ATMOSPHERIC PRESSURE GC (APGC)

Expanding analytical capabilities in food and environmental laboratories

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Brings advanced MS technologies previously unavailable to GC users

Atmospheric Pressure GC (APGC) is one of the universal source options that enables access to Waters' advanced MS technologies including the Xevo™ TQ-S micro, Xevo TQ-XS, Xevo G2-XS QTof, SYNAPT™ G2-S*i*, and Vion™ IMS-MS QTof systems for analyzing GC amenable compounds.

APGC is an soft ionization technique similar to Atmospheric Pressure Chemical Ionization (APCI) where ionization occurs at atmospheric pressure. When compared to EI, APGC yields less fragmentation, often revealing the presence of a molecular ion (or protonated molecular ion). This means sensitivity and selectivity are increased, which can simplify MRM precursor selection. Operating the ionization at atmospheric pressure removes the GC flow rate restriction imposed by a vacuum system, allowing a much wider range of flow rates and carrier gas options for GC separations. APGC is not intended to replace EI, but to compliment it as it provides different advantages.

In APGC, a corona discharge forms a plasma with an incoming stream of nitrogen under atmospheric conditions. After GC separation, compounds exit the chromatographic column into the plasma (Figure 1) and ionization of the compounds takes place by two primary mechanisms: charge transfer (Figure 2) and proton transfer (Figure 3). The molecular ion, protonated molecular ion, or a mixture of both types of ions are usually present. Proton transfer is heavily influenced by the presence of a protic modifier (water, methanol or 2-propanol) in the source that enhances protonation whereas charge transfer reactions are optimum under dry source conditions.

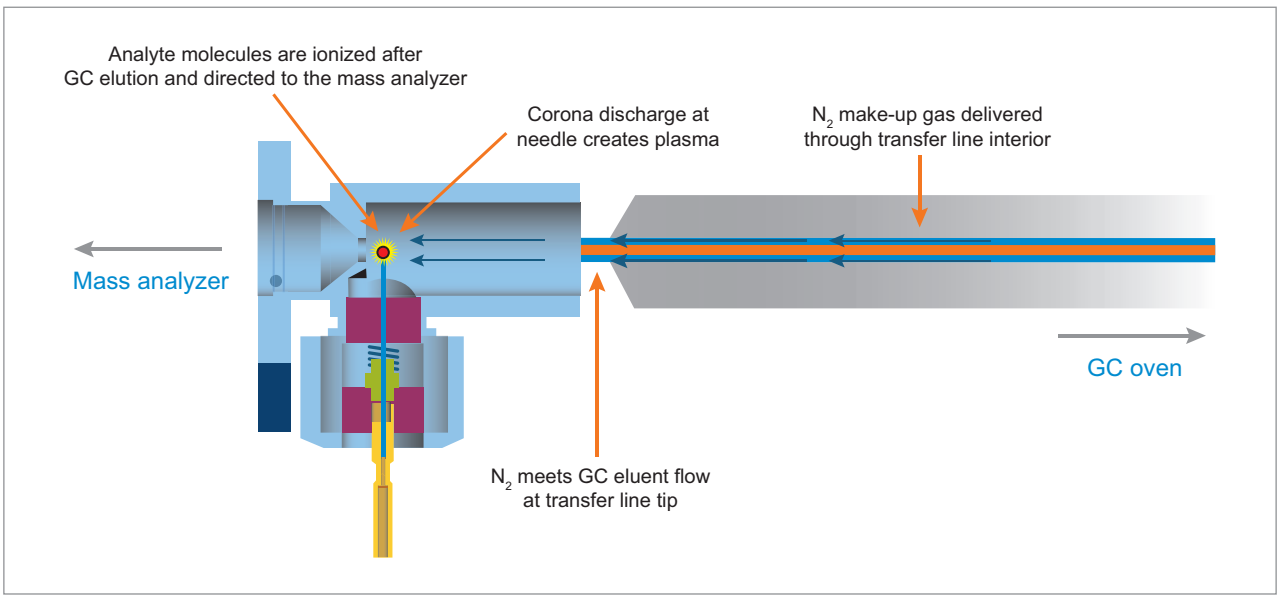

Figure 1. Schematic diagram of the APGC Source.

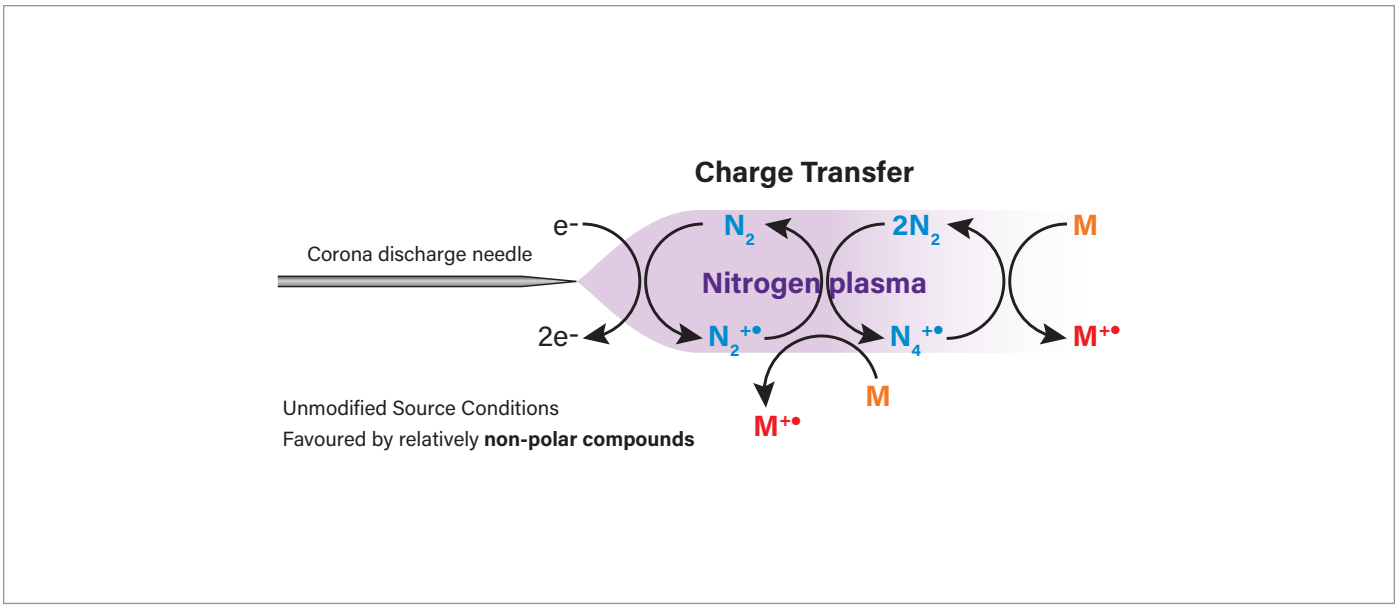

Figure 2. Charge transfer.

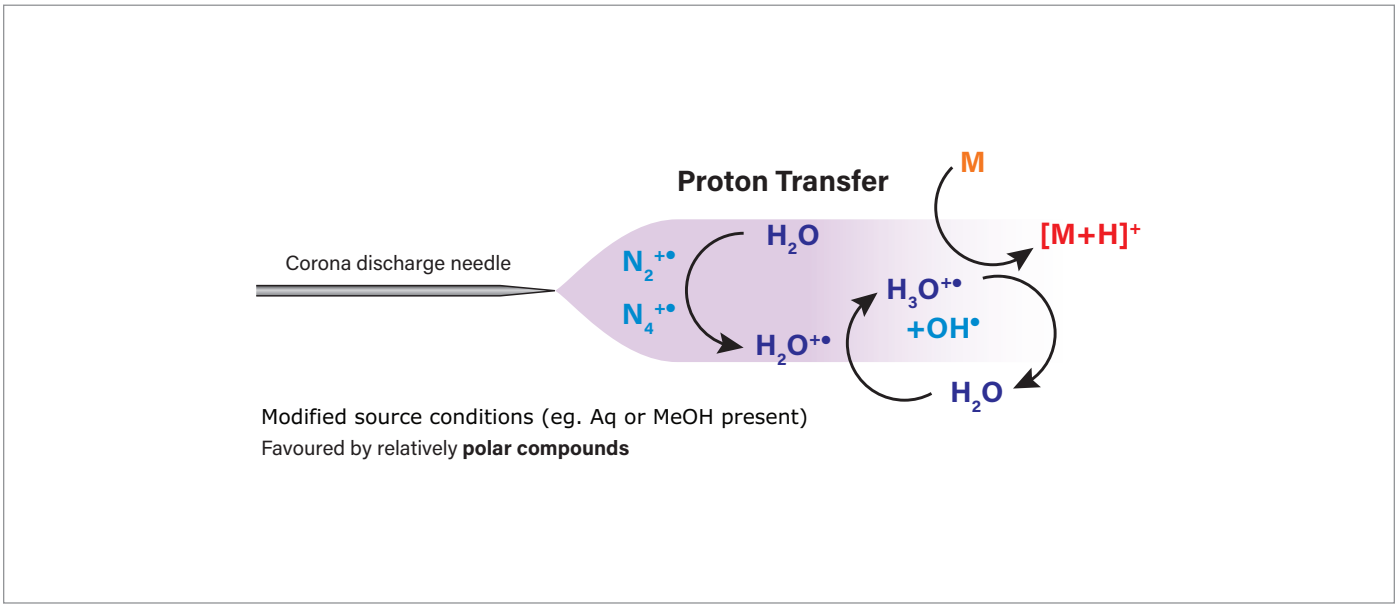

Figure 3. Proton transfer.

[PLATFORM FLEXIBILITY]

[INCREASE CHROMATOGRAPHIC FLEXIBILITY]

[APGC PERFORMANCE]

Platform Flexibility

Analyzing GC and LC amenable compounds often requires two dedicated systems. Waters' universal source design allows utilization of a single MS system for both GC- and LC-MS by switching from electrospray ionization to APGC and back again, quickly and easily. This provides complete compound coverage in applications such as pesticide screening and improves laboratory efficiency while maintaining required levels of sensitivity.

Using interchangeable universal source options, such as APGC, enables access to Waters' advanced MS technologies. For example, Tof capabilities of high-resolution/accurate mass measurements, MS^E, ion mobility separations/collision cross section (CCS), and tandem quadrupole functionalities of MRMs/PICs/RADAR, available on Waters LC-MS instruments, can be used for the analyses of GC-amenable compounds using these interchangeable sources.

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UPLC and APGC Multi Residue Pesticide Analysis on a Single Tandem Quadrupole Mass Spectrometer Platform

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APPLICATION BENEFITS

Using the Xevo® TQ-S micro Tandem Quadupole Mass Spectrometer with the Universal Source for pesticide analysis allows:

- UPLC® and APGC analysis of the sample extracts on a single tandem quadrupole mass spectrometer.
- Analysis of large suites of pesticides in a single injection per chromatographic inlet.
- Analysis of fruit and vegetable matrices at legislatively relevant levels of 0.010 mg/kg.
- Easy generation of methods using the Quanpedia™ Database.

WATERS SOLUTIONS

ACQUITY® UPLC H-Class System

Atmospheric Pressure Gas Chromatography (APGC)

Xevo TQ-S micro

DisQuE™ QuEChERS, AOAC Method Sample Preparation Kit, Pouches

MassLynx® MS Software

Quanpedia Database

TargetLynx™ XS Application Manager

KEYWORDS

LC, GC, pesticide residue analysis, MRL, QuEChERS, GC-MS/MS, LC-MS/MS

AIM

Demonstrate analysis of a large suite of pesticides in fruit and vegetable extracts using both LC and GC on the same tandem quadrupole MS platform at legislatively relevant limits.

INTRODUCTION

Hundreds of pesticides are commercially available and approved for use on various fruit and vegetable plants, to prevent pest infestation and improve shelf-life of fresh produce. Maximum Residue Levels (MRLs) are set at the highest level of pesticide that the relevant regulatory body would expect to find in that crop when it has been treated in line with good agricultural practice. In the EU, if a pesticide is not explicitly mentioned in the MRL legislation, a default MRL is used for enforcement. This default value is set to be equal to the limit of quantification (LOQ) achievable with the analytical methods used for analysis. National authorities control and enforce MRLs by testing samples for pesticide residue levels using analytical surveillance programs. These programs check for compliance with MRLs, assess dietary exposure, and check for use of unauthorized pesticides. The food industry also carries out its own due diligence analyses.

Mass spectrometry coupled with both gas (GC) and liquid chromatography (LC) is needed to provide comprehensive analysis of a wide range of pesticide residues with sufficient sensitivity to meet global MRL regulations. The use of Quick, Easy, Cheap, Efficient, Rugged and Safe (QuEChERS) sample extraction and clean up has streamlined analytical efficiencies for multi residue analyses.[1] The advantage of ultra performance liquid chromatography (UPLC) coupled with tandem quadrupole mass spectrometry (MS/MS) for multi residue pesticide analysis is widely reported.[2] More recently the use of GC-MS/MS operated at atmospheric pressure (APGC) has been shown to offer significant improvements in performance over electron impact (EI) for challenging pesticides, in terms of selectivity, specificity, and speed of analysis.[3,4]

The APGC source ionizes compounds using a corona discharge at atmospheric pressure in an APCI-like manner. Therefore, this ionization mechanism is a much softer technique than classic electron impact (EI) ionization and produces larger amounts of intact parent ions, especially in the case of fragile or easily fragmented compounds. APGC ionization can occur using two mechanisms; proton transfer (wet source) or charge transfer (dry source). In proton transfer ionization, $[M+H]^+$ ions are formed, whereas in charge transfer ionization, $M^{+\cdot}$ ions are formed.

In this application note, a single workflow for the multi residue analysis of pesticides is demonstrated on a variety of fruit and vegetable samples. Utilizing the universal source of Waters® Xevo TQ-S micro allows for LC and GC analyses to be completed on the same tandem quadrupole MS instrument, with less than 30 minutes needed to switch between chromatographic inlets. The performance of the method will be highlighted in terms of sensitivity, repeatability, and linearity for both LC and GC in compliance with the SANTE guidelines (11945/2015) for pesticide analysis.[5]

EXPERIMENTAL

The LC and GC suites of pesticides analyzed in this study (listed in the Appendix) were chosen to cover a wide range of different pesticide classes and chemistries. The multi residue MS/MS methods were generated using Quanpedia, with separate databases utilized for generation of the LC and GC methods. Each database contains MRMs and retention time information for each compound. When the MS method is generated the MRM function windows are automatically set for each compound. For the UPLC method, a window of 1 minute was placed around each compound's expected retention time. For the APGC method, a window of 30 seconds was used due to the narrower peak widths exhibited in GC analysis. In addition to the MS methods, TargetLynx data processing methods and the LC inlet method were also generated through the Quanpedia Database.

Sample extraction and cleanup

Celery, lemon, corn, and kale samples were purchased at a local grocery store. Samples were chosen to be representative of different types of matrix complexity from different commodity groups, including high water content (celery and kale), high acid content (lemon), and high starch/protein with low water content (corn). Samples were immediately homogenized in a food processer and frozen until sample preparation was performed. QuEChERS extraction was performed according to the official AOAC method 2007.01 using the DisQuE QuEChERS, AOAC Method Sample Preparation Kit (P/N 176002922).[6] Figure 1 highlights the sample extraction.

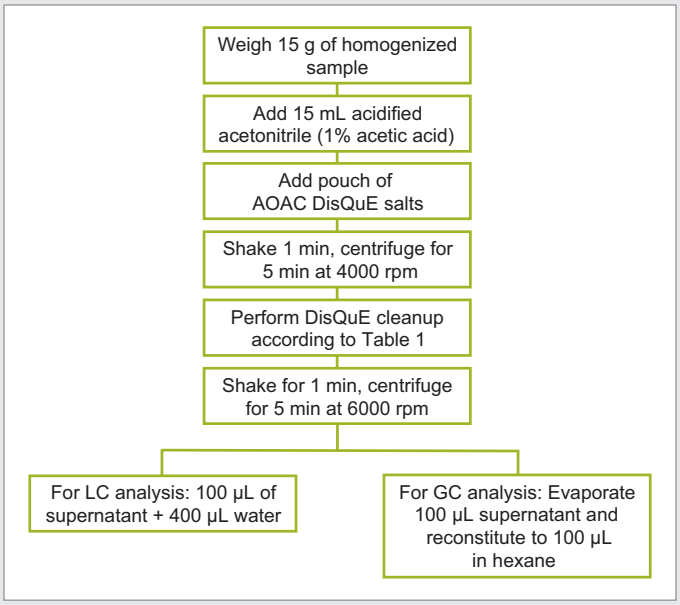

Figure 1. DisQuE sample extraction method.

Sample	MgSO4	PSA	GCB	Volume	Part number
Celery	150 mg	25 mg	7.5 mg	1 mL	186004831 + 186004835
Lemon	150 mg	25 mg	–	1 mL	186004831
Corn	150 mg	25 mg	–	1 mL	186004831
Kale	900 mg	150 mg	150 mg	6 mL	186004833 + 186004835

Table 1. dSPE cleanup conditions used for each sample matrix.

LC-MS/MS conditions

LC system:	ACQUITY UPLC H-Class
Column:	ACQUITY BEH C_{18} 1.7 µm 2.1 x 100 mm
Column temp.:	45 °C
Injection volume:	5 µL
Flow rate:	0.45 mL/min
Mobile phase A:	Water + 10 mM ammonium acetate
Mobile Phase B:	Methanol + 10 mM ammonium acetate

Gradient:

Time (min)	%A	%B
0.00	98	2
0.25	98	2
12.25	1	99
13.00	1	99
13.01	98	2
17.00	98	2

MS system:	Xevo TQ-S micro
Ionization mode:	ESI+
Capillary voltage:	1 kV
Desolvation temp.:	500 °C
Desolvation gas flow:	1000 L/hr
Source temp.:	150 °C

GC-MS/MS conditions

GC system:	7890A
Autosampler:	CTC PAL
Column:	30 m x 0.25 mm x 0.25 µm Rxi-5MS
Carrier gas:	Helium
Flow rate:	2.0 mL/min
Injection:	Splitless
Injector temp.:	280 °C
Injection volume:	1 µL
Makeup gas:	Nitrogen at 250 mL/min
Transfer line temp.:	320 °C

Oven program:

Rate (°C/min)	Temp. (°C)	Hold (min)
–	80	1.00
25	150	0.00
8	270	0.00
20	320	4.10

MS system:	Xevo TQ-S micro
Ionization mode:	API+
Ionization mechanism:	Proton transfer (3 vials of water in source)
Corona current:	20 µA for first 3.5 min 3.0 µA for rest of run
Cone gas flow:	0 L/hr
Auxiliary gas flow:	250 L/hr
Source temp.:	150 °C

RESULTS AND DISCUSSION

METHOD MANAGEMENT USING THE QUANPEDIA DATABASE

Working with methods involving large numbers of compounds can be time consuming when done manually and is prone to errors when setting up time segmented acquisition. Quanpedia is a compound centric database, typically used for method generation, but can also function as a method management tool. Initial methods for this analysis were generated using existing UPLC and APGC databases (Figure 2). Retention time changes resulting from further method development or method changes wereupdated in the database. This allowed for immediate and automatic updates to be made in the MS and processing methods by just re-generating the methods in three simple clicks.

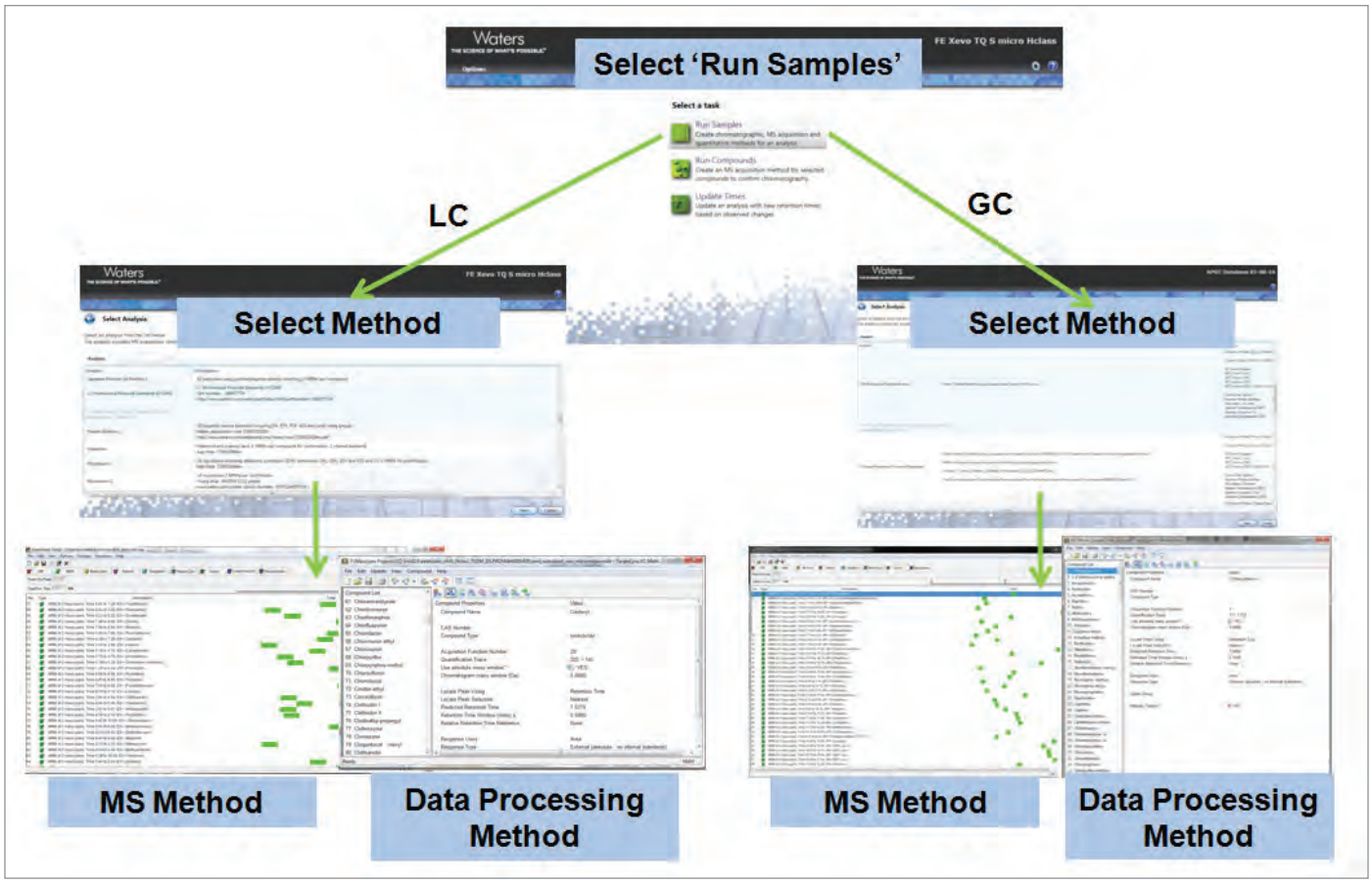

Figure 2. Quanpedia databases that were used to manage the methods for both UPLC and APGC analysis demonstrating the three click workflow of method generation.

RAPID AND ROBUST DATA AQUISITION

For successful analysis of large numbers of pesticides and their metabolites, it is important that the mass spectrometer can maintain sufficient sensitivity while acquiring MRM transitions with a fast scan speed to provide enough data points across each chromatographic peak (e.g. minumum of 12 points per peak). The fast scanning speeds of the TQ-S micro allow for this robust and rapid data acquisition while maintaining large retention time windows to accommodate any shift in retention time due to column maintenance (GC) or chromatography changes caused by the different matrices.[6]

Figure 3 highlights one of the busiest sections of the APGC MS Method. In this example, flutolanil is just one of approximately 30 pesticides (set across 30 channels, each acquiring at least two transitions per compound) eluting in a 1.5 minute time window. The dwell time calculated by the autodwell function to collect a minimum of 12 points per peak was 0.006 s. The resulting chromatogram of three replicate injections of 0.010 mg/kg of flutolanil in celery matrix can be seen in Figure 3. Even with the fast scanning speed, 19 points were collected across the peak and the RSD of three consecutive injections in matrix was 5.2%. The same is true for the UPLC method used for this analysis.

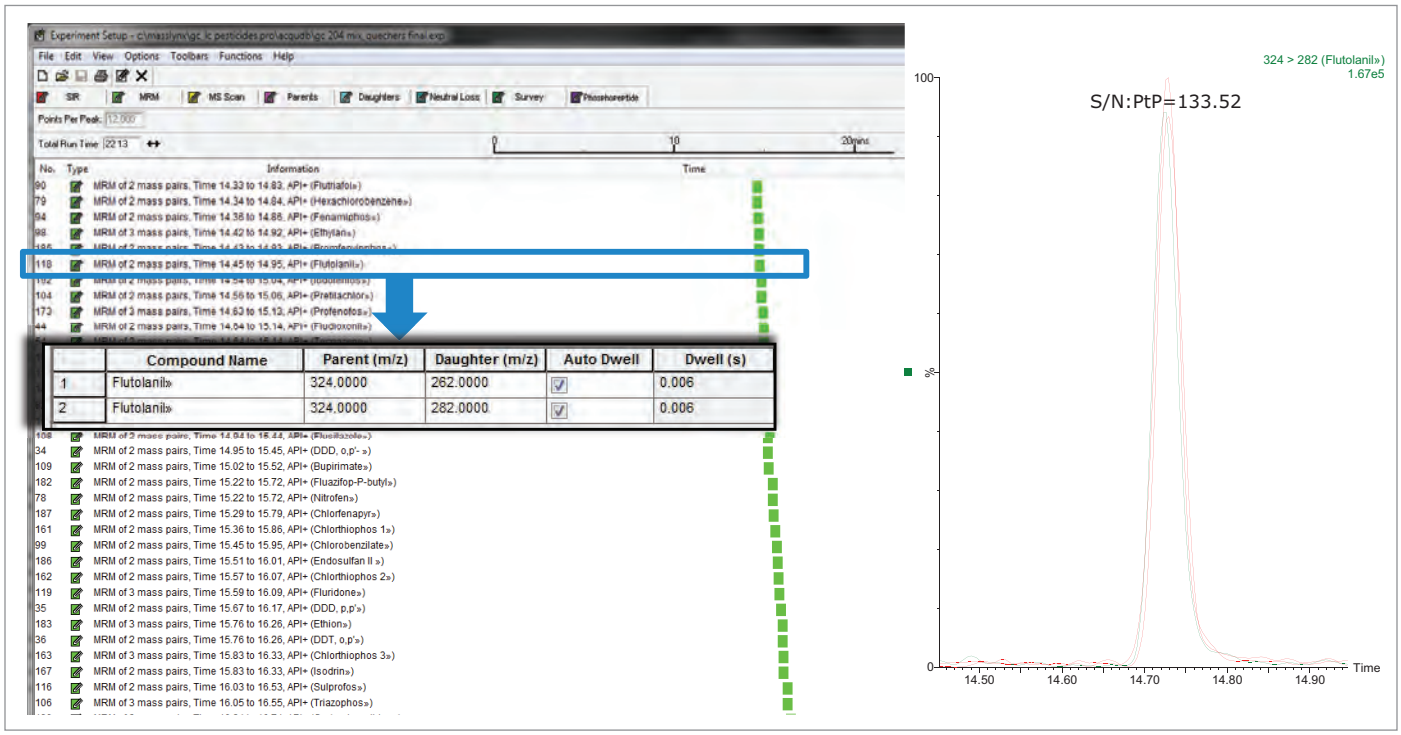

Figure 3. Demonstration of the fast scanning of the Xevo TQ-S micro demonstrating retention of peak quality at a fast scan time.

PESTICIDES IN MATRIX

Matrix matched standards were prepared in celery, lemon, corn and kale over a range of 0.001 to 0.050 mg/kg and replicate injections made using the UPLC and APGC methods. A summed MRM overlay of a selection of pesticides can be seen in Figure 4, showing 0.010 mg/kg in celery extract from both the (A) APGC and (B) UPLC analyses. The data were fitted with the best fit calibration; for the UPLC data, the response was shown to be linear whereas the APGC response over the range investigated was non-linear and so was fitted with a quadratic calibration. The majority of the compounds in both analysis methods had correlation coefficient (R^2) values of 0.995 or greater. Figure 5 shows the matrix matched calibration curves and the peak response at 0.001 mg/kg of a representative pesticide from each analysis method in the four matrices. Residuals from triplicate injections at each calibration point were within ±20%. Ion ratios were also shown to be within 30% tolerance of the reference values.

