

# Citrine Systems

## System User Guide



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# Operational Precautions and Limitations

# 1

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**Note:** Before operating the system, carefully read all of the sections of this guide.

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This section contains general safety-related information and provides regulatory compliance information. It also describes potential hazards and associated warnings for the system and the precautions that should be taken to minimize the hazards.

In addition to this section, for information about the symbols and conventions used in the laboratory environment, on the system, and in this documentation, refer to the section: [Glossary of Symbols](#). For site requirements, including mains supply, source exhaust, ventilation, compressed air, nitrogen, and roughing pump requirements, refer to the document: *Site Planning Guide*.

## General Safety Information

To prevent personal injury or system damage, read, understand, and obey all safety precautions and warnings in this document, the manufacturer chemical safety data sheets (SDSs), and product label information. Labels are shown with internationally recognized symbols. Failure to heed these warnings could result in serious injury.

This safety information is intended to supplement federal, state, provincial, and local environmental health and safety (EHS) regulations. The information provided covers system-related safety information applicable to the operation of the system. It does not cover every safety procedure that should be practiced. Ultimately, the user and the organization are responsible for compliance with federal, state, provincial, and local EHS regulations and maintaining a safe laboratory environment.

Refer to the appropriate laboratory reference material and standard operating procedures.

## Documentation Symbols and Conventions

The following symbols and conventions are used throughout the guide.



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**DANGER!** Danger signifies an action that leads to severe injury or death.

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**WARNING!** Warning signifies an action that could cause personal injury if precautions are not followed.

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**CAUTION:** Caution signifies an operation that could cause damage to the system or corruption or loss of data if precautions are not followed.

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**Note:** Note emphasizes significant information in a procedure or description.

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**Tip!** Tip provides useful information that helps apply the techniques and procedures in the text for a specific need and provides shortcuts, but is not essential to the completion of a procedure.

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## Regulatory Compliance

This system complies with the regulations and standards listed in this section. For dated references, refer to the *Declaration of Conformity* included with the system and the individual system components. Applicable labels have been affixed to the system.

### Canada

- **Electromagnetic Interference (EMI):** CAN/CSA CISPR11. This ISM device complies with Canadian ICES-001. Refer to the section: [Electromagnetic Interference](#).
- **Safety:**
  - CAN/CSA C22.2 No. 61010-1
  - CAN/CSA C22.2 No 61010-2-061
  - CAN/CSA C22.2 No. 61010-2-101

### Europe

- **In Vitro Diagnostic Devices (IVD):** In Vitro Diagnostics Regulation 2017/746
- **Electromagnetic Compatibility (EMC):** Electromagnetic Compatibility Directive 2014/30/EU as implemented in these standards:
  - EN 61326-1
  - EN 61326-2-6
  - EN 55011 (Class A)

Refer to the section: [Electromagnetic Compatibility](#).

## Operational Precautions and Limitations

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- **Safety:** Low Voltage Directives 2014/35/EU as implemented in these standards:
  - EN 61010-1
  - EN 61010-2-061
  - EN 61010-2-101
- **Waste Electrical and Electronic Equipment (WEEE):** Waste Electrical and Electronic Equipment Directive 2012/96/EEC, as implemented in EN 40519. Refer to the section: [Waste Electrical and Electronic Equipment](#).
- **Packaging and Packaging Waste (PPW):** Packaging and Packaging Waste Directive 94/62/EC
- **RoHS Restriction of Hazardous Substances:** RoHS Directive 2011/65/EU and 2015/863/EU

## United States

- **Radio Emissions Interference Regulations:** 47 CFR 15, as implemented in FCC Part 15 (Class A)
- **Safety:** Occupational Safety and Health Regulations, 29 CFR 1910, as implemented in these standards:
  - UL 61010-1
  - IEC 61010-2-061
  - IEC 61010-2-101

## International

- **Electromagnetic Compatibility (EMC):**
  - IEC 61326-1
  - IEC CISPR 11 (Class A)
  - IEC 61000-3-2
  - IEC 61000-3-3

Refer to the section: [Electromagnetic Compatibility](#).

- **Safety:**
  - IEC 61010-1
  - IEC 61010-2-061

- IEC 61010-2-101

## Electrical Precautions



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**WARNING! Electrical Shock Hazard. Do not remove the covers. Removing the covers might cause injury or malfunctioning of the system. The covers need not be removed for routine maintenance, inspection, or adjustment. Contact a SCIEX Field Service Employee (FSE) for repairs that require the covers to be removed.**

---

- Follow required electrical safe work practices.
- Use cable management practices to control electrical cables. This will decrease the chance of a tripping hazard.

For information about system electrical specifications, refer to the section: [Performance Characteristics and Specifications](#) or the document: *Site Planning Guide*.

## Mains Supply

Connect the system to a compatible mains supply as instructed in this guide.



---

**WARNING! Electrical Shock Hazard. Use only qualified personnel for the installation of all of the electrical supplies and fixtures, and make sure that all of the installations adhere to local regulations and safety standards.**

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**WARNING! Electrical Shock Hazard. Make sure that the system can be disconnected from the mains supply outlet in an emergency. Do not block the mains supply outlet.**

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**WARNING! Electrical Shock Hazard. Use only the mains supply cables supplied with the system. Do not use mains supply cables that are not properly rated for the operation of this system.**

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## Protective Earth Conductor

The mains supply must include a correctly installed protective earth conductor. The protective earth conductor must be installed or examined by a qualified electrician before the system is connected.

## Operational Precautions and Limitations

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**WARNING! Electrical Shock Hazard.** Do not intentionally interrupt the protective earth conductor. Any interruption of the protective earth conductor creates an electrical shock hazard.

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**WARNING! Electrical Shock Hazard.** Make sure that a protective earth conductor (grounding cable) is connected between the sample loop and an appropriate grounding point at the ion source. This supplementary grounding reinforces the safety configuration specified by SCIEX.

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## Chemical Precautions

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**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** Determine whether decontamination is required before cleaning or maintenance. If radioactive materials, biological agents, or toxic chemicals have been used with the system, then the customer must decontaminate the system before cleaning or maintenance.

---



**WARNING! Environmental Hazard.** Do not dispose of system components in municipal waste. Follow local regulations when disposing of components.

---



**WARNING! Biohazard or Toxic Chemical Hazard.** Connect the drain tubing to the mass spectrometer and the source exhaust drain bottle properly, to prevent leaks.

---

- Determine which chemicals have been used in the system prior to service and regular maintenance. For the health and safety precautions that must be followed for a chemical, refer to the document: *Safety Data Sheet*. For storage information, refer to the document: *Certificate of Analysis*. To find a SCIEX *Safety Data Sheet* or *Certificate of Analysis*, go to [sciex.com/tech-regulatory](http://sciex.com/tech-regulatory).
  - Always wear assigned personal protective equipment, including powder-free gloves, safety glasses, and a laboratory coat.
- 

**Note:** Nitrile or neoprene gloves are recommended.

---

- Work in a well-ventilated area or fume hood.
  - Avoid ignition sources when working with flammable materials, such as isopropanol, methanol, and other flammable solvents.
  - Take care in the use and disposal of any chemicals. There is a potential risk of personal injury if proper procedures for handling and disposal of chemicals are not followed.
  - Avoid skin contact with chemicals during cleaning, and wash hands after use.
-

- Make sure that all exhaust hoses are connected properly and that all connections are functioning as designed.
- Collect all spent liquids and dispose of them as hazardous waste.
- Comply with all of the local regulations for the storage, handling, and disposal of biohazardous, toxic, and radioactive materials.
- (Recommended) Use secondary containment trays beneath the roughing pump, the solvent bottles, and the waste container to capture potential chemical spills.

## System Safe Fluids

The following fluids can safely be used with the system. For information about safe cleaning solutions, refer to the section: [Required Materials \(Not Supplied\)](#).



**CAUTION:** Potential System Damage. Do not use any other fluid until confirmation is received from SCIEX that it does not present a hazard. This is not an exhaustive list.

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**Note:** Use only new, freshly prepared LC-MS-grade or better solvents for the LC mobile phases.

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- **Organic Solvents**
  - LC-MS-grade acetonitrile; up to 100%
  - LC-MS-grade methanol; up to 100%
  - LC-MS-grade isopropanol; up to 100%
  - LC-MS-grade or higher water; up to 100%
  - Tetrahydrofuran; up to 100%
  - Toluene and other aromatic solvents; up to 100%
  - Hexanes; up to 100%
- **Buffers**
  - Ammonium acetate; less than 100 mM
  - Ammonium formate; less than 100 mM
  - Phosphate; less than 1%
- **Acids and Bases**
  - Formic acid; less than 1%
  - Acetic acid; less than 1%
  - Trifluoroacetic acid (TFA); less than 1%

## Operational Precautions and Limitations

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- Heptafluorobutyric acid (HFBA); less than 1%
- Ammonia/ammonium hydroxide; less than 1%
- Phosphoric acid; less than 1%
- Trimethylamine; less than 1%
- Triethylamine; less than 1%

## Ventilation Precautions

The venting of fumes and disposal of waste must comply with all of the federal, state, provincial, and local health and safety regulations. It is the responsibility of the customer to make sure that the air quality is maintained in compliance with local health and safety regulations.

The source exhaust system and roughing pump must be vented to a dedicated laboratory fume hood or an external exhaust system.



**WARNING! Fire Hazard. Make sure that the source exhaust system is connected and functioning, to prevent flammable vapor from accumulating in the ion source.**

---



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Take care to vent exhaust gases to a dedicated laboratory fume hood or exhaust system and make sure that the ventilation tubing is secured with clamps. Make sure that the laboratory has appropriate air exchange for the work performed.**

---



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Do not operate the mass spectrometer if the source exhaust drain and roughing pump exhaust hoses are not properly connected to the laboratory ventilation system. Examine the exhaust tubing regularly to make sure that there are no leaks. The use of mass spectrometers without proper system ventilation might constitute a health hazard and might result in serious injury.**

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**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Do not use the ion source without knowledge of and training in the proper use, containment, and evacuation of toxic or injurious materials used with the ion source.**

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**WARNING! Puncture Hazard, Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** Discontinue use of the ion source if the ion source window is cracked or broken, and then contact a SCIEX Field Service Employee (FSE). Any toxic or injurious materials introduced in the equipment will be present in the source exhaust output. Exhaust from equipment should be vented from the room. Dispose of sharps following established laboratory safety procedures.

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## Physical Precautions

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**WARNING! Hot Surface Hazard.** Let the IonDrive Turbo V ion source cool for at least 90 minutes before starting any maintenance procedures. Some surfaces of the ion source and vacuum interface become hot during operation.

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**WARNING! Lifting Hazard.** Use a mechanical lifting device to lift and move the mass spectrometer. If the mass spectrometer must be moved manually, then at least six people are required to move it safely. Follow established safe lifting procedures. We recommend the use of a professional moving service. For the weights of system components, refer to the document: *Site Planning Guide*.

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## Environmental Precautions

Use qualified personnel for the installation of electrical mains, heating, ventilation, and plumbing supplies and fixtures. Make sure that all of the installations comply with local bylaws and biohazard regulations. For information about the required environmental conditions for the system, refer to the document: *Site Planning Guide*.

Allow access space around the equipment when setting up the system.



**DANGER! Explosion Hazard.** Do not operate the system in an environment containing explosive gases. The system is not designed for operation in an explosive environment.

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**WARNING! Biohazard.** For biohazardous material use, always comply with local regulations for hazard assessment, control, and handling. This system or any part is not intended to act as a biological containment.

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## Operational Precautions and Limitations

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**WARNING! Environmental Hazard.** Follow established procedures for disposal of biohazardous, toxic, radioactive, and electronic waste. The customer is responsible for disposal of hazardous substances, including chemicals, waste oils, and electrical components, in accordance with local laws and regulations.

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**CAUTION: Potential Mass Shift.** Maintain a stable ambient temperature. If the temperature changes by more than 2 °C per hour, then the resolution and mass calibration might be affected.

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**CAUTION: Potential System Contamination.** When using a gas generator, refer to the documentation that accompanies the gas generator for information from the manufacturer on the use of a gas generator with a compressor. For example, use of a gas generator with a compressor can result in hydrocarbons entering the mass spectrometer, if they are present in the environment.

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## Electromagnetic Environment

### Electromagnetic Compatibility

**Basic Electromagnetic Environment:** Environment existing at locations characterized by being supplied directly at low voltage from the public mains network.

**Performance Criteria A (Criteria A):** Equipment shall operate as intended with no degradation of performance and no loss of function during or after the test.

**Performance Criteria B (Criteria B):** Equipment may experience loss of function (one or more) during the test but shall operate as intended with some degradation of performance and functions self-recoverable after the test.

**Performance Criteria C (Criteria C):** Equipment may experience loss of function (one or more) during the test but shall operate as intended with some degradation of performance and functions recoverable by an operator after the test.

The equipment is intended for use in a basic electromagnetic environment.

The expected performance loss under the electromagnetic immunity conditions is less than 20% change in total ion count (TIC).

**CAUTION: Potential Wrong Result.** Do not use this device in close proximity to sources of strong electromagnetic (EMC) radiation, such as unshielded intentional RF sources, because EMC radiation might interfere with the proper operation.

---

Make sure that a compatible electromagnetic environment for the equipment can be maintained so that the device will operate as intended. If the power supply line is subject to high electrical noise, then install a surge protector.

### Electromagnetic Interference

**Group 1 Equipment:** This equipment is classified as industrial, scientific, and medical (ISM) equipment that might use RF energy for internal operation.

**Class A Equipment:** Equipment which is suitable for use in all establishments other than domestic and those directly connected to a low voltage power supply network which supplies buildings used for domestic purposes. [Derived from CISPR 11:2009, 5.3] Class A equipment shall meet Class A limits.

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**CAUTION: Potential Radio Interference. This equipment is not intended for use in residential environments and may not provide adequate protection to radio reception in such environments.**

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This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to Part 15 of the FCC (Federal Communications Commission) Compliance Rules.

These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the operator's manual, can cause harmful interference to radio communications.

Operation of this equipment in a residential area is likely to cause harmful interference in which case you will be required to correct the interference, at your own expense. Changes or modifications not expressly approved by the manufacturer could void your authority to operate the equipment.

### Decommissioning and Disposal



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**WARNING! Environmental Hazard. Follow established procedures for disposal of biohazardous, toxic, radioactive, and electronic waste. The customer is responsible for disposal of hazardous substances, including chemicals, waste oils, and electrical components, in accordance with local laws and regulations.**

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Before decommissioning, decontaminate the entire system following local regulations.

When removing the system from service, separate and recycle different materials according to national and local environmental regulations. Refer to the section: [Storage and Handling](#).

## Operational Precautions and Limitations

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**Note:** SCIEX will not accept any system returns without a completed Decontamination Form. Contact an FSE to obtain a copy of the form.

---

Do not dispose of system components or subassemblies, including computer parts, as unsorted municipal waste.

## Waste Electrical and Electronic Equipment

Follow local municipal waste ordinances for proper disposal provisions to reduce the environmental impact of waste, electrical, and electronic equipment (WEEE). To safely dispose of this equipment, contact a local Customer Service office for complimentary equipment pick-up and recycling.

## Intended Use

The Citrine system is a liquid chromatography-tandem mass spectrometry (LC-MS/MS) systems intended to identify inorganic or organic compounds in human specimens by ionizing the compound under investigation and separating the resulting ions by means of an electrical field according to their mass. For *In Vitro* Diagnostic Use.

## Limitations of Use

The Citrine system is intended for use in a clinical laboratory environment by qualified laboratory personnel for professional use only.

## Description

The Citrine system includes the following components:

- A Citrine QTRAP mass spectrometer or a Citrine Triple Quad mass spectrometer with a IonDrive Turbo V ion source that uses either the TurbolonSpray probe or the atmospheric pressure chemical ionization (APCI) probe, and roughing pumps.
- A SCIEX-supplied computer and monitor with the Analyst MD software for instrument optimization, acquisition method development, and data acquisition, and the MultiQuant MD software for processing.

## Qualified Personnel

Only qualified SCIEX personnel shall install, inspect, and service the equipment. After installing the system, the Field Service Employee (FSE) uses the *Customer Familiarization Checklist* to orient the customer on system operation, cleaning, and basic maintenance. SCIEX might not cover the damage to a system under warranty if it is serviced by personnel not authorized by SCIEX.

Only personnel qualified by the manufacturer shall maintain the equipment. A laboratory designate can be familiarized with the Qualified Maintenance Person (QMP) procedures during the installation. A QMP is a person who is suitably aware of the electrical and chemical risks associated with servicing laboratory equipment.

# Quality Control Guidelines

It is expected that a clinical laboratory using a Citrine system adheres to procedures that cover issues such as, but not limited to, operator training, assay development and validation, and external audits of laboratory assay performance.

## Laboratory Processes

Verify methods before their implementation in clinical use for reporting results<sup>2,5</sup>. As well, establish standard operating procedures (SOPs) for analytical methods to make sure that pre-analytical, analytical, and post-analytical processes do not vary from their intended use<sup>3</sup>. For example, at a minimum, an SOP must be in place for:<sup>3,4</sup>

- Sample collection methods
- Sample preparation methods
- Liquid chromatography settings and initial conditions
- Mass spectrometer setup and calibration initial conditions
- Liquid chromatography and mass spectrometry maintenance
- Mass spectrometer acquisition methods
- Sample batch list preparation methods
- Data analysis methods
- Data review
- Release protocols once the data is analyzed

We recommend that users manually review all of the integration results to make sure of the quality of the raw data and the correctness of peak integrations performed by the Analyst MD software. The review of chromatographic peaks must be performed by a trained professional. If the Analyst MD software has not correctly integrated a chromatographic peak, which can be caused by the presence of closely eluting peaks, split peaks, noisy data, or high background signal, then the peak integrations should be corrected according to SOPs for analytical methods established by the laboratory.

## Quality Control Samples

Quality control (QC) samples provide feedback on the performance of the analytical method and assess the integrity and validity of the unknown samples analyzed within the run<sup>2</sup>. QC sample data should be monitored day-to-day. Follow appropriate guidance for the inclusion of QC samples in an analytical run.

QC samples can be obtained from a commercial source with the appropriate documentation or from a well characterized pool of patient samples<sup>2</sup>. We recommend that at least two QC samples be included per analytical run of patient samples<sup>1,2</sup>. Use the data review process to confirm that the QC samples are within pre-defined limits for the analytical method. Data acquired in an analytical run where the QCs are outside the pre-defined limits might be invalid<sup>2</sup>. Refer to the laboratory SOP.

## Internal Standards

Internal standards are analytes that are structurally similar analogs or stable, isotopically labeled analytes that are added to all of the sample types, (that is, standards, blanks, QCs, and unknowns) at known and constant concentrations to facilitate quantitation<sup>3</sup>. The internal standard signal intensity can be monitored across an analytical run to confirm the integrity of the analytical method and validity of an individual sample.

SOPs should include criteria for the evaluation of internal standard analytes and QC samples. Samples that fall outside the criteria might indicate issues with the performance of the method, system, or sample<sup>2,5</sup>. Use internal standards (isotopically labeled analytes) for measurements involving critical decisions. Identifying these issues early allows the laboratory to investigate, correct, and potentially repeat the analysis, if necessary<sup>2,5</sup>.

## References

1. Guidance for Industry: Bioanalytical Method Validation; U.S. Department of Health and Human Services Food and Drug Administration; Center for Drug Evaluation (CDER) Centre for Veterinary Medicine (CVM), May 2001 BP
2. CLSI Standard C50–A–Vol. 27, No. 24—Mass Spectrometry in the Clinical Laboratory: General Principles and Guidance: Approved Guideline
3. ISO 17025: 2005—General requirements for the competence of testing and calibration laboratories
4. ISO 15189:2012 Medical laboratories-Requirements for quality and competence
5. CLSI document C62-A: Liquid Chromatography-Mass Spectrometry Methods; Approved Guideline Volume 34 Number 16, October 2014

## Equipment Use and Modification



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**WARNING! Personal Injury Hazard. Contact the SCiEX representative if product installation, adjustment, or relocation is required.**

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## Use and Function

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**WARNING! Electrical Shock Hazard. Do not remove the covers. Removing the covers might cause injury or malfunctioning of the system. The covers need not be removed for routine maintenance, inspection, or adjustment. Contact a SCIEX Field Service Employee (FSE) for repairs that require the covers to be removed.**

---



**WARNING! Personal Injury Hazard. Use SCIEX-recommended parts only. Use of parts not recommended by SCIEX or use of parts for any purpose other than their intended purpose can put the user at risk of harm or negatively impact system performance.**

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**WARNING! Lifting Hazard. Use a mechanical lifting device to lift and move the mass spectrometer. If the mass spectrometer must be moved manually, then at least six people are required to move it safely. Follow established safe lifting procedures. We recommend the use of a professional moving service. For the weights of system components, refer to the document: *Site Planning Guide*.**

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**WARNING! Crushing Hazard. Wear protective footwear when moving heavy objects.**

---

Use the system indoors in a laboratory that complies with the environmental conditions recommended in the mass spectrometer document: *Site Planning Guide*.

If the system is used in an environment or in a manner not prescribed by the manufacturer, then the performance and protection provided by the equipment might be impaired.

Unauthorized modification or operation of the system might cause personal injury and equipment damage, and might void the warranty. Erroneous data might be generated if the system is operated outside the recommended environmental conditions or with unauthorized modifications. Contact an FSE for information on servicing the system.

Warranty terms are available from the [sciex.com/warranty](https://sciex.com/warranty) page. The Citrine system has an expected lifetime of 7 years from manufacture date. This lifetime might be extended when scheduled maintenance procedures are followed.

## Laboratory Conditions

### Safe Environmental Conditions

The system is designed to operate safely under these conditions:

- Indoors

- Altitude: Up to 2,000 m (6,560 ft) above sea level
- Ambient temperature: 5 °C (41 °F) to 40 °C (104 °F)
- Relative humidity: 20% to 80%, non-condensing
- Mains supply voltage fluctuations:  $\pm$  10% of the nominal voltage
- Transient overvoltages: Up to the levels of Overvoltage Category II
- Temporary overvoltages on the mains supply
- Pollution Degree 2

## Performance Specifications

The system is designed to meet specifications under these conditions:

- An ambient temperature of 15 °C to 30 °C (59 °F to 86 °F)

Over time, the temperature must remain within a range of 4 °C (7.2 °F), with the rate of the change in temperature not exceeding 2 °C (3.6 °F) per hour. Ambient temperature fluctuations exceeding the limits might result in mass shifts in spectra.

- Relative humidity from 20% to 80%, non-condensing

# Installation Procedures and Special Requirements

# 3

A SCIEX Field Service Employee (FSE) installs and configures the system.

This section includes procedures to connect and configure the system hardware and software. Refer to these procedures if the system must be moved, replumbed, or reconfigured.



---

**WARNING! Electrical Shock Hazard. Make sure that the system can be disconnected from the mains supply outlet in an emergency. Do not block the mains supply outlet.**

---

For information on installing the Analyst MD software, refer to the document: *Software Installation Guide*.

## Adjust the Integrated Syringe Pump Position



---

**WARNING! Puncture Hazard. Take care when handling the syringe. The tip of the syringe is extremely sharp.**

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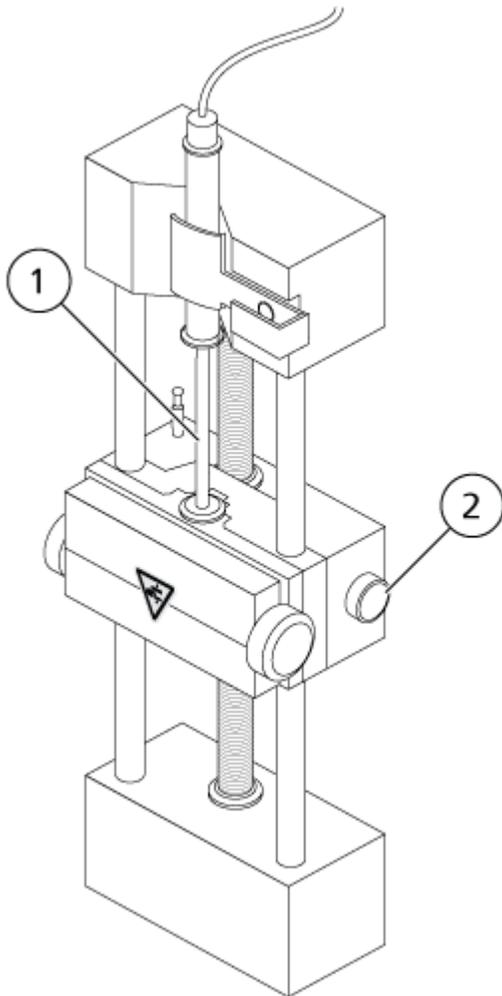
**WARNING! Puncture Hazard. Make sure that the syringe is seated properly in the syringe pump and that the automatic syringe pump stop is adjusted properly to avoid damaging or breaking the glass syringe. If the syringe breaks, follow established safety procedures for sharps disposal.**

---

For the location of the syringe pump on the mass spectrometer, refer to the figure: [Mass Spectrometer Overview](#).

1. Open the syringe cover.
2. Press the Release button on the right side of the syringe pump to lower the base and then insert the syringe.

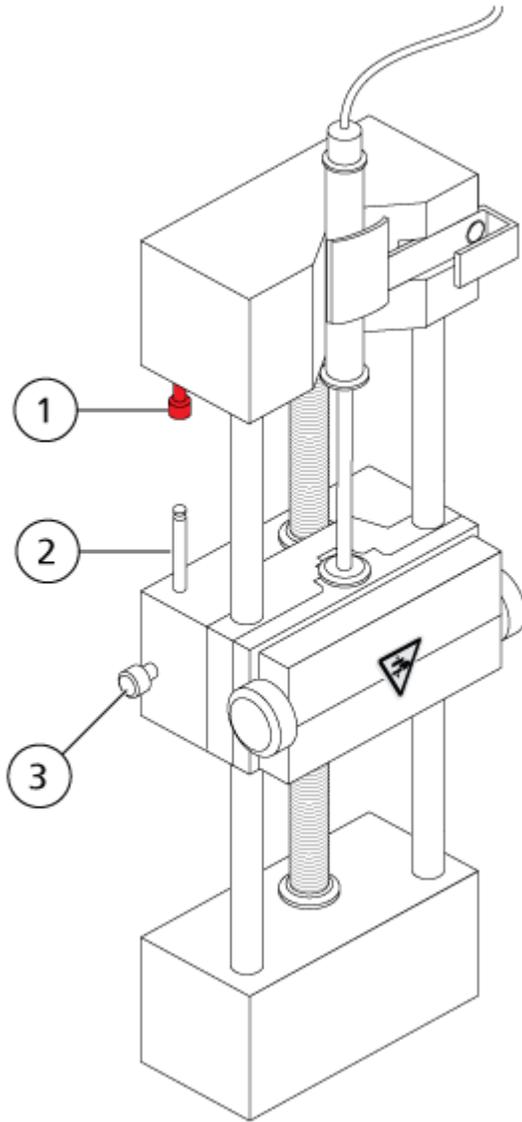
Figure 3-1 Lowering the Syringe



Item	Description
1	Syringe plunger
2	Release button. Press to raise or lower the base.

3. Make sure that the end of the syringe is flush with the base and that the shaft of the syringe rests in the cutout.
4. Adjust the post so that it triggers the automatic syringe stop before the syringe plunger hits the bottom of the glass syringe.

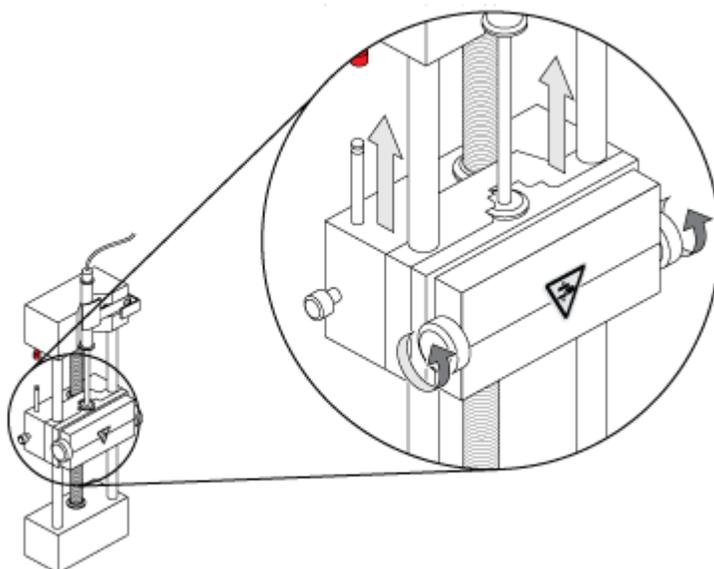
**Figure 3-2 Automatic Syringe Stop**



Item	Description
1	Automatic syringe stop. After the post hits the automatic syringe stop, the syringe pump stops.
2	Post. Adjust the height to prevent the syringe plunger from hitting the syringe during sample infusion.
3	Post lock screw. Tighten the screw after the height of the post is adjusted.

5. Turn the syringe pump screws to secure the syringe.

Figure 3-3 Syringe Pump Screws



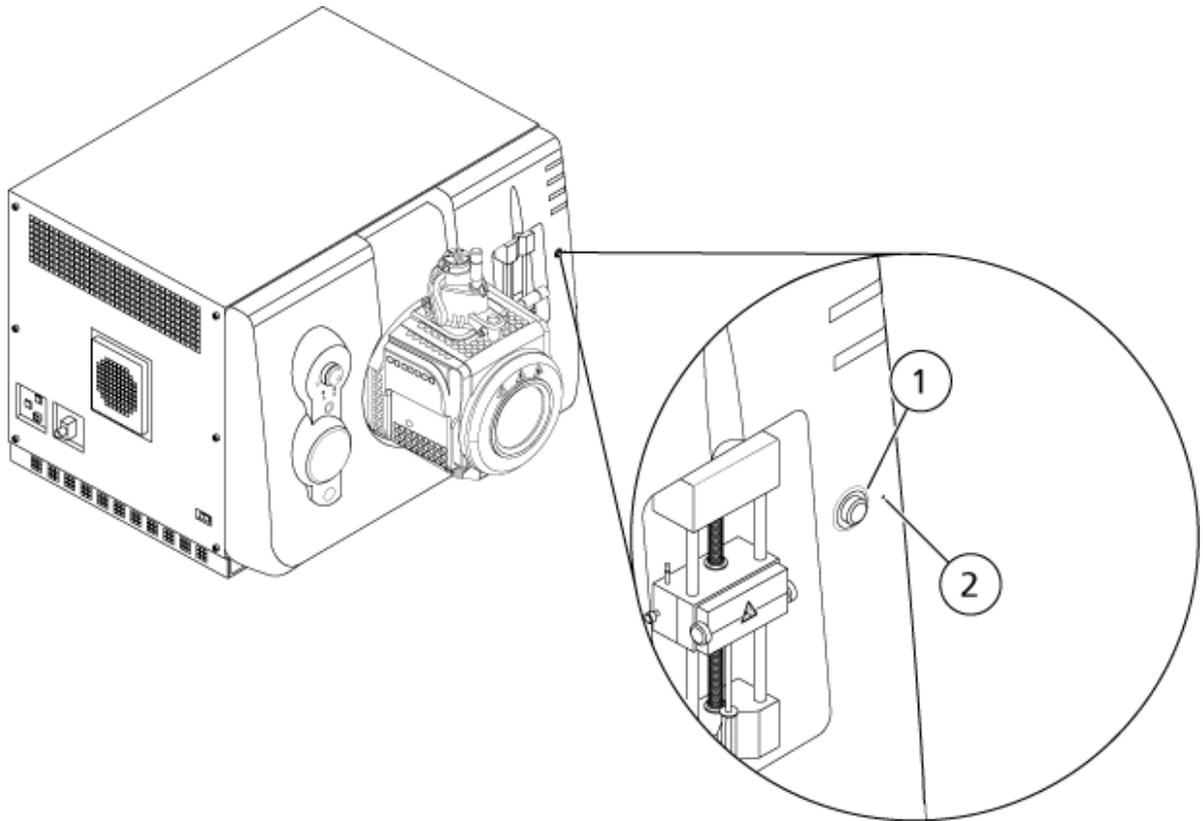
6. Make sure that the mass spectrometer and integrated syringe pump are activated in the software.

---

**Note:** For subsequent manual use, after the mass spectrometer is in Ready state, start the flow by pressing the button on the mass spectrometer that is to the right of the syringe. The LED beside the button flashes when the syringe pump is in use. The syringe pump flow can also be controlled automatically by the Analyst MD software.

---

**Figure 3-4 Syringe Pump LED**



Item	Description
1	Syringe pump on and off button
2	Syringe pump status LED

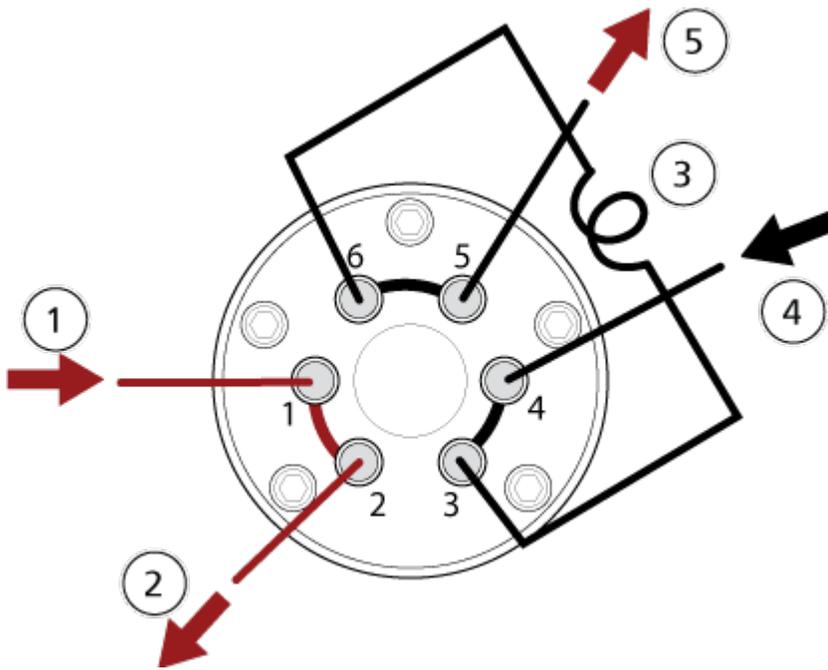
7. In the Analyst MD software, on the Navigation bar, double-click **Manual Tuning**.
8. Click **Start Syringe**.
9. To stop the syringe pump, click **Stop Syringe**.

## Plumb the Diverter Valve

The integrated diverter valve, which is located next to the ion source, can be plumbed in injector mode or diverter mode. To configure the valve, open the Configuration tab and then make sure that the **Use integrated injector/diverter valve** check box is selected. Refer to the section: [Add Devices to a Hardware Profile](#).



**Figure 3-6 Diverter Valve—Injector Mode Position B**



Item	Description
1	Sample in
2	Waste out
3	Sample loop (ports 3 and 6)
4	Mobile phase in
5	To column, or to the mass spectrometer, if a column is not installed

### Plumb the Diverter Valve in Diverter Mode

When the valve is in Position A, the sample flow goes to the mass spectrometer. When the valve switches to Position B, the flow goes to waste.

Plumb the valve for Diverter mode.

Figure 3-7 Diverter Valve—Diverter Mode Position A

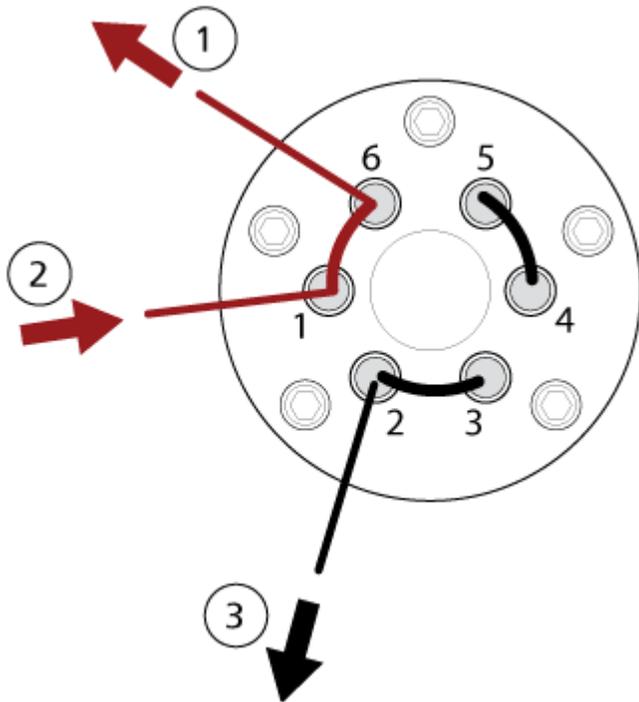
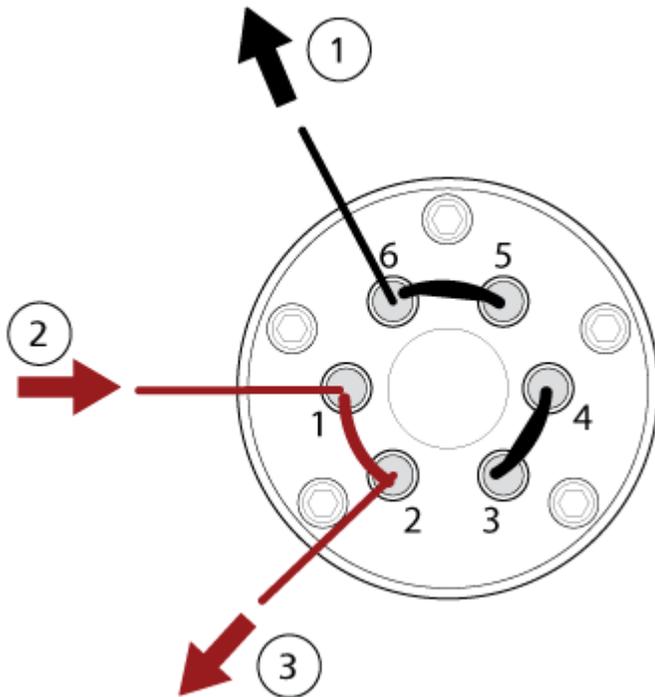


Figure 3-8 Diverter Valve—Diverter Mode Position B



## Installation Procedures and Special Requirements

---

Item	Description
1	To mass spectrometer
2	From column
3	Waste out

## Ion Source Installation



**WARNING! Electrical Shock Hazard.** Install the ion source on the mass spectrometer as the last step in this procedure. High voltage is present when the ion source is installed.

---

**CAUTION: Potential System Damage.** Do not lift or carry the ion source with one hand. The ion source is designed to be lifted or carried using two hands, one on each side of the ion source.

---

The ion source is connected to the vacuum interface and is held in position by two source latches. The interior of the ion source is visible through the windows on the side and front of the ion source.

When the ion source is installed, the software recognizes the ion source and shows the ion source identification.

In this guide, the software that controls the mass spectrometer is referred to as the control software.

### Required Materials

- Ion source
- Red PEEK tubing (0.005 inch bore)

## Prepare for Installation



**WARNING! Puncture Hazard.** Be careful when handling the electrode. The tip of the electrode is extremely sharp.

---

**Tip!** Do not discard the empty package. Use it to store the ion source when it is not in use.

---

Adjust the electrode adjustment nut on the probe to move the electrode tip inside the electrode tube. Refer to the figures: [Figure 4-4](#) and [Figure 4-5](#).

---

For optimum stability and performance, the electrode tip should extend between 0.5 mm and 1.0 mm from the end of the probe. Refer to the section: [Optimize the TurbolonSpray Probe Position](#) or [Optimize the APCI Probe Position](#).

### Install the Probe



**WARNING! Electrical Shock Hazard.** Make sure that the ion source is completely disconnected from the mass spectrometer before proceeding.

---



**WARNING! Puncture Hazard.** Be careful when handling the electrode. The tip of the electrode is extremely sharp.

---

**CAUTION: Potential System Damage.** Do not let the protruding electrode tip or the corona discharge needle touch any part of the ion source housing, to avoid damaging the probe.

---

**CAUTION: Potential System Damage.** Make sure that the corona discharge needle tip is turned away from the aperture if the TurbolonSpray probe is in use.

---

Prerequisite Procedures
<ul style="list-style-type: none"><li>• <a href="#">Remove the Ion Source</a>.</li></ul>



The probe is not pre-installed in the ion source. Always remove the ion source from the mass spectrometer before exchanging probes.

---

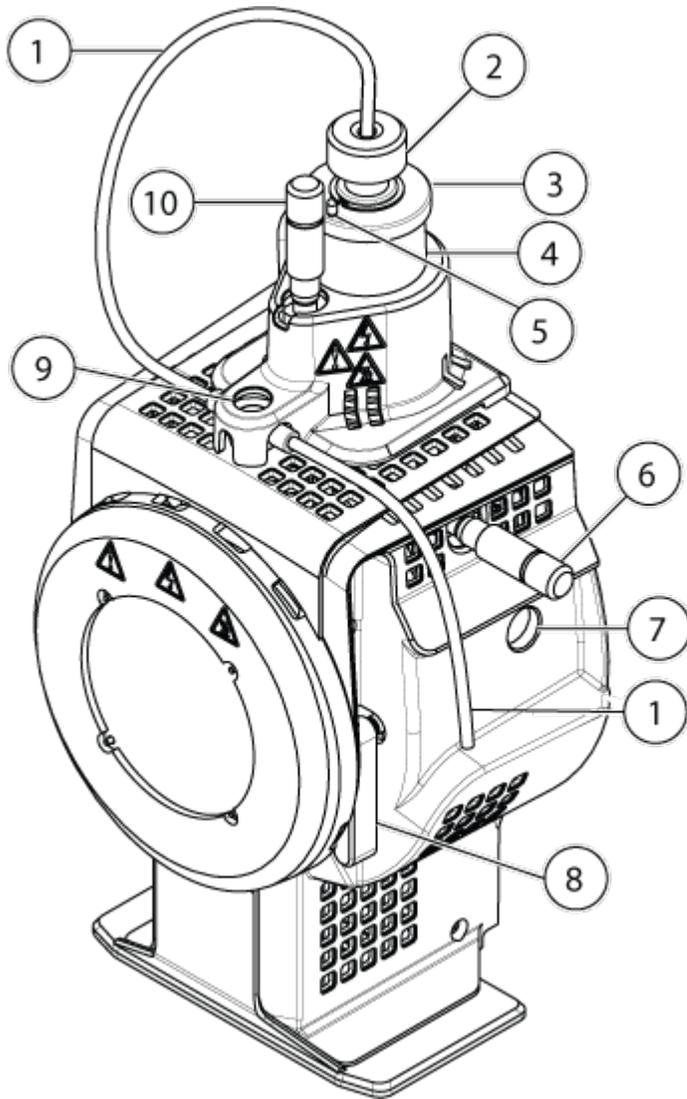
**Note:** If the probe is not properly installed in the ion source, then the high-voltage power for the mass spectrometer and source exhaust system is turned off.

---

## Installation Procedures and Special Requirements

---

**Figure 3-9 Ion Source Components**



Item	Description
1	Sample tubing
2	Electrode adjustment nut
3	Retaining ring
4	Probe tower
5	Corona discharge needle adjustment screw

## Installation Procedures and Special Requirements

---

Item	Description
6	Micrometer used to position the probe on the horizontal axis for ion source sensitivity adjustments
7	Window port
8	One of two source latches that secure the ion source to the mass spectrometer
9	Grounding union, located under the ion source cover
10	Micrometer used to position the probe on the vertical axis for ion source sensitivity adjustments

1. Make sure that the corona discharge needle tip is pointed away from the curtain plate aperture. Refer to the section: [Adjust the Position of the Corona Discharge Needle](#).
2. Insert the probe in the tower. Align the hole on the probe with the corona discharge needle adjustment screw at the top of the ion source. Refer to the section: [Ion Source Components](#).
3. Gently push the probe down until the contacts engage with those in the tower.
4. Turn the retaining ring over the probe, push it down to engage the threads on the probe with the threads on the tower, and then tighten it until it is finger-tight.
5. For the APCI probe only, make sure that the corona discharge needle tip is pointed toward the curtain plate aperture. Refer to the section: [Adjust the Position of the Corona Discharge Needle](#).

### Connect the Ion Source Tubing



---

**WARNING! Electrical Shock Hazard. Do not bypass the grounding union connection. The grounding union provides grounding between the mass spectrometer and the sample introduction device.**

---



---

**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Make sure that the sample tubing nut is tightened properly before operating this equipment, to prevent leakage.**

---

Refer to the section: [Ion Source Components](#).

1. Insert a 30 cm piece of red PEEK tubing in the sample tubing nut.
2. Install the sample tubing nut in the port at the top of the probe, and then tighten the sample tubing nut until it is finger-tight.
3. Connect the other end of the tubing to the grounding union on the ion source.

## Install the Ion Source on the Mass Spectrometer



**WARNING! Electrical Shock Hazard.** Install the probe in the ion source before installing the ion source on the mass spectrometer.



**WARNING! Pinching Hazard.** When installing the ion source, be careful not to pinch fingers between the ion source and the vacuum interface.

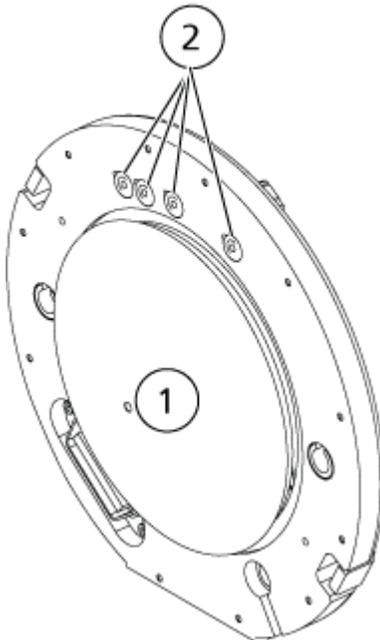
**CAUTION: Potential System Damage.** Do not let the protruding electrode tip or the corona discharge needle touch any part of the ion source housing, to avoid damaging the probe.

**Note:** If the probe is not properly installed in the ion source, then the high-voltage power for the mass spectrometer and source exhaust system is turned off.

### Prerequisites

- Make sure that all of the O-rings are present on the vacuum interface.

**Figure 3-10 O-rings on the Vacuum Interface**



Item	Description
1	Curtain plate
2	O-rings

## Installation Procedures and Special Requirements

---

1. Make sure that the ion source latches on either side of the ion source are pointing up in the 12 o'clock position. Refer to the section: [Ion Source Components](#).
2. Align the ion source with the vacuum interface, making sure that the guide pins on the ion source are aligned with the sockets in the vacuum interface.
3. Push the ion source gently against the vacuum interface and then rotate the ion source latches down to lock the ion source in place.  
The mass spectrometer recognizes the ion source and then shows the ion source identification in the control software.
4. Connect the red PEEK tubing from the sample supply device to the other side of the grounding union on the ion source.

The system is designed for analysis of small molecules in biological samples. Depending on the properties of the analytes or the complexity of the starting sample, sample preparation can include various types of extraction or filtration prior to extraction of analytes in solution. The sample is separated by a liquid chromatograph. The separated fractions are then introduced in a mass spectrometer for further separation based on the specific molecular mass of the compounds.

For information on the computer and software, refer to the document: *Software Installation Guide* for the software.

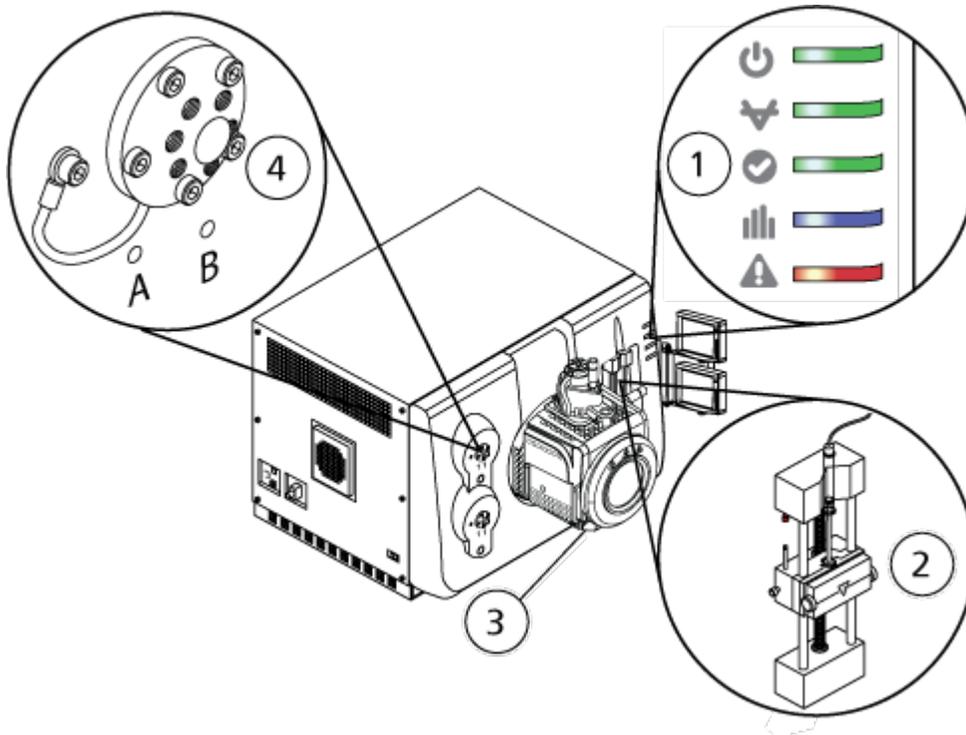
## System Overview

The Citrine system includes the following components:

- Citrine QTRAP or Citrine Triple Quad mass spectrometer with two roughing pumps and a source of compressed air and nitrogen
- A IonDrive Turbo V ion source that uses either the TurbolonSpray probe or the atmospheric pressure chemical ionization (APCI) probe.
- SCIEX-supplied computer and monitor with the Analyst MD software for instrument optimization, acquisition method development, data acquisition, and processing. For computer specifications and requirements, refer to the document: *Software Installation Guide* for the Analyst MD software.

## Mass Spectrometer Overview

Figure 4-1 Front View



Item	Description	Primary Materials	Refer To
1	Panel symbols	Plastic	<a href="#">Panel Symbols.</a>
2	Syringe pump	Paint over steel (body), stainless steel (rails), brass, Cu, Sn, Pb (bearings)	<a href="#">Adjust the Integrated Syringe Pump Position</a>
3	Ion source	N/A	<a href="#">Ion Source Overview</a>
4	Diverter valve	Stainless steel	<a href="#">Plumb the Diverter Valve.</a>

### Panel Symbols

The following table describes the mass spectrometer status LEDs.

## Principles of Operation

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**Table 4-1 Panel Symbols**

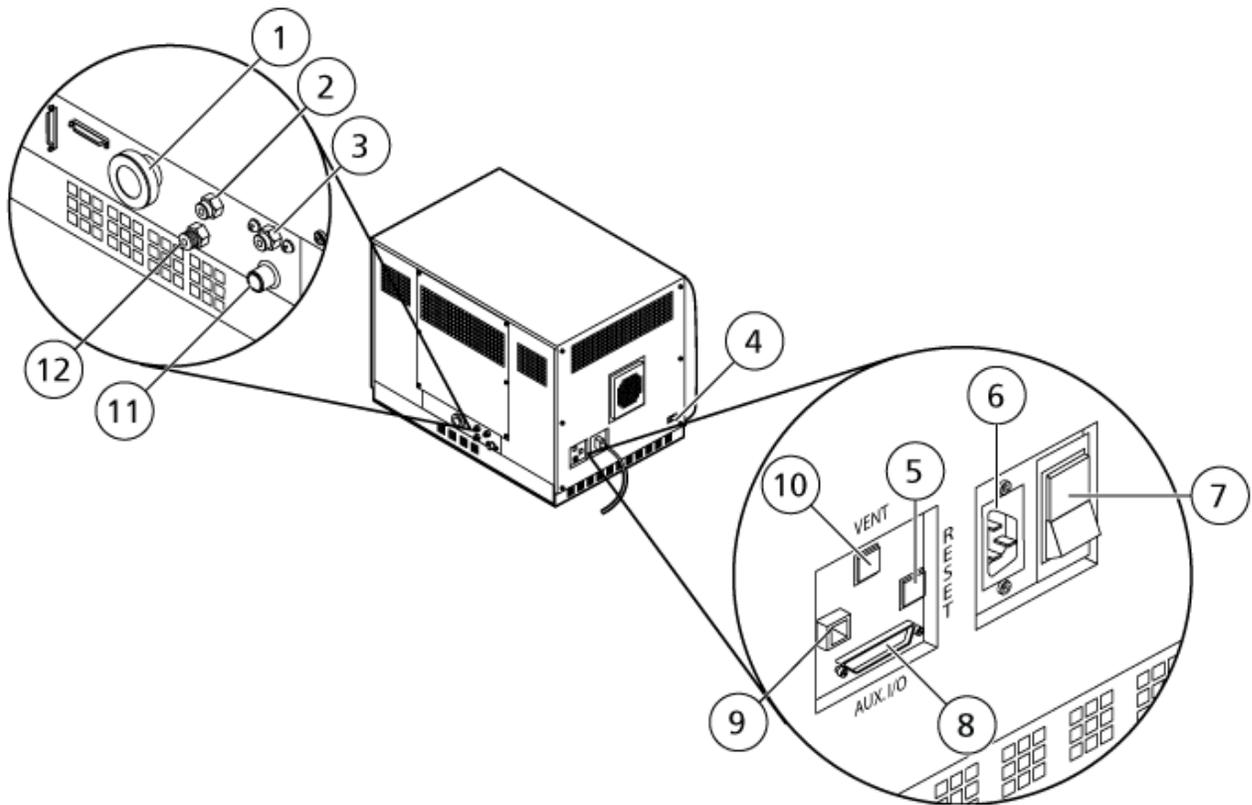
LED	Color	Name	Description
	Green	Power	Illuminated when the system is powered up.
	Green	Vacuum	Illuminated when the operating vacuum level has been achieved. Flashing if the vacuum is not at the correct level, that is, during pump down and venting.
	Green	Ready	Illuminated when the system is in the Ready state. The system must be in the Ready state to operate.
	Blue	Scanning	Flashing when the system is acquiring data.
	Red	Fault	Illuminated when the system encounters a system fault.

After the system is turned on, all of the LEDs illuminate. The power LED remains illuminated. The other LEDs flash for two seconds and then turn off. The vacuum LED starts flashing. After the operating vacuum level is achieved, this LED remains illuminated.

## Connections

The following figure shows the location of the mass spectrometer connections, including the locations of the **RESET** and **VENT** buttons and the mass spectrometer convenience switch.

Figure 4-2 Back and Side Views



Item	Description	Primary Materials	For More Information
1	Roughing pump vacuum connection	Aluminum (hose fitting), zinc-plated steel (hose clamp)	Contact an FSE. This connection is not user serviceable.
2	Air supply (Gas 1/Gas 2)	Plastic	Refer to the <i>Site Planning Guide</i> .
3	Source exhaust supply	Plastic	Refer to the document: <i>Site Planning Guide</i> .
4	Source communication connection	Aluminum	Contact an FSE.
5	<b>RESET</b> button	Plastic	Refer to the section: <a href="#">Reset the Mass Spectrometer</a> .

## Principles of Operation

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Item	Description	Primary Materials	For More Information
6	Mains supply connection	Aluminum/plastic	Refer to the section: <a href="#">Start Up the System</a> or <a href="#">Shut Down and Vent the System</a> .
7	Mass spectrometer convenience switch <ul style="list-style-type: none"><li>• Up = On</li><li>• Down = Off</li></ul>	Plastic	Refer to the section: <a href="#">Start Up the System</a> or <a href="#">Shut Down and Vent the System</a> .
8	Aux I/O connection	Sheet metal (zinc-plated)	Refer to the document: <i>Peripheral Devices Setup Guide</i> .
9	Ethernet connection, connects the mass spectrometer and the computer	Sheet metal (zinc-plated)	Contact an FSE.
10	<b>VENT</b> button	Plastic	Refer to the section: <a href="#">Start Up the System</a> or <a href="#">Shut Down and Vent the System</a> .
11	Source exhaust waste, to the source exhaust drain bottle	Stainless steel	Refer to the document: <i>Site Planning Guide</i> .
12	Nitrogen gas supply (Curtain Gas™ supply, CAD gas)	Stainless steel	Refer to the document: <i>Site Planning Guide</i> .

## Ion Source Overview

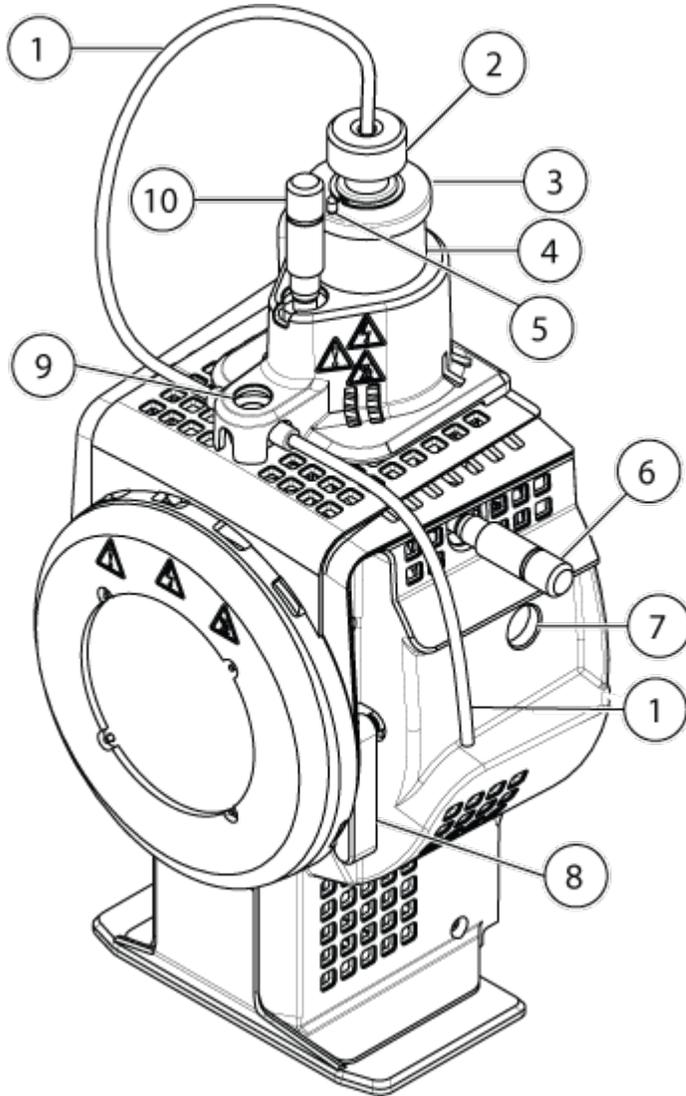
The IonDrive Turbo V ion source can be used for either electrospray ionization (ESI) or atmospheric pressure chemical ionization (APCI).

The TurbolonSpray probe is used for ESI mode operation. The APCI probe is used for APCI mode operation.

The IonDrive Turbo V ion source was designed to provide more heat and better desolvation and ionization, especially at high-flow rates. It features increased heater diameter for improved ionization, a larger sweet spot, and less performance variability.

## Ion Source Components

Figure 4-3 Ion Source Components



Item	Description
1	Sample tubing
2	Electrode adjustment nut
3	Retaining ring
4	Probe tower
5	Corona discharge needle adjustment screw

## Principles of Operation

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Item	Description
6	Micrometer used to position the probe on the horizontal axis for ion source sensitivity adjustments
7	Window port
8	One of two source latches that secure the ion source to the mass spectrometer
9	Grounding union. It is not visible below the ion source cover.
10	Micrometer used to position the probe on the vertical axis for ion source sensitivity adjustments

## Probes

The TurbolonSpray and APCI probes provide a range of capability for testing samples. Select the probe and method most suitable for the compounds in the sample.

**Table 4-2 Ion Source Specifications**

Specification	TurbolonSpray Probe	APCI Probe
Temperature range	From ambient temperature to 750 °C, depending on liquid flow	From ambient temperature to 750 °C, depending on liquid flow
Liquid flow inlet	5 µL/min to 3,000 µL/min	200 µL/min to 3,000 µL/min
Ion source gas 1 / ion source gas 2	Refer to the mass spectrometer document: <i>Site Planning Guide</i>	

The software for the mass spectrometer identifies the installed probe and enables the corresponding user controls. All of the data acquired using the ion source is identified with an abbreviation representing the probe used to acquire the data (TIS for the TurbolonSpray probe and HN for the APCI probe).

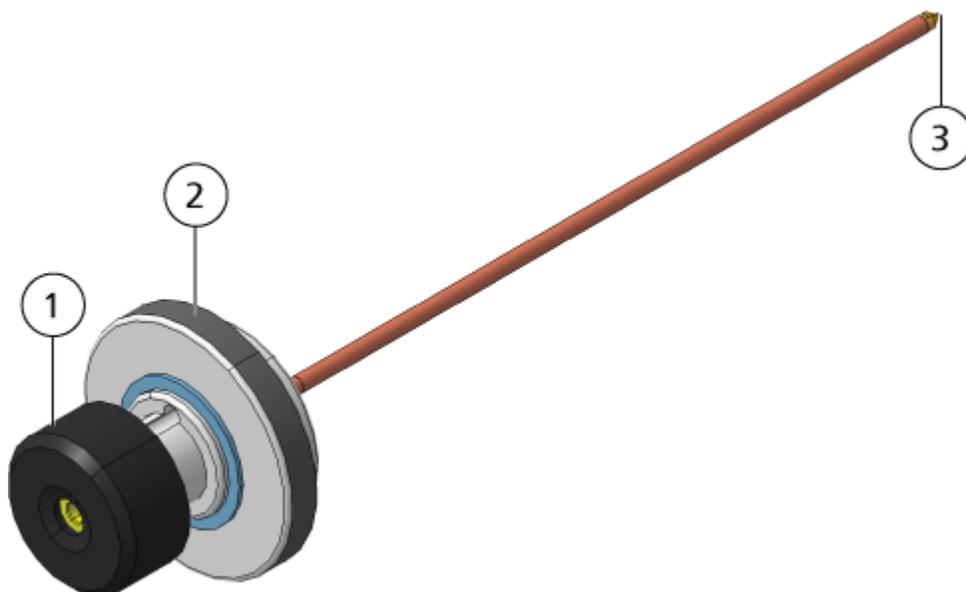
### TurbolonSpray Probe

The TurbolonSpray probe consists of 300 µm outer diameter (o.d.) (0.012 inch) stainless steel tubing. It is located centrally with the two turbo heaters placed at a 45 degree angle to each side. Samples introduced through the TurbolonSpray probe are ionized within the tubing, by the application of high voltage (**IonSpray Voltage**). Then they are nebulized by a jet of hot, dry, Zero-grade air from the turbo heaters, creating a mist of small, highly-charged droplets. The combination of the ion source effluent and the heated dry gas from the turbo sprayer is projected at a 90 degree angle to the ion path. Refer to the section: [Theory of Operation — Ion Source](#).



**WARNING! Puncture Hazard.** Be careful when handling the electrode. The tips of the electrodes are extremely sharp.

Figure 4-4 Parts of the TurbolonSpray Probe



Item	Description
1	Electrode adjustment nut (black collar) that adjusts the extension of the electrode tip
2	Retaining ring that fastens the probe to the probe tower on the ion source housing
3	Electrode tip through which samples are sprayed in the sample inlet area of the ion source

#### APCI Probe

The APCI probe consists of 100 µm inner diameter (i.d.) (0.004 inch) stainless steel tubing surrounded by a flow of nebulizer gas (Gas 1). The liquid sample stream is pumped through the sprayer, where it is nebulized in a ceramic tube containing a heater. The inner wall of the ceramic tube can be maintained at a temperature range of 100 °C to 750 °C and is monitored by the sensor embedded in the heater.

A high-velocity jet of nebulizer gas flows around the electrode tip to disperse the sample as a mist of fine particles. It moves through the ceramic vaporization heater to the reaction region of the ion source and then past the corona discharge needle where the sample molecules are ionized as they pass through the ion source housing. Refer to the section: [Theory of Operation — Ion Source](#).

