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Food Safety and Environmental Analyses
and more...

Director's note

Dear Reader,

I am proud to announce the publication of the first issue of ***Shimadzu Journal***. Guided by our corporate philosophy, "Contributing to Society through Science and Technology," we conduct our business to meet customers' needs in a wide variety of fields throughout the world. Our new brand statement, "Excellence in Science," reflects our desire and attitude to diligently respond to customers' requirements by offering superior, world-class technologies.

The Analytical and Measuring Instruments Division offers state-of-the-art solutions in a wide variety of fields that impact the health and lives of people. These fields include the environment, food safety, pharmaceuticals and medical diagnostics. Our diverse product lineup ranks as the best in the industry. This lineup includes mass spectrometers, chromatographs, life science instruments, spectrophotometers, surface analytical instruments, microscopes, environmental monitors of exhaust gas and water, strength and fatigue testing machines, X-ray inspection devices, balances, and thermal analyzers.

Currently, we are focusing on the fields of pharmaceuticals, life science, and food safety, working toward providing solutions that improve the lives and health of people. As a manufacturer, we believe that collaborating with researchers is the best way to develop new solutions that will deliver true contributions to the world. Therefore, in this Journal, we introduce collaborative research projects and share the latest information and results with readers.

This inaugural issue focuses on food safety and environmental analysis, and contains information on two collaborations. One is with Professor Mondello of University of Messina, Italy. An interview with Professor Mondello, who is becoming the leading light in the field of the Comprehensive GC x GC / LC technology, which is widely applicable to the fields of food, fragrance and petrochemistry, provides details on his current studies and the challenges he faces. The second collaboration is with the Food and Environment Research Agency, UK. This application note presents an analysis of pesticide residues in food. In addition, the journal contains information on other applicable topics, as well as the latest news and applications.

We aim to be a good partner for you. We strive to meet your needs with the highest technological capabilities and valuable platforms and solutions that you can use with confidence. I hope that this journal will be of great help to all of you.

Yours Sincerely,

Teruhisa UEDA, PhD.

General Manager, Analytical & Measurement Instruments Division

CONTENT

Featuring on Food Safety and Environmental analysis

Prof. Luigi Mondello
The Pioneer of Multidimensional Chromatography

We interviewed Professor Luigi Mondello of University of Messina in Italy, who has become the leading light in the field of multidimensional chromatography. Shimadzu has been collaborating with Prof. Mondello and his team since 1996, funding and providing instruments to further advance the techniques.

Shimadzu:

Please let us know the theme and outline of your study, as well as the goal.

Prof. Mondello:

A GC×GC-MS/FID method has been developed for the elucidation of an important group of lipid compounds in vegetable oil and dairy products, namely the unsaponifiable matter. The sensitivity and the formation of group-type patterns were the GC×GC characteristics most exploited in the specific applications reported. Such a method can certainly be used to assess the quality (e.g., presence or absence of degradation products) and genuineness (e.g., vegetable oil adulteration, presence or absence of phytosterols derived from vegetable lipids) in vegetable oil and dairy products.
I now focus on ultra- and very-fast gas chromatography that allows users to perform analyses in greatly reduced times, without losing information. Fast gas chromatography units are often linked with mass spectrometers for qualitative and quantitative objectives. Such methods provide the possibility of obtaining real-time results, and are very useful when analyzing a high number of samples or when needing immediate answers.
I and Shimadzu are also upgrading comprehensive two-dimensional and multidimensional chromatography systems. I apply such approaches to the analysis of samples, ranging from plant extracts, food products, petrochemicals, pharmaceutical and cosmetic products, to various environmental substances.
In particular, the development and introduction of multidimensional instrumentation, as well as innovative software, has contributed greatly toward revealing the unsuspected complexity of many real-world samples.
Ten years from now, the comprehensive chromatography methods we are working with, such as LC×LC, GC×GC and LC×GC, will have a revolutionary effect on the chromatography community. If we succeed in making these powerful technologies more accessible, in terms of both hardware and software, then the impact will be a great one.

Shimadzu:

What stage are you now on and what comes next? What are the challenges?

Prof. Mondello:

I plan to continue developing instrumentation for fast GC, with attention focused on the injection system, and to develop simpler and more effective modulators for comprehensive chromatography. I also plan to engage in the development multidimensional chromatography software and MS spectra databases, which will make compound identification a simpler and more reliable task. Our main common goal, namely the evolution of chromatography-mass spectrometry technologies, will be achieved within the context of our intense collaboration with Shimadzu.

Shimadzu:

What do you expect from Shimadzu as a partner and does Shimadzu meet your expectations?

Prof. Mondello:

Without Shimadzu I could not have achieved all of this. I started working with Shimadzu because of the company's willingness to revolutionize instrument technology. In the last ten years Shimadzu has developed entirely new instrumentation. They did not just remake old instruments. I also like the Japanese way of solving problems–slowly but with excellent results. We tested, advised and validated these powerful devices for speed, sensitivity and selectivity. It has been a very nice and fruitful collaboration.
When I first started my collaboration with Shimadzu, fast GC and GC-MS were barely employed due to the lack of commercial instrumentation. Now, fast methods are routinely employed in many industrial and academic fields. Comprehensive two-dimensional gas, liquid and liquid-gas chromatography hardware and software have been developed and are now exploited by a great number of analysts across the world. GC×GC methods are making greater inroads in the analysis of fatty acid methyl esters in food and biological samples, pesticides and petrochemicals, as well as flavors and fragrances, while LC×LC, linked with mass spectrometry, is increasingly useful for a range of tests related to health, biology and nutrition, including proteomics, lipidomics, and food antioxidant analysis. LC×GC methods have a demonstrated effectiveness for food contaminant analysis (e.g., mineral oil in vegetable oils). In short, analytical horizons have been extended.

Shimadzu:

Do you have any suggestions or expectations for this global technical journal?

Prof. Mondello:

Because the journal is focused on the description and presentation of modern powerful analytical instrumentation, it would also be nice to dedicate a space on how, when and by whom specific methods were introduced to the field. Some topics could be, for example, mass spectrometry in general, triple quad MS, comprehensive 2D LC, 2D GC, etc.

Shimadzu:

Thank you very much.

A GCxGC Handbook and Application Compendium authored by Prof. Mondello are available at:
www.shimadzu.com/an/gcms/gcgc.html

And here are his latest publications:

(1)Peter Quinto Tranchida, Flavio Antonio Franchina, Mariosimone Zoccali, Sebastiano Panto', Danilo Sciarrone, Paola Dugo, Luigi Mondello "Untargeted And Targeted Comprehensive Two-Dimensional GC Analysis Using A Novel Unified High-Speed Triple Quadrupole Mass Spectrometer" J. Chromatogr. A 1278 (2013) 153-159

(2)Peter Q. Tranchida, Simona Salivo, Flavio A. Franchina, Ivana Bonaccorsi, Paola Dugo, Luigi Mondello "Qualitative and quantitative analysis of the unsaponifiable fraction of vegetable oils by using comprehensive 2D GC with dual MS/FID detection" Anal. Bioanal. Chem. 405 (2013) 4655-4663

Analysis of MOSH and MOAH using SPE prior to GC×GC-MS analysis

Luigi Mondello[1]

Abstract

The present work is focused on the development/optimization of a comprehensive two-dimensional gas chromatography method, with dual detection [flame ionization (FID) and mass spectrometric], for the simultaneous identification and quantification of mineral-oil contaminants in a variety of food products. The two main classes of contaminants, namely saturated and aromatic hydrocarbons, were previously fractionated on a manually-packed silver silica solid-phase extraction (SPE) cartridge. The presence of a series of unknown compounds was investigated using the mass spectrometric data, and were tentatively-identified as esterified fatty acids, most probably derived from vegetable oil based ink.

Keyword:Food, MOSH, MOAH, GC×GC, Comprehensive GC, Quadrupole mass spectrometer

1. Introduction

Mineral oil products derive from crude petroleum, through distillation processes and various refining steps, and contain proportions of mineral oil saturated hydrocarbons (MOSH, including *n*-alkanes, isoalkanes and cycloalkanes), and mineral oil aromatic hydrocarbons (MOAH), mainly consisting of alkylated polyaromatic hydrocarbons (PAH)[1].

Mineral oil contamination in foods, deriving from a variety of sources, has been studied for quite a long time[2-4]. One of the major sources of contamination is paperboard packaging, an issue known since 1997[2], even though it has gained great attention only recently[5,6] Such a contamination derives from the printing inks applied directly to the packaging, and/or from the ink used in the newspapers, employed to produce recycled fiber.It has also been demonstrated that mineral oil migrating from paperboard usually contains a large proportion (15-20%) of MOAH[3,4], which are more of a worry from a toxicological viewpoint.

The occurrence and danger of mineral oil products in foods has been discussed widely in recent years[7-10]. The Joint FAO/WHO Expert Committee on Food Additives (JECFA), in 2002, reported a list of admissible daily intake (ADI) values for different white mineral oils[8]; based on such data, an envisioned limit of 0.6 mg kg^{-1} was proposed for MOSH migration (up to C25) in dry foods from paperboard packaging[7]. The European Food Safety Authority (EFSA), published an opinion in June 2012[9], casting doubts on the "JECFA" list, due to the lack of sufficient toxicological information and, as a consequence, the JECFA values were recently withdrawn[10]. Furthermore, even though EFSA emphasized the potential carcinogenic risk of MOAH constituents[9], an official approved evaluation of MOAH is still lacking.

Most of the approaches reported over the last decades have been directed to the analysis of MOSH, exploited as a contamination marker, using both off- and on-line techniques. Off-line methods based on prep LC, or solid-phase extraction (SPE), have been described[11-18]. The lipid fraction can be eliminated either through saponification, followed by silica-gel column chromatography [11,12], or directly through a prep LC silica column[13,14], or an SPE cartridge[15-18]. Several techniques, based on the use of glass SPE, have been described; with regards to packing materials, a variety of solutions have been proposed, such as activated silica gel[16], non-activated silica gel[17], or silver (Ag) silica gel[18].

Considering the application of all methods, it can be affirmed without a doubt, that the most popular technique has been on-line liquid chromatography-gas chromatography (LC-GC), with a silica LC column[6,19-23]. Additionally,and in consideration of the toxicological relevance of MOAH, work has been directed to the clear pre-separation of the MOSH from the MOAH. For example, Biedermann and co-workers exploited the separation efficiency of an LC silica column, in an on-line LC-GC system, to separate the MOAH from the MOSH, and these from the lipidmatrix[20]. It must also be noted that off-line SPE methods, using a Ag silica-gel SPE cartridge, have been developed for MOSH and MOAH determination[24,25].

With regards to detectors, flame ionization (FID) systems have been widely employed for the reliable quantification of the humps of unresolved complex mixtures (UCM), generated in MOSH/MOAH applications; FIDs are useful because they provide virtually the same response *per* mass of hydrocarbons, eventhough the lack of structural information is certainly a major drawback[23]. In fact, the attainment of profound information on the composition of MOSH and MOAH constituents, can provide

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fundamental information on potential toxicity, and on the contamination source. Such an objective was reached by Biedermann and Grob, who used an MS detector, along with the additional information generated by a comprehensive 2D GC (GC×GC) analysis[26]. A pre-separation of the MOSH and MOAH groups was achieved through off-line LC, a process necessary to avoid the overlapping of steranes and hopanes (present in the MOSH fraction), with alkylated (two- and three-ring) aromatics. The GC×GC system was coupled alternatively with an MS system, for qualitative purposes, and with an FID system for quantification, and hence, two applications were required to obtain both information types. A GC×GC-MS method, after an off-line LC pre-separation step, has also been exploited by Mondello and co-workers, to attain a more expanded view on MOSH contamination in homogenized baby foods[27].

The present document describes a GC×GC method,characterized by dual MS/FID detection, for the qualitative and quantitative analysis of MOSH and MOAH in various foods.The pre-separation step was performed by using Ag-SPE.

2. Experimental

2-1. Samples and chemicals

CH_2Cl_2 and *n*-hexane were purchased from Sigma-Aldrich (Milan, Italy), and distilled before use. The C7-C40 standard mixture,the paraffin oil (code 18512), $AgNO_3$, and silica gel 60 (particle size 0.063-0.2 mm, 70-230 mesh) were purchased from Supelco and Sigma-Aldrich (Milan). Glass SPE cartridges (6 mL glass tubes with a frit) were purchased from Macherey-Nagel (Chromabond, Düren, Germany).

