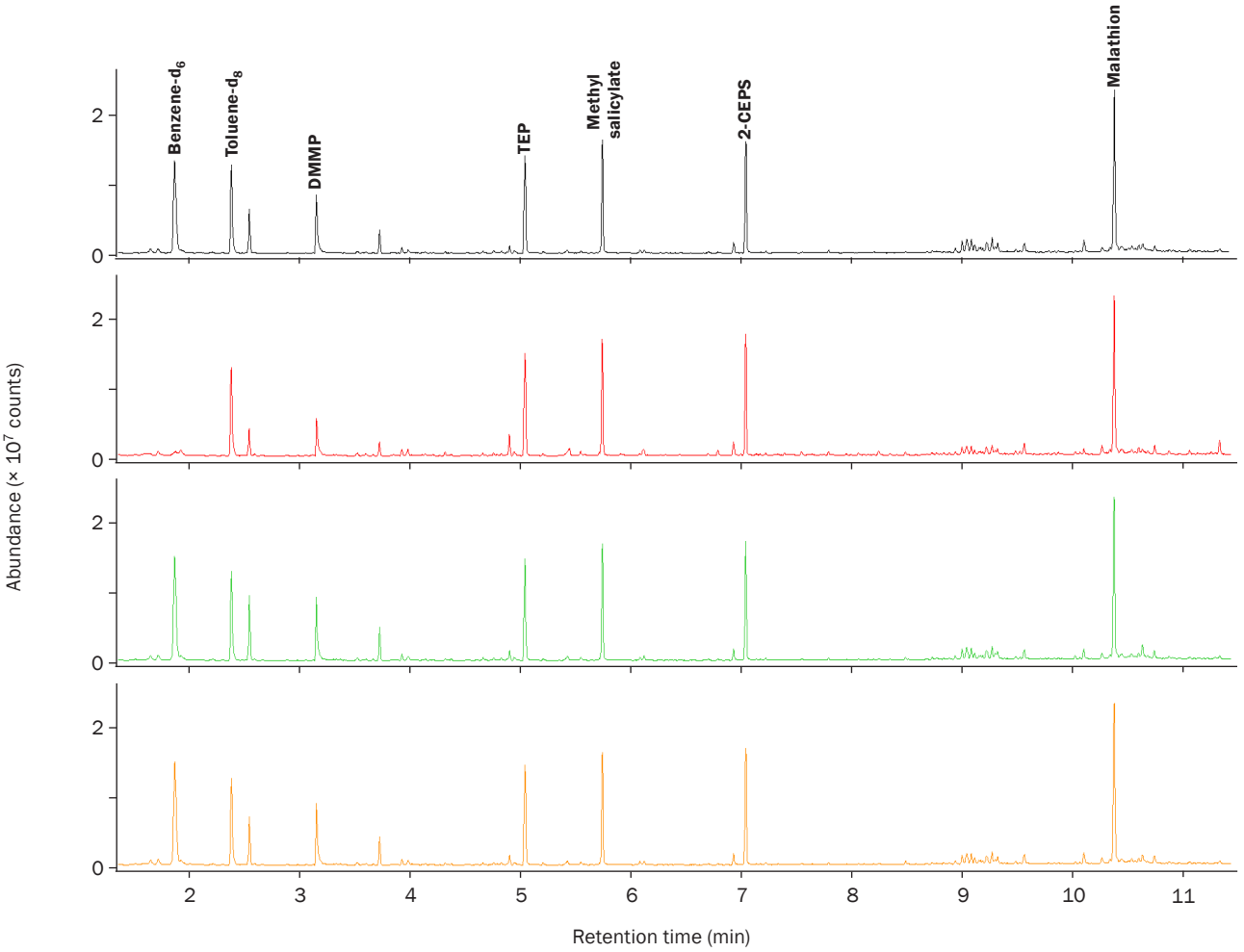

Figure 4: Four replicates of the 100-ng/µL standard prepared using both internal standards: benzene-d_6 (1.9 min) and toluene-d_8 (2.4 min). Benzene-d_6, toluene-d_8, DMMP (3.19), TEP (5.05), methyl salicylate (5.75), 2-CEPS (7.05) and malathion (10.37).