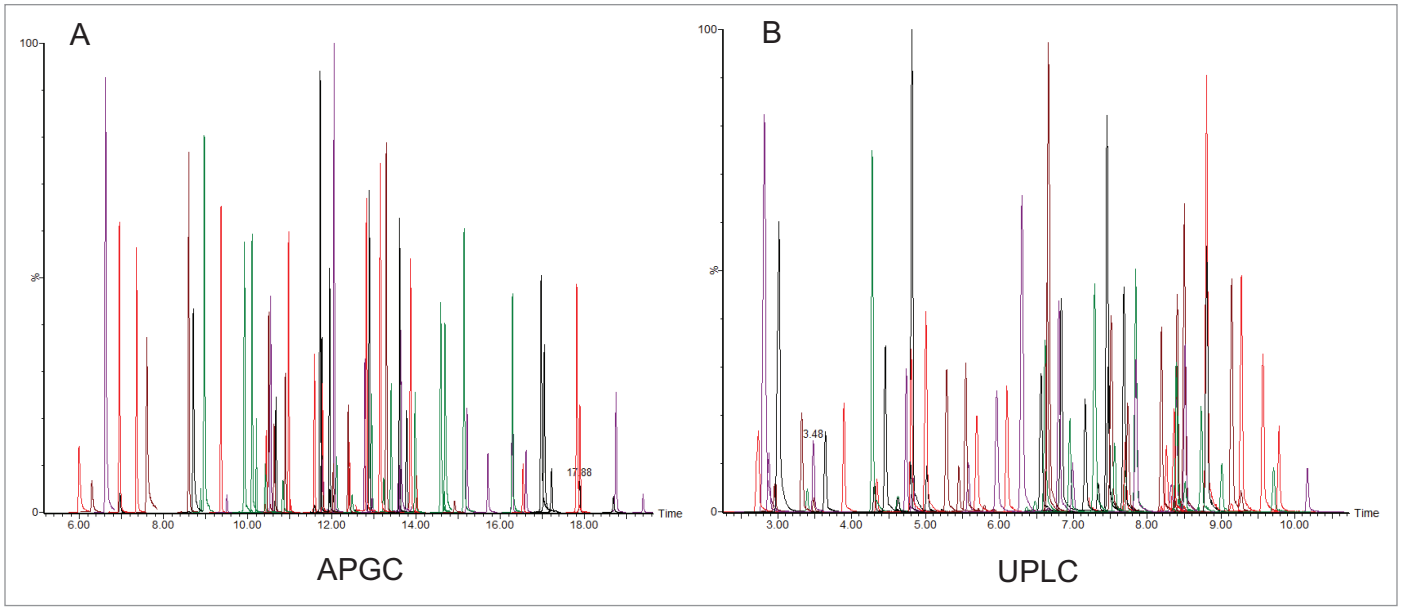

Figure 4. Overlay of a selection of pesticides at 0.010 mg/kg analyzed in a celery extract on A. APGC, and B. UPLC.

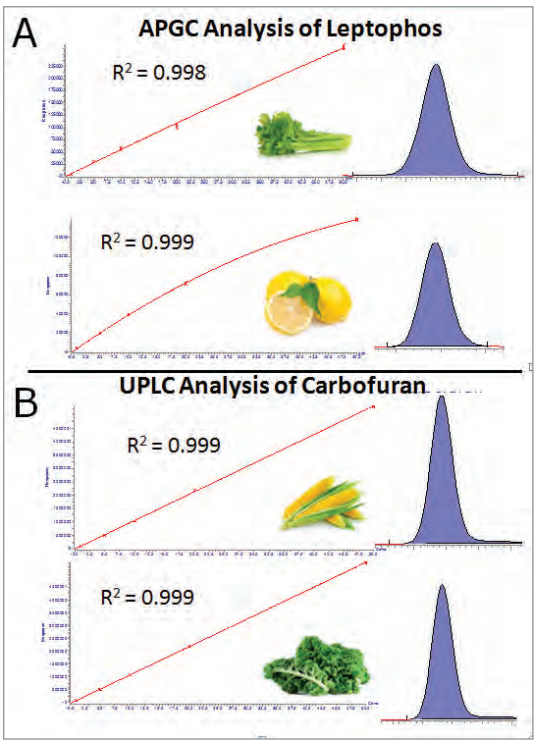

Figure 5. Matrix matched calibration curves and chromatograms for standards at 0.001 mg/kg for peaks from: A. APGC analysis of leptophos in celery and lemon; and B. UPLC analysis of carbofuran in corn and kale.

For convenience, all sample extracts were spiked at the default MRL of 0.010 mg/kg. Figure 6 demonstrates the percentage of pesticides in each method detected in the spiked matrices at 0.010 mg/kg. However many pesticides could also be detected at 0.001 mg/kg as demonstrated in Figure 5 showing leptophos (APGC compound) and carbofuran (UPLC compound) in the different matrices. The precision of the measurements was excellent with more than 90% of the detected pesticides exhibiting RSDs of peak area of less than 10% (n=3). The exception was the APGC analysis of the kale matrix which had more than 80% of pesticides exhibiting RSDs less than 10% (Figure 7).

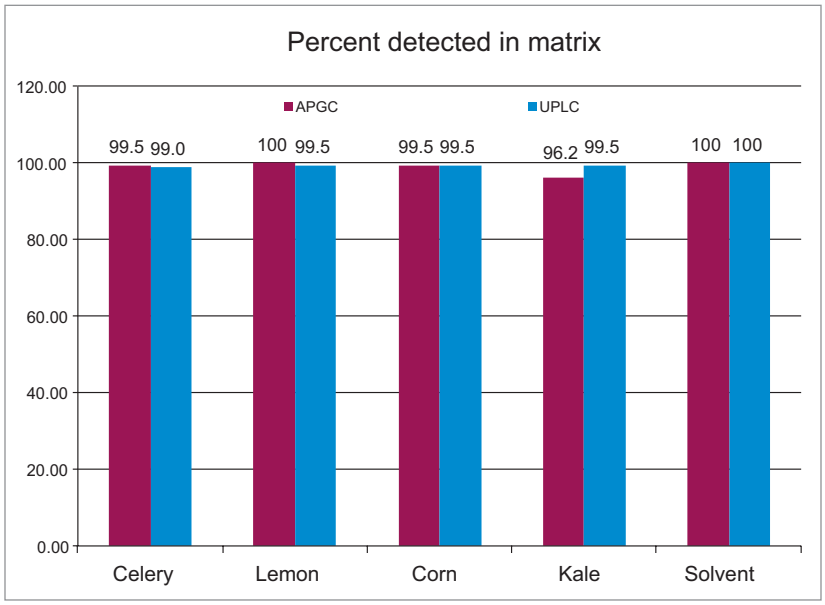

Figure 6. The percentage of pesticides detected in the 0.010 mg/kg standard for each matrix using both APGC and UPLC.

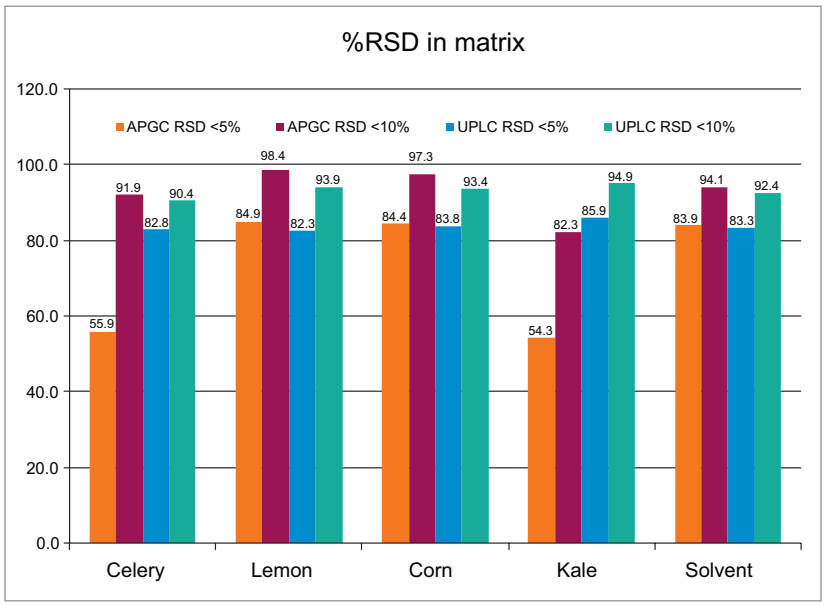

Figure 7. Percentage of compounds detected at 0.010 mg/kg in each matrix and associated RSDs.

CONCLUSIONS

Complex multi residue pesticide analysis was demonstrated using both UPLC and APGC analysis on the same tandem quadrupole instrument (Xevo TQ-S micro). Instrument methods were generated and maintained using Quanpedia databases making method generation and maintenance fast and simple. Although the multi residue methods contained approximately 200 compounds each, the reliable scanning speed of the TQ-S micro produced accurate and precise measurements. The performance for the determination of pesticide residues analyzed in four matrices of varying complexity complied with the SANTE guidelines for pesticide residue analysis. Detection at the EU default maximum residue limit of 0.010 mg/kg was easily achieved for >99% of pesticides analyzed with good precision (RSDs <10%) for most analytes in the food samples. Having the flexibility of the Universal Source architecture to provide access to both UPLC-MS/MS and GC-MS/MS on the same instrument, allows for an increase of laboratory efficiency, while maintaining required sensitivity and repeatability.

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Appendix

Pesticides in APGC Method

2-Phenylphenol	Diclobenil	Oxyfluorfen
4,4'-Methoxychlor olefin	Dicloran	Paclobutrazol
Acetochlor	Dimethachlor	Parathion
Acrinathrin	Diphenamid	Pebulate
Alachlor	Diphenylamine	Penconazole
Allidochlor	Edifenphos	Pendimethalin
Anthraquinone	Endosulfan ether	Pentachloroaniline
Atrazine	Endosulfan II	Pentachlorobenzonitrile
Azinphos-ethyl	Endosulfan sulfate	Pentachlorothioanisole
Azinphos-methyl	Endrin aldehyde	Permethrin, cis-
Benfluralin	EPN	Permethrin, trans-
Bifenthrin	Ethalfluralin	Phenothrin 1
Bioallethrin	Ethion	Phenothrin 2
Biphenyl	Ethylan	Phorate
Bromfenvinphos	Etofenprox	Phosalone
Bromfenvinphos-methyl	Etridazole	Phosmet
Bromophos-ethyl	Fenamiphos	Piperonyl butoxide
Bromophos-methyl	Fenarimol	Pirimiphos-ethyl
Bromopropylate	Fenchlorphos	Pirimiphos-methyl
Bupirimate	Fenitrothion	Prochloraz
Captafol	Fenpropathrin	Procymidone
Captan	Fenson	Prodiamine
Carbophenothion	Fenthion	Profenofos
Carfentrazone ethyl	Fenvalerate 1	Profluralin
Chlorfenapyr	Fenvalerate 2	Propachlor
Chlorfenvinphos	Fipronil	Propanil
Chlorobenzilate	Fluazifop-P-butyl	Propisochlor
Chloroneb	Fluchloralin	Propyzamide
Chlorothalonil	Flucythrinate 1	Prothiofos
Chlorpropham	Flucythrinate 2	Pyraclofos
Chlorpyrifos	Fludioxonil	Pyrazophos
Chlorpyrifos-methyl	Fluquinconazole	Pyridaben
Chlorthal-dimethyl	Flusilazole	Pyridaphenthion
Chlorthiophos 1	Flutolanil	Pyrimethanil
Chlorthiophos 2	Flutriafol	Pyriproxyfen
Chlorthiophos 3	Folpet	Quinalphos
Chlozolinate	Fonofos	Resmethrin 1
Clomazone	Hexachlorobenzene	Sulfotep
Coumaphos	Hexazinone	Sulprofos
Cycloate	Iodofenfos	tau-Fluvalinate 1
Cyfluthrin 1	Iprodione	tau-Fluvalinate 2
Cyfluthrin 2	Isazophos	Tebuconazole
Cyfluthrin 3	Isodrin	Tebufenpyrad
Cyfluthrin 4	Isopropalin	Tefluthrin
Cyhalothrin, lambda-	Lenacil	Terbacil
Cypermethrin 1	Leptophos	Terbufos
Cypermethrin 2	Linuron	Terbutylazine
Cypermethrin 3	Malathion	Tetrachloroaniline, 2,3,5,6-
Cypermethrin 4	Metalaxyl	Tetrachlorvinphos
Cyprodinil	Metazachlor	Tetradifon
DDD, o,p'-	Methacrifos	Tetramethrin 1
DDD, p,p'-	Methoxychlor	Tetramethrin 2
DDE, o,p'-	Methyl parathion	Tolclofos-methyl
DDE, p,p'-	Metolachlor	Tolylfluanid
DDT, o,p'-	Mevinphos	Transfluthrin
DDT, p,p'-	MGK 264 1	Triadimefon
Deltamethrin	MGK 264 2	Triadimenol
Diallate	Myclobutanil	Triallate
Diazinon	N-(2;4-Dimethylphenyl)formamide	Triazophos
Dichlofluanid	Nitralin	Triflumizole
Dichloroaniline, 3,4'-	Nitrofen	Trifluralin
Dichlorobenzophenone, 4,4'-	Oxadiazon	Vinclozolin

Pesticides in UPLC Method

Abamectin	Etoxazole	Nuarimol
Acephate	Famoxadone	Omethoate
Acetamiprid	Fenamidone	Oxadixyl
Acibenzolar-S-methyl	Fenarimol	Oxamyl
Aldicarb	Fenazaquin	Paclobutrazol
Aldicarb sulfone	Fenbuconazole	Penconazole
Aldicarb sulfoxide	Fenhexamid	Pencycuron
Ametryn	Fenobucarb	Phenmedipham
Aminocarb	Fenoxycarb	Picoxystrobin
Amitraz	Fenpropimorph	Piperonyl butoxide
Azoxystrobin	Fenpyroximat	Pirimicarb
Benalaxyl	Fenuron	Procloraz
Bendiocarb	Fipronil	Promecarb
Benfuracarb	Flonicamid	Prometon
Benzoximate	Flufenacet	Prometryn
Bifenazate	Flufenoxuron	Propamocarb
Bitertanol	Fluomethuron	Propargite
Boscalid	Fluoxastrobin	Propham
Bromuconazole I	Fluquinconazole	Propiconazole
Bromuconazole II	Flusilazole	Propoxur
Bupirimate	Flutolanil	Prothioconazole
Buprofezin	Flutriafol	Pymetrozine
Butafenacil	Forchlorfenuron	Pyracarbolid
Butocarboxim	Formetanate HCL	Pyraclostrobin
Butoxycarboxim	Fuberidazole	Pyridaben
Carbaryl	Furalaxyl	Pyrimethanil
Carbendazim	Furathiocarb	Pyriproxifen
Carbetamide	Hexaconazole	Quinoxyfen
Carbofuran	Hexythiazox	Rotenone
Carbofuran-3-hydroxy	Hydramethylnon	Secbumeton
Carboxin	Imazalil	Siduron
Carfentrazone-ethyl	Imidacloprid	Simetryn
Chlorantraniliprole	Indoxacarb	Spinetoram
Chlorfluazuron	Ipconazole	Spinosad A
Chloroxuron	Iprovalicarb I	Spinosad D
Chlortoluron	Iprovalicarb II	Spirodiclofen
Clethodim I	Isocarbofos	Spirotetramat
Clofentezine	Isoprocarb	Spiroxamine I
Clothianidin	Isoproturon	Spiroxamine II
Cyazofamid	Kresoxim-methyl	Sulfentrazone
Cycluron	Linuron	Tebuconazole
Cymoxanil	Lufenuron	Tebufenozide
Cyproconazole I	Mandipropamid	Tebufenpyrad
Cyproconazole II	Mefenacet	Tebuthiuron
Cyprodinil	Mepanipyrim	Teflubenzuron
Cyromazine	Mepronil	Temephos
Desmedipham	Mesotrione	Terbumeton
Diclobutrazol	Metaflumizone	Terbutryn
Dicrotophos	Metalaxyl	Tetraconazole
Diethofencarb	Metconazole	Thiabendazole
Difenoconazole	Methabenzthiazuron	Thiacloprid
Diflubenzuron	Methamidophos	Thiamethoxam
Dimethoate	Methiocarb	Thidiazuron
Dimethomorph I	Methomyl	Thiobencarb
Dimethomorph II	Methoprotryne	Thiophanate-methyl
Dimoxystrobin	Methoxyfenozide	Triadimefon
Diniconazole	Metobromuron	Triadimenol
Dinotefuran	Metribuzin	Trichlorfon
Dioxacarb	Mevinphos I	Tricyclazole
Diuron	Mevinphos II	Trifloxystrobin
Emamectin benzoate	Mexacarbate	Triflumizole
Epoxiconazole	Monocrotophos	Triflumuron
Etaconazole	Monolinuron	Triticonazole
Ethiofencarb	Myclobutanil	Vamidothion
Ethiprole	Neburon	Zoxamide
Ethirimol	Nitenpyram	
Ethofumesate	Novaluron	

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Structural Elucidation of an Unknown Compound in the Fatty Acid Methyl Esters (FAMEs) Extract of Avocado Using APGC-HRMS

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[2]Krueger Food Laboratories Inc., Chelmsford, MA, USA

APPLICATION BENEFITS

- Generation of accurate mass measurements of low- and high-energy spectra across a wide *m/z* range aided in identification of known FAMEs and elucidation of the unknown compound.
- "Soft" ionization using APGC resulted in preservation of the molecular ion followed by fragmentation to provide more comprehensive molecular detail.
- Integrated elucidation tools in UNIFI® Discovery Toolset facilitated the search of hundreds of online databases, facilitating elemental composition and fragment match.

WATERS SOLUTIONS

Xevo® G2-XS QTof

UNIFI Scientific Information System

Atmospheric Pressure Gas Chromatography (APGC)

KEYWORDS

Avocado, fatty acids, FAMEs, HRMS, APGC, Universal Source

INTRODUCTION

Avocado is a major tropical fruit with various known health benefits when consumed as part of a balanced diet.[1] Its high lipid fraction in particular contains phytosterols, tocopherols, and most notably omega fatty acids.[1] Profiling this fatty acid content is often performed via gas chromatography (GC) following derivatization into fatty acid methyl esters (FAMEs). This is done to reduce the fatty acid polarity and neutralize the carboxyl functionality so that structural differences can be exploited via chromatography.[2] Chromatographic resolution is sufficient and reproducible enough for various FAMEs to be identified by retention time alone, and some methods will implement the use of non-mass selective detection such as flame ionization detection (FID). However the appearance of additional unexpected constituents in the derivitized lipid fraction can complicate the chromatogram.

Identification of unknowns in foodstuffs is a major challenge for food companies who need to determine whether an unknown compound may be harmful to consumers. In this application note, we describe a FAMEs analysis of an avocado sample that contained a previously unknown constituent resulting from an unexpected chromatographic peak. Through the use of high resolution mass spectrometry (HRMS) and atmospheric pressure chemical ionization following gas chromatography (APGC), the accurate mass of the molecular ion and generated fragments were used to propose an identification of the compound.

EXPERIMENTAL

GC conditions

GC system:	A7890
Column:	DB-5MS 30 m x 0.25 mm 0.25 µm (J&W)
Injection mode:	Splitless
Liner:	Gooseneck Splitless, deactivated (Restek)
Column pneumatics:	Constant flow
Column flow:	1.2 mL/min
Injector temp.:	280

GC oven temp. ramp:

Temp.	Temp. ramp	Hold time
100 °C		4 min
240 °C	5 °C/min	15 min

Total run time: 47 min

MS conditions

MS system:	Xevo G2-XS QTof
Ionization mode:	API$^+$
Acquisition mode:	MS^E
Acquisition range:	50 to 1000 *m/z*
Low collision energy:	6 eV
High collision energy:	20 to 50 eV
Source temp.:	150 °C
Interface temp.:	310 °C
Corona current:	5.0 µA
Cone voltage:	30 V
Cone gas:	110 L/hr
Auxillary gas:	300 L/hr
Make-up gas:	300 L/hr

Data management

UNIFI Scientific Information System

An avocado extract (NIST, Gaithersburg, MD, described below) was diluted 1:1000 (v/v) in heptanes and 1 µL was injected for analysis on the Waters® APGC-Xevo G2-XS QTof System. For the analysis of relatively polar compounds, as described in this work, a vial of water was placed into the enclosed ionization source in order to generate protons and yield the preferential formation of $[M+H]^+$ ions upon vaporization (Figure 1). Ionization is achieved through coronal discharge in the APGC source. Alternating states of low and elevated collision energy occurred during the chromatographic run, and the resulting accurate mass spectrum is time-aligned for precursor and fragment ions (MS^E). Both spectra were used in the identification of known FAMEs and structural elucidation of the unknown peak. A search list in the analysis method of known FAMEs was generated by importing structures obtained on ChemSpider (www.chemspider.com) for the following compounds: C18:2 methyl lineoleate, C18:1 trans-methyl elaidate, C18:1 cis-methyl oleate, C16:1 methyl palmitoleate, and C16:0 methyl palmitate. Structural import provided the UNIFI Scientific Library search list with the resulting molecular formula and exact mass, which was automatically used to generate extracted ion chromatograms, as well as propose fragment structures from the high collision energy data.

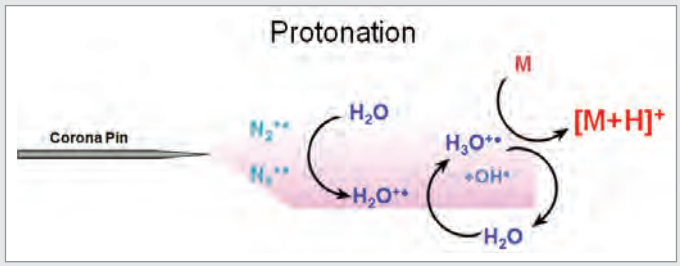


Figure 1. Schematic of protonation as it occurs with the APGC Source.

Sample description

The fat from the samples was prepared by acid hydrolysis Mojonnier extraction according to AOAC 922.06. The recovered fat was converted to FAMEs with NaOH/MeOH/BF_3 according to AOAC 969.33. The final FAME extract was diluted in heptanes.

RESULTS AND DISCUSSION

IDENTIFICATION OF KNOWN FAMEs

In order to initially assess the sample and effectiveness of the analytical approach, previously identified FAMEs (named in the EXPERIMENTAL section) from a GC-FID analysis of the sample were identified. Identification of the 5 FAMEs was based on exact mass measurements within +/- 3 ppm mass error. Figure 2 shows the results from the search list as a table (top) summarizing key pieces of data for each compound, such as retention time, mass error, and adducts observed in the spectrum. Adducts monitored included the predominant $[M+H]^+$ ion, as well as the loss of an electron (displayed as –e) which results from a charge transfer reaction, and the lesser observed $[M+N]^+$ ion. Due to the collection of data independent spectra, the full molecular character can be captured without intervention in the acquisition method; adducts are specified in the processing method which can be revised at any time following analysis. For each compound, the extracted ion chromatogram (XIC) as well as low and high energy spectra can be seen for the predominant ion observed. Structures were also used to interrogate the high energy spectrum (the bottom spectrum shown) producing likely structural fragmentation matches from the high energy spectra.

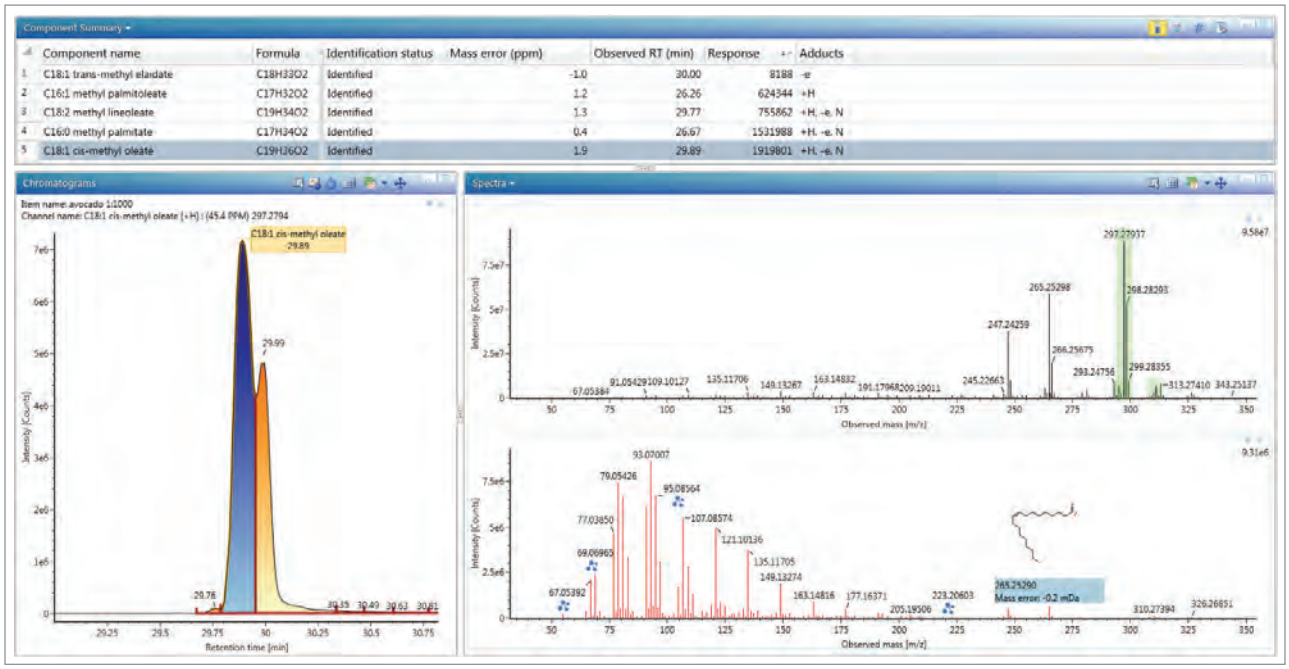


Figure 2. Summary of all identifications based on the target list, screened against the full spectrum data that were acquired. Below the table view, the XIC, low-energy spectrum, and high-energy spectrum for the selected component, C18:1 cis-methyl oleate, are shown. One of the proposed fragment structures is displayed in the spectrum view above the m/z 265.25290 peak.

Further interrogation of the fragmentation of the FAMEs in the high energy spectra yielded some key structural characteristics. Figure 3 shows the high energy spectrum for C18:1 cis-methyl oleate in an expanded and annotated view. The presence of the traditionally observed (i.e. as seen using electron ionization GC-MS) loss of the terminal ether group is seen at *m/z* 265.2531 (mass error -0.2 mDa, or -0.89 ppm). To the left of this fragment, we see a collection of alkyl chain breakages. Within these breakages, clusters spanning ~6 *m/z* indicate the process of hydride abstraction,[3] where the chain will lose two subsequent hydrogen atoms. In combining all of these fragments, the complete structure of the compound can be further confirmed.

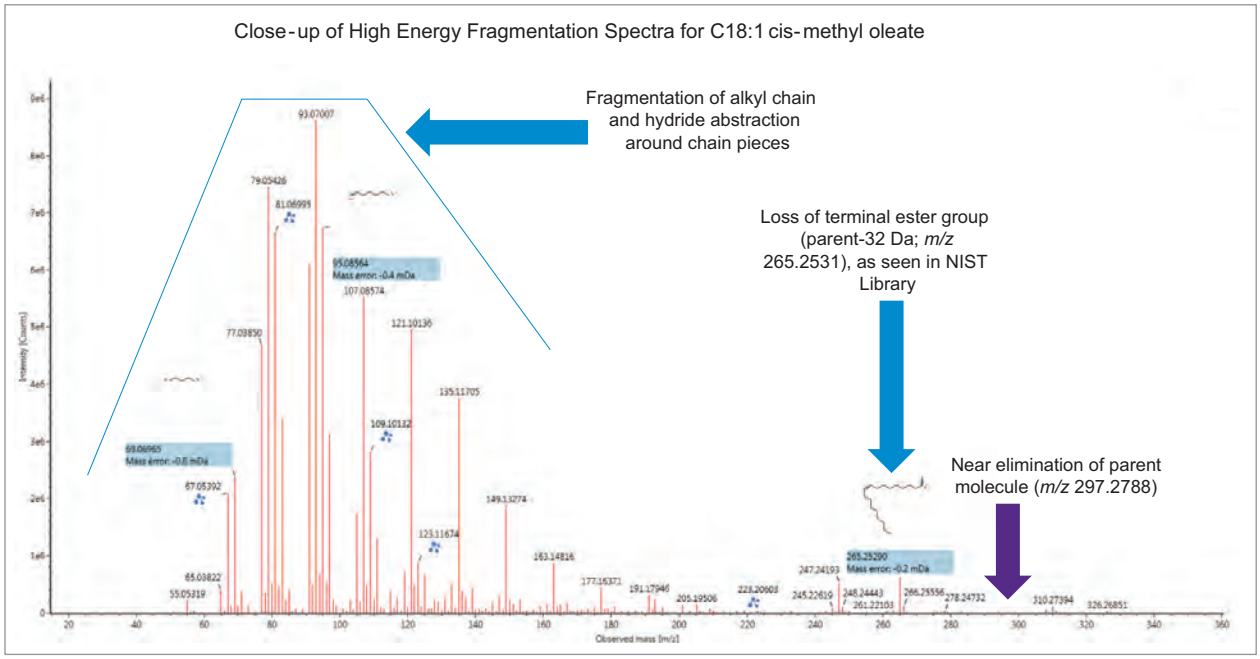


Figure 3. Accurate mass fragments observed for C18:1 cis-methyl oleate.

STRUCTURAL ELUCIDATION OF UKNOWN PEAK

Following the retention order of the known FAMEs, an unknown prominent peak was observed in the BPI (base peak intensity) chromatogram, eluting after the known FAMEs. Investigation of the spectrum under this peak resulted in a proposed molecular mass of 303.2683 *m/z*. Figure 4 shows the overlaid XIC of this mass, relative to the other FAMEs.

