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National Food Institute

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Validation Report 41

Determination of pesticide residues in rapeseeds, rapeseed, rapeseed cake and rapeseed meal by GC-MS/MS

(QuEChERS-µSPE clean-up method)

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CONTENT:

1. Introduction 3
2. Principle of analysis 3
3. Validation 4
4. Results and conclusion 6
5. References 6
Appendix 1 7
Appendix 2. Validation results 11

1. Introduction

Pesticide residues in rapeseed, rapeseed cake, and rapeseed meal were validated using QuEChERS citrate buffer extraction[1] followed by µ-Solid Phase Extraction (µ-SPE) clean-up. A customised workflow was designed and implemented on a stand-alone Thermo Scientific™ TriPlus™ RSH™ multi-purpose autosampler. Mini cartridges containing magnesium sulfate, primary-secondary amine, C_{18}, and graphitized carbon X were used for the automatic clean-up process. Cleaned extracts were diluted with acetonitrile on the robotic system and were analysed on a gas chromatography coupled to a tandem mass spectrometry detector system (GC-MS/MS). The pesticides and/or metabolites included in the validation study are shown in Appendix 3.

2. Principle of analysis

Sample preparation

Samples were each homogenized and blended using an Ultra Centrifugal Mill ZM 200. After homogenization, 2 g test portions were weighted accurately in a 50 mL PP tube and 50 µl. Then, 40 µl of 20 µg L^{-1} of isotopically labelled internal standards (IS) consisting of azoxystrobin-d4, dichlorvos-d6 and etofenprox-d5, were added to all samples prior to extraction. 10 mL of cold water and 10 mL of acetonitrile were added. The tubes were shaked for 1 minute at a speed of 750 rpm on a Geno Grinder 2010 shake. Prepared mixture of salts, containing 4 g $MgSO_4$, 1 g NaCl, 1 g Na_3 citrate dihydrate and 0.5 g Na_2H citrate sesquihydrate, were added to the samples. Tubes were again mechanically shaked for another minute followed by centrifugation for 10 minutes at 4500 rpm. Eight millilitres of supernatant were transferred in a clean tube and placed in -80°C freezer for at least 1 hour. After freezing-out the samples were removed from the freezer, thawed, and centrifuged at 5 °C for 10 minutes at 4500 rpm. One millilitre of clean extract was transferred to a 2 ml glass vial and placed on the stand-alone automated micro SPE for clean-up and dilution. After this step, the extracts were ready to analyse on GC-MS/MS.

µ-SPE clean-up workflow

A stand-alone Thermo Scientific™ TriPlus™ RSH™ multi-purpose autosampler was used for the clean-up. Chromeleon 7 version software was integrated and used to program and operate the

device. The workflow used is similar to the one described by Hakme and Poulsen[2], but a new version was developed where the automatic addition of internal standard and an additional dilution step were added. In brief, the system automatically takes 300 µl of the extract to elute through the µ-SPE column. Later, 10 µl of a 0.2 µg/ml of IS mix consisting of chlorpyriphos-d10 and (TPP) is added as a quality control (QC) standard, prior to GC analysis, which serves to check the stability of the µ-SPE and the GC injection system. Later on, extracts are diluted 1:1 with acetonitrile to make amount of coextracted matrix equal the amount in the calibration standards.

GC-MS/MS parameters

For gas chromatographic separation, a Thermo Scientific™ Trace™ 1310 Gas Chromatograph coupled to a Thermo Scientific™ TriPlus™ RSH autosampler was used. The samples were injected in a programmable temperature vaporizer (PTV) mode through a PTV baffle liner 2×2.75×120 mm for Thermo GCs (Siltek). The injection volume was 1 µL and the injection temperature was set to 70°C. Helium as used as carrier gas at a flow of 1.2 ml.min^{-1}. The analytes were separated on a TG-5SILMS (capillary column of 30 m long, 0.25 mm inner diameter and a film thickness of 0.25 µm). The oven temperature program was as follows: 60°C for 1.5 min, up to 90°C at 25°C/min for 1.5 min, up to 180°C at 25°C /min, then up to 280°C at 5 °C/min and finally up to 300°C at 10°C/min and for 12 min. The total runtime was 42 min. For the mass spectrometric analysis, a Thermo Scientific™ TSQ™ 8000 Evo was used. The instrument has been upgraded with and Advanced Electron ionisation source, (AEI). The AEI source was operated with an electron energy of 50 eV. The source temperature was set at 300°C, and the transfer line, at 280°C. The analyses were performed by a triple quadrupole operating in the MRM mode (Multiple Reaction Monitoring). Retention time (Rt), precursor mass, product mass, and collision energy (CE) for each of the studied compounds are shown in Appendix 1.

