Research Article

Rapid Communications in Mass Spectrometry

Received: 25 September 2012 Revised: 13 November 2012 Accepted: 14 November 2012 Published online in Wiley Online Library

Rapid Commun. Mass Spectrom. **2013**, *27*, 419–429
(wileyonlinelibrary.com) DOI: 10.1002/rcm.6469

Performance of dielectric barrier discharge ionization mass spectrometry for pesticide testing: a comparison with atmospheric pressure chemical ionization and electrospray ionization

Bienvenida Gilbert-López[1], Helma Geltenpoth[1], Cordula Meyer[1], Antje Michels[1], Heiko Hayen[3], Antonio Molina-Díaz[2], Juan F. García-Reyes[2]* and Joachim Franzke[1]**

[1]Leibniz-Institut für Analytische Wissenschaften – ISAS – e.V, Otto-Hahn-Str. 6b, 44227 Dortmund, Germany
[2]Analytical Chemistry Research Group (FQM-323), Department of Physical and Analytical Chemistry, University of Jaén, Campus Las Lagunillas, Edif. B-3, 23071, Jaén, Spain
[3]University of Wuppertal, Department of Food Chemistry, 42119 Wuppertal, Germany

RATIONALE: The present study reports on the evaluation of dielectric barrier discharge microplasma ionization (DBDI) for liquid chromatography/high resolution mass spectrometry (LC/HRMS) analyses of pesticide residues in fruit and vegetables. Ionization, fragmentation, analytical performance and matrix effects displayed by LC/DBDI-MS were critically evaluated and compared with both atmospheric pressure chemical ionization (APCI) and electrospray (ESI), using a set of over 40 representative multiclass pesticides.
METHODS: Sample preparation was accomplished using standard QuEChERS procedure and the identification and quantitation of the pesticides tested accomplished by means of LC/MS with a hybrid linear quadrupole ion trap (LIT)-Fourier transform ion cyclotron resonance (FTICR) mass spectrometer operated in full-scan positive ion mode using DBDI, APCI and ESI sources.
RESULTS: The developed LC/DBDI-MS method allowed the screening of 43 pesticides in three different vegetable matrices: apple, orange and tomato. Minor matrix effects (i.e. signal suppression or enhancement ≤20%) were observed in most of the studied compounds: 95%, 70% and 81% of the studied compounds showed minor matrix effects in extracts of apple, orange and tomato, respectively. The results of the analysis of spiked orange extracts showed that the sensitivity obtained with LC/DBDI-MS is appropriate for multi-residue analysis of pesticide residues in fruit and vegetable samples. The limits of quantitation (LOQs) obtained for most of the studied pesticides were in compliance with the European Regulation 396/2005 (and subsequent updates) on food commodities (default maximum residue level of 10 μg kg^{-1}).
CONCLUSIONS: Comparative studies with commercial sources demonstrate the suitability of DBDI as an ionization technique for residue analysis, because of the combination of the following two advantages: (1) the use of DBDI provides minimized matrix effects compared with APCI, and (2) improved the detection – in terms of sensitivity – of selected compounds that are not easily ionized by ESI, such as parathion.

Pesticides are widely used in current agricultural practice because of their unquestionable benefits for crop protection. Their application involves improved crop yields and also increases the quantity of fresh fruits and vegetables in the diet. However, the persistence of pesticide residues in agricultural products destined for human consumption is one of the most serious problems connected to their use since the presence of pesticide residues in food may negatively affect human health.[1] For this reason, pesticide testing is of paramount importance not only for the protection of human health, but also for trade and official control purposes. For most of these compounds, regulatory guidelines set maximum residue levels (MRLs) in drinking water and food to help protect the community against contamination and potential negative health effects. Therefore, pesticide testing is a target application in these commodities, including a broad range of compounds with different physicochemical properties. Current status on agrochemicals is oriented towards greener approaches, involving the rational use of non-persistent last-generation (polar) pesticides. The increasing number of these polar pesticides, together with the requirement of tedious derivatization steps to analyze some compounds and the issue with non-volatile, thermolabile species, has prompted the use of liquid chromatography/mass spectrometry (LC/MS) to the detriment of gas chromatography/mass spectrometry (GC/MS).[2–4]

* *Correspondence to:* J. F. García-Reyes, Analytical Chemistry Research Group, University of Jaén, 23071 Jaén, Spain. E-mail: jfgreyes@ujaen.es
** *Correspondence to:* J. Franzke, Leibniz-Institut für Analytische Wissenschaften – ISAS – e.V., 44227 Dortmund, Germany. E-mail: franzke@isas.de

LC/MS coupled with an electrospray ionization (ESI) source offers a wider coverage than GC/MS applying electron ionization (EI)[5] for pesticide testing applications. However, there are selected groups of agrochemicals such as organochlorine pesticides that are not efficiently ionized by ESI and still require the use of GC/MS. In order to avoid the need to use both techniques to cover a wide polarity range of pesticide and reduce the costs of duplicated analysis per sample (both LC/MS and GC/MS runs), alternative or complementary ionization techniques have been developed to increase the LC/MS coverage provided by ESI. Atmospheric pressure chemical ionization (APCI) is the widest used complementary ionization source to ESI and many manufacturers offer dual sources or interfaces that allow the change from ESI to APCI in a few minutes. Atmospheric pressure photoionization (APPI) is another interesting technique developed for increasing the range of LC/MS applications to less polar compounds,[6–9] and hybrid APPI/ESI sources have been also studied.[10] Atmospheric pressure laser ionization (APLI)[11,12] and its hybrid ESI/APLI interface,[13] electrochemistry-assisted ESI[14–16] and LC/electron ionization MS[17–19] are amongst other technologies proposed to expand the ionization coverage of electrospray.

Dielectric barrier discharge microplasma ionization (DBDI) for LC/MS has been recently reported as an alternative to increase the range of polarities that can be analyzed in one LC/MS run.[20,21] LC/DBDI-MS was preliminary tested for a wide array of compounds with an special emphasis on non-polar compounds such as polycyclic aromatic hydrocarbons and organochorine pesticides.[20,21] Nevertheless, a thorough investigation of the performance of DBDI compared with ESI and APCI on multi-class compounds including the study of matrix effects in complex samples has not been carried out yet.

A change in ionization efficiency in the presence of other compounds is called the matrix effect. The matrix effect was first described by Kebarle and Tang[22] who demonstrated that the response of one organic base decreased as the concentration of other bases increased. The exact mechanism of ion suppression is not known. It has been found that the matrix effect may be caused by nonvolatile material[23] or by compounds of high surface activity.[24] Matrix effects are associated with the ionization process, so that the effectiveness of the ion-formation process depends on both the ion source and the nature of the matrix. Different behaviours are reported in the literature when using ESI or APCI sources.[25,26] From the literature examples, it can be concluded that, with some exceptions, ESI is more subject to matrix effects than APCI.[27]

The extent of the matrix effect can be different, because of the different ionization mechanisms. While in ESI the analyte is ionized in the liquid phase inside the electrically charged droplets and the analyte ions pass from the liquid to the gas phase, in APCI the neutral analytes are transferred into the gas phase by vaporizing the liquid in a heated gas stream and the ionization occurs through the chemical ionization of the gas phase analyte.[23,28] APCI is by far less investigated than ESI but it is generally reported that the former is less susceptible to matrix effects, because ionization takes place in the gas phase.[27] Analogously, DBDI is also an ionization technique in the vapour phase. Therefore, matrix effects were anticipated to be smaller compared to ESI. The lack of information on matrix effects in DBDI prompted us to carry out a detailed investigation of this phenomenon.