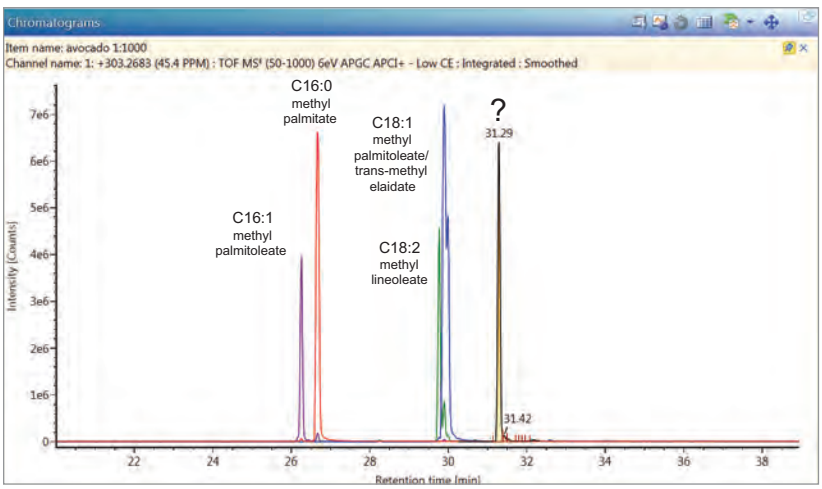


Figure 4. Known FAMEs and an unknown peak at 31.29 minutes.

Structural elucidation of the unknown peak was initialized with UNIFI by selecting the parent ion mass from the list of *m/z* from which an assignment was not made against the screening list of FAMEs (Figure 5). The candidate mass of interest (303.2683 *m/z*) was selected, and following a right-mouse click, and the option to *Elucidate* was chosen, launching the Discovery Toolset. The Discovery Toolset within UNIFI utilizes a combination of elemental composition calculation, isotope fidelity computational scoring, ChemSpider database searching, and fragment matching of high energy data.

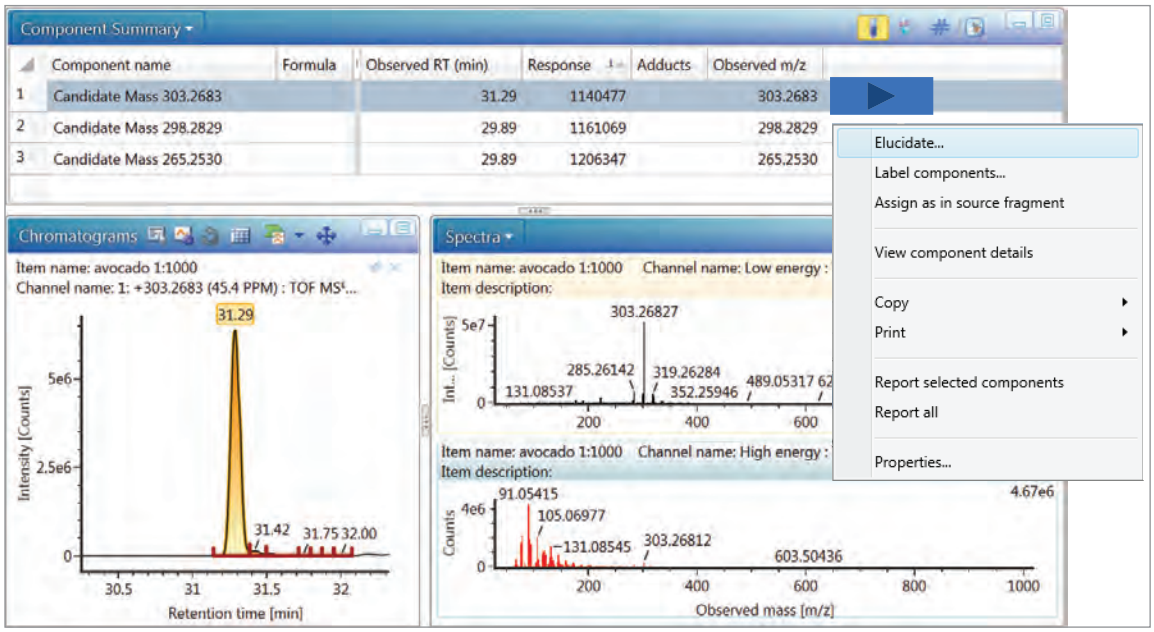


Figure 5. Candidate mass at 303.2683 m/z as it appears prior to structural elucidation. Lock mass correction has already been applied during the initial automated data processing, allowing for elucidation tools in the Discovery Toolset to be immediately applied.

For the unknown peak, the top result for a proposed identification was a 3-pentadecenylphenol species, shown in Figure 6. The proposed structure was found to have a high number of fragment formulas suggested, further supporting the likelihood of this identification. Further interrogation of the high energy spectra (Figure 7) shows similar alkyl chain breakage and hydride abstraction, as seen with the FAMEs – the proposed structure also contains an alkyl chain "tail", increasing confidence in the identification. However, the ester loss was seen for C18:1 cis-methyl oleate (Figure 3). Instead, the loss of H_2O (parent-18 *m/z*; 285.2582 *m/z*) can be seen off of the ring portion of the structure, and most importantly, the intact benzene ring itself is apparent as a fragment (Figure 7).

Online searching of this compound against FooDB (http://foodb.ca/), the world's largest compendium of food related compounds, showed that this compound has been found in the lipid portion of cashew nuts and *Gingko biloba* fruit (Figure 8).[4] Although not indicated to be in avocado explicitly, the nature of the compound identification with highly confirmed spectral matching, combined with the presence of this compound in lipid portions of other fruits supported a tentative match. Assignment of the previously unknown mass with the name from ChemSpider was performed within UNIFI by selecting *Assign* below the ChemSpider result view (Figure 6). Though not commercially available at the time of this writing, final confirmation would typically be performed through the analysis of a pure chemical standard.

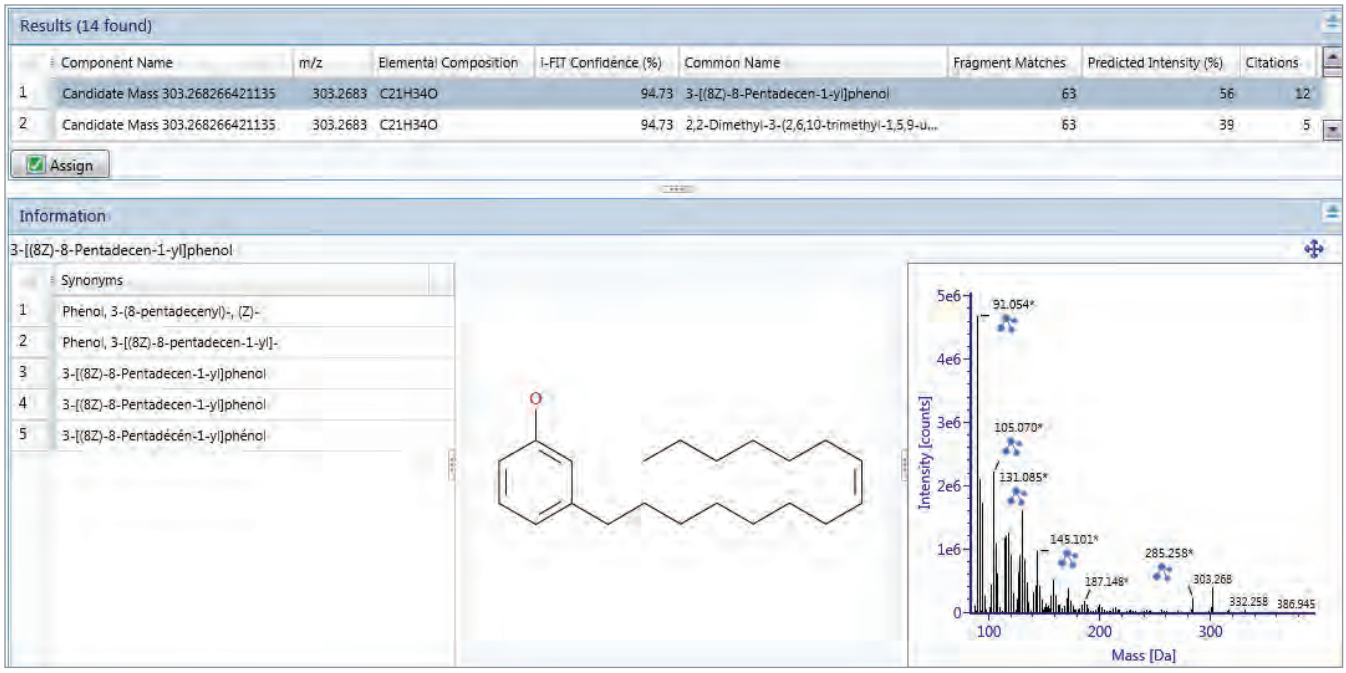


Figure 5. Candidate mass at 303.2683 m/z as it appears prior to structural elucidation. Lock mass correction has already been applied during the initial automated data processing, allowing for the elucidation tools in the Discovery Toolset to be immediately applied.

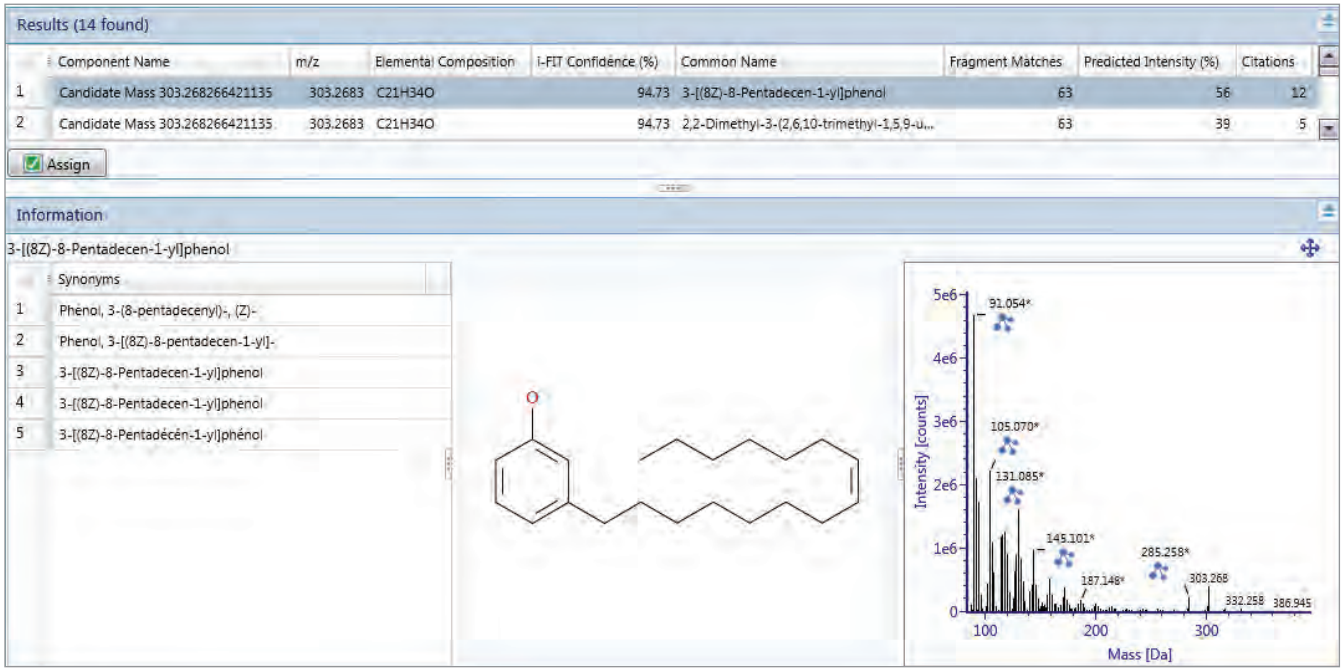


Figure 6. Discovery Toolset results for a proposed structure as found based on elemental composition of spectral peak at 303.2683, ChemSpider search of this composition, and comparison of high energy spectra to possible fragments from the structure. Blue molecule icons in spectral view (high energy spectra only) indicate a fragment match for that mass. A total of 63 proposed fragments were found for the spectra against this structure.

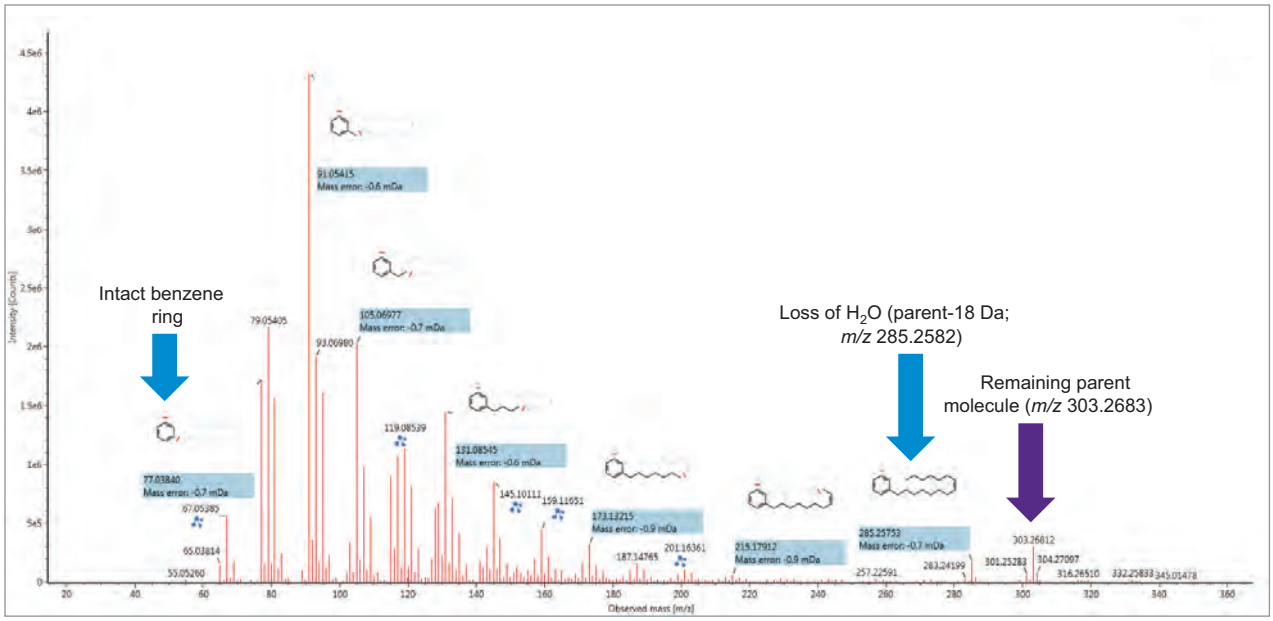
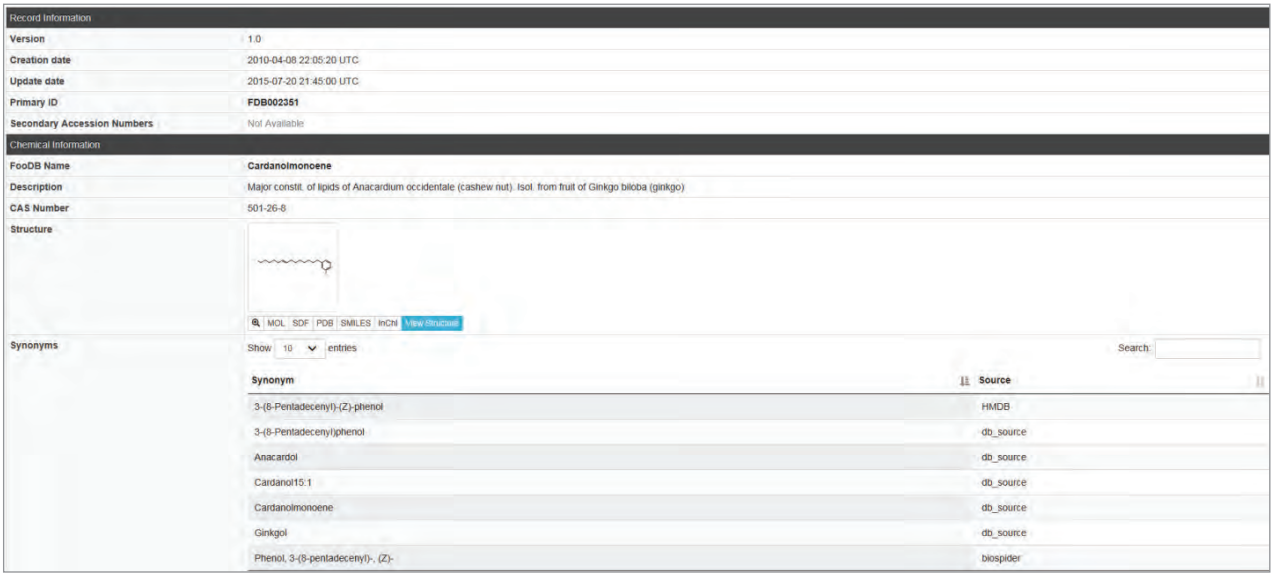


Figure 7. High energy spectrum close-up for unknown peak with proposed structure and fragment assignments.



Record Information	
Version	1.0
Creation date	2010-04-08 22:05:20 UTC
Update date	2015-07-20 21:45:00 UTC
Primary ID	FDB002351
Secondary Accession Numbers	Not Available
Chemical Information	
FooDB Name	Cardanolmonoene
Description	Major constit. of lipids of Anacardium occidentale (cashew nut). Isol. from fruit of Ginkgo biloba (ginkgo)
CAS Number	501-26-8
Structure	MOL SDF PDB SMILES InChI View Structure

Synonyms — Show 10 entries — Search:

Synonym	Source
3-(8-Pentadecenyl)-(Z)-phenol	HMDB
3-(8-Pentadecenyl)phenol	db_source
Anacardol	db_source
Cardanol15:1	db_source
Cardanolmonoene	db_source
Ginkgol	db_source
Phenol, 3-(8-pentadecenyl)-, (Z)-	biospider

Figure 8. FooDB search result for 3[(8Z)-8-pentadecen-1-yl]phenol.[4]

CONCLUSIONS

A lipid extract of avocado was found to contain an unexpected and unknown component. The extract was subsequently analyzed for both known FAMEs and for additional compounds. The use of full spectral acquisition, with alternating collision energy states (MS^E), afforded the ability to both interrogate the data for known compounds using fragment matching, as well as searching for any additional masses of interest. Elucidation of the unknowns was made possible by interrogating the high quality accurate mass data into the easily accessible UNIFI Discovery Toolkit. A proposed identification was made for the unknown peak based on the use of the spectra and assigned within the analysis. Future profiling of avocado samples can be performed with this additional compound in mind, and any future unknown peaks can be elucidated using this same basic approach.

Acknowledgment

The authors would like to thank Melissa Phillips of NIST for providing the avocado powder samples and for her thoughtful review of this work.

References

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3. www.ms-textbook.com/1st/downloads/chap7.pdf
4. http://foodb.ca/compounds/FDB002351

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Demonstration of Collision Cross Section (CCS) Value Conservation Across LC and GC Analyses

Lauren Mullin, Gareth Cleland, and Mike McCullagh

GOAL

To demonstrate the conservation of CCS values for 73 pesticides introduced under both GC and LC conditions.

BACKGROUND

Contaminant identification in food and environmental matrices using both GC and LC-MS techniques are widely implemented, although challenges with matrix effects, false detections, and reproducibility of ion ratios exist. In this technology brief we demonstrate the application of travelling wave ion mobility spectrometry (TW-IMS) coupled to a quadrupole time-of-flight (QTof) MS to generate a robust and unique additional identification point for contaminant analysis. The determination of a collision cross section (CCS) of an ion can be extrapolated from the observed drift time as the ion passes through the drift cell. To demonstrate the robust and precise nature of CCS values, a suite of pesticides were analyzed under both GC and LC conditions, and the CCS values obtained compared.

THE SOLUTION

A comparison of 73 GC and LC amenable pesticides that had been injected as solvent standards five times each during both the LC and GC analyses found CCS values that were strongly correlated to one another (Figure 1).

CCS values are robust and precise values associated with physical properties of an ion, and are conserved regardless of chromatographic technique implemented.

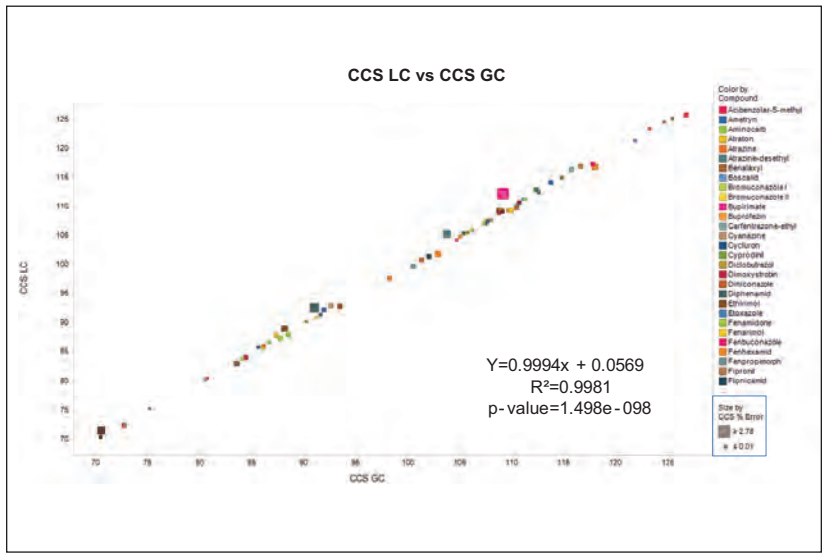

Figure 1. CCS value regression analysis of GC and LC analyses, indicating conversely to the RT comparisons, a strong correlation between CCS measurements obtained under GC and LC analyses for the 73 pesticides.

A regression analysis of the LC and GC results for CCS produced an R^2 value of 0.998, indicating a very high degree of correlation. Moreover, the CCS values across the five injections in each technique showed minimal deviation.

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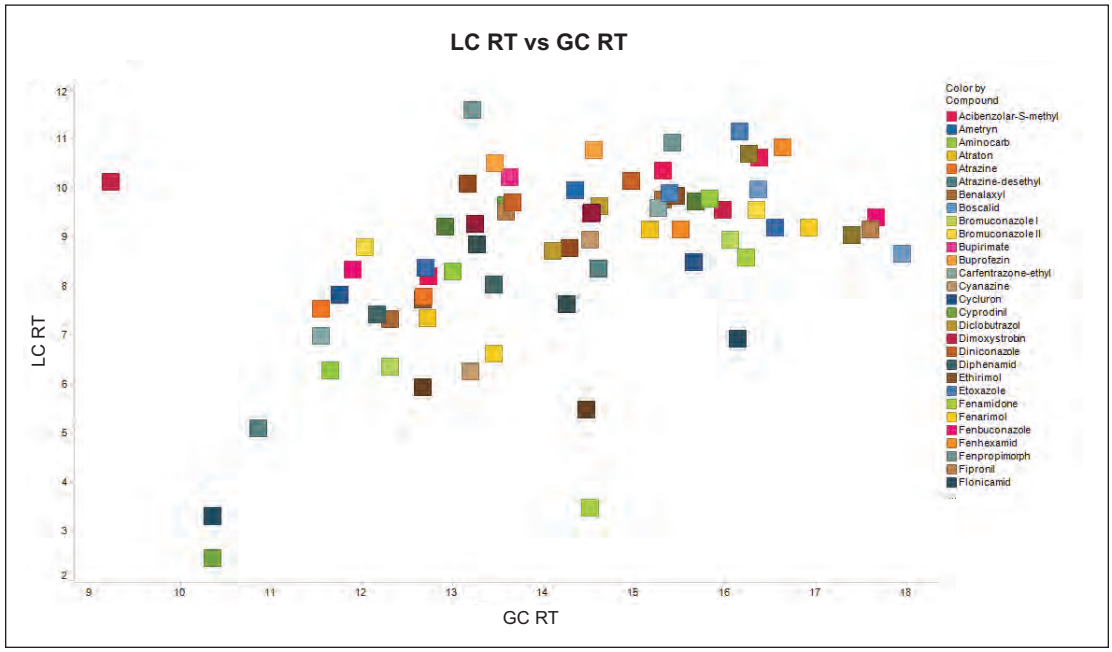

Figure 2. Retention time regression analysis of GC and LC analyses, indicating no correlation between the two approaches for 73 pesticides analyzed.

When retention times under the two techniques were compared, as would be expected, no correlation was observed (Figure 2). From these results, it could be seen that CCS values represented a unique property of the ions generated that was well conserved, regardless of that analytes introduction into the travelling wave ion mobility MS system. These results support the use of CCS values, in addition to mass and characteristic product ions, for compound identification.

SUMMARY

CCS values are unique properties of an ion in the gas phase that are retained regardless of the method used to introduce the analytes into the MS system. Using CCS values in contaminant screening offers a unique point of identification, and allows flexibility around retention time tolerances applied for identification purposes.

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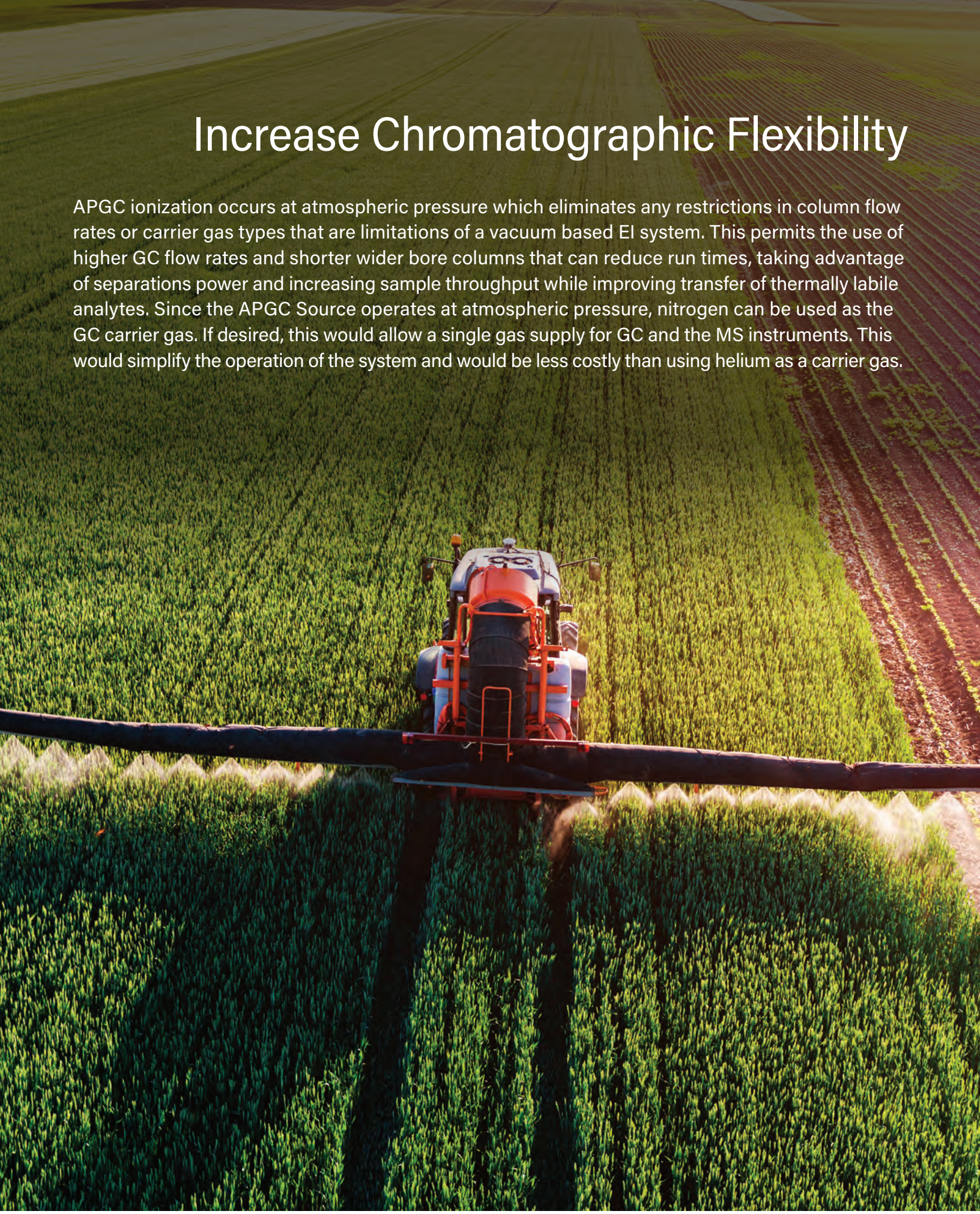
Increase Chromatographic Flexibility

APGC ionization occurs at atmospheric pressure which eliminates any restrictions in column flow rates or carrier gas types that are limitations of a vacuum based EI system. This permits the use of higher GC flow rates and shorter wider bore columns that can reduce run times, taking advantage of separations power and increasing sample throughput while improving transfer of thermally labile analytes. Since the APGC Source operates at atmospheric pressure, nitrogen can be used as the GC carrier gas. If desired, this would allow a single gas supply for GC and the MS instruments. This would simplify the operation of the system and would be less costly than using helium as a carrier gas.



Harnessing the Efficiency of Nitrogen Carrier Gas with the Atmospheric Pressure Gas Chromatography (APGC) Source

Lauren Mullin and Adam Ladak
Waters Corporation, Milford, MA, USA

Implementing nitrogen (N_2) as the carrier gas for GC experiments is a cost-effective approach for atmospheric pressure MS sources, while maintaining critical separations and chromatographic performance.

GOAL

To highlight the ease of implementation and efficient performance of N_2 as a replacement carrier gas for helium (He) in GC separations interfaced with the atmospheric pressure gas chromatography (APGC) MS source.

