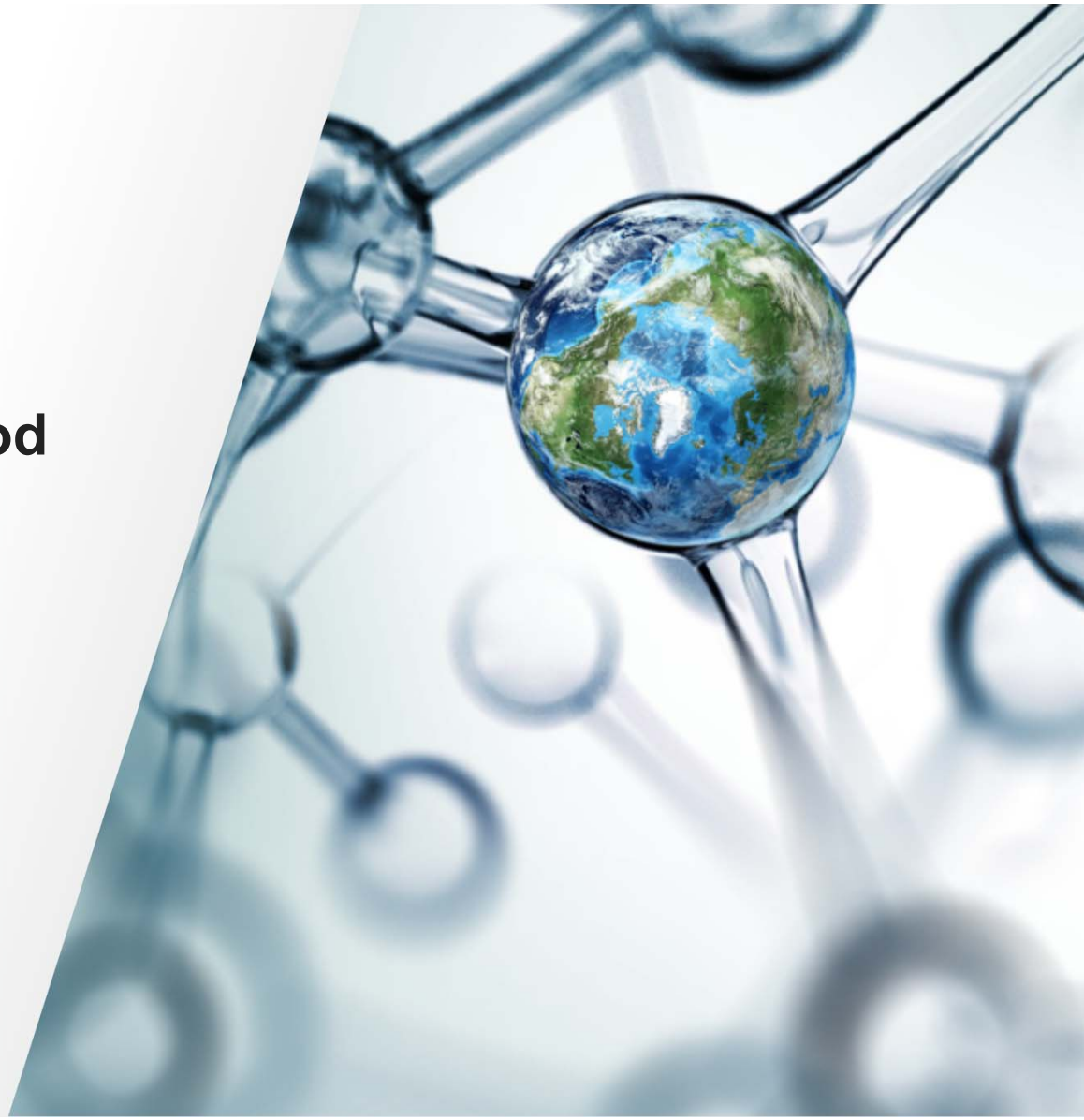
Cationic polar pesticides in food and beverage matrices by IC-MS/MS

John Madden, Ph.D.

