

# Determination of pesticides in foods using phosphorus and sulfur detection by GC-ICP-QQQ

## Application note

### Food testing

#### Authors

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#### Introduction

The determination of pesticide residues in food products is important especially for infants and children, as the quantity of food they ingest per kilogram of body weight is relatively high. Some pesticides are known or suspected endocrine disrupting chemicals (EDCs), so the potential for interference with the function of the naturally occurring hormones can severely impact a growing child, even at low concentrations. The implications of long-term, low-dose exposure through food consumption have led to interest in development of highly sensitive analytical methods that combine rapid sample preparation with hyphenated separation and detection. Most pesticide residue laboratories use some variation of the QuEChERS (Quick, Easy, Cheap, Effective, Rugged and Safe) extraction methods. Often the extracted material is analyzed using gas chromatography-tandem mass spectrometry (GC/MS/MS) for the thermally stable, less polar pesticides, or liquid chromatography-tandem mass spectrometry (LC-MS/MS) analysis for the less volatile and/or more polar ones. A more recently developed alternative technique involves coupling GC to triple quadrupole ICP-MS (GC-ICP-QQQ), which is able to



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determine pesticides by measuring the heteroatoms P and S (also Cl and Br) contained in most pesticides. GC-ICP-QQQ offers very good selectivity and specificity, and sensitivity that can be greater than the established methods. The method described in this work can be applied to other compounds such as organophosphorus chemical warfare agents, brominated flame retardants, polymer additives and trace sulfur compounds in flavors and fragrances.

## Experimental

### Reagents

Three standard pesticide mixes were obtained from Ultra Scientific (Kingstown, RI, USA) and Agilent Technologies (p/n 5190-0468). The standard solutions were diluted with high purity grade acetonitrile to form intermediate solutions, which were then used to prepare calibration standard solutions following serial dilutions in acetonitrile.

### Samples and sample preparation

Several different food matrices were tested for their pesticide content, including baby food fruit purees (purchased in São Paulo, Brazil), and 3 types of tea (purchased in China). All food matrices were homogenized and extracted in triplicate applying the AOAC 2007.1 QuEChERS method [1], and using a QuEChERS Extraction Kit (p/n 5982-5755).

As the first step, approximately  $15 \pm 0.1$  g of baby food sample or  $5 \pm 0.1$  g of tea were weighed into 50 mL centrifuge tubes and 15 mL of acetonitrile was added. Two ceramic bars were added and each sample was homogenized for 1 min using a vortex mixer. Agilent Bond Elut QuEChERS extraction anhydrous salt containing 6 g of  $\text{MgSO}_4$  and 1.5 g of NaCl was added. The mixture was agitated for 1 min and then centrifuged at 2600 g for 5 min.

Secondly, an 8 mL aliquot of the upper acetonitrile layer was transferred to an Agilent Bond Elut QuEChERS General Fruits and Vegetables 15 mL centrifuge tube with dispersive SPE (p/n 5982-5058) containing primary-secondary amine (PSA) and  $\text{MgSO}_4$ . The tube was vortexed for 1 min and centrifuged at 2600 g for 5 min. The resultant upper layer was transferred to an amber glass vial (p/n 5183-4496) for analysis.

### Instrumentation

An Agilent 7890 GC was coupled to an Agilent 8800 ICP-QQQ using the Agilent GC-ICP-MS interface (G3158D). Two Agilent columns were used in series. The first column was a 5 m length cut from a 20 m x 0.18 mm x 0.18  $\mu\text{m}$  film thickness DB-35ms Ultra Inert (UI) capillary column, p/n 121-3822UI (a 5 m column is available as p/n G3903-61005). This column was installed between the inlet and one end of the purged union and was backflushed shortly before the run had ended to prevent high boiling point contaminants from entering the second column. The second column was a 15 m x 0.25 mm i.d. x 0.25  $\mu\text{m}$  film thickness DB-5MS UI capillary column (p/n 19091S-431UI), which was installed between the other end of the purged union and the ICP-MS transfer line connection inside the GC oven. Sample injections of 1  $\mu\text{L}$  volume were made under splitless conditions with the inlet held at 280 °C. Pre-heated make-up gas was used to sweep the GC column effluent efficiently into the plasma. GC operating parameters are detailed in a previous study [2].

