JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Article

pubs.acs.org/JAFC

Rapid and Simple Analysis of Pesticides Persisting on Green Pepper Surfaces Swabbing with Solvent-Moistened Cotton

Eiki Watanabe,* Yuso Kobara, and Yasuhiro Yogo

National Institute for Agro-Environmental Sciences, Tsukuba, Ibaraki 305-8604, Japan

ABSTRACT: A rapid and simple nondestructive extraction (NDE) method that includes wiping off of systemic neonicotinoid insecticides has been developed to streamline sample pretreatment procedures conducted before chromatographic determination. Pesticide residues were extracted from green pepper surfaces by swabbing them with absorbent cotton moistened with acetone or acetonitrile. After spraying of pesticides, the extraction rate decreased gradually, except for thiacloprid. Presumably, extraction rates depend on the physicochemical properties of pesticides, especially water solubility. It was thought that the applicability of the proposed method greatly depended on the systemic speed of each pesticide, and water solubility was placed as the index that was important to making certain. Direct analysis of some insecticides persisting on sample surfaces has been possible only by extraction before chromatographic determination. These findings indicate strongly that the proposed NDE method has collateral conditions, but it appears promising for on-site pretreatment for pesticide residue analysis.

KEYWORDS: *neonicotinoid, systemic pesticide, nondestructive extraction, wiping off*

INTRODUCTION

Pesticide residues are present in foods as a result of their application to agricultural products to prevent losses from weeds, insects, and plant pathogens. Various multiresidue determinations have been developed to analyze agricultural samples for these agrochemicals. A salient disadvantage associated with many of these methods is their requirement for troublesome multistage sample pretreatment procedures and large quantities of hazardous solvents.[1−3] In recent years, high-performance liquid chromatography coupled with mass spectrometry (LC-MS or LC-MS/MS) has become an important tool for highly sensitive quantitative analysis of pesticide residues in various matrices in relation to food safety. Sample pretreatments composed of extraction and cleanup processes are absolutely necessary for accurate determination of pesticide residues in foods consisting of complicated matrices. However, despite rigorous sample pretreatments and practical use of highly accurate analytical instruments such as LC-MS and GC-MS, one is often confronted with a major problem in pesticide residue analyses: compound and matrix-dependent response suppression or enhancement might occur as the so-called matrix effect (or matrix interference), as reported in several reviews.[4,5]

Pesticide residues are distributed heterogeneously on or in plants. For that reason, a sample must be homogenized thoroughly. A sufficiently homogeneous sample is extracted, with the result that matrix components are often coextracted together with the target pesticides. Consequently, cleanup processes are necessary, as described above. However, several reports describe pesticide residue analyses based on nondestructive extraction (NDE) methods. Saranwong and Kawano[6] developed a rapid analytical method for a fungicide, dichlofluanid, persisting on tomato surfaces based on nondestructive washing off with acetone and determination using near-infrared spectroscopy. Edison et al.[7,8] proposed a rapid screening method for several pesticide residues on fruit surfaces. Concretely, swabs were used to extract various pesticides. Then the swabs were analyzed using direct analysis in real-time ambient pressure desorption ionization coupled to a high-resolution mass spectrometer.

Development of a rapid and simple screening method for pesticide residues is very important in contributing to the safe securing of agricultural products. As suggested in these reports, it is thought that NDE, where complicated sample preprocessing after the extraction is unnecessary, is promising as a sample pretreatment screening method for pesticide residues in agricultural products.

As described in this paper, we specifically examine the advantages of NDE in which there might be little coextraction of matrix components; it therefore might be unnecessary to conduct cleanup processes after extraction. To verify the potential applicability of the extraction method, we targeted systemic pesticides, neonicotinoids, which have been attracting interest as promising insecticides because of their high insecticidal activity at very low application rates and their safety for humans and the environment. It is highly possible that pesticide which was sprayed on fruiting vegetables may remain in them at relatively high concentrations because foliar applications to them are feasible until the day before the harvest. We selected one of fruiting vegetables, a green pepper, as a model sample for that reason.

MATERIALS AND METHODS

Chemicals and Reagents. All solvents, which were of pesticide analytical grade and HPLC grade, were acquired from Wako Pure Chemical Industries Ltd. (Osaka, Japan). Water used for HPLC was prepared directly in the laboratory using a water purification system (Milli-Q; Millipore Corp., Bedford, MA). Analytical-grade pesticide and related metabolite standards with purities up to 95% were

Received: June 3, 2012
Accepted: August 21, 2012
Published: August 21, 2012

obtained from Wako Pure Chemical Industries Ltd., Hayashi Pure Chemical Ind., Ltd. (Osaka, Japan), and Kanto Chemical Co., Inc. (Tokyo, Japan). Each stock solution (1 mg/mL) was prepared in acetonitrile or methanol.