3. Validation

Validation design

The QuEChERS-µSPE clean-up method was validated for 132 compounds (pesticides or/and metabolites) in four different matrices (wheat, rice, rye, and oat). The validation was performed on

5-6 replicates at each of the four cereals matrices, and at three spiking levels of 0.005, 0.01 and 0.05 mg/kg. Extraction of a blank sample were included for all commodities.

Calibration curves and linearity

Linearity study was performed by using matrix-matched calibration curve prepared in 5 concentrations for each one of the compounds within the range of 0.33 to 100 µg/L. The calibration curves were fitted to linear function and the deviation of the back–calculated concentration of the calibration standards from the true concentrations were within ±20%.

All quantifications were performed using bracketing matrix matched calibration curves.

Specificity

The ion ratios for sample extracts were within ±30% (relative) of average of relevant calibration standards from same sequence. The ion ratios may vary slightly depending on concentration level and in some cases the average of calibration standard was based on the lower calibration levels for the low spike samples.

Accuracy – Recovery

Recovery values were calculated as average recovery of 5-6 replicates for each level (0.005, 0.01, and 0.05 mg/kg) and matrices. Accepted recovery range was between 70 and 120% (following SANTE document)[3]. Values outside this range have been accepted if the precision data was satisfactory.

Precision – repeatability and internal reproducibility

Repeatability and internal reproducibility were calculated for all pesticides and degradation products on all four spiking levels (0.005, 0.01 and 0.05 mg/kg) as given in ISO 5725-22. Accepted values were ≤ 20%.

Limit of quantification, LOQ

The Limit of quantification (LOQ) was determined as the lowest spiked level for which the acceptance criteria were met (average relative recovery between 70 and 120% and precision lower

than or equal to 20%), and ion ratios for sample extracts were within ±30% (relative) of average of relevant calibration standards.

4. Results and conclusion

A total of 132 compounds were successfully validated using QuEChERS-µSPE clean-up method. All the validation data for pesticides and/or metabolites and three different matrices are presented in appendix 2.

The lowest LOQ of 0.005 mg/kg was achieved for 102 compounds. An LOQ of 0.01 mg/kg was achieved for 20 compounds and 10 compounds could be validate only at 0.05 mg/kg. Acrinathrin, carbosulfan, flufenoxuron, methacrifos, methamidophos, oxycarboxin, phosmet, pyrazophos and simazine could not be validated.

Deltamethrin could be validated only in rapeseed meal at 0.01 mg/kg. Bromuconazole, cyhalothrin-lambda, cypermethrin, hexaconazole, iprodione, lindane, phosalone, pirimicarb-desmethyl could be validated at 0.05 mg/kg and only in rapeseed cake and rapeseed meal. These compounds could not be validated in rapeseed. Monocrotophos could be validated at 0.05 mg/kg in rapeseed and rapeseed cake but to in rapeseed meal.

5. References

1. EN 15662:2008. Foods of plant origin - Determination of pesticide residues using GC-MS and/or LC-MS/MS following acetonitrile extraction/partitioning and clean-up by dispersive SPE - QuEChERS-method

2. Hakme E., & Poulsen, M.E (2021). Evaluation of the automated micro-solid phase extraction clean-up system for the analysis of pesticide residues in cereals by gas chromatography-Orbitrap mass spectrometry. Journal of Chromatography A, 1652 (562384). DOI: 10.1016/j.chroma.2021.462384

3. Guidance document on analytical quality control and method validation procedures for pesticide residues and analysis in food and feed, Document SANTE/ 11312 /2021.

Appendix 1

Retention time (Rt), precursor mass, product mass, and collision energy (CE) of the studied compounds

