

# **Turbo V Ion Source**

# For the X500 QTOF and ZenoTOF 7600/7600+ Systems

**Operator Guide** 



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# **Operational Precautions and Limitations**

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

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

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.

# **Operational Precautions and Hazards**

For regulatory and safety information for the mass spectrometer, refer to the document: *System User Guide*.



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.



WARNING! Hot Surface Hazard. Before any maintenance procedures are started, let the temperature of the Turbo V ion source decrease for at least 30 minutes. Some surfaces of the ion source and vacuum interface become hot during operation.



WARNING! Puncture Hazard, Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. If the ion source window is cracked or broken, then do not use the ion source. 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.



WARNING! Fire and Toxic Chemical Hazard. Before solvent leaks are corrected, make sure that the liquid flow to the ion source is stopped, that the ion spray voltage is off, that there are no open flames or other sources of fire nearby, and that the room is sufficiently ventilated. The fluid from a leak can be highly flammable. If the fluid is exposed to electrical discharges or a source of fire, then ignition can occur. If ventilation is not sufficient, then the fluid might cause poisoning.



WARNING! Toxic Chemical Hazard. Wear personal protective equipment (PPE), including a laboratory coat, gloves, and safety glasses, to avoid skin or eye exposure.



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 applicable personal protective equipment and absorbent wipes to contain the spill and dispose of it following local regulations.



WARNING! Environmental Hazard. Do not discard system components in municipal waste. To discard components correctly, obey 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.

# **Chemical Precautions**



WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Before cleaning or maintenance, identify whether decontamination is required. 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 discard system components in municipal waste. To discard components correctly, obey local regulations.



WARNING! Biohazard or Toxic Chemical Hazard. To prevent leaks, connect the drain tubing to the mass spectrometer and the source exhaust drain bottle correctly.

• Before servicing and regular maintenance, identify the chemicals that have been used in the system. For the health and safety precautions that must be obeyed for a chemical, refer to the

safety data sheet (SDS). For storage information, refer to the certificate of analysis. To find a SCIEX SDS or certificate of analysis, go to sciex.com/tech-regulatory.

• Always wear assigned personal protective equipment, which includes powder-free gloves, protective eyewear, and a laboratory coat.

Note: Nitrile or neoprene gloves are recommended.

- Do work in a well-ventilated area or fume hood.
- When flammable materials such as isopropanol and methanol are in use, do not go near ignition sources.
- Be careful with the use and disposal of any chemicals. If the correct procedures for chemical use and disposal are not obeyed, then personal injury can occur.
- During cleaning, do not let chemicals touch the skin. Wash hands after use.
- Make sure that all exhaust hoses are connected correctly and that all connections are functioning as designed.
- Collect all spent liquids and discard them as hazardous waste.
- Obey all of the local regulations for the storage, use, and disposal of biohazardous, toxic, and radioactive materials.

#### System Safe Fluids

The following fluids can safely be used with the system.

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

- 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%
- 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%

#### Laboratory Conditions Safe Environmental Conditions

The system is designed to operate safely in 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%, noncondensing
- Mains supply voltage fluctuations: ±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:

• Ambient temperature is between 15 °C to 30 °C (59 °F to 86 °F).

Over time, the temperature must remain within a range of 2 °C (3.6 °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 is between 20% to 80%, non-condensing.

# **Equipment Use and Modification**



WARNING! Electrical Shock Hazard. Do not remove the covers. If the covers are removed, then injury or incorrect system operation can occur. Removal of the covers is not required for routine maintenance, inspection, or adjustment. For repairs that require removal of the covers, contact a SCIEX field service employee (FSE).



WARNING! Personal Injury Hazard. Use only parts that are recommended by SCIEX. The use of parts that are not recommended by SCIEX or the use of parts for any purpose other than their intended purpose can put the user at risk of harm or have a negative effect on system performance.

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 conditions or in an environment that are not approved by the manufacturer, then the performance and protection that is supplied by the equipment might be decreased or lost.

Contact an FSE for information about servicing the system. Unauthorized modification or operation of the system might cause personal injury and equipment damage, and might void the warranty. If the system is operated outside the recommended environmental conditions or with unauthorized modifications, then the acquired data might be inaccurate.

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

The twin ESI probe is used for ESI mode operation. The twin APCI probe is used for APCI mode operation. The standard probe supplied with the ion source is the twin ESI probe.

The twin probes enable on-demand introduction of calibrant and sample through independent electrodes.

Applications for the ion source include qualitative method development and qualitative and quantitative analysis.

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

#### **Ionization Modes**

#### 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  $\mu$ L/min to 3,000  $\mu$ L/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.
- 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.

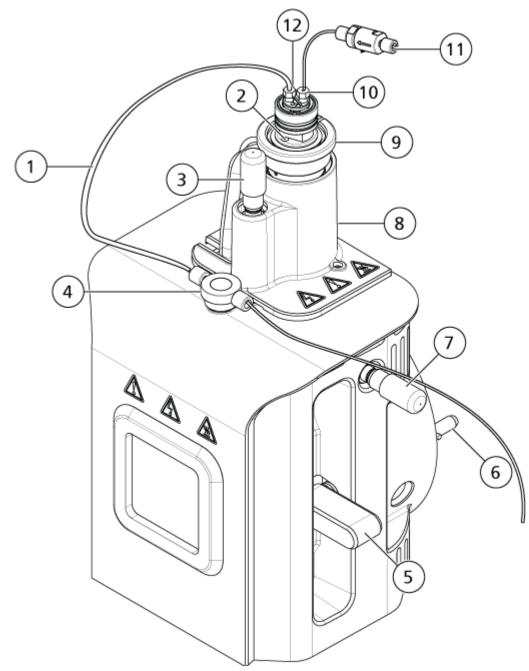
• Rapid sample introduction by flow injection with or without an LC column.