May 31, 2021



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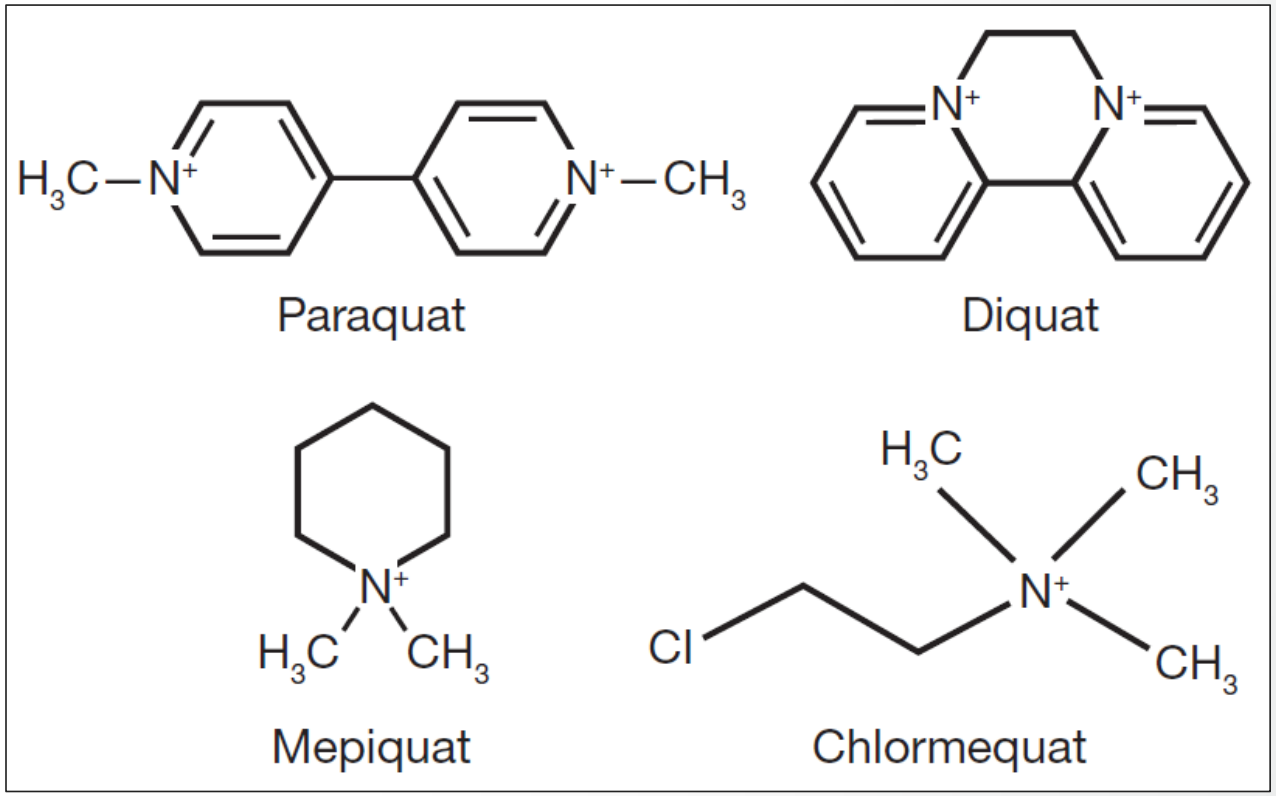
Background

- Ionic polar pesticides include some of the most frequently used pesticides worldwide
- Highly ionic polar pesticides present unique challenges to traditional methods of pesticide analysis
  - Traditional extraction methods typically suffer from low extraction efficiency of highly ionic polar pesticides
  - RPLC methods have very poor retention of highly ionic polar pesticides
- Advances in the analysis of these target analytes has led to an increase in testing and regulation
  - Driven primarily by anionic polar pesticides such as glyphosate and gluphosinate
- The current IC-MS/MS method requires the use of a HRAM instrument due to poor chromatographic resolution of the paraquat – diquat pair
  - Thus, developments in the analysis of quaternary ammonium polar pesticides (quats) has lagged
- This work will review the development of a new method for the analysis of quats in food and beverage matrices
  - Improved chromatographic resolution of PQ-DQ pair negates the need for HRAM

The four target compounds (Quats)



Extraction techniques

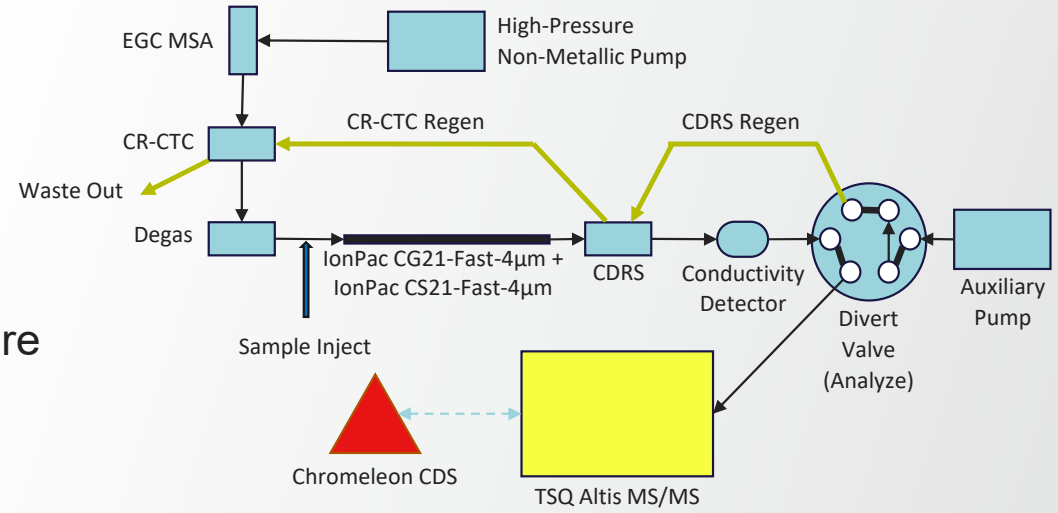
- The most common multiresidue method, QuEChERS, suffers from low extraction efficiency of Quats
  - Developed primarily for extraction of non-polar pesticides
  - Not applicable to this work
- For highly ionic polar pesticides the Quick Polar Pesticides Extraction (QuPPE) method is used
  - Developed by the European Reference Laboratory for Single Residue Methods (EURL-SRM)
  - Based on extraction with methanol/water, followed by centrifugation and filtering
  - No liquid/liquid partitioning or solid phase extraction
  - Extracts contain high levels of co-extractives, such as anions and cations
- In order to evaluate the performance of this new analytical method, the QuPPE extraction technique was applied to two different food samples, carrots and wheat flour, as well as tea extracts
  - The extracts were analyzed, and a synthetic mixture developed to simulate these extracts
  - Quats were spiked into the simulated as well as actual QuPPE extracts as well as a green tea extract
  - Qualitative performance was assessed based on recovery from the spiked synthetic mixture and extracts