Because of the influence of matrix effects on the quantitative approach and the requirements of sensitivity of residue analysis in complex samples such as food, some authors have investigated different techniques to reduce matrix effects besides the change in ionization source, such as dilution of the extracts[26] or post-column addition of analytes, and have proposed some explanations about the mechanisms involved in the ion-formation process.[23,27]

The reduction of matrix effects without losing sensitivity for the analytes of interest is a challenging task, overall when a generic sample treatment is combined with liquid chromatography/high-resolution mass spectrometry (LC/HRMS). This combination is nowadays very common in the field of pesticide residue analysis because of the increasing number of active compounds and derived degradation products to be monitored within a single analytical method.[29] Accordingly, LC/HRMS provides a detailed picture of complexity and composition of the sample extract injected, together with the signals of the trace amounts of the analytes of interest.[30]

Within this context, we present the evaluation of the DBDI source in terms of analytical performance, taking as a reference the 10 μg kg^{-1} established as the most stringent value for pesticides in any commodity. Vegetable samples were treated by the QuEChERS (Quick, Easy, Cheap, Effective, Rugged, and Safe)[31] extraction procedure and analyzed by LC/DBDI-HRMS using a Fourier transform ion cyclotron resonance (FTICR) mass spectrometer. Matrix effects were investigated and compared to those obtained by the standard ionization method based on electrospray ionization (ESI), and also to APCI due to the similarity of the ionization mechanism between the latter and DBDI.

EXPERIMENTAL

Target compounds

A set of 43 multiclass pesticides used in fruit and vegetable production have been selected for this study (see Supplementary Table S1, Supporting Information). They were chosen as representative of many pesticide classes (herbicides, insecticides, fungicides, etc.) because of their relevance in terms of pesticide testing regulations,[32] and presence in previously studied samples.[33]

Chemicals

HPLC-grade acetonitrile for sample treatment and methanol for pesticide stock solutions were obtained from Merck (Darmstadt, Germany). For HPLC/MS analysis, acetonitrile and methanol were obtained from Roth (Karlsruhe, Germany). Formic acid (LC/MS quality) and anhydrous magnesium sulphate (reagent grade) were obtained from Fluka (Buchs, Switzerland). Acetic acid was purchased from Panreac (Barcelona, Spain). Primary-secondary amine (Supelclean™ PSA SPE Bulk packing, 50 μm) was purchased from Supelco (Bellefonte, PA, USA). A Milli-Q-Plus ultra-pure water system (Milford, MA, USA) was used throughout the study to obtain the HPLC-grade water used during the analyses. Sodium acetate (reagent grade) was from Sigma-Aldrich (Madrid, Spain).

Analytical grade standards (PESTANAL® quality) for individual compounds were purchased from Sigma-Aldrich (Madrid, Spain). Individual pesticide stock solution (ca. 500 mg L^{-1}) were prepared in methanol and stored at –20°C. Then, a working solution containing the mixture of standards was prepared (10 mg L^{-1}) in methanol and was also kept at –20°C.

Sample preparation

Extraction procedure

The employed procedure was the so called 'QuEChERS' (acronym of 'quick, easy, cheap, effective, rugged and safe') described elsewhere.[31] The proposed procedure comprised the following steps: a representative 15 g portion of previously crushed (including the peel), blended and homogenized sample was weighed in a 50-mL plastic centrifuge tube. Then 15 mL of acetonitrile containing 1% acetic acid were added, and the tube was vigorously shaken for 1 min. After this time, 6 g of anhydrous magnesium sulfate and 1.5 g of sodium acetate were added, and immediately the shaking process was repeated for 1 min to prevent coagulation of $MgSO_4$. The extract then was centrifuged (3700 rpm) for 1 min. Then, 5 mL of the supernatant (acetonitrile phase) were taken with a pipette and transferred to a 15-mL graduated plastic centrifuge tube containing 250 mg of PSA and 750 mg of $MgSO_4$, that was then energetically shaken for 20 s. The extract was then centrifuged again (3700 rpm) for 3 min. A volume of 1 mL of the acetonitrile extract was evaporated near to dryness and reconstituted with 1 mL of methanol/water (30:70 v/v). Prior to LC/MS analysis the extract was filtered through a 0.45 μm PTFE filter (Millex FG, Millipore, Milford, MA, USA).

Several extracts of representative vegetable and fruit matrices[34] were prepared: orange (as representative fruit with high acid and high water content), tomato and apple (as representative vegetable and fruit with high content of water, respectively); acetonitrile extracts were diluted 1:3 with water. Besides, a concentrated (non-diluted) orange extract was prepared using the same procedure. In this case, the acetonitrile extract was evaporated to near dryness and reconstituted with a mixture of methanol and water (30% and 70% v/v, respectively).

Matrix-matched standards

Matrix-matched standards of the studied pesticides were prepared (for each matrix) by adding known amounts of working pesticide mix solution to the extracts in order to obtain the desired concentration range. For fruit and vegetable extracts, matrix-matched standards were prepared at the following concentration levels: 1, 10, 20, 50, 100, 200 μg L^{-1}. Blank extracts were also measured to ensure the absence of the selected pesticides originally in the sample.

Quantitative evaluation of matrix effects

Matrix effects (ME) were defined as the slope calibration curve of matrix-matched standards divided by the slope calibration curve of solvent-based standards. Values <1 indicate signal suppression and values >1 represent signal enhancement. A value of ME = 1 means that no matrix effect occurred.

Liquid chromatography/hybrid linear ion trap Fourier transform ion cyclotron resonance mass spectrometry (LC-LIT-FTICRMS) analysis

HPLC details

The employed Surveyor™ HPLC system consisted of a vacuum degasser, an autosampler and a quaternary pump (Thermo Fisher Scientific, Bremen, Germany). The separation of the species from the extracts was carried out in a reversed-phase C-8 column (Thermo Hypersil Gold, 150 × 2.1 mm, 5 μm particle diameter, 175 Å pore size) using a binary gradient. In order to prevent column damage, an inline filter was located between the autosampler and the column. The column oven was set to 30°C and 5 μL of extract were injected in each study. The total HPLC run time was 40 min. Mobile phase A composition was: 95% water, 5% acetonitrile (both with 0.1% formic acid). Mobile phase B was acetonitrile with 0.1% formic acid. The chromatographic method held the initial mobile phase composition (5% B) constant for 2 min, followed by a linear gradient to 95% B at 32 min. Then, in 1 min the system returned to initial conditions (5% B at 33 min) and remained constant during 7 min. The flow rate was 150 μL min^{-1}. The total ion chromatograms corresponding to spiked extracts of apple, orange and tomato are shown in Supplementary Fig. S1 (see Supporting Information).

Mass spectrometer

Mass spectrometric detection was carried out using a hybrid linear quadrupole ion trap (LIT)-Fourier transform ion cyclotron resonance (FTICR) mass spectrometer (Thermo Finnigan LTQ FT™; Thermo Fisher Scientific, Bremen, Germany) equipped with an Ion Max™ API source housing. This housing has an adjustable probe mount that allows the adjustment of the probe depth and also the exchange among APCI, APPI and ESI probes. Besides this, the commercial housing has two windows (located in the left and front side) that can be removed to install the PEEK adapter for the DBD plasmajet. This PEEK adapter was manufactured at ISAS (Leibniz-Institut für Analytische Wissenschaften – ISAS – e.V., Dortmund, Germany), as reported by Hayen *et al.*[20] The instrument was operated in the FTICR-MS full-scan mode, acquiring data in the positive ionization mode throughout the range *m/z* 60–600 with a resolution $R = 50\,000$ (full width at half maximum, FWHM). The full-scan data recorded was processed with Xcalibur™ version 2.1 software (Thermo Fisher Scientific, Bremen, Germany).

DBDI, APCI and ESI sources

The DBD microplasma ionization source[20] was realized by modification of a commercial API source (Ion Max™ API source, Thermo Fisher Scientific) so that the HPLC eluent nebulized and vaporized in the same manner as for APCI. This approach has the advantage of being easily connected to the mass spectrometer without the need to modify the vacuum interface. In addition, direct comparison between the proposed DBDI source and the standard APCI source was also facilitated, because their housing is identical. Additionally, the ESI probe head fits into the same housing, which guarantees a similar geometry and hence comparability to DBDI and APCI, respectively.

Ionization was carried out by a DBD with a plasma cone outside the electrode region. The plasma was operated with a helium (99.999% purity) flow of 200 mL/min. The DBD

consisted of a 3-cm long glass capillary with an i.d. of 500 μm and an o.d. of 1.2 mm (ca. 5 μL of gas capacity). Rings with an i.d. of 500 μm are located around the capillary, forming electrodes with a separation distance of 12 mm. The distance of the electrode to the end of the capillary is 2 mm. A periodic voltage pulse (5.4 kV with a frequency of 20 kHz and a pulse width of 2 μs) is applied. The plasma electrodes are enclosed in a Teflon tube not only for safety precautions, but also to prevent a discharge between the electrodes outside the capillary. This Teflon tube containing the DBD is called 'Plasmajet' and was located at the left side of the source housing, in the radial[21] position (Supplementary Fig. S2, see Supporting Information) regarding the MS inlet capillary (heated at 275°C).