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  $\mu$ L/min to 3,000  $\mu$ L/min, through a wide-bore column.

Refer to the section: APCI Mode.

# **Ion Source Components**

Figure 2-1 Ion Source Components



ltem	Description
1	Sample tubing

ltem	Description
2	Corona discharge needle adjustment screw
3	Y-axis micrometer used to position the probe on the vertical axis for ion source sensitivity adjustments
4	Grounding union
5	One of two source latches that secure the ion source to the mass spectrometer
6	Guide pin
7	X-axis micrometer used to position the probe on the horizontal axis for ion source sensitivity adjustments
8	Probe tower
9	Retaining ring
10	Calibrant (CAL) port with fitting
11	Flow module, consisting of calibrant tubing and check valve
12	Sample (LC) port with fitting

#### **Probes**

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

Specification	Twin ESI Probe	Twin 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
lon source gas 1 / ion source gas 2	Refer to the mass spectrometer document: <i>Site Planning Guide</i>	

Table 2-1 Ion Source Specifications

The software for the mass spectrometer identifies the installed probe and enables the corresponding user controls.

#### Twin ESI Probe

The twin ESI probe is 220 mm long. It contains two 100  $\mu$ m (0.004 inch) inner diameter (i.d.) stainless steel electrodes and is located centrally, with the two turbo heaters placed at a 45 degree angle to each side.

The sample supply is connected to the port labeled **LC** and the calibrant is connected to the port labeled **CAL**. Analytes (samples or calibrants) introduced through the twin ESI probe are ionized within the tubing by the application of high voltage (**Spray Voltage**). The ions are then nebulized by a jet of compressed zero air, creating a mist of small, highly-charged droplets. The combination of the spray and the heated dry gas from the turbo heaters is projected at a 90 degree angle to the ion path.

#### Figure 2-2 Parts of the Twin ESI Probe



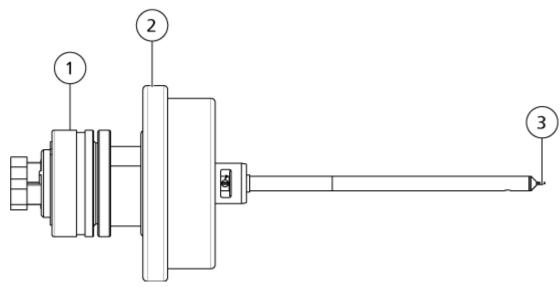
ltem	Description
1	Electrode adjustment nut (black collar) that adjusts the extension of the electrode tips (protrusion)
2	Retaining ring that fastens the probe to the probe tower on the ion source housing
3	Electrode tips through which sample or calibrant are sprayed in the sample inlet area of the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

#### **Twin APCI Probe**

The twin APCI probe is 125 mm long. It contains two 100  $\mu$ m (0.004 inch) inner diameter (i.d.) stainless steel electrodes surrounded by a flow of nebulizer gas (Gas 1).

The sample supply is connected to the port labeled **LC** and the calibrant is connected to the port labeled **CAL**. Analytes (samples or calibrants) are pumped through the sprayer, where they are nebulized in a ceramic tube containing a heater. A sensor embedded in the heater makes sure that the ceramic tube is kept at an appropriate temperature. A high-velocity jet of nebulizer

gas flows around the electrode tip to disperse the sample as a mist of fine particles. The sample 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.



#### Figure 2-3 Parts of the Twin APCI Probe

Item	Description
1	Electrode adjustment nut (black collar) that adjusts the extension of the electrode tips (protrusion)
2	Retaining ring that fastens the probe to the probe tower on the ion source housing
3	Electrode tips through which sample or calibrant 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 incorrectly 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. 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 decrease 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 help the user safely remove and correctly handle 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-supplied exhaust ventilation system. For information about the ventilation requirements for the source exhaust system, refer to the document: *Site Planning Guide*.

**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.



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 tempered glass window on the front of the ion source.

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

#### **Required Materials**

- Ion source
- Twin ESI probe
- (Optional) Twin APCI probe
- 1/4 inch wrench
- 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 figures: Figure 2-2 and Figure 2-3.

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 Twin ESI Probe Position or Optimize the Twin 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 tips 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 ESI probe is in use.

#### **Prerequisite Procedures**

• Remove the Ion Source.

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.

- 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.
- 6. For the twin 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 LC port at the top of the probe, and then tighten the sample tubing nut until it is finger-tight. Use a 1/4-inch wrench to tighten it one quarter turn further.

The twin probe has two ports. Be sure to use the port labeled LC.

- 3. Connect the other end of the tubing to the grounding union on the ion source.
- 4. Connect the calibrant tubing to the port labeled **CAL**. Tighten the hex nut until it is fingertight, and then use a 1/4-inch wrench to tighten it one quarter turn further.

## 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 tips 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 3-1 O-rings on the Vacuum Interface

Item	Description
1	Curtain plate
2	O-rings

- 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.
- 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.