Sample preparation

- Carrot Baby Food and Wheat Flour QuPPE extracts
  - 10 g of sample is mixed with 10 g of deionized water in a 50 mL vial and agitated for 5 mins
    - For cereals 5 g of sample is mixed with 15 mL of deionized water
  - 30 mL of cold methanol was added, and the mixture agitated another 1 min
  - The mixture was centrifuged at 4,000 rpm for 5 mins
  - The supernatant was filtered and diluted 1 in 10
- Green Tea
  - A green tea infusion was prepared by boiling 10.0 g of tea leaves in 100 mL of water for 30 s
  - The mixture was allowed to cool before filtering and diluting 1 in 10
- Simulated Matrix
  - Simulated samples were prepared by diluting a Six-Cation Std in deionized water to obtain the total ionic strength (TIS) desired
  - A TIS of 250 ppm was used to simulate the strongest matrix measured from the QuPPE extracts

Instrumentation

- Thermo Scientific™ Dionex™ ICS-6000 IC System
  - Thermo Scientific™ Dionex™ IonPac™ CG21-Fast-4µm (2 × 30 mm) guard and CS21-Fast-4µm (2 × 150 mm) separator
- Thermo Scientific™ Dionex™ AXP Pump
- Thermo Scientific™ TSQ Altis™ Triple Quadrupole Mass Spectrometer
  - Equipped with a heated electrospray ionization (H-ESI) source
- Thermo Scientific™ Chromeleon™ Chromatography Data System (CDS) Software



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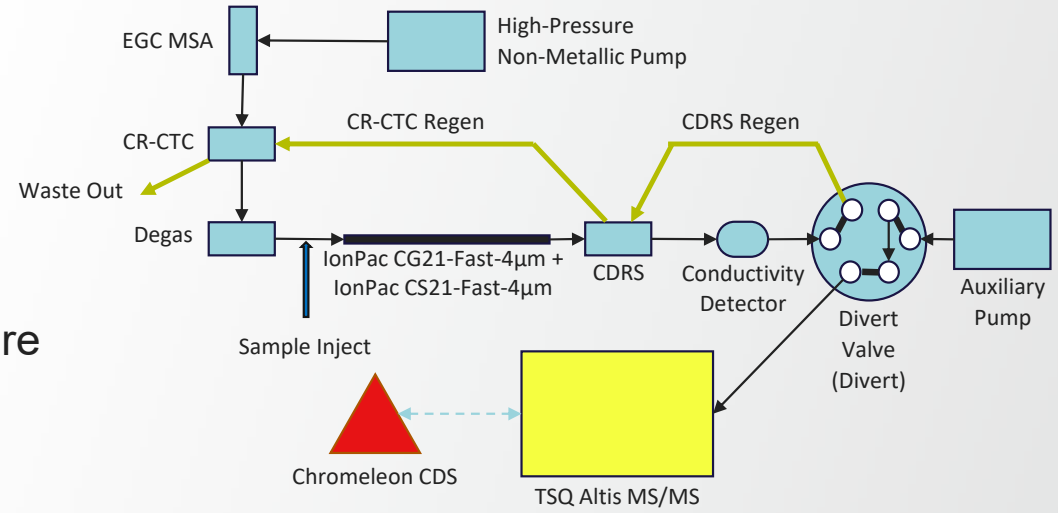

