Breakthrough performance of the Orbitrap Exploris GC for analytical testing and scientific research applications

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Goal

To demonstrate the Thermo Scientific™ Orbitrap Exploris™ GC mass spectrometers deliver exceptional analytical performance for both analytical science and scientific research applications.

Introduction

The Orbitrap Exploris GC mass spectrometers have been designed for analytical testing and scientific research applications. These highly selective high-resolution Orbitrap-based analytical platforms aim to deliver unprecedented and flexible performance with up to 240,000 mass resolving power.

The objective of this study was to further explore the power of high resolution and accurate mass using Orbitrap-based GC-MS[1] by evaluating key analytical parameters that are essential for analytical testing and scientific research applications. These include linear dynamic range, sensitivity, NIST library search matching, spectral fidelity, scan speed, mass accuracy, robustness, compound confirmation using positive chemical ionization, and resolving power.

Experimental

In all experiments, a Thermo Scientific Orbitrap Exploris GC was used. Some additional experiments were performed on the Thermo Scientific Orbitrap Exploris GC 240, which has a maximum resolving power of 240,000 (at *m/z* 200 FWHM). Sample introduction was performed using a Thermo Scientific™ TriPlus™ RSH autosampler, and chromatographic separation of the gas-phase chemical components was achieved using a Thermo Scientific™ TRACE™ 1310 Gas Chromatograph equipped with various capillary columns. The Orbitrap Exploris GC was tuned and calibrated using PFTBA to achieve mass accuracy of <1.0 ppm.

The routine ionization mode was electron ionization (EI) and the mass spectrometers were operated using full scan with default 60,000 mass resolution (FWHM, measured at *m/z* 200). Data acquired was lock-mass corrected using GC column bleed siloxane masses.

Standard and sample preparation

Standard preparation

To assess key analytical parameters, standards were prepared from stock standards in solvent and in matrix:

A) 8270 MegaMix (Restek, catalogue number 31850) was diluted in hexane to produce 12 calibration level standards (ranging from 0.1 ppb to 10,000 ppb)

B) Mixed pesticide calibration standards were prepared from stock standards (Restek, catalogue number 32562), and diluted in matrix to generate 11 calibrations standards (ranging from 0.1 to 500 ppb)

C) Mixed standard, containing octafluoronaphtalene (OFN) (0.1 pg/µL) and hexachlorobenzene (HCB) (10 pg/µL) in iso-octane.

Sample preparation

Locally purchased organic wheat flour or oats (10 g) were weighed into a 50 mL centrifuge tube. Acetonitrile (10 mL), containing 1% (v/v) of acetic acid, was then added to the sample, which was then vortexed for 1 min. To this, 3 g of magnesium sulfate ($MgSO_4$), 1.7 g of sodium acetate, and 0.5 g of disodium hydrogen citrate were added, and the tube was vortexed for 1 min and centrifuged at 4,000 rpm for 10 min. After centrifugation, 5 mL of the supernatant was transferred into a polypropylene tube with 250 mg C18 sorbent, 750 mg $MgSO_4$, and 750 mg primary/secondary amine (PSA). The tube was vortexed for 1 min and centrifuged for 10 min at 4,000 rpm and supernatant used for GC-MS analyses.

For unknown identification and structural elucidation experiments, commercially available oregano samples were purchased locally. Samples were weighed (150 mg) and transferred into 10 mL crimp top headspace vials (vials P/N 10-CV, caps P/N 20-MCBC-ST3) for analysis.

Results and discussion

Scan speed

Fast data acquisition, to allow sufficient data points across narrow GC peaks, is critical in order to achieve accurate and precise compound identification. An example using the Orbitrap Exploris GC is shown in Figure 1, where 15 data points across the 2.3 s wide peak for 2-nitroaniline (extracted ion chromatogram of *m/z* 138.04238) were obtained.

Compound identification using spectral libraries

The Exploris Orbitrap GC, with full scan range mass accuracy and sensitivity, enables accurate and reliable commercial library (e.g. NIST/Wiley) matching. Figure 2 shows the NIST library search results achieved using the Exploris Orbitrap GC for the analysis of aldrin in a mixed pesticide standard, with both forward and reverse library match scores of >890 achieved.

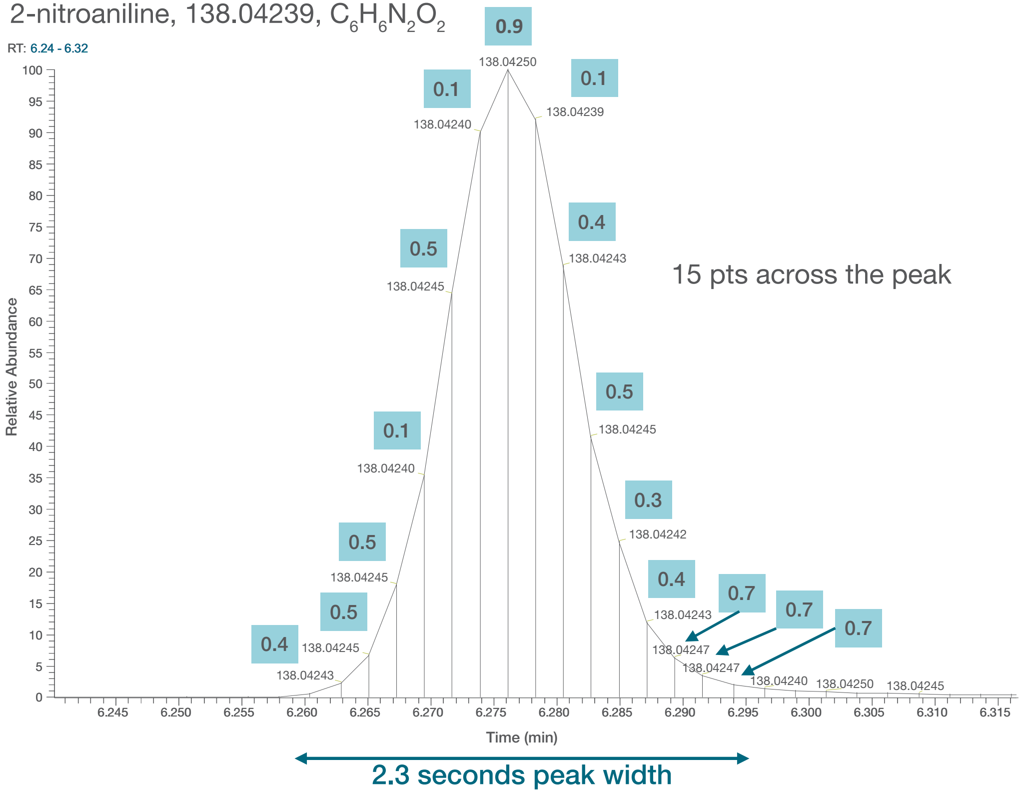
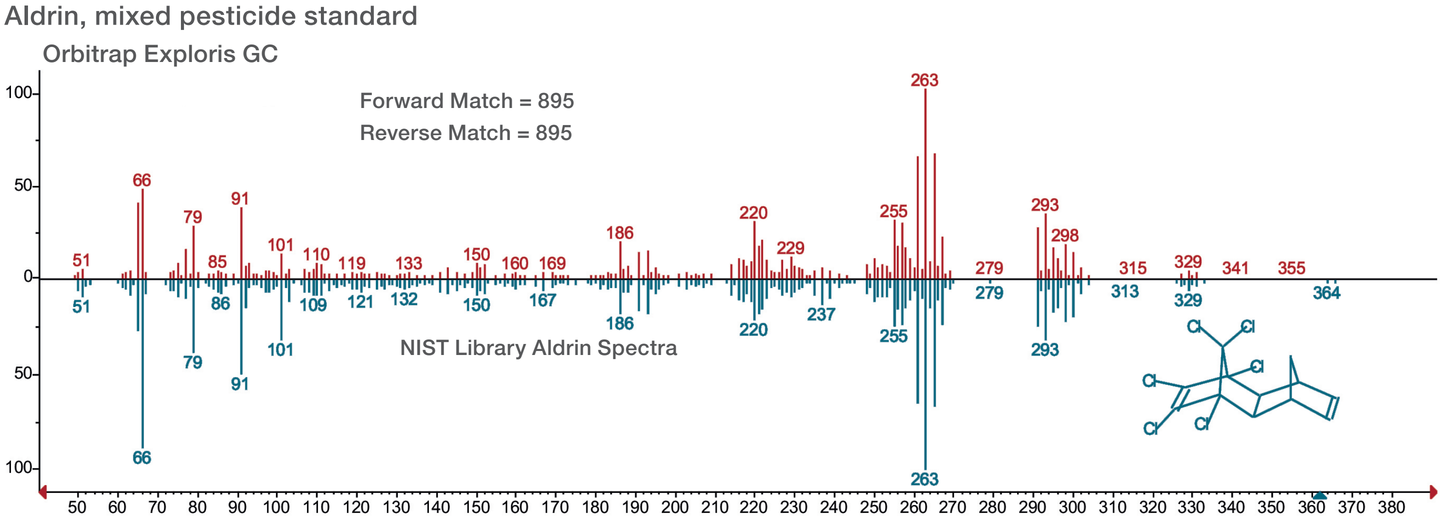