## Principles of Operation

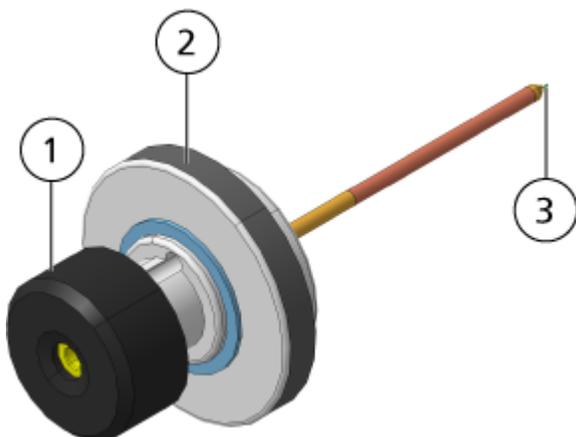
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**WARNING! Puncture Hazard. Be careful when handling the electrode. The tips of the electrodes are extremely sharp.**

---

Figure 4-5 Parts of the APCI Probe



Item	Description
1	Electrode adjustment nut (black collar) that adjusts the extension of the electrode tip
2	Retaining ring that secures the probe in the probe tower
3	Electrode tip through which samples are sprayed in the sample inlet area of the ion source

## Gas and Electrical Connections

Gas and low- and high-voltage electrical connections are provided on the front plate of the vacuum interface and they connect internally through the ion source housing. When the ion source is installed on the mass spectrometer, all of the electrical and gas connections are complete.

## Ion Source Sense Circuit

An ion source sense circuit disables the high-voltage power supply for the mass spectrometer and the source exhaust system under these conditions:

- The ion source is not installed or is improperly installed.
- A probe is not installed.
- The mass spectrometer senses a gas fault.
- A turbo heater has failed.

- The ion source has overheated.

## Source Exhaust System



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** Make sure that the source exhaust system is connected and functioning, to safely remove sample vapor exhaust from the laboratory environment. Emissions from the equipment must be exhausted in the general building exhaust and not allowed to exhaust in the workspace of the laboratory. For requirements for the source exhaust system, refer to the document: *Site Planning Guide*.



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** Vent the source exhaust system to either a dedicated laboratory fume hood or an external ventilation system to prevent hazardous vapors from being released in the laboratory environment.



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** If an LC system is used with the mass spectrometer, and if the source exhaust system is not functioning properly, then shut down the LC system until functionality of the source exhaust system has been restored.



**WARNING! Fire Hazard.** Do not direct more than 3 mL/min of flammable solvent in the ion source. Although the LC components can provide a flow rate of up to 5 mL/min, exceeding the maximum flow rate can cause solvent to accumulate in the ion source. Do not use the ion source if the source exhaust system is not enabled and functioning when the ion source and the probe are properly installed.

**Note:** Make sure that all of the exhaust tubing is securely connected to reduce the chance of equipment exhaust entering the room.

An ion source produces both sample and solvent vapors. These vapors are a potential hazard to the laboratory environment. The source exhaust system is designed to safely remove and allow for the appropriate handling of the sample and solvent vapors. When the ion source is installed, the mass spectrometer does not operate unless the source exhaust system is operating.

A vacuum switch installed in the source exhaust sense circuit measures the vacuum in the source. If the vacuum in the source rises above the set point while the probe is installed, then the system enters an exhaust fault, that is, Not Ready, state.

An active exhaust system removes ion source exhaust, including gases, solvent, and sample vapor, through a drain port, without introducing chemical noise. The drain port connects through a drain chamber and a source exhaust pump to a drain bottle, and from there to a customer-

## Principles of Operation

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supplied exhaust ventilation system. For information about the ventilation requirements for the source exhaust system, refer to the document: *Site Planning Guide*.

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**Note:** Examine the source exhaust system periodically to make sure that the exhaust tubing is intact and that exhaust is not leaking in the room.

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## Analyst MD Software Overview

The Analyst MD software works with the mass spectrometer and the liquid chromatography (LC) system and the associated firmware to control the system and data acquisition. During system operation, the acquired data is sent to the Analyst MD software, where it can be shown as full mass spectra, intensity of single or multiple ions over time, or total ion count over time.

### Different Data Views

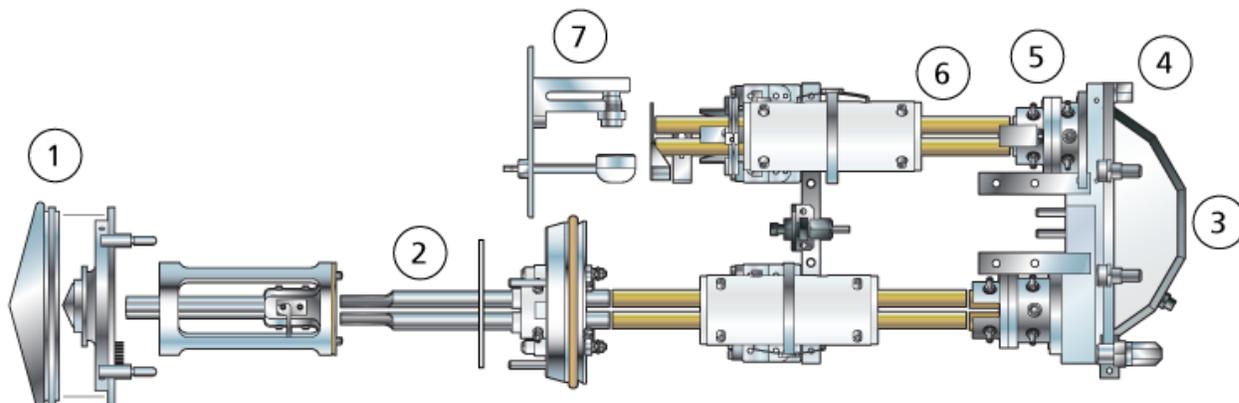
#### MS Parameters

The working parameters are the set of mass spectrometer (MS) parameters currently being used.

The compound parameters and source and gas parameters are stored with the method. The resolution and detector parameters are mass spectrometer-dependent and are stored as instrument data. If the Tune and Calibrate mode is used to create a method, then the working parameters can be optimized for best instrument performance. Alternatively, ramp each parameter, one at a time, while cycling the experiment.

- Source and gas parameters: These parameters can change depending on the ion source used.
- Compound parameters: These parameters consist mostly of voltages in the ion path. Optimal values for compound-dependent parameters vary depending on the compound being analyzed.
- Resolution parameters: These parameters affect the resolution and calibration.
- Detector parameters: These parameters affect the detector.

Figure 4-6 Ion Optics Path and Parameters



Location	Parameter	Parameter Type	Use	Scan Type
1	IonSpray Voltage (IS)	Source and gas	The IS parameter controls the voltage applied to the electrode in the ESI probe that ionizes the sample in the ion source. The parameter depends on the polarity and it affects the spray stability and the sensitivity. The parameter can be compound-dependent and must be optimized for each compound.	All
1	Nebulizer Current (NC)	Source and gas	The NC parameter controls the current applied to the corona discharge needle in the APCI probe. The discharge ionizes solvent molecules, which in turn ionize the sample molecules.	All
1	Ion Source Gas 1 (GS1)	Source and gas	The GS1 parameter controls the nebulizer gas for both the ESI and APCI probes.	All
1	Ion Source Gas 2 (GS2)	Source and gas	The GS2 parameter controls the heater gas for the ESI probe.	All
1	Temperature (TEM)	Source and gas	The TEM parameter controls the temperature of the heater gas for the ESI and APCI probes.	All

## Principles of Operation

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Location	Parameter	Parameter Type	Use	Scan Type
1	Curtain Gas (CUR)	Source and gas	The CUR parameter controls the flow of the gas for the Curtain Gas interface. The Curtain Gas interface is located between the curtain plate and the orifice. It prevents the contamination of the ion optics.	All
1	Declustering Potential (DP)	Compound	<p>The DP parameter controls the voltage on the orifice, which controls the ability to decluster ions between the orifice and the IonDrive QJet ion guide. It is used to minimize the solvent clusters that might remain on the sample ions after they enter the vacuum chamber, and, if required, to fragment ions. The higher the voltage, the higher the energy imparted to the ions. If the DP parameter is too high, then unwanted fragmentation might occur.</p> <p>Use the preset value and optimize for the compound.</p>	All
2	Entrance Potential (EP)	Compound	<p>The EP parameter controls the potential difference between the voltage on Q0 and ground. The entrance potential guides and focuses the ions through the high-pressure in the Q0 region.</p> <p>Use the preset value.</p>	All

Location	Parameter	Parameter Type	Use	Scan Type
2	Q0 Trapping	Compound	<p>The Q0 trapping parameter controls the storage of ions in the Q0 region. It is used to increase sensitivity and duty cycle by trapping ions in the Q0 region while ions are being mass-selectively ejected from the linear ion trap. Use a fixed fill time with this parameter.</p> <p>Either select or clear the feature as required by the experiment.</p> <p>We recommend using a fixed fill time of 20 ms or greater.</p>	EMS, EPI, ER, and MS/MS/MS
3	CAD Gas	Source and gas	<p>The CAD Gas parameter controls the pressure of the CAD gas in the collision cell during Q3, MS/MS, and LIT scans. For Q3 scans, the collision gas helps to focus the ions as they pass through the Q2 collision cell. The preset value for the CAD Gas parameter is in Fixed mode. For MS/MS scan types, the CAD gas helps to fragment the precursor ions. When the precursor ions collide with the collision gas, they dissociate to form product ions.</p> <p>Use the preset value and optimize for the compound.</p>	Q3 MI, Q3 MS, MRM, Prec, NL, EMS, ER, EPI, and MS/MS/MS
3	Collision Energy (CE)	Compound	<p>The CE parameter controls the potential difference between the Q0 region and the Q2 collision cell. It is used only in MS/MS scan types. This parameter is the amount of energy that the precursor ions receive as they are accelerated in the Q2 collision cell, where they collide with gas molecules and fragment.</p> <p>Use the preset value and optimize for the compound.</p>	EPI, MS/MS/MS, MRM, MS2, Prec, NL, and LIT

## Principles of Operation

Location	Parameter	Parameter Type	Use	Scan Type
3	Collision Energy Spread (CES)	Compound	<p>The CES parameter, in conjunction with the CE parameter, determines which three discrete collision energies are applied to the precursor mass in an Enhanced Product Ion (EPI) or MS/MS/MS (MS3) scan when CES is used. When a collision energy spread value is entered, CES is automatically turned on.</p> <p>Use the preset value and optimize for the compound.</p>	EPI and MS/MS/MS
3	Collision Cell Exit Potential (CXP)	Compound	<p>The CXP parameter is only used in Q3 and MS/MS scan types. This parameter transmits the ions in the Q3 quadrupole.</p> <p>Use the preset value and optimize for the compound.</p>	Q3, MRM, MS2, Prec, NL
4	Q3 Entry Barrier	Compound	<p>The Q3 Entry Barrier parameter is used to transfer the ions from the Q2 collision cell in the linear ion trap.</p> <p>Use the preset value.</p>	EMS, EPI, ER, and MS/MS/MS
5	MS/MS/MS Fragmentation Excitation Time	Compound	<p>The MS/MS/MS Fragmentation Time parameter controls the amount of time that the excitation energy is applied. It is used in combination with the excitation energy to fragment the isolated second precursor ion.</p> <p>Use the preset value.</p>	MS/MS/MS

Location	Parameter	Parameter Type	Use	Scan Type
6	Fixed LIT Fill Time	Compound	<p>The Fixed LIT Fill Time parameter controls the amount of time that the LIT fills with ions.</p> <p>Use the preset value and adjust it to achieve the desired signal response based on sample concentration.</p>	EMS, EPI, ER, and MS/MS/MS
6	Dynamic Fill Time (DFT)	Compound	<p>The DFT parameter dynamically calculates the amount of time that ions are collected in the linear ion trap based on the incoming ion signal. When DFT is turned on, the signal is optimized to either increase sensitivity or minimize space-charging.</p> <p>Either select or clear the feature based on the experiment.</p> <p>In the <b>Tools &gt; Settings &gt; Method Options</b> dialog, the DFT settings are optimized for the default scan rate. These settings are also suitable for other LIT scan speeds.</p>	EMS, EPI, ER, and MS/MS/MS
7	CEM	Detector	The CEM parameter controls the voltage applied to the detector. The voltage controls the detector response.	All

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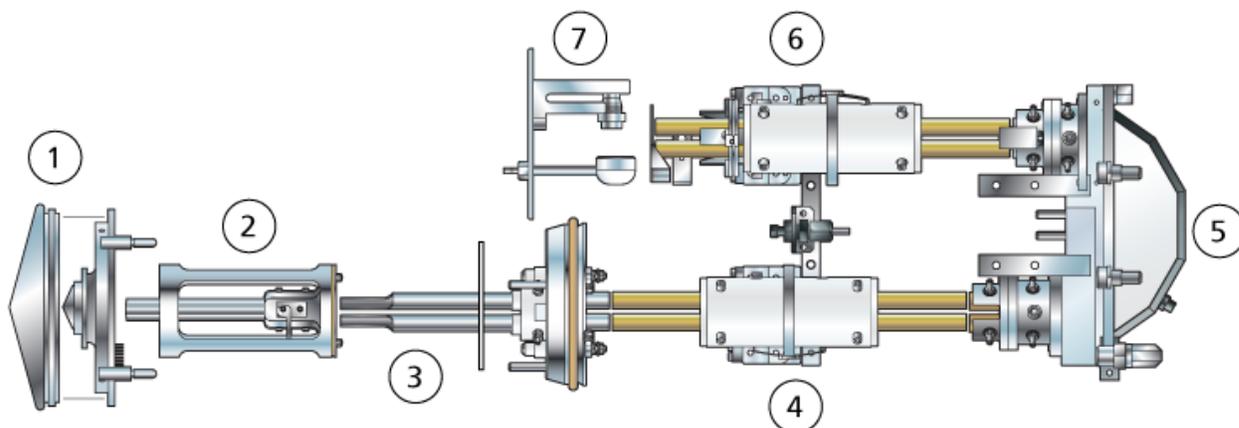
Mass spectrometry measures the mass-to-charge ratio of ions to identify unknown compounds, to quantify known compounds, and to provide information about the structural and chemical properties of molecules.

The mass spectrometer has a series of quadrupole filters that transmit ions according to their mass-to-charge ratio ( $m/z$ ). The first quadrupole in this series is the IonDrive QJet ion guide, located between the orifice plate and the Q0 region. The IonDrive QJet ion guide does not filter ions, but focuses them before they enter the Q0 region. The IonDrive QJet ion guide helps to focus ions into the Q0 region. In the Q0 region, the ions are again focused before they enter the Q1 quadrupole.

## Principles of Operation

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**Figure 4-7 Ion Path**



Item	Description
1	Curtain plate and orifice plate
2	IonDrive QJet ion guide
3	Q0 region
4	Q1 quadrupole
5	Q2 collision cell
6	Q3 quadrupole
7	Detector

The Q1 quadrupole is a filtering quadrupole that sorts the ions before they enter the Q2 collision cell. In the Q2 collision cell the internal energy of an ion is increased through collisions with gas molecules until the molecular bonds break, creating product ions. This technique allows users to design experiments that measure the  $m/z$  of product ions to determine the composition of the parent ions.

After passing through the Q2 collision cell, the ions enter the Q3 quadrupole for additional filtering. Then they enter the detector. In the detector, the ions create a current that is converted into a voltage pulse. The voltage pulses leaving the detector are directly proportional to the quantity of ions entering the detector. The system monitors these voltage pulses and then converts the information into a signal. The signal represents the ion intensity for a particular  $m/z$  value and the system shows this information as a mass spectrum.

The linear ion trap (LIT) functionality provides a number of Enhanced modes of operation. A common factor of the Enhanced modes is that ions are trapped in the Q3 quadrupole region and then scanned out to produce full spectrum data. Many spectra are rapidly collected in a

short period of time and are significantly more intense than spectra collected in a comparable standard quadrupole mode of operation.

During the collection phase, ions pass through the Q2 collision cell where the CAD gas focuses the ions into the Q3 region. The Q3 quadrupole is operated with only the main RF voltage applied. Ions are prevented from passing through the Q3 quadrupole and are reflected back by an exit lens to which a DC barrier voltage is applied. After the fill time elapses, a time defined by the user, or determined by the Dynamic Fill Time feature, a DC barrier voltage is applied to the Q3 entrance lens (IQ3). This voltage confines the collected ions in the Q3 region and stops additional ions from entering. The entrance and exit lens DC voltage barriers and the RF voltage applied to the quadrupole rods confine the ions within the Q3 region.

During the scan out phase, the voltage on the exit lens and the auxiliary RF voltage are ramped simultaneously with the main RF voltage for increased resolution and sensitivity as compared to quadrupole scan types. An auxiliary AC frequency is applied to the Q3 quadrupole. The main RF voltage amplitude is ramped from low to high values, which sequentially brings masses into resonance with the auxiliary AC frequency. When ions are brought into resonance with the AC frequency, they acquire enough axial velocity to overcome the exit lens barrier and are axially ejected towards the mass spectrometer ion detector. Full spectra data can be acquired from the ions collected in the Q3 region by rapidly scanning the main RF voltage.

For information about the available software parameters, refer to the document: *Help*.

## ESI Mode

ESI produces gas phase ions of analytes in a sample by applying a high voltage to the sample effluent flowing through a needle. With the aid of heated gas flow, ESI produces singly and multiply charged ions in a relatively mild condition so that it is suitable for a wide range of compounds including small molecules, such as drugs or pesticides, and larger molecules, such as peptides, proteins, and other biopolymers. The sensitivity depends on the chemical properties of the analyte, the gas flow rate, the temperature, the voltage, and the mobile phase composition.

The ESI technique is mild enough to be used with labile compounds, such as peptides, proteins, and thermally labile pharmaceuticals. It functions with flow rates from 5  $\mu\text{L}/\text{min}$  to 3,000  $\mu\text{L}/\text{min}$  and it vaporizes 100% aqueous to 100% organic solvents.

Refer to the section: [Electrospray Ionization Mode](#).

## APCI Mode

The APCI mode is suitable for:

- Ionization of compounds that do not readily form ions in solution. These are usually non-polar compounds.

## Principles of Operation

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- Creation of simple APCI spectra for LC-MS/MS experiments.
- High-throughput analyses of complex and dirty samples. It is less sensitive to ion suppression effects.
- Rapid sample introduction by flow injection with or without an LC column.

The APCI technique can be used for volatile and thermally labile compounds with minimal thermal decomposition. The rapid desolvation and vaporization of the droplets and entrained analyte minimizes thermal decomposition and preserves molecular identity for ionization by the corona discharge needle. Buffers are readily tolerated by the ion source without significant contamination and the flash vaporization of the sprayed effluent allows up to 100% water to be used. The probe can accept the entire effluent, without splitting, at flow rates from 200  $\mu\text{L}/\text{min}$  to 3,000  $\mu\text{L}/\text{min}$ , through a wide-bore column.

Refer to the section: [APCI Mode](#).

## Data Handling

### Reporter Software

The Analyst MD software is responsible for data acquisition and processing and does not make any clinical decisions. The Reporter software extends the reporting functionality available in the Analyst MD software. The Reporter software can be used to create custom reports.

### MultiQuant MD Software

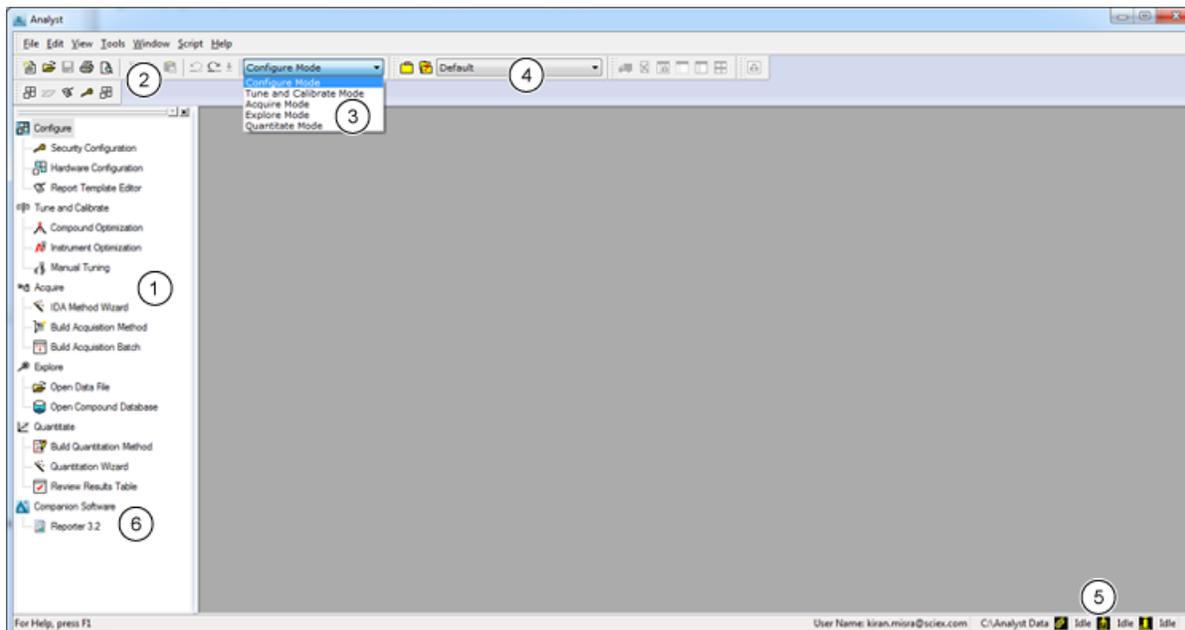
The MultiQuant MD software is quantitative data processing software that enables the processing of multiple analytes and samples at the same time. It is co-installed with the Analyst MD software. The MultiQuant MD software does not make any clinical decisions. The reporting functionality is available in the MultiQuant MD software and it can be used to create custom reports.

## Theory of Operation—Software

This section describes concepts used in the software.

## Analyst MD Software Window

Figure 4-8 Analyst MD Software Window



Item	Description
1	<p>Navigation bar: The Navigation bar gives access to the various software modes. Users can customize some elements of the Navigation bar to suit their preferences. For example, users can resize it, move it, or fix it in place. To hide the Navigation bar, click the × in the top right corner. To view the Navigation bar, click <b>View &gt; Navigation Bar</b>.</p> <p>The top level of the navigation tree has an icon that represents each software mode. Double-click the icon for a particular mode to expand or collapse the tree. This shows or hides the icons for the available functionality within the selected mode.</p>
2	Menu bar: Changes depending on the mode. Some options, such as Cut, Copy, and Paste, are the same in every mode. Other options are specific to certain modes and are unavailable in other modes.
3	Mode list: Click to change modes. Different modes have different toolbar icons available.
4	Project list: Click to change the project in which data is saved.

## Principles of Operation

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Item	Description
5	Instrument and peripheral device status: The Status bar contains information about current activities. It depicts the state of the instrument by color: green (Ready), yellow (Idle), red (Error), or white (no local instrument workstation). An icon indicates the status of a remote instrument. Double-click an icon to open the device status window.
6	Companion Software: Contains any installed companion software that is opened from the software.

## Analyst MD Software Modes

The software is divided into modes, which are discrete functional areas where users can perform a range of activities related to a main task. Users can access modes through the Navigation bar or the Mode list in the toolbar and can switch from one mode to another without losing any work.

**Table 4-3 Modes in the Analyst MD software**

Name	Description
<b>Configure</b>	Use this mode to configure devices and system settings. Set various options and parameters for the software, including hardware configuration and report template settings.
<b>Tune and Calibrate</b>	Use this mode to set options for tuning the instruments to ensure optimal results. In this mode, users can: <ul style="list-style-type: none"><li>• Perform instrument optimization.</li><li>• Perform manual tuning.</li><li>• Change the appearance of graphical views, select the types of information that are shown when file information is opened, and set linking options and other appearance options.</li><li>• Change processing options.</li></ul>

Table 4-3 Modes in the Analyst MD software (continued)

Name	Description
<b>Acquire</b>	Use this mode to set options to decide how samples should be acquired. In this mode, users can: <ul style="list-style-type: none"> <li>• Create an IDA acquisition method with the IDA Method Wizard.</li> <li>• Create an acquisition method with the Acquisition Method Editor.</li> <li>• Create a batch with the Batch Editor.</li> <li>• View the queue with Queue Manager.</li> <li>• Monitor the acquisition status.</li> </ul>
<b>Explore</b>	Use this mode to perform qualitative analysis on samples. In this mode, users can: <ul style="list-style-type: none"> <li>• View a graph.</li> <li>• View a chromatogram.</li> <li>• View a spectrum.</li> <li>• Show data in real time during batch acquisition.</li> </ul>
<b>Quantitate</b>	Use this mode to analyze the acquired data and build a quantitative method to generate a Results Table. Use the Results Table to manually review all of the peaks for each analyte and internal standard within a batch and to view calibration curves, sample statistics, and metric plots.

## Quantitative Analysis

Quantitative analysis is used to find the concentration of a specific substance in a sample. By analyzing an unknown sample and comparing it to standard samples, that is, samples containing the same substance with known concentrations, the software can calculate the concentration of the unknown sample. The process involves creating a calibration curve using the signal response or response ratio of the standards and then calculating the concentrations of the unknown samples. The calculated concentrations of all of the samples are added to a Results Table.

Quantitative analysis is most commonly performed using a Multiple Reactions Monitoring (MRM) scan. In an MRM scan, a precursor ion and a characteristic product ion are used to define an MRM transition that is highly specific to the analyte. The MRM transition, coupled with the retention time associated with the analyte during liquid chromatography, provides the specificity required for quantitation.

## Principles of Operation

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Quantitation is accomplished through the use of validated MRM LC-MS/MS acquisition methods, acquisition of calibration standard curves, and the subsequent integration of the peaks associated with the compounds of interest. The calibration curve relationship between signal response and concentration is used to determine the quantity of a particular analyte in an unknown sample.

## Integration

In LC-MS/MS data, integration refers to obtaining the area under a curve for the peak associated with a specific compound. Using a method that specifies the transitions, expected retention times, internal standards, integration, and regression parameters, the software is able to automatically integrate peaks for a given set of samples.

## Results Tables

Results Tables summarize the calculated concentration of an analyte in each unknown sample, based on the calibration curve. Results Tables also include the calibration curves as well as statistics for the results. The user can customize the Results Tables and view the Results Tables in layouts.

The data from a Results Table can be exported to a txt file for use in other applications, such as Microsoft Excel. The user can also export data in the table or just the data in the visible columns.

We recommend that users do not rename data files after Results Tables are created.

For information on using the Results Table right-click menu, refer to the section: [Results Table](#).

## Calibration Curves

A calibration curve, also known as a standard concentration curve, is a method for determining the concentration of a substance in an unknown sample by comparing the unknown sample to a set of standard samples of known concentration. The calibration curve is a plot of how the instrument responds (the analytical signal) to changes in the concentration of the analyte (the substance to be measured). The operator prepares a series of standards across a range of concentrations near the expected concentration of the analyte in the unknown sample.

Calibration standards are used to build calibration curves. Incorrect readings or missing readings on some of the calibration samples might indicate issues with the analytical run. Follow acceptable methods found in literature and regulatory agency guidances to create a calibration curve. Examples of good practices in the preparation of calibration curves include:

- Preparing calibration standards in a blank matrix in which the analyte is to be measured.
- Generating a calibration curve for each analyte to be measured.

- Making sure of the coverage of the expected concentration range of the analyte, including typical and atypical specimens.
- Using six to eight standards to generate the curve.

This is not a comprehensive list and other guidances should be used in determining the best practice in developing a calibration curve for the laboratory.

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**Note:** In some analytical runs, single-point calibration standards are used. Single-point calibrations are performed using a matrix blank sample and a single standard concentration. The relationship between instrument response and analyte concentration is determined by the line created by these two points. Both the acquisition and quantitation methods should be validated before being accepted for their intended use.

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### Regression Types

- Linear ( $y = mx + b$ )
- Linear through Zero ( $y = mx$ )
- Quadratic ( $y = a^2 + bx + c$ )

# Performance Characteristics and Specifications

# 5

This section includes characteristics and specifications for the Citrine Triple Quad MS/MS system and the Citrine QTRAP MS/MS system .

## Mass Spectrometer Specifications

**Table 5-1 Mass Spectrometer and Roughing Pump Specifications**

Sensitivity (MRM mode)	Reserpine 200 fg on column	S/N greater than 2000 CV less than 5%
Maximum scan speed	12,000 Da/sec	
Maximum linear ion trap scan speed (QTRAP systems only)	20,000 Da/sec	
Polarity switching	Minimum of 5 msec in MRM mode and <i>Scheduled</i> MRM mode	
Minimum MRM dwell time	1 msec	
Mass range ( <i>m/z</i> )	5 Da to 2000 Da	
Cross talk for reserpine 609/195	Less than 0.5% cross talk is detectable with a 2 msec dwell time and a 3 msec inter-MRM pause time	
Scan types	Full scan MS and selected ion MS for both Q1 and Q3, product ion scan, precursor ion scan, neutral loss or gain scan, multiple reaction monitoring (MRM) scan, and scheduled MRM scan types. For QTRAP systems only, enhanced MS scan, enhanced product ion scan, enhanced resolution scan, and MS <sup>3</sup> scan.	
Dynamic range	Five orders of magnitude	
<b>Weight</b>		
Weight—mass spectrometer	130 kg	
Weight—roughing pump	34 kg (75 lbs) per pump	
<b>Dimensions</b>		

## Performance Characteristics and Specifications

**Table 5-1 Mass Spectrometer and Roughing Pump Specifications (continued)**

Mass spectrometer (W × D × H)	79 cm × 79 cm × 59 cm
Roughing pump (W × D × H)	12 inches x 17 inches x 9 inches

**Table 5-2 Probe Specifications**

Parameter	TurbolonSpray Probe	APCI Probe
Ion source temperature range	Probe temperature from ambient temperature to 750 °C, depending on liquid flow	Probe temperature from ambient temperature to 750 °C, depending on liquid flow
Flow rate compatibility	5 µL/min to 3 mL/min	200 µL/min to 3 mL/min
<b>CAUTION: Potential System Damage. Do not exceed the maximum flow rate. Doing so can cause erratic performance or damage to the system.</b>		

**Table 5-3 Electrical Specifications**

<b>Mass Spectrometer</b>	
Nominal input voltage	200 VAC to 240 VAC
Input voltage fluctuation	± 10% of nominal
Frequency	50 Hz or 60 Hz
Maximum input current	10 A
Maximum input power	1,000 VA
<b>Roughing Pump per pump</b>	
Nominal input voltage	200 VAC to 240 VAC
Input voltage fluctuation	±10% of nominal
Frequency	50 Hz or 60 Hz
Maximum input current	6 A
Maximum input power	1,200 VA
<b>Computer</b>	

## Performance Characteristics and Specifications

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**Table 5-3 Electrical Specifications (continued)**

Nominal input voltage	100 VAC to 240 VAC
Input voltage fluctuation	±10% of nominal
Frequency	50 Hz or 60 Hz
Maximum input current	8 A (50 Hz), 6 A (60 Hz)
Maximum input power	460 W
<b>Monitor</b>	
Nominal input voltage	100 VAC to 240 VAC
Frequency	50 Hz or 60 Hz
Maximum input current	2.5 A

# Operating Instructions — Sample Workflows

# 6

**Table 6-1 Instrument Setup Workflow**

Step	To Do This	Find The Information In	What Does It Do?
1	Create a hardware profile.	<a href="#">Create a Hardware Profile</a>	Each hardware profile must include a mass spectrometer and other devices, such as an LC system. Only devices included in the active hardware profile can be used when creating acquisition methods.
2	Create projects to store data.	<a href="#">Create Projects and Subprojects</a>	Using projects and subprojects facilitates data management and makes comparison of results easier.
3	Optimize the mass spectrometer.	<a href="#">Verify Instrument Performance</a>	This is the process of optimizing the resolution and mass spectrometer parameters, and calibrating the mass spectrometer to obtain the best sensitivity and performance from the system.

**Table 6-2 Example Routine Analysis Workflow**

Step	To Do This	Find The Information In	What Does It Do?
1	Activate a hardware profile applicable to the method.	<a href="#">Create a Hardware Profile</a>	Each hardware profile must include a mass spectrometer and other devices, such as an LC system. Only devices included in the active hardware profile can be used when creating acquisition methods.
2	Create projects to store data.	<a href="#">Create Projects and Subprojects</a>	Using projects and subprojects facilitates data management and makes comparison of results easier.

## Operating Instructions — Sample Workflows

**Table 6-2 Example Routine Analysis Workflow (continued)**

Step	To Do This	Find The Information In	What Does It Do?
3	Create and submit a batch.	<a href="#">Add Sets and Samples to a Batch</a> and <a href="#">Submit a Sample or Set of Samples</a>	After creating an acquisition method, run samples by creating an acquisition batch and submitting the batch to the acquisition queue.
4	Equilibrate the system.	<a href="#">Equilibrate the System</a>	Equilibrate the system prior to starting data acquisition. A system that is not equilibrated can result in poor data.
5	Run samples to acquire data.	<a href="#">Acquire Data</a>	Running samples involves managing the acquisition queue and monitoring instrument and device status. To submit samples and acquire data, use the Queue Manager. The Queue Manager shows queue, batch, and sample status, and facilitates management of samples and batches in the queue.
6	Analyze data in Explore mode (optional).	<a href="#">Operating Instructions — Analyze and Explore Data</a>	In Explore mode, many tools are available for viewing and processing the acquired data. Graphs can be customized with peak labels and captions, contour plots can be shown, and spectra can be saved in the library.
<ul style="list-style-type: none"> <li>• For quantitative analysis using the MultiQuant MD software follow steps <a href="#">7</a> to <a href="#">8</a>. The MultiQuant MD software is recommended for larger data sets.</li> <li>• For quantitative analysis using the Analyst MD software follow steps <a href="#">9</a> to <a href="#">10a</a>.</li> <li>• For qualitative analysis using the Analyst MD software follow steps <a href="#">9</a> to <a href="#">10b</a>.</li> </ul>			
7	Analyze quantitative data in the MultiQuant MD software.	<i>MultiQuant MD Software Reference Guide:</i> Chapter 7, 8, 10, 11, 12, 13, 14	Generate and use a Results Table to manually review all of the peaks for each analyte and internal standard within a batch and to view calibration curves, sample statistics, and metric plots.

**Table 6-2 Example Routine Analysis Workflow (continued)**

Step	To Do This	Find The Information In	What Does It Do?
8	Create a report in the MultiQuant MD software.	<i>MultiQuant MD Software Reference Guide: Appendix C</i>	Generate a report using the provided report templates for the generated and reviewed results.
9	Analyze qualitative (or quantitative) data in the Analyst MD software.	<a href="#">Operating Instructions — Analyze and Process Quantitative Data</a>	Generate a Results Table to manually review all of the peaks for each analyte and internal standard within a batch. For quantitative analysis, also review calibration curves, sample statistics, and metric plots.
10a	Create a report in the Reporter software.	<a href="#">Generate Reports</a>	Generate a report using the provided report templates for the generated and reviewed results. For reports specific to qualitative analysis, use the set of report templates labeled with library searching.
10b	Select a library and then create a report using the Reporter software.	<a href="#">Generate Reports</a>	Select the appropriate MS/MS spectra library for the results and generate a report using the provided report templates labeled as library search for the generated and reviewed results. For reports specific to qualitative analysis, use the set of report templates labeled with library search.

**Table 6-3 Example Method Developer Workflow**

Step	To Do This	Find The Information In	What Does It Do?
1	Create a hardware profile.	<a href="#">Create a Hardware Profile</a>	Each hardware profile must include a mass spectrometer and other devices, such as an LC system. Only devices included in the active hardware profile can be used when creating acquisition methods.
2	Create projects to store data.	<a href="#">Create Projects and Subprojects</a>	Using projects and subprojects facilitates data management and makes comparison of results easier.

## Operating Instructions — Sample Workflows

**Table 6-3 Example Method Developer Workflow (continued)**

Step	To Do This	Find The Information In	What Does It Do?
3	Optimize the compound automatically  Or Step 4 - Optimize the compound manually.	<a href="#">Operating Instructions — Automatic Optimization</a>	The software automatically optimizes the compound and mass spectrometer parameters for the compounds of interest.
4	Optimize the compound manually.	<a href="#">Manual Compound Optimization</a>	The user manually optimizes the compound and mass spectrometer parameters for the compounds of interest. Manual optimization provides more experienced users with greater control during the optimization process.
5	Create an acquisition method.	<a href="#">Operating Instructions — Acquisition Methods</a>	To analyze samples, create an acquisition method for the mass spectrometer and any LC devices. An acquisition method indicates which peripheral devices to use, when to use them to acquire data, and the associated parameters.
6	Create and submit a batch.	<a href="#">Add Sets and Samples to a Batch</a> and <a href="#">Submit a Sample or Set of Samples</a>	After creating an acquisition method, run samples by creating an acquisition batch and submitting the batch to the acquisition queue.
7	Equilibrate the system.	<a href="#">Equilibrate the System</a>	Equilibrate the system prior to starting data acquisition. A system that is not equilibrated can result in poor data.

Table 6-3 Example Method Developer Workflow (continued)

Step	To Do This	Find The Information In	What Does It Do?
8	Run samples to acquire data.	<a href="#">Acquire Data</a>	Running samples involves managing the acquisition queue and monitoring instrument and device status. To submit samples and acquire data, use the Queue Manager. The Queue Manager shows queue, batch, and sample status, and facilitates management of samples and batches in the queue.
9	Analyze data in Explore mode (optional).	<a href="#">Operating Instructions — Analyze and Explore Data</a>	In Explore mode, many tools are available for viewing and processing the acquired data. Graphs can be customized with peak labels and captions, contour plots can be shown, and spectra can be saved in the library.
<ul style="list-style-type: none"> <li>• For quantitative analysis using the MultiQuant MD software follow steps <a href="#">10</a> to <a href="#">12</a>. The MultiQuant MD software is recommended for larger data sets.</li> <li>• For quantitative analysis using the Analyst MD software follow steps <a href="#">13</a> to <a href="#">15</a>.</li> <li>• For qualitative analysis, contact Support.</li> <li>• For qualitative analysis using the Analyst MD software follow steps <a href="#">14</a> to <a href="#">15</a>. For general guidance, contact support.</li> </ul>			
10	Create a quantitative method in the MultiQuant MD software.	<i>MultiQuant MD Software Reference Guide: Quantitation Method Editor</i>	Use the various quantitative method creation tools in the software to analyze the acquired data and build a quantitative method to generate a Results Table.
11	Analyze quantitative data in MultiQuant MD software.	<i>MultiQuant MD Software Reference Guide: Chapter 7, 8, 10, 11, 12, 13, 14</i>	Generate and use a Results Table to manually review all of the peaks for each analyte and internal standard within a batch and to view calibration curves, sample statistics, and metric plots.

## Operating Instructions — Sample Workflows

**Table 6-3 Example Method Developer Workflow (continued)**

Step	To Do This	Find The Information In	What Does It Do?
12	Create a report in MultiQuant MD software.	<i>MultiQuant MD Software Reference Guide: Appendix C</i>	Generate a report using the provided report templates for the generated and reviewed results.
13	Create a quantitative method in the Analyst MD software.	<i>MultiQuant MD Software Reference Guide: Quantitation Method Editor</i>	Use the various quantitative method creation tools in the software to analyze the acquired data and build a quantitative method to generate a Results Table.
14	Analyze qualitative (or quantitative) data in Analyst MD software.	<a href="#">Operating Instructions — Analyze and Process Quantitative Data</a>	Generate a Results Table to manually review all of the peaks for each analyte and internal standard within a batch. For quantitative analysis, also review calibration curves, sample statistics, and metric plots.
15	Create a report in the Analyst Reporter.	<a href="#">Generate Reports</a>	Generate a report using the provided report templates for the generated and reviewed results. For reports specific to qualitative analysis, use the set of report templates labeled with library searching.

# Operating Instructions — Mass Spectrometer and Ion Source

# 7



**WARNING! Personal Injury Hazard.** Follow the instructions in the documentation when using the system. The protection provided by the equipment might be impaired if the equipment is used in a manner not specified by SCIEX.

## Start Up the System



**WARNING! Electrical Shock Hazard.** Make sure that the system can be disconnected from the mains supply outlet in an emergency. Do not block the mains supply outlet.

**Note:** Before operating the instrument, read the safety information in the section: [Operational Precautions and Limitations](#).

### Prerequisites

- The site requirements specified in the *Site Planning Guide* are met. The *Site Planning Guide* includes information on the mains supply and connections, compressed air, nitrogen, roughing pump, ventilation, exhaust, and site clearance requirements. Contact SCIEX for a copy of the *Site Planning Guide*, if required. For contact information, go to [sciex.com/contact-us](http://sciex.com/contact-us).
- The source exhaust gas, compressed air, and nitrogen gases are connected to the mass spectrometer.
- The 4 L source exhaust drain bottle is connected to the exhaust waste connection on the back of the mass spectrometer and to the laboratory ventilation system.
- The source exhaust hoses are securely clamped at the mass spectrometer, source exhaust drain bottle, and ventilation connections.
- The mass spectrometer convenience switch is turned off and the mains supply cable is connected to the mass spectrometer.
- The mass spectrometer and roughing pump mains supply cables are connected to the 200 VAC to 240 VAC mains supply.
- The Ethernet cable is connected to both the mass spectrometer and the computer.

1. Turn on the mass spectrometer convenience switch. Refer to the figure: [Figure 4-2](#).
2. Turn on the computer.
3. Open the control software.

## Ion Source Optimization



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**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Do not use the ion source without knowledge of and training in the proper use, containment, and evacuation of toxic or injurious materials used with the ion source.**

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**WARNING! Fire Hazard. Do not direct more than 3 mL/min of flammable solvent in the ion source. Although the LC components can provide a flow rate of up to 5 mL/min, exceeding the maximum flow rate can cause solvent to accumulate in the ion source. Do not use the ion source if the source exhaust system is not enabled and functioning when the ion source and the probe are properly installed.**

---



---

**WARNING! Puncture Hazard, Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Discontinue use of the ion source if the ion source window is cracked or broken, and then contact a SCIEX Field Service Employee (FSE). Any toxic or injurious materials introduced in the equipment will be present in the source exhaust output. Exhaust from equipment should be vented from the room. Dispose of sharps following established laboratory safety procedures.**

---

Optimize the ion source whenever the analyte, flow rate, or mobile phase composition changes.

When optimizing ion source-dependent parameters, introduce the sample at a flow rate that will be used during sample analysis, using either flow injection analysis (FIA) or tee infusion as the method of sample introduction. Optimize the position of the ion source before optimizing the ion source-dependent parameters.

Several parameters affect the performance of the source. Optimize the performance while injecting a known compound and monitoring the signal of the known ion. Adjust the micrometer and gas and voltage parameters to maximize the signal-to-noise ratio and signal stability.

Refer to the section: [TurbolonSpray Probe Optimization](#) or [APCI Probe Optimization](#).

## TurbolonSpray Probe Optimization



---

**WARNING!** Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Make sure that the source exhaust system is connected and functioning, and that good general laboratory ventilation is provided. Adequate laboratory ventilation is required to control solvent and sample emissions, and to provide for the safe operation of the system.

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---

**WARNING!** Fire Hazard. Do not direct more than 3 mL/min of flammable solvent in the ion source. Although the LC components can provide a flow rate of up to 5 mL/min, exceeding the maximum flow rate can cause solvent to accumulate in the ion source. Do not use the ion source if the source exhaust system is not enabled and functioning when the ion source and the probe are properly installed.

---



---

**WARNING!** Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Make sure that the electrode protrudes beyond the tip of the probe, to prevent hazardous vapors from escaping from the source. The electrode must not be recessed within the probe.

---

---

**CAUTION:** Potential System Damage. If the LC system connected to the mass spectrometer is not controlled by the software, then do not leave the mass spectrometer unattended while in operation. The liquid stream from the LC system can flood the ion source when the mass spectrometer goes in Standby state.

---

---

**Note:** To keep the system clean and at optimum performance, adjust the probe position when changing the flow rate.

---

---

**Tip!** It is easier to optimize signal and signal-to-noise with flow injection analysis than with on-column injections.

---

---

**Note:** If the **IonSpray Voltage** is too high, then a corona discharge can occur. A corona discharge is visible as a blue glow at the tip of the probe. It results in decreased sensitivity and stability of the signal.

---

## Flow Rate and Ion Source Temperature

The sample introduction flow rate and the sample solvent composition affect the optimal TurbolonSpray probe temperature. A higher flow rate or a higher aqueous content requires a higher optimal temperature.

The TurbolonSpray probe is often used with sample flow rates of 5  $\mu\text{L}/\text{min}$  to 1,000  $\mu\text{L}/\text{min}$ . The heat is used to increase the rate of evaporation which improves ionization efficiency, resulting

in increased sensitivity. Extremely low flow rates of high organic solvent usually do not require increased temperatures. Refer to the section: [Source Parameters and Voltages](#).

### Method

The liquid sample stream is delivered to the ion source by an LC pump or by a syringe pump. If it is delivered by an LC pump, then the sample can be injected directly in the mobile phase using flow injection analysis (FIA) or tee infusion, through a syringe pump, or through a separation column using a loop injector or autosampler. If it is introduced by a syringe pump, then the sample is injected directly in the ion source. Infusion optimization can only be used for ion path optimization and MS/MS fragment selection.

### Prepare the System

To create a method that is optimized for a compound, refer to the section: [Manual Compound Optimization](#).

1. Open the control software.
2. On the Navigation bar, under **Tune and Calibrate** mode, double-click **Manual Tuning**.
3. Open a previously optimized method or create a method based on the compounds.
4. If the ion source has been allowed to cool, then do the following.
  - a. Set the ion source temperature to 450.
  - b. Let the ion source warm up for 30 minutes.

The 30-minute warm-up stage prevents solvent vapors from condensing in the cold probe.

5. Start the solvent flow and sample injection.

### Set the Starting Conditions

1. In the Tune Method Editor, make sure that the correct **Scan Type** and appropriate compound parameters are selected.
2. Type a starting value for **Ion Source Gas 1**.  
For LC pumps, use a value between 40 and 60 for Gas 1.
3. Type a starting value for **Ion Source Gas 2 (GS2)**.  
For LC pumps, use a value between 30 and 50 for Gas 2.

---

**Note:** Gas 2 is used with higher flow rates typical with an LC system and in conjunction with increased temperature.

---

4. Type 20 in the **Curtain Gas (CUR)** field.
5. Start acquisition.

## Optimize the TurbolonSpray Probe Position



**WARNING!** Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Make sure that the electrode protrudes beyond the tip of the probe, to prevent hazardous vapors from escaping from the source. The electrode must not be recessed within the probe.

---



**WARNING!** Puncture Hazard. Be careful when handling the electrode. The tip of the electrode is extremely sharp.

---

After the probe is optimized, it needs only minor adjustment. If the probe is removed, or if the analyte, flow rate, or solvent composition change, then repeat the optimizing procedure.

Refer to the section: [Ion Source Components](#).

1. Look through the window of the ion source to view the position of the probe.
2. Monitor the signal or signal-to-noise of the analytes in the control software.
3. Use the horizontal micrometer to adjust the probe position in small increments to achieve the best signal or signal-to-noise ratio.

The probe can optimize slightly to either side of the aperture.

---

**Tip!** Adjust the horizontal micrometer setting to direct the liquid spray from the TurbolonSpray probe away from the aperture to prevent contamination of the aperture; to prevent piercing of the flow of the gas for the Curtain Gas interface, which can create an unstable signal; and to prevent electrical shorting due to the presence of the liquid.

---

4. Use the vertical micrometer to adjust the probe position in small increments to achieve the best signal or signal-to-noise ratio.
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**Note:** The vertical position of the probe depends on the flow rate. At lower flow rates, the probe should be closer to the aperture. At higher flow rates, the probe should be farther from the aperture.

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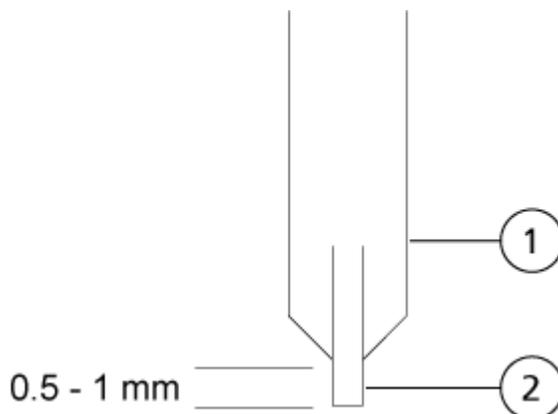
5. Adjust the black electrode adjustment nut on the probe to move the electrode tube in or out of the probe (to adjust the protrusion).
- 

**Note:** The electrode tip should protrude between 0.5 mm and 1.0 mm from the end of the probe.

---

The optimal setting for the electrode tip is compound-dependent. The distance that the electrode tip protrudes affects the shape of the spray cone, and the shape of the spray cone affects mass spectrometer sensitivity.

**Figure 7-1 Electrode Tip Extension Adjustment**



Item	Description
1	Probe
2	Electrode

### Optimize Source and Gas Parameters and Voltage

Optimize ion source gas 1 (nebulizer gas) for the best signal stability and sensitivity. Ion source gas 2 (heater gas) aids in the evaporation of solvent, which helps to increase the ionization of the sample.

Too high a temperature can cause premature vaporization of the solvent at the tip of the TurbolonSpray probe, especially if the probe protrudes too far, which results in signal instability and a high chemical background noise. Similarly, a high heater gas flow can produce a noisy or unstable signal.

---

**Note:** If the **IonSpray Voltage** is too high, then a corona discharge can occur. A corona discharge is visible as a blue glow at the tip of the probe. It results in decreased sensitivity and stability of the signal.

---

1. Adjust ion source gas 1 and ion source gas 2 in increments of 5 to achieve the best signal or signal-to-noise ratio.

---

**Note:** Ion source gas 2 is used with higher flow rates typical with an LC system and in conjunction with increased temperature.

---

2. Increase the flow rate of the gas for the Curtain Gas interface until the signal begins to decrease.

**Note:** To prevent contamination, use the highest possible value for the flow rate of the gas for the Curtain Gas interface that does not sacrifice sensitivity. Do not set the flow rate lower than the values in the table: [Table 7-1](#). This helps to prevent penetration of the flow of the gas for the Curtain Gas interface, which can produce a noisy signal; prevent contamination of the aperture; and increase the overall signal-to-noise ratio.

**Table 7-1 CUR Parameter Values**

Mass Spectrometer	Starting Value
Citrine systems	30

3. Adjust the **IonSpray Voltage (IS)** Spray Voltage in increments of 500 V to maximize signal-to-noise.

### Optimize the Turbo Heater Temperature

The optimal heater temperature is dependent on the compound, flow rate, and mobile phase composition. The higher the flow rate and the higher the aqueous composition, the higher the optimized temperature.

When optimizing the source temperature, make sure that the ion source equilibrates to the new temperature setting.

Adjust the ion source temperature in increments of 50 °C to 100 °C to achieve the best signal or signal-to-noise ratio.

### APCI Probe Optimization



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**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Make sure that the source exhaust system is connected and functioning, and that good general laboratory ventilation is provided. Adequate laboratory ventilation is required to control solvent and sample emissions, and to provide for the safe operation of the system.**

---



---

**WARNING! Fire Hazard. Do not direct more than 3 mL/min of flammable solvent in the ion source. Although the LC components can provide a flow rate of up to 5 mL/min, exceeding the maximum flow rate can cause solvent to accumulate in the ion source. Do not use the ion source if the source exhaust system is not enabled and functioning when the ion source and the probe are properly installed.**

---



**WARNING!** Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Make sure that the electrode protrudes beyond the tip of the probe, to prevent hazardous vapors from escaping from the source. The electrode must not be recessed within the probe.

---

**CAUTION: Potential System Damage.** If the LC system connected to the mass spectrometer is not controlled by the software, then do not leave the mass spectrometer unattended while in operation. The liquid stream from the LC system can flood the ion source when the mass spectrometer goes in Standby state.

---

**Note:** The minimum flow rate supported by the APCI probe is 200  $\mu\text{L}/\text{min}$ . For a complete list of APCI probe parameters, refer to the section: [APCI Probe Parameters](#).

---

**Tip!** It is easier to optimize signal and signal-to-noise with flow injection analysis than with on-column injections.

---

**Note:** When using the APCI probe, make sure that the corona discharge needle is pointing toward the aperture.

---

### Prepare the System

To create a method that is optimized for a compound, refer to the section: [Manual Compound Optimization](#).

1. Open the control software.
2. On the Navigation bar, under **Tune and Calibrate** mode, double-click **Manual Tuning**.
3. Open a previously optimized method or create a method based on the compounds.
4. If the ion source has been allowed to cool, then do the following.
  - a. Set the ion source temperature to 450.
  - b. Let the ion source warm up for 30 minutes.

The 30-minute warm-up stage prevents solvent vapors from condensing in the cold probe.

5. Start the solvent flow and sample injection.

### Set the Starting Conditions

1. In the Tune Method Editor, make sure that the correct **Scan Type** and appropriate compound parameters are selected.
  2. Type 30 in the **Ion Source Gas 1 (GS1)** field.
  3. Type 20 in the **Curtain Gas (CUR)** field.
-

4. Type 1 in the **Nebulizer Current (NC)** field.
5. On the Compound tab, in the **Declustering potential (DP)** field, type 100.
6. Start acquisition.

## Optimize Source and Gas Parameters

1. Adjust ion source gas 1 in increments of five to achieve the best signal or signal-to-noise ratio.
2. Increase the flow rate of the gas for the Curtain Gas interface until the signal starts to decrease.

**Note:** To prevent contamination, use the highest possible value for the flow rate of the gas for the Curtain Gas interface that does not sacrifice sensitivity. Do not set the flow rate lower than the values in the table: [Table 7-2](#). This helps to prevent penetration of the flow of the gas for the Curtain Gas interface, which can produce a noisy signal; prevent contamination of the aperture; and increase the overall signal-to-noise ratio.

**Table 7-2 CUR Parameter Values**

Mass Spectrometer	Starting Value
Citrine systems	30

## Adjust the Position of the Corona Discharge Needle



**WARNING! Electrical Shock Hazard.** Follow this procedure to avoid contact with the high voltages applied to the corona discharge needle, curtain plate, and turbo heaters.

### Required Materials

- Insulated flat-bladed screwdriver

When using the APCI probe, make sure that the corona discharge needle is pointing toward the aperture. When using the TurbolonSpray probe, make sure that the corona discharge needle is pointed away from the aperture.

1. Use an insulated flat-bladed screwdriver to rotate the corona discharge needle adjustment screw on the top of the needle.
2. Look through the glass window to make sure that the needle is aligned with the tip facing the aperture.

## Optimize the APCI Probe Position



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Make sure that the electrode protrudes beyond the tip of the probe, to prevent hazardous vapors from escaping from the source. The electrode must not be recessed within the probe.**

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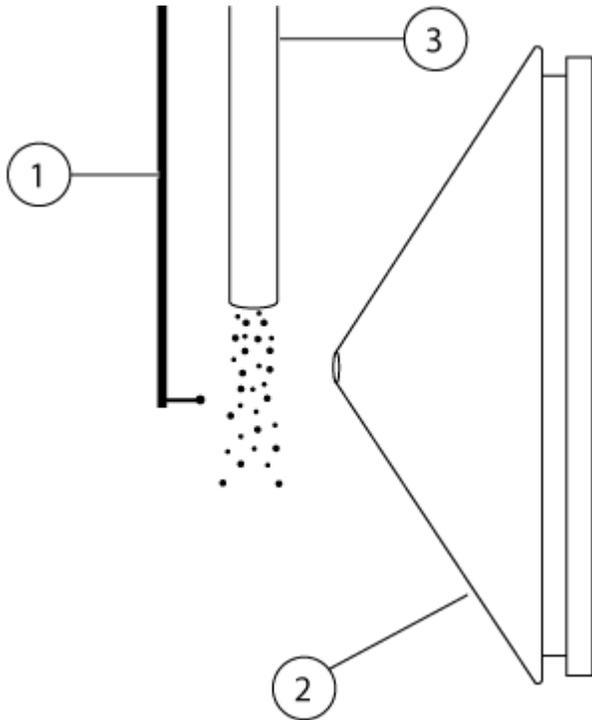
**WARNING! Puncture Hazard. Be careful when handling the electrode. The tip of the electrode is extremely sharp.**

---

Make sure that the curtain plate aperture remains clear of solvent or solvent droplets at all times.

The position of the sprayer nozzle affects sensitivity and signal stability. Adjust the probe position in small increments only. At lower flow rates, move the probe closer to the aperture. For higher flow rates, move the probe farther away from the aperture. After the probe is optimized, it needs only minor adjustment. If the probe is removed, or if the analyte, flow rate, or solvent composition changes, then repeat the optimization procedure.

**Figure 7-2 Sprayer Nozzle Position**



Item	Description
1	Corona discharge needle

## Operating Instructions — Mass Spectrometer and Ion Source

---

Item	Description
2	Curtain plate
3	APCI probe

1. Use the previous horizontal and vertical micrometer settings or set them to 5 as a starting position.

---

**Note:** To avoid reducing the performance of the mass spectrometer, do not spray directly in the aperture.

---

2. In the control software, monitor the signal or signal-to-noise of the analytes.
3. Use the horizontal micrometer to adjust the probe in small increments to achieve the best signal or signal-to-noise ratio.
4. Use the vertical micrometer to adjust the probe in small increments to achieve the best signal or signal-to-noise ratio.
5. Adjust the black electrode adjustment nut on the probe to move the electrode tube in or out of the probe (to adjust the protrusion).

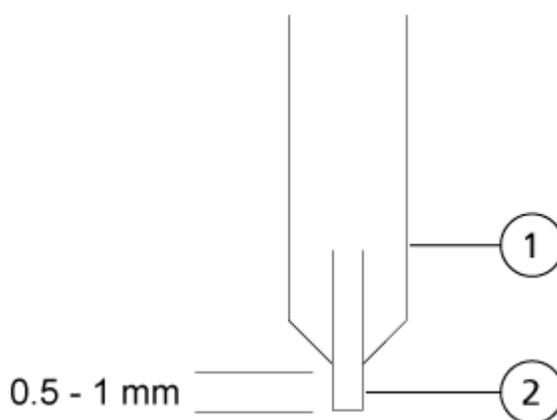
---

**Note:** The electrode tip should protrude between 0.5 mm and 1.0 mm from the end of the probe.

---

The optimal setting for the electrode tip is compound-dependent. The distance that the electrode tip protrudes affects the shape of the spray cone, and the shape of the spray cone affects mass spectrometer sensitivity.

**Figure 7-3 Electrode Tip Extension Adjustment**



Item	Description
1	Probe
2	Electrode

### Optimize the Nebulizer Current

The ion source is controlled by current and not by voltage. Select the appropriate current for the acquisition method, regardless of the ion source selection position.

Start with a nebulizer current value of 3 and then increase or decrease it to achieve the best signal or signal-to-noise ratio.

The nebulizer current applied to the corona discharge needle usually optimizes between 1  $\mu\text{A}$  and 5  $\mu\text{A}$  in either polarity. If no changes in signal are observed when the current is increased, then leave the current at the lowest value that provides the best signal or signal-to-noise ratio.

### Optimize the APCI Probe Temperature

The quantity and type of solvent affects the optimal APCI probe temperature. At higher flow rates, the optimal temperature increases.

---

**Note:** When the ion source is turned on, the ion source heaters require 1 minute to 2 minutes before becoming red, indicating that the source is warming up.

---

Adjust the ion source temperature in increments of 50 °C to 100 °C to achieve the best signal or signal-to-noise ratio.

### Optimization Tips

Optimization of the ion source minimizes the need for cleaning of the ion source and vacuum interface components.

- Use the highest temperature possible when optimizing compounds. A temperature of 700 °C is common for many compounds. High temperatures help keep the ion source clean and reduce background noise.
- Use the highest possible value for the flow rate of the gas for the Curtain Gas interface that does not sacrifice sensitivity. This helps to:
  - Prevent penetration of the flow of gas for the Curtain Gas interface, which can produce a noisy signal.
  - Prevent contamination of the aperture.
  - Increase the overall signal-to-noise ratio.

- Adjust the horizontal micrometer setting to direct the liquid spray from the probe away from the aperture to:
  - Prevent contamination of the aperture.
  - Prevent piercing of the flow of gas for the Curtain Gas interface, which can create an unstable signal.
  - Prevent electrical shorting due to the presence of the liquid.

To do so, use the vertical micrometer to move the probe up.

- Use the lowest IonSpray Voltage possible without losing signal. Focus on signal-to-noise and not just signal.

## Mass Spectrometer Calibration Procedure

Mass calibration can change over time. Verify the mass calibration regularly. At installation, the system is mass calibrated and the resolution of the spectral peaks optimized at both unit and high resolution, in both Positive and Negative mode, to obtain the best sensitivity and performance of the mass spectrometer. Mass calibration makes sure that the signals originating from the ionized compounds are recorded at their true  $m/z$  values.<sup>1</sup> The  $m/z$  scale is calibrated using compounds of a known purity and mass. During resolution optimization, the peak width and peak shape are adjusted. Mass spectral resolution is represented as  $(m/z)/\text{Width}$  where Width is the peak width of a spectral peak at a given  $m/z$  value.<sup>1</sup> The peak width is typically measured at the points where the intensity is half the peak height.

---

**Note:** With increasing resolution, sensitivity decreases. It is important to find a balance between the requirements for sensitivity and resolution.

---

The Instrument Optimization module in the Analyst MD software is used to mass calibrate and optimize the resolution. Verify the mass calibration and resolution weekly or after cleaning the instrument to confirm that the system is working properly. In general, the calibration and resolution for a triple quadrupole mass spectrometer is stable for three months to six months unless the system loses vacuum. If the system loses vacuum, then check the calibration and resolution before using the system. Refer to the section: [Operating Instructions — Tune and Calibrate](#).

---

**CAUTION: Potential Wrong Result. Make sure that the system is calibrated. If the system is not calibrated properly, then a mass might be identified incorrectly or quantitation might be inaccurate.**

---

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<sup>1</sup> CLSI Standard C50–A–Vol. 27, No. 24—Mass Spectrometry in the Clinical Laboratory: General Principles and Guidance: Approved Guideline.

Refer to the sections: [Operational Precautions and Limitations](#) and [Calibration Ions and Solutions](#).

## Reset the Mass Spectrometer

1. Stop any ongoing scans and then turn off sample flow to the mass spectrometer.
2. Close the control software.
3. Press and hold the **Reset** button for five seconds.  
A click is heard when the relay activates. After approximately 3 minutes, the mass spectrometer reaches operating pressure.

## Shut Down and Vent the System

Some procedures require that the system be shut down. Others require that it also be vented. Follow these steps to shut down and, if required, vent the system.

---

**CAUTION: Potential System Damage. Do not turn off the roughing pumps until after the turbo pumps have spun down.**

---

**Note:** If the input gas supply must be disconnected, then relieve the pressure in the gas lines before disconnecting it.

---

**Tip!** If the mass spectrometer will not be used for an extended period, then leave it in Standby state with the ion source in place. If the mass spectrometer must be shut down, then follow these instructions.

---

1. Complete or stop any ongoing scans.
- 

**CAUTION: Potential System Damage. Turn off the sample flow before shutting down the system.**

---

2. Turn off the sample flow to the system.
  3. Close the software.
  4. (If required) Follow these steps to vent the system:
- 

**Note:** Vent the system before performing a full cleaning of the vacuum interface, before cleaning the Q0 region, and before replacing the roughing pump oil. For more information, contact the Qualified Maintenance Person (QMP) or FSE.

---

- a. Press and hold the **Vent** button for three seconds.
-

The Vacuum LED begins flashing more quickly than during pump down. The turbo pump spins down gradually.

- b. Allow the system to vent for 15 minutes.
5. Turn off the mass spectrometer convenience switch.
6. Disconnect the mass spectrometer mains supply cable from the mains supply outlet.
7. (If venting the system) Disconnect the roughing pump mains supply cable from the mains supply outlet.

# Operating Instructions — Hardware Profiles and Projects

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# 8

## Hardware Profiles

A hardware profile tells the software how the mass spectrometer and the devices are configured and connected to the computer. Multiple hardware profiles can be configured, but only one profile can be active at any time.