This ion source, like the unmodified APCI source, works with a heated nebulizer maintained at 450°C. Nitrogen (99.999% purity) was used to nebulize the liquid eluent (sheath gas, flow rate set at 40.0 arbitrary units) and also to transport the finely dispersed sample droplets through the heated ceramic tube in which they were vaporized (auxiliary gas, flow rate set at 5.0 arbitrary units – 4.0 for ESI). Additionally, another nitrogen flow (sweep gas, flow rate of 2.0 arbitrary units) through the opposite direction of ions is used. The discharge current for the APCI experiments was set to 5 μA, while for DBDI experiments this current has to be zero. For ESI the capillary voltage was set to 4 kV.

RESULTS AND DISCUSSION

LC/DBDI-MS mass spectral features of multiclass pesticides. Compound identification and confirmation

A comparative evaluation of mass spectral features and in-source fragmentation was accomplished with DBDI, APCI and ESI sources under the same instrumental parameters except those particular to each individual technique. A standard solution containing the mixture of pesticides (1 μg mL^{-1}) was prepared and analyzed in duplicate by each ionization

Table 1. Fragmentation study of selected pesticides (those which displayed uncommon fragments and/or sodium adducts) with DBDI, APCI and ESI. Relative abundances correspond to a standard of a concentration of 1 μg mL^{-1}. *m/z* values written in bold were used for calibration/quantitation purposes. 'Uncommon' fragments found in the experiment with *m/z* values in italics

				Relative abundance (%)*		
Compound	*m/z* ion	Formula	Exact mass	DBDI	APCI	ESI
Metalaxyl (Rt 14.4 min)	$[M+Na]^+$	$C_{15}H_{21}NO_4Na$	302.13628	8.8	0.5	53.9
	$[M+H]^+$	**$C_{15}H_{22}NO_4$**	**280.15433**	**64.1**	**100.0**	**100.0**
	Frag.	$C_{14}H_{18}NO_3$	248.12812	33.2	3.3	1.8
	Frag.	$C_{13}H_{18}NO_2$	220.13321	77.0	4.9	1.1
	Frag.	$C_{12}H_{18}NO$	192.13829	100.0	83.7	73.8
Linuron (Rt 16.1 min)	**$[M+H]^+$**	**$C_9H_{11}Cl_2N_2O_2$**	**249.01921**	**100.0**	**100.0**	**100.0**
	Frag.	*$C_8H_7N_2Cl_2$*	*200.99808*	*18.0*	*0.3*	–
	Frag.	$C_8H_7ClN_2O$	182.02414	11.9	0.1	0.2
	Frag.	$C_6H_4Cl_2N$	159.97153	1.9	–	–
Prochloraz (Rt 15.4 min)	**$[M+H]^+$**	**$C_{15}H_{17}N_3O_2Cl_3$**	**376.03809**	7.5	**100.0**	**100.0**
	Frag.	**$C_{12}H_{13}NO_2Cl_3$**	**308.00064**	**100.0**	12.4	10.9
	Frag.	*$C_{11}H_{15}NOCl_3$*	*282.02137*	*23.7*	*3.6*	–
Propargite (Rt 21.3 min)	$[M+Na]^+$	$C_{19}H_{26}O_4SNa$	373.14440	3.7	–	93.6
	$[M+H+H_2O]^+$	$C_{19}H_{28}O_5S$	368.16520	–	–	100.0
	$[M+H]^+$	$C_{19}H_{26}O_4S$	350.15463	–	60.0	–
	Frag.	***$C_{16}H_{23}O$***	***231.17434***	***100.0***	***100.0***	***19.6***
	Frag.	*$C_{12}H_{15}O$*	*175.11174*	*25.9*	*21.1*	*1.1*
Simazine (Rt 12.4 min)	**$[M+H]^+$**	**$C_7H_{13}N_5Cl$**	**202.08540**	**100.0**	**100.0**	**100.0**
	Frag.	$C_5H_9N_5Cl$	174.05410	2.3	–	–
	$[M+H\text{-}HCl]^+$	*$C_7H_{12}N_5$*	*166.10872*	*1.8*	*0.7*	–
	Frag.	$C_4H_7N_3Cl$	132.03230	3.1	–	–
Tebuconazole (Rt 17.0 min)	**$[M+H]^+$**	**$C_{16}H_{23}ClN_3O$**	**308.15242**	**100.0**	**100.0**	**100.0**
	$[M+H\text{-}H_2O]^+$	*$C_{16}H_{21}N_3Cl$*	*290.14185*	*45.2*	*34.7*	–
Thiachloprid (Rt 11.9 min)	**$[M+H]^+$**	**$C_{10}H_{10}N_4SCl$**	**253.03092**	**100.0**	**100.0**	**100.0**
	Frag.	$C_{10}H_9N_4S$	217.05424	0.1	–	–
	Frag.	*$C_8H_8N_2Cl$*	*167.03705*	*7.1*	*0.2*	–
	Frag.	C_6H_5NCl	126.01050	0.7	–	–
Triadimenol (Rt 15.8 min)	**$[M+H]^+$**	**$C_{14}H_{19}N_3O_2Cl$**	**296.11603**	**92.6**	**100.0**	**100.0**
	Frag.	$C_{12}H_{16}O_2Cl$	227.08333	52.4	5.3	0.9
	Frag.	*$C_{10}H_8O_3N$*	*190.04987*	*100.0*	*0.8*	–
	Frag.	*$C_8H_{14}ON_3$*	*168.11314*	*67.0*	6.3	–
	Frag.	$C_6H_{11}O$	99.08044	0.3	–	–
	Frag.	$C_2H_4N_3$	70.03997	5.6	–	–

*Acquisition mass range, tune file and all parameters not depending on the source remained constant to avoid any difference in the experiment except the ion source evaluated.

source. For data evaluation, extracted ion chromatograms (EICs) corresponding to the calculated exact mass were obtained using a mass tolerance of 20 ppm (relative mass error).

DBDI and APCI yielded more fragment ions than ESI. Some of the fragments found have not been previously reported by LC/ESI-MS. For fragment identification, the maximum error tolerance was set at 5 ppm. The possible fragment should have also the same retention time as the precursor ion, and a logical fragmentation step from the molecular ion should be followed to achieve the empirical formula obtained. Most of the fragment ions encountered were even-electron ions. The compounds that showed 'uncommon' fragments were linuron ($[M+H-48]^+$), prochloraz ($[M+H-94]^+$), propargite ($[M+H-119]^+$ and $[M+H-175]^+$), simazine ($[M+H-HCl]^+$), tebuconazole ($[M+H-H_2O]^+$), thiacloprid ($[M+H-86]^+$) and triadimenol ($[M+H-128]^+$). Only the 'new' fragments found for propargite were also encountered in ESI experiments (see Table 1).

Except for parathion ethyl, areas obtained by ESI were more intense than those obtained by either DBDI or APCI for all pesticides. Parathion ethyl was detected in solvent standards using ESI at concentration levels from 500 μg L^{-1}

Table 2. Analytical parameters of the developed method using DBDI as ionization source: correlation coefficients and relative standard deviation