Figure 1. Extracted ion chromatogram (XIC) of 2-nitroaniline (*m/z* 138.04238 ± 5 ppm window) in a 1,000 pg/µL mixed solvent standard. Data acquired in full scan at 60k resolution (FWHM at *m/z* 200). Excellent mass accuracy is shown for each individual scan as well as mass difference (in ppm). An average mass difference of 0.4 ppm was measured across the peak.



Aldrin, mixed pesticide standard

Orbitrap Exploris GC

NIST Library Aldrin Spectra

Figure 2. NIST library search mass spectra and match results achieved for the analysis of aldrin in a mixed pesticide standard using an Orbitrap Exploris GC operated in full scan at 60k resolution (FWHM at *m/z* 200)

Additional results achieved using the Orbitrap Exploris GC are shown in Figure 3 for a selection of pesticides in a mixed pesticide standard in whole flour matrix, where similar NIST library results were obtained considering both reverse and forward search results.

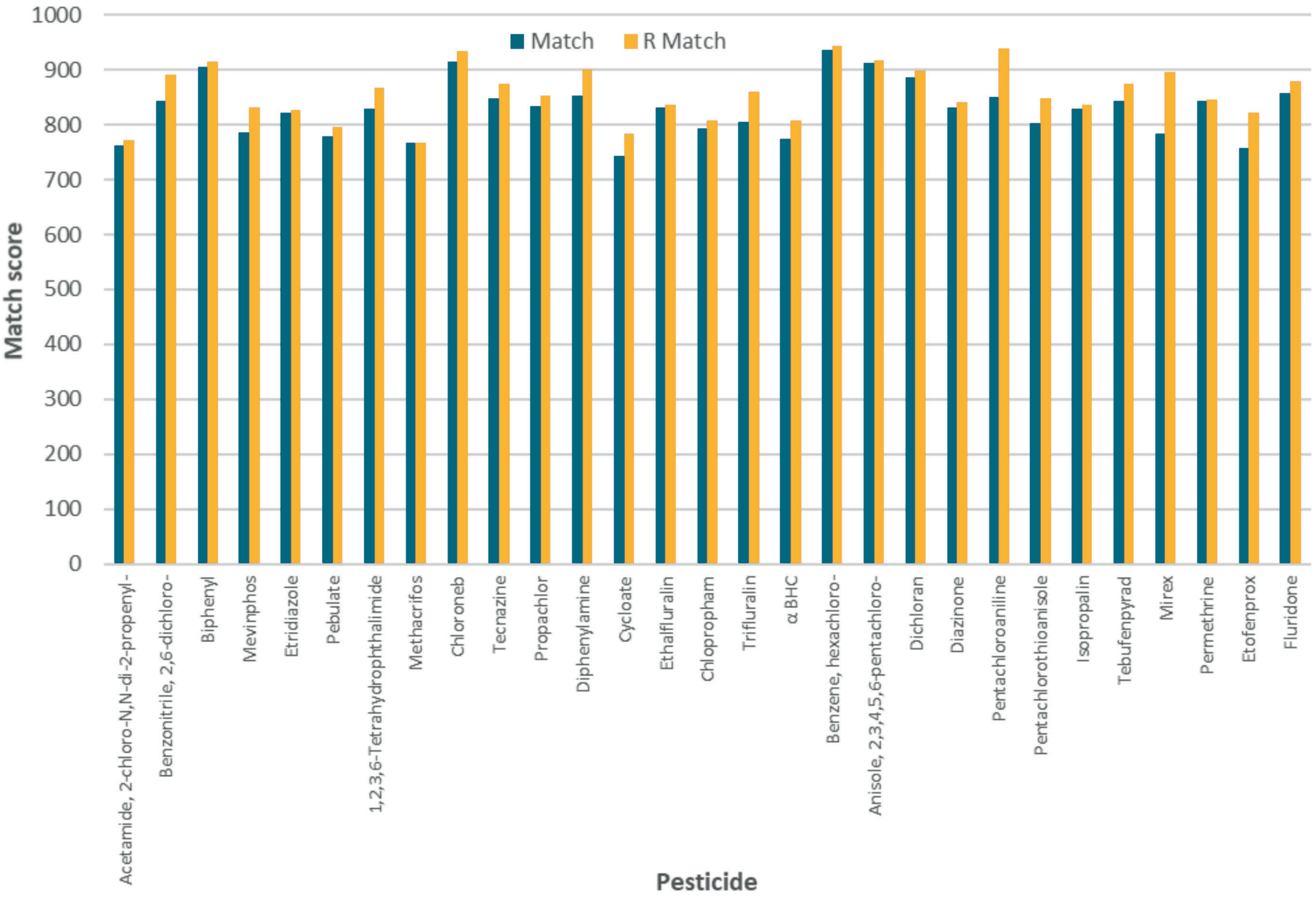


Figure 3. Library search scores achieved using an Orbitrap Exploris GC for a selection of pesticides in a mixed pesticide standard in a whole flour matrix (a score of 1,000 equals a perfect match). Forward search scores (Match) and reverse search scores (R Match) given for each pesticide, when searched against the NIST library. The Orbitrap Exploris GC was operated in full scan at 60k resolution (FWHM at *m/z* 200).