#### Sample Inlet Requirements

- Use appropriate analytical procedures and practices to minimize external dead volumes. The sample inlet transfers the liquid sample to the ion source inlet without loss and with minimal dead volume.
- Prefilter samples so that the capillary tubing in the sample inlets is not blocked by particles, precipitated samples, or salts.
- Make sure that all of the connections are tight enough to prevent leaks. Do not over-tighten.

#### **Examine for Leaks**



WARNING! Toxic Chemical Hazard. Wear personal protective equipment (PPE), including a laboratory coat, gloves, and safety glasses, to avoid skin or eye exposure.

Regularly examine the ion source for leaks.

• Examine fittings and tubing to make sure that there are no leaks.



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.



WARNING! Fire Hazard. Do not direct more than 3 mL/min of flammable solvent in the ion source. 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. If the ion source window is cracked or broken, then do not use the ion source. 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: Twin ESI Probe Optimization or Twin APCI Probe Optimization.

# Sample Introduction

#### Method

The liquid sample stream is delivered to the ion source by an LC pump. The sample can be injected directly in the mobile phase using flow injection analysis (FIA) or tee infusion, through a syringe pump (not supplied), or through a separation column using a loop injector or autosampler.

#### **Flow Rate**

Sample flow rates are determined by the LC system or syringe pump. The twin ESI probe supports flow rates from 5  $\mu$ L/min to 3,000  $\mu$ L/min. The twin APCI probe supports flow rates from 200  $\mu$ L/min to 3,000  $\mu$ L/min.

## **Twin ESI 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 supplied. Adequate laboratory ventilation is required to control solvent and sample emissions, and for safe operation of the system.

WARNING! Fire Hazard. Do not direct more than 3 mL/min of flammable solvent in the ion source. 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:** 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.

**Note:** The **IonSpray Voltage** is always applied to both the TurbolonSpray probe and the APCI probe simultaneously, and the temperature parameter is always applied to both the turbo and APCI heaters simultaneously.

#### Flow Rate and Ion Source Temperature

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

The twin ESI probe is often used with sample flow rates of 5  $\mu$ L/min to 3,000  $\mu$ L/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.

#### Set Up the System

- 1. Configure the LC pump to deliver the mobile phase at the required flow rate. Refer to the section: Source Parameters and Voltages.
- 2. Connect the grounding union on the ion source to an LC pump, through an injector equipped with a loop, or to an autosampler.
- 3. If an autosampler is being used, then configure the autosampler to perform multiple injections.

#### **Prepare the System**

- 1. Open the control software.
- 2. Open a previously optimized method or create a method based on the compounds.
- 3. 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.

4. Start the solvent flow and sample injection.

#### Set the Starting Conditions

- Type a starting value for **Ion Source Gas 1**.
   For LC pumps, use a value between 40 and 60 for Gas 1.
- Type a starting value for **Ion Source Gas 2**.
   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.

3. Type the appropriate value in the Spray Voltage field.

- Positive mode: 5500
- Negative mode: -4500
- 4. Type 25 in the Curtain Gas field.
- 5. Start acquisition.

#### **Optimize the Twin ESI 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. Use the previous horizontal and vertical micrometer settings or set them to **5** as a starting position.
- 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 twin ESI 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.

**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.

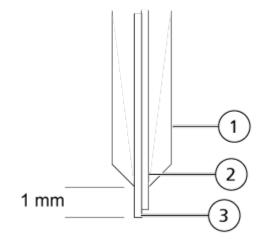
5. Adjust the black electrode adjustment nut on the probe to move the electrode tube in or out of the probe (to adjust the protrusion).

Note: Make sure that both electrodes protrude from the probe.

**Tip!** If the spray is too close to the aperture, then it will interfere with the flow of the gas for the Curtain Gas Interface, resulting in contamination of the vacuum interface. To prevent contamination, move the probe up, using the vertical micrometer.

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 4-1 Electrode Tip Extension Adjustment



ltem	Description
1	Twin probe
2	Calibrant electrode
3	Sample 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 twin ESI 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.

Use the lowest ion source voltage possible without losing signal.

**Note:** 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.
- 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 decrease sensitivity. Do not set the flow rate lower than the values in the table: Table 4-1. This helps to prevent penetration of the flow of the gas for the Curtain Gas interface, which can cause a noisy signal; prevent contamination of the aperture; and increase the overall signal-to-noise ratio.

#### **Table 4-1 CUR Parameter Values**

Mass Spectrometer	Starting Value
X500 QTOF and ZenoTOF 7600/7600+ systems	25

3. Adjust the ion source 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.

#### **Twin APCI 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 supplied. Adequate laboratory ventilation is required to control solvent and sample emissions, and for safe operation of the system. WARNING! Fire Hazard. Do not direct more than 3 mL/min of flammable solvent in the ion source. 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  $\mu$ L/min. For a complete list of APCI probe parameters, refer to the section: Twin APCI Probe Parameters.

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

**Note:** The **IonSpray Voltage** is always applied to both the TurbolonSpray probe and the APCI probe simultaneously, and the temperature parameter is always applied to both the turbo and APCI heaters simultaneously.