Pesticide	Ion	Formula ion	*m/z*	Rt (min)	RSD% Inter-day (n = 4)	Correlation coefficients (r)		
						Apple	Orange	Tomato
Atrazine	$[M+H]^+$	$C_8H_{15}ClN_5$	216.10105	14.29	7.2	0.99990	0.99820	0.99323
Bromuconazole	$[M+H]^+$	$C_{13}H_{13}BrCl_2N_3O$	375.96285	16.41	6.0	0.99995	0.99970	0.99980
Buprofezin	$[M+H]^+$	$C_{16}H_{24}N_3OS$	306.16346	16.78	5.5	0.99750	0.99865	0.99015
Carbofuran	$[M+H]^+$	$C_{12}H_{16}NO_3$	222.11247	13.82	7.1	0.99920	0.99945	0.99760
Chlorfenvinphos E-isomer	$[M+H]^+$	$C_{12}H_{15}Cl_3O_4P$	358.97680	18.21	6.7	0.99945	0.99905	0.99599
Chlorotoluron	$[M+H]^+$	$C_{10}H_{14}N_2OCl$	213.07892	14.05	17.3	0.99970	0.99910	0.99509
Deet (Diethyltoluamide)	$[M+H]^+$	$C_{12}H_{18}NO$	192.13829	14.60	6.7	0.99810	0.99915	0.99925
Desethyl terbuthylazine	$[M+H]^+$	$C_7H_{13}N_5Cl$	202.08540	13.19	4.9	0.99815	0.99925	0.99564
Diazinon	$[M+H]^+$	$C_{12}H_{22}N_2O_3PS$	305.10833	18.85	5.0	0.99970	0.99649	0.99950
Dichlorvos	$[M+H]^+$	$C_4H_8Cl_2O_4P$	220.95318	13.29	5.8	0.99945	0.99920	0.99990
Difenoconazole (2 isomers)	$[M+H]^+$	$C_{19}H_{18}Cl_2N_3O_3$	406.07197	18.38	7.6	0.99990	0.99985	0.99990
Difenoxuron	$[M+H]^+$	$C_{16}H_{19}N_2O_3$	287.13902	14.72	7.6	0.99985	0.99905	0.99975
Dimethoate	$[M+H]^+$	$C_5H_{13}NO_3PS_2$	230.00690	10.85	7.9	0.99995	0.99920	0.99995
Dimethomorph (2 isomers)	$[M+H]^+$	$C_{21}H_{23}ClNO_4$	388.13101	15.35	9.0	0.99980	0.99880	0.99975
Diuron	$[M+H]^+$	$C_9H_{11}Cl_2N_2O$	233.02429	14.62	6.7	0.99990	0.99795	0.99945
Fenamiphos	$[M+H]^+$	$C_{13}H_{23}NO_3PS$	304.11308	16.67	8.9	0.99965	0.99965	0.99860
Fenhexamid	$[M+H]^+$	$C_{14}H_{18}Cl_2NO_2$	302.07091	16.87	5.3	0.99960	0.99875	0.99935
Fenuron	$[M+H]^+$	$C_9H_{13}N_2O$	165.10224	10.11	7.6	0.99975	0.99930	0.99534
Hexythiazox	$[M+H]^+$	$C_{17}H_{22}ClN_2O_2S$	353.10850	20.74	6.3	0.99845	0.99890	0.99960
Imazalil	$[M+H]^+$	$C_{14}H_{15}Cl_2N2O$	297.05559	12.46	8.6	0.98874	0.99975	0.99980
Imidacloprid	$[M+H]^+$	$C_9H_{11}ClN_5O_2$	256.05958	10.10	5.8	0.99865	0.99975	0.99975
Isoproturon	$[M+H]^+$	$C_{12}H_{19}N_2O$	207.14919	14.65	3.6	0.99960	0.99865	0.99985
Kresoxim-methyl	$[M+H]^+$	$C_{18}H_{20}NO_4$	314.13868	18.36	6.4	0.99860	0.99910	0.99990
Linuron	$[M+H]^+$	$C_9H_{11}Cl_2N_2O_2$	249.01921	16.22	9.8	0.99945	0.99800	0.99970
Metalaxyl	$[M+H]^+$	$C_{15}H_{22}NO_4$	280.15433	14.57	7.3	0.99990	0.99915	0.99960
Metobromuron	$[M+H]^+$	$C_9H_{12}BrN_2O_2$	259.00767	14.94	7.4	0.99980	0.99995	0.99990
Monolinuron	$[M+H]^+$	$C_9H_{12}ClN_2O_2$	215.05818	14.60	7.2	0.99970	0.99770	0.99629
Monuron	$[M+H]^+$	$C_9H_{12}ClN_2O$	199.06327	12.78	4.3	0.99965	0.99955	0.99965
Parathion (ethyl)	$[M+H]^+$	$C_{10}H_{15}NO_5PS$	292.04031	18.70	6.1	0.99990	0.99990	0.99975
Pirimiphos-methyl	$[M+H]^+$	$C_{11}H_{21}N_3O_3PS$	306.10357	18.36	4.8	0.99925	0.99649	0.99935
Prochloraz	$[M+H]^+$	$C_{15}H_{17}Cl_3N_3O_2$	376.03809	15.48	5.8	0.99845	0.99292	0.99649
	Frag.	$C_{12}H_{13}NO_2Cl_3$	308.00064		7.3			
Propargite	$[M+Na]^+$	$C_{19}H_{26}O_4NaS$	373.14440					
	Frag.	$C_{16}H_{23}O$	231.17434	21.16	7.3	0.99187	0.99840	0.99514
Propazine	$[M+H]^+$	$C_9H_{17}ClN_5$	230.11670	15.72	8.4	0.99990	0.99960	0.99955
Pyrimethanil	$[M+H]^+$	$C_{12}H_{14}N_3$	200.11822	12.72	8.5	0.99865	0.99905	0.99554
Simazine	$[M+H]^+$	$C_7H_{13}N_5Cl$	202.08540	12.68	7.7	0.99890	0.99990	0.99995
Spiromesifen	Frag.	$C_{17}H_{21}O_3$	273.14852	22.04	8.8	0.99960	0.99720	0.99935
Tebuconazole	$[M+H]^+$	$C_{16}H_{23}ClN_3O$	308.15242	17.15	8.8	0.99985	0.99875	0.99985
Terbuthylazine	$[M+H]^+$	$C_9H_{17}ClN_5$	230.11670	16.06	6.7	0.99995	0.99970	0.99970
Thiabendazole	$[M+H]^+$	$C_{10}H_8N_3S$	202.04334	5.30	8.6	0.99835	0.99935	0.99448
Thiacloprid	$[M+H]^+$	$C_{10}H_{10}ClN_4S$	253.03092	12.06	4.2	0.99965	0.99885	0.99388
Triadimefon	$[M+H]^+$	$C_{14}H_{17}ClN_3O_2$	294.10038	16.94	7.4	0.99990	0.99860	0.99479
Triadimenol	$[M+H]^+$	$C_{14}H_{19}ClN_3O_2$	296.11603	15.84	2.2	0.99995	0.99820	0.99202
Trifloxystrobin	$[M+H]^+$	$C_{20}H_{20}F_3N_2O_4$	409.13697	19.85	5.7	0.99980	1.00000	0.99850

onwards. It was detected in matrix-matched standards only by APCI and DBDI, because the maximum concentration level tested in matrix standards was 200 μg L^{-1}.

In general, except for spiromesifen and propargite, the most abundant ion was the protonated molecule, which was used for quantitation purposes. Spiromesifen, metalaxyl, propargite and prochloraz did not show the same behaviour with each ionization source. (1) The spiromesifen protonated molecule was only found in ESI experiments, being the more abundant ion. On the other hand, a fragment of spiromesifen was found instead of the protonated molecule by either APCI or DBDI. (2) The sodium adduct was found for metalaxyl using all ionization sources, with higher abundance in ESI, followed by DBDI, and not very abundant in the case of APCI. In this case, the area of the protonated molecule of metalaxyl was considered for quantification purposes. (3) The sodium adduct was found for propargite by ESI, and with a very small intensity in DBDI, while it was not observed by APCI. The protonated molecule of propargite was only detected by APCI. $[M+H+H_2O]^+$ was the main ion observed by ESI. However, fragment ions were more intense when DBDI or APCI was used. (4) The fragment of prochloraz was used for quantitation purposes using DBDI as ionization source, while the protonated molecule was more intense with APCI and ESI. All these findings, particularly the increased abundance of some fragment ions, suggests that the ionization conditions of both DBDI and APCI are less gentle than ESI.

The presence of sodium adducts is not desirable and occurs to a higher extent in ESI than in APCI. Only a few publications have reported $[M+Na]^+$ ions when using APCI. Aguilar *et al.*[35] attributed the presence of Na^+ to impurities in the solvent and/or in the transfer capillary, but did not give any explanation about the mechanism of adduct formation. Thurman *et al.*[36] proposed a reaction in the gas phase between hydrated sodium atoms and analyte molecules as the pathway of sodium adduct formation; this reaction would be prompted by the presence of carbonyl or carboxyl groups in the molecule, as in the case of metalaxyl.

