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Application of ACQUITY TQD for the Analysis of Pesticide Residues in Baby Food

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Abstract

This application note assesses the suitability of the Waters ACQUITY TQD for tandem quadrupole-based analysis of pesticide residue in baby food. Polarity switching, differing dwell times and ion ratio robustness will also be assessed.

Introduction

The European Union residue monitoring program, 2005–2007, establishes the need to cover 55 active ingredients in various foods, including baby foods.[1,2] Twenty of these pesticides are suitable for multi-residue LC-MS analysis; only one has a negative polarity in electrospray mode, normally requiring two injections (one in each polarity ion mode). Consequently compounds with negative polarity are often excluded from monitoring programs. Ideally, these should be determined in a single analysis with polarity switching.

Chemists analyzing pesticide residues are under increasing pressure to broaden the range of pesticides determined in a single analysis: to improve limits of detection, precision and quantitation; to increase confidence in the validity of residue data; to provide faster methods and to reduce usage of hazardous solvents while maintaining or reducing costs. In order to meet these demanding requirements, the scope, sensitivity, efficiency and speed of multi-residue methods of analysis must be improved.

The introduction of the ACQUITY TQD featuring the TQ detector (Figure 1) allows scientists to perform UPLC analysis of pesticides while harnessing all the benefits that this new tandem quadrupole instrument brings to the laboratory. With IntelliStart technology, the instrument is designed to remove the burden of complicated operation, time-intensive troubleshooting and upkeep. Its small footprint will give any laboratory an advantage as this powerful tool removes the need for larger instrumentation.

Figure 1. The Waters ACQUITY TQD with the TQ Detector.

Experimental

The sample extraction method has been previously reported.[3] Extracts of blank baby food matrix in acetonitrile were provided along with mixtures of the compounds in acetonitrile by the Central Science Laboratory (CSL), York, UK. Extracts for injection were prepared by spiking the compounds into the baby food matrix. The supernatant was analyzed on the ACQUITY TQD following dilution with water (1:9) v/v.

UPLC Conditions

LC system:	Waters ACQUITY UPLC System
Column:	ACQUITY UPLC BEH C_{18} Column 2.1 x 50 mm, 1.7 µm

Column temp.:	40 °C
Flow rate:	600 µL/min
Mobile phase A:	Water
Mobile phase B:	Methanol
Total run time:	7 min
Injection volume:	20 µL

Gradient

Time 0 min	90% A
Time 4 min	100% B
Time 5 min	100% B

MS Conditions

MS system:	Waters TQ detector
Ionization mode:	ESI positive and negative
Switching time:	0.02 s
Capillary voltage:	2500 V
Cone voltage:	35 V
Desolvation gas:	Nitrogen, 800 L/Hr, 400 ˚C
Cone gas:	Nitrogen, 50 L/Hr

Source temp.:	140 °C
Acquisition:	Multiple Reaction Monitoring (MRM)
Collision gas:	Argon at 4.0 x 10^{-3} mBar

The ACQUITY TQD was tuned so that the precursor and product ions were resolved with a peak width at half height of less than 0.7 Da. The list of pesticide residues and the MRM transitions, along with the retention times, dwell times, cone voltages and collision energies for the method are shown in Appendix 1. Pesticide residues listed in red were acquired in negative ion mode.

Acquisition and Processing Methods

Waters MassLynx Software v4.1 was used for acquisition and its TargetLynx application manager used for data processing.

Results and Discussion

All compounds were separated successfully. Figure 2 shows the Total Ion Chromatogram (TIC) for all compounds.