Field application pesticide formulations are as follows: Mospilan (water-soluble powder) containing 20.0% acetamiprid (Nippon Soda Co., Ltd., Tokyo, Japan); Admire (flowable) containing 20.0% imidacloprid (Bayer Crop Science, Tokyo, Japan); Dantotsu (water-soluble powder) containing 16.0% clothianidin (Sumitomo Chemical Co., Ltd., Tokyo, Japan); Starkle (water-soluble granule) containing 20.0% dinotefuran (Hokko Chemical Ind. Co., Ltd., Tokyo, Japan); Bariard (water-dispersible granule) containing 30.0% thiacloprid (Bayer Crop Science); Actara (water-soluble granule) containing 10.0% thiamethoxam (Syngenta Japan K.K., Tokyo, Japan); Bestguard (water-soluble powder) containing 10.0% nitenpyram (Sumitomo Chemical Co., Ltd.); Amistar 20 (flowable) containing 20.0% azoxystrobin (Syngenta Japan K.K.); Match (emulsifiable concentrate) containing 5.0% lufenuron (Syngenta Japan K.K.). Each pesticide formulation was diluted with water, then used for preparation of mimic samples, and applied to green pepper for preparation of incurred samples (Table 1).

Table 1. Summary of Pesticide Formulations Used in the Present Work

trade name	active ingredient	dilution rate with water	concentration of active ingredient after dilution (μg/mL)
Mospilan	acetamiprid	4000-fold	50
Admire	imidacloprid	4000-fold	50
Dantostu	clothianidin	2000-fold	80
Starkle	dinotefuran	2000-fold	100
Bariard	thiacloprid	2000-fold	150
Actara	thiamethoxam	2000-fold	50
Bestguard	nitenpyram	2000-fold	50
Amistar 20	azoxystrobin	2000-fold	100
Match	lufenuron	2000-fold	25

Chem Elut solid-phase extraction (SPE) cartridges packed with diatomaceous earth material were purchased from Varian Inc. (Harbor City, CA). Envi-Carb/NH_2 SPE cartridges (500 mg of graphitized carbon black and 500 mg of aminopropyl silica gel) and InertSep GC/PSA SPE cartridges (500 mg of graphitized carbon black and 500 mg of ethylenediamine-*N*-propyl silica gel) were from Supelco (Bellefonte, PA) and GL Sciences Inc. (Tokyo, Japan).

Preparation of Mimic Samples for Selection of Extractant and Recovery Tests. Each pesticide formulation diluted with water was applied on green pepper (10 μL/g of sample for recovery test, and 20 μL/g of sample for selection of extractant and recovery test) using a microsyringe (Hamilton Co., Reno, NV). Then the samples were allowed to stand for overnight at room temperature.

Production of Green Peppers with Field-Incurred Residues. Green peppers were grown in a plastic greenhouse on arable land of the National Institute for Agro-Environmental Sciences. Green peppers in the harvesting stage were sprayed individually with each pesticide formulation diluted with water according to the manufacturer's label using a handy sprayer (Dia Sprayer; Furupla, Co. Ltd., Tokyo, Japan). Five incurred green pepper samples at a time were harvested at 1, 3, and 7 days after spraying.

Sample Pretreatment with NDE. After each green pepper was weighed precisely, pesticide was extracted manually for 3 min while thoroughly wiping off the sample surface using absorbent cotton steeped in 20 mL of acetone or acetonitrile. Ten milliliter aliquots of the extract were portioned out; then the solvent was evaporated to dryness using a gentle nitrogen stream. The residue was reconstituted with 1 mL of mobile phase; then the solution was filtered with a PTFE membrane syringe-driven filter unit (0.45 μm, Millipore Corp., Billerica, MA). Before determination with HPLC, the solution was diluted properly with mobile phase.