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

#### Set Up the System

- 1. Configure the LC pump to deliver the mobile phase at the required flow rate. Refer to the section: Source Parameters and Voltages.
- 2. Connect the grounding union on the ion source to an LC pump, through an injector equipped with a loop, or to an autosampler.
- 3. If an autosampler is being used, then configure the autosampler to perform multiple injections.

#### Prepare the System

- 1. Open the control software.
- 2. Open a previously optimized method or create a method based on the compounds.

- 3. 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.

4. Start the solvent flow and sample injection.

#### Set the Starting Conditions

- 1. Type **30** in the **Ion Source Gas 1** field.
- 2. Type 1 in the **Nebulizer Current (NC)** field.
- 3. 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 decrease sensitivity. Do not set the flow rate lower than the values in the table: Table 4-2. This helps to prevent penetration of the flow of the gas for the Curtain Gas interface, which can cause a noisy signal; prevent contamination of the aperture; and increase the overall signal-to-noise ratio.

#### **Table 4-2 CUR Parameter Values**

Mass Spectrometer	Starting Value
X500 QTOF and ZenoTOF 7600/7600+ systems	25

#### 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 twin APCI probe, make sure that the corona discharge needle is pointing toward the aperture. When using the twin ESI 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 Twin APCI Probe Position**



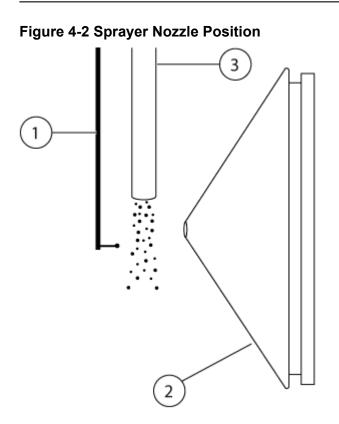
WARNING! Ionizing Radiation Hazard, Biohazard, or Toxic Chemical Hazard. Make sure that the electrode protrudes beyond the tip of the probe, to prevent hazardous vapors from escaping from the source. The electrode must not be recessed within the probe.



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.



ltem	Description
1	Corona discharge needle
2	Curtain plate
3	Twin APCI probe

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

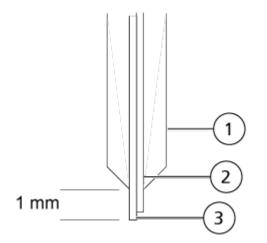
**Note:** To avoid reducing the performance of the mass spectrometer, do not spray directly in the aperture.

- 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 4-3 Electrode Tip Extension Adjustment



ltem	Description
1	Twin probe
2	Calibrant electrode
3	Sample 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  $\mu$ A and 5  $\mu$ 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.

• 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 ion source 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 ion source voltage possible without losing signal. In SCIEX OS, this is the **Spray Voltage** field. Focus on signal-to-noise and not just signal.
- For flow rates greater than 2 mL/min in APCI mode, equilibrate the mass spectrometer before starting the liquid flow, to make sure that the nebulization temperature is reached.

The following warnings apply to all of the maintenance procedures in this section.



WARNING! Hot Surface Hazard. Before any maintenance procedures are started, let the temperature of the Turbo V ion source decrease for at least 30 minutes. 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.



WARNING! Toxic Chemical Hazard. Wear personal protective equipment (PPE), including a laboratory coat, gloves, and safety glasses, to avoid skin or eye exposure.



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 applicable 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. If the ion source window is cracked or broken, then do not use the ion source. 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 identify how often to clean or do 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 examine 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

### **Recommended Maintenance Schedule**

The following table provides a recommended schedule for cleaning and maintaining the ion source. For a list of consumable and spare parts, refer to the document: *Parts and Equipment Guide*.

Tip! Do maintenance tasks regularly to make sure that system performance is optimal.

Contact a Qualified Maintenance Person (QMP) to order consumable parts and for basic service and maintenance requirements. Contact a SCIEX Field Service Employee (FSE) for all other service and maintenance requirements.

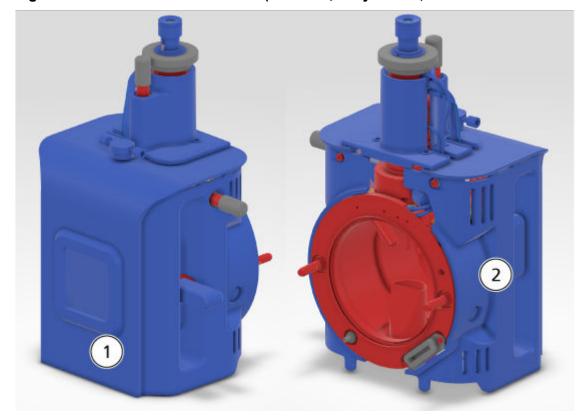
Note: For part numbers, refer to the document: Parts and Equipment Guide.

Component	Frequency	Task	For More Information
lon source probes	As required	Examine and replace	Refer to the section: Remove the Probe and Install the Probe.
Electrodes for the twin ESI or APCI probes	As required	Examine and replace	Refer to the section: Replace the Twin Electrodes.
Corona discharge needle	As required	Replace	Refer to the section: Replace the Corona Discharge Needle.
Turbo heater	As required	Replace	Contact the local QMP or FSE.
Sample tubing	As required	Replace	Refer to the section: Connect the lon Source Tubing.