2-2. Samples and preparations

Samples of pasta, rice and icing sugar, were purchased in a supermarket. The ground samples were extracted overnight using *n*-hexane, and then purified through Ag-SPE. Briefly, a 1:2 food to solvent ratio was employed to extract MOSH and MOAH from the samples. After, an aliquot of the extract was concentrated prior to SPE clean-up, on a Ag silica gel cartridge. Silver silica gel was prepared by adding a $AgNO_3$ solution (0.75 g/mL in Milli-Q water, Millipore, Bedford, MA, USA) to previously activated (400°C overnight) silica gel, blended for about 30 min, and left to rest for 12 h; finally, the mixture was heated at 75°C overnight to eliminate the remaining water. The SPE cartridge was manually packed with 1 g of Ag silica,prior to sample loading (250 µL). First, the sample was eluted with 1 mL of *n*-hexane, which was discharged; then, the MOSH constituents were eluted with 1.5 mL of *n*-hexane, followed by 0.5 mL of *n*-hexane/dichloromethane (50:50 *v/v*); a 0.5 mL *n*-hexane/dichloromethane fraction followed, which was discharged; finally, the MOAH class was eluted with further 7 mL of *n*-hexane/dichloromethane (50:50 *v/v*).

The eluted fractions were concentrated to a final volume of 100 µL to increase sensitivity, since large volume injection (LVI) was not used.

2-3. GC×GC-MS/FID analysis

GC×GC experiments were performed on a system consisting of a GC2010 gas chromatograph, and a QP2010 Ultra quadrupole mass spectrometer (Shimadzu, Kyoto, Japan).

The primary column, an SLB-5ms 30 m × 0.25 mm ID × 0.25 µm d_f [silphenylene polymer, virtually equivalent in polarity to poly (5% diphenyl/95% methyl siloxane)], was connected to an uncoated capillary segment (1.0 m × 0.25 mm ID, used to create a double-loop), and to a 1.0 m × 0.10 mm ID × 0.10 µm d_f Supelcowax-10 (polyethylene glycol) segment (Supelco). The second column was connected through a capillary column splitter (SGE) to two uncoated capillaries, with these linked to the FID (0.5 m × 0.1 mm ID) and to the MS (0.25 m × 0.05 mm ID) systems.

2-4. Method parameters

Modulation was performed every 6000 msec, by using a loop-type modulator (under license from Zoex Corporation, Houston, TX, USA). The duration of the hot pulse (350°C) was 375 msec.

GC oven temperature program: 50°C to 280°C (hold 7.5 min) at 4°C/min. Carrier gas, He, was supplied at an initial pressure of 243 kPa (constant linear velocity mode). Injection temperature: 360°C. Injection mode and volume: pulsed injection (300 kPa hold for 1 min) in the split mode (1:10); 6 µL. The FID was operated as follows: H_2 flow: 40.0 mL/min; air flow: 400.0 mL/min; make up (He): 30.0 mL/min.

MS parameters: samples were analyzed in the full scan mode with a scan speed of 20,000 amu/sec and a mass range of 40-510 *m/z*; spectra generation frequency: 33 Hz; interface and ion source temperatures were 250°C and 200°C, respectively. MS ionization mode: electron ionization.

Bidimensional visualization was carried out by using the ChromSquarev. 1.5 software (Shimadzu Europe, Duisburg, Germany). The MS libraries used for spectral matching were NIST05, FFNSC, and FAME library.

3. Results and Discussion

3-1. GC×GC-MS/FID *optimization and validation*

GC×GC method optimization was achieved by using offset printing ink, which is formed mainly of MOSH (> 90%), and by a minor MOAH fraction. Apart from problems related to co-elution, if the offset ink had been injected neat then the MOSH group would have overloaded the columns and modulator, while the MOAH constituents would have been barely detected; therefore, a pre-separation on the Ag-SPE cartridge was necessary.

Flow division between the FID and MS units was a compromise among different necessities, the main one being the attainment of a satisfactory sensitivity for quantification purposes. Because the detectors employed operate under different pressure conditions, the employment of two branches with equal IDs proved to be a non-ideal choice; the reason was related to the fact that an excessively long "MS" branch was required to generate an adequate flow resistance, to divert the majority of the effluent to the FID. Such a configuration would have led to substantial differences in the second-dimension elution times, between the qualitative and quantitative experiments. A good compromise was found through the use of an MS-linked 0.25 m × 0.05 mm ID branch, and a 0.5 m × 0.1 mm ID FID one.

Such a splitting configuration produced the following flow conditions:about 84% and 16% of the effluent reaching the FID and MS at the initial analysis temperature, respectively. The split ratio changed slightly during the GC run, with about 87% and 13% of the effluent diverted to the FID and MS, at the end. Since the calibration curve was constructed under the same analytical conditions, the quantitative results were not affected.

The GC×GC dual-detection operational conditions were optimized with the aim of maintaining the same chromatography performance, compared to an MS-only system, as shown in Fig. 1. In the MS-only approach, with the same analytical columns, the head pressure (approx. 150 kPa) was selected to generate about 20 cm/sec and 210 cm/sec, in the first and second dimension, respectively. In the dual-detection approach, a 243-kPa pressure produced the same gas velocity in the first dimension (to attain the same elution temperatures), and a slightly lower one in the second (180 cm/sec).

Asix-point (each point was derived through duplicate applications) calibration curve was constructed through the FID trace, using solutions of paraffin oil in *n*-hexane, in the 0.35-24 mg/Kg range. The least squares method was exploited to estimate the regression line, while the linearity and the goodness of the curve were evaluated through the regression coefficient (0.9993), and a visual inspection of the residual plot, and were confirmed using Mandel's fitting test (Fcalc<Ftab). The significance of the intercept ($p = 0.03$) was established running a *t*-test, at the 5% significance level.

Measurement of the limit of quantification (LoQ), in mineral oil analyses, is tightly related to the MW distribution of the contaminants, hence on the hump width. However an approximate estimation of the LoQ was made by considering the standard deviation (n=3), calculated at the lowest calibration point, multiplied by 10. The LoQ was estimated to be approximately 1.2 mg/Kg.

3-2. Food analysis

MOSH and the MOAH fractions, relative to pasta, icing sugar and rice, were quantified up to C25 (as required by the envisioned limit), using the aforementioned method; attention was paid, during integration, to eliminate the natural alkanes from the MOSH compounds, and the "unknown" peaks from the MOAH group. Specifically, for GC×GC-FID quantification, the "polygonal integration function" was applied, which enabled the definition of a polygonal area in which all the integrated peaks are automatically summed, and the data relative to each peak is saved as well. Thus, the undesired peaks can be easily selected, and subtracted from the total area. Quantification information, relative to the three foods, is listed in Table 1.

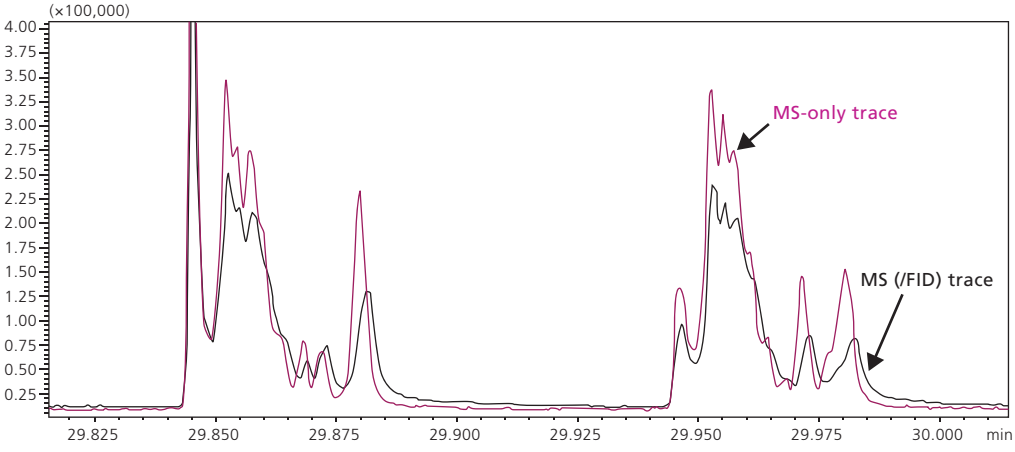

Fig. 1 Comparison of raw TIC chromatogram expansions (printing ink analyses), obtained using a GC×GC-MS and a GC×GC-MS/FID system

Table 1 Quantification values relative to the MOSH and the MOAH fractions, in samples of pasta, icing sugar, and rice, using Ag-SPE-GC×GC-MS/FID

Food	MOSH <C25 (mg/Kg)	MOAH< C25 (mg/Kg)
Pasta	3.5	1.6
Icing sugar	8.4	1.3
Rice	33.8	2.2

3-3. GC×GC-MS results for the MOAH fraction

The peaks present in the GC×GC chromatograms, for the three samples, were tentatively-identified on the basis of MS database similarities (≥ 80%) and in accordance with linear retention indices (LRI), contained in the same database. Since a widely-accepted procedure for the calculation of GC×GC LRI values has not been developed, such data were calculated in a one-dimensional mode; furthermore, a rather wide LRI filter window (± 25 units) was applied (to eliminate wrong matches), to compensate for the retention effects of the polar capillary.The tentatively identified compounds, along with experimental and database LRI, are listed in Table 2.

Two compounds were outside the LRI range; specifically, octyldodecanoate and octyltetradecanoate were characterized by a difference of +56 and +57 units, respectively. It noteworthy that, in these cases, the database LRI values (http://webbook.nist.gov/chemistry/), were attained using a methyl silicon capillary column [(Ultra-1) 25 m × 0.32 mm × 0.25 µm], while in the present research a 30 m × 0.25 mm ID × 0.25 µ m silphenylene polymer phase was used. Since the similarity matches were satisfactory, and the analyte locations in the 2D chromatogram gave a further idea on the chemical structure, these solutes were given a name.

Figures 2, 3 and 4 show GC×GC-MS chromatograms for the pasta, icing sugar and rice samples, respectively.

Table 2 Compounds identified in the "MOAH" GC×GC–MS analysis; database-derived (database LRI) and experimental LRI (defined as LRI) values, and spectral similarities (MS%)

	compound	pasta MS%	ice sugar MS%	rice MS%	LRI	database LRI
1	Isopropyldodecanoate	94	93	95	1622	1627
2	Dioctylether	92	93	94	1667	1688
3	2-Ethylhexyl octanoate	87	85	87	1703	1715
4	Ethyltetradecanoate	-	-	82	1798	1795
5	Isopropyltetradecanoate	93	94	93	1821	1828
6	Isoamyldodecanoate	-	92	-	1846	1844
7	6,10,14-Trimethyl-2-pentadecanone	-	-	96	1846	1846
8	2-Heptadecanone	-	-	95	1896	1906
9	Methylhexadecanoate	95	91	93	1929	1925
10	Ethylhexadecanoate	92	83	94	1992	1993
11	Isopropylhexadecanoate	90	90	90	2025	2024
12	Abietatriene	84	81	83	2085	2075
13	Octyldodecanoate	86	84	83	2102	2158
14	2-Nonadecanone	-	-	92	2108	2106
15	Methyloctadecanoate	90	92	90	2130	2124
16	Dodecyloctanoate	95	91	-	2175	2177
17	n-Butylhexadecanoate	92	93	90	2198	2188
18	Octyltetradecanoate	83	85	-	2302	2359
19	Tetradecyloctanoate	84	90	-	2380	2375
20	n-Butyloctadecanoate	84	88	87	2395	2388
21	Pentadecyloctanoate	-	85	84	2477	2475
22	Octylhexadecanoate	83	84	83	2504	2505
23	Di(ethylhexyl) phthalate	95	93	94	2542	2550
24	1-Tetracosanol	-	-	92	2697	2710
25	Squalene	93	93	-	2828	2847
26	1-Hexacosanol	91	-	92	2884	2877
27	Tetradecyltetradecanoate	81	81	89	2968	2950

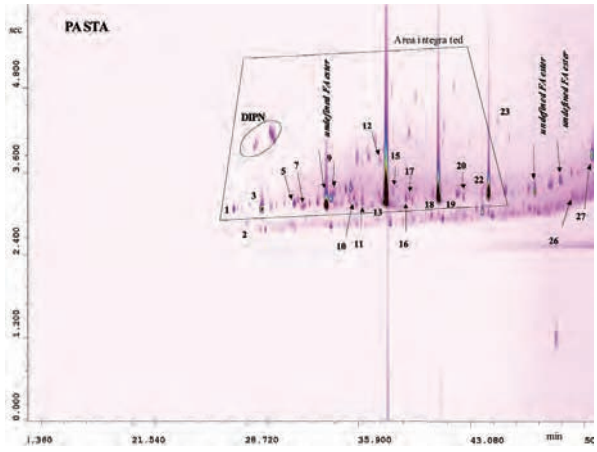

Fig. 2 GC×GC-MS chromatogram, relative to the pasta MOAH fraction. Identification as reported in Table 2. FA: fatty acid; DINP: diisopropylnaphthalenes

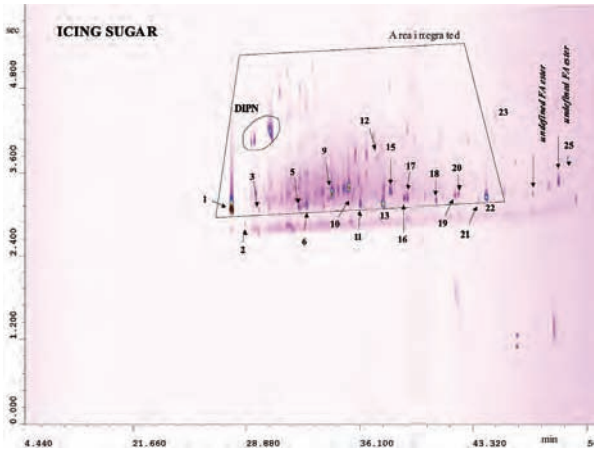

Fig. 3 GC×GC-MS chromatogram, relative to the icing sugar MOAH fraction. Identification as reported in Table 2.

