Utilisation of LC/MSMS (QTRAP) and polarity switching for the quantitative analysis of over 300 pesticides in crude QuEChERS extracts from various fruit and vegetable matrices

Laura M. Melton, Devanshi S. Vyas, Emily E. Flynn, Kirsty B. Reid, Michael J. Taylor, Katie J. Viezens, Andrew T. Wilson
Science and Advice for Scottish Agriculture (SASA), Roddinglaw Road, Edinburgh, UK, EH12 9FJ

Introduction

SASA (Science and Advice for Scottish Agriculture) is an Official UK laboratory that on behalf of the Scottish Government, participates in statutory UK and EU annual surveillance programmes that monitor UK and imported food & drink for residues of pesticides and their metabolites.

LC/MSMS has been a front-line tool in our laboratory for many years but with older instrumentation it was not possible to generate reliable data for all pesticides in a single run. Samples often had to be run several times in order to achieve the required detection limits (typically 10 ppb), to cover all pesticides in positive and negative electrospray ionisation modes and to provide sufficient identification points to confirm positive residues.

Nowadays, we use a highly sensitive LC/MSMS (QTRAP) instrument capable of simultaneous acquisition of hundreds of MRMs incorporating fast polarity switching without compromising data or generation of sufficient data points across each peak for quantitation. We are also able to analyse QuEChERS extracts that are not subjected to a clean-up step due to the excellent robustness of the system.

Experimental

Sample preparation

10g of cryomilled fruit and vegetable samples were extracted using citrate QuEChERS method (matrix concentration ≡ 1g ml^{-1}). No clean-up was employed. Sample extracts were then filtered into vials for LC/MSMS (0.45µm PTFE).
Calibration standards were prepared in appropriate (organic) fruit or vegetable matrix.

Set-up for the SCIEX 6500 QTRAP mass spectrometer and Shimadzu Nexera UHPLC

- Run time: 17 min
- Flow rate: 0.4 mL/min
- Eluent A: Methanol/H_2O 5/95 v/v + 5mM ammonium acetate
- Eluent B: Methanol + 5mM ammonium acetate
- Gradient elution
- Column: Phenomenex Kinetex 2.6 µm, C18, 50 x 4.6 mm with Phenomenex Security Guard cartridge
- Injection volume: 3 µl
- Minimum of 2 MRMs acquired per pesticide
- Polarity switching

Results

The data below demonstrates the versatility of the instrument with a summary of all of the residues detected and quantified from 2016 fruit and vegetable surveillance samples (Table 1). Figure 1 shows our laboratory's consistent performance in FAPAS and EU proficiency tests for LC/MSMS compounds in 2016. Z-scores must be in the range -2 to 2 to be acceptable. The instrument routinely acquires hundreds of MRMs incorporating polarity switching (Figure 2) and maintains peak integrity with excellent signal to noise and sufficient data points in both ionisation modes. The consistent performance and stability of the instrument over time i.e. 100 injections is illustrated in data presented in Figure 3.

Table 1. Summary of residues detected and quantified by LC/MSMS in 2016 samples.

Commodity	Number of samples tested	Number of positive samples	Number of residues detected and quantified > Reporting Level (RL)	Number of pesticides detected (number of pesticides sought)	Number of residues > Maximum Residue Level (MRL)	Minimum Residue (mg/kg)	Maximum Residue (mg/kg)
Beans with Pods	120	62	142	41(306)	40	0.001	0.9
Cabbage	96	17	26	13(306)	0	0.01	0.2
Grapefruit	96	95	403	22(306)	2	0.01	7.8
Leek	96	22	36	10(306)	0	0.01	0.1
Spring onion	48	26	43	11(306)	0	0.01	0.9
Strawberry	96	81	341	36(306)	1	0.01	1.6

Reporting Level (RL) 0.01 mg/kg for most pesticides
Pesticide residues in food: results of monitoring programme can be found at:
https://www.gov.uk/government/collections/pesticide-residues-in-food-results-of-monitoring-programme#annual-monitoring-results

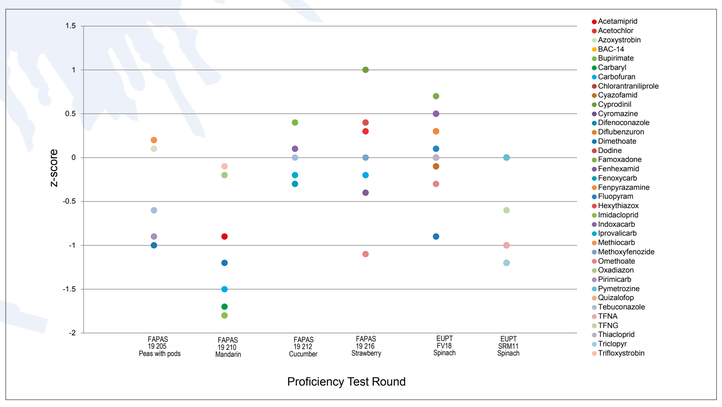

Figure 1. SASA LCMSMS Proficiency Test Data 2016.