Table 3. Limits of quantification (LOQs) obtained for selected analytes in orange by LC/MS using DBDI. Comparison with MRLs established by the EU

Pesticide	Ion	Formula ion	*m/z*	LOQ (μg kg^{-1})	EU MRL[a] (μg kg^{-1})	Amending Regulation[a]
Atrazine	$[M+H]^+$	$C_8H_{15}ClN_5$	216.10105	10.0	50(*)	2008/839
Bromuconazole	$[M+H]^+$	$C_{13}H_{13}BrCl_2N_3O$	375.96285	20.0	50(*)	2008/149
Chlorfenvinphos E-isomer	$[M+H]^+$	$C_{12}H_{15}Cl_3O_4P$	358.97680	7.9	20(*)	2011/310
Chlorotoluron	$[M+H]^+$	$C_{10}H_{14}N_2OCl$	213.07892	10.0	50(*)	2008/149
Deet	$[M+H]^+$	$C_{12}H_{18}NO$	192.13829	8.0	10(**)	
Diazinon	$[M+H]^+$	$C_{12}H_{22}N_2O_3PS$	305.10833	3.3	10(*)	2008/149
Dichlorvos	$[M+H]^+$	$C_4H_8Cl_2O_4P$	220.95318	10.0	10(*)	2008/149
Difenoconazole (2 isomers)	$[M+H]^+$	$C_{19}H_{18}Cl_2N_3O_3$	406.07197	6.8	100	2011/508
Difenoxuron	$[M+H]^+$	$C_{16}H_{19}N_2O_3$	287.13902	7.6	10(**)	
Dimethomorph (2 isomers)	$[M+H]^+$	$C_{21}H_{23}ClNO_4$	388.13101	5.9	50(*)	2011/508
Diuron	$[M+H]^+$	$C_9H_{11}Cl_2N_2O$	233.02429	6.3	100	2008/149
Fenamiphos	$[M+H]^+$	$C_{13}H_{23}NO_3PS$	304.11308	6.9	20(*)	2011/559
Fenhexamid	$[M+H]^+$	$C_{14}H_{18}Cl_2NO_2$	302.07091	4.7	50(*)	2011/508
Fenuron	$[M+H]^+$	$C_9H_{13}N_2O$	165.10224	[b]	10(**)	
Imazalil	$[M+H]^+$	$C_{14}H_{15}Cl_2N_2O$	297.05559	7.1	5000	2010/750
Isoproturon	$[M+H]^+$	$C_{12}H_{19}N_2O$	207.14919	10.0	50(*)	2008/149
Linuron	$[M+H]^+$	$C_9H_{11}Cl_2N_2O_2$	249.01921	8.3	50(*)	2008/149
Metalaxyl	$[M+H]^+$	$C_{15}H_{22}NO_4$	280.15433	19.0	500	2008/839
Metobromuron	$[M+H]^+$	$C_9H_{12}BrN_2O_2$	259.00767	14.7	10(**)	
Monolinuron	$[M+H]^+$	$C_9H_{12}ClN_2O_2$	215.05818	33.0	50(*)	2008/149
Parathion (ethyl)	$[M+H]^+$	$C_{10}H_{15}NO_5PS$	292.04031	10.0	20(*)	2008/149
Pirimiphos-methyl	$[M+H]^+$	$C_{11}H_{21}N_3O_3PS$	306.10357	2.0	1000	2008/149
Propazine	$[M+H]^+$	$C_9H_{17}ClN_5$	230.11670	7.3	10(**)	
Pyrimethanil	$[M+H]^+$	$C_{12}H_{14}N_3$	200.11822	5.1	10000	2011/524
Simazine	$[M+H]^+$	$C_7H_{13}N_5Cl$	202.08540	10.0	10(*)	2011/310
Spiromesifen	Frag.	$C_{17}H_{21}O_3$	273.14852	9.3	20(*)	2008/839
Tebuconazole	$[M+H]^+$	$C_{16}H_{23}ClN_3O$	308.15242	9.2	900	2011/524
Terbuthylazine	$[M+H]^+$	$C_9H_{17}ClN_5$	230.11670	16.8	100	2008/149
Thiabendazole	$[M+H]^+$	$C_{10}H_8N_3S$	202.04334	5.0	5000	2008/149
Thiacloprid	$[M+H]^+$	$C_{10}H_{10}ClN_4S$	253.03092	1.5	20(*)	2011/508
Trifloxystrobin	$[M+H]^+$	$C_{20}H_{20}F_3N_2O_4$	409.13697	11.0	300	2011/508

[a]Regulation (EC) 396/2005.
[b]Not detected at the 10 μg kg^{-1} level, nor in matrix standard, neither in solvent standard.
(*)Indicates the lower limit of analytical determination.
(**)Default limit for non-specifically regulated compounds.

Rapid Communications in Mass Spectrometry

Analytical performance of LC/DBDI-MS

The linearity of the method was evaluated by preparing calibration graphs of matrix-matched standards at six concentration levels in the range 1–200 μg L^{-1}. Exemplarily, the total ion chromatograms (TICs) obtained by LC/DBDI-MS analysis of 100 μg L^{-1} spiked extracts of apple, orange and tomato samples are displayed in Supplementary Fig. S1 (see Supporting Information). Calibration curves were measured using three different ionization sources: ESI, DBDI and APCI. Non-spiked (or blank) extracts were also measured. Obviously, the linearity obtained with the commercial sources is satisfactory in all matrices tested, as it has been demonstrated in many publications so far. The DBDI source also showed an excellent linearity in the studied range, with correlation coefficients (r) higher or equal to 0.998 for most of the studied analytes in all three matrices tested. Detailed data of DBDI method linearity (r) and precision (RSD%) is presented in Table 2. Inter-day relative standard deviation (RSD%) was calculated by the analysis on four non-consecutive days of a solvent-based standard containing the mixture of analytes at a concentration level of 50 μg L^{-1}. The deviation in the reproducibility of the signal was below 10% for all studied analytes, except for chlorotoluron.

Quantitation was carried out by using peak areas of the extracted ion chromatograms (EICs) of the protonated molecules for each pesticide – with a 20 ppm mass window, except for spiromesifen, propargite, – and prochloraz in the case of DBDI, where the most abundant fragment ions were used. The limits of quantitation (LOQs) obtained in the studied fruit and vegetable extracts were very low, being in the range 1–10 μg L^{-1} for most of studied pesticides. In the case of DBDI and ESI, the LOQs for most of the studied analytes, in all three matrices tested, were in the range 1–5 μg L^{-1}. Using APCI, the LOQs for most of the target analytes in the apple extracts were in the range 1–5 μg L^{-1}, while, in orange and tomato extracts, the LOQs were mostly in the range 5–10 μg L^{-1}. Detailed information about the LOQs is shown in Supplementary Tables S2, S3 and S4 (see Supporting Information). The use of accurate mass analysis of the protonated molecule together with that of additional characteristic fragment ion(s) – including characteristic isotopic signals, and retention time (Rt) – enables the unambiguous identification and confirmation of the studied pesticides at low concentration levels in the studied sample extracts.

As an example, in order to test the sensitivity of the method, a matrix-matched standard containing the mixture of 31 selected pesticides was prepared in an orange extract at a low concentration level: 10 μg L^{-1}. This standard was analyzed by DBDI along with a blank extract to check the possible positive findings. The aim of this analysis was to compare the LOQs obtained with the Maximum Residue Levels (MRLs) established for pesticides in any commodity by the European Union.[32] As shown in Table 3, the LOQs were in compliance with the EU MRLs for all studied analytes, except for metobromuron and fenuron, which showed LOQs higher than the default 10 μg kg^{-1} MRL.

Matrix effects

To evaluate the matrix effects, the slopes obtained in the calibration with matrix-matched standards were compared with those obtained with solvent-based standards, calculating slope ratios matrix/solvent for each pesticide. Results showed that in most cases the signal is affected by the matrix (slope ratio $\neq 1$), so matrix-matched standards are necessary for calibration and quantitation. However, as is shown in Fig. 1, signal suppression/enhancement observed in each matrix tested is lower than 20% in most cases (minor matrix effects). Detailed data about matrix effects can be found in Supplementary Tables S2, S3 and S4 (see Supporting Information).

