

Determination of 105 pesticide residues by GC/MS/MS

W. S. Kim[★], J. A. Do¹, H. J. Lee, J. Y. Lee, S. J. Yang² and S.H. Lee³

Test & Analytical Laboratory, Busan Regional Food & Drug Administration, Busan 608-829, Korea

¹*National institute of Food and Drug Safety Evaluation, Korea Food & Drug Administration, Seoul, 122-704, Korea*

²*Agilent Korea, Seoul, 135-280, Korea*

³*Medical Device Safety Bureau, Korea Food & Drug Administration, Seoul, 122-704, Korea*

(Received September 7, 2009; Accepted June 10, 2010)

GC/MS/MS를 이용한 105종의 잔류농약 분석에 관한 연구

김우성[★] · 도정아¹ · 이희정 · 이주영 · 양승주² · 이선희³

부산지방식품의약품안전청 시험분석센터, ¹식품의약품안전평가원 화학물질과

²에질런트 코리아, ³식품의약품안전청 의약품안전국

(2009. 9. 7. 접수, 2010. 6. 10. 승인)

Abstract: A multi-residual method using gas chromatography coupled with mass spectrometry (GC/MS/MS) was developed for the analysis of 105 pesticides. This method was tested on lemons, beans and other vegetables. The pretreatment of these pesticides was performed by liquid-liquid partition followed by cleanup with solid phase extraction cartridge (SPE Florisil), after acetonitrile extraction from matrices and sodium chloride (15 g) addition. The recovery ranged from 71.1% to 126.0% except for azinphos-methyl, famoxadone, fenamidone, flufenoxuron and triadimefon in lemons and from 72.5% to 124.5% in bean. In lemon, the limit of detection (LOD) and limit of quantification (LOQ) were 0.001~150 ng/mL and 0.004~500 ng/mL, respectively.

요 약: 레몬과 콩 그리고 채소류에서 105개의 잔류농약을 다중 다성분 분석법으로 GC/MS/MS 분석하였다. 각 시료로부터 acetonitrile로 1차 추출하고 NaCl 15 g을 첨가한 다음 액-액 분배하였으며, Florisil 고체상(SPE) Cartridge로 정제하여 분석하였다. 레몬과 콩에서의 105개 농약의 회수율은 azinphos-methyl, famoxadone, fenamidone, flufenoxuron, triadimefon을 제외한 모든 농약에서 우수한 것으로 나타났는데 레몬에서 71.1~126.0%, 콩에서는 72.5~124.5% 였다. 또한 레몬에서의 검출한계와 정량한계는 각각 0.001~150 ng/mL와 0.004~500 ng/mL으로 확인되었다.

Key words: multi-residual method, GC/MS/MS, limit of detection, limit of quantification

★ Corresponding author

Phone : +82-(0)51-610-6140 Fax : +82-(0)51-610-6159

E-mail : kwsh1964@kfda.go.kr

1. Introduction

Agricultural chemicals synthesized to avoid damage due to blight and harmful insects are used to ensure the safety of agricultural products. However, with their beneficial characteristics, they also have adverse effects on the human body and environment. Public interest in this problem is high, as people are concerned about the reported effects of pesticides on

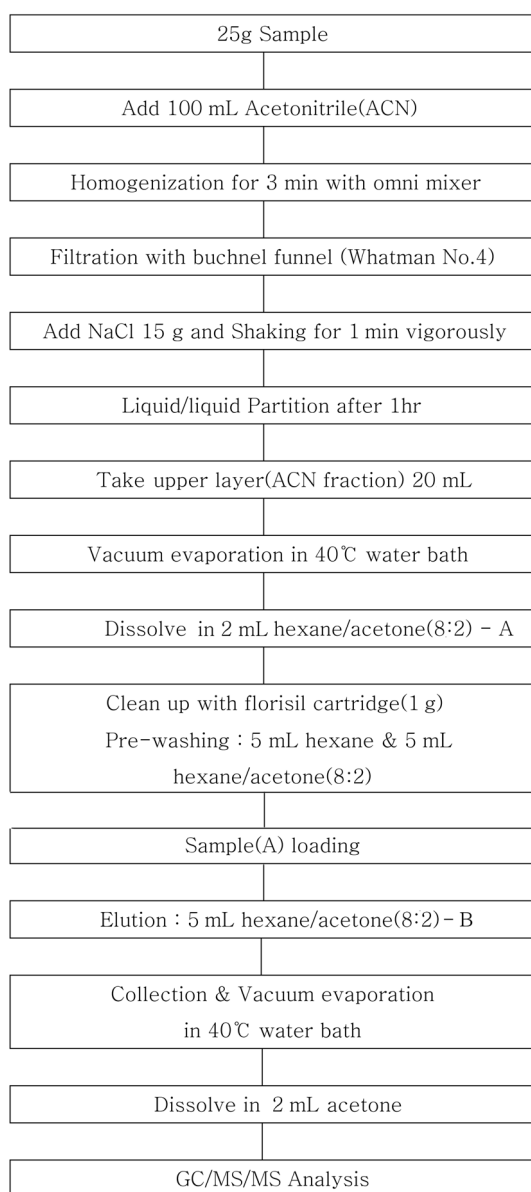


Fig. 1. Flow diagram of sample preparation procedure.

their own bodies.

