Introducing Gas Chromatography – Vacuum Ultraviolet Spectroscopy (GC-VUV)

A Novel Tool for the Identification and Quantitation of Gas-Phase Analytes

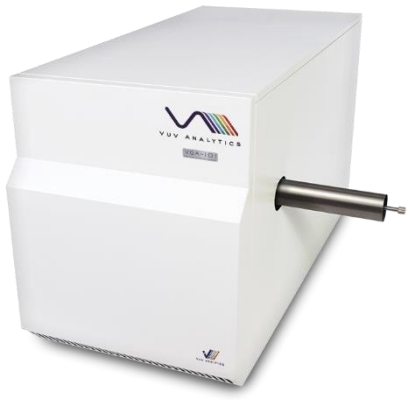
WHITE PAPER

Introduction

Gas Chromatography – Vacuum Ultraviolet (GC-VUV) spectroscopy is a recent innovation introduced in 2014 as a universal detection platform for gas chromatography[1]. The vacuum ultraviolet (VUV) absorbance spectrum measured by VUV detectors (120 – 240 nm) had historically been restricted to bright source synchrotron facilities due to significant background absorption challenges inherent to working within the wavelength range. The VGA-100™ GC-VUV detector is the first bench-top spectrometer capable of full VUV spectrum detection, and the VGA-101™ extends its wavelength detection range to 430 nm. Both VUV spectrometers have the unique capability of providing spectral information that is both qualitative and quantitative for most gas phase compounds.

GC-VUV spectral data is inherently three dimensional (time, absorbance, wavelength) and specific to chemical structure. Nearly all compounds absorb in the VUV region of the electromagnetic spectrum with the exception of carrier gases hydrogen, helium, and argon. The high energy, short wavelength VUV photons probe electronic transitions in virtually all chemical bonds including ground state to excited state $\sigma \rightarrow \sigma^*$ and $\pi \rightarrow \pi^*$. The result is spectral "fingerprints" that are specific to individual compound structure and can be readily identified by the VUV library. Unique VUV spectra enable closely related compounds such as structural isomers to be clearly differentiated. VUV detectors provide the perfect complement to mass spectrometry, which struggles with characterizing constitutional isomers and compounds with low mass quantitation ions. VUV spectra can also be used to deconvolve analyte co-elution, resulting in an accurate quantitative representation of individual analyte contribution to the original response[2]. This characteristic lends itself to significantly reducing GC runtimes through flow rate-enhanced chromatographic compression.

VUV spectroscopy follows the simple linear relationship between absorbance and concentration described by the Beer-Lambert Law. The straightforward nature of VUV spectral data eliminates guesswork related to retention time based identification and makes the technology accessible to users in both R&D and production settings. VUV detector ease of use is further enhanced by the ability to automate compound class data analysis. VUV absorbance spectra exhibit feature similarity within compound classes. Rapid compound class characterization can be achieved in complex samples utilizing compound spectral shape and retention index information. VUV Analyze™ software automation reduces the typical group analysis data processing time from 15 – 30 minutes to <1 minute per sample. This brief introduction to GC-VUV describes how its capabilities can be used to solve analytical challenges across a variety of difficult applications.



VUV ANALYTICS

Discussion

- VGA Gas Chromatography Detectors

The VGA-100 and VGA-101 gas chromatography detectors are compatible with most major GC manufacturers. The detectors can be connected through a heated transfer line which is inserted through a punch-out in the GC oven casing. A makeup flow of carrier gas is introduced at the end of the transfer line. Analytes arrive in the flow cell and are exposed to VUV light from a deuterium lamp. Specially coated reflective optics paired with a back-thinned charged coupled device (CCD) enable the collection of high quality VUV absorption data. Figure 1 shows a schematic of the analyte path from GC to VUV detector.