BACKGROUND

Helium (He) is the most commonly used carrier gas in gas chromatography (GC) applications. However, the finite nature of reserves has resulted in periodic price increases and concern regarding availability.[1] Nitrogen (N_2) is a more affordable and readily available option that has historically been less utilized as a GC carrier gas. Reasons for this are that N_2 has lower diffusivity than He or hydrogen and often requires longer run times to achieve similar separations. In this technology brief we show GC coupled with an atmospheric pressure ionization mass spectrometry, which utilizes N_2 for both ionization and make-up flow. This allows a single gas source to be used for chromatographic separation as well as for ionization. Following automated method transfer calculations available within

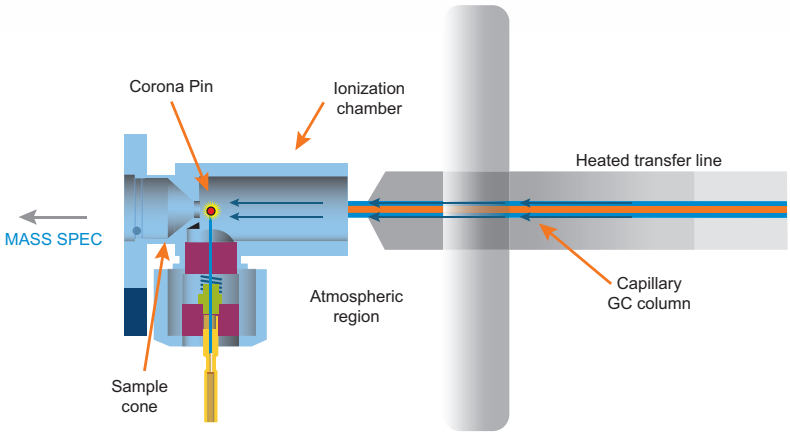


Figure 1. MS and GC atmospheric source interface. Nitrogen is supplied to the source from the heated transfer line, as well as cone and auxiliary gas supplies.

Waters® UNIFI® Software, efficient and comparable chromatography using N_2 carrier gas was achieved for pesticides, polybrominated diphenyl ethers (PBDEs), polychlorinated biphenyls (PCBs), and chlorinated dioxin/furans (PCDD/Fs). Unlike electron ionization, atmospheric pressure ionization remains robust during the introduction of N_2. Also, a higher range of column flows can be used in the APGC source compared to tradition vacuum GC, and more flexibility with regards to method translation such that optimized carrier gas linear velocities could be achieved for N_2.

THE SOLUTION

Experiments were performed on a Xevo® G2-XS QTof with APGC (Figure 1). Ionization was performed using atmospheric pressure chemical ionization, such that protonation (resulting in the [M+H]+ ion) or charge transfer (resulting in the M•+ ion) reactions occurred. The GC method for pesticides is described in Table 1a and 1b, and for the analysis of PBDEs, PCBs, and PCDD/Fs in Table 2a and 2b. Methods were revised using an automated calculator for method transfer to arrive at optimum values for N_2 as a carrier gas, resulting in comparable separations to those achieved using He. Figure 2 shows the calculator as available in Waters UNIFI Software for GC Instrument Control. When using the Speed Gain option of the calculator, the increase of the Outlet Flow rate (mL/min) and resulting Average Velocity (cm/s) resulted in a shortened method when N_2 carrier gas was used as compared to He.

For the PBDEs, PCBs, and PCDD/Fs method, the separation of the closely eluting hexachlorodibenzo-p-furan (HxCDF) congeners 1,2,3,4,7,8– and 1,2,3,6,7,8-HxCDF is shown in Figure 3. A 25% valley is retained for the co-eluting 1,2,3,4,7,8– and 1,2,3,6,7,8-HxCDF congeners using both N_2 and He separations, as specified in the EPA 1613 analytical guidance.[2] In addition to both analytical assays retaining critical separations when using N_2 carrier gas, a faster run time was achieved. The reduced lifetime of the GC filaments traditionally caused during electron ionization when using N_2 carrier gas is eliminated by the use of an atmospheric pressure chemical ionization MS source. Thus far no negative implications from the use of N_2 as a carrier gas have been observed or are expected.

1a

Parameter	Value
Column	DB 5MS 30 m x 0.25 mm, 0.25 µm (J&W)
Carrier gas	Helium or Nitrogen
Injection mode	Splitless
Inlet liner	Single taper splitless, deactivated (Restek)
Column pneumatics	Constant flow
Column flow (mL/min)	1.2 or 1.31
Inlet temperature (°C)	280

1b

Temperature	Temperature ramp (°C/min)	Hold time (min)
40		1.00 or 0.92
320	27 or 29.49	2.63 or 2.41

Table 1a and 1b. GC method for pesticide analysis.

2a

Parameter	Value
Column	Rxi 5Sil 60 m x 0.25 mm, 0.25 µm (Restek)
Carrier gas	Helium or Nitrogen
Injection mode	Splitless
Inlet liner	Single taper splitless, deactivated (Restek)
Column pneumatics	Constant flow
Column flow (mL/min)	1.0 or 1.09
Inlet temperature (°C)	280

2b

Temperature	Temperature ramp (°C/min)	Hold time (min)
120		2.00 or 1.84
200	35 or 38.05	0.00
215	5 or 5.43	18.00 or 16.57
235	5 or 5.43	7.00 or 6.44
290	5 or 5.43	5.00 or 4.60
325	7 or 7.93	2.30 or 2.12

Table 2a and 2b. GC method for PBDEs, PCBs, and PCDD/Fs analysis.

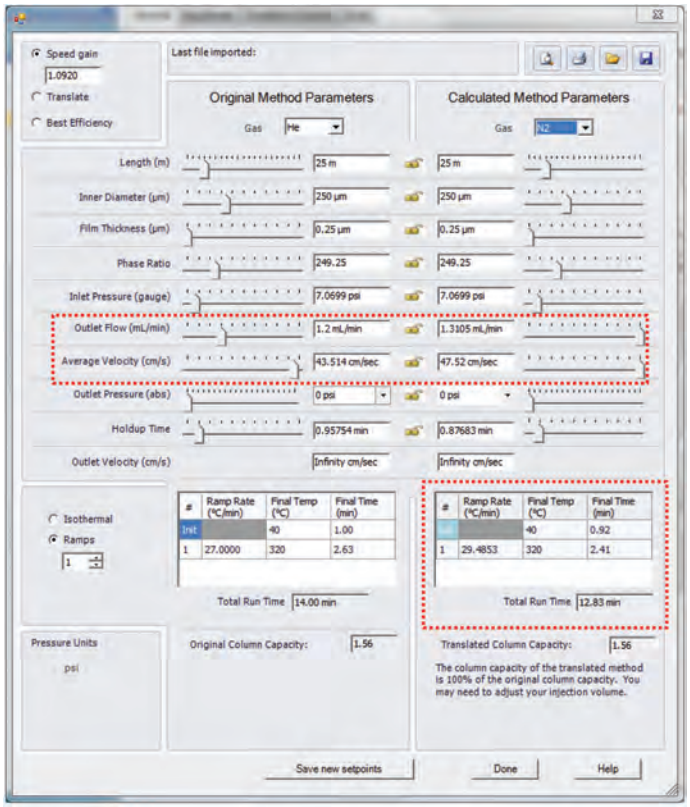


Figure 2. Method converter within UNIFI instrument control updates parameters based on the physical properties of N_2 carrier gas.

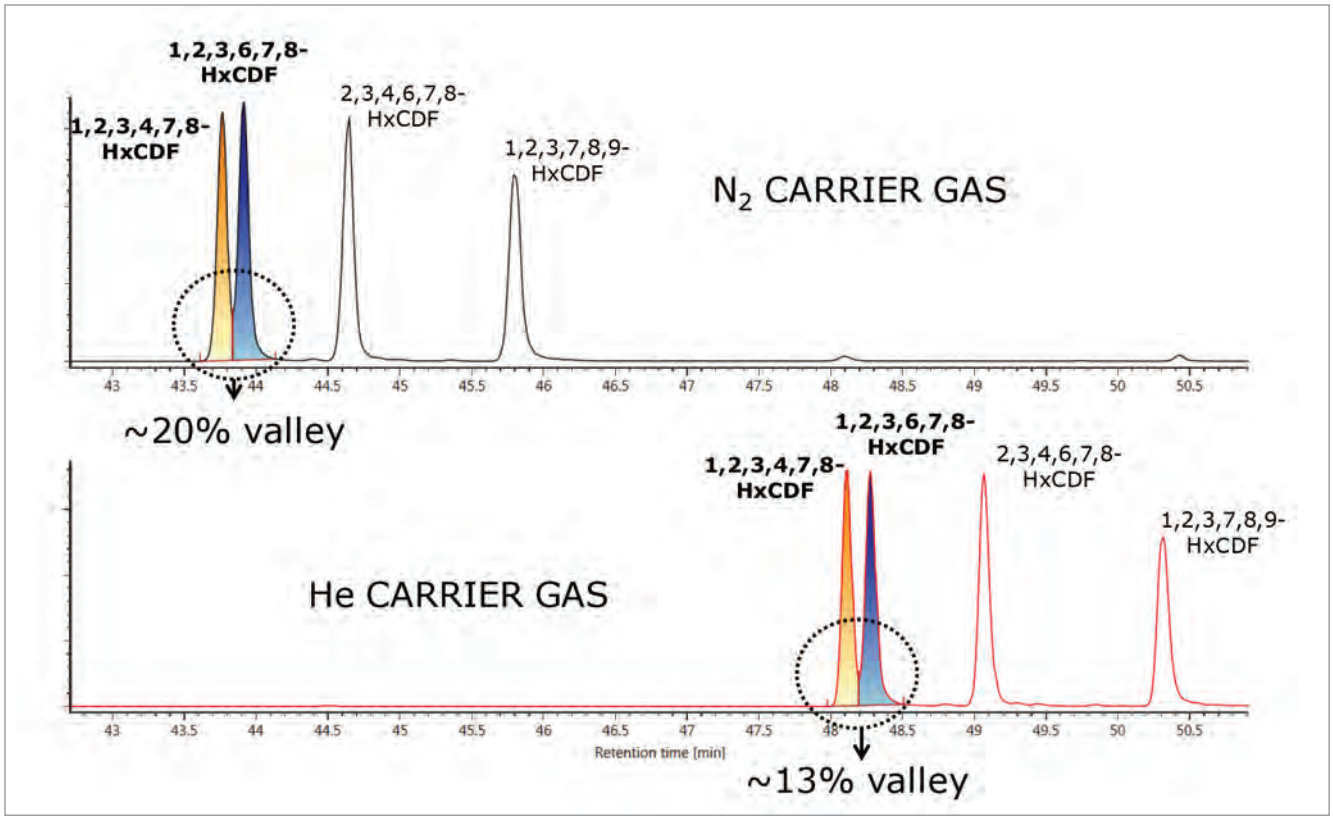


Figure 3. HxCDFs separation contrasting N_2 and He carrier gases, where both gases ensure the method requirement of 25% valley separation is achieved for the congeners specified.

SUMMARY

Nitrogen can be used as a single gas source for both GC carrier and MS source gas flows using the APGC source, and presents a viable replacement option to Helium carrier gas. N_2 optimized GC temperature program methods require less time without sacrificing critical separations for the pesticides and POPs analyses studied.

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Enhancing MRM Experiments in GC/MS/MS Using APGC

Tania Portolés,[1] Laura Cherta,[1] Joaquim Beltran,[1] Antonietta Gledhill,[2] Félix Hernández[1]
[1]University Jaume I, Castellón, Spain
[2]Waters Corporation, Manchester, UK

APPLICATION BENEFITS

- Increased sensitivity and selectivity for GC amenable pesticides.
- Able to analyze both GC and LC compounds on one MS platform.
- Routine system that is suitable for all food testing laboratories.

WATERS SOLUTIONS

APGC

Xevo® TQ-S

ACQUITY UPLC® System

MassLynx® MS Software

TargetLynx™ Application Manager

KEY WORDS

Atmospheric pressure gas chromatography, APGC, gas chromatography, GC, tandem mass spectrometry, pesticides, proton transfer

INTRODUCTION

Gas and liquid chromatography (GC and LC) coupled to mass spectrometry (MS) are the techniques of choice in pesticide residue analysis (PRA) for a wide variety of sample matrices.

GC–MS is commonly applied for non-polar, volatile, and thermally stable compounds and ionization in GC–MS normally occurs under vacuum conditions using either electron ionization (EI) or chemical ionization (CI).

EI is typically employed in the wide majority of GC–MS applications, and is a robust and highly reproducible technique. It is well known that this ionization process produces extensive fragmentation of some molecules, leading in many cases to the absence of the molecular ion $[M]^{\bullet+}$ in EI spectra. CI induces softer ionization and this can lead to better selectivity and sensitivity for some analytes, as well as fewer matrix interferences. But CI ionization is only applicable to specific chemical classes[1-3] and sensitivity is limited.

Many recent methods have been reported in PRA based on EI-GC-tandem quadrupole mass spectrometry, due to its better performance for quantitative multi-residue analysis compared to EI-GC-single quadrupole mass spectrometry. The selection of adequate precursor and product ions enhance selectivity and sensitivity, minimizing or even eliminating matrix interferences. In this way, very low detection limits can be achieved.

However, the extensive fragmentation due to the high energy transferred to the molecules during the ionization process produces little or no molecular ions for many pesticides, as for example organochlorine (OC) pesticides, organophosphorus (OP), pyrethroids, and chloroacetanilides.[4-6] Also, compounds belonging to the same chemical family can show similar EI spectra; so the use of common ions/transitions can complicate the identification and quantification processes, especially if analytes are co-eluting. When the molecular ion is absent or has very low abundance, it is necessary to select a (abundant) fragment ion as precursor. In addition to the loss of sensitivity, the specificity of the method can be also affected.

Atmospheric pressure ionization in GC–MS was first introduced by Horning *et al.*[7] and the technique offers attractive analytical capabilities in GC–MS analysis. APGC is a technique that operates at atmospheric pressure

EXPERIMENTAL

Sample preparation

Different fruits and vegetable (apple, orange, tomato and carrot) fortified extracts were used and prepared using the QuEChERS extraction approach (AOAC Official method)[9]

GC conditions

GC system:	7890A
Injector:	Splitless mode
Injection:	1 µL at 280 °C
Column:	DB-5MS (J&W Scientific, USA)
	30 m I.D. 0.25 mm df 0.25 µm
Column temp.:	70 °C (1 min), 25 °C/min to 150 °C
	10 °C/min to 300 °C (3 min)
Transfer line temp.:	310 °C
Carrier gas flow:	2 mL/min (Helium)
Auxiliary gas:	250 L/h (Nitrogen)
Make-up gas:	320 mL/min (Nitrogen)

MS conditions

MS system:	Xevo TQ-S
APCI corona pin:	1.8 µA
Cone voltage:	25 V
Source offset:	50 V
Cone gas:	170 L/h (Nitrogen)
Data management:	MassLynx Software v4.1, with Targetlynx Application Manager

and uses a soft ionization process which can provide abundant molecular ions that in most cases to be used as precursor ions in MRM experiments for multi-residue GC–MS/MS analysis.

Waters® Xevo TQ-S tandem quadrupole mass spectrometer is a highly sensitive and robust instrument that is compatible with both the ACQUITY UPLC System and APGC.

The aim of the project[8] was to evaluate the potential of APGC–Xevo TQ-S for the quantitative analysis of pesticides in foodstuffs: 25 pesticides with varying chemical properties have been selected and the potential advantages of APGC were evaluated in comparison with EI ionization.

RT (min)	Compounds	Cone voltage (V)	MRMs	Collision energy (eV)
4.70	Dichlorvos	10	Q 221 > 145	10
			q1 221 > 113	30
			q2 221 > 127	20
5.97	Mevinphos	30	Q 225 > 127	10
			q1 225 > 113	30
			q2 225 > 193	10
6.96	Molinate	20	Q 188 > 126	10
			q1 188 > 98	20
			q2 188 > 160	10
8.00	Dicrotophos	40	Q 238 > 112	10
			q1 238 > 127	20
			q2 238 > 193	10
8.24	Monocrotophos	20	Q 224 > 127	10
			q1 224 > 113	30
			q2 224 > 193	10
8.95	Terbufos	10	Q 187 > 131	10
			q1 187 > 97	20
			q2 187 > 113	20
9.80	Phosphamidon	40	Q 300 > 127	20
			q1 300 > 174	10
			q2 300 > 227	10
9.76	Endosulfan ether	30	Q 341 > 217	30
			q1 341 > 170	30
			q2 341 > 205	20
9.94	Chlorpyriphos methyl	40	Q 322 > 125	30
			q1 322 > 212	30
			q2 322 > 290	20
10.77	Chlorpyriphos	20	Q 350 > 198	20
			q1 350 > 294	10
			q2 350 > 322	10
10.85	Aldrin	30	Q 363 > 159	20
			q1 363 > 215	20
			q2 363 > 327	10
11.39	Isodrin	30	Q 363 > 159	20
			q1 363 > 215	20
			q2 363 > 327	10
11.56	Chlorfenvinphos	30	Q 359 > 170	30
			q1 359 > 99	10
			q2 359 > 205	20
11.56	Oxychlordane	10	Q 421 > 151	20
			q1 421 > 115	20
			q2 421 > 285	30
11.56	Heptachlor epox B	20	Q 387 > 217	30
			q1 387 > 251	20
			q2 387 > 252	10
12.23	Endosulfan I	10	Q 405 > 323	10
			q1 405 > 205	20
			q2 405 > 217	30
12.72	Buprofezin	30	Q 306 > 106	20
			q1 306 > 203	10
			q2 306 > 250	10
12.73	Dieldrin	20	Q 379 > 325	10
			q1 379 > 147	20
			q2 379 > 261	20
13.10	Endrin	30	Q 379 > 343	10
			q1 379 > 243	20
			q2 379 > 244	20
13.36	Ethion	10	Q 385 > 125	20
			q1 385 > 97	10
			q2 385 > 143	30
14.01	Endosulfan sulfate	10	Q 323 > 217	30
			q1 323 > 252	20
			q2 323 > 287	10
15.63	Azinphos methyl	20	Q 261 > 125	20
			q1 261 > 167	10
			q2 261 > 183	10
15.66	Pyriproxyfen	10	Q 322 > 185	20
			q1 322 > 129	30
			q2 322 > 227	10
16.04	Fenarimol	40	Q 331 > 268	20
			q1 331 > 139	30
			q2 331 > 259	20
16.17	Azinphos ethyl	20	Q 289 > 137	20
			q1 289 > 233	10
			q2 289 > 261	10

Table 1. Retention times and APGC MRM transitions for the compounds.

RESULTS AND DISCUSSION

In order to evaluate the capabilities of APGC, 25 pesticides were selected on the basis of their mass spectral behavior in the EI source and those that are commonly analyzed using EI-GC-MS/ MS in order to meet food safety regulatory guidelines.

MRM selectivity and specificity

There are many examples of pesticides known to be analytically problematic when using GC–MS/MS methods under EI conditions, due to EI being a highly energetic ionization mechanism. An example of this can be seen in Figure 1, where we show a spectral comparison for EI and APGC.

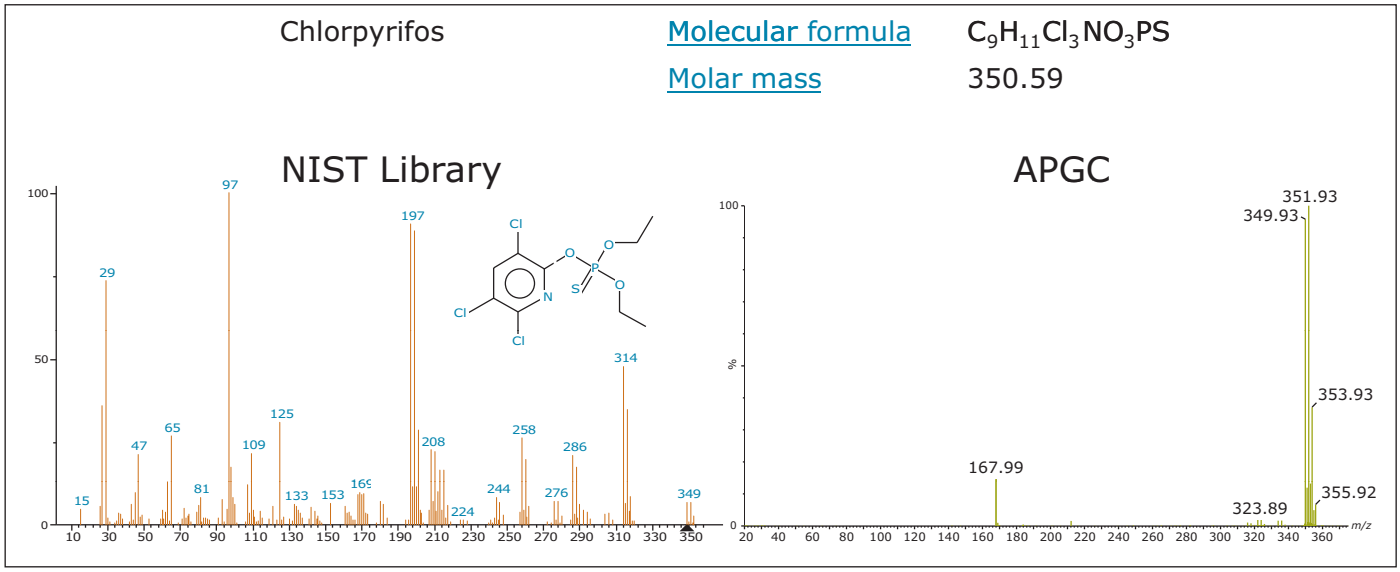

Figure 1. Spectral comparison for chlorpyrifos: EI (left) and APGC (right). The EI spectrum (taken from NIST) is highly fragmented whereas the APGC spectrum shows less fragmentation and provides the $[M+H]^+$ as the base peak.

When a compound undergoes a high degree of fragmentation the resulting MRM is often not specific, and this can affect the selectivity and may lead to a false positive identification.

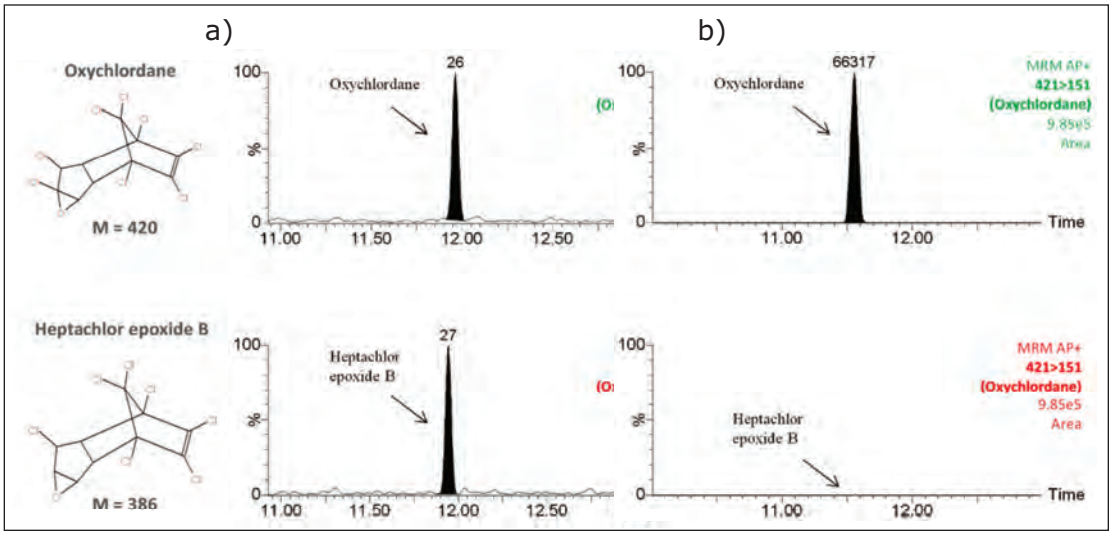

Figure 2. Chromatograms obtained for the selected MRM transitions acquired for heptachlor epoxide B (top), and oxychlordane (bottom) under (a) EI (100 ppb), and (b) APGC (10 ppb).

Figure 2 shows an example of the potential for false positive identification using EI for heptachlor epoxide B and oxychlordane. Pure standards of each compound were analyzed using the typical EI transition for oxychlordane (235 > 141, Figure 2a). Due to the structural similarity of the compounds, the heptachlor epoxide B standard was detected at the same retention time with the same transition as oxychlordane. However, using APGC and a more selective transition (421 > 151: 421 corresponds to the $[M+H]^+$ ion of oxychlordane in APGC), allowed the determination of oxychlordane without interference from heptachlor epoxide B.

Sensitivity

The Xevo TQ-S is a highly sensitive and robust tandem quadrupole mass spectrometer when coupled to an ACQUITY UPLC System,[11-13] and data from this work (and other work carried out) suggests that this is also the case when using APGC.

Using the MRM method developed the instrumental sensitivity and linearity was evaluated, and linearity was studied by injecting solvent standards in duplicate in the range 0.1 to 100 ppb (corresponding to 0.1 to 100 µg/kg in sample). The majority of compounds showed a linear response from 0.1 to 100 ppb with $r^2 > 0.99$ (data not shown).

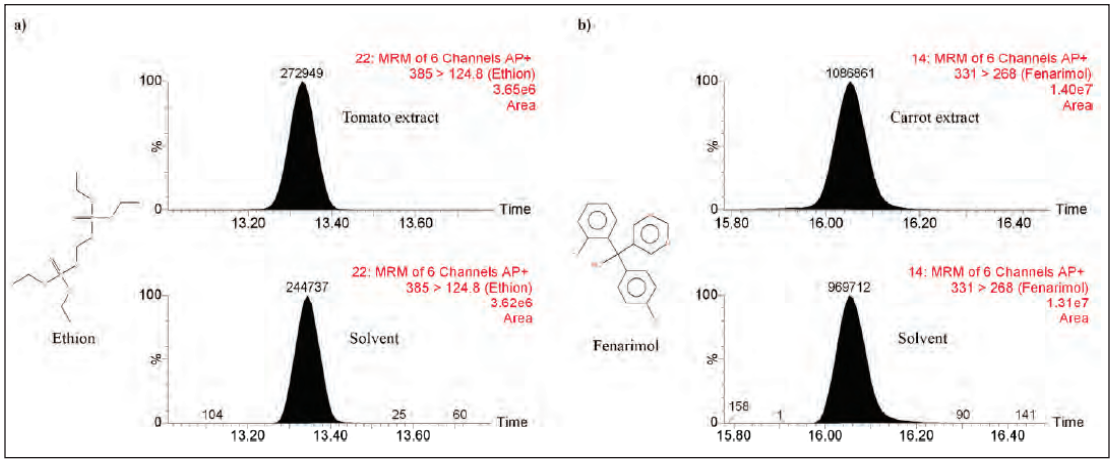

Figure 3. MRM chromatograms for (a) ethion in spiked tomato extract (10 ppb) and in solvent standard (10 ppb), and (b) fenarimol in spiked carrot extract (10 ppb) and in solvent standard (10 ppb),

Fortified extracts of the compounds in various fruits and vegetables were prepared and Figure 3 illustrates representative examples for ethion and fenarimol in tomato and carrot, respectively. The sensitivity of APGC coupled to the Xevo TQ-S led to an estimation of LOQ (calculated as 10 x S/N) between 0.02 and 2 ppb depending on the analyte and matrix under study. Responses for the spiked samples (apple, orange, tomato, and carrot) were compared to those of standards in solvent. No significant matrix effects were observed, although a little enhancement of the response of some analytes in the sample extracts occurred.

The sensitivity of the Xevo TQ-S currently exceeds existing regulations related to pesticide residue analysis for both GC and LC-amenable pesticides. The additional sensitivity enables analysts to dilute samples, significantly reducing matrix interferences and minimize the amount injected on-column. This in turn has major benefits for system cleanliness and reduces instrument maintenance requirements.

CONCLUSIONS

For many food testing labs, both GC and LC are important techniques that enable a broad range of compounds to be analysed. With the introduction of APGC it is now possible to have access to both types of separation on the Xevo TQ-S.

The Xevo TQ-S used in conjunction with an ACQUITY UPLC has been reported to be a very sensitive and robust tandem quadrupole mass spectrometer, and preliminary work using the same MS system, but with APGC also indicates that high sensitivity can also be achieved for GC-amenable compounds.

As shown by Portoles *et al.*,[8] pesticides that demonstrate high fragmentation under EI can be easily analyzed using APGC, where the $[M+H]^+$ became the base peak of the spectrum for the majority of compounds. The soft and reproducible ionization process results in an increase in sensitivity (compared to other GC-MS/MS methods) in the subsequent APGC–Xevo TQ-S method.

The Xevo TQ-S in combination with both the ACQUITY UPLC and APGC makes it an attractive MS for routine food testing laboratories due to its chromatographic versatility, the ability to choose more selective MRMs, and the system sensitivity.

