

MALDI-8020 and 8030 Mass Spectrometer




User Guide

Research use only



Health and safety precautions

For your safety and the safe operation of the instrument, read the following warnings and cautions. Warnings highlight situations that could result in serious injury or death.

	<p>Do not remove any panels from the instrument as it can produce lethal voltages.</p> <p>Do not modify the instrument.</p> <p>Keep liquids and flammable vapours away from the instrument.</p>
	<p>The power supply must be suitable for that on the rating plate (on the rear mains panel).</p> <p>The power supply to the instrument must be provided from an earthed socket outlet.</p> <p>Do not stretch, twist or coil the mains power cable.</p>
	<p>The instrument is a Class 1 Laser product which complies with the requirements of 21CFR1040.10 and 1040.11 except for deviations pursuant to Laser Notice No. 50, dated June 24, 2007, and IEC 60825-1 Ed.3 (2014).</p> <p>Do not remove any panels from the instrument - this may result in laser light exposure greater than Class 1.</p> <p>Use of controls or adjustments or performance of procedures other than those specified herein may result in hazardous radiation exposure.</p>

Chinese RoHS statement

信息提供清单


This information is applicable for People's Republic of China only.

部件名称	有害物质					
	铅(Pb)	汞(Hg)	镉(Cd)	六价铬(Cr(VI))	多溴联苯(PBB)	多溴二苯醚(PBDE)
金属框架, 机壳	×	○	○	×	○	○
电路板	×	×	×	×	×	×
线束·联接电缆	×	×	×	×	×	×
树脂框架与树脂盖之类	×	○	×	×	×	×
电源组件	×	×	×	×	×	×
电气部件	×	×	×	×	×	×
机械驱动机构	×	○	×	×	○	○
电容器	×	○	○	×	○	○
高压发生器	×	×	×	×	×	×
探测器	×	×	×	×	×	×
监视器	×	×	×	×	×	×

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×：表示该有害物质至少在该部件的某一均质材料中的含量超出 GB/T26572 规定的限量要求。

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About this guide

Welcome to the MALDI-8020 and MALDI-8030 user guide. This guide contains all the information you need to understand, maintain and operate these instruments, as well as detailed instructions on using the included application software.

These instruments (and also this guide) are intended for laboratory use by professional users who are trained to use the instrument and familiar with good laboratory practices. As a minimum, this guide assumes you are familiar with the following:

- Standard laboratory procedures and precautions;
- Sample preparation;
- Using PCs and the Windows operating system.

Training courses are available including sample preparation and using the instrument. Contact your local Shimadzu office or distributor for further information.

Notes

- The illustrations provided in the guide are intended as examples only, and must not be interpreted as actual representation of data, results or equipment.
- Some of the software applications described in this guide are optional components which you can only use if you selected these options at time of purchase. For example, *AuraSolution*, *SampleStation* and *QC Reporter* are optional modules.
- While we always try to ensure that the content of our publications is accurate, we know that mistakes can be made. In a continuous effort to improve, we welcome feedback on the accuracy and usability of this guide. Please submit all feedback via the **Email comments** link or directly to Tech.Pubs@kratos.co.uk.

Change history

Issue A (Dec 2020)	All	First issue.
Issue B (Oct 2021)	Update to weight reference on page 19.	-
Issue C (Nov 2021)	Update to the procedure for switching the PC and instrument on.	

Additional information

The MALDI-8020 and MALDI-8030 are manufactured by:

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Kratos Analytical Limited is part of the Shimadzu Corporation, Japan.

Contact details

For servicing and repairs, contact your local Shimadzu office, or distributor.

Disclaimer and copyright

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Trademarks and acknowledgements

MALDI Solutions, MALDI-8020 and MALDI-8030 are trademarks of Kratos Analytical Limited and Shimadzu Corporation, Japan.

Windows is a trademark of Microsoft, Inc.

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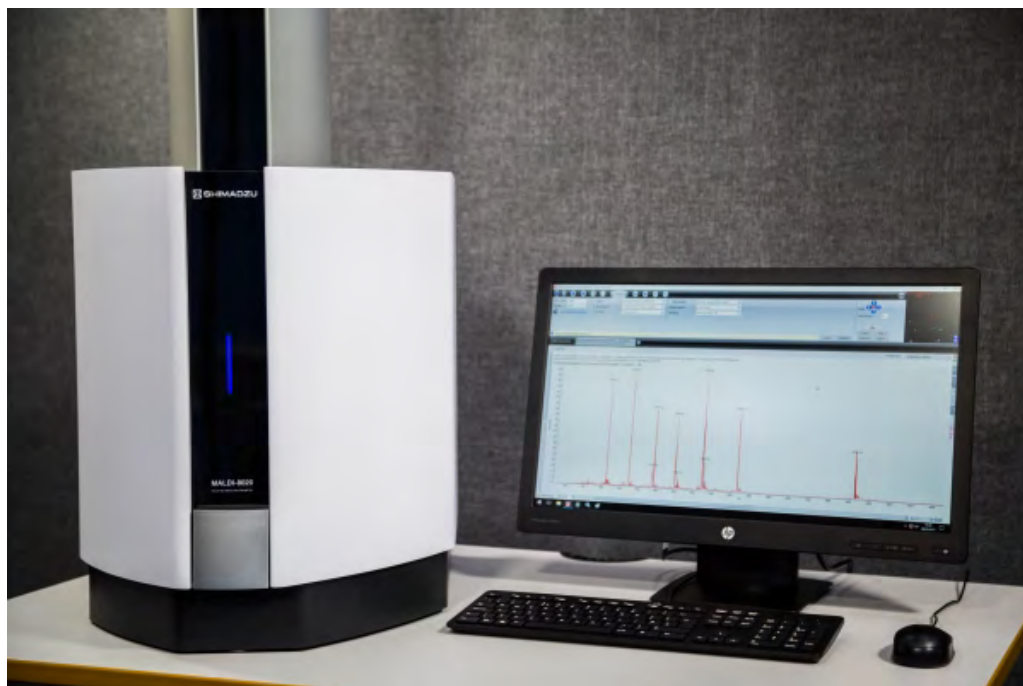
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Introduction

The MALDI-8020 and MALDI-8030 instruments are benchtop-sized, linear MALDI-TOF mass spectrometers which operate in conjunction with the supplied application software to provide high-performance rapid mass measurement of samples. The instruments are typically used for research, quality control and analysis of proteins & peptides, as well as other biological and organic samples.

The MALDI-8030 provides a dual polarity ion source which enables users to perform linear MALDI-TOF analysis in either positive or negative ion mode.



Key features of the instruments




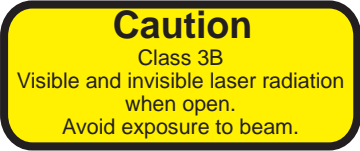
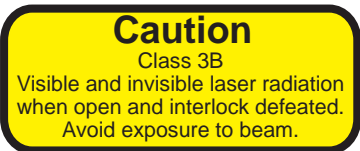

- Linear mode (positive-ion) MALDI-TOF;
- For MALDI-8030 instruments only - Linear mode (negative-ion) MALDI-TOF;
- 200 Hz solid-state laser, 355 nm;
- Load-lock chamber for fast sample introduction;
- UV laser-based source cleaning (patented);
- Small footprint/benchtop design;
- Quiet operation (<55 dB).

Instrument labels

Internal labels




The following label symbols are used within the instrument.

Note: As there is a potential personal injury hazard, access to the inside of the instrument is only allowed by authorised service engineers.

Label	Meaning
	Safety alert symbol - indicates a general potential personal injury hazard.
	Electric shock symbol - indicates a specific potential personal injury hazard.
	Laser hazard symbol - indicates a specific potential personal injury hazard.
	Class 3B invisible and visible radiation - indicates classification of the laser which is enclosed by a protective housing within the instrument. Text on the label reads "Caution: Class 3B invisible and visible laser radiation when open. Avoid exposure to beam."
	Class 3B invisible and visible radiation - indicates classification of the laser which is enclosed by a protective housing within the instrument. Text on the label reads "Caution: Class 3B invisible and visible laser radiation when open and interlock defeated. Avoid exposure to beam."
	Caution moving parts - indicates hazard from exposed lead-screws and moving motors.

Rear mains panel labels

The following label symbols are used on the rear mains panel:

Label	Meaning
	Safety alert symbol - indicates a general potential personal injury hazard.
	Electric shock symbol - indicates a specific potential personal injury hazard.
	Class 1 laser product - indicates that the instrument is safe under all conditions of normal use.

Layout & connections

Layout - front



The target plate door allows you to load and unload target plates from the instrument (see [Loading and unloading target plates](#)).

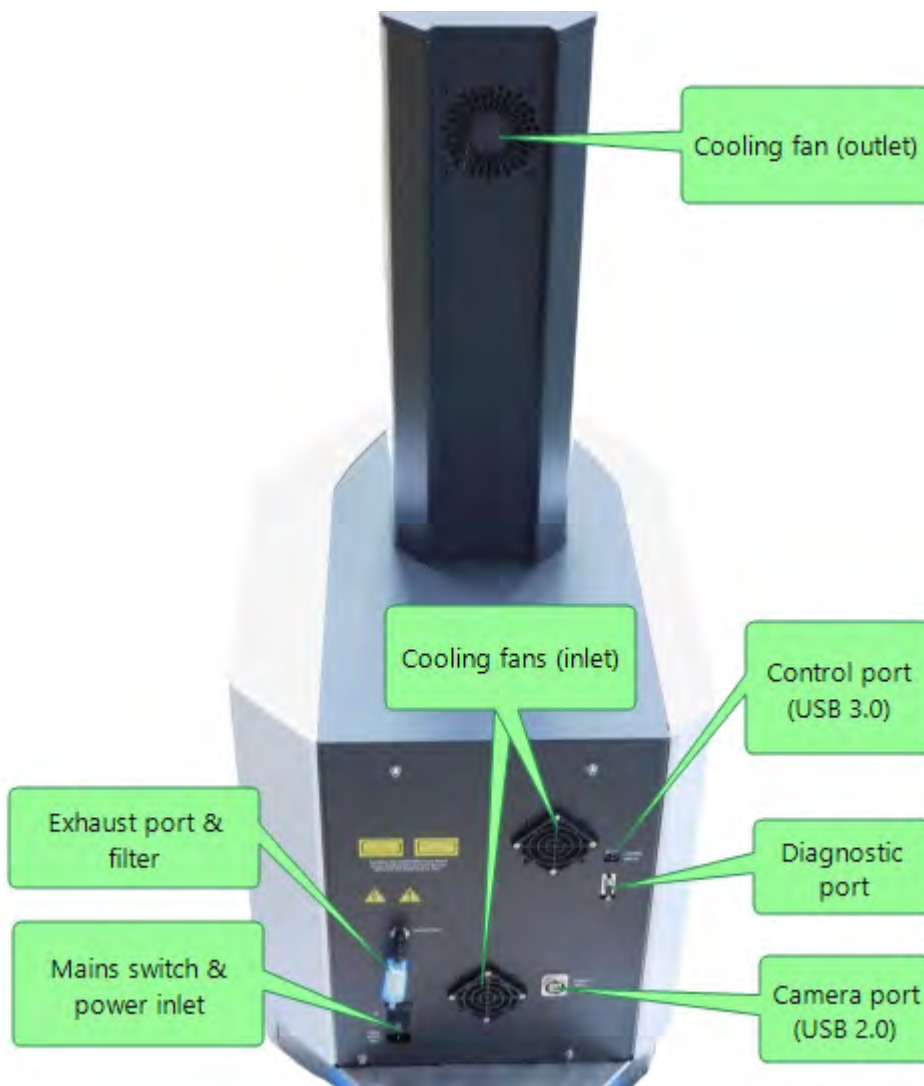
Front Panel LEDs

The front panel LED can also be used to determine the operating status of the instrument as follows:

Colour	Status	Function
Pink	Blinking	Instrument is starting up after reboot
Orange	Blinking	Instrument is venting or pumping down
Green	Blinking	Door is open, instrument ready to load target plate
Green	Steady	Target plate bar code recognised
Cyan	Steady	Instrument is in Standby mode
Cyan	Blinking	High voltage ramping in progress
Blue	Steady	Instrument is in positive ion Operate mode
Blue	Blinking	Instrument is acquiring data from the sample in positive ion mode
White	Steady	Instrument is in negative ion Operate mode (MALDI-8030 only)
White	Blinking	Instrument is acquiring data from the sample in negative ion mode (MALDI-8030 only)
Red	Blinking	Instrument fault
Red	Steady	USB communications error
Yellow	Blinking	Laser cleaning in progress

For more in-depth status information on the instrument, see [Instrument status](#).

Layout - rear

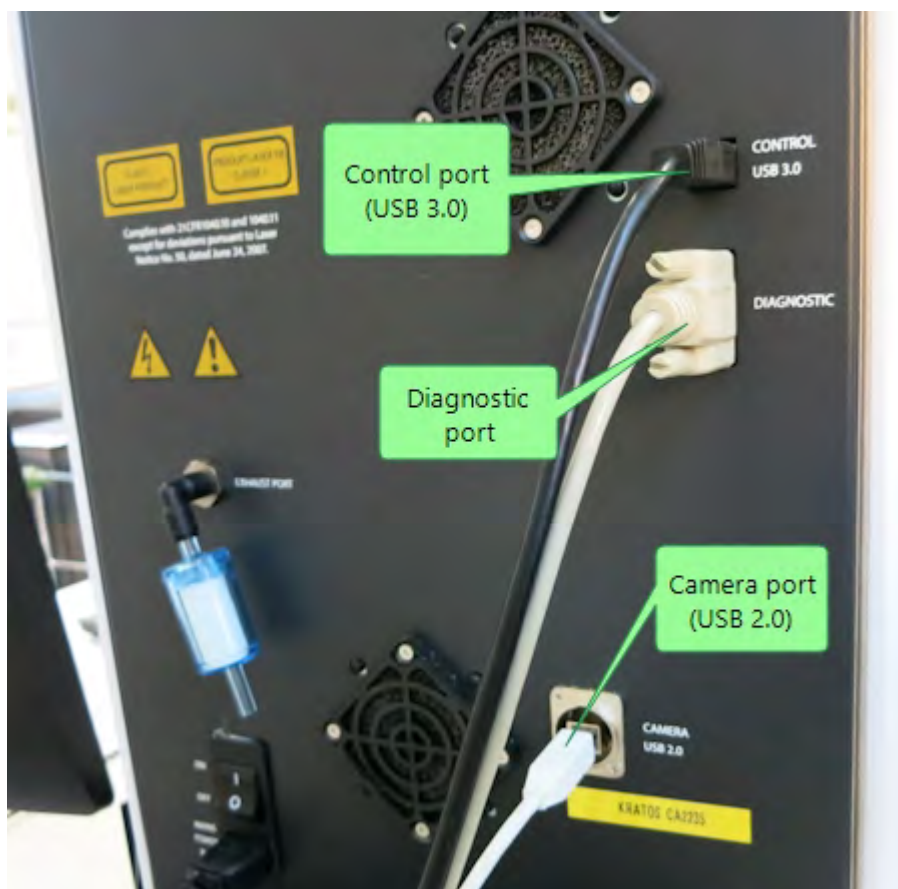


Always ensure that there is clear access to either the rear panel mains switch or to the mains plug (where it is plugged into the wall socket), to allow the instrument power to be easily disconnected from the mains power at any time.

Mains power leads

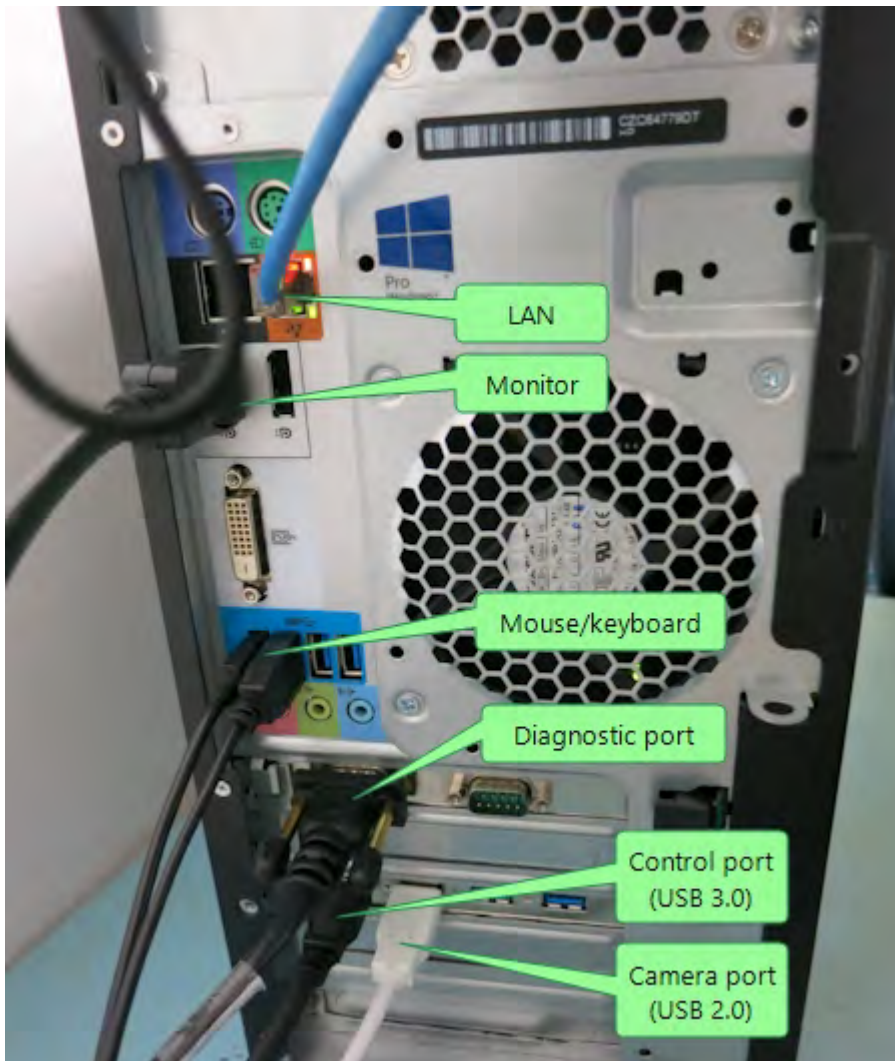
The instrument is supplied with power leads for the instrument, PC and monitor. If alternative leads are used, they must be suitable for the power rating of the equipment.

Connections (instrument side)

**Notes:**

- The control port is used for communications between the instrument and the PC workstation;
- The diagnostic port is used by service engineers to assist servicing and fault diagnosis;
- The camera port is used to feed the video image from the internal camera back to the PC workstation.

Connections (PC workstation side)

**Notes:**

- The control port and camera port connections are made to the additional USB ports on the rear of the PC workstation, not the built-in ports;
- The diagnostic port is connected to the additional serial port connections on the rear of the PC workstation, not the built-in port.

Switching on/off

Switching on

When switching on, follow the procedure as specified below.

1. Switch on the instrument PC and monitor.
2. Before the Windows log in screen displays, set the mains power switch at the back of the instrument to the ON (I) position, and then wait while the instrument powers up and pumps down the vacuum system to the required operating pressure.




3. When the front panel LED blinks **Green**, the instrument is ready to use.

For more information on the LEDs on the front panel, see [Front panel LED indicators](#).

Trouble Shooting

In some instances a connection between instrument and PC is unsuccessful. The table below shows symptoms of the communication error and possible solutions:

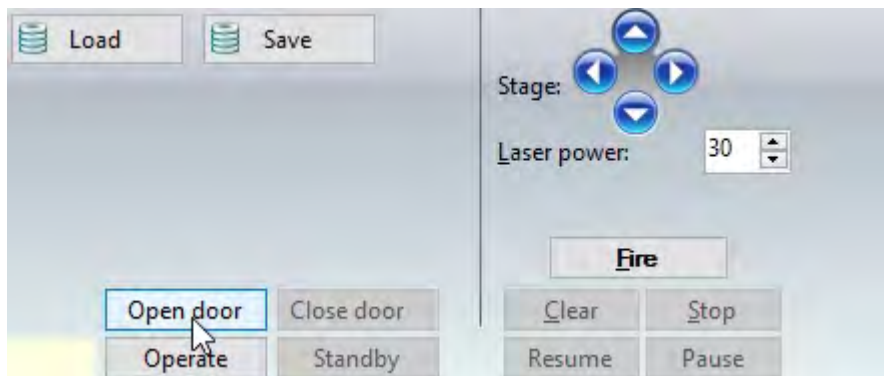
Symptom	Possible Cause/Solution
Instrument front light is red	Communication between PC and instrument unsuccessful. Repeat Steps 1 and 2.
USB Drives not recognised	Instrument switched on before PC. Check Windows Device Manager. Switch instrument off and back on or switch off both PC and Instrument repeating steps 1 and 2.
Services not started	<p>Communication between PC and instrument unsuccessful. Navigate to Control and Status services and try restarting the following services manually:</p> <ul style="list-style-type: none"> • Shimadzu Corporation Automation Service • Shimadzu Corporation Instrument Control Service • Shimadzu Corporation Instrument Status Service <p>To access the Control and Status Services application, click the Windows key  type service.msc and press return.</p>

Switching off

We recommend that the instrument is left switched ON. If it is switched off for an extended period and then switched on, it may take several hours for the instrument to reach its operating vacuum pressure.

If you do need to switch off, follow the steps below in sequence. Doing so ensures that the plate carrier within the instrument is 'parked' safely.

1. In *MALDI Solutions Data Acquisition*, select the **Acquire** tab and then click **Open door**.



2. Wait until the front panel LED blinks **green**.
3. If required, remove any loaded target plate from the instrument then physically shut the door without clicking **Confirm close door** from the pop-up list of values (see [Loading and unloading target plates](#)).
4. If required, close down all MALDI Solutions applications and switch off the PC workstation.
5. Set the mains power switch at the back of the instrument to the OFF (O) position

Mains power failure

If the power supply to the instrument fails, or is switched off, the vacuum system will automatically start when the power is restored.

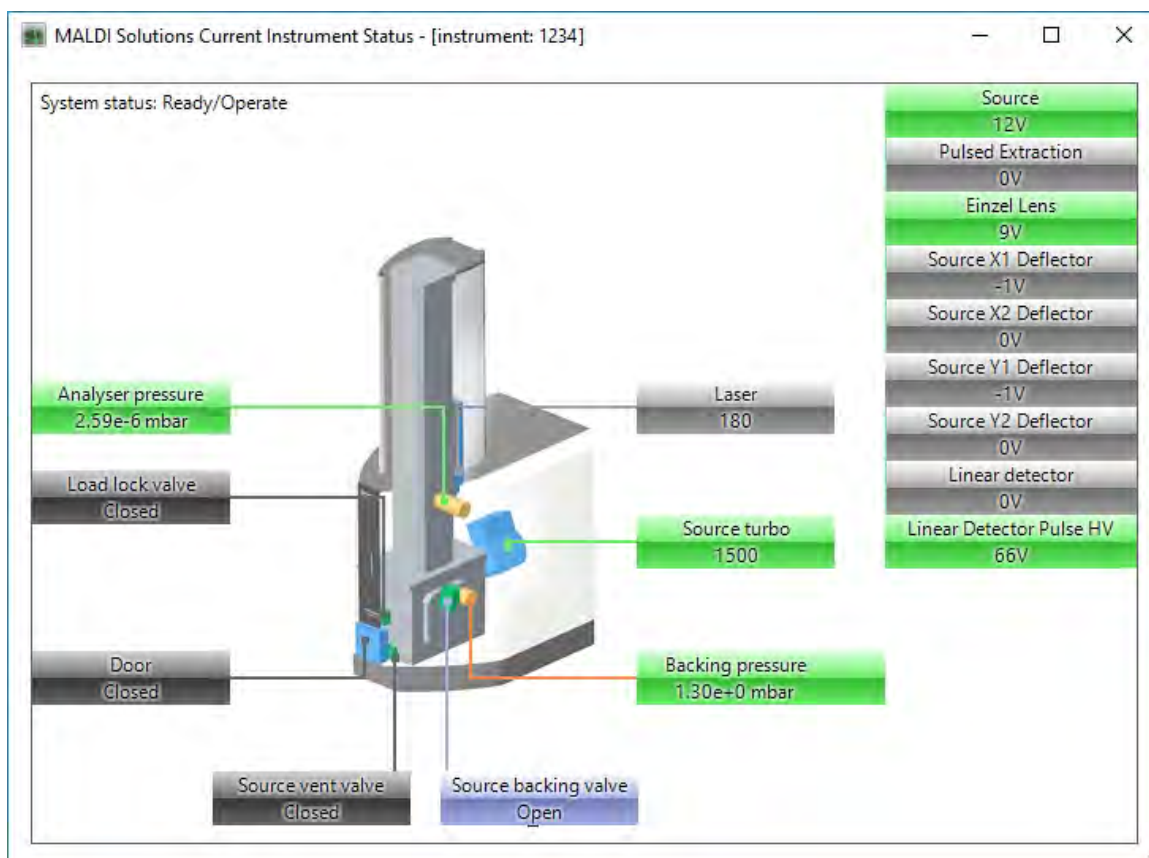
Instrument status

In addition to the front panel LED indicator, more detailed information on the current instrument status can be displayed using the *MALDI Solutions Current Instrument Status* software application. This shows you a graphical representation of the instrument, with colour-coded annotations providing a visual representation of the status of the major components.

As you use the instrument, the viewer reflects those changes, for example, if you open the door.

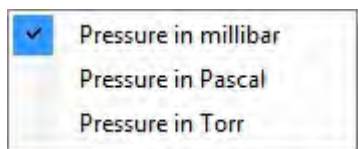
Starting the application

1. On the **Start** menu, locate and select **Shimadzu Corporation > Current Instrument Status**.



Changing the units of pressure

1. Click the right mouse button anywhere in the viewer.



2. From the pop-up menu, select the required unit.

Status

Pressure readings

Displays the current operating pressure of the source (analyser) chamber and backing line.

Colour	Description
Green	At required pressure
Amber	Not at required pressure - progress bar indicates progress to required pressure

Valves/door

Displays the mechanical status of the valves and target plate door.

Colour	Description
Blue	Open
Black	Closed

Laser

Displays the current operating power of the laser in arbitrary units (0-180). This indicator does not change colour.

Source turbo

Displays the current operating speed of the turbo pump (rpm).

Colour	Description
Grey	Turbo is off
Yellow	Turbo is accelerating
Green	Turbo is at full speed
Red	Turbo has failed

Components

Displays the current voltage applied to each component.

Colour	Description
Grey	Component is off
Green	Component is on
Red	Component has failed

Instrument life cycle

Storage

The storage requirements for the instrument are:




- Storage temperatures: Min -20°C (-4°F); Max +60°C (140°F);
- Maximum humidity: 70% relative humidity (non condensing).

Delivery and installation


The instrument is delivered in suitable packaging that includes installation instructions and lifting straps.

Note: Only Shimadzu or Kratos accredited engineers should unpack or install the instrument.

Site requirements


	The MALDI-8020 instrument weighs approximately 86 kg (190 lbs) and the MALDI-8030 weighs approximately 92 kg (202 pounds); use suitable mechanical handling equipment to lift the instrument. Check that the new location can support the weight of the instrument.
	Always ensure that there is clear access to either the rear panel mains switch or to the mains plug (where it is plugged into the wall socket) to allow the instrument power to be easily disconnected at any time.
	Two inlet fans and an outlet fan cool the instrument. Do not obstruct the fans.

Servicing

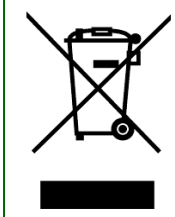
	Only Shimadzu or Kratos accredited service engineers are permitted to service components within the instrument.
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We recommend that the instrument is serviced at least once a year, depending on usage and samples used. After the one-year warranty period, service contracts are available to provide the appropriate level of support. Contact your local Shimadzu office, or distributor, for details.

Moving the instrument

	Do not move the instrument while the turbo pump is running. Switch off and allow the pump to slow down for at least 30 minutes.
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Disposal



The instrument is marked with the adjacent symbol. This means that at the end of its useful life, it must not be disposed of with general household waste. When the instrument has reached the end of its life, contact your local Shimadzu office, or distributor, for details of its disposal.

Specifications

Instrument specifications

Dimensions (including stabiliser)	600 mm (width) x 745 mm (depth) x 1,055 mm (height). (23.6" (width) x 29.5" (depth) x 41.5" (height))
Weight	MALDI-8020: Approximately 86 kg (190 pounds) MALDI-8030: Approximately 92 kg (202 pounds)
Power supply	A clean, stable and continuous mains supply is required; 100 to 240 Vac, 50/60 Hz, 1,000 VA single phase
Electrical insulation	The instrument is classed as an Electrical Installation Category II, pollution degree 2, according to BS EN 61010-1:2012
Operating temperature	15°C (59°F) to 32°C (90°F)
Performance temperature	18°C (64°F) to 28°C (82°F)
Relative humidity	Less than 70% non condensing at 22°C (72°F)
Sound level	<55 dB(A)
Maximum altitude	2,000 m (6,562 feet)
Location	Indoor use only

PC specification

The PC workstation used to control the instrument must comply with IEC 60950 IT equipment (the instrument's camera is powered via the USB cable). This is a regulatory requirement to meet UL (Underwriters Laboratories) approval.

Laser specifications

Wavelength	355 nm
Beam divergence	20 mrad (full angle).
Pulse width	1.5 ns
Repetition rate	50, 100, 200 Hz
Nominal power	80 µJ/pulse

Performance specifications

The performance of the instrument may vary slightly depending on the type of target plate you are using. The following specifications were obtained using FlexiMass-SR48 steel targets.