Table 5-1 Ion Source Maintenance Tasks

# Ion Source Handling

Surfaces of the ion source become hot during operation. The following figure shows 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 5-1 Ion Source Hot Surfaces (Red=Hot, Gray=Warm, Blue=Handle with Care)

ltem	Description
1	Front
2	Back

### **Remove the Ion Source**

Note: Nitrogen continues to flow at a rate of 9 L/min when the mass spectrometer is 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. Stop the CDS.

#### Ion Source Maintenance

- (SCIEX OS) Click Standby ( 4.
- ) on the status panel. 5. Wait at least 30 minutes for the ion source to cool.
- Disconnect the sample tubing from the grounding union. 6.
- 7. Disconnect the calibrant tubing from the check valve.
- 8. Turn the two source latches up to the 12 o'clock position to release the ion source.
- 9. 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.

10. Put the ion source on a clean, secure surface.

### Clean the Ion Source Surfaces



WARNING! Electrical Shock Hazard. Before this procedure is started, remove the ion source from the mass spectrometer. Obey all electrical safe work practices.

#### **Prerequisite Procedures**

Remove the Ion Source.

Clean the surfaces of the ion source after a spill or when they become dirty.

• 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.
  - Set ion source gas 1 and ion source gas 2 to at least 40. C.
  - Set the flow rate of the gas for the Curtain Gas interface to the highest setting possible. d.

- 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. Before this procedure is started, remove the ion source from the mass spectrometer. Obey all electrical safe work practices.

CAUTION: Potential System Damage. Do not let the protruding electrode tips 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 calibrant tubing nut and then disconnect the calibrant tubing from the probe.
- 3. Loosen the retaining ring that secures the probe on the ion source housing.
- 4. Gently pull the probe straight up out of the tower.
- 5. Put the probe on a secure, clean surface.

### **Replace the Twin Electrodes**



WARNING! Electrical Shock Hazard. Before this procedure is started, remove the ion source from the mass spectrometer. Obey 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.

#### Ion Source Maintenance

The probe contains twin electrodes. Replace the twin electrodes 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.

- 1. Remove the electrode adjustment nut and then remove the twin electrodes.
- 2. Install the new twin electrodes inside the probe and then tighten the electrode adjustment nut.
- 3. Install the probe. Refer to the section: Install the Probe.
- 4. Install the ion source on the mass spectrometer. Refer to the section: Ion Source Installation.
- 5. Connect the sample tubing. Refer to the section: Connect the Ion Source Tubing.
- 6. Connect the calibrant tubing.
- 7. Adjust the electrode tip extension. Refer to the section: Optimize the Twin ESI Probe Position or Optimize the Twin APCI Probe Position.

### **Replace the Corona Discharge Needle**



WARNING! Electrical Shock Hazard. Before this procedure is started, remove the ion source from the mass spectrometer. Obey all electrical safe work practices.



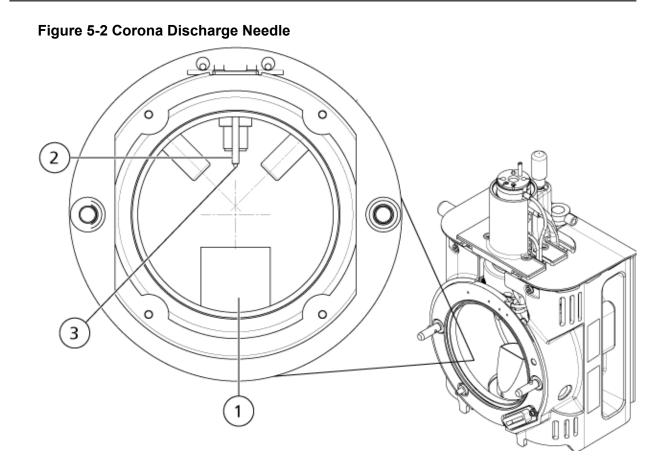
WARNING! Puncture Hazard. Handle the needle with care. The tip of the needle is extremely sharp.

#### **Prerequisite Procedures**

- Remove the Ion Source.
- Remove the Probe.

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.



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: lon 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. Before this procedure is started, remove the ion source from the mass spectrometer. Obey all electrical safe work practices.

Note: To replace the calibrant tubing, refer to the System User Guide.

#### 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 lon Source Tubing.
- 3. Install the ion source. Refer to the section: Ion Source Installation.
- 4. Start the sample flow.

### **Storage and Handling**



WARNING! Environmental Hazard. Do not discard system components in municipal waste. To discard components correctly, obey local regulations.

The environmental requirements for the storage and transport of the ion source:

- Ambient temperature between –30 °C and +60 °C (–22 °F and 140 °F)
- Atmospheric pressure between 75 kPa to 101 kPa
- Relative humidity not exceeding 99%, non-condensing

