Ignite your Routine Testing Methods

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SCIEX, Concord, Ontario (Canada)
Overview

• Update to the SCIEX LC-MS/MS product portfolio
  – New SCIEX ExionLC™ systems
  – New SCIEX Triple Quad™ and QTRAP® 6500+ systems
  – New

• Application in Food Testing
  – Monofluoroacetate (1080) in milk in infant formula
  – Pesticide screening using ultra-fast polarity switching
  – Triazole derivative metabolites in fruits and vegetables
  – Target identification and quantitation using HR-MS/MS
  – Unknown screening using HR-MS/MS
New SCIEX ExionLC™ Systems

• ExionLC™ 100
  - Simple, non-intimidating, all-in-one box
  - Standard quaternary pump at HPLC pressures for more robustness, less downtime

• ExionLC™ AC
  - Consistent UHPLC performance
  - Minimal downtime

• ExionLC™ AD
  - Full UHPLC, high pressure ratings
  - Lowest carryover system

✔ Fully controlled by Analyst® software 1.6.3
✔ Improved software integration for better stability
New SCIEX Triple Quad™ and QTRAP® 6500+ Systems

• Improved IonDrive™ detection system
  – Quantify with lowest limits of quantitation and up to six orders of linear dynamic range
  – Ultra fast polarity switching with Scheduled MRM™ Pro algorithm for multi-target quantitation

• Elevated SelexION® technology
  – Differential Mobility Spectrometry with improved transmission and no effect on resolution

• Audible noise reduction (active exhaust)

✓ Industry proven performance of IonDrive™ Turbo V ion source and QJet® ion guide
✓ Results in less time – fast, high-throughput, simplified data processing tools
✓ Service and support resources for success
Improved IonDrive™ Detection System

- New detector
  - Larger detection area

- New power supply
  - High energy dynode at 15 kV improves sensitivity
  - Enables faster 5 msec polarity switching

- “Floated” design
  - Ions in negative polarity directed straight into the detector

✓ Contributes to significant signal gains in negative mode Q3 fragment ion <100 Da

✓ Improved data quality in screening applications with ultra-fast polarity switching
Detection of 0.1 ng/mL Monofluoroacetate

High Sensitivity in Negative Polarity using the QTRAP® 6500+ system

Sample preparation protocol and LC conditions adopted from the AOAC First Action Official Method 2015.03, MS/MS using QTRAP® 6500+ system
Switching between positive and negative polarity in 5 msec

552 MRM transitions
Positive ESI

Flufenacet CV = 4.37%

Methoxyfenozide CV = 1.52%

58 MRM transitions
Negative ESI

Phenomenex Kinetex™ Biphenyl 2.6u (50 x 2.1mm) column
Gradient of water/methanol + 5 mM ammonium formate
~300 pesticides at 1 ng/mL, MS/MS using QTRAP® 6500+ system
SelexION®+ DMS with Jet Injector Technology

- New DMS cell designed to improve ion transmission by ~2x without loss in selectivity or resolution
  - Addition of lens increases ion velocities into DMS cell
  - Reduces transit times through detrimental fringing fields

✓ Continually driving selectivity improvements
Reduced Noise and Interferences when Using DMS

Example: Triazole in Fruit and Vegetable Extracts

6500+ with DMS

6500+ DMS removed

Original method developed by J. Jasak and R. Schöning, also published as “EURL-SRM QuPPe-Method 8” by M. Anastassiades et al.

MS/MS using QTRAP® 6500+ system
A new high resolution mass spec system is on the horizon

Mission Complete >
What’s New?

SCIEX X500R QTOF System powered by SCIEX OS Software

SCIEX X500R QTOF system

SCIEX ExionLC™ AC system

SCIEX OS software
Introducing the SCIEX X500R QTOF System

• Hardware
  – SCIEX ExionLC™ Systems
    – Fully controlled by SCIEX OS software
    – Improved software integration for better stability
  – SCIEX X500R QTOF System
    – N-optic design
    – Heated TOF path
    – Minimized footprint, engineered for simplicity and service accessibility

• Software
  – SCIEX OS Software
    – New user interface
    – Simultaneous identification and quantitation
    – Automatic unknown identification

• Application data
  – Target identification and quantitation
  – Unknown screening
Introducing the SCIEX X500R QTOF System

Design Improvements and Details

**Engineered for simplicity**
Optimal performance delivered through adjustment of only 2 voltage variables.

**Minimized footprint**
The benchtop stature (110 x 57 x 112 cm)* occupies less lab space than any other HRMS system on the market.