Pesticide	Rt	Precursor	Product	CE	Precursor	Product	CE
2-phenylphenol	9.7	170	115	35	170	141	25
Aldrin	14.8	262.7	192.9	32	292.9	257.9	10
Atrazine	11.7	200	122	8	215	173	10
Azoxystrobin	29.9	344.1	156	34	344.1	171.9	36
Bifenthrin	21.8	181	165.9	10	181.1	153.1	10
Bitertanol	25.1	170	115.1	34	170	141.1	20
Boscalid	26.9	139.9	76	22	139.9	112	10
Bromophos-ethyl	16.5	302.7	284.8	14	358.9	302.9	15
Bromopropylate	21.9	184.9	75.5	30	340.8	185	14
Bromuconazole	21.6	172.9	144.9	16	293	173	10
Bupirimate	17.8	208.1	165	12	273.1	193.2	8
Cadusafos	11	159	96.9	16	159	130.9	8
Carboxin	17.9	87	43	6	143	43	16
Chlorfenapyr	18.1	247	227	15	248.9	112	24
Chlorfenson	17.3	174.9	111	10	302	175	10
Chlorfenvinphos	15.9	266.9	159	16	269	161	15
Chlormephos	9.1	154	65	16	154	121	5
Chlorobenzilate	18.6	139	74.9	26	139	111	12
Chlorpropham	10.8	171	127	8	213	127	8
Chlorpyrifos	14.5	196.7	168.9	12	313.9	257.9	12
Chlorpyrifos-methyl	13.4	285.9	93	20	286	271	15
Clofentezine	24.5	102	50.9	12	102	74.9	12
Clomazone	11.8	125	89	16	138	74.9	24
Cyflutrin	26.5	206	151	12	226	206	10
Cyhalothrin-lambda	23.59	180.9	151.9	22	197	141.1	10
Cypermethrin	27	163	127	10	181	152	20
Cyproconazole	18.4	222	82.1	10	222	89.3	38
Cyprodinil	15.6	224.1	196.9	20	224.1	208	18
Deltamethrin_cis(I+II)	29.7	181	152.1	22	252.8	92.9	16

Demeton-S-methyl	10.5	88	59.8	6	109	79	6
Diazinon	12.1	137.1	84.1	12	199	93	15
Dichlorvos	7.6	185	93	12	220	185	10
Dicloran	11.7	160	124.1	8	176	148	12
Dicofol-pp	20.6	111	74.9	14	139	111	12
Dieldrin	17.8	262.8	192.9	30	276.9	206.9	20
Difenoconazole(I+II)	29.4	265	138.9	60	324.9	267	10
Dimethomorph(I)	30.1	301	139	14	301	165.1	10
Dimethomorph(II)	30.6	301	139	14	301	165.1	10
Diphenylamine	10.7	168.1	139	38	168.1	167.1	14
Disulfoton	12.4	185.9	96.9	16	186	153	5
Endosulfan-alpha	17	158.9	123	12	194.7	125	22
Endosulfan-beta	18.8	158.9	123	12	194.7	125	22
Endosulfan-sulfate	20.1	238.7	203.9	12	271.7	234.9	12
Endrin	18.4	245	173	22	262.8	192.9	30
EPN	21.8	157	77	22	169	77	22
Epoxiconazole	21.2	165	138	8	192	111	22
Ethion	19	153	97	10	230.9	128.9	22
Ethoprophos	10.6	157.9	96.9	16	157.9	113.9	6
Ethoxyquin	11.6	174.1	131.2	18	174.1	146.1	12
Etofenprox	27.5	163.1	77.1	32	163.1	107.1	16
Fenarimol	24	139	74.9	26	139	111	14
Fenazaquin	22.5	145.1	91	24	145.1	117.1	12
Fenbuconazole	26.2	129	77.8	18	129	102	14
Fenitrothion	14.2	125	79	8	277	109	16
Fenoxycarb	22	116	44.1	16	116	88	8
Fenpropathrin	22.1	181	126.8	28	181	151.9	22
Fenpropimorph	14.8	128.1	70.1	12	128.1	110.1	8
Fenson	15.2	141	77	8	268	77	20
Fenthion	14.7	245.3	125	12	278	109	15
Fenvalerate I+	29	125	89	18	167	89	32
Fluazifop-p-butyl	18.3	282	91.1	18	282	238.1	16
Fludioxonil	17.4	153.7	127	8	248	127	26
Fluquinconazole	25.4	340	108.1	36	340	298	16
Flutriafol	17.1	123	75	24	123	95	12
Fenvalerate II	28.9	180.8	152.1	22	208	181	15
HCH-alpha	11.3	180.9	145	8	182.8	146.7	12