Instrument conditions

Ion Chromatograph Conditions

Mobile Phase:	Methanesulfonic Acid
Eluent Source:	Thermo Scientific™ Dionex™ EGC 500 MSA Cartridge with Thermo Scientific™ Dionex™ CR-CTC III Trap Column
Gradient:	t = -4.0 – 0.0 min 3.0 mM t = 0.0 – 3.6 min 3.0 – 6.0 mM t = 3.6 – 6.0 min 6.0 – 22.0 mM t = 6.0 – 15 min 22.0 – 25.0 mM
Analytical Column:	Dionex IonPac CS21-Fast-4µm Separator with Dionex IonPac CG21-Fast-4µm Guard
Suppressor:	Thermo Scientific™ Dionex™ CDRS 600 Suppressor (2 mm), 22 mA
External Flow Pump:	0.3 mL/min
Eluent Flow Rate:	0.3 mL/min
Injection Volume:	10 µL
Column Temperature:	40 °C

Triple Quadrupole Mass Spectrometer Conditions

Ionization Mode:	Heated Electrospray (H-ESI)
Scan Type:	Selected Reaction Monitoring (SRM)
Polarity:	Positive
Spray Voltage:	2,800 V
Sheath Gas:	45 Arb
Auxiliary Gas:	2.5 Arb
Sweep Gas:	2.0 Arb
Ion Transfer Tube Temp:	350 °C
Vaporizer Temp:	300 °C
Cycle Time:	0.8 sec
Q1 Resolution (FWHM):	0.7
Q3 Resolution (FWHM):	1.2

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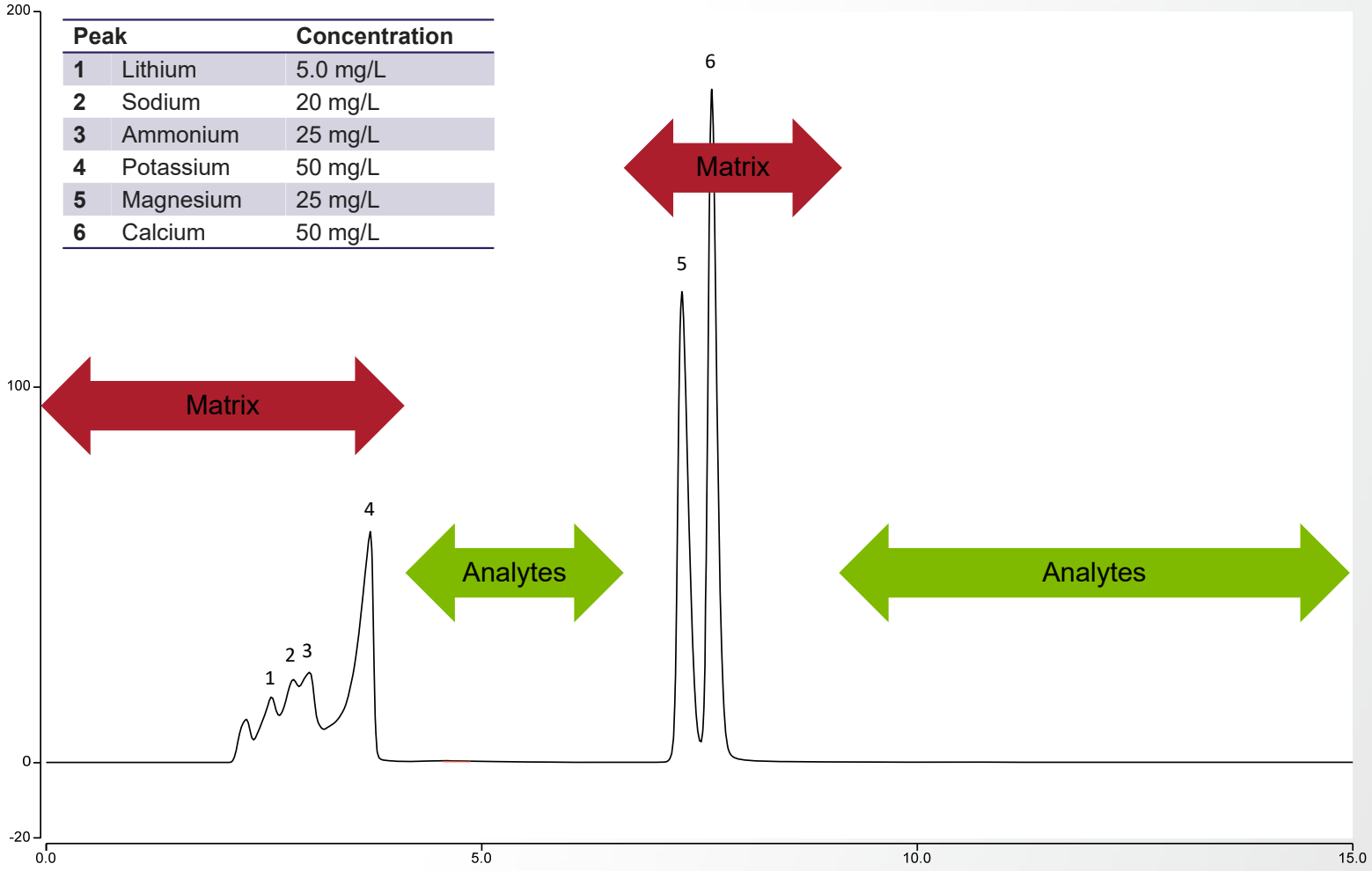
Selected Reaction Monitoring (SRM) transitions