The pesticides that remain in most agricultural products decrease with time and through the pre-treatment process; however, safety problems may occur due to the pesticides remaining in fruits and vegetables. Many procedures for extraction of these pesticides require extensive sample preparation, multiple extractions, extract purification and concentration before chromatographic analysis.¹⁻⁴ In addition, various instruments are used to determine whether the trace amounts of pesticides in foodstuffs comply with the level required by legislation regulation. In recent years, some applications using GC, HPLC and mass selective detectors (GC/MS) have been described as rapid and reliable multi-residual methods for determining pesticide levels.⁵⁻⁷ Mass spectrometry (MS) has become very popular in laboratories for detecting pesticides. Unfortunately, a full scan often does not provide enough sensitivity in real samples, but SIM (Selected Ion Monitoring), which improves sensitivity, considerably reduces the qualitative information, thus increasing the risk of false positives.⁸ The combination of GC with tandem mass spectrometry (GC/MS/MS) has been shown to be applicable to the analysis of trace amounts of contaminants like pesticides or metabolites in complex samples such as biological fluids,⁹⁻¹¹ water¹²⁻¹³ or fruits and vegetables.¹⁴⁻¹⁵ This paper proposes a new method to determine the levels of 105 pesticides in beans and lemon. It is based on fast and simple acetonitrile extraction of the samples with SPE florisil clean up (Fig. 1) and GC/MS/MS analysis.

2. Experimental

2.1. Reagents and standards

Pesticide standards of analytical grade were purchased from Dr. Ehrenstorfer GmbH (Augsberg, Germany), Chem Service (West Chester, USA), Supelco (Bellefonte, USA), Riedel de Haen (Schnellendorf, Germany) or Wako (Japan). Individual stock solutions were prepared by dissolving 5~10 mg of each compound in 10 mL acetone. A mixed solution with approximate concentration of 10 $\mu\text{g/mL}$ in

acetone was also prepared. Working standard solutions were prepared by diluting this mixed solution. Residue analysis grade acetonitrile and anhydrous sodium sulfate were purchased from Merck (Damstadt, Germany). Additionally, sodium chloride (Junsei) and Whatman filter paper, bond elute (MEGA BE-FL 1 g/6 mL, Varian) were used for cleanup.

2.2. Instrumental analysis

A Varian (Walnut Creek, CA, USA) CP-3800 gas chromatograph equipped with a triple Quadrupole mass spectrometer was used. Splitless injection was performed with purge delay time 1.5 min (Purge flow 100 mL/min). A Varian (5% phenyl-95% methyl

silicone) fused-silica column (30 m × 0.25 mm × 0.25 μm film thickness) was used. Other GC/MS/MS analysis conditions are as follows; helium carrier gas flow, 0.8 mL/min; injector temperature, 250 °C; injection volume, 1 μL; ion source temperature, 200 °C; solvent delay time, 4 min; oven temperature condition: 70 °C for 3 min, then 20 °C/min to 180 °C (holding for 8.5 min) and 5 °C/min to 300 °C, held for 7.5 min. The total run time was 40 min. The common parameters for source, manifold and transfer line temperatures were 200 °C, 40 °C and 250 °C, respectively. The optimization of collision-induced dissociation (CID) parameters was carried out in order to generate MS/MS spectra with a

Table 1. Instrumental analysis conditions of the 105 pesticides

No.	Seg	Pesticides	Retention time	M.W.	Precursor ion	MRM1	MRM2	MRM3	Collision energy (eV)
1	1	Propoxur	7.181	209	110	64	92		15
2	2	Dichlobenil	8.577	171	171	100	136		20
3	2	Hexaflumuron	9.013	461	202	110(30)	174(15)	146(20)	
4	2	Lufenuron	9.010	511	203	140(20)	111(30)	175(15)	
5	2	Mevinphos	9.092	224	192	127	164		10
6	2	Nitrapyrin	9.401	229	194	133	158		20
7	3	Tecnazene	10.742	259	203	85	143		25
8	4	BHC-alpha	11.965	288	181	145	109		25
9	4	Chlorpropham	11.301	213	213	171	127		5
10	4	Dicloran	12.241	206	206	148	176		20
11	4	Diphenylamine	11.076	169	169	169	167		10
12	4	Ethalfuralin	11.108	333	276	248	202		10
13	4	Ethoprophos	11.073	242	158	114	97		20
14	4	Simazine	12.282	201	201	173	186		10
15	4	Thiometon	12.051	246	125	79	93		20
16	4	Trifluralin	11.245	335	306	206	264		20
17	5	BHC-beta	12.537	288	181	109	145		30
18	5	BHC-delta	13.434	288	181	145	109		10
19	5	BHC-gamma	12.740	288	181	145	109		10
20	5	Diazinon	12.772	304	304	179	137		30
21	5	Etrimfos	13.220	292	181	153	98		5
22	5	Flufenoxuron(2)	13.424	489	126	98(15)	106(10)		
23	5	Fonofos	12.873	246	246	109(20)	137(10)		
24	5	Isazofos	13.126	313	161	119	146		10
25	5	Quintozene	12.622	293	237	143	119		20
26	5	Tebupirimfos	13.384	318	261	137	153		15
27	5	Tefluthrin	13.067	419	177	127	157		5
28	6	Acetochlor	14.060	269	223	132	147		15
29	6	Alachlor	14.317	269	188	131	160		15
30	6	Chlorpyrifos-methyl	14.136	321	286	93	271		20