Phosphorus and sulfur have high first ionization potentials so they are poorly ionized in the ICP-MS plasma, leading to relatively low sensitivity. They are also difficult to measure by conventional quadrupole ICP-MS (ICP-QMS) due to intense spectral interferences. However, ICP-QQQ operating in MS/MS reaction cell mode can be applied to resolve these spectral interferences, allowing the quantification of P and S at far lower levels than was previously possible using ICP-QMS. In this study, P and S were measured as the reaction product ions  $^{31}\text{PO}^+$  at  $m/z$  47 and  $^{32}\text{SO}^+$  at  $m/z$  48 respectively, using MS/MS mass-shift mode with  $\text{O}_2$  as the cell gas.

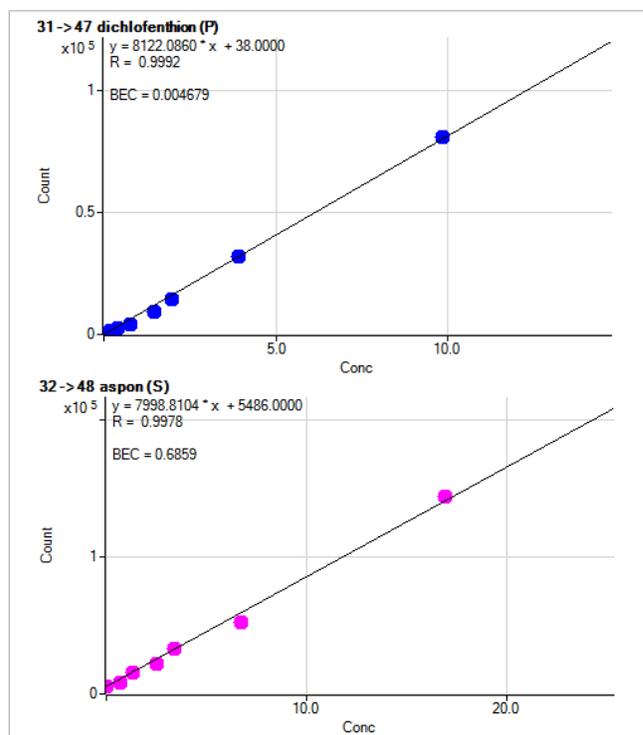
To tune the ICP-QQQ, the helium carrier gas supplying the GC inlet and auxiliary electronic pneumatics control (aux EPC) module was replaced with argon containing  $\text{H}_2\text{S}$  at 100 ppm. Oxygen cell gas flow rate and cell settings were optimized for  $^{32}\text{S}^+$  signal from the  $\text{H}_2\text{S}$ -spiked argon. A high residual sulfur background was observed after switching back to pure helium, suggesting that an alternative position to introduce the tuning gas into the ICP-QQQ would be required for more routine usage. ICP-QQQ operating conditions are shown in Table 1.

**Table 1.** Agilent 8800 ICP-QQQ operating conditions

	P and S measurement
Sampling depth (mm)	3
Make up gas flow rate (L/min)	0.6
Octopole bias (V)	-0.5
Kinetic Energy Discrimination (V)	-13
Cell gas	O <sub>2</sub>
Cell gas flow rate	20% of full scale
Monitored masses	Q1 » Q2 31 » 47; 32 » 48

## Results and discussion

To confirm the linearity of the GC-ICP-QQQ method, calibration curves for P (~0.01–16 µg/L) and S (~0.8–33 µg/L) were generated using 10 and 6 calibration standards plus blanks, respectively. The concentrations represent the range required for the measurement of these elements in pesticides. For routine operation the calibration could be simplified to 3 or 4 calibration levels for each compound. Correlation coefficients (R) over 0.997 were achieved for both elements in all compounds, indicating excellent linearity over the calibration range, as shown in Table 2. Representative calibration curves are given for P and S in Figure 1.