When a hardware profile is created in the Hardware Configuration Editor, the peripheral devices must be configured so that the software can communicate with them. Configuring the peripheral devices requires two procedures:

- Making the physical connections. For information about making the physical connections to the devices, refer to the document: *Peripheral Devices Setup Guide*.
- Configuring the software to communicate with the peripheral devices. For a list of the supported devices, refer to the Analyst MD software document: *Software Installation Guide*.

When the software is installed, the driver required for each peripheral device is also installed. After the peripheral devices are physically connected to the computer, configure the appropriate configuration information.

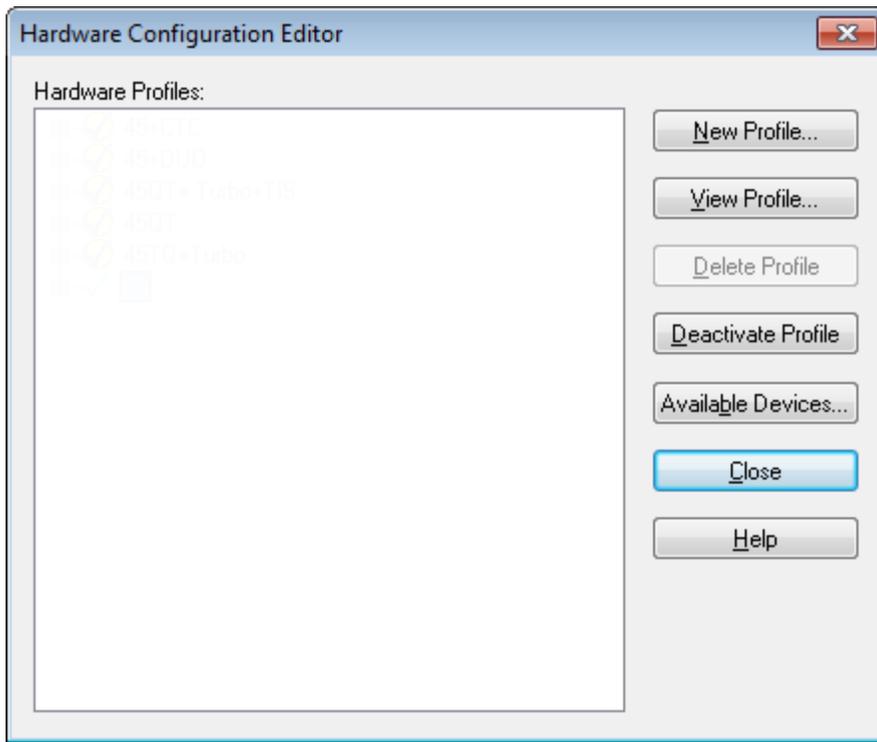
Each hardware profile must include a mass spectrometer. Before creating an acquisition method, make sure that all devices to be used in the method are included in the hardware profile. The devices configured in the active hardware profile and selected in the Add/Remove Device Method dialog are shown as icons in the Acquisition method pane. Only peripheral devices included in the active hardware profile can be used to create acquisition methods.

## Create a Hardware Profile

The user can create multiple hardware profiles, but only one profile can be active at any time.

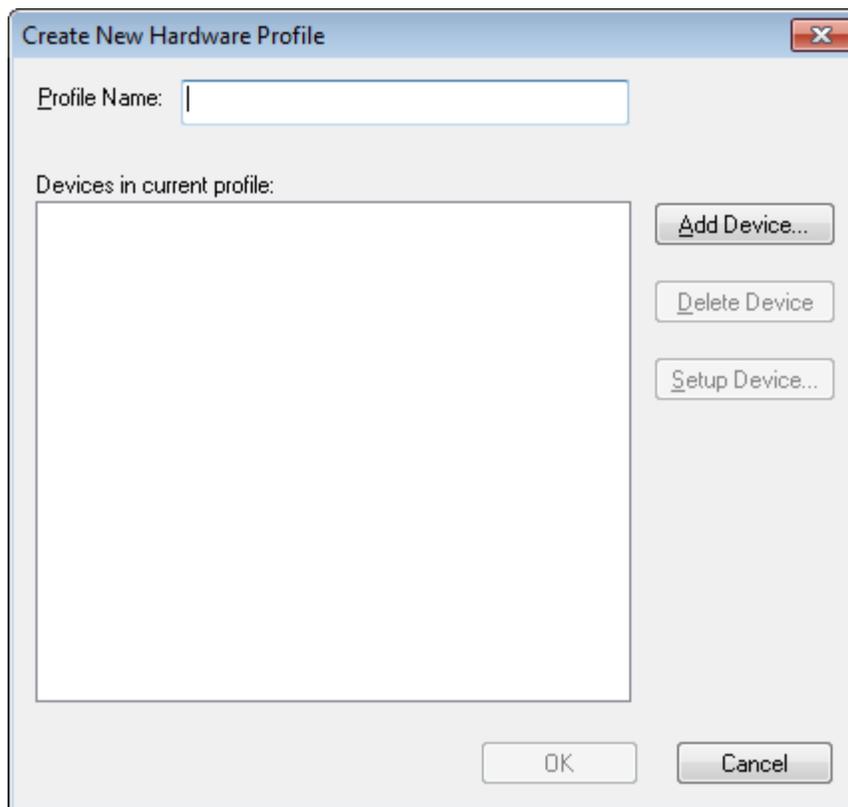
1. On the Navigation bar, under **Configure**, double-click **Hardware Configuration**.

**Figure 8-1 Hardware Configuration Editor Dialog**



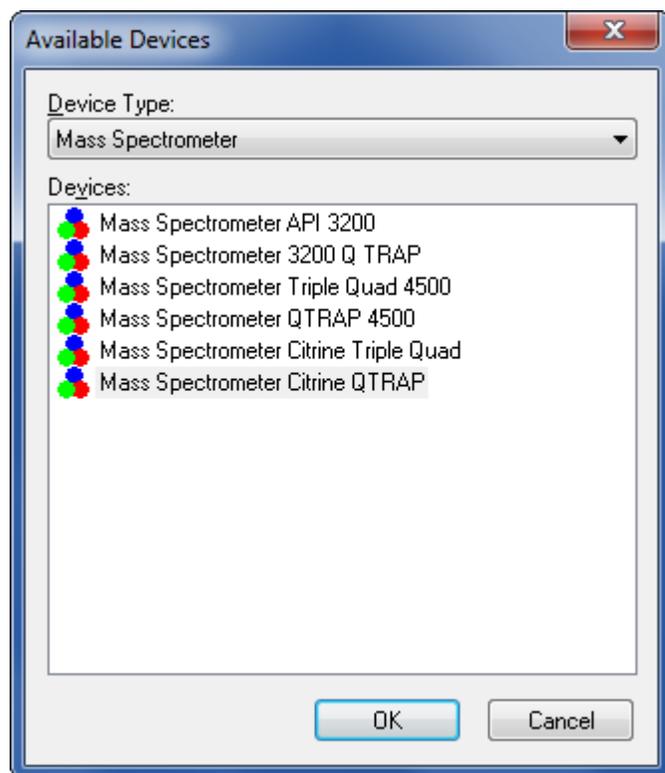
2. Click **New Profile**.

**Figure 8-2 Create New Hardware Profile Dialog**



3. Type a name in the **Profile Name** field.
4. Click **Add Device**.

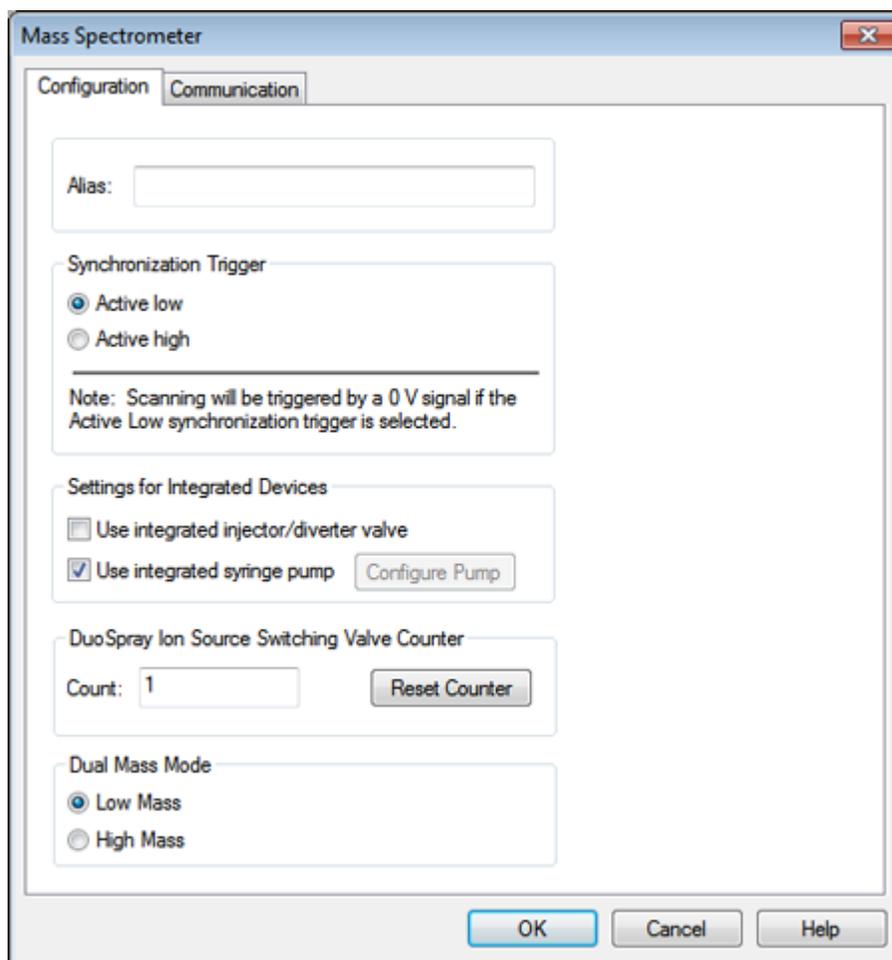
**Figure 8-3 Available Devices Dialog**



In the Available Devices dialog, **Mass Spectrometer** is the preset value for the **Device Type** field.

5. In the **Devices** list, select the appropriate mass spectrometer and then click **OK**.
6. On the Create New Hardware Profile dialog, click **Setup Device**.
7. On the Configuration tab, in the Dual Mass Mode section, select one of the following options:
8. (Optional) To configure mass spectrometers that use the integrated syringe pump, on the Configuration tab, select the **Use integrated syringe pump** check box.

Figure 8-4 Configuration Tab with Syringe Pump Configured



9. (Optional) To configure the mass spectrometer for the diverter valve, on the Configuration tab, select **Use integrated injector/diverter valve**.
10. (Optional) Select additional features on the Configuration and Communication tabs as required.
11. Click **OK**.
12. On the Create New Hardware Profile dialog, click **Add Device** and then add and set up each device that is used with the mass spectrometer. Refer to the section: [Add Devices to a Hardware Profile](#).
13. Click **OK** in the Create New Hardware Profile dialog.
14. Click the hardware profile to be activated in the Hardware Configuration Editor.
15. Click **Activate Profile**.

The check mark turns green. If a red × is shown, then there is an issue with the hardware profile activation.

---

**Tip!** A hardware profile need not be deactivated before another is activated. Click a hardware profile and then click **Activate Profile**. The active profile is deactivated automatically.

---

16. Click **Close**.

---

**Note:** The current operating mode of the active hardware profile can be viewed in the Detailed Status dialog for the instrument by double-clicking the Mass Spec icon on the lower-right section in the Analyst MD software window.

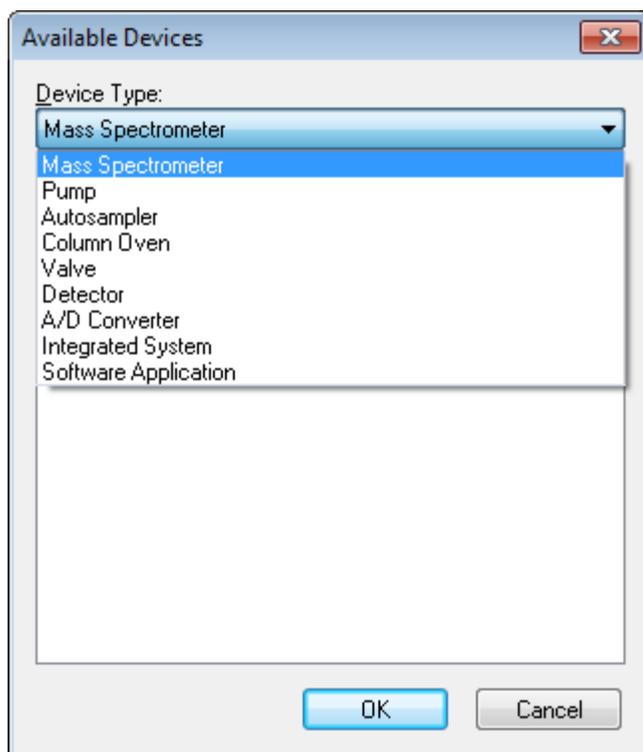
---

## Add Devices to a Hardware Profile

Devices must be configured to enable the software to communicate with them. When the software is installed, the driver required for each device is also installed. Before devices can be configured, they must be physically connected to the computer. For more information, refer to the document: *Peripheral Devices Setup Guide*.

1. Open the Hardware Configuration Editor.
2. In the **Hardware Profiles** list, deactivate the hardware profile.
3. Click **Edit Profile**.
4. Click **Add Device**.  
The Available Devices dialog opens.
5. In the **Device Type** list, select the device and then click **OK**.

**Figure 8-5 Available Devices Dialog**



6. Click **OK**.
7. Select the device from the **Devices** list and then click **OK**.
8. Click **Setup Device**.  
A dialog containing configuration values for the device opens.
9. (Optional) On the Communication tab, in the **Alias** field, type a name or other identifier for the device.

---

**Note:** For devices using serial communication, make sure that the serial port selected matches the serial port to which the device is physically connected.

---

**Note:** The **Alias** field might also be referred to as the **Name** box and might be found on another tab under **Alias**.

---

- If the device uses a **Serial Port** as a communication interface, then in the **COM Port Number** list, select the COM port to which the device is connected.
- If the device uses **Ethernet** as a communication interface, then type the **IP Address** assigned to the device by the administrator or use the corresponding **Host Name** for the address.

- If the device uses **GPIB Board** as a communication interface, then do not change the settings for the GPIB board.

The rest of the preset values for the device are probably appropriate. Do not change them. For information about the Configuration and Communication tabs, refer to the document: *Help*.

10. To restore the device preset values, on the Communication tab, click **Set Defaults**.
11. To save the configuration, click **OK**.
12. Repeat step 4 to step 11 for each device.
13. Click **OK** in the Create New Hardware Profile dialog.
14. To activate the hardware profile, do this:
  - a. In the Hardware Configuration Editor, click the hardware profile.
  - b. Click **Activate Profile**.

The check mark turns green. If a red × is shown, then there is an issue with the hardware profile activation. For more information, refer to the section: [Troubleshoot Hardware Profile Activation](#).

---

**Tip!** An active hardware profile does not have to be deactivated before another one is activated. Click an inactive hardware profile and then click **Activate Profile**. The other profile is deactivated automatically.

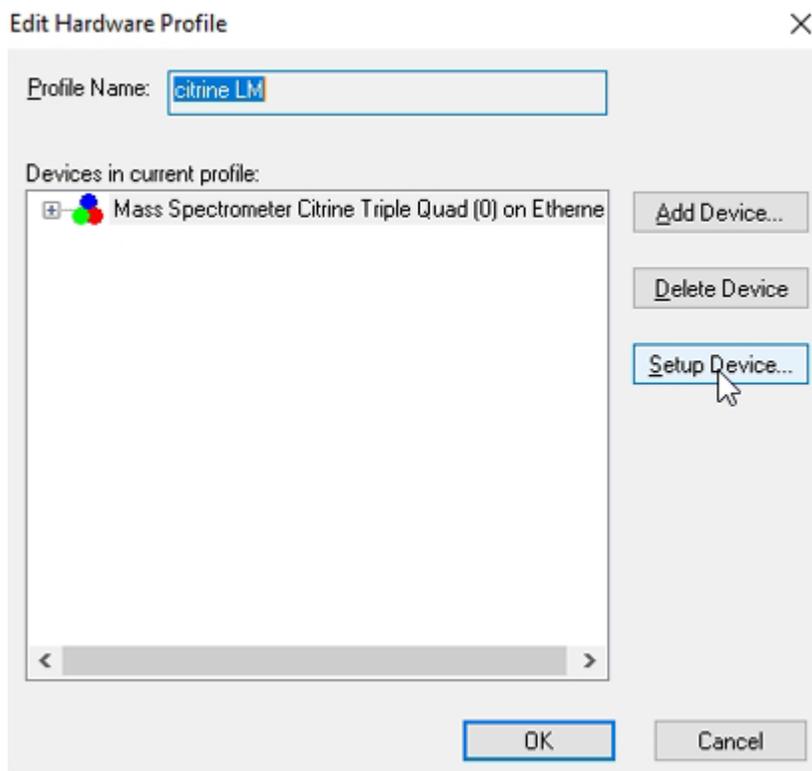
---

15. Click **Close**.

## Edit Devices in a Hardware Profile

1. Open the Hardware Configuration Editor.
2. In the **Hardware Profiles** list, deactivate the hardware profile.
3. Click **Edit Profile**.
4. Select the applicable device and then click **Setup Device**.  
The device configuration dialog opens.

**Figure 8-6 Available Devices Dialog**



5. Edit the configuration and then click **OK**.
6. Click **OK**.
7. Click **Activate Profile**.  
The check mark turns green. If a red x is shown, then there is an issue with the hardware profile activation. For more information, refer to the section: [Troubleshoot Hardware Profile Activation](#).

---

**Tip!** An active hardware profile does not have to be deactivated before another one is activated. Click an inactive hardware profile and then click **Activate Profile**. The other profile is deactivated automatically.

---

8. Click **Close**.

## Troubleshoot Hardware Profile Activation

If a hardware profile fails to become active, then a dialog opens indicating which device in the profile failed to activate. A device might fail to activate because of communications errors.

1. Read the error message generated. Depending on the message, there might be an issue with a device or how the communication is set up.
2. Make sure that the device is connected to the mains supply and is turned on.
3. Make sure that the COM port or IP address assigned to the device is correct.

---

**Tip!** On computers with two built-in serial ports, the first port on the serial port expansion card is usually COM3, even though the cable indicates P1.

---

4. Make sure that the communication settings for the device, for example, the dual inline package (DIP) switch settings, are set correctly and match the settings on the Communication tab.
5. Turn off the device.
6. Wait 10 seconds.
7. Turn on the device.  
Wait until all of the device power-up activities are complete before trying to activate the hardware profile again. Some devices might require 30 seconds or more to complete the power-up activities.
8. Activate the hardware profile.
9. If the issue persists, then delete the failing profile and create a new one.
10. If the issue still persists, then go to [sciex.com/request-support](https://sciex.com/request-support).

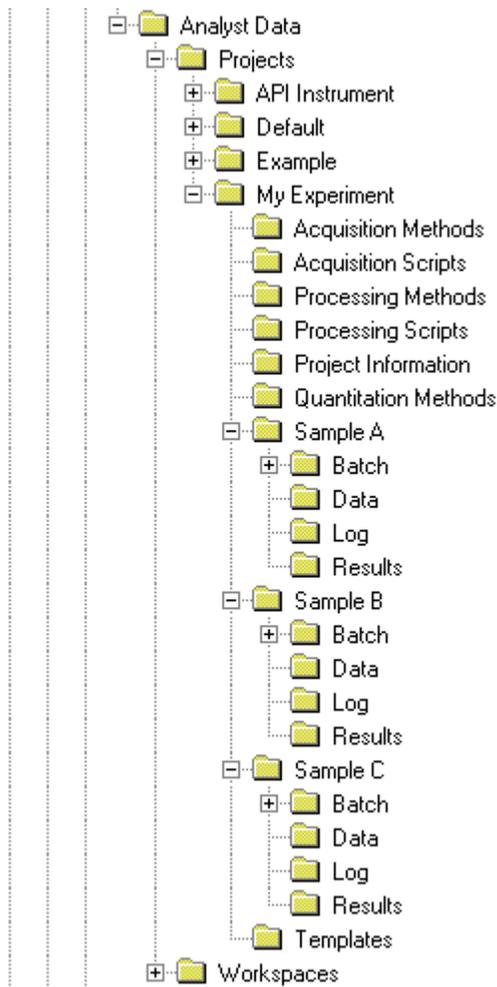
## Projects and Subprojects

Before beginning an experiment, decide where to store the files related to the experiment. Use projects and subprojects for each experiment to better manage data and compare results. For example, use subprojects to store the results for specific dates.

### About Subprojects

A subproject contains a subset of the folders in the project. All subprojects must contain the same folders. Subprojects can be very useful for organizing data. For example, if different laboratories are running samples of various compounds using the same acquisition method, then subprojects can be used to store the results for each laboratory while the acquisition method folder is stored in the project folder. The acquisition method would then be available for use with subproject or laboratory. Alternatively, if samples are run over a period of several weeks, then the results from each day can be stored in a separate subproject.

Figure 8-7 Example of a Project and Subproject Folder Structure

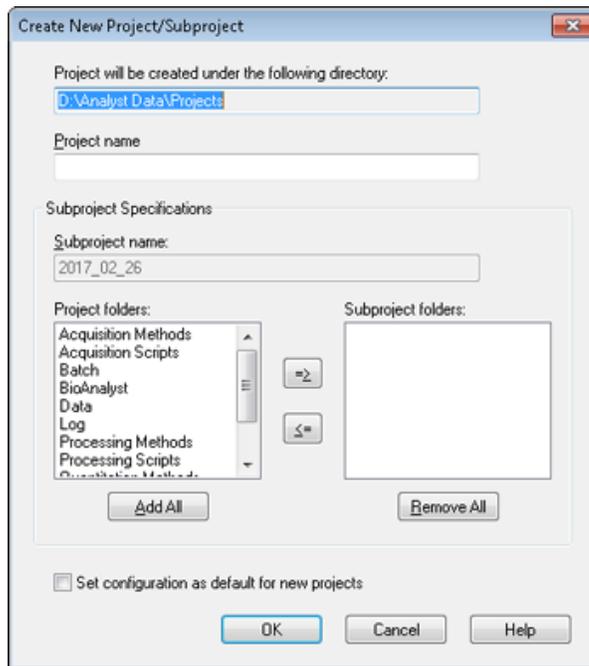


## Create Projects and Subprojects

To use a subproject structure within a project, create the subproject structure when the project is created.

1. Click **Tools > Project > Create Project**.

**Figure 8-8 Create New Project/Subproject Dialog**



---

**Note:** A new subproject cannot be created for a project that was not originally created with a subproject.

---

2. Type a project name in the **Project name** field.
3. (Optional) To use subprojects, do this:
  - a. Select the required folders and then use the arrow buttons to move them to the **Subproject folders** list.
  - b. In the **Subproject name** field, type a name for the first subproject or use the existing date.
4. (Optional) To use this project and subproject folder organization for all new projects, select the **Set configuration as default for new projects** check box.  
All new projects are created with this folder configuration.
5. Click **OK**.

## Create Subprojects

Subprojects can only be created in a project that has an existing subproject structure.

1. On the **Project** tool bar, from the **Project** list, select the project.
2. Click **Tools > Project > Create Subproject**.

3. In the **Subproject name** box, type a name for the subproject or use the existing date.
4. Click **OK**.

### Copy Subprojects

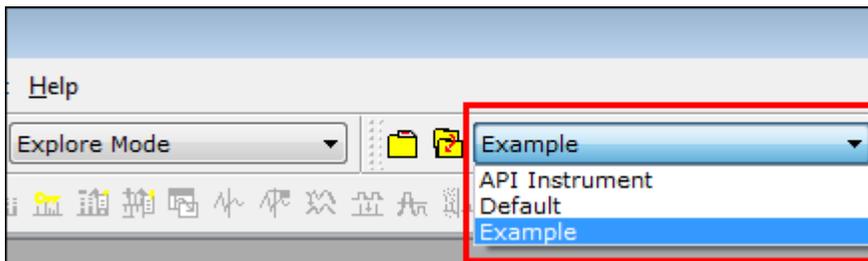
A subproject can be copied from another project that has existing subprojects. If the copied subprojects contain folders that also exist in the destination project folder, then the software uses the project level folders.

1. Click **Tools > Project > Copy Subproject**.  
The Copy Subproject dialog opens.
2. Click **Browse** to browse to the subproject source.
3. Click **OK**.
4. Select the subproject from the **Source Subproject** list.
5. Click **Browse** to browse to the subproject destination.
6. Type the name in the **Target Subproject** field.
7. Click **OK**.
8. Do one of the following:
  - To copy all folders and files from the **Subproject Source** into the **Subproject Destination**, select the **Copy Contents** check box.
  - To copy only the folders in the same structure into the **Subproject Destination**, make sure that the **Copy Contents** check box is cleared.
9. Click **Copy**.

### Change Between Projects and Subprojects

On the software tool bar, from the project list, click the required project or subproject.

**Figure 8-9 Project List**



The project list in this figure shows the **API Instrument**, **Default**, and **Example** folders.

## Installed Project Folders

Three project folders are installed with the software: **API Instrument**, **Default**, and **Example**.

### API Instrument Folder

The API Instrument folder is unique and very important to the correct functioning of the mass spectrometer. The API Instrument folder contains the information required for tuning and calibrating the mass spectrometer. This information includes:

- Parameter settings files
- Reference files
- Instrument data files that contain calibration and resolution information
- Acquisition methods used during automatic tuning

The API Instrument folder also contains data files for manual tuning runs that were performed using the **Start** button rather than the **Acquire** button. These data files are saved automatically in the `API Instrument\Tuning Cache` folder and named with the date and time that they were created. The Tuning Cache folder is automatically cleared periodically.

### Default Folder

The Default folder contains folders that are present in new projects and serves as a template for new projects.

### Example Folder

The Example folder contains sample methods and data files. Users can practice working with the Explore or Quantitate modes using the example data files. The example files are sorted into subfolders by mass spectrometer type and application area.

# Operating Instructions — Tune and Calibrate

# 9

Run the **Verify instrument performance** option weekly or after the mass spectrometer is cleaned to confirm that the system is working properly. In general, for triple quadrupole systems, calibration and resolution is maintained for three to six months unless the system loses vacuum. For QTRAP systems, the resolution should also be maintained for three to six months but the system should be calibrated approximately monthly. If the system loses vacuum, then check the calibration and resolution before using the system. For more information about tuning and calibration, refer to the documents: *Advanced User Guide* and *Manual Tuning Tutorial*.

**Tip!** Perform maintenance tasks regularly to make sure that the mass spectrometer is performing optimally.

## Prerequisites

- The spray is stable and the correct tuning solution is being used.
- A printer is configured.

## Required Materials

- Tuning solutions that are supplied in the Standards Chemical Kit shipped with the system. If required, a new kit can be ordered from SCIEX. Refer to the section: [Calibration Ions and Solutions](#).
- 5 mL, 1 mL, or 250  $\mu$ L serial gas-tight syringes.
- Red PEEK sample tubing.

## About Tuning and Calibrating

Tuning the instrument is the process of optimizing the resolution and instrument parameters to attain the best sensitivity and performance of the mass spectrometer. Optimizing the resolution means adjusting the peak width and peak shape. You can tune and calibrate the instrument either automatically or manually.

**CAUTION: Potential Calibration Error.** If the temperature changes by more than 2 °C, then the resolution and mass calibration might be affected.

**Tip!** Clean the Q0 region regularly to minimize the impact of charging (a significant loss of sensitivity of the ions of interest over a short period of time) on the quadrupoles. Contact the Qualified Maintenance Person (QMP) or FSE.

---

**Automatic tuning:** The software performs resolution optimization and mass calibration, using the Instrument Optimization wizard. For LIT instruments, MS3 optimizations are also performed.

**Manual tuning:** Many of the instrument resolution optimizations and calibrations can be performed manually.

## Back Up the API Instrument Folder

Back up the `API Instrument` folder regularly and after routine maintenance has been performed.

Copy the `API Instrument` folder, paste it to a different location, preferably on another computer, and then rename the folder. Use the date and a mass spectrometer reference when naming the folder, if there is more than one mass spectrometer. For example, `API Instrument_instrument model3_010107`.

## Back up Instrument Parameters

1. On the Navigation bar, under **Tune and Calibrate**, double-click **Instrument Optimization**.
2. Click **File > Backup Instrument Settings Files** in the Instrument Optimization dialog.
3. Type a file name and then click **Save**.
4. Click **Exit**.

## Restore Instrument Parameters

1. On the Navigation bar, under **Tune and Calibrate**, double-click **Instrument Optimization**.
2. Click **File > Restore Instrument Settings Files** in the Instrument Optimization dialog.
3. Navigate to the instrument settings that you want to restore and then click **Open**.
4. Click **Exit**.

## Automatically Tune and Calibrate

Instrument Optimization is automatic instrument tuning software that tunes both quadrupole and LIT modes and performs mass calibration. For quadrupole mode, it adjusts the resolution offsets. For LIT mode, it optimizes AF3 and EXB. For MS3, it adjusts the Excitation and Isolation coefficients. Select one of the instrument performance options:

- **Verify instrument performance:** Tests the instrument performance but leaves the instrument settings unchanged. A report is generated at the end of the test. You can use this option weekly to check how well the instrument is performing.
- **Adjust mass calibration only:** Automatically checks and adjusts the mass calibration. If the mass calibration has changed, then the software corrects it. This option can be used weekly for LIT instruments, or monthly to check and adjust the mass calibration if required.
- **Adjust instrument settings:** Checks and adjusts the instrument settings and mass calibration. The instrument settings are updated from the current settings to optimal settings. Use this option if the instrument performance is poor or if the peak shape is bad. Only experienced users should adjust the instrument settings.

---

**Note:** Old LIT methods must be updated with the new settings. Toggle the LIT speed in the advanced MS tab and then save the method.

---

- **Reset selected scan modes to default values and adjust instrument settings:** Resets the instrument values to the factory preset values. Select this option if a major component of the instrument is replaced or after the first installation. Only FSEs should use this feature.

Back up the current instrument parameters in case they must be restored later.

The preset location for the instrument parameters is <drive>:\Analyst Data\Projects\API Instrument\Instrument Optimization\Instrument Settings Backups\User Created Backups.

## Verify Instrument Performance

Use this procedure to verify or adjust the performance of the mass spectrometer. For information about using the other instrument performance options, refer to the document: *Help*.

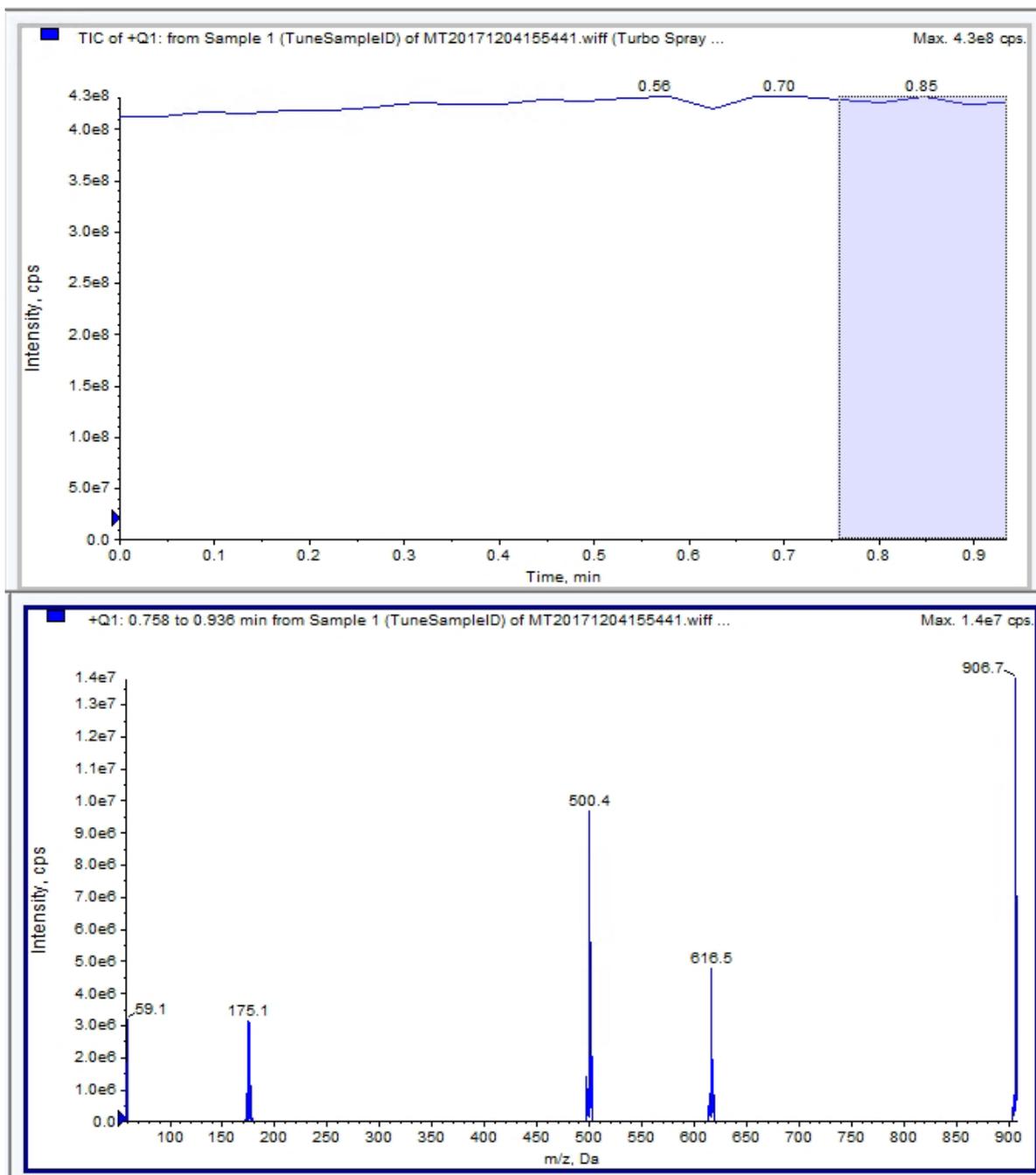
Prerequisites
<ul style="list-style-type: none"><li>• The syringe pump is activated in the hardware profile. If the syringe pump is not activated, then edit the hardware profile. Refer to the section: <a href="#">Add Devices to a Hardware Profile</a>.</li><li>• The API Instrument folder is selected.</li></ul>



1. On the Navigation bar, under **Tune and Calibrate**, double-click **Manual Tuning**.

2. Start the syringe pump, type **5** in the **Duration** field, and then run a calibration method. Confirm that the total ion chromatogram (TIC) is stable and that the peaks of interest are present in the spectrum.

**Figure 9-1 Example of a Stable TIC and Peaks of Interest**



## Operating Instructions — Tune and Calibrate

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3. On the Navigation bar, under **Tune and Calibrate**, double-click **Instrument Optimization**. The Instrument Optimization dialog opens.
4. Click **Verify instrument performance**.
5. Click **Next**.
6. Click **Approved Tuning**.
7. Click **Next**.
8. Select a **Tuning Solution** from the list.  
Depending on the solution selected, different modes are available:
  - a. Click a polarity.
  - b. (If available) Click **Q1** and **Q3** in the Quad section.
  - c. (If available) Click the required scan speeds.
  - d. (If available) Click the scan speeds in the LIT section.
  - e. (If available) Click **Excitation** in the MS/MS/MS section
9. Click **Next**.
10. If the Select a mode page opens, then select **Automatic**.
11. Click **Next**.
12. Click **GO**.  
The Verifying or Adjusting Performance dialog opens. After the process has completed, the Results Summary opens. For more information, refer to the document: *Help*.
13. (If applicable, depending on the options selected) When prompted, change solutions for different scan types and polarities.

## Verifying or Adjusting Performance Dialog

The top left corner shows the part of the instrument that is being tuned.

The Current Spectrum graph shows the spectrum of the current scan, the optimal scan selected by the software, or the scan at the current parameter value when the software results are viewed in interactive mode.

The Instrument Optimization Decision Plots, in the top right graph, dynamically show the intensity-to-voltage curves of the parameters that are currently being optimized.

## Results Summary

The Results Summary is a record of any instrument settings changes that were made by the Instrument Optimization wizard.

The Results Summary includes the location of the data and instrument settings backup files, as well as a step-by-step record of the changes and results during optimization.

The Results Summary also includes a verification report. This report contains a snapshot of the mass spectrum for each relevant mass for the scan modes being verified. The spectrum is labeled with the target mass, where the mass was found, the mass shift, the peak width, and the peak intensity. The spectrum can be used as a visual record of peak shape or scan mode performance. A summary table of results follows the spectra.

The Results Summary is automatically saved in the following path: `<drive>:\Analyst Data\Projects\API Instrument\Data\Instrument Optimization\yyyy-mm-dd\results.pdf`, where `yyyy-mm-dd` is the date on which the report was created. Users can print the Results Summary or open a previously saved Results Summary.

## Recover the API Instrument Folder

Back up the `API Instrument` folder regularly and after routine maintenance has been performed.

1. Rename the current `API Instrument` folder.
2. Copy the backup folder into the `Projects` folder.
3. Change the name of the backup folder to `API Instrument`.

# Operating Instructions — Automatic Optimization

# 10

To tune instrument parameters for particular compounds the following steps are recommended. The mixture of four compounds is used for illustrating the steps of the procedure. The user will have to apply the same procedure while using the appropriate compounds for the assay of interest.

This section describes how to:

- Automatically optimize for the analyte using the Compound Optimization wizard.
- Choose between infusion and flow injection analysis (FIA).
- Optimize parameters:
  - If infusion analysis is chosen, then use an infusion method to optimize compound-dependent parameters.
  - If FIA analysis is chosen, then use FIA to optimize compound- and ion source-dependent parameters.

This procedure uses minoxidil, tolbutamide, reserpine, and rescinnamine as example compounds. Other available compounds can be used, but the methods must be adjusted accordingly.

The user can also optimize compounds manually. Refer to the document: *System User Guide* for the mass spectrometer.

## Prerequisites

- The mass spectrometer is tuned and calibrated.
- (For FIA analysis) An acquisition method template is available.
- (For FIA analysis) An LC pump and an autosampler are connected and configured in the hardware profile.
- All of the required peripheral devices, including the LC components, if required, are configured in the hardware profile.

### Required Materials

- A syringe, preferably a 1.0 mL syringe.
- (For FIA analysis) Mobile phase: 1:1 acetonitrile:water + 2 mM ammonium acetate + 0.1% formic acid.

---

**Note:** Users can choose a different mobile phase based on the experimental properties of the compound.

---

- LC pump and autosampler.
- (For FIA Analysis) Autosampler vials.

## About Automatic Optimization

Automatic optimization first checks for the presence of the compounds. The voltages of the various ion path parameters are gradually increased or decreased to determine the maximum signal intensity of the Q1 scan for each ion. A text file is generated and shown during the optimization process. This file records the experiments performed and the optimum value for each parameter. A file folder containing all of the experiments performed is also generated and can be found by opening the data file folder in Explore mode. For each experiment performed, an acquisition method is also generated and saved in the Acquisition Method folder.

During the optimization process, select how the precursor ion and the corresponding product ions are to be chosen.

## Types of Sample Introduction

### Infusion

Infusion is the continuous flow of the sample at low flow rates into the ion source using a syringe pump. During the infusion optimization process, the software can select precursor and product ions and optimize for declustering potential, collision energy, and collision cell exit potential. The voltages of the ion path parameters are gradually increased or decreased to get the maximum signal intensity for the precursor and product ions.

Use infusion optimization to optimize compound-dependent parameters only, at much lower flow rates than those used during LC-MS/MS analysis.

### FIA

FIA is the injection of a sample by the autosampler into the mass spectrometer using an LC system. During the FIA optimization process, multiple sample injections are performed

## Operating Instructions — Automatic Optimization

---

for various ion source- or compound-dependent parameters that are changed between injections. FIA compound optimization optimizes parameters by performing looped experiments in succession. One compound-dependent parameter is optimized first, followed by the next compound-dependent parameter. FIA optimizes for ion source-dependent parameters by making one injection for each value.

Compound parameters must be narrowed using at least two more FIA cycles.

Use FIA optimization to optimize both compound-dependent and source-dependent parameters using an LC system at higher flow rates.

**Table 10-1 Differences Between Sample Introduction Methods**

Method	Required Devices	Parameters	Typical Flow Rate Range
Infusion	Syringe pump	Compound-dependent	5 $\mu\text{L}/\text{min}$ to 25 $\mu\text{L}/\text{min}$
FIA	LC pump and autosampler	Ion source- and compound-dependent	25 $\mu\text{L}/\text{min}$ to 1000 $\mu\text{L}/\text{min}$

During optimization, a text file is generated and then shown. This file records the experiments performed and the optimal values for each parameter. A file folder containing all the data files is also generated. For each experiment performed, an acquisition method is also generated and saved in the `Acquisition Methods` folder.

## Automatically Optimize for an Analyte Using Infusion

Use this procedure to perform automatic MS/MS optimization using infusion with a known precursor ion and an unknown product ion.

### Confirm the Presence of Compounds

Confirm the presence of compounds of interest before continuing with automatic optimization.

1. In the Analyst MD software, create a project.
2. Activate the hardware profile.
3. Prepare the sample:
  - a. Aspirate the compound solution into a syringe and remove the air from the syringe.
  - b. Use the tubing with the special fitting to connect the syringe to the mass spectrometer.
  - c. Install the syringe in the integrated syringe pump.

4. Infuse the compound in solution at a rate of 5  $\mu\text{L}/\text{min}$  to 10  $\mu\text{L}/\text{min}$ .
5. On the Navigation bar, under **Tune and Calibrate**, double-click **Manual Tuning**.
6. In the method list field, click **Syringe Pump Method**.
7. On the Syringe Pump Method Properties tab, type the appropriate parameter values. Refer to the table: [Table 10-2](#).

**Table 10-2 Syringe Pump Method Properties Tab**

Parameter	Typical Value
<b>Syringe Diameter</b>	Syringe dependent; 1.0 mL syringe is 4.610 mm
<b>Flow Rate</b>	5 to 10
<b>Unit</b>	$\mu\text{L}/\text{min}$

**Figure 10-1 Syringe Pump Method Properties Tab**

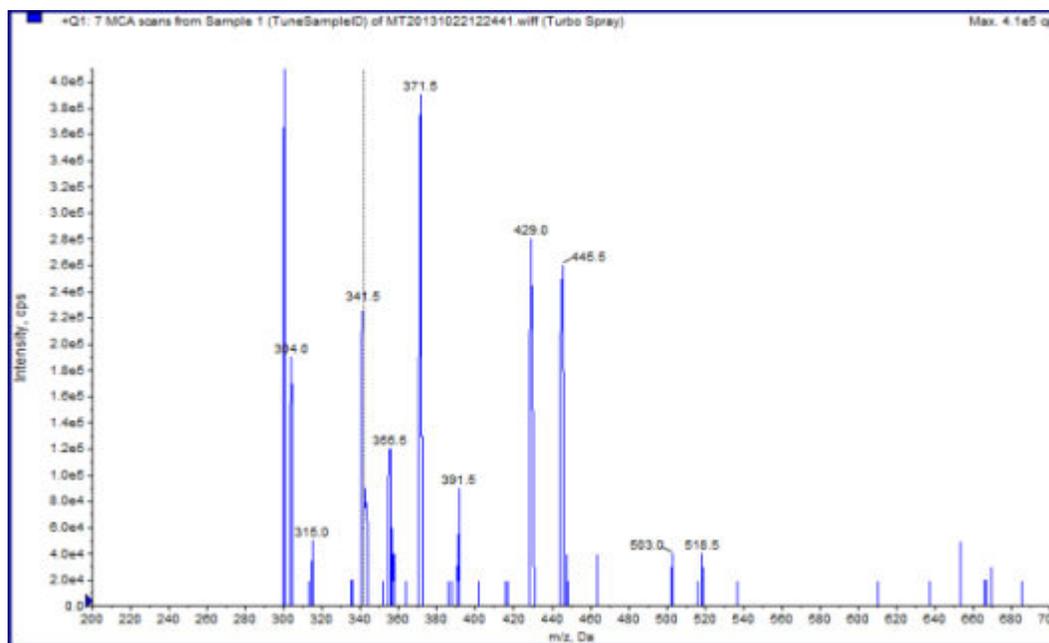
8. Click **Start Syringe Pump**.
9. Click **MS Method** in the method list.
10. Click **Start**.
11. Wait until an even TIC is shown on the left and peaks are shown on the right. Then click **Stop**.
12. Select the **MCA** check box.
13. Type 10 In the **Cycles** field.
14. Click **Start**.

When the ten scans are complete, the graph shows the masses of the four compounds as ions.

**Note:** The intensities of the compounds should be much higher than the smallest noise peaks but not so high that no noise peaks are not seen. In the first case, the peak might not be a real compound. In the second case, the concentration might be too high for the software to optimize properly.

---

**Figure 10-2 Compound Ions**



## Perform Automatic MS and MS/MS Optimization Using Infusion with a Known Precursor Ion and an Unknown Product Ion

Automatic optimization for MS/MS analysis optimizes certain compound-dependent parameters for one or more MRM transitions. The software finds the ion of interest and then optimizes the compound-dependent parameters to get the maximum sensitivity for the compound. The software ramps CE and then selects the most intense fragments that meet all of the product ion selection criteria.

If the initial Q1 scan signal is too high, then the software tries to reduce the CEM to keep ions within the detector range. If the signal is still too high after the CEM is reduced, then the process stops and an error message is shown. Dilute the solution and then restart the optimization. Make sure to purge the infusion line.

The parameters from the last quantitative optimization are stored.

1. Make sure that the proper concentration of solution has been installed in the syringe pump, and that the syringe pump has been started.  
The syringe pump must be started in the Manual Tune window before compound optimization begins.  
If an integrated syringe pump is available and has been started, then the Syringe Pump Status LED flashes.
2. On the Navigation bar, under **Tune and Calibrate**, double-click **Compound Optimization**. The Instrument Settings page opens.
3. In the Inlet section, click **Infusion**.
4. Click **MS/MS Analysis** in the Mass Spectrometer section.
5. Click **Next**.  
The Ions to use in MS/MS Analysis page opens.
6. Select the appropriate parameter values. Refer to the table: [Table 10-3](#).

**Table 10-3 Example Parameters to use in MS/MS Analysis Page**

Parameter	Value
<b>MW Ion: Search Window</b>	2.500
<b>Resolution</b>	Unit
<b>Polarity</b>	Positive or Negative, depending on the properties of the targeted compound
<b>Product Ion</b>	Auto Select
<b>Resolution</b>	Unit

---

**Note:** The optimization algorithm looks for the most intense peak in the specified search window. If the most intense peak in that window is not the mass of interest, then the software optimizes for the wrong ion.

---

7. Click **Criteria** next to the **Auto Select** option.  
The Product Ion Auto Selection Criteria dialog opens.
8. Type the appropriate parameter values. Refer to the table: [Table 10-4](#).

**Table 10-4 Example Product Ion Auto Selection Criteria Dialog Parameters**

Parameter	Value	Description
<b>From the Most Intense (peaks)</b>	3	The number of fragment peaks to be optimized. The algorithm generates a product ion scan spectrum while ramping the CE in MCA mode. In this example, the algorithm takes the three most intense fragment ions from the spectrum and then continues the MS/MS optimization on those fragments only.  For unknown compounds, select more peaks.
<b>Build final method using (most intense peaks)</b>	2	The number of fragment ions per precursor ion (target compound) to be automatically included in the acquisition method. The specified number defines the number of MRM transitions to be included for each target compound in the method. The order of preference is based on the intensity of the fragment ion.  Two is a better starting value than one because typically two product ions are required for quantitation. Start with three in case there is an issue with one of the two best. Go back and the third is already identified.  For unknown compounds, select more peaks for use in case of interference.
<b>Exclude Product Ions within <math>\pm</math> (Da of Precursor Ion <math>m/z</math>)</b>	20.000	The Da value that defines the exclusion window around the precursor ion. Fragment ions that fall within this window are not selected for MRM optimization. For example, if the user types $\pm 5$ Da for a precursor ion that has an $m/z$ value of 500, then any fragment ions within an $m/z$ value of 495 to 505 are excluded. This prevents the precursor ion from being optimized as a product ion.
<b>Min. Mass for Product Ion (Da)</b>	60.000	The lowest fragment mass to be considered for optimization. Use this option to increase or decrease the size of the window that contains fragment ions to be considered from the precursor mass.
<b>Threshold for Product Ion (cps)</b>	100.000	Minimum number of counts for a product ion to be considered.

9. Click **OK** to save the changes to the selection criteria.
10. Click **Next**.  
The Target Components dialog opens.
11. Type the appropriate parameter values. Refer to the table: [Table 10-5](#).

---

**Note:** The compound name must be unique for each compound or transition.

---

**Table 10-5 Example Target Compounds Dialog Parameters**

Target Compound	Field	Value
Reserpine	Compound Name	Reserpine
	MW (Da) <sup>2</sup>	609.3
	No. Charges	1
Minoxidil	Compound Name	Minoxidil
	MW (Da) <sup>2</sup>	210.2
	No. Charges	1
Tolbutamide	Compound Name	Tolbutamide
	MW (Da) <sup>2</sup>	271.1
	No. Charges	1
Rescinnamine (IS)	Compound Name	Rescinnamine
	MW (Da) <sup>2</sup>	635.3
	No. Charges	1

12. Click **Finish** to start the optimization process.

Two windows are shown, a text file window and an acquisition window. The user might need to minimize one of them to see the other. The experiment being run is shown on the top of the acquisition window. The X-axis shows the parameter that is being optimized for each experiment. The text file window is updated as results are generated.

After optimization is complete, an MRM acquisition file is created and named *<compound>\_QOpt\_FinalMRM\_Pos.dam*, where *<compound>* is the first compound in the Target Components page.

---

<sup>2</sup> Type the exact ion mass.

### Review the Optimization Results

When optimization completes, the optimized parameters are saved in an acquisition method. All dam and wiff files generated during the optimization process are saved in the `Acquisition Methods` folder and in a subfolder in the `Data` folder, respectively, in the project. The name of the subfolder is generated using the name of the compound and the date.

1. After completing the optimization, print the text file containing the optimized parameters for each compound.
2. Click **File > Open** and then select the `Reserpine_QOpt_FinalMRM.POS.dam` file.
3. Compare the values in the text file to those in the dam file.
4. Inspect the contents of the following folders:
  - **Data:** Review all of the runs executed during optimization. Compare a wiff file with the optimized value in the method or printed parameters.
  - **Acquisition Method:** `Reserpine_QOpt_FinalMRM.POS.dam` file and other dam files created during optimization.
  - **Log:** Report file (rtf) shown during the optimization process.

### Automatically Optimize for an Analyte Using FIA

Prerequisites
<ul style="list-style-type: none"><li>• Identify the ions for the compounds and save the basic acquisition method.</li><li>• Add an autosampler and LC pump to the basic acquisition method. Using FIA for optimization requires that these devices be active in the hardware profile.</li><li>• Create an LC-MS/MS acquisition method based on the <code>Reserpine_QOpt_FinalMRM.POS.dam</code> file and then name the new method.</li></ul>



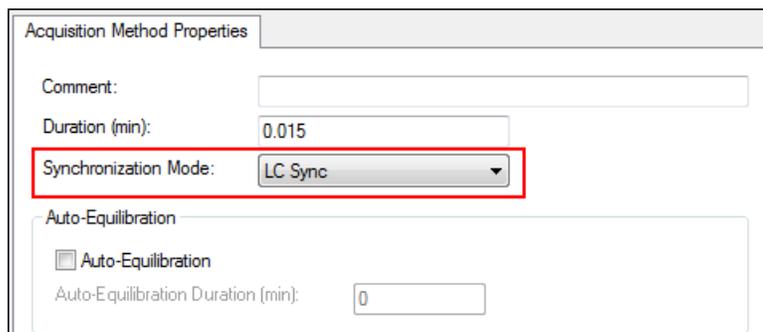
**Note:** Although FIA can be used to optimize compound-dependent parameters, this is typically not done because of the number of cycles required to obtain the optimal parameter values.

---

1. Put a dilution of the four-compound mix in an autosampler.  
Sufficient sample is required to review each variable of each parameter and have sample left over. For example, for the temperature to run at 300 °C, 400 °C, and 500 °C, if the injection volume is 10 µL, then more than a 30 µL (3 × 10 µL injection) is required.
2. Make sure that **LC Sync** is selected in the method.

**Note:** In LC Sync mode, the mass spectrometer coordinates with the action of the LC system to make sure that data is acquired properly.

**Figure 10-3 Acquisition Method with LC Sync Selected**



3. Make sure that the ion source and gas parameters are set to reasonable levels to prevent the contamination of the mass spectrometer while optimizing. Refer to the section: [Ion Source Optimization](#).
4. Set the horizontal micrometer to 5.
5. Set the vertical micrometer on the ion source for the flow rate. As a starting point, use the parameters in the following table.

**Table 10-6 Ion Source Vertical Parameters**

Flow rate	Initial Vertical Parameters
1 $\mu$ L/min to 20 $\mu$ L/min	10 mm
20 $\mu$ L/min to 250 $\mu$ L/min	5 mm
250 $\mu$ L/min to 500 $\mu$ L/min	2 mm
500 + $\mu$ L/min	0 mm

6. Set the values for the LC system and use an autosampler injection volume of 10  $\mu$ L, for example. Use the same concentration for the infusion experiment, or a lower concentration.

The LC pumps must be configured for an isocratic run with no column. The MS and LC times must be the same to collect the proper data.

The flow rate and percent of mobile phases used should be based on the LC column used, the general chromatography, and the approximate mobile phase concentration at which the compounds of interest elute.

7. On the Navigation bar, under **Tune and Calibrate**, double-click **Compound Optimization**. The Instrument Settings page opens.

## Operating Instructions — Automatic Optimization

---

8. Depending on the LC system being used, type the appropriate parameter values. Refer to the table: [Table 10-7](#).

**Table 10-7 Example Instrument Settings Parameters**

Parameter	Value
Inlet	FIA
Rack Code	Autosampler specific
Rack Position	Autosampler specific
Injection Volume	10 µL (example volume)
Mass Spectrometer	MS/MS Analysis

9. Select the appropriate default acquisition method.
10. Click **Next**.
11. Make sure that the **Int. Std.** check box is cleared.  
Selecting the check box indicates which MRM corresponds to the internal standards.  
Internal standards are not optimized during the optimization process.
12. In the Resolution group, select **Unit** in both the **Q1 Resolution** and **Q3 Resolution** fields.

Figure 10-4 Q1 and Q3 Resolution Fields

FIA Target Compounds

These target compounds will be optimized. You may change the compound name. Please specify which one of them are used as internal standards.

	Compound Name	Q1 Mass (Da)	Q3 Mass (Da)	Int. Std.	Vial Pos.
1	Compound 609.300-195.00	609.300	195.000	<input type="checkbox"/>	1
2	Compound 210.200-164.20	210.200	164.200	<input type="checkbox"/>	1
3	Compound 271.100-91.100	271.100	91.100	<input type="checkbox"/>	1
4	Compound 635.300-221.20	635.300	221.200	<input type="checkbox"/>	1

Note: All compounds identified as I.S. (internal standard) will not be used to determine optimum Source / Gas Parameter conditions.

Resolution

Q1 Resolution:

Q3 Resolution:

< Back   Next >   Cancel   Help

13. Click **Next**.
14. On the FIA Source Parameters page, type numbers that are lower or higher than the original value, but within specifications.

**Note:** To keep the system clean, make sure that the values are not too low. For parameters that can be used as a starting point, refer to the table: [Table 10-8](#).

**Tip!** Type the values before selecting the check box.

Table 10-8 Example Parameters for FIA Source Parameters Page

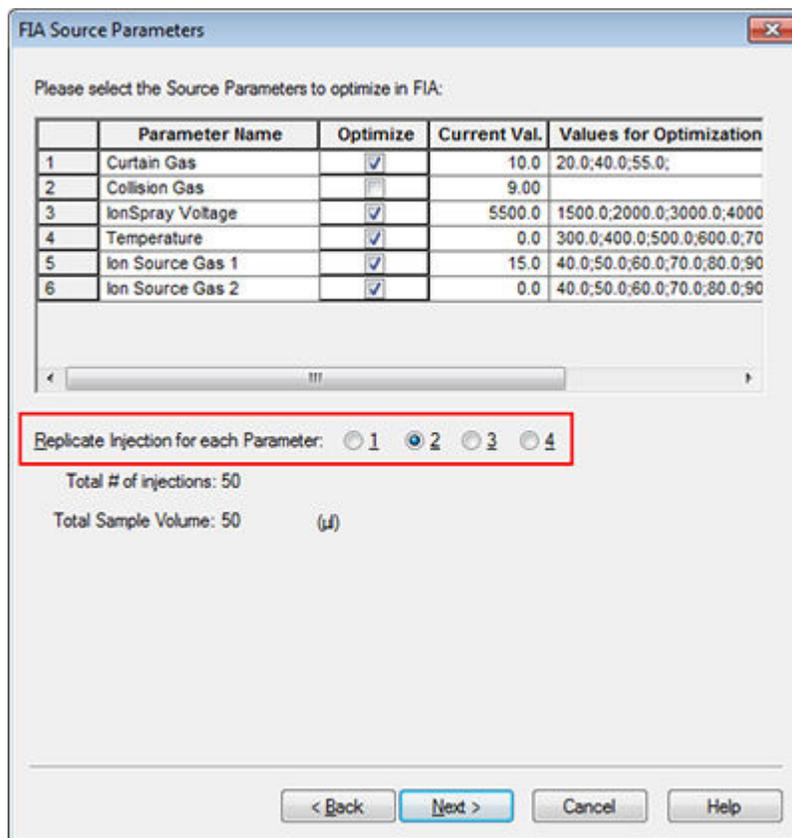
Parameter	Select the Optimize Check Box?	Values for Optimization
Curtain Gas	Yes	20;40;55
Collision Gas	No	—
IonSpray Voltage	Yes	1500;2000;3000;4000;5000

Table 10-8 Example Parameters for FIA Source Parameters Page (continued)

Parameter	Select the Optimize Check Box?	Values for Optimization
Temperature	Yes	300;400;500;600;700
Ion Source Gas 1	Yes	40;50;60;70;80;90
Ion Source Gas 2	Yes	40;50;60;70;80;90

15. Select **1** or **2** beside **Replicate Injection for each Parameter**.  
The total number of injections and the total sample volume are calculated based on the specifications on this dialog. Note the total sample volume needed. The sample volume increases with the number of variables for each parameter that are being optimized, because each variable is a separate method.

Figure 10-5 Example Replicate Injection for each Parameter Field



16. Click **Next**.
17. On the FIA Compound Parameters page for each analyte, use the example parameter values provided as a starting point. Refer to the table: [Table 10-9](#).

**Note:** The values in the following table are suggested values. For more information, refer to the document: *Help*.

---

**Table 10-9 Example FIA Compound Parameters Page**

Parameter	Select the Optimize Check Box?	Values for Optimization
<b>Declustering Potential</b>	Yes	60;80;100;120;200
<b>Entrance Potential</b>	No	—
<b>Collision Energy</b>	Yes	20;30;40;50;70;80;100
<b>Collision Cell Exit Potential</b>	Yes	2;4;6;8;10;12

The total number of injections and the dependent sample volume update automatically. In contrast to ion source parameters, which require one injection per value per replicate, compound-dependent parameters only require one injection per parameter. A looped experiment is performed for each parameter. The values are alternated scan-by-scan within one injection.

---

**Note:** Do not specify too many values. Doing so will prevent proper evaluation of the parameter.

---

18. Using the Compound list, move to another compound and type the parameters to be optimized.
  19. Repeat step 18 until all of the parameters for all of the compounds are provided.
  20. Type the duration of the optimization in the **Mass Spec. Duration** field.
- 

**Note:** This value should be at least the required length of time for each injection.

---

Figure 10-6 Example Mass Spec. Duration Field

FIA Compound Parameters

Please select the Compound Parameters to optimize by FIA:

Compound:  MRM: 609.300 - 195.000

	Parameter Name	Optimize	Current Val.	Values for Optimization
1	Declustering Potential	<input checked="" type="checkbox"/>	120.0	60.0;80.0;100.0;120.0;200.
2	Entrance Potential	<input type="checkbox"/>	10.0	
3	Collision Energy	<input checked="" type="checkbox"/>	30.0	20.0;30.0;40.0;50.0;70.0;80
4	Collision Cell Exit Potential	<input checked="" type="checkbox"/>	15.0	2.0;4.0;6.0;8.0;10.0;12.0;

Total # of Injections: 53  
Total Sample Volume: 53 (µl)  
**Mass Spec. Duration: 1.5 (min)**

< Back Finish Cancel Help

21. Click **Finish** to begin the optimization process.

The software optimizes the specified ion source and compound-dependent parameters to get the maximum sensitivity for the MRM transition of the compound. As the software proceeds through the optimization, it creates a **Compound Optimization** report.

22. To get optimized parameters, repeat this routine.

---

**Note:** Typically, the ion source and gas parameters must be narrowed using one more FIA cycle.

---

23. Open the final optimized FIA method called \*\_FIA\_sample\_1.

---

**Note:** The software generates several acquisition methods.

---

24. Save this method using a simpler name.

An acquisition method consists of experiments and periods. Use the Acquisition Method Editor to create a sequence of periods and experiments for the mass spectrometer and any devices in the active hardware profile.

We recommend that only users who are proficient in method development create or modify acquisition and quantitation methods. For more information about roles and security, refer to the document: *Laboratory Director Guide*.

## Create an Acquisition Method Using the Acquisition Method Editor

---

**Tip!** If users are creating a new acquisition method file from an existing file, then some or all of the peripheral device methods in the acquisition method might be used.

---

Only devices configured in the active hardware profile are shown in the Acquisition method pane. Any devices added to the hardware profile must also be added to existing acquisition methods. For more information about devices, refer to the document: *Peripheral Devices Setup Guide*.

1. Make sure that a hardware profile containing the mass spectrometer and peripheral devices is active.
2. On the Navigation bar, under **Acquire**, double-click **Build Acquisition Method**.
3. Select a **Synchronization Mode** on the Acquisition Method Properties tab.
4. (Optional) Select the **Auto-Equilibration** check box and then type the required equilibration time, in minutes.
5. In the Acquisition Method pane, click the **Mass Spec** icon.
6. On the MS tab, select a **Scan type**.
7. Type values in the other fields as required.
8. On the Advanced MS tab, type values in the fields as required.
9. On the MS tab, click **Edit Parameters**.
10. On the Source/Gas tab, specify values in the fields as required.

## Operating Instructions — Acquisition Methods

---

11. On the Compound tab, specify values in the fields as required.
12. Click **OK**.
13. Click a device icon and then set the parameters for the device.
14. Add any additional periods and experiments. Refer to the sections: [Add an Experiment](#) and [Add a Period](#).
15. Click **File > Save**.

## About LC Methods

Creating an acquisition method using a peripheral device, such as an LC system, includes providing the operating parameters for that device. If a new acquisition method is created from an existing file, some or all of the peripheral device methods can be used in the acquisition method.

## Create Mass Spectrometry Methods

Use the Acquisition Method Editor to create a mass spectrometer (MS) acquisition method. Depending on the type of mass spectrometer configured and the scan type selected, different fields and options are available. The Acquisition Method Editor validates the settings as the parameters are typed.

Create one of the following methods and then use it to acquire data. Refer to the section: [Create and Submit a Batch](#)

- [Create an Acquisition Method using a Q1 MS Scan Type](#)
- [Create an Acquisition Method using a Q1 MI Scan Type](#)
- [Create an Acquisition Method using an MRM Scan Type](#)

### Create an Acquisition Method using a Q1 MS Scan Type

Use the following procedure to create a method using the Q1 MS scan. The ion intensity is returned for every requested mass in the scan range.

Prerequisites
<ul style="list-style-type: none"><li>• Make sure that a hardware profile containing the mass spectrometer and syringe pump is active.</li><li>• On the software toolbar, make sure that the appropriate project is selected.</li></ul>



1. On the Navigation bar, under **Acquire**, double-click **Build Acquisition Method**.

The Acquisition Method Editor is shown with a method template based on the active hardware profile.