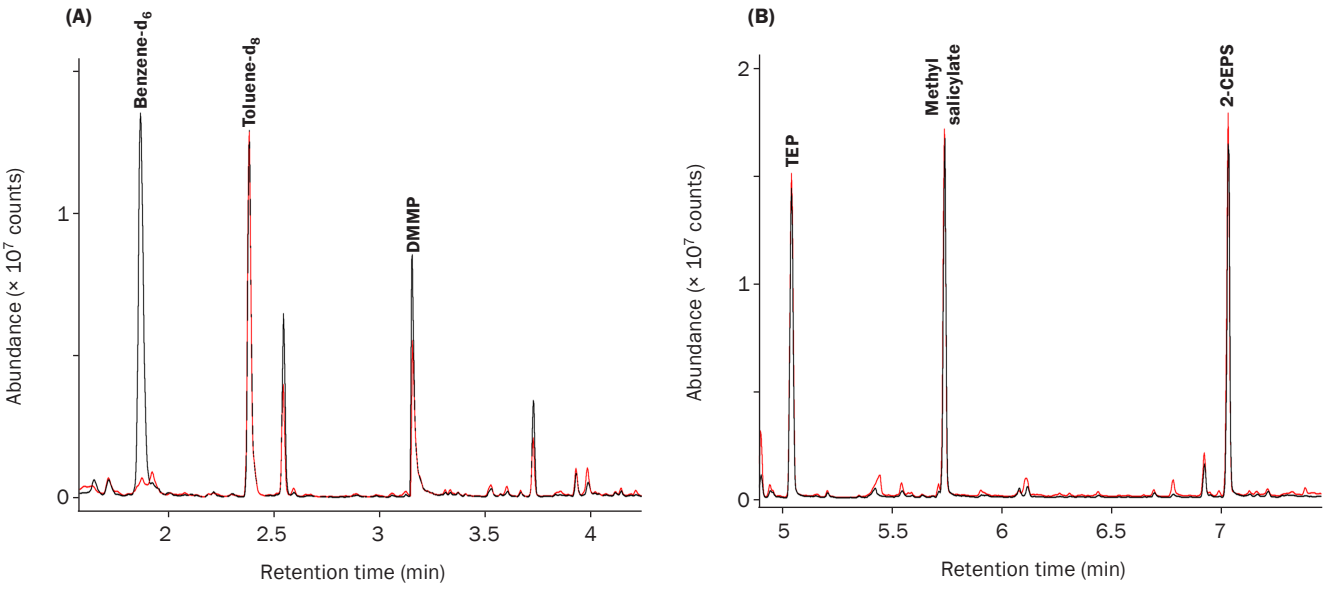

Figure 5: Overlay of a 100-ng standard (black) with analysis of a 100-ng standard that was prepared incorrectly (red). (A) Benzene-d_6, toluene-d_8 and DMMP (3.19). (B) TEP (5.05), methyl salicylate (5.75) and 2-CEPS (7.05).

