Analysis of 10 systemic pesticide residues in various baby foods using liquid chromatography-tandem mass spectrometry

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Abstract

Ten systemic pesticides, including methomyl, thiamethoxam, acetamiprid, carbofuran, fosthiazate, metalaxyl, azoxystrobin, diethofencarb, propiconazole, and difenoconazole were detected in 13 baby foods (cereals, boiled potatoes, fruits, and milk) using the QuEChERS method for sample preparation and liquid chromatography tandem mass spectrometry for analysis. The matrix-matched calibration curves showed good linearity with determination coefficients (R^2) > 0.992. The limits of detection and quantitation were 0.0015–0.003 and 0.005–0.01 mg/kg, respectively. Mean recoveries of three different concentrations ranged from 69.2–127.1% with relative standard deviations < 20. The method was successfully applied to 13 actual samples collected from a local market, and none of the samples contained pesticide residues. In sum, this method is suitable to accurately identify and quantify systemic pesticides with matrix-matched standards in various baby foods.

Keywords: Baby foods; Systemic pesticides; Residue analysis; QuEChERS; Tandem mass spectrometry.

Introduction

- Systemic pesticides act by moving into a plant through its roots, from which they spread via the plant's vascular system throughout cells.
- Special concern has arisen regarding food for infants (<12 months) and young children (1–3 years old) foods as a result of their higher food intake per kilogram body weight than adults.
- In general, babies and infants have food sources that are less varied. To prevent pesticide residues from entering the children's food chain, the European Commission specified an MRL of 0.01 mg/kg for pesticide residues.
- Therefore, sensitive and reliable analytical methods are required to monitor pesticide residues in baby foods to ensure the safety of the infant food supply.

Objective

The aim of this study was to develop a selective and sensitive method for analyzing systemic pesticides (five fungicides and five insecticides) in 13 baby foods by LC/MS/MS.

Chemicals and reagents

Sample

❖ Milk, modified milk powder, soybean milk, and yogurt

Standards

❖ Methomyl, thiamethoxam, acetamiprid, carbofuran, fosthiazate, metalaxyl, azoxystrobin, diethofencarb, propiconazole, and difenoconazole. These pesticides were selected on the basis of the risk to the consumers.

Reagents

❖ Acetonitrile (ACN), methanol (MeOH), Acetic acid (HAc), anhydrous magnesium sulfate ($MgSO_4$) and sodium acetate (NaOAc), Primary secondary amines (PSA) and C18.

Method

10 g homogenized Sample

Add 15 mL of 1% acetic acid in MeCN in a 50 mL Teflon centrifuge tube and vortex-mix for 1 min

Extraction

Add 6 g anhydrous $MgSO_4$ and 1.5 g NaOAc , shake by hand for 1 min and centrifuge for 5 min

Centrifugation

A 6 mL aliquot of the extract was transferred to a 15 mL Teflon centrifuge tube containing 900 mg of anhydrous MgSO4 and 300 mg PSA (+150 mg C18 for fatty food samples – milk, modified milk powder, soybean milk, and yogurt)

Clean-up

The Teflon centrifuge tube was shaken by hand for 1 min followed by centrifugation for 5 min. One milliliter of the extract was transferred to a vial.

Detection LC/MS/MS

LC/MS/MS conditions

Condition	Content
Instrument model	Allience 2695 LC Separations Module (Waters, Milford, MA) Micromass Quattro Micro triplequadrupole tandem mass (Waters)
Column	SunFire C_{18} (2.1 mm i.d × 150 mm, 3.5 µm, Waters, Milford, MA)
Column flow	0.2 mL/min
Mobile phase	A – 0.1% formic acid in water B – 0.1% formic acid in ACN
Injection volume	20 µL
Capillary Voltage	3.2 kV
Mode	Electrospray ionization positive (ESI^+) Multiple reaction monitoring (MRM)
Source temp.	150°C Desolvation temp. 350°C