2. In the Acquisition Method pane, click **Acquisition Method**.
3. On the Acquisition Method Properties tab, in the **Synchronization Mode** list, make sure that **No Sync** is selected. For more information about synchronization modes, refer to the document: *Help*.
4. In the Acquisition method pane, click the **Mass Spec** icon.
5. On the MS tab, in the **Scan type** list, select **Q1 MS (Q1)**.
6. In the Polarity section, click **Positive**.
7. Clear **Center/Width** and **Parameter Range** check boxes if selected.
8. In the mass ranges table, type the values that are shown in the following table.

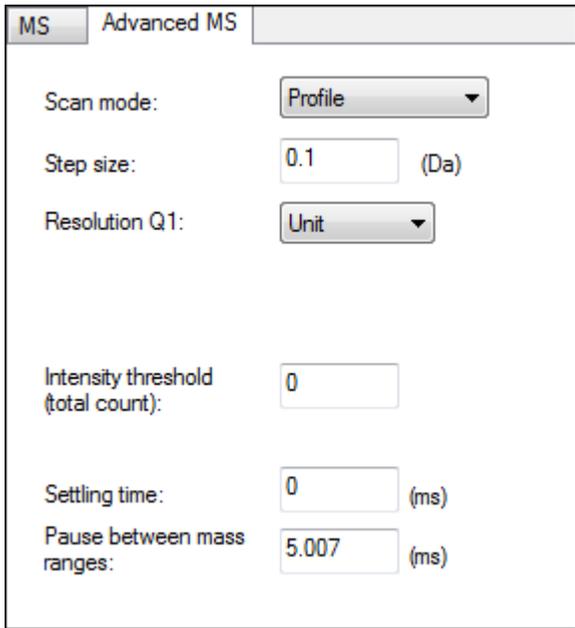
**Figure 11-1 MS Tab Parameter Values**

**Table 11-1 MS Tab Parameter Values**

Field	Example Value
Start (Da)	200
Stop (Da)	700
Time (sec)	2.5
Scan rate (Da/s)	200
Duration (min)	3

- On the Advanced MS tab, note that the **Scan mode** is set to **Profile** and the **Step size** is 0.1.

**Figure 11-2 Advanced MS tab**



MS	Advanced MS
Scan mode:	Profile
Step size:	0.1 (Da)
Resolution Q1:	Unit
Intensity threshold (total count):	0
Settling time:	0 (ms)
Pause between mass ranges:	5.007 (ms)

In this example, the quadrupole (Q1) is scanning a 500 Da range in 0.1 Da steps. Therefore, there are 5000 steps across the mass range. If the scan takes 2.5 seconds, then the dwell time is 0.5 ms per step. This is usually the fastest that Q1 or Q3 should be scanned in a standard calibration procedure. Proper consideration for mass calibration should be taken if Q1 or Q3 are to be scanned faster.

---

**Note:** The step size and the time of the scan control the dwell time per step for the scan. The dwell time is the length of time spent acquiring signal at each step in a scan.

---

- On the MS tab, click **Edit Parameters**. The Parameter Table dialog opens.
- On the Source/Gas tab, type the following values:

**Table 11-2 Source/Gas Tab Parameters**

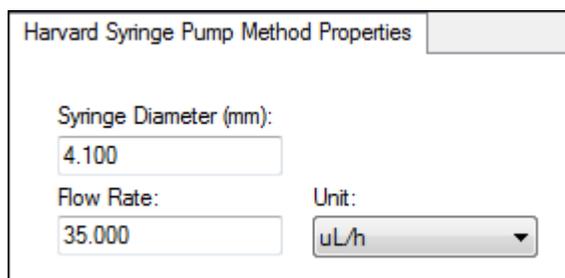
Source/Gas parameters	Typical value
Curtain Gas (CUR)	35
IonSpray Voltage (IS)	5000
Temperature (TEM)	0

**Table 11-2 Source/Gas Tab Parameters (continued)**

Source/Gas parameters	Typical value
Ion Source Gas 1 (GS1)	20
Ion Source Gas 2 (GS2)	0

12. Click the Compound tab and then set the **Declustering Potential (DP)** to 90 and leave the **Entrance Potential (EP)** at 10.  
A value of 90 might not be optimal for the mass spectrometer but it is a good DP to start with.
13. Click **OK**.
14. In the Acquisition method pane, click the **Harvard Syringe Pump** or **Integrated Syringe Pump** icon.

**Figure 11-3 Harvard Syringe Pump Method Properties Tab**



15. Edit the syringe pump method to include **Syringe Diameter**, **Flow Rate**, and **Unit**.
16. Save the acquisition method.  
Next steps: Use the acquisition method to acquire data for preliminary analysis. To create and submit batches, refer to the section: [Create and Submit a Batch](#).

## Create an Acquisition Method using a Q1 MI Scan Type

Use the following procedure to create a method using the Q1 MI scan. The ion intensity is returned for only the specified masses.

Prerequisites
<ul style="list-style-type: none"> <li>• Make sure that a hardware profile containing the mass spectrometer and syringe pump is active.</li> <li>• On the software toolbar, make sure that the appropriate project is selected.</li> </ul>

1. On the Navigation bar, under **Acquire**, double-click **Build Acquisition Method**.  
The Method Editor is shown with a method template based on the active hardware profile.

## Operating Instructions — Acquisition Methods

2. In the Acquisition method pane, click **Acquisition Method**.
3. On the Acquisition Method Properties tab, in the **Synchronization Mode** list, make sure that **No Sync** is selected. For more information about synchronization modes, refer to the document: *Help*.
4. In the Acquisition method pane, click the **Mass Spec** icon.
5. On the MS tab, in the **Scan type** list, select **Q1 Multiple Ions (Q1 MI)**.
6. In the Polarity section, click **Positive**.
7. In the mass ranges table, type the values that are shown in the following table.

**Figure 11-4 MS Tab Parameter Values**

The screenshot shows the 'MS Advanced MS' configuration window. Key parameters are as follows:

- Experiment: 1
- Scan type: Q1 Multiple Ions (Q1 MI)
- Polarity: Positive (selected)
- Duration: 0.000 (min)
- Delay Time: 0 (sec)
- Cycles: 1
- Cycle: 0.0000 (sec)
- Scheduled Ionization:
- Start Time: 0 (min)
- Stop Time: 0 (min)
- Total Scan Time (includes pauses): 0.0000 (sec)

	Q1 Mass (Da)	Dwell Time (msec)
1		

**Table 11-3 MS Tab Parameter Values**

Field	Value
Q1 Mass (Da)	609
Time (msec)	100

8. Click **Edit Parameters**.  
The Parameter table dialog opens.
9. On the Source/Gas tab, type the following values:

**Table 11-4 Source/Gas Tab Parameters**

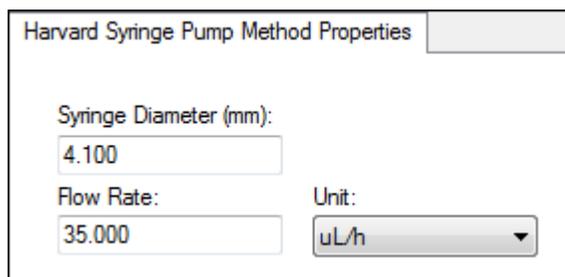
Source/Gas parameters	Typical value
Curtain Gas (CUR)	35

**Table 11-4 Source/Gas Tab Parameters (continued)**

Source/Gas parameters	Typical value
IonSpray Voltage (IS)	5000
Temperature (TEM)	0
Ion Source Gas 1 (GS1)	20
Ion Source Gas 2 (GS2)	0

10. Click the Compound tab and then set the **Declustering Potential (DP)** to 90 and leave the **Entrance Potential (EP)** at 10.  
A value of 90 might not be optimal for the mass spectrometer but it is a good DP to start with.
11. Click **OK**.
12. In the Acquisition method pane, click the **Harvard Syringe Pump** or **Integrated Syringe Pump** icon.

**Figure 11-5 Harvard Syringe Pump Method Properties Tab**



13. Edit the syringe pump method to include **Syringe Diameter**, **Flow Rate**, and **Unit**.
14. **Save** the acquisition method.  
Next steps: Create and submit a batch containing this acquisition method. To create and submit batches, refer to the section: [Create and Submit a Batch](#).

## Create an Acquisition Method using an MRM Scan Type

Use the following procedure to create a method using the MRM scan. This scan type is used in quantitative applications. An MRM scan can be used to determine how much of a compound is in a sample. It is used in pharmacokinetic analysis and increasingly in applied markets and screening applications.

### Prerequisites

- Make sure that a hardware profile containing the mass spectrometer and syringe pump is active.
- On the software toolbar, make sure that the appropriate project is selected.

1. On the Navigation bar, under **Acquire**, double-click **Build Acquisition Method**. The Acquisition Method Editor is shown with a method template based on the active hardware profile.
2. In the Acquisition Method pane, click **Acquisition Method**.
3. On the Acquisition Method Properties tab, in the **Synchronization Mode** list, make sure that **No Sync** is selected. For more information about synchronization modes, refer to the document: *Help*.
4. In the Acquisition method pane, click the **Mass Spec** icon.
5. On the MS tab, in the **Scan type** list, select **MRM (MRM)**.
6. In the Polarity section, click **Positive**.
7. In the mass ranges table, type the values that are shown in the following table.

**Figure 11-6 MRM Scan Type**

The screenshot shows the MS configuration interface for an MRM (MRM) scan. Key settings include:

- Experiment: 1
- Scan type: MRM (MRM)
- Polarity: Positive
- Scheduled MRM: Enabled
- Period Summary: Duration: 0.000 (min), Delay Time: 0 (sec), Cycles: 1, Cycle: 0.0000 (sec)
- Scheduled Ionization: Start Time: 0 (min), Stop Time: 0 (min)
- Total Scan Time (includes pauses): 0.0000 (sec)

	Q1 Mass (Da)	Q3 Mass (Da)	Dwell Time (msec)	ID
1				

**Table 11-5 Mass Range and Dwell Time**

Q1 Mass (Da)	Q3 Mass (Da)	Time (msec)
609	397.2	100

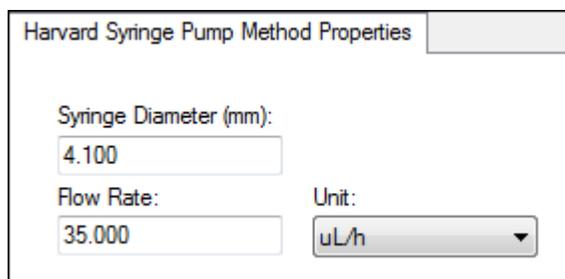
8. On the MS tab, click **Edit Parameters**.  
The Parameter Table dialog opens.
9. On the Source/Gas tab, type the following values:

**Table 11-6 Source/Gas Tab Parameters**

Source/Gas parameters	Typical value
<b>Curtain Gas (CUR)</b>	35
<b>IonSpray Voltage (IS)</b>	5000
<b>Temperature (TEM)</b>	0
<b>Ion Source Gas 1 (GS1)</b>	20
<b>Ion Source Gas 2 (GS2)</b>	0

10. On the Compound tab, set the **Declustering Potential (DP)** to 90 and leave the **Entrance Potential (EP)** at 45.
11. Click **OK**.
12. In the Acquisition method pane, click the **Harvard Syringe Pump** icon.

**Figure 11-7 Harvard Syringe Pump Method Properties Tab**



13. On the Syringe Pump tab, edit the syringe pump method to include **Syringe Diameter**, **Flow Rate**, and **Unit**.
14. **Save** the acquisition method.

Next steps: Create and submit a batch containing this acquisition method. To create and submit batches, refer to the section: [Create and Submit a Batch](#).

## Add or Remove Devices From Acquisition Methods

Use the Acquisition Method Editor to customize the acquisition method by adding or removing LC peripheral device methods. If the required device icon is not in the Acquisition Method Browser pane, then add the peripheral device only if it is included in the active hardware profile.

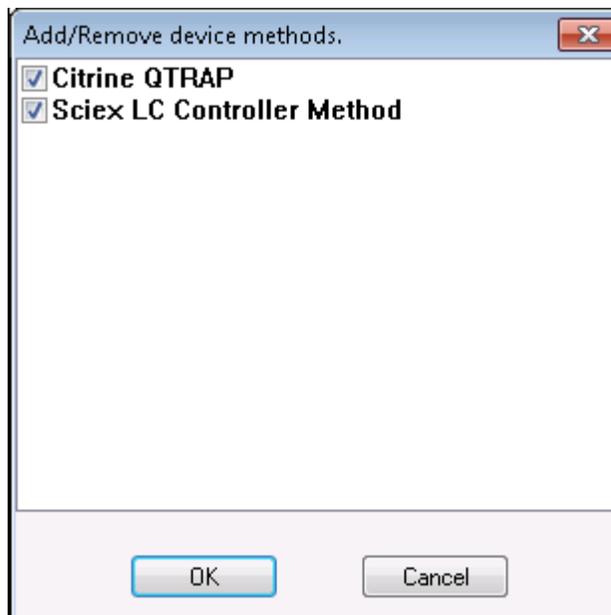
**Note:** The available parameters for the LC devices vary depending on the manufacturer.

---

### Add or Remove an LC Device

1. With a method file open in the Acquisition Method Editor, in the Acquisition method pane, right-click **Acquisition Method** and then click **Add/Remove Device Method**.

**Figure 11-8 Add/Remove Device Method Dialog**



2. Select or clear the check boxes beside the device method to add or remove the device method.
3. Click **OK**.
4. If an LC device is selected, then in the **Acquisition Method Properties** tab, select **LC sync** for the **Synchronization** mode.

### Modify LC Pump Properties

Modify each device in the method so that it functions properly for your method.

1. With a method file open in the Acquisition Method Editor, in the Acquisition method pane, click the LC device icon.  
The LC Pump Gradient tab opens in the Acquisition Method Editor pane.

Figure 11-9 SCIEX DX Pump Parameters

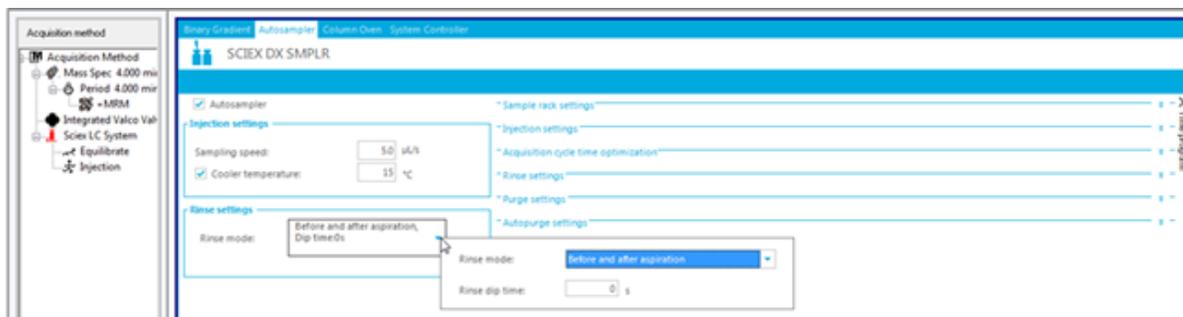


2. Edit the LC pump method as required for the experiments.
3. Save the method.

## Set Autosampler Properties

1. With a method file open in the Acquisition Method Editor, in the Acquisition method pane, click the **Autosampler** device.  
The Autosampler Properties tab opens in the Acquisition Method Editor pane.

Figure 11-10 SCIEX DX Autosampler Parameters



2. If required, edit the injection and wash details.
3. Save the method.

### Set Column Oven Properties

1. With a method file open in the Acquisition Method Editor, in the Acquisition method pane, click the **Column Oven** device.

The Column Oven Properties tab opens in the Acquisition Method Editor pane.

**Figure 11-11 SCIEX DX Column Oven Parameters**



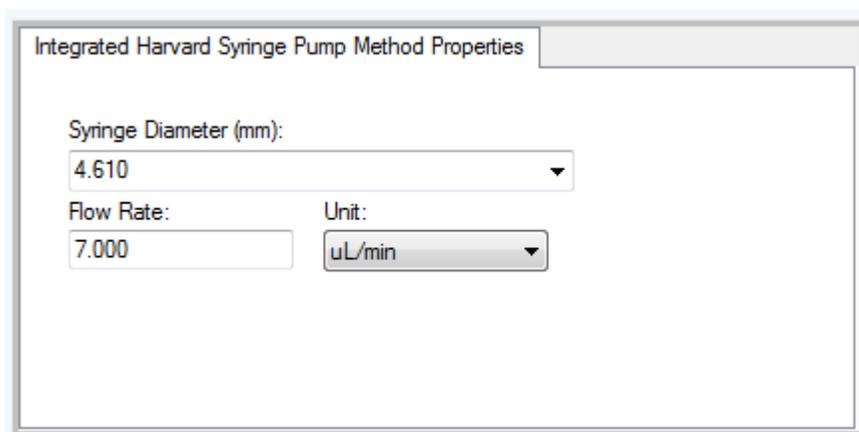
2. Type the temperature of the column oven or column oven compartments in degrees Celsius.
3. Save the file.

### Configure the Integrated Syringe Pump

This procedure is for the integrated syringe pump.

1. Make sure that the integrated syringe pump is selected in the hardware device profile.
2. Click the **Syringe Pump** icon in the Acquisition method pane.  
The Syringe Pump Method Properties tab opens in the Acquisition Method Editor.

**Figure 11-12 Syringe Pump Properties tab**



3. Type the syringe diameter in the **Syringe Diameter (mm)** field.

4. Type the flow rate in the **Flow Rate** field.
5. Select the units of flow from the **Unit** list.

### **Set Diverter Valve Properties**

The switching valve can be used as a diverter or injection valve.

1. With a method file open in the Acquisition Method Editor, in the Acquisition method pane, click the **Valve** icon.  
The Valve Properties tab opens in the Acquisition Method Editor pane.

Figure 11-13 Integrated Valco Valve

The screenshot shows the 'Integrated Valco Valve Method Properties' dialog box. On the left, a tree view under 'Acquisition method' shows the following structure:

- Acquisition Method
  - Mass Spec 4,000 min
    - Period 4,000 min
      - +MRM
      - Integrated Valco Valve** (highlighted)
      - Sciex LC System
        - Equilibrate
        - Injection

The right pane contains the following configuration options:

- Valve Type: Diverter
- Change Position Names: [Dropdown]
- Position Name for Step0: A

Below these options are two paragraphs of instructions:

To use the valve as an injector, select the Synchronization Mode, "Manual Sync with Valve".

To use the valve as a diverter, select any other Synchronization Mode.

(To change the Synchronization Mode, click on "Acquisition Method" tab.)

A table with 20 rows and 2 columns is shown:

	Total Time (mi)	Position
1		
2		
3		
4		
5		
6		
7		
8		
9		
10		
11		
12		
13		
14		
15		
16		
17		
18		
19		
20		

At the bottom right, there are two buttons: 'Add Entry' and 'Remove Entry'.

2. Change the position names from their preset names, if required.

The switching valve is sometimes used to switch the flow of solvent to waste, or to a different column. The preset position names are A and B.

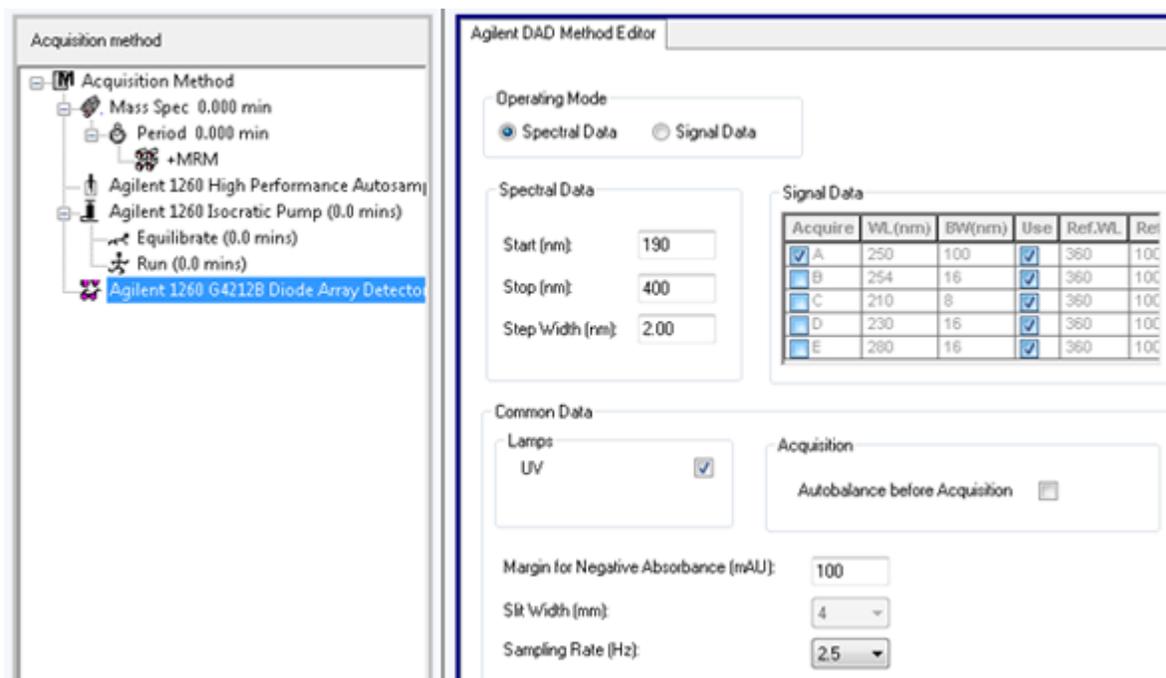
- In the **Change Position Names** list, select a position.
- In the **Change Position Names** list, rename the preset position names A and B to Inject and Divert or to Column and Waste, depending on how the valve is plumbed.

3. In the **Total Time (min)** column, click a cell and then type the total time the valve will remain in this position.
4. In the **Position** column, click a cell and then, in the **Position** list, select the valve position.
5. Repeat step 3 and step 4 for each switch of the valve required during acquisition.
6. Save the file.

## Set Diode Array Detector Properties

1. With a method file open in the Acquisition Method Editor, in the Acquisition method pane, click the **Diode Array Detector (DAD)** icon.  
The DAD Method Editor tab opens in the Acquisition Method Editor pane.

Figure 11-14 DAD Method Editor Tab

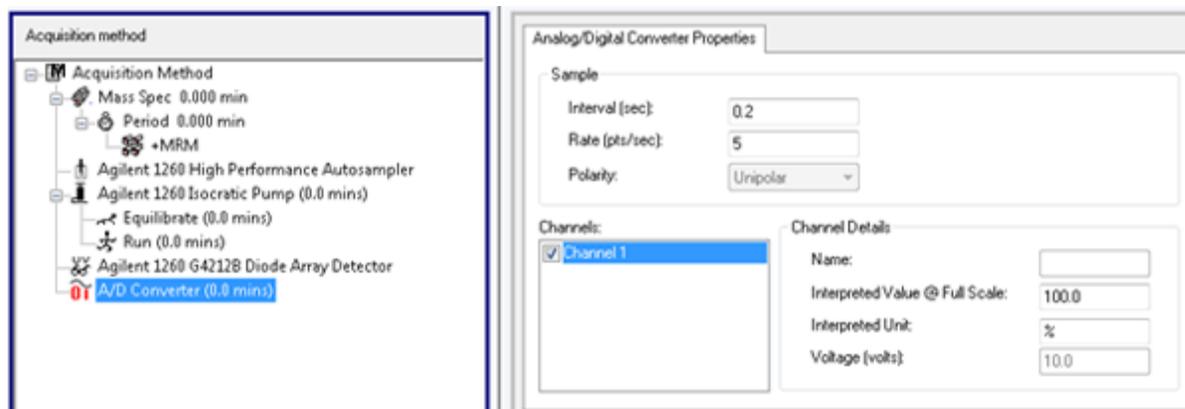


2. Do one of the following:
  - To scan one to five individual wavelengths, in the **Operating Mode** section, click **Signal Data** and then edit the data requirements.
  - To scan over a wavelength range, in the **Operating Mode** section, click **Spectral Data** and then edit the data requirements.
3. Save the file.

### Set Analog to Digital Converter Properties

1. With a method file open in the Acquisition Method Editor, in the Acquisition method pane, click the **Analog to Digital Converter (ADC)** icon.  
The Analog/Digital Converter Properties tab opens in the Acquisition Method Editor pane.

**Figure 11-15 Analog to Digital Converter Properties Tab**



2. In the **Sample** section, in the **Rate (pts/sec)** field, type the rate.

---

**Note:** The interval and rate are proportional to each other. When you change the rate, the software automatically calculates the interval.

---

3. Do the following to set the channel details:
  - In the **Channels** field, click the channel name, and then select the check box beside the name to include it in the method.
  - In the **Interpreted Value @ Full Scale** field, type the appropriate value.
  - In the **Interpreted Unit** field, type the appropriate unit.

The number of available channels is specified when setting up the ADC in the hardware profile.

4. Save the file.

### Change Acquisition Methods

In Acquire mode, users can add or delete periods and experiments to existing acquisition methods.

## Periods

A period can contain one or more looped experiments. In a multi-period acquisition method, experiments are performed for a specified amount of time and then the software switches to another set of experiments. Periods are useful when the elution time of the compounds in an LC run is known. The mass spectrometer can perform different experiments according to when the compounds elute to obtain as much information as possible in the same run.

The following figure shows a three-period method.

**Figure 11-16 Example of a Multi-period Experiment**

The screenshot displays the software interface for configuring an acquisition method. On the left, the 'Acquisition method' tree shows a sequence of three periods: 3.000 min, 2.000 min, and 1.000 min, each containing an MS2 scan. The hardware components listed include an Agilent 1260 High Performance Autosampler, Agilent 1260 Isocratic Pump (6.0 mins), Equilibrate (0.0 mins), Run (6.0 mins), Agilent 1260 G4212B Diode Array Detector, and A/D Converter (0.0 mins). On the right, the 'MS' parameters for the first period are shown. The 'Scan type' is 'Product Ion (MS2)', 'Scan rate' is '200 (Da/s)', and 'Polarity' is 'Positive'. The 'Period Summary' indicates a 'Duration' of 3.000 min, 'Delay Time' of 0 sec, 'Cycles' of 109, and 'Cycle' time of 1.6555 sec. The 'MCA' section shows 'Number of scans to sum' as 1 and 'Product Of' as 609.300 (Da). A table below the MCA section shows the mass range and time for the first two cycles:

	Start (Da)	Stop (Da)	Time (sec)
1	70.000	400.000	1.6555
2			

## Experiments

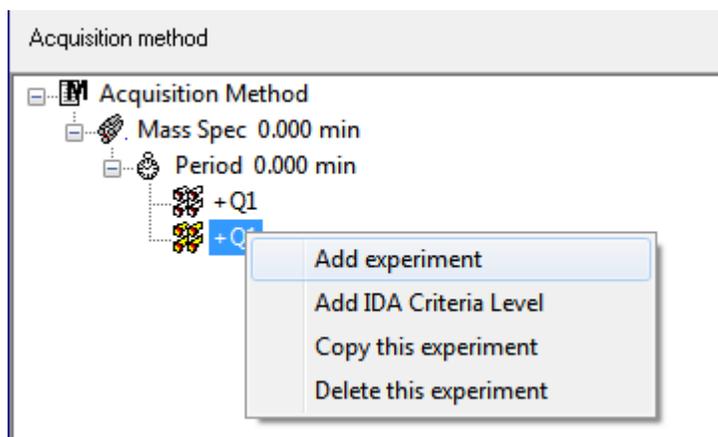
An experiment includes the mass spectrometer settings and the scan type during an MS scan. A set of MS scans performed for a specific amount of time is called a period. An acquisition method in which the MS parameters and actions are the same through the entire duration is called a single-period, single-experiment method.

In looped experiments, MS settings are changed on a scan-by-scan basis. For example, if the sample contains two compounds, A and B, users might want to loop an MS/MS experiment of compound A with an MS/MS experiment of compound B to obtain information about both compounds in the same run. The mass spectrometer method will alternate between the two scan types. Other examples of looped experiments include alternating between positive and negative modes in a run and Information Dependent Acquisition (IDA) methods.

## Add an Experiment

1. In the Acquisition method pane, on the period where the experiment is to be added, right-click and then click **Add experiment**.

**Figure 11-17 Add Experiment**



An experiment is added below the last experiment in the period.

---

**Note:** An experiment cannot be inserted between experiments, IDA criteria, or periods. Users can only add an experiment at the end of the period.

---

2. On the MS tab, select the appropriate parameters.

### Copy an Experiment into a Period

1. Open a multi-period method.
2. In the Acquisition method pane, press **Ctrl**, and then drag the experiment to the period. The experiment is copied below the last experiment in the period.

### Copy an Experiment within a Period

Use this procedure to add the same or similar experiments to a period if most or all of the parameters are the same.

Right-click the experiment and then click **Copy this experiment**.

A copy of the experiment is added below the last experiment created.

### Add a Period

In the Acquisition method pane, right-click the **Mass Spec** icon, and then click **Add period**. A period is added below the last period created.

---

**Note:** Users cannot use multiple periods in an IDA experiment.

---

## Scan Techniques

**MS:** In MS scans, also referred to as single MS scans, ions are separated according to their mass-to-charge ratio ( $m/z$ ). A single MS scan might be used to determine the molecular weight of a compound. Single MS scans can also be referred to as survey scans. MS scans do not provide any information about the chemical composition of the ions, other than the mass. Perform MS/MS or MS/MS/MS scans to obtain more information about the ions.

**MS/MS:** MS/MS scans are used to determine structural information.

- For MS/MS scans on triple quadrupole systems, selected ions enter the Q2 collision cell where they are collisionally activated to fragment, producing characteristic product ions.
- For MS/MS scans on QTRAP systems, precursor ion fragmentation can occur in the Q2 collision cell or the linear ion trap.

If enough energy is used, then the precursor ion fragments to produce characteristic product ions.

**MS/MS/MS:** The linear ion trap (LIT) system MS/MS/MS scans go one step further than MS/MS scans. A fragment that is produced in the collision cell is fragmented further in the LIT to give more structural information about the molecular ion.

## Scan Types

Triple quadrupole instruments have high-sensitivity Multiple Reaction Monitoring (MRM) capabilities required for quantitation experiments. In addition, they have highly specific scan types, such as precursor ion and neutral loss scans, which allow a more advanced search to be performed on the components of the samples.

**Q1 MS (Q1):** A full scan type using the first quadrupole (Q1). The ion intensity is returned for every mass in the scan range.

**Q1 Multiple Ions (Q1 MI):** A selective scan type using the Q1 quadrupole. The ion intensity is returned for the specified masses only.

**Q3 MS (Q3):** A full scan type using the third quadrupole (Q3). The ion intensity is returned for every mass in the scan range.

**Q3 Multiple Ions (Q3 MI):** A selective scan type using the Q3 quadrupole. The ion intensity is returned for the specified masses only.

**MRM (MRM):** An MS/MS scan in which a user-defined ion is isolated in the Q1 quadrupole, then fragmented in the Q2 collision cell. The Q3 quadrupole is then used to isolate a user-defined fragment ion that is recorded by the detector. This scan type is used primarily for quantitation.

**Product Ion (MS2):** An MS/MS full scan where the Q1 quadrupole is used to isolate and transmit a specific precursor ion and the Q3 quadrupole scans a defined mass range. This scan type is used to identify all of the fragment ions of a particular precursor ion.

**Precursor Ion (Prec):** An MS/MS scan where the Q3 quadrupole is fixed at a specified  $m/z$  value to transmit a specific product ion and the Q1 quadrupole scans a mass range. This scan type is used to confirm the presence of a precursor ion or, more commonly, to identify compounds sharing a common product ion.

**Neutral Loss (NL):** An MS/MS scan where both the Q1 quadrupole and the Q3 quadrupole scan a mass range, a fixed mass apart. A response is observed if the ion chosen by the Q1 quadrupole fragments by losing the neutral loss, the fixed mass, specified. This scan type is used to confirm the presence of a precursor ion or, more commonly, to identify compounds sharing a common neutral loss.

## LIT-Mode Scan Types

The LIT-mode scans use the Q3 quadrupole as a linear ion trap. Ions are trapped and stored in the Q3 quadrupole before being scanned out, giving increased sensitivity. In addition, MS/MS/MS analysis can be performed in the linear ion trap, providing more information about the sample. LIT-mode scan types are typically used for qualitative measurements.

**Enhanced MS (EMS):** Ions are scanned in the Q1 quadrupole and then collected in the linear ion trap. These ions are scanned out of the Q3 quadrupole to produce single MS type spectra.

**Enhanced Product Ion (EPI):** This scan type is used to obtain a high-quality MS/MS spectrum for a specific ion. The fragmentation is done in the Q2 collision cell and thus provides the information-rich MS/MS spectrum typical of collisionally activated dissociation (CAD) fragmentation. In this scan mode, the precursor ion to be fragmented is first selected in the Q1 quadrupole with a mass window that is 1 Da to 4 Da wide, filtering out all of the other ions. The precursor ion is fragmented by CAD gas in the Q2 collision cell. The fragment ions generated are captured in the linear ion trap and then scanned out at one of three scan speeds, depending on the required fragment ion resolution.

For IDA experiments, the **Product Of** field is set to 30 Da by default, and this value should not be changed.

**Enhanced Resolution (ER):** This scan type is similar to the EMS scan except that a small 30 Da mass window around the precursor mass is scanned out of the linear ion trap at the slowest scan rate to produce a narrow window of the best-resolved spectra.

**MS/MS/MS (MS3):** A precursor ion is selected by the Q1 quadrupole and fragmented with collisionally activated dissociation in the Q2 collision cell. The resulting product ions are all transmitted to the linear ion trap, where a single product ion is then isolated. The isolated ion is further fragmented in the linear ion trap, and the resulting product ions are scanned out of the trap at one of three scan speeds. As with any in-trap Collision Induced Dissociation (CID)

technique, there is a low mass cut-off for the second MS/MS step because the lowest mass fragment and precursor must be simultaneously stable in the trap. For QTRAP systems, this results in the loss of ions lower than 28 percent of the mass of the precursor ion during MS3 experiments. This phenomenon is often referred to as the one-third cut-off rule.

## About Spectral Data Acquisition

For a description of the modes in which spectral data can be acquired, refer to the table: [Table 11-7](#).

**Table 11-7 Spectral Data**

Mode	Description
Profile	The preset value is 0.1 Da. Profile data is the data generated by the mass spectrometer and corresponds to the intensity recorded at a series of evenly spaced discrete mass values. For example, for a mass range from 100 Da to 200 Da and a step size of 0.1 Da, the mass spectrometer scans 99.95 to 100.05 (recorded as value 100), 100.95 to 101.05 (recorded as value 101)...199.95 to 200.05 (recorded as value 200).
Peak Hopping	The preset value is 1.0 Da. Peak hopping is a mode of operating a mass spectrometer in which large steps (approximately 1 Da) are made. It has the advantage of speed (fewer data steps are made) but with the loss of peak shape information.
Centroid	The mass spectrometer scans as in profile mode, but creates a centroid of the data, replacing found peaks with the intensity-weighted center of gravity for each peak. Centroid data has the advantage of significantly reducing file size. The disadvantage is that peak shape information is lost and if data has been collected as a centroid then it cannot be altered. We recommend the use of profile mode and centroiding of the data post-acquisition.

---

**CAUTION: Potential System Damage.** If the LC system connected to the mass spectrometer is not controlled by the software, then do not leave the mass spectrometer unattended while in operation. The liquid stream from the LC system can flood the ion source when the mass spectrometer goes in Standby state.

---

A batch is a collection of information about the samples to be analyzed. Samples are usually grouped into sets to simplify submission. Grouping the samples into a set also reduces the amount of data that must be typed manually. A set can consist of a single sample or multiple samples. All of the sets in a batch use the same hardware profile. However, samples in a set can have different acquisition methods. A batch can be submitted only from an acquisition computer.

Batches include the following information:

- Sample information, such as name, ID, and comment
- Autosampler rack information, vial position, and injection volume
- Acquisition methods
- Processing method (optional)
- Quantitation information (optional)
- Custom sample data (optional)
- Set information

## Create and Submit Batches

A batch is a collection of information about the samples to be analyzed. Batches tell the software the order in which to analyze the samples. For information about importing batches, refer to the *Advanced User Guide*.

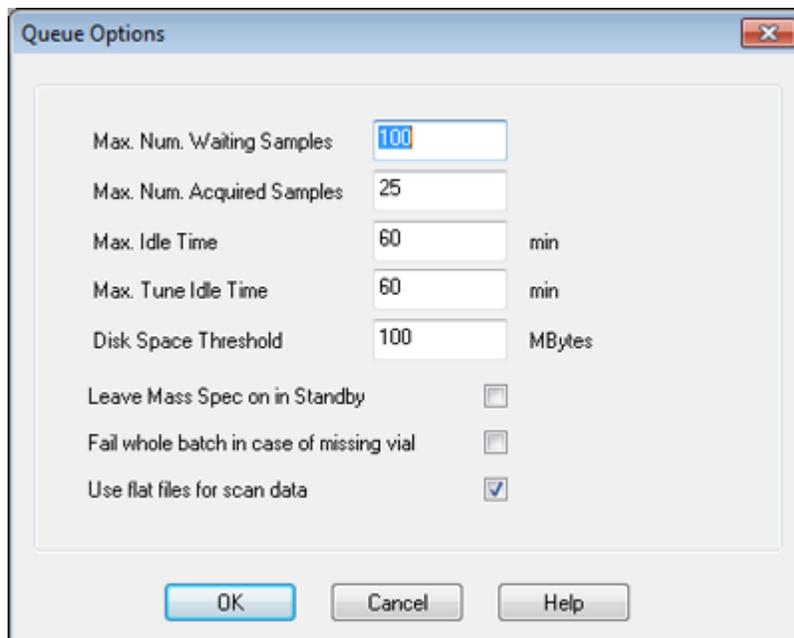
## Set Queue Options

The software goes one-by-one through the list of samples in the queue, acquiring each sample with the selected acquisition method. After all of the samples have been acquired, the acquisition stops and the mass spectrometer goes into Standby state after the **Max. Idle Time** set in the Queue Options has elapsed. In the Standby state, the LC pumps stop and some instrument voltages are turned off.

The user can change the amount of time between acquisition of the last sample and the change to Standby state. For information about the other fields in the Queue Options dialog, refer to the document: *Help*.

1. On the Navigation bar, click **Configure**.
2. Click **Tools > Settings > Queue Options**.

**Figure 12-1 Queue Options Dialog**



3. In the **Max. Num. Waiting Samples** field, set the maximum number of samples to a value that is greater than the number of samples that will be submitted to the queue.
4. In the **Max. Idle Time** field, type the amount of time the software will wait after acquisition is completed before going to Standby state. The preset value is 60 minutes.

If gas cylinders are used, then adjust this time to make sure that the gas in the cylinders is not depleted.

If an LC method is used, then before the run is started, make sure that there is enough solvent in the reservoirs for all of the sample runs at the primary flow rate and the maximum idle time.

5. Select the **Leave Mass Spec on in Standby** check box to keep the mass spectrometer running after analysis has been completed.

This feature allows the heaters and gases to continue running, even after devices have entered Idle state, so that the ion source and entrance to the mass spectrometer are kept free of contaminants.

## Operating Instructions — Batches

---

6. Select the **Fail Whole Batch in Case of Missing Vial** check box to make the entire batch fail when a missing vial is encountered.

If this option is not selected, then only the current sample will fail and the software will continue to the next sample.

## Create and Submit a Batch

Use this workflow to create a batch.

Do not use the automatically generated Quick Quant method to perform quantitation if the Quick Quant feature is used to store Sample Types and Concentrations in the data file. This quantitation method does not use compound and sample-specific integration parameters that have been optimized for peak selection.

## Add Sets and Samples to a Batch

A set can consist of a single sample or multiple samples.

---

**Note:** For more information about adding quantitation information to a batch, refer to the document: *Advanced User Guide*.

---

1. On the Navigation bar, under **Acquire**, double-click **Build Acquisition Batch**.

**Figure 12-2 Batch Editor Dialog**

Sample Name	Rack Code	Rack Position	Plate Code	Plate Position	Vial Position	Data File	Inj. Volume (µl)
-------------	-----------	---------------	------------	----------------	---------------	-----------	------------------

2. On the Sample tab, in the **Set** list, type a name.
3. Click **Add Set**.
4. Click **Add Samples** to add samples to the new set.

Figure 12-3 Add Sample Dialog

5. In the Sample name section, in the **Prefix** field, type a name for the samples in this set.
6. To add incremental numbering to the end of the sample name, select the **Sample number** check box.
7. If the **Sample number** check box is selected, then in the **Number of digits** field, type the number of digits to include in the sample name.  
For example, if 3 is typed, then the sample names would be samplename001, samplename002, and samplename003.
8. In the Data file section, in the **Prefix** field, type a name for the data file that will store the sample information.
9. Select the **Set name** check box to use the set name as part of the data file name.
10. Select the **Auto Increment** check box to increment the data file names automatically.

When the mass spectrometer is used as an *in vitro* diagnostic device, it is recommended that only one sample be acquired per data file.

---

**Note:** The data for each sample can be stored in the same or a separate data file. The names of the data file will have numerical suffixes starting from 1.

---

11. Type a name in the **Sub Folder** field.  
The folder is stored in the `Data` folder for the current project. If the **Sub Folder** field is left blank, then the data file is stored in the `Data` folder and a subfolder is not created.

## Operating Instructions — Batches

---

12. In the New samples section, in the **Number** field, type the number of new samples to add.
13. Click **OK**.  
The sample table fills with the sample names and data file names.

---

**Tip! Fill Down** and **Auto Increment** options are available in the right-click menu after a single column heading or several rows in a column are selected.

---

14. On the Sample tab, in the Acquisition section, select a method from the list.  
Depending on how the system is set up, specific information for the autosampler must be entered. Even if the injection volume is set in the method, the user can change the injection volume for one or more samples by changing the value in the injection volume column.

---

**Note:** To use different methods for some of the samples in this set, select the **Use Multiple Methods** check box. The **Acquisition Method** column is shown in the Sample table. Select the acquisition method for each sample in this column.

---

15. In the **Sample** tab, from the **Set** list, select the set.
16. For each sample in the set, do the following if applicable:
  - In the **Rack Code** column, select the rack type.
  - In the **Rack Position** column, select the position of the rack in the autosampler.
  - In the **Plate Code** column, select the plate type.
  - In the **Plate Position** column, select the position of the plate on the rack.
  - In the **Vial Position** column, type the position of the vial in the plate or tray.

---

**Note:** To automatically fill in the samples from the Locations tab, click on the first and last vial within a set while pressing the **Shift** key. These vials appear as red circles. On the Locations tab, multiple injections from the same vial can be done by pressing the **Ctrl** key while clicking the vial location. The red circle turns green.

---

17. Alternatively, to use the location tab to select the vial positions, refer to the section: [Select Vial Positions Using the Locations Tab \(Optional\)](#).
18. (Optional) Use the procedures in section: [Table 12-1](#) as required.

**Table 12-1 Batch Editor Tips**

Tp Do This	Do This
To change all the values in a column simultaneously	Click a column heading and then right-click. From the menu, select the <b>Auto Increment</b> and <b>Fill Down</b> commands to change the values in the column.  This feature also works for multiple cells in the same column.
To change an existing acquisition method	Select the method and then click <b>Method Editor</b> from the list. To create a new acquisition method, select <b>None</b> from the list and then click <b>Method Editor</b> . Only experienced users should use this feature.  Do not use this feature if the <b>Use Multiple Methods</b> option is selected.
To apply a previously created quantitation method	Select the method from the <b>Quantitation</b> list.
To select more than one well or vial at a time	Press the <b>Shift</b> key while clicking the first and last well or vial in the range.

19. (Optional) To define quantitation details prior to submitting the batch, refer to the section: [Set Quantitation Details in the Batch Editor \(Optional\)](#).
20. Open the Submit tab.

---

**Note:** The order of samples can be edited before the samples are submitted to the queue. To change the order of samples, on the Submit tab, double-click any of the numbers at the far left of the table (a very faint square box is shown), and then drag them to the new location.

---

21. If the Submit Status section contains a message about the status of the batch, then do one of the following:
  - If the message indicates that the batch is ready for submission, then go to step [22](#).
  - If the message indicates that the batch is not ready for submission, then make the changes as indicated by the message.
22. After confirming that all of the batch information is correct, click **Submit**. The batch is submitted to the queue and can be viewed in the Queue Manager.
23. Save the file.

### Submit a Sample or Set of Samples

---

**Note:** Run the sample again if an abnormal termination occurs during sample acquisition. If the abnormal termination is caused by a power failure, then the temperature of the autosampler tray is not maintained and sample integrity might be compromised.

---

1. Select one sample or a set of samples.
2. Open the Submit tab in the Batch Editor.
3. If the Submit Status group contains a message about the status of the batch, then do one of the following:
  - If the message indicates that the batch is ready for submission, then go to the next step.
  - If the message indicates that the batch is not ready for submission, then make the changes as indicated by the message.
4. Click **Submit**.

### Change Sample Order

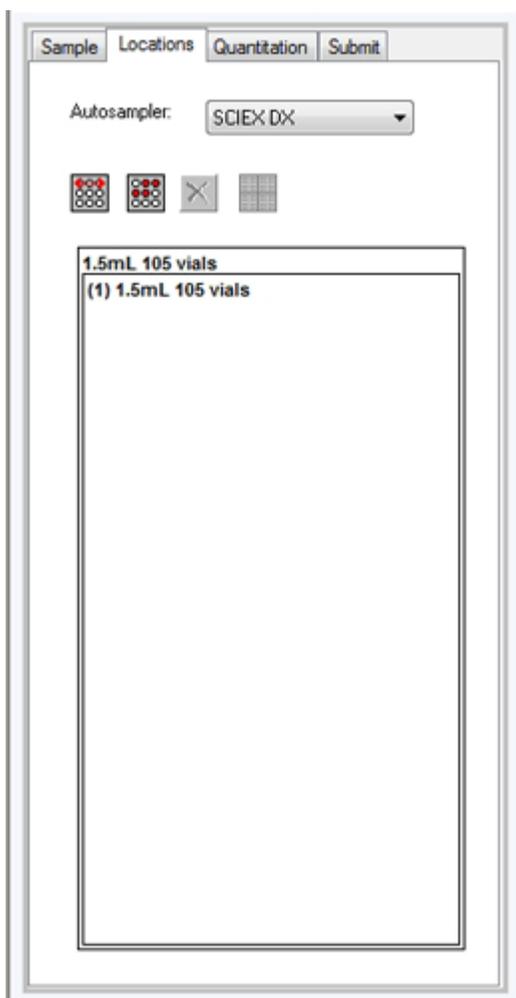
The order of the samples can be changed before the samples are submitted to the **Queue**.

On the Submit tab, double-click any of the numbers at the far left of the table (a very faint square box is visible), and then drag them to the new location.

### Select Vial Positions Using the Locations Tab (Optional)

1. In the Batch Editor, open the Locations tab.
2. In the **Set** list, select the set.
3. From the **Autosampler** list, select the autosampler.
4. In the space associated with the rack, right-click and then select the rack type. The plates or trays are shown in the rack.
5. Double-click in the white space labeled rack type. A visual sample rack layout is shown. The appropriate number of rack spaces for the autosampler is shown in the graphic rack view.

Figure 12-4 Visual Sample Location



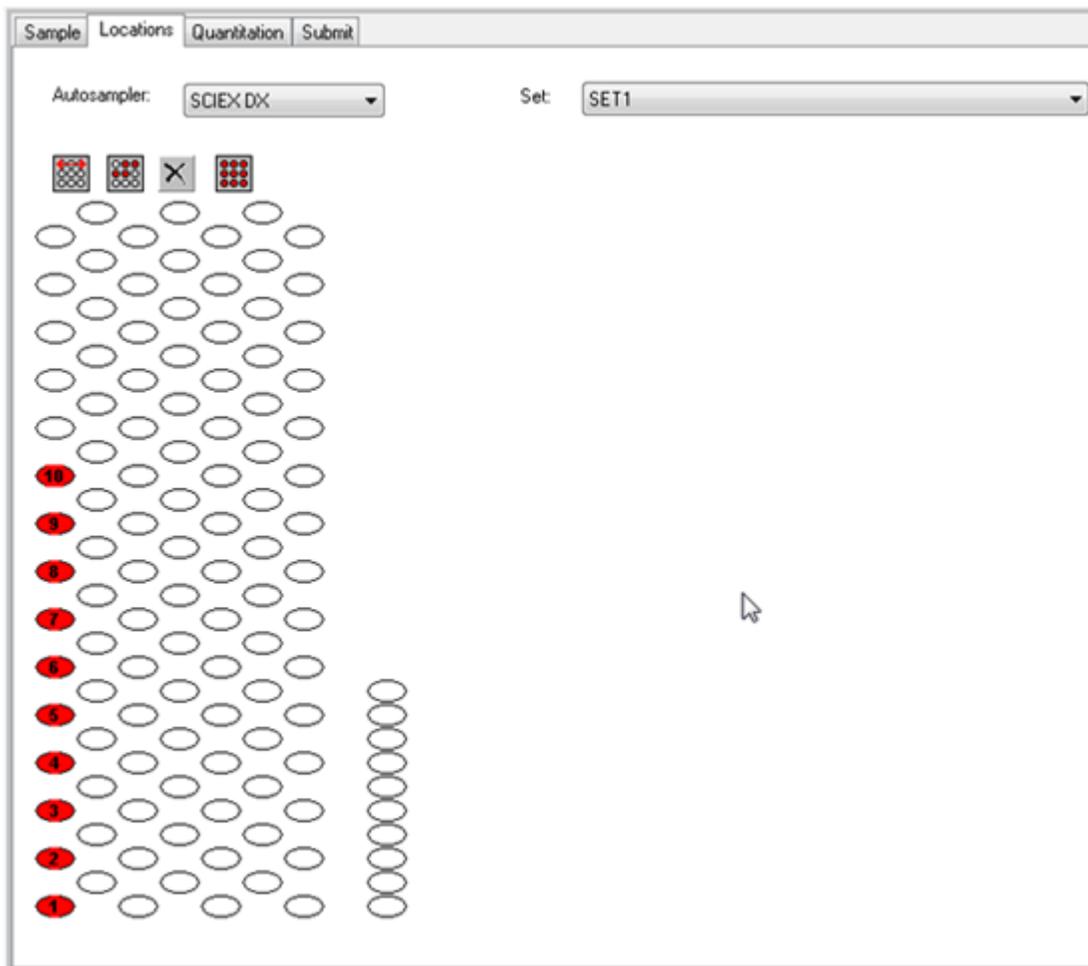
6. Double-click one of the rectangles.  
The circles depicting the wells or vials for the plate or tray are shown.

---

**Tip!** To see the corresponding vial number in the graphical representation, move the cursor over the sample position. Use this information to confirm that the vial positions in the software match the vial positions in the autosampler.

---

Figure 12-5 Locations Tab



---

**Note:** Depending on the autosampler being used, it might not be necessary to type details in additional columns.

---

7. To select whether samples are marked by row or column, click the **Row/Column selection** selector button.  
If the button shows a red horizontal line, then the Batch Editor marks the samples by row. If the button shows a red vertical line, then the Batch Editor marks the samples by column.
8. Click the sample wells or vials in the order in which they should be analyzed.

---

**Tip!** Click a selected well or vial again to clear it.

---

**Tip!** To fill in the samples automatically, press **Shift** while clicking the first and last vial within a set. To perform multiple injections from the same vial, press **Ctrl** while clicking the vial location. The red circle changes to a green circle.

---

## Set Quantitation Details in the Batch Editor (Optional)

If a quantitation method is used with a batch and if the user does not want to select quantitation details after acquisition, then the quantitation details (sample type, sample concentration) must be defined before the batch is submitted.

The appropriate **Internal Standard** and **Standard** columns are shown in the Quantitation tab according to the quantitation method selected in the Sample tab.

1. With a batch file open in the Batch Editor window, open the Quantitation tab.
2. Select the set containing the samples.
3. Select a **Quant Type**, **Dilution Factor**, and **Weight/Volume** for all of the samples from the list in the cell.
4. (If required) In the **Analyte** column, type the analyte concentration.
5. (If required) In the **Internal Standard** column, type the internal standard concentration.
6. Repeat this procedure for each set in the batch.

## Equilibrate the System

Equilibrate the system before submitting a batch. Equilibration warms up and prepares the mass spectrometer for the next sample or batch.

1. Click **Equilibrate** ()  
The Equilibrate dialog opens.
2. Select the acquisition method used for the submitted batch.
3. Type the equilibration time in the **Time (min)** field, in minutes.
4. Click **OK**. The system starts the equilibration.  
After the equilibration is completed, the system state changes to Ready.

## Operating Instructions — Batches

---

**Tip!** If the equilibration does not finish as expected, or if the system state does not change to Ready after the equilibration finishes, then make sure that the following conditions are true:

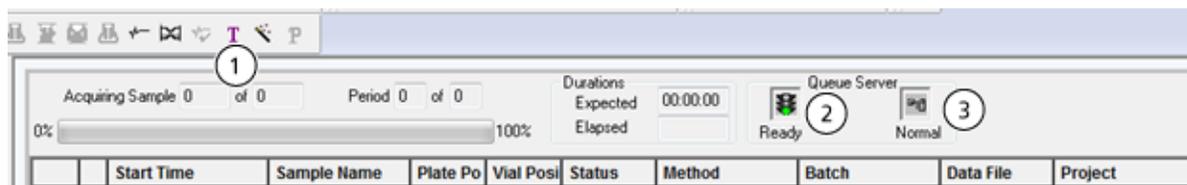
- The activated hardware profile is appropriate for the acquisition method.
  - The LC system is turned on.
  - The LC system has correctly established communication with the software.
- 

## Acquire Data

The software must not be in Tune and Calibrate mode when sample acquisition is started. Also, if the system has been previously run that day and has not yet been set to Standby state, then sample acquisition will start automatically.

1. Make sure that the column oven temperature is reached.
2. Make sure that the **Reserve Instrument for Tuning** (  ) icon is not pressed in.
3. On the Navigation bar, click **Acquire**.
4. Click **View > Sample Queue**.  
The Queue Manager opens with all of the submitted samples.

**Figure 12-6 Queue Manager**



Item	Description
1	The <b>Reserve Instrument for Tuning</b> icon should not be pressed in.
2	Queue state must be Ready.
3	<b>Queue Server</b> state must be Normal. Refer to the section: <a href="#">Queue States</a> .

5. Click **Acquire > Start Sample**.

---

**Note:** We recommend that the sample be run again if an abnormal termination occurs during sample acquisition.

---

## Import Batch Files

Users can import a text file containing batch information instead of creating a batch in the Batch Editor. If all of the sample details are in a spreadsheet, then rearranging and importing the data in the spreadsheet is faster than manually typing the data in the Batch Editor.

Before importing batch information from a text file, make sure that the data in the file is organized and formatted correctly. In particular, the column headings in the spreadsheet must match the Batch Editor column headings. To make sure that the text file includes the proper headings, create a batch using the Batch Editor, export it as a text file, type the appropriate values in a spreadsheet editor, and then import the file back into the Batch Editor.

For examples of correctly formatted files, refer to the Batch folder in the Example project.

The information in a batch file can also be exported for use with other applications, such as Microsoft Excel, Microsoft Access, and certain Laboratory Information Management System (LIMS) software.

## Build a Batch as a Text File

Prerequisites
Make sure that the active hardware profile includes all of the devices to be used to acquire the samples.

To make sure that the text file includes the proper headings, create a batch using the Batch Editor, export it as a text file, type the appropriate values in a spreadsheet editor, and then import the file back into the Batch Editor. Users can export a batch only if it contains at least one set with at least one sample. The saved text file can be used again later as a template.

1. In the Batch Editor, create a single-set, single-sample batch.
2. Click **File > Export**.  
The Save As dialog opens.
3. Type a name for the text file in the **File name** field and then click **Save**.
4. Open the text file in a spreadsheet program such as Excel.
5. Type, or copy and paste, the details for the samples: one sample per row, with the details under the appropriate headings.

---

**Note:** Do not delete any of the columns. The columns in the spreadsheet must match the columns in the Batch Editor.

---

6. Save the modified text file as a txt or csv file and then close the spreadsheet program.  
The text file is now ready to be imported into the Batch Editor.

### Import a Batch from a Text File

1. In the Batch Editor, on the Sample tab, right-click, and then click **Import From > File**. The Open dialog opens.
2. Click the required text file and then click **Open**. If an autosampler is used, then the Select Autosampler dialog opens.

---

**Note:** If the saved text file is not visible in the **Files of type** list, then select **Microsoft Text Driver (\*.txt; \*.csv)**. Files with the extension .txt are shown in the field.

---

3. In the autosampler list, select the autosampler and then click **OK**. The sample table fills with the details from the text file.
4. Submit the batch.

### Batch and Acquisition Method Editor Tips

To Do This	Do This
To change the values in the table	(For example, to change a sample name) Click in a cell and then type the new value.
To change all the values in a column simultaneously	Click a column heading and then right-click. From the menu that is shown, select the <b>Auto Increment</b> and <b>Fill Down</b> commands to change the values in the column.  This feature also works for multiple cells in the same column.
To change an existing acquisition method	From the list, select the method and then click <b>Method Editor</b> . To create a new acquisition method, from the list, select <b>None</b> and then click <b>Method Editor</b> . Only experienced users should use this feature.  Do not use this feature when you are using the Use <b>Multiple Methods</b> option.
To apply a previously created quantitation method	Select the method from the <b>Quantitation</b> menu.
To select more than one well or vial at a time	Press <b>Shift</b> and click the first and last well or vial of the range you want to select.

For information on using the Batch Editor right-click menu, refer to the section: [Batch Editor](#).

## Batch Troubleshooting Tips

Symptom	Possible Cause	Corrective Action
In the Analyst MD software, Batch editor, the injection volume (uL) column is shown as -1.	The acquisition method is not selected in the batch.	Select the correct acquisition method for the batch.
	LC device in the selected acquisition method does not match the activated LC device in hardware profile.	Open and review the intended acquisition method. Correct the LC device information and then save the method.
	The activated hardware profile does not contain an LC device or it contains a different LC device.	Open the Hardware Configuration Editor and then deactivate the hardware profile. Edit the hardware profile, correct the LC device information, and then activate the hardware profile.

## Stop Sample Acquisition

When a sample acquisition is stopped, the software completes the current scan before stopping acquisition.

1. In the Queue Manager, click the sample in the queue after the point where acquisition should stop.
2. On the Navigation bar, click **Acquire**.
3. Click **Acquire > Stop Sample**.  
The acquisition stops after the current scan in the selected sample is acquired. The sample status in the **Queue Manager (Local)** window changes to **Terminated**, and all other samples following in the queue are **Waiting**.
4. To continue processing the batch, click **Acquire > Start Sample**.

Use the sample files installed in the Example folder to learn how to view and analyze data using the most common analysis and processing tools. For more information about the following topics, refer to the document: *Advanced User Guide*.

- Labeling graphs
- Overlaying and summing spectra or chromatograms
- Performing background subtractions
- Smoothing algorithms
- Working with smoothed data
- Working with centroid data
- Working with contour plots
- Working with the fragment interpretation tool
- Working with library databases and library records

## Overview of Spectral and Chromatographic Data

Spectral data provides mass-specific information about a compound. A chromatogram provides a general view of the data, usually time dependent when using an LC column, but it does not provide any information about the components of a peak. A spectrum, however, looks at a particular peak and provides the molecular weight of the corresponding compound, which can be used to find more specific information. For example, while a chromatogram might show only one peak, that peak can represent more than one compound, that is, different masses. A spectrum shows all of the masses that make up a peak, including the intensity of each mass.

Chromatographic data can change in both time and intensity if there is a change in the chromatographic conditions in a given sample. Spectral intensities might change, but the masses are fixed because the mass of a compound does not change.

There are two ways to generate spectral data:

- If only one scan is acquired, by default the data is shown as a spectrum.
- From a chromatogram.

A typical spectrum is shown with the molecular weight, labeled with the mass/charge ratio ( $m/z$ ), on the X-axis. The intensity is shown on the Y-axis.

## Analyze Data

When a data file is opened, different panes are shown in windows, depending on the type of experiment performed. The software stores data in files with a wiff extension. Wiff files can contain data for more than one sample. In addition to wiff files, the software can open txt files. However, txt files contain data for only one sample.

The information contained in a data file can be viewed in tabular or graphical form. Graphical data is presented either as a chromatogram or as a spectrum. Data from either of these can appear as a table of data points, and various sorting operations can be performed on the data.

Users can open files containing existing data or data that is currently being acquired. Users can also view all experiment-related data in tabular form. The table pane consists of two tabs, the Data List tab and the Peak List tab. The Data List tab contains experiment-related information, such as acquisition time and scan intensity. The Peak List tab displays peak-related information such as peak height, peak area, and baseline type.

If the data contains results from multiple experiments, users can create individual TICs (Total Ion Chromatograms) for each experiment, and another TIC that represents the sum of all experiments.

The preset TIC that represents the sum of all of the experiments is shown with a splitter tool below the center of the X-axis.

## Open Data Files

---

**Tip!** To turn off the automatic update on the mass spectrum, right-click the mass spectrum and then click **Show Last Scan**. If there is a check mark beside **Show Last Scan**, then the spectrum will update in real-time.

---

1. On the Navigation bar, under **Explore**, double-click **Open Data File**. The Select Sample dialog is shown.
2. In the **Data Files** list, navigate to the data file to open, select a sample, and then click **OK**. The data acquired from the sample is shown. If data is still being acquired, then the mass spectrum, DAD/UV trace, and TIC continue to update automatically.

## Navigate Between Samples in a Data File

---

**Note:** If samples were saved in separate data files, then open each file individually.

---

## Operating Instructions — Analyze and Explore Data

---

For descriptions of the navigation icons used in this procedure, refer to the table: [Table D-4](#).

Open a data file that contains multiple samples and then do one of the following:

- To skip to the next sample in the data file, click the **Show Next Sample** icon (.
- To skip to a non-sequential sample, click the **Go to Sample** icon (.
- In the Select Sample dialog, from the **Sample** list, select the sample to view.
- To go to the previous sample in the data file, click the **Show Previous Sample** icon (.

## View Experimental Conditions

The experimental conditions used to collect data are stored in the data file with the results. The information contains the details of the acquisition method used: the MS acquisition method, that is, the number of periods, experiments, and cycles, including instrument parameters, and the LC device method, including the LC pump flow rate. In addition, it also contains the MS resolution and mass calibration tables used for the sample acquisition. For the software functionality available when the user views the file information, refer to the section: [Show File Information Pane Right-click Menu](#).

---

**Note:** If data is acquired from more than one sample into the same wiff file, then the file information pane does not refresh automatically when the user scrolls through the samples. Close the file information pane and then open it again to view the details for the next sample in the wiff file.

---

Click **Explore > Show > Show File Information**.  
The File Information pane opens below the graph.

---

**Tip!** To create an acquisition method from the File Information pane, right-click the File Information pane and then click **Save Acquisition Method**.

