

응용 자료

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Abstract

In this application note, pyrolysis gas chromatography coupled to high-resolution mass spectrometry (HRMS) is used with soft ionization to analyze virgin and recycled plastics. Applying statistical techniques to the acquired data, differences between the plastics can be found. These differences are subsequently characterized using the accurate mass of the molecular ions together with the fragmentation data.

Benefits

- Simple sample preparation with material added directly to pyrolysis sample cups
- Soft ionization using APGC allows for molecular ions to be detected from which elemental compositions are derived to aid compound identification and confirmation
- Identification of unique markers to distinguish between different samples through statistical techniques

Introduction

With the implementation of strategies in the EU to move towards a circular plastic economy, there is a drive to reduce or reuse plastic waste leading to the adoption of recycled and reclaimed materials into different industrial applications.¹ This includes the development of innovative materials for plastic production to increase

sustainability, and the improvement of the economics and quality of plastic recycling. To maintain value for use in high-performance applications, there is a greater need to be able to characterize these recycled materials to ensure they are safe to use.

In the field of polymer research, pyrolysis coupled to gas chromatography-mass spectrometry (GC-MS) has been extensively used and is becoming increasingly prominent for materials such as plastics.² However, the high energy of electron impact (EI) ionization can lead to insufficient sensitivity and selectivity for undertaking full characterization. Consequently, the identification of additives and potential contaminants is often not possible by this technique.

Pyrolysis-GC with soft ionization, together with high-resolution mass spectrometry (HRMS) is a complementary technique used in this field to help address some of these limitations. Atmospheric Pressure Gas Chromatography (APGC) is an atmospheric pressure chemical ionization technique, which uses a corona discharge enabling softer ionization. This results in molecular ion detection which can help with the confirmation of a molecular formula for identification. APGC can be coupled to a quadrupole time-of-flight mass spectrometer (QToF MS) which can acquire data in MS^E mode, whereby both low and high collision energy spectra are simultaneously acquired.³ Using this technique, the accurate mass of both precursor and fragment ions are available, both of which aid structural elucidation and, ultimately, compound identification.⁴ To further understand the differences between virgin and recycled materials, statistical analyses can be used in conjunction with the data acquired from these analytical instruments.⁵

In this study, an analytical workflow was developed to analyze virgin and recycled industrial plastics (Figure 1). The recycled plastics were regrinds from reclaimed plastic off cuts. Simple steps were undertaken to characterize these complex materials starting with data acquisition using Pyrolysis-APGC coupled to a QToF MS in MS^E mode. The acquired data were screened against a library to discover trends across the samples. To understand the differences between the virgin and recycled materials, statistical analyses of the data were used. The chemical markers found to be either unique to, or elevated in, each sample were characterized using structural elucidation tools.



Figure 1. The analytical approach used for the analysis of virgin vs. recycled plastics, and the determination of chemical markers responsible for differences.

Experimental

Sample Description

Samples were prepared from a selection of recycled pellets, virgin pellets, and molded gears made from PrestaAMID™, which consists of 95% high-molecular-weight polyamide (thyssenkrupp Presta, Liechtenstein). The samples were weighed to approximately 0.2 mg and loaded into stainless steel sample cups with a plug of quartz wool. Triplicates of each sample were prepared, and sample cups filled with quartz wool alone were used as blanks. The sample list was randomized before the samples were placed into the autosampler.

Sample type	Percentage recycled	Label
Recycled pellet	0%	0% RP
Recycled pellet	100%	100% RP
Recycled gear	0%	0% RG
Recycled gear	100%	100% RG

Table 1. Sample description and label.

