Chromatography Technical Note No AS132

Determination of pesticides in QuEChERs extracts using the Multiflex GC/Q-TOF

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Introduction

QuEChERS is a Quick-Easy-Cheap-Effective-Rugged-Safe multi-analyte method that uses acetonitrile to extract pesticide residues from various agricultural commodities. Injecting acetonitrile QuEChERS extracts onto a HP-5 MS column can cause chromatographic peak splitting due to a mismatch in polarity between the column and diluent. Peak splitting is normally worse for the more volatile analytes such as dichlorvos and biphenyl. This affect can be minimised either, by using a retention gap or by injecting the solvent cold and then using a thermal gradient to desorb analytes onto the GC.

The Cool Inlet System (CIS) shown in Figure 1 is a Programmable Temperature Vaporiser which allows samples to be injected cold. The CIS can then be heated rapidly with a controlled temperature ramp to vent the solvent and then volatilize the pesticides onto the GC column. The CIS conditions have been optimized to achieve good peak shape for early eluting analytes.

The Agilent 7200 is a GC accurate mass -Time of Flight Mass Spectrometer (GC/Q-TOF) which is shown below in Figure 2. Mass accuracy is typically 5ppm. By extracting a known accurate mass with a narrow mass window *e.g.* 20ppm from the total ion current (TIC) improved signal:noise and selectivity is observed.

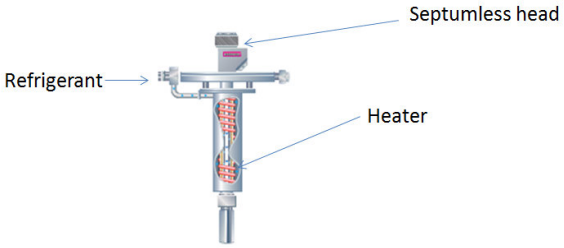

Figure 1: Gerstel Cold Inlet System (CIS)

Figure 2: Photo of Multiflex GC/-Q-TOF

Instrumentation

Agilent GC 7890B and Agilent 7200 Q-TOF
Gerstel MPS 2 XL-*xt*
Gerstel CIS4,
Agilent MassHunter software (version B07.00)
Maestro software integrated

Method

GC-MS parameters:

CIS parameters:	(baffled liner) PTV Solvent Vent 40 °C ramped to 240 °C
Column:	HP5ms 30 m x 0.25 mm x 0.25 µm
Injection volume:	1 µl
Oven program:	40 °C thermal gradient to 300 °C over 30 minutes
MS:	70 eV EI Q-TOF (mass range 50 – 500 *m/z*)

Acetonitrile extracts of apples were used to prepare matrix-matched calibration standards. Blank and spiked extracts were cleaned-up using PSA—(primary and secondary amine) solid phase extraction sorbent in dispersive mode.

A seven point calibration ranging from 10 ppb to 200 ppb was performed. TPP (triphenyl phosphate) was used as internal standard.

Results

Figure 3 shows the calibration plot for pirimphos-methyl. A correlation coefficient of 0.9980 was achieved and the detection limit for pirimiphos-methyl was calculated to be 0.2 ppb . Detection limits have been calculated by determining the chromatographic peak (signal) height and baseline noise height. The detection limit has been defined as three times signal to baseline noise. Detection limits have been calculated from the lowest standard at 10ppb within this application note.

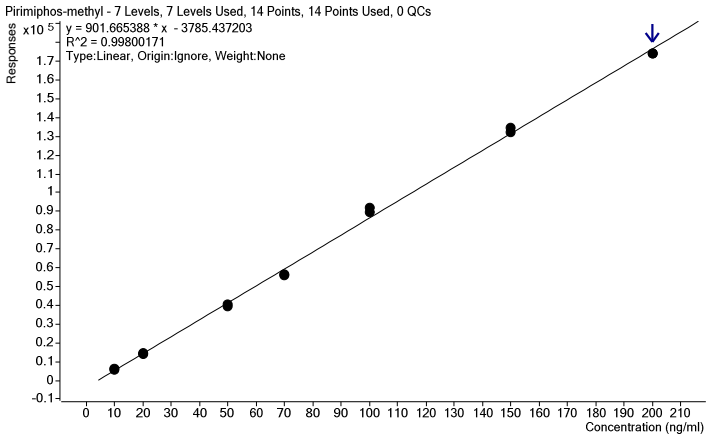

Figure 3 Calibration plot for pirimphos –methyl

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Figure 4 shows a comparison of duplicate injections of a 10 ppb spike with an unspiked apple extract (extracted ion chromatogram of 305.0963 *m/z*). Peak at 12.5 minutes is pirimiphos methyl.

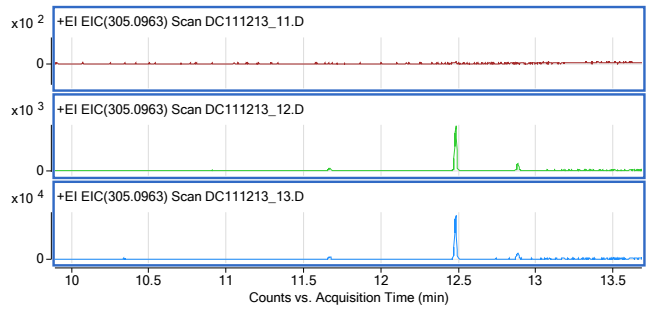

Figure 4 Extracted ion Chromatogram for pirimiphos methyl (*m/z* 305.0963)

An early eluting analyte, dichlovos, can be challenging to obtain good linearity. Figure 5 shows the calibration plot for dichlovos. The correlation coefficient was 0.9860 and the detection limit for dichlovos was calculated to be 0.5 ppb.