Mass range	1 to 350,000 Da and 10,000 to 500,000 Da
Mass resolution	> 5,000 FWHM (full width half-height maximum) on ACTH 18-39 (2,465 Da)
Accuracy	<20 ppm with internal calibration <150 ppm with external calibration (nearest neighbour external calibration on Fleximass-SR48 steel target, within 30 minutes)
Sensitivity	250 fmol on bovine serum albumin (on-target) 250 amol on Glu-1-Fibrinopeptide B (on-target)

Compliance

- The instrument conforms to the relevant European regulation for electrical safety and electromagnetic compatibility (EMC).
- The instrument complies with the emissions and immunity requirements of IEC 61326.

Chapter 2 - Working with samples

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Introduction

Samples are introduced into the instrument using target plates which can be 'spotted' with a range of different samples up to the maximum the plate allows. Target plates are inserted and removed via the sample door on the front of the instrument.



Listed below are the two types of target plate supported on the MALDI-8020 and MALDI-8030 instruments.

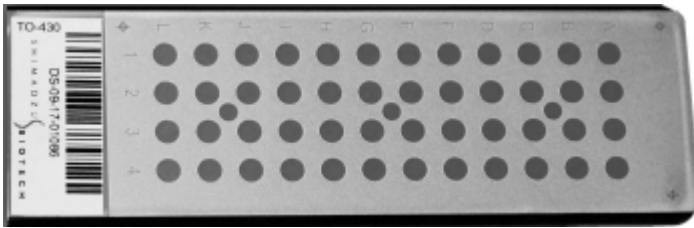
FlexiMass-DS disposable targets

These polymeric target plates are disposable (single use) alternatives to stainless steel re-usable plates. They can be used as supplied and do not require cleaning prior to use. Each plate has a unique bar code which allows its usage to be tracked and recorded. The unique bar code can be read directly by the barcode reader integrated in the door of the instrument.

Note: After using this type of target plate, it is strongly recommended that you do not attempt to clean or otherwise re-use them. Doing so will compromise the surface characteristics and performance of the plate and may damage the instrument.

Please dispose of used plates in accordance with local, state and federal regulations.

Number of sample positions:	Sample = 48 Calibrant = 3
Internal diameter of sample positions:	Sample = 2.8 mm Calibrant = 1.5 mm
Order number:	TO-430R00 (four packs of four targets).



FlexiMass-SR48 steel targets

These stainless steel target plates are durable and designed to be re-used after cleaning (see [Cleaning target plates](#)).

Number of sample positions:	Sample = 48 Calibrant = 3
Internal diameter of sample positions:	Sample = 2.8 mm Calibrant = 2.2 mm
Order number:	TO-431R00 (pack of four targets).



Preparing samples

The procedures in this section are examples of typical sample preparation procedures, and should be used for guidance only. Adapt them to suit your requirements.

Notes:

- Ensure personnel are fully trained in the handling of chemicals and use of laboratory equipment;
- Use standard laboratory precautions when preparing samples, for example, in a wet laboratory or designated preparation area;
- Ensure that samples are not prepared within the vicinity of the instrument;
- Always wear powder-free chemically resistant gloves, lab coat and protective goggles when handling target plates.
- Ensure samples are thoroughly dry on the target plate before loading the plate into the instrument.

Serial dilutions of samples

The procedures below assume that you have low concentration starting samples. However, if you have a high concentration starting sample, you may wish to make 100 fold or greater dilutions, especially if you require fmol/ μL concentrations. Adapt the procedures below to suit.

Make 10-fold dilutions from stock concentrations ($10^{-3} \text{ M} = 1,000 \text{ pmol}/\mu\text{L}$) :

1. Add 90 μL of 0.1% trifluoroacetic acid to a 0.5 ml micro-centrifuge tube, or similar, using a pipette.
2. Take 10 μL of the stock solution (1,000 pmol/ μL) using a pipette. Equilibrate pipette tip by filling and emptying 3 times. Add to the micro-centrifuge tube, mix up and down 5 times. The result is a concentration of 100 pmol/ μL .
3. Vortex the mix.
4. Repeat this procedure until you reach the required concentration, using a new pipette tip at each stage. Typical concentrations for peptides/proteins are 10 and 1 pmol/ μL .

Where a 5 pmol/ μL or 500 fmol/ μL concentration is required, make 20-fold dilutions. For example, take 95 μL of 0.1% trifluoroacetic acid and to this add 5 μL of sample (e.g. 100 pmol/ μL) to give a concentration of 5 pmol/ μL .

Calibration samples

A calibration mix must cover the mass range of interest. Typically, a calibration would contain evenly spaced calibrants across the mass range of interest, using a minimum of three calibrants.

Calibrants (amino acid sequence)	Calculated Monoisotopic Mass	
	(M+H)+	*(M-H)-
Angiotensin 2 human (DRVYIHPF)	1,046.54 Da	1044.54 Da
Angiotensin 1 human (DRVYIHPFHL)	1,296.69 Da	1294.69 Da
ProteoMass™ P14R MALDI-MS standard (PPPPPPPPPPPPPPR)	1,533.86 Da	1531.86 Da
[Glu1]-Fibrinopeptide B human (EGVNDNEEGFFSAR)	1,570.68 Da	1568.68 Da
N-Acetyl-Renin Substrate Tetradecapeptide porcine (C2H2O-DRVYIHPFLLVYS)	1,800.94 Da	1798.94 Da
ACTH fragment 1-17 human (SYSMEHFRWGKPVGKKR)	2,093.09 Da	2091.09 Da
ACTH fragment 18-39 human (RPVKVYPNGAEDESAAEAFPLEF)	2,465.20 Da	2463.20 Da
Adrenocorticotrophic Hormone Fragment 7-38 human (FRWGKPVGKKRRPVKYPNGAEDESAAEAFPLE)	3,657.93 Da	3655.93 Da
	Calculated Average Mass	
	(M+H)+	*(M-H)-
Bovine serum albumin	66,430.20 Da	66,428.20 Da

***NOTE:** (MALDI-8030 only) In negative ion mode analysis, the corresponding (M-H)- mass is used.

Peptide/protein samples

Preparation

1. Use 10 pmol/μL concentrations.
2. Take 100 μL of each peptide/protein into a micro-centrifuge tube, or similar.
3. Vortex the mix.

For calibrants, you may need to adjust the concentrations in the mix to produce a spectrum of similar intensity peaks, i.e. the monoisotopic peaks are of similar height.

Spotting peptides - general

Add matrix and sample to the wells using a pipette.

1. Spot 0.5 μL of sample.
2. Add 0.5 μL matrix of α-cyano 4-hydroxycinnamic acid 5 mg/mL in acetonitrile/0.1% trifluoroacetic acid (1:1).

3. Leave the spotted target plate for several minutes at room temperature for the samples to dry.
4. Check that the samples are visibly dry before inserting the target plate into the instrument.

Spotting peptides - low concentrations

For very low concentrations, pre-coat the required wells.

1. Spot 0.5 μL of matrix, leave for few seconds, remove the surplus using a pipette and leave to dry.
2. Spot 0.5 μL of sample.
3. Add 0.5 μL matrix of a-cyano 4-hydroxycinnamic acid 5 mg/mL in acetonitrile/0.1% trifluoroacetic acid (1:1).
4. Leave the spotted target plate for several minutes at room temperature for the samples to dry.
5. Check that the samples are visibly dry before inserting the target plate into the instrument.

Spotting proteins

1. Spot 0.5 μL matrix of sinapinic acid, saturated solution (20 mg/mL) in acetonitrile/0.1% trifluoroacetic acid (1:1).
2. Add 0.5 μL of sample.
3. Allow to dry.

Glycan samples

The procedures in this section are for guidance only. Adapt to suit your requirements.

1. Spot 0.5 μL matrix of 2,5-dihydroxybenzoic acid (DHB) at 10 mg/mL in acetonitrile/0.1% trifluoroacetic acid (1:1).
2. Add 0.5 μL of sample.
3. Allow to dry.
4. Optionally, pipette 0.3 μL of chilled ethanol onto the dried spot and allow to dry.

For Negative mode analysis (MALDI-8030 only)

In negative mode analysis, generally a similar procedure of sample preparation and spotting to above is used except 0.1% trifluoroacetic acid is replaced with water.

Sample records

Keep detailed records of all substances which have been used in this instrument. Service engineers are not allowed to service instruments contaminated with any substance which is radioactive or biologically active unless thorough decontamination has first been performed.

Cleaning target plates

Follow the procedures below for cleaning of FlexiMass-SR48 steel targets ONLY. Do not attempt to clean the single-use polymeric FlexiMass-DS targets. Doing so will compromise the characteristics and performance of the product and may ultimately lead to damage of the instrument.

Shimadzu accepts no liability for any damage caused by improper handling of FlexiMass-DS targets.

The following instructions can also be found at: <http://shimadzu.com/an/lifescience/maldi/support.html>

Basic cleaning

- Wipe off any fibres or dust particles using a dry lint-free cloth;
- Remove any finger marks or other light oil/grease marks using a lint-free cloth moistened with HPLC grade acetone.

Intensive cleaning

Notes:

- For targets intended for lipid or low molecular weight (<800Da) analyses, omit all steps that include Micro 90 solution;
- When handling solvents, wear suitable personal protective equipment (e.g. safety glasses, lab coat and gloves);
- Refer to manufacturer's MSDS (Material Safety Data Sheet) for instructions on safe handling and disposal.

Items required

- Ultrasonic bath for target cleaning;
- Cleaning solution concentrate: Micro 90 (available from Sigma Aldrich) - Micro 90 steps are optional, but useful for heavy contamination;
- Hotplate or oven capable of heating a 5% Micro 90 solution to approximately 60°C (140°F);
- HPLC grade methanol and acetone;
- Deionised or HPLC grade water;
- Small oven for drying targets, up to 50°C (122°F).

Cleaning procedure

1. Prepare 100 mL of a 5% (v/v) solution of Micro 90 in deionised or HPLC grade water (5% (v/v) = 5 mL of Micro 90 concentrate + 95 mL water).
2. Heat the 5% Micro 90 solution to approximately 60°C (140°F).
3. Using a tissue and a suitable solvent (e.g. methanol), remove any visible signs of samples from the surface of the target.
4. Place the target face up into a small container and immerse (cover) with acetone.
5. Cover the container and place into an ultrasonic bath.
6. Sonicate the target in acetone for 15 minutes at room temperature.

7. Discard the used acetone. Rinse the target (x2) with 100 mL deionised or HPLC grade water. Discard rinsing solutions.
8. Place the target face up into a small container and immerse (cover) with the heated 5% (v/v) Micro 90 solution.
9. Cover the container and place into an ultrasonic bath.
10. Sonicate the target in the 5% Micro 90 cleaning solution for 15 minutes at room temperature.
11. Discard the used 5% Micro 90 solution. Rinse the target (x2) with 100 mL deionised or HPLC grade water. Discard rinsing solutions.
12. Cover the target with approximately 100 mL of deionised or HPLC grade water. Sonicate target for 5 minutes at room temperature.
13. Cover the target with approximately 100 mL of methanol. Sonicate target for 15 minutes at room temperature. Discard the rinsing solution.
14. Cover the target with approximately 100 mL of deionised or HPLC grade water. Sonicate target for 15 minutes at room temperature. Discard the rinsing solution.
15. Shake the target to remove excess water from the rinsing step.
16. Flood the target with methanol and allow excess to evaporate.
17. Place the target in an oven at 50°C (122°F) for at least 6 hours (e.g. overnight). This oven drying step is necessary to minimise unwanted sample spreading during use.
18. Prior to use, allow the target to adjust to room temperature.

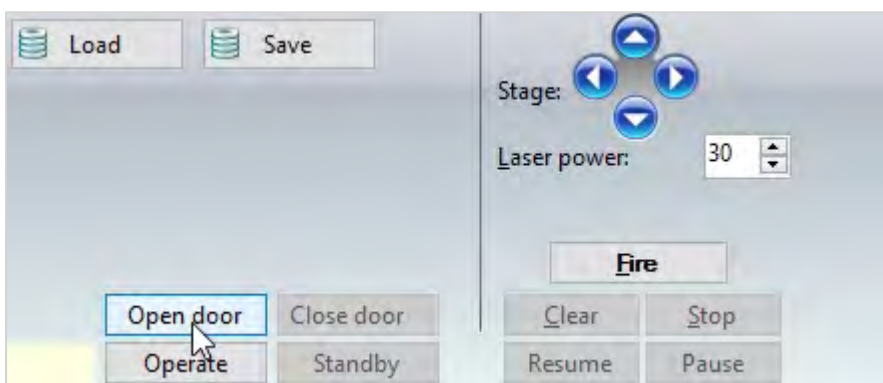
Loading and unloading target plates

When loading and unloading target plates, observe the following precautions:

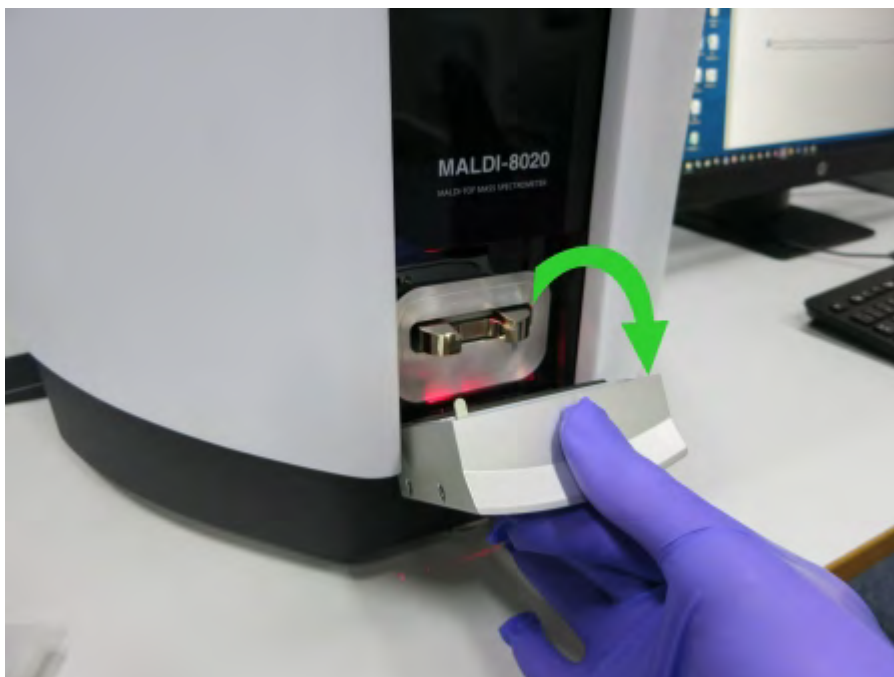
- All samples must be prepared according to standard laboratory techniques (see [Preparing samples](#)).
- Use powder-free gloves while handling target plates.
- Avoid touching the spotted areas of the plate.
- Do not put wet samples or any other element such as fibre into the instrument. Moisture or other particles within the vacuum system may damage the instrument.

Loading a target plate

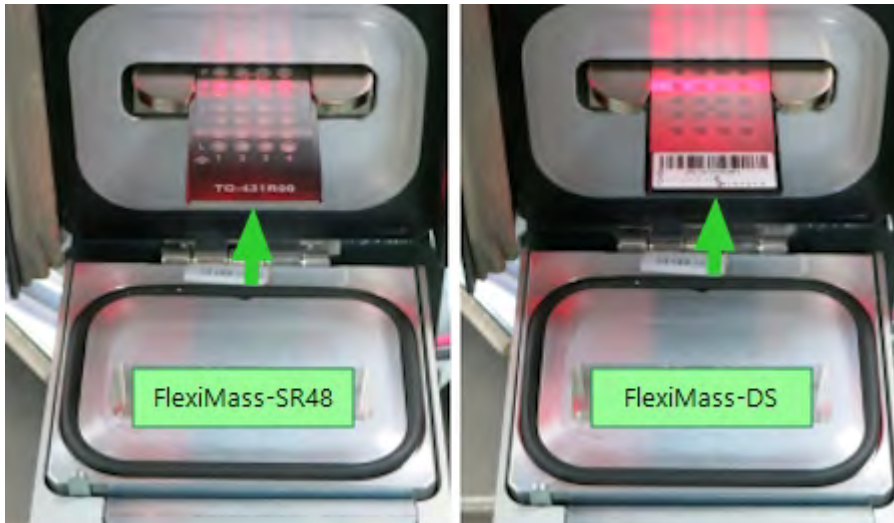
1. In *MALDI Solutions Data Acquisition*, select the **Acquire** tab and then click **Open Door**.



2. When the front panel LED blinks **green**, pull back gently from the top of the door to open.



- Slide a target plate into the slot using the orientation shown below, pressing firmly on the front edge of the plate until it can go no further.

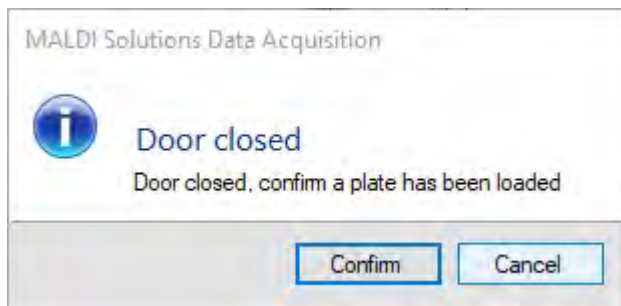


- If any dust or debris is visible on the inner surface of the door, gently remove it with a dry lint-free cloth.
- Close the front door by lifting upwards and applying gentle pressure until the door latch clicks into place.



Note: The front door mechanism automatically seals the sample chamber when the door is closed. However, when the door is closed, the door cover can move by several millimetres; this is normal and does not affect the performance of the instrument.

- Wait until the following dialog displays and click **Confirm**.



Note: At this stage you can click **Cancel** to abort the loading operation and load a fresh plate. If you click **Cancel** and then decide to proceed with loading the plate you originally inserted, click **Close door** on the **Acquire** tab to manually confirm loading.

- Once the target plate is loaded, proceed with the instructions below for aligning the plate before use.

Aligning a target plate

This procedure may be required to account for small differences in the plate dimensions, to ensure that the camera crosshairs are correctly aligned with the wells on the plate. Normally you will only need to do this once if changing plate type.

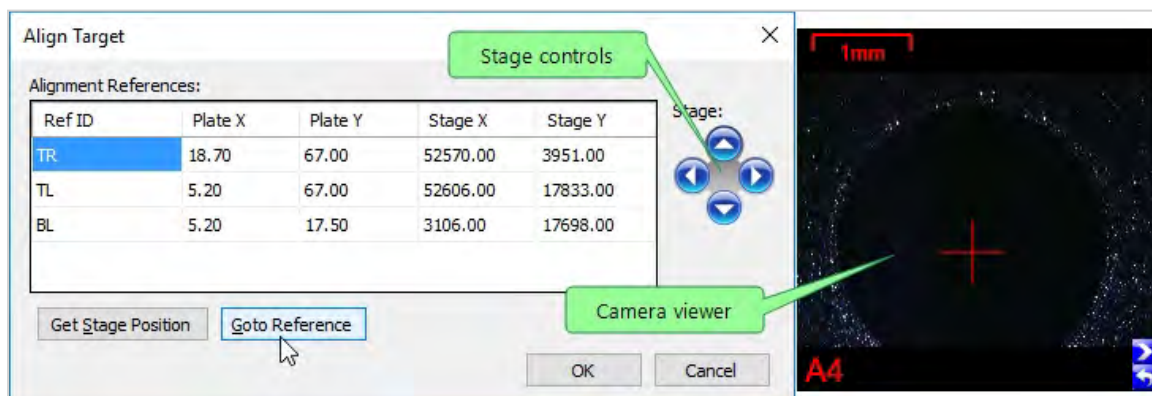
- In *MALDI Solutions Data Acquisition*, click the **Align Target** icon:



- The **Align Target** dialog displays the approximate reference sample positions for the currently loaded plate. The reference sample positions are enumerated as follows:

- TR = Top Right (corresponds to sample position A4)
- TL = Top Left (corresponds to sample position A1)
- BL - Bottom Left (corresponds to sample position L1)

- In the **Ref ID** column, select reference **TR** and then click **Goto Reference**. The sample stage will move so that position A4 is roughly centred in the camera viewer.



- Use the stage controls to make small adjustments to the crosshair position so that it is as close to the centre of the sample position as possible. You can use the following keyboard shortcuts to control the units of

movement as follows:

Shortcut	Movement unit
Click	0.05 mm
Alt+Click	0.01 mm
Shift+Click	0.1 mm
Ctrl+Click	0.15 mm
Shift+Ctrl+Click	0.25 mm

5. Once you are satisfied with the crosshair alignment, click **Get Stage Position** to record the reference point for the sample positions.
6. Repeat steps 3 through 5 for the TL and BL reference points.
7. Click **OK** to record all changes to the reference points for the loaded target plate.

Unloading a target plate

1. Follow steps 1 and 2 of the loading procedure above.
2. With the door open, grip the target plate by its front edge and then carefully withdraw it from the slot.

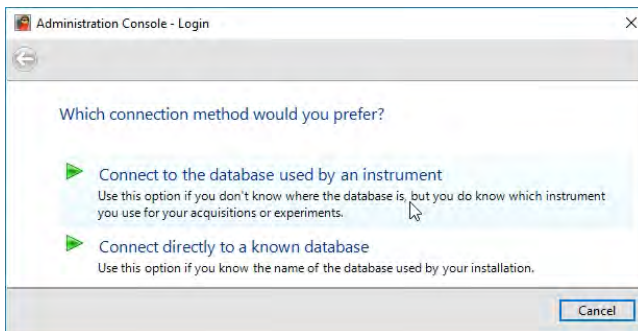
Note: If you do not intend to load another target plate immediately, close the door after unloading. You should not leave the door open for extended periods of time in order to avoid contamination of the inner surface.

Chapter 3 - Administration

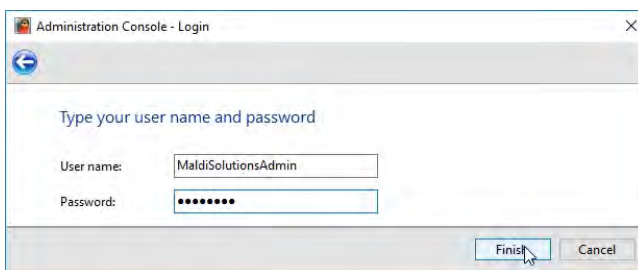
Introduction	37
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Starting the application

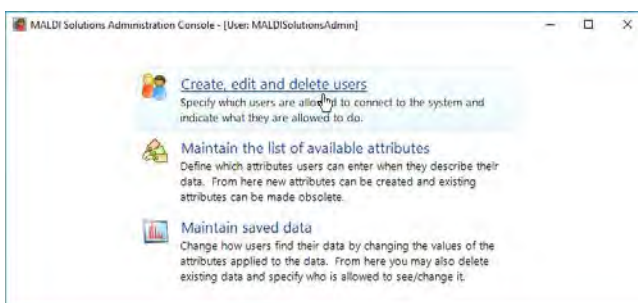
1. From the **Start** menu, locate and select **Shimadzu Corporation > Administration Console**.



2. If you wish to use the default instrument database, click **Connect to the database used by an instrument** and then proceed to step 4.
3. If you wish to connect to a remote database (i.e. on another instrument) click **Connect directly to a known database**, however, this option not currently supported.
4. Enter your **User name** and **Password**, and then click **Finish**.



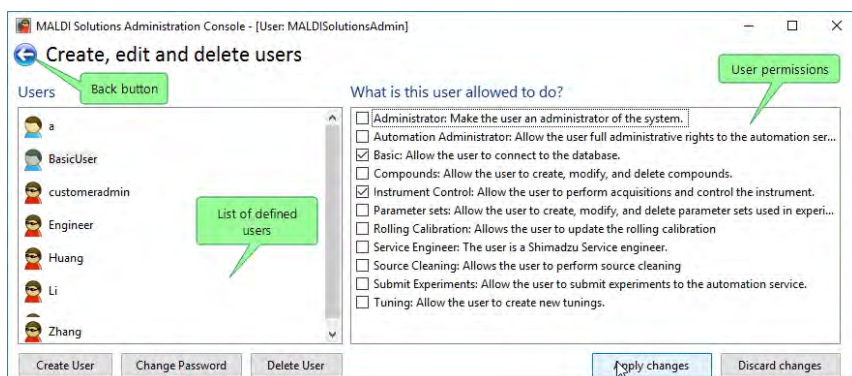
You will see the main menu of the application as shown below.



Note: Depending on your [user account privileges](#), you may not see all the options shown above.

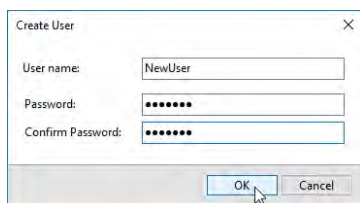
Managing users

1. In *MALDI Solutions Administration Console*, click **Create, edit and delete users**.



Creating a new user

1. Click the **Create User** button.



2. Enter the **User name** and **Password** for the new user.
3. Enter the same password you specified in step 2 in the **Confirm Password** box and then click **OK**.

The new user is added to the list with the **Basic** and **Instrument Control** permissions selected.

Modifying user permissions

1. In the list of defined users, select the entry for the user you wish to modify.
2. In the **What is this user allowed to do** pane, select or clear the permissions as required.

Notes:

- Do not select the **Service Engineer** or **Tuning** permissions - these are for service engineers only.
 - Selecting the **Administrator** permission will give the selected user access to all features of the Administration Console and should be assigned with caution. Users with administrator-level permissions are indicated by a red icon in the list of defined users.
3. Repeat steps 1 and 2 for any other users you wish to modify.
 4. Click **Apply changes** to save modified permissions for all user accounts, or **Discard changes** to revert to the previously saved permissions.
 5. Click the **Back** button to return to the main menu.

Deleting a user

1. In the list of defined users, select the entry for the user you wish to delete and then click **Delete user**.
2. When the confirmation dialog appears, click **Yes**.

Note: There must be at least one user in the list who has the **Administrator** permission assigned. The application will not allow you to delete all users with this permission assigned.

3. Repeat step 1 for any other users you wish to delete.

Managing attributes

Attributes are a form of metadata which allow entries in the instrument database to be categorised for search purposes. These are typically displayed during database operations, for example when [loading or saving acquisitions](#).

You can use the application to modify, create or make obsolete attributes which are associated with each type of data item in the instrument database. Obsolete attributes are no longer displayed in any application which uses the database, but are not removed from it.

Note: Once created, attributes cannot be deleted. However, if you erroneously create a new attribute for a data type, you can make it obsolete, in which case it will no longer be displayed when loading, saving or searching the instrument database.

Modifying attributes

1. In *MALDI Solutions Administration Console*, click **Maintain the list of available attributes**.
2. In the **Select the type of item to modify** drop-down list, select the category of data item you wish to modify, e.g. APPLICATIONDATA.

MALDI Solutions Administration Console - [User: Engineer]

Maintain the list of available attributes

Select the type of item to modify

APPLICATIONDATA

Customise the names of the system attributes

Unique Name	Display Name
Name	Parameter Set Name
Date	Creation Date
User	Creator
Instrument	Instrument
InstrumentMode	Instrument Mode
InstrumentSerialNumber	Instrument Serial Number

Modify the user-definable attributes

Unique Name	Required	Display Name	Default Value	Obsolete
BatchID	<input type="checkbox"/>	BatchID		<input type="checkbox"/>
Project	<input type="checkbox"/>	Project		<input type="checkbox"/>

Create New

Apply changes Discard changes

3. In the **Customise the names of the system attributes** list, make any required changes to the display names associated with the listed attributes.

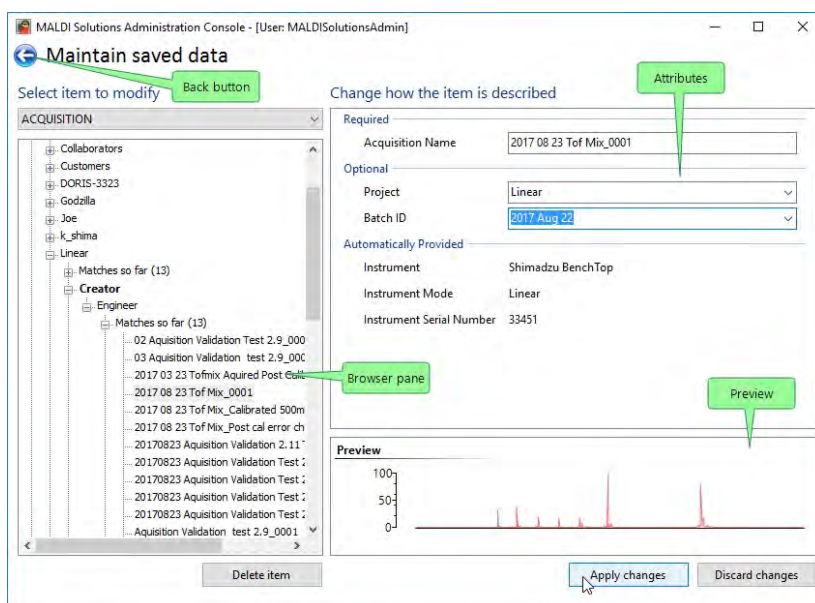
4. In the **Modify the user-definable attributes** list, make the required changes to the attributes:
 - Select the **Required** check box to make the attribute mandatory;
 - Select the **Obsolete** check box to make the attribute obsolete - this will hide the attribute from applications but will not remove it from existing data items;
 - Click **Create New** to add a new attribute to the data type - note that once created, attributes cannot be deleted.
5. Click **Apply changes** to save the changes you made to the attributes, or **Discard changes** to undo any changes.
6. Click the **Back** button to return to the main application window.

Managing data

You can use the application to delete entries from the instrument database, or to modify the values associated with their attributes. This allows you to make more efficient use of the instrument database, for example retrospectively adding attributes which will aid in later searches, or removing unwanted entries which are no longer required.

Modifying attributes

1. In *MALDI Solutions Administration Console*, click **Maintain saved data**.
2. In the **Select item to modify** drop-down list, select the category of data item you wish to modify, e.g. ACQUISITION.
3. Locate and click on the item in the browser pane you wish to modify.