Pyrolysis Conditions

Pyrolyzer: EGA/PY-3030D (FrontierLab)

Interface temp.: 320 °C

Pyrolyzer temp.: 600 °C

Analysis type: Single-shot analysis

GC Conditions

GC system: Agilent 7890

Inlet mode: Split/Splitless

Split ratio: 50:1

Split flow: 50 mL/min

Inlet temp.: 310 °C

GC interface temp.: 300 °C

Column: Rtx-5MS, 30 m x 0.25 mm x 0.25 µm, RESTEK

Column flow: 1.4 mL/min

Septum purge flow: 3 mL/min

Oven gradient: 45 °C for 5 min, ramp to 330 °C at a rate of 20 °C/min, final hold for 18 min

Total GC run time: 37.25 min

MS Conditions

MS system: Xevo™ G2-XS QTof*

Ionization mode: APGC™

Corona current: 3 µA

Sampling cone: 40 V

Source temp.: 150 °C

Mass range: m/z 50–1200

Scan time: 0.2 s

Cone gas: 150 L/h

Auxillary gas: 350 L/h

MS^E collision energy: Low energy: 6 V; High energy: 15–35 V

**(Equivalent or better performance is expected with the Xevo G3 QTof.)*

Data Management

Data were acquired using Waters™ MassLynx™ 4.2. Software. Data processing and reporting were performed using the UNIFI™ application within the waters_connect™ platform. All statistical analyses were undertaken with EZInfo™ 3.0.

Results and Discussion

Base peak intensity (BPI) pyrograms were obtained for all samples, and examples of BPIs for the 100% recycled pellets and 0% recycled pellets are shown in Figure 2. The pyrograms demonstrate the complexity of the materials.

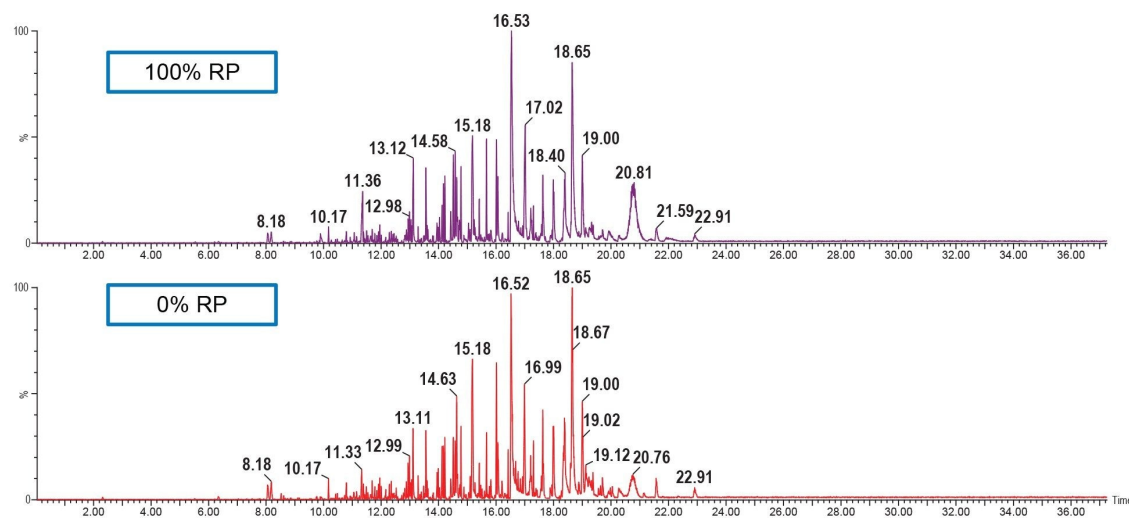


Figure 2. Base peak intensity (BPI) pyrograms from full scan data of 100% (top) and 0% (bottom) recycled pellets.

A library containing typical pyrolyzates of these types of polymer was created by importing .mol files into the UNIFI application.^{3,6} The samples were screened against this library for accurate mass matches to evaluate any trends across the dataset. Figure 3, for example, shows that 6-acetamido-N-(5-cyanopentyl) hexanamide was detected in each sample of the 0% and 100% recycled gears. This is a tentative identification because the match was made based on the accurate mass alone.