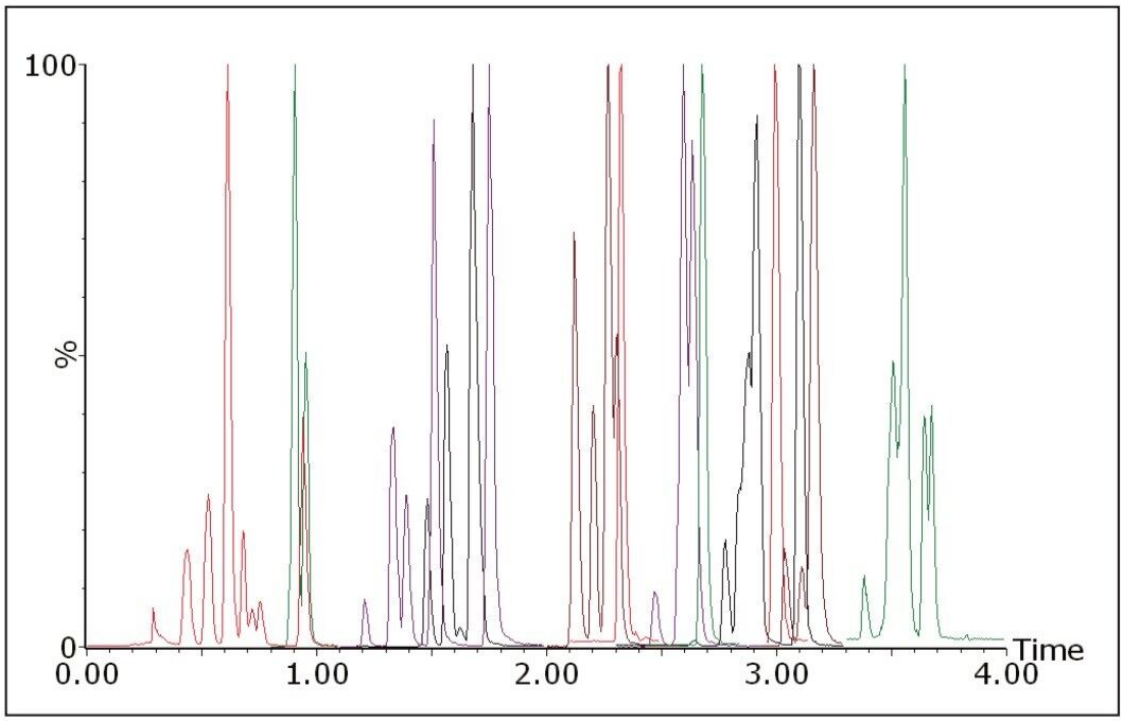

Figure 2. Positive, negative switching TIC for all pesticides.

The polarity switching capability of the ACQUITY TQD is demonstrated when comparing two co-eluting compounds, lenacil (negative ion compound) and phorate sulfone (positive ion compound). Figure 3 shows the chromatography achieved with 12 data points over each peak. Both compounds show good linearity and give good correlation coefficients (r^2), illustrated in Figure 4, while using the 20 millisecond inter-scan delay for polarity switching.