# Ion Source Troubleshooting

Symptom	Possible Cause	Corrective Action	
The control software reports that the mass spectrometer has gone to a Fault state.	<ol> <li>The probe is not installed.</li> <li>The probe is not connected securely.</li> </ol>	<ol> <li>Install the probe. Refer to the section: Install the Probe.</li> <li>Reinstall the probe:         <ul> <li>a. Remove the probe. Refer to the section: Remove the Probe.</li> <li>b. Install the probe, making sure to tighten the retaining ring securely. Refer to the section: Install the Probe.</li> </ul> </li> </ol>	
The spray is not uniform.	The electrode is blocked.	Replace the electrode. Refer to the section: Replace the Twin Electrodes.	
The ion source temperature is not reached or temperature is too high or unstable.	The turbo heater is faulty.	Contact the local QMP or FSE.	
Sensitivity is poor.	<ol> <li>The interface components (front end) are dirty.</li> <li>Solvent vapor or other unknown compounds are present in the analyzer region.</li> <li>The shorter electrode is not protruding from the probe.</li> </ol>	<ol> <li>Clean the interface components and then install the ion source.</li> <li>Optimize the flow rate of the gas for the Curtain Gas interface. Refer to the section: Ion Source Optimization.</li> <li>Adjust the electrode tip extension. Refer to the section: Optimize the Twin ESI Probe Position or Optimize the Twin APCI Probe Position.</li> </ol>	

Symptom	Possible Cause	Corrective Action	
During testing, the ion source fails to meet specifications.	<ol> <li>The test solution was not prepared correctly.</li> <li>The mass spectrometer has not passed the installation tests.</li> </ol>	<ol> <li>Confirm that the test solution was prepared correctly.</li> <li>If the problem cannot be resolved, then contact the FSE to perform the installation tests.</li> </ol>	

Symptom Possible 0		ssible Cause	Co	Corrective Action		
Background noise is high.	1.	The ion source temperature is too high.	1.	-	timize the ion source nperature.	
	2.	The ion source is contaminated.	2.	Op flov	timize the heater gas <i>N</i> .	
			3.	sou the	ean or replace the ion urce components, and en condition the ion urce and front end:	
				a.	Move the probe to the furthest position from the aperture (vertically and horizontally).	
				b.	Make sure that the interface heater is on.	
				C.	Infuse or inject 50:50 methanol:water with a pump flow rate of 1 mL/min.	
				d.	In the control software, set the ion source temperature to 650, ion source gas 1 to 60, and ion source gas 2 to 60.	
				e.	Set the flow rate of the gas for the Curtain Gas interface to 45 or 50.	
				f.	Acquire data for a minimum of 2 hours, or preferably overnight, for best results.	

Symptom	Possible Cause	Corrective Action
lon source performance has degraded.	<ol> <li>The probe is not optimized.</li> <li>The sample was not prepared correctly or the sample has degraded.</li> <li>The sample inlet fittings are leaking.</li> </ol>	<ol> <li>Optimize the probe. Refer to the section: Twin ESI Probe Optimization or Twin APCI Probe Optimization.</li> <li>Make sure that the sample was prepared correctly.</li> <li>Make sure that the fittings are tight and replace the fittings if leaks continue. Do not overtighten the fittings.</li> <li>Install and optimize an alternate ion source. If the issue persists, then contact an FSE</li> </ol>
Arcing or sparks occur.	The position of the corona discharge needle is incorrect or the electrode tip is damaged.	Turn the corona discharge needle toward the curtain plate, and away from the stream of heater gas. Refer to the section: Adjust the Position of the Corona Discharge Needle.
Calibrant signal is low.	<ol> <li>The CDS is not connected.</li> <li>The CDS tubing is blocked.</li> </ol>	<ol> <li>Inspect the CDS connections.</li> <li>Inspect the calibrant tubing for blockages or leaks.</li> </ol>

# **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.

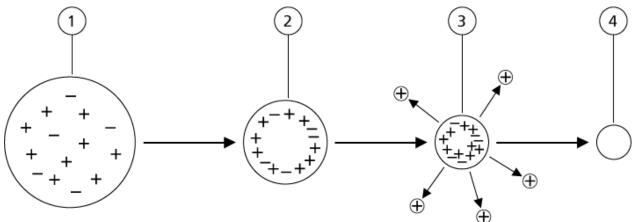
The interaction of the **Spray voltage** and the turbo heaters helps focus the stream and increases the rate of droplet evaporation, resulting in an increased ion signal. The heated gas increases the efficiency of ion evaporation, resulting in increased sensitivity and the ability to handle higher liquid sample flow rates.

A high-velocity flow of nebulizer gas shears droplets from the liquid sample stream in the **Spray 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 A-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.





ltem	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 involatile molecules in solution in a liquid to a molecular gas without inducing excessive decomposition. The twin APCI probe process of gently nebulizing the sample in 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,  $H_3O^+[H_2O]_n$ .

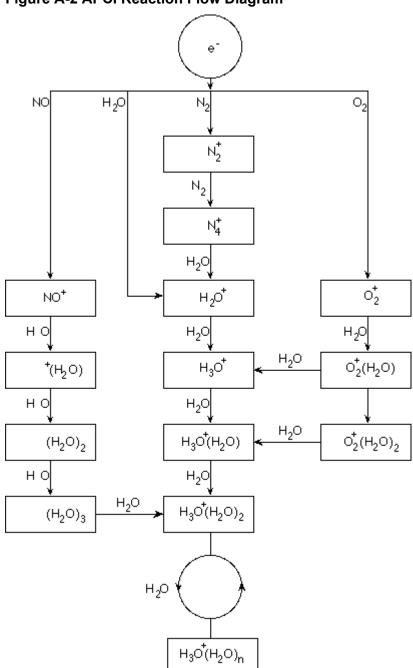


Figure A-2 APCI Reaction Flow Diagram

The major primary ions  $N_2^+$ ,  $O_2^+$ ,  $H_2O^+$ , and  $NO^+$  are formed by the electron impact of coronacreated 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.

