Pesticide analysis in fruit and vegetables by microflow liquid chromatography coupled to mass spectrometry

CONTENTS

1. Aim and scope

This study is aimed at the development and validation a new method to decrease matrix effects in the quantification of pesticide residues in fruits and vegetables using a microflow-liquid chromatography system coupled to a triple quadrupole mass spectrometer and an electrospray ionization source with an ESI emitter to provide a microflow rate (microflow-LC-ESI-QqQ-MS). Multiresidue method validation and quality control were carried out for 90 pesticide residues including degradation products in three commodities (tomato, pepper and orange), within the framework of european regulatory controls on pesticide residues.[1]

2. Short description

The samples homogeneously crushed were extracted with QuEChERS procedure, previously published[2]. Blank samples were examined to confirm absence of the target pesticide residues and used for preparing matrix-matched standards. The internal standards dichlorvos-d^6 and malathion-d^{10} were used to control the recovery experiments. Linuron-d^6 and Dimethoate-d^6 were used at the step of dilution and injection of the samples into the microflow-LC-MS system to test robustness of the whole analysis. The diluted extract is analysed by injecting 3 µL into the microflow-LC-ESI-QqQ-MS system.

3. Apparatus and consumables

- Automatic pipettes, suitable for handling volumes of 30 µL to 500 µL and 1 mL to 3 mL.
- 50 ml PTFE centrifuge tubes with screw caps
- 15 ml PTFE centrifuge tubes with screw caps
- Vortex
- Automatic axial extractor
- Centrifuge, suitable for the centrifuge tubes employed in the procedure and capable of achieving at least 3700 rpm
- Concentration workstation
- Syringes, e.g. 2 mL disposable syringes
- Syringe filters PTFE, 0.2 µm pore size
- Injection vials, 2 ml, suitable for LC auto-sampler
- Volumetric flasks

4. Chemicals

- Acetonitrile ultra-gradient grade
- Formic acid
- Trisodium citrate dihydrate
- Sodium chloride
- Disodium hydrogencitrate sesquihydrate
- Anhydrous magnesium sulphate
- Primary secondary amine bonded silica (PSA), bulk material
- Bondesil-C18
- Ultra-pure water
- Pesticides standards

5. Procedure

5.1. Sample preparation

The samples were homogeneously crushed and stored at 4◦C before spiking and sample extraction. Samples were prepared and processed according to the SANCO Guideline for pesticide residues analysis in food[1].

5.2. Recovery experiments for method validation

The samples employed in validation studies did not contain any of the pesticides analysed.

Individual pesticide stock solutions (1000–2000 mg/L) were prepared in acetonitrile and ethyl acetate and were stored in amber screw-capped glass vials in the dark at -20 °C.

60g of previously homogenised sample (tomato, pepper and orange) were weighed and transferred to a crystalliser, where they were fortified homogenously with the working standard solution in acetonitrile.

The validation method was performed at two fortification levels (5 and 50 µg/Kg). Five replicates were analysed at each level.

5.3. Extraction

1. Weigh 10 g ± 0.1 g of sample in 50 mL PTFE centrifuge tube.
2. Add 10 mL of acetonitrile and 50 µL of 10 mg/L of dichlorvos-d^6 and malathion-d^{10} (internal surrogate standards)
3. Shake in automatic axial extractor for 4 minutes.

4. Add 4 g of anhydrous magnesium sulphate, 1 g of sodium chloride, 1g of trisodiumcitrate dehydrate and 0.5 g of disodium hydrogencitrate sesquihydrate.
5. Shake in automatic axial extractor for 4 minutes.
6. Centrifuge for 5 min at 3500 rpm.
7. Transfer 5 mL of supernatant into 15 mL PTFE centrifuge tube containing 750mg of anhydrous magnesium sulphate, 125 mg of PSA and 125 mg of bondesil-C18 and shake in a vortex 30 s.
8. Centrifuge for 5 min at 3500 rpm.
9. Add to the extract 50 µL of 5% formic acid in acetonitrile.
10. An aliquot is diluted 30 times with a mixture of AcN: H2O (20:80) and filtered using a 0.2 µm PTFE syringe filter.