Compound	Transition Type	Parent Ion (m/z)	Product Ion (m/z)
Chlormequat	Quantifier	122.1	57.9
	Qualifier	122.1	62.9
Chlormequat-d4	Quantifier	126.0	57.9
Mepiquat	Quantifier	114.1	98.1
	Qualifier	114.1	58.0
Mepiquat-d16	Quantifier	130.0	110.0
Paraquat	Quantifier	93.0	171.0
	Qualifier	93.0	85.0
Paraquat-d8	Quantifier	97.0	179.0
Diquat	Quantifier	92.0	84.5
	Qualifier	92.0	157.1
Diquat-d8	Quantifier	96.0	88.5

IC separation of the components in the simulated matrix

Peak		Concentration
1	Lithium	5.0 mg/L
2	Sodium	20 mg/L
3	Ammonium	25 mg/L
4	Potassium	50 mg/L
5	Magnesium	25 mg/L
6	Calcium	50 mg/L



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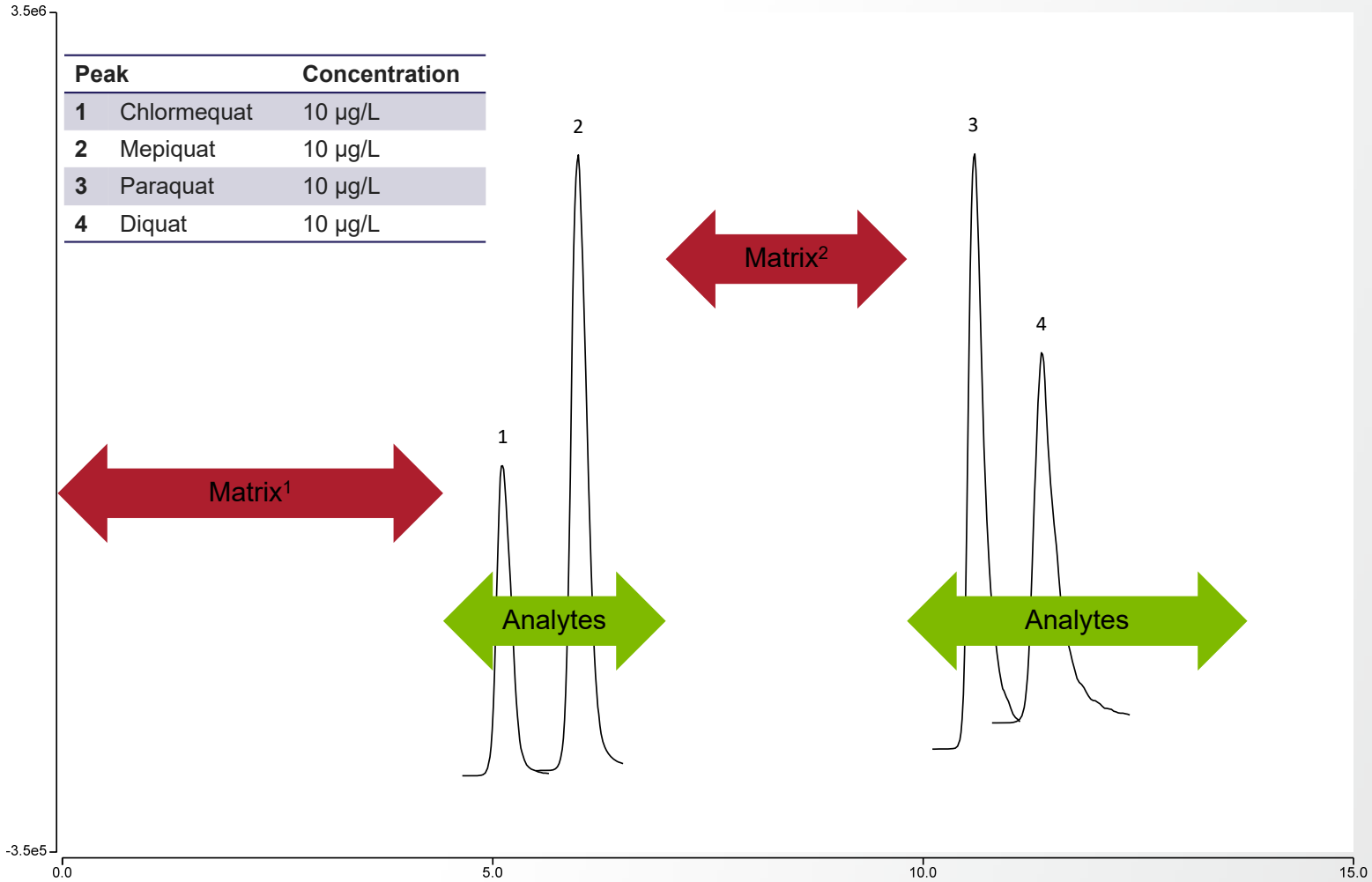
IC-MS separation of the Quats in the simulated matrix