Linear dynamic range

A wide linear dynamic range is essential, especially when dealing with applications where the samples analyzed contain a complex chemical background that could potentially interfere with the analytes of interest (e.g., pesticide screening and quantification, metabolomics). To test the linear dynamic range using the Orbitrap Exploris GC, repeat injections (n=3) of increasing concentration levels (0.1 pg to 10,000 pg on column) of mixed solvent standards were performed. An example of compound linearity obtained using log(10) is shown in Figure 4 for hexachloroethane, the results demonstrating linear dynamic range extending to six orders of magnitude (0.1–10,000 pg on-column) making the Orbitrap Exploris GC an ideal platform for quantitative analysis.

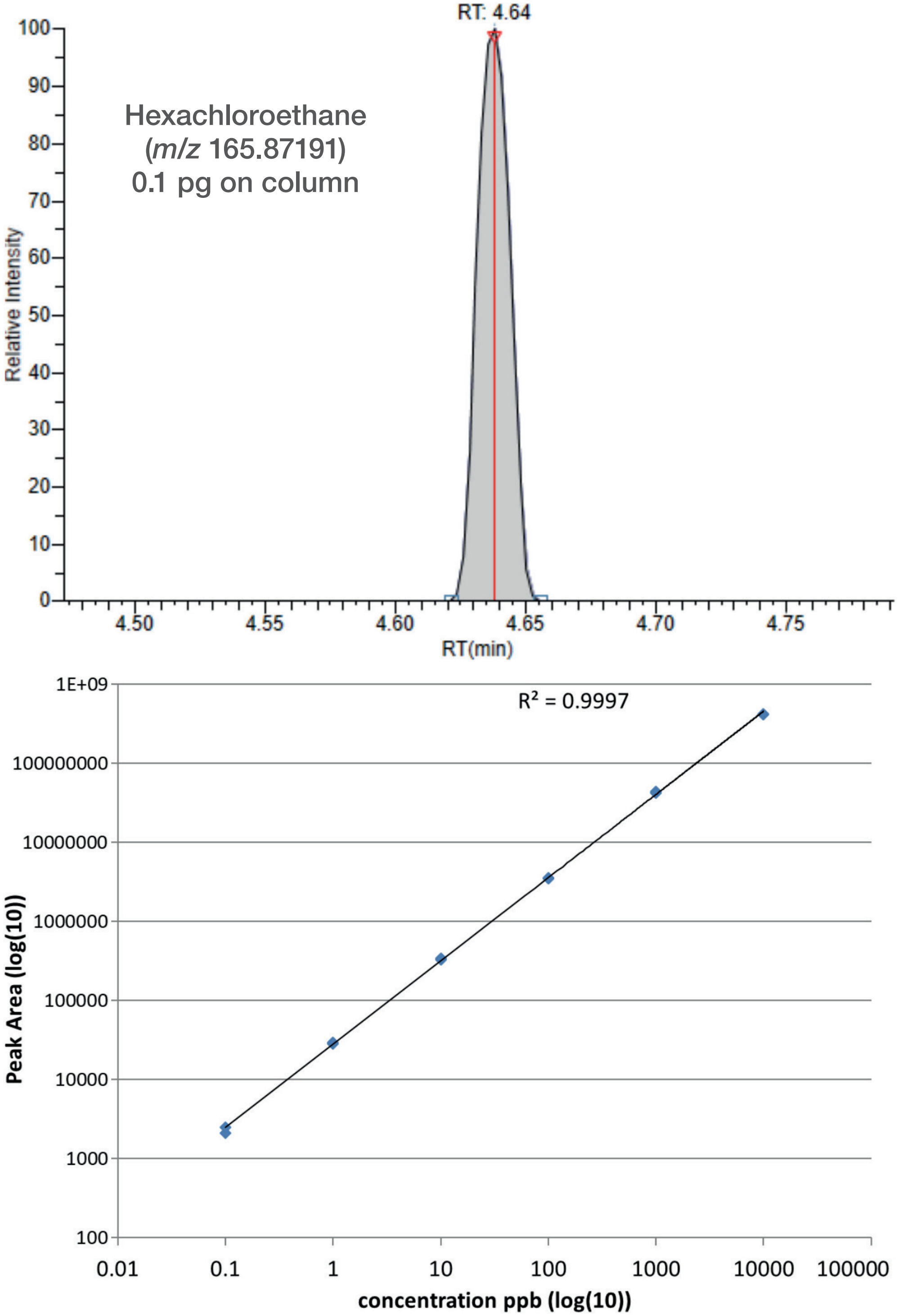


Figure 4. Linear dynamic range of the Orbitrap Exploris GC is demonstrated using hexachloroethane solvent standards injected over six orders of magnitude. The extracted ion chromatogram (*m/z* 165.87191) corresponding to hexachloroethane at 0.1 pg on column is shown together with the coefficient of determination (R^2) values determined over a concentration range of 0.1–10,000 pg on column.

Moreover, excellent peak area repeatability (n=3 injections) was obtained at each concentration level as demonstrated for hexachloroethane in Table 1 which shows %RSD ranged from 1.0 to 4.9 across the six orders of magnitude.

Table 1. Calculated %RSD from n=3 repeat injections of hexachloroethane solvent standard at various on column concentrations. Data from the Orbitrap Exploris GC shown.

Hexachloroethane concentration (pg on column)	Orbitrap Exploris GC %RSD (n=3)
10000	1.7
1000	1.5
100	1.0
10	2.4
1	2.4
0.1	4.9

Sensitivity

The sensitivity achievable with the Orbitrap Exploris GC was evaluated for the analysis of whole flour spiked with pesticides. For this a whole flour sample extract was spiked with pesticides at 10 pg/µL level (equivalent to the European Union (EU) default maximum residue level (MRL) set at 10 µg/kg) and repeat injections (n=10) were performed.