With this treatment, 1 mL of sample extract represents 1 g of sample.

5.4. Measurement

The microflow-LC-ESI-QqQ-MS system was operated in SRM (selected reaction monitoring) mode with a resolution set to unit for Q1 and Q3. The best sensitivity in SRM mode was achieved under time-scheduled conditions and with a time window of 30 s. Identification was based on the EU guideline for LC–MS/MS analysis [3]: acquisition of two SRM transitions, retention time (tolerance of ±0.2 min) and compliance of the SRM ratio (relationship between the abundance of transitions selected for identification and for quantification, SRM2/SRM1 with a tolerance of ±30%). The mass transitions used are presented in Appendix I.

5.5. Instrumentation and analytical conditions for the microLC- MS/MS system

5.5.1. Eksigent ekspert™ microflow-LC 200 system.

- Column: Halo-C18 of 50 x 0.5 mm i.d. and 2.7µm, 90Å particle size
- Mobile phase A: 0.1% formic acid in ultra-pure water
- Mobile phase B: acetonitrile
- Flow rate: 30 µL/min
- Injection volume: 3 µL

Mobile phase gradient for pesticides analyse in positive mode

Time [min]	Mobile phase A	Mobile phase B
0	80%	20%
1	80%	20%
10	2%	98%
13	2%	98%
14	80%	20%

5.5.2. 4500 QTRAP

- TEM(ESI source gas temperature): 300 °C
- GS1(Nebulizer Gas): 30psi
- GS2(Turbo Gas): 30psi
- CUR (Curtin gas): 20 psi
- IS (Ion spray voltaje): 5000v
- CAD (Collision gas): Medium
- EX: 10 psi
- CXP (Collision Cell Exit Potencial): 12 psi

Operational parameters for pesticides are presented in Appendix I.

6. Validation of the method

6.1. Recoveries and within-laboratory reproducibility

The results corresponding to the mean recovery (n=5) and within-laboratory reproducibility in terms of relative standard deviation (RSD) at both fortification levels are summarized in Appendix II. Acceptable mean recoveries are those within the range of 70-120 %, with an associated repeatability RSD < 20%.[1]

6.2. Limits of quantitation

Document N° SANCO/12495/2011 defines limit of quantitation as the lowest validated spike level meeting the method performance acceptability criteria. LOQs are summarized in Appendix II.

6.3. Linearity

Linearity of the systems was evaluated by assessing the signal responses of the target analytes from matrix-matched calibration solutions prepared by spiking blank extracts at eight concentration levels, from 0.16 to 13.3 µg L^{-1}, corresponding to 5–400 µg kg^{-1} in the sample. In almost all cases, coefficient of determination (r^2) was higher than 0.99. Linearity ranges for all pesticides in all matrices are summarized in Appendix II.

6.4. Matrix effects

Matrix effects were evaluated based on the slopes of regression lines plotted from results obtained in matrices versus standard solutions. Values of matrix effects are summarized in Appendix II.

This report aims to provide information to laboratories that analyse pesticide residues in vegetables and fruit using a micro-flow system.

7. References

- SANCO/12571/2013. Guidance document on analytical quality control and validation procedures for pesticide residues analysis in food and feed. Avalailable at: http://ec.europa.eu/food/plant/plant_protection_products/guidance_documents/docs/qualcontrol_en.pdf.

- EURL-FV multiresidue method using QuEChERS followed by GC-QqQ/MS/MS and LC-QqQ/MS/MS for fruits and vegetables. http://www.crlpesticides.eu/library/docs/fv/CRLFV_Multiresidue_methods.pdf.