Results and discussion

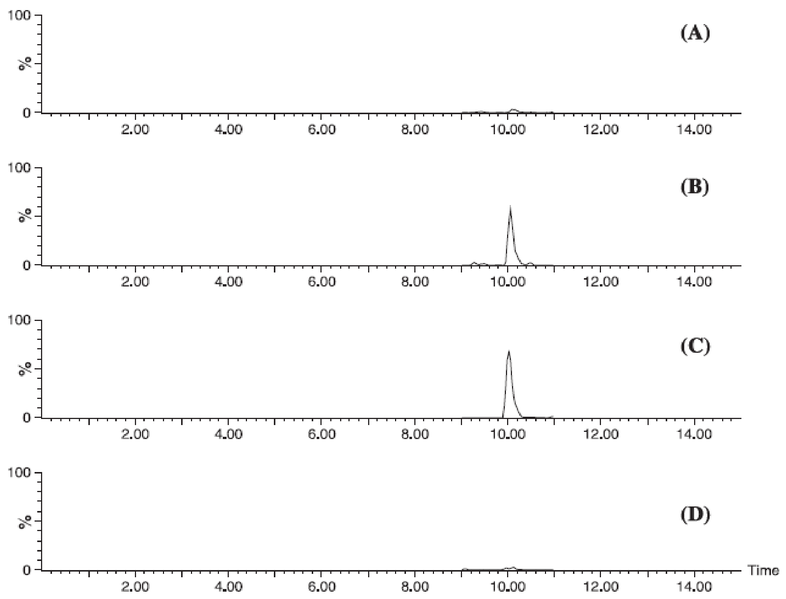

Figure 1. Liquid chromatography–tandem mass spectrometry chromatograms of carbofuran (quantifier ion); a blank apple sample (A), in a solvent standard of 0.005 mg/kg (B), a fortified sample of 0.005 mg/kg (C), and actual sample (D).

Table 1. Liquid chromatography-tandem mass spectroscopy multiple reaction monitoring transition data for the 10 systemic pesticides in baby foods

No.	Pesticide	Action	Formula	Retention time (min)	Precursor ion (*m/z*)	Product ion (*m/z*)	Collision energy (eV)
1	Methomyl	Insecticide	$C_5H_{10}N_2O_2S$	8.431	163.0	88.0	10
						106.0	10
2	Thiamethoxam	Insecticide	$C_8H_{10}ClN_5O_3S$	8.967	292.0	211.0	12
						131.9	22
3	Acetamiprid	Insecticide	$C_{10}H_{11}ClN_4$	9.393	223.1	121.0	18
						56.0	18
4	Carbofuran	Insecticide	$C_{12}H_{15}NO_3$	10.090	222.1	165.0	13
						123.0	13
5	Fosthiazate	Insecticide	$C_9H_{18}NO_3PS_2$	10.162	284.1	104.0	12
						228.0	12
6	Metalaxyl	Fungicide	$C_{15}H_{21}NO_4$	10.233	280.3	220.1	14
						160.1	14
7	Azoxystrobin	Fungicide	$C_{22}H_{17}N_3O_5$	10.575	404.1	372.0	13
						344.1	30
8	Diethofencarb	Fungicide	$C_{14}H_{21}NO_4$	10.647	268.1	180.0	20
						220.0	20
9	Propiconazole	Fungicide	$C_{15}H_{17}Cl_2N_3O_2$	11.099	342.1	69.0	25
						158.8	25
10	Difenoconazole	Fungicide	$C_{19}H_{17}Cl_2N_3O_3$	11.196	406.1	251.0	23
						111.0	23

Table 2. Infant food samples for pesticide analysis

No.	Food	Form	Group
1	Rice	Boiled	Cereal grains
2	Glutinous rice	Boiled	
3	Potato	Boiled	Potatoes
4	Sweet potato	Boiled	
5	Apple	Directly	Fruits
6	Banana	Directly	
7	Strawberry	Directly	
8	Pear	Directly	
9	Citrus	Directly	
10	Milk	Directly	Milks
11	Modified milk powder	Mixing with water	
12	Soybean milk	Directly	
13	Yogurt	Directly	

Table 3. Linearity, limit of detection (LOD), limit of quantification (LOQ) and the lowest maximum residue limits (MRLs) of the target pesticides

No.	Compound	Linearity (r^2)	LOD (mg/kg)	LOQ (mg/kg)	Lowest MRL[a]
1	Methomyl	0.998	0.0030	0.010	0.05
2	Thiamethoxam	0.997	0.0030	0.010	0.05
3	Acetamiprid	0.994	0.0030	0.010	0.10
4	Carbofuran	0.993	0.0015	0.005	0.03
5	Fosthiazate	0.999	0.0015	0.005	0.05
6	Metalaxyl	0.992	0.0015	0.005	0.05
7	Azoxystrobin	0.998	0.0015	0.005	0.05
8	Diethofencarb	0.998	0.0030	0.010	0.05
9	Propiconazole	0.999	0.0025	0.008	0.05
10	Difenoconazole	0.999	0.0015	0.005	0.05

[a]MRLs have established by the KFDA (2011).