HCH-beta	11.9	180.9	145	14	218.7	146.6	18
Heptenophos	10	124	62.9	28	124	89	12
Hexaconazole	17.2	213.9	123.5	28	213.9	159	18
Hexythiazox	16.7	184	59	20	184	149	6
Indoxacarb	29.5	203	106.1	22	203	134	20
Iprodione	21.6	314	245	10	315.7	247	10
Iprovalicarb(I)	17.8	118.9	117.1	8	134.1	42	20
Iprovalicarb(II)	17.9	118.9	117.1	8	134.1	42	20
Isofenphos-methyl	15.4	199	65	34	199	121	10
Isoprothiolane	17.4	204	85	28	204	118	8
Jodofenfos	17.3	125	47	12	376.8	361.8	16
Kresoxim-methyl	17.8	130.9	130.1	10	206	116	4
Lindane	12	180.9	109	26	180.9	145	14
Linuron	14.5	159.8	133	12	187	124	20
Metalaxyl	13.7	131.9	117	12	160.1	130	18
Methidathion	16.5	145	58	14	145	85	6
Methoxychlor	22	227.1	141.1	32	227.1	169.1	22
Metribuzin	13.3	198	82.1	16	198	110	10
Mevinphos	8.9	127	95	14	127	109	10
Monocrotophos	11	127	95	14	127	109	10
Myclobutanil	17.7	179	90	28	179	125	14
Nuarimol	20.5	139	111	12	235	139	14
Ofurace	19.6	232.1	158.1	18	232.1	186.1	8
Oxadixyl	18.8	163.1	117	24	163.1	132.1	8
Paclobutrazol	16.7	125	89	18	236	125	12
Parathion	14.8	235	139	8	291	81	20
Parathion-methyl	13.5	124.9	47	12	124.9	79	6
Penconazole	15.8	158.9	89	28	248	157	22
Pencycuron	11.8	125	89	16	125	99	16
Pendimethalin	15.6	252.1	161	14	252.1	162	8
Permethrin(I+II)	25.3	183.1	153	12	183.1	165.1	12
Phenthoate	16	246	121	8	274	121	10
Phosalone	23	182	74.8	30	182	111	14
Pirimicarb	12.7	166.1	55	18	166.1	96	12
Pirimicarb-desmethyl	13	152.1	42	25	152.1	96	10
Pirimiphos-methyl	14.1	290.1	125	20	290.1	233	8
Prochloraz	25.5	180.1	138.1	12	310	268	5

Procymidone	16.2	95.9	67.1	8	283	96.1	8
Profenofos	17.5	296.7	268.9	10	336.9	266.9	12
Propiconazole(I+II)	20.1	172.9	74	38	172.9	109	26
Propoxur	10.3	110	62.9	24	110	64.1	16
Propyzamide	12.1	172.9	74	38	172.9	109	26
Prothiofos	17.4	266.7	220.9	18	266.7	238.9	8
Pyridaben	25.4	147.1	117.1	20	147.1	119.1	8
Pyridaphenthion	21.5	199	77.1	24	199	92.1	14
Pyrimethanil	12.4	198.1	117.9	30	198.1	157.6	18
Pyriproxyfen	23.3	136.1	78	20	136.1	96	10
Quinoxyfen	20.1	237	208	26	271.8	237.1	12
Tebuconazole	20.6	125	89	16	125	99	16
Tebufenpyrad	22.4	276.1	171	10	318.1	131.1	14
Tecnazene	10.3	214.8	143.6	20	214.8	178.7	10
Tefluthrin	12.4	177	127	14	177	137	16
Tetraconazole	14.9	100.9	51	10	171	136	10
Tetradifon	22.8	159	74.8	32	159	111	20
Thiometon	11.5	125	79	8	158	125	10
Tolclofos-methyl	13.6	265	219.9	20	265	250	12
Triadimefon	14.8	208	111	20	208	126.7	12
Triadimenol	16.3	128	65	18	128	100	10
Triallate	12.6	86.1	43.3	6	268	183.9	18
Triazophos	19.5	161	105.7	12	161	134.1	8
Trichlorfon	7.6	145	109	10	185	93	12
Tricyclazole	17.4	162	84.9	18	162	133.9	8
Trifloxystrobin	19.9	116.1	89	8	186	145	10
Trifluralin	10.7	306.1	159.7	20	306.1	206	10
Triticonazole	23	217	167	18	235.1	181.9	12
Vinclozolin	13.4	198	145	15	285	212	5

Appendix 2. Validation results

Recoveries (Rec), repeatability (RSD_r), internal reproducibility (RSDR), expanded uncertainty (U) without correcting for recoveries and Limit of Quantification (LOQ) for pesticides validated on rapeseed (RS), Rapeseed cake (RSC) and Rapeseed meal (RSM) using QuEChERS-µSPE clean-up method.

