Waters™

Applikationsbericht

Non-Targeted Screening of Extractables and Leachables in E-Cigarettes Using UPLC and GC Coupled to QTof-MS

Narendra Meruva, Baiba Cabovska, Dimple D. Shah, Kari L. Organtini, Gareth E. Cleland

Waters Corporation

Abstract

This study demonstrates an integrated workflow for targeted and non-targeted screening using UPLC and GC on a single MS platform with UNIFI informatics for extractable and leachable screening in e-cigarettes, food, cosmetics, and pharmaceutical packaging applications.

Benefits

- Comprehensive characterization of extractables and leachables using UPLC and GC which can be configured to a single QTof-MS
- Accurate mass screening using MS^E data acquisition combined with scientific libraries streamlines identification of potential extractables
- Sample comparison workflows and structure elucidation toolkits for characterization of unknown compounds
- Metabolite ID workflow can be used to evaluate possible degradation or transformation products of formulation components

Introduction

Characterization of extractables and leachables is essential for ensuring the safety, quality, and efficacy of inhalation tobacco products such as e-cigarettes. The initial step for characterizing extractables from e-cigarettes involves targeted screening where you analyze the extract and quantify against known impurity standards. This is a well-established process that can be performed using analytical techniques such as GC-MS, LC-MS/MS and ICP-MS. However the finished products (e-liquids, refill cartridges, and e-cigarette aerosol) may have impurities present from the starting materials and other packaging and device components that need to be further evaluated by non-targeted screening analysis.

E-cigarette regulations are still evolving due to a lack of scientific information and lack of product quality and safety standards. Both the US FDA regulation and the revised EU Tobacco Products Directive (TPD2; 2014/40/EU) subject e-cigarette manufacturers to product and ingredient disclosures and good manufacturing practices to ensure e-cigarette products are appropriate for the protection of the public health.[1,2] In the UK, the MHRA (Medicines and Healthcare Products Regulatory Agency) regulates e-

cigarettes as nicotine delivery devices and requires manufacturers to provide complete quality information for licensing e-cigarette devices including the composition of the e-cigarette device, the plastic, polymer, and metal components used, the quality of the nicotine and excipients, data from extractables and leachables studies, and product stability data during use, and shelf-life.[3]

In this study, the various components of an e-cigarette device (end caps, mouth piece, gauze, heating element, and flavor formulation) were extracted individually and subjected to non-targeted high resolution screening using UPLC and GC which can be configured to the same QTof-MS. Accurate mass data for precursor and fragment ions was acquired using alternating high and low collision energy states (MS^E) across the full analytical mass range. Data from the sample component extracts was compared to the reagent blank to determine differences and identify potential extractables. In this application note, we describe a workflow on how non-targeted screening for extractables and leachables testing can be performed in e-cigarettes. The workflow demonstrated here is also applicable to nontargeted screening for extractables and leachables in packaging for food, cosmetics, and pharmaceuticals.

Experimental

The various components of a closed system e-cigarette cartridge (outer and inner end caps, mouth piece, gauze with flavor formulation, paper wrap, and metal shell) were extracted separately using isopropanol solvent for 30 minutes and subjected to non-targeted high resolution screening using UPLC and GC coupled to QTof-MS. As part of the batch QC analysis, Waters Extractables and Leachables Screening Standard [p/n: 186008063], that includes 18 common polymer additives, was used to evaluate and benchmark the high resolution UPLC-QTof-MS system. The Extractables and Leachables Screening Standard covers a mass range of up to 1176 Da, supporting both positive and negative ionization modes.

UPLC conditions

UPLC system:	ACQUITY UPLC I-Class
Column:	ACQUITY UPLC BEH C_{18}, 130Å, 1.7 µm, 2.1 × 100 mm
Column temp.:	45 °C

Sample temp.:	4 °C
Mobile phase A:	10 mM ammonium acetate (pH 5.0) in water
Mobile phase B:	10 mM ammonium acetate (pH 5.0) in water
Flow rate:	0.45 ml/min
Needle wash:	50:50 water:methanol (v/v)
Syringe purge:	10:90 methanol:water (v/v)
Total run time:	17 min
Injection volume:	10 µL

Gradient:

Time (min)	%A	%B
0.00	98	2
0.025	98	2
12.25	1	99
13.00	1	99
13.01	98	2
17.00	98	2