**TwinSpray**
An independent calibrant delivery path for reliable auto-calibration.

**Integrated calibration**
Maintains mass calibration through long runs without effect on sample flow.

**Service accessibility**
Easy QJet access for fast and efficient maintenance and single three stage split flow pump for increased system uptime.
Introducing the SCIEX X500R QTOF System

Design Improvements and Details

**N-optic design**
4 mm orifice leading into TOF accelerator tube delivers resolution without compromise in sensitivity.

**Heated TOF path**
Six heater drones throughout the TOF path to maintain robustness and mass accuracy.
Introducing the SCIEX X500R QTOF System

Design Improvements and Details

Legendary Turbo V source with optional IonDrive and Curtain Gas interface
Renowned ionization performance and ruggedness now delivered with a high resolution accurate mass analyzer.
Perfect Balance to Elevate Your Lab’s Performance

SCIEX X500R QTOF System

• The first robust, high performance high resolution MS/MS system designed for routine use.
  – Sensitivity to easily detect maximum residue levels
  – Resolving power to remove interference from complex food matrices
  – Linearity to quantify over up to 4 orders of magnitude
  – Mass accuracy to identify compounds following regulatory guidelines
  – Confident identification based MS/MS (IDA and SWATH™ MS/MS<sup>ALL</sup>, ion ratios and MS/MS spectra)
  – Industry leading robustness of Turbo V™ source and Curtain Gas™ interface
The SCIEX X500R QTOF System – Performance

Sensitivity and Resolution (20 µg/kg Pesticides in Fruit, 5 µL injected)

Cyromazine @ 167.104
Carbendazime @ 192.077
Imazalil @ 297.056
Trifloxystrobin @ 409.137
Spinosyn A @ 732.468

Data Acquisition

Mass Spec

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The SCIEX X500R QTOF System – Performance

Linearity and Mass Accuracy (Paclobutrazol 0.1 to 10,000 ng/mL)

- **Mass error**: -0.2 to 0.91 ppm
- **r²**: 0.9993
- **4 orders linear dynamic range**
Information Dependent Acquisition of MS/MS (IDA)

Provides MS/MS Spectra with High Selectivity (Q1 Resolution unit)

IDA can be used with and without inclusion list for target or non-target screening
Confident Identification of Isomers using MS/MS (Prometon vs. Terbumeton)

Prometon
\( \text{C}_{10}\text{H}_{19}\text{N}_{5}\text{O}+\text{H}^+ \)
RT = 5.44 min

MS: 226.1664 (0.63 ppm)
MS/MS: 98.5%

Terbumeton
\( \text{C}_{10}\text{H}_{19}\text{N}_{5}\text{O}+\text{H}^+ \)
RT = 5.53 min

MS 226.1665 (0.99 ppm)
MS/MS: 95.7%

Identification based on MS/MS library searching (Q1 at unit resolution)
Increased Selectivity using MRM\textsuperscript{HR}

Feed Sample Tested Positive for NP-Semicarbazide

MRM 209/192 and 209/166

TOF-MS XIC of 209.0675 ± 10 ppm

MRM\textsuperscript{HR} 209/192.0404 and 209/166.0611
Perfect Balance to Elevate Your Lab’s Performance

SCIEX X500R QTOF System Powered by SCIEX OS Software

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  ‒ Resolving power to remove interference from complex food matrices
  ‒ Linearity to quantify over up to 4 orders of magnitude
  ‒ Mass accuracy to identify compounds following regulatory guidelines
  ‒ Confident identification based MS/MS (IDA and SWATH™ MS/MS ALL, ion ratios and MS/MS spectra)
  ‒ Industry leading robustness of Turbo V™ source and Curtain Gas™ interface

• Simple software workflows that deliver reliable results, it’s the solution that’s ready to meet your challenges today and for the future, from the trusted LC-MS/MS industry leader.
Introducing the SCIEX OS Software

Single Software Platform for MS Control, Data Processing and Reporting
SCIEX OS Software

Software Improvements and Details

New user interface
Thoughtfully designed for faster learning and improved productivity

Acquire, analyze, report
Run the mass spectrometer and perform data processing, review and reporting in the same software
SCIEX OS Software – Home