Table 1. Continued

No.	Seg	Pesticides	Retention time	M.W.	Precursor ion	MRM1	MRM2	MRM3	Collision energy (eV)
31	6	Dimethenamid	13.990	276	230	154	137		20
32	6	Dithiopyr	14.570	401	286	210	238		15
33	6	Formothion	13.742	257	170	93(15)	125>79(10)		
34	6	Heptachlor	14.615	370	272	237	239		20
35	6	Metalaxyl	14.502	279	206	132	162		10
36	6	Parathion-methyl	14.326	263	263	109(10)	127(20)	246(5)	
37	6	Propanil	14.034	217	161	99	126		30
38	6	Propisochlor	14.435	284	162	119	144		20
39	6	Tolclofos-methyl	14.340	300	265	250	93		20
40	6	Vinclozolin	14.215	285	285	213	186		10
41	7	Chlorpyrifos	15.434	349	314	258	285		15
42	7	Dichlofluanid	15.236	332	224	123	77		25
43	7	Diethofencarb	15.426	267	225	196	168		5
44	7	Dimethylvinphos	15.513	332	295	109	280		20
45	7	Esprocarb	15.232	265	222	162	151		5
46	7	Fenitrothion : MEP	14.997	277	277	260	109		10
47	7	Fosthiazate(2)	16.187	283	195	103	139		10
48	7	Malathion	15.195	330	173	99	127		10
49	7	Methyl pentachlorophenyl sulfide	15.241	296	296	246	263		30
50	7	Metolachlor	15.407	283	162	133	134		10
51	7	Pirimiphos-ethyl	16.014	333	333	180	163		10
52	7	Pirimiphos-methyl	14.864	305	305	180	233		15
53	7	Tetraconazole	15.721	372	336	218	191		15
54	7	Thiazopyr	15.239	396	128	128(5)	85(25)		
55	7	Triadimefon	15.765	293	208	111(30)	181(15)		
56	8	Chlorfenvinphos	16.744	358	267	159	203		15
57	8	Heptachlor-epoxide	16.818	386	353	263	282		15
58	8	Isofenphos	16.667	345	213	121	185		10
59	8	Mecarbam	16.811	329	131	74	86		15
60	8	Penconazole	16.659	283	248	157(30)	248		5
61	8	Phenthoate : PAP	16.920	320	274	121	246		10
62	8	Procymidone	17.088	283	283	96	283		10
63	8	Tolyfluanid	16.754	346	238	91	137		35
64	9	o,p-DDE	17.593	316	246	176	246		30
65	9	Paclbutrazol	17.681	293	236	125	167		10
66	10	Carboxin	18.855	235	143	114	143	87	10
67	10	Chinomethionat	18.797	234	206	148	121		25
68	10	Dieldrin	18.880	378	263	193(30)	228(20)		
69	10	Endosulfan-alpha	17.986	404	195	125	159		25
70	10	Myclobutanil	18.749	288	179	125	152		10
71	10	Napropamide	18.149	271	271	72	128		10
72	10	o,p-DDD	18.883	318	235	199	165		20
73	10	p,p-DDE	18.634	316	246	176	211		25
74	10	Prothiofos	18.323	344	267	221	239		20
75	10	Thifluzamide	18.597	528	194	125	166		20
76	11	Chlorobenzilate	19.698	324	251	111	139		30
77	11	Endosulfan-beta	19.919	404	241	170	206		20

Table 1. Continued

No.	Seg	Pesticides	Retention time	M.W.	Precursor ion	MRM1	MRM2	MRM3	Collision energy (eV)
78	11	Endrin	19.576	378	263	193	228		30
79	11	Fenoxanil	19.370	329	189	125	154		15
80	11	Fluacrypyrim	20.186	426	145	115	102		15
81	12	Carbophenothion	20.893	342	342	157(10)	296(5)		
82	12	Edifenphos	21.012	310	173	109	155		10
83	12	Norflurazon	20.965	303	145	95	125		15
84	12	p,p-DDT	21.295	352	235	165	199		20
85	12	Triazophos	20.509	313	161	106	92		15
86	13	Azinphos-methyl	24.292	317	160	132	104		5
87	13	Bifenthrin	22.868	422	181	166	167		15
88	13	Bromopropylate	22.985	426	341	183	185		10
89	13	EPN	22.967	323	157	77	110		15
90	13	Etoazole	23.179	359	141	112(20)	113(15)	141(20)	
91	13	Fenamidone	23.326	311	238	103	238		30
92	13	Fenoxycarb	23.083	301	186	109	157		15
93	13	Fenpropathrin	23.235	349	265	210(10)	181(20)		
94	13	Indanofan	23.482	341	139	75	111		30
95	13	Phosalone	24.111	367	182	111(25)	138(10)	102(25)	
96	13	Phosmet (PMP) : Imidan	22.891	317	160	132	104		15
97	13	Tetradifon	23.939	354	356	159	229		20
98	14	Fenarimol	25.149	330	251	139	111		25
99	14	Fluquinconazole	26.571	376	340	286	298		25
100	14	Pyrazophos	25.085	373	221	149	193		15
101	15	Cypermethrin(3)	27.964	415	181	152	127		30
102	15	Famoxadone	31.782	374	196	167	153		15
103	15	Flucythrinate	28.242	451	199	107	157		15
104	15	Halfenprox	28.030	477	265	265(10)	131(25)	237(15)	
105	15	Indoxacarb	30.821	528	150	123	114		25