Conventional Sample Pretreatment of Green Pepper Residue after NDE for Estimation of Residue Contents. Sample pretreatments for residue samples were performed according to the method described by Watanabe et al.[9] with slight modifications. The frozen green pepper residue sample after NDE was crushed roughly with a stainless steel spatula before extraction. Then it was homogenized with 100 mL of acetonitrile for 3 min using a homogenizer (Polytron PT2100; Kinematica AG, Lucerne, Switzerland). After homogenization, the resulting mixture was filtered through a funnel by suction. Then the residue on the funnel was treated similarly with 50 mL of acetonitrile. Both extracts were accurately made up to 200 mL with acetonitrile in a volumetric flask. For all targeted pesticides aside from nitenpyram and its metabolites, 50 mL aliquots of the extract, equivalent to a quarter of residue sample, was concentrated to about 5 mL. Then 5 mL of water was added to the concentrated extract. After the aqueous extract was applied to a Chem Elut SPE cartridge, it stood for 10 min. The cartridge was washed with 80 mL of *n*-hexane. Subsequently, the target pesticide was eluted with 100 mL of dichloromethane. Eluate was concentrated to about 1 mL. Residue dissolved in 5 mL of acetonitrile/toluene (3:1, v/v) was applied to an Envi-Carb/NH_2 SPE cartridge preconditioned with 10 mL of acetonitrile/toluene (3:1, v/v). After pesticide was eluted with 20 mL of acetonitrile/toluene (3:1, v/v), the eluate was concentrated to about 1 mL. It was then evaporated to dryness by a gentle nitrogen stream. Residue was treated similarly as described above.

For nitenpyram and its metabolites, after concentration of 50 mL aliquots of the extract, 10 mL of acetonitrile was added to the concentrated residue. Then the solution was applied to an InertSep GC/PSA SPE cartridge preconditioned with 5 mL of acetone and 5 mL of *n*-hexane. Compounds were eluted with 5 mL of acetonitrile, and the eluate was concentrated to about 1 mL. Residue dissolved in 5 mL of acetonitrile was applied to an Envi-Carb SPE cartridge preconditioned with 10 mL of acetonitrile. Nitenpyram and its metabolites were eluted with 10 mL of acetonitrile. Thereafter, the procedure was conducted similarly as described above.

Estimation of NDE Rate. The NDE rate for each pesticide was estimated by (a) absolute content (μg) extracted with NDE and (b) residue content (μg) after NDE as follows.

$$\text{NDE rate (\%)} = \text{(a)} \times 100/\sum \text{(a)} + \text{(b)}$$

Absolute and residual contents of nitenpyram were estimated from total concentrations of nitenpyram and its two major metabolites, 2-[*N*-(6-chloro-3-pyridylmethyl)-*N*-ethylamino]-2-methyliminoacetic acid (CPMA) and N^1-(6-chloro-3-pyridylmethyl)-N^1-ethyl-N^2-methylformamidine (CPMF) according to the following expression

$$[\text{total concentrations of nitenpyram containing CPMA and CPMF}] = A + B \times 1.06 + C \times 1.28$$

where *A*, *B*, and *C* are concentrations of nitenpyram, CPMA, and CPMF, respectively, and conversion factors 1.06 for CPMA and 1.28 for CPMF were estimated by dividing the molecular weight of nitenpyram (270.72) by those of metabolites (255.70 for CPMA, 211.69 for CPMF).

Evaluation of Matrix Effects. For verification of matrix effects, the extract of nonspiked sample with the NDE method was also prepared according the above-described procedure, and then each pesticide standard (0.5 μg) was added to the extract dissolved in 1 mL of mobile phase. Matrix effects were calculated by comparison of the peak area obtained for each pesticide in the sample with the NDE method with that of the pure solvent (mobile phase) according to the equation[10]

$$\text{matrix effect (\%)} = \left[\frac{\text{peak area of extract spiked with 0.5 μg of each pesticide}}{\text{peak area of pure solvent spiked with 0.5 μg of each pesticide}} - 1\right] \times 100$$

dx.doi.org/10.1021/jf3035429 | J. Agric. Food Chem. 2012, 60, 9000−9005

HPLC. The HPLC (1100 series; Agilent Technologies Japan Ltd.) was equipped with a quaternary pump, an autosampler, a column oven, and a diode array detector. The analytical column was a reversed-phase column (250 mm × 4.6 mm, 5 μm particle size, SunFire C18; Waters, Milford, MA) used in conjunction with a security guard column (20 mm × 4.6 mm, 5 μm particle size). Column oven temperature was kept at 40 °C for neonicotinoid insecticides and some metabolites and at 20 °C for azoxystrobin and lufenuron. Sample injection volume was 20 μL. For neonicotinoid insecticides aside from nitenpyram and its metabolites, the mobile phase was acetonitrile/water (25:75, v/v) and the flow rate was 0.85 mL/min. For nitenpyram and its metabolites, methanol/0.05 M KH_2PO_4 (30:70, v/v) as the mobile phase was run at 0.6 mL/min. For azoxystrobin and lufenuron, acetonitrile/water (70:30, v/v) was used as the mobile phase. Flow rate was 0.7 mL/min. Detection wavelength was set at 230 nm. Detection wavelengths were 230 (for azoxystrobin and lufenuron), 246 (for acetamiprid and thiacloprid), 254 (for thiamethoxam), and 270 nm (for clothianidin, dinotefuran, imidacloprid, and nitenpyram and its metabolites).