---

## Show Data in Tables

1. Open a data file.
2. Click **Explore > Show > Show List Data**.  
The data is shown in a pane below the graph.

Figure 13-1 Peak List Tab (QTRAP systems)

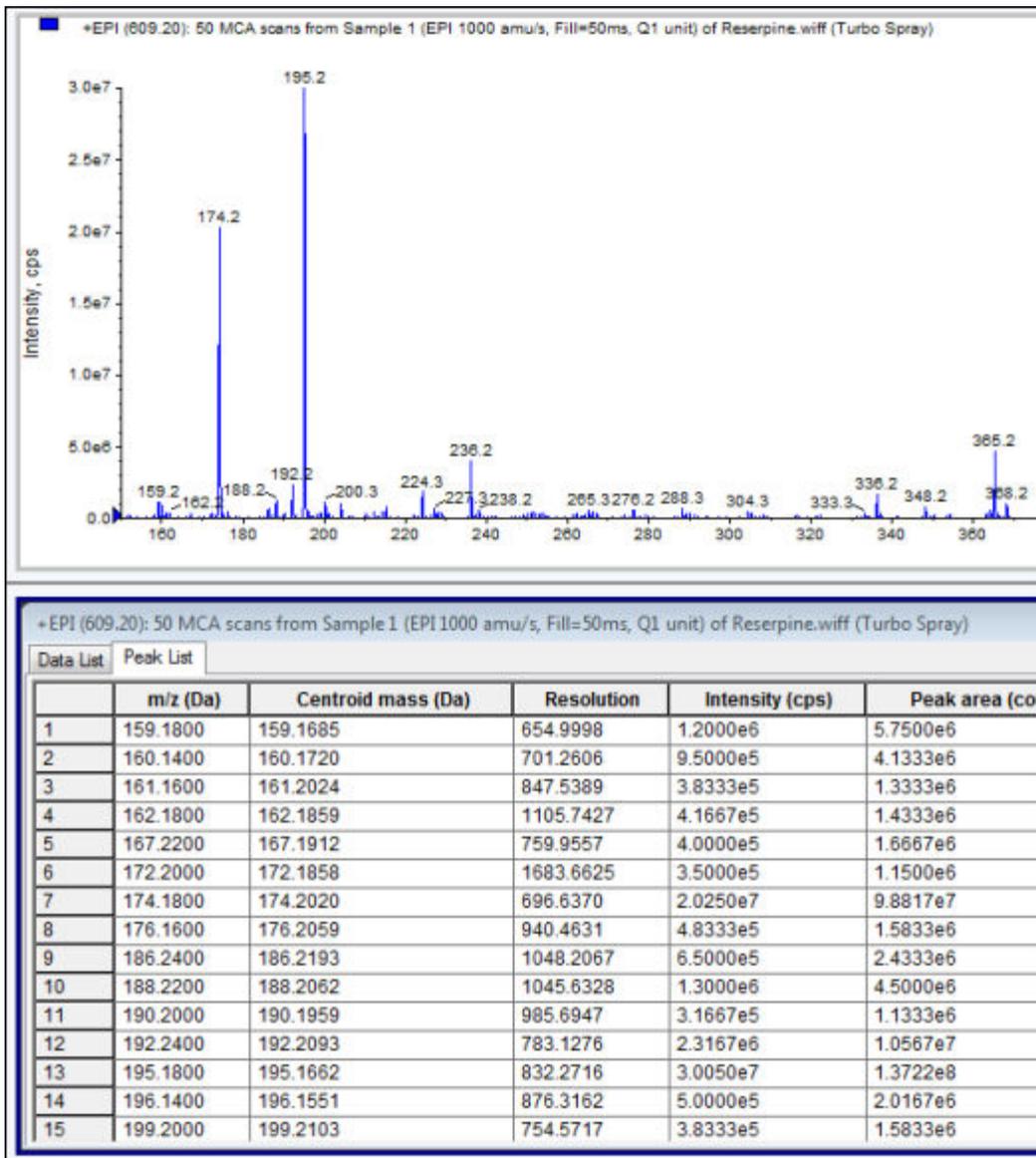


Table 13-1 Right-click Menu for the Spectral Peak List Tab

Menu	Function
Column Options	Opens the <b>Select Columns for Peak List</b> dialog.
Save As Text	Saves the data as a .txt file.
Delete Pane	Deletes the selected pane.

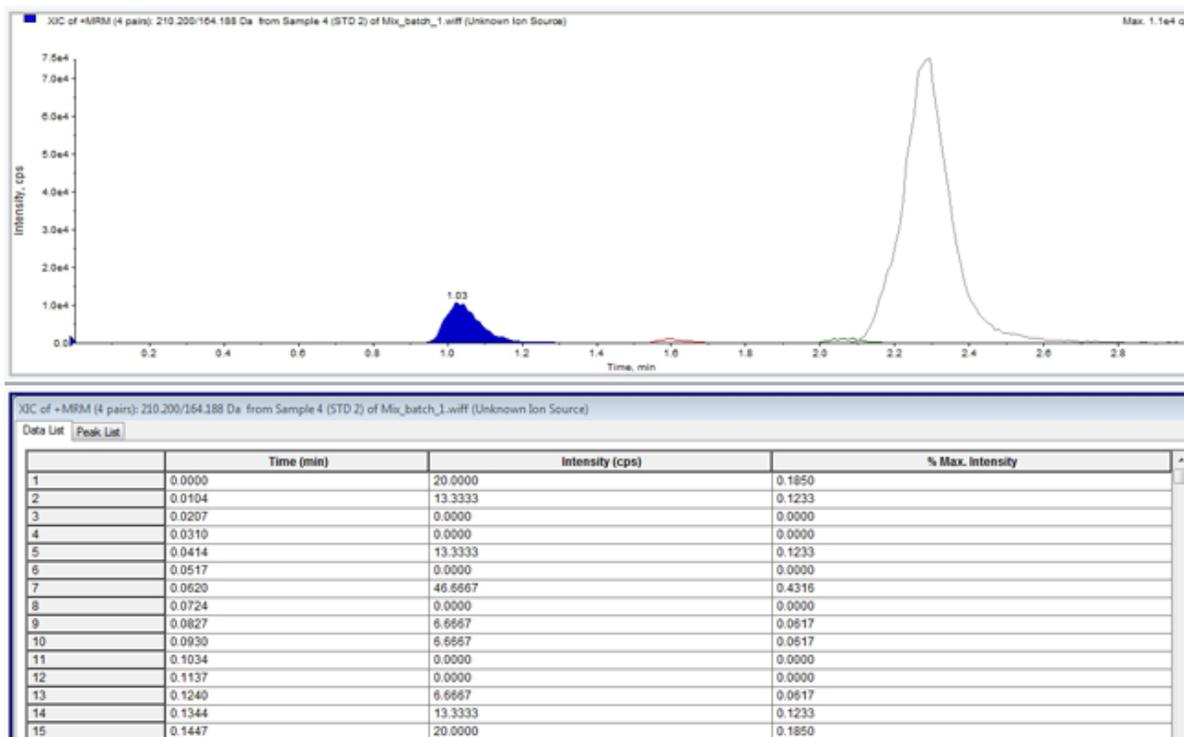
**Table 13-2 Right-click Menu for the Chromatographic Peak List Tab**

Menu	Function
Show Peaks in Graph	Show the peaks in two colors in the graph.
IntelliQuan Parameters	Opens the <b>IntelliQuan</b> dialog.
Save As Text	Saves the data as a txt file.
Delete Pane	Deletes the selected pane.

## Show Basic Quantitative Data

1. Open a data file.
2. Click **Explore > Show > Show List Data**.

**Figure 13-2 List Data**



3. On the Peak List tab, right-click and then select **Show Peaks in Graph**. Peaks are shown in two colors.
4. To change the peak finding algorithm settings, right-click and then select either **Analyst Classic Parameters** or **IntelliQuan Parameters**, whichever is active.

- (Optional) To remove the colored peaks, right-click in the Peak List tab and then clear **Show Peaks in Graph**.

## Spectra

A spectrum is the data that is obtained directly from the mass spectrometer and normally represents the number of ions detected with particular mass-to-charge ( $m/z$ ) values. It is shown as a graph with the  $m/z$  values on the X-axis and intensity (cps) represented on the Y-axis. For more information on how to work with spectra, refer to [Table G-4](#).

In the case of MS/MS data, the intensity is associated with two masses, the precursor ion mass (Q1) and the product ion mass or masses (Q3).

## Chromatograms

A chromatogram is a graphical view of the data obtained from the analysis of a sample. It plots the signal intensity along an axis that shows either time or scan number. For more information about software functionality available for chromatograms, and on using the Chromatogram Panes right-click menu, refer to the section: [Chromatogram Panes](#).

The software plots intensity, in counts per second (cps), on the Y-axis against time on the X-axis. Peaks above a set threshold are labeled automatically. In the case of LC-MS, the chromatogram is often shown as a function of time. For a description of the types of chromatograms, refer to the table: [Table 13-3](#).

For more information about using the available icons, refer to the table: [Table 13-5](#).

**Table 13-3 Types of Chromatograms**

Types of Chromatograms	Purpose
Total Ion Chromatogram (TIC)	<p>A chromatogram generated by plotting the intensity of all ions in a scan against time or scan number.</p> <p>When a data file is opened, it is preset to open as a TIC. If the experiment contains only one scan, then it is shown as a spectrum.</p> <p>If the <b>MCA</b> check box is selected during acquisition of the data file, then the data file opens to the mass spectrum. If the <b>MCA</b> check box is not selected, then the data file opens as the TIC.</p>

**Table 13-3 Types of Chromatograms (continued)**

Types of Chromatograms	Purpose
Extracted Ion Chromatogram (XIC)	An chromatogram created by taking intensity values at a single, discrete mass value, or a mass range, from a series of mass spectral scans. It indicates the behavior of a given mass, or mass range, as a function of time.
Base Peak Chromatogram (BPC)	A chromatogram that shows the intensity of the most intense ion within a scan versus time or scan number.
Total Wavelength Chromatogram (TWC)	A chromatogram created by summing all of the absorbance values in the acquired wavelength range and then plotting the values against time. It consists of the summed absorbances of all ions in a scan plotted against time in a chromatographic pane.
Extracted Wavelength Chromatogram (XWC)	A subset of TWC. An XWC shows the absorbance for a single wavelength or the sum of the absorbance for a range of wavelengths.
Diode Array Detector (DAD)	A chromatogram that shows the absorption spectrum of eluting compounds at one or more wavelengths.

## Show TICs from a Spectrum

To see an example data file, make sure that the `Example` project is selected.

To see an LIT spectrum, open the `LIT` folder, and then open the `Reserpine.wiff` file.

Click **Explore > Show > Show TIC**.

The TIC opens in a new pane.

---

**Tip!** Right-click inside a pane containing a spectrum and then click **Show TIC**.

---

For information on using the **Spectra Panes** right-click menu, refer to the section: [Spectra Panes](#)

## Show a Spectrum from a TIC

A TIC is created by summing the intensity contributions of all ions from a series of mass scans. Use the TIC to view an entire data set in a single pane. It consists of the summed intensities of all of the ions in a scan plotted against time in a chromatographic pane. If the data contains

results from multiple experiments, then a TIC for each experiment can be created below the TIC that represents the sum of all of the experiments.

When a data file is opened, it is preset to be shown as a TIC. However, if the experiment contains only one scan, it is shown as a spectrum. If the user selects the **MCA** check box before acquiring the data file, then the data file opens to the mass spectrum. If the **MCA** check box is not selected, then the data file opens with the TIC.

For information on using the Spectra Panes right-click menu, refer to the section: [Spectra Panes](#).

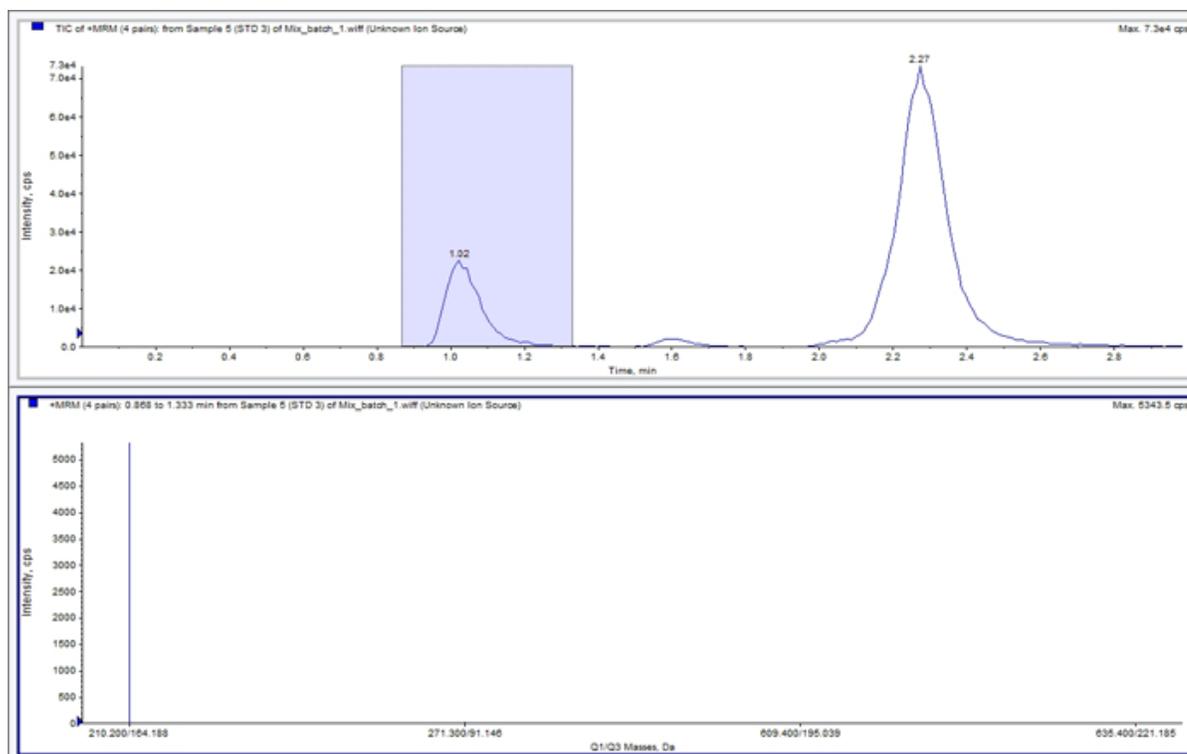
1. In a pane containing a TIC, select a range.
2. Click **Explore > Show > Show Spectrum**.  
The spectrum opens in a new pane.

---

**Tip!** Double-click in the TIC pane at a particular time to show the spectrum.

---

**Figure 13-3 Example of a TIC**



## XICs

An XIC is an extracted ion chromatogram created by taking intensity values at a single, discrete mass value, or a mass range from a series of mass spectral scans. It shows the behavior of a given mass, or mass range, as a function of time. The intensity of the ion, or the summed intensities of all ions in a given range, is plotted in a chromatographic pane.

## Generate XICs

XICs can be generated only from single-period, single-experiment chromatograms or spectra. To obtain an XIC from multi-period or multi-experiment data, split the data into separate panes by clicking the triangle under the X-axis. For more information about using the available icons, refer to the table: [Table 13-5](#).

Several methods are available for extracting ions to generate an XIC, depending on whether chromatographic or spectral data is used. For a summary of methods that can be used with chromatograms and spectra, refer to the following table.

**Table 13-4 Summary of XIC Generation Methods**

Method	Use with Chromatogram	Use with Spectrum	Extraction
Selected range	No	Yes	Extracts ions from a selected area in a spectrum.
Maximum	No	Yes	Extracts ions from a selected area in a spectrum using the most intense peak in the selected area. This option creates an XIC using the maximum mass from the selected spectral range.
Base peak masses	Yes	Yes	Can be used only with Base Peak Chromatograms (BPCs). Use the <b>Use Base Peak Masses</b> command to extract ion results in an XIC with a different colored trace for each mass. If the selection includes multiple peaks, then the resulting XIC will have an equal number of colored traces, one for each mass.
Specified masses	Yes	Yes	Extracts ions from any type of spectrum or chromatogram. Select up to ten start and stop masses for which to generate XICs.

## Generate an XIC Using a Selected Range

1. Open a data file containing spectra.
2. Select a range by pressing the left mouse button at the start of the range, dragging the cursor to the end of the range, and then releasing the left mouse button.  
The selection is shown in blue.
3. Click **Explore > Extract Ions > Use Range**.  
An XIC of the selection opens in a pane below the spectrum pane. The experiment information at the top of the pane contains the mass range and the maximum intensity in counts per second.

## Generate an XIC Using the Maximum Peak

1. Open a data file containing spectra.
2. Select a range in a spectrum.  
The selection is shown in blue.
3. Click **Explore > Extract Ions > Use Maximum**.  
An XIC of the maximum peak specified selection opens below the spectrum pane. The experiment information at the top of the pane contains the mass range and the maximum intensity in counts per second.

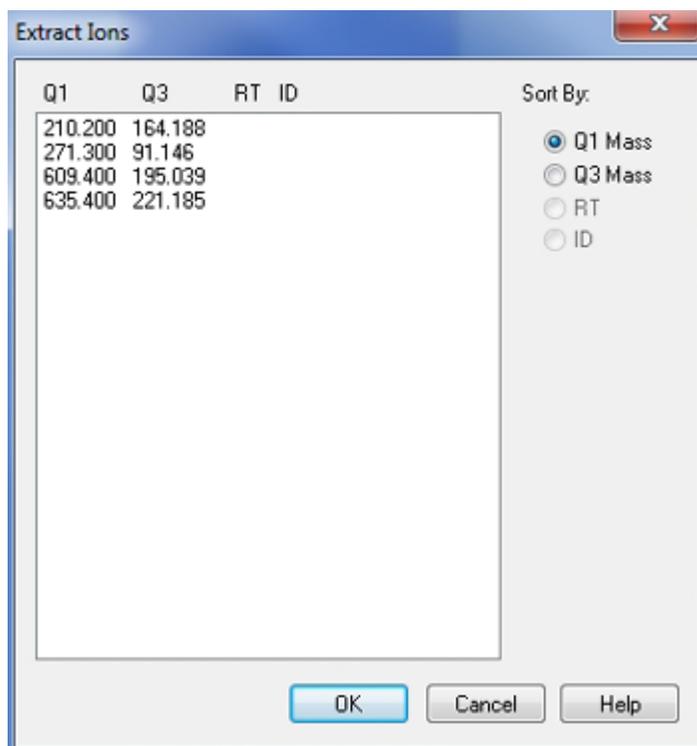
## Generate an XIC Using Base Peak Masses

1. Open a data file containing spectra.
2. In a BPC, select the peak from which to extract ions.  
The selection is shown in blue.
3. Click **Explore > Extract Ions > Use Base Peak Masses**.  
An XIC of the specified selection opens below the spectrum pane. The experiment information at the top of the pane shows the mass range and the maximum intensity in counts per second.

## Extract Ion by Selecting Masses

1. Open a spectrum or chromatogram.
2. Click **Explore > Extract Ions > Use Dialog**.

**Figure 13-4 Extract Ions Dialog**



3. Type the values for each XIC to be created.
  - In the **Start** field, type the start value (lower value) for the mass range.
  - In the **Stop** field, type the stop value (higher value) for the mass range.

---

**Note:** If a stop value is not typed, then the range is defined by the start value.

---

4. Click **OK**.

An XIC of the selection opens below the chromatogram pane. The experiment information at the top of the pane includes the masses and the maximum intensity in counts per second.

## BPCs

A BPC shows the intensity of the most intense ion in every scan as a function of scan number or retention time. It is useful in instances where the TIC is so dominated by noise that there is a large offset and chromatographic peaks are hard to distinguish. It also helps to distinguish between co-eluting components. BPCs are generated only from single period, single experiment data.

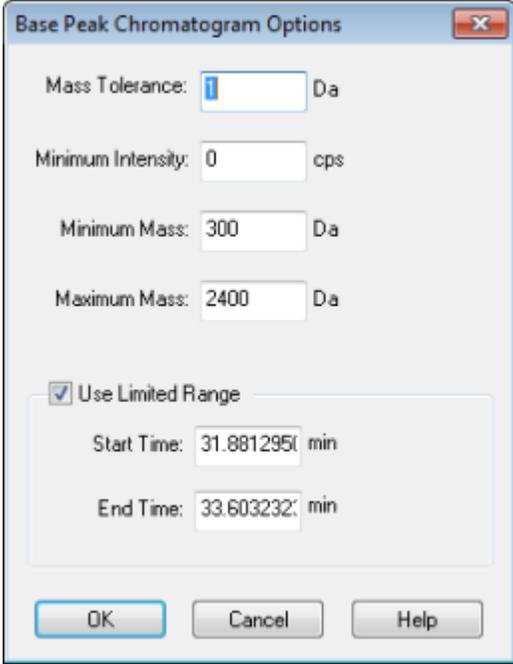
The graph uses two colors, alternating each time the mass of the base peak changes. The color changes are maintained when the data is manipulated by scrolling or zooming. For information about selecting the colors used in the graph, refer to the document: *Help*.

### Generate BPCs

BPCs can be generated only from single-period, single-experiment data.

1. Open a data file.
2. Select an area within a TIC.  
The selection is shown in blue.
3. Click **Explore > Show > Show Base Peak Chromatogram**.  
The selections are shown in the **Start Time** and **End Time** fields.

**Figure 13-5 Base Peak Chromatogram Options**



The screenshot shows a dialog box titled "Base Peak Chromatogram Options" with a close button (X) in the top right corner. The dialog contains several input fields and a checkbox. The fields are: "Mass Tolerance" with a value of 1 and unit "Da"; "Minimum Intensity" with a value of 0 and unit "cps"; "Minimum Mass" with a value of 300 and unit "Da"; "Maximum Mass" with a value of 2400 and unit "Da"; "Start Time" with a value of 31.881295 and unit "min"; and "End Time" with a value of 33.603232 and unit "min". There is a checkbox labeled "Use Limited Range" which is checked. At the bottom of the dialog are three buttons: "OK", "Cancel", and "Help".

4. In the **Mass Tolerance** field, type the value to indicate the mass range used to find a peak. The software finds the peak using a value twice the typed range ( $\pm$  the mass value).
5. In the **Minimum Intensity** field, type the intensity below which peaks are ignored by the algorithm.
6. In the **Minimum Mass** field, type the mass at the beginning of the scan range.
7. In the **Maximum Mass** field, type the mass at the end of the scan range.

- To set the start and end times, select the **Use Limited Range** check box and do the following:
  - In the **Start Time** field, type the time when the targeted range of the experiment starts.
  - In the **End Time** field, type the time when the targeted range of the experiment ends.
- Click **OK**.  
The BPC is generated in a new pane.

## Adjust the Threshold

The threshold is an invisible line drawn parallel to the X-axis of a graph that sets a limit below which the software will not include peaks in a spectrum. The line has a handle, represented by a blue triangle to the left of the Y-axis. Click the blue triangle to view a dotted line that represents the threshold. The threshold can be raised or lowered, but changing the threshold value does not change the data. The software does not label any peaks in the region that lies below the threshold.

- Open a data file.
- Do one of the following:
  - To increase the threshold, drag the blue triangle up the Y-axis.
  - To decrease the threshold, drag the blue triangle down.
  - Click **Explore > Set Threshold**. In the Threshold Options dialog that opens, type the threshold value and then click **OK**.
  - Click **Explore > Threshold**.

The graph updates to show the new threshold. Peak labeling and the peak list are also updated.

---

**Tip!** To view the current threshold value, move the pointer over the threshold handle.

---

## Generate TWCs

A TWC is a less commonly used chromatogram. It shows the total absorbance (mAU) as a function of time. The TWC provides a way of viewing an entire data set in a single pane. It consists of the summed absorbances of all ions in a scan plotted against time in a chromatogram. If the data contains results from multiple experiments, then a TWC for each experiment can be created below the TWC that represents the sum of all of the experiments.

A TWC shows total absorbance (mAU) on the Y-axis plotted against time on the X-axis. For more information about using the available icons, refer to the table: [Table 13-5](#).

1. Open a data file that contains a DAD spectrum.
2. Click **Explore > Show > Show DAD TWC**.  
The TWC is shown in a pane below the DAD spectrum.

---

**Tip!** Right-click inside the pane containing the DAD spectrum and then click **Show DAD TWC**.

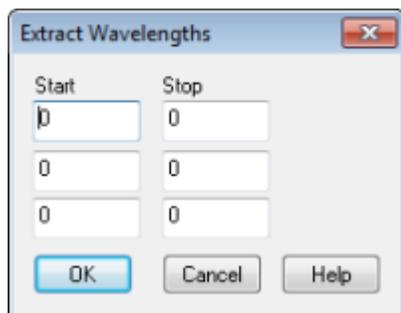
---

## Generate XWCs

An XWC is a wavelength chromatogram created by taking intensity values at a single wavelength, or by taking the sum of the absorbance for a range of several wavelengths. Up to three ranges can be extracted from a DAD spectrum to generate the XWC. For more information about using the available icons, refer to the table: [Table 13-5](#).

1. Open a data file that contains a DAD spectrum.
2. Right-click anywhere in the pane and then click **Extract Wavelengths**.

**Figure 13-6 Extract Wavelengths Dialog**



3. Type **Start** and **Stop** values.
4. Click **OK**.  
The XWC is shown in a pane below the DAD spectrum.

## Show DAD Data

Like mass spectrometer data, DAD data can be viewed in chromatogram or spectrum form. Users can view the DAD spectrum for a single point in time, or for a range of time as a Total Wavelength Chromatogram (TWC).

1. Open a data file containing data acquired with a DAD.  
The TWC, which is analogous to a TIC, opens in a pane below the TIC.
2. In the TWC pane, click a point to select a single point in time, or highlight an area of the spectrum to select a range of time.

## Operating Instructions — Analyze and Explore Data

---

### 3. Click **Explore > Show > Show DAD Spectrum**.

The DAD spectrum opens in a pane below the TWC. The Y-axis shows the absorbance and the X-axis shows the wavelength.

---

**Tip!** If the pane with the TWC is closed, then click a point anywhere in the TWC to open it again. Click **Explore > Show > Show DAD TWC**.

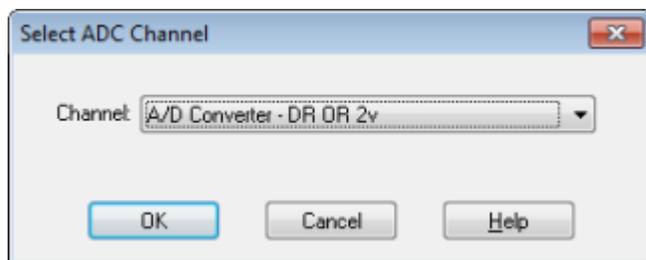
---

## Show ADC Data

Analog-to-digital converter (ADC) data is acquired from a secondary detector, for example from a UV detector through an ADC card, and is useful for comparison with mass spectrometer data. To make ADC data available, acquire the ADC data and the mass spectrometer data simultaneously. Both types of data are then saved in the same file.

1. Make sure that the project folder where the ADC data is stored is selected. For example, click the `Example` folder.
2. On the Navigation bar, under **Explore**, double-click **Open Data File**.  
The Select Sample dialog opens.
3. In the **Data Files** field, double-click the sub-data folder (if applicable), and then click the data file to be opened. For example, in the `Example` folder, double-click **Devices** and then click **Adc16chan.wiff**.
4. In the **Samples** list, select a sample, and then click **OK**.
5. Click **Explore > Show > Show ADC Data**.

**Figure 13-7 Select ADC Channel Dialog**



6. In the **Channel** list, select a channel, and then click **OK**.  
The ADC data is shown in a new pane beneath the active pane.

## Graphical Data Processing

Graphical data can be processed many ways. This section provides information and procedures for using some of the most commonly used tools.

The user can zoom in on part of a graph to view a particular peak or an area in greater detail in both spectra and chromatograms. The user can also zoom in repeatedly to view smaller peaks.

## Manage Data

Use the following menu options or icons to manage data in graphs.

**Table 13-5 Graph Options**

To Do This	Use This Menu Option	Or Click This Icon
Copy a graph to a new window	Select the graph to copy. Click <b>Explore &gt; Duplicate Data &gt; In New Window.</b>	
Rescale a graph to its original size	Select the graph. Click <b>Explore &gt; Home Graph.</b>	
Move a pane	<ul style="list-style-type: none"> <li>Select the graph. Click <b>Window &gt; Move Pane.</b></li> <li>Select the pane or window and then drag it to the new position. This position can be inside the same window or within another window.</li> </ul> <p>A four-headed arrow is shown when the cursor is on the boundary of the active window or pane.</p> <ul style="list-style-type: none"> <li>If the pane is at the top or bottom of the target pane, then the pane moves above or below that pane, respectively.</li> <li>If the pane is at the left or right of the target pane, then the pane moves to the left or right of that pane, respectively.</li> <li>If the pane is at any other position, then the pane moves to the target row. The drop shadow of the pane as the pane is moved indicates its new position.</li> </ul>	
Link panes	<ol style="list-style-type: none"> <li>With the two graphs open, click one to make that pane active.</li> <li>Click <b>Explore &gt; Link</b> and then click the other pane.</li> </ol>	
Remove linking	Close one of the panes. Click <b>Explore &gt; Remove Link.</b>	

Table 13-5 Graph Options (continued)

To Do This	Use This Menu Option	Or Click This Icon
Delete a pane	Select the graph. Click <b>Window &gt; Delete Pane.</b>	
Lock a pane	Select the graph. Click <b>Window &gt; Lock Panes.</b>	
Hide a pane	Select the graph. Click <b>Window &gt; Hide Pane.</b>	
Maximize a pane	Select the graph. Click <b>Window &gt; Maximize Pane.</b>	
Tile panes	Select the graph. Click <b>Window &gt; Tile all Panes.</b>	

## Zoom in on a Graph

Zoom in on part of a graph to view a particular peak or area in greater detail in both spectra and chromatograms. Zoom in repeatedly to view smaller peaks.

### Zoom In on the Y-axis

1. Move the pointer to the left of the Y-axis on either side of the area to be expanded and then drag away from the starting point in a vertical direction while holding the left mouse button. A box is drawn along the Y-axis representing the new scale.

---

**Note:** Take care when zooming in on the baseline. Zoom in too far and the zoom-in box closes.

---

2. Release the mouse button to draw the graph to the new scale.

---

**Tip!** To return the Y-axis of the graph to the original scale, double-click either axis. To return the entire graph to the original scale, click **Explore > Home Graph.**

---

### Zoom In on the X-axis

1. Move the pointer under the X-axis to either side of the area to be expanded and then drag away from the starting point in a horizontal direction while holding the left mouse button.
2. Release the mouse button to draw the graph to the new scale.

---

**Tip!** To return the X-axis of the graph to the original scale, double-click the X-axis. To return the entire graph to the original scale, click **Explore > Home Graph.**

---

## Label Graphs

The preset style for labels on graphs and chromatograms can be customized. Select the fonts to use for peak and axis labels, and the colors to use for the traces. Add axis labels and the type of label and precision for the peaks.

### Add Captions to a Graph

Use captions to label peaks of interest or significant points on the graph. A caption placed beside a peak stays with the peak when the peak is zoomed in or out. Captions also stay with the original sample when the user navigates between samples in a data file. A caption contains one line of text, with a maximum of 128 characters.

1. On the spectrum, right-click, and then click **Add Caption**.  
The Add Caption dialog opens.
2. In the **Caption** box, type the text.
3. To change the size and style of the caption, click **Font**.
4. To place the caption, click **OK**.

---

**Tip!** To change the position of the caption, drag it to a different position. The caption stays in the same place relative to the X- and Y-axes when the peak is zoomed in or out. To edit or delete the caption, right-click the caption and then click the appropriate command.

---

### Add Text to a Graph

Use text to add multiple lines of information to a graph. Unlike captions, which are associated with a specific peak and move with it as the user zooms the graph, text labels remain in their original location as the user zooms the graph. They do not stay with the original sample when the user navigates between samples in a data file.

1. On the graph, right-click and then click **Add User Text**.  
The Add User Text dialog opens.
2. In the **User Text** field, type the text.
3. To center the text, select the **Center Text** check box.
4. To change the size and style of the caption, click **Font**.
5. To insert the text, click **OK**.

---

**Tip!** To change the position of the text, drag the text to a different position. To edit or delete the text, right-click the text and then choose the appropriate command.

---

## Overlay and Sum Spectra or Chromatograms

Overlay graphs created using similar methods to visually compare two or more sets of data. Each individual spectrum is distinguished by the color of its trace. For full scan data, this allows the user to visualize the differences between several sample spectra.

After two or more graphs are overlaid, sum the graphs to get a new trace. Each point on the new trace is the sum of the points from the graphs. Summing several overlays of similar data type can make subsequent processing operations easier and faster. For example, overlay several XICs, sum them, and then smooth the summed overlay to remove noise.

Summing overlays is similar to generating a TIC with the benefit of being able to choose which graphs to overlay. For example, if the user is looking at ten experiments, the TIC will add all ten experiments together. If the overlays are summed, then the user has the option of adding only nine of the ten overlaid graphs. This is useful if the data collected in the one experiment is just noise.

### Overlay Graphs

Two or more sets of data can be visually compared by overlaying graphs created by similar methods. Each individual spectrum is distinguished by the color of its trace. For full scan data, this allows users to visualize the differences between several sample spectra.

If one or more panes are chosen, then each XIC opens in a separate pane.

---

**Tip!** To overlay fewer than four graphs in the same pane, press **Ctrl**, right-click in a pane, and then click **Appearance Options**. In the Appearance Options dialog, on the Multiple Graph Options tab, select **Yes** for the **Overlay Multiple Panes** fields for **Spectrum** and **Chromatogram**.

---

1. Select the first pane to be overlaid.
2. Click **Explore > Overlay**.
3. Click in the second pane.

The graphs are overlaid showing the two traces in different colors.

---

**Tip!** To view a color-coded list of the overlaid graphs, right-click the title bar of the pane.

---

### Cycle Between Overlaid Graphs

1. Select a pane that contains overlaid graphs.
2. Click **Explore > Cycle Overlays**.

The view changes so that the next graph in the sequence is shown in the foreground.

## Sum Overlays

1. Overlay the graphs to sum.
2. Click **Explore > Sum Overlays**.  
The overlaid graphs are added together.

**Table 13-6 Explore Toolbar Quick Reference: Overlaying Graphs**

Icon	Name	Function
	Home Graph	Click to return the graph to the original scale.
	Overlay	Click to overlay graphs.
	Cycle Overlays	Click to cycle between overlaid graphs.
	Sum Overlays	Click to add the graphs together.

## Perform Background Subtractions

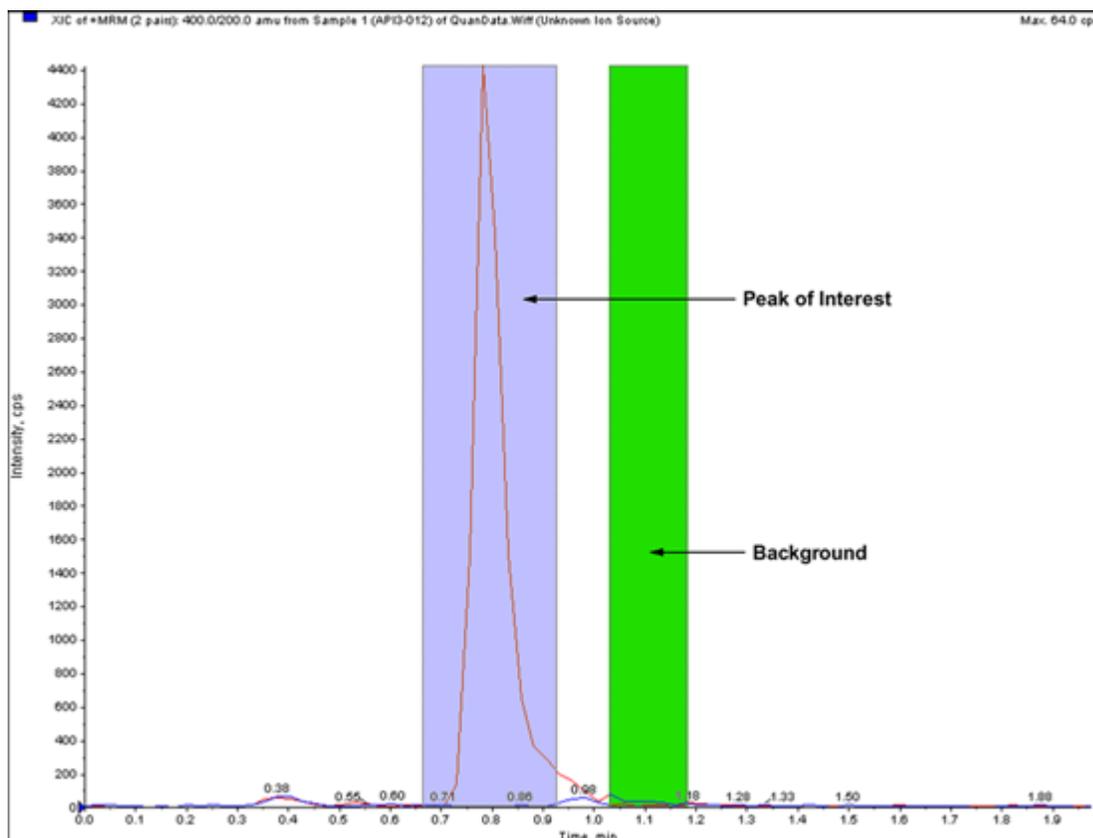
Background subtraction reduces the amount of noise in a spectrum by subtracting either one or two ranges that contain noise from a range that contains a peak. Move the ranges independently, or lock them and then move them as a single entity within the graph. Locked Background Subtraction is the preset setting.

**Background Subtract:** Use background subtract to isolate a peak of interest. Highlight and then subtract up to two selected ranges from the peak. Lock the ranges and then move them within the graph to optimize peak isolation, or to isolate another peak.

### Perform a Background Subtraction from a Spectrum

1. Open a data file.
2. Select a background range.

**Figure 13-8 Background Range Selected**



3. Hold down the **Shift** key and then select another background range.
4. To set the subtract range, click **Explore > Background Subtract > Set Subtract Range**.
5. Select the peak of interest.
6. Click **Explore > Background Subtract > Perform Background Subtract**.  
The background is subtracted from the peak and a new spectrum is generated.
7. To isolate another peak, drag the locked ranges in the chromatogram and repeat the background subtract.

---

**Note:** To clear the background subtract region, click **Explore > Background Subtract > Clear Subtract Range**.

---

8. To save the background subtracted spectrum as a processed data file, click **File > Save**.

## Unlock the Ranges

The selected subtraction range is set to locked.

- Click **Explore > Background Subtract > Subtract Range Locked**.

The ranges are unlocked and you can move each one independently.

**Table 13-7 Explore Toolbar Quick Reference: Background Subtract**

Icon	Name	Function
	Perform Background Subtract	Click to perform a background subtract after the background ranges are selected.
	Subtract Range Locked	Click to lock the selected background ranges. Unlock the background ranges to move each range independently.

## Smoothing Algorithms

Smoothing a data set removes local variations that are most likely due to noise. Several smoothing cycles can be applied to the data, but users can only undo the most recent smooth. Smoothing is not available for MI/MRM spectra. Choose the smooth algorithm or the Gaussian smoothing algorithm as the preset smoothing method.

### Smooth Algorithm

When this algorithm is used to smooth data, the point weighting values are set for three data points: the current data point, the preceding data point, and the following data point. The smooth algorithm multiplies the data points by the assigned weighting values, sums these values, and then divides the total by the sum of the point weight values. It is a gentler smooth than the Gaussian algorithm, and it takes a long time to smooth very noisy data.

### Gaussian Smoothing Algorithm

Gaussian smoothing involves replacing each data point with the weighted average of a number of data points on either side of it. The weighting for each new data point is calculated on the basis of a Gaussian curve. It is a coarser smooth than the smooth algorithm, but it is good for smoothing very noisy data.

You set two values when using the Gaussian smoothing method:

**Gaussian filter width (% of minimal distance between points):** This value shows the width used to calculate the weighting of neighboring points. The width is described in terms of percentages of the distance between two points in the scan, where the preset width of 100% gives a distribution that is as wide as the distance between data points.

**Limit of Gaussian filter (number of minimal distance between points):** This value corresponds to the limits of the Gaussian curve, shown in multiples of the distance between points. For example, the preset value of 10 creates a Gaussian curve that truncates after ten data point widths on either side of the center.

## Smooth Data

You can choose the Analyst MD software smoothing method or the Gaussian smoothing method.

---

**Tip!** To undo smoothing, click **Edit > Undo**. The software supports one level of undo.

---

## Smooth Data using the Smooth Algorithm

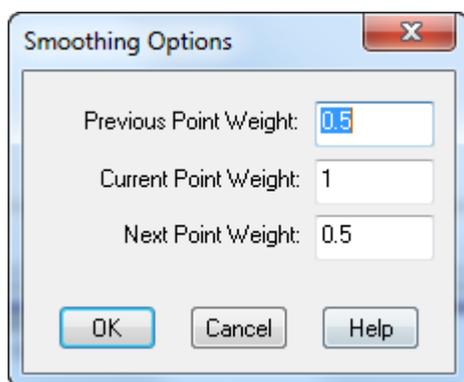
---

**Tip!** To undo smoothing, click **Edit > Undo**. The software supports one level of undo.

---

1. Select a pane containing a chromatogram or spectrum.
2. Click **Explore > Smooth**.  
The Smoothing Options dialog opens.

**Figure 13-9 Smoothing Options Dialog**



3. In the **Previous Point Weight** field, type the weighting factor to be applied to the previous data point.
4. In the **Current Point Weight** field, type the weighting factor to be applied to the center data point.
5. In the **Next Point Weight** field, type the weighting factor to be applied to the following data point.
6. Click **OK**.

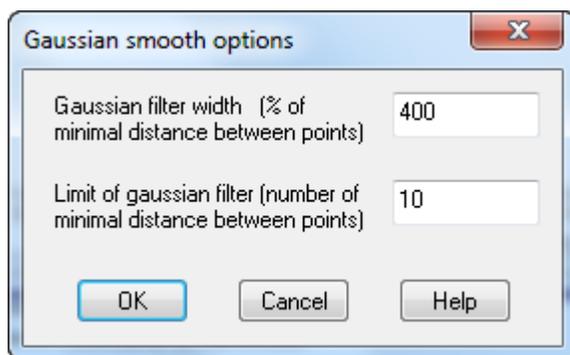
The data set is smoothed, replacing the current data set in the pane.

## Smooth Data using Gaussian Smoothing

1. Select a pane containing a chromatogram or spectrum.
2. Click **Explore > Gaussian Smooth**.

The Gaussian smooth options dialog is shown.

**Figure 13-10 Gaussian smooth options dialog**



3. In the **Gaussian filter width** field, type the width used to find the weighting of neighboring points as a percentage of the distance between the two points.
4. In the **Limit of gaussian filter** field, type the limit of the Gaussian curve, given in multiples of the distance between points.
5. To smooth the data, click **OK**.

The data set is smoothed, replacing the current data set in the pane.

**Table 13-8 Explore Toolbar Quick Reference: Smoothing Data**

Icon	Name	Function
	Smooth	Click to smooth data using the smooth algorithm.
	Gaussian smooth	Click to smooth data using Gaussian smoothing.

## Centroid Data

Centroiding converts peak distribution values into a single value of  $m/z$  and intensity that represents the peak. Centroiding data collected in profile mode simplifies the data and reduces

## Operating Instructions — Analyze and Explore Data

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the file size. Centroiding provides more accurate peak assignment and reduces the amount of data, but it also removes the information about the peak shape.

The centroiding algorithm converts peaks to single values by using an intensity weighted average to calculate the center of gravity of the peak. The output of the algorithm is a list of peaks with parameters, as shown in the table: [Table 13-9](#).

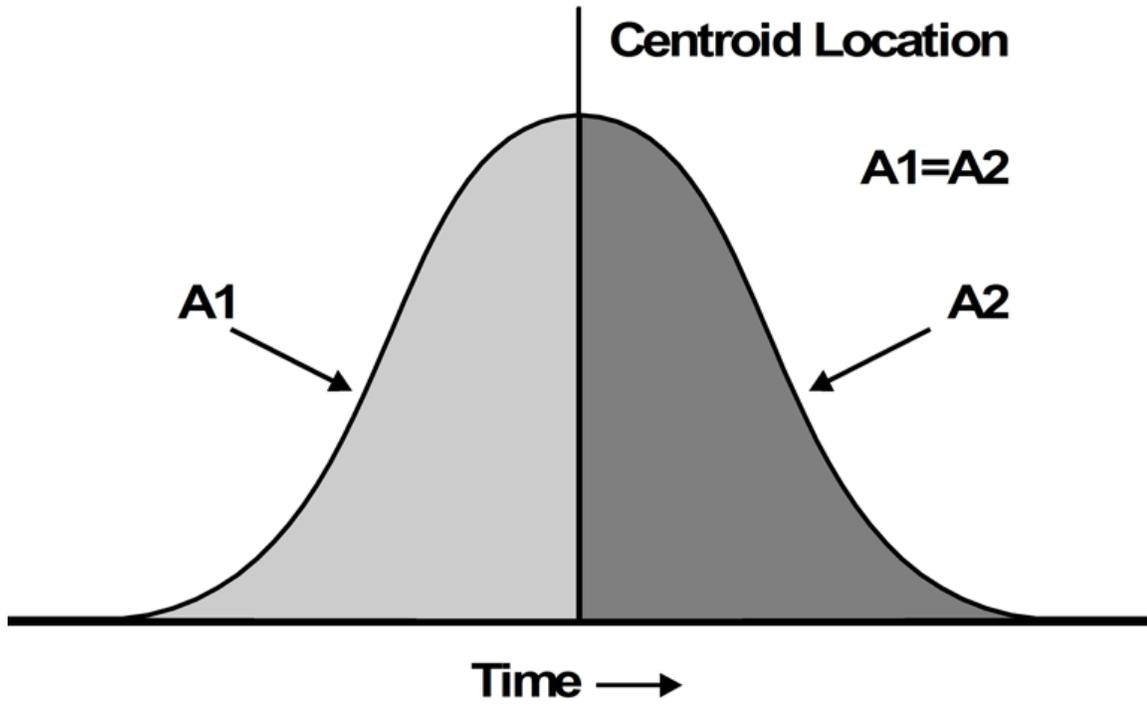
**Table 13-9 Peak Parameters**

Parameter	Definition
Centroid Value	The value of the centroided data in units of mass or time.
Intensity	The intensity of each peak in cps.
Width	The width of the centroided peak in amu.

Data is automatically centroided when added to a library or when a search is conducted.

1. Select a pane containing a spectrum.  
Centroiding data changes the appearance of the existing graph. To compare the result with the original data, make a copy of the graph before centroiding.
2. Click **Explore > Centroid**.  
The data is centroided.

Figure 13-11 Analyte Centroid Location



Icon	Name	Function
	Centroid	Click to centroid data.

## Save and Open Processed Data Files

Users can save processed data, such as specific layouts and captions, that can be re-opened in Explore mode only. These files also contain relevant history information and are similar to data files except they will contain only the data from the active pane in Explore. These files have the pdt extension and are stored in the Data folder in the current project.

### Save a Processed Data File

1. Select the pane of data to be saved.
2. Click **File > Save Processed Data File**.
3. In the **File name** field, type a name.

4. Click **Save**.

### Open a Processed Data File

1. In Explore mode, click **File > Open Processed Data File**.  
The Load Processed Data File dialog opens.
2. Select a file and then click **Open**.

## Work with Contour Plots

A Contour Plot is a color-coded plot of a complete data set that uses color to represent a third dimension in the plot. In a Contour Plot of a TIC, the X-axis represents retention time or scan number, the Y-axis represents mass, and the color represents the intensity of the data at that point. In a Contour Plot of a TWC for DAD data, the X-axis represents retention time or scan number, the Y-axis represents wavelength, and the color represents absorbance. Contour Plot is a post-acquisition tool that does not function in a real-time scan acquisition.

---

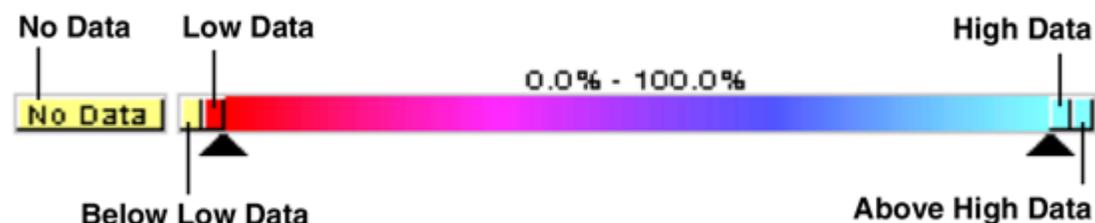
**Note:** Contour Plot does not support MI or MRM scans, but it does support DAD scans.

---

Color is the third axis in Contour Plot, and it represents either intensity or absorbance. Change the high and low intensity or absorbance values in Contour Plot using the control triangles on the color bar above the Contour Plot. The percentage parameters at the top of the Contour Plot pane indicate the values held by the low and high sliders. The actual values are based on a percentage of the maximum intensity or absorbance within the selected area. The value is shown in the top right corner of the Contour Plot pane.

The controls shown in [Figure 13-12](#) change the colors in a Contour Plot.

**Figure 13-12 Buttons Controlling Contour Plot Colors**



Define the colors on a Contour Plot graph to provide better contrast and show data specifications according to requirements. For example, setting the intensity/wavelength and changing the color of the values for Below Low Data and Above High Data can eliminate background noise in Contour Plot.

The Below Low Data and Above High Data buttons shrink and expand on the color bar if the slider controls are moved. When the contour plot colors are changed, the new colors become the preset colors for all subsequent graphs.

### View a Contour Plot

Users can view a Contour Plot from TIC, XIC, TWC, or XWC graphs only after acquisition. TICs and XICs are available for all wiff data files. TWCs and XWCs are available only for data acquired by a DAD.

1. In **Explore** mode, open a data file as a TIC, XIC, TWC, or XWC graph.
2. Highlight the range to view in the Contour Plot. If a selection is not made, then the entire range is viewed.
3. Click **Explore > Show > Show Contour Plot**.  
A Contour Plot of the selected area is shown in a separate pane.

### Select an Area in a Contour Plot

Users can zoom in on a particular selection or view the corresponding mass spectrum for that selection.

Do one of the following:

- To select a standard area within a box, drag the pointer to create a box around an area in the Contour Plot.
- To make a vertical selection, press Ctrl and drag the pointer vertically.
- To make a horizontal selection, press the space bar and drag the pointer horizontally.

### Set the Intensity and Absorbance in a Contour Plot

Do one of the following:

- To set the low intensity/absorbance value in Contour Plot, drag the left triangular slider from the color bar above the Contour Plot to the required position.

Contour Plot automatically adjusts the color of values below the setting to indicate they are outside the range.

- To set the high intensity/absorbance value in Contour Plot, drag the right triangular slider from the color bar above the Contour Plot to the required position.

Contour Plot automatically adjusts the color of values above the setting to indicate they are outside the range.

## Change Colors in a Contour Plot

1. In the Contour Plot pane, click one of the color buttons.  
The Color dialog opens.
2. Click a color, and then click **OK**.

The graph changes to reflect the color change.

---

**Tip!** Use the **Define Custom Colors** palette to create customized colors for use in a Contour Plot.

---

## Fragment Interpretation

The Fragment Interpretation tool helps the user interpret MS/MS data. The Fragment Interpretation Tool generates a list of theoretical fragment masses from single, non-cyclic bond cleavage of a molecular structure. The molecular structure can be created in a third-party drawing program and then saved as a mol file. The tool can then match the theoretical list with peaks in the current mass spectrum. Fragment Interpretation shows the theoretical fragments in the fragment list and compares the fragment masses to peaks in the mass spectrum. Peaks above the threshold intensity and within the user-defined mass tolerance (maximum 2 Da) of fragment masses are considered matched and are shown in bold text in the fragment list.

---

**Note:** The Fragment Interpretation tool cannot be used with the following scan types:

- Precursor Ion
  - Neutral Loss
  - Q1 Multiple Ion
  - Q3 Multiple Ion
  - Multiple Reaction Monitoring (MRM)
- 

## Work with the Fragment Interpretation Tool

If multiple spectrum panes are viewed, then the Fragment Interpretation tool connects to the active spectrum. If the data file contains more than one sample, then the Fragment Interpretation tool connects to the active spectrum.

The tool automatically calculates the single, non-cyclic bond cleavage fragments from a mol file. When the Fragment Interpretation tool is connected to a spectrum, theoretical fragments in bold text, indicate a matching peak in the spectrum within the specified mass tolerance and intensity threshold.

When a single, non-cyclic bond is selected in the molecular structure, the Fragment Interpretation tool highlights the two fragments created when the bond is cleaved, and then shows matching peaks in the connected spectrum.

### Connect the Fragment Interpretation Tool to a Spectrum

If a spectrum is open when the Fragment Interpretation tool is opened, then the active panel links to the open spectrum automatically.

1. Click **Explore > Show > Show Fragment Interpretation Tool**.
2. From the lower right corner of the Fragment Interpretation pane, click the connect button. The pointer changes to the connecting tool.
3. Click the spectrum graph to be connected to the Fragment Interpretation tool. The connected graph indicator in the lower left corner contains the name of the graph connected to the Fragment Interpretation pane. The connection is broken when either the graph or Fragment Interpretation is closed. If the connected wiff file has more than one sample, then the Fragment Interpretation pane updates automatically as the user scrolls through the samples.

### Match Fragments with Peaks

1. Click **Explore > Show > Show Fragment Interpretation Tool**.
2. With a mol file in the Fragment Interpretation pane, select a cell in the **Fragment List** that is shown in bold.

In the spectrum, the software highlights the matching spectral peak in the color selected under the **Options** tab. In the molecular structure, the bond is highlighted.

If a row that has more than one matching fragment is clicked, then the spectral peak that is closest to its monoisotopic mass is highlighted in the mass spectrum in the color specified in the **Options** tab.

### Select a Bond in a Molecular Structure

1. Click **Explore > Show > Show Fragment Interpretation Tool**.
2. With a mol file opened in the Fragment Interpretation pane, click a single, non-cyclic bond in the molecular structure.

The two resulting fragments are shown as highlights in the fragment list. The masses of the two fragments are shown on either side of the bond.

If a spectrum is connected, then the Fragment Interpretation tool shows any matching peaks in the graph. If a fragment in the list is selected and the fragment is matched to a peak, then the Fragment Interpretation window zooms in on that peak.

### View Isotopes

The Fragment Interpretation tool can show the theoretical isotopic distribution for a peak matching a fragment in the fragment list.

1. Click **Explore > Show > Show Fragment Interpretation Tool**.
2. In the Fragment Interpretation pane, click the **Options** tab.
3. Select the **Show Isotopes** check box.
4. Click **Apply**.
5. From the **Fragment List**, select a fragment that matches a peak.  
The isotopic distribution for matched peaks is shown in the spectrum.

### Show Formula Differences for Fragments

The formula and monoisotopic mass difference between two related hypothetical fragments can be shown in the software. The formula difference is shown when two peaks are selected. The formula and monoisotopic mass difference is shown when two fragments are selected, or when two single, non-cyclic bonds are selected.

#### Show a Formula Difference in a Spectrum

1. Click a fragment peak.
2. Press **Shift** and then click another fragment peak.  
If the formula difference is equal to a fragment from the fragment list, the fragment highlights in the list. Otherwise, the formula difference between the matching fragments of the peaks is shown in a message box.

#### Show a Formula Difference in the Fragment List

1. Click the row number for one fragment.
2. Press **Shift** and then click another fragment peak.  
The formula and monoisotopic mass difference is shown in a message box if the fragments are related.

#### Show a Formula Difference in a Molecular Structure

1. Click a single, non-cyclic bond. The default fragment (of the two highlighted fragments) is selected. To select the other fragment of the cleaved bond, press **Ctrl** and then click the bond.

2. Select a second non-cyclic bond. To select the default fragment, press **Shift** and then click the bond. To select the other fragment of the cleaved bond, press **Ctrl+Shift** and then click the bond.

Fragment Interpretation calculates the formula and monoisotopic mass difference between the fragment selected in step 1 and the fragment selected in step 2, if the fragments are related. The formula and monoisotopic mass difference is shown in a message box.

## Library Databases

The Library Search feature compares unknown spectra to known MS spectra contained in the library database and generates a list of possible matches.

Use Library Search to do the following:

- Compare library contents against an unknown spectrum.
- Add records to the library.
- Edit existing records.

Library data is stored in the following locations:

- MS Access on a local database
- MS SQL Server

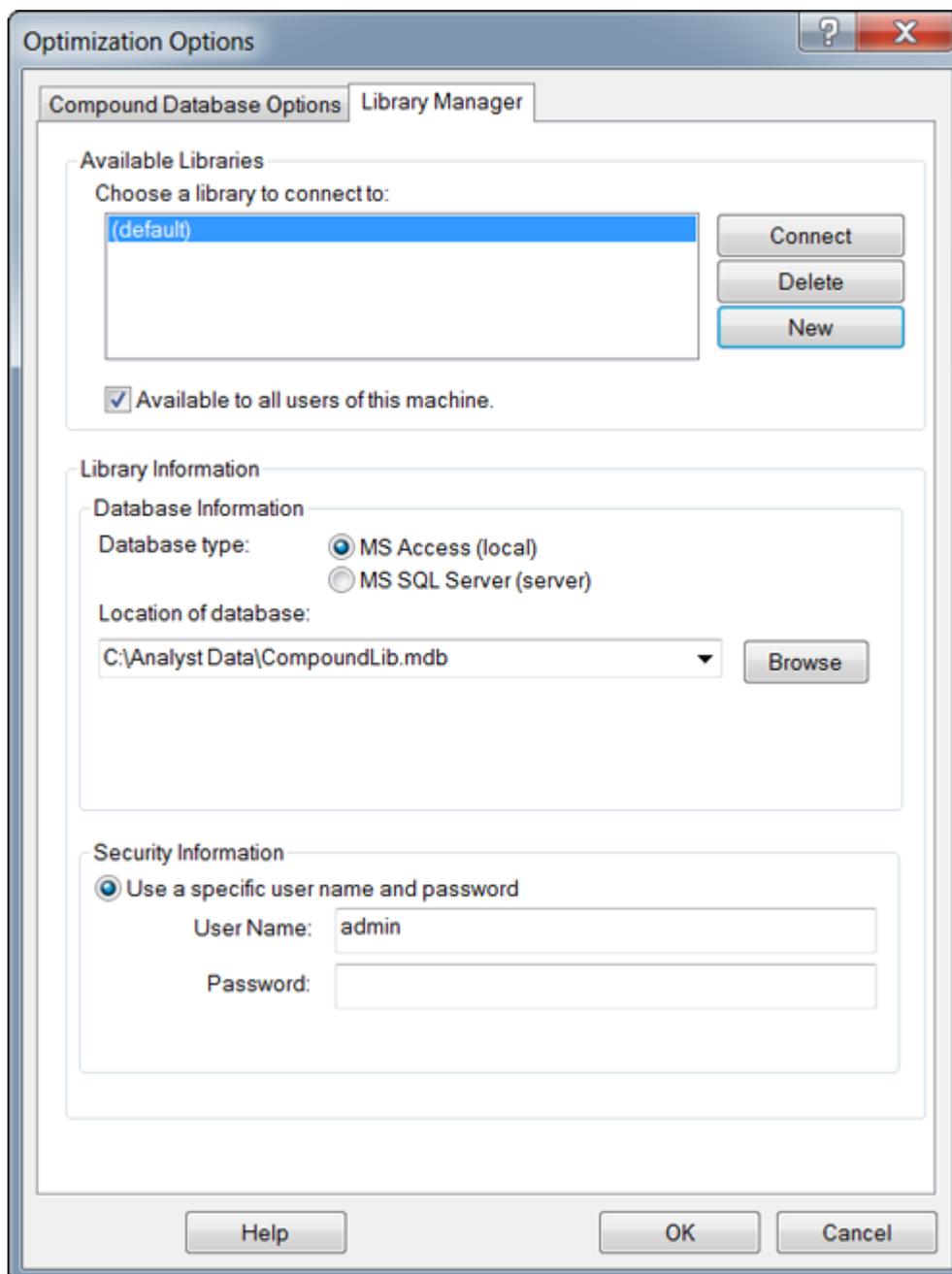
Before using the Library Search feature, locate where the library data is stored and then connect the computer to that location. Library databases can be stored locally on the computer or on a server and accessed over a network.

## Switch between Existing Library Databases

Users can connect to any databases that have aliases that are already set up.

1. Click **Tools > Settings > Optimization Options**.  
The Optimization Options dialog opens.
2. Click the Library Manager tab.

Figure 13-13 Library Manager Tab



3. In the **Available Libraries** section, click the alias of the database to connect to and then click **Connect**.
4. To allow other users to access the database, select the **Available to all users of this machine** check box.

5. Click **OK**.

## Connect to a Local Library Database

1. Click **Tools > Settings > Optimization Options**.  
The Optimization Options dialog opens.
2. Click the Library Manager tab.
3. In the **Available Libraries** section, click **New**.

**Figure 13-14 Add Library Dialog**

**Add Library**

Library Information

Enter a Name for the Library

Database Information

Database type:  MS Access (local)  
 MS SQL Server (server)

Enter the location of the database:

Browse

Security Information

Use a specific user name and password

User Name:

Password:

Save Cancel

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4. Type a name for the library.
5. In the **Database Information** section, select **MS Access (local)**.
6. Type the database location.
7. In the **Security Information** section, if a user name and password are required to access this database, then type the user name and password.
8. Click **Save**.

## Connect to a Server Library Database

1. Click **Tools > Settings > Optimization Options**.  
The Optimization Options dialog opens.
2. Click the Library Manager tab.
3. In the **Available Libraries** section, click **New**.
4. Type a name for the library.
5. In the **Database Information** section, select **MS SQL Server (server)**.

Figure 13-15 Add Library Dialog

The screenshot shows a dialog box titled "Add Library" with a close button (X) in the top right corner. The dialog is organized into three main sections:

- Library Information:** Contains a text input field with the label "Enter a Name for the Library".
- Database Information:** Contains two radio buttons for "Database type": "MS Access (local)" and "MS SQL Server (server)". The "MS SQL Server (server)" option is selected. Below this is a dropdown menu labeled "Enter the name of the database server:" with a "Refresh" button to its right. Below the dropdown is a text input field labeled "Enter the name of the database on the server:".
- Security Information:** Contains two radio buttons: "Use Windows integrated security" and "Use a specific user name and password". The "Use a specific user name and password" option is selected. Below these are two text input fields labeled "User Name:" and "Password:".

At the bottom right of the dialog are two buttons: "Save" and "Cancel".

6. Type the name of the database server.
7. Type the name of the database.
8. Do one of the following in the **Security Information** section:
  - If a specific user name and password are required to access this database, then type the user name and password.
  - To use Windows security, select the **Use Windows integrated security** option.
9. Click **Save**.

## Work with Library Records

If the library contents are searched without constraints, then all records are listed. If the library contents are searched with constraints, then only those records that match the specified constraints are listed. The number of records shown depends on the number of constraints selected. If many restraints are selected, then few records are listed.

### View all Library Records

Click **Explore > Library Search > List**.

The Librarian dialog opens with all records in the database.

### Search Library Records with Constraints

1. Click **Explore > Library Search > List With Constraints**.

**Figure 13-16 List Constraints Dialog**

The screenshot shows a dialog box titled "List Constraints". At the top, there is a "Conditions" section with three fields: "Field Name:" (a dropdown menu showing "Formula"), "Relation:" (a dropdown menu showing "Contains"), and "Value:" (an empty text box). Below these fields is a large empty rectangular area. To the right of this area are several buttons: "Add", "Modify", "Remove", "Group", "Ungroup", and "Or". At the bottom of the dialog, there are two tables. The first table is labeled "Elements Included:" and has columns "Element", "Min.", and "Max." with rows numbered 1, 2, and 3. The second table is labeled "Excluded:" and has a column "Element" with rows numbered 1, 2, and 3. On the right side of the dialog, there are three buttons: "Help", "Cancel", and "List".

2. In the **Field Name** list, select a field on which you want to base a constraint.

3. In the **Relation** list, select the relation (operator) that applies to the field name.
4. In the **Value** field, type the value of the field name based on the relation.
5. To add the selected constraint to the **Conditions** list, click **Add**.
6. Continue adding constraints to the conditions list as required.
7. Coupling distinct constraints within the **Conditions** list creates more specific conditions that enhance the search.
  - To group constraints, select the constraints and then click **Group**.
  - To separate grouped constraints, click the group, and then click **Ungroup**.
8. To change the relationship between constraints, click the relationship, and then click **And** or **Or**.
9. To exclude compounds containing a certain number of atoms of specific elements, select or type the elements in the **Elements Included** table, and then type a minimum and maximum number of atoms of the element.

---

**Note:** Element symbols are case-sensitive. For example, Hydrogen is H, not h and Sodium is Na, not NA or na.

---

10. To exclude compounds containing certain elements, select or type the elements in the **Excluded** table.
11. To search for compounds fitting your criteria, click **List**.  
Records that match all the constraints are shown in the Records table. Listing constraints are saved.

### Add a Record to the Library

1. Right-click an active spectrum, and then click **Add a Record**.  
The spectrum will be centroided automatically if it has not been centroided already.  
The Add a Record dialog opens with data from the spectrum.
2. On the **Mass Spectral Information** tab, type a name in the **Compound Name** field. The compound name is mandatory and must uniquely identify the compound within the library.
3. Edit any of the other fields. Many of the fields are filled in automatically from the data associated with the spectrum.
4. Click the **General Information** tab, edit the fields as required, and then click **OK**.