Pesticide	Spike level 0.005 mg/kg					Spike level 0.01 mg/kg					Spike level 0.05 mg/kg					LOQ (mg/kg)	Matrices
	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %		
2-phenylphenol	88	7	37	80	38	96	5	19	41	20	96	5	8	18	8	0.005	RS,RSC,RSM
Aldrin	43	14	38	138	39	47	11	21	115	22	46	7	31	125	32	0.005	RS,RSC,RSM
Atrazine	91	11	29	62	30	96	8	17	36	17	95	6	6	16	6	0.005	RS,RSC,RSM
Azoxystrobin						81	21	22	59	23	100	6	7	14	7	0.01	RS[1],RCS[1],RSM[1]
Bifenthrin	58	13	26	100	26	65	7	15	76	15	57	8	34	110	35	0.005	RS,RSC,RSM
Bitertanol	94	11	11	26	12	105	6	17	36	17	96	5	7	17	7	0.005	RS,RSC,RSM
Boscalid	80	8	18	55	19	88	8	13	36	13	98	5	7	16	7	0.005	RS RSC,RSM
Bromophos-ethyl	72	10	30	84	31	74	7	21	68	22	66	6	25	86	26	0.005	RS,RSC,RSM
Bromopropylate	88	10	22	51	23	87	7	12	36	12	81	7	12	44	12	0.005	RS,RSC,RSM
Bromuconazole											109	6	13	33	13	0.05	RSC[2],RSM[2]
Bupirimate	88	8	17	42	18	96	10	11	24	11	97	5	6	14	6	0.005	RS,RSC,RSM
Cadusafos	89	7	22	50	22	92	10	12	30	12	93	5	5	19	5	0.005	RS,RSC,RSM
Carboxin	91	7	17	39	17	95	12	14	30	15	98	4	12	24	12	0.005	RS,RSC,RSM
Chlorfenapyr						93	17	24	52	25	93	8	12	28	12	0.01	RSC[1],RSM[1]
Chlorfenson	68	9	15	71	16	82	9	9	41	9	86	5	8	31	8	0.005	RS,RSC,RSM

Pesticide	Spike level 0.005 mg/kg					Spike level 0.01 mg/kg					Spike level 0.05 mg/kg					LOQ (mg/kg)	Matrices
	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %		
Chlorfenvinphos	108	5	18	40	18	97	13	15	32	16	95	5	9	21	9	0.005	RS,RSC,RSM
Chlormephos	81	21	23	60	23	90	11	11	29	11	96	9	11	23	11	0.005	RS,RSC,RSM
Chlorobenzilate	79	11	15	53	16	88	8	9	31	10	93	5	8	22	8	0.005	RS,RSC,RSM
Chlorpropham	82	20	36	82	37	90	11	14	35	14	97	6	7	15	7	0.005	RS,RSC,RSM
Chlorpyrifos	78	12	23	65	24	81	13	17	51	17	83	10	14	45	15	0.005	RS,RSC,RSM
Chlorpyrifos-methyl						89	17	19	45	20	94	8	18	38	18	0.01	RS[1],RCS[1],RSM[1]
Clofentezine	120	10	53	116	54	94	29	46	96	48	98	12	40	81	41	0.005	RS,RSC,RSM
Clomazone	83	8	14	44	15	93	7	8	21	8	95	5	7	17	7	0.005	RS,RSC,RSM
Cyflutrin	62	17	19	86	20	69	30	32	90	33	97	6	26	53	27	0.005	RS,RSC,RSM
Cyhalothrin-lambda											72	18	48	114	50	0.05	RSM2
Cypermethrin											89	6	21	49	22	0.05	RSC2,RSM2
Cyproconazole	78	13	17	56	18	94	6	16	35	16	101	4	12	24	12	0.005	RS[1],RSC,RSM
Cyprodinil						63	21	21	87	22	82	8	12	43	12	0.01	RS[1],RCS[1],RSM[1]
Deltamethrin_cis (I+II)						55	18				103	11	12	27	13	0.01	RSM1
Demeton-S-methyl	90	9	14	35	15	91	15	14	34	15	95	5	12	26	12	0.005	RS,RSC,RSM
Diazinon	85	12	19	49	19	89	12	12	32	12	88	5	7	29	8	0.005	RS,RSC,RSM
Dichlorvos	73	12	27	77	28	80	15	18	54	18	89	9	15	38	16	0.005	RS,RSC,RSM
Dicloran	75	15	15	60	16	87	9	9	32	9	95	5	8	20	9	0.005	RS,RSC,RSM
Dicofol-pp	91	8	17	39	18	97	8	9	19	9	100	5	6	13	6	0.005	RS,RSC,RSM