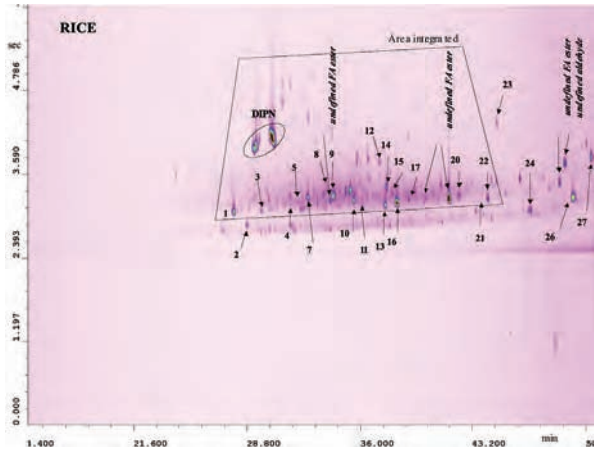

Fig. 4 GC×GC-MS chromatogram, relative to the rice MOAH fraction. Identification as reported in Table 2.

The identification of the specific aromatic compounds, present in the MOAH "cloud", was outside the scope of the investigation; however, even if desired, the identification of such constituents could not have been performed with satisfactory reliability, because of the low amounts of such constituents. However, it was possible to determine the MOAH quantities (FID trace) and patterns, which are highly important to define the contamination source.

A series of peaks, present in the MOAH fraction, were identified as esterified fatty acids. However, their presence did not affect reliable quantification because these compounds were subtracted from the total MOAH area. The esterified fatty acids derived from the paperboard packaging. In fact, in a sample of pasta analyzed prior to box packing, no sign of MOAH contamination was observed.

The possibility to use offset printing ink, based on vegetable oils, has been known for more than fifteen years, though its use has become more frequent since contamination from paperboard packaging has become an issue of worry.

A series of "unknowns" in the GC×GC-MS chromatograms were labeled as "undefined FA esters", since the relative spectra were clearly that of FA esters, although the database searches gave different possible "homologue" matches with good similarities, but not always with a correspondent LRI value. Hence, it was not possible to identify such compounds with sufficient reliability, even though they were marked in the figures, since their chemical nature was evident. It could also not be excluded that such FA esters were not contained in the MS database. For example, in the pasta sample (Fig. 2), only three out of the four main peaks were identified, namely octyldodecanoate, octyltetradecanoate, and octylhexadecanoate. However, it can be deduced from its 2D position that the "undefined FA ester" was most probably octyldecanoate, even though such a compound was not present in the MS databases used. A good "visual" similarity was observed with the spectrum reported in the NIST web site, however no LRI information was found, thus this compound remained unidentified.

It is noteworthy that practically the same compounds were found in all the samples subjected to analyses; however, different quantitative profiles were observed, probably due to a different ink-type and/or to a different contamination source. It can be hypothesized that the vegetable oil offset printing ink was directly used in the pasta packaging (highly contaminated), while it was present, in different amounts, in the recycled fiber used for the packaging of the other two food sample.

Reference

[1] K.Grob, M. Biedermann, A. Caramaschi, B. Pacciarelli, J. High Resolut. Chromatogr. 14 (1991) 33-39.
[2] Ch. Droz, K. Grob, Z. Lebensm. Unters. Forsch. 205 (1997) 239-241.
[3] M. Biedermann, Y. Uematsu, K. Grob, Packag. Tech. Sci. 24 (2011) 61-73.
[4] M. Biedermann, K. Grob, Eur. Food Res. Technol. 230 (2010) 785-796.
[5] A. Vollmer, M. Biedermann, F. Grundböck, J.E. Ingenhoff, S. Biedermann-Brem, W. Altkofer, K. Grob, Eur. Food Res. Technol. 232 (2011) 175-182.
[6] M. Biedermann, K. Grob, J. Chromatogr. A 1255 (2012) 76-99.
[7] German Federal Institute for Risk Assessment (BfR). Stellungnahme Nr. 008/2010 des BfR vom 03, December, 2009.
[8] Joint FAO/WHO Expert Committee of Food Additives (JECFA) 2002, 59th report, 11-20; WHO Technical report Series 913, http://whqlibdoc.who.int/trs/WHO_TRS_913.pdf.
[9] European Food Safety Authority (EFSA), Scientific opinion on Mineral Oil Hydrocarbons in Food, EFSA Journal 10(6) (2012) 2704 1-185.
[10] Summary and conclusion of the 76th Meeting of the Joint FAO/WHO Expert Committee on Food Additives, 29 June 2012.
[11] A. Guinda, A. Lanzòn, T. Albi, J. Agric. Food Chem. 44 (1996) 1723-1726.
[12] O. Koprivnjak, G. Procida, L. Favretto, Food Technolog. Biotechnolog. 95 (1997) 125-131.
[13] Y.A. Tan, and A. Kuntom. J.AOAC Int. 76 (1993) 371-376.
[14] A.S. McGill, C.F. Moffat, P.R. Mackie, P. Cruickshank, J. Sci. FoodAgric. 61 (1993) 357-363
[15] C. Wagner, H.-P. Neukom, K. Grob, S. Moret, T. Populin, L.S. Conte, Mitt. Lebensm. Hyg. 92 (2001) 499-514.
[16] K. Grob, Workshop EU-DG-SANCO and the KLZH, Switzerland (2008).
[17] D. Fiorini, A. Paciaroni, F. Gigli, R. Ballini, Food Control, 21 (2010) 1155-1160.
[18] S. Moret, L. Barp, K. Grob, L.S. Conte, Food Chem. 129 (2011) 1898-1903.
[19] K. Grob, A. Artho, M. Biedermann, J. Egli, Food Addit. Contam. 8 (1991) 437-446.
[20] M. Biedermann, K. Fiselier, K. Gob, J. Agric. Food Chem. 57 (2009) 8711-8721.
[21] P.Q. Tranchida, M. Zoccali, G. Purcaro, S. Moret, L. Conte, M. Beccaria, P. Dugo, L. Mondello, J. Chromatogr. A, 2011, 1218, 7476-7480.
[22] G. Purcaro, S. Moret, L.S. Conte, J. Chromatogr. A, 1255 (2012) 100-111.
[23] M. Biedermann, K. Grob, J. Chromatogr. A 1255 (2012) 56-75.
[24] S. Moret, L. Barp, G. Purcaro, L.S. Conte, J. Chromatogr. A, 1243 (2012) 1-7.
[25] http://www.bfr.bund.de/cm/349/determination-of-hydrocarbons-from-mineral-oil-or- plastics.pdf, BfR (2012).
[26] M. Biedermann, K. Grob, J. Sep. Sci. 32 (2009) 3726-3737.
[27] L. Mondello, M. Zoccali, G. Purcaro, F.A. Franchina, D. Sciarrone, S. Moret, L. Conte, P.Q. Tranchida, J. Chromatogr. A, 1259 (2012) 221-226.

Multi-Residue Analysis of 210 Pesticides in Food Samples by Triple Quadrupole UHPLC-MS/MS

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Abstract

Pesticides and their metabolites are of great concern to society as they are harmful to human health, pollute natural resources and disturb the equilibrium of the ecosystem. Consequently, stricter food safety regulations are being enforced around the world, placing pesticide analysis laboratories under increasing pressure to expand the list of targeted pesticides, detect analytes at lower levels and with greater precision, reduce analysis turnaround times, and all the while maintaining or reducing costs. In this study a method was successfully developed for the quantitation of 210 commonly analysed pesticides in food samples using the Nexera UHPLC and LCMS-8040. Initial validation was performed to demonstrate instrument capabilities. Limits of detection (LOD) for 90 % of compounds were less than 0.001 mg kg-1 (1 ppb) and all compounds were less than 0.01 mg kg^{-1} (10 ppb) for both the quantifying and qualifying transitions using only a 2 µL injection. Repeatability at the 0.01 mg kg^{-1} reporting level was typically less than 5 %RSD for compounds and correlation coefficients were typically greater than 0.997 in a variety of studied food extracts. Consequently, the LCMS-8040 is ideally suited for routine monitoring of pesticides below the 0.01 mg kg^{-1} default level set by EU and Japanese legislation.

Keywords : Pesticides; Multi-residue analysis; LCMS-8040; Food safety; Fruit; Vegetables

1. Introduction

Pesticide residues in food continue to be the target of studies due to the uncertainty concerning adverse effects that those residues may have on human health after a lengthy exposure at low levels. More than 1000 active ingredients have been utilised and are formulated in thousands of different commercial products. They include a variety of compounds, mainly insecticides, herbicides and fungicides, as well as their metabolites, with very different physico-chemical characteristics and large differences in polarity, volatility and persistence.[1] Consequently, in order to ensure food safety for consumers and to facilitate international trade, regulatory bodies around the world have established maximum residue levels (MRLs) for pesticide residues in food commodities; that is, the maximum amount of pesticide residue and its toxic metabolites that might be expected on a commodity if good agricultural practice was adhered to during the use of the pesticide.[2]

In the European Union regulation 396/2005/EC was implemented in 2008 harmonising pesticide MRLS in all member states for 435 pesticide active substances in 378 commodities.[3] This EU regulation covers pesticides both currently and formerly used in agriculture in or outside the EU. For pesticide and food commodity combinations not listed in the regulation a default MRL of 0.01 mg kg^{-1} applies (Art 18(1b) of European Union Regulation No 396/2005).[3] In general, MRLs in the European Food regulation are in the range 0.01 - 10 mg kg^{-1} depending on the pesticide-commodity combination, with the lowest levels set for banned pesticides. For vegetables, fruits and cereals intended for the production of baby foods, Directive 2006/141/EC requires that baby food contains no detectable levels of pesticide residues defined as < 0.01 mg kg^{-1} and prohibits the use of certain very toxic pesticides in the production of infant foods and establishes even lower MRLs for a few other very toxic pesticides.[4] Regulatory bodies around the world, as in the EU, have produced similar guidelines. In the US, tolerances for more than 450 pesticides and other ingredients are stated in the electronic Code of Federal Regulations (US Environmental Protection Agency Office of Pesticide Programs) and are enforced by the US FDA.[5] Japan's positive list system for agricultural chemical residues in foods, introduced in 2006, contains MRLs for over 400 pesticides in various commodities.[6] China published national standard GB 2763-2005 in 2005 and more recently GB 28260-2011 which was introduced in 2012 and specifies 181 MRLS for 85 pesticides in food.[7,8]

Consequently, pesticide analysis laboratories are under increasing pressure to expand the list of targeted pesticides, detect analytes at lower levels and with greater precision, reduce analysis turnaround times and reduce usage of hazardous solvents while maintaining or reducing costs. Pesticide residues were traditionally analysed mainly by GC-based multi-residue methods often with MS detection. However, many modern (semi)polar compounds and/or ionic compounds could not

be analysed in this way due to poor thermal stability or volatility without the need for derivatisation.[9] Recent improvements in liquid chromatography - tandem mass spectrometry, combined with the discussed pitfalls of GCMS, have meant LCMSMS has become a vital technique. LC-triple quadruple mass spectrometry enables highly selective and sensitive analysis and is well suited to the multi-class analysis of large numbers of pesticides at trace levels.