Under the chromatographic conditions described, calibration graphs were constructed by plotting peak areas vs concentrations. Excellent lineality and coefficient of regressions (r) were achieved for the investigated nine pesticides in the current work (Table 2). The limit of detection (LOD) for each pesticide was determined as the lowest concentration of each pesticide that gave a signal-to-noise ratio of 3. This was as low as 5 ng/mL for neonicotinoid insecticides except nitenpyram and azoxystrobin and as high as 15 ng/mL for nitenpyram and lufenuron (Table 2).

Table 2. Analytical Data for Neonicotinoid Insecticides, Azoxystrobin, and Lufenuron Investigated in the Current Work Using the HPLC Method

pesticide	equation of calibration curve	linearity (μg/mL)	r	LOD (ng/mL)
acetamiprid	$y = 71.8x + 1.348$	0.01−4	1.0000	5
imidacloprid	$y = 67.8x + 0.473$	0.01−4	1.0000	5
clothianidin	$y = 65.0x + 0.137$	0.01−4	1.0000	5
dinotefuran	$y = 50.6x + 1.775$	0.01−4	0.9999	5
thiacloprid	$y = 63.1x + 0.550$	0.01−4	0.9999	5
thiamethoxam	$y = 43.2x - 0.044$	0.01−4	1.0000	5
nitenpyram	$y = 47.7x - 0.350$	0.02−4	0.9999	15
azoxystrobin	$y = 51.1x - 0.500$	0.01−4	0.9999	5
lufenuron	$y = 33.7x + 0.350$	0.02−4	0.9999	15

RESULTS AND DISCUSSION

In the preliminary experiments, water-miscible solvents, acetone, acetonitrile, and methanol, which are commonly used in extraction of neonicotinoid insecticides, were tested. The most suitable extractant for NDE was selected. As Figure 1 shows, acetone exhibited the best extraction rate (shown as recovery rate) for all pesticides in mimic samples applied with diluted formulations containing a known amount of active ingredient. In this experiment, the extraction rate of nitenpyram with acetone was about 80%. However, the insecticide is metabolized in plants into two major metabolites, CPMA and CPMF, which must be analyzed as regulated compounds together with the parent nitenpyram.[11] Furthermore, the former metabolite is unstable in acetone. It is susceptible to transformation to CPMF.[12,13] Accordingly, it seems that, if possible, acetone should not be used as an extractant for nitenpyram to analyze three compounds individually. On the basis of these findings, acetone was chosen in subsequent experiments because this solvent yielded satisfactorily results and showed a higher extraction rate for all pesticides aside from nitenpyram, for which acetonitrile showed fair efficiency.

A recovery test was conducted using mimic samples applied with each diluted pesticide formulation at two concentration levels according to the optimum conditions for NDE described above. Figure 2 summarized the accuracy of the proposed NDE in mimic green pepper samples. Recovery rates ranged from 79.7% to 108.6%, and the results indicated that the proposed NDE quantitatively extracted the applied pesticides in all cases, with % CV ranging from 1.9% to 10.3%. These results are considered to be reasonable as the recovery of the proposed NDE.[14,15]

We investigated time-dependent changes of NDE rates from real field-incurred green pepper samples. Figure 3 shows that NDE rates of six neonicotinoid insecticides pesticides aside from thiacloprid decreased remarkably, although imidacloprid in the green peppers sampled at 0, 1, and 3 days after spraying and clothianidin, dinotefuran, and nitenpyram in them sampled at 0 and 1 day after spraying were generally extracted (more than 70%). It is noteworthy that thiamethoxam and acetamiprid were extracted at only about 60%, respectively, and it was readily apparent that the proposed method was not suited to extraction from the samples harvested even at 0 and 1 day after