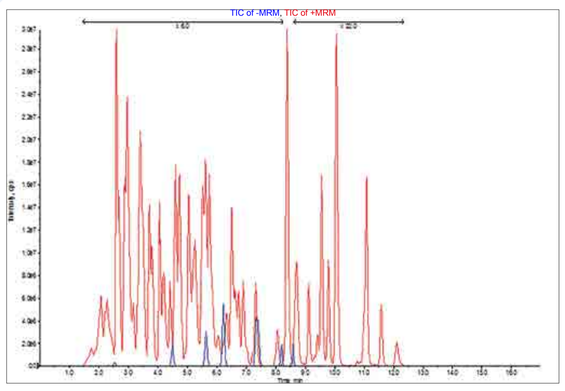

Figure 2a. TIC of strawberry matrix standard at reporting level (0.01 mg/kg for most pesticides).

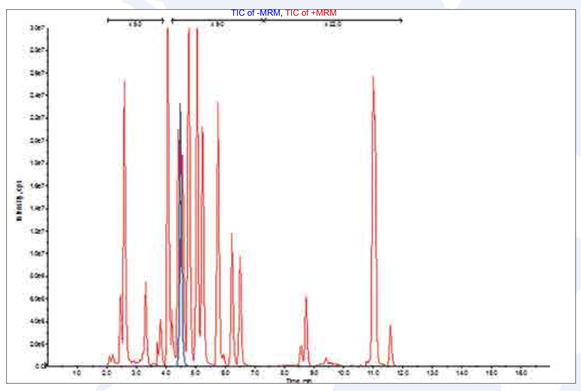

Figure 2b. TIC of 2016 strawberry sample containing 16 pesticide residues (range 0.01 – 0.3 mg/kg) i.e. 15 positive MRM in red and 1 negative MRM in blue.

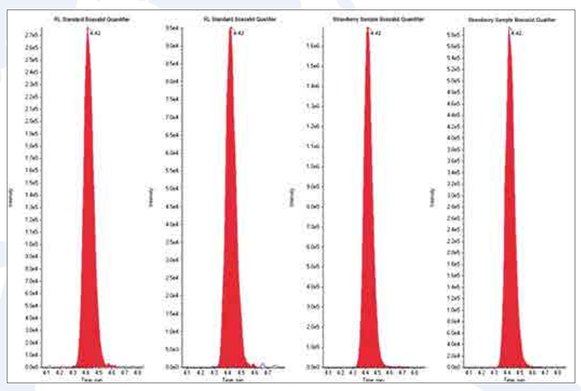
Figure 2c. XIC of boscalid (+MRM) quantifier and qualifier transitions for strawberry matrix standard at reporting level (0.01 mg/kg) and 2016 strawberry sample containing 16 pesticide residues including boscalid (+MRM) at 0.08 mg/kg.

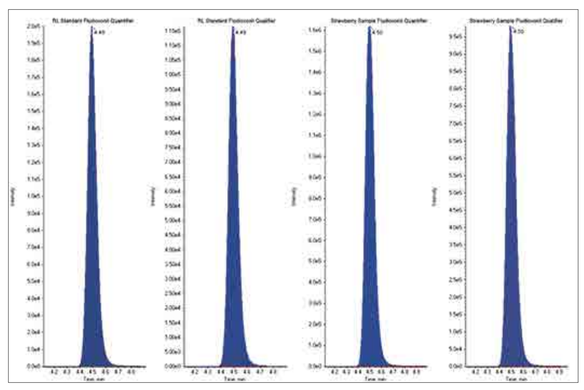
Figure 2d. XIC of fludioxonil (-MRM) quantifier and qualifier transitions for strawberry matrix standard at reporting level (0.01 mg/kg) and 2016 strawberry sample containing 16 pesticide residues including fludioxonil (-MRM) at 0.1 mg/kg.

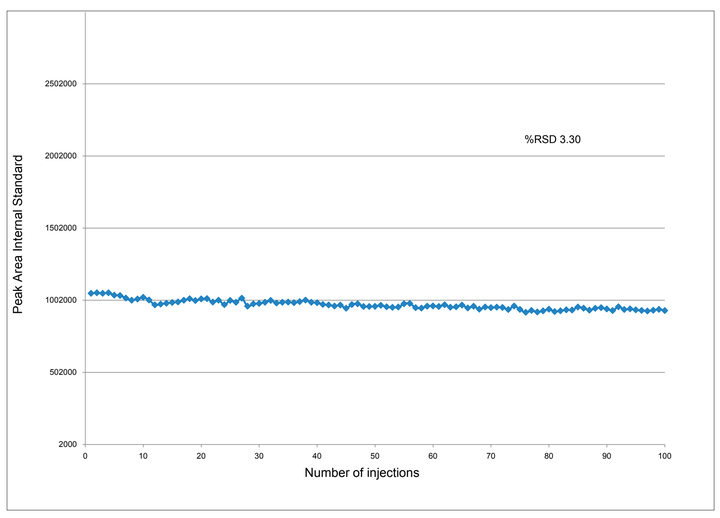

Figure 3. Performance Data (100 injections over 28 hours run time using full acquisition method described above).

Conclusions

The instrument's robust and reliable performance has allowed our laboratory to detect and quantify hundreds of pesticide residues in various fruit and vegetable surveillance samples in a single run. Results were from crude extracts and no clean-up step was used. Furthermore, all of our UK and EU proficiency test results were acceptable for LC/MSMS compounds present in each test commodity. The sensitivity and robust performance of the instrument meant that repeat runs were only required if residues exceeded the calibration range. System performance was consistent despite minimal time spent on maintenance of the instrument.