- Commission Decision 2002/657/EC of 12 August 2002 implementing Council Directive 96/23/EC establishes criteria and procedures for the validation of analytical methods to ensure the quality and comparability of analytical results generated by official laboratories. Official Journal of the European Communities L221/8, 17.8.2002.

APPENDIX I: MASS TRANSITIONS

Table1. SRM transitions of identification and quantification (SRM1 and SRM2). Operational parameters of the multiresidue method for pesticide residue analysis by microflowLC–ESI-QTRAP-MS/MS.

Pesticide residues	t_R (min)	DP (V)	SRM_1	CE1 (eV)	SRM_2	CE2 (eV)
Acetamiprid	1.02	77	223.0/126.0	30	223.0/56.0	28
Azinphos-Methyl	4.15	55	318.0/132.1	21	318.0/261	8
Azoxystrobin	4.48	57	404.0/372.0	20	404.0/344.1	33
Bitertanol	4.94	60	338.0/269.1	14	338.0/99.1	24
Boscalid	4.55	105	343.0/307.0	27	343.0/140.0	24
Bromuconazole	4.24	100	378.0/159.0	40	378.0/70.0	59
Bupirimate	3.57	100	317.0/166.1	32	317.0/272.1	27
Buprofezin	4.60	62	306.0/201.0	17	306.0/116.0	23

Carbaryl	2.92	56	202.0/145.0	13	202.0/127.0	37
Carbendazim	0.45	113	192.0/160.0	26	192.0/132.0	43
Chlorpyrifos	6.92	66	352.0/200.0	28	352.0/125.0	27
Cyprodinil	3.22	130	225.6/93.1	48	225.6/108.1	33
Diazinon	5.60	102	304.8/169.2	29	304.8/153.1	28
Diclorvos-d_6	2.18	66	226.8/115.0	25	226.8/83.0	37
Dicrotophos	0.49	63	238.0/112.0	17	238.0/193.1	13
Diethofencarb	4.13	61	268.0/226.1	13	268.0/180.1	25
Difenoconazole	5.44	105	406.0/251.0	35	406.0/337.0	25
Dimethoate	0.94	50	230.0/199.0	12	230.0/171.0	19
Dimethoate-d_6	0.91	62	236.0/205.0	11	236.0/177.0	21
Diniconazole	4.99	85	326.0/70.0	60	326.0/159.0	45
Epoxiconazole	449	90	330.0/121.1	22	330.0/141.1	23
Ethirimol	0.48	100	210.0/140.0	28	210.0/98.0	34
Ethofenprox	8.35	60	394.0/177.1	20	394.0/359.2	15
Ethoprophos	4.43	65	243.0/131.0	27	243.0/215.0	16
Fenamidone	4.45	75	312.0/236.0	21	312.0/92.1	40
Fenarimol	4.24	110	331.0/268.0	32	331.0/259.3	35
Fenbuconazole	4.86	90	337.0/125.0	50	337.0/70.0	50
Fenhexamid	4.52	100	302.0/97.0	30	302.0/55.0	60
Fenitrothion	4.99	86	278.0/125.0	26	278.0/109.0	21
Fenpropathrin	6.92	80	350.0/125.0	21	350.0/97.1	44
Fenpropimorph	3.06	150	304.0/147.0	39	304.0/130.0	35
Fenpyroximate	7.03	110	422.0/366.0	21	422.0/215.2	34
Fenthion	5.59	80	279.0/247.0	17	279.0/169.1	23
Fenthion-Oxon	3.52	85	263.0/231.0	21	263.0/216.0	31
Fenthion-Oxonsulfone	1.31	95	295.0/217.0	25	295.0/104.0	32
Fenthion-Oxonsulfoxide	0.78	85	279.0/264.0	27	279.0/104.0	35
Fenthion-Sulfone	3.73	95	311.0/125.0	95	311.0/279.0	89
Fenthion-Sulfoxide	2.86	90	295.0/280.0	25	295.0/232.0	29
Flusilazole	4.77	97	316.0/247.0	25	316.0/165.0	39
Flutriafol	3.09	80	302.0/70.0	55	302.0/123.0	35
Fosthiazate	2.99	62	284.0/228.0	13	284.0/104.0	32
Hexaconazole	4.77	95	314.0/70.0	55	314.0/159.1	45
Imazalil	2.29	110	297.0/159.0	29	297.0/201.0	25