4. Amend the values for the attributes displayed in the **Required** and/or **Optional** sections of the **Change how the item is described** pane.

Notes:

- For some data items e.g. acquisitions, you can click on the link in the **Preview** pane to display a graphical preview of the data item.
- You cannot modify attributes in the **Automatically Provided** section of the dialog.

5. Click **Apply changes** to save the changes you made to the attributes, or **Discard changes** to undo any changes.
6. Click the **Back** button to return to the main application window.

Deleting an item

1. In *MALDI Solutions Administration Console*, click **Maintain saved data**.
2. In the **Select item to modify** drop-down list, select the category of data item you wish to delete, e.g. ACQUISITION.
3. Locate and click on the item in the browser pane you wish to modify.
4. Click **Delete item**.
5. Click **Yes** on the confirmatory dialog to delete the item.
6. Click the **Back** button to return to the main application window.

Chapter 4 - Acquiring data

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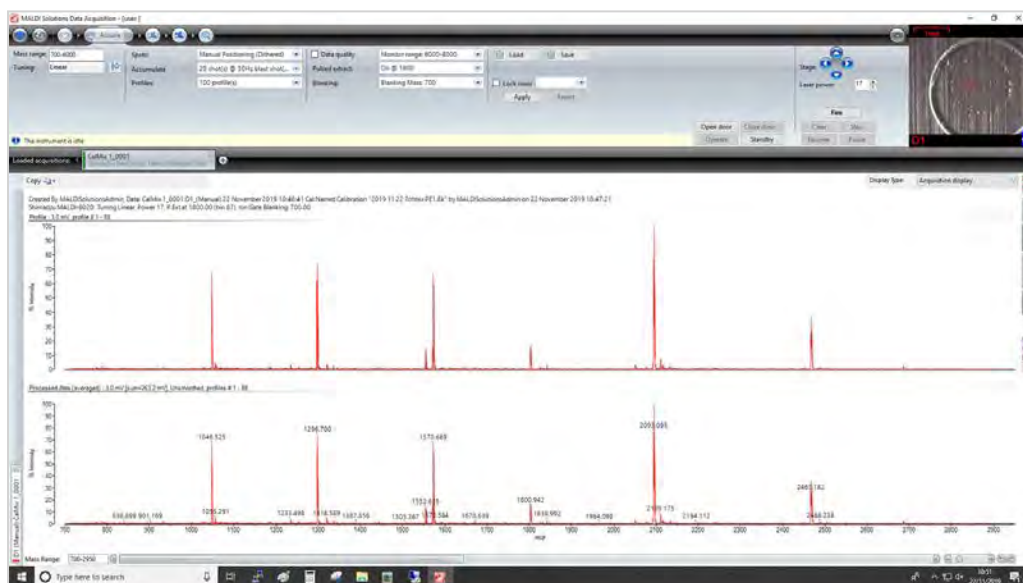
Introduction

Acquiring and analysis of data is primarily achieved through the *MALDI Solutions Data Acquisition* software application. This applications allows you to:

- Acquire data from the sample in the form of a spectrum;
 - Note:** This may be done on single target spots or multiple spots as a batch experiment.
- Calibrate the acquisition for accurate results;
- Process the spectral data for easier analysis;
- Correct mass drift using the Lock Mass feature;
- Load or save acquired data in the application database.

In addition to these primary functions, *MALDI Solutions Data Acquisition* also allows you to:

- Control the camera through an integrated viewer application;
- Manage the usage of target plates;
- Perform comparative searches on the Mascot database;
- Customise the user interface for more efficient working.

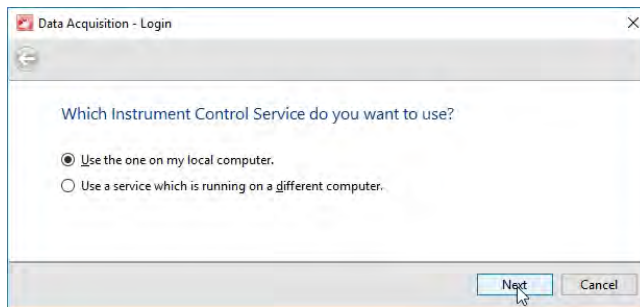


Starting the application

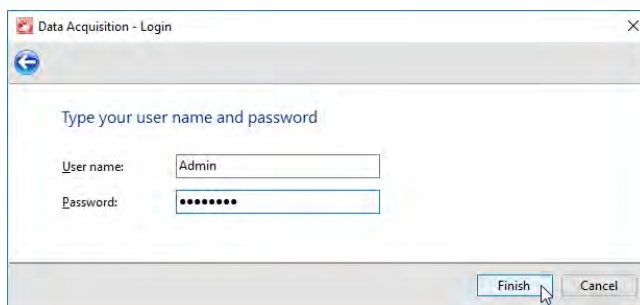
1. On the **Start** menu, locate and select **Shimadzu Corporation > Data Acquisition**.
2. On the **Login** dialog, select the Instrument Control Service on the local computer you want to use and then click **Next**.

Note: The option for a service on different computer is not supported

3.



4. Select **Use the one on my local computer**.
5. Enter your **User name** and **Password**, and then click **Finish**.



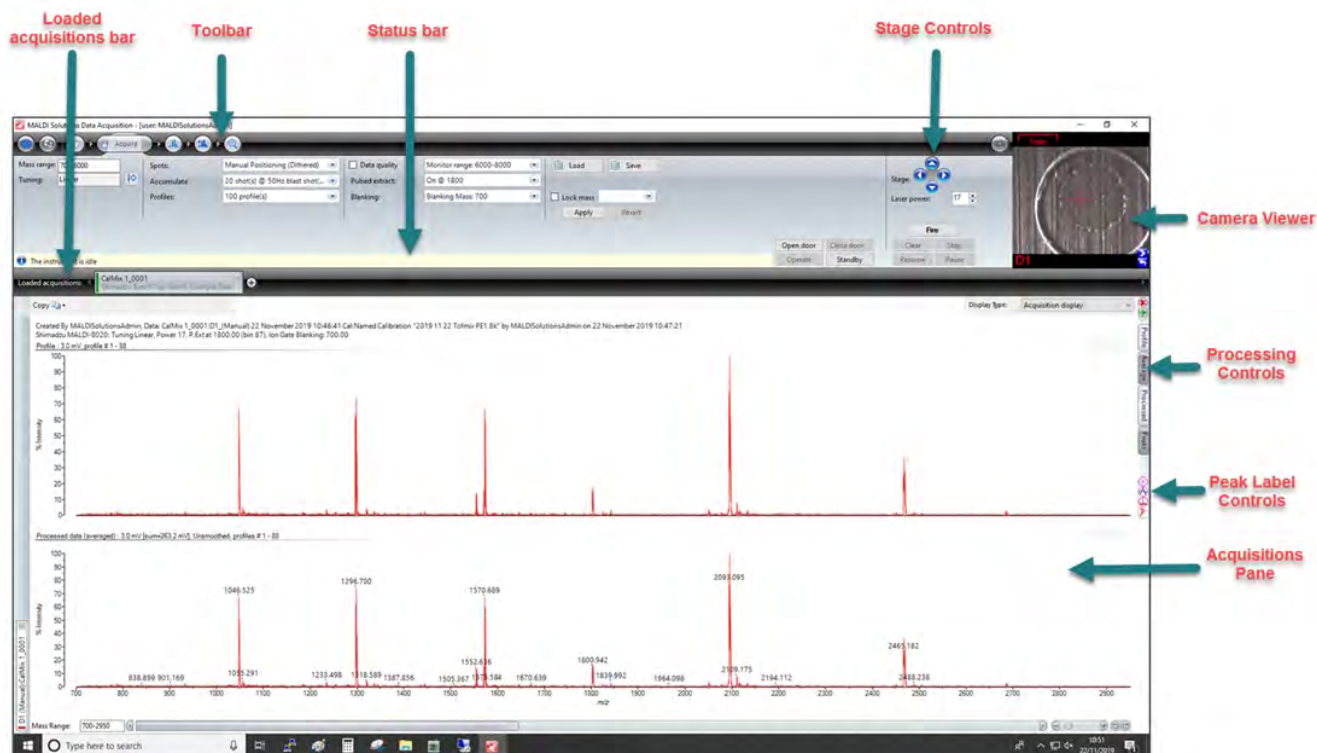
Note: User names and passwords are set by your designated administrator using the [Administration Console](#).

Using the interface

The *MALDI Solutions Data Acquisition* application is designed to guide you through an acquisition. As you work through the application, you only see fields, or features, that are relevant to you at that time.

Application window



The main application window contains all the software controls and can be minimised, resized or tiled on the desktop like any other windowed application. Note that to shut down the software, you should use the standard window close control (X) in the top right corner. Alternatively, use the minimise (-) to hide the top window.








Toolbar

The toolbar contains the following buttons.












General functions




Button	Function
	Settings - use to change your personal settings (see Printing & settings).
	Print Analysis - use to print the display of a spectrum (see Printing & settings).

Workflow functions

Button	Function
	Target tab - provides controls for specifying the type of target plate in use (see Working with samples).
	Acquire tab - displays the main controls and parameter settings for acquiring sample data (see Acquiring data).
	Process tab - displays the functions you can use to process acquired data (see Processing data).
	Calibrate tab - displays the controls for calibrating the instrument for a particular acquisition type (see Calibration).
	Search - controls for searching the Mascot database (see Searching with Mascot).

Other controls

Button	Function										
	Show/hide - minimises the information displayed in the application window.										
	Load acquisition - allows you to load a previously saved acquisition. Multiple acquisitions may be loaded which will appear on different tabs in the Loaded acquisitions bar.										
	Load parameters - allows you to load a previously saved set of acquisition parameters.										
	Save parameters - allows you to save the current set of acquisition parameters.										
	Open door - prepares the instrument for unloading of the current target plate.										
	Close door - locks the sample door and begins the process of loading a target plate (if inserted).										
	Operate - brings the instrument out of standby ready to acquire data.										
	Standby - places the instrument in standby mode (switches off the laser and the high-voltage supplies).										
	<p>Stage controls - allow manual movement of the camera crosshairs around the loaded target plate. You can use shortcut keys in combination with mouse clicks as shown below to vary the units of movement.</p> <table> <tbody> <tr> <td>Click</td> <td>0.05 mm</td> </tr> <tr> <td>Alt+Click</td> <td>0.01 mm</td> </tr> <tr> <td>Shift+Click</td> <td>0.1 mm</td> </tr> <tr> <td>Ctrl+Click</td> <td>0.15 mm</td> </tr> <tr> <td>Shift+Ctrl+Click</td> <td>0.25 mm</td> </tr> </tbody> </table>	Click	0.05 mm	Alt+Click	0.01 mm	Shift+Click	0.1 mm	Ctrl+Click	0.15 mm	Shift+Ctrl+Click	0.25 mm
Click	0.05 mm										
Alt+Click	0.01 mm										
Shift+Click	0.1 mm										
Ctrl+Click	0.15 mm										
Shift+Ctrl+Click	0.25 mm										
	Begins the specified laser firing sequence, acquiring data until the sequence ends.										
	Clear - removes accumulated data from the acquired spectrum.										

Button	Function
	Stop - stops the current acquisition (cannot resume).
	Resume - resumes a paused acquisition.
	Pause - temporarily halts an acquisition.

Loaded acquisitions bar

The **Loaded acquisitions** bar is a tabbed container, with each tab displaying data in the acquisition pane for a newly acquired spectrum or loaded acquisition. Multiple acquired or loaded spectra can be browsed using this control. For more information, see [Loading & saving data](#) and [Acquiring a spectrum](#).

Status bar

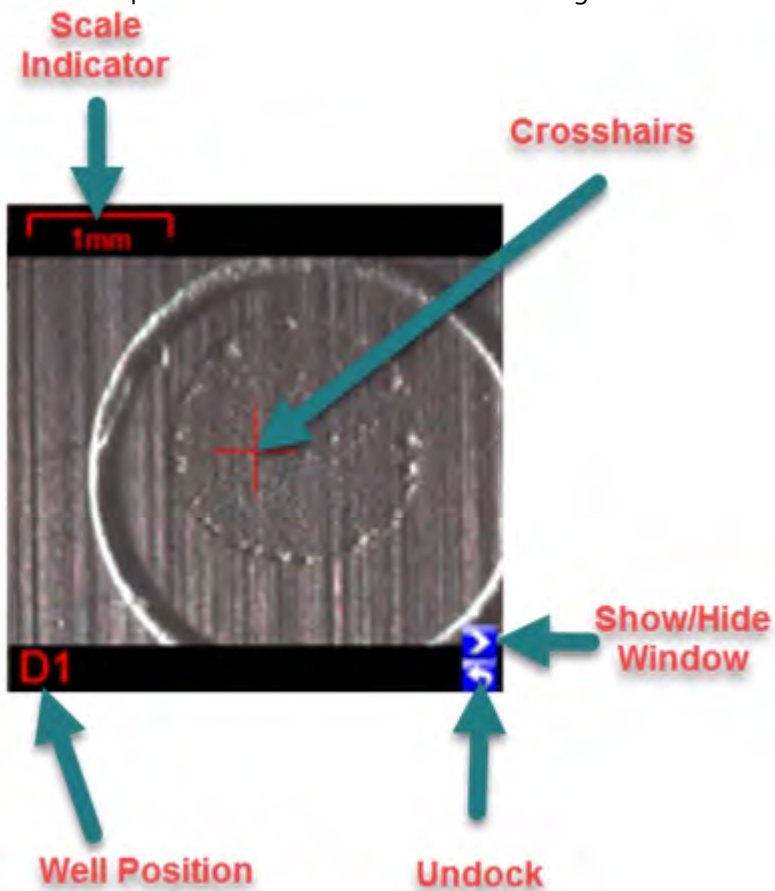
The status bar displays the current status of the instrument. Information including the operating mode, vacuum state and door status appears in this bar as the instrument is used. You should use this display (together with the front panel LED) to determine the current status of the instrument during use.

Camera viewer

The camera viewer provides a magnified view of the area on a loaded target plate where the instrument laser is currently positioned. For more details, see [Using the camera viewer](#).

Using the camera viewer

The camera viewer allows you to directly observe the loaded target plate at high magnification. The crosshairs indicate the point where the laser will strike the target when fired.



Positioning the crosshairs

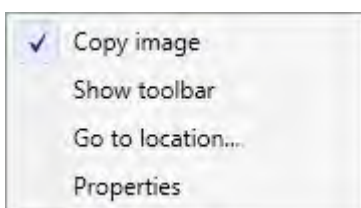
You can position the crosshairs on the Target slide by left-clicking inside the camera viewer. To position the crosshairs more precisely, use the stage controls (see [Using the interface](#)).

Note: The crosshairs are always centred in the camera view. When you use the controls above, the sample stage moves, altering the position on the Target slide the crosshairs point to.

Copying the image

You can copy the image of the camera view to the clipboard and subsequently paste it into another application.

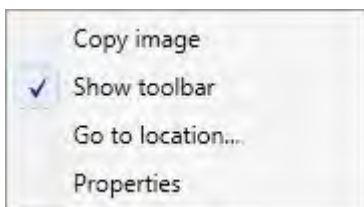
1. Right-click inside the camera viewer window and then select **Copy image** from the pop-up menu.



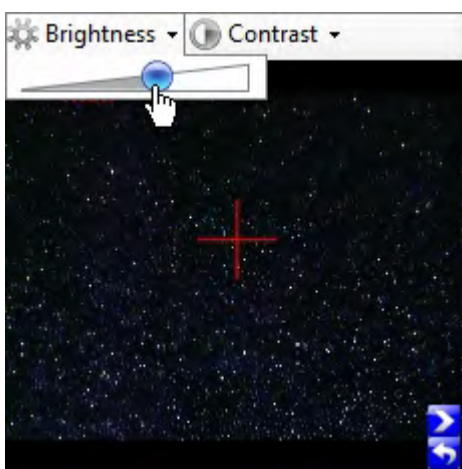
This will copy the camera viewer image to the Windows clipboard. The image can now be pasted into other applications using the standard Windows paste command (Ctrl+V).

Changing the brightness and contrast

1. In the camera viewer window, right-click and then select **Show toolbar** from the pop-up menu.



2. Use the sliders to adjust the **Brightness** and/or **Contrast** settings.

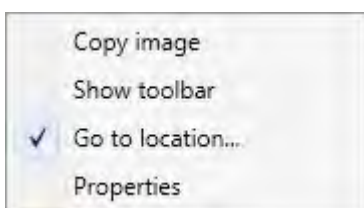


3. Click inside the camera viewer window to dismiss the toolbar.

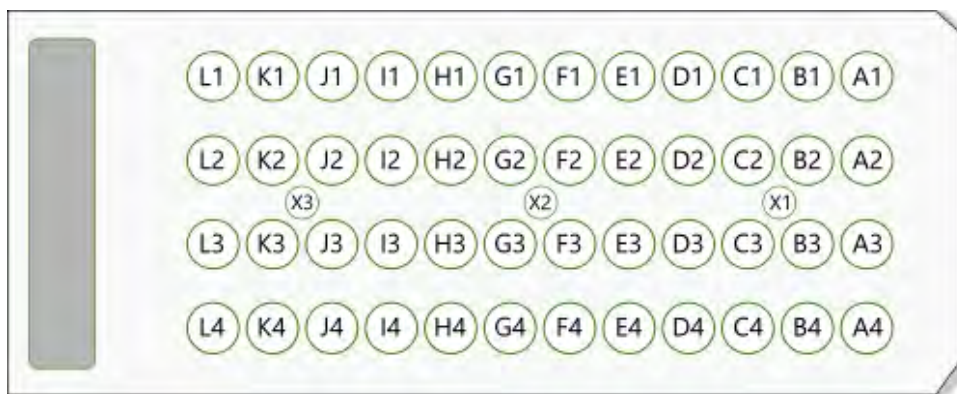
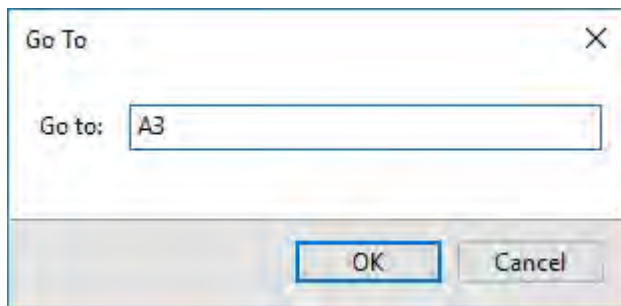
Go to location

Once the target plate has been aligned (see [Loading and unloading target plates](#)), you can use this feature to move to a specific sample position on the plate.

1. In the camera viewer window, right-click and then select **Go to location** from the pop-up menu.

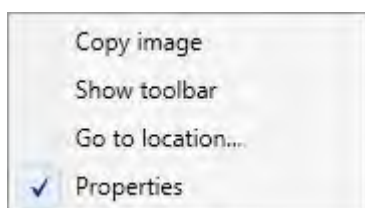


2. In the **Go To** dialog, enter the required sample position using the two-letter codes shown below, and then click **OK** to position the camera crosshairs at the specified location.

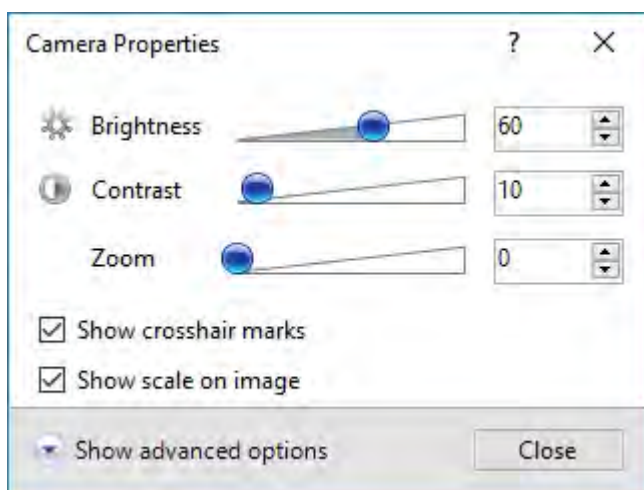


Displaying camera properties

1. In the camera viewer window, right-click and then select **Properties** from the pop-up menu.



2. Adjust the parameter settings as required and then click **Close**.

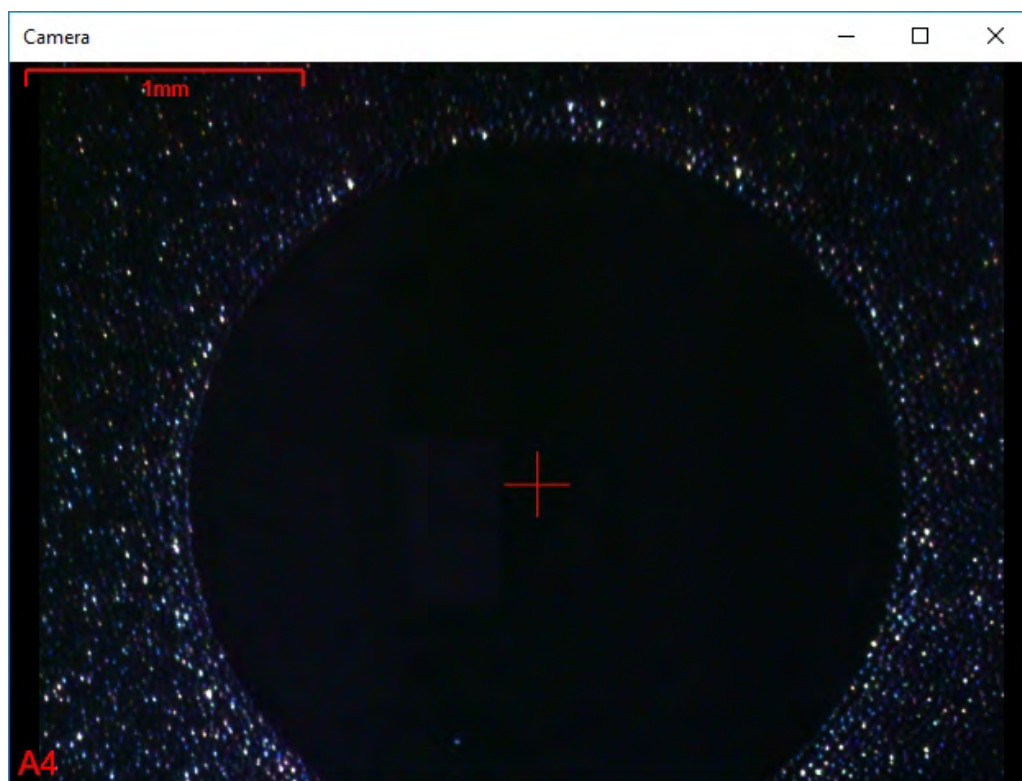


Note: Advanced options are intended for use by for service engineers only and should not be altered.

Undocking the camera viewer

Undocking the camera viewer provides you with a larger image.

1. Click the **Undock** icon to expand the camera viewer into a separate window.



2. Close the window (X) to automatically re-dock the camera viewer.

Loading & saving data

The *MALDI Solutions Data Acquisition* application loads and saves all data to an internal database - there are no external files to manage unless you export data to a different format.

The controls used to load or save data differ depending on which application or dialog is used to initiate the operation. However, the mechanism remains the same. For example, loading and saving of parameter sets and experiments in the optional *QC Reporter* application uses the same basic method as for the acquisitions and parameter sets described below.

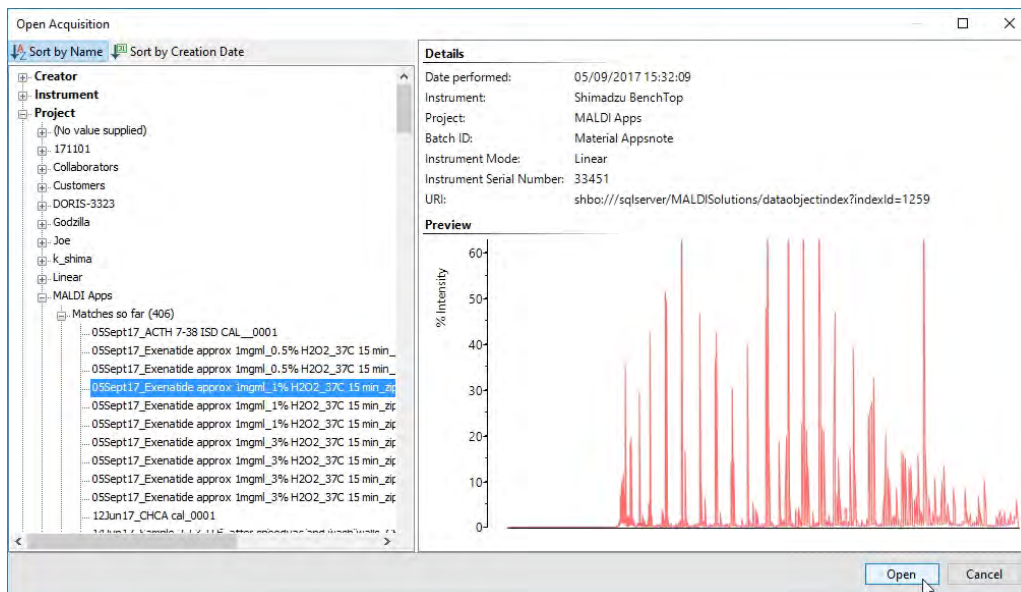
Attributes

When saving data, it is good practice to assign attributes which help to describe the data, as this will make searches easier. Attributes may be mandatory, optional, or are provided automatically. Examples of attributes include "acquisition name", "customer", "project", "batch ID", "part no", etc. Increasing the number of attributes makes finding data easier. Therefore, if you are saving data, provide as many attributes as possible - try not to leave optional fields blank.

Loading & saving acquisitions

Loading an acquisition

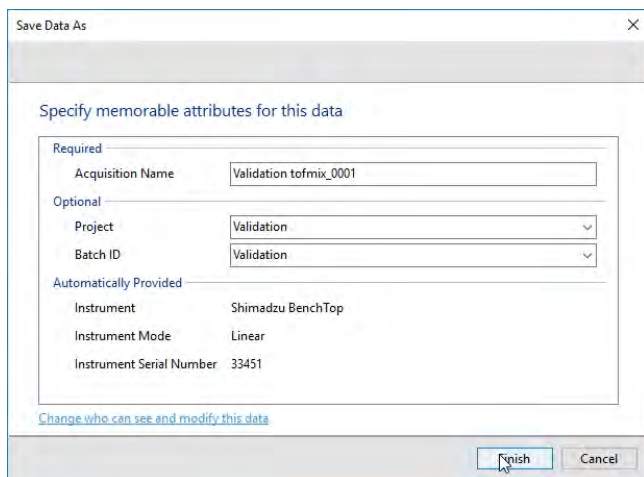
1. On the **Loaded acquisitions** bar, click the **Load acquisition** button.
2. In the **Open Acquisition** browser, select the desired acquisition and then click **Open**.



Note: You can sort the entries by name or creation date by clicking the appropriate button. Click on an entry to reveal the available choices. As you drill down through the tree structure, the number of entries reduces, leaving you with the required entry.

Saving an acquisition

1. In the **Loaded acquisitions** bar, right-click and then select **Save acquisition** from the pop-up menu.



The screenshot shows the 'Save Data As' dialog box with the 'Specify memorable attributes for this data' section. It is divided into three categories: Required, Optional, and Automatically Provided.

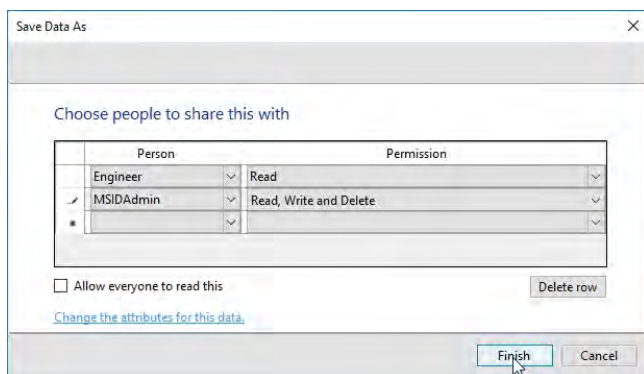
Category	Attribute	Value
Required	Acquisition Name	Validation tofmix_0001
Optional		
Project	Validation	▼
Batch ID	Validation	▼
Automatically Provided		
Instrument	Shimadzu BenchTop	
Instrument Mode	Linear	
Instrument Serial Number	33451	

At the bottom of the dialog, there is a link: [Change who can see and modify this data](#). Buttons for 'Finish' and 'Cancel' are also present.

2. In the **Save Data As** dialog, enter the required attributes.

Note: When saving data, it is good practice to assign attributes which help to describe the data, as the browser used to load data uses attributes to organise the tree structure. Attributes may be required, optional, or automatically provided.

3. Click the **Change who can see and modify this data** link to set user permissions for the saved data.



The screenshot shows the 'Save Data As' dialog box with the 'Choose people to share this with' section. It features a table with columns for 'Person' and 'Permission'.