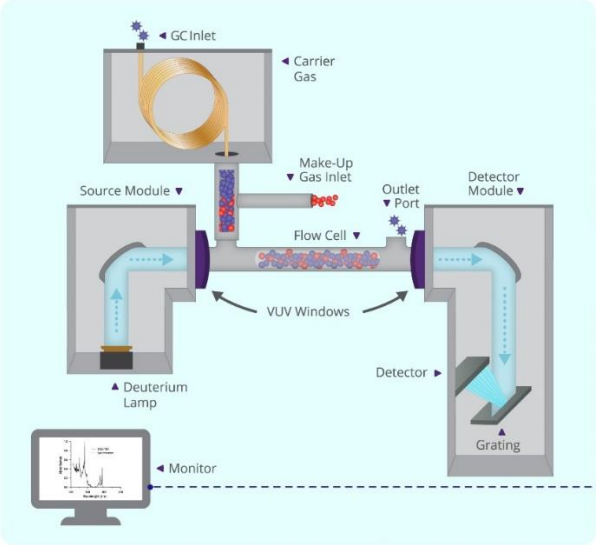


Figure 1: Schematic of the GC-VUV instrumental setup (not to scale). Dimensions of the detector are 30" x 13" x 17" or 76.2 x 33 x 43.2 cm. Flow cell volume is ~40 µL. Path length is 10 cm.

- VUV Spectral Identification

Gas phase species absorb and display unique spectra between 120 – 240 nm where high energy $\sigma \rightarrow \sigma^*$, $n \rightarrow \sigma^*$, $\pi \rightarrow \pi^*$, $n \rightarrow \pi^*$ electronic transitions can be excited and probed. VUV spectra reflect the absorbance cross section of compounds and are specific to their electronic structure and functional group arrangement. The ability of VUV detectors to produce spectra for most compounds results in universal and highly selective compound identification. VUV spectroscopy data is highly characteristic while also providing quantitative information. Many commonly used GC detectors such as the electron capture detector (ECD), flame ionization detector (FID), and thermal conductivity detector (TCD) produce quantitative but not qualitative detail. Gas chromatography – mass spectrometry (GC-MS) generates qualitative and quantitative data but has difficulty characterizing labile and low mass compounds, as well as differentiating between isomers. GC-VUV complements MS by overcoming its limitations and providing a secondary method of confirmation. It also offers a single instrument alternative to the use of multiple detectors for qualitative and quantitative analysis.

Naphthols, xylenes, and cis- and trans-fatty acids are compounds that are prohibitively difficult to distinguish according to their electron ionization mass spectral profiles[1]. Xylenes present the additional challenge of natural co-elution that makes separating their isoforms problematic. Figure 2 shows the distinct VUV spectra of m-, p-, and o-xylene. These compounds can be differentiated despite their only difference being the position of two methyl groups around a benzene ring. As will be seen later, the spectral differences of these isomers enable their co-elution to be resolved through spectral deconvolution.

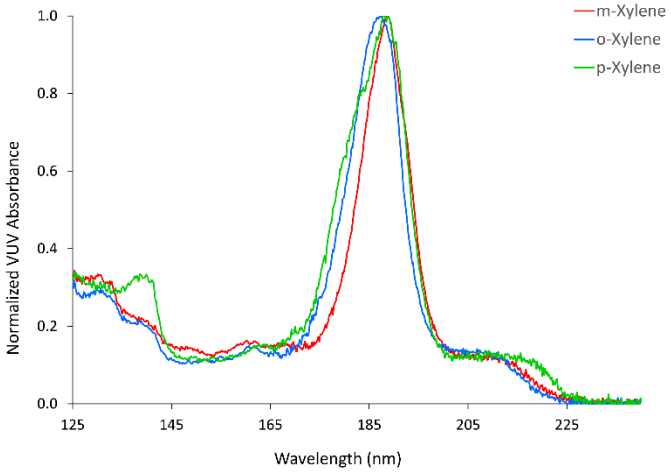


Figure 2: The distinct VUV spectra of m-, p-, and o-xylene. The compounds differ by only the positions of two methyl groups on a benzene ring, and are virtually impossible to distinguish by gas chromatography – mass spectrometry (GC-MS).