Peak		Concentration
1	Chlormequat	10 µg/L
2	Mepiquat	10 µg/L
3	Paraquat	10 µg/L
4	Diquat	10 µg/L

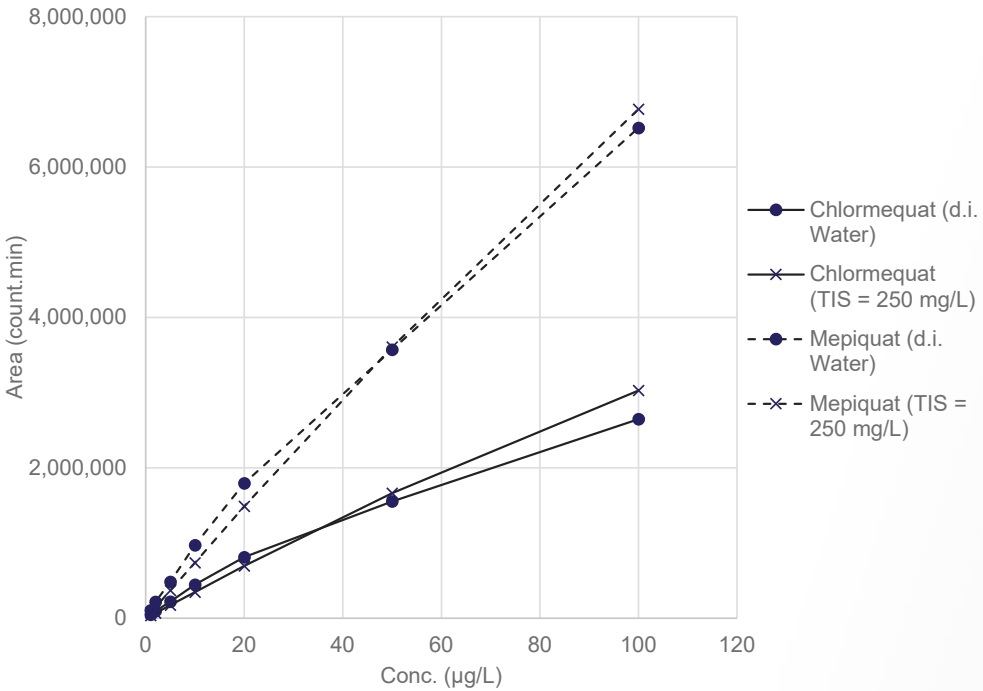


Matrix Effects

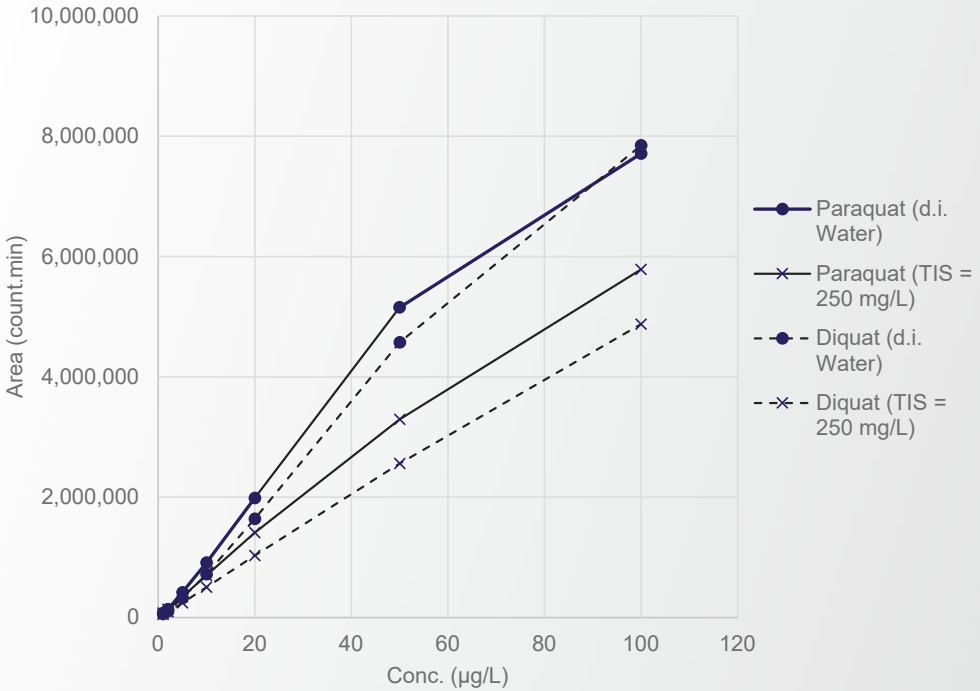
- Matrix-effect signal suppression in the HESI probe must be considered
  - Significant suppression was noted in the high ionic strength matrices upon the divalent species
- Isotopically-labeled internal standard addition was used to compensate
  - Isotopically labeled CQ, MQ, DQ and PQ are all readily available
- Isotopically-labeled Quats were spiked into the sample at the same concentration as the naturally occurring Quats

