

# Rapid Multiresidue Method for the Determination of more than 300 Pesticide Residues in Food

Masahiro Okihashi<sup>1\*</sup> • Yoko Kitagawa<sup>1</sup> • Hirotaka Obana<sup>1</sup> • Yukio Tanaka<sup>1</sup> • Yoko Yamagishi<sup>2</sup> • Kuniyo Sugitate<sup>2</sup> • Kaori Saito<sup>2</sup> • Masayuki Kubota<sup>2</sup> • Michiko Kanai<sup>2</sup> • Taisuke Ueda<sup>3</sup> • Syuichi Harada<sup>3</sup> • Yoshio Kimura<sup>3</sup>

<sup>1</sup> Osaka Prefectural Institute of Public Health, Nakamichi 1-3-69, Higashinari-ku, Osaka, 537-0025, Japan

<sup>2</sup> Thermo Fisher Scientific K.K., C-2F, 3-9 Moriya-cho, Kanagawa-ku, Yokohama, 221-0022, Japan

<sup>3</sup> Hayashi Pure Chemical. IND., LTD, 3-2-12 Uchihiranomachi, Chuo-ku, Osaka, 540-0037, Japan

Corresponding author: \* okihasi@iph.pref.osaka.jp

## ABSTRACT

The aim of this study was to develop a simple and efficient sample preparation methodology in pesticide multiresidue analysis that shortens the analytical process during extraction and cleanup. We have conducted recovery tests of about 300 pesticides in foods with a modified method reported previously. Ten g of sample was extracted with 20 ml of acetonitrile using a high-speed homogenizer. One gram of NaCl and 4 g of anhydrous MgSO<sub>4</sub> were added and shaken immediately. The tube was centrifuged to separate the sediment and water from the acetonitrile extract. The acetonitrile layer obtained after salting out was loaded into the double-layered SPE cartridge with a graphitized carbon black and primary secondary amine, followed by elution with acetonitrile-toluene (3:1). The eluate was evaporated and the residue was dissolved in acetone-hexane (1:9) or methanol. The test solution was determined by a GC-FPD for organophosphorous pesticides, GC-MS in the NCI mode for organochlorines, pyrethroids and other halogenated pesticides, and GC-MS in the EI mode for other pesticides. LC-MS/MS was also used to determine less volatile pesticides. Recovery studies were performed by fortifying 3 or 5 matrices at 0.05 or 0.1 µg/g. Recoveries of about 300 pesticides were mainly 70-110% and the relative standard deviations were below 20%. Limits of detection ranged between 0.1 and 50 ng/g for tested pesticides.

**Keywords:** analysis, fruits, QuEChERS, vegetables, pesticides, residues

**Abbreviations:** EI, electron ionization; ESI, electrospray ionization; FPD, flame photometric detector; GC, gas chromatography; LC, liquid chromatography; MRL, maximum residue limit; MRM, multiple reaction monitoring; MS, mass spectrometry; MS/MS, tandem mass spectrometry; NCI, negative chemical ionization; RSD, relative standard deviation; SIM, selected ion monitoring; SPE, solid phase extraction

## INTRODUCTION

The Japanese Ministry of Health, Labour and Welfare has issued MRLs of about 800 pesticides and veterinary drugs and introduced the positive list system (Notification No. 497-499, 2005). In this system, all agricultural chemicals are regulated under a uniform limit (0.01 µg/g) except for MRLs. Foods in which any agricultural chemical residues are found in excess of MRLs or the uniform limit will be excluded from the market as illegal. This system does not require analyzing all pesticides before distribution, but the demands for pesticide residue analysis of commodities are increasing for various pesticides. The Quarantine Station in Japan has been monitoring imported foods to ensure that pesticide residues do not exceed the MRLs (Hirahara *et al.* 2005), local governments have monitored their regional markets as a usual policy. Food distributors are eager to monitor their commodities to reduce risks of violation, farm cooperatives want to warrant their products safe, some food companies and supermarkets show “no pesticide” as a value-added product, and consumers’ cooperatives want to keep away from residual pesticides. Thus, numerous governmental and private laboratories have an interest to performed residue analysis of foods at early stages of the distribution chain. These institutions monitoring pesticide residues in foods require fast and efficient multiresidue methods with a broad scope of application in order to maximize the coverage of their monitoring targets. To date, many multiresidue analytic methods have been reported (Fillion *et al.* 2000; Obana *et al.* 2001; Klein *et al.* 2003;

Pang *et al.* 2006). In a previous paper, we proposed a rapid multiresidue method for the determination of 180 pesticides in foods using GC-FPD and GC-MS (Okihashi *et al.* 2005) based on the QuEChERS method (Anastassiades *et al.* 2003). The positive aspects of our method against the original QuEChERS were the powerful extraction with the homogenizer and the efficient cleanup with the double-layered SPE. This SPE step involved evaporation and reconstitution by hexane that could remove water-soluble substances, such as sugars and salts, indirectly. In this study, the method was slightly modified and the targets were extended to 380 pesticides and metabolites according to commercially pre-mixed standard solutions. These pesticides were detected by GC-MS in EI and NCI, and by GC-FPD and LC-MS/MS in the positive and negative ESI modes.

## MATERIALS AND METHODS

### Apparatus

#### GC-MS (EI) instrument

The extracts were analyzed with a Thermo Fisher (Waltham, MA) TRACE GC Ultra and POLARIS Q ion trap mass spectrometer. EI was applied in the MS instrument. The system was equipped with a split/splitless injection inlet, electronic pressure control, and an AS-2000 auto sampler. Xcalibur software was used for instrument control and data analysis.

### GC-MS (NCI) instrument

The extracts were analyzed with a Shimadzu (Kyoto, Japan) GCMS-QP2010 gas chromatograph mass spectrometer. NCI was applied in the MS instrument. The system was equipped with a split/splitless injection inlet, electronic pressure control, and AOC-20i auto injector. GCMSolutions software was used for instrument control and data analysis.

### GC-FPD instrument

The extracts were analyzed with a Shimadzu GC-17A gas chromatograph equipped with a FPD, a split/splitless injection inlet, and an AOC-14 auto injector. C-R7A was used for instrument control and data analysis.

### LC-MS/MS instrument

The extracts were analyzed with a Thermo Fisher TSQ Quantum Discovery MAX mass spectrometer using ESI mode. The LC instrument was a Finnigan Surveyor Plus with a quaternary pump and an autosampler. Xcalibur software was used for instrument control and data analysis.

### Food processor, mill and homogenizer

A Toshiba (Tokyo, Japan) QS-7 food processor was used to comminute fruit and vegetable samples. A Matsushita (Osaka, Japan) MX-X61 was used to mill rice and soybean. A Hitachi (Tokyo, Japan) HG30 homogenizer was used to blend sample and acetonitrile in the extraction step.

### Tube and centrifuge

For the extraction step, Becton Dickinson (Franklin Lakes, NJ) BLUE MAX 50 ml polypropylene conical tubes were employed. A Hitachi Himac SCR 20B centrifuge was utilized for these tubes.

### Analytical balance

A Sartorius (Westbury, NY) BP2100S top-loading balance was used to weigh the chopped samples and solid reagents.

### Solvent evaporator

An Iwaki (ASAHI TECHNO GLASS, Chiba, Japan) REN-1000 and REN-1 rotary evaporator was employed to concentrate eluates.

### Reagents

**Acetonitrile, toluene, acetone, n-hexane, methanol, ammonium acetate, anhydrous magnesium sulfate, sodium chloride and water**

The organic solvents were of pesticide analysis grade from Wako Pure Chemical Ind. (Osaka, Japan). All solid reagents were of analytical grade from Wako. Ultrapure water from a Millipore (Billerica, MA) MILLI-Q SP TOC water purification system was used for preparing aqueous solutions.

### Pesticide standard mixture solutions

GC/MS-I, GC/MS-II, GC/MS-III, GC/MS-IV, GC/FPD-V, GC/MS-VI, LC/MS\_PLMix-1, LC/MS\_PLMix-2, and LC/MS\_PLMix-3 were obtained from Hayashi Pure Chemical (Osaka, Japan). The mixtures contained 50, 49, 49, 59, 65, 36, 60, 12, 31 pesticides respectively and were dissolved in acetone (GC series) or acetonitrile (LC/MS series) at concentrations of 10 µg/ml each. These solutions were used for spiking the samples and also for calibration purposes after appropriate dilution.

### SPE cartridges

Octadecylsilane 500 mg cartridges (ODS) and double-layered cartridges with 500 mg of graphitized carbon black and 500 mg of

primary secondary amine (GCB/PSA) were obtained from Supelco (Bellefonte, PA) as ENVI-C18 and ENVI-Carb/PSA, respectively. ODS was preconditioned with 10 ml of acetonitrile and GCB/PSA was preconditioned with the elution of 30 ml mixture of acetone-toluene (3:1).