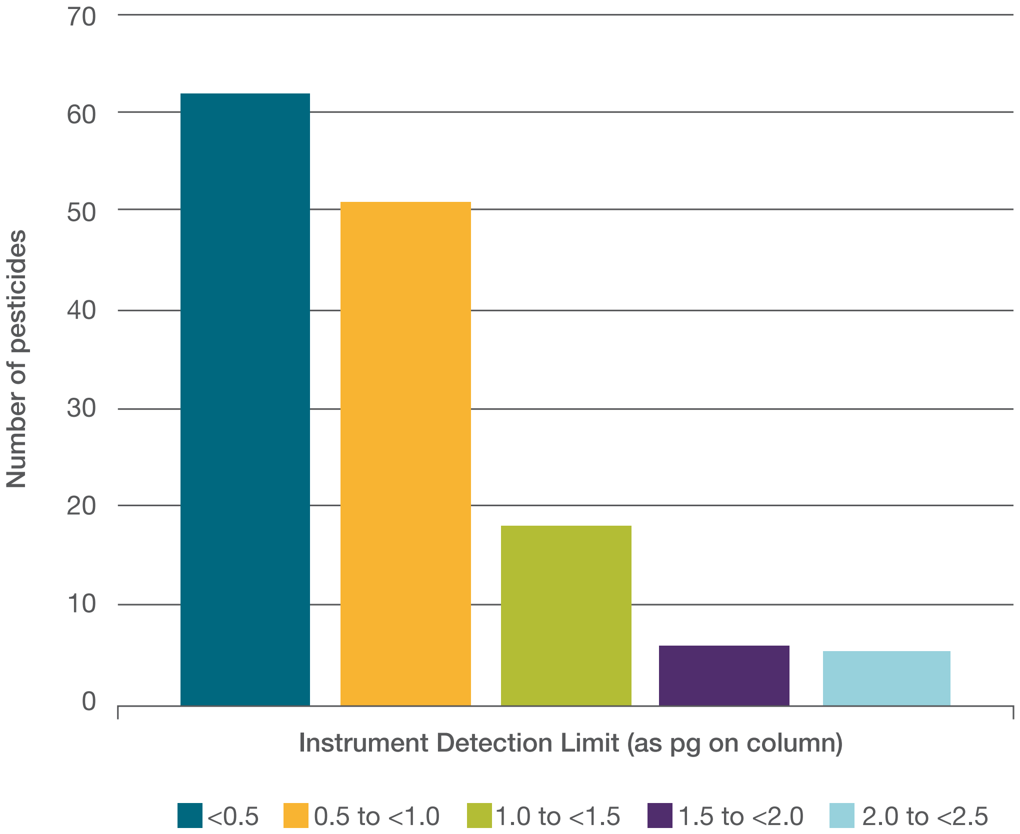
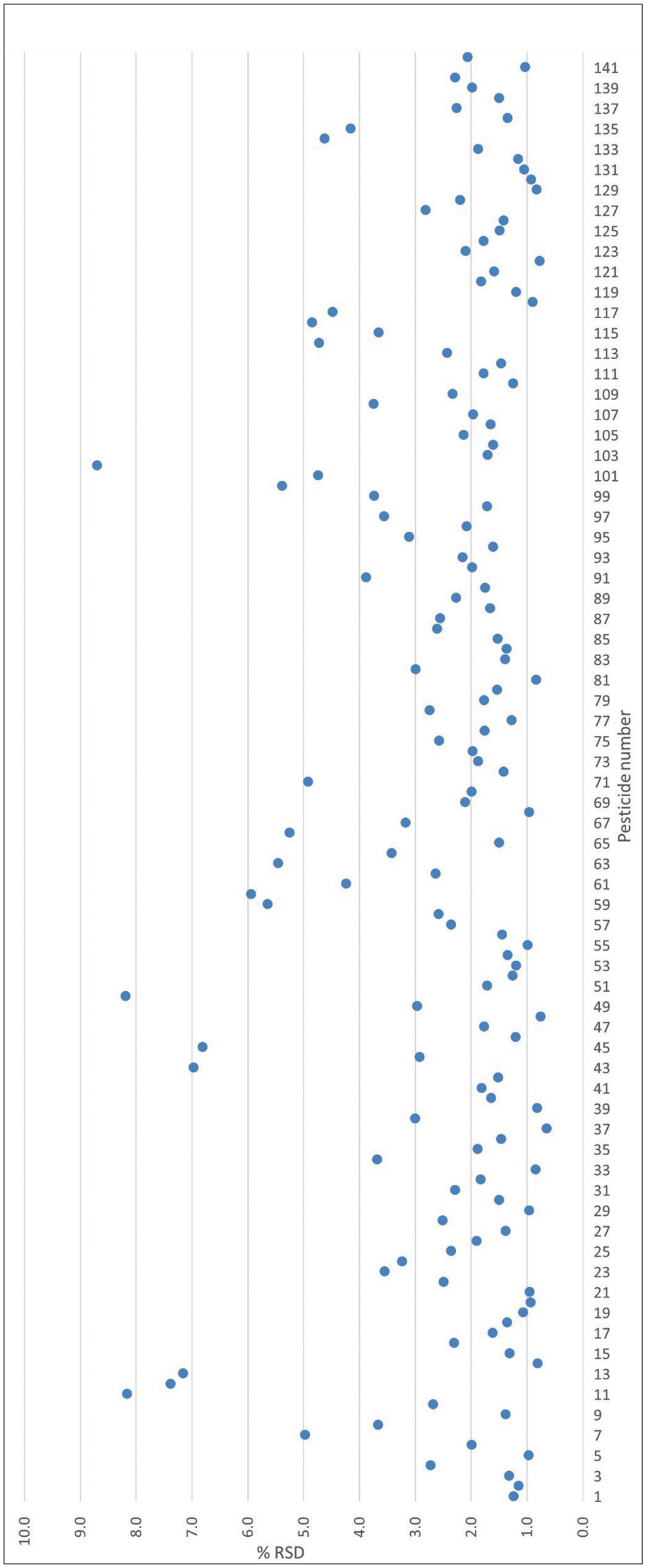


Figure 5. Sensitivity instrument detection limit (IDL as pg on column) determined for 142 pesticides in whole flour using an Orbitrap Exploris GC. Data was obtained by taking into account the amount on column, the Student's-*t* value corresponding degrees of freedom for repeat injections (n=10) of a 10 pg/µL whole flour matrix matched pesticide standard.

Sensitivity expressed as instrument detection limits (IDL) for all the pesticides analyzed was calculated and is reported in Figure 5, and the associated %RSD values are shown in Figure 6. The IDL was calculated taking into account the Student's-*t* critical values for the corresponding degrees of freedom (99% confidence). Excellent sensitivity with IDL values ranging from 0.18 to 2.45 pg/µL was achieved, with an average value of 0.7 pg/µL. The results confirmed that the Orbitrap Exploris GC has the sensitivity levels to meet the regulatory analysis of pesticides in matrix matched standards.