relative abundance of the parent ion, and CID gas pressure was 1.5 m Torr. In EI mode (Ionization energy : 70 eV), the filament current was set to 50 μ A. The MRM parameters are summarized in Table 1.¹⁴ MRM 1 was used for quantification, and the intensity ratio of MRM 1, MRM 2 and MRM 3 was used as the confirmatory parameter.

2.3. Sample preparation

The extraction method was proposed by KFDA (Food Code No. 83).¹⁵ It is summarized in Fig. 1. The sample was dissolved in 2 mL (A) of hexane/acetone (8:2) and loaded on Bond elute (florisil, 1 g/6 mL), and then eluted with hexane/acetone (8:2) 5 mL (B), which was previously conditioned with 5

mL of hexane, and 5 mL of hexane/acetone (8:2). 7 mL of hexane/acetone (8:2) (A+B=C) were combined. C solutions were dried on a rotary evaporator. An Omni Macro Homogenizer with high-speed was used.

3. Results and Discussion

3.1. Optimization of MS/MS experimental conditions

The GC/MS/MS measurement method is usually composed of some groups of MRM ions by the system software from Varian instrumental equipment (GC/MS/MS). That is time segment of start and end times of each function can be defined as retention

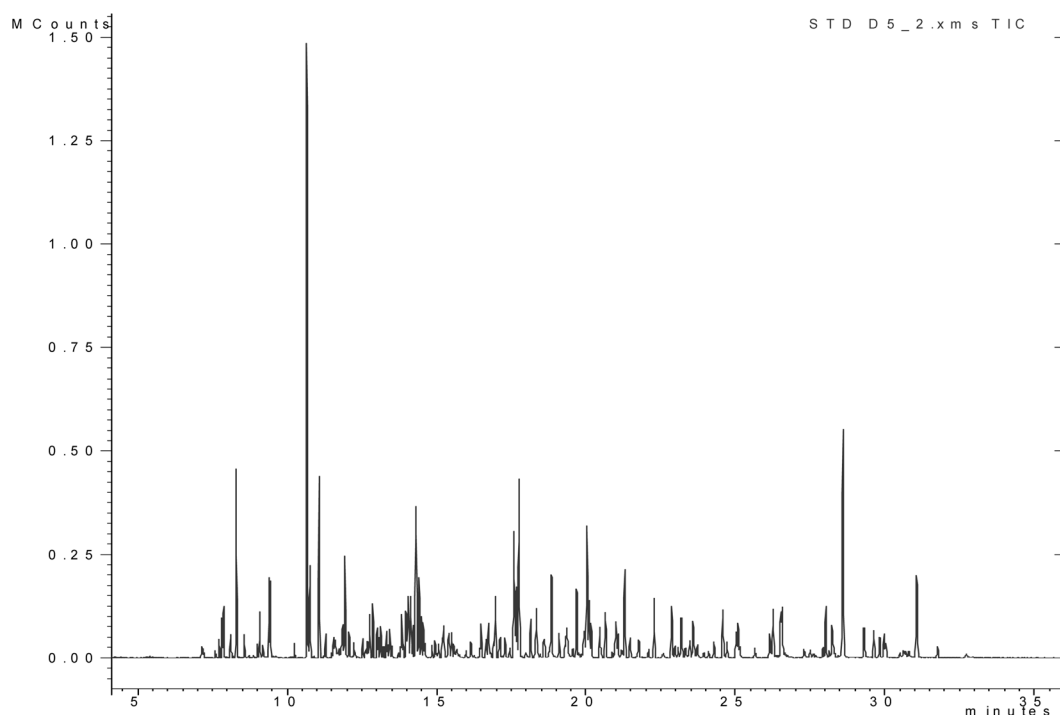


Fig. 2. Typical chromatogram of a standard mixture solution including 105 pesticides.