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Identification of Non-Intentionally Added Substances (NIAS) in Food Contact Materials Using APGC-Xevo G2-XS QTof and UNIFI Software

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Waters Corporation, Wilmslow, UK

APPLICATION BENEFITS

- Reliable GC-MS method for screening and structural elucidation of non-intentionally added substances (NIAS) in food packaging materials
- Atmospheric Pressure Gas Chromatography (APGC) is a soft ionization technique that produces lower levels of fragmentation than EI, enabling improved detection of challenging molecular ions and the avoidance of possible erroneous identification
- UNIFI® Software provides customized workflows to streamline and simplify elucidation of unknown compounds from food packaging

WATERS SOLUTIONS

Atmospheric Pressure Gas Chromatography (APGC)

Xevo® G2-XS QTof Mass Spectrometer

UNIFI Scientific Information System

KEYWORDS

High resolution mass spectrometry, HRMS, food contact materials, leachables, non-targeted analysis, GC-MS, migration, componentization, elucidation, electron ionization, EI, MS^E

INTRODUCTION

Food comes into contact with many materials and articles during its production, processing, storage, preparation, and serving before its eventual consumption. Such materials and articles are called food contact materials (FCMs). Recently, concern about the wholesomeness and safety of food products has increased dramatically. Most of the concern usually focuses on food additives, monomers, oligomers, and non-intentionally added substances (NIAS). A non-intentionally added substance is defined in the European Union (EU) Regulation No 10/2011 as "an impurity in the substances used or a reaction intermediate formed during the production process or a decomposition or reaction product."[1,2] FCMs can, therefore, be considered materials containing a complex mixture of substances of known or unknown identity/origin. Depending on their physico-chemical properties and chemical composition, FCMs may transfer some constituents, both Intentionally Added Substances (IAS) and NIAS to foodstuffs. This mass transfer phenomenon is called migration, and may lead to high exposure to certain chemicals, which might cause a risk for human health.[3] Therefore, migration must be evaluated and controlled. Furthermore, where migration brings about an unacceptable change in the composition of food or brings about deterioration in the organoleptic properties of the food, it must be avoided.[4]

Before performing a migration study, a screening analysis of the packaging material is required to identify the chemicals that are present in the material and those that are more likely to migrate. This initial step usually involves a strong extraction of the material with an organic solvent or a mixture of solvents. The extract is then injected via LC-MS and/or GC-MS for non-targeted screening analysis of non-volatiles, and volatiles/semi-volatiles, respectively. With respect to semi-volatiles and volatiles analyses, a GC coupled to a quadrupole mass spectrometer equipped with electron ionization using 70 eV in the ion source is typically employed, since it allows the analyst to use scientific libraries, such as NIST, for comparing acquired spectra with those in the library. However, the identification process becomes almost impossible when the compound of interest is not listed in the library, or when the sensitivity of the quadrupole MS is not sufficient for reliable mass confirmation. Waters® Atmospheric Pressure Gas Chromatography (APGC) and Xevo G2-XS quadrupole time-of-flight (QTof) mass spectrometer, along with the UNIFI Scientific Information System provides an advantageous solution to overcome this hurdle.

APGC is a soft ionization technique which enables molecular ions to be observed.[5] Furthermore, the use of high resolution mass spectrometry (HRMS) and its proprietary MS^E mode[6] allows analysts to simultaneously acquire data containing the accurate mass of precursor and fragment ions. Finally, UNIFI's Discovery tool utilizes accurate mass and fragment information to facilitate the decision-making process towards the eventual identification of unknown compounds. To illustrate the benefits of APGC-QTof against electron ionization (EI)-single quadrupole MS, a polymer extracted sample was injected into both systems using the same chromatographic conditions in order to perform a comparative study of the chromatographic traces.

EXPERIMENTAL

Sample preparation

The sample, consisting of novel starch-based biopolymer pellets (0.5 g), was extracted three times with 2.5 mL of methanol in an ultrasonic bath for 1 hour at 40 °C. The total extraction solution (7.5 mL) was concentrated to 1 mL under a gentle nitrogen flow at room temperature before injection.

GC conditions

GC system:	Agilent 7890A
Autosampler:	7683B
Column:	DB-5MS, 30 m x 0.25 mm I.D. x 0.25 µm film thickness
Injection type:	1 µL pulsed splitless
Pulse time:	1.2 min
Pulsed pressure:	32 psi
Inlet temp.:	250 °C
Carrier gas:	He at 1 mL/min
Oven temp. program:	50 °C held for 2 min, ramp 50 to 300 °C 10 °C/min, 300 °C held for 10 min

MS conditions

MS system:	Xevo G2-XS QTof, sensitivity mode
Scan range:	50 to 650 *m/z*
Corona current:	2.2 µA
Sample cone:	30 V
Source temp.:	150 °C
Cone gas flow:	140 L/h
Auxiliary gas flow:	225 L/h
Make-up gas:	N_2 300 mL/min at 300 °C
Collision ramp for MS^E:	20 to 30 eV
Lock mass:	Persistent column bleed peak, 207.0324 *m/z*
EI solvent delay:	4 min
Data management:	UNIFI Scientific Information System

RESULTS AND DISCUSSION

Data were acquired using dry conditions, where nitrogen charge transfer occurs and gives rise to the (radical cation) molecular ion $M^{+\cdot}$ information.

First, Total Ion Current (TIC) chromatograms acquired with EI (using an Agilent 6890N gas chromatograph with a MS 5975B detector) and APGC were compared. It is notable that APGC showed a higher number of peaks (Figure 1). This is due to the higher sensitivity of the QTof versus the single quadrupole, and to the intrinsic characteristics of the two different types of ionization techniques.

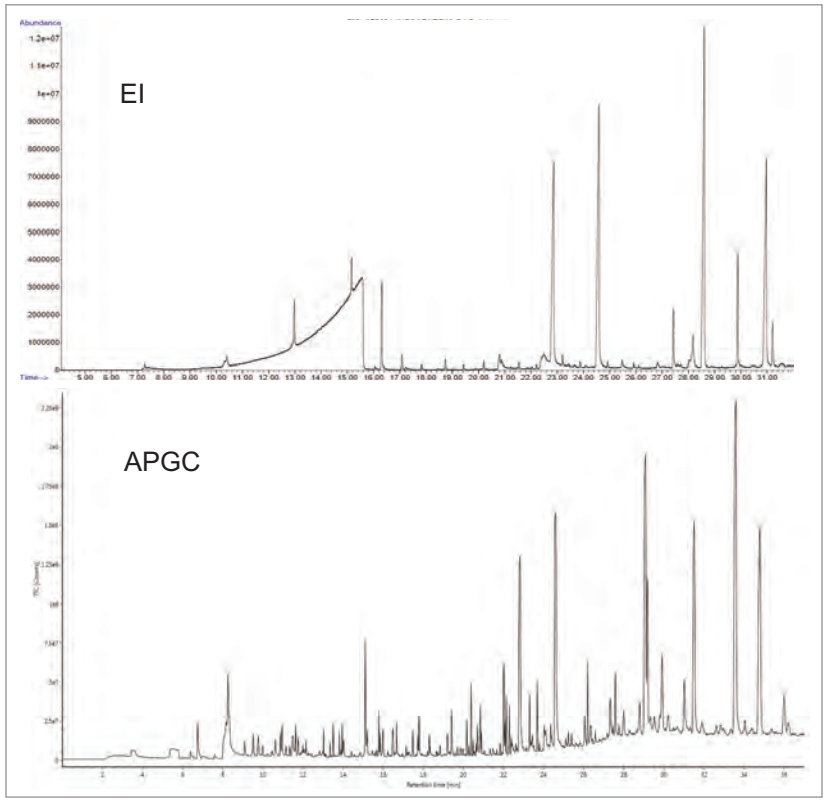

Figure 1. TIC chromatograms of the polymer extract acquired with EI (top), and with APGC at low collision energy (bottom).

BINARY COMPARISON

It is important to determine whether a peak comes from the tested material or from external contamination. Therefore, the analysis of a sample must always be accompanied by the analysis of its blank extract. UNIFI Software's Binary Compare feature allows direct comparison of the analysis results of an unknown sample with those of a reference (blank) sample, and to display the results in a mirror-image plot (Figure 2).

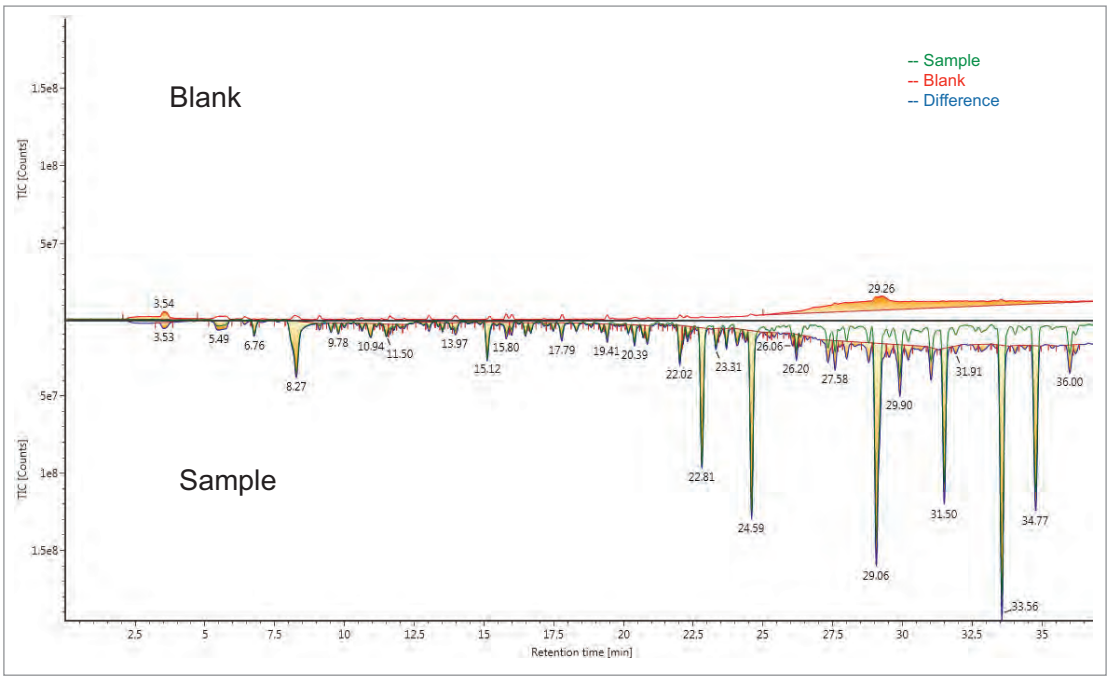

Figure 2. UNIFI's Binary Compare window shows the unknown sample and blank chromatographic profiles.

In addition, after specifying the mass tolerance, retention time tolerance, and intensity threshold of the unknown and reference samples in the comparison settings, UNIFI returns a Component Summary, where it is easy to identify the ions that are present in the unknown sample only, sorted by the intensity of response (Figure 3).

Component Summary

	Unknown component name	Unknown RT (min)	Unknown m/z	Match type	Unknown intensity (Counts)	Unknown/Reference	Reference m/z
1	Candidate Mass 480.4893	34.78	480.4893	Unknown Unique	4817260		0.0000
2	Candidate Mass 421.1843	33.57	421.1843	Common	3552189	104.3914	421.1836
3	Candidate Mass 452.4577	31.50	452.4577	Common	3513449	430.0651	452.4577
4	Candidate Mass 401.2153	29.07	401.2153	Common	3393889	66.9177	401.2153
5	Candidate Mass 481.4937	34.78	481.4937	Unknown Unique	3079423		0.0000
6	Candidate Mass 450.1754	33.57	450.1754	Common	2885954	160.5685	450.1748
7	Candidate Mass 430.2064	29.06	430.2064	Common	2880768	112.7112	430.2060
8	Candidate Mass 420.1770	33.57	420.1770	Common	2702121	97.1917	420.1765
9	Candidate Mass 400.2085	29.06	400.2085	Common	2615383	60.3286	400.2082
10	Candidate Mass 435.1651	33.58	435.1651	Common	2391867	137.5356	435.1639
11	Candidate Mass 453.4619	31.50	453.4619	Unknown Unique	2183887		0.0000
12	Candidate Mass 256.2635	22.85	256.2635	Common	2115949	83.8246	256.2634
13	Candidate Mass 285.2981	24.60	285.2981	Common	2100727	208.3426	285.2978

Figure 3. Excerpt of Component Summary table.

UNIFI's Binary Compare function is particularly useful when the blank samples present a high level of contamination, as well as when some of the peaks are not perfectly resolved. Furthermore, some components were not visible in the TIC chromatogram due to the trace-level nature of some NIAS from the packaging materials. In these circumstances, UNIFI Software helps the user to determine the unique compounds in the sample extract despite their low intensity, which would be labelled as "unknown unique".

CONFIRMING IDENTIFICATION

The first step is testing the applicability of APGC for the confirmation of compounds that are associated to a candidate in the NIST library with a high *match* value. By way of example, the peak at retention time 16.3 min was identified by EI as 1,6-Dioxacyclododecane-7,12-dione (molecular formula $C_{10}H_{16}O_4$, monoisotopic molecular mass 200.1049 amu, CAS number 777-95-7) with a match of 917 (Figure 4A).

The same peak was processed via APGC, and its spectrum showed a base peak at *m/z* 201.1120, which is attributed to the $[M+H]^+$ ion (Figure 4B).

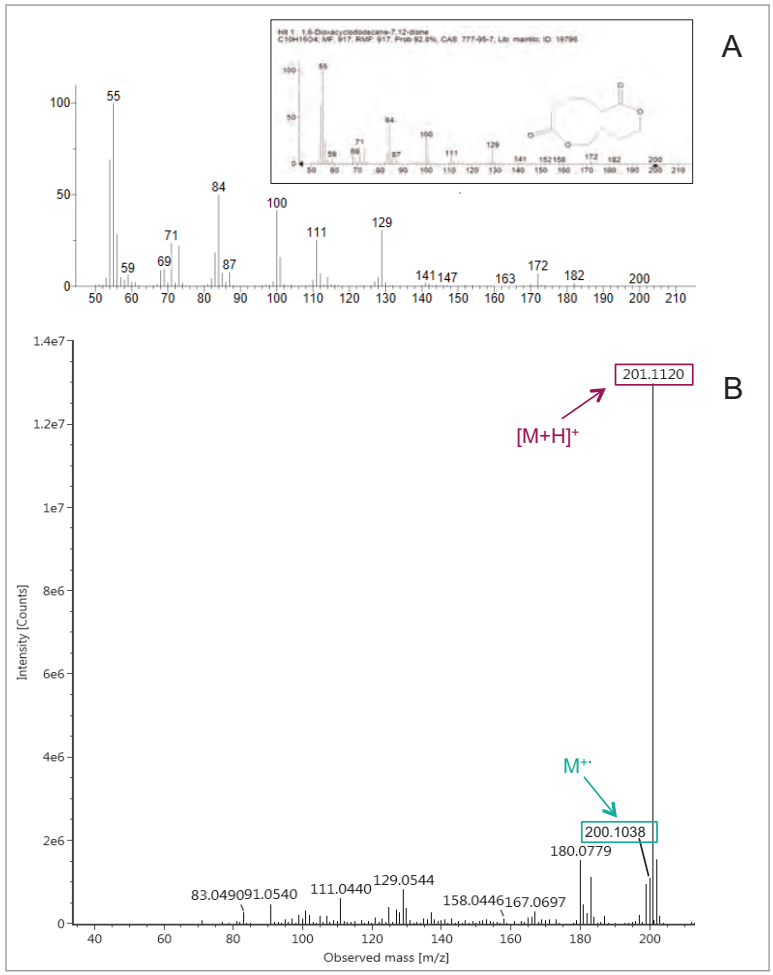

Figure 4. Comparison between the unknown and the reference for peak Rt = 16.2 min, showing (a) EI spectra, and (b) APGC low collision energy spectrum of the same chromatographic peak.

Using UNIFI's Mass Calculator feature, it is possible to obtain the exact mass of the adduct candidate molecular formula proposed by the EI library $[C_{10}H_{16}O_4+H]^+$. Hence, the mDa and ppm errors can be calculated. In the current example, the candidate molecular formula presents -0.14 mDa error and -0.7 ppm error. In APGC, the molecular ion $M^{+\cdot}$ at *m/z* 200.1038 is also present; in this case, the errors are -0.48 mDa and -2.4 ppm. Even though the presented APGC spectrum was obtained under dry conditions, protonation prevails over charge transfer because the structure of the investigated molecule favors accepting a proton, since even under dry conditions, the complete elimination of moisture in the ion source cannot be reached. The results demonstrate that the molecular formula of the candidate could be confirmed by the accurate mass of the molecular ion and the protonated adduct.

While linear adipates are usually employed as plasticizers in many plastic materials, 1,6-Dioxacyclododecane-7,12-dione is a cyclic adipate that was previously also found as a NIAS in biodegradable polyesters,[7] printing inks,[8] and polyurethane plastics.[9]

This example highlights the usefulness of APGC coupled with high resolution mass spectrometry when confirmation of the molecular formula is needed.

CORRECTING AN INCORRECT IDENTIFICATION

At the retention time 17.2 min in EI there was a very low intensity and broad peak that NIST attributed to 3,4-altrosan or beta-D-glucopyranose, 1,6-anhydro-, with a *match* value of 787. Both compounds have a molecular weight of 162 amu. However, by analyzing the same peak in APGC, a base ion peak at *m/z* 232.1817 appeared.

UNIFI Software allows users to create a customized workflow through the introduction of filters in order to get better visualization of data, and to save time by focusing on the most relevant components. For example, it is possible to select a specific Rt window to be analyzed and an ion intensity threshold. Applying this filter (Rt window 17.16–17.27 min and response >5000 counts) for peak Rt 17.2 min in APGC, UNIFI returns the component list that fits those settings. In this example, we displayed the processed and non-processed high collision energy spectra of the same component, shown in Figure 5. The processed spectrum appears "cleaner" because it focuses only on the component under investigation, without ions coming from other compounds that could partially coelute with the compound of interest.

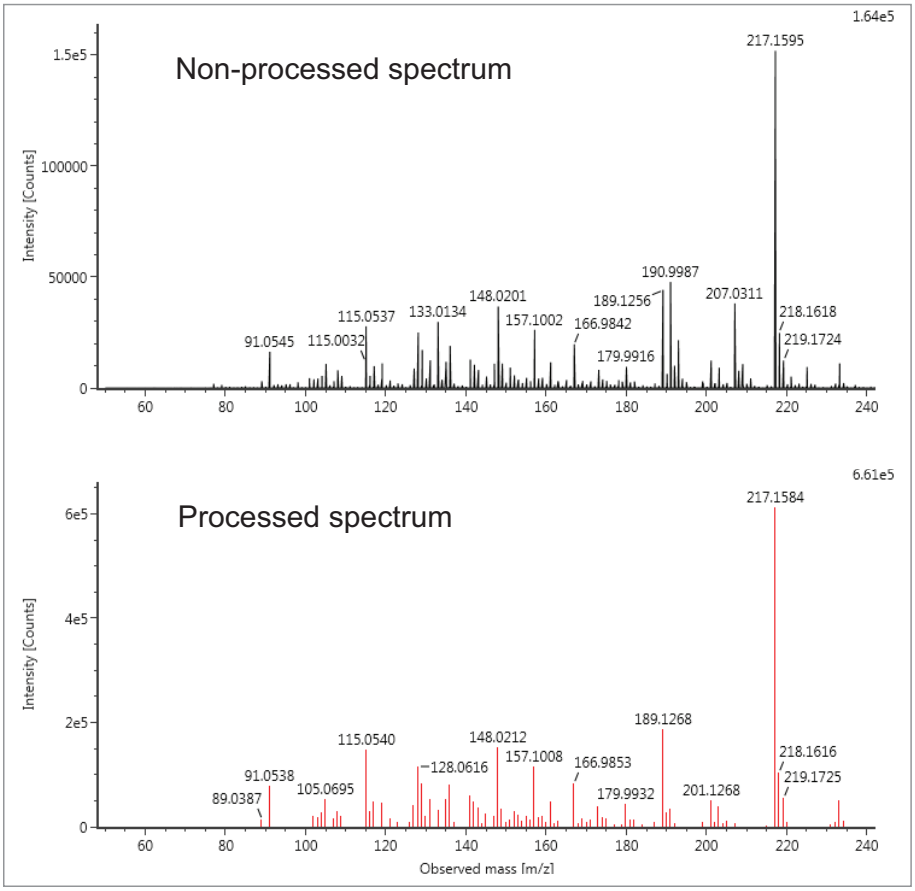

Figure 5. APGC high collision energy spectra of peak Rt 17.2 min. Non-processed spectrum (top) and processed spectrum based on component m/z 232.1817 (bottom).

UNIFI's filters, views, and workflow steps allow users to review data in a more timely, consistent, and accurate way. The componentization feature in UNIFI allows interrogation of entire datasets without having to interact with the raw data. Componentization also facilitates the selection of candidate components, which may represent unexpected substances within a sample; this is possible with UNIFI's 3D peak detection algorithm.[10]

When screening complex samples, the UNIFI Elucidation toolset can be used to investigate and potentially identify candidate components. The Elucidation toolset includes an elemental composition calculator that determines a number of possible formulas for an accurate mass peak. Elemental Composition uses an algorithm, i-FIT,™ to score each formula by the likelihood that the theoretical isotope pattern of the formula matches a cluster of peaks in the spectrum. To restrict the number of possible formulas, the i-FIT model can take into account fragment ion mass spectral peaks, the number of atoms of elements specified, valence state, the number of double bonds in a formula, the type of isotope pattern, and a series of chemical rules.

By applying the Elemental Composition tool to mass 232.1817 UNIFI proposed the molecular formula $C_{16}H_{24}O$ ($M^{+\cdot}$) with the lowest mDa error and the highest i-FIT confidence (%), as shown in Figure 6.

After searching ChemSpider, PubChem, and SciFinder, the suggested molecular formula was attributed to 1,2,3,4-tetrahydro-1-methoxy-1,6-dimethyl-4-(1-methylethyl) naphthalene (CAS number 60698-94-4). The Elemental Composition tool was also used to check the molecular formula of the most abundant fragments in the processed high collision energy spectrum, and to deduce their structures. In Figure 7 the proposed fragmentation pathway is shown, which confirmed the candidate structure of the molecular ion.

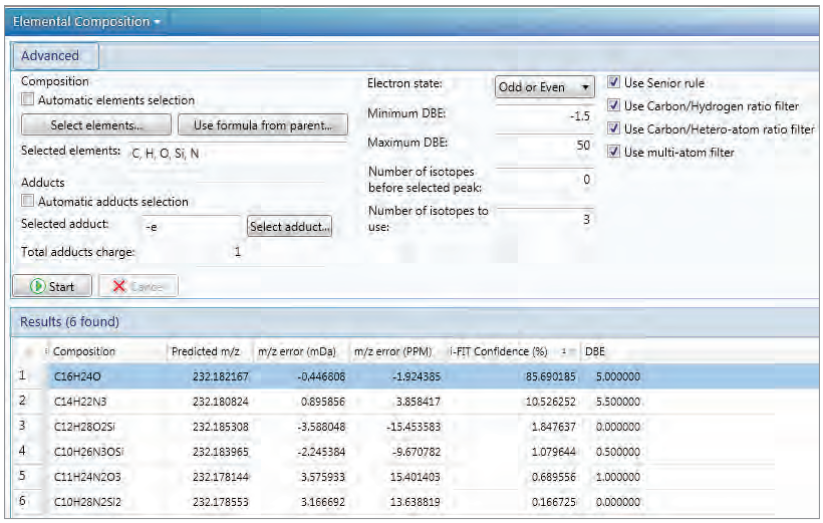

Figure 6. Results from UNIFI Software's Elemental Composition tool for the ion m/z 232.1817.

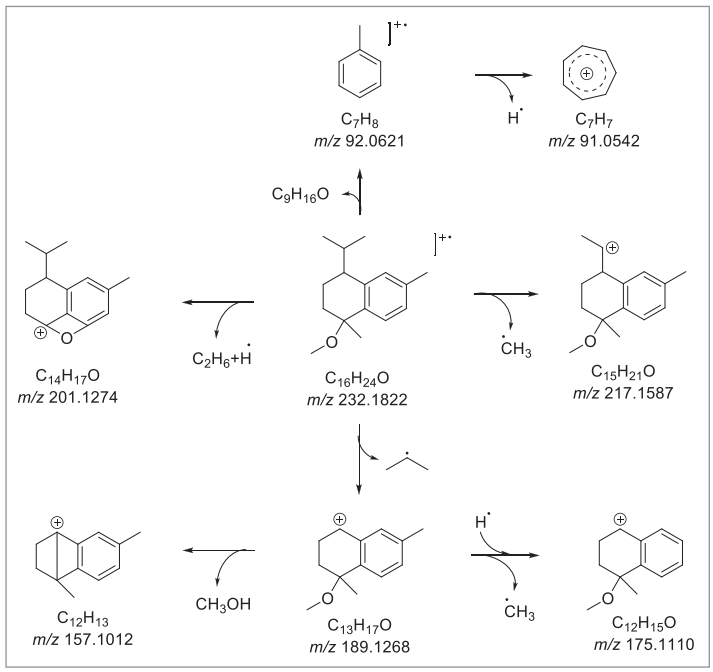

Figure 7. Proposed fragmentation pathway of the molecular ion $M^{+\cdot}$. Fragment ions are defined by their molecular formula and exact mass-to-charge ratio.

1,2,3,4-tetrahydro-1-methoxy-1,6-dimethyl-4-(1-methylethyl) naphthalene was also found in essential oil extracts of several plants, such as hops, pine and Japanese spicebush,[11-13] as well as in propoli extracts[14] as a component of the volatile profile.

Here, we were able to correct the EI identifications of components that presented a low match value or that were not listed in the libraries using APGC and UNIFI.

IDENTIFYING PREVIOUSLY NON-DETECTABLE PEAKS

Since the APGC-QTof MS system delivers enhanced sensitivity compared to EI-MS, APGC spectra lead to a significantly higher number of detected peaks. Consequently, it is possible to extend the identification process to a wider range of compounds. By way of example, the compound represented by the peak at Rt 27.3 min in the APGC spectrum was not present in the EI spectrum (Figure 8).

In this step, the Discovery tool in UNIFI was employed on the base ion peak *m/z* 410.3169.

In Figure 9 it can be noted that UNIFI attributed the component of interest to a predicted list of chemicals, recognized to be likely by an automatic search in ChemSpider. The table shows a list of possible compounds sorted by Predicted Intensity, i-FIT Confidence, Fragment Match, or number of citations.

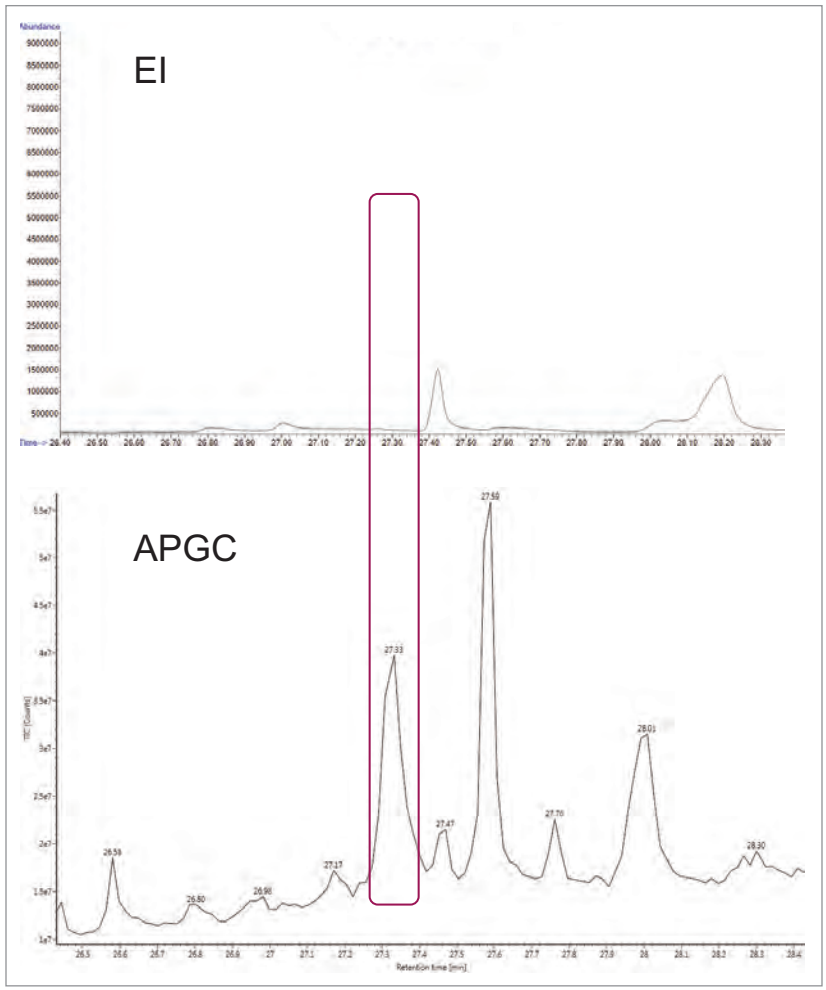

Figure 8. Comparison between the EI and APGC chromatograms within the range 26.4–28.4 min, highlighting the peak at 27.3 min in APGC, not detected with EI.

Discovery

Parameters

Results (90 found)

	Component Name	Elemental Composition	Predicted m/z	Predicted Intensity...	i-FIT Confidence (%)	DBE	Fragment Matches	Citations	Common Name
4	Candidate Mass 410.31690...	C28H42O2	410.318	60	47.62	8.0	79	40	e-Tokoferol
5	Candidate Mass 410.31690...	C28H42O2	410.318	60	47.62	8.0	79	36	gamma-Tocotrienol
6	Candidate Mass 410.31690...	C28H42O2	410.318	60	47.62	8.0	79	11	2,5,8-Trimethyl-2-(4,8,12-trimethyltridec
7	Candidate Mass 410.31690...	C28H42O2	410.318	60	47.62	8.0	79	10	2,7,8-Trimethyl-2-[(3E,7E)-4,8,12-trimeth
8	Candidate Mass 410.31690...	C28H42O2	410.318	60	47.62	8.0	79	4	2,5,8-Trimethyl-2-[(3E,7E)-4,8,12-trimeth
9	Candidate Mass 410.31690...	C28H42O2	410.318	60	47.62	8.0	79	2	2,7,8-Trimethyl-2-(4,8,12-trimethyl-3,7,1
10	Candidate Mass 410.31690...	C28H42O2	410.318	59	47.62	8.0	88	13	Phenylacetaldehyde digeranyl acetal
11	Candidate Mass 410.31690...	C28H42O2	410.318	59	47.62	8.0	88	5	(2,2-Bis[(3,7-dimethyl-2,6-octadien-1-yl)o
12	Candidate Mass 410.31690...	C28H42O2	410.318	41	47.62	8.0	42	4	(3beta,22E,24S)-3-Hydroxyergosta-5,8,22
13	Candidate Mass 410.31690...	C28H42O2	410.318	41	47.62	8.0	42	4	(22E,24xi)-3-Hydroxyergosta-5,8,22-trien
14	Candidate Mass 410.31690...	C28H42O2	410.318	41	47.62	8.0	42	3	(3beta,22E,24xi)-3-Hydroxyergosta-5,8,2
15	Candidate Mass 410.31690...	C28H42O2	410.318	41	47.62	8.0	39	5	Ergosta-4,24(28)-diene-3,6-dione

Figure 9. Results from UNIFI's Discovery tool for component m/z 410.3169 at Rt 27.33 min.