Orbitrap Exploris GC pesticide %RSD in whole flour



No	Compound	No	Compound
1	2-phenylphenol	72	Fenthion
2	3,4-Dichloroaniline	73	Fluquinconazole
3	4,4'-Dichlorobenzophenone	74	Fluridone
4	9,10-Anthracenedione	75	Flutolanil
5	Acetamide, 2-chloro-N-(2,6-dimethylphenyl)-N-(2-methoxyethyl)-	76	Heptachlor epoxide
6	Acetamide, 2-chloro-N-(ethoxymethyl)-N-(2-ethyl-6-methylphenyl)-	77	Hexazinone
7	Acetamide, 2-chloro-N,N-di-2-propenyl-	78	Iodofenphos
8	Acrinathrin	79	Isazofos
9	Alachlor	80	Isodrin
10	Aldrin/Aldrin-r	81	Isopropalin
11	Azinphos-ethyl	82	Lenacil
12	Azinphos-methyl	83	Leptophos
13	Benfluralin	84	Metazachlor
14	Benzenamine, 2,3,5,6-tetrachloro-	85	Methacrifos
15	Benzene, hexachloro-	86	Methyl parathion
16	Benzonitrile, 2,6-dichloro-	87	Mevinphos
17	Benzonitrile, pentachloro-	88	Mirex
18	BHC, alpha	89	Mitotane
19	BHC, beta	90	N-(2-Ethylhexyl)-5-norbornene-2,3-dicarboximide
20	BHC, delta	91	Nitralin
21	BHC, gamma	92	Nitrofen
22	Bifenthrin	93	Norflurazon
23	Bioallethrin	94	o,p'-DDE
24	Biphenyl	95	o,p'-Methoxychlor
25	Bromfenvinphos-methyl	96	Oxadiazone
26	Bromophos	97	Oxyfluorfen
27	Bromophos-ethyl	98	p,p'-DDE
28	Bromopropylate	99	p.p'-DDT
29	Bupirimate	100	Parathion
30	Carbamothioic acid, bis(1-methylethyl)-, S-(2,3-dichloro-2-propenyl) ester	101	Pebulate
31	Carbophenothion	102	Penconazole
32	Carfentrazone-ethyl	103	Pendimethalin
33	Chlorbenside	104	Pentachloroaniline
34	Chlorfenapyr	105	Pentachlorothioanisole
35	Chlorfenson /Ovex	106	Permethrin
36	Chlorfenvinphos	107	Phosalone
37	Chlorobenzilate	108	Phosmet
38	Chloroneb	109	Phosphonothioic acid, phenyl-, O-ethyl O-(4-nitrophenyl) ester
39	Chlorpropham	110	Piperonyl butoxide
40	Chlorpyrifos(-ethyl)	111	Pirimiphos methyl
41	Chlorpyriphos-methyl	112	Pirimiphos-ethyl
42	Chlorthiophos	113	Pretilachlor
43	Chlorthiophos II	114	Prochloraz
44	Chlozolinate	115	Prodiamine
45	Cis-1,2,3,6-Tetrahydrophthalimide	116	Profenofos
46	cis-Chlordane	117	Profluralin
47	cis-Nonachlor	118	Propanil
48	Clomazone/Dimethazone	119	Propyzamide
49	Coumaphos	120	Pyrazophos
50	Cycloate	121	Pyridaphenthion
51	Cyprodinil	122	Pyrimethanil
52	Diazinone	123	Pyriproxyfen
53	Dichloran	124	Quintozene
54	Diphenamid	125	Sulfotep
55	Diphenylamine	126	Sulprofos
56	Disulfoton	127	Tebuconazole
57	Endosulfan (beta)	128	Tebufenpyrad
58	Endosulfan ether	129	Tecnazene
59	Endosulfan sulfate	130	Tefluthrin
60	Endosulfan, (alpha)	131	Terbacil
61	Endrin	132	Terbufos
62	Endrin ketone	133	Terbuthylazine
63	Endrin-aldehyde	134	Tetrachlorvinphos
64	Ethalfluralin	135	Tetramethrin
65	Ethion	136	Tolclofos-methyl
66	Etofenprox	137	trans-Chlordane
67	Fenamiphos	138	Transfluthrin
68	Fenarimol	139	trans-Nonachlor
69	Fenchlorphos/Ronnel	140	Triadimefon
70	Fenitrothion	141	Triallate
71	Fenson	142	Vinclozolin

Figure 6. IDL repeatability expressed as % RSD achieved for the analysis of 142 pesticides using an Orbitrap Exploris GC, for repeat injections (n=10) of a 10 pg/μL whole flour matrix matched pesticide standard

Spectral fidelity irrespective of concentration

Maintaining spectral fidelity over the full analytical concentration range in matrix is critical to maintain confidence in compound identification, even at low levels, as illustrated in Figure 7 for pentachloroaniline in whole flour matrix. The mass accuracy for every ion in the isotopic cluster is <1 ppm giving high confidence in the identification.