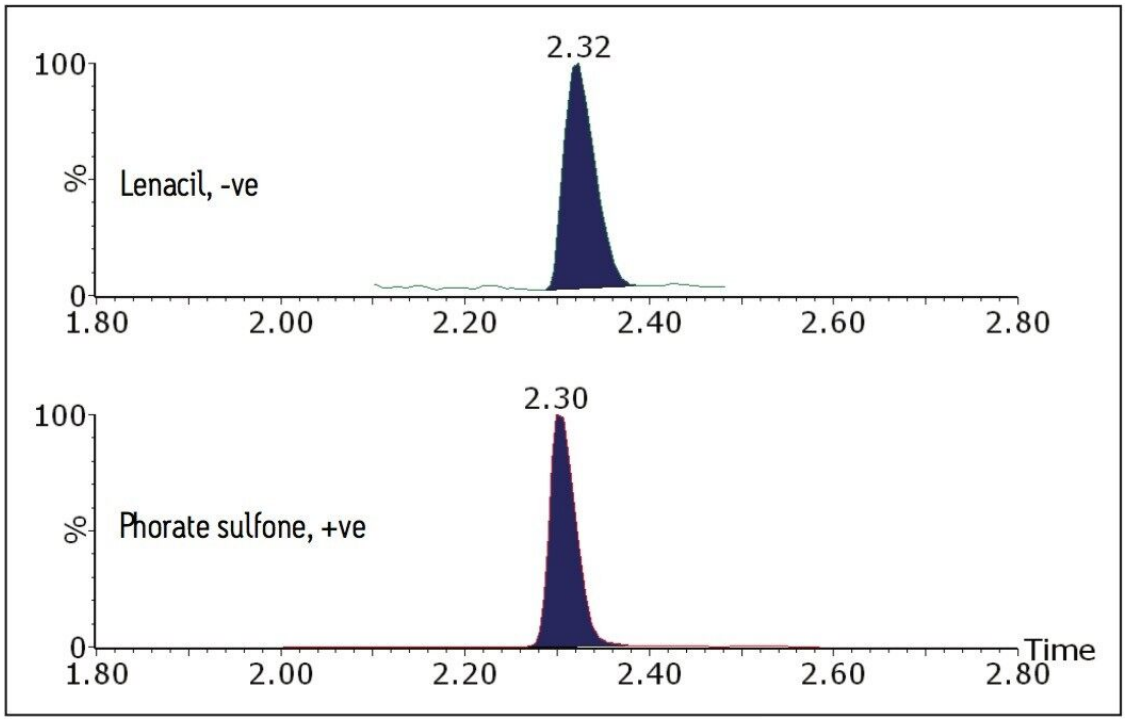

Figure 3. Chromatography achieved by two co-eluting compounds undertaking polarity switching.

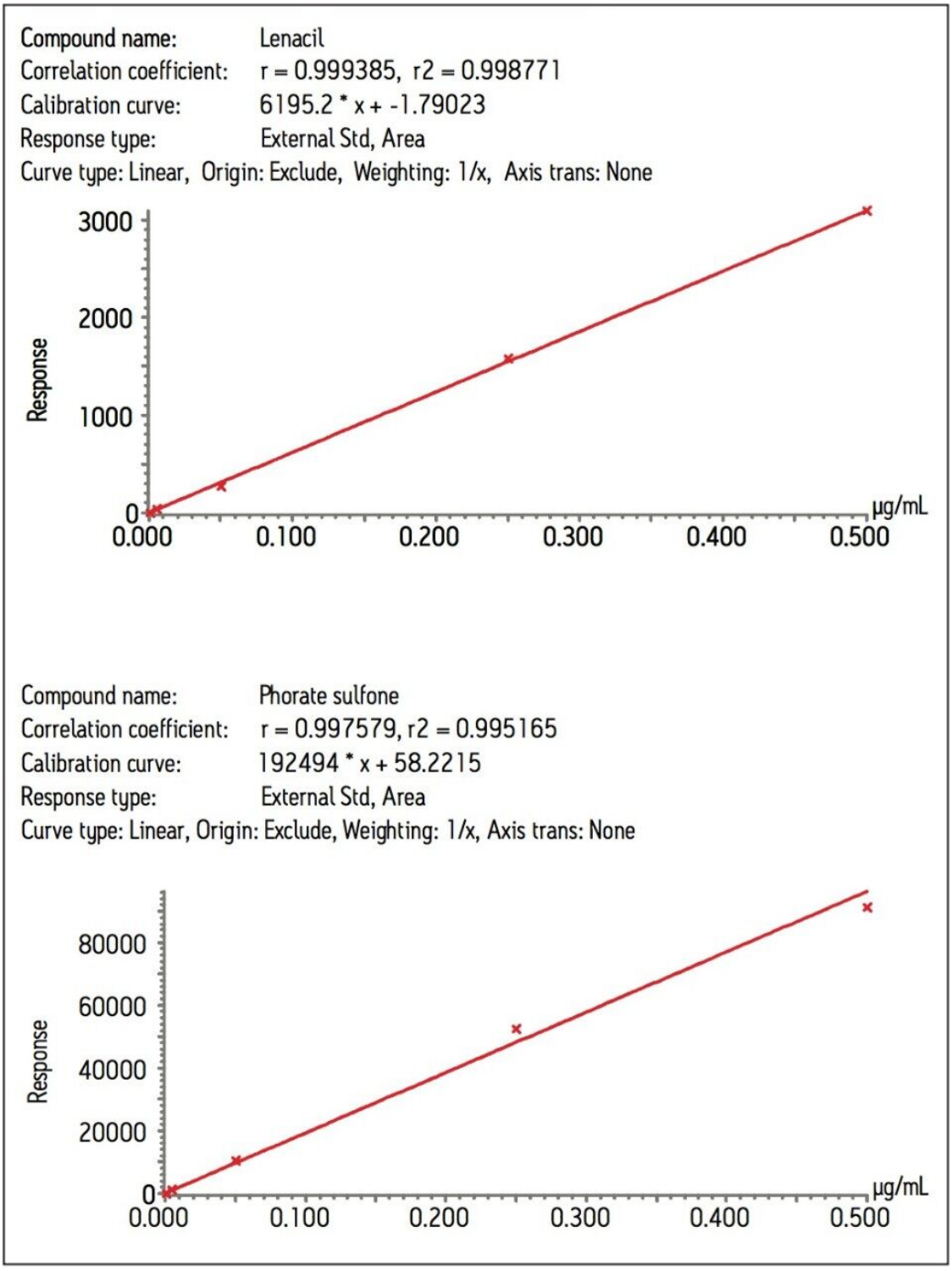

Figure 4. Calibration curves for lenacil and phorate sulfone in fruit-based baby food while polarity switching is on-going.