Imidacloprid	0.84	65	256.0/209.0	22	256.0/175.0	27
Iprodione	4.90	80	330.0/245.0	21	330.0/288.0	25
Iprovalicarb	4.33	60	321.1/119.0	30	321.1/203.1	12
Kresoxim-Methyl	5.41	64	314.0/267.0	10	314.0/282.1	11
Linuron-d_6	4.05	74	255.0/160.0	26	255.0/185.0	24
Malathion-d_{10}	4.86	76	341.0/132.0	19	341.0/100.0	37
Mandipropamid	4.67	80	412.0/328.0	20	412.0/356.0	15
Metconazole	4.90	90	320.0/70.0	65	320.0/125.0	60
Methidathion	4.06	55	303.0/145.0	14	303.0/85.0	28
Methiocarb	3.99	60	226.0/169.0	13	226.0/121.1	25
Methiocarb-Sulfone	1.34	78	258.0/201.0	12	258.0/122.0	23
Methiocarb-Sulfoxide	0.71	70	242.0/185.0	18	242.0/122.0	37
Methomyl	0.51	37	163.0/106.0	14	163.0/88.1	12
Methoxyfenozide	4.81	50	369.0/313.2	11	369.0/149.1	28
Omethoate	0.45	60	214.0/183.0	16	214.0/155.0	21
Oxadixyl	2.14	67	279.0/219.2	14	279.0/102.0	14
Oxydemeton-Methyl	0.46	48	247.0/169.0	18	247.0/105.0	17
Paclobutrazol	3.91	90	294.0/70.0	52	294.0/125.0	55
Parathion	5.59	60	292.0/236.0	20	292.0/264.1	13
Parathion-Methyl	4.58	70	264.0/232.0	23	264.0/125.0	23
Penconazole	4.78	77	284.0/70.0	42	284.0/159.0	45
Pencynuron	5.90	95	329.0/125.0	55	329.0/218.0	22
Pendimethalin	6.89	40	282.0/212.1	16	282.0/194.0	26
Phenthoate	5.65	66	321.0/163.0	15	321.0/247.0	15
Phosalone	6.04	75	368.0/182.0	23	368.0/322.1	14
Phoxim	6.01	65	299.0/129.0	16	299.0/153.1	9
Pirimicarb	0.52	70	239.0/182.1	21	239.0/72.1	39
Pirimiphos-Methyl	5.28	110	306.0/164.1	30	306.0/108.1	43
Prochloraz	3.87	53	376.0/308.0	16	376.0/266.0	23
Propargite	7.29	60	368.0/231.0	15	368.0/175.0	21
Propiconazole	5.01	100	342.0/159.0	41	342.0/69.0	35
Propoxur	2.48	50	210.0/168.0	11	210.0/111.1	20
Propyzamide	4.51	70	256.0/190.0	21	256.0/173.0	33
Prothiofos	7.92	80	344.8/241.0	25	344.8/269.0	16
Pyraclostrobin	5.79	64	388.0/194.0	16	388.0/164.0	24