In this work, we discuss the development of a multi-residue pesticide method for 210 pesticides using the Nexera UHPLC and LCMS-8040 triple quadruple. Pesticides were matrix-matched in food matrix (lettuce, pear and dried fruit) following QuEChERS sample preparation. The method was evaluated in matrix to ensure that the necessary reporting limits were obtained according to the various regulatory guidelines around the world with acceptable precision, in addition to ensuring chromatographic resolution of pesticide isomers with identical SRM transitions.

2. Experimental

A stock of pesticides was obtained from the Food and Environment Agency, UK, at a concentration of 0.01 mg kg^{-1} (for each pesticide) in acetone:acetonitrile 1:1. Linearity was investigated over a nine-point calibration with samples ranging from 0.005〜0.2mgkg-1 (5〜200ppb) analysed in duplicate; calibration samples were injected once in increasing order and once in decreasing order. Linearity was assessed with four calibration curves prepared by serial dilution of: (1) acetonitrile, (2) dried fruit extract, (3) lettuce extract and, (4) pear extract. Instrumental area repeatability was determined by replicate (n=6) injection of pear matrix at 0.01 mg kg^{-1}. LC-MS mobile phase solvents and additives were all of LC–MS quality and purchased from Sigma–Aldrich.
Food extracts were supplied by the Food and Environment Agency, UK, following established QuEChERS protocols. QuEChERS is acronym for Quick Easy Cheap Effective Rugged Safe and is a widely used sample preparation technique for the extraction of pesticides from food. Food samples included dried fruit, lettuce and pear, with the final extracts prepared in 100% acetonitrile.

LC Parameters

UHPLC:	Nexera UHPLC system		
Column:	Shim-pack XR-ODS III (150 x 2 mm, 2.2 µm particle size)		
Column temp.:	40 °C		
Mobile phase:	A = Water with 5 mM ammonium formate and 0.01 % formic acid B = Methanol with 5 mM ammonium formate and 0.01 % formic acid		
Gradient:	Time (min)	A%	B%
	0	95	5
	16	0	100
	18	0	100
	18.1	95	5
	20	95	5
Flow rate:	0.4 mL min^{-1}		
Injection volume:	32 µL (stacked injection: 2 µL sample + 30µL water)		
Needle wash:	1000 µL Methanol		

MS Parameters

MS:	LCMS-8040 triple quadrupole mass spectrometer
Ionisation:	ESI-Positive and negative (15 msec. polarity switch)
SRM:	Dwell time 5 msec. Pause time 1 msec.
Desolvation line:	250 °C
Heating block:	400 °C
Drying gas:	15 L min^{-1}
Nebulising gas:	2 L min^{-1}
SRM optimisation:	1:1 water:methanol with 10 mM ammonium acetate Flow rate: 0.5mL min^{-1} Flow injection analysis (No column fitted) 0.2 µL (0.01 mg kg^{-1} pesticide standard solution)
Mobile phase screening:	Carrier 1:1 water:methanol Flow rate: 0.3mL min^{-1} Flow injection analysis (No column fitted) 5 µL injection (0.01 mg kg^{-1} pesticide standard solution) 1 µL air gap (see text for mobile phase compositions)

Pesticide limits of detection were calculated based on the method described by the US-EPA in Title 40 Code of Federal Regulation Part 136,[10] using a standard deviation of 7 replicates in pear matrix at a concentration value that corresponds to an instrument signal to noise ratio in the range of 2.5 to 5 and a Student's t 99% confidence interval:

$$MDL = St(n-1,1-\alpha=0.99) \quad x\,s.d.$$

Where, t(n-1,1-α=0.99) = Student's t value for the 99% confidence level with n-1 degrees of freedom (t = 3.14 for 7 replicates), n = number of replicates, and s.d. = standard deviation of the replicate analyses.

3. Results and discussion

3.1 SRM optimisation

Target precursor and product ions were selected based on recommendations from the Food and Environment Agency, UK, and data from the EURL DataPool.[11] Typically the protonated or deprotonated molecule was used for the precursor ion. In order to try to prevent interference of SRM transitions from matrix, product ions greater than m/z 100 were selected where possible as they are typically more diagnostic.[12] Analyte specific MS parameters (Q1 pre-bias (V), Q3 pre-bias (V) and collision energy) were optimised using automated flow injection analysis. Briefly, this involves placing pesticide standards into the auto-sampler, from where they are then rapidly injected into the MS with a different parameter optimised on each injection. Each compound was optimised in only a few minutes using the automated software provided in LabSolutions. This allowed large numbers of compounds to be optimised overnight; this is in stark contrast to traditional time-consuming infusion in order to optimise parameters. The compounds studied and their associated transitions are shown in Table-1.

Table 1 Studied compounds and their chemical formulas, CAS numbers, SRMs, retention times, limits of detection and R^2
(You can find the whole table linked to the shortened one below.)

Compound	Formula	CAS	Transition 1	Transition 2	Pear extract				
					RT (min.)	Transition 1 LOD (ppb)	Transition 2 LOD (ppb)	%RSD (10ppb)	R^2
Avermectin B1a	C48H72O14	71751-41-2	891 > 305	891 > 567	16.4	0.35	0.56	5.0	0.9975
Acephate	C4H10NO3PS	30560-19-1	184 > 143	184 > 49	3.0	0.17	0.31	1.0	0.9999
Acetamiprid	C10H11ClN4	135410-20-7	223 > 126	223 > 99	7.2	0.50	1.00	1.1	0.9979
Acrinathrin	C26H21F6NO5	101007-06-1	559 > 208	559 > 181	16.1	1.32	2.36	4.4	0.9990
Alachlor	C14H20ClNO2	15972-60-8	270 > 238	270 > 162	13.4	0.09	0.26	1.5	0.9995
Aldicarb	C7H14N2O2S	116-06-3	208 > 116	208 > 89	8.5	0.05	0.10	1.7	0.9998
Aldicarb sulfone	C7H14N2O4S	1646-88-4	240 > 223	240 > 86	4.3	0.17	0.13	1.8	0.9999
Aldicarb sulfoxide	C7H14N2O3S	1646-87-3	207 > 89	207 > 132	3.9	0.22	0.36	2.3	1.0000
Amidosulfuron	C9H15N5O7S2	120923-37-7	370 > 261	370 > 139	9.3	0.14	0.22	2.8	0.9984
Asulam	C8H10N2O4S	3337-71-1	231 > 156	231 > 92	3.4	0.72	2.03	3.8	0.9979
Atrazine	C8H14ClN5	1912-24-9	216 > 174	216 > 104	11.1	0.10	0.22	2.4	0.9989
Azinphosmethyl	C10H12N3O3PS2	86-50-0	318 > 132	318 > 77	11.8	0.50	0.50	2.7	0.9903
Azoxystrobin	C22H17N3O5	131860-33-8	404 > 372	404 > 344	12.1	0.03	0.30	2.1	0.9989
Bendiocarb	C11H13NO4	22781-23-3	224 > 109	224 > 167	9.8	0.10	0.09	1.5	0.9996
Benthiavalicarb-isopropyl	C18H24FN3O3S	177406-68-7	382 > 180	382 > 116	12.7	0.12	0.41	0.9	0.9997
Bispyribac sodium	C19H17N4NaO8	125401-92-5	453 > 297	453 > 179	12.1	1.41	5.43	7.4	0.9954
Boscalid	C18H12Cl2N2O	188425-85-6	343 > 307	343 > 140	12.5	0.81	1.19	4.6	0.9968
Bromoxynil*	C7H3Br2NO	1689-84-5	274 > 79	276 > 81	9.9	2.24	2.61	4.5	0.9968
Bromuconazole	C13H12BrCl2N3O	116255-48-2	376 > 159	376 > 70	13.0	0.72	1.79	2.9	0.9994
Butachlor	C17H26ClNO2	23184-66-9	312 > 238	312 > 57	15.3	0.29	0.39	1.6	0.9998
Butocarboxim	C7H14N2O2 S	34681-10-2	208 > 75	208 > 191	8.4	0.13	0.87	3.1	0.9999
Butocarboxim sulfone	C7H14N2O4S	34681-23-7	223 > 106	223 > 166	4.1	2.63	3.23	9.7	0.9949
Butocarboxim sulfoxide	C7H14N2O3S	34681-24-8	207 > 88	207 > 75	3.7	0.22	0.21	1.9	0.9999
Carbaryl	C12H11NO2	63-25-2	202 > 145	202 > 127	10.3	0.13	0.22	2.4	0.9988
Carbendazim	C9H9N3O2	10605-21-7	192 > 160	192 > 132	7.1	0.50	1.00	1.1	0.9996
Carbofuran	C12H15NO3	1563-66-2	222 > 165	222 > 123	11.1	0.12	0.18	0.7	0.9993
Carboxin	C12H13NO2S	5234-68-4	236 > 143	236 > 87	10.2	0.09	0.25	0.9	0.9991
Chlorantraniliprole*	C18H14BrCl2N5O2	500008-45-7	482 > 284	482 > 177	11.8	0.50	1.00	2.3	0.9979
Chlorfenvinfos	C12H14Cl3O4P	470-90-6	361 > 155	361 > 99	14.0	0.28	0.49	2.3	0.9966
Chloridazon	C10H8ClN3O	1698-60-8	222 > 92	222 > 104	7.2	0.20	0.18	3.2	0.9990
Chlorotoluron	C10H13ClN2O	15545-48-9	213 > 72	213 > 46	10.8	0.05	0.13	1.3	0.9967
Chromafenozide	C24H30N2O3	143807-66-3	395 > 175	395 > 91	13.0	0.05	0.60	1.0	0.9977
Clethodim	C17H26ClNO3S	99129-21-2	360 > 164	360 > 268	14.7	0.08	0.45	0.7	0.9970
Clofentezine	C14H8Cl2N4	74115-24-5	303 > 138	303 > 102	14.4	4.03	5.76	9.5	0.9967
Clothianidin	C6H8ClN5O2S	210880-92-5	250 > 132	250 > 169	6.5	0.25	0.12	1.6	0.9978
Tricyclazole	C9H7N3S	41814-78-2	190 > 136	190 > 163	8.3	0.10	0.20	2.3	0.9993
Trifloxystrobin	C20H19F3N2O4	141517-21-7	409 > 186	409 > 145	14.6	0.02	0.05	1.2	0.9994
Triflumizole	C15H15ClF3N3O	68694-11-1	346 > 278	346 > 43	14.8	0.09	0.09	1.3	0.9996
Triflumuron*	C15H10ClF3N2O3	64628-44-0	357 > 154	357 > 176	14.2	1.76	3.12	4.6	0.9991
Triforine	C10H14Cl6N4O2	26644-46-2	435 > 390	437 > 392	11.7	0.92	3.53	4.8	0.9963
Triticonazole	C17H20ClN3O	131983-72-7	318 > 70	320 > 70	13.2	0.40	0.41	1.9	0.9993
Zoxamide	C14H16Cl3NO2	156052-68-5	336 > 187	336 > 159	14.0	0.09	0.29	1.3	0.9951
2,4-D*	C8H6Cl2O3	94-75-7	219 > 161	219 > 125	10.3	1.09	5.00	9.7	0.9980

* Negative electrospray ionisation

3.2 Rapid screening of different mobile phase compositions on signal response

The signal intensity in LCMS can be strongly influenced by the mobile phase composition. In order to optimise the signal intensity, pesticides were added into vials containing different mobile phase compositions and injected into the interface with no column installed. The Nexera auto-sampler was setup to inject an air gap both before and after the injected sample in order to prevent the sample mixing with carrier mobile phase. This approach enables a large number of potential mobile phase compositions to be screened in a short automated period of time and without the need to manually change mobile phases. Ten different mobile phase compositions were tested, including: ammonium acetate, ammonium formate, formic acid, acetic acid, and ammonium formate with formic acid in water:methanol or acetonitrile 1:1. A total of 23 different pesticides were assessed, selected to include a range of different polarities and both positively and negatively ionised compounds. The different mobile phases tested and their peak area response, relative to the highest peak area response obtained for that compound, are shown in Table 2.