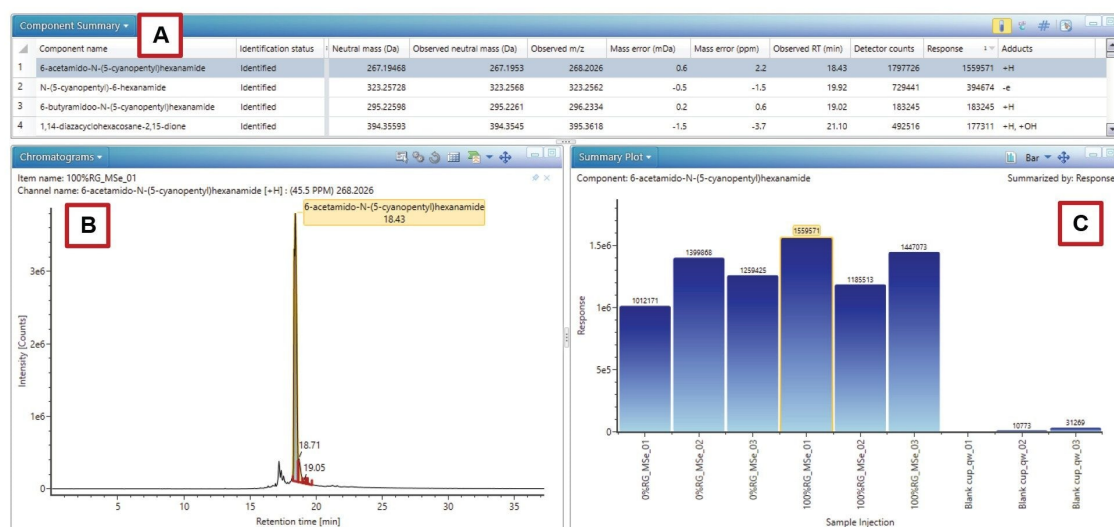


Figure 3. Screening workflow in UNIFI highlighting the results for 6-acetamido-N-(5-cyanopentyl)hexanamide. [A] Component summary table. [B] Extracted ion chromatogram for 6-acetamido-N-(5-cyanopentyl)hexanamide. [C] Trend plot of the response for this compound across all injections.

Due to the complexity of the pyrograms, to find the differences between the virgin and the recycled materials, the data were transferred to EZInfo, and Principal Component Analysis (PCA) was performed. Figure 4A shows the data from the blanks, 0%, and 100% recycled gear samples in a PCA scores plot. There is clear separation between the three groups of samples, which highlights that there are differences between the samples.

To establish the markers responsible for the differences between the 0% and 100% recycled gears, an Orthogonal Projections to Latent Structures Discriminant Analysis (OPLS-DA) was performed on data from these groups of samples. The resulting S-Plot is shown in Figure 4B, and the markers highlighted in the red regions are up-regulated in one of the sample types. These markers were selected and transferred back into the UNIFI application for further investigation.

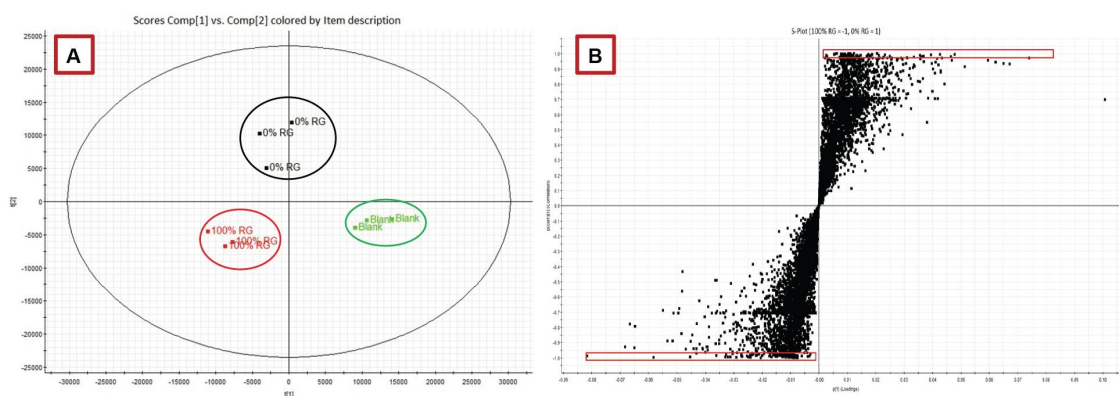


Figure 4. [A] PCA scores plot of the blanks, 0%, and 100% recycled gears showing clear separation. [B] S-plot from the OPLS-DA of the 0% vs 100% recycled gears, indicating statistically significant markers for the virgin (upper right) and the 100% recycled (lower left) material.