The ACQUITY TQD has a greater dynamic range than 0.0005 to 0.5 µg/mL, a good practical working range

for quantitation that is equivalent to 3 orders of magnitude, as illustrated in Figure 5. Here, the range is extended from 0.0005 µg/mL to 2.5 µg/mL, equivalent to 4.5 orders of magnitude. This wider range shows the instrument can perform beyond the usual range of quantification.

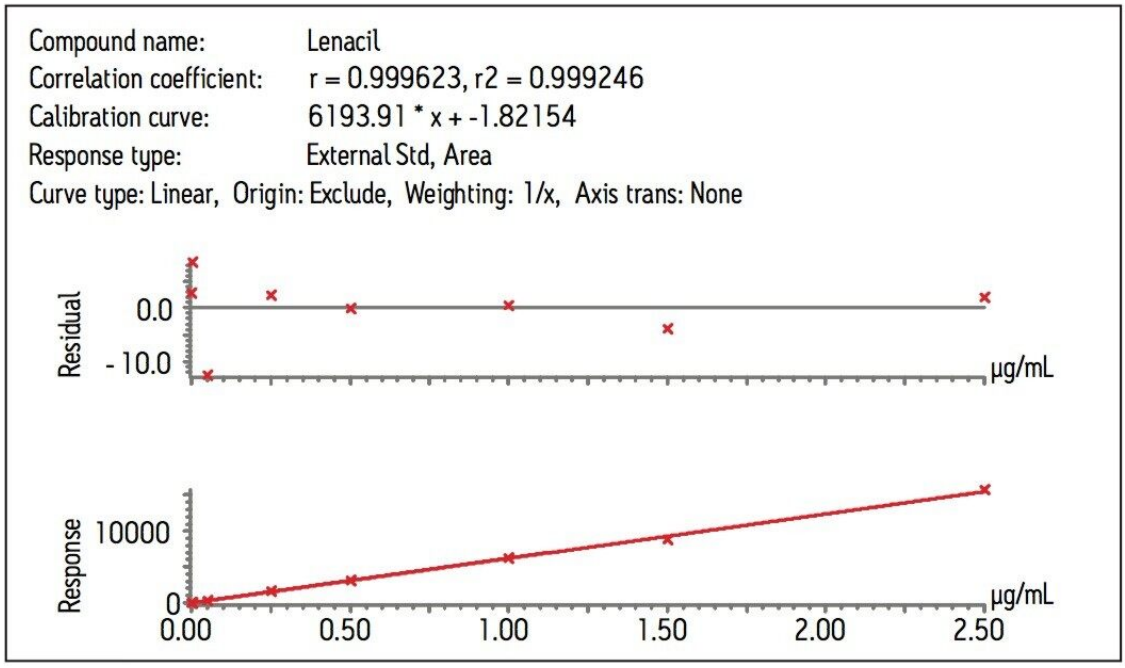

Figure 5. Calibration curve for lenacil, 0.0005 to 2.5 µg/mL in fruit-based baby food.

The ACQUITY TQD was tested over a 100 injection sequence to assess its robustness to matrix. This 12-hour batch analysis contained a series of five matrix-matched standards, between 0.005 and 0.250 µg/mL, delivering approximately 200 mg of matrix. The results for three compounds are illustrated in Figure 6, where the peak area/concentration ratio is plotted against injection number.