Pesticide	Spike level 0.005 mg/kg					Spike level 0.01 mg/kg					Spike level 0.05 mg/kg					LOQ (mg/kg)	Matrices
	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %		
Dieldrin						73	19	24	74	25	72	7	13	62	13	0.01	RSC[1],RSM[1]
Difenoconazole (I+II)	88	7	7	28	7	91	9	9	25	9	95	5	6	15	6	0.005	RS,RSC,RSM
Dimethomorph (I)	91	8	16	38	16	95	9	11	24	11	99	5	6	13	7	0.005	RS,RSC,RSM
Dimethomorph (II)	88	7	17	43	18	95	7	12	27	13	100	5	6	13	7	0.005	RS,RSC,RSM
Diphenylamine	80	11	19	57	20	84	8	12	40	13	84	6	6	34	6	0.005	RS,RSC,RSM
Disulfoton	74	20	23	71	24	82	10	13	45	14	82	6	12	43	12	0.005	RS,RSC,RSM
Endosulfan-alpha	64	21	43	114	44	75	11	24	70	24	71	9	29	83	29	0.005	RS,RSC,RSM
Endosulfan-beta						88	10	13	37	14	94	9	12	27	12	0.01	RS[1],RCS[1],RSM[1]
Endosulfan-sulfate						71	16	22	75	24	80	11	26	68	27	0.01	RSC[1],RSM[1]
Endrin	67	13	15	73	15	75	10	13	57	14	71	6	16	66	17	0.005	RS,RSC,RSM
EPN	77	20	29	77	30	86	20	19	49	20	109	7	20	45	21	0.005	RS[1],RSC,RSM
Epoxiconazole	86	10	20	49	20	94	7	12	27	12	99	4	6	12	6	0.005	RS,RSC,RSM
Ethion	85	7	12	38	12	84	16	16	46	17	93	6	16	35	16	0.005	RS,RSC,RSM
Ethoprophos	84	12	17	47	17	92	8	8	24	8	97	6	6	14	6	0.005	RS,RSC,RSM
Ethoxyquin	112	5	19	46	20	91	7	14	34	14	72	5	14	64	15	0.005	RS,RSC,RSM
Etofenprox	62	9	23	89	23	67	5	15	73	15	58	7	28	102	29	0.005	RS,RSC,RSM
Fenarimol	96	7	9	19	9	101	7	9	19	10	97	3	3	9	4	0.005	RS,RSC,RSM
Fenazaquin	60	12	34	107	35	67	9	20	78	21	64	6	17	79	17	0.005	RS,RSC,RSM
Fenbuconazole	91	6	14	35	14	97	6	10	21	10	100	5	5	10	5	0.005	RS,RSC,RSM

Pesticide	Spike level 0.005 mg/kg					Spike level 0.01 mg/kg					Spike level 0.05 mg/kg					LOQ (mg/kg)	Matrices
	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %		
Fenitrothion	126	8	16	61	17	108	13	13	32	14	109	7	19	43	20	0.005	RSC,RSM
Fenoxycarb	87	11	16	41	16	88	9	9	31	9	94	6	10	24	10	0.005	RS, RSC,RSM
Fenpropathrin						109	14	33	71	34	87	8	11	35	11	0.01	RSC[1], RSM[1]
Fenpropimorph	65	7	13	75	13	64	6	12	76	13	62	11	11	80	11	0.005	RS, RSC,RSM
Fenson	84	6	24	59	25	93	9	11	28	12	95	9	8	19	8	0.005	RS,RSC,RSM
Fenthion	99	8	21	44	22	98	10	11	23	12	102	8	11	23	11	0.005	RS,RSC,RSM
Fenvalerate(I+II)	68	17	28	86	29	71	13	28	82	29	94	9	30	64	31	0.005	RS[2],RSC,RSM
Fluazifop-p-butyl	85	11	14	42	14	90	10	13	34	14	94	5	9	22	9	0.005	RS,RSC,RSM
Fludioxonil	78	12	18	58	19	92	11	14	33	15	101	5	7	14	7	0.005	RS,RSC,RSM
Fluquinconazole	98	6	18	36	18	108	5	9	24	10	111	6	11	32	11	0.005	RS,RSC,RSM
Flutriafol	92	11	17	40	18	99	13	12	25	12	105	6	8	19	8	0.005	RS,RSC,RSM
Fluvalinate-tau(I+II)						66	21	69	160	72	70	13	59	136	61	0.01	RCS[1],RSM[1]
HCH-alpha	92	15	28	60	29	93	11	21	45	21	98	9	15	32	16	0.005	RS,RSC,RSM
HCH-beta	86	12	22	52	22	93	8	9	24	10	88	6	13	35	13	0.005	RS,RSC,RSM
Heptenophos	106	6	18	39	18	95	13	15	33	16	96	6	9	21	10	0.005	RS,RSC,RSM
Hexaconazole											89	9	13	35	13	0.05	RSC2,RSM2
Hexythiazox	86	10	18	46	19	87	5	14	39	15	78	6	18	58	19	0.005	RS,RSC,RSM
Indoxacarb											112	16	31	69	32	0.05	RS[2],RSC[2],RSM[2]
Iprodione											138	14	60	145	62	0.05	RSC2,RSM2