The selected markers were investigated using the Discovery Tool in UNIFI's elucidation toolkit.⁷ Since the pyrolysis data were collected in MS^E mode, where high and low collision energies are alternated, the accurate mass of both precursor and fragments ions were available for the interpretation of each marker.

The Discovery Tool combines elemental composition determination with database searching and *in silico* fragmentation. The elemental composition calculator determines the most likely chemical formulae for a precursor ion. The i-FIT algorithm is used to rank formulae by the likelihood that the theoretical isotope pattern of the formula matches the isotope pattern in the measured spectrum. Each elemental composition that explains a given precursor ion is automatically submitted to a ChemSpider database search which returns prospective compounds, together with their structures, for a given composition. The structure of each compound is automatically submitted to an *in silico* fragmentation algorithm and the m/z values of the theoretical fragments are compared to m/z values of the high energy ions of the marker. Figure 5 demonstrates the information displayed by the Discovery Tool for an elucidated marker.

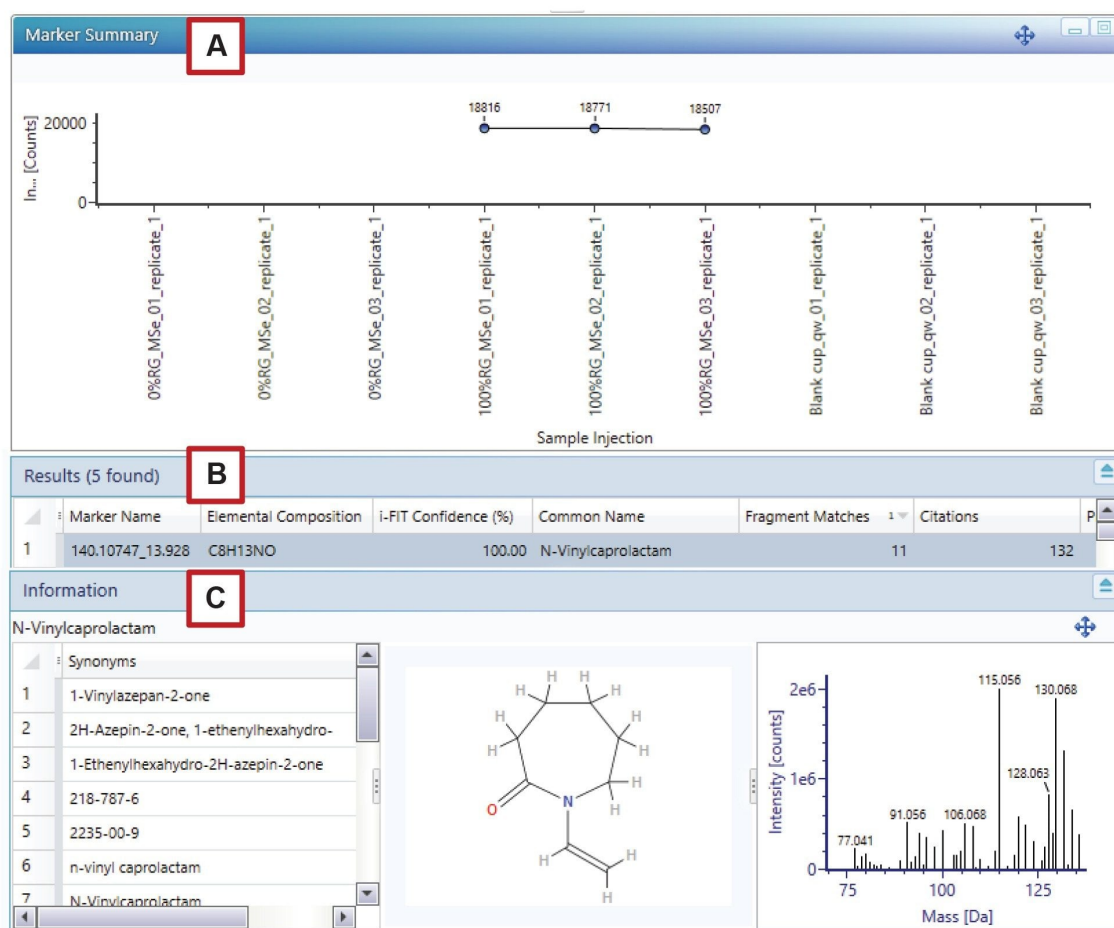


Figure 5. The information displayed by the Discovery Tool for the putative identification of a marker unique to the 100% RG sample as N-vinylcaprolactam at retention time 13.93 minutes with protonated m/z 140.1075 ($[C_8H_{13}NO]^+$ mass accuracy 0.2 ppm). [A] Marker abundance (response) across all samples. [B] Results for this marker; the predicted elemental composition, i-FIT confidence, common name for the compound, number of fragment matches, and the number of citations found. [C] Synonyms for this compound, the structure, and high energy spectrum.