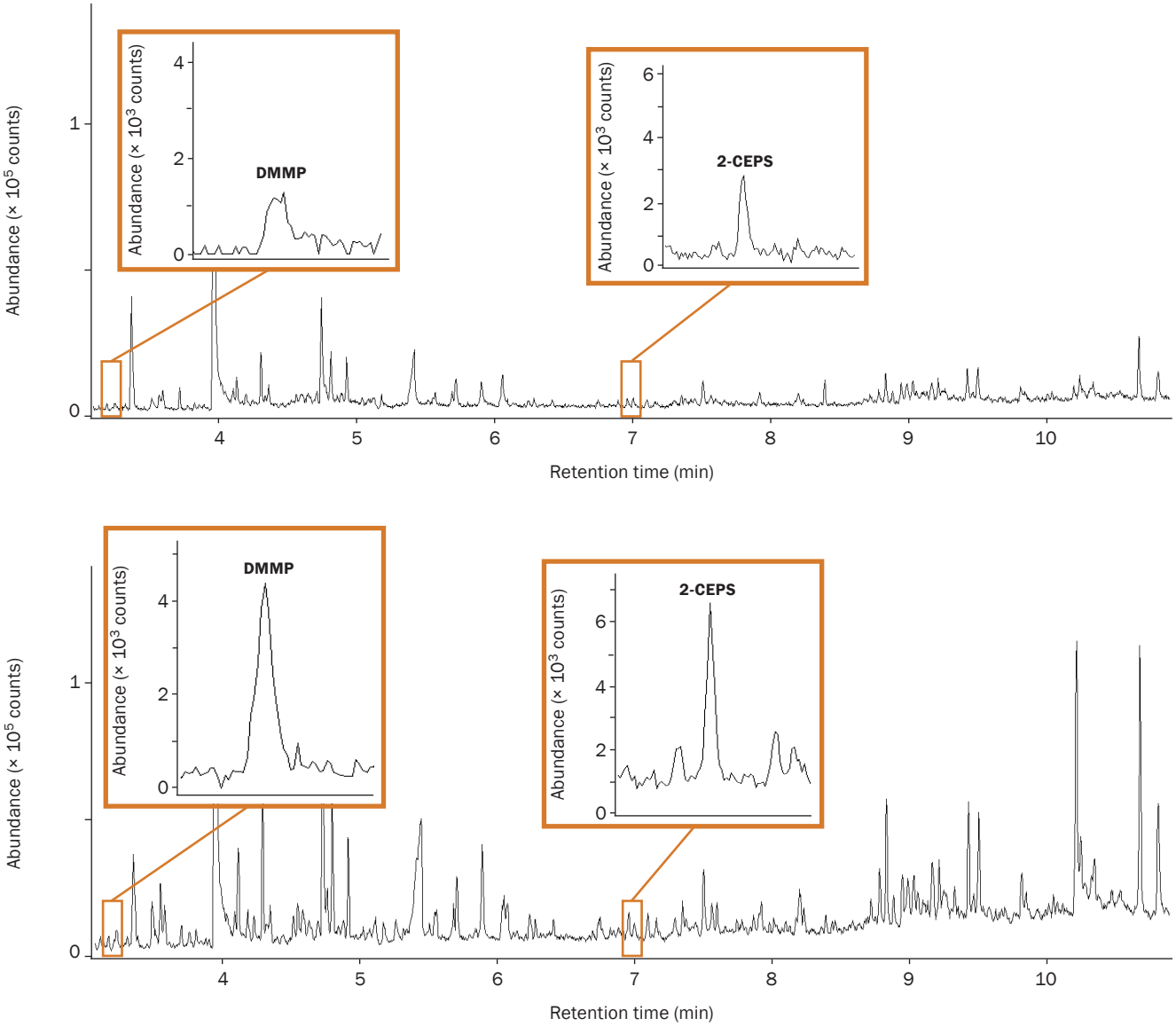

Figure 6: Comparison of results from the 5-pg standard (EIC data) without stacking (top) and with the stacking of three replicate samples (bottom).

To test the efficacy of trap stacking, a trace-level standard was produced (0.005 ng/µL) and four clean and conditioned sorbent tubes were each spiked with 1 µL.

One of the tubes was analysed using the regular analytical method and the remaining three tubes were analysed by stacking the desorbed vapour from all three tubes onto the focusing trap before trap desorption under normal conditions. The results are shown in Figure 6.

2-CEPS is clearly detected in both chromatograms at a good resolution and peak shape at 7 minutes. However, DMMP exhibits a poor response and is hard to see in the single-tube analysis. A much clearer peak is generated when three tubes are desorbed into the focusing trap before analysis, allowing all compounds, including DMMP, to be detected.

Anything that robustly enhances sensitivity is invaluable for CWA monitoring applications, particularly around agent destruction ('de-mil') facilities where the safety of site personnel is at stake.

Using re-collection to validate analyte recovery through the TD flow path under method conditions

While most of this work was carried out splitless to maximise sensitivity, all Markes' thermal desorbers allow the quantitative re-collection of split flows onto a conditioned sorbent tube. This is an invaluable feature for TD that enables repeat analysis and validation of analyte recovery through the sample flow path.

For these experiments, a split flow of 9 mL/min was applied during trap desorption with all other parameters remaining the same. Figure 7 displays the initial analysis (top), which was quantitatively re-collected and re-analysed (below).