Some differences among the ionization techniques have been observed. Unexpectedly, APCI showed the worst matrix effects in all studied matrices, causing mainly signal enhancement. For instance, 30 of the studied compounds showed signal enhancement in tomato extracts (see Fig. 2). Matrix-induced response enhancement in GC has been explained as a blockage by matrix components of active sites (mainly free silanol groups) in the GC inlet and column, thus reducing losses of susceptible analytes caused by adsorption or degradation on these active sites.[37] In contrast, scarce information can be found in the literature about the causes of signal enhancement in HPLC/MS; only signal suppression has been studied more in detail, and a major

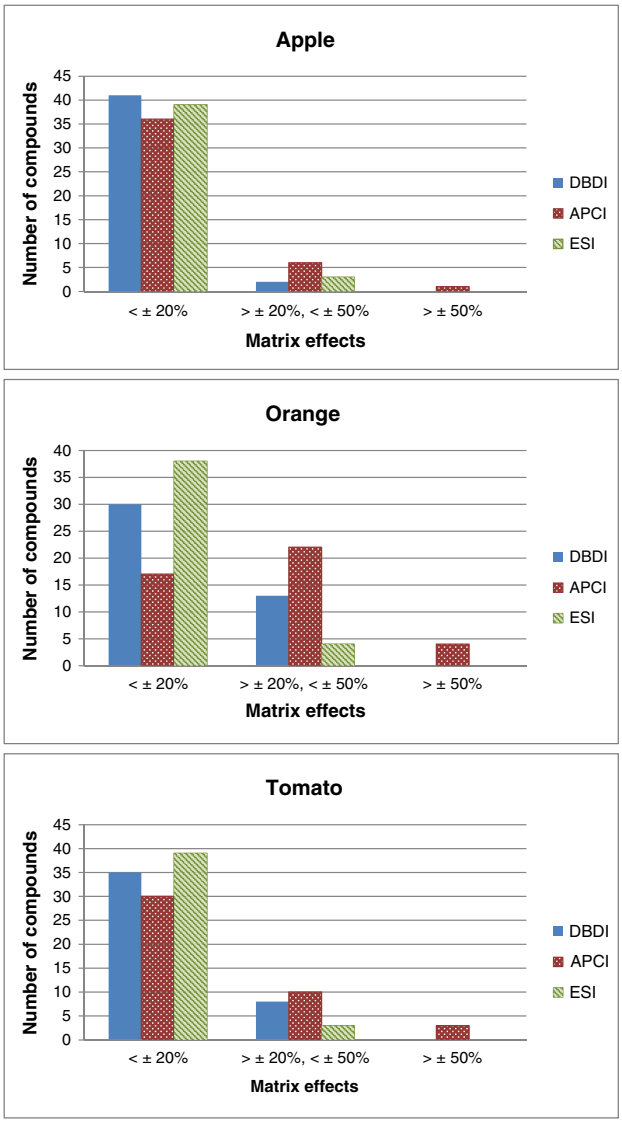

Figure 1. Evaluation of the matrix effects displayed by DBDI, APCI and ESI sources during LC/MS analyses of the studied pesticides in each individual vegetable matrix tested (apple, orange and tomato).

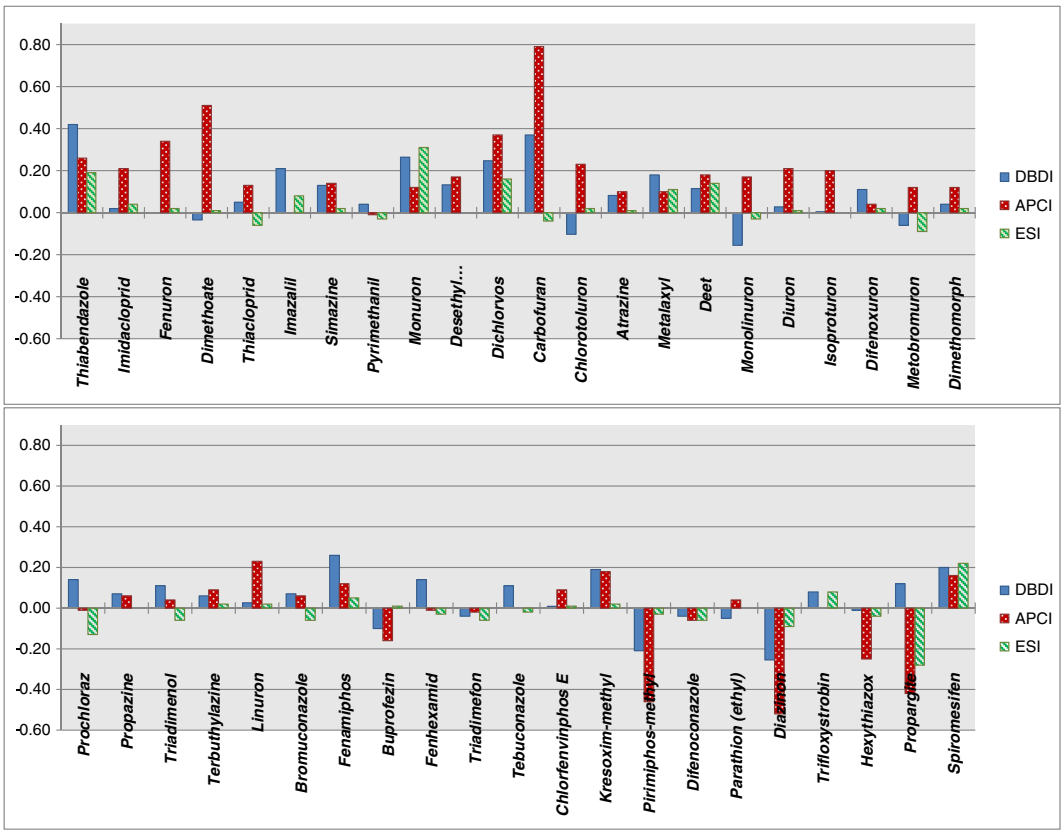

Figure 2. Absolute signal suppression or enhancement obtained in tomato extract during LC/MS analyses of the studied pesticides using the different ionization sources tested.

extent on signal decrease has been reported for ESI than for APCI.[23,38] Both ion suppression and enhancement have been observed for LC/MS/MS using APCI.[39,40] Gas-phase neutralization processes and coprecipitation of the analytes with nonvolatile materials may be responsible for the matrix effects observed with APCI.

The case of carbofuran is particularly interesting. This compound showed minor signal enhancement (<20%, in apple extract) or moderate signal enhancement (<40%, in orange and tomato extracts) by DBDI ionization, a strong signal enhancement (>50% in all tested matrices) by APCI ionization, and a small signal suppression (<20% in all tested matrices) by ESI ionization. A carefully study of the mass spectrum of carbofuran revealed the presence of the ion *m/z* 295.22677 in all matrices tested (apple, orange and tomato) within a mass error below of 2 ppm in all cases. Additionally, *m/z* 277.21551 was detected by APCI and DBDI, whereas *m/z* 317.20910 was observed solely by ESI with the same elution profile. The first mass corresponds to a neutral loss of water from *m/z* 295.22677, the second mass to the sodium adduct $[M+Na]^+$ of *m/z* 295.22677. The absence of this species in solvent standards and its presence in all matrices tested at a similar intensity suggested that it could be an interfering species related to the sample treatment. This presumption was confirmed by checking the list of common interfering species in mass spectrometry elaborated by Keller *et al.*,[41] where the ion at *m/z* 295.22677 is assigned to a fragment of Triton: $[(C_{14}H_{22}O)(C_2H_4O)_2+H]^+$. The presence of the detergent Triton (i.e. octylphenol ethoxylate) is corroborated by detection of higher oligomers with up to 14 ethoxylate moieties. Furthermore, masses related to reduced Triton $[(C_{14}H_{28}O)(C_2H_4O)_n$ were present in the samples. The relative abundances of the ions in the spectrum suggested that the compound of *m/z* 295.22677 is the main reason for the observed matrix effects. Thus, greater signal enhancement is observed at higher abundances of the interferent, and this effect is more pronounced in APCI than in DBDI. In ESI, neither the interference abundance nor matrix effect is relevant (see Fig. 3). Possible explanations are altered vaporization efficiency by the abovementioned coeluting surface active compound or proton transfer reactions that occurs in the gas phase, facilitating the ionization of carbofuran molecules.