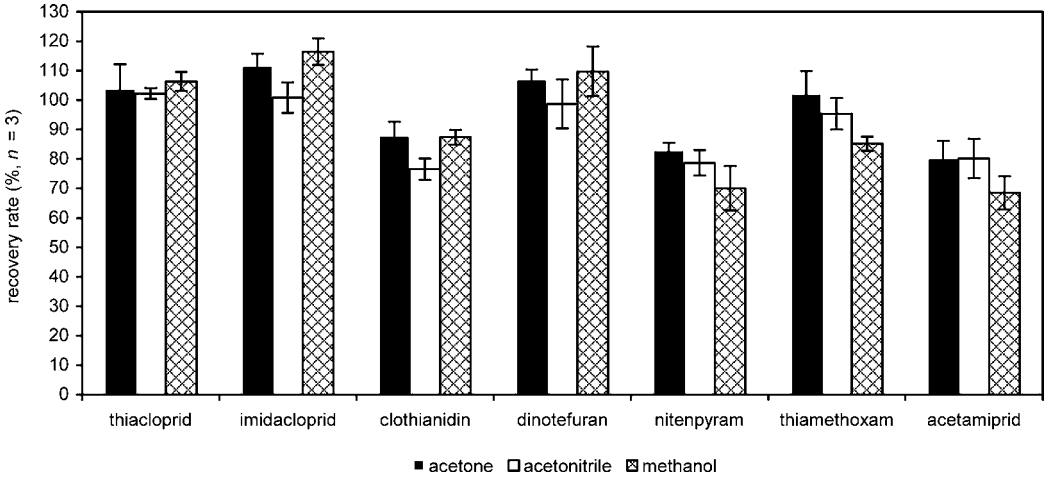

Figure 1. Selection of optimal extractant for NDE by mimic green pepper samples. Error bars indicate the standard deviation about the average (n = 3).

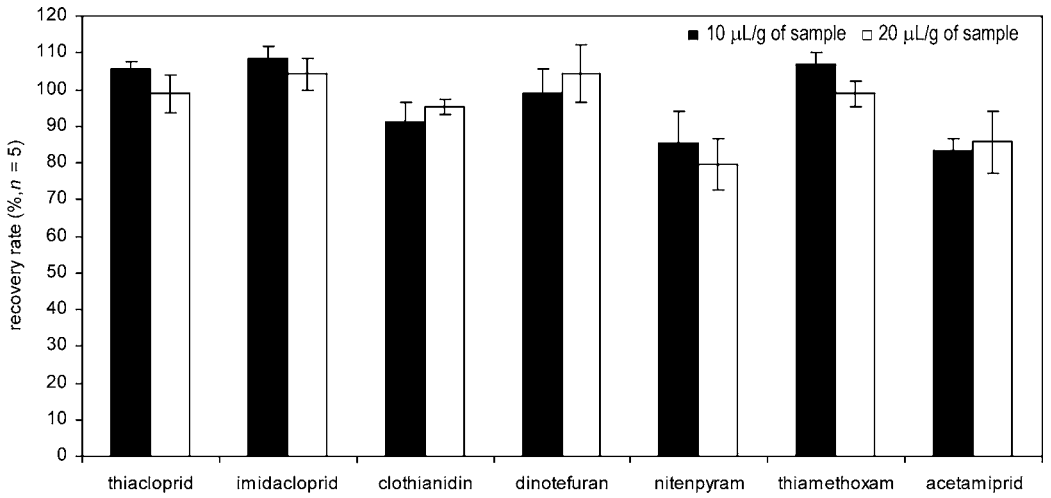

Figure 2. Recovery rates of neonicotinoid insecticides using NDE from mimic green pepper samples applied with each diluted formulation at two levels. Error bars indicate the standard deviation about the average (n = 5). Acetone was used as extractant for neonicotinoid insecticides except for nitenpyram for which acetonitrile was used.