Compound	Area Response (count.min @ 10 µg/L)	Ratio
Chlormequat	373,966	0.7964
Chlormequat-d4	469,565	
Mepiquat	833,089	0.8413
Mepiquat-d16	990,237	
Paraquat	492,443	0.9330
Paraquat-d8	527,811	
Diquat	379,658	1.0498
Diquat-d8	361,662	

Detector response curves



Detector response curves of CQ (solid lines) and MQ (dashed lines) obtained in two different matrices (deionized water and simulated matrix with Total Ionic Strength = 250 mg/L)



Detector response curves of PQ (solid lines) and DQ (dashed lines) obtained in two different matrices (deionized water and simulated matrix with Total Ionic Strength = 250 mg/L)

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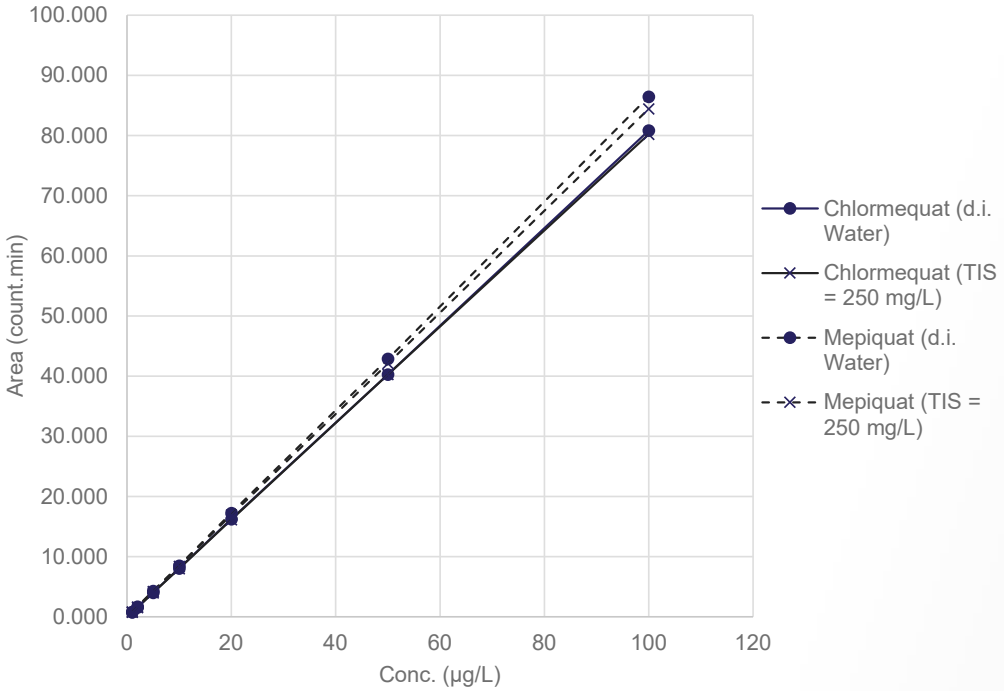
Normalized response curves



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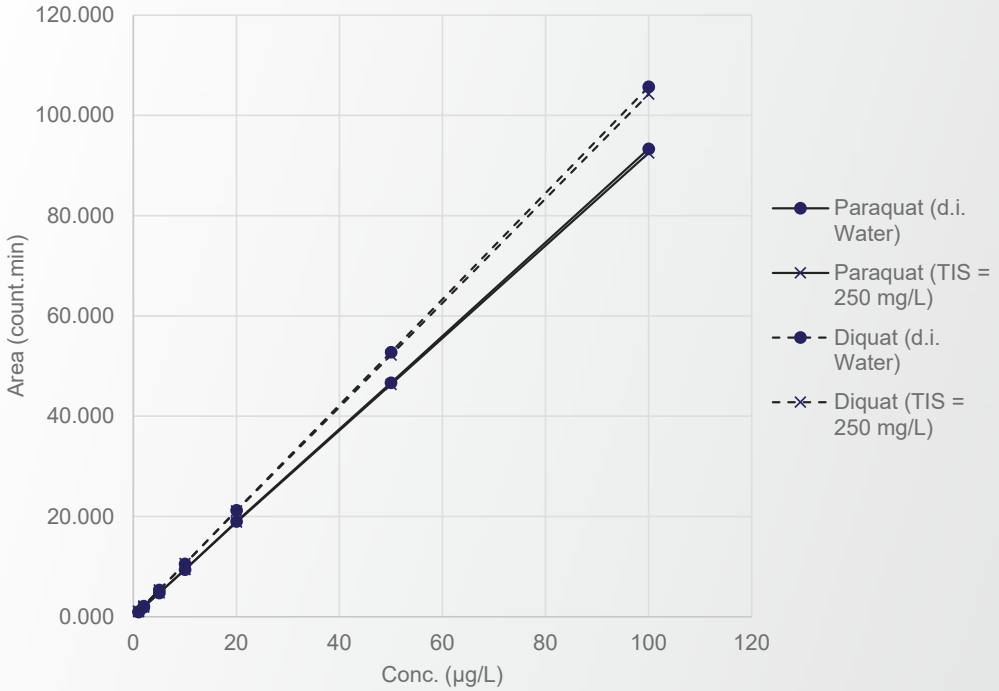