### Food samples

Blank samples of rice, soybean, grapefruit, spinach, cabbage used in experiments were purchased from a local market in Osaka. About 500-1000 g of fruit and vegetable samples were well homogenized in the food processor. Rice and soybean were comminuted in the mill. All homogenous samples were placed in plastic storage bags, and stored at -20°C until their use for fortification experiments.

### Extraction and cleanup procedure

Appropriate numbers of 50 ml tubes with caps containing  $4.0 \pm 0.2$  g anhydrous  $\text{MgSO}_4$  +  $1.0 \pm 0.1$  g NaCl were prepared in advance. An aliquot of 10 g of sample homogenate was weighed into a 50 ml tube. For samples of grain, 5 g of sample was weighed and 5 ml of water was added. The sample was extracted with 20 ml of acetonitrile by a HG30 homogenizer for 1 min. The  $\text{MgSO}_4$  and NaCl in the tube prepared were added and shaken immediately for about 30 s with the screw cap on. The extract was centrifuged for 10 min at  $1000 \times g$  to separate the sediment and water from the acetonitrile. Next, 16 ml (equivalent to 8 g or 4 g of sample) of the acetonitrile layer obtained after centrifugation was loaded into a GCB/PSA SPE tube. In the case of fatty samples including rice and soybean, the acetonitrile extract was passed through an ODS SPE tube, followed by washing with 10 ml of acetonitrile, and all eluates were loaded to a GCB/PSA tube. Pesticides were eluted gravitationally with 30 ml of acetonitrile-toluene (3:1). The eluate was evaporated and the residue was dissolved in 8 ml or 4 ml of acetone-hexane (1:9), based on sample size, for GC-FPD and GC-MS analysis, or methanol for LC-MS/MS analysis. The concentration of the sample represented by the test solution was 1 g/ml. Fig. 1 summarizes the procedure.

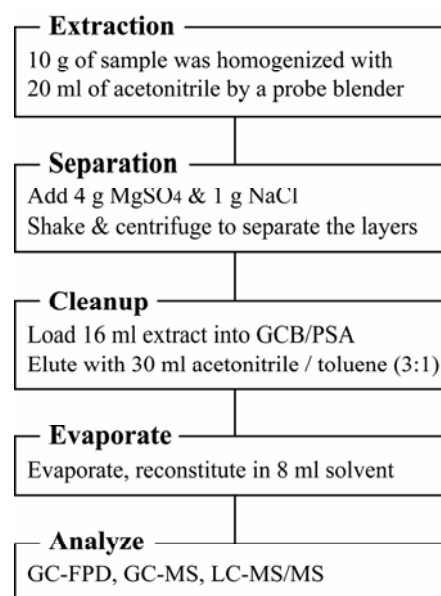


Fig. 1 Flow chart of the sample analysis method used in this study.

### Fortifications

In recovery studies, 100 µl of GC standard mixture solutions or 50 µl of LC-MS/MS standard mixture solutions were added to each 10 g of fruit and vegetable samples. In the case of rice and soybean, 50 µl of GC standard mixture solutions or 25 µl of LC-MS/MS standard mixture solutions were added to 5 g of samples. The tubes containing fortified sample were left for 30 min to give them time to interact with the matrix.

## Preparation of matrix matched calibration standards

Calibration was achieved by preparing matrix-matched calibration standards from the extracts of blank samples in order to compensate for the matrix effect (Erney *et al.* 1993). Analytes were quantified by using a 3-point calibration with those matrix-matched calibration standards corresponding to the spiked concentration.

## GC-MS (EI) analysis

GC-MS (EI) analysis was conducted on a Rtx-5ms (Restek, Bellefonte, PA) capillary column of 30 m, 0.25 mm I.D., 0.25  $\mu$ m film thickness, and the following conditions were used: helium carrier gas flow 1.5 ml/min, injection temperature 250°C, injection volume 1  $\mu$ l (splitless), MS transfer line temperature 280°C, ion source temperature 250°C, oven temperature program 60°C for 1 min; then 8°C/min ramp to 280°C and held for 5 min. Total run time was 33.5 min. Full scan analysis (80-450 m/z) was used.

## GC-MS (NCI) analysis

GC-MS (NCI) analysis was conducted on a DB-5 (Agilent, Folsom, CA) capillary column of 30 m, 0.25 mm I.D., 0.25  $\mu$ m film thickness, and the following conditions were used: helium carrier gas flow 1.7 ml/min, injection temperature 250°C, injection volume 1  $\mu$ l (splitless), MS transfer line temperature 250°C, ion source temperature 200°C, oven temperature program 60°C for 1 min; then 20°C/min ramp to 170°C and 6°C/min ramp to 300°C and held for 7 min. Total run time was 35.17 min. The SIM mode was used for recovery experiments.

## GC-FPD analysis

GC-FPD analysis was conducted on a DB-1701 (Agilent) capillary column of 30 m, 0.32 mm I.D., 0.25  $\mu$ m film thickness, and the following conditions were used: helium carrier gas flow 2.0 ml/min, injection temperature 250°C, injection volume 2  $\mu$ l (splitless), detector temperature 280°C, oven temperature program 80°C for 2 min; then 20°C/min ramp to 180°C followed by 4°C/min ramp to 260°C and 10°C/min ramp to 280°C and held for 5 min. Total run time was 34 min.

## LC-MS/MS analysis

LC-MS/MS analysis was conducted on a Hypersil GOLD (Thermo Fisher) column of 150 mm, 2.1 mm I.D., 5  $\mu$ m particle size. The injection volume was 2  $\mu$ l and oven temperature was 40°C. A gradient elution program at 0.2 ml/min flow, in which both reservoirs contained 5 mM ammonium acetate in (A) water and (B) methanol, was used as follows; 2% solution B ramped to 95% linearly over 15 min then held for 10 min, then returned to 2% solution B and allowed to equilibrate for 10 min. The ESI source was used in the positive and negative mode, and ion transfer tube temperature was 330°C, ion spray voltages were 4200 V/positive and 3250 V/negative respectively.

## RESULTS AND DISCUSSION

### Method modification

In QuEChERS method, PSA particles were used for cleanup procedure as dispersive-SPE (Anastassiades *et al.* 2003). This cleanup was very quick because evaporation and reconstitution steps were not required. Saito *et al.* examined removal efficiencies of various SPE cartridges against pigments and fatty acids, and they reported that the combination of GCB and PSA provided excellent results (Saito *et al.* 2004). We chose traditional SPE of GCB/PSA taking account of cleanup efficiencies. In our previous paper (Okihashi *et al.* 2005), we used acetic acid to control the PSA capacity and improve recoveries of chinomethionat and chlorothalonil. In that case, the weakened PSA allowed the elution of such pesticides as well as many food co-extracted matrix components that deteriorate instrument performance

in terms of resolution and detector sensitivity. In routine analysis, it is significant to maintain instruments that work well for accurate results. Frequent maintenance requires much time and consequently decreases total throughput of routine analysis. In this work, we postponed a few pesticides, such as chinomethionat and chlorothalonil, to save time in sample preparation. The extraction and cleanup steps were performed without acetic acid to reduce food matrices and protect instruments. In the case of rice and soybean, an ODS SPE was additionally used to remove excess fat.

## Measurement

As most GC-MS-instruments are restricted in their capacity of simultaneously recording SIM-ions, the number of pesticides that can be effectively measured within one run is limited for example to max. 200 compounds (Fillion *et al.* 2000; Pang *et al.* 2006). In the proposed method, three kinds of GC instruments were used instead of several injections to a GC-MS and it enabled reciprocal confirmation analysis. The standard mixtures of all GC series (GC/MS-I, II, III, IV, VI, GC/FPD-V) were measured by GC-MS (EI), at first. A scan test of each standard mixture solution was conducted initially to obtain its mass spectrum and retention time. Based on the mass spectrum, one ion was selected to quantify each compound. In GC-MS (NCI), the standard mixtures of GC/MS series (GC/MS-I, II, III, IV, VI) were measured. Some pesticides that contained halogen atoms, especially organochlorines and pyrethroids, could be detected at lower levels than those with GC-MS (EI). A scan test of each standard mixture solutions was conducted to choose one quantitative ion for each pesticide. Two SIM measurements that consisted of 4 or 6 segments were designed for these pesticides. In both measurements, 6 to 13 ions were monitored simultaneously in a segment with 0.2 sec. In contrast to GC-MS (EI) no serious interference was observed with GC-MS (NCI), while food matrices were frequently detected and sometimes interfered with the results in GC-MS (EI). Organophosphorous pesticides (GC/FPD-V) were detected using GC-FPD with a DB-1701, a mid-polarity phase column. In routine analysis, it saves more time to recognize the negative results for various organophosphorous pesticides from one copy of a flat chromatogram. The data from GC-MS is composed of many mass chromatograms and takes some time to confirm in data analysis. GC-FPD is useful to shorten the time needed for identification. But some pairs of pesticides overlapped in a FPD chromatogram. To resolve these pesticides, the data from GC-MS (EI) was used. The quantification ions of these compounds that are amenable to GC analysis are listed in **Table 1**. The limits of detection were defined based on the noise levels on the chromatograms of the blank sample solution and the respective standard peaks.