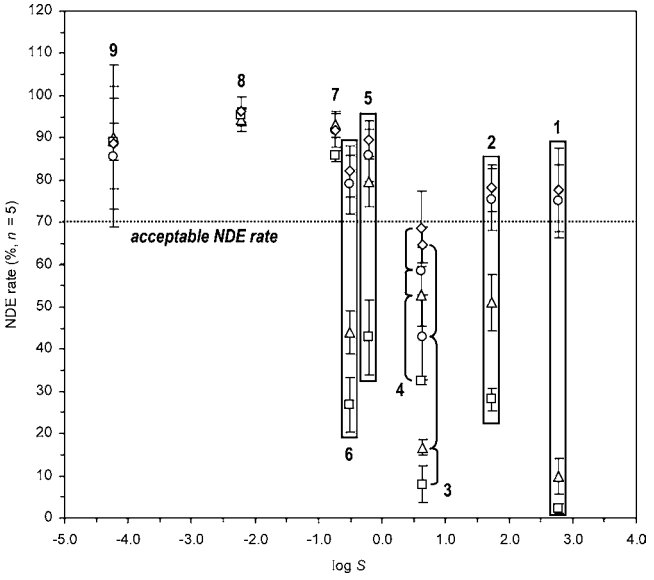

Figure 3. Relationship between the NDE rates of applied pesticides residues from field-incurred green pepper samples and their water solubilities. Error bars indicate the standard deviation about the average (n = 5). Refer to Table 1 for each number in the graph.

spraying. However, the NDE rate of thiacloprid was excellent over the test period.

As described above, the method proposed in this work is originally an extraction method that was specialized for adherent pesticide residues on sample surfaces. Therefore, in principle, the extraction method cannot be applicable to systemic pesticides such as neonicotinoid insecticides which move into plants over time. Nevertheless, only systemic thiacloprid showed a quantitative extraction rate (higher than 85%) (Figure 3).

Yukimoto and Hamada described that the systemic degree of applied pesticides into plants depends on their water solubilities,[16] that is, because adherent pesticide residues on plant surfaces transfer from hydrophobic epicuticular wax layer to inside of plant tissue and then are distributed into hydrophilic plant cells, it is thought that a difference occurs at the distribution speed because of the difference in water solubility of pesticide. We guessed that the difference would influence the diachronic decrease in NDE rate.

The proposed NDE using acetone as extractant was applied to green pepper samples that had been treated with a systemic fungicide, azoxystrobin, and a nonsystemic insecticide, lufenuron, showing lower water solubilities than neonicotinoid insecticides (Table 3),[17] with the result that more than 90% of azoxystrobin as well as thiacloprid were quantitatively extracted during the test period (Figure 3). However, because highly hydrophobic lufenuron shows virtually no systemicity and because it might persist on the fruit surface, it was extracted well using this method. The results suggested that the applicability of the NDE method does not depend on systemicity having merely pesticide or not; it greatly depends on the systemic speed (distribution speed in plant cell) of pesticide, and we conclude that the water solubility of each pesticide becomes the important index in ascertaining the applicability.

Table 3. Physicochemical Properties for the Investigated Pesticides[a]

	pesticide	water solubility, S (g/L)	log K_{ow}
1	nitenpyram	>590 (20 °C)	−0.66 (25 °C)
2	dinotefuran	54.3 ± 1.3 (20 °C)	−0.644
3	acetamiprid	4.25 (25 °C)	0.80 (25 °C)
4	thiamethoxam	4.1 (25 °C)	−0.13 (25 °C)
5	imidacloprid	0.61 (20 °C)	0.57 (21 °C)
6	clothianidin	0.304 (pH 4)~0.340 (pH 10) (20 °C)	0.7 (25 °C)
7	thiacloprid	0.185 (20 °C)	0.74 (unbuffered water)
8	azoxystrobin	0.006 (20 °C)	2.5 (20 °C)
9	lufenuron	$<0.06 \times 10^{-3}$ (25 °C)	5.12 (25 °C)

[a]Each value was referred to *The Pesticide Manual*, 14th ed; ref 17.

Since the relative responses (peak area of sample extract with NDE/peak area of pure solvent) for all of the investigated pesticides were in the range of −10−8%, it may be concluded that although a minor matrix effect appeared, the influence on the analytical data is extremely small. Therefore, fundamentally troublesome cleanup procedures were not needed after