### Search for a Similar Spectrum

Users can search the library for a spectrum (and its related compound information) that matches (or is similar to) an active spectrum. Searches can be performed with or without constraints.

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When searches are performed with constraints, only those records that match all the criteria are listed. The results are shown in a ranked list. The first item on the list is the best fit to the active spectrum. Entries lower in the list do not match as well.

The more constraints selected, the more precise the list becomes and fewer, more relevant matches are listed. After a set of constraints is defined, they will apply to all subsequent searches, unless they are edited.

Only peaks above the threshold are used in the search. When selecting search constraints, peaks can be added or removed from the active spectrum. For example, if a peak is considered to be background or noise spike, then it should not be used in the search because it could produce inaccurate results.

When searches are performed without constraints, a much larger list of suggested spectra is returned because the library makes fewer specific matches to the spectral data.

### Search for a Similar Spectrum

1. Right-click on an active spectrum and then click **Search With Constraints**.  
The software will centroid the spectrum automatically, if required.

Figure 13-17 Search Constraints Dialog

Constraint	Tolerance	Unit
Mass Tolerance	+/- 0.2	Da
Intensity Factor	+/- 2	
1st Precursor m/z	+/- 0.25	Da
Collision Energy	+/- 5	
2nd Precursor m/z	+/- 0.25	Da
Excitation Energy	+/- 5	
Retention Time	+/- 0.1	min

2. In the **Maximum Number of Match** field, type the maximum number of compounds to be returned by the search.
3. In the **Preselect Constraints** section, select the check boxes for the constraints to apply.
4. For each constraint selected, in the **Preset Tolerance** section, type the tolerance.

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5. If required, select a method of sorting records from the **Result Sorted by** list.
6. If required, type text in the **Comment Contains** field.
7. If required, type text in the **Keyword Contains** field.
8. To apply peak constraints by adding and removing peaks, click **Peak Constraints**. The **Peaks Included** table opens.
9. To add peaks to the list you want to search against, click **Add** and then type the *m/z* and the corresponding intensity in the empty cell.
10. To remove peaks so they will not be included in the search, select the peaks to exclude in the search and then click **Remove**.
11. Click **Search** to save the constraints and begin the search.

### View a Compound from the Search Results

If several spectra match the unknown spectrum, then other spectra can be viewed and compared to the unknown.

1. In the Search Results dialog, in the list of compounds, select the row number of the compound.
2. Click the spectrum pane of one of the known compounds. The spectrum of the selected compound opens.

### Library Search Tips

To Do This	Do This
Library searches: To group conditions	Select the conditions to group and then click <b>Group</b> . This function behaves like parentheses in formulas.
Library searches: To search without using constraints	Right-click an active spectrum, and then click <b>Search Library</b> . The Search Results dialog opens.
Table-specific queries: To view the entire table again	Right-click anywhere in the Results Table and then click <b>Query &gt; Show All</b> . You Queries can be applied again or edited.  It is recommended that the user validate any queries that are used to analyze data in a Results Table.

To Do This	Do This
Peak Integration: To review peaks	<p>To review all peaks, make sure that all samples are listed in the Results Table.</p> <p>The Peak Review window contains the peaks listed in the Results Table. If some samples are hidden in the table (for example, if a query was applied), then they are also hidden in peak review.</p> <p>It is recommended that the user validate any queries that are used to analyze data in a Results Table.</p>
Peak Integration: To move to the first peak in the batch	<p>Right-click anywhere in the Peak Review pane and then click <b>Show First Page</b>. To move to the last peak in the batch, right-click anywhere in the Peak Review pane and then click <b>Show Last Page</b>.</p>
To examine calibration curves	<p>Right-click anywhere in the curve, click <b>Active Plot</b>, and select the curve to be plotted on top.</p>
Sample statistic review: To review an individual peak	<p>Select the <b>Display the Data Set(s)</b> check box, and then, in the <b>Data Point</b> column, double-click the data point that represents the peak. The software shows the Peak Review window with the selected peak.</p>
Results Tables: To return the Results Table to its original order	<p>Right-click on the Results Table and click <b>Sort &gt; Sort By Index</b>.</p>
Acquisition Methods: To create an acquisition method from the file information pane	<p>Right-click the file information pane and then click <b>Save Acquisition Method</b>.</p>

# Operating Instructions — Analyze and Process Quantitative Data

---

# 14

This section describes how to use the Analyst MD software to analyze and process quantitative data. Data can also be processed using the MultiQuant MD software. We recommend that the MultiQuant MD software be used to quantitate data. Refer to the documentation that comes with the MultiQuant MD software.

Use the sample files found in the `Example` folder to learn how to select samples for quantitation, how to select preset queries and create table-specific queries, and how to analyze the acquired data. For more information about the following topics, refer to the document: *Advanced User Guide*.

We recommend that the user validate any queries that are used to analyze data in a Results Table.

- Metric Plots
- Layout of a Results Table

## Quantitative Analysis

Quantitative analysis is used to find the concentration of a specific substance in a sample. By analyzing an unknown sample and comparing it to standard samples, that is, samples containing the same substance with known concentrations, the software can calculate the concentration of the unknown sample. The process involves creating a calibration curve using the signal response or response ratio of the standards and then calculating the concentrations of the unknown samples. The calculated concentrations of all of the samples are added to a Results Table.

Quantitative analysis is most commonly performed using a Multiple Reactions Monitoring (MRM) scan. In an MRM scan, a precursor ion and a characteristic product ion are used to define an MRM transition that is highly specific to the analyte. The MRM transition, coupled with the retention time associated with the analyte during liquid chromatography, provides the specificity required for quantitation.

Quantitation is accomplished through the use of validated MRM LC-MS/MS acquisition methods, acquisition of calibration standard curves, and the subsequent integration of the peaks associated with the compounds of interest. The calibration curve relationship between signal response and concentration is used to determine the quantity of a particular analyte in an unknown sample.

## Quantitation Methods

A quantitation method is a set of parameters used to generate peaks in a sample. The quantitation method can include parameters used to locate and integrate peaks, generate standard curves, and calculate unknown concentrations. A previously saved quantitation method can be selected from the **Quantitation** menu in the batch.

Three tools can be used to create a quantitation method: the Quantitation Wizard, the Build Quantitation Method, and Quick Quant.

### Build Quantitation Method

The Build Quantitation Method does not generate a quantitation Results Table, although the method can subsequently be used in the Quantitation Wizard to create a Results Table. The Build Quantitation Method can also be used to change existing quantitation methods. This is the most flexible way of creating a quantitation method. Refer to the section: [Create a Method Using the Quantitation Method Editor](#).

### Quantitation Wizard

The Quantitation Wizard generates a Results Table at the same time as the quantitation method. Alternatively, an existing quantitation method can be used to quantitate different sets of data.

### Quick Quant

Quick Quant is not recommended for quantitation of results.

Quick Quant is part of the Batch Editor. Use Quick Quant to add compound concentrations before acquiring data. Because a sample has not been acquired, a representative sample cannot be selected, nor can peaks be reviewed. With this feature, only the method components are defined.

Do not use the automatically generated Quick Quant method to perform quantitation if the Quick Quant feature is used to store sample types and concentrations in the data file. This quantitation method does not use compound and sample-specific integration parameters that have been optimized for peak selection.

To use a previously saved quantitation method, select it from the **Quantitation** menu in the batch. For instructions on creating a batch, refer to the section: [Create and Submit a Batch](#).

## Quantitation Methods and Results Tables

For the following procedures, use the sample data that is installed in `Example/Triple Quad` folder. The folder contains the data files, `Mix_Batch_1` and `Mix_Batch_2`. These sample files are used to demonstrate the usefulness of metric plots to isolate problematic samples. The ions scanned were reserpine (609.3/195.0), minoxidil (210.2/164.2), tolbutamide (271.1/91.1) and rescinnamine (635.3/221.2), which is the internal standard. `Mix_Batch_1` contains no errors, but `Mix_Batch_2` contains a QC sample to which the internal standard was added twice (sample QC2).

### Create a Method Using the Quantitation Method Editor

---

**Note:** We recommend that the user validate any queries that are used to analyze data in a Results Table.

---

Prerequisites
<ul style="list-style-type: none"><li>Select the project or subproject that contains the data to be quantified. Refer to the section: <a href="#">Change Between Projects and Subprojects</a></li></ul>



1. Make sure that the `Example` project is selected.
2. On the Navigation bar, under **Quantitate**, double-click **Build Quantitation Method**. The Select Sample dialog opens.
3. Double-click the **Triple Quad** folder in the **Data Files** list.
4. Select **Mix\_Batch\_2.wiff**. The samples in the selected data file are shown in the **Samples** list.

---

**Note:** If the **Compound ID** field was populated for the samples and internal standards in the acquisition method, then in the Internal Standards table, when a value is selected in the **Q1/Q3** field, the **Name** field is automatically populated.

---

5. Select a sample that provides a detectable signal to select integration parameters that fit the entire batch and then click **OK**.
6. In the Internal Standards table, in the **Name** column, select **rescinnamine**. In **Q1/Q3** column, select **635.3/221.2**.
7. In the Analytes table, do the following:
  - a. In the **Name** column, select **minoxidil** for the **Q1/Q3** column masses of **210.2/164.188**, **tolbutamide** for **271.3/91.146**, and **reserpine** for **609.4/195.039**.
  - b. In the **Internal Standard** column, from the list, select the **rescinnamine** as internal standard to be associated with each analyte.

- c. Delete **635.4/221.185** from the **Q1/Q3** column in the Analytes table.

---

**Note:** If the **Compound ID** field was populated for the samples and internal standards in the acquisition method, then in the Analytes table, the **Name** field and **Q1/Q3** field are populated.

---

8. Open the Integration tab.  
The preset integration parameters are suitable for most peaks.
9. If the integration is not suitable, then change the algorithm. Refer to the section: [Manually Integrate Peaks](#).
10. Click **Show or Hide Parameters** () to show the additional integration algorithms.
11. Open the Calibration tab.  
The preset parameters are suitable for these samples. User can change the fit, weighting, and regression parameter depending on the specific applications.
12. Save the quantitation method.  
The new method can be used when a batch is created in the Batch Editor or when the Quantitation Wizard is used to generate a Results Table.

---

**Tip!** The quantitation method can only be used in the current project unless it is copied to another project. To do this, click **Tools > Project > Copy Data**. A new project must be created and selected to be available for use.

---

### Create a Results Table Using the Quantitation Wizard

---

**Note:** We recommend that the user validate any queries that are used to analyze data in a Results Table.

---

Prerequisites
<ul style="list-style-type: none"><li>• Select the project or subproject that contains the data to be quantified. Refer to the section: <a href="#">Change Between Projects and Subprojects</a></li></ul>



1. On the Navigation bar, under **Quantitate**, double-click **Quantitation Wizard**.  
The Create Quantitation Set - Select Samples page opens.
2. In the **Available Data Files** list, double-click the **Triple Quad** folder.
3. Select **Mix\_batch\_2.wiff**.
4. Click **Add All**.

---

**Note:** We recommend that users do not process or report results from any sample for which acquisition was abnormally or unexpectedly terminated.

---

## Operating Instructions — Analyze and Process Quantitative Data

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5. Click **Next**.  
The Create Quantitation Set - Select Settings & Query page opens.
6. Click **Select Existing: Query** in the **Default Query** section.
7. Select **Accuracy 15%** from the **Query** list.

---

**Note:** To create a query at the same time, refer to the section: [Create a Standard Query \(Optional\)](#).

---

**Note:** It is the responsibility of the user to evaluate and validate the query to be used for specific applications.

---

8. Click **Next**.  
The Create Quantitation Set - Select Method page opens.
9. Click **Choose Existing Method**.
10. Select **PK Data\_Mix.qmf** from the **Method** list.
11. Click **Finish**.  
The Results Table opens.

---

**Tip!** To add or remove samples in the Results Table, click **Tools > Results Table > Add/Remove Samples**.

---

12. Review the sample type, actual concentration, peak integration, calibration curves, statistics pane, metric plot for the internal standard, and other information related to data quantitation.
13. Save the Results Table.

---

**Note:** We recommend that users do not change datafile (wiff) names if a Results Table includes samples from that file.

---

---

**Tip!** Well-formatted reports can be created from a Results Table using the Reporter software. We recommend that the user validate the results if a Reporter template that contains a query is used. Refer to the section: [Reporter Software](#).

---

### Create a Standard Query (Optional)

Advanced users can create a query and a standard query numerous ways. The following is one example. For more information about creating queries, refer to the document: *Help*.

We recommend that the user validate any queries that are used to analyze data in a **Results Table**.

1. On the Navigation bar, under **Quantitate**, double-click **Quantitation Wizard**.  
The Create Quantitation Set - Select Samples page opens.

2. Select samples to be used as part of the quantitation set.
3. Click **Next**.  
The Select Settings & Query page opens.
4. In the **Default Query** section, select **Create New Standard Query**.
5. Type a query name.

**Figure 14-1 Create Quantitation Set — Select Settings & Query Page**

Create Quantitation Set - Select Settings & Query

Please select the settings for the new results table and the default query (if any). Integration Algorithm: IntelliQuan

Settings to Use: Default

Default Query

None

Select Existing:

Query: Accuracy 15%  Execute Query as Standard Query

Create New Standard Query

Name:

< Back Next > Finish Cancel Help

6. Click **Next**.

Figure 14-2 Create Quantitation Set — Create Default Query Page

Please specify the concentrations/sample names and the corresponding allowed accuracy variations (in percent). You can leave any of the "variation" fields empty as desired.

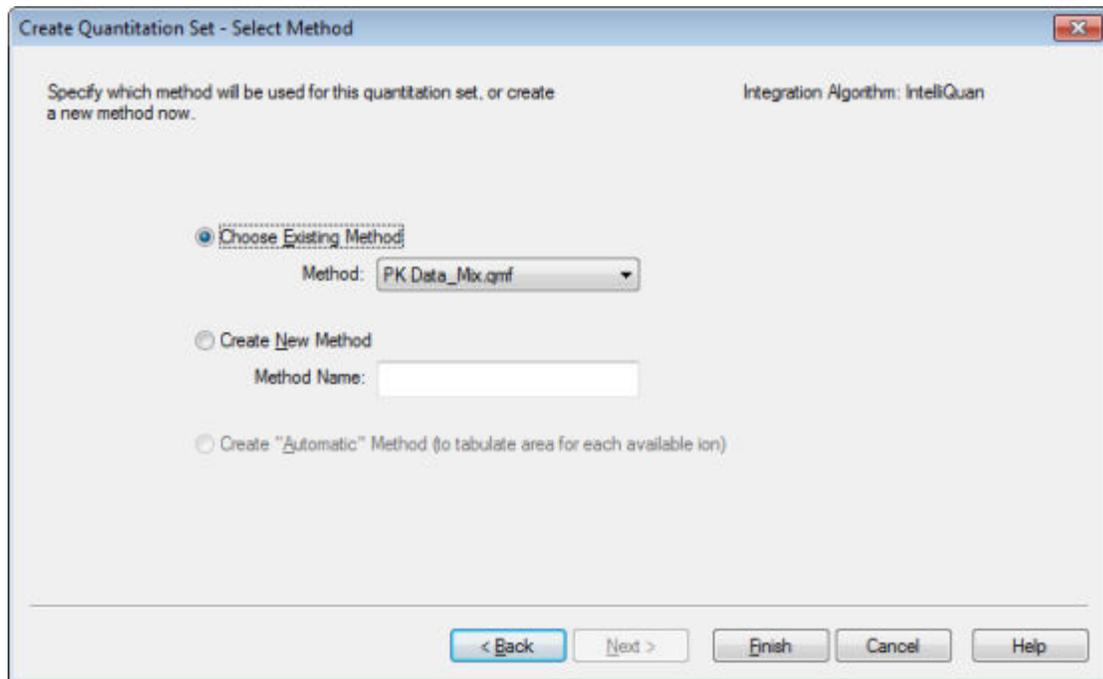
Maximum Allowed Accuracy Variation for QCs (%)		Maximum Allowed Accuracy Variation for Standards (%)	
Concentration	Max. Variation	Concentration	Max. Variation
4.000000		0.120000	
40.000000		0.240000	
400.000000		0.490000	
4000.000000		0.980000	
12000.000000		1.950000	
		3.910000	
		7.810000	
		15.630000	
		31.250000	
		62.500000	
		125.000000	

Query By Name

< Back   Next >   Finish   Cancel   Help

7. In the **Maximum Allowed Accuracy Variation for QCs (%)** table in the **Max. Variation** column, type the maximum allowable percent of variation for each QC, for example 5 is  $\pm 5\%$ , in the same row as the corresponding concentration. If the concentrations were not specified during acquisition, then they are not shown. In this case, type them in the **Concentration** column.
8. In the **Maximum Allowed Accuracy Variation for Standards (%)** table, in the **Max. Variation** column, type the maximum allowable percent of variation for each standard, for example 10 is  $\pm 10\%$ , in the same row as the corresponding concentration. If the concentrations were not specified during acquisition, then they are not shown. Type the concentrations in the **Concentration** column.
9. Click **Next**.

**Figure 14-3 Create Quantitation Set — Select Method Page**



10. Select or create a method.

11. Click **Finish**.

The query is applied as a standard query. The query results are shown as a Pass or Fail entry in the **Standard Query Status** column of the Results Table.

---

**Tip!** To return to the full view, right-click and then click **Full**.

---

## Define the Layout of Results Tables

Predefined views of the Results Table are available.

Right-click in the Results Table and then click one of the following:

- To view the Full layout, click **Full**.  
All the analytes are shown.
- To view the Summary layout, click **Summary** and then click a field name.
- To view the Analyte layout, click **Analyte** and then click a single analyte if more than one analyte exists.
- To view the Analyte Group layout, click **Analyte Group** and then click an analyte group.

## Operating Instructions — Analyze and Process Quantitative Data

---

**Tip!** A new analyte group must be created first. To do this, right-click in the Results Table and then click **Analyte Group > New**.

---

**Tip!** To go back to the full view, right-click and then click **Full**.

---

The table is shown with the selected layout.

## Sort Data in Results Tables

Data in a Results Table can be sorted in the following ways:

- Quickly sort the table on one to three columns, using one of the Sort buttons. This sort criteria cannot be saved.
- Create a table-specific sort to save the sort criteria with the current table. Table-specific sorts allows the user to sort the current table on one to three columns and save the criterion for use with that table.
- Use a previously created preset sort. Users can create and save a sort and then later apply it to a Results Table.

---

**Tip!** To save a sort or any other table setting, right-click in the table and then click **Table Settings > Export To New Table Settings**. The sort and other parameters can be used in the current project. To use the table settings in a different project, copy it to another project. Click **Tools > Project > Copy Data**. A new project must be created and selected to be available for use.

---

## Sort a Results Table

1. Select up to three columns in the Results Table in the order to be sorted.
2. Do one of the following:
  - To sort in ascending order, click **A-Z**.
  - To sort in descending order, click **Z-A**.

## Sort a Results Table and Save the Sort Criteria

1. Right-click in the Results Table and then click **Sort > New**.

**Figure 14-4 Sort Dialog**

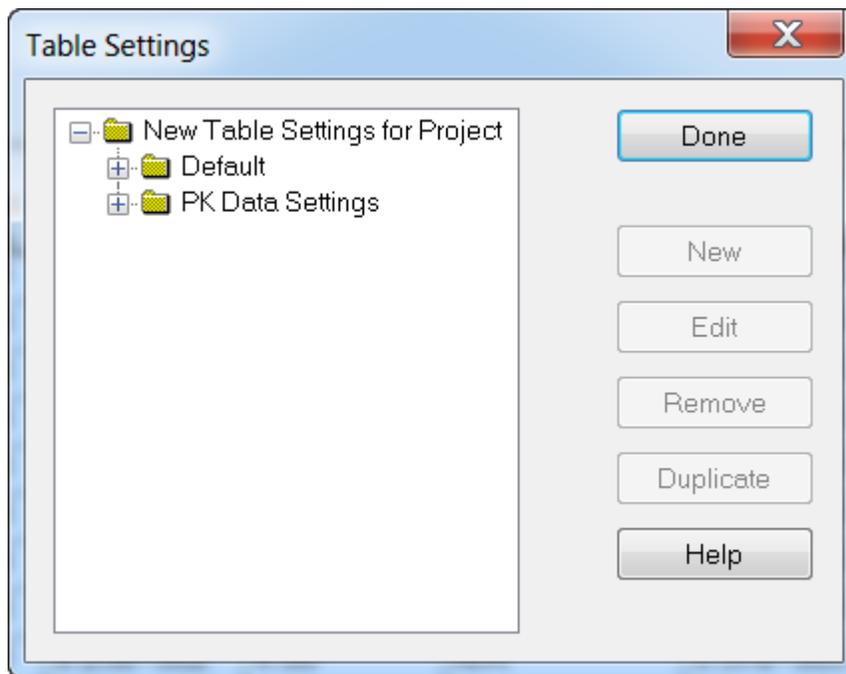


2. In the **Name** field, type the name for the new sort.
3. For each sorting rule, in the **Sort By** and **Then By** sections, do the following:
  - In the **Group** list, select the type of column to sort on.
  - In the **Column** list, select the column to sort on.
  - Select the direction of the sort: **Ascending** or **Descending**.
4. Do one of the following:
  - To perform the sort, save the sort criteria, and close the **Sort** dialog, click **Save/Execute**.
  - To perform the sort and close the **Sort** dialog without saving the sort criteria, click **Execute**.

## Save Default Sort Criteria for Future Results Tables

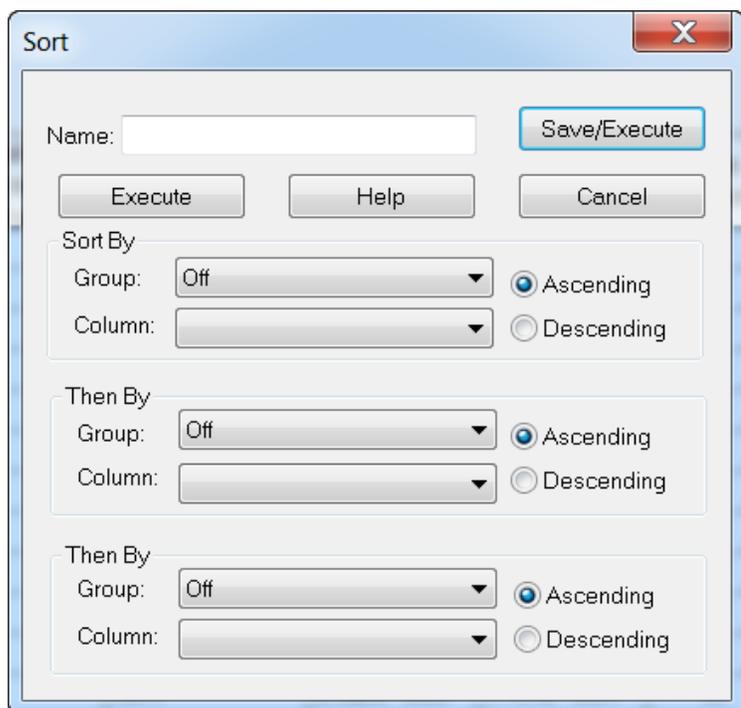
1. Click **Tools > Settings > New Quantitation Results Table Settings**.

Figure 14-5 Table Settings Dialog



2. Expand the **Table Settings** folder and then double-click the **Default** folder.
3. From the expanded **Default** folder, select the **Sorts** folder, and then click **New**.

Figure 14-6 Sort dialog



4. In the **Name** field, type the name for the new sort.
5. For each sorting rule, in the **Sort By** and **Then By** sections, do the following:
  - In the **Group** list, select the type of column to sort on.
  - In the **Column** list, select the column to sort on.
  - Select the direction of the sort: **Ascending** or **Descending**.
6. To save the criteria and close the **Sort** dialog, click **OK**.
7. To close the **Table Settings** dialog, click **Done**.

## Sort a Results Table using Preset Sort Criteria

- Right-click in the Results Table, click **Sort** and then select the name of the sort.

## Peak Review and Manual Integration of Peaks

Use peak review to survey the peaks that the software has identified and then redefine the peak or the start and end points where required.

After identifying the analytes and internal standards that the software must find, the software searches for the peaks in the samples. When the software identifies a peak, it shows the chromatograms for each analyte and internal standard in the Create Quantitation Method: Define Integration page of the Standard Wizard or on the Integration tab of the Full Method Editor. The user can confirm the peaks that are found or change the quantitation method to better define the peaks. We recommend that users manually review all integration results.

For information on using the Peak Review right-click menu, refer to the section: [Peak Review](#).

### Review Peaks

During peak review, the user might want to view a peak in its entirety or to examine the baseline to find out how well the software found the start and end points of the peak. The automatic zooming feature can be used to do either.

To help the software find a peak, define the exact start and end points of the peak and background manually. These changes will apply only to that individual peak unless the global method is updated.

---

**Note:** We recommend that manually integrated results be validated.

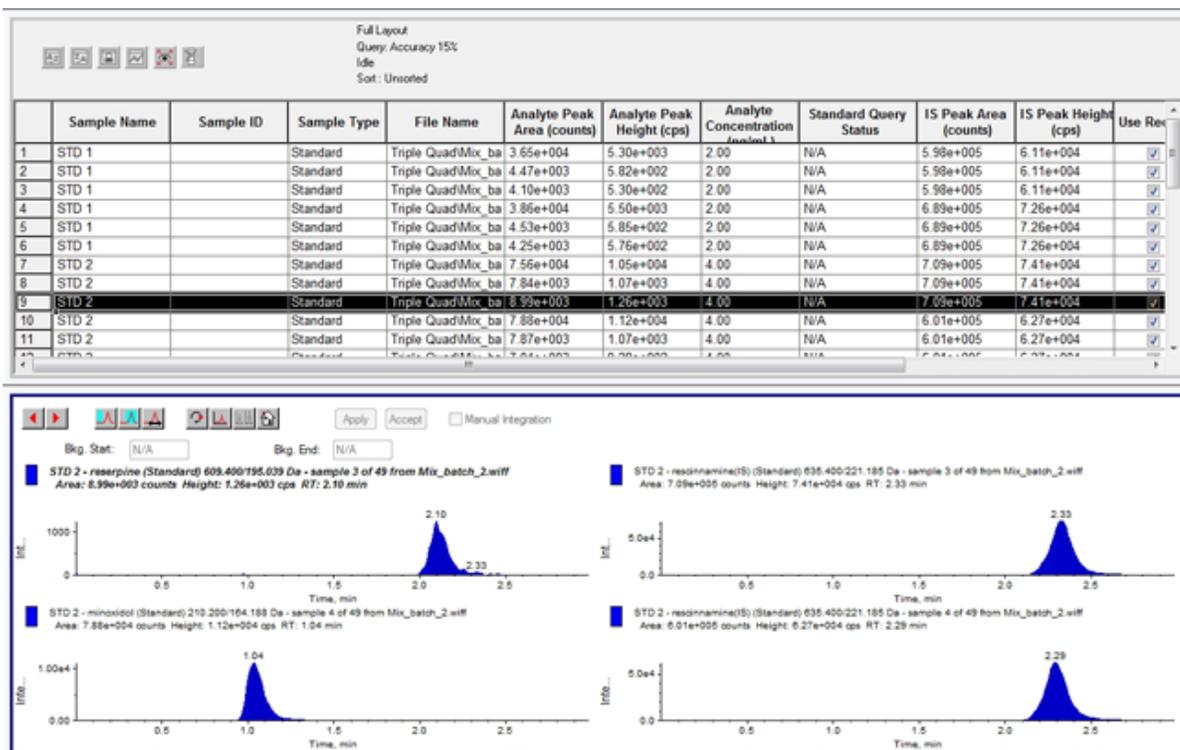
---

**Tip!** To review an individual peak, right-click on a point on the curve and then click **Show Peak**. The software opens the Peak Review window with the selected peak.

---

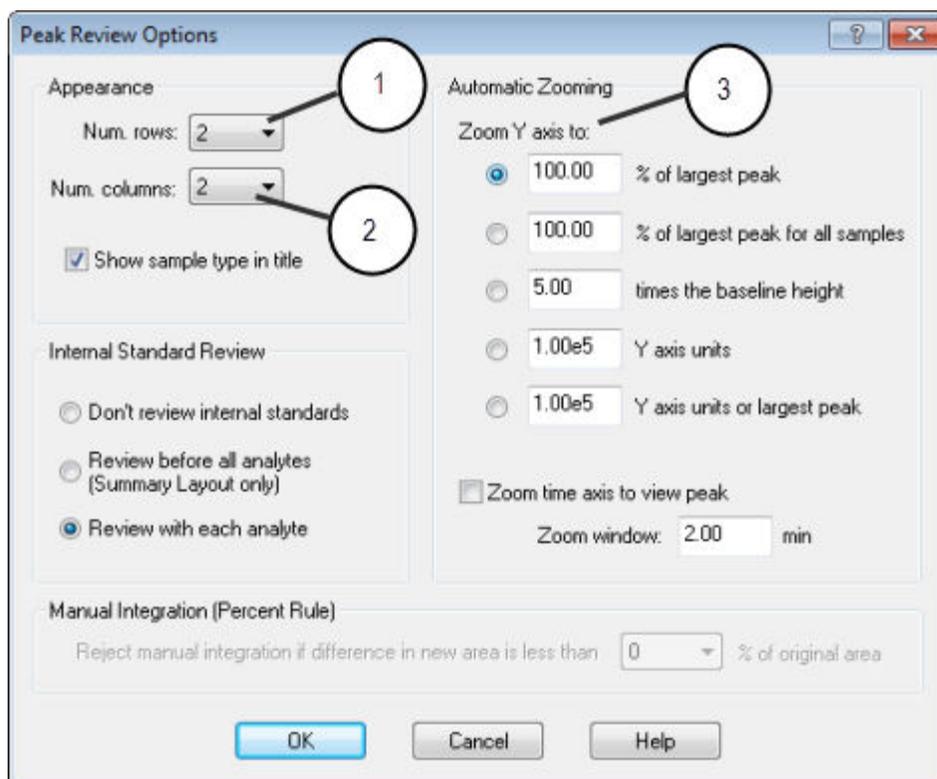
1. Right-click the Results Table and then click **Analyte**.
2. Select an analyte.
3. Click **Tools > Peak Review > Pane**.  
The peaks are shown below the Results Table with only the peaks listed in the Results Table.

Figure 14-7 Peak Review



4. Right-click the pane and then click **Options**. The Peak Review Options dialog opens.
5. In the **Appearance** section, change **Num. rows** to **1** and **Num. columns** to **2**.
6. In the **Automatic Zooming** section, click **Zoom Y axis to: 100% of largest peak** to show the entire peak.

Figure 14-8 Peak Review Options Dialog



Item	Definition
1	Number of rows
2	Number of columns
3	<b>Zoom Y-axis to 100% of largest peak</b> to show the entire peak

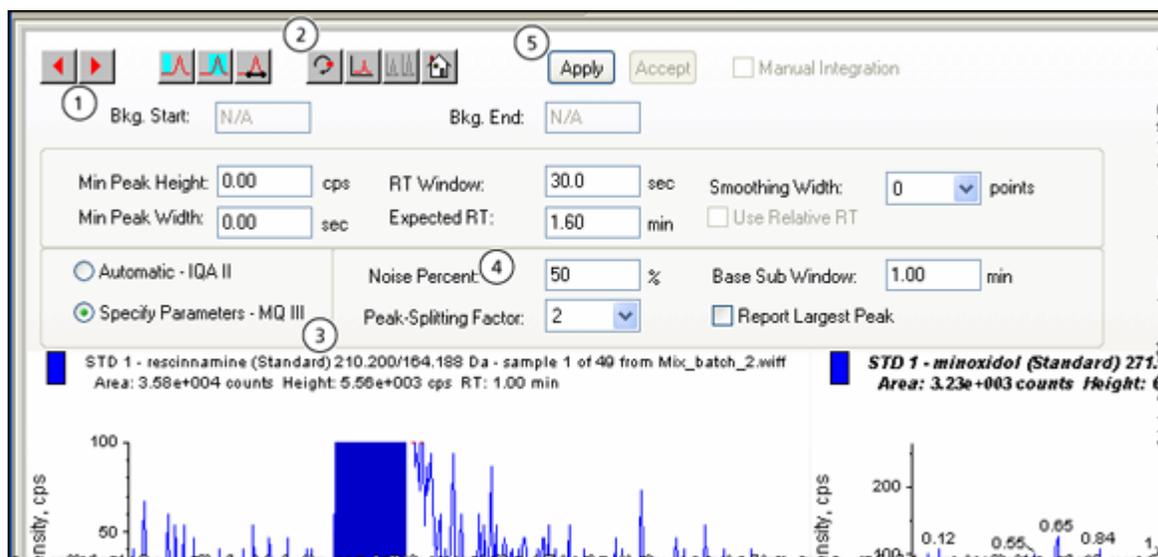
7. Click **OK**.
8. To move through the peaks, click the right-pointing arrow. Refer to the figure: [Figure 14-9](#).
9. Go to the second injection of standard 3.  
In this example, the peak can be integrated closer to the baseline by selecting the **Specify Parameters** option.

---

**Tip!** To move to a specific peak in the Peak Review pane, select the corresponding row in the Results Table.

---

Figure 14-9 Peak Review Pane



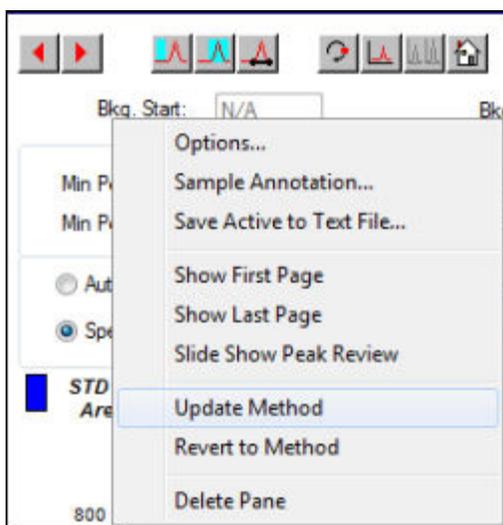
Item	Definition
1	Arrows: Click to move through the peaks.
2	<b>Show or Hide Parameters:</b> Click to show the integration parameters.
3	Integration parameters: Click to change the parameters.
4	<b>Noise Percentage:</b> Type a noise percentage.
5	<b>Apply:</b> Click to integrate the parameters.

10. Click **Show or Hide Parameters** twice.
11. Click **Specify Parameters - MQ III**.
12. Change the **Noise Percent** value.
13. Click **Apply**.  
The peak is integrated closer to the baseline.
14. If the change does not improve the peak integration, then adjust the **Noise Percent** parameter until the optimal value is found.

**Note:** The **Update Method** option only updates the algorithm values for that specific analyte (or internal standard) and not all analytes.

15. To update the algorithm for all peaks, right-click in the pane and then click **Update Method**.

Figure 14-10 Update Method



## Manually Integrate Peaks

Manually integrating peaks should be done last, to limit person-to-person variability. Manually integrate peaks only if all the peaks have not been found after the algorithm parameters have been adjusted and updated. We recommend that users validate the results to determine whether manual integration is acceptable for specific applications.

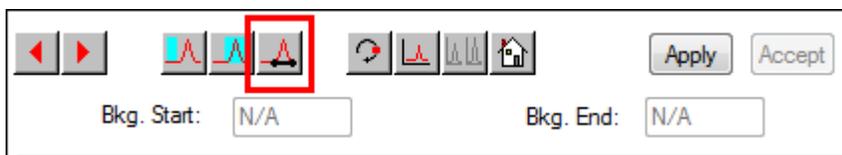
---

**Note:** Peaks that are manually integrated, or where the algorithm was changed for only that peak, are identified in the **Record Modified** column of the Results Table, as are peaks that have algorithm parameter changes for a sample that are not applied to the entire analyte group.

---

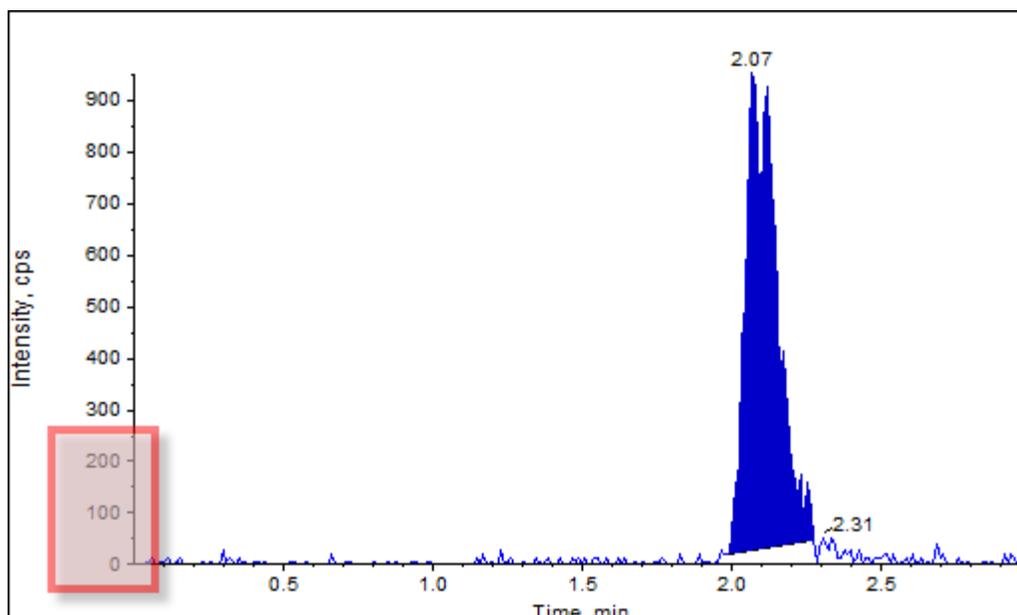
1. In the **Peak Review** pane, click **Manual Integration Mode**.

Figure 14-11 Peak Review Pane: Manual Integration Mode



2. Zoom in on the lower 10% of the peak.

Figure 14-12 Peak Review Pane: Zooming in on a Peak



3. Move the cross-hair to where the start of the peak is to be defined and then drag the cross-hair to where the end of the peak is to be defined. The software shades the area bounded by the base and sides of the peak. Peak parameters are gray because they are no longer applicable because the peak was drawn manually.
4. Do one of the following:
  - To make this change permanent, click **Accept**.
  - To discard the changes, clear the **Manual Integration** check box.

---

**Tip!** If a peak was correct as originally selected, right-click the peak and then click **Revert to Method**.

---

## Calibration Curves

Use calibration curves to find the calculated concentration of samples, including quality control (QC) samples. QC samples are added to a batch to estimate the data quality and accuracy of standards in the batch. QC samples have known analyte concentrations but are treated as unknowns so that the measured concentrations can be compared to the actual value.

The calibration curve is generated by plotting the concentration of the standard against its area or height. If an internal standard is used, then the ratio of the standard concentration or internal standard is plotted against the ratio of the standard peak height or area to the internal standard peak height or area. The area or height ratio of a sample is then applied to this curve to find the

## Operating Instructions — Analyze and Process Quantitative Data

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concentration of the sample, as shown in the Results Table. A regression equation is generated by this calibration curve according to the regression that was specified. The regression equation is used to calculate the concentration of the unknown samples.

Linear regression is recommended for the calibration curve.

We recommend that the user does not report quantitative values that fall outside the concentration range covered by the calibration curve.

For information on using the Calibration Curve right-click menu, refer to the section: [Calibration Curve](#).

## View Calibration Curves

The user can view the calibration curve and change the regression options in an open Results Table. If two or more Results Tables are open, then the calibration curves can be overlaid. To overlay curves, make sure that the method used to create the tables is the same.

Plot a calibration curve to see the curve used for regression. The **Calculated Concentration** column in the Results Table reflects any changes resulting from the fit of the curve to the points of the standard.

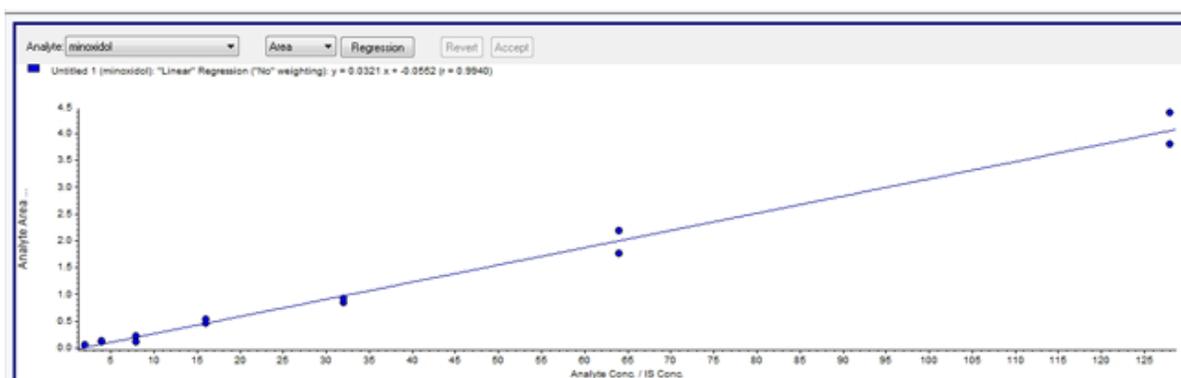
---

**Note:** This option is available only when a Results Table is open.

---

1. Open a Results Table.
2. Click **Tools > Calibration > Pane**.  
The Calibration Curve pane containing the calibration curve opens.

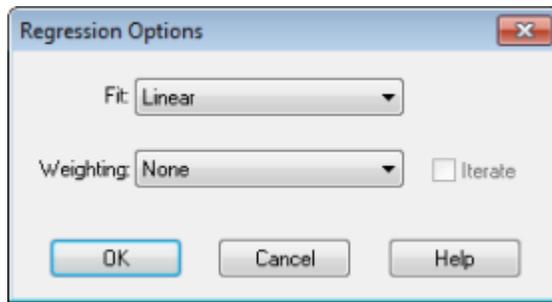
**Figure 14-13 Calibration Curve**



3. If there is more than one analyte, then use the following steps to view the calibration curve for another analyte:
  - a. From the **Analyte** list, select an analyte.

- b. If required, from the next list, select **Area** or **Height**.
4. To change the regression options for the calibration curve, do the following:
  - a. Click **Regression**.

**Figure 14-14 Regression Options Dialog**



- b. Select **Linear** in the **Fit** list.
- c. Select **1 / x** in the **Weighting** list.
- d. Click **OK**.

The calibration curve opens. The user can review individual peaks on the curve or exclude points from the curve to produce a better curve.

5. If required, repeat these steps to create a more appropriate curve.
6. To save the changes, click **Accept**.

## Overlay Calibration Curves

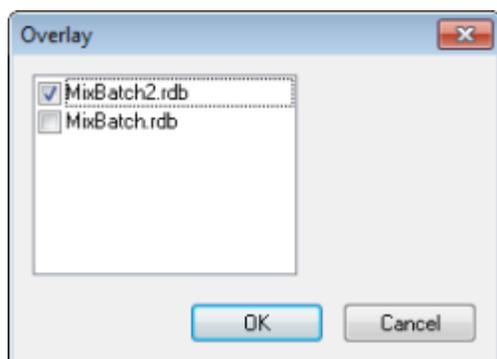
---

**Tip!** To examine the curve for one table more closely, right-click the curve and then click **Active Plot**. Select the curve to be plotted on top.

---

1. With two or more open Results Tables, view a calibration curve for one of the tables.
2. Right-click the calibration curve and then click **Overlay**.

**Figure 14-15 Overlay Dialog**



3. Select the tables to overlay with the current curve.
4. Click **OK**.  
The software plots the curves for all selected tables on the same graph.

## Sample Statistics

Use the Statistics window to view the statistics for samples, typically for standards and quality controls (QCs). The data from each available batch in the Results Table opens in tabular form in the grid and a row of data is shown for each standard or QC concentration.

We recommend that batch statistics be included in reports to be sure of the validity of the results.

## View the Statistics for Standards and QCs

When more than one Results Table is open, statistical information about the standards and QCs for additional batches can be shown in the Statistics window. This facilitates comparison of results between batches and identification of trends in the standards or QCs.

1. Open a Results Table.
2. Click **Tools > Statistics**.
3. Select **Concentration** from the **Statistics Metric** list.
4. Select an analyte in the **Analyte Name** field.
5. Select **Standard** in the **Sample Type** field.  
The results are shown.
6. Examine the **%CV** and **Accuracy** columns.

The **%CV** shows the coefficient of variance between the measurements of a single parameter, for example the area. **Accuracy** shows how close the plotted point is to the interpolated value.

7. If required, select the **Display Low/High values** check box and then examine the **Low**, **High** values, and **Mean** for each row in the grid. Each row represents standards that have the same concentration levels.
8. Select another analyte.  
The results are shown on a per-analyte basis.
9. To check for quality control (QC) variations at the same concentration levels, select **QC** in the **Sample Type** field.

## Metric Plots

A metric plot graphically shows the data in a Results Table column plotted against the X-axis or the Y-axis, or the data in two columns plotted against each other. This section describes how to generate and work with metric plots.

A few predefined metric plots are also included:

- Int\_Std\_Response (to locate problem sample)
- Analyte\_Area versus Height (to verify chromatography behavior)
- PK profile (conc. versus time point, to run after Sample query)

## Generate Metric Plots

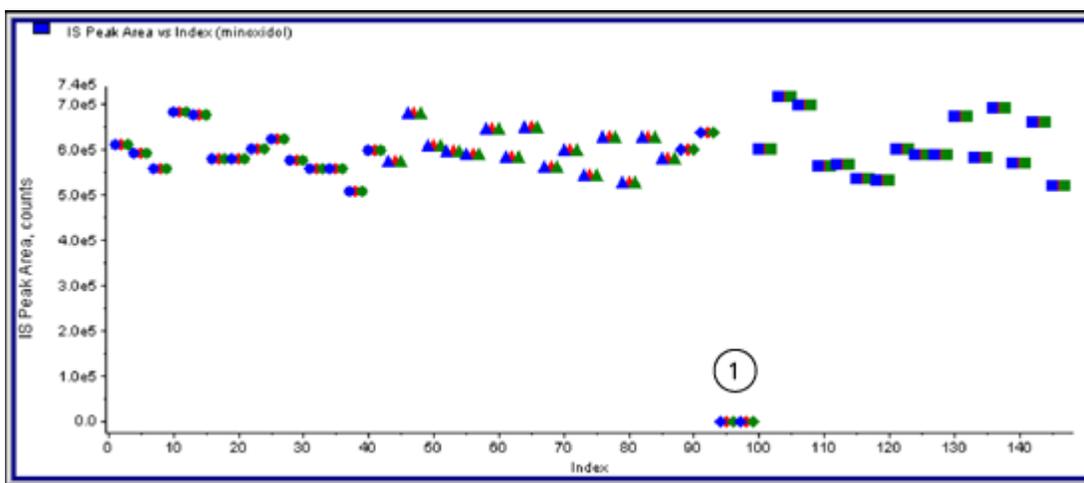
Use metric plots to plot a given column, such as Analyte Peak Area, Accuracy, or Calculated Concentration, from the Results Table. Two Results Table fields can also be plotted against each other and the plot then used to investigate points that are shown outside of the normal range. Metric plots are often used with queries. For more information about queries, refer to the document: *Help*.

Generate metric plots in the following ways:

- Use the Plot button to plot a column or columns of the current Results Table, but not save the plotting criteria.
- Create a table-specific plot to save the plot criteria with the current table.
- Create a global plot to save the plotting criteria for use with future Results Tables.

QC, unknown, blanks, double blanks, or solvents are not shown on the calibration curve, but metric plots of them can be generated.

Figure 14-16 Example of a Metric Plot



Item	Description
1	Double blanks

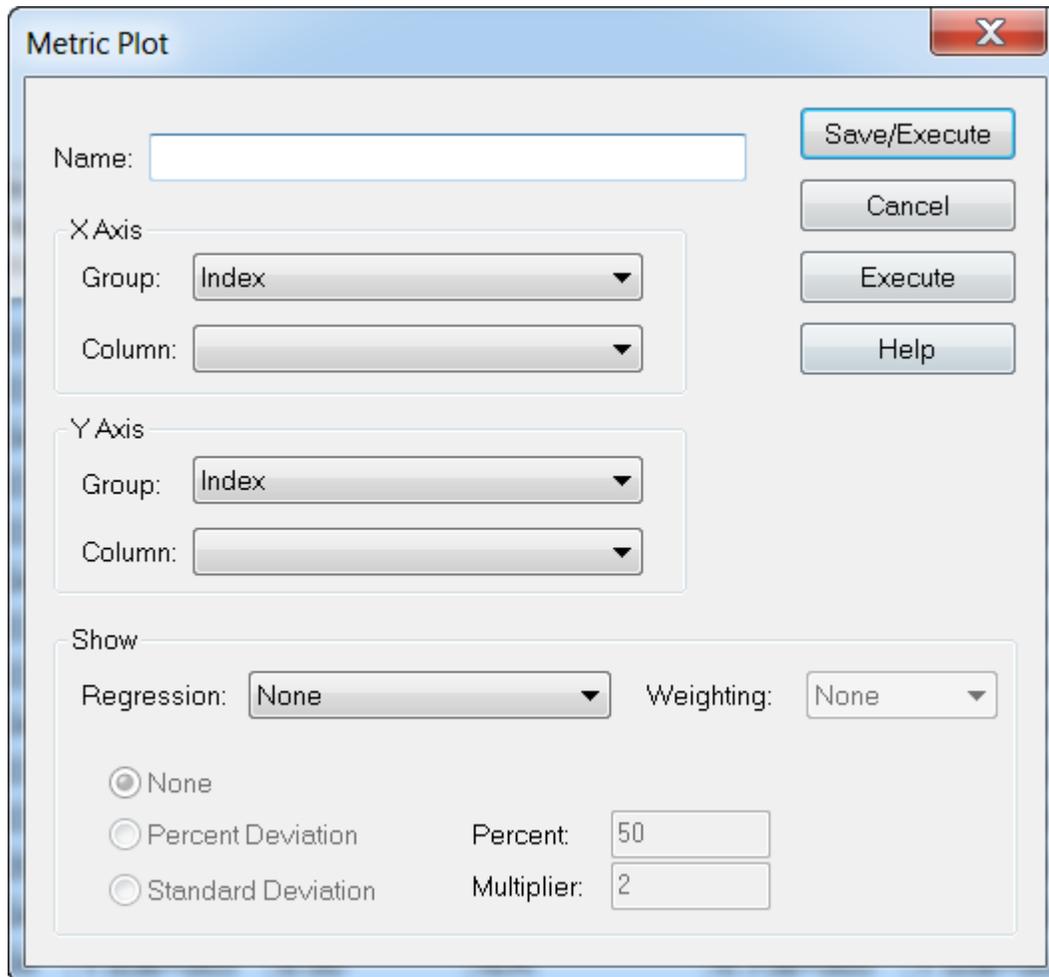
### Generate a Metric Temporary Plot

- With a Results Table open, do one of the following:
  - To plot the data on the Y-axis with the X-axis as an index, click the heading of the column for the data to be plotted.
  - To plot the data from the first selected column on the X-axis and the second selected column on the Y-axis, select two columns by pressing the **Ctrl** key and then clicking the column headings.
- Above the Results Table, click the **Metric Plot by Selection** icon. Refer to [Table D-9](#). The metric plot opens.
- Right-click in the plot pane and then click **Data Legend** to view an explanation of the colors used by the plot.
- Right-click in the plot pane and then click **Point Legend** to view an explanation of the symbols used by the plot.

### Generate a Metric Plot and Save the Plot Criteria

- Open the appropriate Results Table.
- Right-click in the Results Table and then click **Metric Plot > New**.

Figure 14-17 Metric Plot Dialog



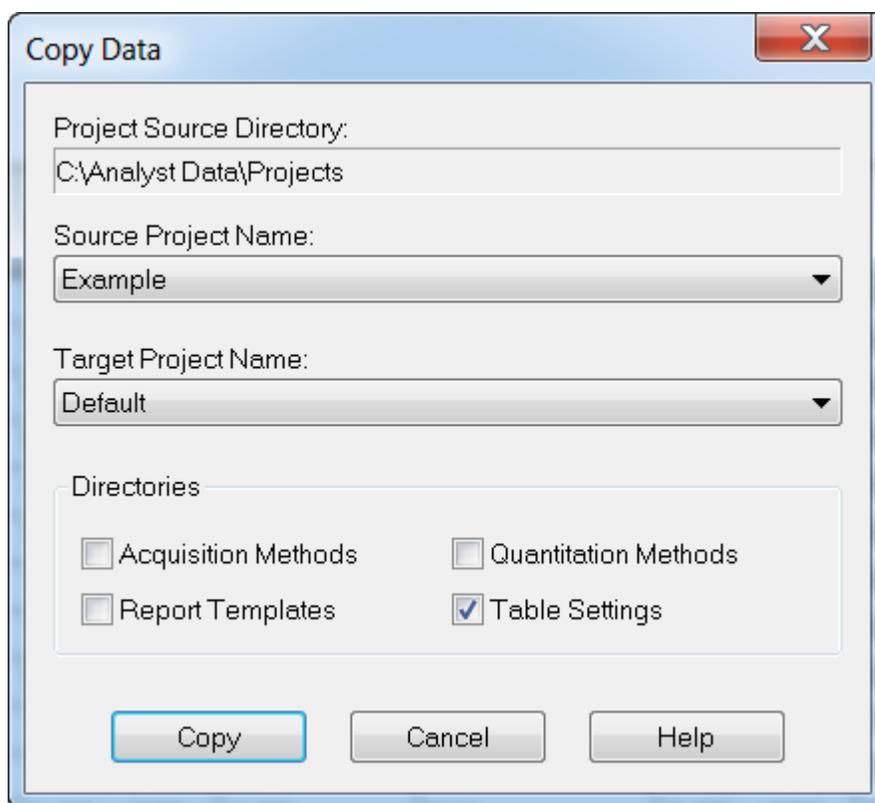
3. In the **Name** field, type the name for the new plot criteria.
4. In the **X-Axis** section, in the **Group** list, to plot a field in the Y-axis using the X-axis as an index, select **Index** and leave the **Column** list blank.
5. To plot two columns against each other, in the **Y-axis** section, in the **Group** list, select **Internal Standard**, and then, in the **Column** list, select **IS Peak Area**.
6. If required, in the **Regression** list, select the type of regression to use, and then select the appropriate regression settings.
7. To generate the plot and save the plot criteria, click **Save/Execute**.  
The metric plot opens. For more information, refer to [Figure 14-16](#).
8. Right-click in the plot pane and then click **Data Legend** to view an explanation of the colors used by the plot.

9. Right-click in the plot pane and then click **Point Legend** to view an explanation of the symbols used by the plot.  
This set of criteria is now available for future plots of this Results Table in the right-click menu of the Results Table. Users can also edit the plotting criteria.
10. To view the problem sample, try plotting the concentration of the unknown against time or plotting the area of the internal standard against the index.

### Save Default Plot Criteria for Future Results Tables

1. Right-click in the Results Table and then click **Table Settings > Export To New Table Settings**.  
This will export the table settings from the rdb so that it can be reused in other quantitation runs within the project.
2. To export table settings to another project, click **Tools > Project > Copy Data**.

**Figure 14-18 Copy Data Dialog**



---

The Reporter software extends the reporting functionality available in the Analyst MD software.

---

**CAUTION: Potential Wrong Results. To avoid incorrect results:**

- **Validate all Reporter queries before using them.**
  - **Validate the results if a modified Reporter template or one that contains a query is used.**
  - **Make sure that validation is completed on Reporter templates.**
- 

The Reporter software can be used to create custom reports with Microsoft Word and Excel (2013, 2016, or Office 365). The Reporter software has the following features:

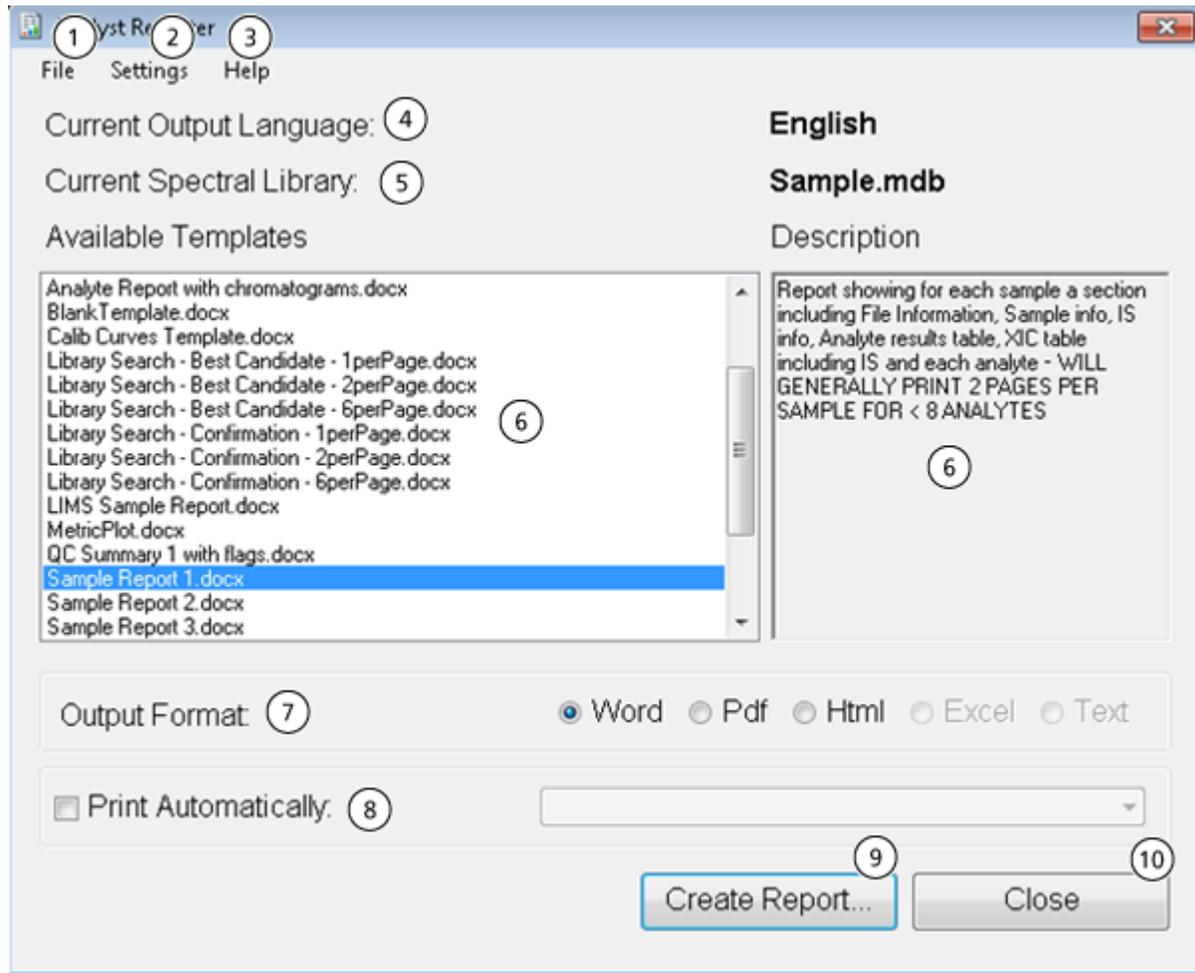
- Provides a variety of reports that use the data available in a Results Table, in file information, and in quantitative peak review windows.
- Provides a variety of reports that present MS/MS library search results. The user can configure the Reporter software to search against any MS/MS spectral library that uses the Analyst MD software (mdb) format.
- Uses Microsoft Word templates to provide the format information needed when generating reports. These templates can be created or modified to provide customized report formats. For information about creating or editing the Report Template Editor refer to the document: *Help*.
- Contains a blank starting template that can be used in the Reporter software editing environment to design report templates to meet most reporting requirements.
- Automatically prints, exports to Adobe Portable Document Format (pdf), and delivers results by e-mail.
- Generates reports from custom software applications that use the available Analyst MD software programming libraries.

Reporter software can be used as follows:

- Within the Analyst MD software to manually generate a report or set of reports.
- By applications that do not use the Analyst MD software.

# Analyst Reporter User Interface

Figure 15-1 Analyst Reporter



Item	Option	Description
1	File > Exit	Exits the program and releases all resources.

Item	Option	Description
2	<b>Settings &gt; Select Output Language</b>	Sets the language dictionary that will be used to replace language tags within a report template. Templates that contain language tags can be used to generate reports in any language. The language tags are replaced with text from a matching tag in the dictionary file for the selected language. These dictionary files are contained in the folder: C:\Program Files\AB SCIEX\AnalystReporter\Resources\Languages on Windows 7, 32-bit operating systems or C:\Program Files (x86)\AB SCIEX\AnalystReporter\Resources\Languages on Windows 7, 64-bit or Windows 10, 64-bit operating systems.
2	<b>Settings &gt; Select Library</b>	Browse to a spectral library. This library will be used for matching and scoring MS/MS data from Results Tables that contain data from information dependent acquisition (IDA) triggered MS/MS scan types.
2	<b>Settings &gt; Select Template Folder</b>	Sets the folder from which the available templates will be read. To return to the default template folder, select the <b>Default</b> option.
3	<b>Help &gt; About</b>	Shows information about the version of Reporter software currently installed.
4	<b>Current Output Language</b>	Shows the currently selected language dictionary used for replacing language tags within a report template. To select a language dictionary, click <b>Settings &gt; Select Output Language</b> .
5	<b>Current Spectral Library</b>	Shows the currently selected spectral library. To select a spectral library, click <b>Settings &gt; Select Library</b> .
6	<b>Available Templates and Description</b>	Shows a list of available report templates. Select a template to show a description of the template. To change the folder from which available templates are read, select <b>Settings &gt; Select Template Folder &gt; Browse</b> .

## Reporter Software

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Item	Option	Description
7	<b>Output Format</b>	<p>Shows the output formats that are supported by the Reporter software. Only formats that are compatible with the selected report template are enabled.</p> <ul style="list-style-type: none"><li>• <b>Word:</b> Microsoft Word document (docx) is produced. This document can be viewed by Microsoft Word 2010 and above.</li><li>• <b>PDF:</b> A report is created directly in PDF format.</li><li>• <b>HTML:</b> Microsoft Word is used to generate an HTML file. Associated image files are stored in a folder with the same name as the HTML file.</li><li>• <b>Excel:</b> A plain text file (csv) is produced. Report templates that contain values separated by commas can be opened in Microsoft Excel, where each value will be shown in a separate cell. Only templates that are specifically marked as text-compatible can be used for this output format.</li><li>• <b>Text:</b> A plain text document (txt) is produced. Only templates that are specifically marked as text compatible can be used for this output format.</li></ul>
8	<b>Print Automatically</b>	After the report has been created it is printed to the selected printer. Select any available printer.
9	<b>Create Report</b>	Creates the report in the selected output format using the selected report template.
10	<b>Close</b>	Exits the program and releases all resources.

## Generate Reports

The Reporter software extracts numerical data from the Results Table and sample and graphical information from the wiff file.

1. Open a **Results Table**.
2. Under **Companion Software**, double-click **Reporter**.  
The **Analyst Reporter** opens.
3. In the **Available Templates** field, select the applicable report template.
4. Click the **PDF** output format.

The Word option is pre-selected and the report is automatically saved in the current project Results folder. If the PDF output format is not selected, then the report is created and opened in Word or printed as selected, but the report is not saved. This lets the user edit the report in Word before saving the original report.

5. Optional for a library search (qualitative) workflow when a Results Tables that contains data from information dependent acquisition (IDA) triggered MS/MS scan types is used: Click **Settings > Select Library**, navigate to the applicable MS/MS library database (mdb format), and then click **Open**.
6. (Optional) Select the **Print Automatically** check box to print the reports automatically on a pre-selected printer.  
The default printer set in Windows is used unless a different printer is selected. The Reporter software keeps the selected printer between operations. If the printer is set to a PDF printer driver, then the Reporter software generates PDF file versions of the created reports automatically.
7. Click **Create Report**.  
The screen shows various progress indicators as the software opens the template and populates it with data from the Results Table. Some reports can take seconds to generate, others can take longer. A large data set with many MRM transitions or a large number of graphics might result in reports of several hundred pages that take hours to generate.