Everything in a Single Software Platform

Acquisition
Build MS and LC methods
Create batches
Run samples

Processing
Simultaneous Identification and quantitation

Management
Adjust hardware, software, and user settings
1. Separate Method Editor for LC and MS Methods

<table>
<thead>
<tr>
<th>Source and Gas Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ion source gas 1: 50 psi</td>
</tr>
<tr>
<td>Ion source gas 2: 70 psi</td>
</tr>
<tr>
<td>Temperature: 450 °C</td>
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<table>
<thead>
<tr>
<th>TOF-MS</th>
</tr>
</thead>
<tbody>
<tr>
<td>TOF start mass: 100 Da</td>
</tr>
<tr>
<td>TOF stop mass: 1000 Da</td>
</tr>
<tr>
<td>Accumulation time: 0.2 sec</td>
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</table>

<table>
<thead>
<tr>
<th>IDA Criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maximum candidate ions: 10</td>
</tr>
<tr>
<td>Intensity threshold exceeds: 100 cps</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>TOF-MS/MS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Presursor ion: 100 Da</td>
</tr>
<tr>
<td>TOF start mass: 50 Da</td>
</tr>
<tr>
<td>TOF stop mass: 1000 Da</td>
</tr>
<tr>
<td>Accumulation time: 0.05 sec</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>TOF-MS/MS criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dynamic background subtraction</td>
</tr>
<tr>
<td>Exclude former candidate ions</td>
</tr>
<tr>
<td>For: 10 sec, After: 15 occurrences</td>
</tr>
<tr>
<td>Declustering potential: 80 V</td>
</tr>
<tr>
<td>Collision energy: 10 V</td>
</tr>
<tr>
<td>DP spread: 0 V</td>
</tr>
<tr>
<td>CE spread: 0 V</td>
</tr>
</tbody>
</table>
Auto-calibration using the built-in Calibrant Delivery System (CDS)
### 3. Batch Editor and Setup for Auto-Calibration

Smart grid to assist in building the batch

- Copy/paste, fill down, auto increment, import/export
SCIEX OS Software – Queue, Live View and Status

4. Queue Manager with Inserted Auto-Cal Samples

Detailed instrument status

Live TIC

Live Spectrum
SCIEX OS Software – Identification and Quantitation

Software Improvements and Details

- Triple quadrupole like quantitation
- Simultaneous identification and quantitation
  Powerful new tools to deliver quantitative and qualitative results from high resolution MS and MS/MS data.
- Processing of large compound lists
- Intuitive Filtering for easy data review
Targeted Data Processing Workflow

XIC generation for each target

Identification

Quantitation

RT ✓  Mass Isotopes ✓  MS/MS search ✓

Above reporting level
### Identification and Quantitation in SCIEX OS Software

1. Define Retention Time and Accurate Mass for Each Target Analyte

![Image of SCIEX OS Software interface showing the Modify Method window with a table of analytes and their properties.]
2. Define Identification Criteria and Confidence Settings
3. Review Quantitative and Quantitative Results

Flagging when above MRL and traffic lights for confidence

Review of peak integration, spectra and calibration line
Pesticides in Store-bought Fruits and Vegetables

QuEChERS Extracts with 10x Dilution (5 µL Injection)

- Organic strawberry
- Strawberry
- Lemon
- Grapes
- Spinosyn A
- Boscalid
- Imazalil
- Cyprodinil
# Pesticides (> 5 µg/kg) in Store-bought Fruits and Vegetables