dx.doi.org/10.1021/jf3035429 | J. Agric. Food Chem. 2012, 60, 9000−9005

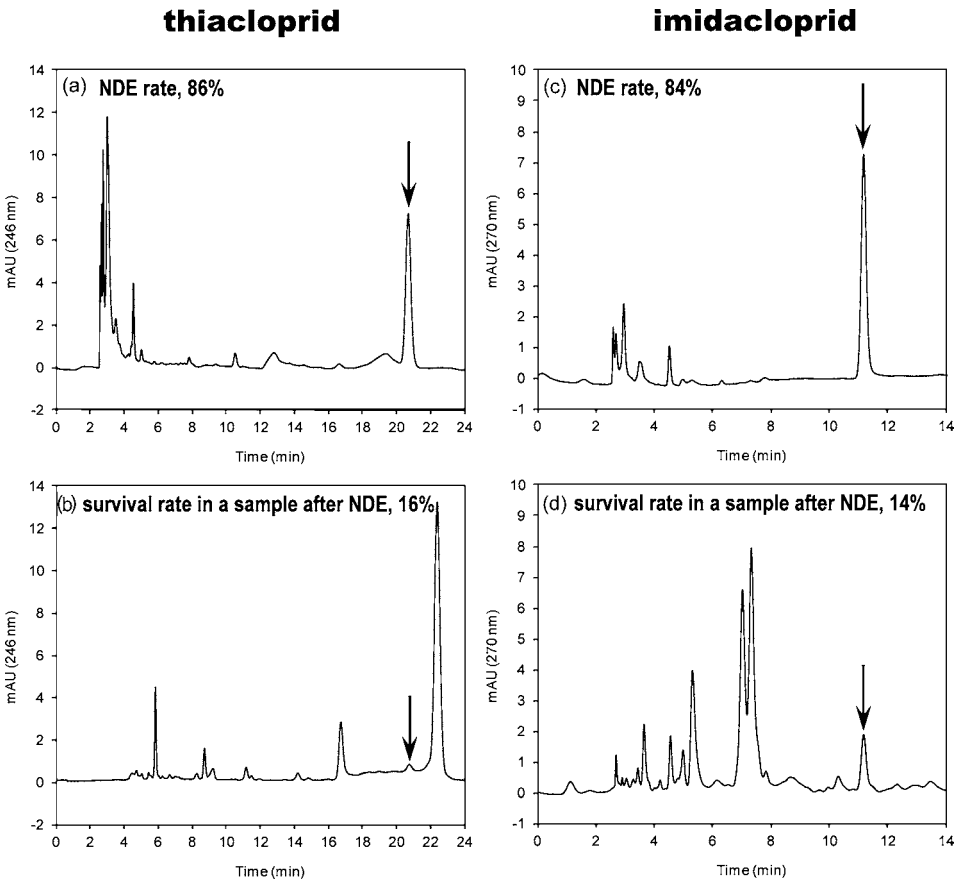

Figure 4. HPLC chromatograms of extracts from field-incurred green pepper samples. (a) NDE with acetone for thiacloprid residue in a sample harvested 7 days after spraying. (b) Cleaned-up extract after NDE of a sample harvested 7 days after spraying. (c) NDE with acetone for imidacloprid residue in a sample harvested 1 day after spraying. (d) Cleaned-up extract after NDE of a sample harvested 1 day after spraying.

extraction. In fact, the chromatograms exhibit an advantage over commonly used extraction methods for homogenized samples: there are no peaks of interference around the target pesticides. Moreover, the chromatograms of the samples extracted using the proposed method were considerably clean, drawing a comparison with those of conventional extraction methods (Figure 4).

The method presented here demonstrates that NDE is an efficient tool for rapid and simple extraction of pesticide residues on fruit surfaces without cleaning up sample extracts normally required in conventional pesticide residue analyses. The proposed method involves the use of small amounts of solvent (20 mL or less per sample) and is therefore environmentally friendly. Furthermore, because specialized apparatus for extraction is dispensable for the proposed method, it might be useful as an extraction method in the production of agricultural products. However, some collateral conditions are that (1) the proposed NDE method can be effective for extraction of pesticides showing high water solubility and (2) it can be limited to extraction for pesticides that are applied directly and principally to the stems and leaves of plants. Lastly, we will make an effort to direct the applicability of the proposed method to agricultural products and pesticides of other types.

AUTHOR INFORMATION

Corresponding Author

*Phone/fax: +81 29 838 8306. E-mail: eikiw@affrc.go.jp.

Funding

This work was financially supported by the Pesticide Science Society of Japan (FY2011).

Notes

The authors declare no competing financial interest.

ACKNOWLEDGMENTS

We sincerely express our gratitude to Dr. Masako Ueji (Japan Plant Protection Association, Tokyo, Japan) and Dr. Kazuaki Iijima (Institute of Environmental Toxicology, Ibaraki, Japan) for their valuable suggestions and to Mr. Takahiro Ara (National Institute for Agro-Environmental Sciences) for support of the preparation of the field-incurred green pepper samples.

REFERENCES

(1) Specht, W.; Pelz, S.; Gilsbach, W. Gas-chromatographic determination of pesticide residues after clean-up by gel-permeation chromatography and mini-silica gel-column chromatography. *Fresenius J. Anal. Chem.* **1995**, *353*, 183−190.