MS (ESI) conditions

MS system	Xevo G2-XS QTof
Capillary voltage	0.8 kV
Sampling cone	20.0
Source temp.	120 °C
Source offset	80
Carrier gas	Nitrogen
Cone gas flow	50 L/Hr
Desolvation gas flow	1000 L/Hr
Acquisition range	50–1200 *m/z*
Scan time	0.25 sec
Lockmass	Leucine enkephalin (556.2771 *m/z*)

GC Conditions

GC system:	A7890 (with APGC Interface)
Column:	DB-5MS 0.25 µm, 30 m × 0.25 mm
Desolvation temp.:	550 °C
Flow rate:	1.2 mL/min
Initial temp.:	35 °C (1.6 min)
Ramp:	25 °C/min

Final temp.:	320 °C (7 min)
Run time:	20 min
Inlet mode:	Splitless
Inlet type:	Multimode
Temp.:	280 °C
Injection volume:	1 µL
Make-up gas:	Nitrogen
Make-up gas flow:	250 mL/min
Transfer line temp.:	310 °C

MS (API) conditions

QTof System:	Xevo G2-XS QTof MS (with APGC interface)
Corona current:	3.0 µA
Sampling cone:	20.0
Source temp.:	120 °C
Source offset:	80
Cone gas flow:	175 L/Hr
Auxiliary gas flow:	50 L/Hr

Acquisition range:	50–1200 *m/z*
Scan time:	0.25 sec
Lockmass	Siloxane bleed (281.0517 *m/z*)

Data acquisition and processing

Accurate mass data from both the GC and UPLC–QTof-MS analysis of the e-cigarette component extracts were acquired and processed using the UNIFI Scientific Infomation System.

Results and Discussion

The Xevo G2-XS QTof-MS couples to either UPLC or GC to provide a full system solution for chemical profiling. Accurate mass data from both the GC and UPLC-QTof-MS analysis of e-cigarette component extracts were acquired and processed using the extractables and leachables workflow in the UNIFI Scientific Information System. Precursor and fragment ions were acquired simultaneously using alternating low- and high-collision energy states (MS^E) across the full analytical mass range. Potential candidate markers were screened against a library of known extractables and leachables compounds in UNIFI, and automatically interrogated using multiple matching criteria including accurate mass for precursor and fragment ions, adducts, and isotopic fit.

The GC-QTof-MS profiles of e-cigarette component extracts are shown in Figure 1. Potential extractables were short-listed based on the following criteria: detector response >1000, mass error ± 5 ppm and the number of expected fragments detected >0. The established UNIFI workflow utilizes accurate mass precursor and fragment ion data, and applied criteria to simplify data review and facilitate the decision-making process. It allows analysts to evaluate complex data in a more efficient way and enables rapid identification of known and unknown compounds.