<table>
<thead>
<tr>
<th>Sample</th>
<th>Pesticide</th>
<th>Concentration (µg/kg)</th>
<th>RT error (%)</th>
<th>Mass error (ppm)</th>
<th>Isotope ratio error (%)</th>
<th>MS/MS PUR (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Organic strawberry</strong></td>
<td>Spinosyn A</td>
<td>13.9</td>
<td>0.01</td>
<td>0.55</td>
<td>9.1</td>
<td>100.0</td>
</tr>
<tr>
<td></td>
<td>Spinosyn D</td>
<td>33.3</td>
<td>0.01</td>
<td>1.63</td>
<td>6.0</td>
<td>99.4</td>
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<tr>
<td><strong>Strawberry</strong></td>
<td>Acetamiprid</td>
<td>19.2</td>
<td>0.08</td>
<td>-0.35</td>
<td>6.5</td>
<td>98.7</td>
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<tr>
<td></td>
<td>Boscalid</td>
<td>161</td>
<td>0.00</td>
<td>-0.49</td>
<td>4.9</td>
<td>99.3</td>
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<tr>
<td></td>
<td>Myclobutanil</td>
<td>85.0</td>
<td>0.00</td>
<td>-0.31</td>
<td>13.9</td>
<td>100.0</td>
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<tr>
<td></td>
<td>Pyraclostrobin</td>
<td>40.5</td>
<td>0.00</td>
<td>1.33</td>
<td>16.3</td>
<td>99.0</td>
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<tr>
<td></td>
<td>Pyrimethanil</td>
<td>391</td>
<td>0.00</td>
<td>0.32</td>
<td>4.7</td>
<td>97.3</td>
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<tr>
<td><strong>Blueberry</strong></td>
<td>n.d.</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
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<tr>
<td><strong>Organic Banana</strong></td>
<td>Spinosyn D</td>
<td>12.6</td>
<td>0.00</td>
<td>2.33</td>
<td>19.8</td>
<td>100.0</td>
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<tr>
<td><strong>Banana</strong></td>
<td>Buprofezin</td>
<td>341</td>
<td>0.01</td>
<td>0.32</td>
<td>3.5</td>
<td>100.0</td>
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<tr>
<td></td>
<td>Imazalil</td>
<td>565</td>
<td>0.02</td>
<td>0.79</td>
<td>15.1</td>
<td>91.5</td>
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<tr>
<td></td>
<td>Thiabendazole</td>
<td>444</td>
<td>0.01</td>
<td>-1.51</td>
<td>13.9</td>
<td>97.6</td>
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<tr>
<td><strong>Lemon</strong></td>
<td>Imazalil</td>
<td>1080</td>
<td>0.02</td>
<td>0.74</td>
<td>7.3</td>
<td>94.7</td>
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<tr>
<td></td>
<td>Pyrimethanil</td>
<td>164</td>
<td>0.01</td>
<td>-0.77</td>
<td>1.0</td>
<td>99.2</td>
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<td>Pyriproxyfen</td>
<td>31.6</td>
<td>0.01</td>
<td>0.43</td>
<td>11.4</td>
<td>95.3</td>
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<tr>
<td><strong>Spinach</strong></td>
<td>n.d.</td>
<td>-</td>
<td>-</td>
<td>-</td>
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<tr>
<td><strong>Grapes</strong></td>
<td>Boscalid</td>
<td>115</td>
<td>0.01</td>
<td>-0.80</td>
<td>8.8</td>
<td>97.2</td>
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<tr>
<td></td>
<td>Buprofezin</td>
<td>17.3</td>
<td>0.01</td>
<td>0.22</td>
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<td>99.6</td>
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<td>Cyprodinil</td>
<td>412</td>
<td>0.01</td>
<td>-0.87</td>
<td>3.3</td>
<td>94.8</td>
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<td>Imadacloprid</td>
<td>82.5</td>
<td>0.01</td>
<td>-0.58</td>
<td>14.6</td>
<td>96.1</td>
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<tr>
<td></td>
<td>Pyraclostrobin</td>
<td>46.7</td>
<td>0.00</td>
<td>-1.31</td>
<td>4.8</td>
<td>100.0</td>
</tr>
</tbody>
</table>
SCIEX OS Software – Unknown Identification

Software Improvements and Details

Automatic sample-control comparison

ChemSpider searching
Sorting by number of references and automatic structure elucidation to identify unknowns

Non-Target peak finding

Automatic MS/MS library searching and formula finding
Non-Targeted Data Processing Workflow

Sample-control comparison

Mass and RT

Identification

Formula finding ✓  MS/MS search ✓
1. Define Non-Target Peak Finding and Identification Parameters
Unknown Identification in SCIEX OS Software

2. Automatic Sample-Control Comparison and Compound Identification

Automatic formula finding and MS/MS library searching
Unknown Identification in SCIEX OS Software

3. ChemSpider Searching and Structure Elucidation

ChemSpider searching of found formulae
Automatic structure elucidation using HR-MS/MS spectra
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ChemSpider searching of found formulae

Automatic structure elucidation using HR-MS/MS spectra
Summary

• Hardware and Software
  – New SCIEX ExionLC™ systems
    – Fully controlled by SCIEX OS software
  – New SCIEX Triple Quad™ and QTRAP® 6500+ systems
    – Improved IonDrive™ detection system
    – Elevated SelexION® technology
  – New SCIEX X500R QTOF System
    – N-optic design
    – Heated TOF path
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• Application data
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