The standard mixtures of the LC/MS series (LC/MS PLMix-1, 2, 3) were mixed and measured by LC-MS/MS using the MRM modes. Suitable transitions from precursor to product ions (MRM transitions) were identified. At first, full scan mass spectra of each pesticide were recorded to obtain retention times and determine precursor ions. Next, the product ions were examined through the daughter scan mode. Then, the most abundant product ion for each compound was chosen for MRM mode. Almost all of the compounds were measured by the positive ESI mode; however, some were also measured by the negative ESI mode. The MRM transition and collision energy of each compound are summarized in **Table 2**. The dwell time used for each of these analytes was 20 ms. In the positive ESI mode, analytes were divided into two segments to shorten the cycle time. These MRM chromatographic peaks indicated sufficient intensity to detect at a level of 0.1 ng/g except for a few pesticides.

**Table 1** Mean recovery (%) and relative standard deviation (%).

Mix	Compound	Detector	Ion (m/z)	LOD (ng/g)	Recovery (%) and RSD (%), (n=5)									Frequency	
					Cabbage	Grapefruit	Spinach	Rice	Soybean						
I	Acrinathrin	NCI	333	0.5	88	4	89	16	72	3	48	17	28	39	3
IV	Allethrin	NCI	256	0.5	101	3	90	10	102	3	86	7	83	7	5
I	Benfluralin	NCI	335	0.1	91	3	74	9	73	5	72	7	77	7	5
III	BHC, $\alpha$ -	NCI	71	0.2	83	3	92	3	70	2	89	6	76	10	5
III	BHC, $\beta$ -	NCI	71	0.5	87	3	94	4	76	1	96	3	87	5	5
III	BHC, $\delta$ -	NCI	71	0.2	90	3	93	5	75	1	95	2	117	12	5
II	BHC, $\gamma$ -	NCI	71	0.2	89	4	88	7	82	2	91	5	78	6	5
I	Bifenox	NCI	341	0.2	80	4	87	7	90	3	84	11	76	5	5
I	Bifenthrin	NCI	205	2	91	3	89	12	95	2	80	5	63	6	4
IV	Bromacil	NCI	262	2	83	3	80	16	109	1	86	6	84	4	5
III	Butafenacil	NCI	390	10	77	3	92	16	85	2	71	9	42	59	4
I	Cafenstrole	NCI	250	1	94	1	84	9	92	3	61	24	46	26	3
I	Captafol	NCI	150	0.2	NC	-	69	8	29	5	NC	-	NC	-	0
I	Captan	NCI	150	2	16	6	69	7	38	6	NC	-	NC	-	0
III	Carfentrazone-ethyl	NCI	375	0.5	74	3	96	9	83	3	85	6	80	6	5
IV	Chinomethionat	NCI	206	0.1	NC	-	35	15	NC	-	NC	-	NC	-	0
I	Chlorfenapyr	NCI	349	0.1	78	4	87	10	86	3	88	8	77	5	5
VI	Chlormefos	NCI	185	1	90	4	93	7	77	3	75	15	29	75	4
I	Chlornitrofen	NCI	281	0.5	78	4	88	8	89	3	91	5	71	4	5
I	Chlorobenzilate	NCI	278	10	84	4	90	10	89	4	90	8	73	3	5
I	Chlorothalonil	NCI	266	2	NC	-	24	25	NC	-	39	25	NC	-	0
VI	Cyflufenamid	NCI	340	0.2	97	2	90	8	94	5	76	15	77	6	5
I	Cyfluthrin	NCI	207	0.5	82	4	90	10	88	1	76	13	63	8	4
III	Cyhalothrin	NCI	205	0.2	82	2	96	9	80	3	77	7	67	19	4
I	Cypermethrin	NCI	207	1	82	4	90	11	90	1	77	12	64	6	4
VI	DEF	NCI	257	0.1	93	2	88	7	91	3	70	9	63	6	4
I	Deltamethrin	NCI	79	2	88	3	89	8	75	4	69	12	71	17	4
VI	Dialifos	NCI	185	1	87	3	89	10	93	2	71	18	49	17	4
I	Dichlofluanid	NCI	207	0.5	83	3	74	9	80	3	75	14	28	8	4
I	Dicofol	NCI	250	0.2	83	3	86	7	77	2	86	4	68	4	4
I	Dimethipin	NCI	211	2	86	3	82	5	86	5	76	25	95	5	5
VI	Diniconazol	NCI	289	0.2	101	1	79	11	102	6	77	11	77	6	5
VI	Ditalimfos	NCI	299	0.2	51	18	78	5	25	19	NC	-	NC	-	1
I	Dithiopyr	NCI	386	0.1	88	3	87	7	82	2	92	8	86	4	5
I	Endosulfan- $\alpha$	NCI	242	0.5	81	3	88	8	83	2	92	3	72	5	5
I	Endosulfan- $\beta$	NCI	242	2	86	3	87	8	89	5	85	7	69	6	4
I	Fenarimol	NCI	276	0.2	89	3	80	12	94	3	81	12	73	7	5
VI	Fenchlorphos	NCI	211	0.1	84	3	95	8	82	2	83	7	70	5	5
III	Fenoxanil	NCI	201	0.5	86	3	92	14	84	3	86	6	83	5	5
II	Fenpropathrin	NCI	141	0.5	89	3	86	10	101	4	76	10	66	6	4
II	Fenvalerate	NCI	211	0.2	82	4	90	15	86	1	73	15	60	9	4
II	Fipronil	NCI	384	0.2	85	3	84	8	79	2	66	22	61	7	3
IV	Fluacrypyrim	NCI	221	0.5	78	2	93	9	119	1	93	5	86	4	5
II	Flucythrinate	NCI	243	0.2	84	4	88	11	90	1	71	21	73	6	5
II	Flutolanil	NCI	257	0.5	98	1	75	12	94	4	80	9	81	6	5
II	Fluvalinate	NCI	294	0.2	86	4	89	11	79	3	61	18	26	41	3
IV	Folpet	NCI	146	1	28	12	64	13	45	11	NC	-	NC	-	0
II	Fthalide	NCI	272	0.1	79	5	84	7	78	1	84	9	71	5	5
III	Indanofan	NCI	174	2	75	4	92	16	82	2	81	11	80	7	5
II	Iprodione	NCI	329	5	90	2	82	17	96	3	69	17	70	5	4
II	Isoprothiolane	NCI	262	1	98	2	84	6	96	3	83	10	79	6	5
II	Kresoxim-methyl	NCI	174	1	97	2	85	11	95	4	75	12	76	6	5
VI	Leptophos	NCI	241	0.5	77	4	92	11	89	2	71	14	47	6	4
II	Metribuzin	NCI	198	0.2	102	3	70	8	88	3	82	13	77	6	5
III	Nitralin	NCI	345	0.5	81	3	72	10	59	7	72	10	82	6	4
IV	Pacllobutrazol	NCI	256	0.5	85	4	89	10	113	6	94	6	88	9	5
II	Pendimethalin	NCI	281	0.5	98	4	83	7	87	2	85	5	78	5	5
II	Pentoxazone	NCI	240	1	90	2	84	14	95	3	79	15	65	6	4
II	Procymidone	NCI	282	20	51	11	82	5	94	5	84	14	74	7	4
II	Propyzamide	NCI	255	0.5	103	4	85	7	91	3	88	9	84	7	5
II	Pyraflufen-ethyl	NCI	349	2	76	3	85	9	114	5	72	18	74	7	5
VI	Pyrazophos	NCI	373	2	93	2	84	10	95	3	74	15	72	7	5
III	Pyridaben	NCI	217	5	75	3	93	9	84	2	81	10	72	5	5
II	Pyrifenox E	NCI	226	1	94	2	77	10	95	4	81	8	84	9	5
II	Pyrifenox Z	NCI	384	5	103	5	86	4	75	3	66	21	87	6	4
IV	Quintozene	NCI	249	0.2	88	2	93	8	106	2	93	17	68	4	4
III	Quizalofop-ethyl	NCI	372	5	83	4	93	6	80	2	82	12	78	6	5
VI	Spirodiclofen	NCI	311	0.5	65	11	86	11	42	11	14	13	NC	-	1
IV	Tecnazene	NCI	215	0.1	80	3	93	8	99	3	77	11	59	11	4
III	Tefluthrin	NCI	241	0.1	92	2	93	4	72	2	93	4	81	5	5
IV	Tetraconazole	NCI	117	0.2	91	2	88	11	105	2	96	4	90	3	5
IV	Tetramethrin	NCI	331	5	83	3	98	10	113	3	83	7	80	5	5
III	Thifluzamide	NCI	85	0.5	84	3	90	17	83	3	84	7	82	5	5
III	Thiobencarb	NCI	132	5	95	2	91	3	75	3	85	4	78	7	5
III	Thiocyclam	NCI	136	0.5	58	7	72	9	55	4	43	16	NC	-	1

Table 1 (cont.)