As expected with multi residue methods, there was not one optimum mobile phase for all pesticides. Overall, the lowest signal was achieved for mobile phases containing water:methanol only, and the mobile phase containing water:acetonitrile 10 mM ammonium acetate. Negatively ionised compounds (fludioxinil and ioxynil) provided superior responses in water:methanol 10mM ammonium acetate, while the addition of either formic acid or acetic acid decreased response. The highest signals were typically found in 10 mM ammonium formate, 10mM ammonium acetate, and 10 mM ammonium formate with 0.1 % formic acid. The effect of methanol and acetonitrile in the mobile phase was also investigated. Comparison of 10mM ammonium formate in methanol and acetonitrile showed that intensities were typically lower with the use of acetonitrile. Similarly the use of ammonium acetate in methanol and acetonitrile presented the same trend. The same observation with regards to methanol and acetonitrile for pesticide analysis have been reported by others.[13]

Table 2 Results of rapid mobile phase screening using flow injection analysis for 23 pesticides. All peaks areas were normalised against the maximum peak area achieved for that compound. Accordingly, 100 % indicates the highest peak area achieved and is highlighted.

Compound	H2O:MeOH	H2O:MeOH 0.05% Formic acid	H2O:MeOH 0.1% Formic acid	H2O:MeOH 0.2% Formic acid	H2O:MeOH 5mM Ammonium acetate	H2O:MeOH 10mM Ammonium acetate	H2O:MeOH 20mM Ammonium acetate	H2O:MeOH 50mM Ammonium acetate	H2O:MeCN 10mM Ammonium acetate	H2O:MeOH 10mM Ammonium formate	H2O:MeCN 10mM Ammonium formate	H2O:MeOH 0.1% Acetic acid	H2O:MeOH 0.1% Formic acid 50mM ammonium formate
Atrazine	52	**100**	99	88	52	71	66	62	48	80	50	52	87
Azinphos-methyl	14	32	32	27	75	98	87	59	26	**100**	26	30	96
Azoxystrobin	27	30	29	25	69	87	77	58	65	**100**	82	29	99
Carbendazim	66	**100**	91	92	37	42	38	32	26	71	36	64	81
Chlorantraniliprole	**100**	46	52	41	69	81	92	69	27	91	60	94	56
Cyprodinil	66	94	88	86	55	63	57	41	51	**100**	82	67	78
Difenoconazole	27	85	90	72	70	**100**	92	73	59	99	62	61	90
Fludioxinil	69	42	38	37	74	**100**	95	84	60	94	81	55	76
Imazalil	85	69	62	63	66	78	73	62	51	**100**	68	58	74
Ioxynil	**100**	47	41	43	41	60	60	51	34	62	53	55	53
Isoproturon	28	34	34	30	74	93	84	75	78	**100**	90	30	98
Metalaxyl	30	31	31	25	68	92	81	76	79	**100**	87	31	92
Myclobutanil	15	71	75	57	65	**100**	91	73	23	86	25	58	84
Pirimicarb	82	85	76	78	66	90	80	68	68	**100**	80	66	78
Pirimicarb-desmethyl	72	90	81	83	64	85	74	67	64	**100**	82	70	86
Prochloraz	38	**100**	94	89	47	65	56	45	45	61	46	64	64
Pyraclostrobin	33	32	30	27	62	78	70	55	61	**100**	82	26	93
Pyrimethanil	54	**100**	92	91	54	65	54	31	48	92	74	62	76
Tebufenozide	28	40	40	36	70	88	78	65	73	96	84	33	**100**
Thiabendazole	96	**100**	91	89	58	69	61	48	37	99	60	67	84
Thiacloprid	16	28	28	25	53	59	45	32	34	86	49	18	**100**
Thiophanate methyl	24	21	24	17	62	77	62	44	34	98	43	31	**100**
Triadimenol	17	96	**100**	81	56	88	86	74	44	79	46	66	74
Minimum	14	21	24	17	37	42	38	31	23	61	25	18	53
Maximum	100	100	100	92	75	100	95	84	79	100	90	94	100
Average	**50**	**64**	**62**	**57**	**61**	**80**	**72**	**58**	**49**	**91**	**63**	**52**	**83**

3.3 Performance Optimising Injection Sequence (POISe)

In reversed phase UHPLC, early eluting compounds typically display the greatest peak distortion. Peak distortion is a particular problem is pesticide analysis as samples are typically extracted by QuEChERS, with samples diluted in 100% acetonitrile (a strong eluting solvent). To solve this issue, laboratories may decide to dilute the acetonitrile extracts in water before LCMS injection. However, doing so adds an additional sample preparation step and dilution in water can also negatively affect the stability of some analytes.[14]

To minimise peak dispersion with the injection of acetonitrile extracts, one potential solution is the use of a band compression technique.[15] Band compression is achieved by injecting a band of weak eluting solvent onto the column after the analytes. As the analyte and the weak eluting solvent bands travel towards the column, minute mixing occurs. Therefore, the analytes are dissolved in a weak eluting solvent when they reach the column leading to isocratic band compression.

The performance optimising injection sequence (POISe) was evaluated by injecting between 5 – 40 µL of water following a 3 µL injection of pear extract in 100% acetonitrile. This was achieved using the Nexera auto-sampler (SIL-30AC) pre-treatment program to perform this function.

Fig.1 shows the injection of pear extract *with* and *without* the performance optimising injection sequence. Using POISe, band dispersion was minimised considerably for early eluting pesticides, with peak widths reduced by 5-69%. The optimum amount of water to inject following the sample was found to be 30 µL. Increasing this volume to 40 µL did not provide any significant improvements. Early eluting compounds are affected by the injection of a weak eluting solvent band to a much larger extent in comparison to analytes with higher retention factors. This improvement is due to the reduction in the sample solvent elution strength, which has a large impact on the early eluting compounds. Whereas, analytes with higher retention factors will experience some degree of band compression in the mobile phase already. Table 3 lists the peak width for 11 early eluting compounds. Compounds are arranged in retention time order to show the improvement using the POISe on early eluting analytes.

(A) 3 µL pear extract injection without the POISe

(B) 3 µL pear extract injection with the POISe (30 µL water)

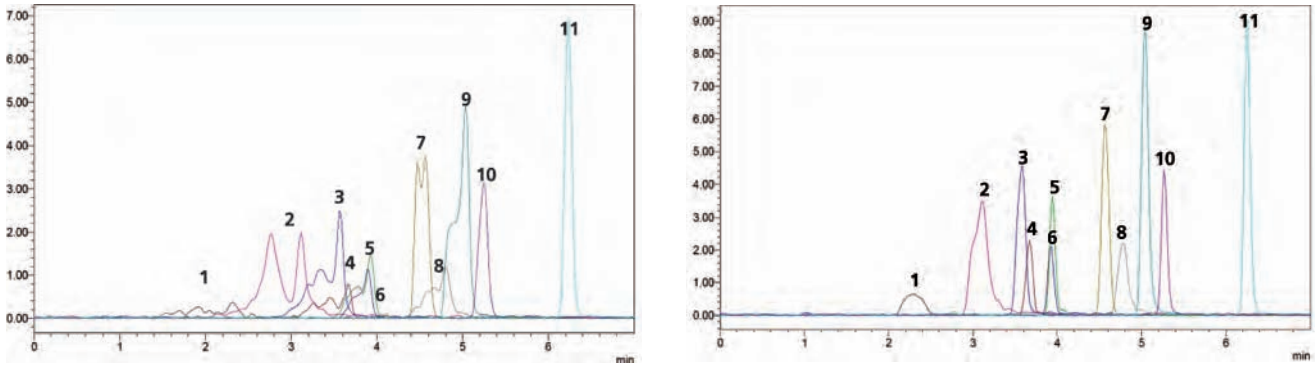


Fig.1 Pear extract (0.050 mg kg^{-1}) injected without (A) and with (B) the performance optimising injection sequence

Table 3 Peak widths obtained with and without the performance optimising injection sequence

No.	Compound	Peak width (min.)		Peak width change (%)
		Without POISe	With POISe	
1	Methamidophos	1.193	0.466	**-60.9**
2	Propamocarb	0.937	0.473	**-49.5**
3	Omethoate	0.773	0.247	**-68.0**
4	Butocarboxim sulfoxide	0.664	0.205	**-69.1**
5	Aldicarb sulfoxide	0.545	0.195	**-64.2**
6	Dinotefuran	0.460	0.247	**-46.3**
7	Oxamyl	0.317	0.248	**-21.8**
8	DMPF	0.309	0.254	**-17.8**
9	Demeton-S-methyl sulfoxide	0.418	0.271	**-35.2**
10	Demeton-S-methyl sulphone	0.277	0.248	**-10.5**
11	Ethiofencarb sulphone	0.233	0.220	**-5.6**

3.4 UHPLC gradient optimisation

Based on the results of the mobile phase screening investigation (section 3.2) the three superior compositions were tested: 1) 10 mM ammonium formate, 2) 10 mM ammonium acetate and 3) 10 mM ammonium formate with 0.1 % formic acid. Separation was achieved using a Shim-Pack XR-ODS III, 2.0 x 150 mm, 2.2 µm particle size. Ammonium formate was found to be the most effective compromise for all 210 compounds in terms of signal to noise ratios and peak shapes.

However two problems with ammonium formate were observed; early elution of asulum and poor peak shape of propamocarb. Consequently, 0.01 % formic acid was tested and found to increase the retention of asulum, and improve the peak shape of propamocarb. The addition of acid was found to shorten the retention time of cyromazine (RT 2.2 min.), yet this retention time was still in excess of 2 column volumes as required in quality control procedures for pesticide residues analysis in food and feed.[13]

A number of pesticide isomers have identical transitions and consequently must be separated chromatographically. Employing a 16 minute gradient resulted in resolution greater than 1 between all necessary pesticides including: butocarboxim sulphoxide / aldicarb sulphoxide, ethiofencarb sulphone / methiocarb sulphone, diuron / fluometronsulam and desmedipham / phenmedipham. Figure 2 highlights the excellent peak shapes achieved on the Nexera UHPLC.

3.5 Final method performance

In order to assess the performance of the LCMS-8040 for real samples, limits of detection, linearity and repeatability were determined in food extracts. Linearity was assessed from 0.5 – 200 ppb in four types of sample: (1) acetonitrile, (2) dried fruit extract, (3) lettuce extract and, (4) pear extract. All 210 pesticides achieved excellent correlation coefficients greater than 0.99 in all four types of matrix with typical values greater than 0.997. Correlation coefficients are listed in Table 1 for all pesticides in pear extract, and the calibration curves of eight selected pesticides shown in Fig. 3.

Pesticide limits of detection were calculated based on the method described by the US-EPA (see experimental section). Limits of detection were assessed for both the quantifying transition and the qualifying transition and are listed in Table 1. All of the studied pesticides presented LODs less than the 0.01 mg kg^{-1} reporting level for both transition 1 and 2.

A limit of detection less than 0.001 mg kg^{-1} (1ppb) was achieved for the quantifying transition and less than 0.002 mg kg^{-1} (2 ppb) for the qualifying transition for 90 % of compounds: thereby highlighting the excellent sensitivity of the LCMS-8040 for pesticide analysis. Furthermore, these limits of detection were achieved with an injection volume of only 2 µL. Therefore, detection limits could be reduced even further with larger injection volumes. An injection volume of 2 µL was used in the study to allow the injection of 100 % acetonitrile extracts without detriment to early eluting peak shapes.

Repeatability was assessed at the 0.01 mg kg^{-1} reporting level as peak area %RSD for six replicate injections in pear extracts. Repeatability less than 5 %RSD was achieved for 92 % of the 210 pesticides studied. All of the studied compounds presented repeatability less than 10 %RSD, with exception of haloxyfop acid (13.4 %).