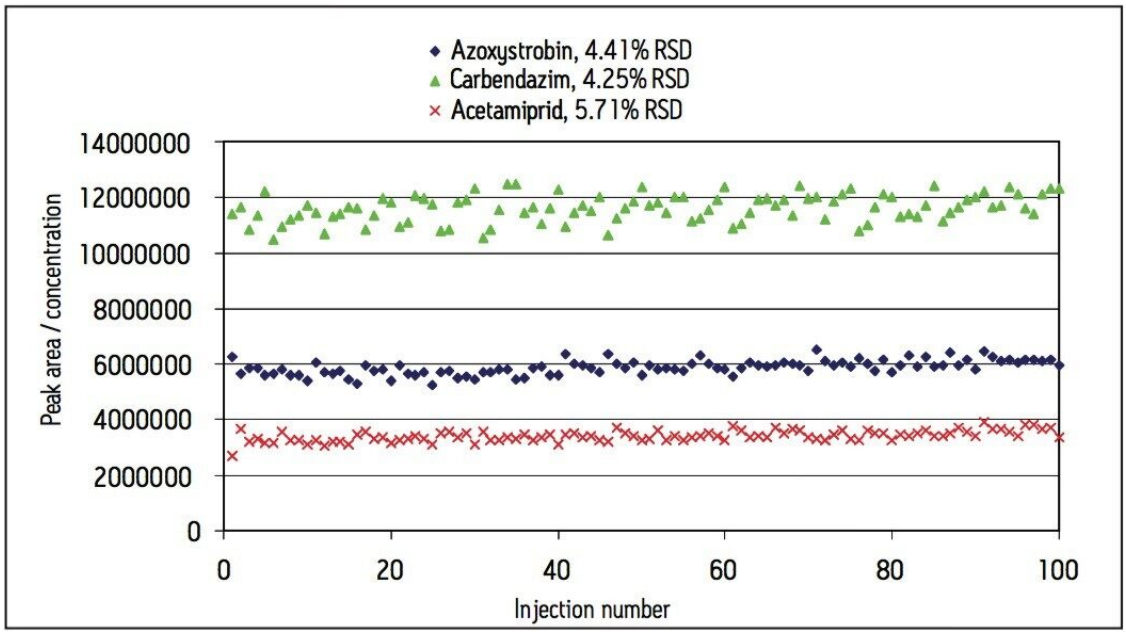

Figure 6. Robustness of ACQUITY TQD over 100 injections of three compounds in fruit-based baby food.

All three compounds display excellent percent relative standard deviations (%RSD) over the injection sequence indicating good instrument robustness.

The ACQUITY TQD's travelling wave collision cell was tested for use at short dwell times for the multi residue method. Figure 7 shows methiocarb in fruit-based baby food at a concentration of 0.05 µg/mL. The sample was injected seven times in succession using the same MRM transition, at differing dwell times from 5 to 500 milliseconds. The data shows that the signal intensity remains consistent over the range of dwell times. As the dwell time decreases the number of points per peak increases from 8 points for the dwell time of 500 milliseconds to 220 points for the shortest dwell time of 5 milliseconds.