Mix	Compound	Detector	Ion (m/z)	LOD (ng/g)	Recovery (%) and RSD (%), (n=5)								Frequency		
					Cabbage	Grapefruit	Spinach	Rice	Soybean	NC	NC	NC			
III	Tolylfluanid	NCI	212	5	18	9	69	4	37	5	NC	-	NC	-	0
III	Triadimefon	NCI	127	0.5	90	4	90	7	77	1	90	4	82	5	5
III	Trichlamide	NCI	230	2	77	4	89	7	80	2	86	4	82	5	5
III	Uniconazole P	NCI	291	2	87	3	83	11	83	3	77	9	75	4	5
III	Vinclozolin	NCI	241	0.1	83	3	93	4	78	1	91	3	82	8	5
VI	2,6-Dichlorobenzamide	EI	173	5	90	7	67	15	122	4	68	25	92	15	2
IV	Acetochlor	EI	223	5	84	3	93	5	111	4	99	5	87	5	5
I	Alachlor	EI	188	2	87	6	87	8	114	1	87	6	88	2	5
IV	Ametryn	EI	227	5	88	9	114	9	110	4	101	4	91	4	5
IV	Amitraz	EI	162	50	60	37	35	55	62	7	53	13	51	8	0
I	Atrazine	EI	200	5	88	5	85	5	117	2	86	8	93	4	5
VI	Azaconazole	EI	217	5	85	7	90	24	114	1	75	14	85	6	4
VI	Azamethiphos	EI	199	20	86	3	72	11	95	3	NC	-	NC	-	3
VI	Azinphos-ethyl	EI	132	5	82	6	100	15	118	10	64	32	92	7	4
I	Azoxystrobin	EI	344	10	84	4	85	4	111	4	65	25	93	9	4
IV	Benalaxyl	EI	148	2	66	3	107	9	111	2	99	6	87	5	4
IV	Benfuresate	EI	163	2	83	3	90	10	106	3	98	4	94	5	5
VI	Benzylaminopurine	EI	225	-	NC	-	NC	-	NC	-	NC	-	NC	-	0
III	Bifenazate	EI	258	2	60	8	101	18	76	6	74	18	99	10	4
I	Bioresmethrin	EI	143	5	68	6	70	15	77	9	66	16	72	6	3
IV	Biphenyl	EI	154	2	56	16	74	23	62	10	52	25	16	57	0
IV	Bitertanol	EI	170	5	52	5	NC	-	111	3	102	4	91	4	3
I	Bromobutide	EI	232	2	89	6	88	10	109	3	83	7	85	3	5
VI	Bromophos	EI	331	2	90	5	87	11	113	2	79	5	76	8	5
I	Bromopropylate	EI	341	2	86	5	87	10	112	5	86	6	79	6	5
VI	Bromuconazole	EI	295	20	85	8	76	30	113	4	83	9	80	13	4
I	Buprofezin	EI	175	5	82	8	NC	-	117	3	87	5	81	6	4
IV	Butachlor	EI	160	2	74	4	106	21	103	4	95	6	85	5	4
IV	Butylate	EI	146	5	57	11	75	55	70	11	66	22	41	22	1
IV	Carbetamide	EI	119	20	75	5	89	16	113	3	98	6	99	6	5
IV	Chlormethoxynil	EI	266	5	72	5	136	14	105	5	105	7	89	4	4
IV	Chloridazon	EI	221	5	NC	-	NC	-	107	10	95	9	89	10	3
I	Chlorpropham	EI	127	5	93	7	80	8	118	4	88	6	73	16	5
IV	Chlorpropylate	EI	251	2	78	4	112	10	113	2	98	6	87	10	5
VI	Chlorthiophos	EI	325	2	83	5	85	7	113	6	80	7	80	9	5
IV	Cinmethylin	EI	105	5	56	7	92	19	112	4	92	10	80	5	4
I	Clofentezine	EI	137	20	86	6	57	29	108	6	20	42	7	55	2
I	Clomeprop	EI	288	2	86	7	89	14	116	4	87	6	80	9	5
VI	Crimidine	EI	142	2	89	4	87	3	117	3	78	10	75	16	5
I	Cyanazine	EI	225	5	79	6	85	7	115	2	80	12	87	6	5
I	Cyhalofop-butyl	EI	357	2	85	6	90	7	111	4	78	12	87	7	5
I	Cyproconazole	EI	222	5	82	6	82	17	113	4	79	8	93	4	5
I	Cyprodinil	EI	224	2	89	6	76	8	116	4	82	5	81	4	5
I	Desmedipham	EI	181	5	NC	-	NC	-	124	3	77	21	115	4	1
I	Dichlobenil	EI	171	2	73	3	66	16	115	5	69	14	30	63	2
VI	Diclobutrazol	EI	270	5	84	5	84	8	106	3	77	9	82	8	5
IV	Diclocymet	EI	277	5	75	2	96	7	100	13	100	7	88	4	5
IV	Diclofluanid metabolite	EI	200	50	86	4	NC	-	111	5	102	5	98	5	4
I	Dicloran	EI	176	20	95	2	46	7	110	4	68	11	81	4	3
V	Dicrotophos	EI	127	5	90	6	77	30	104	2	86	6	98	6	4
I	Diethofencarb	EI	225	2	87	4	87	10	116	2	80	12	87	6	5
I	Difenoconazole	EI	323	5	89	8	80	8	105	4	77	15	94	9	5
I	Diflufenican	EI	266	2	85	5	81	7	110	4	84	7	86	7	5
I	Dimepiperate	EI	91	50	NC	-	83	10	115	8	87	3	87	7	4
I	Dimethametryn	EI	212	2	89	5	75	4	116	2	82	5	83	4	5
III	Dimethenamid	EI	154	2	81	2	98	5	117	2	87	5	89	2	5
I	Dimethenamid	EI	154	2	87	5	85	14	105	2	100	5	90	3	5
IV	Dimethomorph	EI	301	5	73	3	89	34	114	5	111	6	93	7	4
IV	Dioxathion	EI	270	50	95	9	115	30	99	7	104	5	117	6	4
I	Diphenamid	EI	167	2	91	4	84	12	115	1	82	12	91	4	5
IV	Diphenylamine	EI	169	2	94	6	73	32	82	3	103	6	50	7	3
I	EPTC	EI	128	10	63	8	61	16	90	4	61	21	29	52	1
I	Esprocarb	EI	222	2	86	6	85	4	114	1	85	5	78	4	5
IV	Ethychlozate	EI	238	5	75	5	96	10	NC	-	NC	-	NC	-	2
IV	Etobenzanid	EI	304	50	NC	-	77	21	106	1	46	22	79	9	2
I	Etofenprox	EI	163	2	93	4	84	6	107	3	85	3	72	6	5
IV	Etoxazole	EI	330	5	76	6	134	44	113	5	92	5	87	4	4
IV	Etoxazole metabolite	EI	246	20	56	24	NC	-	35	13	NC	-	18	7	0
I	Etridiazole	EI	211	5	78	4	80	7	97	4	62	33	33	46	3
IV	Famoxadone	EI	330	5	72	3	93	9	102	4	103	4	90	6	5
II	Fenbuconazole	EI	129	5	84	7	57	42	116	2	72	16	92	7	4
IV	Fenothiocarb	EI	160	10	48	9	92	35	100	8	93	7	88	7	3
II	Fenoxaprop-ethyl	EI	288	2	91	4	84	25	111	6	87	5	79	5	4
VI	Fenoxycarb	EI	255	10	NC	-	NC	-	94	6	50	42	87	17	2
IV	Fenpropimorph	EI	128	2	78	1	91	10	107	2	89	7	67	2	4

Table 1 (cont.)