Pesticide	Spike level 0.005 mg/kg					Spike level 0.01 mg/kg					Spike level 0.05 mg/kg					LOQ (mg/kg)	Matrices
	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %		
Iprovalicarb(I)	84	20	32	74	33	91	13	22	49	22	105	6	10	23	11	0.005	RS,RSC,RSM
Iprovalicarb(II)						99	20	20	41	20	107	6	12	27	12	0.01	RS[1],RCS[1],RSM[1]
Isofenphos-methyl	81	8	15	49	15	90	9	10	28	10	97	7	7	16	7	0.005	RS,RSC,RSM
Isoprothiolane	88	9	15	40	16	97	8	9	19	9	99	4	3	7	3	0.005	RS,RSC,RSM
Jodofenfos	113	16	57	120	58	95	19	34	70	35	97	4	27	56	28	0.005	RS,RSC,RSM
Kresoxim-methyl						82	14	15	48	15	94	6	7	18	7	0.01	RSC[1],RSM[1]
Lindane											84	17	55	118	57	0.05	RSC2,RSM2
Linuron						94	14	21	44	21	96	6	17	36	18	0.01	RS[1],RCS[1],RSM[1]
Metalaxyl	79	20	25	68	26	95	13	21	43	21	105	7	9	22	9	0.005	RS[1], RSC,RSM
Methidathion	136	10	42	112	43	111	23	31	67	32	144	8	50	135	51	0.005	RS, RSC,RSM
Methoxychlor	68	11	11	67	11	81	14	14	49	15	92	6	16	36	16	0.005	RS, RSC,RSM
Metribuzin	134	14	22	82	23	112	10	17	43	18	101	12	25	52	26	0.005	RS,RSC,RSM
Mevinphos	111	10	50	106	52	84	21	46	99	47	81	7	27	67	28	0.005	RS,RSC,RSM
Monocrotophos											112	8	50	107	52	0.05	RS[2], RSC[2]
Myclobutanil	87	8	15	42	16	98	7	9	19	10	102	4	7	15	7	0.005	RS, RSC,RSM
Nuarimol	87	9	17	44	17	94	8	10	24	10	98	5	6	13	6	0.005	RS, RSC,RSM
Ofurace	108	14	36	75	37	121	9	14	51	15						0.005	RS, RSC,RSM
Oxadixyl	88	6	20	47	20	98	7	12	24	12	106	5	11	26	12	0.005	RS,RSC,RSM
Paclobutrazol	87	6	11	34	11	99	8	10	21	10	99	5	5	10	5	0.005	RS,RSC,RSM

Pesticide	Spike level 0.005 mg/kg					Spike level 0.01 mg/kg					Spike level 0.05 mg/kg					LOQ (mg/kg)	Matrices
	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %		
Parathion	101	12	24	49	24	96	15	16	34	16	106	10	14	32	15	0.005	RS,RSC,RSM
Parathion-methyl						99	11	11	24	12	111	9	24	54	25	0.01	RSC[1],RSM[1]
Penconazole	94	7	14	31	14	96	5	6	15	6	94	5	5	15	5	0.005	RS,RSC,RSM
Pencycuron	83	8	14	44	15	93	7	8	21	8	95	5	7	17	7	0.005	RS,RSC,RSM
Pendimethalin	80	11	17	54	17	81	7	8	42	8	78	7	14	53	15	0.005	RS,RSC,RSM
Permethrin(I+II)	60	21	68	162	70	63	18	51	129	53	55	10	62	157	64	0.005	RS[2],RSC,RSM
Phenthoate	128	11	28	80	29	100	15	28	57	29	110	5	22	50	23	0.005	RS,RSC,RSM
Phosalone											144	12	48	132	49	0.05	RSC[2],RSM[2]
Pirimicarb	84	10	15	44	15	92	10	14	33	14	97	5	11	22	11	0.005	RS,RSC,RSM
Pirimicarb-desmethyl											106	15	40	83	41	0.05	RSC2,RSM2
Pirimiphos-methyl						90	9	10	28	10	94	8	11	25	11	0.01	RS[1],RCS[1],RSM[1]
Prochloraz	78	21	43	98	44	78	20	35	84	36	87	6	8	32	9	0.005	RS,RSC,RSM
Procymidone	78	11	26	69	27	93	7	13	31	14	100	7	6	13	6	0.005	RS,RSC,RSM
Profenofos	86	14	26	60	26	83	22	22	55	22	93	6	22	47	22	0.005	RS,RSC,RSM
Propiconazole(I+II)	76	8	22	66	22	89	7	12	33	12	98	6	7	15	7	0.005	RS,RSC,RSM
Propoxur	118	17	18	52	18	87	11	25	59	26	86	10	32	72	33	0.005	RSC,RSM
Propyzamide	84	16	15	45	16	91	11	11	29	11	92	5	9	24	9	0.005	RS, RSC,RSM
Prothiofos	68	9	32	91	33	71	10	24	77	25	62	7	27	94	28	0.005	RS, RSC,RSM
Pyridaben	87	10	17	43	17	85	5	7	33	7	73	6	19	66	19	0.005	RS, RSC,RSM