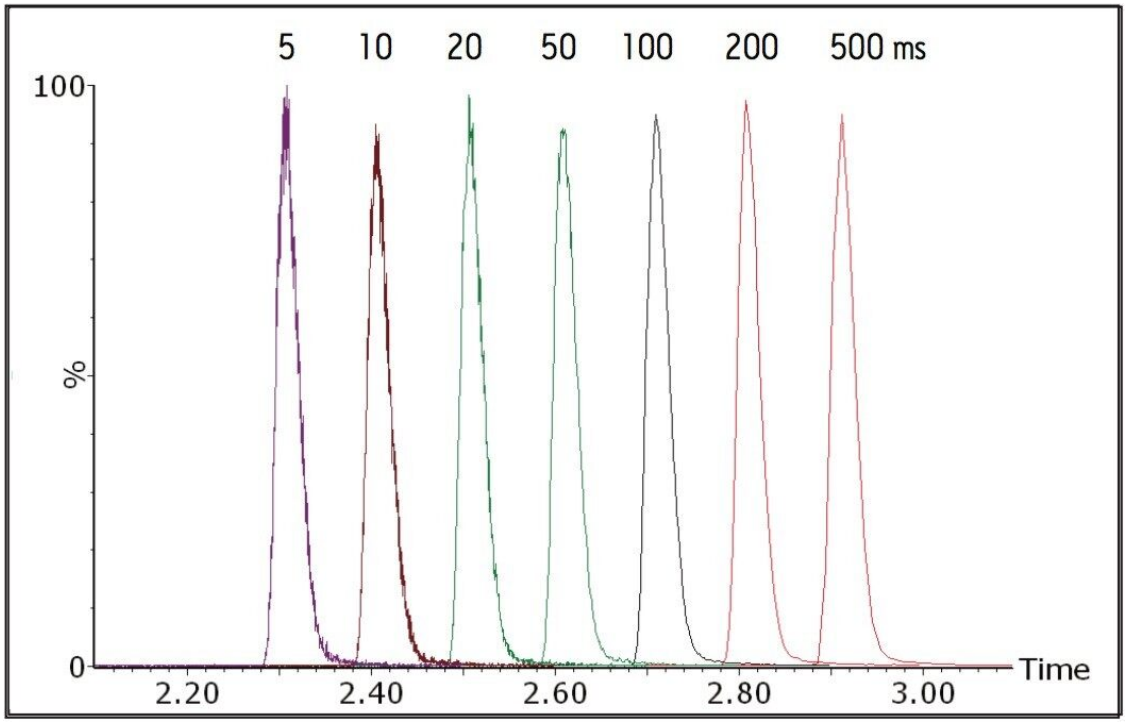

Figure 7. Methiocarb 0.05 µg/mL on the ACQUITY TQD, showing that dwell time does not affect signal intensity.

TargetLynx was used to provide automatic quantification and confirmation with two MRM transitions processed for each residue. The browser produced by TargetLynx for thiabendazole at a spiked concentration of 0.01 µg/mL in fruit-based baby food is illustrated in Figure 8.

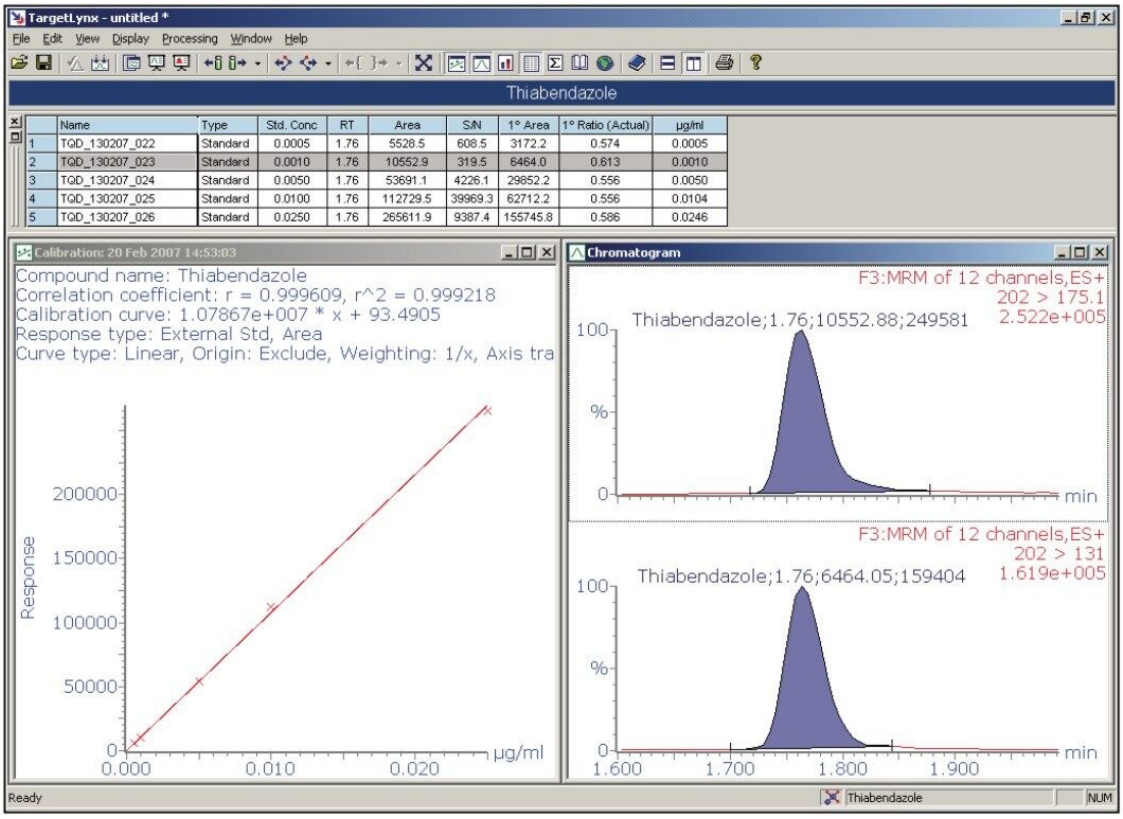

Figure 8. Example TargetLynx browser for thiabendazole in fruit-based baby food (0.01 µg/mL).

All residues could be screened and confirmed to a concentration of 0.01 µg/mL.

Two MRM transitions were monitored for each compound. The primary transition is used for quantification and the secondary transition is used for conformation purposes.