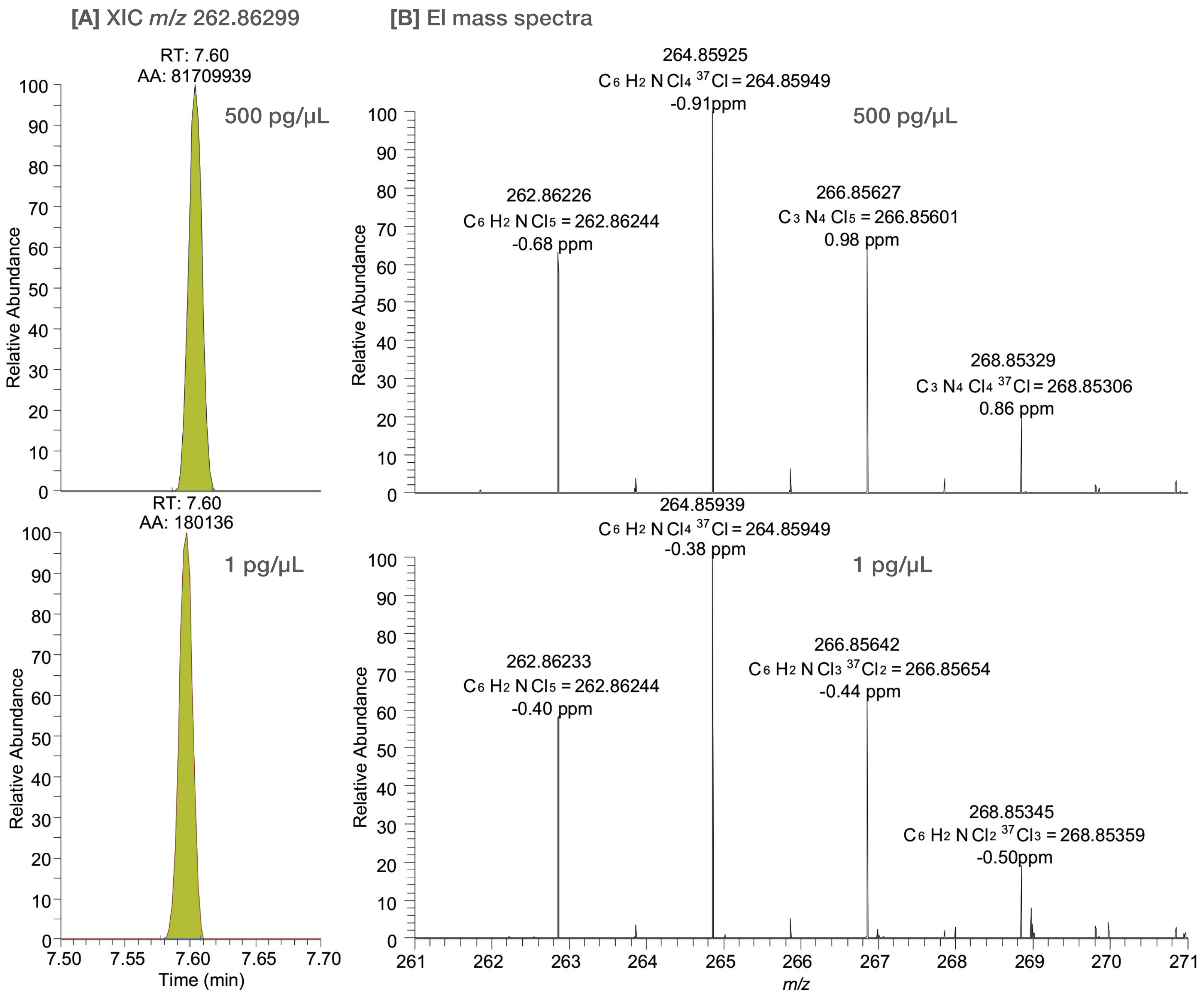
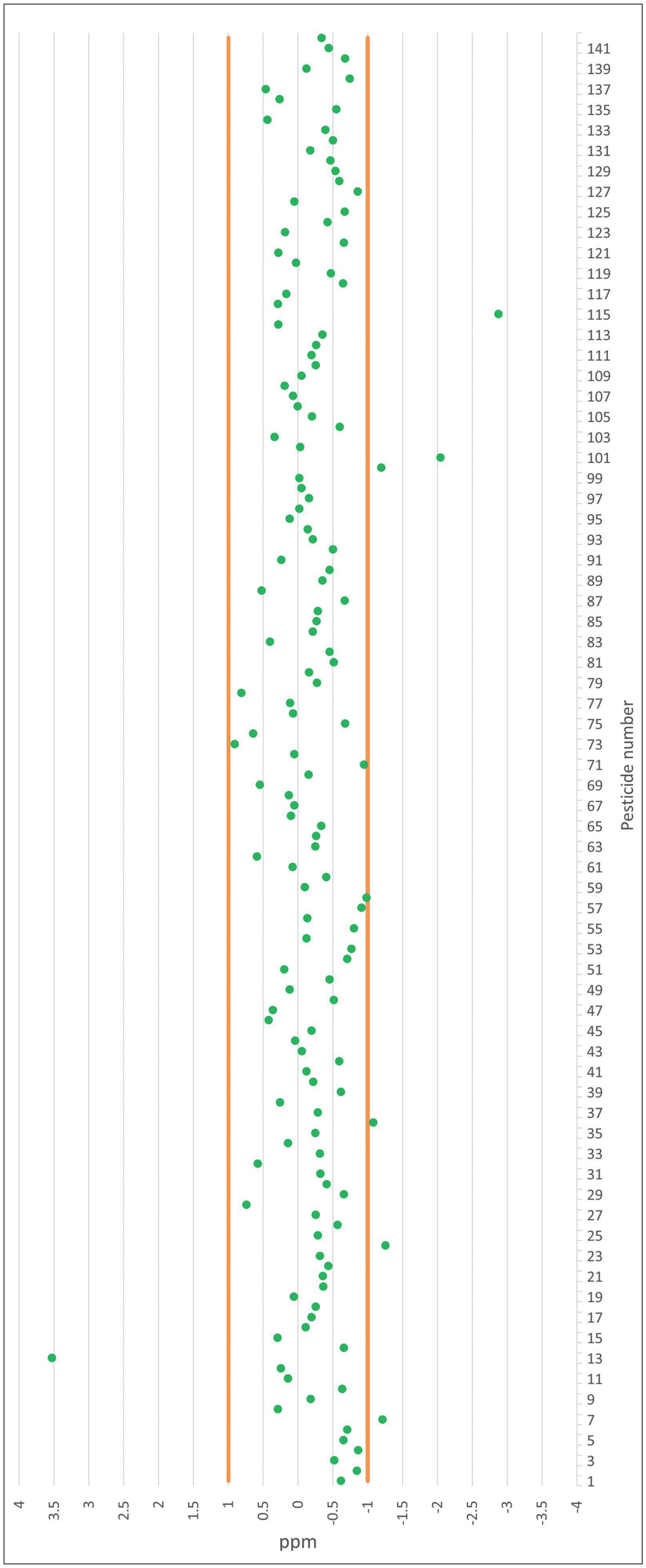


Figure 7. Spectral fidelity illustrated for pentachloroaniline for two levels (1 and 500 pg/µL) in whole flour matrix using an Orbitrap Exploris GC. [A]: Extracted ion chromatograms (XIC) for pentachloroaniline at each level annotated with peak retention time (RT) and peak area (AA); [B]: EI mass spectra zoomed at the molecular ion cluster at each level, annotated with measured mass, elemental composition, theoretical mass and mass accuracy (ppm).

Mass accuracy

To have a high degree of confidence in compound identification, low (<1 ppm) mass accuracy is critical. To test the mass accuracy that can be achieved using the Orbitrap Exploris GC, repeat injections (n=10) of mixed pesticides standards (10 pg/µL) were carried out. Examples are reported in Figure 8, with an average mass accuracy of -0.2 ppm.

Orbitrap Exploris GC pesticide accurate mass in whole flour



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24	Biphenyl	95	o,p'-Methoxychlor
25	Bromfenvinphos-methyl	96	Oxadiazone
26	Bromophos	97	Oxyfluorfen
27	Bromophos-ethyl	98	p,p'-DDE
28	Bromopropylate	99	p.p'-DDT
29	Bupirimate	100	Parathion
30	Carbamothioic acid, bis(1-methylethyl)-, S-(2,3-dichloro-2-propenyl) ester	101	Pebulate
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49	Coumaphos	120	Pyrazophos
50	Cycloate	121	Pyridaphenthion
51	Cyprodinil	122	Pyrimethanil
52	Diazinone	123	Pyriproxyfen
53	Dichloran	124	Quintozene
54	Diphenamid	125	Sulfotep
55	Diphenylamine	126	Sulprofos
56	Disulfoton	127	Tebuconazole
57	Endosulfan (beta)	128	Tebufenpyrad
58	Endosulfan ether	129	Tecnazene
59	Endosulfan sulfate	130	Tefluthrin
60	Endosulfan, (alpha)	131	Terbacil
61	Endrin	132	Terbufos
62	Endrin ketone	133	Terbuthylazine
63	Endrin-aldehyde	134	Tetrachlorvinphos
64	Ethalfluralin	135	Tetramethrin
65	Ethion	136	Tolclofos-methyl
66	Etofenprox	137	trans-Chlordane
67	Fenamiphos	138	Transfluthrin
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69	Fenchlorphos/Ronnel	140	Triadimefon
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Figure 8. Mass accuracy (ppm) in matrix is illustrated for repeat injections (n=10) of mixed pesticides standards (10 pg/µL)

Maintaining sub-ppm mass accuracy irrespective of compound concentration

Sub-ppm mass accuracy was maintained across compound concentrations using an Orbitrap Exploris GC, as exemplified for hexachloroethane (Table 2). In all cases, irrespective of the *m/z* and concentration level, <1 ppm values were observed. This is essential as any compromise in accuracy of mass measurements can result in false identification and non-detection of toxic chemicals such as pesticides in a screening experiment.[2] It is also necessary to maintain this performance at all concentration levels as any level can be encountered in real world samples.