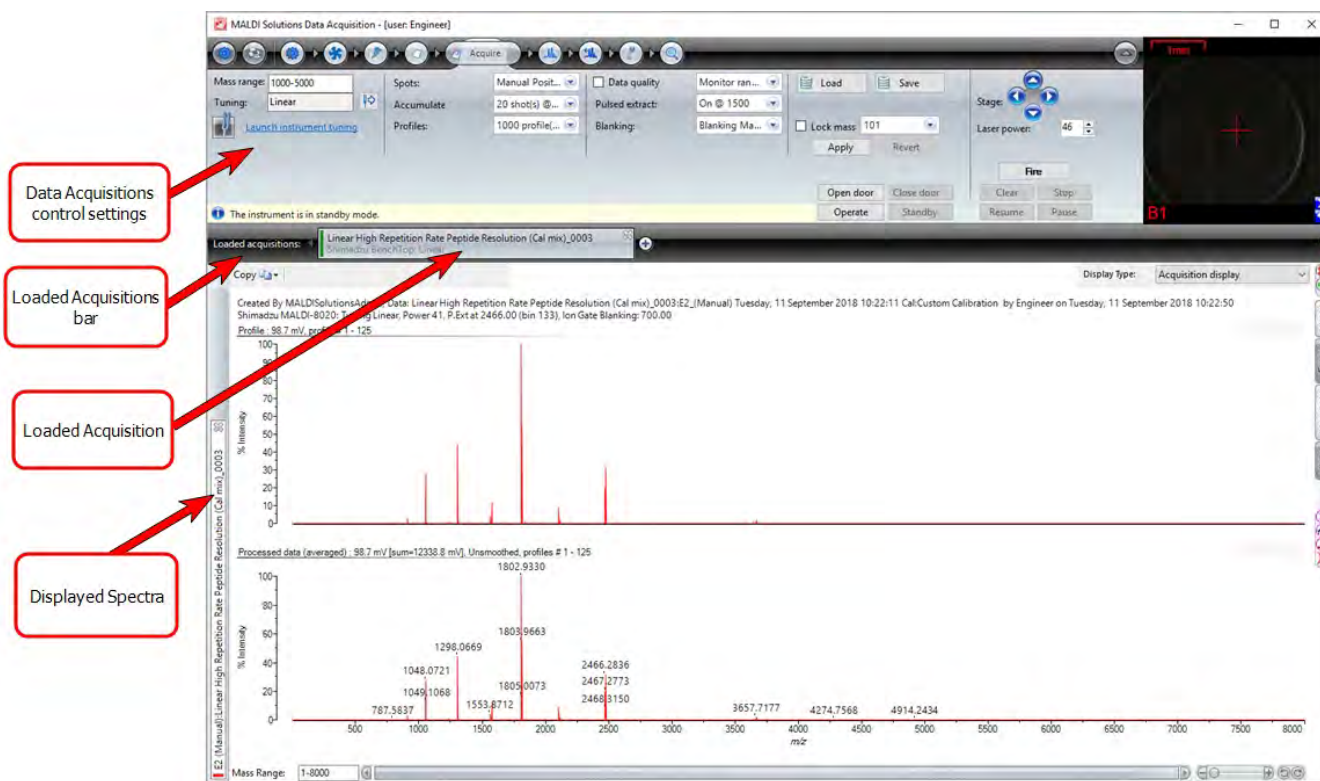
Person	Permission
Engineer ▼	Read ▼
MSIDAdmin ▼	Read, Write and Delete ▼
▼	▼

Below the table, there is a checkbox: Allow everyone to read this. A 'Delete row' button is also present. At the bottom, there is a link: [Change the attributes for this data](#). Buttons for 'Finish' and 'Cancel' are also present.

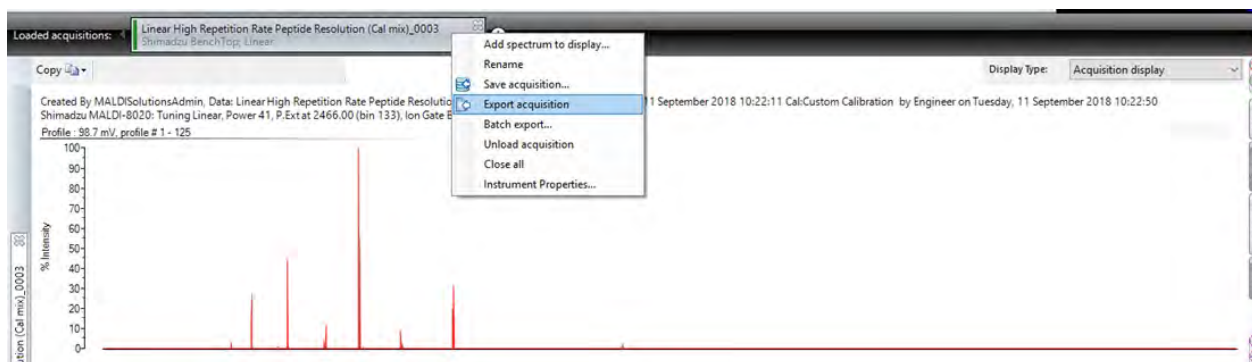
4. To allow everyone to read your preferences, select the **Allow everyone to read this** check box.
5. To add a user, in the **Person** drop-down list, select the required name.
6. In the **Permissions** drop-down list, select the required permissions for each user in the **Person** list.
7. To delete a row, select it and click **Delete row**.
8. Click **Finish**.

Exporting Acquisitions

This feature allows you to export all, or specific, displayed acquisitions to an XML file. You can only export an acquisition that has been saved.

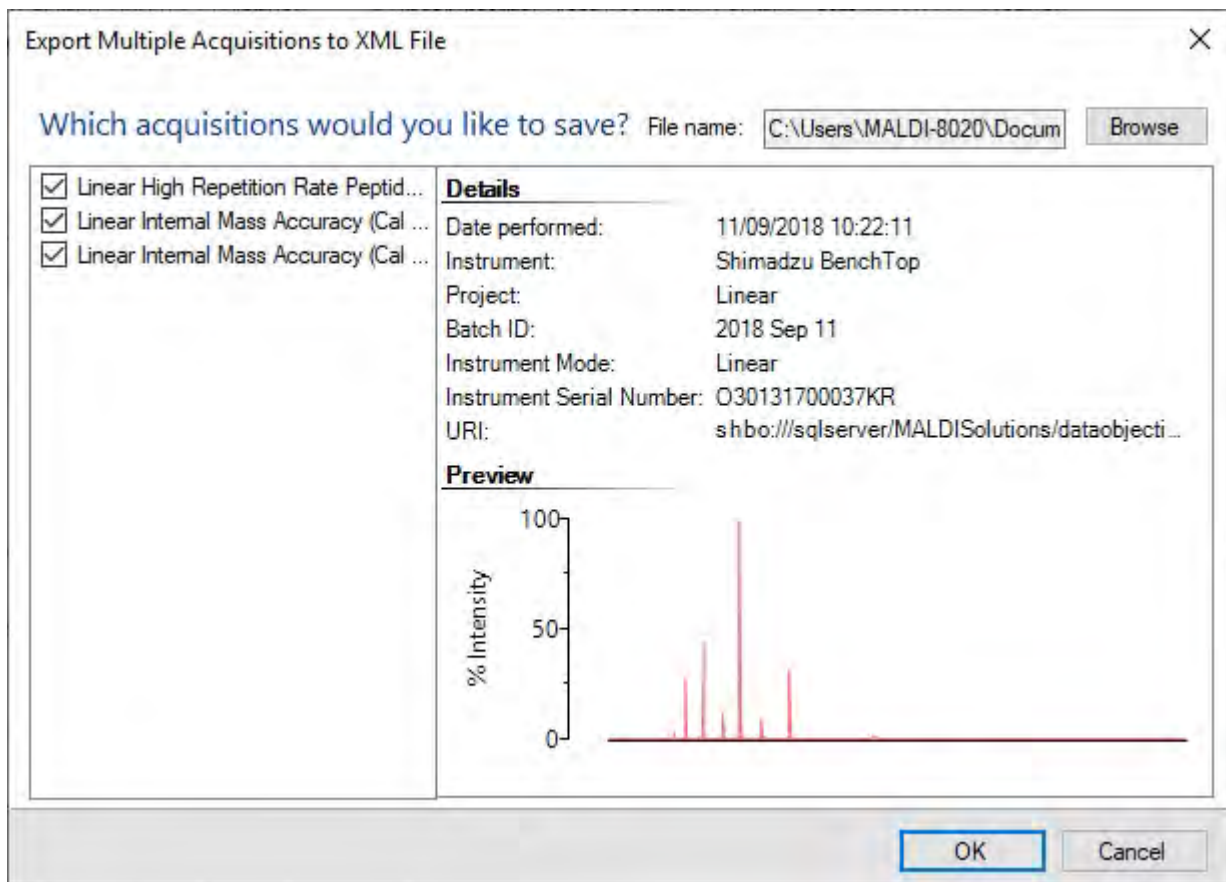


1. Right-mouse click on the required acquisition in the Loaded Acquisitions bar.



2. From the drop-down list of values, select **Export Acquisition**.

The Multiple Acquisitions to XML File windows displays.



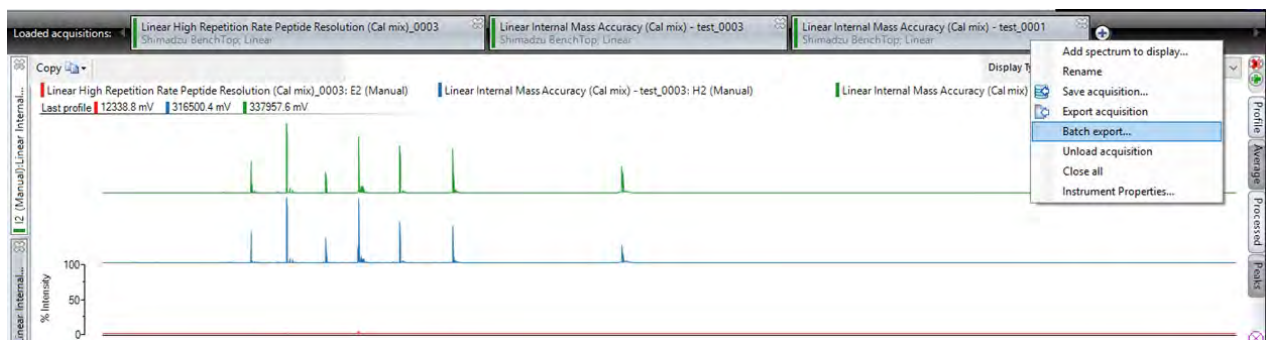
1. Select, or clear, the check boxes for the required acquisitions.
2. Click **Browse** and select the path and file name for the export.
3. Click **OK** to create a file.

The XML file is created.

Batch exports

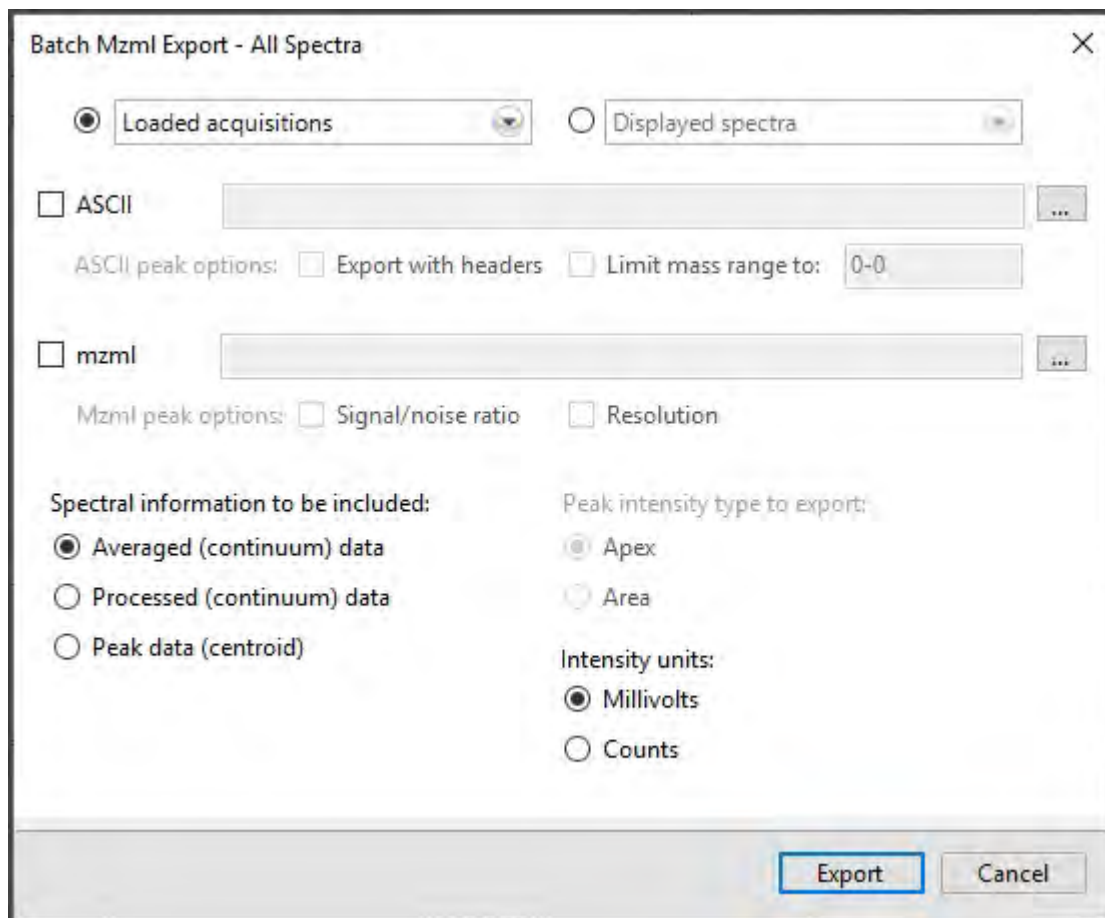
This feature allows you to export numerous acquisitions to mzML or ASCII based files. For example, if the dataset contains several spots, each spot will be exported as a separate file (one file per spot). If the dataset is for a single spot, only one mzML or ASCII file will be created.

1. Right-mouse click in the acquisition tool bar.



2. Select **Batch export**.

The Batch mzML Export - All Spectra window displays.




"The Batch mzML Export - All Spectra" window provides you with options such as the type of export, whether the spectra to be exported is from loaded acquisitions or from a displayed spectra and to specify what spectral information is to be included in the export file to be created.

3. Do one of the following:

- If the export is to contain one or more previously loaded acquisitions, select the Loaded acquisitions option and choose the acquisition or acquisitions from the drop-down options list.
- If the export is to contain one or more of the displayed spectra, select the Displayed spectra option and choose one or more spectra from the drop-down options list.

4. Select the file type

5. Click the browse icon  and define the folder for the export.

6. Select the required spectral information (radio button).

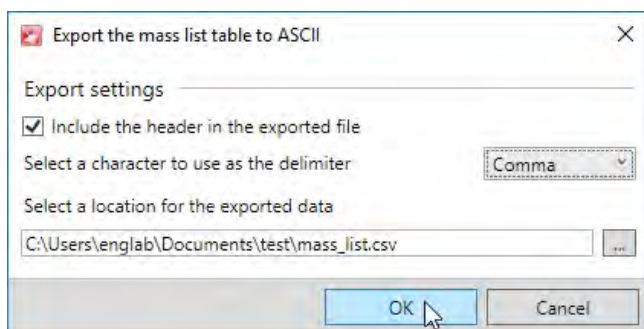
7. Select any additional required peak information (checkboxes).

8. Click **Export** to create a new file within the defined folder.

An output file is created and can be viewed in the specified destination.

Exporting mass lists

1. With the mass list displayed in the acquisition tab (see [Working with spectra](#)), click the **Export mass list** button.



2. To add a header row to the exported data file, select **Include the header in the exported file**.
3. From the drop-down list, select **Comma, Space** or **Tab** as the delimiter character.
4. Use the browse button (...) to select a destination folder and file name for the exported file.
5. Click **OK** to export the mass list as a CSV (comma separated values) formatted file.

Loading & saving calibration references

Loading calibration references

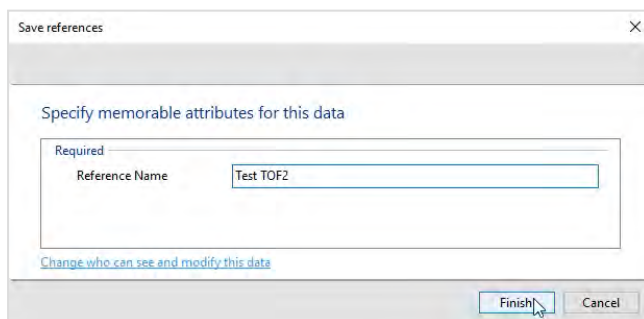
1. On the **Calibrate** tab, click the **Load references** link, select the desired reference and then click **Open**.



Note: The loaded calibration reference will replace any currently entered reference masses for the current calibration.

Saving calibration references

1. On the **Calibrate** tab, click the **Save references** link



2. Enter a name that best describes the references.

- If required, click the **Change who can see and modify this data** link to set user permissions for the saved data (see [Saving an acquisition](#)).
- Click **Finish**.

Loading & saving acquisition parameters

Loading acquisition parameters

- On the **Acquire** tab, click the **Load** button, select the desired parameter set and then click **Open**.



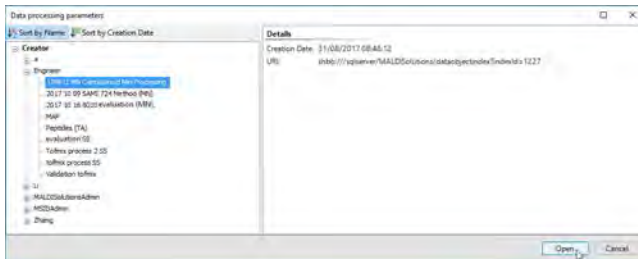
Saving acquisition parameters

- On the **Acquire** tab, click the **Save** button.
- Follow the procedure as described in steps 2 through 8 of [Save acquisition](#).

Loading & saving processing parameters

Loading processing parameters

- On the **Process** tab, click the **Load** button, select the desired parameter set and then click **Open**.



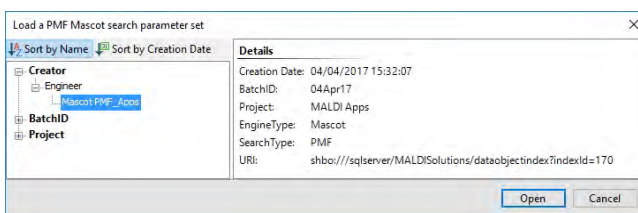
Saving processing parameters

- On the **Process** tab, click the **Save** button.
- Follow the procedure as described in steps 2 through 8 of [Saving an acquisition](#).

Loading & saving Mascot search parameters

Loading Mascot search parameters

- On the **Search** tab, click the **Load** button.



2. Navigate to the required search parameter set and then click **Open**.

Saving Mascot search parameters

Note: Before saving, you must have already created or loaded a Mascot search parameter set (see [Searching with Mascot](#))

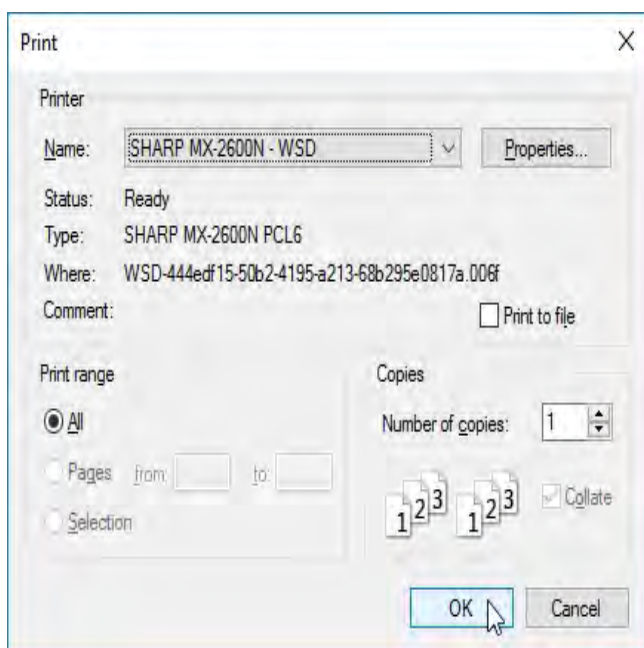
1. On the **Modifying parameters dialog** tab, click the **OK** button.
2. Follow the procedure as described in steps 2 through 8 of [Saving an acquisition](#).

Printing & settings

The general functions on the toolbar (see [Using the interface](#)) allow you to print the currently selected acquisition spectrum, and also to customise certain aspects of the application to suit your requirements.

Printing a spectrum

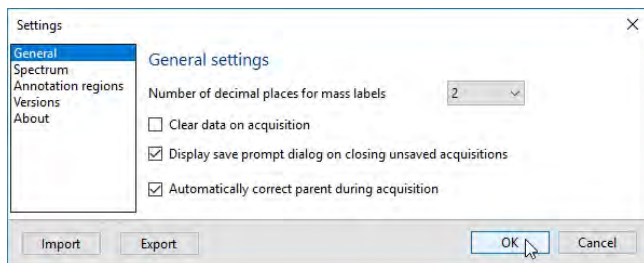
1. From the toolbar, click the **Print analysis** button.
2. On the **Print** dialog, select the desired print options and then click **OK**.



The currently selected spectrum will be printed to your selected output device.

Changing the settings

1. From the toolbar, click the **Settings** icon.
2. Select the settings group you require from the left column, make the desired changes in the right pane of the dialog, and then click **OK**.



The available settings groups are as follows:

General

Number of decimal places for mass labels

Set the number of decimal places for the mass label you wish to display on the spectrum.

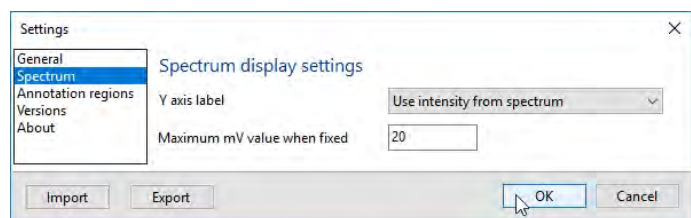
Clear data on acquisition

During an experiment, when you click the **Fire** button you will acquire a set of acquisition data. If you click **Fire** again, you will acquire a second set of acquisition data. If this field is not selected, the first set of acquisition data remains displayed with the new acquisition. If this field is selected, the first set of acquisition data is overridden by the data from the second acquisition.

Automatically correct parent during acquisition

Not used on this instrument.

Spectrum



Y axis label

This field determines how the spectra are displayed:

- **Use the largest intensity from all spectra** - If you have more than one spectrum displayed, the Y axis % intensity is adjusted to use the largest intensity from all displayed spectra. All the other spectra become normalised relative to the largest intensity.
- **Use intensity from spectrum** - The Y axis is set to % intensity.
- **Use fixed millivolt value** - This field operates in conjunction with the millivolt value set in the next field. The Y axis is set to mV.

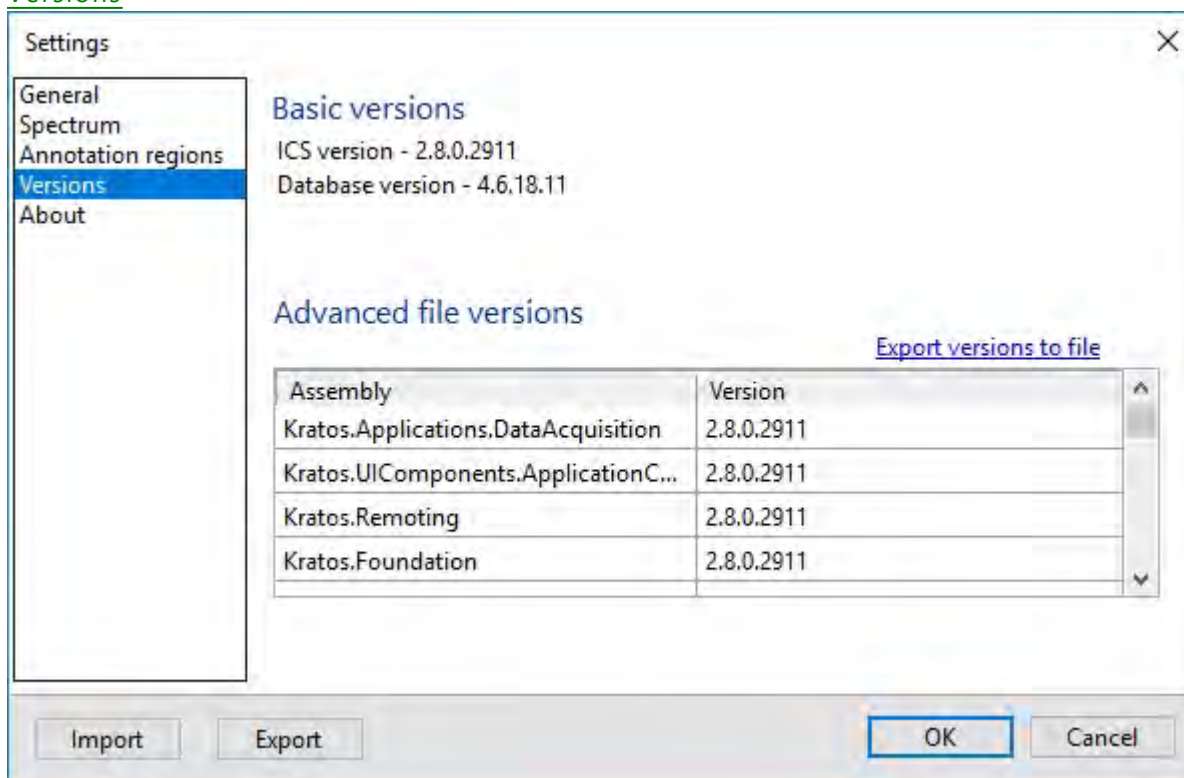
Maximum mV value when fixed

This field operates in conjunction with the **Use fixed millivolt value** field above. Type in the required value.

Annotation regions

For details, see [Annotations](#).

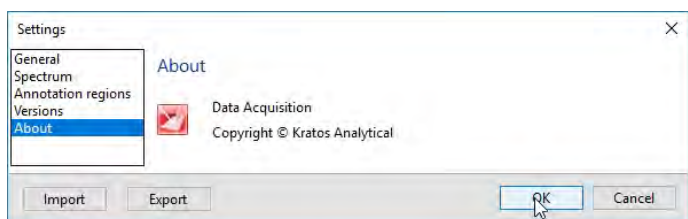
Versions



This feature lists all the version numbers of the software modules. You may also export the version number information to an XML format file by clicking **Export versions to file**.

About

This settings group provides information about the copyright owner.



Import/export

The **Import** and **Export** buttons allow you to load or save the current settings from all groups within the dialog to an XML format file.

Acquiring a spectrum

Notes:

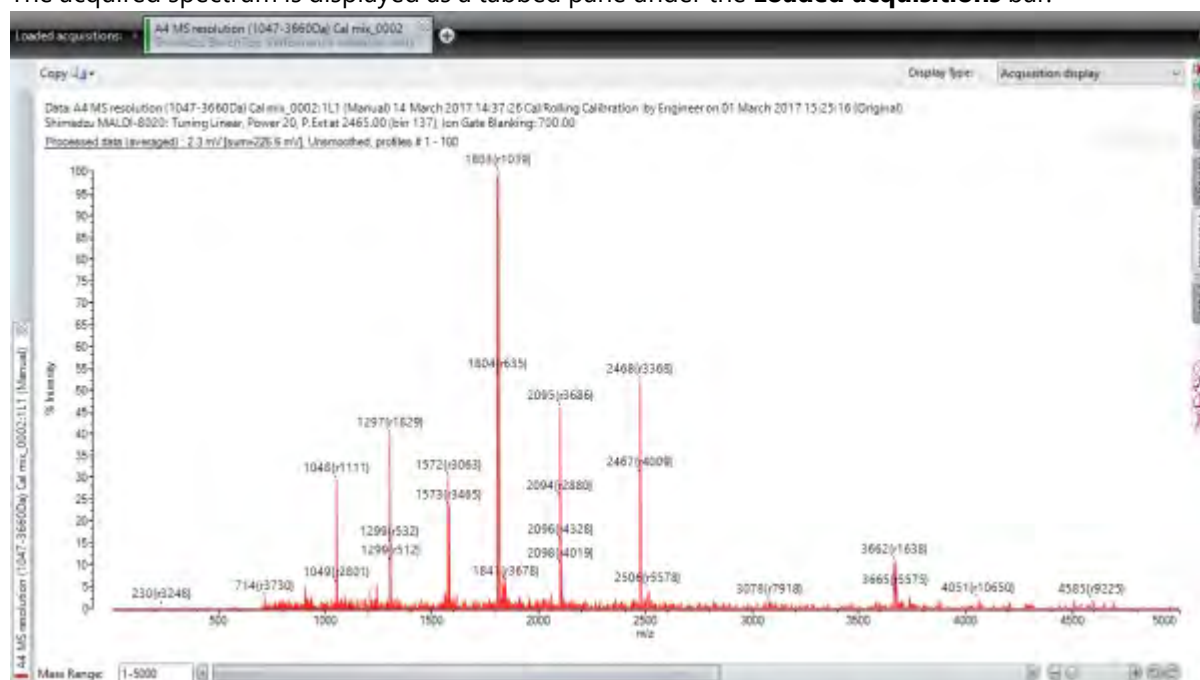
- The term "sample position" refers to a defined area on a target plate where you can place a sample spot, referenced by a row/column ID (eg **A4**). In some applications, the sample position is also referred to as a "well".
- The terms "sample spot" or "spot" refer to a sample position (or well) on which a sample has been deposited (or "spotted").
- The term "raster point" refers to the precise area targeted by the laser as it fires a single shot onto a target plate. Acquisitions may use a single raster point, or incorporate multiple raster points across one or more sample positions on the target plate.
- The term "lock mass" refers to the a function that allows you to compensate for systemic drifts between calibrations and allow the spectra to be of higher mass accuracy. For a more detailed description, (see [Lock Mass](#)).

Performing an acquisition

This procedure describes the basic process for acquiring data from one or more samples on a loaded target plate and displaying the acquired spectrum.

1. If you have not already done so, load a suitably prepared target plate into the instrument (see [Loading and unloading target plates](#)).
2. In *MALDI Solutions Data Acquisition*, select the **Acquire** tab.
3. In the **Acquire** tab, set the required parameters for the acquisition (see [Setting acquisition parameters](#)).
4. If you have not already done so, click the **Operate** button.
5. If you have already specified the required raster point(s) to fire on in the acquisition parameters, proceed to step 7.
6. Manually position the crosshairs in the camera viewer on the raster point you wish to fire on, either by left-clicking within the viewer (see [Using the camera viewer](#)) or by using the stage movement controls (see [Using the interface](#)).
7. If required, specify a lock mass value. If not specified, but is required, the lock mass value can be applied after the acquisition is complete.
8. Click the **Fire** button to begin the acquisition.
9. If you have selected **Continuous firing** in the **Profiles** section of the acquisition parameters, click the **Stop** button to halt the acquisition.