Confirmation is achieved by calculating the ion ratio between the primary and secondary transition. All other injections must have a ratio that lies within 20 percent of the standard for them to be positively confirmed by this technique. Figure 9 shows the ion ratios of three example compounds at concentrations between 0.005 and 0.250 µg/mL in baby food over 100 injections. All injections lie within the ±20% boundary required for confirmation with the average difference shown in the header of the graph.

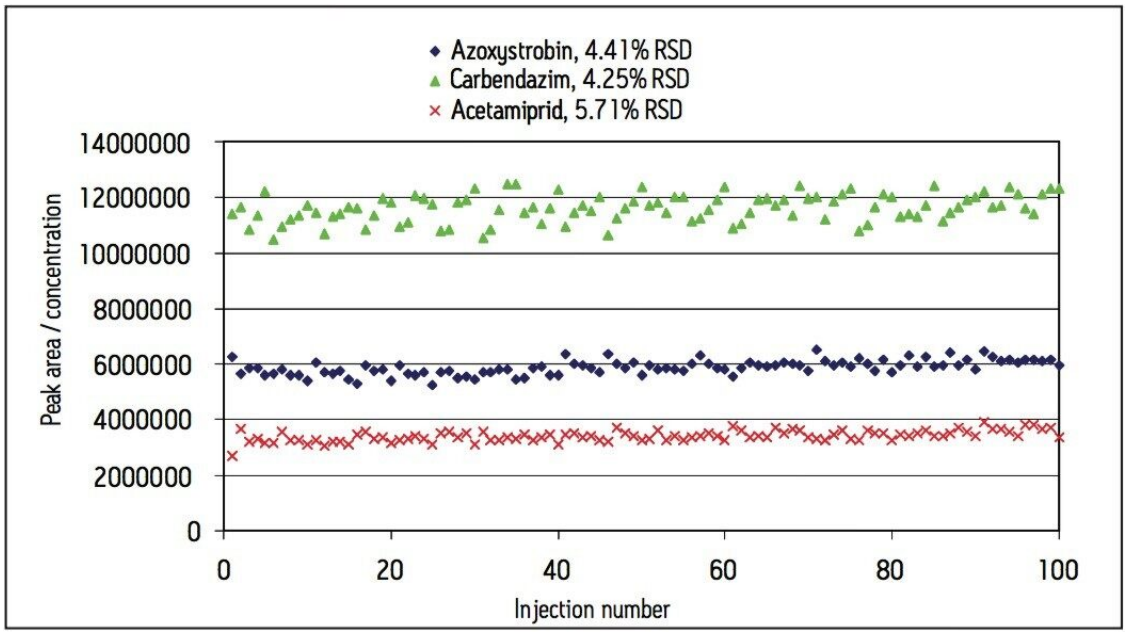

Figure 9. Plot showing confirmatory ion ratio over 100 injections.

Conclusion

A fast and simple UPLC method involving polarity switching of MRM transitions has been successfully transferred to the ACQUITY TQD for the determination of 52 pesticides. Of these, 21 pesticides and 7 metabolites are included in the EU residue monitoring program, 2005–2007.

The ACQUITY TQD was capable of very fast polarity switching, allowing the analysis of positive and negative compounds in a single injection.

The use of very short dwell times of 5 ms was found to have no effect on signal intensity, indicating that sensitivity can be maintained as the number of residues is increased.

Robustness of the ACQUITY TQD was proven with a 100 injection sequence after delivery of approximately 200 mg of matrix to the instrument.

Confirmation was achieved using a secondary MRM transition over a 100 injection sequence, where the variance was less than 20 percent in all cases.

Acknowledgements

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References

1. AGROW Magazine 468 (2005) 9.
2. C.C. Leandro, P. Hancock, R.J. Fussell, B.J. Keely, *J. Chromatogr. A* 1144 (2007) 161-169. doi: 10.1016/j.chroma.2007.01.030.
3. Determination of Pesticides in Food using UPLC with Polarity Switching Tandem Quadrupole LC-MS/MS, Waters Application Note 720001995EN < https://www.waters.com/webassets/cms/library/docs/720001995en.pdf> .

Appendix 1. ACQUITY TQD MRM Parameters.