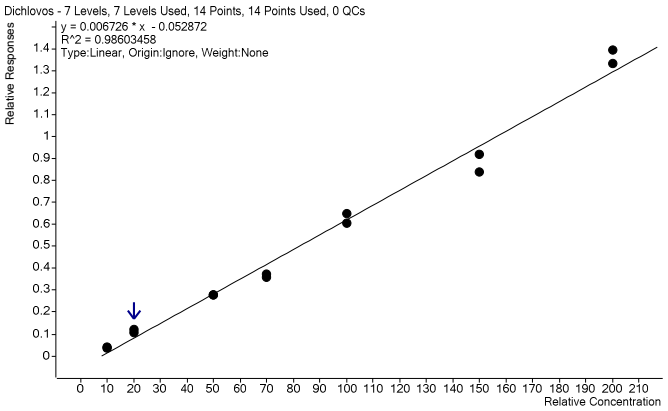

Figure 5 Calibration plot for dichlovos

Figure 6 shows a comparison of duplicate injections of an extract of apple containing 10 ppb dichlorvos with an unspiked apple extract (extracted ion chromatogram of *m/z* 109.0055). Dichlorvos eluted at 8.15 minutes.

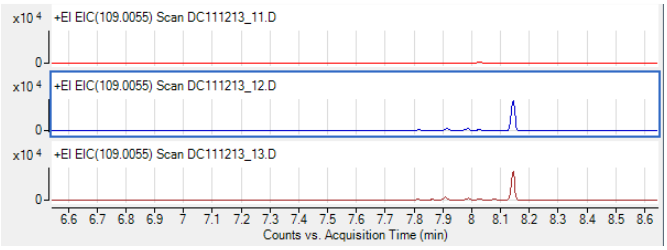

Figure 6 Extracted ion Chromatogram of dichlorvos (*m/z* 109.0055)

Repeatability experiments for the different pesticides have not yet been carried out. However, internal standard was present in all samples. % Relative Standard Deviation (%RSD) of triphenyl phosphate (TPP) was calculated to be 4.5% based on 14 replicate injections. Further work will be performed to investigate precision each analyte. Table 1 shows linearity achieved (seven point calibration) for a number of the pesticides present in the test mixture. All detection limits were all calculated to be 2 ppb or below.

Analyte	**Retention time (min)**	Correlation Coefficient (R^2)
Dichlorobenil	8.9	0.9920
Biphenyl	9.1	0.9910
Propachlor	10.6	0.9981
Diphenylamine	10.7	0.9984
Dichloran	11.3	0.9970
Propanil	12.1	0.9975
Flutolanil	13.5	0.9981
Etoxazole	14.8	0.9945
Coumaphos	16.0	0.9964

Table 1: Correlation coefficients for spiked pesticides in apple extract

The extracted ion chromatograms within this application note have been performed with a 20ppm mass window. Figure 7 shows a comparison of an extracted ion chromatogram, at unit mass resolution (giving resolution comparable to a single quad instrument) and extracted ion chromatogram at 20ppm resolution. (Figure 7) As you can see, with a resolution of 20 ppm only one chromatographic peak can be detected within this retention window (improved selectivity). Whereas extracting with unit mass resolution, many chromatographic peaks can be observed making identification of known analytes problematic.

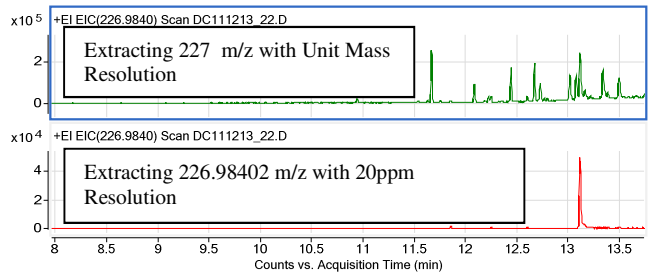

Figure 7 Comparison of extracting unit mass resolution verses 20ppm resolution

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In combination with obtaining accurate mass information, the NIST library can also be used to obtain a good library match using Q-TOF data. A match factor of 8 out of 10 analytes in table 1 was"good". Two of the matches were "fair" matches. These have been defined using a report provided by NIST [1].

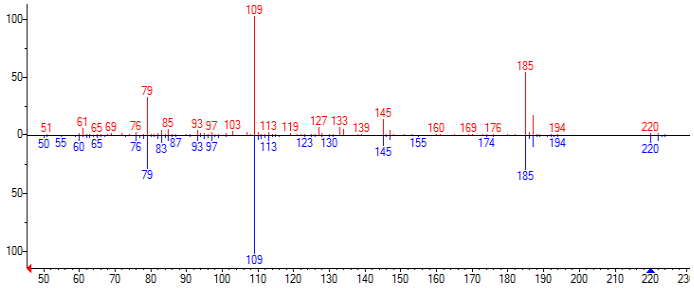

Figure 8 comparison of NIST library (blue) and standard of dichlorvos (red)

Discussion

Low detection limits (ppb level) and high selectivity for known analytes can be achieved using the GC/Q-TOF fitted with a cooled CIS inlet. Good EI library matching (NIST) can also be obtained on the GC/Q-TOF. Please contact us if you require any further information.

I would like to thank Richard Fussell and Mike Hetmanski at The Food and Environmental Agency (Fera) for providing the extracts for this study.

[1] NIST standard reference database NIST/EPA/NIH Mass Spectral Library (2008)

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