The acquired spectrum is displayed as a tabbed pane under the **Loaded acquisitions** bar:



- Once you have acquired a spectrum, you may perform various operations including saving, exporting or changing the way it is displayed on screen. For more details, see [Working with spectra](#).

During an acquisition

While acquiring, you can:

- adjust the laser power for optimal results;
- for manually selected raster points, move the point where the laser hits;
- clear the acquired data;
- pause & resume the acquisition;
- stop the acquisition (and change the processing parameters).

Adjusting the laser power

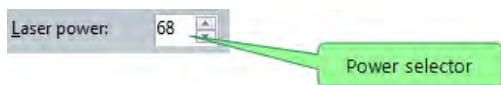
The arbitrary laser power range is 0 to 180.

Select the required laser power:

- too low - sample will not ionise resulting in no or low intensity peaks;
- too high - peaks will be "off the scale" where the detector becomes saturated resulting in poor resolution.

To set the power you can:

- Type in the required power;
- Use the power selector to raise/lower the power in increments of one unit.



See [Setting the optimal laser power](#) below for more details.

Managing an acquisition

- At the start of an acquisition, you may raise/lower the laser power to find the optimal setting. In this scenario, profiles accumulated during optimisation of the laser power may not be required; click **Clear** to remove these spectra.
- You can also **Pause** an acquisition, adjust the peak processing parameters, and **Resume** the acquisition.
- The **Stop** button stops the acquisition, you cannot then resume the acquisition.
- The **Standby** button switches off the laser and the high-voltage supplies.

Setting the optimal laser power

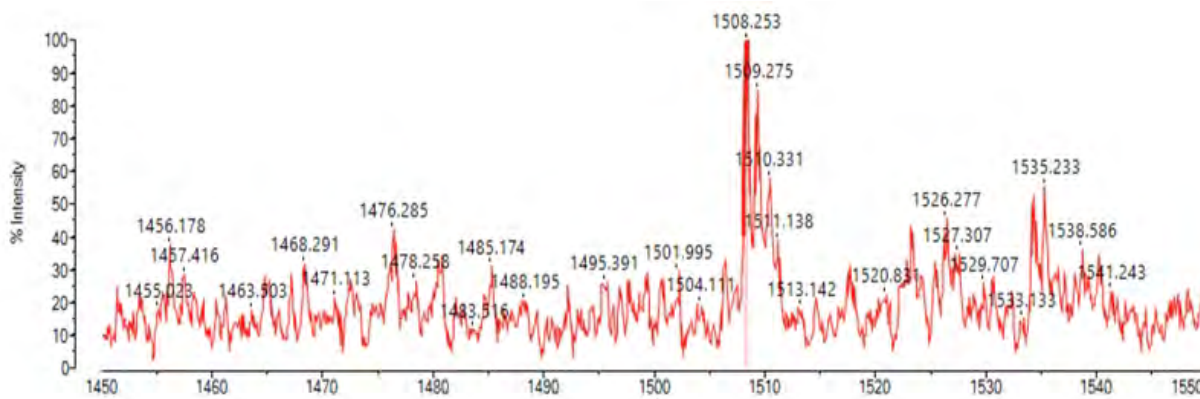
If the laser power is too high, resolution may be reduced as a result of increased initial spatial distribution of the ions and high signal intensity. Additionally, detection of background noise may also increase.

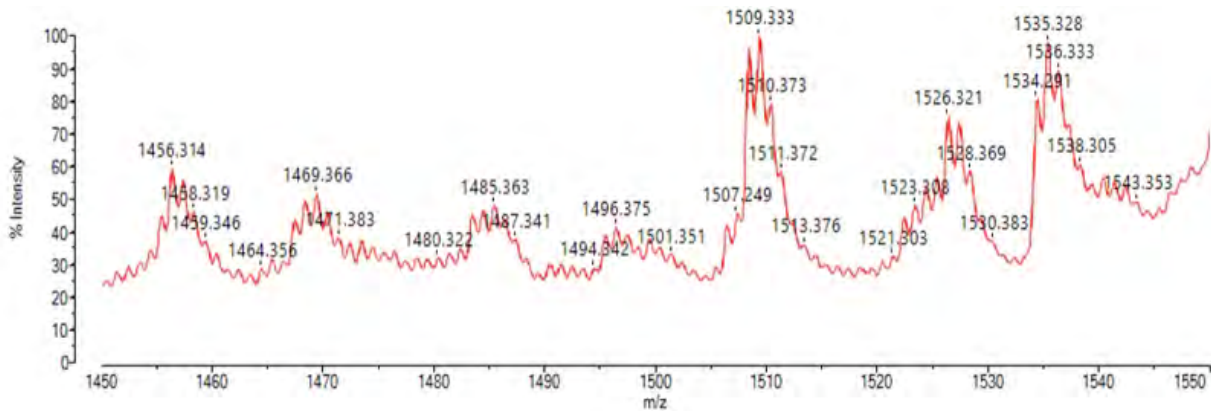
To obtain the best resolution and reduce the detection of background noise, set the laser power to a value slightly above the minimum laser power required for ion generation/detection. The actual laser power required will depend on various factors including the matrix type, concentration of the analyte and sample purity.

If the laser power is too low, the ion signal will be unstable and the signal intensity will be low. Increase the laser power until a signal for the target analyte is observed in the majority of profiles acquired.

If the laser power is too high, the noise peaks may be observed as a regular 'ripple', with a peak observed every 1 Da. This ripple appears only in the specific m/z region depending on the pulsed extraction setting. These ripple peaks are derived from the actual ion fragment. Reduce the laser power to remove this unwanted effect.

Laser power is appropriate



Laser power is too high

Setting acquisition parameters

The parameter settings and controls used to define the acquisition process are displayed on the **Acquire** tab.

Mass range

The mass range parameter controls the upper and lower limits for the masses which are included in the acquired spectrum.

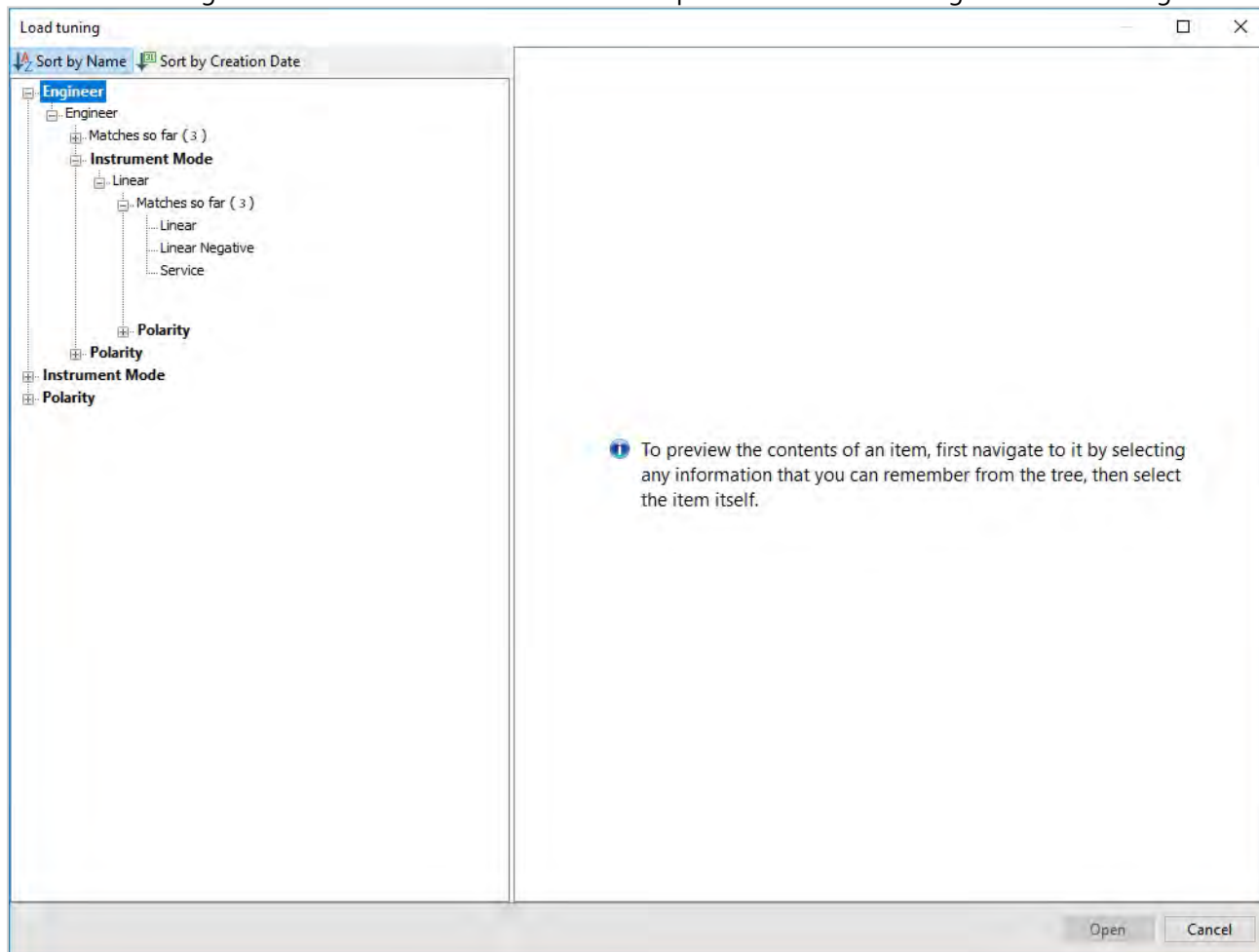
- For masses up to 350 kDa, enter the range in Da (for example 1-5,000).
- For masses above 350 kDa, set a lower limit of 10 kDa (for example, for an upper mass range of 500 kDa, enter 10,000 - 500,000).

Tuning



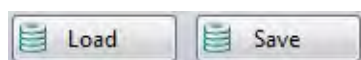
This button is used to select the tuning mode to use for an acquisition. The tuning parameter settings are saved to named data sets, e.g. "Linear". You should only use Linear mode tuning files with the instrument.

Note: When using the MALDI-8030 there is an additional option to select Linear negative mode tuning.



Click the button to display the available tuning modes, select the required mode and then click **Open**.

Load/save



You can load or save acquisition parameter sets by clicking on one of the buttons shown above. See [Loading & saving data](#).

Spots

This option provides more control over the selection of sample spots for the laser to fire on.

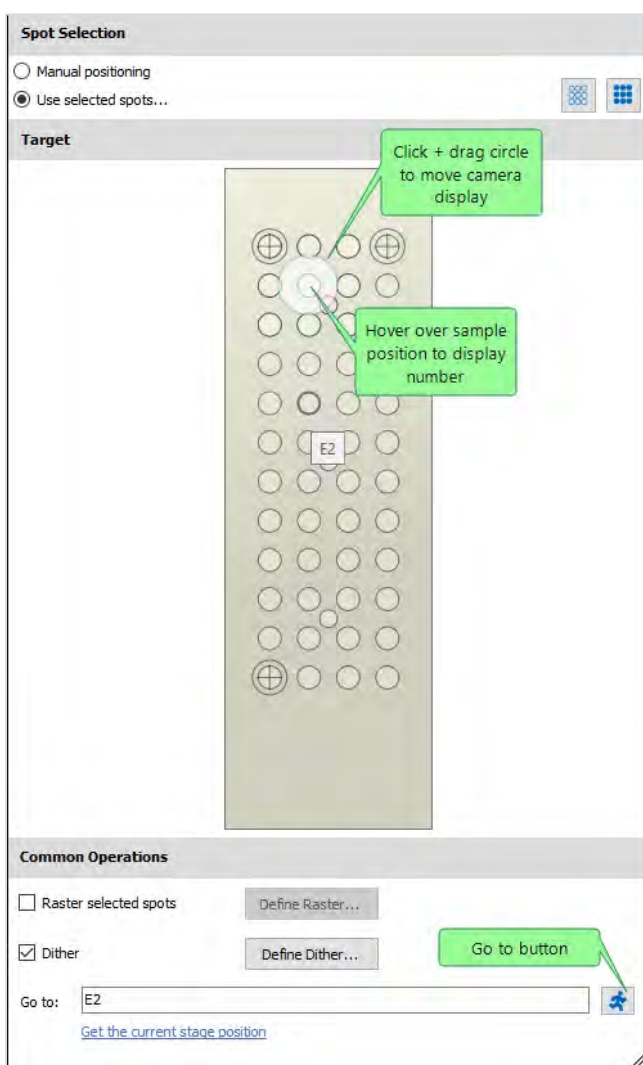
To access the **Spot Selection** dialog, click the down arrow next to the **Spots** field and then select either the **Manual positioning** or **Use selected spots** radio buttons. To dismiss the dialog, click outside the window or click the down arrow control again.

Manual positioning

If you select **Manual positioning**, you can define the spot(s) to be used either by:

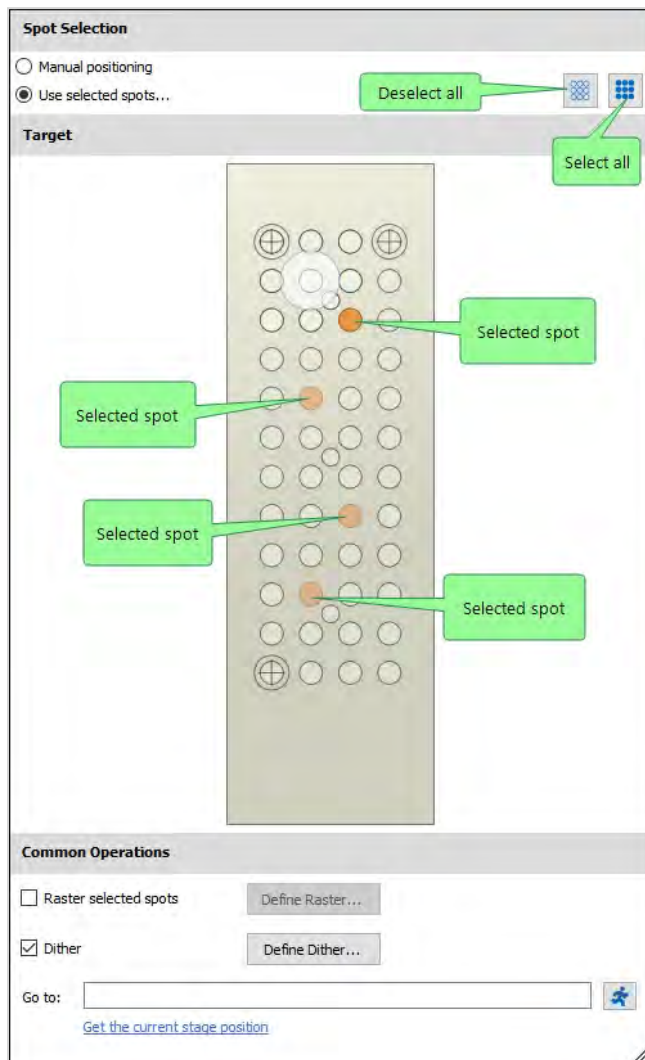
- Clicking and dragging the target circle onto a sample position (the camera crosshair will move to the selected area);
- Enter a sample position number in the **Go to** box and then click the **Go to** button (this works in the same way as the **Go to location** function in the [camera viewer](#)).

Note: You can identify a sample position number by hovering over the desired position.



Use selected spots

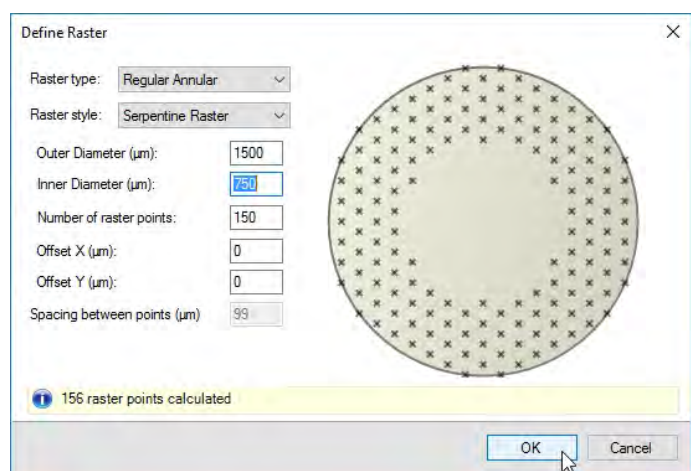
1. If you choose **Use selected spots**, you can select or deselect a spot by left-clicking on it. Multiple spots may be selected, with each chosen spot highlighted on the **Target** display area. You can also use the **Select all** and **Deselect all** buttons to control the selection.



Defining a raster pattern

Defining a raster pattern causes the laser to fire in a sequential pattern over the selected spot(s) rather than a single raster point. Choosing the correct raster pattern will depend on your sample composition. For example, the **Regular circle** type is most suited for samples using a CHCA matrix, as this tends to form small crystals which are evenly distributed across the sample well. The **Regular annular** type is better suited to samples using a DHB matrix, as this tends to form larger crystals at the outer well edges. The **Regular square** type is an alternative to **Regular circle**, but

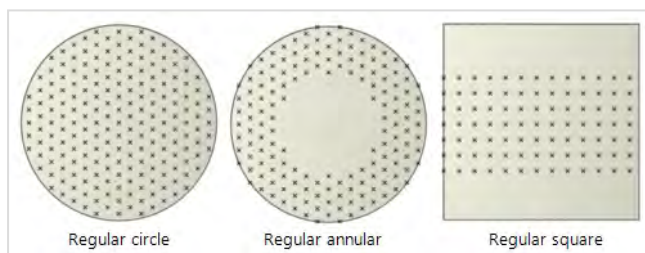
will acquire in a square or rectangular pattern.



To define a raster pattern:

1. On the **Spot Selection** dialog, select the **Raster selected spots** check box and then click **Define Raster**.
2. On the **Define Raster** dialog, select the **Raster type** from the drop-down list.

Note: You can choose from the following predefined types:



3. Select the **Raster style** from the drop-down list.



Note: The **TV raster** style always scans from right to left, while the **Serpentine raster** style scans alternately from left and right.

4. If you selected a regular square raster pattern, enter the height and width. Alternatively, if you selected a circular raster pattern, type in the required **Outer diameter** of the raster pattern (no points outside of this area will be scanned) up to a maximum of 2800 μm .
5. If you selected **Regular annular** as the raster type, enter an **Inner diameter** value (no points inside this area will be scanned).
6. Enter the required **Number of raster points**.

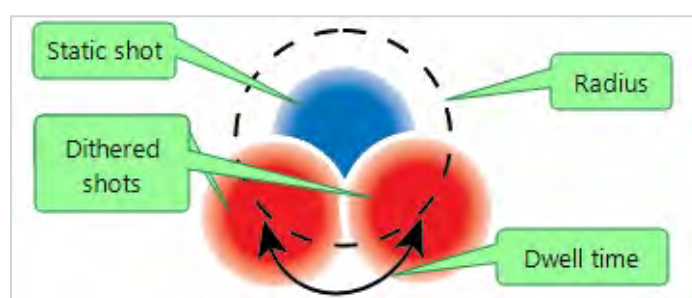
Note: A raster point is the position where the laser fires once at the sample. You can specify any number of

raster points (up to 200 for the Regular Circle and Regular Annular type). The software will use this number, and the available raster area, to calculate the effective number of raster points. Therefore, the number of raster points that you specify may be different from the actual number of points calculated - as displayed in the information bar.

7. If you wish to define an offset from the centre, enter a value in the **Offset X** and/or **Offset Y** fields.
8. Click **OK** to complete the raster definition.

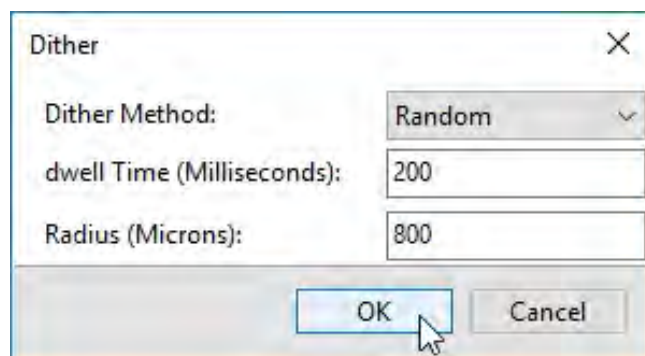
Defining dithering

During dither acquisition there is constant firing of the laser as the stage moves independently. This results in acquisition from various random positions. The region of sampling is based on the user defined radius and the dwell time determines how long the stage remains at each random location. Instead of static shots seen in a raster acquisition there are dithered shots.



To define dithering parameters:

1. On the **Spot Selection** dialog, select the **Dither** check box and then click **Define Dither**.



Note: Only the random **Dither Method** is currently supported.

2. Set the **Dwell time** in milliseconds (this is the time from one dither position to the next).
3. Set the **Radius** in microns (this is the maximum distance of the dither position from the static position).
4. Click **OK**.

Accumulate

Each laser shot produces an individual spectrum. This option controls how these spectra are combined into profiles. A profile is a composite spectrum made up of the combined data produced when a number of laser shots are fired. For example, accumulating 5 shots will collect 5 spectra and then automatically combine them into a single spectrum (the profile).

To set the accumulation parameters:

1. Click the down arrow next to the **Accumulate** field.

2. Set the **Shots to accumulate** value (up to a maximum of 128 shots) either by typing a value or using the spin controls.
3. If required, select the **Laser rep rate** (the number of times the laser fires per second) from the drop-down list.
4. If required, enter the number of **Blast Shots**.

Note: This option instructs the application to discard the data from the specified number of (first) shots at each defined spot. This may help to improve the quality of the acquired spectra in cases where contaminants accumulate in the top layer of the spotted sample.

Profiles

This parameter allows you to specify the number of profiles (spectra) accumulated before the acquisition stops automatically.

To adjust the profile parameters:

1. Click the down arrow control next to the **Profiles** field.

2. Enter a value in the **Set number of profiles** (up to a maximum of 30.000) or select the **Continuous firing** radio button.

Note: Selecting the latter option will cause the laser to fire repeatedly until you click the **Stop** button in the stage controls area (see [Interface](#)).

3. If required, select the **Store Individual Profiles** check box to allow you to view each profile separately.

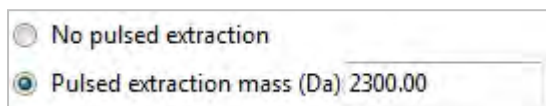
Note: This option will rapidly fill the database with profile entries. Do not select it unless you need to use this feature.

Data quality

See [Data quality](#).

Pulsed extract

This parameter may improve the mass resolution depending on the type of sample. For samples containing a single mass, as a starting point the pulsed extraction (PE) value should be set to the same mass, but it is recommended to optimise up to 1.6x of the mass value. The pulsed extract (PE) value should be set to the same mass. For broad-range samples, set the PE value to approximately $\frac{2}{3}$ of the largest expected mass.



To adjust the PE value, click the down arrow next to the **Pulsed extract** field and enter the required **Pulsed extraction mass** value in Daltons, or select **No pulsed extraction**.

Blanking

This option allows you to filter out ions below a certain mass range, or allow all ions to be detected regardless of mass. For example, a blanking mass of 700 Da filters out ions between 1 and 700 Da, including most matrix ions.

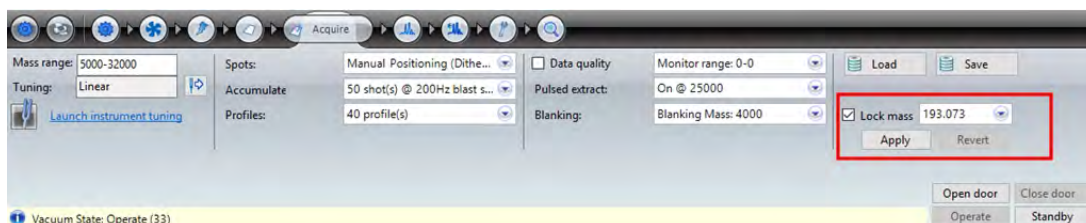
Note: There wont be complete filtering of all ions across the range below the blanked mass.



To adjust the blanking parameter, click the down arrow next to the **Blanking** field and enter the required **Blanking mass** value in Daltons, or select **Gate off**.

Lock Mass

The lock mass function is accessed from the Acquire tab and allows the user to specify a mass to correct mass drifts. This allows the user to compensate for systemic mass drifts between calibrations and allow the spectra to obtain higher mass accuracy. The specified lock mass value, accepted tolerance level and lock mass method are entered in the lock mass parameters. Lock mass is applied to the spectrum either automatically at the end of the acquisition or it can be applied manually to an already acquired spectrum.



Lock Mass Parameters

To set the lock mass parameters, perform the following:

1. Select the **Lock Mass** option.
2. Click the lock mass drop-down menu.
3. In the Lock Mass field, enter the mass value.
4. Enter the tolerance level value and select one of the following corresponding units from the drop-down menu:
 - Da - To apply the tolerance levels using Daltons.
 - mDa - To apply the tolerance levels using Millidaltons.
 - ppm - To apply the tolerance levels using part per millions.

NOTE: By default, if the tolerance level is left blank, a 1 Da value is applied.

5. Lock mass is a method of moving a peak of a known mass to its theoretical mass in the spectrum. Both of the provided lock mass methods do this, but they differ in how they scale the rest of the spectrum. From the Method drop-down menu, select one of the following correction methods:
 - MassCorrection - The mass correction method uses a proportional shift in the masses after the calibration is applied. That is, peaks that are further away from the lock mass peak will be shifted more and vice versa.
 - TimeCorrection - The time correction method uses a linear shift in time, before the calibration is applied. This correction will only provide good performance in the region close to lock mass peak (roughly within 10% of the mass), as the mass actually has a quadratic dependence on time, not linear.
6. To use the selected lock mass parameters, click **Apply**. The Lock Mass parameters are applied to all the displayed spectra in the Acquire tab when a peak is found within the tolerance range at the specified mass.
7. Click **Revert** to roll back the Lock Mass parameters and return the spectra to the previous calibration.

NOTE: When Lock Mass is applied post acquisition, the dataset must be (re)saved in order to retain the lock mass correction.

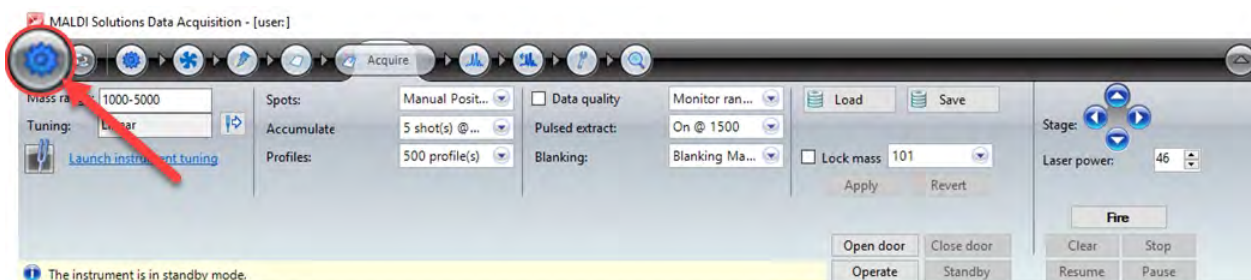
Annotation

This topic provides details on how to customise annotation regions and how to use them on an acquired spectrum.

Annotation region settings

The annotation region is set up using the **Settings** menu option within the Data Acquisitions software.

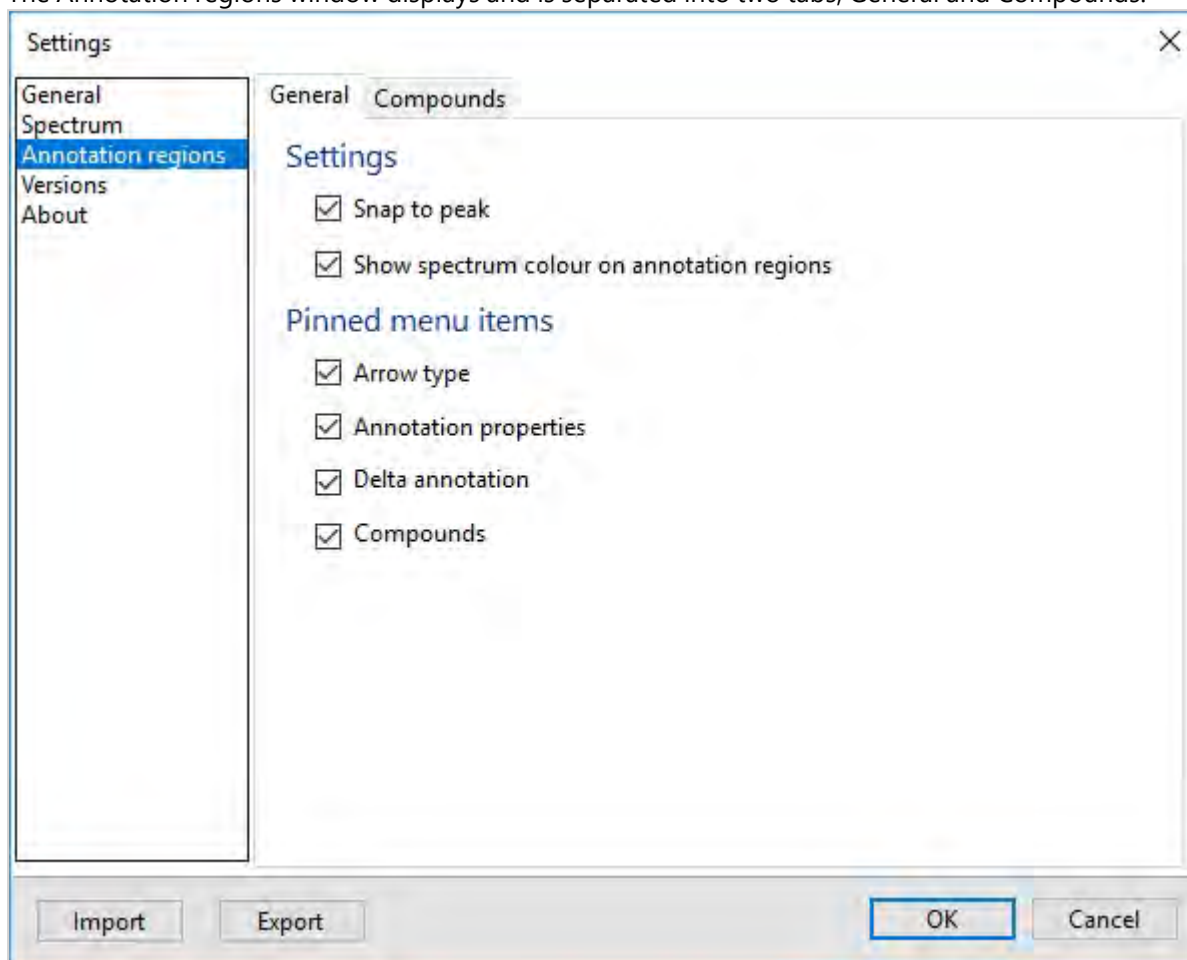
1. Click the **Settings** icon.