# Service and Maintenance Information — Mass Spectrometer

# 16

Regularly clean and maintain the system for optimal performance.



**WARNING! Electrical Shock Hazard. Do not remove the covers. Removing the covers might cause injury or malfunctioning of the system. The covers need not be removed for routine maintenance, inspection, or adjustment. Contact a SCIEX Field Service Employee (FSE) for repairs that require the covers to be removed.**



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Determine whether decontamination is required before cleaning or maintenance. If radioactive materials, biological agents, or toxic chemicals have been used with the system, then the customer must decontaminate the system before cleaning or maintenance.**

## Recommended Maintenance Schedule

The following tables provide a recommended schedule for cleaning and maintaining the system.

**Tip!** Perform maintenance tasks regularly to make sure that the system is performing optimally.

- Perform periodic gas leakage tests and general maintenance inspections to be sure of safe operation of the system.
- Clean the system regularly to keep it in good working condition.
- During system maintenance, carefully examine parts of the external gas supply system, including tubing connected to the equipment, to confirm that the condition is satisfactory. Replace any cracked, pinched, or collapsed tubing.

To determine how often to clean or perform maintenance on the mass spectrometer and ion source, consider the following factors. These factors can cause changes in mass spectrometer performance, indicating that maintenance is required.

- Compounds tested
- Cleanliness of the samples and the sample preparation methods
- Amount of time that the probe is exposed to the sample

## Service and Maintenance Information — Mass Spectrometer

- Overall system run time

For information about tuning frequency, refer to the section: [Calibration Ions and Solutions](#).

To order consumable parts and for basic service and maintenance requirements, contact a QMP or refer to the document: *Parts and Equipment Guide*. Contact a SCIEX FSE for all other service and maintenance requirements.

**Table 16-1 Mass Spectrometer Maintenance Tasks**

Component	Frequency	Task	For More Information
System	Daily	Inspect for leaks	Refer to the section: <a href="#">Chemical Precautions</a> .
Curtain plate	Daily	Clean	Refer to the section: <a href="#">Clean the Curtain Plate</a> .
Roughing pump oil	Weekly	Inspect the level	Refer to the section: <a href="#">Inspect the Roughing Pump Oil Level</a> . Contact the local QMP or FSE to add oil, if required.
Roughing pump oil	Every 2 years, or as needed	Replace	Contact the local QMP or FSE.
Roughing pump oil	As needed	Refill	Contact the local QMP or FSE.
Orifice plate (front)	As needed	Clean	Refer to the section: <a href="#">Clean the Front of the Orifice Plate</a> .
Orifice plate (front and back)	As needed	Clean	Contact the local QMP or FSE.
Mass spectrometer air filter	As needed	Replace	Contact the local QMP or FSE.
IonDrive QJet ion guide and IQ0 lens	As needed	Clean	Contact the local QMP or FSE.
Q0 rod set and IQ1 lens	As needed	Clean	Contact the local QMP or FSE.
Instrument surfaces	As needed	Clean	Refer to the section: <a href="#">Clean the Surfaces</a> .
Source exhaust drain bottle	As needed	Empty	Refer to the section: <a href="#">Empty the Source Exhaust Drain Bottle</a> .
Interface heater	As needed	Replace	Contact the local QMP or FSE.

**Table 16-2 Ion Source Maintenance Tasks**

Component	Frequency	Task	For More Information
TurbolonSpray and APCI probes	As needed	Examine and replace	Refer to the section: <a href="#">Remove the Probe</a> and <a href="#">Install the Probe</a> .
Electrodes for the TurbolonSpray and APCI probes	As needed	Examine and replace	Refer to the section: <a href="#">Replace the Electrode</a> .
Corona discharge needle	As needed	Replace	Refer to the section: <a href="#">Replace the Corona Discharge Needle</a> .
Turbo heater	As needed	Replace	Contact the local QMP or FSE.
Sample tubing	As needed	Replace	Refer to the section: <a href="#">Connect the Ion Source Tubing</a> .

For “As needed” tasks, follow these guidelines:

- Clean the mass spectrometer surfaces after a spill or when they become dirty.
- Empty the source exhaust drain bottle before it becomes full.
- Clean the orifice plate, IonDrive QJet ion guide, and Q0 region if system sensitivity degrades.

---

**Tip!** Clean the Q0 region regularly to minimize the impact of charging, a significant loss of sensitivity of the ions of interest over a short period of time, on the quadrupoles and lenses. Contact a QMP or FSE.

---

- Refill the roughing pump oil when it falls below the minimum oil level.
- Periodically inspect all exhaust connections to make sure that the integrity is maintained, and that any exhaust is removed from the customer lab.

## Clean the Surfaces

Clean the external surfaces of the mass spectrometer after a spill or when they become dirty.

---

**CAUTION: Potential System Damage. Use only the recommended cleaning methods and materials to avoid damaging the equipment.**

---

1. Wipe the external surfaces with a soft cloth dampened with warm, soapy water.
2. Wipe the external surfaces with a soft cloth dampened with water to remove any soap residue.

## Empty the Source Exhaust Drain Bottle



**WARNING! Hot Surface Hazard.** Let the IonDrive Turbo V ion source cool for at least 90 minutes before starting any maintenance procedures. Some surfaces of the ion source and vacuum interface become hot during operation.

---



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** Deposit hazardous materials in appropriately labeled waste containers and dispose of them according to local regulations.

---



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** Take care to vent exhaust gases to a dedicated laboratory fume hood or exhaust system and make sure that the ventilation tubing is secured with clamps. Make sure that the laboratory has appropriate air exchange for the work performed.

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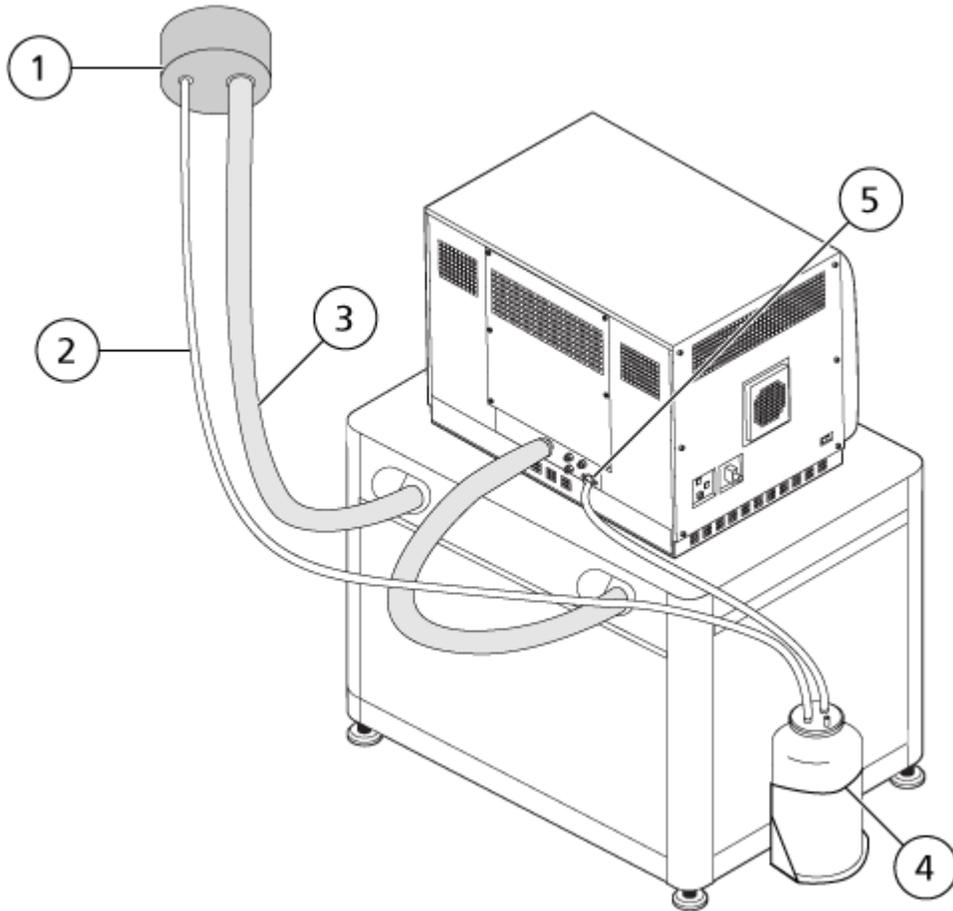
**Note:** Make sure that there are no kinks, sags, or twists in the source waste line.

---

Inspect the source exhaust drain bottle regularly, and empty it before it becomes full. Also inspect the bottle and the fitting for leaks, and tighten connections or replace components, if required. Follow the steps in this procedure to empty the bottle.

1. Remove the ion source.
2. Loosen the clamps connecting the hoses to the cap of the source exhaust drain bottle.

Figure 16-1 Source Exhaust Drain Bottle



Item	Description
1	Connection to vent
2	Source exhaust drain tubing: 2.5 cm (1.0 inch) inner diameter (i.d.)
3	Roughing pump exhaust hose: 3.2 cm (1.25 inch) i.d.
4	Source exhaust drain bottle Make sure that the bottle is secured to prevent spillage.
5	Source exhaust connection to the mass spectrometer: 1.6 cm (0.625 inch) i.d.

**Note:** Source exhaust hose connections at the drain bottle, mass spectrometer, and the lab vent are secured with hose clamps.

3. Lift the drain bottle out of the holder.
4. Disconnect the hoses from the cap.
5. Remove the cap from the drain bottle.
6. Empty the drain bottle and then dispose of the waste according to laboratory procedures and local waste regulations.
7. Install the cap on the bottle and then put the bottle in the holder.
8. Attach the hoses to the cap and then secure them tightly with clamps.

## Clean the Front-End

The following warning applies to all of the procedures in this section:



---

**WARNING! Hot Surface Hazard. Let the IonDrive Turbo V ion source cool for at least 90 minutes before starting any maintenance procedures. Some surfaces of the ion source and vacuum interface become hot during operation.**

---

Clean the mass spectrometer front-end using the routine cleaning method, to:

- Minimize unscheduled system downtime.
- Maintain optimum sensitivity.
- Avoid more extensive cleaning that requires a service visit.

When contamination occurs, perform an initial routine cleaning. Clean up to and including the front of the orifice plate. If routine cleaning does not resolve issues with sensitivity, then a full cleaning might be necessary. Contact the local QMP or FSE.

This section provides instructions for performing routine cleaning without breaking vacuum.

---

**Note:** Follow all of the applicable local regulations. For health and safety guidelines, refer to the section: [Chemical Precautions](#).

---

## Symptoms of Contamination

The system might be contaminated if any one of the following is observed:

- Significant loss in sensitivity
- Increased background noise
- Additional peaks that are not part of the sample in full scan or survey scan methods

## Service and Maintenance Information — Mass Spectrometer

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If any of these issues are observed, then clean the mass spectrometer front-end.

### Required Materials (Not Supplied)

---

**Note:** For consumables ordering information and inquiries, refer to the *MD Instrument Family Parts and Equipment Guide*. Contact a local FSE or visit [sciex.com](http://sciex.com) for more information.

---

- Powder-free gloves, nitrile or neoprene recommended
- Safety glasses
- Laboratory coat
- Fresh LC-MS-grade water. Old water can contain contaminants that can further contaminate the mass spectrometer.
- LC-MS-grade methanol, isopropanol (2-propanol), or acetonitrile
- Cleaning solution. Use one of:
  - 100% methanol
  - 100% isopropanol
  - 1:1 acetonitrile:water solution, freshly prepared
  - 1:1 acetonitrile:water with 0.1% acetic acid solution, freshly prepared
- Clean 1 L or 500 mL glass beaker to prepare cleaning solutions
- 1 L beaker to catch used solvent
- Organic waste container
- Lint-free wipes. Refer to the section: [Tools and Supplies Available from the Manufacturer](#).
- (Optional) Polyester (poly) swabs

### Tools and Supplies Available from the Manufacturer

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**Note:** For part numbers, refer to the document: *Parts and Equipment Guide*.

---

- Small poly swab, thermally bonded. Also available in the Cleaning kit.
- Lint-free wipe (11 cm x 21 cm, 4.3 inches x 8.3 inches). Also available in the Cleaning kit.
- Cleaning kit. Contains the small poly swab, lint-free wipes, Q0 cleaning tool, tapered IonDrive QJet ion guide cleaning brush, Q0 cleaning brush, and Alconox.

## Cleaning Best Practices



**WARNING! Hot Surface Hazard.** Let the IonDrive Turbo V ion source cool for at least 90 minutes before starting any maintenance procedures. Some surfaces of the ion source and vacuum interface become hot during operation.

---



**WARNING! Toxic Chemical Hazard.** Refer to the chemical product safety data sheets and follow all of the recommended safety procedures when handling, storing, and disposing of chemicals.

---



**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** Determine whether decontamination is required before cleaning or maintenance. If radioactive materials, biological agents, or toxic chemicals have been used with the system, then the customer must decontaminate the system before cleaning or maintenance.

---



**WARNING! Environmental Hazard.** Do not dispose of system components in municipal waste. Follow local regulations when disposing of components.

---

- Allow the ion source to cool before removing it.
- Always wear clean, powder-free gloves, nitrile or neoprene recommended, for the cleaning procedures.
- After cleaning the mass spectrometer components, and before reassembling them, put on a new, clean pair of gloves.
- Do not use cleaning supplies other than those specified in this procedure.
- If possible, prepare cleaning solutions just before cleaning.
- Prepare and store all of the organic solutions and organic-containing solutions in very clean glassware only. Never use plastic bottles. Contaminants can leach from these bottles and further contaminate the mass spectrometer.
- To avoid contaminating the cleaning solution, pour the solution on the wipe or swab.
- Allow only the center area of the wipe to contact the mass spectrometer surface. Cut edges can leave fibers behind.

---

**Tip!** Wrap the wipe around a thermally-bonded poly swab.

---

**Figure 16-2 Example: Folding the Wipe**



- To avoid cross-contamination, discard the wipe or swab after it has touched the surface once.
- If required, perform multiple cleanings, using multiple wipes for larger parts of the vacuum interface, such as the curtain plate.
- Only dampen the wipe or swab slightly when applying water or cleaning solution. Water, more often than organic solvents, might cause the wipe to deteriorate, leaving residue on the mass spectrometer.
- Do not rub the wipe across the aperture. Wipe around the aperture to prevent fibers from the wipes from entering the mass spectrometer.
- Do not insert the brush in the aperture on the curtain plate or orifice plate.

## Prepare the Mass Spectrometer



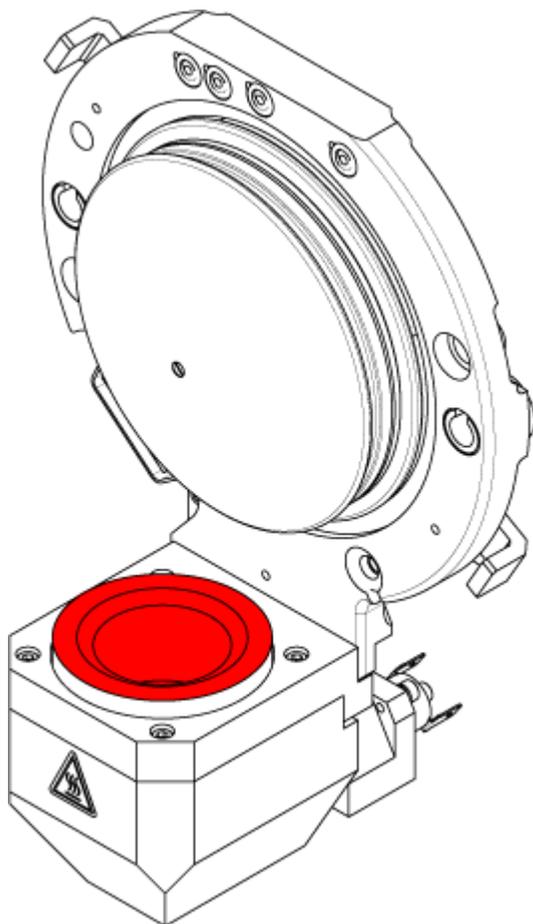
---

**WARNING! Hot Surface Hazard. Let the IonDrive Turbo V ion source cool for at least 90 minutes before starting any maintenance procedures. Some surfaces of the ion source and vacuum interface become hot during operation.**

---

**CAUTION: Potential System Damage. Do not drop anything in the source drain when the ion source is removed.**

**Figure 16-3 Source Drain on the Vacuum Interface**



1. Deactivate the hardware profile. Refer to the document: *Software User Guide*.
2. Remove the ion source. Refer to the section: [Remove the Ion Source](#).  
When the ion source is not in use, store it to protect it from damage and to maintain operating integrity.

## Clean the Curtain Plate

**CAUTION: Potential System Damage. Do not rest the curtain plate or orifice plate on the aperture tip. Make sure that the conical side of the curtain plate faces up.**

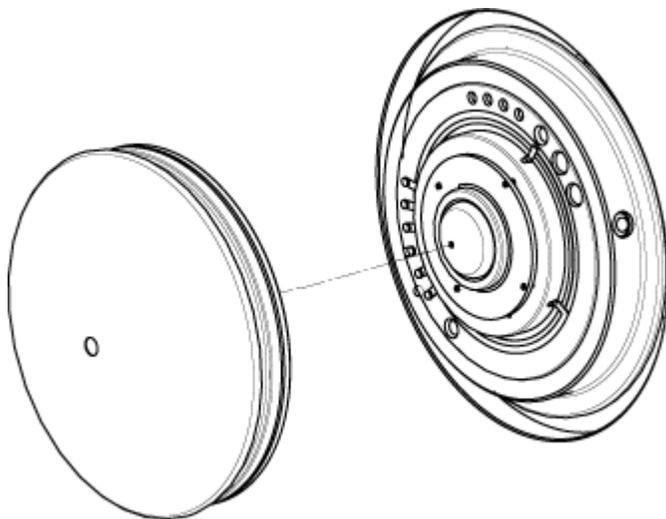
---

**CAUTION: Potential System Damage.** To avoid damaging the aperture, do not insert a wire or metal brush in the aperture on the curtain plate, orifice plate, or interface heater.

---

1. Pull the curtain plate off of the vacuum interface and then put it, conical side up, on a clean, stable surface.

**Figure 16-4 Curtain Plate Removal**



The curtain plate is held in place by three retaining ball catches mounted on the orifice plate.

---

**Tip!** If the curtain plate does not immediately separate from the orifice plate, then turn the curtain plate slightly, less than 90 degrees, to release the ball spring latches.

---

2. Dampen a lint-free wipe with LC-MS-grade water and then clean both sides of the curtain plate.
- 

**Note:** Use multiple wipes, as required.

---

3. Repeat step 2 using the cleaning solution.
  4. Using a dampened wipe or small poly swab, clean the aperture.
  5. Wait until the curtain plate is dry.
  6. Inspect the curtain plate for solvent stains or lint, removing any residue with a clean, slightly damp, lint-free wipe.
- 

**Note:** Persistent spotting or filming is an indicator of contaminated solvent.

---

## Clean the Front of the Orifice Plate

---

**CAUTION: Potential System Damage.** When cleaning the surface of the orifice plate, do not remove the interface heater. Frequent removal of the interface heater can result in damage to the interface heater. Surface cleaning of the interface heater is adequate for routine cleaning.

---

**CAUTION: Potential System Damage.** To avoid damaging the aperture, do not insert a wire or metal brush in the aperture on the curtain plate, orifice plate, or interface heater.

---

1. Dampen a lint-free wipe with LC-MS-grade water and then wipe the front of the orifice plate, including the interface heater.
  2. Repeat step 1 using the cleaning solution.
  3. Wait until the orifice plate is dry.
  4. Inspect the orifice plate for solvent stains or lint, removing any residue with a clean, slightly damp, lint-free wipe.
- 

**Note:** Persistent spotting or filming is an indicator of contaminated solvent.

---

## Put the Mass Spectrometer Back in Service

1. Install the curtain plate.
2. Install the ion source on the mass spectrometer. Refer to [Install the Ion Source on the Mass Spectrometer](#).  
Tighten the ion source by turning the source latches down in the locking position.
3. If the mass spectrometer is connected to the LC system, then restore all of the connections to the LC system.
4. Activate the hardware profile. Refer to the document: *Software User Guide*.

## Storage and Handling



**WARNING! Environmental Hazard.** Do not dispose of system components in municipal waste. Follow local regulations when disposing of components.

---

If the mass spectrometer must be stored for a long time or prepared for shipping, then contact a SCIEX FSE for decommissioning information. To disconnect power from the mass spectrometer, remove the mains supply connector from the AC mains supply.

**Note:** The ion source and mass spectrometer must be transported and stored at a temperature between  $-30\text{ }^{\circ}\text{C}$  to  $+60\text{ }^{\circ}\text{C}$  ( $-22\text{ }^{\circ}\text{F}$  to  $140\text{ }^{\circ}\text{F}$ ) and relative humidity not exceeding 99%, non-condensing. Store the system at an altitude not exceeding 2,000 m (6,562 feet) above sea level.

---

## Inspect the Roughing Pump Oil Level

Inspect the sight glass on the roughing pump to verify that the oil is above the minimum level.

If the oil is below the minimum level, then contact the Qualified Maintenance Person (QMP) or SCIEX Field Service Employee (FSE).

## Service and Maintenance — Ion Source

The following warnings apply to all of the maintenance procedures in this section.



---

**WARNING! Hot Surface Hazard.** Let the IonDrive Turbo V ion source cool for at least 90 minutes before starting any maintenance procedures. Some surfaces of the ion source and vacuum interface become hot during operation.

---



---

**WARNING! Fire and Toxic Chemical Hazard.** Keep flammable liquids away from flame and sparks and use them only in vented chemical fume hoods or safety cabinets.

---



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**WARNING! Toxic Chemical Hazard.** Wear personal protective equipment, including a laboratory coat, gloves, and safety glasses, to avoid skin or eye exposure.

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**WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard.** In the event of a chemical spill, review product safety data sheets for specific instructions. Make sure that the system is in Standby state before cleaning a spill near the ion source. Use appropriate personal protective equipment and absorbent wipes to contain the spill and dispose of it following local regulations.

---



---

**WARNING! Electrical Shock Hazard.** Avoid contact with the high voltages applied to the ion source during operation. Put the system in Standby state before adjusting the sample tubing or other equipment near the ion source.

---



**WARNING! Puncture Hazard, Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Discontinue use of the ion source if the ion source window is cracked or broken, and then contact a SCIEX Field Service Employee (FSE). Any toxic or injurious materials introduced in the equipment will be present in the source exhaust output. Exhaust from equipment should be vented from the room. Dispose of sharps following established laboratory safety procedures.**

---

**CAUTION: Potential System Damage. Do not lift or carry the ion source with one hand. The ion source is designed to be lifted or carried using two hands, one on each side of the ion source.**

---

This section contains general maintenance procedures for the ion source. To determine how often to clean or perform maintenance on the ion source, consider the following:

- Compounds tested
- Cleanliness of the samples and sample preparation techniques
- Amount of time an idle probe contains a sample
- Overall system run time

These factors can cause changes in ion source performance, indicating that maintenance is required.

Make sure that the installed ion source is fully sealed to the mass spectrometer with no evidence of gas leaks. Regularly inspect the ion source and its fittings for leaks. Clean the ion source components regularly to keep the ion source in good working condition.

**CAUTION: Potential System Damage. Use only the recommended cleaning methods and materials to avoid damaging the equipment.**

---

### Required Materials

- 1/4 inch open-ended wrench
- Flat-bladed screwdriver
- LC-MS-grade methanol
- LC-MS-grade deionized water
- Safety glasses
- Breathing mask and filter
- Powder-free gloves, nitrile or neoprene recommended
- Lab coat

## IonDrive Turbo V Ion Source

Surfaces of the ion source become hot during operation. The following figures show surfaces that are cooler (blue and gray) and surfaces that remain hot for an extended period of time (red). Do not touch the surfaces shown in red while using or removing the ion source.

**Figure 16-5 Hot Surfaces: ESI Mode (Red=Hot, Gray=Warm, Blue=Handle with Care)**

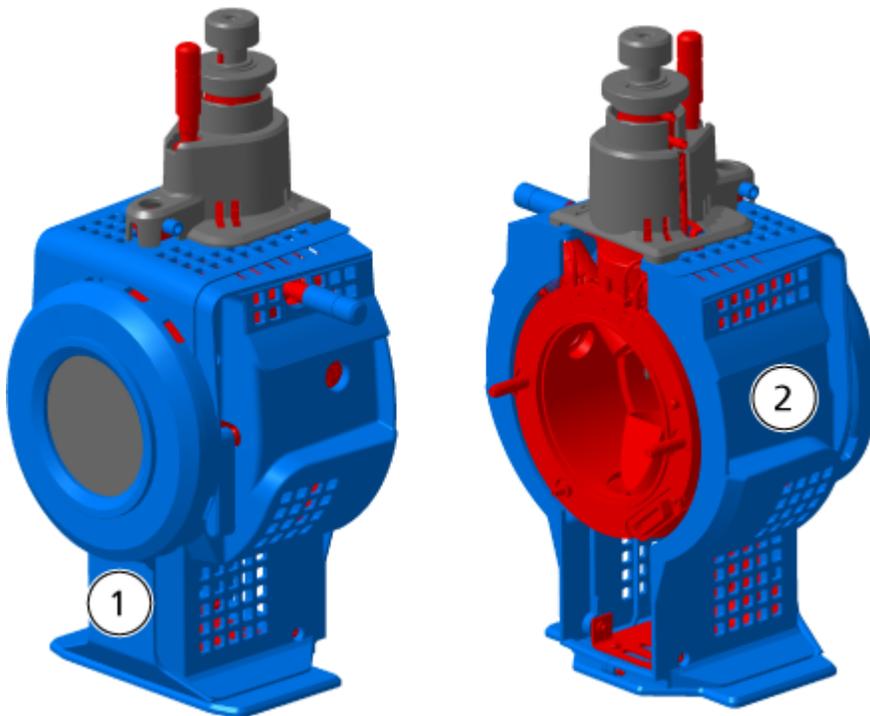
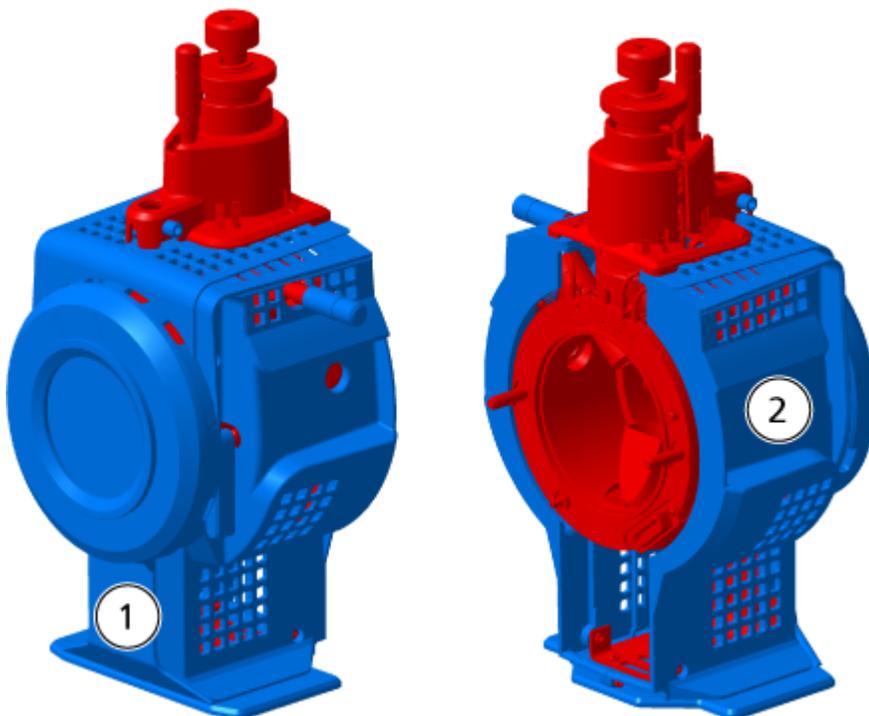


Figure 16-6 Hot Surfaces: APCI Mode (Red=Hot, Blue=Handle with Care)



Item	Description
1	Front
2	Back

## Remove the Ion Source

**Note:** Nitrogen continues to flow at a rate of 5.3 L/min when the mass spectrometer is off or when the ion source is removed from the system. To minimize nitrogen gas consumption and to keep the mass spectrometer clean when it is not in use, leave the ion source installed on the mass spectrometer and leave the system on.

The ion source can be removed quickly and easily, without tools. Always remove the ion source from the mass spectrometer before performing any maintenance on the ion source or exchanging the probes.

1. Stop any ongoing scans.
2. Turn off the sample stream.
3. Set the ion source **Temperature** to 0, if the heaters are in use.

## Service and Maintenance Information — Mass Spectrometer

---

4. Wait at least 90 minutes for the ion source to cool.
5. Disconnect the sample tubing from the grounding union.
6. Turn the two source latches up to the 12 o'clock position to release the ion source.
7. Pull the ion source gently away from the vacuum interface.

---

**Note:** Be careful not to lose the O-rings that are installed on the vacuum interface.

---

8. Put the ion source on a clean, secure surface.

## Clean the Surfaces



**WARNING! Electrical Shock Hazard. Make sure that the ion source is completely disconnected from the mass spectrometer before proceeding.**

---

Clean the surfaces of the ion source after a spill or when they become dirty.

Prerequisite Procedures
<ul style="list-style-type: none"><li>• <a href="#">Remove the Ion Source</a></li></ul>



Wipe the surfaces of the ion source with a soft, damp, cloth.

## Clean the Probe

Flush the ion source periodically, regardless of the type of compounds sampled. Do this by setting up a method in the control software specifically for performing a flushing operation.

1. Change to a mobile phase that is 1:1 water:acetonitrile or 1:1 water:methanol.
2. Adjust the position of the probe so that it is as far from the orifice as possible.
3. In the control software, do the following:
  - a. Create an MS method.
  - b. Set the ion source temperature between 500 °C and 600 °C.
  - c. Set ion source gas 1 and ion source gas 2 to at least 40.
  - d. Set the flow rate of the gas for the Curtain Gas interface to the highest setting possible.
4. Wait until the temperature setpoint is reached.
5. Make sure that the probe and sample tubing are flushed thoroughly.

## Remove the Probe



**WARNING! Electrical Shock Hazard.** Remove the ion source from the mass spectrometer before starting this procedure. Follow all electrical safe work practices.

---

**CAUTION: Potential System Damage.** Do not let the protruding electrode tip or the corona discharge needle touch any part of the ion source housing, to avoid damaging the probe.

---

### Prerequisite Procedures

- [Remove the Ion Source.](#)

The probe can be removed quickly and easily, without tools. Always remove the ion source from the mass spectrometer before changing probes or performing maintenance on the probe.

1. Loosen the sample tubing nut and then disconnect the sample tubing from the probe.
2. Loosen the retaining ring that secures the probe on the ion source housing.
3. Gently pull the probe straight up out of the tower.
4. Put the probe on a secure, clean surface.

## Replace the Electrode



**WARNING! Electrical Shock Hazard.** Remove the ion source from the mass spectrometer before starting this procedure. Follow all electrical safe work practices.

---



**WARNING! Puncture Hazard.** Be careful when handling the electrode. The tip of the electrode is extremely sharp.

---

### Prerequisite Procedures

- [Remove the Ion Source.](#)
- [Remove the Probe.](#)

The probe contains an electrode. Replace the electrode when there is a decrease in performance.

**Note:** After replacing the electrode, evaluate the effect of the change on system performance.

---

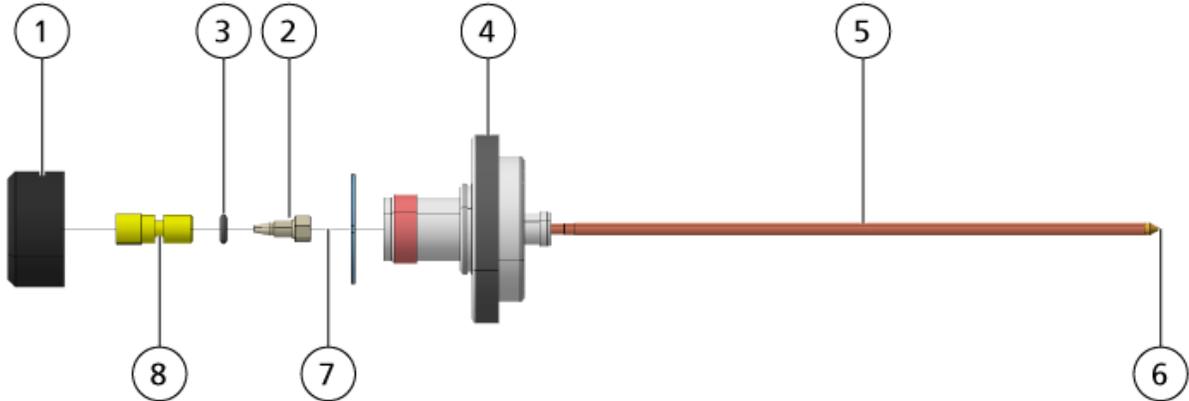
This procedure applies to both probes.

---

## Service and Maintenance Information — Mass Spectrometer

1. Remove the electrode adjustment nut and then remove the electrode.
2. Holding the probe with the tip pointing down, so that the spring remains inside the probe, install a sample fitting in the PEEK union, tightening until it is finger-tight.

**Figure 16-7 Probe, Expanded View**



Item	Description
1	Electrode adjustment nut
2	1/4 inch retaining nut
3	Spring
4	Retaining ring
5	Sprayer tube
6	Electrode tip
7	Electrode tube
8	PEEK union

3. Pull the PEEK union and the attached electrode tube from the probe.
4. Remove the sample fitting from the PEEK union.
5. Use the 1/4 inch open-ended wrench to remove the retaining nut that holds the electrode tube in the PEEK union.
6. Remove the electrode tube from the retaining nut.
7. Insert the new electrode tube in the retaining nut and then in the PEEK union.  
Make sure that the electrode tube is inserted as far as possible in the PEEK union. If there is a gap between the electrode tube and its seat inside the union, a dead volume may occur.
8. Tighten the retaining nut.

Do not cross-thread or over-tighten the retaining nut or the tubing might leak.

9. Make sure that the spring is still inside the probe and then tighten the electrode adjustment nut.
10. Align the electrode tube with the narrow opening in the sprayer tube and then insert the PEEK union and attached electrode tube in the probe. Be careful not to bend the electrode tube.
11. Install and then tighten the electrode adjustment nut.
12. Install the probe. Refer to the section: [Install the Probe](#).
13. Install the ion source on the mass spectrometer. Refer to the section: [Ion Source Installation](#).
14. Connect the sample tubing. Refer to the section: [Connect the Ion Source Tubing](#).
15. Adjust the electrode tip extension. Refer to the section: [Optimize the TurbolonSpray Probe Position](#) or [Optimize the APCI Probe Position](#).

## Replace the Corona Discharge Needle



---

**WARNING! Electrical Shock Hazard.** Remove the ion source from the mass spectrometer before starting this procedure. Follow all electrical safe work practices.

---



---

**WARNING! Puncture Hazard.** Handle the needle with care. The tip of the needle is extremely sharp.

---

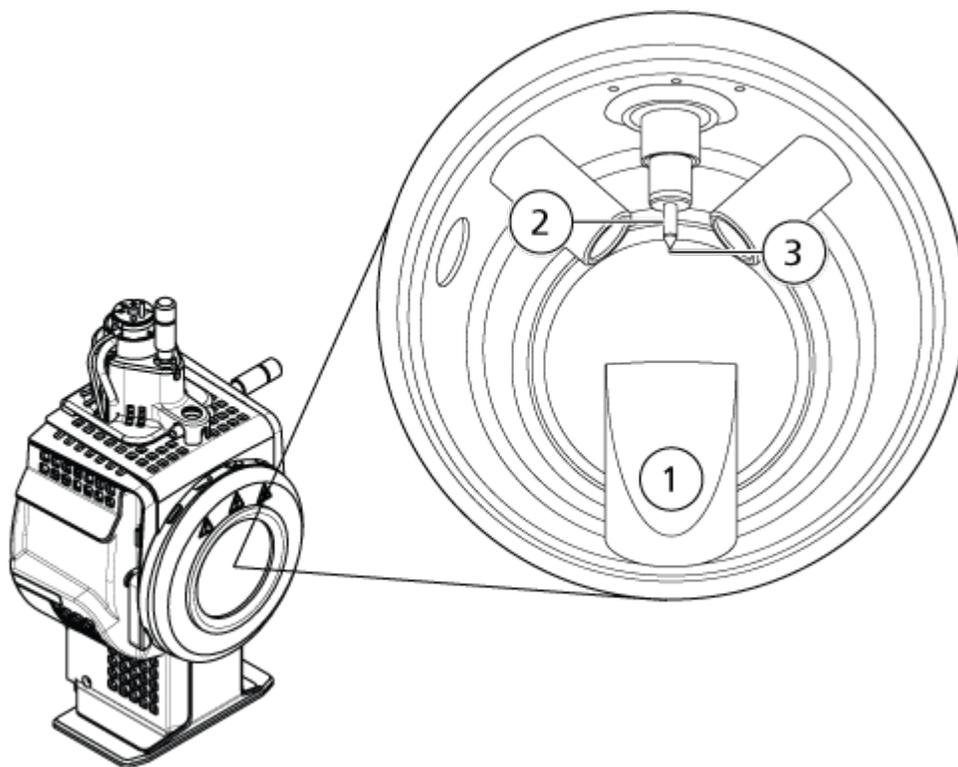
Prerequisite Procedures
<ul style="list-style-type: none"><li>• <a href="#">Remove the Ion Source</a>.</li><li>• <a href="#">Remove the Probe</a>.</li></ul>



If the corona discharge needle tip becomes corroded, then it might not be removable by hand. If it cannot be removed, then cut off the needle tip to remove it and replace the entire corona discharge needle.

1. Rotate the ion source so that the open side is accessible.

**Figure 16-8 Corona Discharge Needle**



Item	Description
1	Exhaust chimney
2	Ceramic sleeve
3	Corona discharge needle tip

2. While holding the corona discharge needle adjustment screw between the thumb and forefinger of one hand and the corona discharge needle with the other hand, rotate the corona discharge needle tip counter-clockwise to loosen and then gently remove the tip. Refer to the section: [Ion Source Components](#).
3. Gently pull the corona discharge needle down through the exhaust chimney to remove it.
4. Insert the new needle through the exhaust chimney in the ceramic sleeve as far as possible.
5. Holding a new tip between the thumb and forefinger of one hand and the corona discharge needle adjustment screw with the other hand, rotate the corona discharge needle tip clockwise to install the tip.
6. Insert the probe and then install the ion source on the mass spectrometer. Refer to the section: [Ion Source Installation](#).

## Replace the Sample Tubing



**WARNING! Electrical Shock Hazard.** Remove the ion source from the mass spectrometer before starting this procedure. Follow all electrical safe work practices.

---

### Prerequisite Procedures

- Stop the sample flow and make sure that any remaining gas has been removed through the source exhaust system.
- Remove the ion source. Refer to the section: [Remove the Ion Source](#).

Use the following procedure to replace the sample tubing if it has a blockage.

1. Disconnect the sample tubing from the probe and the grounding union.
2. Replace the sample tubing with an appropriate length of tubing, cut with a proper tubing cutter. Refer to the section: [Connect the Ion Source Tubing](#).
3. Install the ion source. Refer to the section: [Ion Source Installation](#).
4. Start the sample flow.

# Mass Spectrometer Troubleshooting 17

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This section contains information for troubleshooting basic system issues. Certain activities can only be carried out by a SCIEX-trained Qualified Maintenance Person (QMP) in the laboratory. For advanced troubleshooting, contact a SCIEX Field Service Employee (FSE).

**Table 17-1 System Issues**

Symptom	Possible Cause	Corrective Action
The IonDrive QJet ion guide is extremely dirty or frequently becomes dirty.	The flow rate of the gas for the Curtain Gas interface is too low.	Examine the setting for the gas for the Curtain Gas interface and increase it, if applicable.
A system fault has occurred because the vacuum pressure is too high.	<ol style="list-style-type: none"><li>1. The oil level is too low.</li><li>2. There is a leak.</li><li>3. The wrong orifice plate is installed.</li></ol>	<ol style="list-style-type: none"><li>1. Inspect the oil level in the roughing pump, and then contact the local QMP or an FSE to add oil. Refer to the section: <a href="#">Inspect the Roughing Pump Oil Level</a>.</li><li>2. Inspect and repair leaks.</li><li>3. Install the correct orifice plate.</li></ol>
A system fault has occurred because the QPS exciter module temperature is too high.	<ol style="list-style-type: none"><li>1. The mass spectrometer air filter is blocked.</li><li>2. Ambient temperature is too high.</li></ol>	<ol style="list-style-type: none"><li>1. Contact the local QMP or FSE.</li><li>2. For the ambient temperature specifications, refer to the document: <i>Site Planning Guide</i> for the system.</li></ol>

Table 17-1 System Issues (continued)

Symptom	Possible Cause	Corrective Action
The control software reports that the mass spectrometer is in Fault status because of the ion source.	<ol style="list-style-type: none"> <li>1. The probe is not installed.</li> <li>2. The probe is not connected securely.</li> </ol>	<p>Confirm the fault in the Status panel of the device details page.</p> <ol style="list-style-type: none"> <li>1. Install the probe. Refer to the section: <a href="#">Install the Probe</a>.</li> <li>2. Remove and then install the probe. Tighten the retaining ring securely. Refer to the section: <a href="#">Remove the Probe and Install the Probe</a>.</li> </ol>
The control software indicates that the APCI probe is in use, but the TurbolonSpray probe is installed.	The F3 fuse is blown.	Contact an FSE.
The spray is not uniform.	The electrode is blocked.	Clean or replace the electrode. Refer to the section: <a href="#">Replace the Electrode</a> .
The interface heater is not ready.	The interface heater is faulty.	Contact the local QMP or FSE.
The mass spectrometer resolution is poor.	The mass spectrometer is not tuned.	Use the Instrument Optimization wizard to optimize the mass spectrometer. Refer to the documents: <i>Software User Guide</i> or <i>Help</i> .

## Mass Spectrometer Troubleshooting

**Table 17-1 System Issues (continued)**

Symptom	Possible Cause	Corrective Action
The mass spectrometer performance has degraded.	<ol style="list-style-type: none"> <li>1. The ion source conditions are not optimized.</li> <li>2. The sample was not prepared correctly or the sample has degraded.</li> <li>3. The sample inlet fittings are leaking.</li> </ol>	<ol style="list-style-type: none"> <li>1. Optimize the ion source conditions. Refer to the section: <a href="#">Optimize the TurbolonSpray Probe Position</a> or <a href="#">Optimize the APCI Probe Position</a>.</li> <li>2. Confirm that the sample was prepared correctly.</li> <li>3. Verify that the fittings are the right size and type and make sure that they are tight. Do not overtighten the fittings. Replace the fittings if leaks continue.</li> <li>4. Install and optimize an alternate ion source.</li> <li>5. Contact an FSE if the issue persists.</li> </ol>
Arcing or sparks occur.	The position of the corona discharge needle is incorrect.	If the TurbolonSpray probe is being used, then turn the corona discharge needle toward the curtain plate and away from the stream of heater gas. Refer to the section: <a href="#">Adjust the Position of the Corona Discharge Needle</a> .

**Table 17-2 Sensitivity Issues**

Possible Cause	Corrective Action
<b>Sensitivity is Decreased</b>	
The ion source parameters are not optimized.	Optimize the ion source parameters.
The mass spectrometer is not optimized.	Use the Instrument Optimization wizard to optimize the mass spectrometer.

**Table 17-2 Sensitivity Issues (continued)**

Possible Cause	Corrective Action
The curtain plate is dirty.	Clean the curtain plate. Refer to the section: <a href="#">Clean the Curtain Plate</a> .
The orifice plate is dirty.	Refer to the section: <a href="#">Clean the Front of the Orifice Plate</a> or contact the local QMP or FSE.
The IonDrive QJet ion guide or IQ0 lens is dirty.	Clean the IonDrive QJet ion guide and IQ0 lens. Contact the local QMP or FSE.
Q0 region is dirty.	Test for contamination of the Q0 region. Contact the local QMP or FSE.
The syringe or sample line is leaking.	Inspect the syringe or sample line for leaks and repair any leaks found. Make sure that all fittings are the correct type and size.
The sample has degraded or has a low concentration.	Verify the sample concentration. Use a fresh sample.
The probe is not installed properly.	Remove and install the probe.
The ion source is not installed properly or it is faulty.	Remove and install the ion source, making sure that the latches are properly secured. If this does not resolve the issue, then install and optimize an alternate ion source.
One or more of the O-rings on the vacuum interface is missing.	If the O-rings are on the ion source, then install them on the vacuum interface. If they are missing, then replace them.
There is an issue with the LC system or connections.	Troubleshoot the LC system.
The Declustering Potential (DP) is not optimized.	Optimize DP.
The electrode is dirty or blocked.	Replace the electrode. Refer to the section: <a href="#">Replace the Electrode</a> .
<b>There is No Signal or the Signal is Unstable</b>	
The tubing is blocked.	Replace the sample tubing. Refer to the section: <a href="#">Connect the Ion Source Tubing</a> .

## Mass Spectrometer Troubleshooting

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**Table 17-3 Background Noise Issues**

Possible Cause	Corrective Action
The <b>Temperature (TEM)</b> , IonSpray voltage (IS) or heater gas flow rate (GS2) is too high.	Optimize the ion source parameters. Refer to the section: <a href="#">TurbolonSpray Probe Optimization</a> or <a href="#">APCI Probe Optimization</a> .
The syringe or sample line is dirty.	Clean or replace the syringe or sample line.
The curtain plate is dirty.	Clean the curtain plate. Refer to the section: <a href="#">Clean the Curtain Plate</a> .
The orifice plate is dirty.	Clean the front of the orifice plate. Refer to the section: <a href="#">Clean the Front of the Orifice Plate</a> .
The IonDrive QJet ion guide or IQ0 lens is dirty.	Do a full cleaning of the front end components of the mass spectrometer. Contact the local QMP or FSE.
The Q0 region is dirty.	Clean the Q0 region. Contact the QMP or FSE.
Mobile phase is contaminated.	Replace the mobile phase.
The ion source is contaminated.	Clean or replace the ion source components and then condition the ion source and front end: <ol style="list-style-type: none"><li>1. Move the probe to the furthest position from the aperture, vertically and horizontally.</li><li>2. (Analyst MD software) Make sure that the interface heater is on.</li><li>3. Infuse or inject 50:50 methanol:water with a pump flow rate of 1 mL/min.</li><li>4. In the control software, set the temperature to 650, ion source gas 1 to 60, and ion source gas 2 to 60.</li><li>5. Set the flow rate for the gas for the Curtain Gas interface to 45 or 50.</li><li>6. Run for a minimum of 2 hours or preferably overnight for best results.</li></ol>

For sales, technical assistance, or service, contact an FSE or visit the SCIEX website at [sciex.com](http://sciex.com) for contact information.

# Citrine System Parameters

# A

Citrine systems use the IonDrive Turbo V ion source.

The first number under each scan type is the preset value. The range of numbers is the accessible range for each parameter.

**Table A-1 Citrine System Parameters**

Parameter ID	Access ID	Positive Polarity			Negative Polarity		
		Q1	Q3	MS/MS	Q1	Q3	MS/MS
CUR	CUR	20 20 to 55	20 20 to 55	20 20 to 55	20 20 to 55	20 20 to 55	20 20 to 55
CAD <sup>3 4</sup>	CAD	0 N/A	6 N/A	Med Low, Medium, High	0 N/A	6 N/A	Med Low, Medium, High
CAD <sup>5 6</sup>	CAD	0 N/A	6 N/A	9 0 to 12	0 N/A	6 N/A	9 0 to 12

<sup>3</sup> Citrine QTRAP MS/MS system, Low Mass (LM)

<sup>4</sup> Citrine QTRAP MS/MS system, High Mass (HM)

<sup>5</sup> Citrine Triple Quad MS/MS system, Low Mass (LM)

<sup>6</sup> Citrine Triple Quad MS/MS system, High Mass (HM)

## Citrine System Parameters

Table A-1 Citrine System Parameters (continued)

Parameter ID	Access ID	Positive Polarity			Negative Polarity		
		Q1	Q3	MS/MS	Q1	Q3	MS/MS
IS	IS	5500 0 to 5500	5500 0 to 5500	5500 0 to 5500	-4500 -4500 to 0	-4500 -4500 to 0	-4500 -4500 to 0
NC <sup>7</sup>	NC	3 0 to 5	3 0 to 5	3 0 to 5	-3 -5 to 0	-3 -5 to 0	-3 -5 to 0
TEM	TEM	0 0 to 750	0 0 to 750	0 0 to 750	0 0 to 750	0 0 to 750	0 0 to 750
OR (DP = OR)	DP	100 0 to 300	100 0 to 300	100 0 to 300	-100 -300 to 0	-100 -300 to 0	-100 -300 to 0
Q0 (EP = -Q0)	EP	10 2 to 15	10 2 to 15	10 2 to 15	-10 -15 to -2	-10 -15 to -2	-10 -15 to -2
IQ1 (IQ1 = Q0 + offset)	IQ1	Q0 + (-0.5) -0.1 to -2	Q0 + (-0.5) -0.1 to -2	Q0 + (-0.5) -0.1 to -2	Q0 + 0.5 0.1 to 2	Q0 + 0.5 0.1 to 2	Q0 + 0.5 0.1 to 2
ST (ST = Q0 + offset)	ST	Q0 + (-8) -12 to -5	Q0 + (-8) -12 to -5	Q0 + (-8) -12 to -5	Q0 + 8 5 to 12	Q0 + 8 5 to 12	Q0 + 8 5 to 12

<sup>7</sup> APCI probe

Table A-1 Citrine System Parameters (continued)

Parameter ID	Access ID	Positive Polarity			Negative Polarity		
		Q1	Q3	MS/MS	Q1	Q3	MS/MS
RO1 (IE1 = Q0 – RO1)	IE1	1 0 to 3	N/A	1 0 to 3	–1 –3 to –0	N/A	–1 –3 to –0
IQ2 (IQ2 = Q0 + offset)	IQ2	Q0+ (–10) –30 to –8	Q0+ (–10) –30 to –8	Q0+ (–10) –30 to –8	Q0 + 10 8 to 30	Q0 + 10 8 to 30	Q0 + 10 8 to 30
RO2	RO2	–20 N/A	–20 N/A	N/A	20 N/A	20 N/A	N/A
RO2 (CE = Q0 – RO2)	CE	N/A	N/A	30 5 to 180	N/A	N/A	–30 –180 to –5
ST3 (ST3 = RO2 + offset)	ST3	RO2 – 10 –30 to –5	N/A	N/A	RO2 + 10 5 to 30	N/A	N/A
ST3 (CXP = RO2 – ST3)	CXP	N/A	15 0 to 55	15 0 to 55	N/A	–15 –55 to 0	–15 –55 to 0

## Citrine System Parameters

**Table A-1 Citrine System Parameters (continued)**

Parameter ID	Access ID	Positive Polarity			Negative Polarity		
		Q1	Q3	MS/MS	Q1	Q3	MS/MS
RO3	RO3	-50 N/A	N/A	N/A	50 N/A	N/A	N/A
RO3 (IE3 = RO2 – RO3)	IE3	N/A	1 0 to 5	1 0 to 5	N/A	-1 -5 to 0	-1 -5 to 0
CEM	CEM	1700 0 to 3300					
GS1	GS1	20 0 to 90					
GS2	GS2	0 0 to 90					

**Table A-2 Citrine System Parameters for LIT Scan Types Only**

Parameter ID	Access ID	Positive Polarity	Negative Polarity
CAD	CAD	High Low, Medium, High	High Low, Medium, High

Table A-2 Citrine System Parameters for LIT Scan Types Only (continued)

Parameter ID	Access ID	Positive Polarity	Negative Polarity
AF2 <sup>8</sup>	AF2	0.1 0 to 1	0.1 0 to 1
AF3	AF3	Mass-Speed Dependent 0 to 10	Mass-Speed Dependent 0 to 10
EXB	EXB	Mass-Speed Dependent -165 to 0	Mass-Speed Dependent 0 to 165
CES	CES	0 0 to 50	0 0 to 50
ROS (Q0 - ROS)	CE	10 5 to 180	-10 -5 to -180

<sup>8</sup> MS/MS/MS only

# Source Parameters and Voltages

# B

## TurbolonSpray Probe Parameters

The following table shows the recommended operating conditions for the TurbolonSpray probe at three different flow rates. For each flow rate, the flow rate of the gas for the Curtain Gas interface should be as high as possible. The solvent composition used for optimization was 1:1 water:acetonitrile. These conditions represent a starting point from which to optimize the probe. Using an iterative process, optimize the parameters using flow injection analysis to achieve the best signal or signal-to-noise for the compound of interest.

**Table B-1 Parameter Optimization for the TurbolonSpray Probe**

Parameters	Typical Values			Operational Range
	Low	Medium	High	
LC flow rate	5 $\mu\text{L}/\text{min}$ to 50 $\mu\text{L}/\text{min}$	200 $\mu\text{L}/\text{min}$	1,000 $\mu\text{L}/\text{min}$	5 $\mu\text{L}/\text{min}$ to 3,000 $\mu\text{L}/\text{min}$
Ion source gas 1 (nebulizer gas)	20 psi to 40 psi	40 psi to 60 psi	40 psi to 60 psi	0 psi to 90 psi
Ion source gas 2 (heater gas)	0 psi	50 psi	50 psi	0 psi to 90 psi
<b>IonSpray Voltage</b>	5500 V	5500 V	5500 V	5500 V
Gas for the Curtain Gas interface	30 psi	30 psi	30 psi	20 psi to 50 psi
Ion source temperature <sup>9</sup>	Ambient to 200 °C	200 °C to 650 °C	400 °C to 750 °C	Up to 750 °C
Declustering potential (DP) <sup>10</sup>	Positive: 70 V Negative: -70 V	Positive: 70 V Negative: -70 V	Positive: 100 V Negative: -100 V	Positive: 0 V to V Negative: V to 0 V

<sup>9</sup> Optimum temperature values depend on the compound and mobile phase composition. Higher aqueous content requires a higher temperature. Zero (0) means no temperature is applied.

<sup>10</sup> DP values depend on the compound.

Table B-1 Parameter Optimization for the TurbolonSpray Probe (continued)

Parameters	Typical Values			Operational Range
	Low	Medium	High	
Probe horizontal micrometer setting	3 to 8	3 to 8	3 to 8	0 to 10

## APCI Probe Parameters

Table B-2 Parameter Optimization for the APCI Probe

Parameter	Typical Value	Operational Range
LC flow rate	1,000 $\mu\text{L}/\text{min}$	200 $\mu\text{L}/\text{min}$ to 3,000 $\mu\text{L}/\text{min}$
Ion source gas 1 (nebulizer gas)	30 psi	0 psi to 90 psi
Gas for the Curtain Gas interface	30 psi	20 psi to 50 psi
Ion source temperature <sup>11</sup>	400 $^{\circ}\text{C}$	100 $^{\circ}\text{C}$ to 750 $^{\circ}\text{C}$
Nebulizer current	Positive: 3 $\mu\text{A}$ Negative: -3 $\mu\text{A}$	Positive: 0 mA to 5 $\mu\text{A}$ Negative: -5 mA to 0 $\mu\text{A}$
Declustering potential (DP)	Positive: 60 V Negative: -60 V	Positive: 0 V to 300 V Negative: -300 V to 0 V
Probe vertical micrometer setting	5	Scale 0 to 13

## Probe Position

The position of the probe can affect the sensitivity of the analysis. For more information on how to optimize the position of the probe, refer to the section: [Ion Source Optimization](#).

<sup>11</sup> Temperature values depend on the compound.

# Solvent Composition

The standard concentration of ammonium formate or ammonium acetate is from 2 mmol/L to 10 mmol/L for positive ions and 2 mmol/L to 50 mmol/L for negative ions. The concentration of organic acids is 0.1% to 0.5% by volume for the TurbolonSpray probe and 0.1% to 1.0% by volume for the APCI probe.

Commonly used solvents are:

- Acetonitrile
- Methanol
- Propanol
- Water

Commonly used modifiers are:

- Acetic acid
- Formic acid
- Ammonium formate
- Ammonium acetate

The following modifiers are not commonly used because they complicate the spectrum with their ion mixtures and cluster combinations. They might also suppress the strength of the target compound ion signal.

- Triethyl amine (TEA)
- Sodium phosphate
- Trifluoroacetic acid (TFA)
- Sodium dodecyl sulfate

# Calibration Ions and Solutions

# C

**CAUTION: Potential Wrong Result. Do not use expired solutions or solutions that have not been stored at the indicated storage temperature.**

**Table C-1 Tuning Frequency**

Calibration			Resolution Optimization	
Scan Type	Frequency	Manual/Automated	Frequency	Manual/Automated
Q1 and Q3	3 months to 6 months	Both	3 months to 6 months	Both
LIT	Every 2 weeks, as required	Both	3 months to 6 months	Automated only

**Table C-2 Suggested Tuning Solutions for the SCIEX Citrine Systems**

System	Q1 and Q3		LIT
	Positive	Negative	Positive and Negative
Citrine Triple Quad system	POS PPG, 2e-7 M	NEG PPG, 3e-5 M	N/A
Citrine QTRAP MS/MS system	POS PPG, 2e-7 M	NEG PPG, 3e-5 M	ES Tuning Solution (1:100 dilution)

**Table C-3 Q1 and Q3 Scans for SCIEX Citrine Systems**

Polarity	Masses
<b>Low Mass</b>	

## Calibration Ions and Solutions

---

**Table C-3 Q1 and Q3 Scans for SCIEX Citrine Systems (continued)**

<b>Polarity</b>	<b>Masses</b>							
Positive	59.05	175.133	500.38	616.464	906.673	1080.799	1196.883	N/A
Negative	44.998	585.385	933.636	1223.845	N/A	N/A	N/A	N/A
<b>High Mass</b>								
Positive	59.05	175.133	500.38	616.464	906.673	1254.925	1545.134	1952.427
Negative	44.998	411.259	585.385	933.636	1223.845	1572.097	1863.306	1979.389

**Table C-4 LIT Scans for the SCIEX Citrine QTRAP Systems**

<b>Polarity</b>	<b>Masses</b>				
<b>Low Mass</b>					
Positive	118.087	322.049	622.029	922.01	N/A
Negative	112.985	431.982	601.978	N/A	N/A
<b>High Mass</b>					
Positive	118.087	322.049	622.029	922.01	1521.972
Negative	112.987	431.982	601.978	1033.988	1633.949

# Toolbar Icons

# D

For additional toolbar icons, refer to the document: *Advanced User Guide*.

**Table D-1 Tool Bar Icons**

Icon	Name	Description
	<b>New Subproject</b>	Creates a subproject. Subprojects can only be created later in the process if the project was originally created with subprojects.
	<b>Copy Subproject</b>	Copies a subproject folder.  Subprojects can be copied only from another project that has existing subprojects. If the same folders exist at both the project and subproject levels, then the software uses the project level folders.

**Table D-2 Acquisition Method Editor Icons**

Icon	Name	Description
	<b>Mass Spec</b>	Shows the MS tab in the Acquisition Method editor.
	<b>Period</b>	Adds an experiment, an <b>IDA Criteria Level</b> , or deletes the period.
	<b>Autosampler</b>	Opens the Autosampler Properties tab.
	<b>Syringe Pump</b>	Opens the Syringe Pump Properties tab.
	<b>Column Oven</b>	Opens the Column Oven Properties tab.
	<b>Valve</b>	Opens the Valve Properties tab.
	<b>DAD</b>	Opens the DAD Method Editor. Refer to the section: <a href="#">Show DAD Data</a> .
	<b>ADC</b>	Opens the ADC Properties tab. Refer to the section: <a href="#">Show ADC Data</a> .

## Toolbar Icons

Table D-3 Acquire Mode Icons

Icon	Name	Description
	<b>View Queue</b>	Shows the sample queue.
	<b>Instrument Queue</b>	Shows a remote instrument station.
	<b>Status for Remote Instrument</b>	Shows the status of a remote instrument.
	<b>Start Sample</b>	Starts the sample in the queue.
	<b>Stop Sample</b>	Stops the sample in the queue.
	<b>Abort Sample</b>	Aborts a sample acquisition in the middle of the processing of that sample.
	<b>Stop Queue</b>	Stops the queue before it has completed processing all of the samples.
	<b>Equilibrate</b>	Selects the method to be used to equilibrate the devices. This method should be the same as the method that was used with the first sample in the queue.
	<b>Standby</b>	Puts the instrument in <b>Standby</b> state.
	<b>Ready</b>	Puts the instrument in <b>Ready</b> state.
	<b>Reserve Instrument for Tuning</b>	Reserves the mass spectrometer for tuning and calibrating.
	<b>IDA Method Wizard</b>	Starts the <b>IDA Method Wizard</b> .
Icon	Name	Description
	<b>Calibrate from spectrum</b>	Opens the Mass Calibration Option dialog and uses the active spectrum to calibrate the mass spectrometer.
	<b>Manual Tune</b>	Opens the Manual Tune Editor.

Icon	Name	Description
	<b>Compound Optimization</b>	Optimizes for a compound using infusion by FIA.
	<b>Instrument Optimization</b>	Verifies the instrument performance, adjusts the mass calibration, or adjusts mass spectrometer settings.
	<b>View Queue</b>	Shows the sample queue.
	<b>Instrument Queue</b>	Shows a remote instrument.
	<b>Status for Remote Instrument</b>	Shows the status of a remote instrument.
	<b>Reserve Instrument for Tuning</b>	Reserves the instrument for tuning and calibrating.
	<b>IDA Method Wizard</b>	Starts the IDA Method Wizard.

Table D-4 Explore Quick Reference: Chromatograms and Spectrum

Icon	Name	Description
	<b>Open Data File</b>	Opens files.
	<b>Show Next Sample</b>	Goes to the next sample.
	<b>Show Previous Sample</b>	Goes to the previous sample.
	<b>Go To Sample</b>	Opens the Select Sample dialog.
	<b>List Data</b>	Shows the data in tables.
	<b>Show TIC</b>	Generates a TIC from a spectrum.
	<b>Extract Using Dialog</b>	Extracts ions by selecting masses.
	<b>Show Base Peak Chromatogram</b>	Generates a BPC.

## Toolbar Icons

**Table D-4 Explore Quick Reference: Chromatograms and Spectrum (continued)**

Icon	Name	Description
	<b>Show Spectrum</b>	Generates a spectrum from a TIC.
	<b>Copy Graph to new Window</b>	Copies the active graph to a new window.
	<b>Baseline Subtract</b>	Opens the Baseline Subtract dialog.
	<b>Threshold</b>	Adjusts the threshold.
	<b>Noise Filter</b>	Shows the Noise Filter Options dialog, which can be used define the minimum width of a peak. Signals below this minimum width are regarded as noise.
	<b>Show ADC</b>	Shows ADC data.
	<b>Show Auxiliary Traces</b>	Opens the Select Auxiliary Trace Channel dialog
	<b>Show File Info</b>	Shows the experimental conditions used to collect the data.
	<b>Add arrows</b>	Adds arrows to the X-axis of the active graph.
	<b>Remove all arrows</b>	Removes arrows from the X-axis of the active graph.
	<b>Offset Graph</b>	Compensates for slight differences in the time during which the ADC data and the mass spectrometer data were recorded. This is useful when overlaying graphs for comparison.
	<b>Force Peak Labels</b>	Labels all of the peaks.
	<b>Expand Selection By</b>	Sets the expansion factor for a portion of a graph to be viewed in greater detail.
	<b>Clear ranges</b>	Returns the expanded selection to normal view.
	<b>Set Selection</b>	Defines start and stop points for a selection. This feature provides more accurate selection than is possible by selecting the region using the cursor.