Mix	Compound	Detector	Ion (m/z)	LOD (ng/g)	Recovery (%) and RSD (%), (n=5)								Frequency		
					Cabbage	Grapefruit	Spinach	Rice	Soybean						
III	Ferimzone	EI	239	20	77	5	99	16	96	8	111	8	106	11	5
II	Fludioxonil	EI	248	2	90	5	116	9	111	2	85	16	102	4	5
IV	Flumioxazin	EI	354	5	80	6	86	29	107	7	76	14	66	18	3
VI	Fluquinconazole	EI	340	2	85	3	71	13	114	4	65	17	49	21	3
II	Flusilazole	EI	233	2	87	6	89	19	109	0	76	13	89	4	5
IV	Flusilazole metabolite	EI	235	2	82	1	119	7	99	6	104	4	85	4	5
IV	Furametpyr	EI	298	2	75	5	97	19	107	1	102	5	89	6	5
IV	Furametpyr metabolite	EI	296	2	45	7	93	18	109	4	96	6	84	6	4
II	Halfenprox	EI	265	5	83	5	87	16	113	4	78	4	73	9	5
II	Hexaconazole	EI	214	5	100	17	75	12	115	10	79	7	90	6	5
III	Indoxacarb MP	EI	264	5	72	5	104	9	111	1	95	7	NC	-	4
II	Iprobenfos	EI	204	5	94	6	79	16	114	2	82	6	89	5	5
IV	Lenacil	EI	153	2	74	4	103	19	113	3	77	8	97	5	5
III	MCPB	EI	142	5	NC	-	NC	-	90	3	59	11	36	43	1
III	MCPB-ethyl	EI	211	50	70	3	80	37	103	6	87	18	78	6	4
II	Mefenacet	EI	192	5	90	5	85	19	149	11	12	90	105	7	3
II	Mepronil	EI	269	5	77	8	62	50	115	6	79	11	81	8	4
II	Metalaxyl	EI	160	5	88	6	82	13	119	5	78	11	91	4	5
IV	Metminostrobin	EI	191	5	77	3	97	7	112	4	93	4	91	5	5
III	Metribuzin DA	EI	184	2	84	3	NC	-	87	5	84	17	72	25	3
III	Metribuzin DADK	EI	154	50	79	12	NC	-	74	5	NC	-	NC	-	2
VI	Molinate	EI	126	5	76	4	80	7	107	3	72	11	51	41	4
II	Myclobutanil	EI	179	5	87	6	87	23	114	2	74	12	93	4	4
VI	Napropamide	EI	128	5	75	7	61	38	107	2	62	18	87	6	3
VI	Nereistoxin	EI	149	-	NC	-	NC	-	NC	-	NC	-	NC	-	0
II	Nitrofen	EI	283	5	76	4	84	13	109	4	66	8	82	11	4
V	Omethoate	EI	156	20	78	10	52	36	95	5	72	12	62	22	3
IV	o-Phenylphenol	EI	170	5	87	2	101	9	95	4	96	4	78	3	5
III	Oxabetrinil	EI	73	50	84	6	80	7	96	4	99	5	89	3	5
IV	Oxadiazon	EI	175	2	78	2	100	14	113	3	96	5	87	3	5
II	Oxadixyl	EI	132	5	90	5	75	45	117	4	72	15	93	6	4
III	Oxpoconazole	EI	179	10	66	9	NC	-	102	6	102	5	84	8	3
III	Oxpoconazole fumarate	EI	114	5	78	4	115	8	108	5	103	7	86	7	5
II	Penconazole	EI	248	2	93	6	76	17	115	3	67	6	90	5	4
II	Permethrin	EI	183	5	81	4	78	16	109	5	83	5	72	8	5
III	Phenmedipham	EI	167	5	92	6	75	15	99	3	100	7	95	15	5
III	Phenothiol	EI	244	5	81	6	87	18	90	4	60	6	36	38	3
IV	Phenothrin	EI	183	5	73	4	107	29	108	6	98	5	71	3	4
V	Phorate	EI	231	5	85	8	80	22	83	3	82	4	84	5	4
V	Phosphamidon	EI	127	5	88	5	97	14	102	4	75	7	93	7	5
III	Piperonyl-butoxide	EI	176	2	75	4	99	4	106	4	97	5	87	5	5
II	Pretilachlor	EI	238	2	85	7	88	20	108	2	92	24	84	5	4
VI	Prohydrojasmon	EI	83	20	92	7	76	27	101	7	82	8	79	6	4
IV	Prometryn	EI	241	2	83	4	97	9	108	2	99	4	88	3	5
IV	Propachlor	EI	120	5	71	9	101	20	106	3	93	5	69	8	4
II	Propanil	EI	161	10	80	2	82	16	126	7	88	13	93	5	4
II	Propargite	EI	135	20	80	5	89	20	114	5	87	12	84	10	5
II	Propiconazole	EI	259	10	77	7	73	26	112	14	88	7	91	6	4
VI	Pyraloclostrobin	EI	132	20	NC	-	NC	-	117	8	95	24	92	8	2
II	Pyrazoxyfen	EI	367	50	84	7	NC	-	129	22	NC	-	NC	-	1
II	Pyributicarb	EI	165	2	87	4	83	21	112	5	77	5	72	7	4
II	Pyrimethanil	EI	198	2	92	6	78	14	113	2	82	5	87	3	5
III	Pyrimidifen	EI	184	2	70	7	103	15	104	2	75	21	71	8	4
II	Pyriminobac-methyl E	EI	302	2	90	6	79	10	110	4	75	13	88	7	5
III	Pyriminobac-methyl Z	EI	302	2	77	4	110	8	108	3	98	6	91	5	5
II	Pyriproxyfen	EI	136	2	98	11	91	8	111	1	84	6	77	7	5
II	Pyroquilon	EI	130	5	97	4	63	25	113	3	78	8	88	7	4
I	Quinoclamine	EI	207	5	NC	-	NC	-	111	1	72	14	38	10	2
II	Silaflluofen	EI	286	2	97	4	81	20	108	4	70	4	67	9	4
II	Simazine	EI	201	5	88	6	91	3	120	3	84	8	92	4	5
VI	Simeconazole	EI	195	20	90	6	78	15	117	3	86	10	87	5	5
II	Simetryn	EI	213	2	NC	-	76	8	NC	-	79	8	94	5	3
VI	Sulfotep	EI	322	2	92	6	87	6	113	2	80	7	71	12	5
III	Swep	EI	187	5	92	3	89	20	105	5	98	3	94	3	5
III	Tebuconazole	EI	250	5	75	5	129	9	93	9	90	5	91	5	4
III	Tebuufenpyrad	EI	333	2	76	2	101	14	107	6	98	4	81	6	5
IV	Terbacil	EI	161	5	80	5	NC	-	111	2	103	4	96	4	4
IV	Terbucarb	EI	205	2	82	2	98	10	106	2	100	3	90	4	5
IV	Tetradifon	EI	356	5	84	3	98	10	109	6	96	5	79	6	5
IV	Thenylchlor	EI	288	2	76	4	105	12	114	4	96	9	88	6	5
III	Thiamethoxam	EI	212	5	57	13	67	23	89	6	95	8	76	15	3
IV	Tolfenpyrad	EI	383	5	70	3	98	21	108	4	97	6	90	7	4
III	Tolylfluamid metabolite	EI	214	20	132	6	148	24	147	5	150	8	136	3	0
III	Triadimenol	EI	168	5	NC	-	122	20	NC	-	101	15	91	9	2
IV	Trifloxystrobin	EI	116	5	70	5	104	33	107	4	95	6	92	4	4