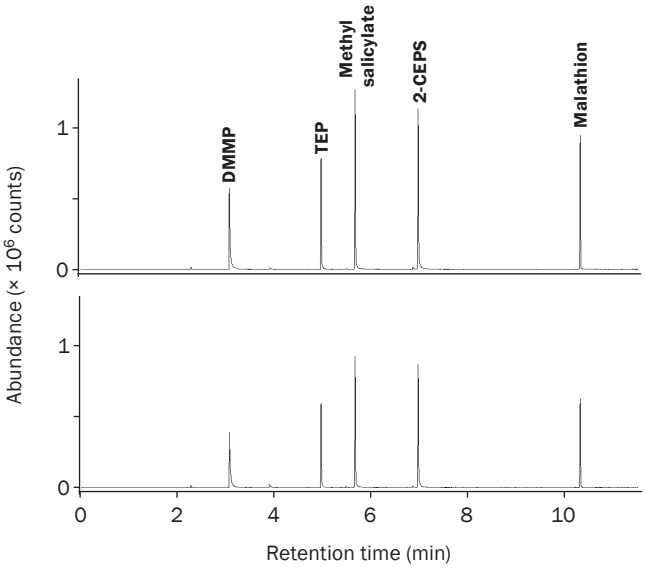

Figure 7: Comparison of initial and repeat analysis of a 100-ng standard shows good recovery for all five target compounds.

The chromatograms show a matching profile for both results with peaks in the re-collected sample at levels predicted from the split ratio indicating excellent recovery across the target analyte range.

When CW applications can be run with a small split, re-collection also allows archiving of critical samples for further analysis by the same or different methods. It also allows samples to re-collected and shipped to secondary laboratories for verification.

Analysis of chemical warfare agents

In Phase 2 of the study, Markes' system's performance was evaluated using live agents. For this part of the study, a UNITY–ULTRA-xr system for standard (3.5-inch) tubes was connected to a GC–MS/dual FPD instrument configured for both S and P modes for simultaneous full characterisation of agents with variable splits for higher sensitivity to specified detectors. A mixed 10-ng standard of live agents was spiked onto an inert-coated sorbent tube (C1-CAXX-5003) that had been used to sample air. The results are shown in Figure 8.

The total ion chromatogram (TIC) shows significant interferences and illustrates the difficulty in identifying trace chemical warfare agents in real samples. The merged extracted ion profiles (EIC masses 86, 99, 106, 114, 127 and 133 m/z) identify multiple agents including sarin (4.9 minutes), soman (8.4 minutes), tabun (9.76 minutes), mustard gas (10.71 minutes), VX (18.8 minutes) and Russian VX (19.05 minutes) and demonstrate the excellent performance of Markes' TD systems for live agent analysis.

Key aspects of Markes' TD technology, which facilitate analysis of such challenging compounds, include:

- The short, inert and uniformly-heated flow path, including proprietary heated TD valve.
- The exceptional performance of the focusing trap with low thermal mass and heating rates of 100°C/s, delivering fast GC injection and optimum sensitivity (narrow peaks) even with low or zero split flows.
- The proprietary heated valve, which enables analytes to enter and leave the trap from the same end (backflush operation), allowing the highest-boiling, stickiest compounds to be trapped and released from the front of the trap – quickly and easily. This allows target compounds over a wide volatility range to be analysed simultaneously and ensures good recovery and peak shape.
- Selection of hydrophobic sorbents and precise control of trapping temperatures allow any atmospheric humidity (water) to be purged out of the system before analysis.