Mass accuracy repeatability

To evaluate repeatability of mass accuracy over long analysis runs at low concentrations when using the Orbitrap Exploris GC, 140 injections of mixed solvent standards containing octafluoronaphtalene (OFN) (0.1 pg oc) and hexachlorobenzene (HCB) (10 pg oc) were performed, as reported in Figure 9. The results confirmed that by using the Orbitrap Exploris GC, high levels of mass accuracy (<1.1 ppm) over 2 days unattended analysis can confidently be achieved, without instrument maintenance, calibration, or tuning.

Table 2. Mass accuracy (ppm) over six orders of magnitude for five selected ions of hexachloroethane measured using the Orbitrap Exploris GC

Level	**Orbitrap Exploris GC**				
ppb on column	***m/z* 118.90306**	***m/z* 116.90601**	***m/z* 120.90011**	***m/z* 165.87191**	***m/z* 202.83781**
0.1	0.1	0.4	0.7	0.1	-0.3
1	0.0	0.0	0.2	0.1	-0.1
10	0.4	0.6	0.5	0.6	0.3
100	0.4	0.5	0.4	0.5	0.3
1000	0.7	0.8	0.7	0.8	0.5
10000	0.5	0.6	0.5	0.6	0.3
average Δppm	**0.4**	**0.5**	**0.5**	**0.4**	**0.2**

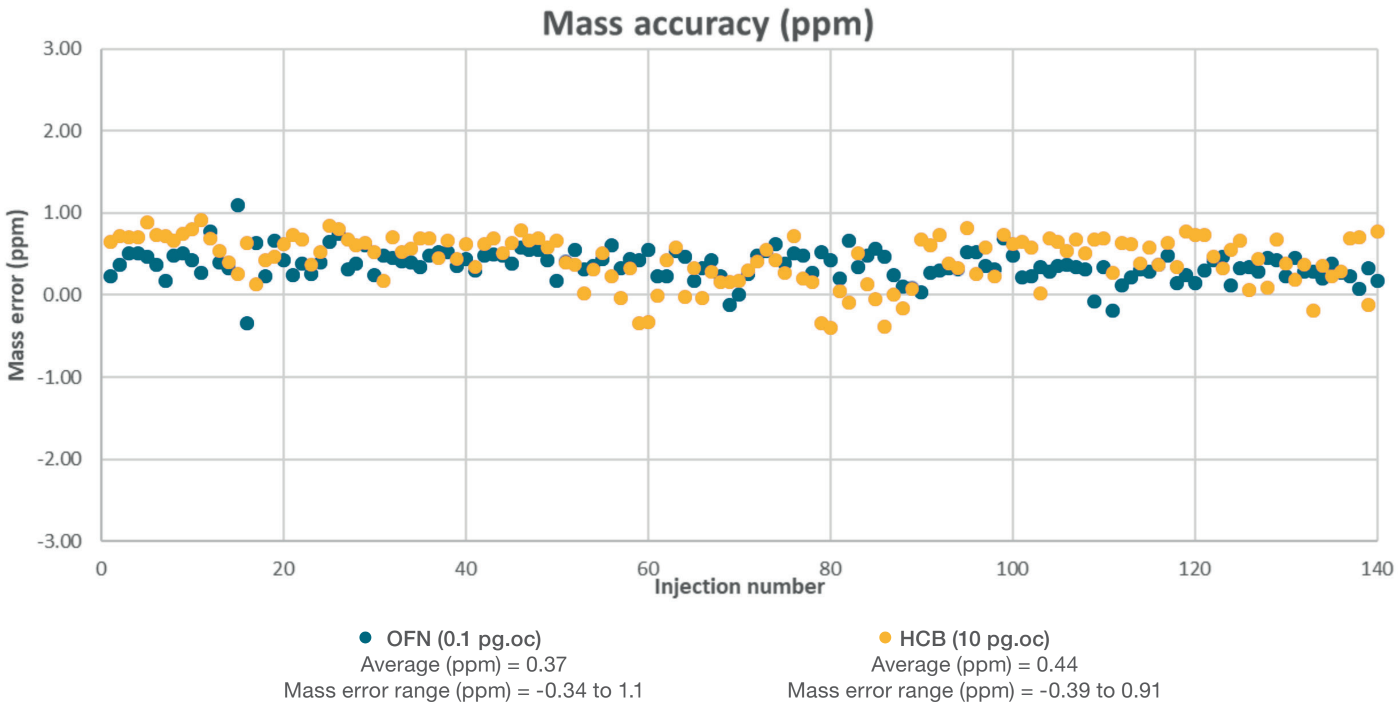

Figure 9. Repeatability of mass accuracy (ppm), illustrated with 140 injections of a solvent standard, containing OFN (0.1 pg on-column) and HCB (10 pg on-column), annotated with average mass accuracy and range achieved using an Orbitrap Exploris GC

Compound confirmation using PCI

When the spectral library match from the EI spectrum is inconclusive, or additional confirmation is required, positive chemical ionization (PCI) data can be used to confirm the elemental composition of the parent molecule using accurate mass information. In PCI experiments using methane as the reagent gas three adducts are typically observed: $[M+H]^+$, $[M+C_2H_5]^+$, and $[M+C_3H_5]^+$. As an example, EI and PCI spectra of camphene in a sample of thyme are reported in Figure 10. The observed molecular ion corresponding to *m/z* 136.12468 is present in the EI spectrum with a mass difference of 0.2 ppm from the theoretical *m/z* 136.12465 for the formula $C_{10}H_{16}$. The presence of the methane adducts in the PCI spectrum with sub-1 ppm mass accuracy confirmed *m/z* 136.12468 as the molecular ion for camphene (RT=9.04 min) and supported the elemental composition of the proposed molecule.

Resolving power

Acquiring reliable accurate mass measurements is critical when detecting low level pesticides in complex matrices. Through the high-mass resolving power of the Orbitrap Exploris GC 240, discrimination between matrix interferences and target analyte ions can be confidently achieved. When the resolution is insufficient, the mass profile of ions overlaps, which could result in high mass error and incorrect assignment of the mass of the target ion. This is demonstrated in Figure 11 where an oat matrix standard (500 pg/µL) was analyzed at resolving powers of 30K, 60K, 120K, and 240K using the Orbitrap Exploris GC 240. The zoomed in mass spectra show the quantifier ion and a matrix ion of a similar mass causing interference. The diphenylamine ion at 30K and 60K is not resolved resulting in poor mass accuracies of -6.38 and -6.18 ppm, respectively, and is resolved at 120K and even further resolved at 240K with mass accuracies of 1.10 and 0.56 ppm, respectively.