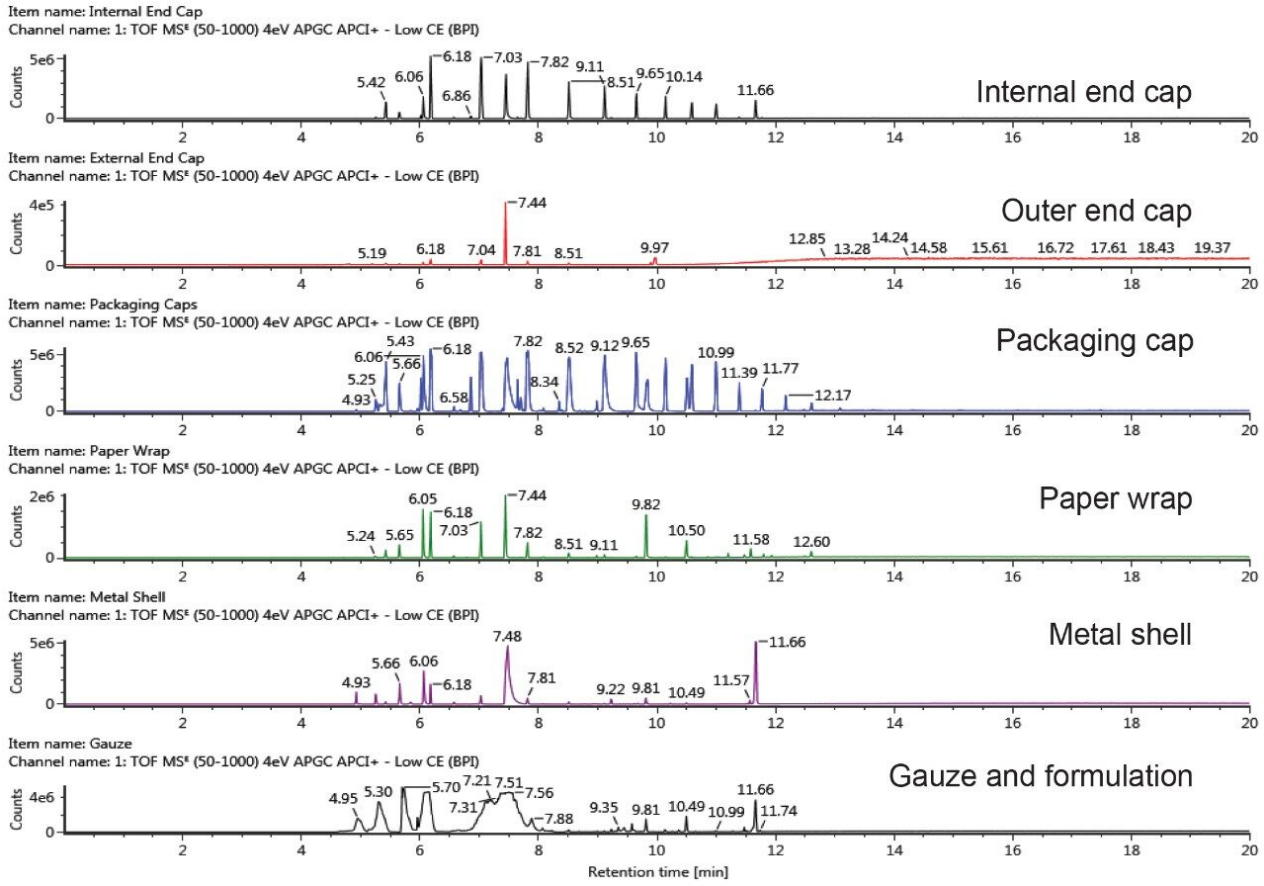

Figure 1. GC-QTof-MS profiles of e-cigarette component extracts.

Figure 2 exhibits the identification of dibutyl phthalate (DBP), a common plasticizer, in the internal end cap, metal shell, and gauze extracts using GC-QTof-MS analysis. The DBP peak had a high detector response (>11,000) in the component extracts compared to the solvent blank, one identified fragment ion, and a low measured mass error (<2.5 ppm). The migration of DBP across the internal end cap, metal shell, and gauze is possible as these components come in contact with each other in the e-cigarette cartomizer assembly.