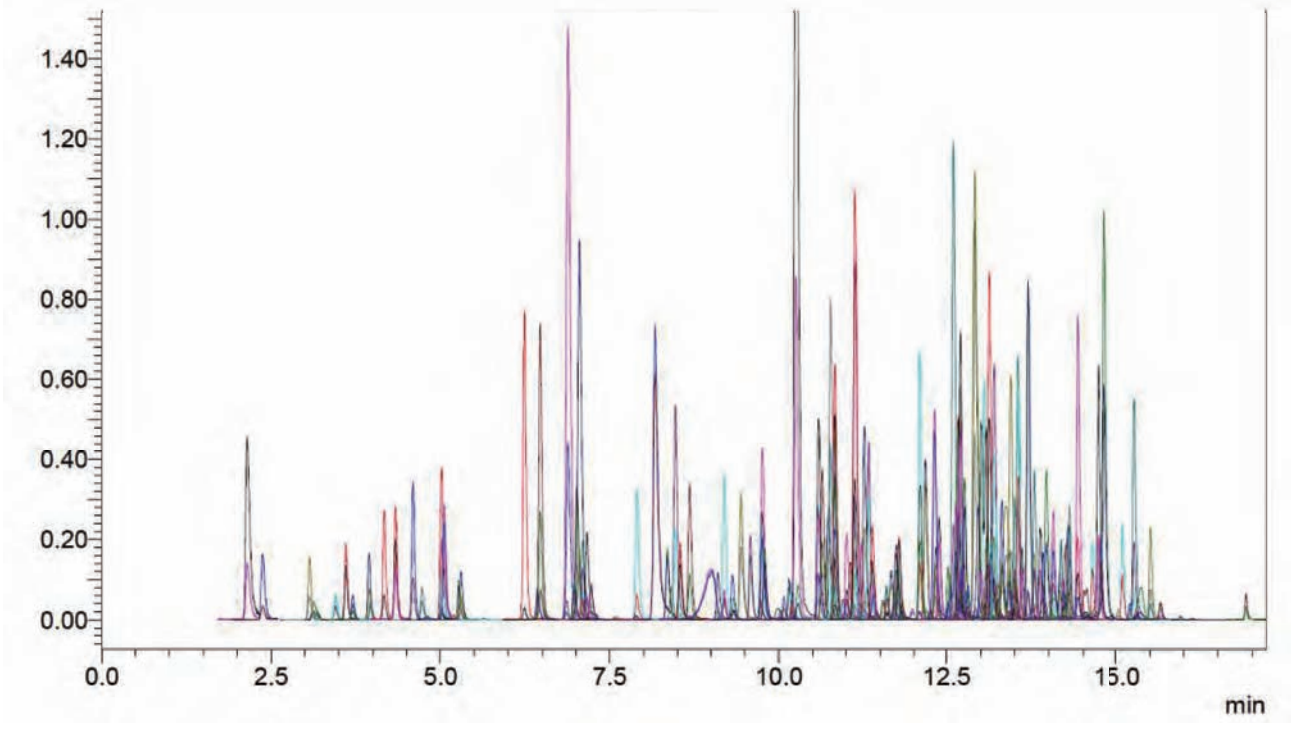


Fig. 2 Extracted ion chromatogram of 210 pesticides using the Shimadzu Nexera UHPLC and the Shimadzu LCMS-8040; 2 µL injection of a 0.05 mg kg-1 standard solution.

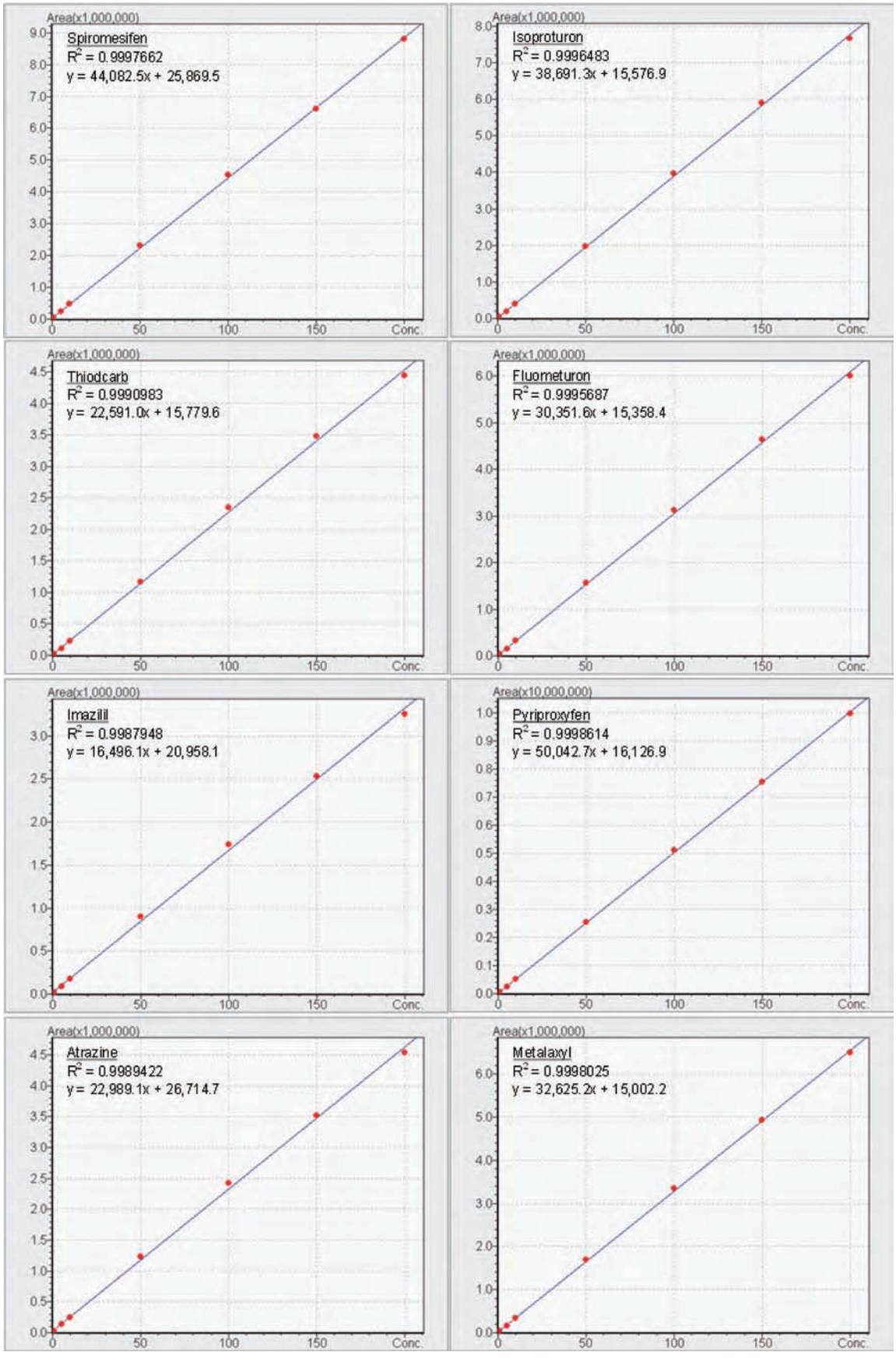


Fig. 3 Calibration curves, 0.005～0.2mgkg-1 (5～200ppb), of eight pesticides in pear matrix

4. Conclusion

The results of the developed methodology show that the Shimadzu LCMS-8040 triple quadrupole can achieve excellent sensitivity, linearity and repeatability in food extracts for over 200 commonly analysed pesticides. Limits of detection were less than 0.01 mg kg^{-1} (10 ppb) for both the quantifying and qualifying transitions for all compounds studied, while for 90% of compounds was less than 0.001 mg kg^{-1} (1ppb) (quantifying transition) and 0.002 mg kg^{-1} (2 ppb) (qualifying transition); therefore providing excellent response, especially given that the injection volume was only 2 µL. The sensitivity of the LCMS-8040 was able to meet the 0.01 mg kg^{-1} (10 ppb) requirements of regulatory guidelines such as those established by the EU and Japan. Repeatability at the 0.01 mg kg^{-1} reporting level was less than 5% for nearly all compounds and correlation coefficients greater than 0.99 for all compounds in a variety of food samples. Consequently the LCMS-8040 is ideally suited for routine monitoring of pesticides in regulatory laboratories.

Acknowledgements

The authors wish to thank the staff at the Food and Environment Agency, UK, for providing food sample extracts and pesticide reference standards.

5. References

1. H. V. Botitsi, S. D. Garbis, A. Economou and D. F. Tsipi, Mass Spectrometry Reviews 2011, 30, 907-939.
2. C. Solera, J. Mañesa and Y. Picó, Critical Reviews in Analytical Chemistry 2008, 38, 93-117.
3. Commission Regulation (EC). 2005. No 396/2005 of the European Parliament and of the Council, maximum residue levels of pesticides in or on food and feed of plant and animal origin. Official Journal of the European Union, L 70: 1-16
4. Commission Directive 2006/141/EC of 22 December 2006 on infant formulae and follow-on formulae and amending Directive 1999/21/EC. Official Journal of the European Union L401: 1-33.
5. US Environmental Protection Agency, Electronic code of federal regulation: Title 40: Part 180 - tolerances and exemptions for pesticide chemical residues in food. **http://www.ecfr.gov/cgi-bin/text-idx?c=ecfr&tpl=/ecfrbrowse/Title40/40cfr180_main_02.tpl**
6. Japanese Ministry of Health, Labour and Welfare, Department of Food Safety. 2006. Director Notice about Analytical Methods for Residual Compositional Substances of Agricultural Chemicals, Feed Additives, and Veterinary Drugs in Food (Syoku-An No. 0124001 January 24, 2005; amendments May 26, 2006).
7. Republic of China National Standard GB 2763-2005. 2005. Maximum residue limits for pesticides in food, Ministry of Health of the People's Republic of China and Ministry of Agriculture of the People's Republic of China.
8. Republic of China National Standard GB 28260-2011. 2011. Maximum residue limits for 85 pesticides in food, Ministry of Health of the People's Republic of China and Ministry of Agriculture of the People's Republic of China
9. L. Alder, K. Greulich, G. Kempe and B. Vieth, Mass Spectrometry Reviews 2006, 25, 838-865
10. US Environmental Protection Agency, Procedure 40 CFR, Part 136, Appendix B.
11. EURL datapool, EU Reference Laboratories, **http://www.eurl-pesticides-datapool.eu**
12. European Commission, SANCO. 2011. Method validation and quality control procedures for pesticide residues analysis in food and feed, Document SANCO/12495/2011
13. C. Jansson, T. Pihlström, B.-G. Österdahl and K. E. Markides, Journal of Chromatography A 2004, 1023, 93-104.
14. K. Maštovská and S. J. Lehotay, Journal of Chromatography A 2004, 1040, 259-272
15. A. C. Sanchez, J. A. Anspach and T. Farkas, Journal of Chromatography A 2012, 1228, 338-348

Shimadzu Selection

These are the articles selected by Shimadzu for this issue. The articles are from application notes relating to Food safety and Environment with a variety of instruments we produce. The cutting edge technologies are also included. Please obtain the articles of your interest through the links on the titles.

Selection 1 Food Safety

Rapid and Highly Sensitive Quantitative Analysis and Screening of Aflatoxins in Foods Using Liquid Chromatography Triple Quadrupole Mass Spectrometry

Aflatoxins (AFs) are the most harmful mycotoxins produced by the fungi Aspergillus flavus and Aspergillus parasiticus and can contaminate foods such as cereals and nuts. To reduce the risk of the ingestion from foods, analyses of the AFs are carried out in many countries. In this study, we examined two alternative high-throughput LC-MS/MS methods.

Selection 2 Food safety

High Throughput LC-MS/MS Analysis of Carbendazim in Orange Juice

A new high throughput LC-MS/MS method was developed to facilitate increased testing for carbendazim in orange juice at low ppb levels. A gradient reversed phase high speed method was developed for the analysis of carbendazim on a 2.1×30 mm 2.6 micron core shell column coupled to a tandem quadrupole mass spectrometer. The use of UHPLC with a high speed injector allowed an analysis to be completed within a one minute timeframe.

Selection 3 Food Identification

DNA Analytical Technology to Identify the Actual Composition of Foods Application of the “MultiNA” Microchip Electrophoresis System

Recently, methods to identify the actual composition of various foods have been developed because of progress in gene-analysis technology. An electrophoretic pattern required for this type of analysis can be obtained by using MCE-202 MultiNA Microchip Electrophoresis System. Examples of analysis data obtained using MultiNA to identify species of meat, tuna, and rice are shown.

Selection 4 Food Safety

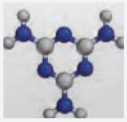
High-Speed Analysis of Amino Acids and Histamine in Fish Sauce via Automated OPA Pre-column Derivatization

Processed foods and fish containing a significant amount of histamine have resulted in several cases of allergic food poisoning. In Japan, no standard values have been set for histamine concentrations in foods. However, the FDA has specified levels of 50 mg/kg max. for foods in general, the EU has specified 100 mg/kg max. for marine products, and the Codex Alimentarius standard specifies 400 mg/kg max. for fish sauce.

Selection 5 Food Evaluation

Evaluation of Foods for People with Dysphagia

In recent years, there has been an increase in people with dysphagia due to an aging population, so the demand for these commodities has increased. This article introduces a system for measuring hardness, adhesion, and agglomeration, evaluation items based on the Consumer Affairs Agency of Japan's Food Labeling Notification No. 277 (Permission of Labeling for Foods for Special Dietary Uses).