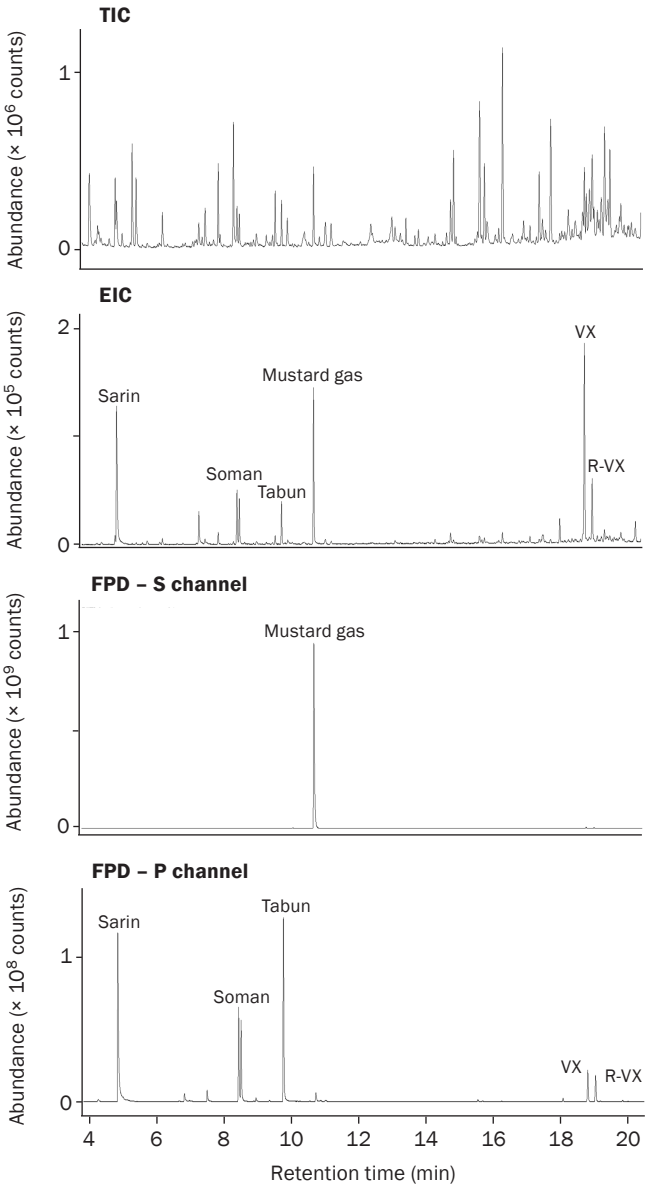

Figure 8: TIC (top) of a 10-ng mixed agent standard on a tube with interferences from the atmospheric sample. Trace 2 shows the merged EIC for target compounds. Trace 3 shows mustard gas (HD) in the S channel of the FPD and Trace 4 (bottom) shows the P channel for the remaining agents (sarin, soman, tabun, VX and Russian VX). Analysis performed using a TD–GC–MS/dFPD with a 70:30 split to the MS.

Results from the FPD detector in S mode show a much higher response than MS for mustard gas. Similarly, using the FPD in P mode showed a much higher response than MS for the other five agents.

Although 10 ng is a relatively high level for the analysis of CWAs, subsequent re-desorption of the same sorbent tube showed no carryover for all compounds except VX, which is notoriously sticky. However, carryover for VX was below 0.25%, even with 70% of the sample sent to the dFPD for higher sensitivity (Figure 9).

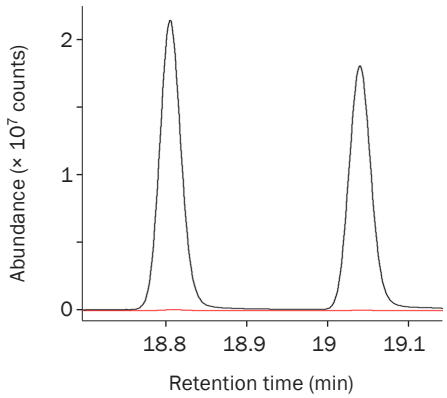

Figure 9: Overlay of FPD P channel for VX and Russian VX, respectively, showing the original 10-ng desorption (black) and subsequent re-desorption (red). Carryover was very low, producing no visible peaks even with a higher sensitivity setting.

Real-world air monitoring of agents requires detection at picogram levels to protect the public and military personnel. Analysis of a 10-pg standard of live agent showed no response for the target compounds using the mass spectrometer in full scan, but good response and peak shape with the FPD channels as shown in Figure 10.

VX and Russian VX can be seen in the inset and show an excellent response and peak shape for a 10-pg standard. The analysis was again carried out splitless on the TD system but did include capillary flow splitting technology to divide the sample between the mass spectrometer and flame photometric detector, meaning only ~6 pg of each analyte reached the selective detector for measurement.