The candidates highlighted in yellow present a Predicted Intensity >50%. After analyzing the most important fragment ions, applying the common organic chemistry rules, and checking their molecular formula and mDa errors, the unknown compound was identified as e-tokoferol, more commonly called beta-tocotrienol, IUPAC name: [*R*-(*E*,*E*)]-3,4-dihydro-2,5,8-trimethyl-2-(4,8,12-trimethyl-3,7,11-tridecatrienyl)-2H-1-benzopyran-6-ol (CAS number 490-23-3). In Figure 10, the Discovery information output is illustrated. On the left side of the figure there is a list of synonyms for the candidate, while on the right side, the software shows the chemical structure and the high collision energy mass spectrum, where the most important fragments are pointed out.
It is possible to check out the molecule's cleavage points by clicking the fragment marker on the ion peak; the fragment *m/z* 191.1062 was chosen as an example.

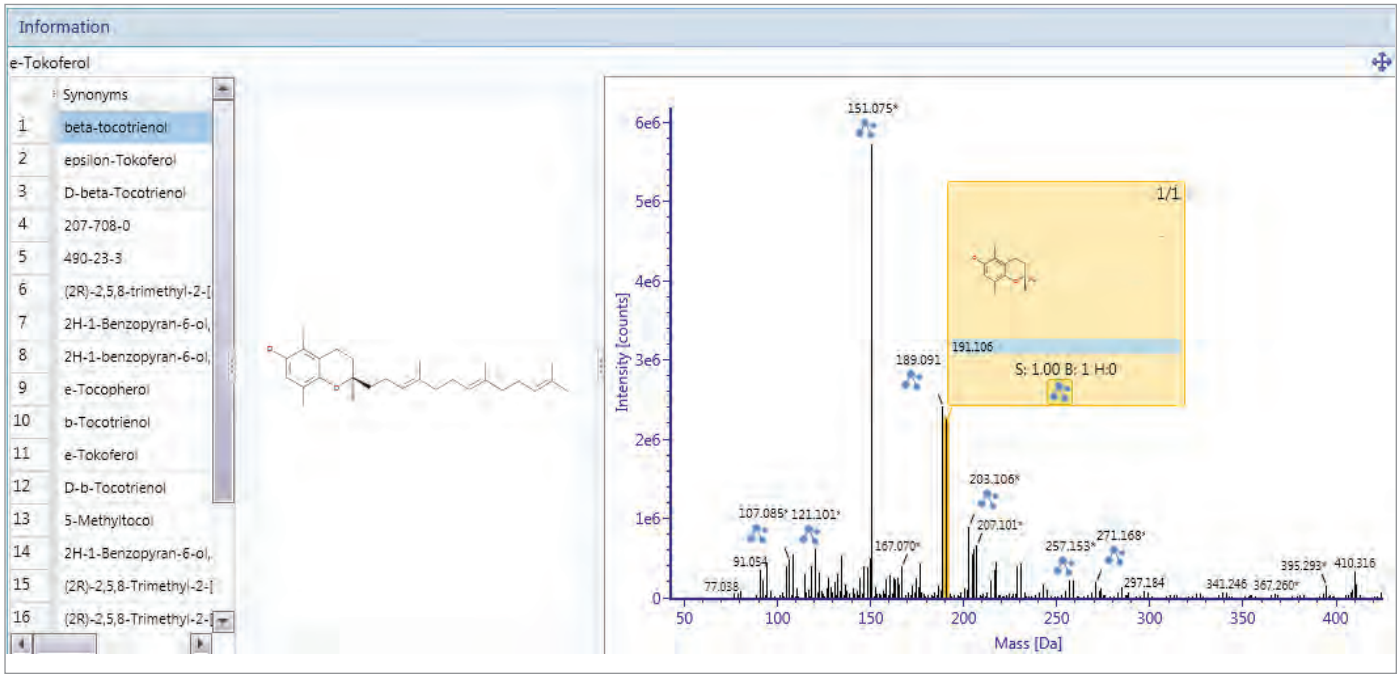

Figure 10. UNIFI's Discovery tool information output of beta-tocotrienol. Highlighted is one of the major fragments (m/z 191.1062).

Tocotrienols are members of the Vitamin E family, characterized by an unsaturated isoprenoid side chain (farnesyl isoprenoid tail) with three double bonds; their presence in the polymer could be due to their employment as antioxidant additives. In addition, tocotrienols are bioactive compounds normally present in many fatty foodstuff (such as vegetable oils), that have been used in many nutritional and pharmaceutical applications.[15]

UNIFI's Discovery tool saves analyst's time in the elucidation process and provides comprehensive high-quality information by sorting the possible candidates, based on several parameters set by the user. However, it should be noted that to reach a confidence level closer to 100% in the identification of an unknown compound, the candidate compound must be confirmed with a standard by verifying retention time, accurate mass, and common fragments.

CONCLUSIONS

Identifying unknown compounds in food contact materials is usually a challenging process. The UNIFI Scientific Information System simplifies the process by providing customizable workflows and achieving data containing accurate mass precursor and fragment ions information acquired by the MS^E functionality.

EI-MS and APGC-QTof MS systems have been proven to be complementary when the compounds of interest are described in commercially available libraries, whereas APGC-QTof MS is particularly advantageous when the elucidation is required for volatile and semi-volatile components not listed in the libraries, or for those at trace or ultra-trace levels. APGC-Xevo G2-XS QTof with UNIFI can determine possible erroneous identifications and also facilitate component identification for peaks that are not detected using an EI quadrupole MS system.

Finally, UNIFI componentization eases the burden of data interpretation for the analyst, reducing potential false-positive assignments, and allowing results to be presented clearly and concisely.

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APGC Performance

The soft ionization of APGC leads to increased molecular ion abundance compared to EI where substantial fragmentation takes place. APGC has demonstrated improved sensitivity over EI in GC-MS and GC-MS/MS applications. When APGC is coupled with the Xevo TQ-XS (Tandem Quadrupole) Mass Spectrometer, ultimate sensitivity for a GC-MS/MS system becomes accessible, meeting even the most difficult limits of detection for regulatory analyses such as confirmation of dioxins in food and feed in the EU. When coupled with Time-of-Flight (ToF) technology, the reduced fragmentation assists in compound coverage and elucidation of compounds due to the higher abundance of the precursor ion.

Confirmation of PCDDs and PCDFs at Sub-Femtogram Levels Using Atmospheric Pressure Gas Chromatography (APGC) with Xevo TQ-XS

Rhys Jones,[1] Keith Hall,[2] David Douce,[1] Jody Dunstan,[1] Ken Rosnack,[3] and Adam Ladak[3]
[1]Waters Corporation, Wilmslow, UK; [2]GC^2, Wythenshawe, UK; [3]Waters Corporation, Milford, MA, USA

GOAL

To determine the limits of detection for dioxins and furans in solvent standards, and to confirm their presence and accurate quantitation in a QC fly ash samples.

BACKGROUND

Polychlorinated dibenzo-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs) are a group of chemically related compounds that are toxic and persistent organic pollutants (POPs). These compounds are restricted internationally under the Stockholm Convention[1] and due to the bioaccumlative nature of these compounds, it is essential to monitor them at ultra trace levels in food and environmental samples. Traditionally these compounds have been analyzed using magnetic sectors with electron ionization sources which require expert users to obtain consistent results. As there is a growing concern for the analysis of these compounds, more user-friendly technology is essential to analyze potentially contaminated samples. Atmospheric Pressure Gas Chromatography (APGC), coupled with a highly sensitive tandem quadrupole mass spectrometer (Xevo® TQ-S),

Coupling APGC to Xevo TQ-XS takes sensitivity to the next level – Confirm dioxins in complex samples at concentrations that are unachievable by traditional magnetic sector GC systems.

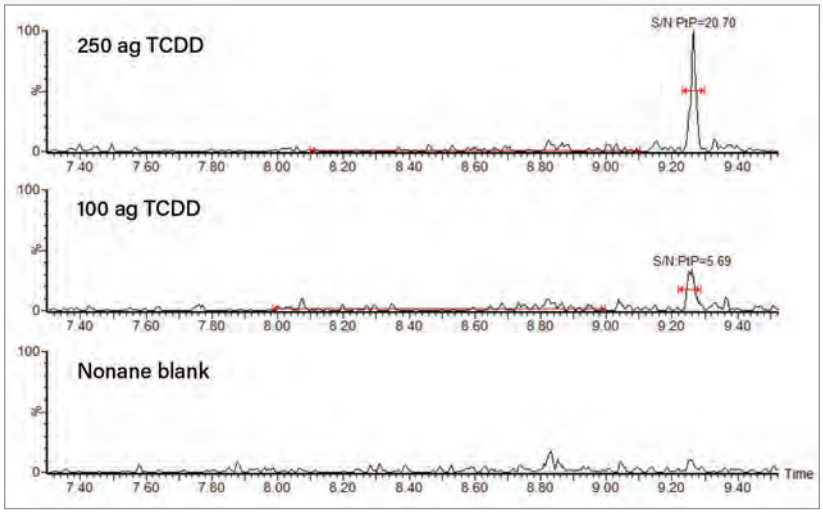

Figure 1. 2,3,7,8 TCDD at 100 ag on-column.

has already been demonstrated to be a sensitive and robust option for confirmatory analysis of PCDDs and PCDFs by GC-MS/MS in compliance with 589/2014/EU.[2] The recent introduction of the Xevo TQ-XS from Waters has allowed lower limits of detection to be reached. This can help reduce time spent on sample preparation/preconcentration, as well as reducing the cost of analysis as diluted standards can be utilized.

THE SOLUTION

GC Method for TCDD assessment

Agilent DB-5MS column, 30 m x 0.25 mm I.D. x 0.25 µm film, helium at 1 mL/min. 7890A GC Oven and Agilent autosampler, split/splitless injector at 290 °C operating in pulsed splitless mode (32 psi for 0.5 min) with a 1.0 µL injection volume. GC program, initial temp. of 130 °C, hold for 1.2 min, ramp at 20 °C/min to 320 °C, and hold for 3.3 min.

GC method for full PCDD and PCDF assessment

Zebron ZB-5MS column, 60 m x 0.25 mm I.D. x 0.25 µm film, helium at 1.4 mL/min. 7890A GC oven and Agilent autosampler, split/splitless injector at 290 °C, operating in pulsed splitless mode (50 psi for 1.8 min) with a 1.0 µL injection volume. GC program: initial temp. of 130 °C, hold for 1.8 min, ramp at 40 °C/min to 200 °C; ramp 2 at 2 °C/min to 235 °C; ramp 3 at 3 °C/minute to 305 °C; ramp 4 at 20 °C/min to 320 °C, and hold for 5 min. Total run time of 49.85 min.

MS parameters for both assessments

Corona pin at 2.0 µA, cone gas 260 L/hr, auxiliary gas 200 L/hr, makeup gas 300 mL/min, quad resolutions at 0.7 Da.

In order to assess the sensitivity of the APGC coupled with the Xevo TQ-XS, a standard of 2,3,7,8-TCDD was diluted in nonane giving a calibration range between 100 ag to 100 pg on column. In order to perform this test, two MRM transitions for TCDD were utilized. Figure 2 shows the linearity of 2,3,7,8 TCDD, which was excellent.

An on-column standard concentration of 100 fg was injected over 20 days in order to assess the reproducibility of the system. Figure 3 shows the outstanding reproducibility of the response, and Figure 4 shows the stability of the isotopic measurements over this series of injections.

Once the initial sensitivity of the system had been verified, a full suite of TCDDs and TCDFs was acquired on the system. A series of EPA 1613 standards were used from CSL to CS5, diluted 1 in 10 with nonane. Figure 5 shows that the isotope ratio assessment for each congener was consistent at all concentrations. This is essential for the confirmation of dioxins and furans in a sample. Legislation states that these ratios are required to be <15%.[3,4,5]

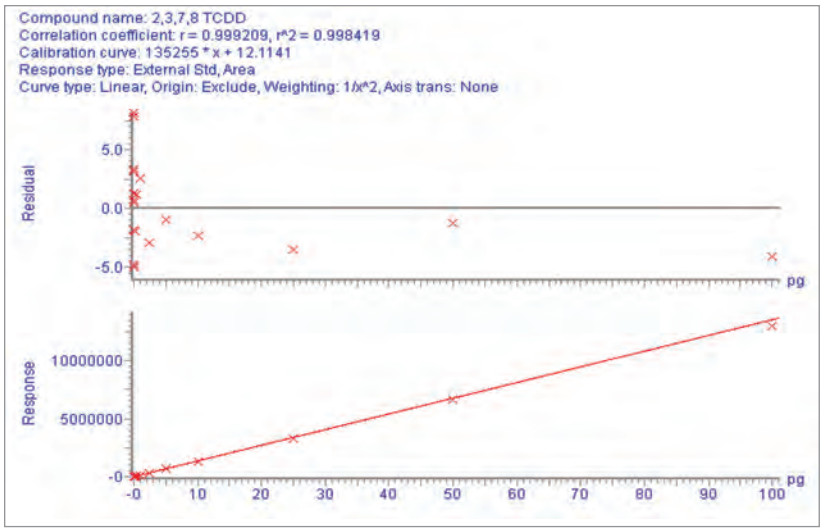

Figure 2. Linearity of 2,3,7,8 TCDD between 100 ag to 100 pg.

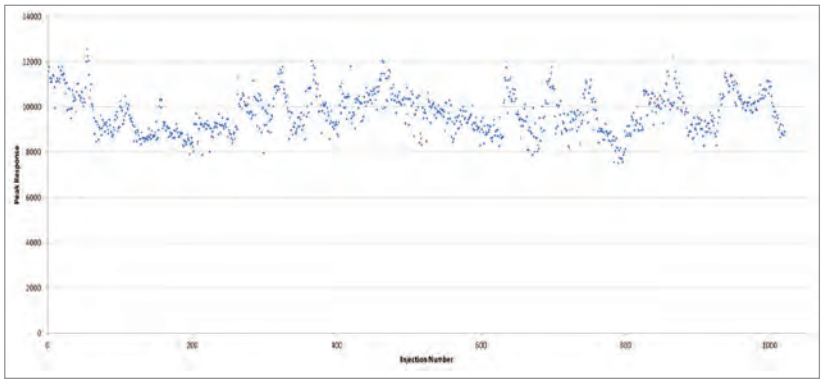

Figure 3. Stability of the response of 100 fg of 2,3,7,8 TCDD over 1000 injections with an RSD of 9.2% (no internal standard correction).

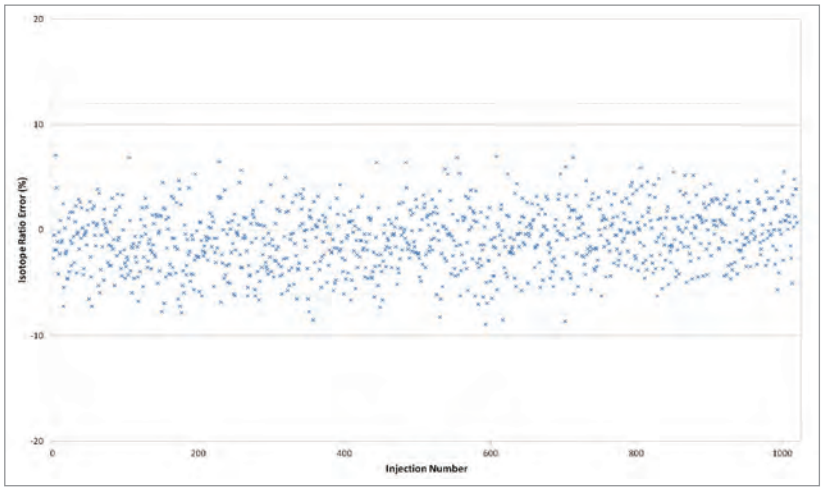

Figure 4. Stability of the isotope ratio of 100 fg of 2,3,7,8 TCDD over 1000 injections.

The final assessment was the analysis of a QC fly ash sample. These types of samples are very complex and often used as proficiency tests for dioxin labs in order to ensure that they are producing accurate results. Figure 6 shows the complexity of the samples and demonstrates the ability of APGC and Xevo TQ-XS to quantify the compound of interest, as highlighted in Figure 6. Figure 7 shows that the value obtained with APGC coupled with the Xevo TQ-XS were consistent with that of the QC sample.

SUMMARY

Utilizing APGC coupled with the Xevo TQ-XS allows sub-femtogram levels of dioxins to be analyzed in complex samples. The added sensitivity enables the dilution of expensive dioxin standards, reduces the need to pre-concentrate sample extracts (prior to analysis), and minimizes the amount of sample required for testing. Not only is this system exceptionally sensitive, it is also robust and produces consistent results over thousands of injections. The APGC coupled with the Xevo TQ-XS far surpasses the regulatory requirements for dioxin testing.

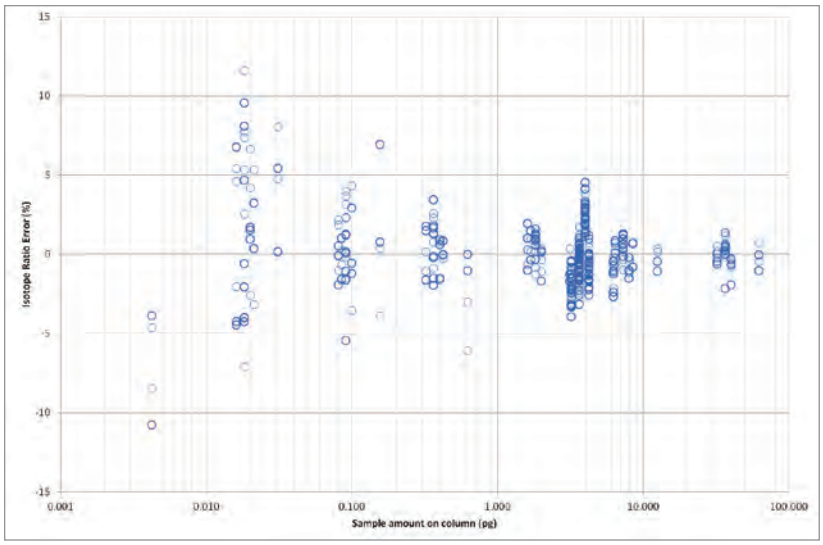

Figure 5. Consistency of the isotope ratio for 1 in 10 dilution of CSL to CS5.

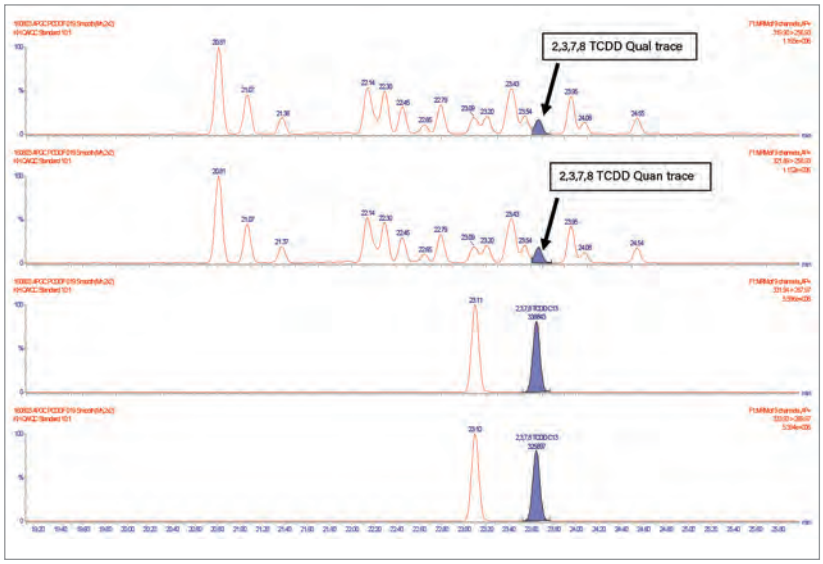

Figure 6. Complex fly ash sample chromatogram showing the identification of 2,3,7,8 TCDD.

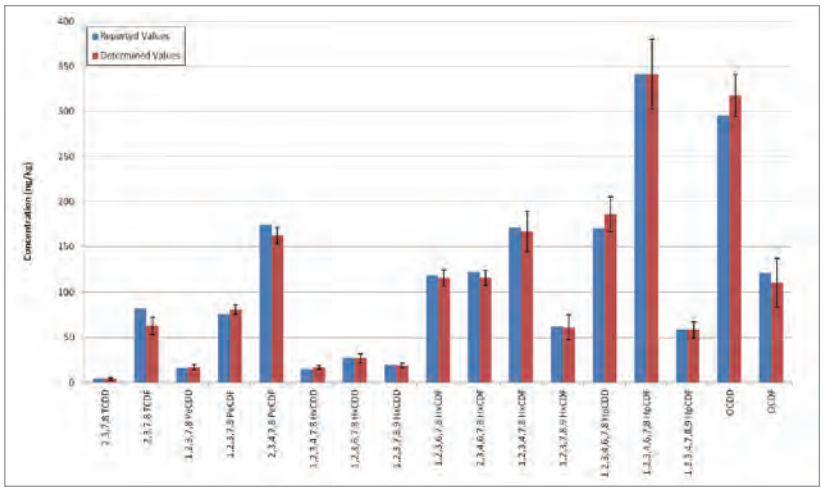

Figure 7. Results from fly ash QC sample.

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Analysis of Dioxins and Furans on a Xevo G2-XS QTof with APGC Using a QuEChERS Extraction Method

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[2]McMaster University, Hamilton, ON, Canada; [3]University of Toronto, Toronto, ON, Canada;
[4]VUV Analytics Inc., Austin, TX, USA; [5]Waters Corporation, Milford, MA, USA

TECHNOLOGY BENEFITS

- Exceeds minimum performance limits for EPA method 1613
- >15X faster sample throughput over traditional techniques
- Less expensive using QuEChERS sample preparation

WATERS SOLUTIONS

Xevo® G2-XS QTof

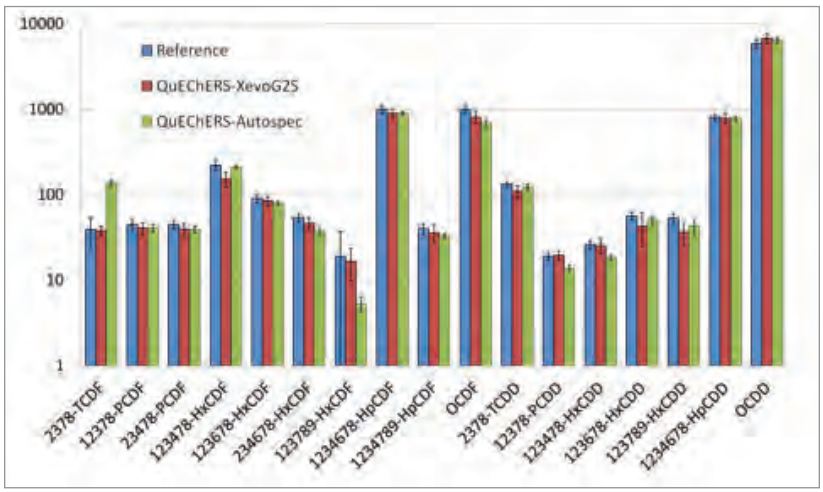
Atmospheric Pressure GC (APGC) Source

MassLynx® MS Software

KEYWORDS

Dioxins, furans, QuEChERS, QuanTof, charge transfer, APGC, persistent organic pollutants, POPs

QuEChERS extraction, combined with APGC and QTof allows dioxins analysis to be performed without the need for an expert operator, quicker and cheaper than traditional dioxin sample preparation and analysis.



Results from dioxins analysis using QuEChERS extraction followed by APGC-QTof MS are in good agreement with the NIST 1944 Standard Reference Material.

INTRODUCTION

The objective of this work was to develop a dioxin method that was faster and more cost effective than the traditional magnetic sector technique using APGC high resolution mass spectrometry (HRMS) analysis on a Waters® Xevo G2-XS QTof while exceeding the minimum performance limits required for EPA method 1613.

Dioxins and dioxin-like compounds are ubiquitous persistent organic pollutants (POPs) linked to various diseases including cancer.[1] They are restricted under the Stockholm Convention[2] and are monitored for their occurrence and toxicity by regulatory agencies worldwide.

The classical analytical method for testing dioxins in sediment using magnetic sector instruments is considered the "gold" reference standard. However it requires an expert operator and specialized instrumentation.[3] Traditional sample preparation times can exceed several days and use a large amount of costly and hazardous solvents.

Since sediment chemistry can vary spatially and temporally, it is necessary to analyze a large number of samples to properly characterize any site being evaluated for dioxin contamination.[4] This translates to an extreme expenditure of time for sample prep and massive solvent usage. Within the last decade, a single phase acetonitrile extraction known as QuEChERS (Quick, Easy, Cheap, Effective, Rugged, and Safe) has been employed to prepare food samples for pesticide analysis in as little as 30 minutes.[5] This technique was modified and adapted as a rapid extraction and cleanup for the analysis of dioxins and furans in sediment samples and was used in the preparation of samples in this study. In this new approach, samples were investigated using the Xevo G2-XS QTof equipped with Atmospheric Pressure GC (APGC).

DISCUSSION

A modified QuEChERS sample preparation method for the screening of dioxins and furans in sediment was developed which reduced sample preparation time from 10 samples in four to five days, to as many as 30 samples in one day.[6] This study also exploits the use of an APGC source (Figure 1a) coupled to the Xevo G2-XS QTof (Figure 1b) as an alternative to a traditional magnetic sector instrument.

Wet sediment samples were fortified with ^{13}C-labeled standards and extracted using a modified QuEChERS method. The separated organic layer was solvent exchanged to hexane by liquid-liquid extraction. The extract was cleaned by a carbon column and then concentrated for instrumental analysis using a magnetic sector GC HRMS system and a Xevo G2-XS QTof equipped with an APGC source. The column used in this analysis was a Restek Rtx-Dioxin2 at 20 m, 30 m, and 40 m lengths.

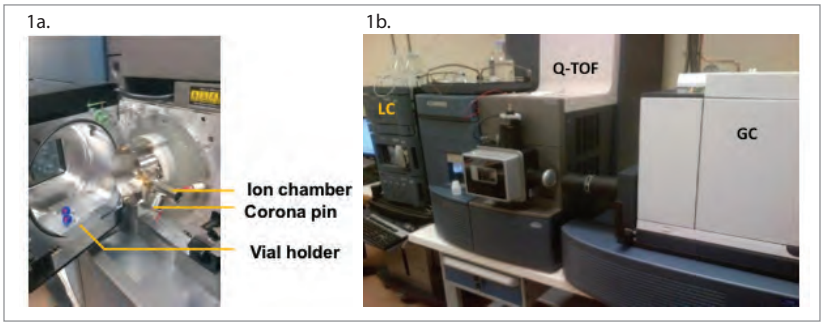


Figure 1a. Atmospheric Pressure Gas Chromatography (APGC) source on a 1b. Xevo G2-XS QTof Mass Spectrometer.

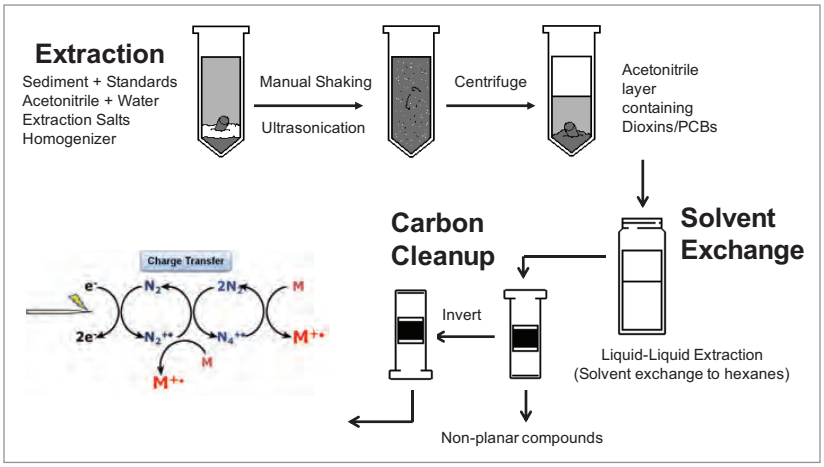


Figure 2. QuEChERS sample preparation schematic.

Analysis of Dioxins and Furans in Sediment by Xevo G2-XS QTof with APGC using a QuEChERS Extraction Method

The capabilities and performance of the APGC-Xevo G2-XS QTof proved to be similar or better than the magnetic sector MS for the analysis of dioxins. Unlike conventional EI (electron ionization) systems, the APGC source allows for higher flow rates to improve analysis times. The effect of increased flow rate on the chromatographic resolution for four different congener classes are shown in Figure 3. Although chromatographic resolution decreases with increased column flow rates, adequate separation is maintained for quantitative analysis, in large part due to the selectivity of the stationary phase (Rtx-Dioxin2). Only the pair of HxCDD congeners appear to co-elute, but given their identical TEFs (toxic equivalency factors), the impact of the reduced chromatographic resolution on TEQ (toxic equivalency quantity) is expected to be negligible.