**Table 1** (cont.)

Mix	Compound	Detector	Ion (m/z)	LOD (ng/g)	Recovery (%) and RSD (%), (n=5)									Frequency	
					Cabbage	Grapefruit	Spinach	Rice	Soybean						
III	Trifluralin	EI	264	2	90	3	105	3	93	5	84	7	79	7	5
IV	Xylycarb	EI	122	5	89	4	100	5	110	4	98	8	100	4	5
V	Anilofos	FPD	226	20	114	10	77	7	102	6	92	3	97	6	5
V	Azinphos-methyl	FPD	132	10	117	17	77	8	107	7	88	4	101	7	5
V	Butamifos	FPD	286	10	102	2	88	4	88	5	86	2	87	5	5
V	Cadusafos	FPD	159	5	97	3	93	1	95	4	86	3	82	7	5
V	Carbophenothion	FPD	342	10	98	2	84	7	100	3	89	3	85	5	5
V	Chlorpyrifos	FPD	314	5	102	2	94	3	94	3	86	3	86	4	5
V	Chlorpyrifos-methyl	FPD	286	5	96	3	91	2	94	3	80	6	86	3	5
V	Cyanofenphos	FPD	169	10	110	8	87	6	100	6	90	2	94	7	5
V	Cyanophos	FPD	243	5	95	5	94	6	87	8	85	4	95	7	5
V	Demeton-S-methyl	FPD	109	10	97	4	90	2	85	4	79	8	83	9	5
V	Diazinon	FPD	179	5	99	3	89	1	94	3	87	3	88	3	5
V	Dichlofenthion	FPD	279	5	102	2	92	1	95	3	88	3	85	3	5
V	Dichlorvos	FPD	185	10	86	6	126	2	117	3	54	65	107	6	3
V	Dimethoate	FPD	87	5	112	4	93	5	114	4	131	4	129	4	3
V	Dimethylvinphos	FPD	295	10	101	1	90	4	98	2	87	4	97	7	5
V	Dioxabenzofos	FPD	216	5	97	3	93	1	91	2	77	8	93	6	5
V	Disulfoton	FPD	186	10	98	4	91	1	75	3	90	3	95	4	5
V	Edifenphos	FPD	310	10	110	8	87	8	96	6	82	6	91	6	5
V	EPN	FPD	169	10	110	9	83	8	100	5	88	4	114	11	5
V	Ethion	FPD	231	5	104	2	90	6	97	4	91	2	98	6	5
V	Ethoprophos	FPD	158	5	97	3	92	1	95	4	87	2	91	4	5
V	Etrimfos	FPD	181	10	98	4	91	1	75	3	85	3	81	5	5
V	Fenamiphos	FPD	303	10	103	9	90	6	104	6	89	2	94	8	5
V	Fenitrothion	FPD	260	5	100	1	90	3	95	3	87	3	92	5	5
V	Fensulfotthion	FPD	293	10	114	11	83	9	104	6	91	2	98	6	5
V	Fenthion	FPD	278	5	101	2	91	2	82	3	88	3	84	6	5
V	Fonofos	FPD	246	5	100	2	91	4	95	3	85	3	86	3	5
V	Formothion	FPD	93	5	87	3	86	2	73	4	31	67	42	66	3
V	Fosthiazate	FPD	227	20	105	1	92	4	103	3	95	3	103	8	5
V	Isazophos	FPD	172	5	97	6	77	12	97	4	90	3	95	4	5
V	Isocarboxophos	FPD	136	10	98	3	91	2	91	2	83	12	97	9	5
V	Isofenphos	FPD	213	10	103	1	90	4	98	3	90	3	93	5	5
V	Isoxathion	FPD	177	10	104	4	90	5	101	4	89	3	94	6	5
V	Malathion	FPD	127	10	100	1	91	3	95	3	86	6	94	6	5
V	Mecarbam	FPD	160	10	105	7	89	6	96	6	85	5	93	7	5
V	Methacrifos	FPD	208	5	85	5	92	2	87	4	80	4	82	3	5
V	Methidathion	FPD	145	10	103	3	91	5	99	4	88	4	95	6	5
V	Mevinphos	FPD	127	5	94	4	93	2	90	4	76	10	86	6	5
V	Monocrotophos	FPD	127	5	100	2	91	2	95	3	77	8	93	6	5
V	Naled	FPD	145	10	80	3	67	2	69	3	71	6	70	7	3
V	Parathion	FPD	291	5	102	1	90	3	95	4	89	3	95	6	5
V	Parathion-methyl	FPD	263	5	99	1	90	3	96	3	88	3	91	5	5
V	Phenthoate	FPD	274	10	103	2	91	4	94	3	88	3	95	5	5
V	Phosalone	FPD	182	20	115	13	79	10	109	7	88	4	94	7	5
V	Phosmet	FPD	160	20	116	11	82	8	103	6	68	23	87	14	4
V	Piperophos	FPD	320	10	110	9	83	8	100	5	90	3	96	6	5
V	Pirimiphos-methyl	FPD	290	5	101	2	94	2	94	3	89	3	90	4	5
V	Profenophos	FPD	337	10	105	3	91	7	97	6	79	11	90	6	5
V	Propaphos	FPD	220	5	104	3	59	7	87	5	92	3	90	6	4
V	Prothiophos	FPD	309	5	105	4	92	4	117	4	89	2	79	4	5
V	Pyraclufos	FPD	360	20	123	14	79	7	123	8	92	3	99	6	3
V	Pyridafenthion	FPD	340	10	114	11	83	7	104	5	88	3	96	9	5
V	Quinalphos	FPD	156	5	103	1	90	4	98	3	87	2	90	5	5
V	Sulprophos	FPD	322	10	103	7	88	6	82	4	90	3	88	7	5
V	Terbufos	FPD	231	5	91	5	78	7	85	4	87	3	88	3	5
V	Tetrachlorvinphos	FPD	329	10	101	1	91	5	97	4	88	4	95	6	5
V	Thiometon	FPD	125	5	94	3	90	2	57	4	80	5	68	6	3
V	Tolclofos-methyl	FPD	265	5	99	2	91	2	94	4	88	3	91	5	5
V	Triazophos	FPD	162	5	112	9	86	7	113	5	90	3	88	7	5

NC = Not calculated because of low sensitivity or recovery, or matrix interference.

Frequency = Number of pesticide-commodity combinations where recoveries and RSDs within the acceptable range.

**Table 2** Mean recovery (%) and relative standard deviation (%).

Mix	Compound	Detector	Precursor>daughter ion (m/z) collision energy (eV)	LOD (g/g)	Recovery (%) and RSD (%), (n=5)				Frequency		
					Cabbage	Grapefruit	Rice				
2	Acephate	ESI+	184>143(10)	0.1	29	11	64	9	57	9	0
3	Acetamidiprid	ESI+	223>126(22)	0.1	81	6	63	2	77	5	2
2	Acibenzolar-S-methyl	ESI+	211>136(32)	1	101	13	91	18	91	8	3
1	Aldicarb	ESI+	208>116(10)	0.1	98	15	60	15	66	19	1
1	Aminocarb	ESI+	209>137(25)	0.1	93	8	65	6	80	11	2
1	Bendiocarb	ESI+	224>167(10)	0.1	91	10	60	4	75	14	2
1	Benfuracarb	ESI+	411>195(25)	0.1	48	44	NC	-	71	40	0
3	Benzobicyclon	ESI+	447>257(26)	0.1	34	21	41	3	25	22	0

Table 2 (cont.)