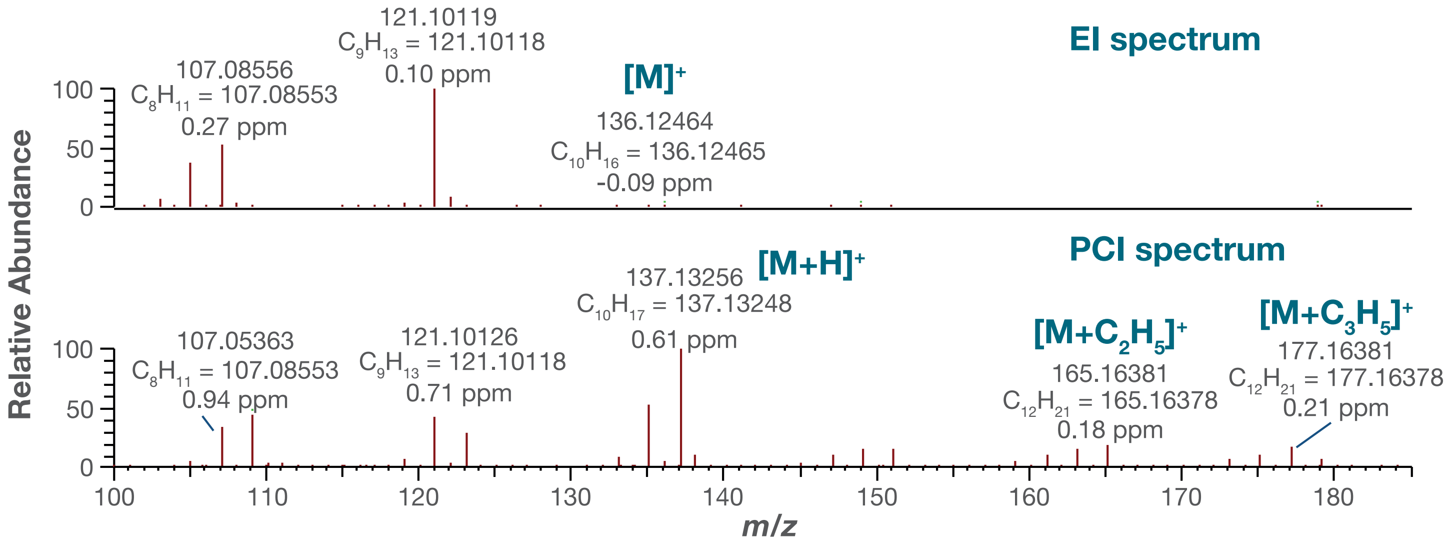

Figure 10. Comparison between EI and PCI spectrum for camphene in a sample of thyme (RT=9.04 min). The molecular ion (*m/z* 136.12468) is visible in the EI spectrum with sub ppm mass accuracy annotated. In the PCI spectrum the typical adducts observed when methane gas is used are clearly visible confirming the molecular ion and the proposed molecular formula for camphene.

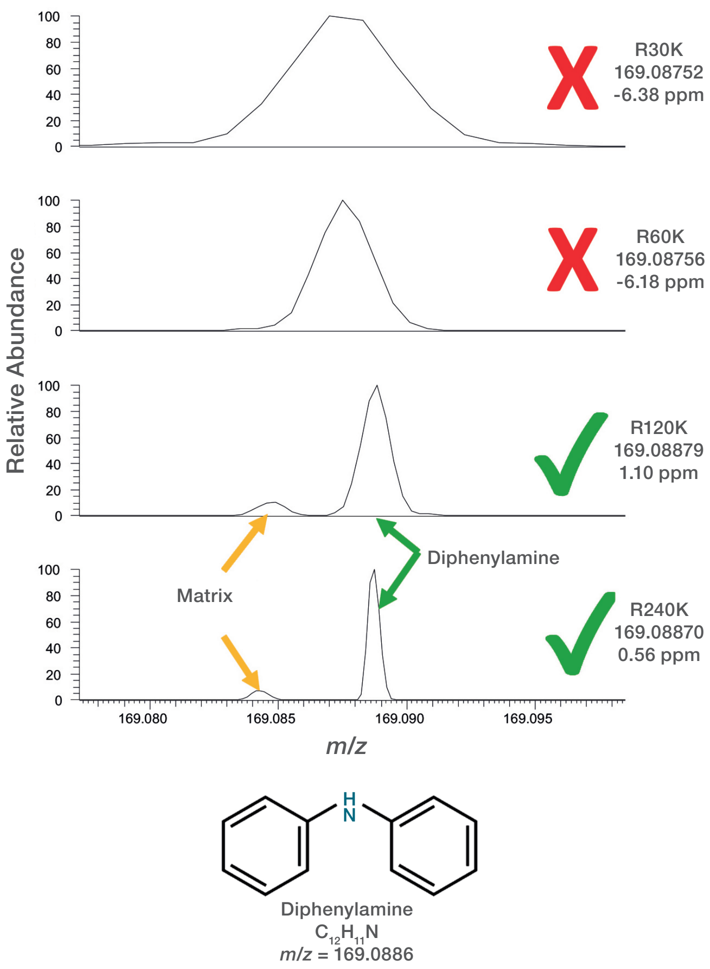

Figure 11. Effects of resolving power on the mass accuracy using the Orbitrap Exploris GC 240 for the detection of diphenylamine in an oat matrix standard (500 pg/µL) acquired at 30K, 60K, 120K, and 240K. The diphenylamine ion at 30K and 60K is not resolved; however, at 120K and 240K the quantifier ion for diphenylamine and a matrix interference ion are sufficiently resolved. Zoomed in mass spectra are annotated with the resolving power, the measured mass, as well as the mass accuracy (ppm).

Conclusions

The data shown here demonstrate that the Orbitrap Exploris GC mass spectrometers deliver exceptional high-quality analytical performance using full scan acquisition for both analytical testing and scientific research applications.

The fast scan speeds available allow sufficient data points across narrow chromatographic peaks to accurately describe the peak area and to ensure signal and spectra reproducibility. The linear dynamic range extending to six orders of magnitude and the sensitivity demonstrated for >140 pesticides in whole flour (IDL values of between 0.18 and 2.45 pg/µL) make the Orbitrap Exploris GC ideal for analytical testing.

NIST library searchable accurate mass spectra are achievable, enabling confident identification of unknown compounds and confirmation of knowns. Spectral fidelity irrespective of concentration can also be achieved, which is critical to maintain confidence in compound identification.

When the spectral library match from the EI spectrum is inconclusive, or additional confirmation is required, the Orbitrap Exploris GC positive chemical ionization (PCI) data can be used to confirm the elemental composition of the parent molecule using accurate mass information.

Sub-ppm mass accuracy is achievable for the analysis of pesticides in whole flour spiked with pesticides. In addition, the high mass resolving power of the Orbitrap Exploris GC 240 enables superior discrimination between matrix interferences and target analyte ions.

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