Selection 6 Environment

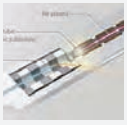
Analysis of Melamine and Its Related Substances in Fertilizers

Recently, it has been confirmed that some granulated products of hydrated calcium cyanamide, which are created by adding water to calcium cyanamide, contain a significant amount of melamine. This article introduces an example of the analysis of melamine and related substances in fertilizers using an HPLC system, with reference to the fertilizer test methods (2012) under the supervision of the Japan's Food and Agricultural Materials Inspection Center (FAMIC).

Selection 7 Environment

Development of Solid-Phase Extraction Method for Simultaneous Analysis of Semi-Volatile Organic Compounds Using a GC-MS Database System

Environmental pollution caused by a variety of chemicals is drawing attention to the need for a more efficient method of simultaneous multicomponent analysis. In this research, we investigated the use of solid-phase SVOC extraction, which requires the use of less solvent. Here, an analytical method developed for comprehensive analysis of SVOCs in water samples using AIQS-DB is reported.



Selection 8 New Technology

Development of New Ionization Detector for Gas Chromatography by Applying Dielectric Barrier Discharge

A new gas chromatographic detector, dielectric barrier discharge ionization detector (BID), has been developed by applying atmospheric non-equilibrium plasma to a photo ionization source. The relation between the type of dielectric barrier discharge and its stability, and the influence on the baseline stability by outgassing from the discharge cell were clarified.

Selection 9 New Technology

Development of Iodoacetyl-Based Ionic Mass Tags for Improved Sensitivity in the Detection of Cysteine-Containing Peptides by MALDI-TOF MS

We developed and characterized six new ionic mass tags that bond to cysteine residues and enable high-sensitivity analysis of peptides. We analyzed peptides for their MS sensitivity using the new mass tags and achieved improvements in sensitivity of approx. 2 to 200 times the sensitivity obtained with a carbamide-methylated peptide control sample.

Selection 10 New Technology

Development of HyperVision HPV-X High-Speed Video Camera

We developed the HPV-X high-speed video camera, which incorporates the FTCMOS high-speed sensor based on the technology of CMOS image sensor. It can capture images without decreasing the spatial resolution at high-speed capturing. This paper describes the HPV-X and applications of high-speed photography.

Selection 11 New Technology

Development of new fNIRS-EEG system for seamless whole brain study

Functional near-infrared spectroscopy (fNIRS) is a versatile functional neuroimaging technology with non-invasive method of monitoring of brain activity. fNIRS has high potentials for such research topics according to the high temporal/spatial resolution compare to fMRI or EEG. We developed a new fNIRS-EEG system which allows seamless whole-brain measurement of the surface of the human brain, high spatial resolution measurement, multi-distance measurement, and high temporal resolution measurement.

Shimadzu Solutions for Pesticide Analysis

Pesticides are used around the world to achieve stable crop production. They are used not only at the site of production but, in these days of extensive distribution, post-harvest pesticides are applied to stop crops spoiling during transport. If the usage and dosage are appropriate, modern pesticides leave little residue in crops and have little effect on humans or the environment. However, many consumers hold a negative image of pesticides due to repeated incidents and accidents related to them. As the types of chemicals and their actual status of use differ from country to country, there are never ending cases of residual pesticides in foods not meeting the regulations at the export destination due to the widespread distribution in the modern world. Against this background there is an extremely strong demand in all countries of the world for the analysis of pesticides in foods.

Chromatography is the major method used to analyze pesticides in foods. The actual instrument used differs according to the physicochemical properties of the target pesticide, the type of food, and the aim of the inspection. Pesticides can be broadly categorized by their properties as low-polarity pesticides that are analyzed by a gas chromatograph (GC) or gas chromatograph mass spectrometer (GC-MS(/MS)) or as high-polarity and heat-sensitive pesticides that are analyzed by a liquid chromatograph (LC) or liquid chromatograph mass spectrometer (LC-MS(/MS)).

Individual pesticides or pesticide groups can be analyzed using a normal LC or GC detector after pretreatment to adequately eliminate the effects of impurities. This acquires stable data with excellent reproducibility. Some detectors offer high selectivity and detection sensitivity with respect to the target pesticide. For example, organochlorine or pyrethroid pesticides are measured with a GC-ECD electron capture detector, organophosphate pesticides with a GC-FPD flame photometric detector, nitrogen-based pesticides with a GC-FTD flame thermionic detector, and N-methyl carbamate pesticides with an LC-RF fluorescence detector using post-column detection.

However, as these normal LC and GC detectors provide only retention times as qualitative information, there is a risk of incomplete separation of components, misidentification, and misquantitation during multicomponent simultaneous analysis or the analysis of samples with high levels of impurities. In these cases, use a mass spectrometer (MS, MS/MS). A mass spectrometer (MS, MS/MS) offers the benefit of high selectivity, as it acquires mass information in addition to retention time information. As a mass spectrometer presents lower risk of misidentification and misquantitation than a normal LC or GC detector, it is suitable for multicomponent simultaneous analysis and the analysis of samples with high levels of impurities. Due to the wide range of libraries available, GC-MS(/MS) is able to screen for unknown substances in addition to the target component, making it extremely advantageous for risk avoidance.

As shown above, the identification of residual pesticides is difficult in the highly developed distribution system of today's world. From the viewpoint of risk avoidance, it is necessary to perform the simultaneous analysis of as many pesticides as possible. Highly selective MS or MS/MS is often used for this purpose.

Shimadzu offers the chromatograph instruments required for residual pesticide analysis to meet all analysis requirements.

The GC-2010 Plus Series capillary GC instruments provide excellent reproducibility and can use an FPD or other high-sensitivity detector to achieve highly reliable and highly accurate analysis. The HPLC and UHPLC product ranges from Prominence to NexeraX2 provide superb reproducibility and their extremely low carryover permits accurate analysis of pesticide components that readily adhere to the instrument. The rapid performance of the mass spectrometer is ideal for multicomponent simultaneous analysis and fast analysis.

The single-quadrupole LC-MS (LCMS-2020) and triple-quadrupole LC-MS/MS (LCMS-8030, 8040, 8080) offer rapid positive/negative switching and high-speed scanning for fast and accurate simultaneous analysis of samples containing both positive and negative ions. To avoid the problems associated with GC-MS analyses, such as helium consumption and the time and effort required for pretreatment, components conventionally analyzed by GC-MS are increasingly analyzed by LC-MS recently. For simultaneous analysis of multiple components including those conventionally analyzed by GC-MS, the Shimadzu LC-MS(/MS) high-speed performance is indispensable*. The GCMS-QP2010 series single quadrupole GC-MS and the GCMS-TQ8030 triple quadrupole GC-MS/MS not only provide excellent high-speed performance, but also achieve improved sensitivity at the time of high-speed sampling using the patented ASSP technology that minimizes the reduction in sensitivity with high-speed scanning. Thanks to this excellent high-speed performance, it is possible to carry out quantitative analysis of trace components by SIM (MRM), and simultaneously carry out qualitative scan analysis, to obtain more reliable data. Also, by using the simultaneous analysis database with the scan data, semi-quantitative analysis of untargeted components is possible**. Screening for untargeted components this way enables rapid response to unexpected incidents, which is useful in terms of risk management.

Applications

General

C180-E059B Analysis Guidebook Food Product Analyses
C219-E007A Shimadzu Food Safety Management Data Book

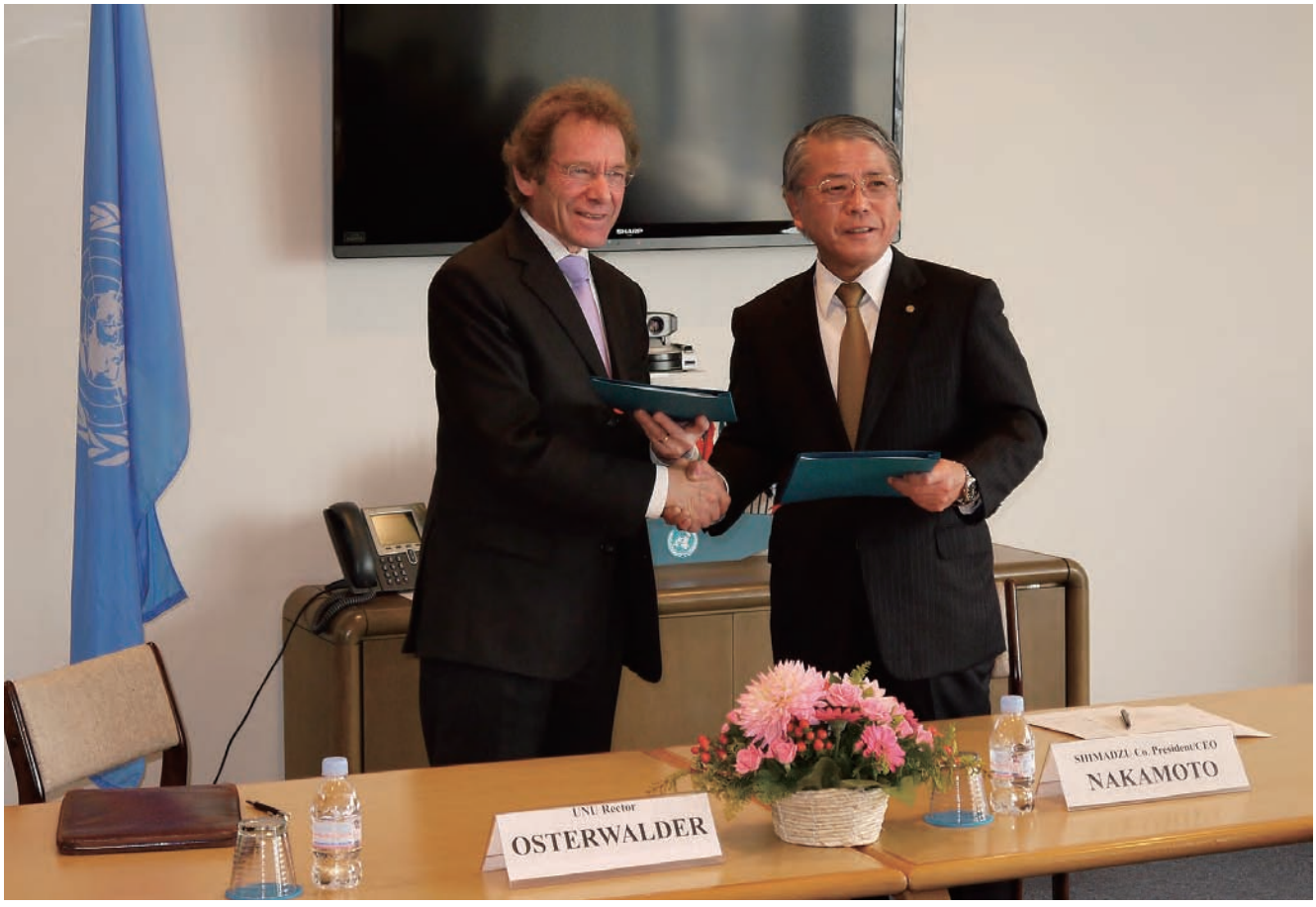
LC-MS/MS

ASMS2012 Poster
* WP27-575 Multi-class pesticides analysis in challenging vegetable matrices using fast 5 msec MRM with 15 msec polarity switching
WP27-574 Exploring the application of a universal method for pesticide screening in foods using a high data acquisition speed MS/MS

GC-MS/MS

Application Data Sheet
No.71 Simultaneous Analysis of Residual Pesticides in Foods via the QuEChERS Method Utilizing GC-MS/MS
**No.64, 65, 72 Scan/MRM Analysis of Residual Pesticides in Foods Using GC-MS/MS (1) (2) (3)

More than 100 application notes of pesticides analysis by LC, GC, LC-MS(/MS), and GC-MS(/MS) are available at:
http://search1.shimadzu.co.jp/search?site=EM0AJRZ6&design=2&group=1



Continued Support for Environmental Projects with the United Nations University

To Cultivate Engineers and Environmental Protection in Asia

Aiming for Environmental Conservation in the Asian Region

The "Environmental Monitoring and Analysis in the East Asian Region: Technology Transfer and Environmental Governance" project has been implemented by the United Nations University since 1996. This project involves the monitoring of chemical pollutants in the coastal environments of 10 Asian countries, for the purpose of creating a sustainable global environment. To date, investigative research focusing on volatile organic compounds (VOC), endocrine disrupting chemicals, and persistent organic pollutants (POP) has been implemented in five phases, with each phase consisting of three years. With the corporate philosophy of "Contributing to Society through Science and Technology," Shimadzu has provided comprehensive support for this project from its commencement in 1996, utilizing the expertise that we have cultivated over our long history. In addition to providing liquid chromatograph mass spectrometers and other instruments manufactured in-house, Shimadzu has supported the convening of international symposia, and trained researchers in analytical technologies. In addition to improvements to capabilities of monitoring in detail the environments in various countries, definitive results have been amassed over these 15 years including improvements to detailed data analysis techniques, the construction of a social network across the major research institutions in Asia, the development of new analysis methods, and the accumulation of data.