The Settings window displays.

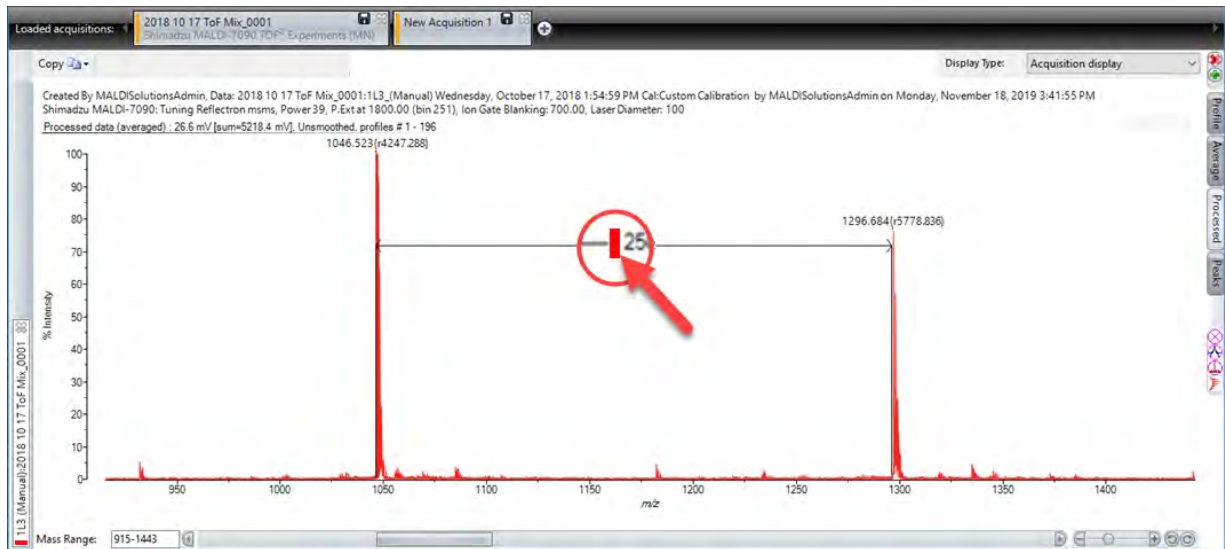
2. Select **Annotation regions**.

The Annotation regions window displays and is separated into two tabs; General and Compounds.



The General tab is split into two regions, Settings and Pinned menu items.

3. In the Settings region, select the **Snap to peak option** to make peaks the starting point for annotation comments.
4. The **Show spectrum colour on annotation region** option, when selected, applies a coloured tag to the annotation which matches the spectra colour. For example, if you have a spectrum in red and you add an annotation to it, that annotation will be identified by a red tag.

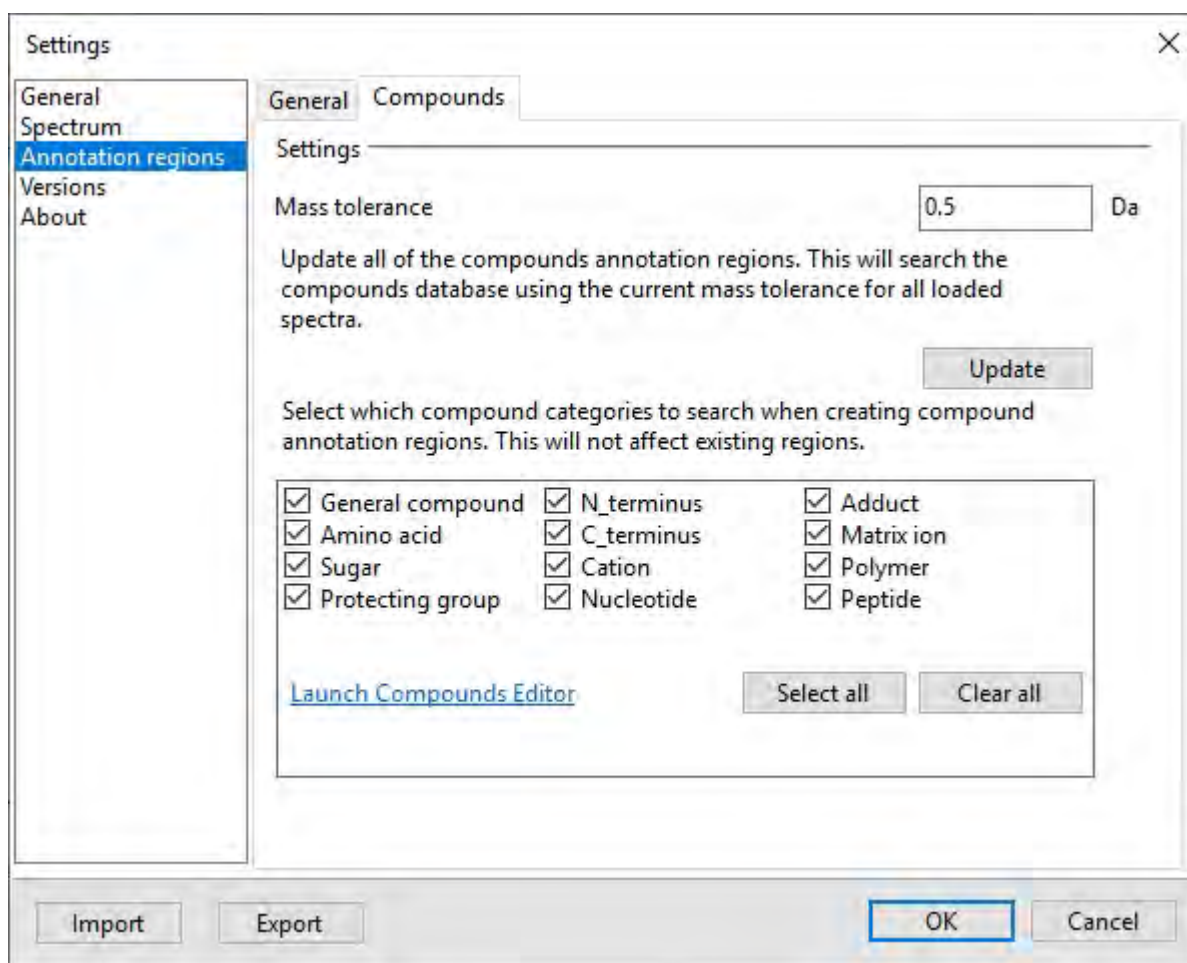


5. In the Pinned menu items region, select which annotation tools are to be made available from the Annotation Toolbar when applying annotation to a spectrum:

- Arrow type
- Annotation properties
- Delta annotation
- Compounds.

To configure the compound settings, do the following:

6. Click the **Compounds** tab.



7. In the Mass tolerance field, set the Mass tolerance value in Daltons.
8. Identify which compound categories to search when creating annotation regions by select one or more compounds.
9. If required, click Launch Compounds Editor to manually edit the compound values

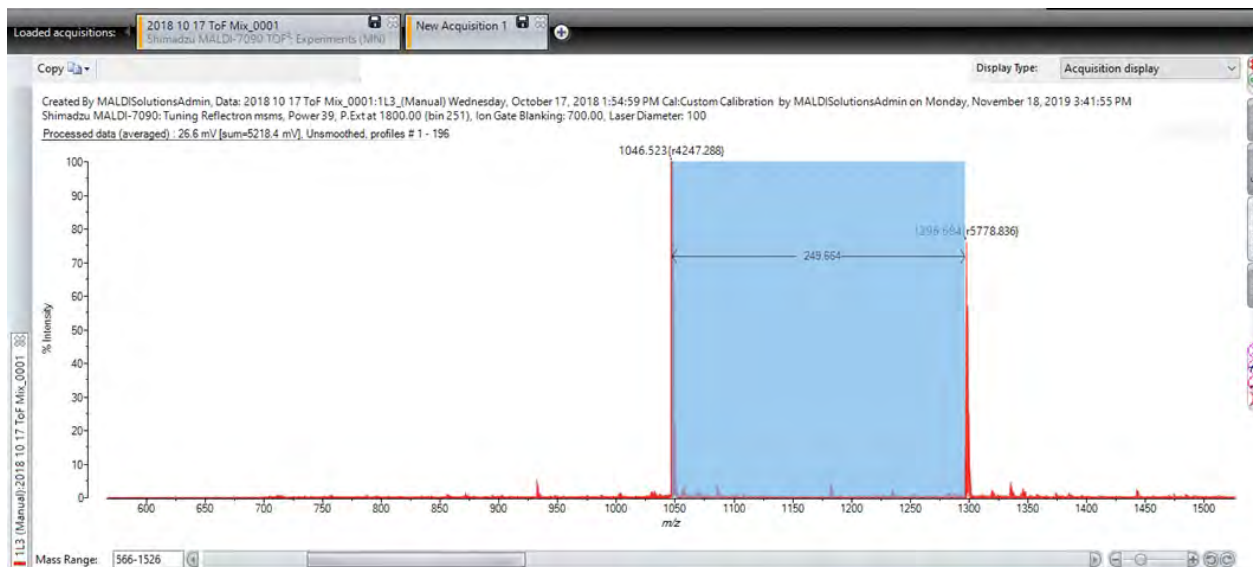
When the settings are specified, click **OK** to apply the changes and close the Settings window.

Annotating Peaks

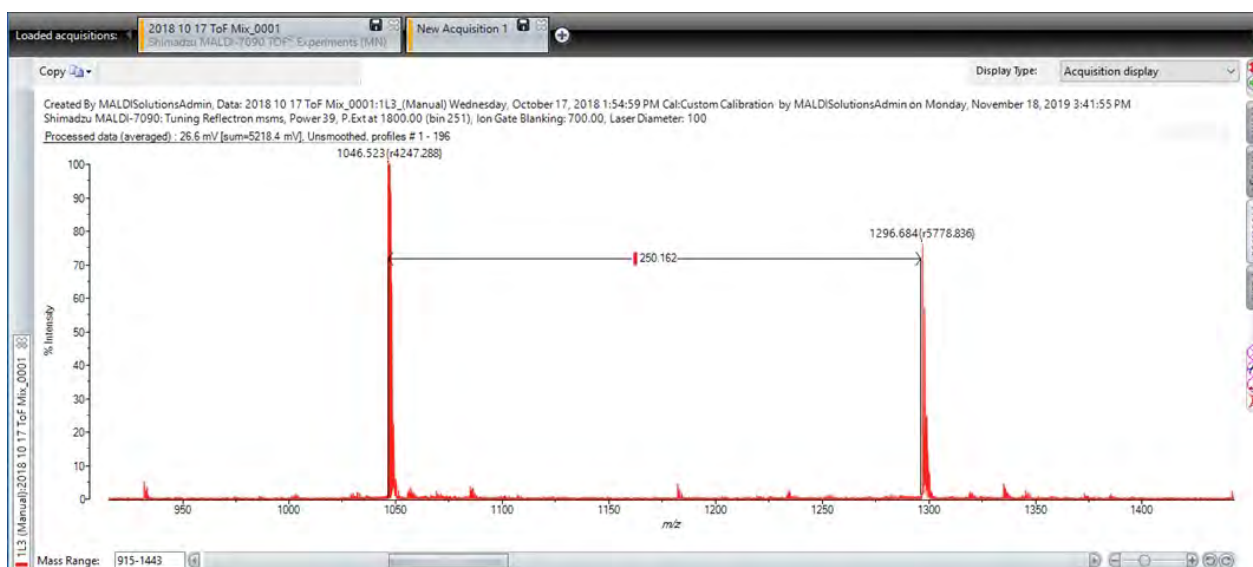
The following section provides details of how annotations are added to a spectrum when data has been acquired.

To annotate a peak, do the following:

1. In an acquired acquisition, hover the mouse cursor over the peak to be annotated and hold down the **Ctrl** key. The selected peak will blink.
2. Using the mouse, click on the peak and drag the cursor along the spectrum and release the mouse button.

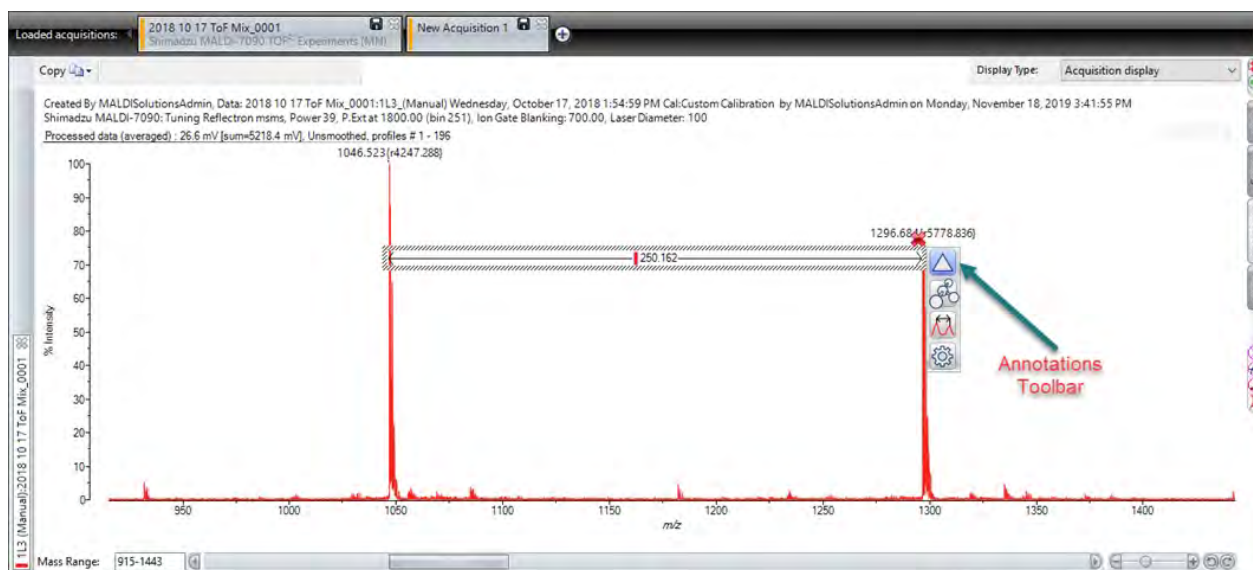


The annotation shows the mass covering the length of the line as it is pulled along. If the annotation tool is set to display compounds, the line will suggest the names of the compounds as it passes through the masses of the enabled compounds in **Compounds Editor**.



3. When the pointer reaches a peak and an expected compound ID appears, release the mouse button to apply the annotation.
4. When the annotation area is set, press the **Ctrl** key and double-click on the annotation box.
5. With the annotation highlighted and the **Ctrl** pressed, the user can move and stretch the box or access the Annotations toolbar.
6. To access the toolbar, with the Ctrl button pressed, hover over the top right hand side of the annotations box.

The Annotation Toolbar displays.



7. The Toolbar provides the following options:



The Delta Mass option sets the value to display only the difference in mass.



The Molecule option sets the tool to display compounds stored in the Compound Editor tool.

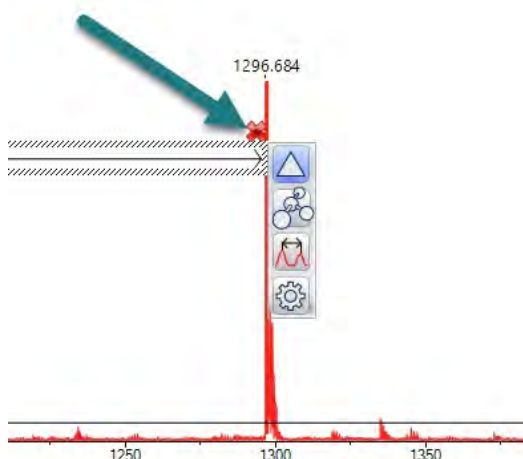


The Peak Top tool changes how the annotation regions are displayed. Click on the option to amend how the annotation lines display.



The Annotation region properties tool allows users to customise items such as the text colour, arrow type and how the annotation region is to be applied.

8. To remove annotations, select the annotations box and click the cross.



Working with spectra

Each acquired or loaded spectrum is presented in a separate window (the acquisition pane) and is accessed by clicking on the relevant tab on the **Loaded acquisitions** bar. The acquisition pane contains a number of controls which allow you to work with the spectrum in different ways.

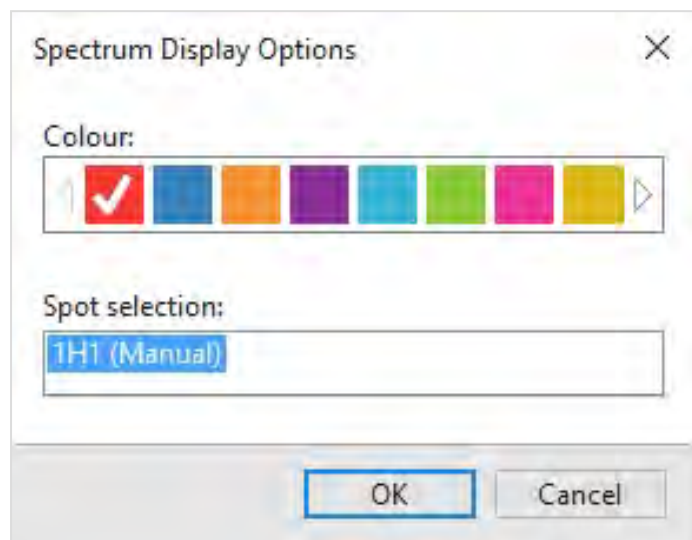


Using acquisition tabs

- Left-click on an acquisition tab to select the desired acquisition pane. If there are more tabs than can be displayed on screen, use the scroll arrows to move through the list of tabs.
- Hover over the tab to display the pop-up **Acquisition information** box which displays a summary of the acquisition attributes.
- To add another tab, follow the procedure for loading an acquisition (see [Loading & saving data](#)).
- To remove a tab, click the **Close tab** control or right-click on the tab and then select **Unload spectrum** from the pop-up menu.
- Use the scroll bar to scroll the currently selected spectrum left/right.
- Use the zoom control to magnify or shrink the currently selected spectrum. You can also zoom in/out using the spectrum controls and clicking/dragging with the left mouse button (see [Zooming on peaks](#)).
- Enter an upper and lower value into the **Mass Range** field to zoom in to a specific mass range.
- Use the undo/redo buttons to undo any changes you have made to the display.
- Use the Copy control to copy either the currently selected spectrum or all displayed spectra to the Windows clipboard. This will create a graphic (in Windows Metafile format) you can paste into other applications.
- Right-click on the tab to display a pop-up menu with the following options:

Add spectrum to display

Adds a spectrum which is displayed in the same acquisition pane. You can add multiple spectra to the acquisitions pane, and switch between them on the displayed acquisitions bar. When you select this option, you can choose the spectrum colour and spot selection (if multiple spots were included in the spectrum).



To remove a tab from the displayed acquisitions bar, right-click and then select **Remove** from the pop-up menu, or click the **Close** control. You can also change the display options from the pop-up menu.

Rename spectrum

Allows you to respecify the attributes and permissions for the currently selected spectra. This procedure is identical to that used when saving an acquisition (see [Loading & saving data](#)).

Save acquisition

Saves the currently selected acquisition to the database (see [Loading & saving data](#)).

Export acquisition

Exports one or more loaded acquisitions to an external XML format file (see [Loading & saving data](#)).

Batch export

Exports all loaded acquisitions to an external mzML format file (see [Loading & saving data](#)).

Unload acquisition

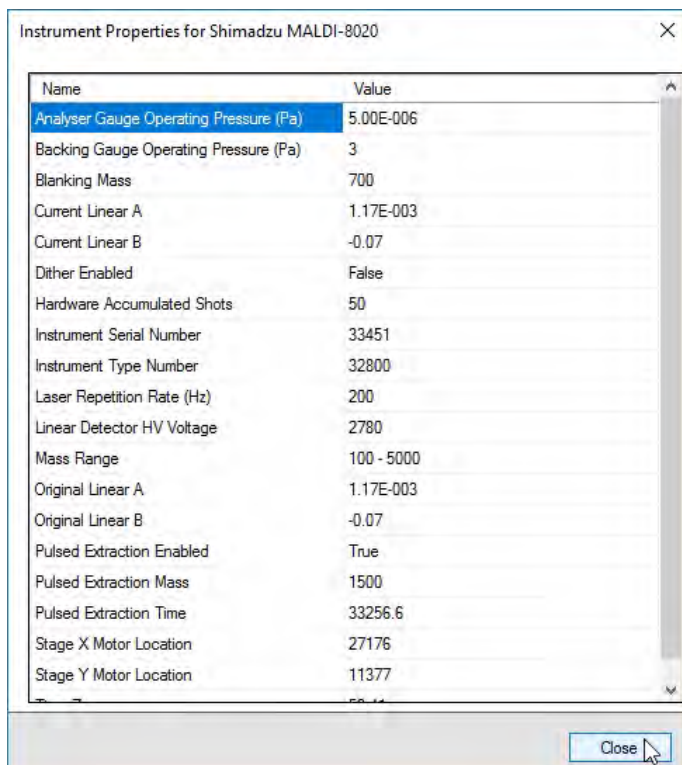
Removes the currently selected acquisition from the **Loaded acquisitions** bar.

Unload all

Removes all currently loaded acquisitions from the **Loaded acquisitions** bar.

Instrument properties

Displays the parameters used by the instrument to perform the currently selected acquisition.

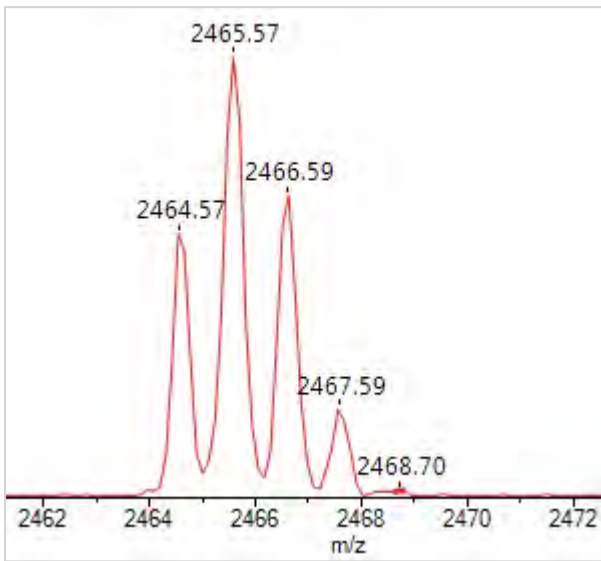


Click **Close** to dismiss the dialog.

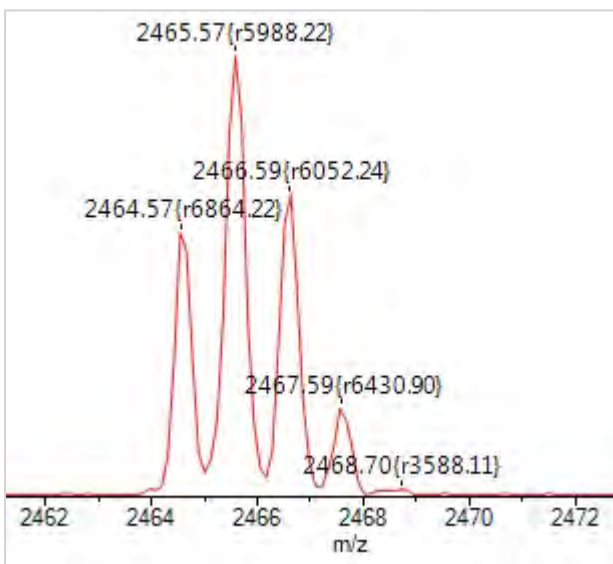
Using the peak label controls


Assuming that the peak processing parameters are set correctly, in the processed spectrum, the peaks are automatically labelled. You can change the way added labels are displayed on a spectrum via the peak label controls as described below.

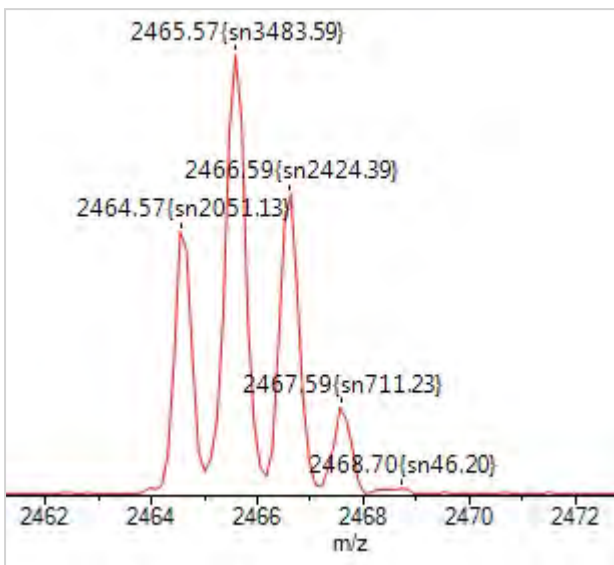
⊗ Click **None** to show only masses (default).




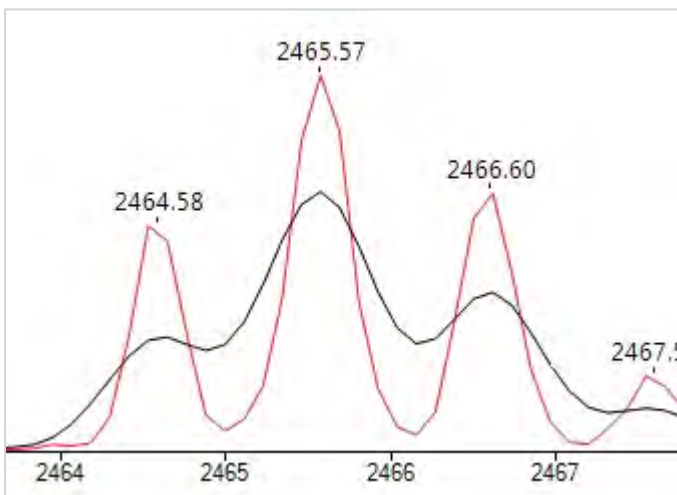
⊗ Click **Resolution** to show masses and resolution values.



 Click **SignalToNoise** to show masses and SNR values.

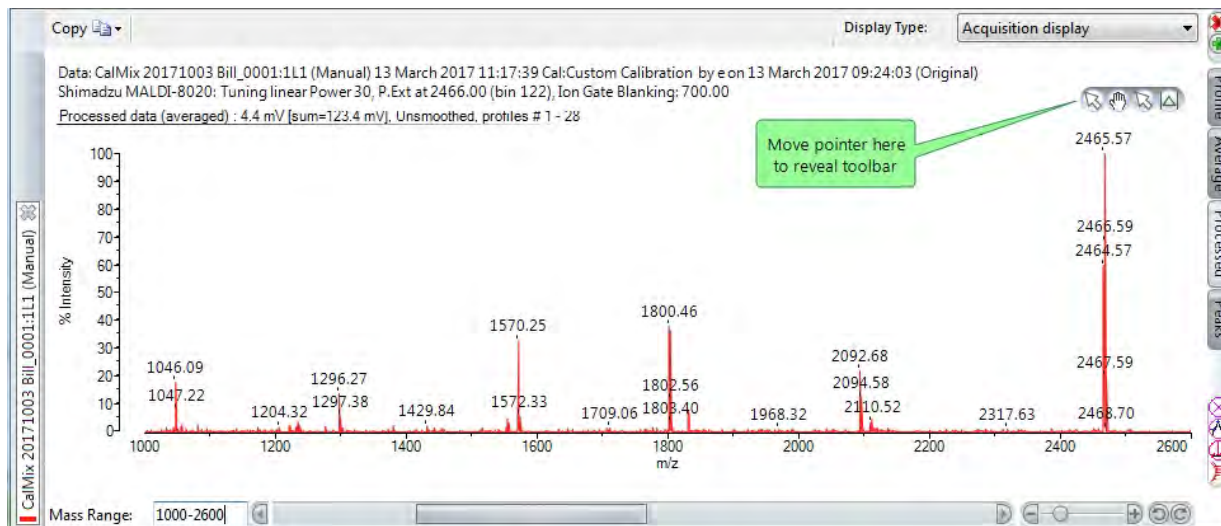


 Click **Threshold** to add threshold plot data to the spectrum (only applicable if you set the **Threshold** type in the **Process** tab).



Using the spectrum controls

The display has a toolbar in the top right-hand corner that becomes visible when you move the pointer closer to it.