The markers of interest detected in the samples and isolated using statistics, can be identified with different levels of confidence. While a reference standard would be needed for full confirmation of an identity, the UNIFI Software utilizes a complete workflow that significantly reduces the burden of lengthy data interpretation. Table 2 indicates the tentative identity of a selection of markers from the 100% recycled gears together with the

corresponding level of confidence (explained below) for each assignment.

Sample	<i>m/z</i>	Retention time (min)	Common name	Chemical formula	Fragment matches	i-FIT confidence (%)	Level
100% RG	182.1541	12.27	4-(2-Pyrrolidinyl)-1,6-heptadien-4-ol	C ₁₁ H ₁₉ NO	21	83	3
100% RG	187.1452	13.54	(4-Isopropyl-1-piperazinyl)acetic acid	C ₉ H ₁₈ N ₂ O ₂	7	100	3
100% RG	140.1075	13.93	N-Vinylcaprolactam	C ₈ H ₁₃ NO	11	100	3
100% RG	158.0959	14.16	1-Naphthylmethylamine	C ₁₁ H ₁₁ N			1
100% RG	186.1467	14.55	(2R)-N2-(1-Aminoethyl)-2,2,4,5(3H)-pyrimidinetetramine	C ₆ H ₁₅ N ₇	5	100	3
100% RG	166.1586	18.39	10-Undecenitrile	C ₁₁ H ₁₉ N	5	100	3
100% RG	256.2017	19.9	1,1-Dimethylethyl 2-[1-(cyclopentylimino)propyl]hydrazinecarboxylate	C ₁₃ H ₂₅ N ₃ O ₂	3	54	2
100% RG	166.1593	19.29	1-Piperidino-1-cyclohexene	C ₁₁ H ₁₉ N	9	100	3
100% RG	156.1384	18.92	1-Tert-butyl-4-piperidone	C ₉ H ₁₇ NO	2	100	3
100% RG	224.1760	18.74	4-(1-Methyl-1H-pyrazol-4-yl)-4-propoxypiperidine	C ₁₂ H ₂₁ N ₃ O	8	35	2

Table 2. Putative identifications by the Discovery Tool of a selection of chemical markers unique to the 100% recycled gears. In this case, level 1 is accurate mass alone, level 2 is accurate mass and fragment matches, and level 3 is accurate mass, fragment matches, and a high i-FIT confidence.

Once confirmed, chemical components can be added into the screening library in UNIFI for future screening analyses. The experimental data obtained, for example the retention time and the *m/z* values of fragment ions, can also be incorporated. As an example, a chemical marker that was found to be unique to the 100% recycled gear through the statistical analysis, and elucidated using the Discovery Tool, was added to the library. The results of screening for this compound in the 0% and 100% recycled gears are shown in Figure 6.