Fatty acid screening and profiling is an application that commonly requires the use of multiple detectors to achieve quantitative and qualitative results[2]. FID is a quantitative detector that is suitable for routine screening when guided by retention index information. GC-MS has traditionally been used for qualitative compound profiling, but falls short where isobaric analytes are prevalent. It especially struggles with differentiating *cis* and *trans* fatty acid isomers. Electron impact ionization can also cause double bond migration and lead to ambiguous fatty acid structural data.

Determining *cis* and *trans* fatty acid distribution in oils and fats is important in assessing their potential health impacts. VUV spectra of *trans*-containing fatty acid methyl ester (FAME) isomers typically found in butter and vegetable oils are shown in Figure 3. These *trans*-containing isomers separate chromatographically from *cis*-containing isomers and have the tendency to co-elute with each other, and in some cases, with select C20:1 isomers. GC-VUV is not only able to differentiate the C18:3 FAME variants, but is also capable of telling *cis* isomers apart from *trans* isomers. Degrees of unsaturation such as C20:1 vs. C18:3 can additionally be distinguished. Previous work has demonstrated how distinct VUV spectra enable straightforward deconvolution and accurate quantitation of *cis* and *trans* FAME isomers[1,3].

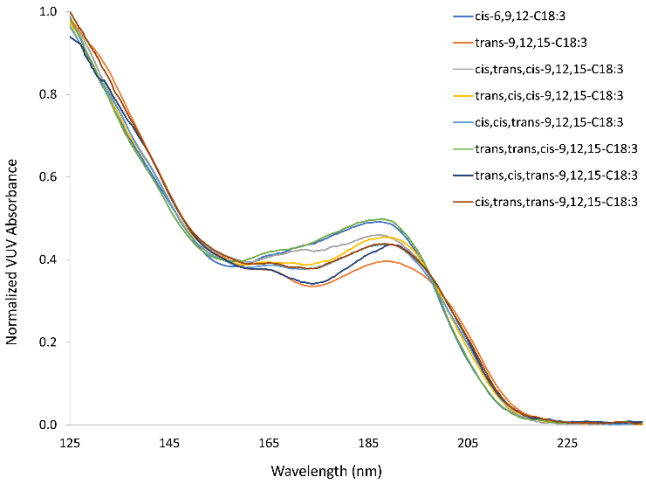


Figure 3: VUV spectra of fatty acid methyl ester (FAME) *cis* and *trans* isomers that are commonly found in butter and vegetable oils. GC-VUV can readily distinguish between the C18:3 FAME isomers, *cis* and *trans* classification, and the degree of unsaturation.

- Fast GC-VUV and Deconvolution

Unique VUV absorbance spectra not only enable unambiguous compound identification, but also allow GC runtimes to be deliberately compressed. VUV detectors operate at ambient pressure and are thus not flow rate limited. Chromatography can be compressed by increasing the GC column flow and oven temperature program rates.

Flow rate-enhanced chromatographic compression utilizes VUV spectral deconvolution to resolve any co-elution that may result from shortening GC runtimes. VUV absorption is additive, meaning that overlapping peaks give a spectrum that corresponds to the sum absorbance of each compound. The individual contribution of each analyte can be determined if the VUV spectra for co-eluting compounds are stored in the VUV library[4]. The ability to differentiate coeluting analyte spectra and use them to deconvolve the overlapping signals is demonstrated in Figure 4. The individual spectra of terpenes limonene and p-Cymene are shown in Panel A along with the

summed absorbance of the selected retention time window (blue region in Panel B) and the fit with VUV library spectra. The R^2 >0.999 fit result confirms their identities, and enables the deconvolution of these and other terpenes analyzed by fast GC-VUV as featured in Panel B.