Mix Compound	Detector	Precursor>daughter ion (m/z); collision energy (eV)	LOD (g/g)	Recovery (%) and RSD (%), (n=5)			Frequency			
				Cabbage	Grapefruit	Rice				
2 Benzobicyclon metabolite	ESI+	355>165(23)	0.1	45	8	88	1	61	14	1
2 Benzofenap	ESI+	431>105(36)	0.1	92	3	84	5	83	11	3
1 Butocarboxim	ESI+	208>75(10)	0.1	106	18	58	30	55	32	1
1 Butocarboxim sulfoxide	ESI+	207>132(10)	0.1	59	10	65	22	42	10	0
1 Carbaryl	ESI+	202>145(12)	0.1	95	11	55	6	75	14	2
2 Carbendazim	ESI+	192>160(20)	0.1	85	5	94	4	52	16	2
1 Carbofuran	ESI+	222>165(15)	0.1	101	11	NC	-	84	7	2
1 Carbofuran-3-hydroxy	ESI+	238>181(12)	0.1	96	9	83	9	93	6	3
1 Carbosulfan	ESI+	381>160(15)	0.1	19	46	NC	-	95	23	0
2 Carpropamid	ESI+	334>139(18)	0.1	101	10	57	7	44	12	1
2 Cumyruon	ESI+	303>185(14)	0.1	99	18	86	9	83	10	3
2 Cyazofamid	ESI+	325>108(15)	0.1	99	20	37	9	68	8	1
2 Cycloprothrin	ESI+	499>181(38)	0.1	93	23	59	22	52	12	0
3 Diflubenzuron	ESI+	311>141(31)	0.1	100	7	29	12	38	8	1
2 Dimethirimol	ESI+	210>71(36)	0.1	88	6	43	3	91	8	2
1 Dioxacarb	ESI+	224>123(18)	0.1	90	6	70	10	74	8	3
3 Dymron	ESI+	269>151(14)	0.1	100	16	37	10	80	10	2
1 Ethiofencarb	ESI+	226>107(14)	0.1	92	15	31	27	63	21	1
3 Ethoxyquin	ESI+	218>174(32)	1	18	52	103	8	NC	-	1
2 Fenbutatin oxide	ESI+	519>351(38)	0.1	48	8	NC	-	55	10	0
1 Fenobucarb	ESI+	208>95(17)	0.1	98	12	81	15	80	8	3
2 Fenpyroximate E	ESI+	422>366(15)	0.1	95	10	69	3	77	15	2
2 Fenpyroximate Z	ESI+	422>366(15)	0.1	99	6	79	4	84	12	3
3 Fentrazamid	ESI+	350>197(10)	0.1	118	21	70	14	79	14	2
3 Flufenoxuron	ESI+	489>158(20)	0.1	94	6	47	6	64	12	1
1 Furathiocarb	ESI+	383>195(20)	0.1	100	10	82	10	83	16	3
2 Hexythiazox	ESI+	353>228((16)	0.1	93	10	38	5	74	16	2
3 Imazalil	ESI+	297>159(24)	1	91	4	75	7	115	13	3
3 Imibenconazole	ESI+	411>125(36)	0.1	93	2	75	5	74	11	3
3 Imidacloprid	ESI+	256>209(16)	0.1	55	6	70	4	55	3	1
1 Isoprocarb	ESI+	194>95(16)	0.1	91	7	74	5	84	9	3
3 Isouron	ESI+	212>167(18)	0.1	96	7	61	4	87	9	2
2 Linuron	ESI+	249>182(18)	0.1	99	6	73	10	75	9	3
2 Mepanipyrim	ESI+	224>77(40)	0.1	97	4	57	6	85	9	2
2 Mepanipyrim metabolite	ESI+	244>226(21)	0.1	96	5	69	3	89	8	2
3 Methabenzthiazuron	ESI+	222>165(15)	0.1	91	6	74	5	79	9	3
3 Methamidophos	ESI+	142>94(14)	0.1	62	3	57	7	59	11	0
1 Methiocarb	ESI+	226>169(10)	0.1	99	15	25	15	65	10	1
1 Methomyl	ESI+	163>88(10)	0.1	110	6	73	9	81	11	3
1 Metolcarb	ESI+	166>109(12)	0.1	91	10	45	7	80	13	2
2 Nitenpyram	ESI+	271>126(34)	0.1	58	9	64	9	53	8	0
1 Oxamyl	ESI+	237>72(15)	0.1	80	3	68	20	54	25	1
2 Oxaziclomefone	ESI+	376>190(15)	0.1	103	11	84	8	84	20	3
3 Pencycuron	ESI+	329>125(30)	0.1	101	4	70	4	89	9	3
2 Phoxim	ESI+	299>129(12)	0.1	93	22	70	13	75	10	2
1 Pirimicarb	ESI+	239>182(16)	0.1	94	7	78	6	87	8	3
2 Probenazole	ESI+	224>41(13)	1	21	17	6	18	NC	-	0
2 Prochloraz	ESI+	376>308(14)	0.1	104	28	63	12	76	22	1
1 Promecarb	ESI+	208>151(10)	0.1	105	15	51	12	81	8	2
3 Propamocarb	ESI+	189>102(19)	0.1	98	3	72	6	96	9	3
1 Propoxur	ESI+	210>168(11)	0.1	85	8	61	5	74	18	2
3 Pymetrozin	ESI+	218>105(22)	0.1	70	3	NC	-	43	12	1
3 Pyrazolate	ESI+	439>173(20)	0.1	7	89	14	11	2	71	0
3 Sethoxydim	ESI+	328>178(20)	0.1	76	19	41	17	66	14	1
2 Spinosyn A	ESI+	732>142(35)	0.1	91	2	80	8	71	8	3
3 Spinosyn D	ESI+	746>142(34)	0.1	88	1	81	6	69	10	2
2 Tebufenozide	ESI+	353>133(18)	0.1	117	19	78	9	88	12	3
2 Tepraloxydim DMP	ESI+	245>117(23)	0.1	95	5	86	7	92	8	3
2 Tepraloxydim OH-DMP	ESI+	261>211(10)	0.1	93	10	88	6	91	6	3
1 Terbucarb	ESI+	278>222(10)	0.1	98	11	86	5	84	19	3
3 Thiabendazole	ESI+	202>175(28)	0.1	95	2	NC	-	87	11	2
3 Thiabendazole metabolite	ESI+	218>191(27)	0.1	NC	-	NC	-	NC	-	0
2 Thiocyclam	ESI+	182>137(17)	0.1	86	7	84	11	90	10	3
1 Thiodicarb	ESI+	355>88(16)	0.1	48	31	57	9	46	40	0
1 Thiofanox	ESI+	219>145(15)	0.1	92	23	60	18	60	22	0
1 Thiofanox sulfone	ESI+	251>76(10)	0.1	82	10	74	19	77	7	3
1 Thiofanox sulfoxide	ESI+	235>104(10)	0.1	101	12	90	17	85	13	3
3 Tribenuron methyl	ESI+	396>155(15)	1	69	45	68	40	58	45	0
3 Tricyclazole	ESI+	190>163(24)	0.1	89	2	70	3	88	10	3
2 Triflumizole	ESI+	346>278(12)	0.1	104	21	68	12	80	24	0
2 Triflumizole metabolite	ESI+	295>215(24)	0.1	95	9	56	8	87	8	2
2 Triforin	ESI+	435>390(12)	0.1	NC	-	45	11	54	10	0
1 Trimethacarb, 2,3,5-, 3,4,5-	ESI+	194>137(12)	0.1	95	9	51	7	86	5	2
3 Vamidothion	ESI+	288>146(14)	0.1	87	11	65	14	69	11	1
1 XMC	ESI+	180>123(12)	0.1	97	12	34	6	77	13	2



**Table 2** (cont.)

Mix Compound	Detector	Precursor>daughter ion (m/z); collision energy (eV)	LOD (g/g)	Recovery (%) and RSD (%), (n=5)			Frequency			
				Cabbage	Grapefruit	Rice				
1 Xylylcarb	ESI+	180>123(12)	0.1	93	13	67	6	83	11	2
3 1-Naphthylacetic acid	ESI-	185>141(15)	1	NC	-	NC	-	NC	-	0
3 Acequinocyl	ESI-	387>341(31)	0.1	NC	-	NC	-	129	51	0
3 Acequinocyl hydroxy	ESI-	341>313(28)	0.1	8	55	60	19	46	40	0
2 Acibenzolar acid	ESI-	179>107(20)	0.1	NC	-	37	18	11	14	0
3 Chlorfluazuron	ESI-	538>518(12)	0.1	83	20	135	18	122	7	1
3 Diuron	ESI-	231>186(18)	0.1	91	4	40	6	96	4	2
3 Fluazinam	ESI-	463>416(20)	0.1	78	9	72	14	65	6	2
3 Inabenfide	ESI-	337>122(18)	0.1	71	6	38	13	90	3	2
3 Lufenuron	ESI-	509>326(18)	0.1	77	16	67	19	111	4	2
2 Methoxyfenozide	ESI-	367>149(22)	0.1	77	7	56	8	72	2	2
3 Teflubenzuron	ESI-	379>339(13)	0.1	80	50	58	19	147	3	0
2 Tepraloxymid	ESI-	340>248(18)	0.1	22	61	52	14	61	16	0

NC = Not calculated because of low sensitivity or recovery, or matrix interference.

Frequency = Number of pesticide-commodity combinations where recoveries and RSDs within the acceptable range.

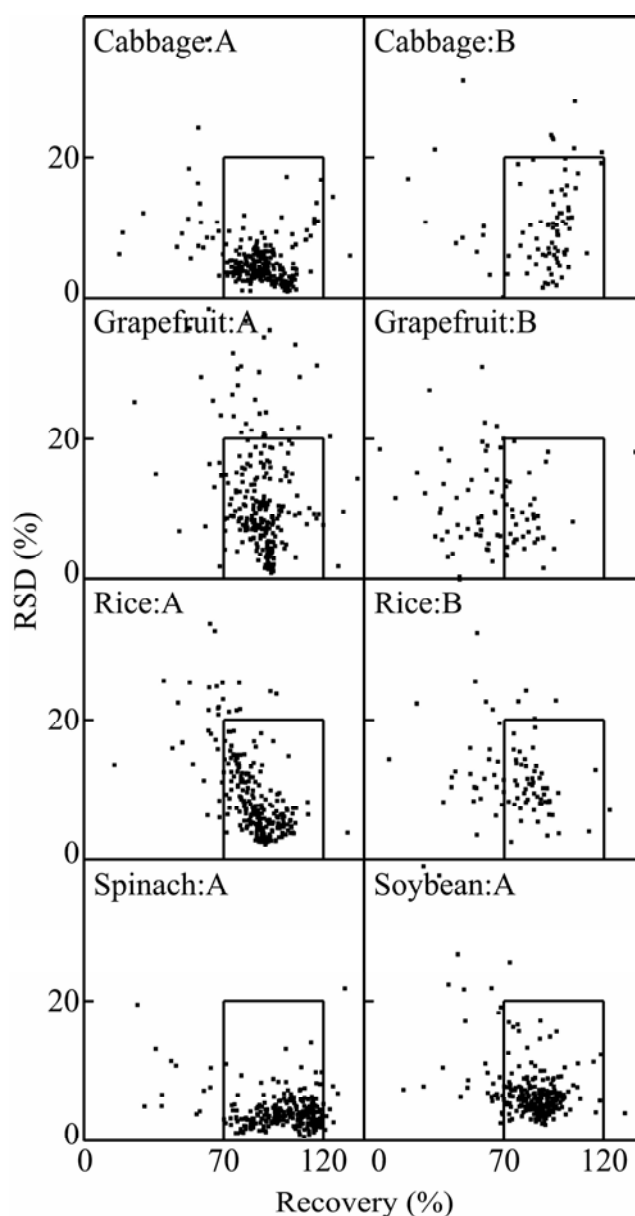
## Recovery test

The recovery tests of GC series of mixtures were conducted in quintuplicate for each sample of cabbage, grapefruit, spinach, rice and soybean at a level of 0.1 µg/g, and the recovery tests for LC/MS series mixtures were also conducted as in quintuplicate for each sample of cabbage, grapefruit and rice at a level of 0.05 µg/g. The recovery data obtained with the matrix-matched standards are listed in **Table 1** (GC series) and **Table 2** (LC/MS series).