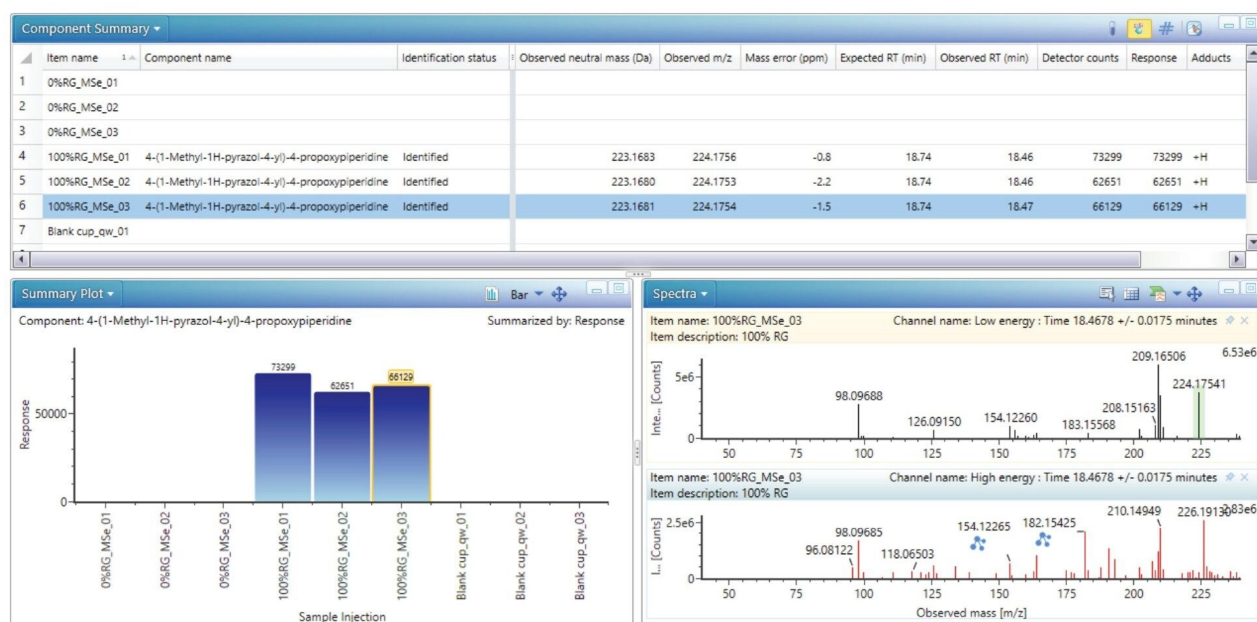


Figure 6. 4-(1-Methyl-1H-pyrazol-4-yl)-4-propoxypiperidine screening results after being added to the library.

Conclusion

Screening for unknown markers using py-APGC-QToF HRMS in combination with multivariate statistical analysis is a powerful tool for detecting compounds specific to a sample for differential characterization. In this case, the approach proved to be beneficial for an in-depth analysis of virgin and recycled (regrind) plastic samples. The analytical workflow encompasses simple steps to characterize these complex materials, including data acquisition, screening against a library, statistical analysis, and structural elucidation.

To understand differences between virgin and recycled material, multivariate statistical analysis can be applied. Utilizing PCA reveals the clustering of sample types and the OPLS-DA S-plot highlights significant markers responsible for differences between the 0% and 100% recycled samples. The Discovery Tool within the structural elucidation toolkit of the UNIFI application can then be applied to find tentative compound identifications using matches to accurate mass precursor and fragment ions.

The use of soft ionization and high-resolution, accurate mass measurements aid this process by retaining both

precursor and fragment ion information. Confirmed compounds can be added to an in-house library or database with their associated experimental data, thus enhancing the scope of such libraries for future screening analyses of a multitude of sample types.

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