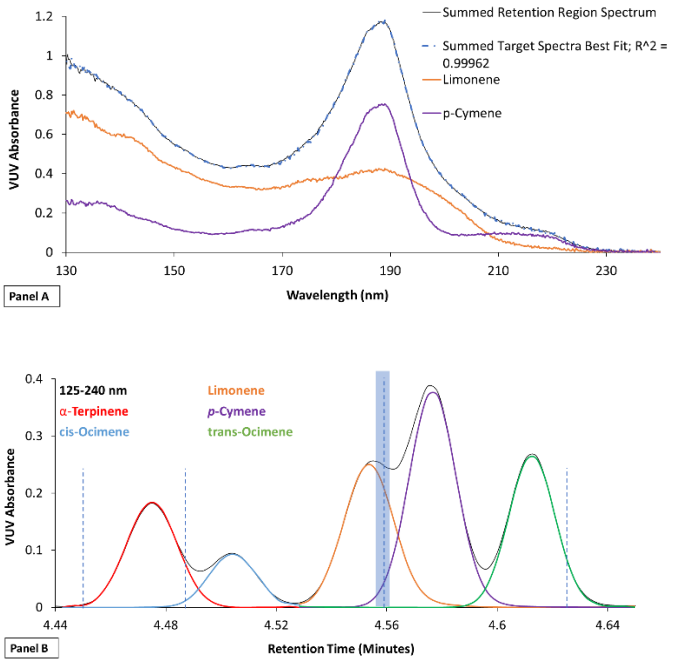


Figure 4: Panel A shows the individual spectra of limonene and p-Cymene along with the summed absorbance of the selected retention time (blue region in Panel B) and the fit with VUV library spectra. The deconvolution of these and other terpenes analyzed by fast GC-VUV is featured in Panel B.

Testing for the presence of residual solvents in Active Pharmaceutical Ingredients (APIs) is critical for patient safety and commonly follows United States Pharmacopeia (USP) Method <467> guidelines, or more broadly, International Council for Harmonization (ICH) Guideline Q3C(R6). The gas chromatography (GC) runtime suggested by USP Method 467 is approximately 60 min. A generic method for residual solvent analysis by GC-MS describes conditions that include a runtime of approximately 30 minutes[5]. A GC-VUV and static headspace method was developed using a chromatographic compression strategy that resulted in a GC runtime of 8 minutes. The GC-VUV method uses a flow rate of 4 mL/min and an oven ramp of 35°C (held for 1 min), followed by an increase to 245°C at a rate of 30°C/min.

Figure 5 compares the results when the general conditions of the GC-MS method were followed against the GC-VUV method run with Class 2 residual solvents. Tetralin eluted at approximately 35 minutes using the GC-MS method conditions, whereas the analyte had a retention time of less than 7 minutes when the GC-VUV method was applied. The co-elution of m- and p-xylene occurred in both GC-MS and GC-VUV method runs. VUV software matched the analyte absorbance of both isomers with VUV library spectra (Figure 2) to deconvolve the overlapping signals as displayed in Figure 6. Goodness of fit information ensures that the correct compound assignment takes place during the post-run data analysis.