Normalized response curves of PQ (solid lines) and DQ (dashed lines) obtained in two different matrices (deionized water and simulated matrix with Total Ionic Strength = 250 mg/L)

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Confirmation

- Confirmation was based on the presence of the transition ions
  - Quantifier (most abundant) and qualifier (second most abundant)
- The measured peak area ratios of qualifier/quantifier were assessed
  - All ratios were within a range of 3%

Compound	Ion Ratio (qualifier / quantifier)					
	d.i. Water			Synthetic Matrix (TIS = 250 mg/L)		
	1.0 µg/L	10 µg/L	100 µg/L	1.0 µg/L	10 µg/L	100 µg/L
Chlormequat	21.2%	21.8%	22.3%	21.4%	21.4%	21.8%
Mepiquat	33.0%	34.3%	34.1%	33.5%	34.3%	34.1%
Paraquat	17.6%	18.0%	17.5%	17.7%	17.1%	17.2%
Diquat	32.3%	31.9%	33.2%	32.4%	31.8%	33.3%

Detection limits

Compound	RSD (Non-Corrected Response) @ 1 µg/L	
	Reagent Water	Simulated Matrix (TIS = 250 mg/L)
Chlormequat	6.77%	5.44%
Mepiquat	5.93%	4.00%
Paraquat	6.73%	40.56%
Diquat	6.28%	35.29%

Compound	RSD (Corrected Response) @ 1 µg/L	
	Reagent Water	Simulated Matrix (TIS = 250 mg/L)
Chlormequat	0.36%	0.46%
Mepiquat	0.46%	1.12%
Paraquat	1.36%	2.74%
Diquat	1.79%	2.84%

Compound	Detection Limits (µg/L)			
	Reagent Water		Simulated Matrix (TIS = 250 mg/L)	
	LOD	LOQ	LOD	LOQ
Chlormequat	0.01	0.03	0.01	0.04
Mepiquat	0.02	0.04	0.04	0.11
Paraquat	0.05	0.14	0.10	0.31
Diquat	0.06	0.18	0.10	0.31

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Recoveries

Compound	Corrected apparent recoveries in deionized water (%)		
	1.0 µg/L	10 µg/L	100 µg/L
Chlormequat	96%	100%	100%
Mepiquat	97%	101%	101%
Paraquat	99%	100%	99%
Diquat	99%	101%	100%

Compound	Corrected apparent recoveries in simulated matrix (250 mg/L) (%)		
	1.0 µg/L	10 µg/L	100 µg/L
Chlormequat	95%	99%	100%
Mepiquat	98%	99%	101%
Paraquat	103%	101%	99%
Diquat	103%	100%	100%

Compound	Corrected apparent recoveries in green tea (1/10) (%)		
	1.0 µg/L	10 µg/L	100 µg/L
Chlormequat	94%	97%	98%
Mepiquat	100%	99%	100%
Paraquat	102%	99%	98%
Diquat	98%	98%	98%

Compound	Corrected apparent recoveries in QuPPE extracted Carrot Baby Food (1/10) (%)		
	1.0 µg/L	10 µg/L	100 µg/L
Chlormequat	96%	97%	97%
Mepiquat	103%	97%	97%
Paraquat	102%	99%	97%
Diquat	102%	100%	98%

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Conclusion

- The SRM mode used to detect ions of interest was effective for qualitative and quantitative determinations in selected food and beverage samples
- The Dionex IonPac CS21-Fast-4µm column was effective at separating the four quaternary amine cationic polar pesticides from the matrix and each other
  - This made it possible to identify and quantitate all four target compounds using nominal mass selectivity
- The IC method demonstrated high accuracy (80–120% instrument precision values) for all samples
- Sample preparation was simplified by using the already established simplified QuPPe method
  - Re-evaluation with the acidified method recommended for paraquat and diquat is recommended
- This method can be recommended as a reliable and cost-effective addition to any routine lab dealing with the determination of the target cationic pesticides in a wide range of samples