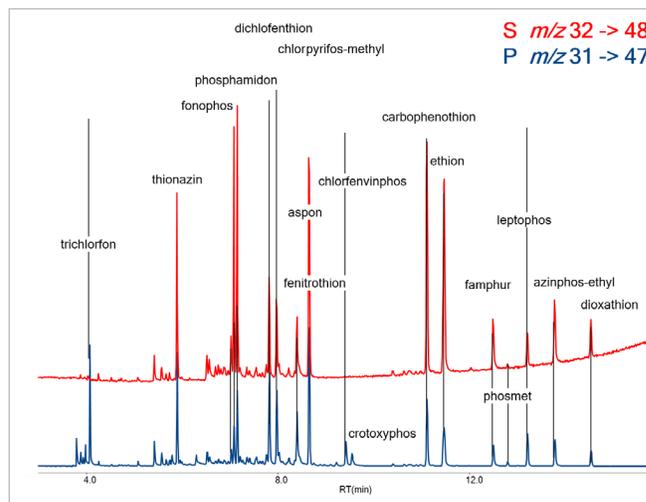
**Figure 1.** Upper: calibration curve for dichlofenthion, based on response for P (measured as PO<sup>+</sup>). Lower: calibration curve for aspon, based on response for S (measured as SO<sup>+</sup>).

**Table 2.** Calibration range and correlation coefficient (R) for 18 pesticides as compounds based on PO<sup>+</sup> and SO<sup>+</sup> measurements

Compound	Retention Time ± 0.3 (min)	P		S	
		Calibration Range as Compound (µg/L)	Correlation Coefficient (R)	Calibration Range as Compound (µg/L)	Correlation Coefficient (R)
Trichlorfon	4.103	0.02 - 12	0.999		
Thionazin	5.926	0.02 - 12	0.999	1 - 12	0.996
Terbufos	7.071	0.02 - 11	0.999	1.3 - 33	0.997
Fonofos	7.185	0.02 - 13	0.999	2 - 26	0.997
Phosphamidon	7.299	0.02 - 21	0.998		
Dichlofenthion	7.858	0.01 - 10	0.999	0.8 - 10	0.999
Chloropyrifos-methyl	7.973	0.01 - 10	0.997	0.8 - 10	0.995
Fenitrothion	8.44	0.02 - 11	0.999	2 - 11	0.999
Aspon	8.705	0.02 - 16	0.999	0.7 - 16	0.998
Chlorfenvinphos	9.486	0.01 - 9	0.996		
Crotoxyphos	9.541	0.01 - 10	0.999		
Carbophenothion	11.158	0.08 - 9	0.999	1.1 - 28	0.997
Ethion	11.527	0.02 - 15	0.999		
Famphur	12.547	0.01 - 10	0.998	4 - 20	0.991
Phosmet	12.851	0.14 - 10	0.996		
Leptophos	13.263	0.01 - 8	0.999	1 - 8	0.999
Azinphos-ethyl	13.827	0.01 - 9	0.999	3 - 19	0.996
Dioxathion	14.587	0.02 - 14	0.998		

Detection limits (DLs) for pesticides using current GC/MS/MS instrumentation typically vary from about 0.1 to 10 µg/L depending on the pesticide and instrument used [3, 4]. The data in Table 3 suggests that GC-ICP-QQQ offers similar or slightly lower DLs than GC/MS/MS for the determination of organophosphorus pesticides. For S-containing pesticides, detection limits are similar to, or slightly higher than those achieved by GC/MS/MS. The majority of the pesticides listed in Table 3 could be detected via their P content and quantified well below the 10 µg/L limit of quantitation (LOQ) required by most food safety laboratories.