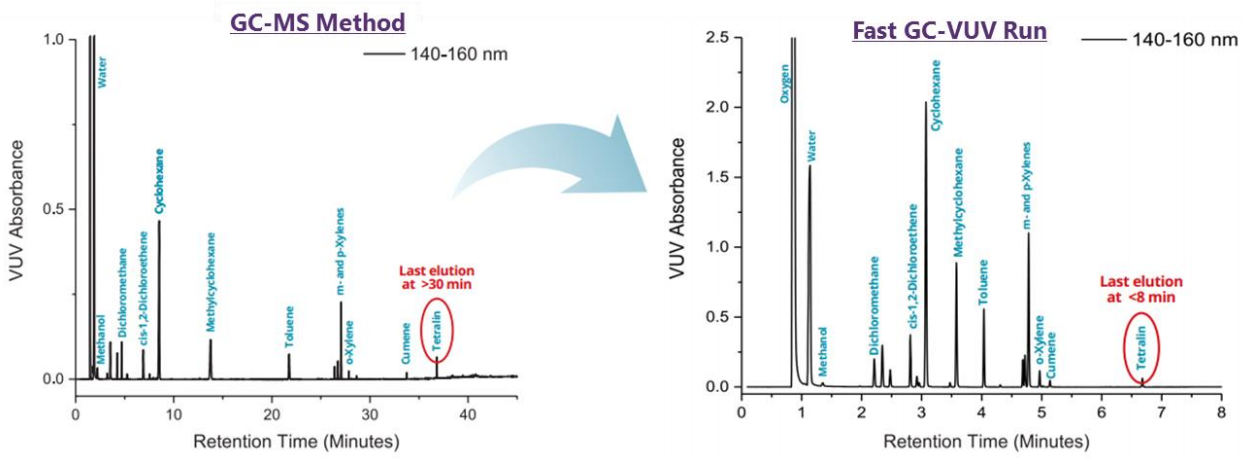


Figure 5: Comparison of legacy GC-MS and fast GC–VUV method runtimes for residual solvent analysis. Tetralin elutes at >30 minutes using the GC-MS method conditions, whereas the fast GC-VUV method elutes it at <7 minutes.

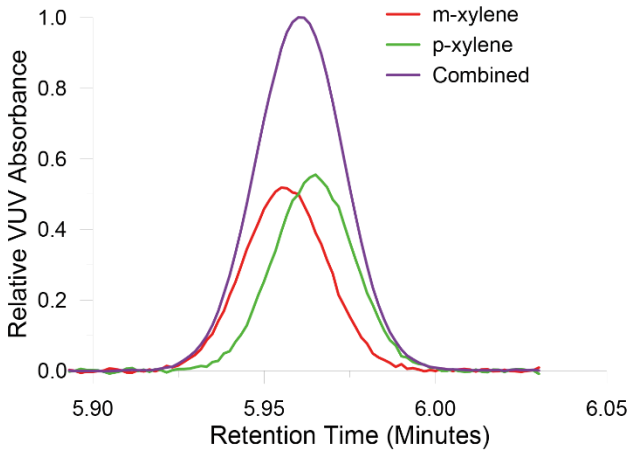


Figure 6: The deconvolution of m- and p-xylene co-elution. The relative contribution of each analyte is shown relative to the sum absorbance.

The flow rate-enhanced chromatographic compression strategy has been applied to a diverse set of applications since the development of the GC-VUV method for residual solvents analysis. The fast GC-VUV approach reduced GC runtimes for terpene analysis from 30 minutes to 9 minutes (the deconvolution of monoterpene isomers is shown in Figure 4). It has also been demonstrated that GC runtimes as short as 14 minutes can be used for PIONA compound analysis of gasoline samples. Typical GC separation times range between 1 – 2 hours using alternative methods. VUV Analyze™ software enables the faster GC-VUV approach by performing PIONA compound class characterization and deconvolving co-eluting analytes during its automated data analysis procedure.

- Compound Class Characterization

GC-VUV is well suited for use in production settings where compound compositional analysis is desired. Because compounds share spectral shape characteristics within a class, VUV Analyze™ software is able to apply its fitting procedure to quickly determine the relative contribution of each compound category present in a sample. Retention index information is used to limit the amount of VUV library searching and fitting performed for each analyte, enabling analysis times of <1 minute per sample. This automated data processing requires the user to simply locate a run file and then initialize the analysis once the initial setup has been completed. Compound class or specific compound concentrations can be reported as either mass or volume percent.