**Operator Guide** RUO-IDV-05-1828-C Samples that are introduced through the twin 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]<sup>+</sup> in Positive polarity and [X-H]<sup>-</sup> in Negative polarity. Refer to the figure: Figure A-3. It is these reagent ions that produce stable sample ions when they collide with the sample molecules.

ltem	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		

#### Figure A-3 Atmospheric Pressure Chemical Ionization

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.

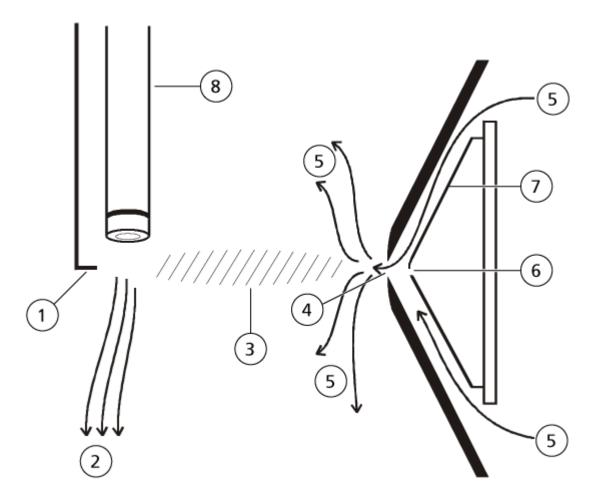
For reverse phase applications, the reagent ions consist of protonated solvent molecules in Positive polarity and solvated oxygen ions in Negative polarity. With favorable thermodynamics, the addition of modifiers changes the reagent ion composition. For example, the addition of acetate buffers or modifiers 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 twin 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.





ltem	Description
1	Discharge needle tip
2	Sample flow

ltem	Description
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 twin 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 twin 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 twin 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 twin APCI probe at temperatures above the optimal temperature might cause thermal decomposition of the sample.

# **Twin ESI Probe Parameters**

The following table shows the recommended operating conditions for the twin ESI 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.

Parameters		Operational			
	Low	Low Medium High		Range	
LC flow rate	5 μL/min to 50 μL/min	200 µL/min	1,000 µL/min	5 μL/min to 3,000 μL/min	
lon 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	
lon source gas 2 (heater gas)	0 psi	50 psi	50 psi	0 psi to 90 psi	
Spray voltage	5500 V	5500 V	5500 V	5500 V	
Gas for the Curtain Gas interface	25 psi	25 psi	25 psi	25 psi to 50 psi	
lon source temperature <sup>1</sup>	Ambient to 200 °C	200 °C to 650 °C	400 °C to 750 °C	Up to 750 °C	
Declustering potential (DP) <sup>2</sup>	Positive: 70 V Negative: –70 V	Positive: 70 V Negative: –70 V	Positive: 100 V Negative: –100 V	Positive: 0 V to 400 V Negative: –400 V to 0 V	
Probe vertical micrometer setting	7 to 10	2 to 5	0 to 2	0 to 13	

Table B-1 Parameter Optimization for the Twin ESI Probe

<sup>1</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>2</sup> DP values depend on the compound.

Parameters	Typical Values Operational			
	Low	Medium	High	Range
Probe horizontal micrometer setting	4 to 6	4 to 6	4 to 6	0 to 10

### Table B-1 Parameter Optimization for the Twin ESI Probe (continued)

# **Twin APCI Probe Parameters**

Parameter	Typical Value	Operational Range
LC flow rate	1,000 µL/min	200 μL/min to 3,000 μL/min
lon source gas 1 (nebulizer gas)	30 psi	0 psi to 90 psi
Gas for the Curtain Gas interface	25 psi	25 psi to 50 psi
lon source temperature <sup>3</sup>	400 °C	100 °C to 750 °C
Nebulizer current	Positive: 3 μΑ Negative: –3 μΑ	Positive: 0 mA to 5 μA Negative: –5 mA to 0 μA
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	4	Scale 0 to 13

### Table B-2 Parameter Optimization for the Twin APCI Probe

### **Parameter Descriptions**

### Table B-3 Source-Dependent Parameters

Parameter	Description
lon source gas 1	Controls the nebulizer gas for the twin ESI and twin APCI probes. Refer to the section: Principles of Operation — Ion Source.

<sup>&</sup>lt;sup>3</sup> Temperature values depend on the compound.

Parameter	Description
lon source gas 2	Controls the heater gas for the ESI probe . The best sensitivity is achieved when the combination of temperature and heater gas flow rate causes the LC solvent to reach a point at which it is nearly all vaporized. To optimize the ion source gas 2, increase the flow to obtain the best signal or signal-to-noise ratio if there is a significant increase in background noise. Too high a gas flow can produce a noisy or unstable signal. Refer to the section: Principles of Operation — Ion Source.
Curtain gas	Controls the flow rate of the gas for the Curtain Gas interface. The Curtain Gas interface is located between the curtain plate and the orifice. It prevents ambient air and solvent droplets from entering and contaminating the ion optics, while permitting direction of sample ions in the vacuum chamber by the electrical fields generated between the vacuum interface and the spray needle. Contamination of the ion entrance optics reduces Q0 transmission, stability, and sensitivity, and increases background noise. Maintain the flow rate of the gas for the Curtain Gas interface as high as possible without losing sensitivity.
lon source temperature	Controls the heat applied to the sample to vaporize it. The optimal ion source temperature is the lowest temperature at which the sample is vaporized completely.
	Optimize in increments of 50 °C.
Ion source	Controls the temperature of the heater gas in the ESI probe .
temperature (ESI probe)	The best sensitivity is achieved when the combination of temperature and ion source gas 2 flow rate causes the LC solvent to reach a point at which it is nearly all vaporized.
	As the organic content of the solvent increases, the optimal probe temperature decreases. With solvents consisting of 100% methanol or acetonitrile, the probe performance might optimize as low as 300 °C. Aqueous solvents consisting of 100% water at flows of approximately 1,000 µL/min require a maximum probe temperature of 750 °C.
	If the ion source temperature is set too low, then vaporization is incomplete, and large, and visible droplets are expelled into the ion source housing.
	If the ion source temperature is set too high, then solvent might vaporize prematurely at the probe tip, especially if the probe is set too low (5 to 13).