Loans of New Models in the Plans for the Next Term

The fall of 2012 saw the implementation of "Monitoring and Governance of Persistent Organic Pollutants (POPs) in Asia -- Monitoring of PFCs --," a 3-year plan for the 6th phase of the project. As Shimadzu had decided to continue its support, a signing ceremony for a support-related agreement for the 6th phase of the project was held on November 12, 2012 at the United Nations University headquarters (Aoyama, Tokyo). The agreement was signed by the two highest officials of the respective organizations: Akira Nakamoto, President of Shimadzu Corporation, and Dr. Konrad Osterwalder, Rector (at the time) of the United Nations University.
At the signing ceremony, President Nakamoto noted that this agreement can be expected to further strengthen the established network of researchers, thereby elevating analytical technologies to a higher level, and he promised even stronger support than conventionally provided. The objective of the 6th phase is to study and monitor the status of FPOS (perfluorooctane sulfonate) and POA (perfluorooctanoic acid) pollutants in the aquatic environment. In this phase, Shimadzu is providing the latest ultra fast liquid chromatograph mass spectrometers (LCMS) that are capable of monitoring compounds with larger molecular weights. In addition, Shimadzu will provide personnel training as per usual, capitalizing on our technical skills and expertise with respect to environmental analysis.
The results of this project at the United Nations University are expected to promulgate through the Asian countries and beyond to emerging nations.

First Phase (1996-1999)

Environmental Monitoring and Analysis in the East Asian Region -- Technology Transfer and Environmental Governance --

Research studies were conducted on monitoring substances, such as pesticides in foods and volatile organic compounds (VOCs) in drinking water and the atmosphere, in order to standardize the analytical technologies for monitoring the concentrations of chemical pollutants in drinking water, soil, food products, and air in the East Asian region.

Second Phase (1999-2002)

Environmental Monitoring and Governance in the East Asian Coastal Hydrosphere -- Endocrine Disruptors in River and Coastal Waters --

Studies and monitoring of endocrine disruptors (pesticides, bisphenol A, alkylphenol, phthalic acid) in river and coastal waters were conducted to determine the degree of environmental pollution and allow the analysis of pollutants in the East Asian Coastal Hydrosphere.

Third Phase (2002-2005)

Environmental Monitoring and Governance in the East Asian Hydrosphere -- POPs in the East Asian Coastal Hydrosphere --

Studies and monitoring of the pollution status in rivers and soil were conducted, and the analytical expertise and human networking in the participating research organizations were reinforced in order to restrict and prevent persistent organic pollutants (POPs) in the East Asian Hydrosphere.

Fourth Phase (2005-2008)

Environmental Monitoring and Governance in the Asian Coastal Hydrosphere -- Monitoring of Persistent Organic Pollutants (POPs) in the East Asian Hydrosphere --

Studies and monitoring of the pollution status in aquatic organisms such as crustaceans and fish were conducted, and the analytical expertise and human networking in the participating research organizations, including two additional countries, were reinforced in order to restrict and prevent persistent organic pollutants (POPs) in the East Asian Hydrosphere.

Fifth Phase (January 2009 to December 2011)

Environmental Monitoring and Governance in the Asian Coastal Hydrosphere -- Monitoring of PCBs and Other Persistent Organic Pollutants (POPs) in the Asian Region --

Studies and monitoring of the pollution status were conducted with respect to PCB in the aquatic environment and brominated flame retardants in the sediment, in order to restrict and prevent persistent organic pollutants (POPs) in the Asian environment. At the same time, analytical expertise and human networking in the participating research organizations were reinforced.

Agilent Technologies and Shimadzu to Exchange Chromatography Instrument Drivers

Standardization Will Reduce Costs, Enhance Interoperability, Broaden Customer Choice

Agilent Technologies Inc. and Shimadzu Corp. have announced they will exchange RapidControl. NET (RC.Net) instrument drivers. Shimadzu's adoption and support of Agilent's RC.Net driver standard strengthens RC.Net as an emerging open industry standard for instrument control across multiple data systems.

Through this exchange, Shimadzu LabSolutions and Agilent OpenLAB Chromatography Data Systems will control both manufacturers' instruments, providing customers more freedom of choice in instrumentation for their laboratories, regardless of which CDS they use. In addition, customers can preserve their investment in workflow definition and supporting operating procedures.

"At Agilent, we are fully committed to bringing instrument control solutions for the most popular chromatography instruments into OpenLAB CDS," said Bruce von Herrmann, general manager of Agilent Software and Informatics. "Our agreement with Shimadzu represents the continued cooperation of both companies. OpenLAB CDS customers will now be able to utilize the features and functions of their Shimadzu instrumentation on our newest CDS product."

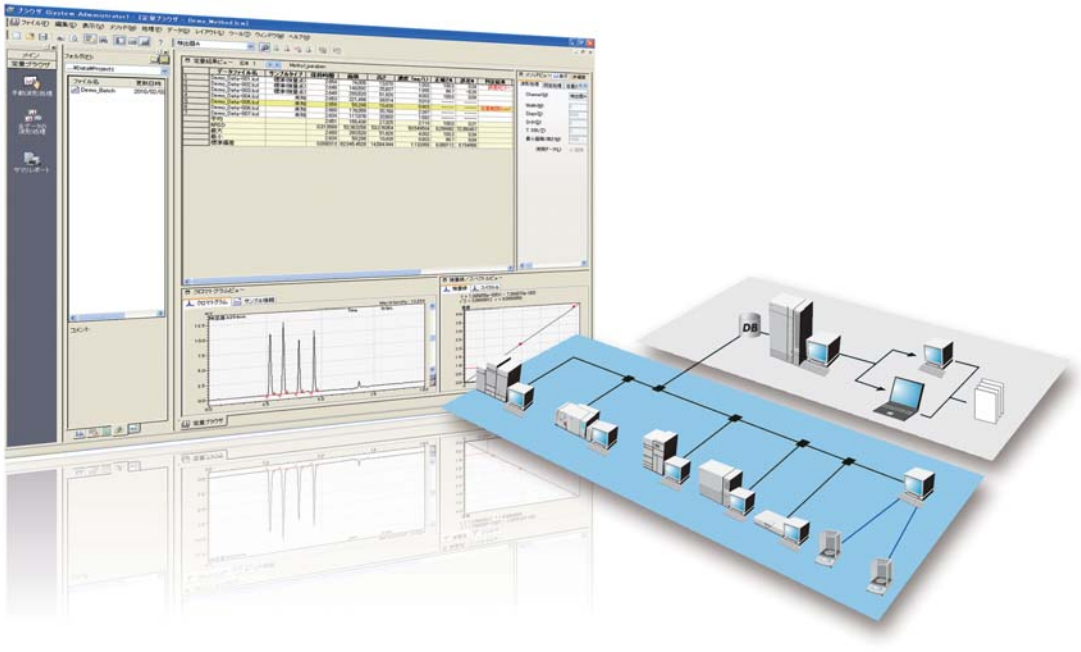
"At Shimadzu, we are dedicated to providing flexible instrumentation and software solutions for our customers," said Masami Tomita, Shimadzu general manager of LC Business Unit, Life Science Business Department. "We are pleased to announce that Shimadzu instruments are now able to be controlled by Agilent OpenLAB CDS. Our collaboration will provide a more integrated solution for customers who require a single CDS product to provide seamless multi-vendor control of all instruments in their laboratory. This will allow any CDS which supports RC.Net to control our instruments. Shimadzu's adoption and implementation of the RC.Net standard will also enable Agilent instruments to be controlled by Shimadzu LabSolutions CDS."

The new Shimadzu RC.Net drivers will initially support Shimadzu's Nexera and Prominence HPLC lines. These drivers will be made available on Agilent's OpenLAB CDS later this month. Shimadzu's LabSolutions CDS will gain the Agilent HPLC RC.Net drivers, supporting Agilent's 1100, 1200, 1260 and 1290 series instruments. This integration is expected to occur in August.

For GCs, the new Shimadzu RC.Net drivers will initially support Shimadzu's GC-2010/Plus and GC-2014 product lines in exchange for Agilent's GC RC.Net drivers supporting Agilent 6890, 7820 and 7890 series instruments. The expected timing for completing this development is late 2013.

For more information about Shimadzu's LabSolutions chromatography systems, visit www.shimadzu.com/an/data-net.

For more information about Agilent OpenLAB CDS, visit www.chem.agilent.com/en-US/Products-Services/Software-Informatics.



New Products

MALDI-7090™

New MALDI TOF-TOF Mass Spectrometer Combining High Throughput, High Resolution and High Energy for the Ultimate in Tandem MS Capabilities

The combination of Shimadzu's extensive MALDI-TOF-TOF expertise with novel patented technology provides the MALDI-7090 with ultimate capabilities for identification and structural characterization of biomolecules.

Features
- Market-leading high-resolution MALDI MS/MS capability
- Unique high-energy CID fragmentation
- Flexible platform for both novice and expert users
- Targeted software modules within MALDI Solutions address a wide variety of research applications including proteomics and tissue imaging

HyperVision HPV-X

A World's First – Journey to Unknown Realms of High Resolution and Ultra-High Speeds

In a variety of scientific applications, phenomena elapse in an instant, leaving no clue to their solution. High-speed video cameras record such moments, enabling visualization through slow-motion replay.

Features
- Ultra-high-speed continuous recording at up to 10 million frames per second
- Increased recording capacity
- Capable of high-resolution recording at the highest speeds

Tracera

Highly Versatile GC Analyzer for Trace Gas Analysis Features a New Barrier Discharge Ionization Detector

The new Tracera GC System is available to solve your trace analysis needs. This system utilizes the new Barrier Discharge Ionization Detector coupled with a GC-2010 Plus capillary gas chromatograph to create a GC system that makes it possible to reveal trace components that are difficult to see by other GC detectors.

Features
- High sensitivity
- Novel universal detector
- Long-term stability

DSC-60 Plus

Differential Scanning Calorimeter Addresses a Wide Range of Applications

The DSC-60 Plus is an indispensable thermal analyzer for materials characterization in R&D and quality control applications in such areas as polymers, pharmaceuticals, electronic parts, and foods. It offers the sensitivity and easy operation required for the development of high-performance, highly functional new materials.

Features
- High-performance general-purpose DSC
- Diverse measurements using simple operations
- Complies with analytical laboratory regulations

Call for
Papers for Shimadzu Young Scientist Award

Shimadzu Asia Pacific invites nominations of scientific papers to recognize exceptional achievements by young Asian scientists under the age of 40 in the field of the Mass Spectrometry. The thesis/research should provide a significant contribution to analytical science development and application towards contributing to Society through Science and Technology.

Eligibility

- Candidate is expected to be employed at a government institution of research / higher education in Asia - India, Singapore, Thailand, Vietnam, Indonesia, Malaysia – and should have completed his/her Master's or PhD degree
- Demonstrable outstanding contribution to the field of analytical science using Mass Spectrometry
- Should be 40 years or younger in age by December 2013

Required documents

- A letter outlining the eligibility of the nominee
- Curriculum vitae of the nominee
- Publication/thesis document (published not earlier than January 1, 2012)
- Number of citations

All qualified applications will be evaluated by an international panel of five recognized Mass Spectrometry experts from different fields and regions.

Awards:

1. Cash Reward of **USD5000 (Gold), USD4000 (Silver)** and **USD3000 (Bronze)**
2. A trophy of recognition
3. Research paper published on Shimadzu website
4. Research paper published in Shimadzu e-Magazine
5. Chance to present their work in a public symposium
6. Joint project with Shimadzu Asia Pacific funded by the company

Deadlines

Nominations and applications are open until October 15, 2013, Singapore time. The award will be presented in a public symposium in Singapore in November/December 2013.

For further details of the application procedure, please visit ***http://www.shimadzuysa.com/***

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