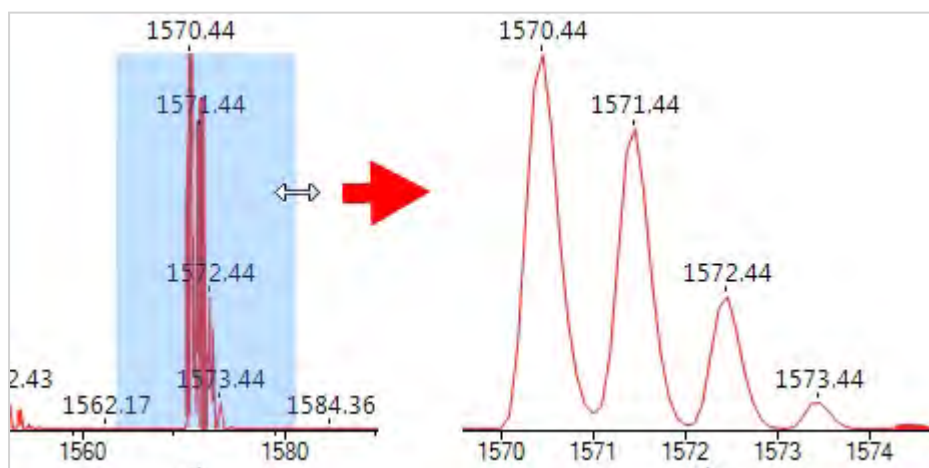


You can use the spectrum controls toolbar to drag, zoom or display information on selected peaks, as well as set the gain region and change the way peak labels are displayed.

	Peak tool - click and drag over a spectrum to zoom, for example, to examine peaks.
	Drag tool - click and drag to scroll the spectrum.
	Move peak label tool - click a peak mass label and drag it to the required location.
	Define gain regions tool - click and drag the required region and set the required magnification.

Zooming on peaks

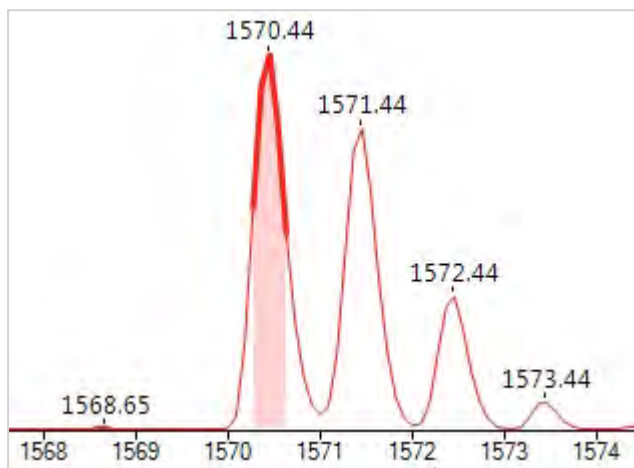
1. Click the **Peak** tool on the spectrum controls toolbar.
2. Move the pointer to the start of the zoom.
3. Click and hold the left mouse button and drag the pointer to the end of the zoom:



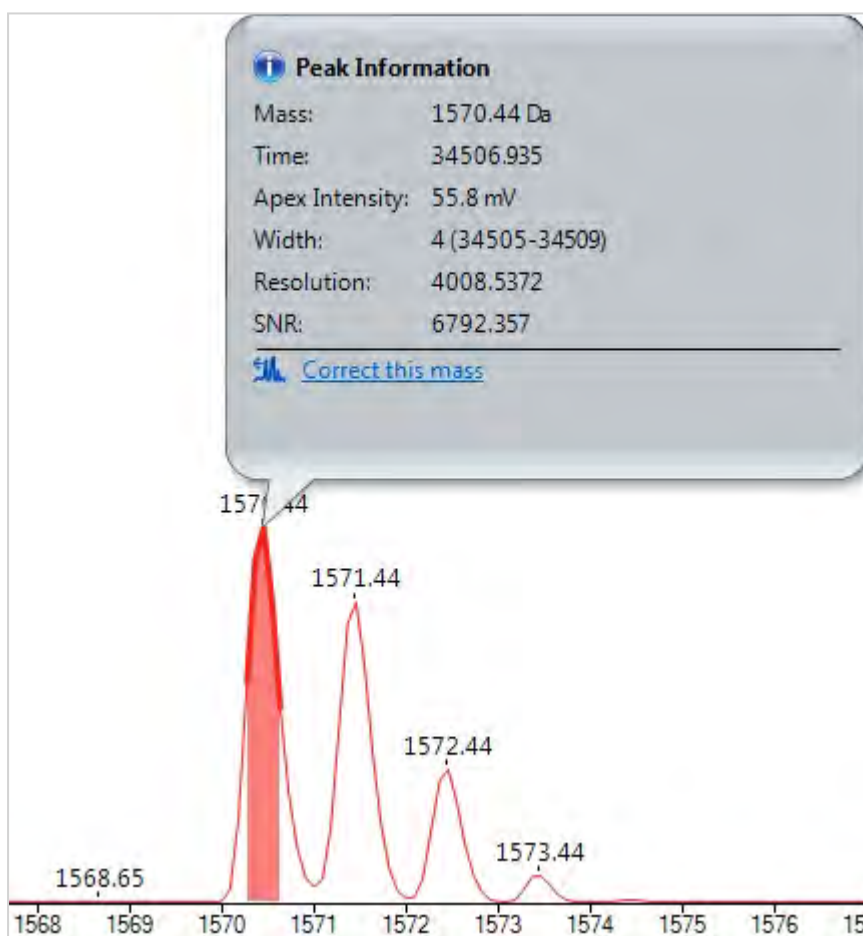
4. Release the mouse button; the spectrum zooms on the selected area.
5. Repeat the zoom to get the required display.

Displaying peak information

1. Click the **Peak** tool on the toolbar:
2. To highlight the area of a peak, move the pointer over the peak:



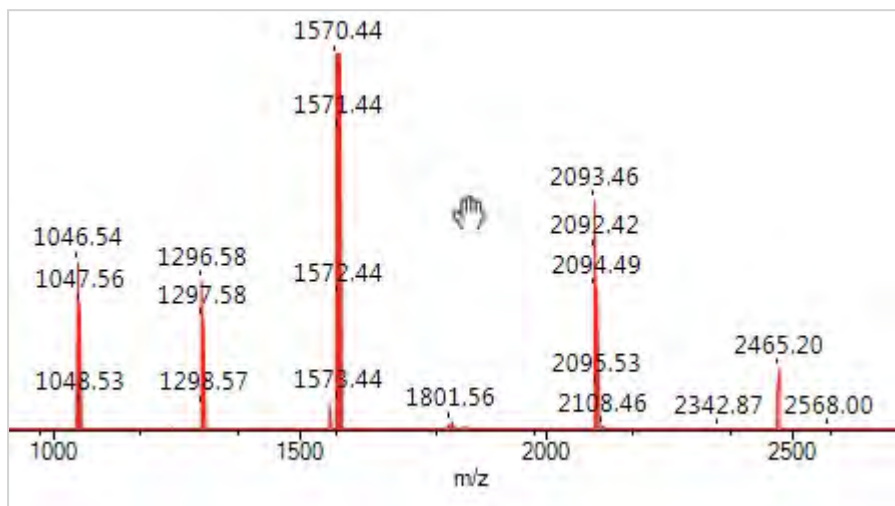
3. Click on the required peak; information about the peak is displayed:



For details about correcting the peak mass, see [Calibration](#).

Dragging the spectrum

1. Click the **Drag** tool on the toolbar; the mouse pointer changes to a hand.

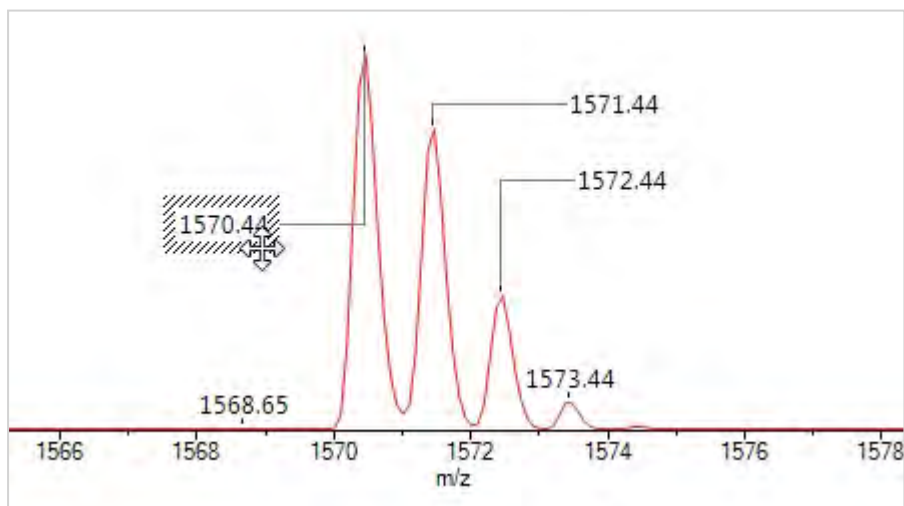


2. Press and hold the left mouse button.
3. Drag the spectrum to the required location and release.
4. To switch off the drag, click the **Peak** tool on the toolbar:

Moving peak labels

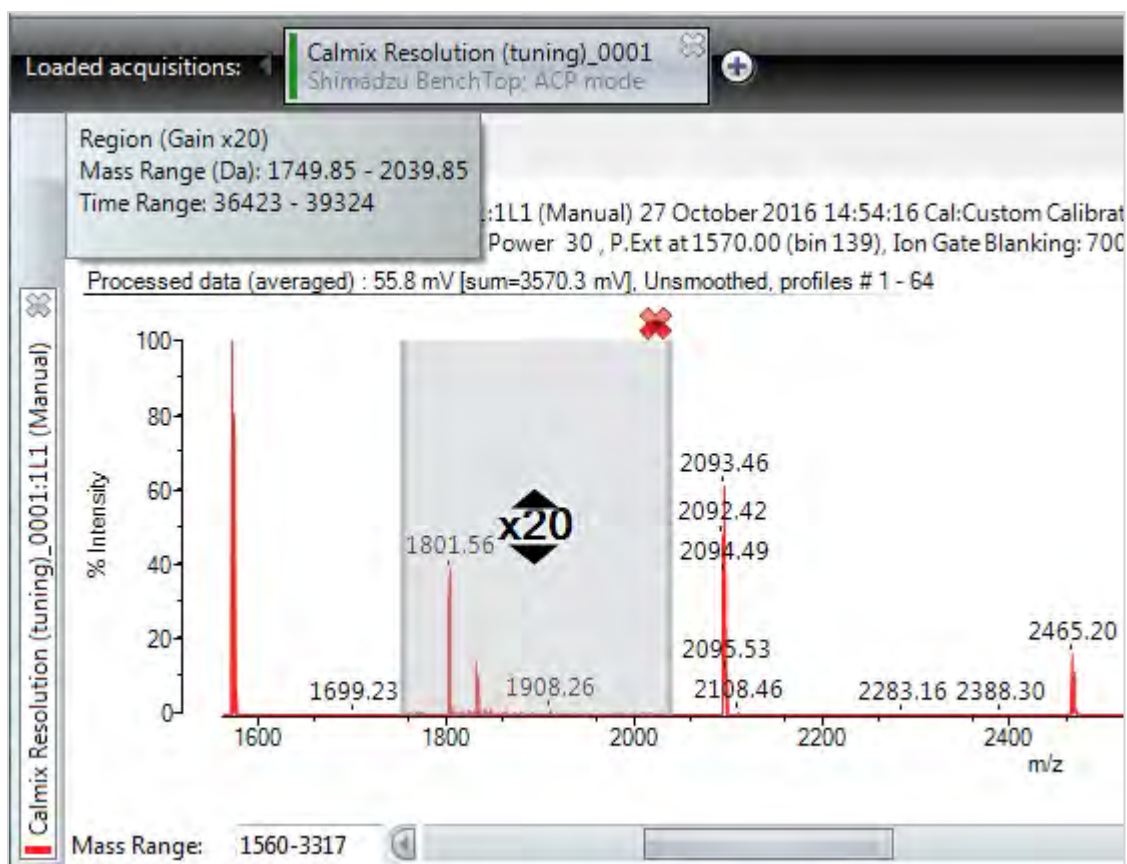
If, for example, a peak label is obscuring the spectrum, you can move the label; a dotted line references the peak.

1. Click the **Move** peak label tool on the toolbar:
2. Click the required spectrum label; it is bounded by a striped box.
3. Click and drag the label (pointer changes to an arrow-tipped cross) to the required location:




4. Repeat for any other labels.
5. To switch off, click the **Peak** tool on the toolbar:

Defining gain regions



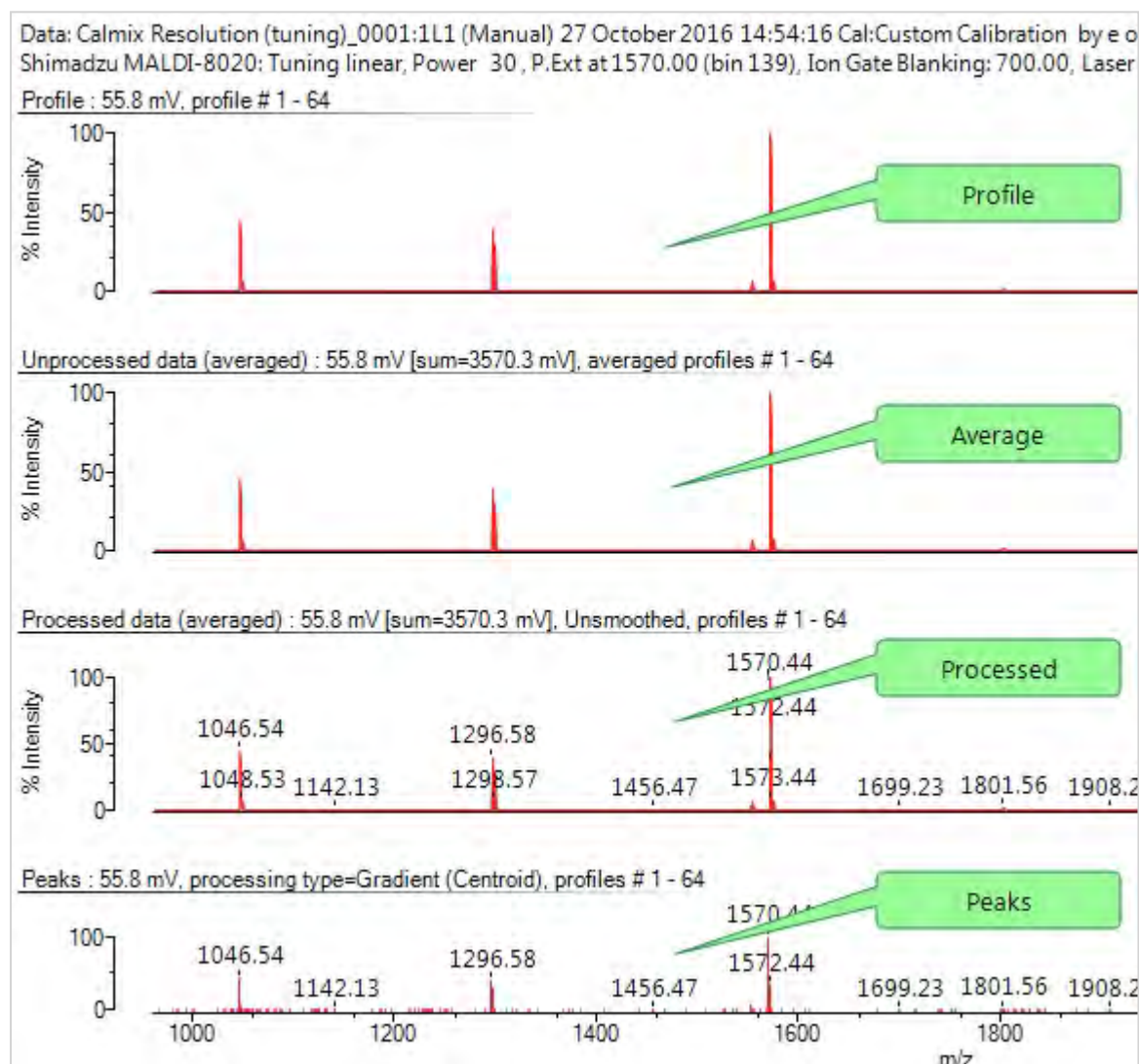
This feature allows you to select a mass range and increase its signal amplitude. You can specify several non-overlapping gain regions for a spectrum.

1. Select the **Define gain regions** tool from the toolbar:
2. Click and drag the mouse pointer over the required region; the display will mark the selected region with a vertical bar at each end and a fill-colour between the bars.
3. Click the up and down arrows to alter the gain of the region. You can set: $\times 1$, $\times 2$, $\times 5$, $\times 10$, $\times 20$, $\times 50$, $\times 100$... $\times 5,000$.
4. Hover the mouse pointer over the region, the tooltip contains information about the range including start and end points in mass (Da) or time range (bins).
5. Adjust the width of the region by dragging the sides.
6. To remove the gain region either:
 - Click the remove icon .
 - Right-mouse click and select either **Delete region** or **Delete all regions** from the pop-up menu.

Using the processing controls

This feature allows you to view multiple viewpoints on the loaded data. You can also view several different mass ranges simultaneously. Select one or more tabs to add a view to the displayed spectrum. The **Profile** and **Processed** views are most common used.

Profile	Displays data collected from the sample for the last profile. Note: Scroll through profiles (using arrow button on handle at top of spectrum) if all profiles were saved during acquisition
Average	Displays the average of all the profiles collected from the sample.
Processed	Displays the averaged data after the application of any smoothing, baseline subtraction, and peak detection to the data.
Peaks	Displays the centroid/apex mass peaks found in the processed data as sticks.

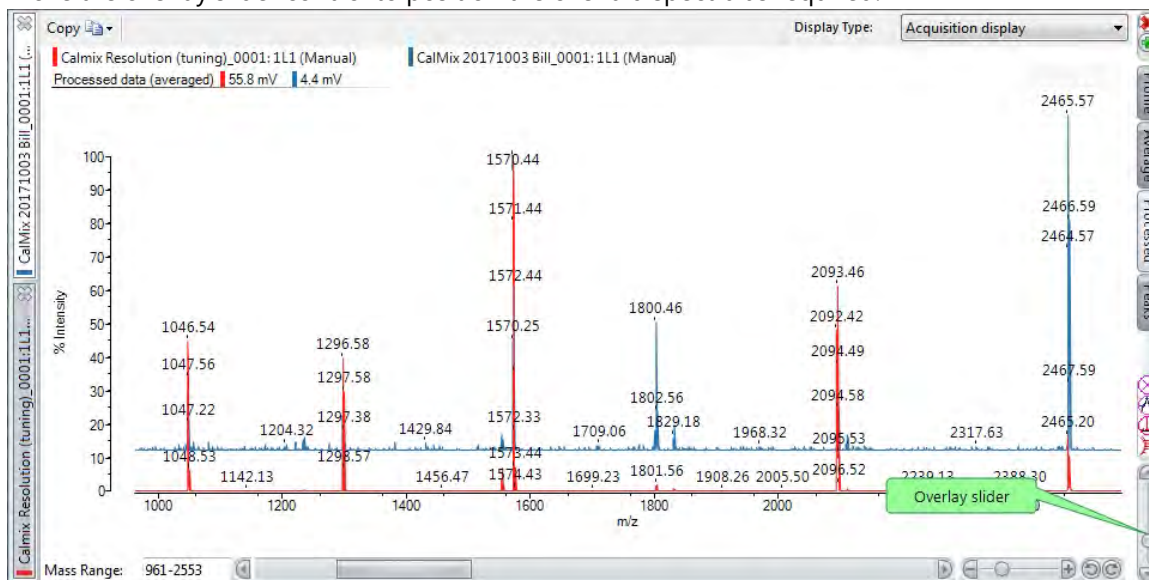


Comparing spectra

Using overlays

This feature allows you to display several spectra and overlay them so that you can compare them.

1. Load each acquisition containing the spectra you wish to compare.
2. Move the overlay slider control to position the overlaid spectra as required.

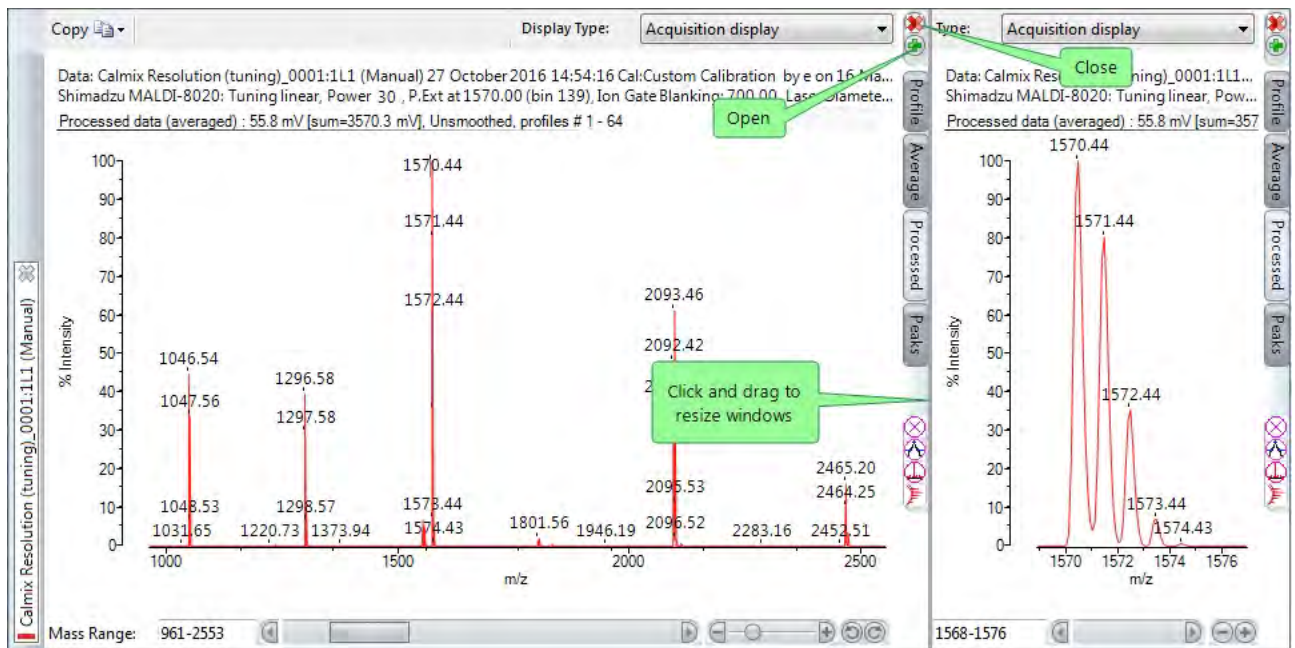


Using split screen controls

Split screen controls allows you to, for example, focus on specific peaks while keeping the main spectrum visible.

1. To add a column, click the split screen icon ; the display divides into two columns (realistically you can display up to 3 columns).
2. To remove a column, click the close screen icon.

3. To resize each column, drag the splitter bar between each column.



Displaying mass lists

This feature allows you to display a mass list for the acquisition being displayed. To display the mass list, select **Mass list** from the Display Type drop-down menu.

Copy Limit maximum peaks by intensity Update Display Type: **Mass list**

Data: Calmix Resolution (tuning)_0001:1L1 (Manual) 27 October 2016 14:54:16 Cal:Custom Calibration by e on 16 March 2017 12:51:07
Shimadzu MALDI-8020: Tuning linear, Power 30, P.Ext at 1570.00 (bin 139), Ion Gate Blanking: 700.00, Laser Diameter: -2147483648

Total peaks in spectrum: 265 Displayed peaks in table: 265 Hidden peaks: 0

Select columns Export mass list

Mass (Da)	Intensity (mV)	Intensity (%)	Area (mV)	Area (%)	Resolution	S / N
209.08	0.02	0	0.05	0	2097	0
382.1	0.03	0	0.08	0	4725	1
382.59	0.03	0	0.08	0	2562	1
383.67	0.03	0	0.07	0	3788	1
384.06	0.02	0	0.08	0	2369	0
719.81	0.01	0	0.02	0	9338	0
742.93	0.08	0	0.22	0	4833	5
745.35	0.02	0	0.06	0	9503	0

Limiting the number of rows

Tick the **Limit maximum peaks by intensity** check box, enter the required value, and click **Update**.

Copy Limit maximum peaks by intensity 10 Display Type: Mass list

Data: Calmix Resolution (tuning)_0001:1L1 (Manual) 27 October 2016 14:54:16 Cal:Custom Calibration by e on 16 March 2017 12:51:07
Shimadzu MALDI-8020: Tuning linear, Power 30, P.Ext at 1570.00 (bin 139), Ion Gate Blanking: 700.00, Laser Diameter: -2147483648

Total peaks in spectrum: 265 Displayed peaks in table: 10 Hidden peaks: 0

Select columns

Mass (Da)	Intensity (mV)	Intensity (%)	Area (mV)	Area (%)	Resolution	S / N
1046.54	24.92	45	122.94	58	2327	1899
1047.56	18.74	34	87.44	41	2486	1426
1296.58	22.09	40	93.61	44	3517	4431
1297.58	16.45	29	67.99	32	3682	3643
1570.44	55.79	100	213.07	100	4009	6792
1571.44	44.72	80	170.26	80	4059	4918
1572.44	19.74	35	72.62	34	4347	1979
2092.42	26.76	48	111.1	52	3365	1099
2093.46	34.08	61	145.99	69	3225	1410
2094.49	20.88	37	89.77	42	3105	870

Selecting which columns are displayed

Select the required columns from the **Select columns** drop-down list:

Select columns

- Mass (Da)
- Intensity (mV)
- Intensity (%)
- Area (mV)
- Area (%)
- Resolution
- S / N

Hiding peaks on the mass list

1. Select one or more rows in the list, and then right-click on the selection.

Note: You can select multiple rows using Shift to select a range, or Ctrl to select individual entries.

Total peaks in spectrum: 265 Displayed peaks in table: 10 Hidden peaks: 0

Select columns

Mass (Da)	Intensity (mV)	Intensity (%)	Area (mV)	Area (%)	Resolution	S / N
1046.54	24.92	45	122.94	58	2327	1899
1047.56	18.74	34	87.44	41	2486	1426
1296.58	22.09	40	93.61	44	3517	4431
1297.58			67.99	32	3682	3643
1570.44			213.07	100	4009	6792
1571.44			170.26	80	4059	4918
1572.44	19.74	35	72.62	34	4347	1979
2092.42	26.76	48	111.1	52	3365	1099
2093.46	34.08	61	145.99	69	3225	1410
2094.49	20.88	37	89.77	42	3105	870

Hide selected peaks
Reset hidden peaks

2. Select **Hide selected peaks** from the pop-up menu. The list is updated and a count of the hidden entries added to the display.

Total peaks in spectrum: 265 Displayed peaks in table: 8 Hidden peaks: 2

Select columns

Mass (Da)	Intensity (mV)	Intensity (%)	Area (mV)	Area (%)	Resolution	S / N
1046.54	24.92	45	122.94	58	2327	1899
1047.56	18.74	34	87.44	41		1426
1296.58	22.09	40	93.61	44		4431
1570.44	55.79	100	213.07	100		6792
1571.44	44.72	80	170.26	80	4059	4918
2092.42	26.76	48	111.1	52	3365	1099
2093.46	34.08	61	145.99	69	3225	1410
2094.49	20.88	37	89.77	42	3105	870

Hidden peak count displayed

Export mass list

See [Loading & saving data](#).

Copy mass list

You can use the **Copy** tool to copy the mass list to the Windows clipboard as either a tab-separated data file (for spreadsheets), or an image metafile for inclusion in documents.

The screenshot shows the 'Copy' menu with three options: 'Copy this display (tab separated)', 'Copy this display (metafile)', and 'Copy all displays (metafile)'. The 'Display Type' is set to 'Mass list'. Below the menu, there is a table with the following data:

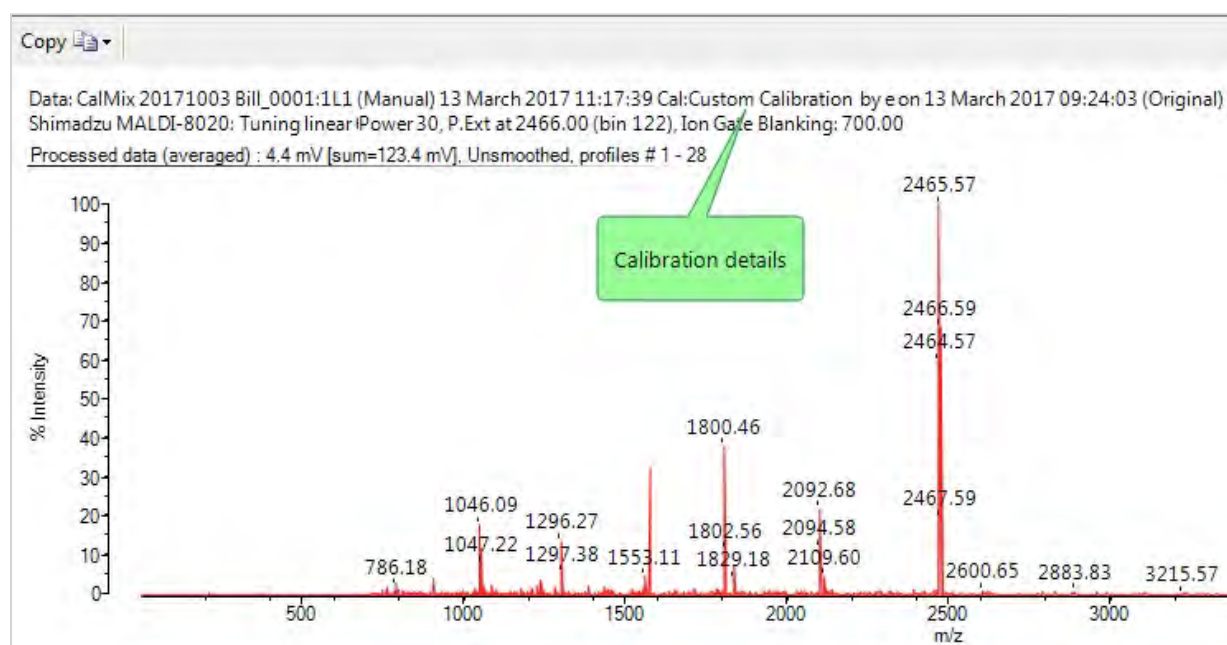
Mass (Da)	Intensity (mV)	Intensity (%)	Area (mV)	Area (%)	Resolution	S / N
1046.54	24.92	45	122.94	58	2327	1899
1047.56	18.74	34	87.44	41	2486	1426
1296.58	22.09	40	93.61	44	3517	4431
1297.58	16.45	29	67.99	32	3682	3643
1570.44	55.79	100	213.07	100	4009	6792
1571.44	44.72	80	170.26	80	4059	4918
1572.44	19.74	35	72.62	34	4347	1979
2092.42	26.76	48	111.1	52	3365	1099
2093.46	34.08	61	145.99	69	3225	1410
2094.49	20.88	37	89.77	42	3105	870

Calibration

Calibrating a spectrum ensures that the mass scale and peak intensities of a spectra are displayed correctly, by comparing the acquired data with a known set of reference values. The primary calibration types used with the instrument are:

- Rolling calibration - this is the default calibration supplied with the instrument which will be used if no other calibration type is selected. If you make changes to the rolling calibration and then save it, the new parameter settings will be applied whenever any user acquires data using the same tuning mode.
- Named calibration - a calibration which has been created by a user and saved to the instrument database. All users can load and use this type of calibration.
- Preferences calibration - If you change a calibration, it is automatically saved and stored with your user account preferences. All subsequent acquisitions will use this calibration unless you specify another.