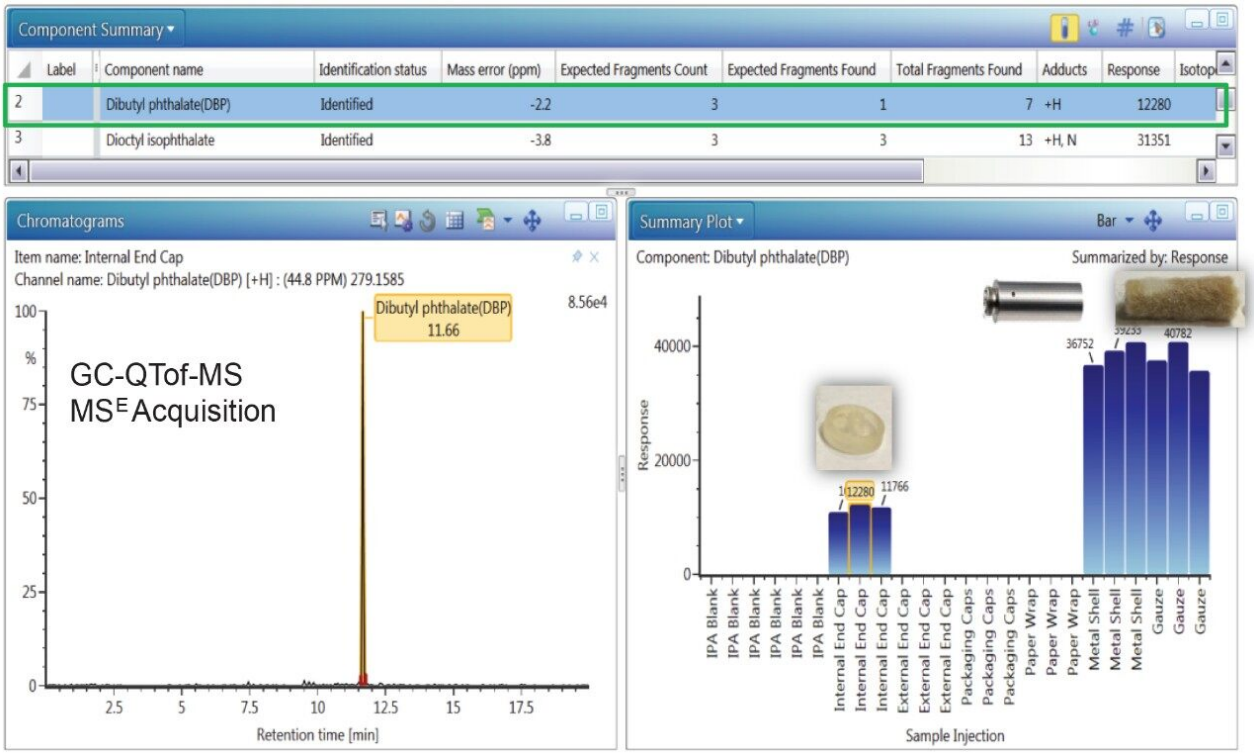

Figure 2. Identification of dibutyl phthalate (DBP) in the internal end cap, metal shell, and gauze using GC-QTof-MS.

Figure 3 shows the identification of HMBTAD, a light stabilizer in the internal end cap, metal shell, and gauze extracts using UPLC-QTof-MS analysis. The HMBTAD peak had a high detector response (>42,000), low mass error (<1.5 ppm) and was not identified in solvent blanks. The relative levels of HMBTAD are higher in the gauze containing the flavor formulation, potentially to increase the product shelf-life stability.