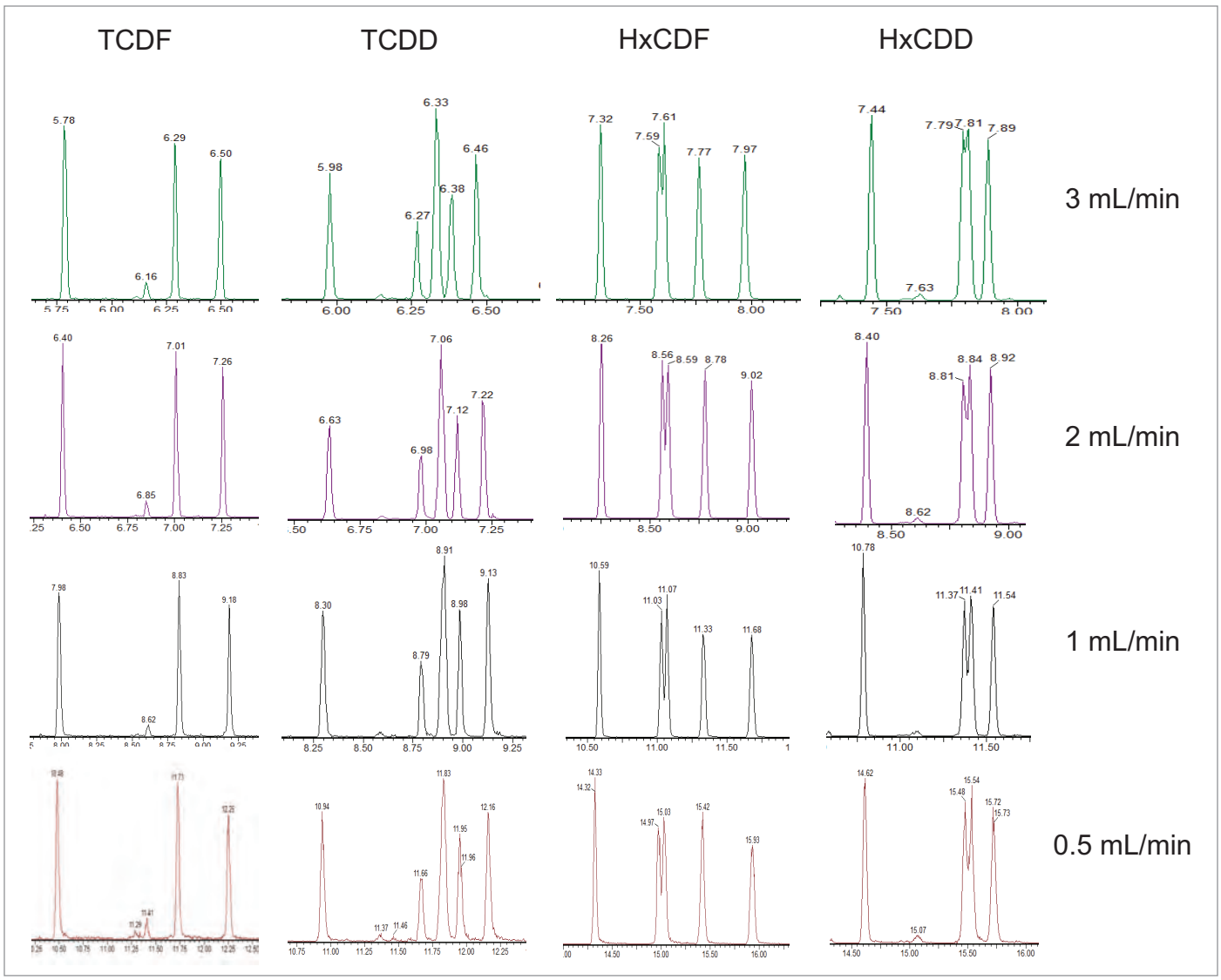


Figure 3. Chromatographic resolution as a function of flow using a mid-level calibration standard CS3WT. Closely eluting congeners were analyzed by APGC-QTof on an Rtx-Dioxin2 column. The higher flow rates possible with APGC will reduce run time while still maintaining adequate separation for quantitation.

Column length was also evaluated and the results are summarized in Figure 4. Shorter columns reduced backpressure, resulting in higher flow rates, and further reduced runtimes (<15 min/sample) with minimal loss in separation. APGC is sufficiently versatile to provide ultimate chromatographic performance (using a 40 m Rtx-Dioxin2 column at optimum flows) that satisfies the regulation, despite the method requirement for EI ionization and magnetic sector MS. When needed, high throughput and increased capacity is possible (using a 20 m Rtx-Dioxin2 column at flows >3 mL/min) while preserving separation of critical isomers.

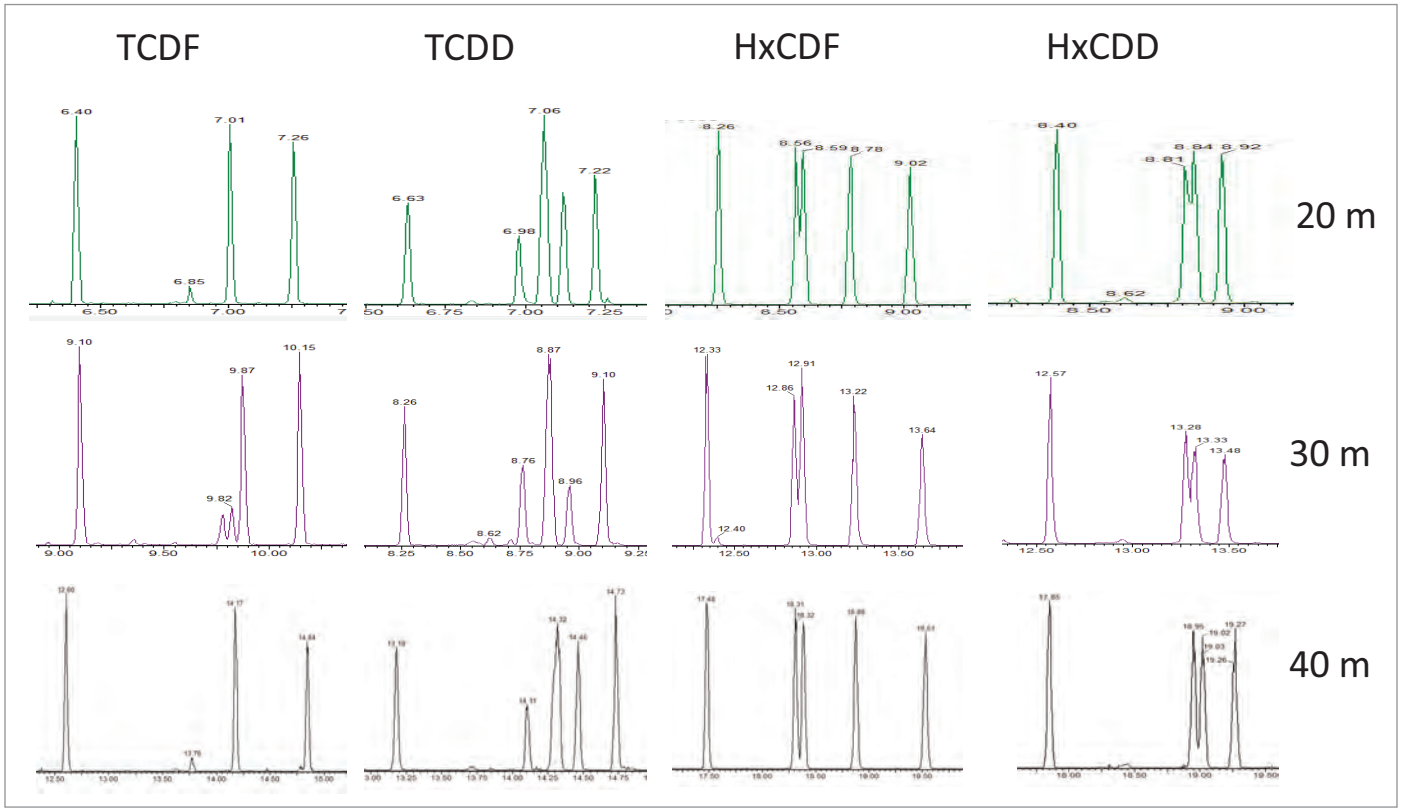


Figure 4. Chromatographic resolution as a function of column length in APGC-QTof on an Rtx-Dioxin2 column (flow rate 2 mL/min) using a mid-level calibration standard CS3WT. Shorter column lengths can reduce run time and provide less resistance to higher flows while maintaining separation.

Figure 5a shows results from a calibration curve of 2,3,7,8-TCDD from 0.5 to 200 pg with good linearity and R^2 of 0.9993 using APGC-Xevo G2-XS QTof.

Shown in Figure 5b are the results of the certified reference material compared to the APGC-Xevo G2-XS and magnetic sector. Results from the APGC-Xevo G2-XS compare favorably to the reference and the magnetic sector. It is worth noting, however, that the magnetic sector results for 2,3,7,8-TCDD and 1,2,3,7,8,9-HxCDF differ from the reference while the APGC results for those same congeners compare more favorably to the reference (Figure 5b).

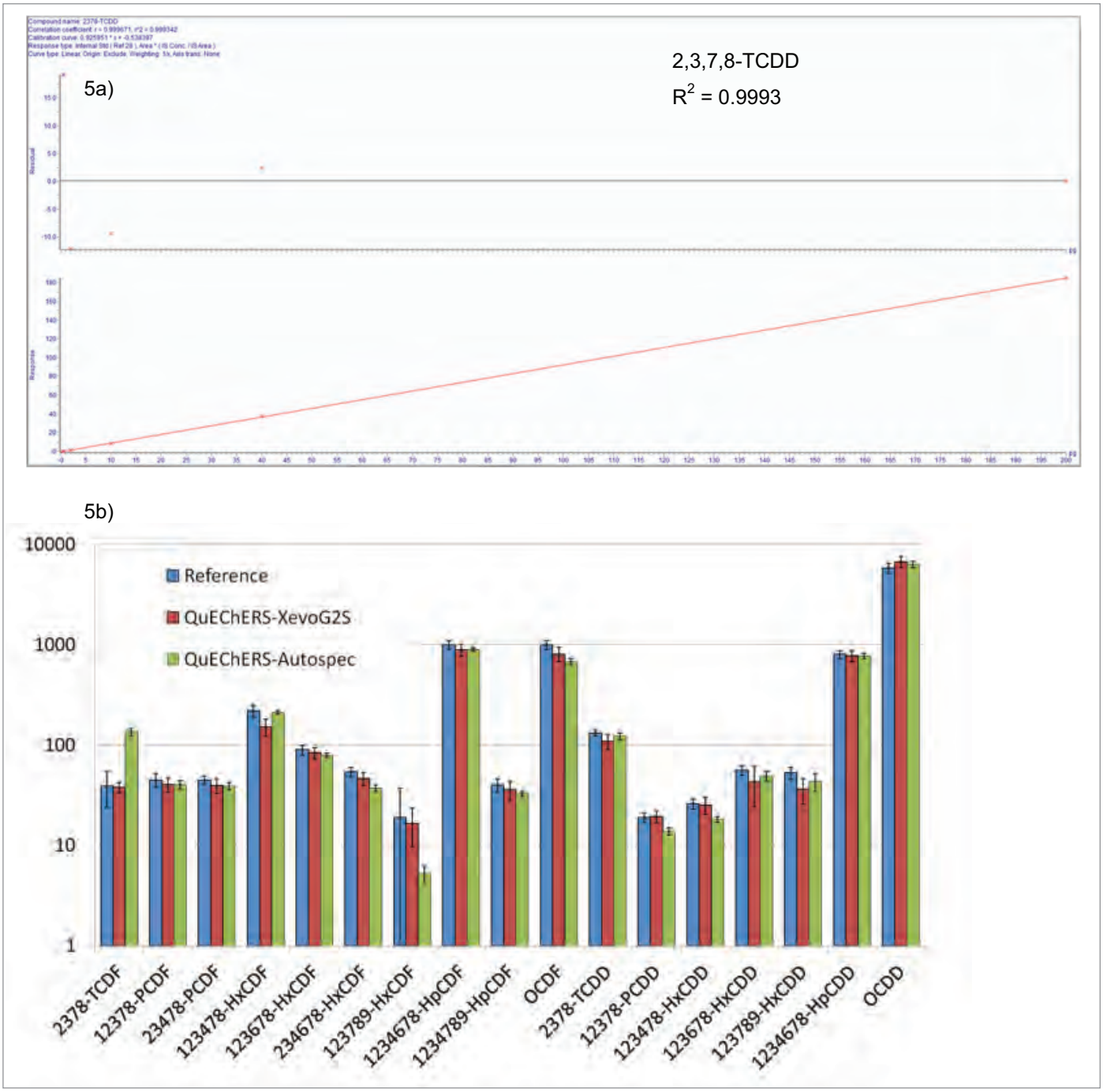


Figure 5a. Residuals plot and linear regression of a five point calibration of 2378-TCDD ranging from 0.5 pg to 200 pg run on APGC-QTof. 5b. Standard reference sediment NIST1944 (ng/kg dry-mass) extracted by the modified QuEChERS method and analyzed by GC-HRMS and with APGC-QTof. Comparison run on a 40 m Rtx-Dioxin2 column with a flow rate of 1 mL/min.

CONCLUSIONS

QuEChERS has been proven to be an effective sample extraction/clean-up method for the analysis of a large number of sediment samples from site remediation activities,[7] hence reducing the time and solvent as compared to the classic preparation. APGC along with the Xevo G2-XS QTof decreased instrumental run time due to its ability to handle higher flow rates than the GC-HRMS system. The combined method of QuEChERS extraction with APGC-QTof analysis provided a sample throughput increase of 15x over traditional techniques. The Xevo G2-XS QToF offers a flexible platform with inlet options including APGC, ESI, APCI, and UniSpray™ to name a few, thus permitting the instrument to perform other analysis when needed. It can operate in a non-targeted acquisition mode that can meet the limits of detection of dioxin regulatory method EPA1613, and can provide additional analytical information such as elemental composition on non-target analytes that can be encountered with both classical and generic sample preparation approaches.

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Analyzing Multi-Class Persistent Organic Pollutants (OCPs, PCBs, PBDEs, and PAHs) in Food Matrices in a Single Injection by APGC-MS/MS

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APPLICATION BENEFITS

- Increased sensitivity and selectivity for GC amenable POPs (OCPs, PAHs, PBDEs and PCBs).
- Improved response of quasi-molecular ion when compared with traditional EI spectra.
- Accurate and sensitive quantification of 141 POPs, including coronene and dibenzo pyrene compounds by APGC.
- Ability to analyze multi-class compounds in a single injection with generic sample preparation.
- The sensitive Xevo® TQ-S is suitable for quantification and confirmation of food and environmental contaminants, and readily interchangeable with UPLC® and APGC.

WATERS SOLUTIONS

Atmospheric Pressure Gas Chromatography (APGC)

Xevo® TQ-S

MassLynx® Software

KEY WORDS

Atmospheric pressure gas chromatography, APGC, organochlorine pesticides, OCPs, polycyclic aromatic hydrocarbons, PAHs, polychlorinated biphenyls, PCBs, polybrominated diphenyl ethers, PBDEs, persistent organic pollutants, POPs

INTRODUCTION

The application of gas chromatography coupled to mass spectrometry (GC-MS) is well established and documented for the analysis of ubiquitous environmental contaminants, such as persistent organic pollutants (POPs). Four classes of globally regulated POPs are polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), and organochlorine pesticides (OCPs).

Common physicochemical characteristics of these POPs include resistance to chemical and biological degradation and high lipophilicity, thus resulting in their persistence and significant potential to bioaccumulate. Many of these compounds are classified as toxic, carcinogenic, and probable carcinogenic. Such is the concern regarding these pollutants that international treaties, in association with regional legislation, requires continued monitoring to ensure human safety.

Due to the structural similarities of many POP congeners and the complexity of these compounds, their analysis can prove challenging. Traditional GC-MS and MS/MS techniques by electron impact (EI) ionization have been favored, due to the volatility, thermal stability, and non-polarity of the compound. However, given the hard ionization associated with EI, extensive fragmentation can impact the abundance of the molecular ion and compound specific spectra. Atmospheric pressure gas chromatography (APGC) allows for a softer ionization technique, thus providing a more abundant molecular ion. Operated at atmospheric pressure, compounds are ionized by corona discharge in the presence of nitrogen. This ionization reaction, depending on the analytes of interest, can occur by two processes: charge transfer (under dry conditions) and proton transfer (in the presence of a protic solvent).

In this application note, we describe the development and validation of a quantitative method for 141 multi-class POP compounds in a variety of foodstuffs to ensure continued monitoring and consumer safety in Quebec, Canada. The sensitivity, selectivity, and quantification capability of APGC, when coupled with the Waters® Xevo TQ-S will be determined, using a generic sample preparation method for the satisfactory extraction of all analytes from a variety of food matrices.

EXPERIMENTAL

GC conditions

GC system:	7890A
Injector:	Splitless
Injection:	1 µL
Temp.:	300 °C
Column:	DB-5 (J&W, USA) 30 m x I.D. 0.25 mm x df 0.25 µm
Guard column:	120 cm fused silica hi-temp
Interface:	55 cm fused silica hi-temp
Temp. gradient:	70 °C (hold for 1 min), 12 °C.min^{-1} to 250 °C, 5 °C.min^{-1} to 280 °C, 4 °C.min^{-1} to 310 °C (hold for 4.5 min)
Transfer line temp.:	340 °C
Carrier gas flow:	1.5 mL.min^{-1} (helium)
Auxiliary gas:	350 l.h^{-1} (nitrogen)
Make-up gas:	250 mL.min^{-1} (nitrogen)

MS conditions

MS system:	Xevo TQ-S
APCI corona pin current:	2.5 µA
Cone gas:	250 l.h^{-1} (nitrogen)
Acquisition mode:	multiple reaction monitoring (MRM) in positive ionization for all four classes of POPs shown in Table 3
Data management:	MassLynx Software with TargetLynx Application Manager

Sample preparation

Milk, infant formula, beef, pork, chicken, and fish were analyzed for PCB, PBDE, OCP, and PAH compounds using the following generic extraction procedure. Homogenized sample (12 g) was placed in a 50-mL glass centrifuge tube and fortified with internal standard, as described in Table 1. A smaller sample portion (10 g) was weighed for foods with high fat content (*e.g.* beef). Water (5 mL) was added to the solid food samples and reconstituted powder milk formulae. Samples were vortexed and allowed to stand for 20 minutes. Ethyl acetate (10 mL) was added and samples were shaken vigorously for 1 minute. QuEChERS salts, magnesium sulphate (4 g), and sodium chloride (2 g) were added to the tubes and shaken vigorously for an additional minute.

Following centrifugation, the supernatant (5 mL) was removed, evaporated, and reconstituted in dichloromethane (2.4 mL). It was then filtered through 0.45 µm PTFE filters in preparation for gel permeation chromatography (GPC). EnvirosepABC GPC pre column (60 x 21.2 mm) and column (350 x 21.2 mm) were used, with dichloromethane as eluent (5 mL.min^{-1}). The resultant extract was transferred to a suitable tube for evaporation, where the GPC collection tube was rinsed three times with dichloromethane. These rinses were combined with the original extract and evaporated to 750 µL. The volume was then made up to 1 mL in hexane and silica gel cleanup was performed. Silica columns were prepared by adding silica (2 g) into a 1-cm wide borosilicate glass column with a glass wool frit. These columns were conditioned with 3:1 hexane: dichloromethane solution (12 mL), followed by hexane (8 mL). The samples were loaded and eluted using 3:1 hexane: dichloromethane solution (20 mL). These extracts were evaporated to <0.5 mL, and fortified once more with the internal standard (25 µL, compounds marked with ** in Table 1). All samples were made up to 500 µL volume with isooctane, vortexed, and analyzed using the Xevo TQ-S with APGC.

Validation procedure

Method efficiency was determined and validated based on an in-house document that was inspired by several internationally recognized documents.[5-15] The limits of detection (LODs) and limits of quantification (LOQs) were determined using fortified replicates (n= 10) for all analytes in each matrix. This was carried out in accordance with the IUPAC method, *i.e.* LOD= 3 x std deviation of experimental noise, and LOQ= 10 x standard deviation of experimental noise.

The lowest limit (LL) was determined for all analytes where subsequent validation work and statistical analysis were based on this. Analyte recovery, repeatability (%RSD), and linearity were investigated. Replicate samples (n= 9) were prepared for each matrix (n= 6) at three fortification levels: 0.2 x LL, LL, and 2 x LL. From these replicates, the recovery and method repeatability were determined individually for each matrix.

Table 1. Internal standards for each class of POP, in order of retention time.

***Signifies internal standards added prior to injection.*

	Compound	RT min	*MRM m/z*	Cone V	Collision eV	*MRM m/z*	Cone Voltage	Collision eV	Spiking level ($\mu g.kg^{-1}$)
1	Naphtalene d-8	6.38	136>108	55	20	136>84	55	20	0.5
2	2-methylnaphtalene-d10	7.65	152>150	40	18	152>122	40	28	0.5
3	Acenaphtylene-d8	9.44	160>158	65	28	160>156	65	30	0.5
4	Acenaphtene-d10	9.78	164>162	40	20	164>160	40	30	0.5
5	PCB #3 C-13	10.75	200>164	30	25	202>164	30	25	0.5
6	Fluorene-d10	10.78	176>174	35	20	176>172	35	35	0.5
7	Hexachlorobenzene C-13	12.11	294>222	30	35	294>257	30	30	0.5
8	BHC beta C-13	12.42	225>189	30	15	225>187	30	15	0.5
9	BHC gamma C-13	12.54	225>189	30	15	225>187	30	15	0.5
10	Phenanthrene-d10	12.69	188>186	65	25	188>160	65	40	0.5
11	PCB #15 C-13	12.75	234>164	30	25	236>164	30	25	0.5
12	Anthracene-d10	12.79	188>186	65	25	188>160	65	40	0.5
13	PCB #31 C-13	13.49	268>198	30	25	270>198	30	25	0.5
14	PCB #52 C-13	14.1	304>232	30	25	304>234	30	25	0.5
15	Heptachlore epoxyde cis C-13	15.05	363>262	30	20	363>292	30	20	0.5
16	Oxychlordane C-13	15.07	397>296	30	18	397>262	30	40	0.5
17	Fluoranthene-d10	15.1	212>210	70	35	212>208	70	35	0.5
18	Pyrene-d10	15.53	212>210	70	35	212>208	70	35	0.5
19	Nonachlor trans C-13	15.76	417>310	30	25	417>270	30	30	0.5
20	PCB #81 C-13	15.96	304>232	30	25	304>234	30	25	0.5
21	DDE p,p C-13	15.98	328>258	30	25	330>260	30	25	1.25
22	Dieldrine C-13	16.06	313>242	30	22	313>278	30	18	0.5
23	PCB #77 C-13	16.12	304>232	30	25	304>234	30	25	0.5
24	PCB #123 C-13	16.5	337.9>268	30	25	337.9>266	30	25	0.5
25	PCB #118 C-13	16.54	337.9>268	30	25	337.9>266	30	25	1
26	PBDE #28 C-13	16.56	417.8>258	35	25	417.8>260	35	25	0.5
27	PCB #114 C-13	16.73	337.9>268	30	25	337.9>266	30	25	0.5
28	DDT o,p C-13	16.74	247>211	30	20	249>213	30	20	0.5
29	PCB #153 C-13	16.91	371.8>301.9	30	30	371.8>299.9	30	30	0.5
30	PCB #105 C-13	16.99	337.9>268	30	25	337.9>266	30	25	0.5
31	DDT p,p C-13	17.32	247>211	30	20	249>213	30	20	0.5
32	PCB #138 C-13**	17.41	371.8>301.9	30	30	371.8>299.9	30	30	0.5
33	PCB #126 C-13	17.57	337.9>268	30	25	337.9>266	30	25	0.5
34	PCB #167 C-13	17.93	371.8>301.9	30	30	371.8>299.9	30	30	0.5
35	Benz[a]anthracene-d12	18.22	240>240	30	15	240>236	30	30	0.5
36	Chrysene-d12	18.31	240>240	30	15	240>236	30	30	0.5
37	PCB #156 C-13	18.38	371.8>301.9	30	30	371.8>299.9	30	30	0.5
38	PCB #157 C-13	18.5	371.8>301.9	30	30	371.8>299.9	30	30	0.5
39	PCB #180 C-13	18.71	405.8>335.9	30	30	405.8>333.9	30	30	0.5
40	PBDE #47 C-13	18.77	497.7>337.9	35	25	497.7>339.9	35	25	0.5
41	PCB #169 C-13	19.15	371.8>301.9	30	30	371.8>299.9	30	30	0.5
42	Mirex C-13	19.39	277>242	30	20	277>240	30	20	0.5
43	5-methylchrysene-d3**	19.59	245>244	45	25	245>242	45	40	0.5
44	PCB #189 C-13	19.95	405.8>335.9	30	30	405.8>333.9	30	30	0.5
45	PBDE #100 C-13	20.73	575.6>415.8	35	25	575.6>417.8	35	25	0.5
46	PCB #194 C-13	20.76	439.8>369.8	30	35	439.8>367.8	30	35	0.5
47	Benzo[b,k]fluoranthene-d12	21.3	264>264	30	15	264>260	30	30	1
48	PBDE #99 C-13	21.32	575.6>415.8	35	25	575.6>417.8	35	25	0.5
49	PCB #206 C-13	21.67	475.7>405.8	30	35	475.7>403.8	30	35	0.5
50	Benzo[e]pyrene-d12	21.99	264>264	30	15	264>260	30	30	0.5
51	Benzo[a]pyrene-d12	22.14	264>264	30	15	264>260	30	30	0.5
52	Perylene-d12	22.38	264>264	30	15	264>260	30	30	0.5
53	PCB #209 C-13	22.44	509.7>439.8	30	35	509.7>437.8	30	35	0.5
54	PBDE #154 C-13	23.15	655.5>495.7	35	25	655.5>493.7	35	25	1
55	PBDE #153 C-13	24.09	655.5>495.7	35	25	655.5>493.7	35	25	1
56	PBDE #138 C-13**	25.5	655.5>495.7	35	25	655.5>493.7	35	25	1
57	Indeno[1,2,3-cd]pyrene-d12	25.59	288>288	40	15	288>284	40	40	0.5
58	Dibenzo[a,h]anthracene-d14	25.68	292>292	40	15	292>288	40	30	0.5
59	Benzo[g,h,i]perylene-d12	26.34	288>288	40	15	288>284	40	40	0.5
60	PBDE #183 C-13	27.23	733.4>573.6	35	25	733.4>575.6	35	25	1
61	Coronene-d12	31.16	312>312	70	15	312>308	70	55	0.5
62	Dibenzo[a,e]pyrene 6C13	31.32	308>308	70	15	308>306	70	40	0.5

RESULTS AND DISCUSSION

The Xevo TQ-S with APGC was evaluated as an accurate and sensitive instrument for the detection of multi-class POP compounds (PAHs, PCBs, PBDEs, and OCPs). Over 140 analytes (excluding internal standards) were targeted in this method and represent the most common congener mixes and regulated POP compounds, covering low, medium, and high boiling compounds.

The accurate detection and quantification of certain compounds, including coronene, dibenzo pyrenes, and BDE #183 can often prove challenging during traditional GC-EI-MS analysis. Despite their complex structures and higher boiling points, these compounds are readily analyzed by APGC, where excellent LODs were achieved, as shown in Table 3 on pages 5-6.

A generic extraction method was developed for all analytes using the internal standards described in Table 1. Excellent recoveries, linearity, and LODs were determined for all analytes across the six different matrices. Comparable recoveries, with satisfactory repeatability, were achieved for the analytes in all matrices, as shown in the Table 2, using the recovery of PCB #126 as a representative example.

Table 2. Excellent recoveries achieved for PCB #126 from all matrices, showing robust efficiency of the generic sample cleanup procedure.

	Sample matrix	Recovery (%)	Repeatability (%RSD)
1.	Milk	104	3.1
2.	Infant formula	106	6.1
3.	Beef	103	5.9
4.	Chicken	103	6.7
5.	Pork	106	8.9
6.	Fish	109	2.8

The validated method using TQ-S with APGC was submitted and successfully accredited in accordance with international standard ISO 17025. For ease of discussion, the method's results for multi-class analytes will be demonstrated from here on using pork meat. The results shown in Table 3 focus on analyte recoveries and repeatability at the lowest fortification level in order to demonstrate system sensitivity and robustness at trace levels. Furthermore, the linearity and limits of detection, as summarized in Table 3 are discussed.

Table 3. MRM transitions for all POPs analyzed. Satisfactory recovery and repeatability results obtained for the four classes of POP compounds at the fortified levels shown in pork. Excellent limits of detection were achieved for all analytes.