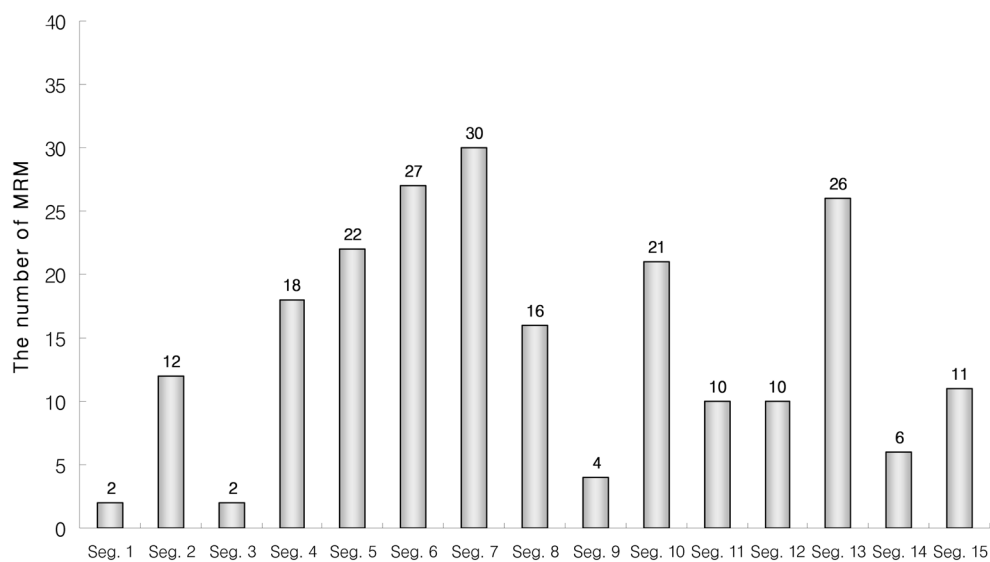


Fig. 3. MRM distribution of the 15 acquisition groups.

times of individual peaks. A measurement method can be constructed with 1 to 32 functions that can allow 40 transition ions to be monitored simultaneously (same segment time) without significant loss of sensitivity (although the maximum number of

transition ions is 200). In practical terms, the chromatographic peak shape, which depends on scan speed, restricts the number of simultaneous analyses. Following conditions from a previous paper (Fernandez Moreno *et al.* 2008), we established a rapid and

Table 2. Recovery and validation results of 105 pesticides in lemon and bean (n=3)

No.	Pesticide	Treatment conc.(ppm)	Bean		Lemon		Bean		Lemon	
			Recovery (%)	RSD (%)	Recovery (%)	RSD (%)	ng/g	ng/g	ng/g	ng/g
1	Acetochlor	1.20	88.6	4.1	108.7	3.0	0.007	0.023	0.005	0.017
2	Alachlor	0.53	89.1	4.6	108.8	2.3	0.012	0.039	0.456	1.521
3	Azinphos-methyl	0.53	77.9	6.2	67.5	6.3	4.660	15.534	9.430	31.434
4	BHC-alpha	0.27	86.8	2.4	103.8	4.8	0.003	0.010	0.002	0.008
5	BHC-beta	0.40	83.5	3.1	109.0	3.9	0.010	0.034	0.480	1.599
6	BHC-delta	0.40	78.2	3.8	102.2	4.5	0.013	0.043	0.616	2.055
7	BHC-gamma	0.27	82.2	3.5	101.8	2.3	0.010	0.035	0.600	2.002
8	Bifenthrin	0.13	85.0	6.2	99.0	1.5	0.158	0.525	0.213	0.711
9	Bromopropylate	0.27	112.5	6.9	129.0	2.7	0.005	0.016	0.004	0.012
10	Carbophenothion	0.40	98.5	3.0	117.7	1.8	0.012	0.039	0.008	0.027
11	Carboxin	0.13	72.5	2.3	85.0	1.7	15.385	51.282	7.792	25.974
12	Chionomethionat	0.67	79.7	3.7	72.8	1.4	0.087	0.288	10.830	36.101
13	Chlorfenvinphos	0.53	93.5	4.3	106.6	1.8	0.006	0.019	0.005	0.015
14	Chlorobenzilate	0.40	108.8	5.1	127.7	2.0	0.001	0.005	0.001	0.004
15	Chlorphopham	0.80	90.8	6.5	119.6	4.0	0.054	0.180	0.028	0.093
16	Chlorpyrifos	0.40	89.5	3.5	113.5	2.2	0.005	0.016	0.003	0.011
17	Chlorpyrifos-methyl	1.33	86.1	4.4	113.7	2.1	0.004	0.013	0.148	0.494
18	Cypermethrin	1.07	106.9	3.9	124.9	0.5	7.565	25.217	4.272	14.241
19	Diazinon	2.00	92.4	2.3	109.6	2.5	0.009	0.031	0.007	0.023
20	Dichlobenil	0.13	81.0	2.6	101.5	2.3	0.001	0.002	0.001	0.003
21	Dichlofluanid	0.53	84.9	6.6	104.5	5.1	0.007	0.024	0.016	0.053
22	Dichloran	0.67	100.6	6.4	128.4	4.0	0.014	0.046	0.013	0.043
23	Dieldrin	0.67	79.8	0.0	96.7	1.2	0.026	0.085	0.019	0.064
24	Diethofencarb	0.40	88.8	4.0	93.2	3.3	0.249	0.829	0.187	0.623
25	Dimethenamid	2.00	89.1	3.9	105.3	2.4	0.005	0.015	0.004	0.012
26	Dimethylvinphos	0.67	86.9	5.0	94.4	3.2	0.004	0.012	0.003	0.012
27	Diphenylamine	0.13	97.5	4.0	110.5	2.3	0.025	0.084	0.037	0.123
28	Dithiopyr	2.00	85.9	3.0	104.4	3.1	0.007	0.024	0.006	0.019
29	Edifenphos	0.27	102.3	3.0	111.0	1.3	0.003	0.009	0.003	0.011
30	Endosulfan-alpha	0.67	82.3	2.2	95.4	3.9	0.059	0.197	0.044	0.147
31	Endosulfan-beta	0.67	82.2	2.9	93.5	3.0	0.086	0.285	0.057	0.191
32	Endrin	0.67	92.3	3.3	108.1	2.0	0.020	0.068	0.015	0.050
33	EPN	0.27	93.5	6.4	111.3	1.6	0.030	0.100	0.773	2.576
34	Esprocarb	0.53	85.1	2.9	101.0	4.8	0.182	0.606	0.075	0.249
35	Ethalfuralin	0.40	86.5	6.5	116.7	2.0	0.019	0.064	0.010	0.034
36	Ethoprophos	0.27	96.5	5.0	88.5	0.8	0.288	0.962	0.959	3.197
37	Etoxazole	0.27	104.3	3.8	115.3	1.6	200.000	666.667	4.196	13.986
38	Etrimfos	1.33	87.4	6.7	113.2	2.5	0.101	0.337	0.060	0.201
39	Famoxadone	1.47	88.6	4.5	49.9	1.2	1.974	6.580	2.510	8.368
40	Fenamidone	0.67	122.0	3.3	41.1	0.3	0.013	0.043	0.013	0.045
41	Fenarimol	0.40	100.0	3.5	96.7	2.8	0.010	0.032	0.012	0.041
42	Fenitrothion	0.27	85.5	4.5	109.8	3.5	0.015	0.051	0.011	0.035
43	Fenoxanil	0.40	91.2	4.0	100.7	2.6	0.035	0.115	0.049	0.165
44	Fenoxycarb	1.47	105.8	3.0	113.3	2.6	0.071	0.237	3.766	12.553
45	Fenpropathrin	0.80	86.4	3.3	115.1	0.9	0.025	0.085	0.019	0.063
46	Fluacrypyrim	0.53	89.6	4.5	104.9	1.3	1.443	4.811	1.612	5.373