Pesticide	RT	MRM Transitions	Dwell time(s)	Cone Voltage (V)	Collision Energy (eV)
Methamidophos	0.43	142>94	0.015	28	14
		142>125			13
Acephate	0.53	184>143	0.015	22	8
		184>125			18
Omethoate	0.6	214>183	0.015	26	12
		214>155			15
Butocarboxim sulfoxide	0.62	207>75	0.015	23	12
		207>132			6
Aldicarb sulfoxide	0.68	207>89	0.015	22	14
		207>132			10
Butoxycarboxim	0.74	223>106	0.015	23	10
		223>166			7
Aldicarb sulfone	0.76	223>86	0.015	29	12
		223>76			7
Methomyl	0.88	163>88	0.01	21	8
		163>106			10

Pesticide	RT	MRM Transitions	Dwell time(s)	Cone Voltage (V)	Collision Energy (eV)
Oxydemeton-methyl	0.9	247>169	0.01	26	14
		247>109			28
Pymetrozine	0.93	218>105	0.015	31	17
		218>79			36
Demeton-S-methyl sulfone	0.95	263>169	0.01	32	17
		263>121			17
Imidacloprid	1.2	256>209	0.01	28	16
		256>175			20
Carbendazim	1.5	192>160	0.01	27	18
		192>132			30
Methiocarb sulfoxide	1.32	242>185	0.01	28	13
		242>168			24
Dimethoate	1.33	230>125	0.01	23	20
		230>171			15
Acetamiprid	1.38	223>126	0.01	33	20
		223>56			15
Cymoxanil	1.47	199>128	0.01	23	8
		199>111			18
Methiocarb sulfone	1.47	258>122	0.01	28	20
		258>107			37
Thiacloprid	1.55	253>126	0.01	34	20
		253>90			37
Butocarboxim	1.66	213>75	0.01	30	15
		213>156			10
Aldicarb	1.69	208>116	0.01	13	7
		208>89			7
Thiabendazole	1.74	202>175	0.01	46	25
		202>131			32
Carbaryl	2.1	202>145	0.02	24	10
		202>127			28
Thiodicarb	2.19	355>88	0.02	21	16
		355>108			16
Phorate sulfoxide	2.24	277>97	0.01	24	32
		277>143			20
Phorate sulfone	2.28	293>97	0.01	24	30
		293>115			24
Lenacil (-ve)	2.31	233>151	0.03	50	24
		233>107			32

Pesticide	RT	MRM Transitions	Dwell time(s)	Cone Voltage (V)	Collision Energy (eV)
Azinphos-methyl	2.45	318>160	0.01	20	8
		318>261			8
Linuron	2.56	249>160	0.01	34	16
		249>182			15
Azoxystrobin	2.58	404>372	0.01	28	15
		404>329			30
Methiocarb	2.61	226>169	0.01	22	10
		226>121			19
Fludioxonil (-ve)	2.65	247>180	0.03	51	28
		247>126			35
Iprovalicarb	2.84	321>119	0.01	21	18
		321>203			8
Triadimenol	2.85	296>70	0.01	14	10
		296>99			16
Dichlofluanid	2.86	333>123	0.01	22	24
		333>224			10
Fenhexamid	2.87	302>97	0.01	35	25
		302>55			35
Fenoxycarb	2.87	302>88	0.01	21	20
		302>116			12
Flufenacet	2.89	364>152	0.01	17	20
		364>194			10
Diflubenzuron (-ve)	2.98	309>156	0.03	20	11
		309>289			9
Cyprodinil	3.08	226>93	0.01	45	33
		226>108			25
Tolylfluanid	3.09	347>137	0.01	19	28
		347>238			10
Zoxamide	3.13	336>187	0.01	25	24
		336>159			41
Imazalil	3.15	297>159	0.01	36	20
		297>69			20
Phorate	3.19	261>75	0.01	11	12
		261>97			32
Hexaflumuron (-ve)	3.37	459>276	0.02	22	22
		459>175			39
Fluazinam (-ve)	3.5	463>416	0.02	26	21
		463>398			17

Pesticide	RT	MRM Transitions	Dwell time(s)	Cone Voltage (V)	Collision Energy (eV)
Teflubenzuron (-ve)	3.54	379>196	0.02	18	25
		379>339			15
Lufenuron (-ve)	3.56	509>175	0.02	22	40
		509>326			22
Flucycloxuron (-ve)	3.64	482>156	0.02	34	14
		482>462			13
Flufenoxuron (-ve)	3.67	487>156	0.02	27	16
		487>329			22

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