	Compound	RT min	*MRM m/z*	Cone V	Collision eV	*MRM m/z*	Cone V	Collision eV	Spiking level ($\mu g.kg^{-1}$)	Recovery %	RSD %	LOD ($\mu g.kg^{-1}$)
1	Naphtalene	6.42	128>102	55	20	128>78	55	20	1	114	14.7	0.2
2	2-methylnaphtalene	7.72	142>141	40	18	142>115	40	28	1	97	10.9	0.2
3	Acenaphtylene	9.46	152>151	65	28	152>150	65	30	0.05	114	13.4	0.02
4	Acenaphtene	9.83	154>153	40	20	154>152	40	30	0.2	96	16.0	0.04
5	PCB IUPAC #1	9.96	188>152	30	25	190>152	30	25	0.005	97	3.6	0.0006
6	Pentachlorobenzene	10.22	250>215	30	25	250>179	30	30	0.05	97	3.1	0.005
7	PCB IUPAC #3	10.76	188>152	30	25	190>152	30	25	0.005	96	7.9	0.001
8	Fluorene	10.84	166>165	35	20	166>164	35	35	0.2	92	9.6	0.05
9	PCB IUPAC #10 (#4)	11.17	222>152	30	25	224>152	30	25	0.01	99	4.4	0.001
10	PCB IUPAC #8	11.94	222>152	30	25	224>152	30	25	0.005	113	4.8	0.0009
11	BHC alpha	11.96	217>181	30	15	219>183	30	15	0.05	100	5.0	0.008
12	Hexachlorobenzene	12.11	284>214	30	35	284>249	30	30	0.05	103	10.4	0.02
13	PCB IUPAC #19	12.32	256>186	30	25	258>186	30	25	0.005	103	4.2	0.0006
14	BHC beta	12.42	217>181	30	15	219>183	30	15	0.05	96	8.2	0.01
15	BHC gamma	12.55	217>181	30	15	219>183	30	15	0.05	94	6.6	0.009
16	PCB IUPAC #18 (#17)	12.71	256>186	30	25	258>186	30	25	0.005	116	7.9	0.001
17	Phenanthrene	12.74	178>177	65	25	178>151	65	40	0.2	91	17.0	0.04
18	PCB IUPAC #15	12.75	222>152	30	25	224>152	30	25	0.005	97	5.6	0.0009
19	Anthracene	12.83	178>177	65	25	178>151	65	40	0.05	93	12.4	0.007
20	HCH delta	12.95	217>181	30	15	219>183	30	15	0.05	99	4.1	0.006
21	PCB IUPAC #54	13.29	292>220	30	25	292>222	30	25	0.005	108	6.5	0.001
22	PCB IUPAC #28 (#31)	13.52	256>186	30	25	258>186	30	25	0.05	122	4.7	0.001
23	PCB IUPAC #33	13.69	256>186	30	25	258>186	30	25	0.005	97	4.5	0.0006
24	Heptachlore	13.81	337>266	30	20	337>302	30	15	0.05	105	6.5	0.009
25	Kepone	13.81	272>237	30	20	272>235	30	20	0.05	111	3.0	0.005
26	PCB IUPAC #22	13.82	256>186	30	25	258>186	30	25	0.005	100	4.5	0.0006
27	PCB IUPAC #52	14.1	292>220	30	25	292>222	30	25	0.05	118	4.8	0.004
28	PCB IUPAC #49 (#47)	14.18	292>220	30	25	292>222	30	25	0.005	118	10.0	0.002
29	PCB IUPAC #104	14.39	325.9>256	30	25	325.9>254	30	25	0.005	112	4.3	0.0006
30	Aldrin	14.41	293>186	30	40	293>220	30	25	0.05	92	6.6	0.009
31	PCB IUPAC #44	14.45	292>220	30	25	292>222	30	25	0.05	113	2.9	0.002
32	PCB IUPAC #37	14.52	256>186	30	25	258>186	30	25	0.005	105	7.5	0.001
33	PCB IUPAC #41	14.67	292>220	30	25	292>222	30	25	0.005	118	6.2	0.001
34	PCB IUPAC #40	14.8	292>220	30	25	292>222	30	25	0.005	106	6.6	0.001
35	PCB IUPAC #74	15.04	292>220	30	25	292>222	30	25	0.05	112	3.1	0.002
36	Heptachlore epoxyde cis	15.05	353>253	30	20	353>282	30	20	0.05	100	5.8	0.008
37	Oxychlordane	15.08	387>287	30	18	387>253	30	40	0.05	103	6.0	0.009
38	PCB IUPAC #70	15.09	292>220	30	25	292>222	30	25	0.05	110	5.5	0.003
39	Fluoranthene	15.13	202>201	70	35	202>200	70	35	0.2	106	3.4	0.02
40	Heptachlore epoxyde trans	15.13	353>253	30	20	353>282	30	20	0.05	101	5.2	0.01
41	PCB IUPAC #66	15.16	292>220	30	25	292>222	30	25	0.05	112	3.9	0.002
42	PCB IUPAC #95	15.17	325.9>256	30	25	325.9>254	30	25	0.05	114	8.1	0.003
43	PCB IUPAC #155	15.39	359.8>289.9	30	30	359.8>287.9	30	30	0.005	94	5.9	0.0009
44	PCB IUPAC #60	15.44	292>220	30	25	292>222	30	25	0.05	120	6.0	0.002
45	Chlordane cis	15.44	373>266	30	25	373>301	30	20	0.05	103	4.1	0.007
46	DDE o,p	15.49	316>246	30	25	318>248	30	25	0.05	101	2.4	0.004
47	PCB IUPAC #101 (#90)	15.53	325.9>256	30	25	325.9>254	30	25	0.1	114	6.0	0.007
48	Pyrene	15.56	202>201	70	35	202>200	70	35	0.2	98	10.7	0.02
49	PCB IUPAC #99	15.62	325.9>256	30	25	325.9>254	30	25	0.005	108	13.3	0.002
50	Endosulfane I	15.65	339>160	30	20	339>196	30	20	0.05	102	7.7	0.01
51	Chlordane trans	15.69	373>266	30	25	373>301	30	20	0.05	103	4.2	0.007
52	PCB IUPAC #119	15.72	325.9>256	30	25	325.9>254	30	25	0.005	99	8.5	0.001
53	Nonachlor trans	15.77	407>300	30	25	407>263	30	30	0.05	104	7.0	0.01
54	PCB IUPAC #87	15.96	325.9>256	30	25	325.9>254	30	25	0.05	118	4.6	0.003
55	PCB IUPAC #81	15.97	292>220	30	25	292>222	30	25	0.005	107	7.6	0.002
56	DDE p,p	15.98	316>246	30	25	318>248	30	25	0.05	113	11.7	0.02
57	Dieldrine	16.07	303>232	30	22	303>268	30	18	0.05	122	5.6	0.01
58	PCB IUPAC #77	16.12	292>220	30	25	292>222	30	25	0.05	111	3.0	0.0006
59	PCB IUPAC #110	16.12	325.9>256	30	25	325.9>254	30	25	0.05	120	3.9	0.003
60	DDD o,p	16.13	235>199	30	20	237>201	30	20	0.05	90	4.7	0.006
61	PBDE #17 C-12	16.29	405.8>246	35	25	405.8>248	35	25	0.005	100	6.9	0.0006
62	PCB IUPAC #151	16.32	359.8>289.9	30	30	359.8>287.9	30	30	0.05	101	5.8	0.003
63	Endrin	16.43	380>345	30	12	380>279	30	20	0.05	116	7.2	0.01
64	PCB IUPAC #123	16.5	325.9>256	30	25	325.9>254	30	25	0.005	100	8.3	0.001
65	PCB IUPAC #149	16.51	359.8>289.9	30	30	359.8>287.9	30	30	0.05	103	9.0	0.003
66	PCB IUPAC #118	16.54	325.9>256	30	25	325.9>254	30	25	0.05	117	3.1	0.002
67	PBDE #28 C-12	16.56	405.8>246	35	25	405.8>248	35	25	0.005	106	4.9	0.0009
68	Endosulfane II	16.57	339>160	30	20	339>196	30	20	0.05	90	10.9	0.02
69	DDD p,p	16.66	235>199	30	20	237>201	30	20	0.05	92	5.4	0.008
70	PCB IUPAC #114	16.74	325.9>256	30	25	325.9>254	30	25	0.005	108	5.8	0.0009
71	DDT o,p	16.74	235>199	30	20	237>201	30	20	0.05	99	4.5	0.007
72	Nonachlor cis	16.79	407>300	30	25	407>263	30	30	0.05	105	4.2	0.0066
73	PCB IUPAC #188	16.82	393.8>323.9	30	30	395.8>325.9	30	30	0.005	106	5.4	0.0009
74	PCB IUPAC #153	16.92	359.8>289.9	30	30	359.8>287.9	30	30	0.05	114	6.6	0.004
75	PCB IUPAC #168 (#132)	16.97	359.8>289.9	30	30	359.8>287.9	30	30	0.005	106	18.8	0.003

	Compound	RT min	*MRM m/z*	Cone V	Collision eV	*MRM m/z*	Cone V	Collision eV	Spiking level ($\mu g.kg^{-1}$)	Recovery %	RSD %	LOD ($\mu g.kg^{-1}$)
76	PCB IUPAC #105	17	325.9>256	30	25	325.9>254	30	25	0.005	106	10.8	0.002
77	PCB IUPAC #141	17.15	359.8>289.9	30	30	359.8>287.9	30	30	0.05	105	2.9	0.002
78	PCB IUPAC #137	17.27	359.8>289.9	30	30	359.8>287.9	30	30	0.005	98	7.0	0.0009
79	DDT p,p	17.33	235>199	30	20	237>201	30	20	0.05	103	14.6	0.02
80	PCB IUPAC #138	17.42	359.8>289.9	30	30	359.8>287.9	30	30	0.05	112	3.9	0.003
81	PCB IUPAC #158	17.47	359.8>289.9	30	30	359.8>287.9	30	30	0.005	97	4.6	0.0006
82	PCB IUPAC #129	17.57	359.8>289.9	30	30	359.8>287.9	30	30	0.005	108	4.9	0.0009
83	PCB IUPAC #178	17.57	393.8>323.9	30	30	395.8>325.9	30	30	0.005	105	9.1	0.001
84	PCB IUPAC #126	17.58	325.9>256	30	25	325.9>254	30	25	0.005	106	8.9	0.001
85	PCB IUPAC #187	17.72	393.8>323.9	30	30	395.8>325.9	30	30	0.05	111	5.0	0.003
86	Benzo[c]phenanthrene	17.77	228>228	30	15	228>226	30	30	0.05	101	6.1	0.004
87	PCB IUPAC #183	17.82	393.8>323.9	30	30	395.8>325.9	30	30	0.005	104	14.3	0.002
88	PCB IUPAC #167 (#128)	17.94	359.8>289.9	30	30	359.8>287.9	30	30	0.01	107	3.6	0.001
89	Cyclopenta[cd]pyrene	18.25	226>225	60	40	226>224	60	45	0.05	81	6.2	0.007
90	Benz[a]anthracene	18.27	228>228	30	15	228>226	30	30	0.05	107	8.7	0.005
91	PCB IUPAC #177	18.29	393.8>323.9	30	30	395.8>325.9	30	30	0.005	104	10.1	0.002
92	PCB IUPAC #202	18.36	427.8>357.8	30	30	427.8>355.8	30	30	0.005	101	5.9	0.0009
93	PBDE #49 C-12	18.36	485.7>325.9	35	25	485.7>327.9	35	25	0.005	105	5.2	0.0009
94	Chrysene	18.37	228>228	30	15	228>226	30	30	0.05	107	16.7	0.01
95	PCB IUPAC #171	18.38	393.8>323.9	30	30	395.8>325.9	30	30	0.005	101	4.3	0.0006
96	PCB IUPAC #156	18.39	359.8>289.9	30	30	359.8>287.9	30	30	0.005	106	6.6	0.001
97	PCB IUPAC #157	18.5	359.8>289.9	30	30	359.8>287.9	30	30	0.005	108	7.1	0.001
98	PCB IUPAC #201	18.52	427.8>357.8	30	30	427.8>355.8	30	30	0.005	103	6.0	0.0009
99	PCB IUPAC #180	18.72	393.8>323.9	30	30	395.8>325.9	30	30	0.05	113	4.4	0.003
100	PCB IUPAC #193	18.77	393.8>323.9	30	30	395.8>325.9	30	30	0.05	115	4.8	0.0009
101	PBDE #47 C-12	18.78	485.7>325.9	35	25	485.7>327.9	35	25	0.05	108	3.7	0.003
102	PCB IUPAC #191	18.86	393.8>323.9	30	30	395.8>325.9	30	30	0.005	96	6.7	0.0009
103	PCB IUPAC #200	19	427.8>357.8	30	30	427.8>355.8	30	30	0.005	104	6.8	0.001
104	PBDE #66 C-12	19.11	485.7>325.9	35	25	485.7>327.9	35	25	0.005	98	7.3	0.001
105	PCB IUPAC #169	19.15	359.8>289.9	30	30	359.8>287.9	30	30	0.005	102	6.8	0.0009
106	PCB IUPAC #170	19.34	393.8>323.9	30	30	395.8>325.9	30	30	0.005	99	9.2	0.002
107	Mirex	19.4	272>237	30	20	272>235	30	20	0.05	109	2.1	0.003
108	PCB IUPAC #199	19.53	427.8>357.8	30	30	427.8>355.8	30	30	0.005	97	5.9	0.0009
109	5-methylchrysene	19.62	242>241	45	25	242>239	45	40	0.05	99	4.8	0.005
110	PCB IUPAC #203	19.64	427.8>357.8	30	30	427.8>355.8	30	30	0.005	95	9.9	0.001
111	PBDE #77 C-12	19.67	485.7>325.9	35	25	485.7>327.9	35	25	0.005	95	12.7	0.0003
112	PCB IUPAC #189	19.96	393.8>323.9	30	30	395.8>325.9	30	30	0.005	101	7.3	0.001
113	PCB IUPAC #208	20.29	463.7>393.8	30	35	463.7>391.8	30	35	0.005	106	3.8	0.0006
114	PCB IUPAC #195	20.32	427.8>357.8	30	30	427.8>355.8	30	30	0.005	100	3.9	0.0006
115	PBDE #100 C-12	20.74	563.6>403.8	35	30	563.6>405.8	35	30	0.005	109	17.0	0.003
116	PCB IUPAC #194	20.77	427.8>357.8	30	30	427.8>355.8	30	30	0.005	111	6.3	0.001
117	PCB IUPAC #205	20.9	427.8>357.8	30	30	427.8>355.8	30	30	0.005	96	5.2	0.0009
118	PBDE #119 C-12	20.94	563.6>403.8	35	30	563.6>405.8	35	30	0.005	101	5.6	0.0006
119	PBDE #99 C-12	21.33	563.6>403.8	35	30	563.6>405.8	35	30	0.05	107	2.9	0.004
120	Benzo[b,j,k]fluoranthene	21.34	252>252	30	15	252>250	30	30	0.15	98	8.1	0.01
121	7,12-Dimethylbenzo[a]anthracene	21.38	256>241	45	25	256>239	45	40	0.05	100	3.8	0.007
122	PCB IUPAC #206	21.68	463.7>393.8	30	35	463.7>391.8	30	35	0.005	103	4.4	0.0006
123	Benzo[e]pyrene	22.06	252>252	30	15	252>250	30	30	0.05	93	11.9	0.004
124	Benzo[a]pyrene	22.2	252>252	30	15	252>250	30	30	0.05	93	12.9	0.005
125	PCB IUPAC #209	22.45	497.7>427.8	30	35	497.7>425.8	30	35	0.005	105	5.3	0.0009
126	Perylene	22.45	252>252	30	15	252>250	30	30	0.05	100	6.4	0.007
127	PBDE #85 C-12	22.51	563.6>403.8	35	30	563.6>405.8	35	30	0.005	100	5.0	0.0009
128	PBDE #154 C-12	23.16	643.5>483.7	35	25	643.5>481.7	35	25	0.01	101	6.4	0.002
129	3-Methylcholanthrene	23.44	268>252	50	40	268>253	50	30	0.05	105	11.6	0.006
130	PBDE #153 C-12	24.1	643.5>483.7	35	25	643.5>481.7	35	25	0.01	102	8.0	0.002
131	PBDE #138 C-12	25.51	643.5>483.7	35	25	643.5>481.7	35	25	0.01	100	5.7	0.002
132	Indeno[1,2,3-cd]pyrene	25.66	276>276	40	15	276>274	40	40	0.05	96	10.6	0.005
133	Dibenzo[a,h]anthracene	25.79	278>278	40	15	278>276	40	30	0.05	94	3.2	0.005
134	Benzo[g,h,i]perylene	26.42	276>276	40	15	276>274	40	40	0.05	103	13.7	0.007
135	Anthanthrene	26.83	276>276	40	15	276>274	40	40	0.05	66	8.3	0.006
136	PBDE #183 C-12	27.25	721.4>561.6	35	35	721.4>563.6	35	35	0.01	108	8.7	0.003
137	Dibenzo[a,l]pyrene	30.14	302>302	70	15	302>300	70	40	0.05	91	5.4	0.008
138	Coronene	31.25	300>300	70	15	300>298	70	55	0.05	104	4.9	0.01
139	Dibenzo[a,e]pyrene	31.33	302>302	70	15	302>300	70	40	0.05	83	5.7	0.007
140	Dibenzo[a,i]pyrene	31.79	302>302	70	15	302>300	70	40	0.05	95	5.2	0.007
141	Dibenzo[a,h]pyrene	32.05	302>302	70	15	302>300	70	40	0.05	86	12.6	0.005

Sensitivity and selectivity

Given the softer ionization associated with APGC, more abundant molecular ions can be observed compared with traditional EI spectra. During method development stages, all of the analytes showed better sensitivity for the molecular ion by charge transfer (in dry source) in comparison with protonation, given the electronegativity associated with chlorinated and brominated compounds.

The increased sensitivity observed for many POP analytes, in comparison with traditional GC-EI-MS methods can be seen in Figure 1. Here the signal-to-noise ratio (S/N) determined for BDE #17 and #28 shows a significant increase when analyzed by APGC. This further reduces matrix loading, thus improving liner, column life, and instrument robustness, while reducing instrument maintenance.

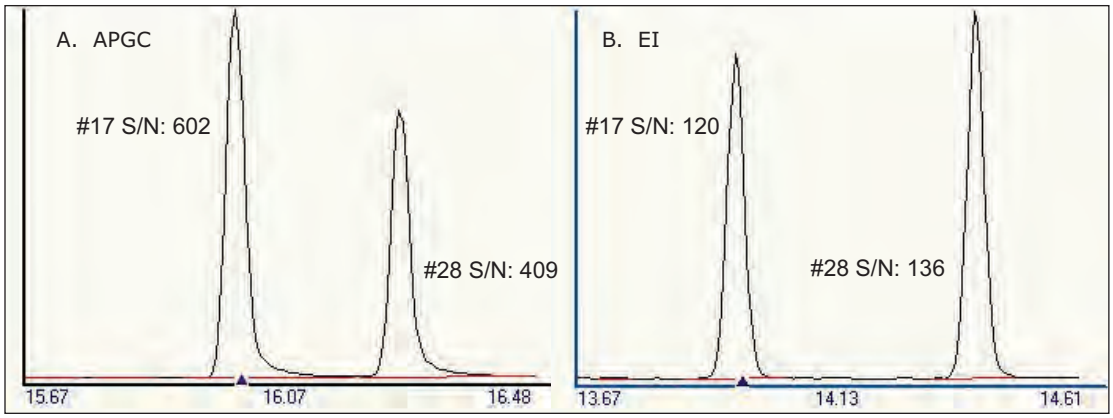

Figure 1. Increased sensitivity achieved for BDE #17 and #28 in extracted pork by A. APGC (2 pg on column) compared with traditional B. EI (20 pg on column).

The medium and high concentration replicates were combined (n= 18) to allow for an averaged and more representative statistical analysis of the method recovery and repeatability for each matrix. Further statistical analysis was completed to allow for validation of a robust method at low concentration levels. To this end, the method recovery and repeatability were determined separately for all analytes in each matrix at the lowest level of fortification (0.2 x LL, where n= 9). These results are shown for pork matrix in Table 3.

Limits of detection

The detection of POPs in food and environmental samples are challenging, because of their ubiquitous presence and the increasingly low detection levels required to meet regulatory limits in complex matrices. However, the analysis of PBDEs, PCBs, PAHs, and OCPs can be achieved below all the required levels of detection using APGC-MS.

The limits determined for all of the analytes are summarized in Table 3, where concentrations of <1 µg.kg^{-1} were achieved for all analytes in pork extract. The excellent sensitivity achieved is further demonstrated in Figure 2, where the S/N ratio was determined for 50 fg on column. An example of each class of POP analyzed was compared to its matrix blank, thus demonstrating the selectivity afforded.

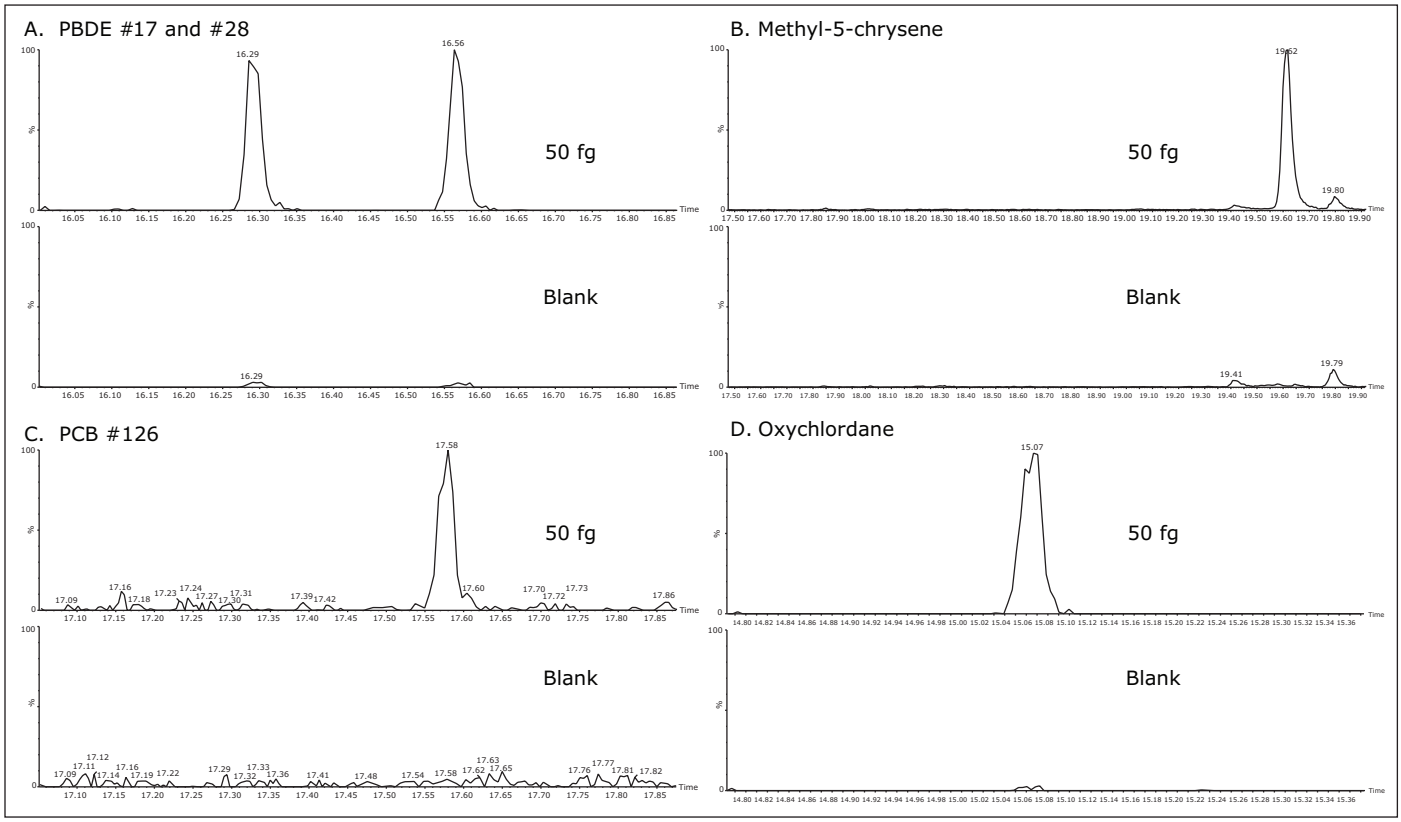

Figure 2. Excellent sensitivity and selectivity determined for 50 fg on column for: A. PBDE #17 and #28; B. methyl-5-chrysene; C. PCB #126; and D. oxychlordane, in comparison to a blank extracted pork sample.

The developed method allowed for excellent repeatability for the multi-class components fortified at low levels in a variety of matrices. This is well demonstrated by the validation data shown in Table 3, where excellent recoveries and method repeatability are shown for all analytes fortified in pork meat (n= 9) at levels between 50 to 1000 $ng.kg^{-1}$.

Using the optimized generic sample preparation and cleanup method, the percentage recoveries ranged from 65% to 122% in pork matrix. Percentage relative standard deviations (%RSD) were found to be <20% for all analytes. This is an acceptable level for multi-residue analysis in complex matrices, showing low variance for all of the PBDEs, PCBs, PAHs, and OCPs when spiked at parts per trillion (ppt, equating to $ng.kg^{-1}$) levels in the complex matrix.

Linearity

Linearity was investigated for all analytes utilizing the internal standards described in Table 1. Good correlation was achieved (R^2>0.99) over a satisfactory working range of 2 to 25 $\mu g.kg^{-1}$. This working range was deemed most appropriate, allowing for the accurate quantification for all analytes at legislated levels where applicable. An example of the calibration curves achieved utilizing the internal standard is provided in Figure 3 for each class of POP in pork meat.

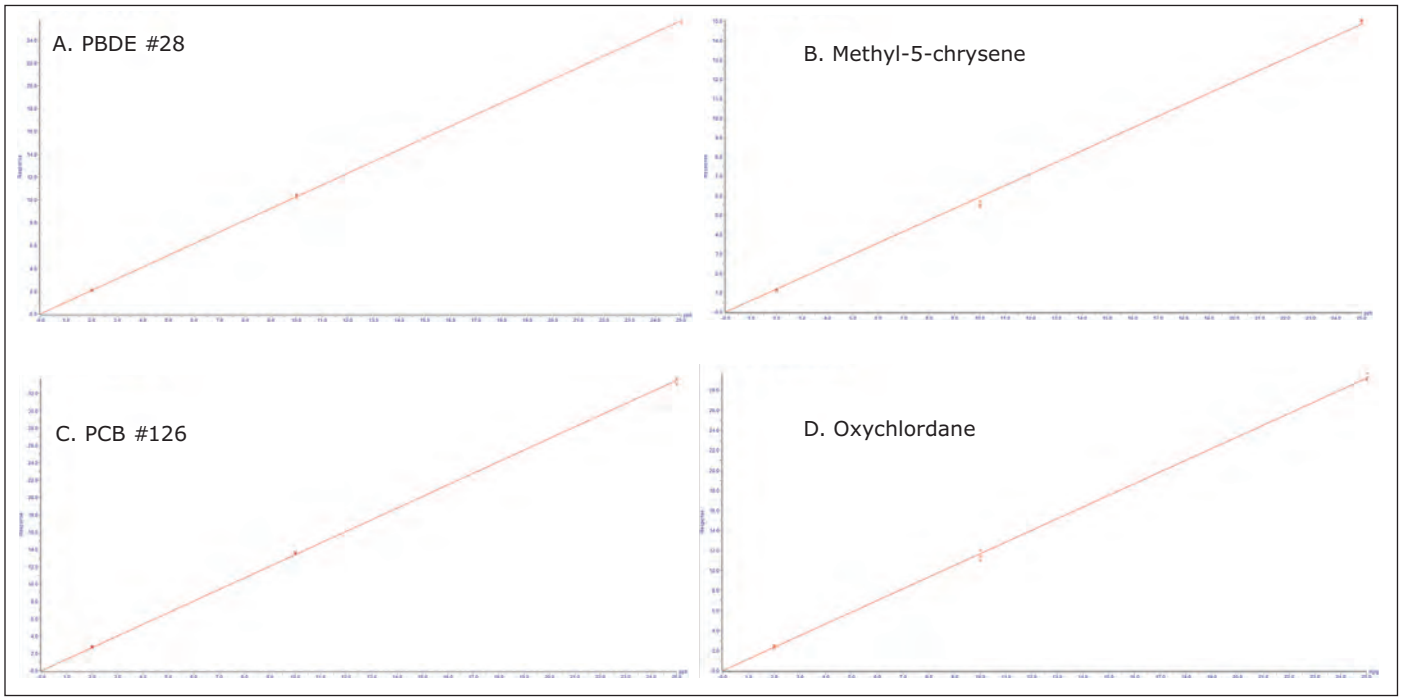

Figure 3. Example of excellent calibration curve correlation for each of the four POP classes over the concentration range 2 to 25 $\mu g.kg^{-1}$ in pork meat for: A. polybrominated diphenyl ethers; B. polycyclic aromatic hydrocarbons; C. polychlorinated biphenyls; D. organochlorine pesticides.

CONCLUSIONS

Time-consuming and costly analyses are a major drain on food and environmental testing laboratories, where multi-analyte methods are preferred for efficient use of resources. While the robust Xevo TQ-S can be coupled with UPLC to provide sensitive analysis of LC-amenable compounds, this work shows the ability of the Xevo TQ-S to analyze multi class POPs by atmospheric pressure gas chromatography with excellent robust sensitivity.

The optimization of a single cleanup method for a variety of analytes has been shown to achieve satisfactory recoveries, while allowing the Xevo TQ-S with APGC to quantify analytes below the regulatory limit. Taking pork as an example, excellent recoveries, in the range of 66% to 122% were determined, where the repeatability was <20% for all 141 POP analytes.

This validated and accredited method has been implemented by the MAPAQ for the routine analysis of a multitude of meats, fish, milk, and infant formula to ensure consumer safety in Quebec, Canada. When compared with traditional GC-EI-MS methods, increased sensitivity, less maintenance, and routine cleaning has been required for the Xevo TQ-S with APGC, further improving laboratory efficiency.

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