Pyrethrins	7.32	62	329.0/161.0	13	329.0/143.0	23
Pyridaben	7.57	90	365.1/309.0	19	365.1/147.0	35
Pyrimethanil	2.04	135	200.0/107.1	32	200.0/168.1	40
Pyriproxyfen	6.71	70	322.0/96.0	20	322.0/227.0	20
Quinoxyfen	5.99	120	308.0/197.0	44	308.0/272.1	40
Rotenone	5.04	120	395.0/213.0	35	395.0/192.1	36
Tebuconazole	4.63	96	308.0/70.1	51	308.0/125.0	53
Tebufenpyrad	6.23	110	334.0/145.0	35	334.0/117.0	60
Tetraconazole	4.68	100	372.0/159.0	43	372.0/70.0	65
Thiodicarb	2.81	63	355.0/88.0	27	355.0/108.0	21
Thiophanate-Methyl	2.50	75	343.0/151.0	27	343.0/311.0	16
Tolclofos-Methyl	5.92	84	301.0/125.0	26	301.0/269.0	22
Triadimefon	4.49	70	294.0/197.0	22	294.0/225.1	18
Triadimenol	3.95	45	296.0/70.0	38	296.0/227.0	13
Triticonazole	4.06	80	318.0/70.0	55	318.0/125.0	50
Zoxamide	5.64	107	336.0/187.0	31	336.0/204.0	23

APPENDIX II: VALIDATION RESULTS

Table 2. Validation of microflow-LC-ESI-QTRAP-MS based method: linearity, concentration range, matrix effect (ME) and recovery for the selected matrices studied.

Pesticide residues	Recovery (%) 5 µgKg^{-1};50 µgKg^{-1}			Correlation coefficient (R2)			Linear range (µgkg^{-1})			ME (%)			LOQ (µgKg^{-1})		
	Tomato	Pepper	Orange	Tomato	Pepper	Orange	Tomato	Pepper	Orange	Tomato	Pepper	Orange	Tomato	Pepper	Orange
Acetamiprid	92;86	92;96	102;106	0.9993	0.9971	0.9992	5-400	5-400	5-400	-7	6	-1	5	5	5
Azinphos-Methyl	88;103	103;121	121;101	0.9964	0.9906	0.9956	10-400	5-400	5-400	15	12	15	5	5	5
Azoxystrobin	102;103	104;95	119;120	0.9990	0.9984	0.9978	5-400	5-400	5-400	-10	8	-2	5	5	5
Bitertanol	111;104	112;94	120;114	0.9980	0.9969	0.9950	5-400	5-400	5-400	-20	-16	-14	5	5	5
Boscalid	94;92	96;100	106;106	0.9991	0.9964	0.9981	5-400	5-400	5-400	-6	-1	3	5	5	5
Bromuconazole	109;87	94;101	94;120	0.9959	0.9964	0.9991	5-400	5-400	5-400	7	15	-8	5	5	5
Bupirimate	110;93	107;98	119;120	0.9991	0.9970	0.9960	5-400	5-400	5-400	-5	0	-7	5	5	5
Buprofezin	96;73	89;85	101;106	0.9969	0.9969	0.9983	5-400	5-400	5-400	1	9	16	5	5	5
Carbaryl	108;102	105;103	106;105	0.9983	0.9961	0.9987	5-400	5-400	5-400	-1	12	6	5	5	5
Carbendazim	102;93	120;120	81;81	0.9994	0.9958	0.9990	5-400	5-400	5-400	-18	5	-23	5	5	5
Chlorpyrifos	95;89	101;90	120;112	0.9967	0.9939	0.9837	5-400	5-400	5-400	-3	-3	-6	5	5	5
Cyprodinil	83;92	92;88	120;104	0.9977	0.9997	0.9902	5-400	5-400	5-400	-8	8	-40	5	5	5
Diazinon	93;78	94;94	93;103	0.9992	0.9983	0.9968	5-400	5-400	5-400	-8	-5	-7	5	5	5