Pesticide	Spike level 0.005 mg/kg					Spike level 0.01 mg/kg					Spike level 0.05 mg/kg					LOQ (mg/kg)	Matrices
	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %		
Pyridaphenthion	122	15	15	55	16	114	15	17	44	17	111	7	18	45	19	0.005	RSC,RSM
Pyrimethanil	89	9	14	35	14	90	7	12	33	13	85	5	10	36	11	0.005	RS,RSC,RSM
Pyriproxyfen	90	9	11	31	12	84	8	9	37	10	76	8	15	57	16	0.005	RS,RSC,RSM
Quinoxyfen	81	5	19	54	19	77	6	15	55	15	71	6	18	69	18	0.005	RS,RSC,RSM
Tebuconazole	92	11	11	28	12	91	7	10	28	10	97	6	7	16	7	0.005	RS,RSC,RSM
Tebufenpyrad	69	11	20	75	21	84	8	10	38	10	88	5	8	29	8	0.005	RS,RSC,RSM
Tecnazene	80	11	24	63	24	82	10	17	49	17	77	8	15	55	15	0.005	RS,RSC,RSM
Tefluthrin	91	8	13	31	13	88	5	12	34	12	72	6	20	70	21	0.005	RS,RSC,RSM
Tetraconazole						105	9	21	45	22	105	9	9	20	9	0.01	RS[1],RCS[1],RSM[1]
Tetradifon						96	8	15	31	15	86	5	9	33	9	0.01	RS[1],RCS[1],RSM[1]
Thiometon						86	19	18	47	19	87	9	11	35	12	0.01	RS[1],RCS[1],RSM[1]
Tolclofos-methyl	91	8	9	25	9	88	9	9	30	9	94	8	11	25	11	0.005	RS,RSC,RSM
Triadimefon	89	13	23	53	24	99	8	11	23	11	103	8	7	16	8	0.005	RS,RSC,RSM
Triadimenol						96	12	11	24	12	91	13	23	51	24	0.01	RS[2],RSC[1],RSM[1]
Triallate	60	17	40	114	41	65	11	20	80	20	69	7	16	71	17	0.005	RS,RSC,RSM
Triazophos	96	16	22	46	23	96	12	21	44	21	107	5	23	50	24	0.005	RSC,RSM
Trichlorfon	75	11	28	77	29	80	15	18	55	19	89	9	15	38	16	0.005	RS,RSC,RSM
Tricyclazole	102	8	33	68	34	102	8	18	38	19	100	4	4	9	4	0.005	RS,RSC,RSM
Trifloxystrobin						61	20				81	7	30	73	31	0.01	RCS[2], RSM[1]

Pesticide	Spike level 0.005 mg/kg					Spike level 0.01 mg/kg					Spike level 0.05 mg/kg					LOQ (mg/kg)	Matrices
	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %	Rec %	RSDr %	RSDR %	U %	Cu %		
Trifluralin	87	7	20	50	21	89	6	14	37	15	84	9	15	45	15	0.005	RS, RSC,RSM
Triticonazole	92	12	19	43	20	93	7	12	27	12	102	6	10	20	10	0.005	RS, RSC,RSM
Vinclozolin	82	14	19	54	20	88	7	10	32	10	97	5	6	13	6	0.005	RS, RSC,RSM

[1] **LOQ = 0.01 mg/kg**

[2] **LOQ = 0.05 mg/kg**