Table B-3 Source-Dependent Parameters (continued)

Parameter	Description
lon source	Controls the temperature of the APCI probe.
temperature (APCI probe)	As the organic content of the solvent increases, the optimal probe temperature should decrease. With solvents consisting of 100% methanol or acetonitrile the probe performance might optimize at temperatures as low as 400 °C at flow rates of 1,000 $\mu$ L/min. Aqueous solvents consisting of 100% water set at flows of approximately 2,000 $\mu$ L/min require a minimum probe temperature of 700 °C.
	If the ion source temperature is set too low, then vaporization is incomplete, and large, and visible droplets are expelled into the ion source housing.
	If the ion source temperature is set too high, then thermal degradation of the sample occurs.
Nebulizer current	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. For the APCI probe, the current applied to the corona discharge needle usually optimizes over a broad range of about 1 $\mu$ A to 5 $\mu$ A, in Positive polarity. To optimize, start at a value of 1 and then increase to achieve the best signal or signal-to-noise ratio. If, when the current is increased, no changes in signal are observed, then leave the current at the lowest setting that provides the best sensitivity, for example, 2 $\mu$ A.
lon source voltage	Controls the voltage applied to the sprayer in the ESI probe , which ionizes the sample in the ion source. The parameter value depends on the polarity, and affects the stability of the spray and the sensitivity.
Interface heater	Turns the interface heater on and off. Heating the interface helps maximize the ion signal and prevents contamination of the ion optics. Unless the compound the user is analyzing is extremely fragile, we recommend that the user heat the interface.

Table B-3 Source-Dependent Parameters (continued)

# **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.

# **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 twin ESI probe and 0.1% to 1.0% by volume for the twin 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

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 and Electrical Safety Requirements.
$\sim$	Alternating current
A	Amperes (current)
	Asphyxiation Hazard
EC REP	Authorized representative in the European community
	Biohazard
CE	CE Marking of Conformity
C S P US	cCSAus mark. Indicates electrical safety certification for Canada and USA.
REF	Catalog number

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 show the product is recyclable. The date code on the label or product indicates the date of manufacture.
O	China RoHS logo. The device does not contain toxic and hazardous substances or elements above the maximum concentration values and the device is an environmentally-friendly product that can be recycled and reused.
Ĩ	Consult instructions for use.
	Crushing Hazard
C BATH American US	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

Symbol	Description
뤔	Ethernet connection
	Explosion Hazard
	Eye Injury Hazard
	Fire Hazard
	Flammable Chemical Hazard
Ţ	Fragile
-	Fuse
Hz	Hertz
<u>۸</u>	International safety symbol
<u>_</u>	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
IVD	In Vitro Diagnostic Device

Symbol	Description
	Ionizing Radiation Hazard
<u></u>	Keep dry.
Ţ.	Do not expose to rain.
	Relative humidity must not exceed 99%.
<u>     1 1     1     1     1     1 </u>	Keep upright.
	Lacerate/Sever Hazard
	Laser Radiation Hazard
	Lifting Hazard
	Magnetic Hazard
	Manufacturer
	Moving Parts Hazard
	Pacemaker Hazard. No access to people with pacemakers.

Symbol	Description
	Pinching Hazard
	Pressurized Gas Hazard
	Protective Earth (ground)
	Puncture Hazard
	Reactive Chemical Hazard
SN	Serial number
	Toxic Chemical Hazard
66 kPa	Transport and store the system within 66 kPa to 103 kPa.
75 kPa	Transport and store the system within 75 kPa to 101 kPa.
min% max%	Transport and store the system within the specified minimum ( <b>min</b> ) and maximum ( <b>max</b> ) levels of relative humidity, noncondensing.
_30 -30 -45	Transport and store the system within –30 °C to +45 °C.

Symbol	Description
-30°C	Transport and store the system within –30 °C to +60 °C.
•-€-> ss-€->	USB 2.0 connection
ss-(♣	USB 3.0 connection
	Ultraviolet Radiation Hazard
UK CA	United Kingdom Conformity Assessment Mark
UKRP	United Kingdom Responsible Person
VA	Volt Ampere (apparent power)
V	Volts (voltage)
	WEEE. Do not dispose of equipment as unsorted municipal waste. Environmental Hazard
W	Watts (power)
M	<i>yyyy-mm-dd</i> Date of manufacture

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To find hardware product documentation, refer to the documentation that comes with the system or component.

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