Dicrotophos	103;101	110;112	98;95	0.9987	0.9981	0.9981	5-400	5-400	5-400	-18	-15	-14	5	5	5
Diethofencarb	113;100	100;98	100;120	0.9986	0.9959	0.9987	5-400	5-400	5-400	-7	8	10	5	5	5
Difenoconazole	93;87	94;94	105;109	0.9992	0.9981	0.9986	5-400	5-400	5-400	-2	3	-5	5	5	5
Dimethoate	97;97	103;102	119;104	0.9993	0.9969	0.9996	5-400	5-400	5-400	-6	5	4	5	5	5
Diniconazole	101;94	93;95	115;106	0.9989	0.9977	0.9942	5-400	5-400	5-400	1	0	2	5	5	5
Epoxiconazole	93;89	99;104	105;117	0.9955	0.9965	0.9959	5-400	5-400	5-400	0	15	15	5	5	5
Ethirimol	90;96	72;70	119;89	0.9945	0.9943	0.9933	5-400	5-400	5-400	-17	5	-29	5	5	5
Ethofenprox	87;87	92;91	120;120	0.9993	0.9992	0.9926	5-400	5-400	5-400	5	15	16	5	5	5
Ethoprophos	108;99	102;101	116;112	0.9979	0.9967	0.9987	5-400	5-400	5-400	-10	-2	3	5	5	5
Fenamidone	98;100	105;104	110;114	0.9973	0.9985	0.9972	5-400	5-400	5-400	-6	-6	-3	5	5	5
Fenarimol	108;87	96;92	121;110	0.9981	0.9918	0.9959	5-400	5-400	5-400	-2	7	0	5	5	5
Fenbuconazole	110;94	93;103	112;120	0.9977	0.9968	0.9947	5-400	5-400	5-400	-6	2	4	5	5	5
Fenhexamid	101;66	109;80	99;92	0.9983	0.9975	0.9988	5-400	5-400	5-400	-4	2	12	5	5	5
Fenitrothion	120;91	120;111	90;105	0.9903	0.9350	0.9918	50-400	50-400	50-400	12	10	11	50	50	50
Fenpropathrin	65;93	95;95	120;119	0.9995	0.9976	0.9863	5-400	5-400	5-400	-10	-2	-1	5	5	5
Fenpropimorph	98;87	94;92	101;105	0.9972	0.9986	0.9992	5-400	5-400	5-400	13	13	9	5	5	5
Fenpyroximate	93;93	105;92	120;120	0.9992	0.9931	0.9801	5-400	5-400	5-400	-15	7	2	5	5	5
Fenthion	96;91	97;93	103;115	0.9996	0.9940	0.9959	5-400	5-400	5-400	7	7	6	5	5	5
Fenthion-Oxon	69;75	65:63	71:69	0.9998	0.9986	0.9957	5-400	5-400	5-400	-5	-7	-14	5	5	5