(2) Fillion, J.; Sauvé, F.; Selwyn, J. 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. *J. AOAC Int.* **2000**, *83*, 698−713.

(3) Ueno, E.; Oshima, H.; Saito, I.; Matsumoto, H. Determination of nitrogen- and phosphorus-containing pesticide residues in vegetables by gas chromatography with nitrogen-phosphorus and flame photometric detection after gel permeation chromatography and two-step minicolumn cleanup. *J. AOAC Int.* **2003**, *86*, 1241−1251.

(4) Nielssen, W. M. A.; Manini, P.; Andreoll, R. Matrix effects in quantitative pesticide analysis using liquid chromatography-mass spectrometry. *Mass Spectrom. Rev.* **2006**, *25*, 881−899.
(5) LeDoux, M. Analytical methods applied to the determination of pesticide residues in foods of animal origin. A review of the past two decades. *J. Chromatogr. A* **2011**, *1218*, 1021−1036.
(6) Saranwong, S.; Kawano, S. Rapid determination of fungicide contaminated on tomato surfaces using the DESIR-NIR: a system for ppm-order concentration. *J. Near Infrared Spectrosc.* **2005**, *13*, 169−175.
(7) Edison, S. E.; Lin, L. A.; Gamble, B. M.; Wong, J.; Zhang, K. Surface swabbing technique for the rapid screening for pesticides using ambient pressure desorption ionization with high-resolution mass spectrometry. *Rapid Commun. Mass Spectrom.* **2011**, *25*, 127−139.
(8) Edison, S. E.; Lin, L. A.; Parrasles, L. Practical considerations for the rapid screening for pesticides using ambient pressure desorption ionisation with high-resolution mass spectrometry. *Food Add. Contam.* **2011**, *28*, 1393−1404.
(9) Watanabe, E.; Baba, K.; Eun, H. Simultaneous determination of neonicotinoid insecticides in agricultural samples by solid-phase extraction cleanup and liquid chromatography equipped with diode-array detection. *J. Agric. Food Chem.* **2007**, *55*, 3798−3804.
(10) Stahnke, H.; Kittlaus, S.; Kempe, G.; Alder, L. Reduction of matrix effects in liquid chromatography-electrospray ionization−mass spectrometry by dilution of the sample extracts: How much dilution is needed? *Anal. Chem.* **2012**, *84*, 1474−1482.
(11) Ministry of Health, Labour and Welfare of Japan. Notification No. 111 of Ministry of Health and Welfare (Sept 1, 1997); http://wwwhourei.mhlw.go.jp/cgi-bin/t_docframe2.cgi?MODE=tsuchi&DMODE=SEARCH&SMODE=NORMAL&KEYWORD=%83%6a%83%65%83%93%83%73%83%89%83%80&EFSNO=6715&FILE=FIRST&POS=0&HITSU=5 (in Japanese) (accessed Mar 28, 2012).
(12) Tsumura, Y.; Nakamura, Y.; Tonogai, Y.; Kakimoto, Y.; Tanaka, Y.; Shibata, T. Determination of neonicotinoid pesticide nitenpyram and its metabolites in agricultural products. *J. Food Hyg. Soc. Jpn.* **1998**, *39*, 127−134 (in Japanese).
(13) Kobayashi, H. Development of residue analysis for pesticides by LC/MS and LC/MS/MS methods. *Bunseki Kagaku* **2009**, *58*, 985−997 (in Japanese).
(14) European Commission. Method validation and quality control procedures for pesticide residues analysis in food and feed (Document No. SANCO/10684/2009); http://ec.europa.eu/food/plant/protection/resources/qualcontrol_en.pdf#search=%27SANCO/10684/2009%27EC (accessed Aug 4, 2012).
(15) Japanese Ministry of Health, Labour and Welfare, Notification Syoku-An No. 1224-1 of Pharmaceutical and Food Safety Bureau (Dec 24, 2010). http://www.mhlw.go.jp/topics/bukyoku/iyaku/syoku-anzen/zanryu3/dl/101224-1.pdf (in Japanese) (accessed Aug 4, 2012).
(16) In *Genshoku Sakumotsu No Yakugai*; Yukimoto, M.; Hamada, K., Eds.; Zenkoku Noson Kyoiku Kyokai: Tokyo, Japan, 1985 (in Japanese).
(17) In *The e-Pesticide Manual*, 14th ed.; Tomlin, C. D. S., Ed.; British Crop Protection Council: Hampshire, U.K., 2006.