CONCLUDING REMARKS

A study to evaluate the usefulness of LC/DBDI-MS for quantitative analyses of pesticides in fruit and vegetables was carried out. The results of the analysis of spiked concentrated extracts showed that the sensitivity obtained with LC/DBDI-MS is appropriate for multi-residue analysis of pesticide residues in fruit and vegetable samples. The LOQs obtained for most of the studied pesticides are in compliance with the European regulation 396/2005 (and its subsequent updates) on food commodities (default MRL of 10 μg kg^{-1}). Comparative studies with commercial sources demonstrate the suitability of DBDI as an ionization source

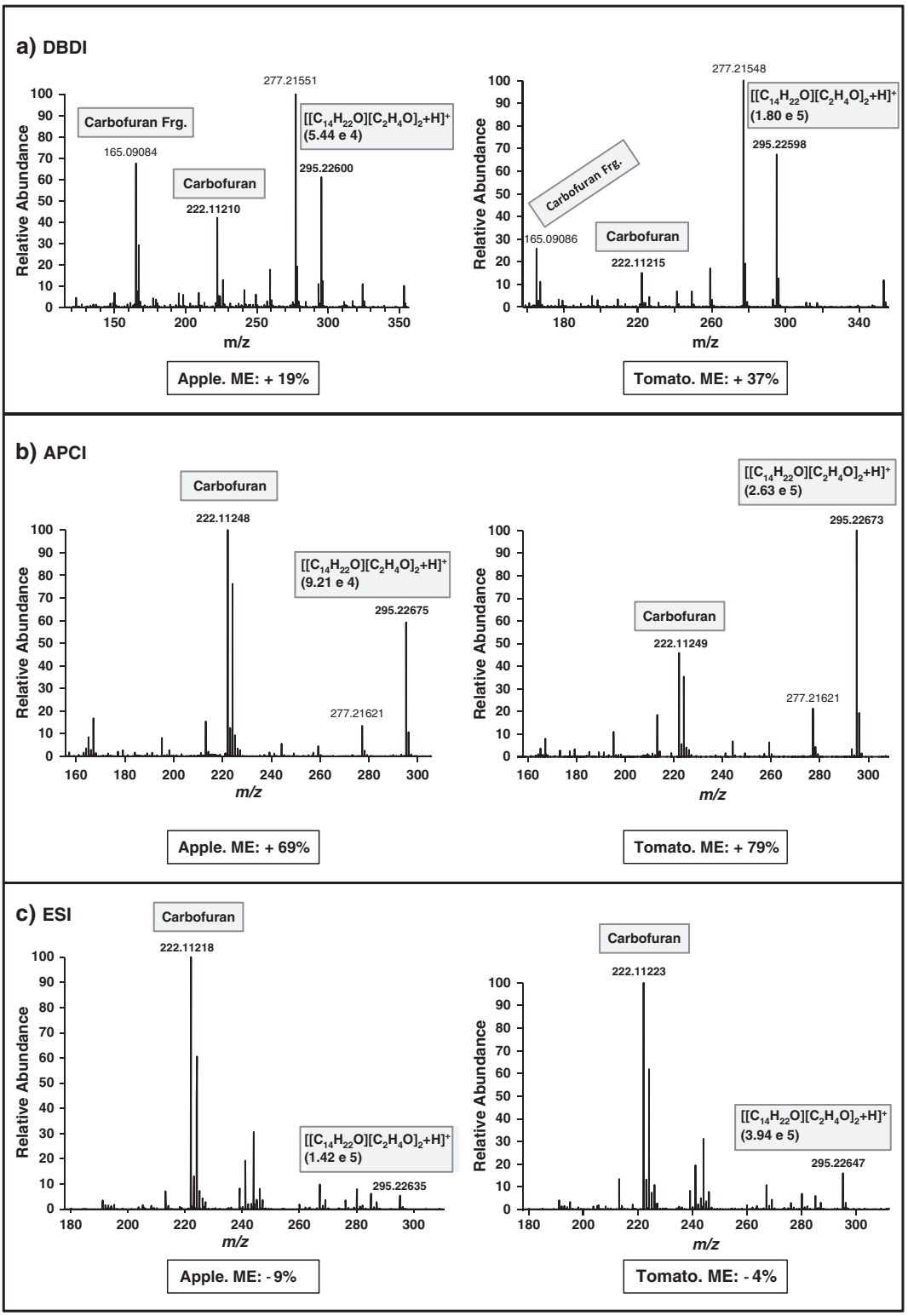

Figure 3. Mass spectrum corresponding to the peak of carbofuran (200 μg L^{-1}; Rt at 13.82 min) obtained from matrix-matched standards of apple and tomato extracts: (a) by DBDI; (b) by APCI; and (c) by ESI. Matrix effect (ME) is the percentage of signal enhancement (+) or suppression (-).

for residue analysis, because (1) the use of DBDI decreased the matrix effects obtained by APCI, and (2) improved the detection – in terms of sensitivity – of selected compounds that are not easily ionized by ESI.

The investigation of matrix effects demonstrated that their extent is dependent on the ionization source. ESI is mainly characterized by signal suppression, whereas APCI and DBDI showed mainly signal enhancement. The knowledge

of these differences is particularly important at values close to the MRLs because matrix enhancement may lead to increased false positive and matrix suppression to higher false negative rates, respectively. The extent of formation of the desired ions was observed to be different even in the presence of the same co-eluting compounds (i.e. Triton fragments) since it depends on the mutual positive or negative effects that the co-eluting species play on the ion formation. The effect of surface-active compounds was not directly tested. However, the importance of surface activities of the analyte and interfering compounds may also play an important role in matrix effects. More work is needed to obtain a thorough understanding of the causes of these phenomena.

SUPPORTING INFORMATION

Additional supporting information may be found in the online version of this article.

Acknowledgements

Financial support from the "Ministerium für Innovation, Wissenschaft und Forschung des Landes Nordrhein-Westfalen" and by the "Bundesministerium für Bildung und Forschung" is gratefully acknowledged. The authors also acknowledge funding support from the Regional Government of Andalusia (Spain) "*Junta de Andalucía*" (Research Group FQM-323) and the Spanish Ministerio de Ciencia e Innovación (Project CTQ-2012-34297). B.G.-L. also acknowledges a scholarship from the German Academic Exchange Service (Postdoctoral Leibniz-DAAD program, PKZ: A/11/94543).