Conclusions

This study demonstrates the excellent performance of Markes' TD systems for analysing both live chemical warfare agents and simulants at trace and higher levels.

The inert flow path contained within every Markes TD system has proven its compatibility with sticky compounds such as malathion and VX and with highly reactive and toxic compounds such as Russian VX and mustard gas without degradation or losses. The value of advanced techniques such as trap stacking has also been demonstrated, enabling trace toxic compounds to be detected at picogram levels even when using a split method. Further, the advantages of enhanced internal standard addition options, allowing better quality control of both sampling and analytical procedures, have also been demonstrated and discussed.

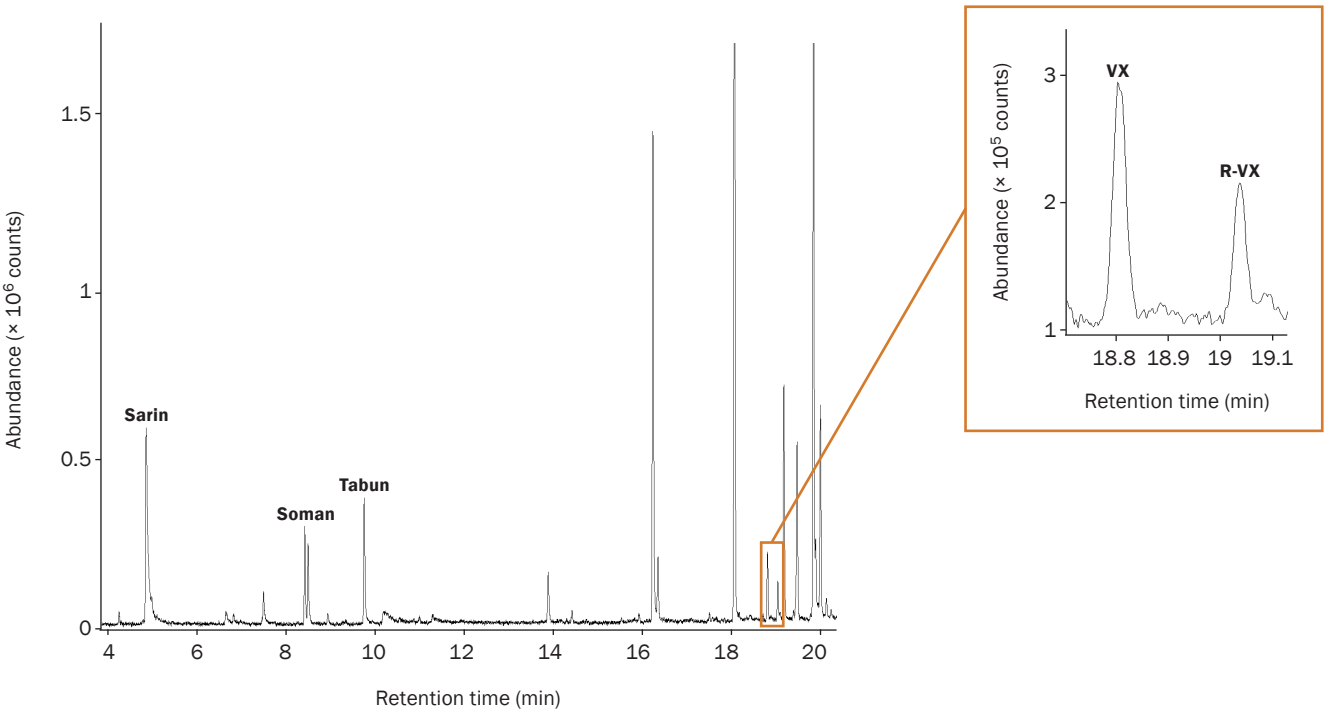

Figure 10: (Left) Analysis of an air sample with an FPD P channel showing sarin, soman, tabun, VX and Russian VX at the 10-pg level on the tube. (Right) Inset image shows a close-up of VX and Russian VX identified.

References

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Trademarks

Applications were performed under the stated analytical conditions. Operation under different conditions, or with incompatible sample matrices, may impact the performance shown.