Fenthion-Oxonsulfone	72;76	71;69	70;65	0.9993	0.9998	0.9929	5-400	5-400	5-400	-2	-8	-9	5	5	5
Fenthion-Oxonsulfoxide	78;71	75;81	81;80	0.9976	0.9991	0.9967	5-400	5-400	5-400	-3	-4	-12	5	5	5
Fenthion-Sulfone	75;69	76;79	79;80	0.9931	0.9986	0.9943	5-400	5-400	5-400	-6	-4	-14	5	5	5
Fenthion-Sulfoxide	76;63	69;71	65;72	0.9997	0.9991	0.9911	5-400	5-400	5-400	-9	-10	-15	5	5	5
Flusilazole	99;100	94;98	113;116	0.9984	0.9988	0.9969	5-400	5-400	5-400	-8	0	0	5	5	5
Flutriafol	98;99	101;100	112;106	0.9991	0.9959	0.9986	5-400	5-400	5-400	-9	1	4	5	5	5
Fosthiazate	101;98	108;101	112;113	0.9992	0.9961	0.9989	5-400	5-400	5-400	-2	8	10	5	5	5
Hexaconazole	83;102	118;94	108;114	0.9974	0.9986	0.9965	5-400	5-400	5-400	-4	4	9	5	5	5
Imazalil	78;81	81;81	105;102	0.9975	0.9986	0.9992	5-400	5-400	5-400	8	11	15	5	5	5
Imidacloprid	101;90	105;108	120;120	0.9988	0.9964	0.9935	5-400	5-400	5-400	-7	4	12	5	5	5
Iprodione	n.d;104	n.d;98	n.d;107	0.9992	0.9941	0.9989	50-400	50-400	50-400	-1	-3	-3	50	50	50
Iprovalicarb	111;93	109;99	112;119	0.9990	0.9948	0.9993	5-400	5-400	5-400	-2	15	12	5	5	5
Kresoxim-Methyl	98;96	116;104	114;113	0.9992	0.9934	0.9988	5-400	5-400	5-400	1	16	14	5	5	5
Mandipropamid	103;98	115;98	102;113	0.9992	0.9955	0.9993	5-400	5-400	5-400	-5	8	9	5	5	5
Metconazole	103;95	102;99	108;100	0.9983	0.9991	0.9979	5-400	5-400	5-400	-5	3	3	5	5	5
Methidathion	89;120	120;120	120;113	0.9955	0.9950	0.9929	5-400	5-400	5-400	-7	-18	-57	5	5	5
Methiocarb	103;98	103;100	115;112	0.9993	0.9966	0.9987	5-400	5-400	5-400	-2	2	-3	5	5	5

Methiocarb-Sulfone	99:98	95:101	111;99	1.0000	0.9998	0.9992	5-400	5-400	5-400	-12	-11	-15	5	5	5
Methiocarb-Sulfoxide	85;81	92:96	110:95	0.9988	0.9971	0.9984	5-400	5-400	5-400	-8	-5	-9	5	5	5
Methomyl	118;97	120;103	93;82	0.9994	0.9988	0.9981	5-400	5-400	5-400	-9	-7	-14	5	5	5
Methoxyfenozide	118;88	106;80	106;98	0.9907	0.9995	0.9984	5-400	5-400	5-400	9	7	17	5	5	5
Omethoate	74;80	82;86	70;71	0.9995	0.9998	0.9987	5-400	5-400	5-400	2	5	4	5	5	5
Oxadixyl	111;99	104;101	120;120	0.9985	0.9946	0.9990	5-400	5-400	5-400	-10	0	18	5	5	5
Oxydemeton-Methyl	101;90	98;90	120;94	0.9995	0.9953	0.9985	5-400	5-400	5-400	-17	-8	-16	5	5	5
Paclobutrazol	96;91	98;102	111;109	0.9985	0.9993	0.9953	5-400	5-400	5-400	1	5	8	5	5	5
Parathion	99;93	86;101	120;120	0.9986	0.9970	0.9350	5-400	5-400	5-400	-12	-13	-15	5	5	5
Parathion-Methyl	68;91	114;110	120;120	0.9919	0.9902	0.9995	5-400	5-400	5-400	-5	10	0	5	5	5
Penconazole	89;96	101;100	100;113	0.9982	0.9965	0.9982	5-400	5-400	5-400	-4	-2	-7	5	5	5
Pencycuron	74;72	67;72	81;80	0.9994	0.9964	0.9984	5-400	5-400	5-400	-4	8	11	5	5	5
Pendimethalin	93;95	104;100	120;120	0.9975	0.9998	0.9757	5-400	5-400	5-400	-5	-2	1	5	5	5
Phenthoate	100;92	78;99	120;120	0.9986	0.9986	0.9932	5-400	5-400	5-400	3	9	16	5	5	5
Phosalone	112;102	99;95	112;116	0.9988	0.9956	0.9925	5-400	5-400	5-400	-10	2	2	5	5	5
Phoxim	107;97	93;100	111;120	0.9989	0.9917	0.9853	5-400	5-400	5-400	-2	16	5	5	5	5
Pirimicarb	105;90	92;94	87;94	0.9977	0.9946	0.9915	5-400	5-400	5-400	-7	-1	-22	5	5	5