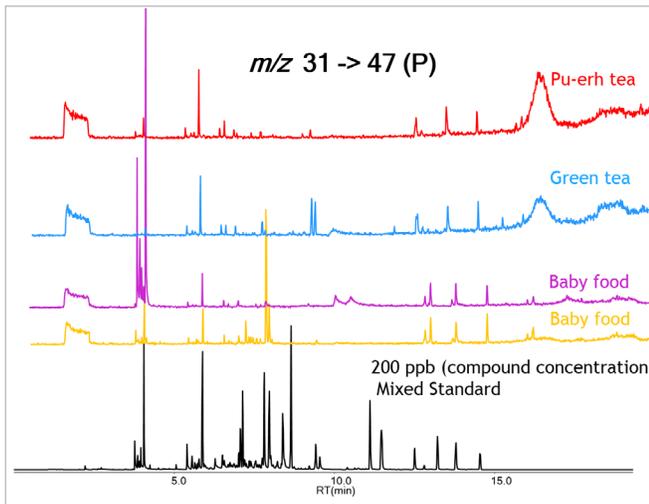
Figure 2 shows overlaid chromatograms for P and S in a mixed pesticide standard. The pesticides that contain more than one heteroelement can easily be identified. Real food samples were measured for pesticide content and the chromatograms for P signal in four foods and the 200 µg/L mixed pesticide standard are shown in Figure 3. Traces of organophosphorus pesticides can be seen in all of the food samples tested.



**Figure 2.** Chromatograms showing the heteroatom traces for P and S in the mixed pesticide standard, with identified pesticide compounds. Reprinted with permission from *J. Agric. Food Chem.*, 2015, 63, 4478–4483. Copyright 2015 American Chemical Society.

**Table 3.** Detection limits as the compound and as the heteroatom element for pesticides. The limits of detection (LOD) were calculated from 3x the signal-to-noise (S/N).

Pesticide	RT ± 0.3 (min)	P		S	
		Compound DL (µg/L)	Element DL (µg/L)	Compound DL (µg/L)	Element DL (µg/L)
Trichlorfon	4.103	0.178	0.021		
Thionazin	5.926	0.221	0.028	11.93	1.580
Terbufos	7.071	0.718	0.077	9.708	3.231
Fonofos	7.185	0.455	0.057	7.917	2.057
Phosphamidon	7.299	0.923	0.095		
Dichlofenthion	7.858	0.362	0.036	15.80	1.604
Chlorpyrifos-methyl	7.973	0.613	0.059	24.18	2.399
Fenitrothion	8.440	0.907	0.101	19.52	2.253
Aspon	8.705	0.200	0.033	9.912	1.676
Chlorfenvinphos	9.486	2.020	0.174		
Crotoxyphos	9.541	3.338	0.329		
Carbophenothion	11.158	0.583	0.053	9.585	2.684
Ethion	11.527	0.707	0.114	11.51	3.832
Famphur	12.547	2.206	0.210	20.61	4.054
Phosmet	12.851	3.829	0.374		
Leptophos	13.263	1.125	0.085	18.35	1.425
Azinphos-ethyl	13.827	1.812	0.163	21.33	3.952
Dioxathion	14.587	1.392	0.189	7.840	2.198



**Figure 3.** Example chromatogram showing the P signal measured in four food samples, together with a 200 µg/L calibration standard.  
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## Conclusions

The GC-ICP-QQQ method is suitable for the selective and sensitive detection of organophosphorus and organosulfur pesticides by measurement of their heteroatoms. Due to the significantly lower background of the Agilent 8800 ICP-QQQ, GC-ICP-QQQ provides good sensitivity performance for the determination of organophosphorus pesticides compared to GC/MS/MS. Positive traces of pesticides were determined in all of the real food samples surveyed.

## More information

For a full account of this application, see Jenny Nelson, Helene Hopper, Fabio Silva, Steve Wilbur, Jianmin Chen, Kumi Shiota Ozawa, and Philip L. Wylie, Evaluation of GC-ICP-MS/MS as a New Strategy for Specific Heteroatom Detection of Phosphorus, Sulfur, and Chlorine Determination in Foods, *J. Agric. Food Chem.*, 2015, DOI: 10.1021/jf506372e

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