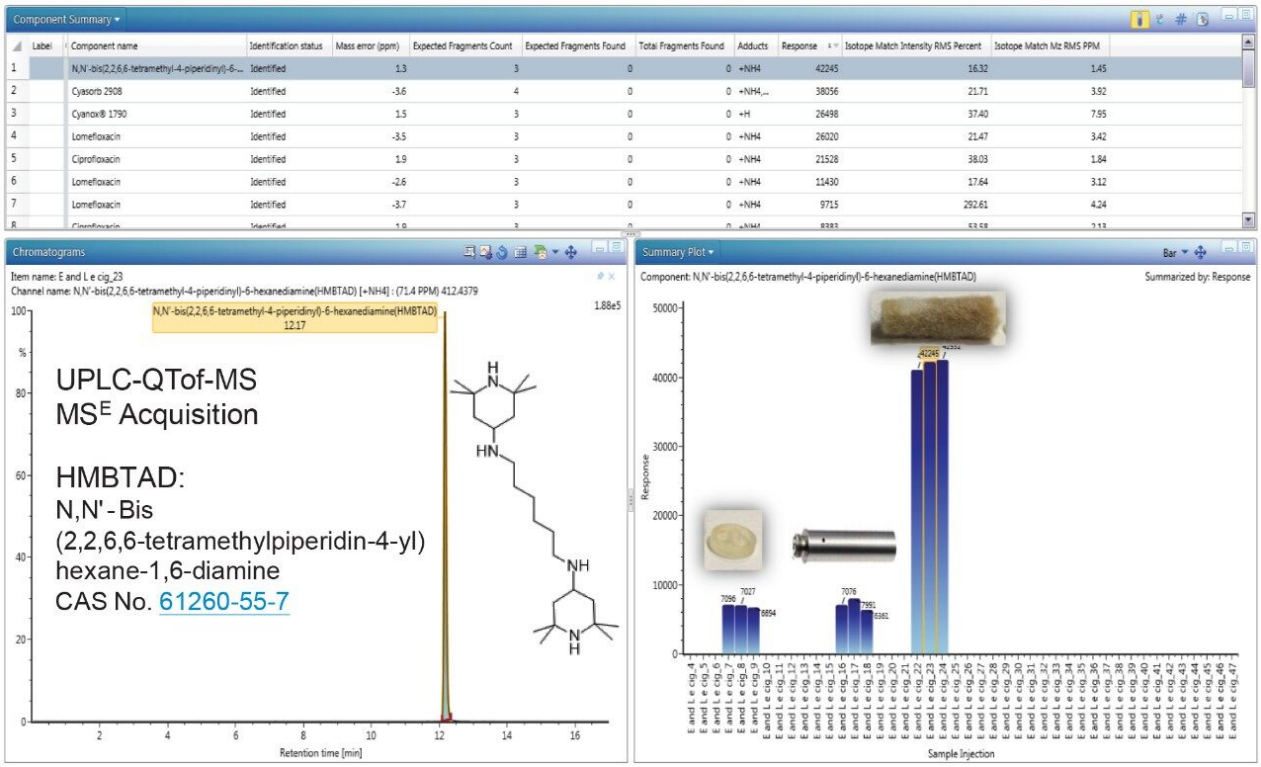

Figure 3. Identification of HMBTAD in inner end cap, metal shell, and gauze using UPLC-QTof-MS.

Table 1 lists the potential extractables detected in various e-cigarette component extracts analyzed by GC-QTof-MS and UPLC-QTof-MS. These compound identifications are based on the targeted match between the experimental data and the UNIFI Scientific Library for the accurate mass precursor and fragment ions, low mass error (± 5 ppm) and relatively high detector response (>1000).

Analysis	Extractables ID	Function	Internal end cap	Outer end cap	Packaging cap	Paper wrap	Metal shell	Gauze
GC-QTof-MS	Dibutyl phthalate (DBP)	Plasticizer	✓				✓	✓
	Octadecanoic acid	Surfactant/ softening agent			✓	✓		✓
	Dioutyl sebacate	Plasticizer			✓	✓		
	4-methyl benzophenone (4-MBP)	Stabilizing agent			✓	✓	✓	✓
	Sorbic acid	Food preservative					✓	✓
	N,N-Dimethyl-p-phenylenediamine	Polymer additive				✓	✓	
UPLC-QTof-MS	HMBTAD	Light stabilizer	✓				✓	✓
	Disperse red 11	Dye			✓			
	Uvinul 120	Anti-oxidant			✓			
	Irgafos 168	Light stabilizer			✓			

Table 1. Tentative identifications of potential extractables using UPLC-GC-QTof-MS analysis.

Conclusion

Comprehensive characterization of extractables and leachables requires evaluation using multiple chromatographic techniques (UPLC and GC), multiple modes of ionization, and an integrated informatics workflow (UNIFI). Accurate mass screening using MS^E data acquisition, combined with scientific libraries can be used to automatically identify target components.

UNIFI's sample comparison and elucidation toolsets are useful for quickly identifying known targets and characterizing unknown compounds. A metabolite identification workflow can be used to evaluate possible degradation or transformation products of formulation components in e-cigarette products. This study demonstrates an integrated workflow for targeted and non-targeted screening using UPLC and GC on a single MS platform with UNIFI informatics for extractable and leachable screening in e-cigarettes, food, cosmetics, and pharmaceutical packaging applications.

References

1. FDA Deeming Regulation (May 2016) – FDA's New Regulations for E-Cigarettes, Cigars, and All Other Tobacco
   Products. https://www.fda.gov/tobaccoproducts/labeling/rulesregulationsguidance/ucm394909.htm.
2. EU Tobacco Products Directive (TPD2;
   2014/40/EU) https://ec.europa.eu/health/sites/health/files/tobacco/docs/dir_201440_en.pdf.
3. Medicines and Healthcare Products Regulatory Agency (2016). E-cigarettes: Regulations for Consumer Products. Relevant guidance documents available via https://www.gov.uk/guidance/ecigarettes-regulations-for-consumer-products.

Featured Products

ACQUITY UPLC I-Class PLUS System <https://www.waters.com/134613317>

Waters Atmospheric Pressure Gas Chromatography (APGC) <https://www.waters.com/10100362>

Xevo G2-XS QTof Quadrupole Time-of-Flight Mass Spectrometry <https://www.waters.com/134798222>

UNIFI Scientific Information System <https://www.waters.com/134801359>

720006387, September 2018