Table 2. Continued

No.	Pesticide	Treatment conc.(ppm)	Bean		Lemon		Bean		Lemon	
			Recovery (%)	RSD (%)	Recovery (%)	RSD (%)	ng/g	ng/g	ng/g	ng/g
47	Flucythrinate	0.53	107.3	5.7	115.8	0.8	0.017	0.058	0.369	1.231
48	Flufenoxuron	2.67	92.5	3.1	66.5	1.5	0.015	0.049	0.024	0.081
49	Fluquinconazole	0.53	109.6	4.3	93.0	4.1	0.003	0.010	0.004	0.013
50	Fonofos	0.67	92.0	4.3	116.9	2.3	0.002	0.006	0.001	0.004
51	Formothion	0.27	85.5	4.6	88.5	1.5	1.599	5.330	1.548	5.161
52	Fosthiazate	0.53	83.5	3.9	80.0	2.8	0.053	0.178	0.059	0.197
53	Halfenprox	0.53	109.4	2.8	128.0	1.2	0.535	1.783	0.623	2.076
54	Heptachlor	0.40	88.3	4.4	112.2	3.2	0.003	0.010	0.003	0.008
55	Heptachlor-epoxide	0.27	76.8	3.0	95.0	4.3	0.019	0.064	0.012	0.041
56	Hexaflumuron	0.67	107.6	13.1	130.4	5.7	0.009	0.030	0.304	1.013
57	Indanofan	0.67	95.3	5.9	113.4	14.8	181.818	606.061	1.255	4.183
58	Indoxacarb	1.07	101.0	3.9	116.8	11.5	1.043	3.477	1.186	3.954
59	Isazofos	0.27	85.5	3.3	78.3	1.6	0.010	0.035	0.621	2.070
60	Isofenphos	0.27	93.8	6.9	117.5	1.1	0.005	0.016	0.004	0.013
61	Lufenuron	0.67	106.1	11.6	128.1	2.4	0.037	0.123	0.175	0.583
62	Malathion	0.27	92.5	9.9	104.8	0.9	0.008	0.027	0.007	0.023
63	Mecarbam	0.40	85.8	5.3	102.7	4.5	2.431	8.103	3.377	11.257
64	Metalaxyl	2.67	77.7	3.9	71.1	0.8	0.028	0.092	0.152	0.508
65	Methyl pentachlorophenyl sulfide	0.67	87.7	3.5	104.7	2.6	0.007	0.022	0.005	0.017
66	Metolachlor	0.27	92.3	3.3	84.3	4.1	0.520	1.734	0.894	2.978
67	Mevinphos	0.67	88.8	4.8	82.9	1.8	0.004	0.013	0.035	0.116
68	Myclobutanil	0.27	89.8	2.4	89.0	2.3	0.015	0.051	0.647	2.157
69	Napropamide	0.80	102.3	4.8	104.6	3.3	0.009	0.031	0.009	0.029
70	Nitrapyrin	0.67	88.8	2.9	110.9	2.0	0.001	0.004	0.069	0.228
71	Norflurazon	0.40	92.2	2.8	84.7	0.3	0.020	0.065	0.018	0.061
72	o,p-DDD	0.13	99.0	5.4	107.0	10.0	0.001	0.005	0.001	0.004
73	o,p-DDE	0.27	98.5	3.5	121.5	1.5	0.001	0.005	0.002	0.005
74	p,p-DDE	0.13	95.5	3.8	116.5	1.7	0.003	0.011	0.006	0.021
75	p,p-DDT	0.53	99.3	3.2	123.8	0.8	0.002	0.006	0.001	0.004
76	Paclotbutrazol	0.27	109.3	9.6	95.3	3.0	0.004	0.013	0.010	0.033
77	Parathion-methyl	2.00	91.7	5.7	123.0	4.0	0.010	0.034	0.007	0.024
78	Penconazole	0.13	87.5	4.6	88.0	2.3	0.006	0.021	0.015	0.052
79	Phenthoate	0.40	88.8	5.5	112.2	1.4	0.006	0.019	0.004	0.014
80	Phosalone	0.27	100.0	5.0	117.0	0.8	0.028	0.094	0.012	0.041
81	Phosmet	0.27	101.8	3.7	108.5	1.7	0.986	3.285	0.924	3.079
82	Pirimiphos-ethyl	0.27	90.0	0.8	91.3	3.0	0.012	0.040	0.014	0.047
83	Pirimiphos-methyl	0.27	88.0	7.5	81.4	2.0	0.005	0.017	0.005	0.016
84	Procymidon	0.27	91.3	3.7	102.8	1.5	0.912	3.041	1.123	3.742
85	Propanil	0.93	91.5	4.1	103.6	1.7	0.013	0.044	0.013	0.044
86	Propisochlor	0.80	80.3	2.2	98.6	2.9	0.022	0.072	1.862	6.208
87	Propoxur	0.13	96.5	5.7	103.5	2.6	0.073	0.244	0.263	0.875
88	Prothiofos	0.40	88.3	13.1	117.2	19.7	0.009	0.031	0.422	1.406
89	Pyrazophos	0.67	108.6	4.2	125.6	0.6	0.006	0.020	0.136	0.453
90	Quintozene	0.40	86.8	4.4	114.8	0.8	0.021	0.071	0.012	0.039
91	Simazine	0.53	86.6	3.6	65.9	0.9	0.037	0.123	0.122	0.405
92	Tebupirimfos	0.40	79.2	2.0	119.7	6.3	0.017	0.056	0.010	0.034