GC-VUV bulk compound characterization was first applied to the analysis of paraffin, iso-paraffin, olefin, naphthene, and aromatic (PIONA) hydrocarbons in gasoline streams. The associated method eliminates the need for multiple column use and complex instrumental setup while reducing the GC runtime from 1 – 2 hours to approximately 34 minutes. Approved as ASTM D8071, it is suitable for use with finished gasoline, reformate, reformer feed, FCC, light naphtha, and heavy naphtha samples. A typical VUV Analyze™ chromatographic analysis is displayed in Figure 7. The inset shows how the analyte spectral response is fit with VUV library spectra for the selected time slice. VUV Analyze™ provides a report detailing the carbon number breakdown within each PIONA compound class, as well as the relative mass or volume percent of classes within the sample. A table with mass % and carbon number data from a gasoline sample run using ASTM D8071 can be seen in Figure 8.

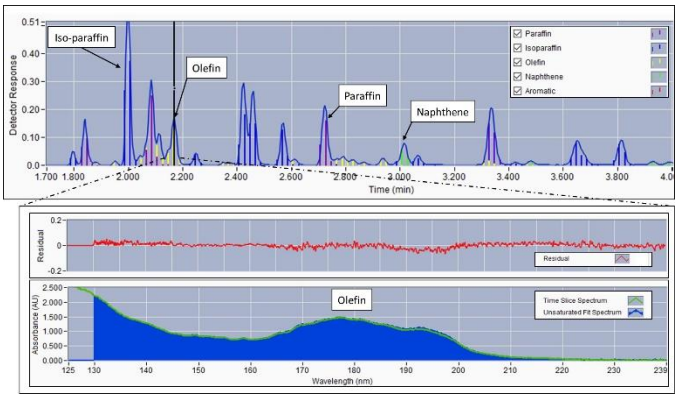
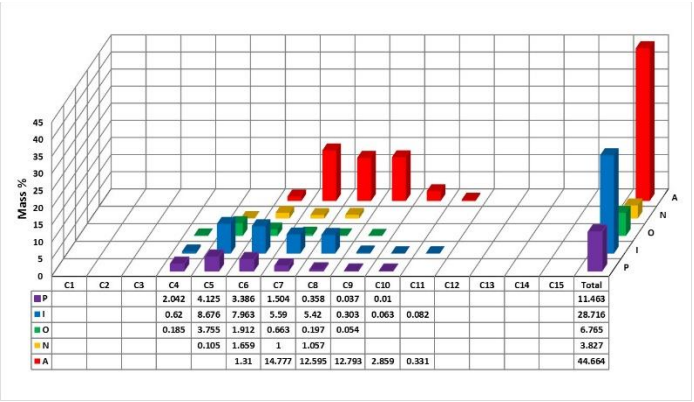


Figure 7: Zoomed-in chromatogram of gasoline sample with key PIONA compound class representative peaks labeled. Inset figure shows analyte spectral features fit with VUV library olefin compound class spectral response information. The residual fit statistical data indicating a good fit is also shown.



	C1	C2	C3	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C15	Total
P				2.042	4.125	3.386	1.504	0.358	0.037	0.01						11.463
I				0.62	8.676	7.963	5.59	5.42	0.303	0.063	0.082					28.716
O				0.185	3.755	1.912	0.663	0.197	0.054							6.765
N					0.105	1.659	1	1.057								3.827
A						1.31	14.777	12.595	12.793	2.859	0.331					44.664

Figure 8: Compositional analysis of gasoline sample run using ASTM D8071. Carbon number and mass % or volume % composition of PIONA compounds are reported by VUV Analyze™ automated software.

Conclusion

VUV spectroscopy has unique capabilities that address many of the challenges inherent to gas chromatographic separation and detection. VUV light probes electronic transitions that are unique to individual compound structure. VUV spectral fingerprints are used to differentiate closely related compounds, including structural isomers. GC runtimes can be significantly reduced through flow rate enhanced chromatographic compression and by resolving co-elution with VUV spectral deconvolution. GC-VUV data is both qualitative and quantitative, reducing the burden of multiple detector approaches to fully characterize sample analytes. The simplicity of VUV data and the ability to automate compound class analysis results in a detector that is suitable for both R&D and production settings.

For more detailed information please visit our website at www.vuvanalytics.com, or contact us at info@vuvanalytics.com.

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