**Table D-4 Explore Quick Reference: Chromatograms and Spectrum (continued)**

Icon	Name	Description
	<b>Normalize To Max</b>	Scales a graph to maximum size, so that the most intense peak is scaled to full scale, whether or not it is visible.
	<b>Show History</b>	Shows a summary of data processing operations performed on a particular file, such as smoothing, subtraction, calibration, and noise filtering.
	<b>Open Compound Database</b>	Opens the compound database.
	<b>Set Threshold</b>	Adjusts the threshold.
	<b>Show Contour Plot</b>	Shows selected data as either a spectrum graph or an XIC. Additionally, for data acquired by a DAD, a contour plot can show selected data as either a DAD spectrum or an XWC.
	<b>Show DAD TWC</b>	Generates a TWC of the DAD spectrum.
	<b>Show DAD Spectrum</b>	Generates a DAD spectrum.
	<b>Extract Wavelength</b>	Extracts up to three wavelength ranges from a DAD spectrum to view the XWC.

**Table D-5 Explore Toolbar Quick Reference: Overlaying Graphs**

Icon	Name	Description
	<b>Home Graph</b>	Return the graph to the original scale.
	<b>Overlay</b>	Overlays graphs.
	<b>Cycle Overlays</b>	Cycles between overlaid graphs.
	<b>Sum Overlays</b>	Adds the graphs together.

## Toolbar Icons

**Table D-6 Explore Toolbar Quick Reference: Fragment Interpretation Tool**

Icon	Name	Description
	<b>Show Fragment Interpretation Tool</b>	Opens the Fragment Interpretation tool, which calculates the single, non-cyclic bond cleavage fragments from a mol file.

**Table D-7 Navigation Icons on the Explore Toolbar**

Icon	Name	Function
	<b>Open File</b>	Opens files.
	<b>Show Next Sample</b>	Navigates to the next sample.
	<b>Show Previous Sample</b>	Navigates to the previous sample.
	<b>GoTo Sample</b>	Opens the Select Sample dialog.
	<b>List Data</b>	Shows the data in tables.
	<b>Show TIC</b>	Generates a TIC from a spectrum.
	<b>Extract Using Dialog</b>	Click to extract ions by selecting masses.
	<b>Show Base Peak Chromatogram</b>	Generates a BPC.
	<b>Show Spectrum</b>	Generates a spectrum from a TIC.
	<b>Copy Graph to new Window</b>	Copies the active graph to a new window.
	<b>Baseline Subtract</b>	Opens the Baseline Subtract dialog.
	<b>Threshold</b>	Adjusts the threshold.
	<b>Noise Filter</b>	Opens the Noise Filter Options dialog that defines the minimum width of a peak. Signals below this minimum width are regarded as noise.
	<b>Show ADC</b>	Shows ADC data.

Table D-7 Navigation Icons on the Explore Toolbar (continued)

Icon	Name	Function
	<b>Show Auxiliary Traces</b>	Opens the Select Auxiliary Trace Channel dialog
	<b>Show File Info</b>	Shows the experimental conditions used to collect the data.
	<b>Add arrows</b>	Adds arrows to the X-axis of the active graph.
	<b>Remove all arrows</b>	Removes arrows from the X-axis of the active graph.
	<b>Offset Graph</b>	Compensates for slight differences in the time during which the ADC data and the mass spectrometer data were recorded. This is useful when overlaying graphs for comparison.
	<b>Force Peak Labels</b>	Labels all the peaks.
	<b>Expand Selection By</b>	Sets the expansion factor for a portion of a graph to view in greater detail.
	<b>Clear ranges</b>	Returns the expanded selection to normal view.
	<b>Set Selection</b>	Sets start and stop points for a selection. This provides more accurate selection than is possible by highlighting the region using the cursor.
	<b>Normalize to Max</b>	Scales a graph to maximum, so that the most intense peak is scaled to full scale, whether or not it is visible.
	<b>Show History</b>	Shows a summary of data processing operations performed on a particular file, such as smoothing, subtraction, calibration, and noise filtering.
	<b>Open Compound Database</b>	Opens the compound database.
	<b>Set Threshold</b>	Adjusts the threshold.

## Toolbar Icons

**Table D-7 Navigation Icons on the Explore Toolbar (continued)**

Icon	Name	Function
	<b>Show Contour Plot</b>	Shows selected data as either a spectrum graph or an XIC. Additionally, for data acquired by a DAD, a contour plot can display selected data as either a DAD spectrum or an XWC.
	<b>Show DAD TWC</b>	Generates a TWC of the DAD.
	<b>Show DAD Spectrum</b>	Generates a DAD spectrum.
	<b>Extract Wavelength</b>	Extracts up to three wavelength ranges from a DAD spectrum to view the XWC.

**Table D-8 Integration Tab and Quantitation Wizard Icons**

Icon	Name	Description
	<b>Set parameters from Background Region</b>	Uses the selected peak.
	<b>Select Peak</b>	Uses the selected background.
	<b>Manual Integration Mode</b>	Manually integrates peaks.
	<b>Show or Hide Parameters</b>	Shows or hides the peak-finding parameters.
	<b>Show Active Graph</b>	Shows the analyte chromatogram only.
	<b>Show Both Analyte and IS</b>	Shows the analyte and its associated chromatogram. Available only when an associated internal standard exists.
	<b>Use Default View for Graph</b>	Returns to the preset, view all data, view if, for example, the user has zoomed in on a chromatogram.

Table D-9 Results Table Icons

Icon	Name	Description
	<b>Sort Ascending by Selection</b>	Sorts the selected column in ascending order.
	<b>Sort Descending by Selection</b>	Sorts the selected column in descending order.
	<b>Lock Or Unlock Column</b>	Locks or unlocks the selected column. A locked column cannot be moved.
	<b>Metric Plot By Selection</b>	Creates a metric plot from the selected column.
	<b>Show all Samples</b>	Shows all of the samples in the Results Table.
	<b>Delete Formula Column</b>	Deletes formula columns.
	<b>Report Generator</b>	Opens the Reporter software.

Table D-10 Icon Quick Reference: Quantitate Mode

Icon	Name	Description
	<b>Add/Remove Samples</b>	Adds or removes samples from the Results Table.
	<b>Export as Text</b>	Saves the Results Table as a text file.
	<b>Modify Method</b>	Opens a wiff file.
	<b>Peak Review - Pane</b>	Opens peaks in a pane.
	<b>Peak Review - Window</b>	Opens peaks in a window.
	<b>Calibration - Pane</b>	Opens the calibration curve in a pane.
	<b>Calibration - Window</b>	Opens the calibration curve in a window.
	<b>Show First Peak</b>	Shows the first peak in the pane or window.

## Toolbar Icons

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**Table D-10 Icon Quick Reference: Quantitate Mode (continued)**

Icon	Name	Description
	<b>Show Last Peak</b>	Shows the last peak in the pane or window.
	<b>Show Audit Trail</b>	Shows the audit trail for the Results Table.
	<b>Clear Audit Trail</b>	Clears the audit trail for the Results Table. This functionality is not available.
	<b>Statistics</b>	Opens the Statistics window.
	<b>Report Generator</b>	Opens the Reporter software.

**Table D-11 Acquire Mode Icons**

Icon	Name	Function
	<b>Start Sample</b>	Click to start the sample in the queue.
	<b>Stop Sample</b>	Click to stop the sample in the queue.
	<b>Equilibrate</b>	Click to select a method to use to equilibrate the mass spectrometer, which includes the ion source, LC column if used, and any peripheral devices. This method should be the same as the one used with the first sample in the queue.

## Electrospray Ionization Mode

The probe is located centrally between the two turbo heaters, which are located at a 45-degree angle on either side of the probe. The combination of the spray and the heated dry gas from the turbo heaters is projected at a 90-degree angle to the aperture in the curtain plate.

Only compounds that ionize in the liquid solvent can be generated as gas phase ions in the source. The efficiency and rate of ion generation depends on the solvation energies of the specific ions. Ions with lower solvation energies are more likely to evaporate than ions with higher solvation energies.

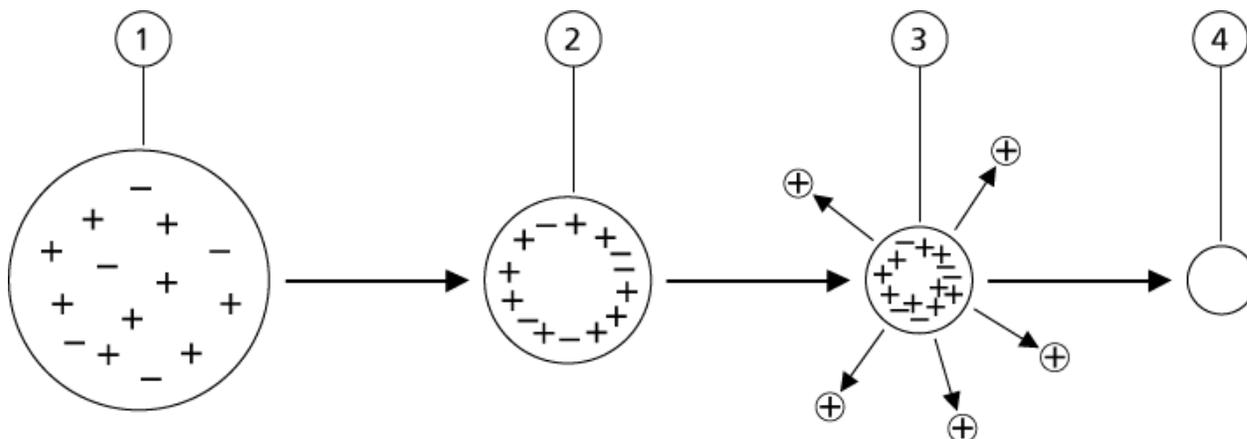
The interaction of the **IonSpray Voltage** and the turbo heaters helps focus the stream and increases the rate of droplet evaporation, resulting in an increased ion signal. The heated gas increases the efficiency of ion evaporation, resulting in increased sensitivity and the ability to handle higher liquid sample flow rates.

A high-velocity flow of nebulizer gas shears droplets from the liquid sample stream in the **IonSpray Voltage** inlet. Using the variable high voltage applied to the sprayer, the ion source applies a net charge to each droplet. This charge aids in the droplet dispersion. Ions of a single polarity are preferentially drawn in the droplets by the high voltage as they are separated from the liquid stream. However, this separation is incomplete and each droplet contains many ions of both polarities. Ions of one polarity are predominant in each droplet, and the difference between the number of positively or negatively charged ions results in the net charge. Only the excess ions of the predominant polarity are available for ion evaporation, and only a fraction of these actually evaporate.

The probe can generate multiply-charged ions from compounds that have multiple charge sites, such as peptides and oligonucleotides. This is useful during analysis of high-molecular-weight species where the multiple charges produce ions of a mass-to-charge ratio ( $m/z$ ) within the mass range of the mass spectrometer. This allows routine molecular-weight determinations of compounds in the kiloDalton (kDa) range.

Each charged droplet contains solvent and both positive and negative ions, but with ions of one predominant polarity. Refer to the figure: [Figure E-1](#). As a conducting medium, excess charges reside at the surface of the droplet. As the solvent evaporates, the electrical field at the surface of the droplet increases due to the decreasing radius of the droplet.

Figure E-1 Ion Evaporation



Item	Description
1	Droplet contains ions of both polarities with one polarity being predominant.
2	As the solvent evaporates, the electrical field increases and the ions move to the surface.
3	At some critical field value, ions are emitted from the droplets.
4	Nonvolatile residue remains as a dry particle.

If the droplet contains excess ions and enough solvent evaporates from the droplet, a critical field is reached at which ions are emitted from the surface. Eventually, all of the solvent will evaporate from the droplet, leaving a dry particle consisting of the nonvolatile components of the sample solution.

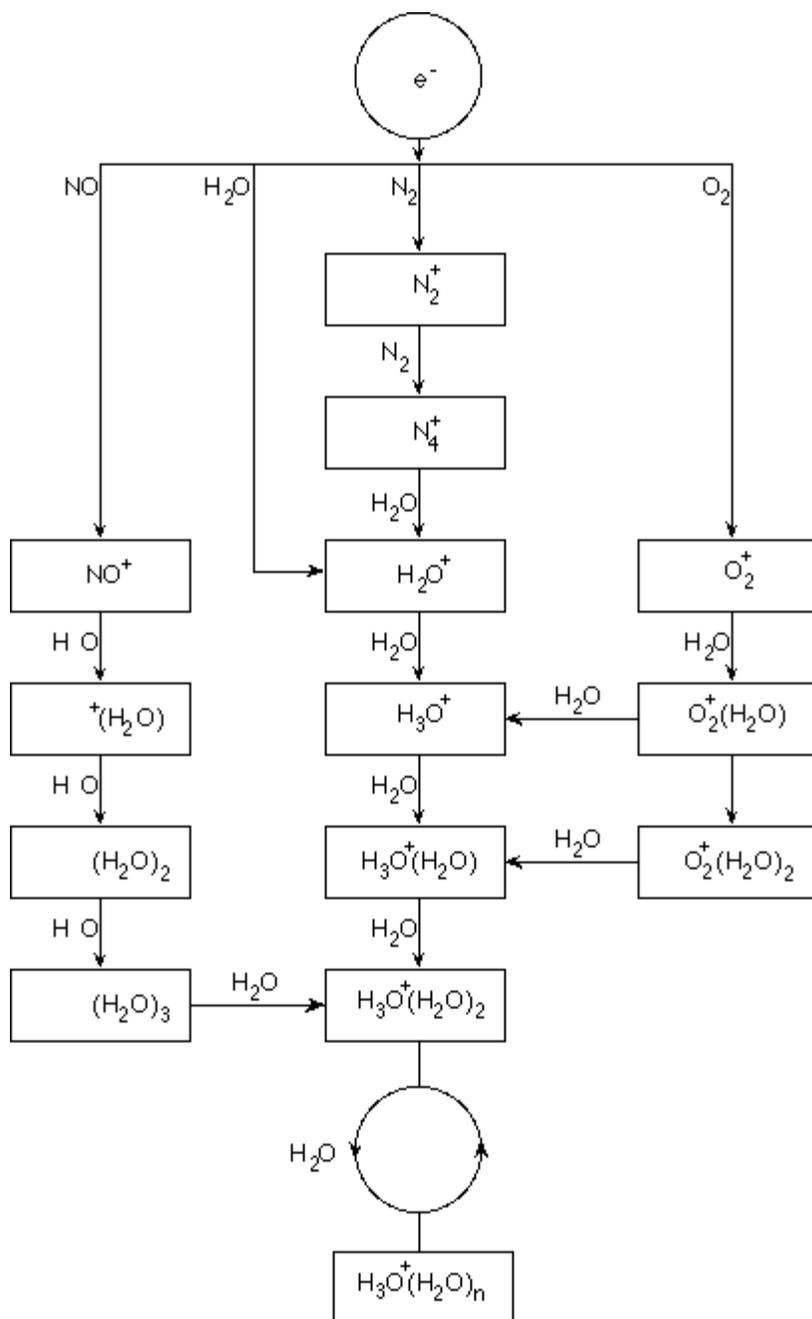
Because the solvation energies for most organic molecules are unknown, the sensitivities of any given organic ion to ion evaporation are difficult to predict. The importance of solvation energy is evident because surfactants that concentrate at the surface of a liquid can be detected very sensitively.

## APCI Mode

The basis for past incompatibilities in linking liquid chromatography with mass spectrometry arose from difficulties converting relatively nonvolatile molecules in solution in a liquid into a molecular gas without inducing excessive decomposition. The APCI probe process of gently nebulizing the sample to finely dispersed small droplets in a heated ceramic tube results in the rapid vaporization of the sample so that the sample molecules are not decomposed.

The following figure shows the reaction flow of the APCI process for reactant positive ions, the proton hydrates,  $H_3O^+[H_2O]_n$ .

Figure E-2 APCI Reaction Flow Diagram



The major primary ions  $N_2^+$ ,  $O_2^+$ ,  $H_2O^+$ , and  $NO^+$  are formed by the electron impact of corona-created electrons on the major neutral components of air. Although  $NO^+$  is normally not a major constituent of clean air, the concentration of this species in the ion source is enhanced due to neutral reactions initiated by the corona discharge.

## Theory of Operation — Ion Source

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Samples that are introduced through the APCI probe are sprayed, with the aid of a nebulizer gas, in the heated ceramic tube. Within the tube, the finely dispersed droplets of sample and solvent undergo a rapid vaporization with minimal thermal decomposition. The gentle vaporization preserves the molecular identity of the sample.

The gaseous sample and solvent molecules pass to the ion source housing where the ionization by APCI is induced by a corona discharge needle connected to the end of the ceramic tube. The sample molecules are ionized by colliding with the reagent ions that are created by the ionization of mobile phase solvent molecules. The vaporized solvent molecules ionize to produce the reagent ions  $[X+H]^+$  in Positive polarity and  $[X-H]^-$  in Negative polarity. Refer to the figure: [Figure E-3](#). It is these reagent ions that produce stable sample ions when they collide with the sample molecules.

**Figure E-3 Atmospheric Pressure Chemical Ionization**

Item	Description
1	Sample
2	Primary ions are created in the vicinity of the corona discharge needle
3	Ionization produces predominantly solvent ions
4	Reagent ions react with sample molecules, forming clusters
5	Curtain plate
6	Interface

x = solvent molecules; M=sample molecules

The sample molecules are ionized through a process of proton transfer in Positive polarity and by either electron transfer or proton transfer in Negative polarity. The energy for the APCI ionization process is collision-dominated because of the relatively high atmospheric pressure of the ion source.

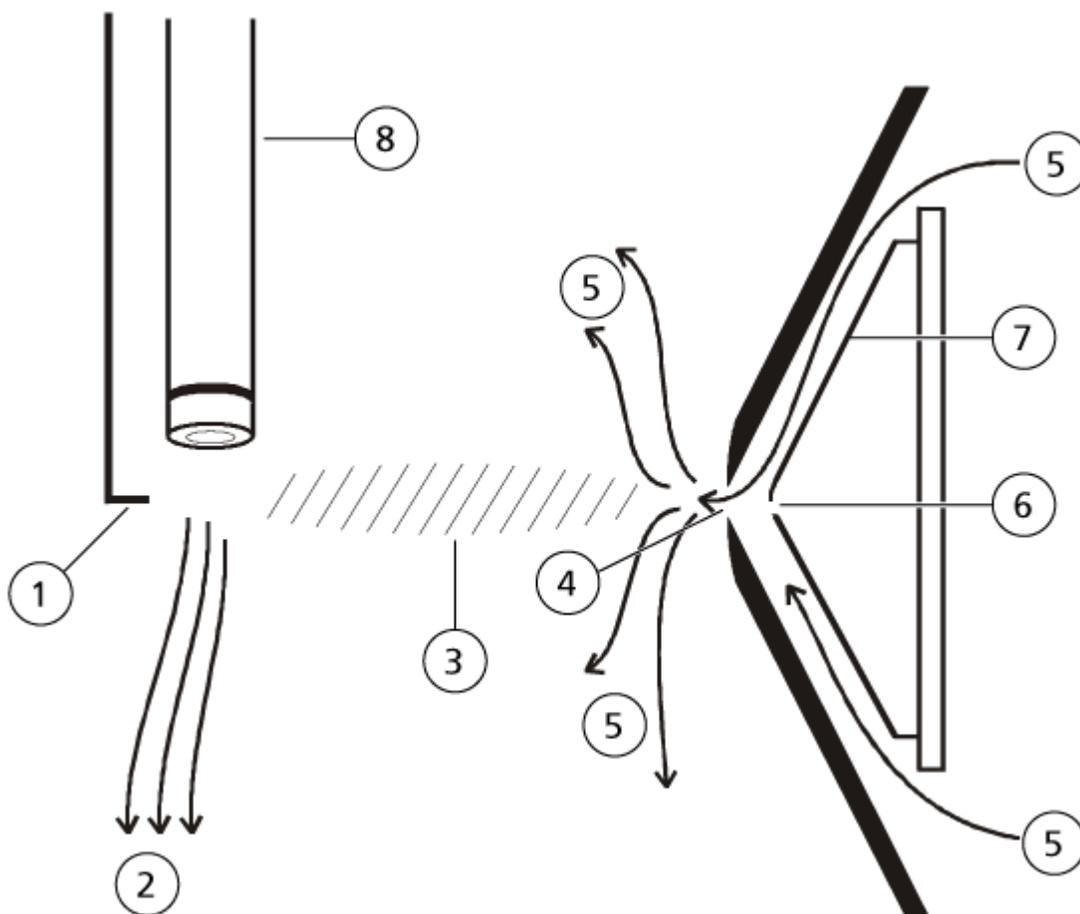
For reverse phase applications, the reagent ions consist of protonated solvent molecules in Positive polarity and solvated oxygen ions in Negative polarity. With favorable thermodynamics, the addition of modifiers changes the reagent ion composition. For example, the addition of acetate buffers or modifiers can make the acetate ion  $[CH_3COO]^-$  the primary reagent in Negative polarity. Ammonium modifiers might make protonated ammonia  $[NH_4]^+$  the primary reagent in Positive polarity.

Through collisions, an equilibrium distribution of certain ions, such as protonated water cluster ions, is maintained. The likelihood of premature fragmentation of the sample ions in the ion source is reduced because of the moderating influence of solvent clusters on the reagent ions and the relatively high gas pressure in the ion source. As a result, the ionization process yields primarily molecular product ions for mass analysis in the mass spectrometer.

## APCI Ionization Region

The following figure shows the general location of the ion-molecule reactor of the APCI probe. The slanted lines indicate a wall-less reactor. A self-starting corona discharge ion current in the microampere range is created as a result of the electric field between the discharge needle and the curtain plate. Primary ions, for example,  $N_2^+$  and  $O_2^+$ , are created by the loss of electrons that originate in the plasma in the immediate vicinity of the discharge needle tip. The energy of these electrons is moderated by a number of collisions with gas molecules before attaining an energy where their effective ionization cross-section allows them to ionize neutral molecules efficiently.

Figure E-4 APCI Ionization Region



Item	Description
1	Discharge needle tip

## Theory of Operation — Ion Source

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Item	Description
2	Sample flow
3	Wall-less reactor
4	Curtain plate aperture
5	Gas for the Curtain Gas interface
6	Orifice
7	Orifice plate
8	Ceramic tube

The primary ions, in turn, generate intermediate ions that lead to the formation of sample ions. Ions of the chosen polarity drift under the influence of the electric field in the direction of the curtain plate and through the gas curtain to the mass analyzer. The whole ion formation process is collision-dominated because of the relatively high atmospheric pressure of the APCI probe. Except in the immediate vicinity of the discharge needle tip, where the strength of the electric field is greatest, the energy imparted to an ion by the electric field is small in comparison with the thermal energy of the ion.

Through collisions, an equal distribution of certain ions (for example, protonated water cluster ions) is maintained. Any excess energy that an ion might acquire in the ion-molecule reaction process is thermalized. Through collisional stabilization, many of the product ions are fixed, even though many subsequent collisions occur. The formation of both product ions and reactant ions is governed by equilibrium conditions at 760 torr (atmospheric) operating pressure.

The APCI probe functions as a wall-less reactor because the ions that pass from the ion source to the vacuum chamber and eventually to the detector never experience collisions with a wall—only collisions with other molecules. Ions are also formed outside the designated ion source, but are not detected and are eventually neutralized by interacting with a wall surface.

The temperature of the probe is an important factor for APCI probe operation. To preserve the molecular identity, the temperature must be set high enough to ensure a rapid evaporation. At a sufficiently high operating temperature, the droplets are vaporized quickly so that organic molecules are desorbed from the droplets with minimal thermal degradation. If, however, the temperature is set too low, the evaporation process is slower and pyrolysis, or decomposition, might occur before vaporization is complete. Operating the APCI probe at temperatures above the optimal temperature might cause thermal decomposition of the sample.

# Manual Compound Optimization

# F

The user must control the autosampler and injection valve manually, because these devices cannot be controlled through the system while it is in Tune and Calibrate mode.

## Prerequisites

- The mass spectrometer is tuned and calibrated.
- Conditions for an LC separation are known.
- All of the required peripheral devices, including the syringe pump, if required, and LC components are in the hardware profile.

## Required Materials

To tune instrument parameters for particular compounds the following solutions are recommended. The mixture of four compounds is used for illustrating the steps of the procedure. The user must apply the same procedure while using the appropriate compounds for the assay of interest.

- Mobile phase: 1:1 acetonitrile:water + 2 mM ammonium acetate + 0.1% formic acid.
- LC pump and autosampler.
- Autosampler vials.
- Four-compound mix (50 ng/mL), consisting of reserpine, minoxidil, tolbutamide, and rescinnamine. The solution can be used for infusion and flow injection analysis (FIA). The concentration is system-dependent. Use a solution that is 49.9% acetonitrile, with 50% deionized water and 0.1% formic acid as diluent. Other compounds can be substituted as long as their molecular weight is known and the compound is reasonable to be ionized by the ion source.

**Table F-1 Compounds and *m/z* Values**

Compound	<i>m/z</i> Value
Minoxidil	210.2
Tolbutamide	271.1
Reserpine	609.3
Rescinnamine	635.3

# About Manual Compound Optimization

Manual compound optimization is used to optimize compound- and ion source-dependent parameters for an analyte. When the user manually optimizes for an analyte, an MS acquisition method is created in Tune and Calibrate mode. Depending on the method of sample introduction selected, add an LC method to the acquisition method so that infusion or LC can be used.

Optimizing to give the highest signal does not always give the highest signal-to-noise ratios. Noise can scale with signal for some parameters and should be verified during optimization if the goal is to maximize the signal-to-noise ratio.

When optimizing ion source-dependent parameters, introduce the sample at flow rates that will be used during sample analysis, using either flow injection analysis (FIA) or tee-infusion as the method of sample introduction. The CAD gas parameter is the only compound-dependent parameter that is shown on the Source/Gas tab and can be easily optimized while infusing the analyte.

Optimize the position of the ion source before optimizing the ion source-dependent parameters. Refer to the section: [Ion Source Optimization](#).

## About Scan Types

For this example, use the Q1 MS, Q1 MI, Product Ion, and MRM scan types. The Q1 MS scan type is used to confirm the presence of compounds of interest. The Q1 MI scan is used to optimize MS or pre-collision cell voltages. The Product Ion scan type is used to determine the product ions of each compound. The MRM scan type is used to optimize the collision energy (CE) and collision cell exit potential (CXP) for each product ion or fragment. Use the methods created in this section for quantitative or qualitative analysis.

## Manually Optimize an Analyte

After the acquisition method is created, optimize compound-dependent parameters using the **Edit Ramp** function or by manually editing the parameters in the Tune Method Editor. Ion source-dependent parameters can be optimized only by manually adjusting the parameters in the Tune Method Editor. Depending on the scan type used, different parameters are available to optimize.

Follow the procedures in the order given:

1. [Confirm the Presence of Compounds](#)
2. [Optimize MS-Specific Parameters](#)

3. [Determine the Product Ions for Optimization](#)
4. [Optimize Collision Cell Exit Potential for each Product Ion](#)

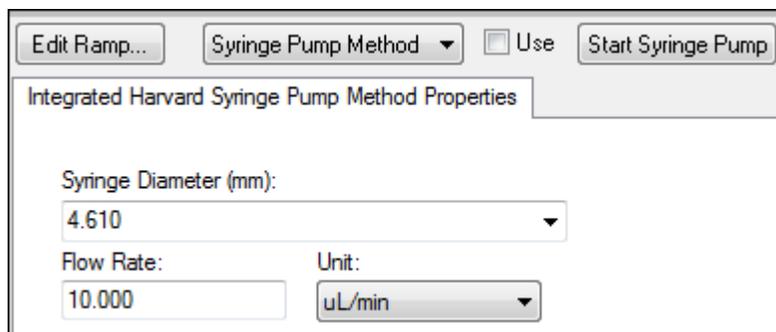
## Confirm the Presence of Compounds

1. Create a project.
2. Activate the hardware profile.
3. Prepare the sample:
  - a. Aspirate the compound solution into a syringe and remove the air from the syringe.
  - b. Use the tubing with the special fitting to connect the syringe to the mass spectrometer.
  - c. Install the syringe in the integrated syringe pump.
4. Infuse the compound in solution at a rate of 5  $\mu\text{L}/\text{min}$  to 10  $\mu\text{L}/\text{min}$ .
5. On the Navigation bar, under **Tune and Calibrate**, double-click **Manual Tuning**.
6. In the method list field, click **Syringe Pump Method**.
7. On the Syringe Pump Method Properties tab, type the parameter values shown in the following table.

**Table F-2 Syringe Pump Method Properties Tab**

Parameter	Value
Syringe Diameter	Syringe dependent. 4.610 mm if a 1.0 mL syringe is used
Flow Rate	10
Unit	$\mu\text{L}/\text{min}$

**Figure F-1 Syringe Pump Method Properties Tab**



8. Click **Start Syringe Pump**.

## Manual Compound Optimization

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9. Select **MS Method** from the method list.
10. On the MS tab, type the parameter values shown in the following table.

**Table F-3 MS Tab**

Parameter	Value
Scan type	Q1 MS (Q1)
Start (Da)	200
Stop (Da)	700
Scan rate (Da/s)	200
Duration (min)	3

11. Click **Start**.
12. Wait until an even TIC is shown on the left and peaks are shown on the right and then click **Stop**.
13. Select the **MCA** check box.
14. Type 10 in the **Cycles** field.
15. Click **Start**.  
When ten scans are complete, the masses of the four compounds are shown as ions.

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**Tip!** If the previous or expected ion chromatogram pane is hidden, then select the window to be shown from the **Window** menu.

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**Note:** Ion intensities of the compounds can show large variances. To facilitate moving to a solution of higher or lower concentration as needed during the optimization, have several concentration levels prepared before beginning optimization.

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16. Right-click the bottom-right spectral pane and then click **Open File**.
17. Find the compounds of interest and then write down the  $m/z$  values for the highest peaks. These values should be within 0.1 Da to 0.2 Da of the expected  $m/z$  value. In the next procedure, use the  $m/z$  values.

## Optimize MS-Specific Parameters

The DP is the difference between the orifice and the ground. The higher the potential difference, the greater the amount of declustering.

The DP parameter has a significant effect on analyte signal. Typical DP values range from 20 V to 150 V. A DP value that is too low can result in lower ion intensity and potential interferences

from clusters. A DP value that is too high can cause fragmentation of the analyte in the source. Generally, the DP should be set to the value that provides the highest intensity.

The EP parameter controls the entrance potential, which guides and focuses the ions through the high-pressure Q0 region. It is typically set at 10 V for positive ions or –10 V for negative ions. The EP has a minor effect on compound optimization and can generally be left at default values without impact on analyte detection limits.

1. Return to the Tune Method Editor and change the method to the **Q1 Multiple Ions (Q1 MI)** scan type.
2. In the mass table, type the appropriate parameter values. Refer to the following table.

**Table F-4 Mass Table Parameters—Q1 Multiple Ions (Q1 MI)**

Compound	Q1 Mass	Time
Reserpine	609.3	1
Minoxidil	210.2	1
Tolbutamide	271.1	1
Rescinnamine	635.3	1

Start with reserpine for a simple case. Repeat the manual optimization process for the remaining compounds.

3. Click **Edit Ramp**.
4. Select **Declustering Potential (DP)** in the Ramp Parameter Settings dialog.

**Note:** Start with the DP parameter and then optimize the other parameters in the order in which they are shown in the dialog. The parameters might not be optimized correctly if they are optimized out of order.

5. Type the required **Start**, **Stop**, and **Step** values.

**Tip!** The existing values are good starting points. Use the **Edit Ramp** function to change these values to be more efficient.

6. Click **OK**.
7. Click **Start**.
8. Right-click the bottom-right XIC pane and then click **Open File** to maximize the XIC view.
9. Monitor the XICs.

**Note:** The value that gives the best signal per second for the ion of interest is the optimal value.

## Manual Compound Optimization

---

10. Note the optimal value for the ion of interest.
11. Move the cursor to the mass table, right-click, and then add the parameter just optimized. A column is added to the table.
12. Add the optimized value to the appropriate row.
13. Repeat these steps for each mass in the acquisition method until there is a list of optimal values for all of the masses.
14. Repeat these steps to optimize for the other MS-specific parameters.

**Table F-5 MS-Specific Parameters**

Parameter	Comment
<b>DP</b>	Set the DP to the value that provides the highest intensity.
<b>EP</b>	Rarely optimize this parameter because it has a smaller effect.
<b>CEP</b>	Select the CEP value with the largest intensity.

## Determine the Product Ions for Optimization

The collision energy (CE) controls the amount of energy that the precursor ions receive as they are accelerated into the collision cell.

Perform this procedure, one compound at a time, using the MS-specific optimized values that were obtained previously. The product ions provide the Q3 mass of MRM transitions.

In this example, the compound reserpine is used.

1. In the Tune Method Editor, close the XIC panes.
2. Click **Product Ion (MS2)** in the **Scan type** field.
3. On the Compound tab, type the optimal value noted in section: [Optimize MS-Specific Parameters](#).
4. On the MS tab, in the **Product Of** field, type 609.4.  
This value is the mass assignment for reserpine that was noted in the section: [Confirm the Presence of Compounds](#).
5. Make sure that the **Center / Width** check box is not selected.
6. In the mass table, type the appropriate **Start**, **Stop**, and **Time** values. Refer to the table: [Table F-6](#).

Table F-6 Mass Table Parameters (Product Ion Scan)

Field	Value
Start (Da)	100
Stop (Da)	650
Time (sec)	2

7. Click **Edit Ramp**.  
The Ramp Parameter Settings dialog opens.
8. Select **Collision Energy** and then type the required **Start**, **Stop**, and **Step** values.

---

**Note:** The existing values are good starting points. Use the **Edit Ramp** function to change these values to be more efficient.

---

9. Click **OK**.
10. Select the **MCA** check box.
11. Click **Start**.
12. Right-click the bottom-right XIC pane and then click **Open File**.
13. Select the product ions with the greatest intensity and note the product ion  $m/z$  value to the first decimal point, such as 195.1.  
We recommend that two or three product ions be optimized for each compound. The additional transitions can be used for confirmation, or to avoid the need to re-optimize a compound in case an interference is found.

---

**Note:** Make sure that the highest peaks selected for optimization do not represent a common loss from the precursor ion, such as water or carbon dioxide. Also make sure that the product ion is not too low in mass or interferences might occur for that transition in real samples or clusters from the mobile phase when analyzing on column.

---

14. Repeat this procedure for the remaining compounds.

## Optimize Collision Cell Exit Potential for each Product Ion

1. In the Tune Method Editor, close the XIC panes.
2. Open the previously saved method.
3. In the mass table, verify the Q1 and Q3  $m/z$  values for the compound.
4. Click the **Compound** tab and then type the optimal DP and CE values noted in section: [Optimize MS-Specific Parameters](#).
5. Click **Edit Ramp**.

## Manual Compound Optimization

---

6. Select **Collision Cell Exit Potential (CXP)** in the Ramp Parameter Settings dialog.
7. Type the required **Start**, **Stop**, and **Step** values.

---

**Tip!** The existing values are good starting points. Use the **Edit Ramp** function to change these values to be more efficient.

---

8. Click **OK**.
9. Click **Start**.
10. Right-click the bottom-right XIC pane and then click **Open File**.
11. Note the optimal value for the ion of interest.  
The value that gives the best signal is the optimal value.
12. In the mass table, right-click and then select the parameter just optimized.  
This adds a column to the table.
13. Repeat if other product ions were monitored.
14. Add the optimized values to the appropriate row.
15. Save the method.
16. Repeat this procedure for any other compounds that were previously optimized.

## Manually Optimize the Ion Source and Gas Parameters

The ion source and gas settings must be set correctly to prevent contamination of the mass spectrometer and to make sure that the compounds of interest are optimally put into the gas phase as ions.

---

**Note:** The optimal ion source and gas settings are related to the solvent composition and the flow rate.

---

The ion source and gas settings must be adjusted when the LC conditions change significantly.

To optimize the ion source and gas parameters, set up a syringe pump with the compounds of interest and then connect the sample tubing with a tee to the LC device. Controlling the LC pump can be done manually or through the software.

Another way to manually optimize ion source and gas settings is to use the autosampler to manually inject the compound of interest while manually varying the parameters in manual tuning to find the optimal settings.

---

## Prepare the Ion Source

1. Set the horizontal micrometer to 5.
2. Set the vertical micrometer on the ion source for the flow rate.  
Use the parameters in the following table.

**Table F-7 Turbo V Ion Source Vertical Parameters**

Flow Rate	Initial Vertical Parameters
1 $\mu$ L/min to 20 $\mu$ L/min	10
20 $\mu$ L/min to 250 $\mu$ L/min	5
250 $\mu$ L/min to 500 $\mu$ L/min	2
500 + $\mu$ L/min	0

Depending on how the optimization is run, a hardware profile with the LC pumps might need to be configured.

Refer to the section: [Ion Source Optimization](#).

## Optimize the Ion Source Parameters

Ion source parameters are optimized for best signal-to-noise for the compound of interest. The flow rate for the gas for the Curtain Gas interface is optimized at the highest setting without losing sensitivity. Refer to the section: [Ion Source Optimization](#).

Use the following procedure to optimize the **Curtain Gas (CUR)** parameter. The main function of this parameter is to prevent the contamination of the ion optics. The CUR parameter should always be maintained as high as possible without losing sensitivity. The value depends on the type of mass spectrometer and ion source.

Do not set the parameter below the starting value.

1. On the Navigation bar, under **Tune and Calibrate**, double-click **Manual Tuning**.
2. Click **File > Open**.
3. In the **Files** list, click the acquisition method used to optimize the compound parameter and then click **OK**.  
The method opens in the Tune Method Editor.
4. Open the Source/Gas tab.
5. Using the ion source and gas flow guide, set all of the ion source and gas parameters so that they are appropriate for the flow rate.

## Manual Compound Optimization

---

6. Set the run time long enough so that many parameters can be adjusted. A recommended starting time is 15 minutes.
7. Click **Start**.  
Data is shown in panes below the Tune Method Editor.
8. Note the signal of the peak of interest.
9. In the **Curtain Gas (CUR)** field, increase the value by five.
10. Continue increasing the **Curtain Gas (CUR)** value until the highest value without losing sensitivity is found.  
As with most Source/Gas parameters, if two values give the same result, then use the higher value.
11. Repeat this procedure for the other Source/Gas parameters.  
When optimizing for these parameters, look for the value that gives the highest signal-to-noise value.

## Advanced Parameters

The following parameters should only be optimized by an experienced operator.

### Optimize AF2

The AF2 parameter controls the fragmentation of the second precursor ion in an MS3 scan. The amount of excitation energy used depends on the compound and the desired amount of fragmentation.

1. For both positive and negative ion modes, ramp **AF2** as follows: 70 mV to 300 mV, with a **Step** value of 10 mV. The allowable range is 0 mV to 1000 mV.
2. Select the AF2 value that gives the best amount of fragmentation.

### About Collision Energy Spread (CES)

A unique feature of QTRAP mass spectrometers is the ability to perform AutoFrag experiments. A composite spectrum can be obtained by filling the linear ion trap with ions from three distinct collision energies, which eliminates the need for optimizing the collision energy.

Under the compound-dependent parameters in Enhanced Product Ion (EPI) and MS3 experiments, the collision energy spread (CES) field allows users to specify the difference in collision energies that will be applied in the experiment. For example, if CE value of 30 and a CES value of 15 are used, then collision energies of 15, 30, and 45 are used.

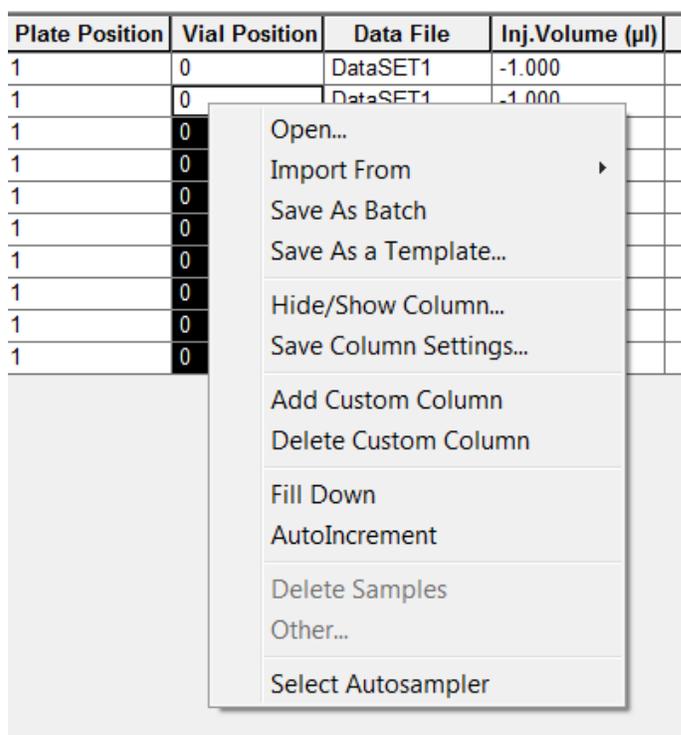
# Right-Click Menus

# G

## Batch Editor

Right-click the Batch Editor table to access the options.

**Figure G-1 Batch Right-Click Menu**



Menu	Function
Open	Opens a batch file.
Import From	Imports a batch from a file.
Save As Batch	Saves the batch with a different name.
Save As a Template	Saves the batch as a template.

## Right-Click Menus

---

Menu	Function
Hide/Show Column	Hides or shows a column.
Save Column Settings	Saves the batch column settings.
Add Custom Column	Adds a custom column.
Delete Custom Column	Deletes a custom column.
Fill Down	Copies the same data into the selected cells.
AutoIncrement	Automatically increments data in the selected cells.
Delete Samples	Deletes the selected row.
Select Autosampler	Selects an autosampler.

## Queue States and Device Status

The Queue Manager shows queue, batch, and sample status. Detailed information about a particular sample in the queue can also be viewed.

---

**Tip!** Click **View Queue** () to view the queue.

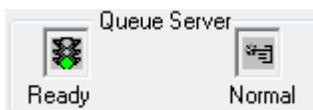
---

For information on using the Queue right-click menu, refer to the section: [Queue](#).

## Queue States

The current state of the queue is indicated in the Queue Server group.

**Figure G-2 Queue Server Indicator Showing Normal Mode**



**Figure G-3 Queue Server Indicator Showing Tune Mode**



The first icon indicates the queue state. The second icon indicates whether the queue is in Tune mode (for tuning) or Normal mode (for running samples). For descriptions of the icons and queue states, refer to the table: [Table G-1](#).

**Table G-1 Queue States**

Icons	State	Definition
<p>The image shows a 'Queue Server' indicator with two icons. The first icon is a traffic light with the red light illuminated, labeled 'Not Ready'. The second icon is a document with a tuning fork symbol, labeled 'Normal'.</p>	<b>Not Ready</b>	The hardware profile is deactivated and the queue is not accepting any sample submissions.
<p>The image shows a 'Queue Server' indicator with two icons. The first icon is a clock face, labeled 'Stand By'. The second icon is a document with a tuning fork symbol, labeled 'Normal'.</p>	<b>Stand By</b>	The hardware profile has been activated, but all devices are idle. Pumps are not running and gases are turned off.
<p>The image shows a 'Queue Server' indicator with two icons. The first icon is a document with a flame symbol, labeled 'Warming Up'. The second icon is a document with a tuning fork symbol, labeled 'Normal'.</p>	<b>Warming Up</b>	The mass spectrometer and devices are equilibrating, columns are being conditioned, the autosampler needle is being washed, and column ovens are reaching temperature. The duration of equilibration is selected by the operator. From this state, the system can go to the <b>Ready</b> state.
<p>The image shows a 'Queue Server' indicator with two icons. The first icon is a traffic light with the green light illuminated, labeled 'Ready'. The second icon is a document with a tuning fork symbol, labeled 'Normal'.</p>	<b>Ready</b>	The system is ready to start running samples and the devices have been equilibrated and are ready to run. In this state, the queue can receive samples and will run after samples are submitted.
<p>The image shows a 'Queue Server' indicator with two icons. The first icon is an hourglass, labeled 'Waiting'. The second icon is a document with a tuning fork symbol, labeled 'Normal'.</p>	<b>Waiting</b>	The system will automatically begin acquisition when the next sample is submitted.
<p>The image shows a 'Queue Server' indicator with two icons. The first icon is a person with a red exclamation mark, labeled 'PreRun'. The second icon is a document with a tuning fork symbol, labeled 'Normal'.</p>	<b>PreRun</b>	The method is being downloaded to each device and device equilibration is occurring. This state occurs before the acquisition of each sample in a batch.
<p>The image shows a 'Queue Server' indicator with two icons. The first icon is a document with a tuning fork symbol, labeled 'Acquiring'. The second icon is a document with a tuning fork symbol, labeled 'Normal'.</p>	<b>Acquiring</b>	The method is running and data acquisition is occurring.

## Right-Click Menus

**Table G-1 Queue States (continued)**

Icons	State	Definition
 <p>The icon shows a 'Queue Server' window with two buttons: 'Paused' (with a red stop sign) and 'Normal' (with a green play button).</p>	<b>Paused</b>	The system has been paused during acquisition.

## View Instrument and Device Status Icons

Icons representing the mass spectrometer and each device in the active hardware configuration are shown on the status bar in the bottom right corner of the window. The user can view the detailed status of an LC pump to make sure the LC pump pressure is appropriate or view the detailed status of the mass spectrometer to monitor the temperature of the ion source.

**Note:** For each status, the background color can be red. A red background indicates that the device encountered an error while in that state.

On the status bar, double-click the icon for the device or mass spectrometer. The Instrument Status dialog opens.

**Table G-2 Instrument and Device Status Icons**

Status	Icon	Background Color	Description
Idle		Green or yellow	The device is not running. If the background color is yellow, then the device should be equilibrated before it is ready to run. If the background color is green, then the device is ready to run.
Equilibrating		Green or yellow	The device is equilibrating.
Waiting		Green	The device is waiting for a command from the software or another device, or for some action by the operator.
Running		Green	The device is running a batch.
Aborting		Green	The device is aborting a run.
Downloading		Green	A method is being transferred to the device.

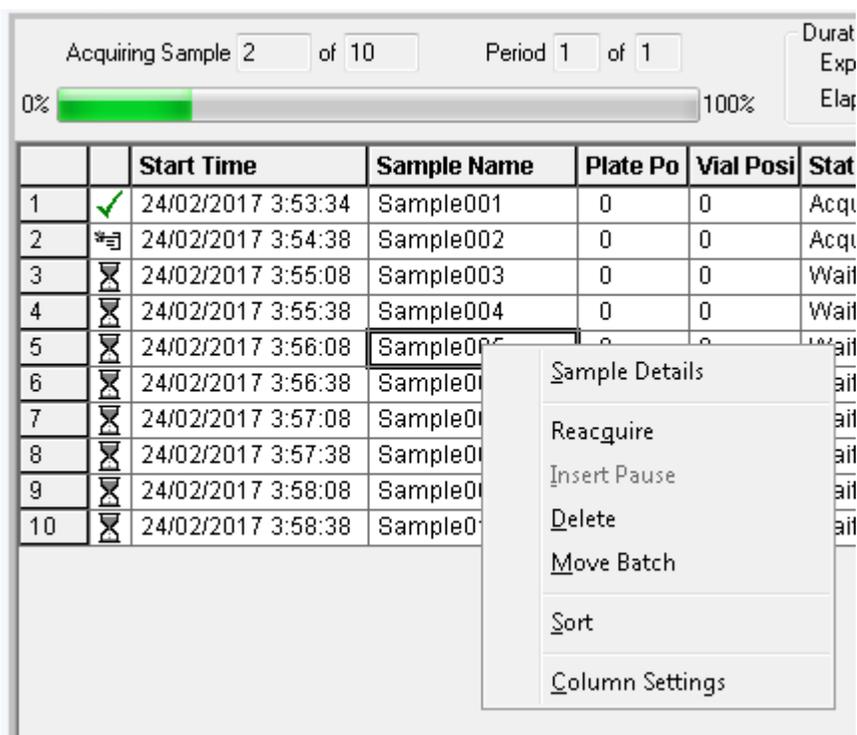
Table G-2 Instrument and Device Status Icons (continued)

Status	Icon	Background Color	Description
Ready		Green	The device is not running, but is ready to run.
Error		Red	The device has encountered an error that should be investigated.

## Queue

Right-click the Queue table to access the options.

Figure G-4 Queue Manager Right-Click Menu



Menu	Function
Sample Details	Opens the Sample Details dialog.
Reacquire	Acquires a sample again.

## Right-Click Menus

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Menu	Function
<b>Insert Pause</b>	Inserts a pause, in seconds, between two samples. This feature is not available in this version of the software.
<b>Delete</b>	Deletes either the batch or the selected samples.
<b>Move Batch</b>	Moves the batch within the queue.
<b>Sort</b>	Sorts on the preselected column.
<b>Column Settings</b>	Changes the column settings.

## Right-Click Menu for Contour Plot Panes

Menu	Function
Show DAD Spectrum	Opens a new pane with the DAD spectrum.
Extract Wavelengths (Use Range)	Extracts up to three wavelength ranges from a DAD spectrum to display the XWC.
Extract Wavelengths (Use Maximum)	Extracts wavelength ranges using the maximum wavelengths.
Zoom to selection	Zooms in on the selected area.
Add User Text	Adds a text box at the position of the cursor.
Undo Zoom	Returns the graph to the original scale
Delete Pane	Deletes the selected pane
Show Cross-Hair	Shows the Cross-Hair (nm/min)

## Show File Information Pane Right-click Menu

Table G-3 Show File Information Pane Right-click Menu

Menu	Function
<b>Copy</b>	Copies the selected data.
<b>Paste</b>	Pastes data.

Table G-3 Show File Information Pane Right-click Menu (continued)

Menu	Function
Select All	Selects all of the data in the pane.
Save To File	Saves data as an rtf file.
Font	Changes the font.
Save Acquisition Method	Saves the acquisition method as a dam file.
Save Acquisition Method to CompoundDB	Opens the Specify Compound Information dialog. Select the IDs and molecular weights to be saved in the compound database.
Delete Pane	Deletes the selected pane.

## Spectra Panes

Table G-4 Right-click Menu for Spectra Panes

Menu	Function
List Data	Lists the data points and integrates chromatograms.
Show TIC	Generates a new pane containing the TIC.
Extract Ions (Use Range)	Extracts a specific ion or set of ions from a selected pane and then generates a new pane containing a chromatogram for the specific ions.
Extract Ions (Use Maximum)	Extracts ions using the most intense peak in a selected area.
Save to Text File	Generates a text file of the pane, which can be opened in Microsoft Excel or other programs.
Save Explore History	Saves information about changes to processing parameters, also called Processing Options, that were made when a wiff file was processed in Explore mode. The processing history is stored in a file with an eph (explore processing history) extension.
Add Caption	Adds a caption at the cursor location in the pane.
Add User Text	Adds a text box at the cursor location in the pane.
Show Last Scan	Shows the scan prior to the selection.

## Right-Click Menus

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**Table G-4 Right-click Menu for Spectra Panes (continued)**

Menu	Function
Select Peaks For Label	In this dialog, select the parameters to reduce peak labeling.
Delete Pane	Deletes the selected pane.
Add a Record	Adds records and compound-related data, including spectra, to the library. An active spectrum is required to perform this task.
Search Library	Searches the library without constraints or with previously saved constraints.
Set Search Constraints	Searches the library using the criteria typed in Search Constraints dialog.

## Chromatogram Panes

**Table G-5 Right-click Menu for Chromatogram Panes**

Menu	Function
List Data	Lists the data points and integrates the peaks found in chromatograms.
Show Spectrum	Generates a new pane containing the spectrum.
Show Contour Plot	Shows a color-coded plot of a dataset, where the color represents the intensity of the data at that point. Only certain MS modes are supported.
Extract Ions	Extracts a specific ion or set of ions from a selected pane and then generates a new pane containing a chromatogram for the specific ions.
Show Base Peak Chromatogram	Generates a new pane containing a base peak chromatogram.
Show ADC Data	Generates a new pane containing the ADC data trace, if acquired.
Show Auxiliary Traces	Opens the Select Auxiliary Trace Channel dialog.
Show UV Detector Data	Generates a new pane containing the UV data trace, if acquired.
Save to Text File	Generates a text file containing the data in a pane, which can be opened in Microsoft Excel or other programs.

Table G-5 Right-click Menu for Chromatogram Panes (continued)

Menu	Function
<b>Save Explore History</b>	Saves information about changes to processing parameters, also called processing options, that were made when a wiff file was processed in Explore mode. The processing history is stored in a file with an eph (explore processing history) extension.
<b>Add Caption</b>	Adds a caption at the cursor location in the pane.
<b>Add User Text</b>	Adds a text box at cursor location in the pane.
<b>Set Subtract Range</b>	Sets the subtract range in the pane.
<b>Clear Subtract Range</b>	Clears the subtract range in the pane.
<b>Subtract Range Locked</b>	Locks or unlocks the subtract ranges. If the subtract ranges are not locked, then each subtract range can be moved independently. The subtract ranges are preset to locked.
<b>Delete Pane</b>	Deletes the selected pane.

## Results Table

Right-click the Results Table to access the options shown in the following table.

Table G-6 Results Table Right-click Menu

Menu	Function
<b>Full</b>	Shows all the columns.
<b>Summary</b>	Shows specific columns.
<b>Analyte</b>	Shows a specific analyte.
<b>Analyte Group</b>	Creates an analyte group.
<b>Sample Type</b>	Shows samples of a specific type or all samples.
<b>Add Formula Column</b>	Adds a formula column. It is recommended that the user validate the results if a formula column is used.
<b>Table Settings</b>	Edits or selects a table setting.

## Right-Click Menus

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**Table G-6 Results Table Right-click Menu (continued)**

Menu	Function
<b>Query</b>	Creates or selects a query.  We recommend that the user validate any queries that are used to analyze data in a Results Table.
<b>Sort</b>	Creates a sort or sorts by index.
<b>Metric Plot</b>	Creates a metric plot.
<b>Delete Pane</b>	Deletes the active pane.
<b>Fill Down</b>	Copies the same data into the selected cells.
<b>Add Custom Column</b>	Adds a custom column.
<b>Delete Custom Column</b>	Deletes the selected custom column.

## Peak Review

Right-click the **Peak Review** window or pane to access the options shown in [Table G-7](#).

**Table G-7 Peak Review Right-click Menu**

Menu	Function
<b>Options</b>	Opens the Peak Review Options dialog.
<b>Sample Annotation</b>	Opens the Sample Annotation dialog.
<b>Save Active to Text File</b>	Saves the selected peak as a text file.
<b>Show First Page</b>	Goes to the first sample.
<b>Show Last Page</b>	Goes to the last sample.
<b>Slide Show Peak Review</b>	Opens the slide show.
<b>Update Method</b>	Updates the algorithm for all peaks.

Table G-7 Peak Review Right-click Menu (continued)

Menu	Function
<b>Revert to Method</b>	Selects a redefined peak based on the current quantitation method.
<b>Delete Pane</b>	Deletes the active pane.

## Calibration Curve

Right-click the Calibration window or pane table to access the options shown in the following table.

Table G-8 Calibration Curve Right-click Menu

Menu	Function
<b>Exclude (Include)</b>	Right-click a point and then click <b>Exclude</b> to exclude the point from the curve. Right-click a point and then click <b>Include</b> to include the point.
<b>Exclude All Analytes (Include All Analytes)</b>	Right-click a point and then click <b>Exclude All Analytes</b> to exclude all of the analytes from the curve. Right-click a point and then click <b>Include All Analytes</b> to include the points.
<b>Show Peak</b>	Reviews an individual peak.
<b>Overlay</b>	Overlays two graphs.
<b>Active Plot</b>	Determines which plot is active.
<b>Legend</b>	Shows the graph legend.
<b>Log Scale X Axis*</b>	Uses a log scale for the X-axis.
<b>Log Scale Y Axis*</b>	Uses a log scale for the Y-axis.
<b>Delete Pane</b>	Deletes the active pane.
<b>Home Graph</b>	Scales the graph to its original size
* A log scale arranges the data points in a more manageable view so that the effect of all points can be monitored simultaneously. For this view, select <b>Log Scale Y Axis</b> versus <b>Log Scale X</b> and not just the log of one axis.	

# Glossary of Symbols

# H

**Note:** Not all of the symbols in the following table are applicable to every instrument.

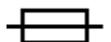
Symbol	Description
	Australian Regulatory Compliance Mark. Indicates that the product complies with Australian Communications Media Authority (ACMA) EMC Requirements.
	Alternating current
A	Amperes (current)
	Asphyxiation Hazard
	Authorized representative in the European community
	Biohazard
	CE Marking of Conformity
	cCSAus mark. Indicates electrical safety certification for Canada and USA.
	Catalog number

## Glossary of Symbols

Symbol	Description
	Caution. Consult the instructions for information about a possible hazard. <b>Note:</b> In SCIEX documentation, this symbol identifies a personal injury hazard.
	China RoHS Caution Label. The electronic information product contains certain toxic or hazardous substances. The center number is the Environmentally Friendly Use Period (EFUP) date, and indicates the number of calendar years the product can be in operation. Upon the expiration of the EFUP, the product must be immediately recycled. The circling arrows indicate the product is recyclable. The date code on the label or product indicates the date of manufacture.
	China RoHS logo. The device does not contain toxic and hazardous substances or elements above the maximum concentration values and it is an environmentally-friendly product that can be recycled and reused.
	Consult instructions for use.
	Crushing Hazard
	cTUVus mark for TUV Rheinland of North America
	Data Matrix symbol that can be scanned by a barcode reader to obtain a unique device identifier (UDI)
	Environmental Hazard
	Ethernet connection

## Glossary of Symbols

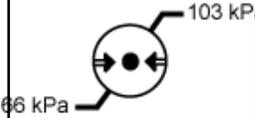
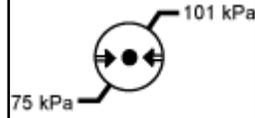
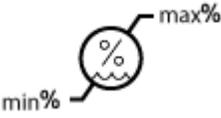
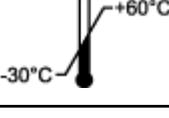
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Symbol	Description
	Explosion Hazard
	Eye Injury Hazard
	Fire Hazard
	Flammable Chemical Hazard
	Fragile
	Fuse
Hz	Hertz
	International safety symbol "Caution, risk of electric shock" (ISO 3864), also known as High Voltage symbol If the main cover must be removed, then contact a SCIEX representative to prevent electric shock.
	Hot Surface Hazard
	In Vitro Diagnostic Device
	Ionizing Radiation Hazard

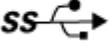
## Glossary of Symbols

Symbol	Description
	Keep dry. Do not expose to rain. Relative humidity must not exceed 99%.
	Keep upright.
	Lacerate/Sever Hazard
	Laser Radiation Hazard
	Lifting Hazard
	Magnetic Hazard
	Manufacturer
	Moving Parts Hazard
	Pacemaker Hazard. No access to people with pacemakers.
	Pinching Hazard

## Glossary of Symbols

Symbol	Description
	Pressurized Gas Hazard
	Protective Earth (ground)
	Puncture Hazard
	Reactive Chemical Hazard
	Serial number
	Toxic Chemical Hazard
	Transport and store the system within 66 kPa to 103 kPa.
	Transport and store the system within 75 kPa to 101 kPa.
	Transport and store the system within the specified minimum ( <b>min</b> ) and maximum ( <b>max</b> ) levels of relative humidity, noncondensing.
	Transport and store the system within $-30\text{ }^{\circ}\text{C}$ to $+45\text{ }^{\circ}\text{C}$ .
	Transport and store the system within $-30\text{ }^{\circ}\text{C}$ to $+60\text{ }^{\circ}\text{C}$ .

## Glossary of Symbols

Symbol	Description
	USB 2.0 connection
	USB 3.0 connection
	Ultraviolet Radiation Hazard
	United Kingdom Conformity Assessment Mark
VA	Volt Ampere (power)
V	Volts (voltage)
	WEEE. Do not dispose of equipment as unsorted municipal waste. Environmental Hazard
W	Watts
	<i>yyyy-mm-dd</i> Date of manufacture

# Glossary of Warnings



**Note:** If any of the labels used to identify a component become detached, contact a Field Service Employee (FSE).

Label	Translation (if applicable)
EN61326—1, EN61326—2-6, CLASS A, GROUP 1, ISM-IVD EQUIPMENT	EN61326—1, EN61326—2-6, CLASS A, GROUP 1, ISM-IVD EQUIPMENT
IMPACT INDICATOR  SENSITIVE PRODUCT WARNING	IMPACT INDICATOR  SENSITIVE PRODUCT WARNING  <b>Note:</b> If the indicator is tripped, then this container has been dropped or otherwise mishandled. Make a note on the Bill of Lading and then check for damage. Any claims for shock damage require a notation.
IMPORTANT!  RECORD ANY VISIBLE CRATE DAMAGE INCLUDING TRIPPED “IMPACT INDICATOR” OR “TILT INDICATOR” ON THE WAYBILL BEFORE ACCEPTING SHIPMENT AND NOTIFY YOUR LOCAL AB SCIEX CUSTOMER SUPPORT ENGINEER IMMEDIATELY.  DO NOT UNCRATE. CONTACT YOUR LOCAL CUSTOMER SUPPORT ENGINEER FOR UNCRATING AND INSTALLATION.	IMPORTANT!  RECORD ANY VISIBLE CRATE DAMAGE INCLUDING TRIPPED “IMPACT INDICATOR” OR “TILT INDICATOR” ON THE WAYBILL BEFORE ACCEPTING SHIPMENT AND NOTIFY YOUR LOCAL AB SCIEX CUSTOMER SUPPORT ENGINEER IMMEDIATELY.  DO NOT UNCRATE. CONTACT YOUR LOCAL CUSTOMER SUPPORT ENGINEER FOR UNCRATING AND INSTALLATION.
MINIMUM OF SIX PERSONS REQUIRED TO SAFELY LIFT THIS EQUIPMENT	MINIMUM OF SIX PERSONS REQUIRED TO SAFELY LIFT THIS EQUIPMENT  <b>Note:</b> Consult instructions for use.

## Glossary of Warnings

Label	Translation (if applicable)
TIP & TELL	<p>Tilt Indicator</p> <hr/> <p><b>Note:</b> Indicates whether the container was tipped or mishandled. Write on the Bill of Lading and inspect for damage. Any claims for tipping require a notation.</p>
TiltWatch PLUS ShockWatch	<p>Tilt Indicator</p> <hr/> <p><b>Note:</b> Indicates whether the container was tipped or mishandled. Write on the Bill of Lading and inspect for damage. Any claims for tipping require a notation.</p>
WARNING: DO NOT OPERATE WITHOUT FIRST ENSURING BOTTLE CAP IS SECURED.	<p>WARNING: DO NOT OPERATE WITHOUT FIRST ENSURING BOTTLE CAP IS SECURED.</p> <hr/> <p><b>Note:</b> This warning is attached to the source exhaust waste bottle.</p>
WARNING: NO USER SERVICEABLE PARTS INSIDE. REFER SERVICING TO QUALIFIED PERSONNEL.	<p>WARNING: NO USER SERVICEABLE PARTS INSIDE. REFER SERVICING TO QUALIFIED PERSONNEL.</p> <hr/> <p><b>Note:</b> Consult instructions for use.</p>

# Contact Us

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## Customer Training

- In North America: [NA.CustomerTraining@sciex.com](mailto:NA.CustomerTraining@sciex.com)
- In Europe: [Europe.CustomerTraining@sciex.com](mailto:Europe.CustomerTraining@sciex.com)
- Outside the EU and North America, visit [sciex.com/education](http://sciex.com/education) for contact information.

## Online Learning Center

- [SCIEX Now Learning Hub](#)

## SCIEX Support

SCIEX and its representatives maintain a staff of fully-trained service and technical specialists located throughout the world. They can answer questions about the system or any technical issues that might arise. For more information, visit the SCIEX website at [sciex.com](http://sciex.com) or contact us in one of the following ways:

- [sciex.com/contact-us](http://sciex.com/contact-us)
- [sciex.com/request-support](http://sciex.com/request-support)

## CyberSecurity

For the latest guidance on cybersecurity for SCIEX products, visit [sciex.com/productsecurity](http://sciex.com/productsecurity).

## Documentation

This version of the document supersedes all previous versions of this document.

To view this document electronically, Adobe Acrobat Reader is required. To download the latest version, go to <https://get.adobe.com/reader>.

To find software product documentation, refer to the release notes or software installation guide that comes with the software.

To find hardware product documentation, refer to the *Customer Reference* DVD that comes with the system or component.

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**Note:** To request a free, printed version of this document, contact [sciex.com/contact-us](http://sciex.com/contact-us).

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