Table 2. Continued

No.	Pesticide	Treatment conc.(ppm)	Bean		Lemon		Bean		Lemon	
			Recovery (%)	RSD (%)	Recovery (%)	RSD (%)	ng/g	ng/g	ng/g	ng/g
93	Tecnazene	0.27	83.3	2.6	103.5	1.5	0.014	0.047	0.008	0.028
94	Tefluthrin	0.67	89.5	1.1	112.7	2.3	0.026	0.085	0.017	0.056
95	Tetraconazole	0.40	85.0	5.1	83.0	7.1	0.017	0.057	0.027	0.089
96	Tetradifon	0.53	96.8	3.0	111.3	1.6	0.017	0.056	0.012	0.041
97	Thiazopyr	0.40	113.8	12.3	72.5	3.5	116.129	387.097	150.000	500.000
98	Thiifluzamide	0.27	104.8	4.4	113.0	2.8	0.204	0.680	0.200	0.666
99	Thiometon	0.53	80.8	0.8	98.6	3.6	0.266	0.888	7.373	24.578
100	Tolclofos-methyl	0.40	87.5	4.1	112.5	1.7	0.006	0.019	0.168	0.560
101	Tolyfluamid	1.07	85.8	4.2	110.7	3.0	0.008	0.028	0.006	0.021
102	Triadimefon	0.40	87.7	5.1	58.0	50.2	0.034	0.113	0.139	0.464
103	Triazophos	0.27	108.3	5.7	107.5	3.4	0.981	3.271	1.647	5.491
104	Trifluralin	0.67	90.7	5.7	125.3	2.3	0.019	0.063	0.010	0.033
105	Vinclozolin	2.67	87.5	2.9	107.1	2.7	0.012	0.040	0.010	0.032

reliable multi-residual method to determine the levels of 105 pesticides. In this study, we used the MRM parameters previously proposed. A typical chromatogram of mixture standards is shown in Fig. 2. For the MS, sensitivity was maximized by means of optimizing the amount of target ions. The parent ion was selected for each pesticide considering its *m/z* and relative abundance in order to improve sensitivity and selectivity. To obtain the sensitive peak of each pesticide, all pesticides were sorted by their retention times and divided into 15 groups. Acquisition was conducted in 15 acquisition groups according to Fig. 3. The dwell time of all MRM ions was 0.15~0.29 sec. In this method, the scan speed of MRM ions ranged from 0.2 to 0.82 sec/scan.