REFERENCES

[1] R. M. González-Rodríguez, R. Rial-Otero, B. Cancho-Grande, C. Gonzalez-Barreiro, J. Simal-Gándara. A review on the fate of pesticides during the processes within the food-production chain. *Crit. Rev. Food Sci.* **2011**, *51*, 99.
[2] Food Contaminants and Residue Analysis, (Ed: Y. Picó). Comprehensive Analytical Chemistry, (Series Ed: D. Barceló), vol. 51, Elsevier, **2010**.
[3] Chromatographic-Mass Spectrometric Food Analysis for Trace Determination of Pesticide Residues, (Ed: A. R. Fernández-Alba). Comprehensive Analytical Chemistry, (Series Ed: D. Barceló), vol. 43, Elsevier, **2005**.
[4] A. K. Malik, C. Blasco, Y. Picó. Liquid chromatography–mass spectrometry in food safety. *J. Chromatogr. A* **2010**, *1217*, 4018.
[5] L. Alder, K. Greulich, G. Kempe, B. Vieth. Residue analysis of 500 high priority pesticides: Better by GC–MS or LC–MS/MS? *Mass Spectrom. Rev.* **2006**, *25*, 838.
[6] D. B. Robb, T. R. Covey, A. P. Bruins. Atmospheric pressure photoionization: An ionization method for liquid chromatography–mass spectrometry. *Anal. Chem.* **2000**, *72*, 3653.
[7] A. Raffaelli, A. Saba. Atmospheric pressure photoionization mass spectrometry. *Mass Spectrom. Rev.* **2003**, *22*, 318.
[8] S. J. Bos, S. M. Van Leeuwen, U. Karst. From fundamentals to applications: recent developments in atmospheric pressure photoionization mass spectrometry. *Anal. Bioanal. Chem.* **2006**, *384*, 85.
[9] H. Moriwaki, M. Ishitake, S. Yoshikawa, H. Miyakoda, J. F. Alary. Determination of polycyclic aromatic hydrocarbons in sediment by liquid chromatography-atmospheric pressure photoionization-mass spectrometry. *Anal. Sci.* **2004**, *20*, 375.
[10] L. C. Short, K. A. Hanold, S. S. Cai, J. A. Syage. Electrospray ionization/atmospheric pressure photoionization multimode source for low-flow liquid chromatography/mass spectrometric analysis. *Rapid Commun. Mass Spectrom.* **2007**, *21*, 1561.
[11] M. Constapel, M. Schellenträger, O. J. Schmitz, S. Gäb, K. J. Brockmann, R. Giese, T. Benter. Atmospheric-pressure laser ionization: a novel ionization method for liquid chromatography/mass spectrometry. *Rapid Commun. Mass Spectrom.* **2005**, *19*, 326.
[12] R. Schiewek, M. Lorenz, R. Giese, K. Brockmann, T. Benter, S. Gäb, O. J. Schmitz. Development of a multipurpose ion source for LC-MS and GC-API MS. *Anal. Bioanal. Chem.* **2008**, *392*, 87.
[13] P. Schmitt-Kopplin, M. Englmann, R. Rossello-Mora, R. Schiewek, K. J. Brockmann, T. Benter, O. J. Schmitz. Combining chip-ESI with APLI (cESILI) as a multimode source for analysis of complex mixtures with ultrahigh-resolution mass spectrometry. *Anal. Bioanal. Chem.* **2008**, *391*, 2803.
[14] G. Diehl, U. Karst. On-line electrochemistry – MS and related techniques. *Anal. Bioanal. Chem.* **2002**, *373*, 390.
[15] G. Diehl, A. Liesener, U. Karst. Liquid chromatography with post-column electrochemical treatment and mass spectrometric detection of non-polar compounds. *Analyst* **2001**, *126*, 288.
[16] S. M. Van Leeuwen, H. Hayen, U. Karst. Liquid chromatography–electrochemistry–mass spectrometry of polycyclic aromatic hydrocarbons. *Anal. Bioanal. Chem.* **2004**, *378*, 917.
[17] A. Cappiello, G. Famiglini, P. Palma. Electron ionization for LC/MS. *Anal. Chem.* **2003**, *75*, 496A.
[18] G. Famiglini, P. Palma, E. Pierini, H. Trufelli, A. Cappiello. Organochlorine pesticides by LC–MS. *Anal. Chem.* **2008**, *80*, 3445.
[19] A. Cappiello, G. Famiglini, E. Pierini, P. Palma, H. Trufelli. Advanced liquid chromatography – mass spectrometry interface based on electron ionization. *Anal. Chem.* **2007**, *79*, 5364.
[20] H. Hayen, A. Michels, J. Franzke. Dielectric barrier discharge ionization for liquid chromatography/mass spectrometry. *Anal. Chem.* **2009**, *81*, 10239.
[21] B. Gilbert-López, J. F. García-Reyes, C. Meyer, A. Michels, J. Franzke, A. Molina-Díaz, H. Hayen. Simultaneous testing of multiclass organic contaminants in food and environment by liquid chromatography/dielectric barrier discharge ionization-mass spectrometry. *Analyst* **2012**, *137*, 5403.
[22] P. Kebarle, L. Tang. From ions in solution to ions in the gas phase – the mechanism of electrospray mass spectrometry. *Anal. Chem.* **1993**, *65*, 972A.
[23] R. King, R. Bonfiglio, C. Fernandez-Metzler, C. Miller-Stein, T. Olah. Mechanistic investigation of ionization suppression in electrospray ionization. *J. Am. Soc. Mass Spectrom.* **2000**, *11*, 942.
[24] N. B. Cech, C. G. Enke. Relating electrospray ionization response to nonpolar character of small peptides. *Anal. Chem.* **2000**, *72*, 2717.
[25] B. K. Matuszewski, M. L. Constanzer, C. M. Chavez-Eng. Strategies for the assessment of matrix effect in quantitative bioanalytical methods based on HPLC–MS/MS. *Anal. Chem.* **2003**, *75*, 3019.
[26] H. Stahnke, S. Kittlaus, G. Kempe, L. Alder. Reduction of matrix effects in liquid chromatography–electrospray ionization–mass spectrometry by dilution of the sample extracts: How much dilution is needed? *Anal. Chem.* **2012**, *84*, 1474.

[27] F. Gosetti, E. Mazzucco, D. Zampieri, M. C. Gennaro. Signal suppression/enhancement in high-performance liquid chromatography tandem mass spectrometry. *J. Chromatogr. A* **2010**, *1217*, 3929.
[28] H. R. Liang, R. L. Foltz, M. Meng, P. Bennet. Ionization enhancement in atmospheric pressure chemical ionization and suppression in electrospray ionization between target drugs and stable-isotope-labeled internal standards in quantitative liquid chromatography/tandem mass spectrometry. *Rapid Commun. Mass Spectrom.* **2003**, *17*, 2815.
[29] A. Kaufmann. The current role of high-resolution mass spectrometry in food analysis. *Anal. Bioanal. Chem.* **2012**, *403*, 1233.
[30] J. F. García-Reyes, M. D. Hernando, A. Molina-Díaz, A. R. Fernández-Alba. Comprehensive screening of target, non-target and unknown pesticides in food by LC-TOFMS. *Trends Anal. Chem.* **2007**, *26*, 828.
[31] M. Anastassiades, S. J. Lehotay, D. Stajnbaher, F. J. Schenck. Fast and easy multiresidue method employing acetonitrile extraction/partitioning and "dispersive solid-phase extraction" for the determination of pesticide residues in produce. *J. AOAC Int.* **2003**, *86*, 412.
[32] European Parliament and the Council. Regulation (EC) 396/2005 on maximum residue levels in or on food and feed of plant and animal origin amending Council Directive 91/414/EEC. *Off. J. Eur. Union* **2005**, *L-70*, 1.
[33] M. Mezcua, O. Malato, J. F. García-Reyes, A. Molina-Díaz, A. R. Fernández-Alba. Accurate-mass databases for comprehensive screening of pesticide residues in food by fast liquid chromatography time-of-flight mass spectrometry. *Anal. Chem.* **2009**, *81*, 913.
[34] SANCO/12495/2011. Method validation and quality control procedures for pesticide residue analysis in food and feed (Implemented by 01/01/2012). Available: http://ec.europa.eu/food/plant/protection/pesticides/docs/qualcontrol_en.pdf.
[35] C. Aguilar, I. Ferrer, F. Borrull, R. M. Marcé, D. Barceló. Comparison of automated on-line solid-phase extraction followed by liquid chromatography–mass spectrometry with atmospheric pressure chemical ionization and particle beam mass spectrometry for the determination of a priority group of pesticides in environmental waters. *J. Chromatogr. A* **1998**, *794*, 147.
[36] E. M. Thurman, I. Ferrer, D. Barceló. Choosing between atmospheric pressure chemical ionization and electrospray ionization interfaces for the HPLC/MS analysis of pesticides. *Anal. Chem.* **2001**, *73*, 5441.
[37] K. Maštovská, S. J. Lehotay, M. Anastassiades. Combination of analyte protectants to overcome matrix effects in routine GC analysis of pesticide residues in food matrices. *Anal. Chem.* **2005**, *77*, 8129.
[38] N. C. Marangou, N. S. Thomaidis, M. A. Koupparis. Optimization and comparison of ESI and APCI LC-MS/MS methods: A case study of Irgarol 1051, diuron, and their degradation products in environmental samples. *J. Am. Soc. Mass Spectrom.* **2011**, *22*, 1826.
[39] J. Smeraglia, S. F. Baldrey, D. Watson. Matrix effects and selectivity issues in LC-MS-MS. *Chromatographia* **2002**, *55*, S95.
[40] M. W. J. van Hout, H. A. G. Niederländer, R. A. de Zeeuw, G. J. de Jong. Ion suppression in the determination of clenbuterol in urine by solid-phase extraction atmospheric pressure chemical ionisation ion-trap mass spectrometry. *Rapid Commun. Mass Spectrom.* **2003**, *17*, 245.
[41] B. O. Keller, J. Sui, A. B. Young, R. M. Whittal. Interferences and contaminants encountered in modern mass spectrometry. *Anal. Chim. Acta* **2008**, *627*, 71.