Xylylcarb was contained in both the mixtures of GC/MS-IV and LC/MS\_PLMix-1. The results of xylylcarb with GC-MS (EI) showed good recoveries in all investigated food while the result of LC-MS/MS showed poor recovery in grapefruit. It could not be explained why LC-MS/MS showed such low recovery despite matrix-matched standards. Klein *et al.* and Pang *et al.* also reported about 30 of the compounds determined by LC-MS/MS respectively (Klein *et al.* 2003; Pang *et al.* 2006). Their results of recovery tests showed similar to our results, i.e., some of tested compounds sometimes indicated poor recoveries. Some kinds of interaction might have happened which could not be corrected with the matrix matching. Furthermore, many compounds showed low recoveries in LC-MS/MS analysis and relatively high RSDs in GC-MS (EI) analysis. The LC/MS series mixtures contain polar compounds and their recoveries might be affected by the change of pH. High RSDs in GC-MS (EI) analysis might have been caused by the high amount of co-extracts that interfered in GC-MS (EI) chromatograms. In contrast, organophosphorous pesticides measured by GC-FPD had lower RSDs. It was speculated that GC-FPD was more accurate than GC-MS.

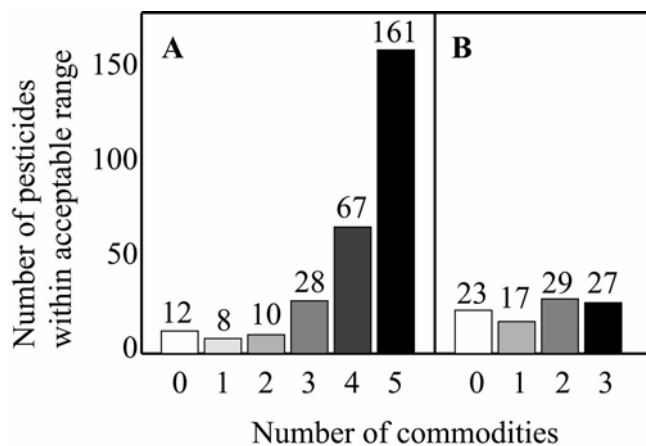
The tolylfluanid metabolite showed very high levels of recovery while its original compounds showed lower levels of recovery. It was thought that tolylfluanid decomposed through extraction and/or cleanup and/or evaporation. No recovery for thiabendazole in grapefruit was determined because the blank sample contained thiabendazole residues at levels which would mask the low-spiked pesticide. In this case, the matrix matched calibration curve had an extremely high y-intercept compared to the spiking level. Some compounds, such as benzylaminopurine and nereistoxin, could not be monitored at tested concentrations including matrix matched standards because of low sensitivity and/or interferences from matrices.

**Fig. 2** shows the results of **Table 1** and **2** in a graphic plot of recovery against RSD. A result of a compound that showed from 70 to 120% of recovery and 20% RSD or less was considered good, and this square range was regarded as an acceptable range. The frequencies of plotting within acceptable ranges were counted for all analytes. **Fig. 3** shows the number of analyte fixed with the frequency. It shows 161 compounds determined by GC and 27 compounds determined by LC-MS/MS plotted within acceptable range for all tested foods. An additional 67 + 28 com-



**Fig. 2** Plot of mean recovery against RSD of 382 pesticides and metabolites. (A) Determined by GC-MS (EI) (147), GC-MS (NCI) (80) and GC-FPD (59) spiked at 0.1 µg/g, n=5. (B) Determined by LC-MS/MS (ESI+) (84) and LC-MS/MS (ESI-) (12) spiked at 0.05 µg/g, n=5. Frames indicate the acceptable range of 70-120%/recovery and 0-20%/RSD.

pounds measured by GC and 29 compounds measured by LC-MS/MS also indicated acceptable results in the majority of tested foods (see **Table 1** and **2** for details). It was concluded that 256 compounds for GC analysis and 56 com-



**Fig. 3 Summary of recovery tests.** Each bar shows the number of pesticides giving mean recoveries and RSDs within the acceptable range. (A) Determined by GC, tested for 5 foods. (B) Determined by LC, tested for 3 foods.

pounds for LC-MS/MS analysis were applicable for screening analysis in the proposed method. Approximately 80% of initially planned pesticide/matrix combinations investigated showed good recoveries and precision.

## CONCLUSION

A rapid multiresidue method to determine more than 300 pesticide residues in food has been studied. The method is based on a simple acetonitrile extraction with SPE cleanup and determination by GC-FPD, GC-MS and LC-MS/MS. The proposed method exhibited good sensitivity and recovery and allows for rapid analysis. A single chemist could prepare from 6 homogenized samples to 6 test solutions within 4 hours. The method requires only a small volume of solvent per sample and needs no special equipments in sample preparation. It covers a wide range of pesticides, is applicable to various fruits and vegetables, and is ideally suited for use in a regulatory laboratory.

## REFERENCES

- Anastassiades M, Lehotay SJ, Štajnbaher D, Schenck FJ (2003) Fast and easy multiresidue method employing acetonitrile extraction/partitioning and "dispersive solid-phase extraction" for the determination of pesticide residues in produce. *Journal of AOAC International* **86**, 412-431
- Erney DR, Gillespie AM, Gilvydis DM, Poole CF (1993) Explanation of the matrix-induced chromatographic response enhancement of organophosphorus pesticides during open tubular column gas chromatography with splitless or hot on-column injection and flame photometric detection. *Journal of Chromatography A* **638**, 57-63
- Fillion J, Sauvé F, Selwyn J (2000) Multiresidue method for the determination of residues of 251 pesticides in fruits and vegetables by gas chromatography/mass spectrometry and liquid chromatography with fluorescence detection. *Journal of AOAC International* **83**, 698-713
- Hirahara Y, Kimura M, Inoue T, Uchikawa S, Otani S, Haganuma A, Matsumoto N, Hirata A, Maruyama S, Iizuka T, Ukuyo M, Ota M, Hirose H, Suzuki S, Uchida Y (2005) Validation of multiresidue screening methods for the determination of 186 pesticides in 11 agricultural products using gas chromatography (GC). *Journal of Health Science* **51**, 617-627
- Klein J, Alder L (2003) Applicability of gradient liquid chromatography with tandem mass spectrometry to the simultaneous screening for about 100 pesticides in crops. *Journal of AOAC International* **86**, 1015-1037
- Notification No. 497-499 (November 29, 2005) Positive List System for Agricultural Chemical Residues in Foods. Ministry of Health, Labour and Welfare, Japan
- Obana H, Akutsu K, Okihashi M, Hori S (2001) Multiresidue analysis of pesticides in vegetables and fruits using two-layered column with graphitized carbon and water absorbent polymer. *The Analyst* **126**, 1529-1534
- Okihashi M, Kitagawa Y, Akutsu K, Obana H, Tanaka Y (2005) Rapid method for the determination of 180 pesticide residues in foods by gas chromatography/mass spectrometry and flame photometric detection. *Journal of Pesticide Science* **30**, 368-377
- Pang GF, Fan CL, Liu YM, Cao YZ, Zhang JJ, Li XM, Li ZY, Wu YP, Guo TT (2006) Determination of residues of 446 pesticides in fruits and vegetables by three-cartridge solid-phase extraction-gas chromatography-mass spectrometry and liquid chromatography-tandem mass spectrometry. *Journal of AOAC International* **89**, 740-771
- Saito Y, Kodama S, Matsunaga A, Yamamoto A (2004) Multiresidue determination of pesticides in agricultural products by gas chromatography/mass spectrometry with large volume injection. *Journal of AOAC International* **87**, 1356-1367