Pirimiphos-Methyl	87;92	97;98	98;97	0.9989	0.9964	0.9917	5-400	5-400	5-400	-10	-1	2	5	5	5
Prochloraz	88;89	99;94	114;111	0.9982	0.9967	0.9957	5-400	5-400	5-400	4	10	14	5	5	5
Propargite	95;101	104;97	144;140	0.9987	0.9957	0.9889	5-400	5-400	5-400	-9	7	7	5	5	5
Propiconazole	84;99	101;96	119;113	0.9966	0.9972	0.9947	5-400	5-400	5-400	1	12	6	5	5	5
Propoxur	105;101	93;107	111;103	0.9992	0.9945	0.9987	5-400	5-400	5-400	-5	6	0	5	5	5
Propyzamide	104;104	99;112	100;108	0.9978	0.9984	0.9990	5-400	5-400	5-400	3	13	14	5	5	5
Prothiofos	89;85	96;99	129;119	0.9984	0.9952	0.9937	5-400	5-400	5-400	-7	-3	-2	5	5	5
Pyraclostrobin	87;98	106;101	115;118	0.9994	0.9989	0.9961	5-400	5-400	5-400	-1	16	3	5	5	5
Pyrethrins	87;101	105;96	120;120	0.9984	0.9991	0.9839	5-400	5-400	5-400	-8	4	6	5	5	5
Pyridaben	88;96	102;93	120;118	0.9989	0.9986	0.9920	5-400	5-400	5-400	-15	4	2	5	5	5
Pyrimethanil	99;96	97;99	89;102	0.9984	0.9968	0.9983	5-400	5-400	5-400	0	9	0	5	5	5
Pyriproxyfen	94;99	105;99	120;120	0.9979	0.9964	0.9778	5-400	5-400	5-400	-1	9	3	5	5	5
Quinoxyfen	91;96	90;94	120;117	0.9978	0.9981	0.9910	5-400	5-400	5-400	-3	3	-7	5	5	5
Rotenone	95;91	87;102	107;120	0.9954	0.9982	0.9981	5-400	5-400	5-400	1	4	9	5	5	5
Tebuconazole	90;95	102;97	102;103	0.9977	0.9998	0.9977	5-400	5-400	5-400	-11	-2	4	5	5	5
Tebufenpyrad	98;97	104;100	113;115	0.9979	0.9972	0.9842	5-400	5-400	5-400	-6	5	4	5	5	5
Tetraconazole	106;103	106;108	111;115	0.9973	0.9959	0.9970	5-400	5-400	5-400	-3	10	2	5	5	5
Thiodicarb	101;103	99;102	105;115	0.9997	0.9994	0.9973	5-400	5-400	5-400	2	6	4	5	5	5

Thiophanate-Methyl	84;87	54;59	97;104	0.9990	0.9969	0.9987	5-400	50-400	5-400	12	14	15	5	50	5
Tolclofos-Methyl	91;88	112;106	75;119	0.9938	0.9993	0.9949	5-400	5-400	5-400	1	7	-1	5	5	5
Triadimefon	99;98	94;104	116;110	0.9971	0.9935	0.9994	5-400	5-400	5-400	-4	12	7	5	5	5
Triadimenol	95;94	91;99	96;90	0.9992	0.9986	0.9965	5-400	5-400	5-400	-2	-5	-10	5	5	5
Triticonazole	113;98	106;97	112;107	0.9979	0.9948	0.9946	5-400	5-400	5-400	-6	6	8	5	5	5
Zoxamide	109;93	96;98	113;110	0.9973	0.9958	0.9992	5-400	5-400	5-400	-6	6	4	5	5	5

ME (%) = ((slope matrix/slopesolvent)-1)×100