Table 4. Recoveries and relative standard deviations (RSDs) of 10 systemic pesticides in representative baby food samples

No.	Compound	Fortified concentration (mg/kg)	Rice		Potato		Apple		A milk		B milk	
			Recovery	RSD (%)	Recovery	RSD (%)	Recovery	RSD (%)	Recovery	RSD (%)	Recovery	RSD (%)
1	Methomyl	0.01	114.4	4.5	109.4	3.5	111.0	3.9	115.0	4.6	85.7	2.9
		0.02	118.3	13.5	106.3	2.0	86.6	5.0	89.0	5.1	80.7	0.5
		0.1	117.5	9.6	95.4	7.3	80.2	2.9	94.0	4.9	98.0	1.8
2	Thiamethoxam	0.01	115.6	9.1	106.2	17.5	94.2	9.1	94.1	7.0	85.1	4.4
		0.02	77.1	12.2	95.8	9.2	76.4	3.3	76.9	7.8	70.1	10.3
		0.1	90.1	13.5	126.1	9.8	79.5	7.0	73.8	6.5	84.3	5.5
3	Acetamiprid	0.01	69.0	5.1	71.0	6.0	99.4	14.0	113.2	7.7	81.4	7.1
		0.02	91.8	9.5	102.9	5.8	76.6	0.3	125.1	4.3	93.0	4.3
		0.1	94.8	12.0	96.6	4.2	69.8	5.3	70.3	2.3	76.5	15.1
4	Carbofuran	0.005	120.4	11.0	76.6	9.3	69.9	6.3	81.6	7.2	105.0	5.4
		0.01	112.4	13.5	83.0	11.3	73.1	10.5	113.5	15.1	80.0	6.7
		0.05	107.1	10.7	72.4	5.7	69.2	4.0	83.1	3.9	107.0	2.5
5	Fosthiazate	0.005	109.9	7.5	122.3	8.5	125.8	10.9	92.7	5.9	79.8	3.7
		0.01	113.9	8.3	115.8	12.0	116.4	9.7	114.0	5.3	75.8	10.5
		0.05	98.8	2.9	122.2	2.5	90.2	10.9	112.0	8.4	107.4	16.1
6	Metalaxyl	0.005	82.6	5.4	115.8	8.1	103.7	16.4	87.4	3.8	74.1	3.9
		0.01	96.7	9.5	72.3	7.6	95.1	4.0	71.6	10.1	87.5	8.4
		0.05	111.9	4.4	90.2	0.1	94.6	11.5	70.2	4.6	82.9	1.1
7	Azoxystrobin	0.005	113.9	0.2	99.0	12.0	90.7	13.9	102.5	9.6	83.5	11.7
		0.01	82.8	9.0	94.1	10.4	75.9	3.8	96.8	3.8	127.1	0.6
		0.05	115.2	6.4	89.0	7.2	87.7	3.8	98.7	6.9	71.2	15.6
8	Diethofencarb	0.01	81.6	1.1	72.5	16.9	70.3	4.0	99.9	13.6	100.6	4.2
		0.02	95.7	5.3	80.8	5.5	89.2	10.1	114.0	8.1	91.3	11.4
		0.1	93.3	6.7	88.1	7.7	75.9	11.6	82.6	3.2	114.1	2.9
9	Propiconazole	0.008	120.2	6.6	105.3	14.6	87.9	5.2	88.7	17.2	85.3	9.1
		0.016	122.2	9.0	118.6	7.8	107.7	5.5	96.3	4.1	99.4	3.1
		0.08	111.1	0.6	108.0	1.0	93.5	3.2	84.9	5.4	87.2	4.2
10	Difenoconazole	0.005	101.8	10.0	102.8	2.3	84.3	3.9	77.2	7.6	78.8	11.0
		0.01	102.6	5.5	111.7	9.1	86.7	4.2	94.8	6.1	73.7	1.9
		0.05	93.9	2.5	107.6	2.1	85.7	1.8	78.4	4.7	82.8	5.5

Milk A and B denote two different companies.

Table 5. Comparison of percentage recovery in pure solvent standards and matrix matched standards

No.	Compound	Fortified concentration (mg/kg)	Rice		Potato		Apple		A-milk		B-milk	
			SC[a]	MC[b]	SC	MC	SC	MC	SC	MC	SC	MC
1	Acetamiprid	0.01	111.4	69.0	120.8	71.0	73.2	99.4	182.6	68.7	172.4	81.4
		0.02	92.9	91.8	93.2	102.9	40.4	76.6	136.4	74.5	176.4	96.4
		0.1	93.4	94.8	99.5	96.6	52.8	69.8	146.4	70.3	159.3	76.5
2	Carbofuran	0.005	132.5	120.4	89.0	76.6	131.2	69.9	90.0	81.6	109.4	105.0
		0.01	129.9	112.4	104.1	83.0	112.3	73.1	93.7	113.5	128.8	80.0
		0.05	115.7	107.1	93.9	72.4	132.2	69.2	136.3	83.1	134.1	107.0

[a]SC, Recoveries for solvent standard calibration.
[b]MC, Recoveries for matrix-matched standard calibration.

Conclusions

The developed method was sensitive and selective enough to detect the analytes below the MRL established by the European Commission and the Korea Food and Drug Administration. The combination of QuEChERS sample preparation and LC/MS/MS analysis provided acceptable quantitative and qualitative results in baby food. This method achieved excellent validation results which could be used on a wide scale to detect pesticides in various baby foods.

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