3.2. Validation of the method

Suggested method was validated for bean and lemon in order to assess its applicability to the analysis of real samples. The identification of target pesticides was carried out by searching in the appropriate retention time windows (retention time \pm 0.1 min). The retention time data is shown in Table 1. The limits of detection (LOD : $S/N > 3$) and quantification (LOQ : $S/N > 10$) were evaluated. In lemons, the LOD and LOQ were 0.001~150 ng/mL and 0.004~500 ng/mL, respectively. In case of bean sample,

the LOD was 0.001~116 ng/mL and LOQ was 0.005~388 ng/mL. Three uncontaminated samples of bean and lemon were spiked with the pesticides at each fortification level. Average recovery data and relative standard deviations (RSD) are shown in Table 2. The recovery of most pesticides were between 71.1~126.0% except for azinphos-methyl, famoxadone, fenamidone, flufenoxuron and triadimefon in lemons and 72.5~124.5% in beans, and RSD was lower than 15% in all cases. In samples used recovery test, any pesticides were not detected. The linearity of the analytical response across the studied range is excellent, with correlation coefficients higher than 0.99 in most cases, except for certain pesticides (fenoxycarb, indoxacarb, famoxadone, propisochlor and propanil).

3.3. Application of the method to real samples

For three years (2005~2007), the proposed method was applied to routine analysis of approximately 2,800 agricultural products in author's laboratory (KFDA, Busan, Korea). Table 3 summarizes the results found that were above the limits permitted by the legislation of Korea. A very large number of the analyzed samples (pepper leaves, *chamnamul*, chard, perilla leaves, *shinsuncho*, red mustard, welsh onion, *chwinamul*, crown daisy, radish leaves, marsh mallow

Table 3. Detection frequency of pesticides residue in agricultural products

No.	Pesticide	No. of frequency detected	No. over MRLs Korea
1	Bifenthrin	14	2
2	Chlorpyrifos	9	4
3	Cypermethrin	25	2
4	Diazinon	8	2
5	Endosulfan	18	1
6	EPN	1	1
7	Ethoprophos	8	3
8	Flufenoxuron	16	2
9	Isazofos	1	1
10	Metalaxyl	8	1
11	Procymidon	63	3
12	Vinclozolin	1	1
	Total	172	23

and carrot) exceeded the MRLs (Maximum Residual Levels) of Korea. The pesticides most frequently found at levels over Korea's MRLs were lufenuron, kresoxim-methyl, chlorpyrifos, procymidone, ethoprophos, diazinone, cypermethrin and metalaxyl. Other agricultural products analyzed presented less positive results.

4. Conclusions

A simultaneous multi-residual method was developed for rapid and precise confirmation of the levels of 105 pesticides using GC/MS/MS. The results obtained showed a good analytical performance of sensitivity and selectivity of the suggested method. The use of the MRM parameters supports the analytes identified. For all pesticides, the sensitivity of this method was high enough to ensure a reliable determination at levels lower than the respective MRL.

References

1. F. J. Egea Gonzalez, A. Mena Granero, C. R. Glass, A. Garrido Frenich and J. L. Martinez Vidal, *Rapid Communications in mass Spectrometry*, **18**, 537-543(2004).
2. S. K. Cho, A. M. Abd El-Aty, Y. S. Park, J. H. Choi, S. Khay, C. A. Kang, B. J. Park, S. J. Kim and J. H. Shim, *Biomed. Chromatogr.* **21**, 602-609(2007).
3. J. Boer and R. J. Law, *J. Chromatogr. A.*, **1000**, 223-251(2003).
4. G. F. Pang, Y. M. Liu, C. L. Fan, J. J. Zhang, Y. Z. Cao, X. M. Li, Z. Y. Li, Y. P. Wu and T. T. Guo, *Anal. Bioanal. Chem.*, **384**, 1366-1408(2006).
5. Z. Lv, L. Gao, H. Gao, Z. Hou and B. Zhang, *J. Food Sci.*, **74**, 37-41(2009).
6. X. G. Chu, X. Z. Hu and H. Y. Yaho, *J. Chromatogr. A.*, **1063**, 201-210(2005).
7. P. Sandra, B. Tienpont and F. David, *J. Chromatogr. A.*, **1000**, 299-309(2003).
8. F. J. Arrebola, J. L. Martinez Vidal, A. Fernandez Gutierrez and M. A. Akhtar, *Anal. Chim. Acta.*, **401**, 45(1999).
9. E. Pitarch, R. Serrano, F. J. Lopez, F. Hernandez, *Anal. Bioanal. Chem.*, **376**, 189-197(2003).
10. E. Jaquerdeo, J. E. Schaff, M. A. Montgomery, M. A. Lebeau, *Rapid Commun. Mass Spectrum.*, **23**, 2697-2705(2009).
11. D. Tsikas, F. M. Gutaki, F. M., J. Sandmann, E. Schwedhelm and J. C. Frolich, *J. Chromatogr. B. Biomed. Sci. Appl.*, **731**, 285-291(1999).
12. N. Sauret-Szczepanski, P. Mirabel, H. Wortham, *Environ. Pollut.* **139**, 133-142(2006).
13. J. L. Martinez Vidal, M. C. Pablos-Espada, A. Garrido Frenich and F. J. Arrebola, *J. Chromatogr. A.*, **867**, 235-245(2000).
14. J. L. Fernandez Moreno, A. Garrido Frenich, P. Plaza Bolanos and J. L. Martinez Vidal, *J. Mass Spectrom.*, **43**, 1235-1254(2008).
15. M. Gamon, C. Lleo and A. Ten, F. Mocholi, *J. AOAC Int.*, **84**, 1209-1216(2001).