When you save an acquisition, the currently applied calibration is saved with the data. Calibration details are displayed in the spectrum header display.



Applying a calibration

1. Acquire a spectrum (see [Acquiring a spectrum](#)) - this will apply a rolling calibration based on the tuning parameters you set for the acquisition (see [Setting acquisition parameters](#)).
2. Select the **Calibrate** tab - this will set the current preferences calibration to the same settings used in the previous step. The calibration data will be applied to the currently loaded acquisition, and to all future acquisitions until you load another calibration or change the tuning parameters settings.
3. If necessary, make any required changes to the calibration (see [Using calibration references](#) below).
4. To save the current calibration as a named calibration, click **Save** and then select **Save a named calibration** (see [Loading & saving data](#)).

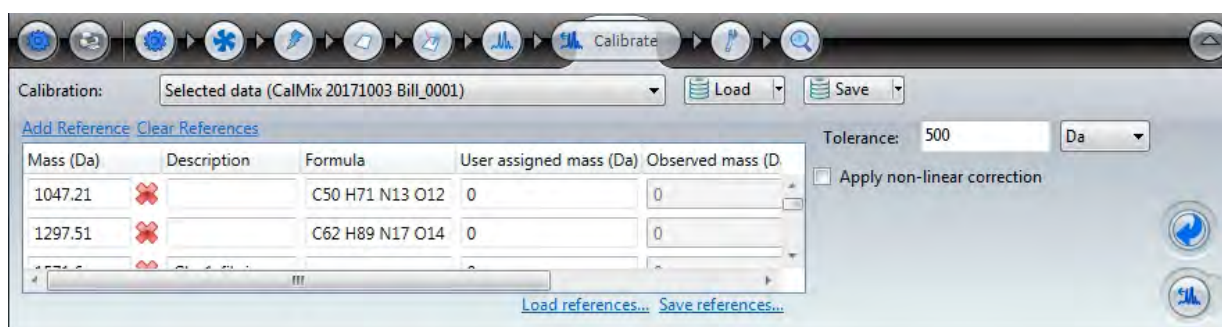
- To save the current calibration as the default rolling calibration for the selected tuning parameter set, click **Save** and then select **Save to the rolling calibration**.

Note: If a named or rolling calibration is changed, saved acquisitions using that calibration are not updated automatically. However, you can open the acquisition and load the revised calibration.

- To apply a different calibration, click **Load** and then select **Load a named calibration**, **Load the current rolling calibration** or **Load original calibration**. In the latter case, the calibration data which was last saved with the acquisition will be re-applied.

Using calibration references

A calibration reference is a list of reference masses with optional fields for a description and formula. You can add, delete or amend references within the list. Calibration references are associated with the current calibration, but may be loaded or saved independently if required.

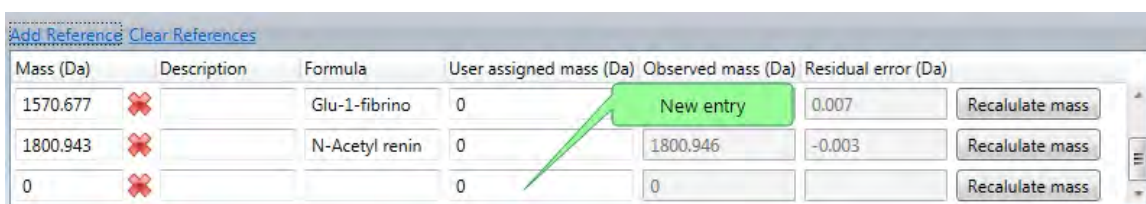


Loading & saving calibration references

See [Loading & saving data](#).

Adding a reference


- On the **Calibrate** tab, click the **Add Reference** link to add a new (blank) entry.



- Fill in the **Mass (Da)** (expected m/z) and optional **Description** and **Formula** fields.

Note: If you entered a formula in the **Formula** field, click the **Recalculate mass** button to automatically enter the expected m/z in the **Mass (Da)** field.

Deleting a reference

- To delete a single reference, in the row for the selected mass, click the delete  button.
- To delete all references, click the **Clear references** link.

Amending a reference

In the row for the selected reference, amend the **Mass (Da)** (expected m/z) and optional **Description** and **Formula** fields.

Note: If you entered a formula in the **Formula** field, click the **Recalculate mass** button to automatically enter the expected m/z in the **Mass (Da)** field.

Creating a calibration

If there are no suitable calibrations available for your acquisition, you can create an entirely new calibration.

Note that when selecting calibration references, these should:

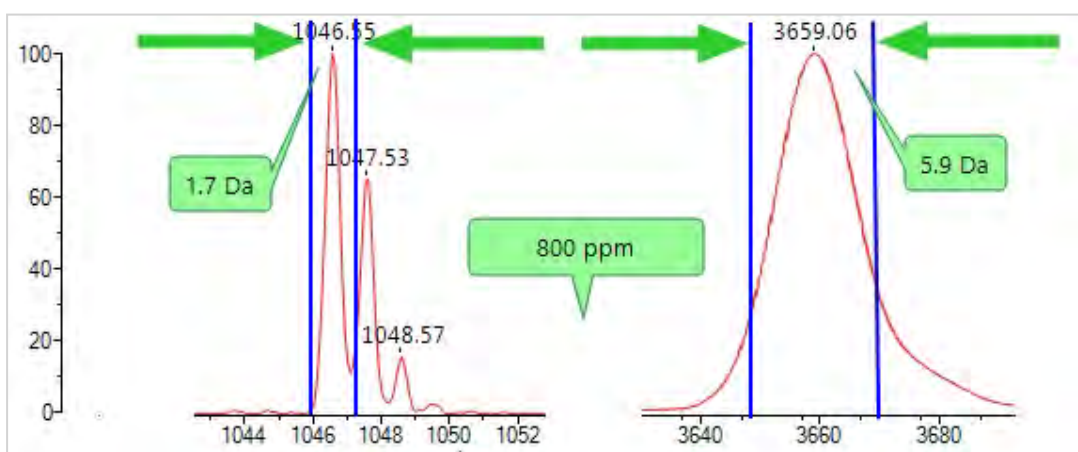
- Cover the whole range of masses you expect from your acquisition;
- Use the same matrix that you will use for the acquisition;
- use the same pulsed extraction value as used in the acquisition parameter settings.

To create a calibration:

1. Acquire a spectrum from a sample containing all the references you wish to include in the calibration (see [Acquiring a spectrum](#)).
2. On the **Calibrate** tab, load or add the required references to the displayed list (see [Using calibration references](#)).
3. In the **Tolerance** box, enter the tolerance value, units and optionally choose the **Apply non-linear correction** option.

Notes:

- To specify absolute mass accuracy, select units of Da or mDa;
- To specify relative mass accuracy, select units of ppt (parts per thousand) or ppm (parts per million). For example, entering 800 ppm results in a tolerance at 1,000 Da of ± 0.8 Da and at 7,000 Da of ± 5.6 Da. This is useful as peaks at low masses are sharp and well defined whereas peaks at high mass are usually less well defined and a higher tolerance is more applicable:



- The instrument operates on the principle that ions are accelerated into a field-free region, and the final velocity of the ions is dependent on their mass. It is the flight times of the ions, i.e. the time from

applying the acceleration voltages to the detection of the ions, that are actually measured. The flight times (t) are converted to mass-to-charge ratios (m/z) via a calibration that uses the theoretical principle that m/z is proportional to t^2 .

However, the observed relationship between t^2 and m/z is non linear. In the mass range up to approximately 2500 Da, this can produce errors of around 50 ppm.

Therefore, to improve the mass accuracy, with internal mass calibration, you can apply a "non-linear correction factor" to correct for deviations and reduce the errors to typically less than 10 ppm. For the correction factor to work, you must have three or more calibrants.


4. Do one of the following:
 - For manual calibration, go to step 5
 - For non-manual calibration, step 9
5. In the acquisition pane, left-click on the spectral peak corresponding to the first reference mass.
6. In the **Peak information** box, click **Correct this mass** and then select the correct mass value from the list of references.

The screenshot shows the 'Peak Information' dialog box with the following data:

Mass:	1045.81 Da
Time:	28169.447
Apex Intensity:	23.8 mV
Width:	5 (28167-28172)
Resolution:	2328.5055
SNR:	998.633


The 'Correct mass' dropdown menu is open, showing the following list of reference masses:

- 1045.81 (selected)
- 1046.54
- 1296.69
- 1570.68
- 1800.94
- 2093.09
- 2465.20
- 3657.93

7. Click the tick button  - the selected reference will be updated with the **User assigned mass** value.

Mass (Da)	Description	Formula	User assigned mass (Da)	Observed mass (Da)	Residual error (Da)	
1046.54			1045.81	0		Recalculate mass
1296.69			1295.87	0		Recalculate mass
1570.68			1569.84	0		Recalculate mass

8. Repeat steps 5 and 6 for the remaining reference masses. If there are no remaining reference masses, go to step 11.

9. To perform a non-manual calibration, click **Load References** to display the mass list.
10. In the Tolerance field, enter the tolerance window value. The most intense peaks within the tolerance window are adjusted to masses in the reference list.
11. When the calibration references are complete, either click the **Calibrate now** link in the **Peak Information** window or click the **Calibrate** button ; the spectrum masses will adjust as required.




- If the calibration is successful, the button shows green:



- If the calibration is not successful, the button shows as either **amber** (passed with residuals outside tolerance) or **red** (fail):



12. If the calibration is successful, the User assigned mass fields in the reference list will be cleared, and the **Observed mass** and **Residual error** fields will be populated. You may now proceed to step 14.

Mass (Da)	Description	Formula	User assigned mass (Da)	Observed mass (Da)	Residual error (Da)	
1046.54			0	1046.56	-0.02	Recalculate mass
1296.69			0	1296.6	0.09	Recalculate mass
1570.68			0	1570.55	0.13	Recalculate mass

[Load references...](#) [Save references...](#)

13. If the calibration is not successful, click the **amber/red** area of the button to view the calibration results.

Calibration Result	
Result:	Passed
Total number of references:	6
Number of unmatched references:	0
RMS Error:	0.272 Da

Calibration Result	
Result:	Failed
Reason for failure:	One or more references have the same (or similar) times
Possible remedy:	Ensure that the you set a suitable observed mass.
Total number of references:	7
Number of unmatched references:	1
RMS Error:	0.272 Da
Largest (unmatched) difference:	989.974 Da (3456.000 Da)
Unmatched references:	3456.000 Da (3456.000 Da)

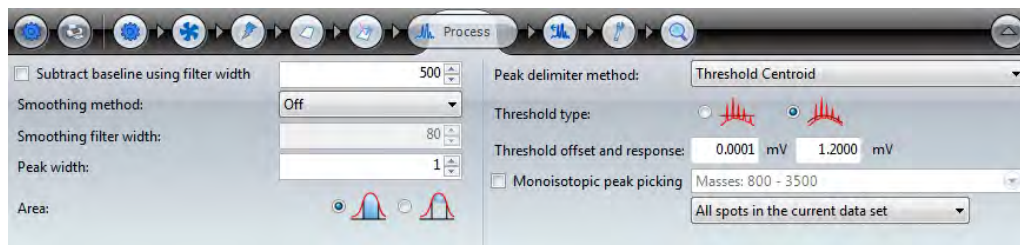
Return to step 3, increasing the **Tolerance** value so that all peaks are matched to the corresponding reference values. If the calibration succeeds, reduce the tolerance down to the required value in steps (click the **Calibrate** button between step changes). For example, set the field to 5 Da, then 1 Da, etc. For isotopically resolved peaks, set the tolerance to 500 mDa.

14. Save the calibration (see [Loading & saving data](#)).

Processing data

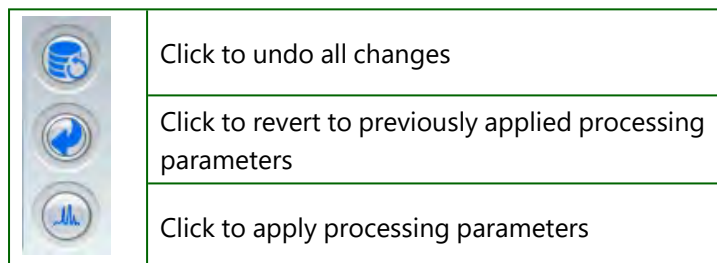
After you have acquired a spectrum, you can apply processing parameters to label peaks, subtract baseline, smooth and/or apply monoisotopic peak picking.

Click the **Process** tab to display the available processing parameters:



Applying the process parameters

You can experiment with the parameters at any time to see the effect on the spectrum. Use the following buttons to apply changes and to undo changes:

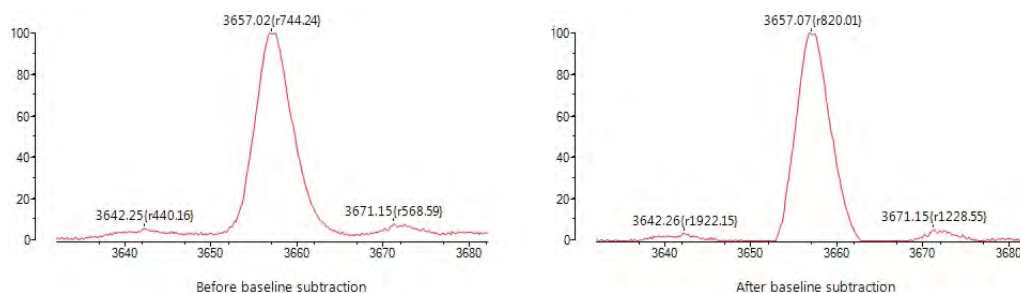


Load/save

Use the Load and Save buttons to load or save the current processing parameters to the instrument database (see [Loading & saving data](#)).

Subtract baseline using filter width

Using a high laser power and/or "chemical" noise (from the sample/matrix) can result in the spectrum not being resolved to the baseline. The **Subtract baseline using filter width** parameter allows you to move the baseline so that the spectrum rests on the x-axis, and improves the signal-to-noise ratio and resolution of the peaks.



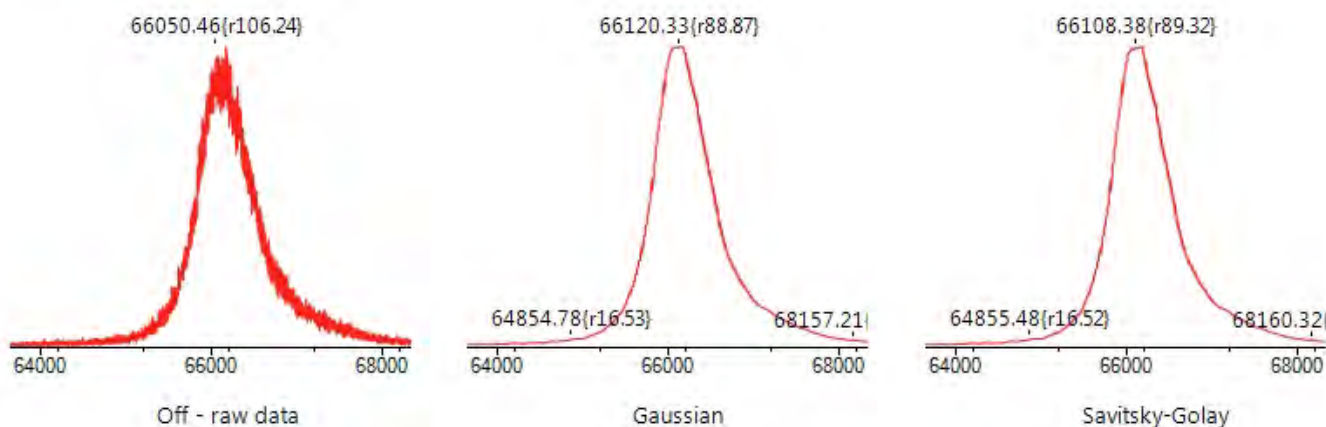
This field defines the cut-off frequency used for filtering the baseline signal (the higher the value, the lower the frequency). Use the suggested values listed below as a guide when entering this parameter value.

Application	Suggested values
Proteins (>20 kDa)	Not selected
Proteins (<20 kDa)	30 to 300
Peptides	3 to 15
Small molecule	3 to 15
Polymers	30 to 500

Select the checkbox to turn the feature on, and then enter a baseline filter width.

Smoothing method & Smoothing filter width

Smoothing reduces the "spikiness" of data caused by background electronic noise. Three smoothing methods are provided, which produce the following effects:



Gaussian

Gaussian smoothing produces a characteristic symmetric "bell curve" shape that quickly falls off towards plus/minus infinity.

Savitsky-Golay

Savitsky-Golay smoothing tends to preserve features of the distribution such as relative maxima, minima and width, which are usually "flattened" by other models.

Smoothing filter width

Use the **Smoothing filter width** field to increase/decrease the amount of smoothing. Generally, the higher the number, the greater the amount of smoothing. Use the following table for guidance:

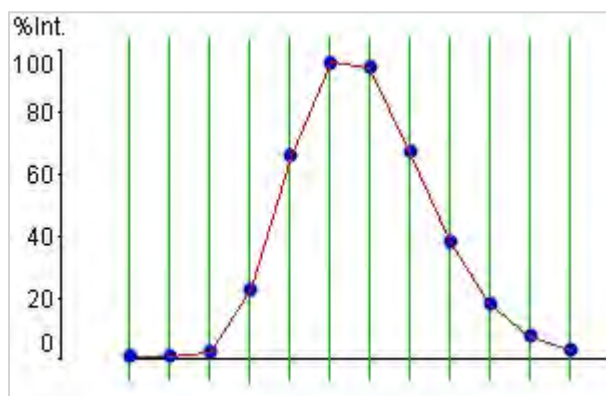
Application	Suggested values
Proteins (>20 kDa)	10 to 200
Proteins (<20 kDa)	3 to 30
Peptides	1 to 5
Small molecule	1 to 5

Application	Suggested values
Polymers	3 to 50

Peak width

The **Peak width** parameter allows you to specify the number of channels that are used during peak detection. A value of 1 means that all peaks will be labelled, even spikes caused by noise. Therefore, the higher the number of channels you set, the fewer (but significant) number of peaks are labelled. As a guide, a peak width between 5 and 10 channels will result in only significant peaks being labelled.

Although you see the spectrum as an analogue trace, it is actually made up of thousands of points joined together by straight lines. Each point (**blue**) is a value on a channel (**green**). Channels are also known as "bins".



Area

The **Area** parameter defines the peak area used to calculate the position of the peak.



Calculates the peak area down to the baseline.



Calculates the area using the limits of the peak.

You may need to experiment with this parameter setting to find the method which gives the best results for your samples.

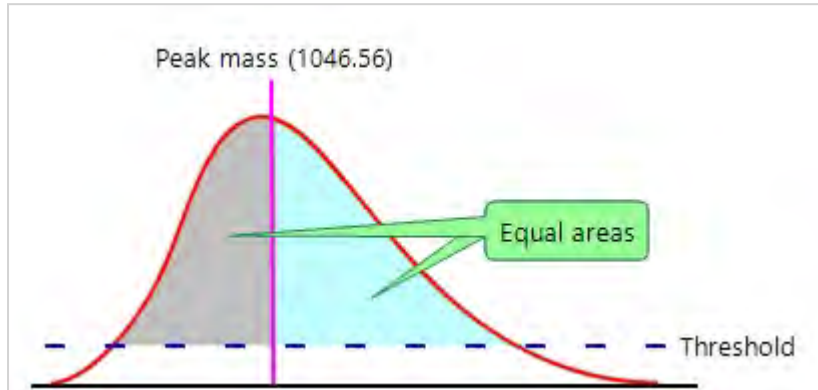
Peak delimiter method

The software supports the following methods for determining the peak mass:

- Threshold Centroid;
- Threshold Centroid 25%;
- Threshold Apex;
- Gradient Centroid;
- Gradient Centroid 25%.

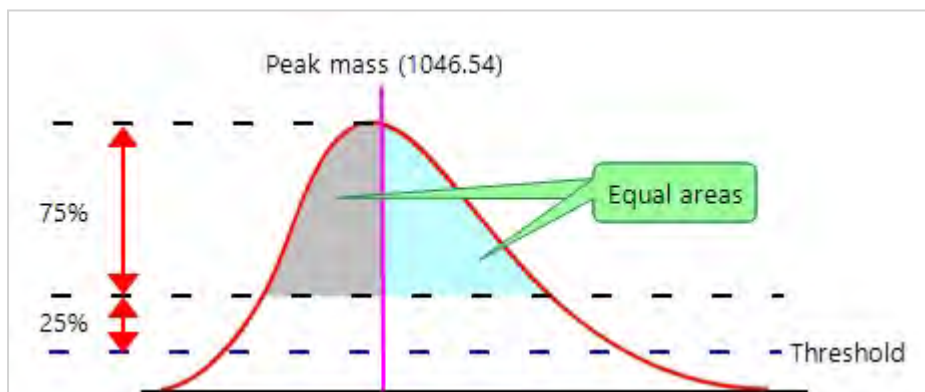
Threshold Centroid

The start and end points of a peak are defined by a constant threshold or an adaptive threshold (see "Threshold type & Threshold offset and response" on page 109). The peak mass is calculated using the area above the threshold. The area is then vertically divided into two equal areas; the peak mass is where the vertical line crosses the curve.



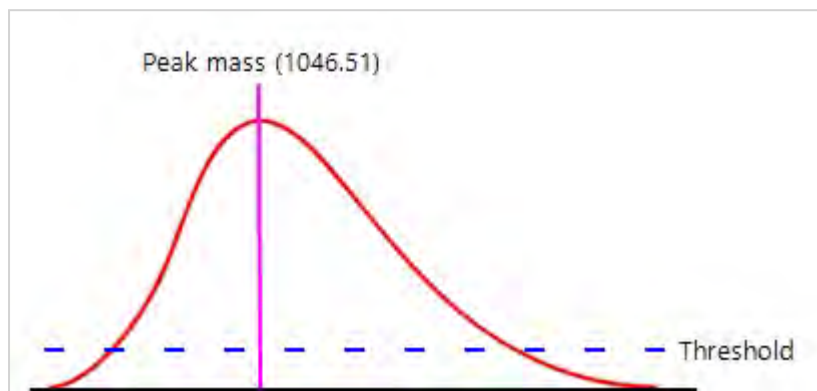
Threshold Centroid 25%

The start and end points of a peak are defined by a constant threshold or an adaptive threshold (see "Threshold type & Threshold offset and response" on page 109). The peak mass is calculated using the area above the threshold; only the top 75% is used. The area is then vertically divided into two equal areas; the peak mass is where the vertical line crosses the curve. This method is recommended for asymmetrical peaks, peptides or low mass samples (<10,000 Da).



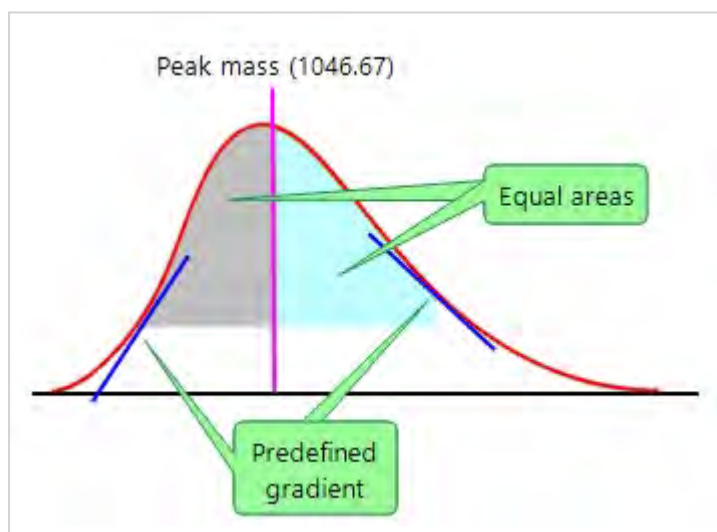
Threshold Apex

The start and end points of a peak are defined by a constant threshold or an adaptive threshold, see "Threshold type & Threshold offset and response" on the facing page. The peak mass is calculated as the highest point above the threshold. This is the recommended method for symmetrical peaks, highly smoothed data or high mass samples (>10,000 Da).



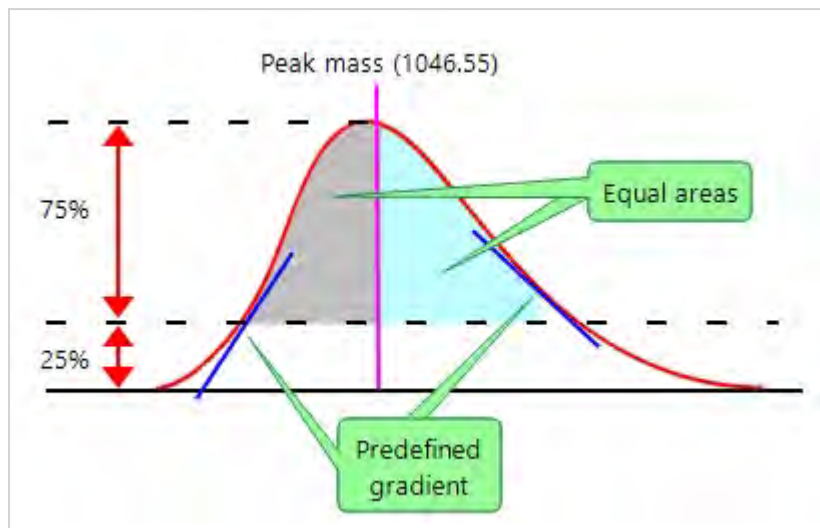
Gradient Centroid

The start and end points of the peak are determined by a predefined gradient. The peak mass is calculated using the area of the curve above the baseline and between the gradient start/end points. The area is then vertically divided into two equal areas; the peak mass is where the vertical line crosses the curve.



Gradient Centroid 25%

The peak start and end points are determined by a predefined gradient. The peak mass is calculated using the area of the curve above the baseline and between the gradient start/end points; only the top 75% is used. The area is then vertically divided into two equal areas; the peak mass is where the vertical line crosses the curve. This method is recommended for asymmetrical peaks, peptides or low mass samples (<10,000 Da).



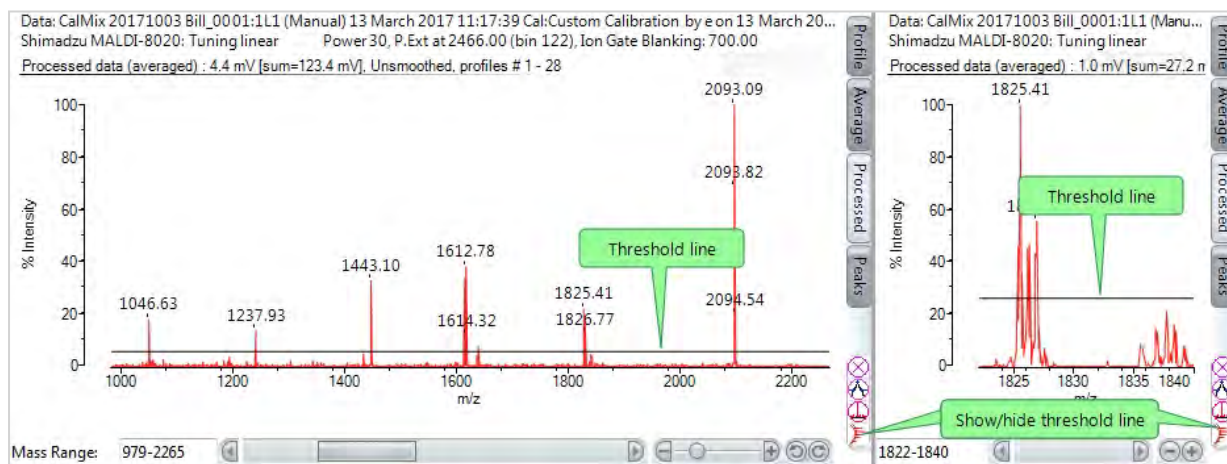
Threshold type & Threshold offset and response

These parameters are only available for the threshold peak delimiter methods.

Constant threshold



This threshold type provides a horizontal line; peak masses above are labelled, peaks below are ignored. If selected, you can use the peak label control to show/hide the threshold line.

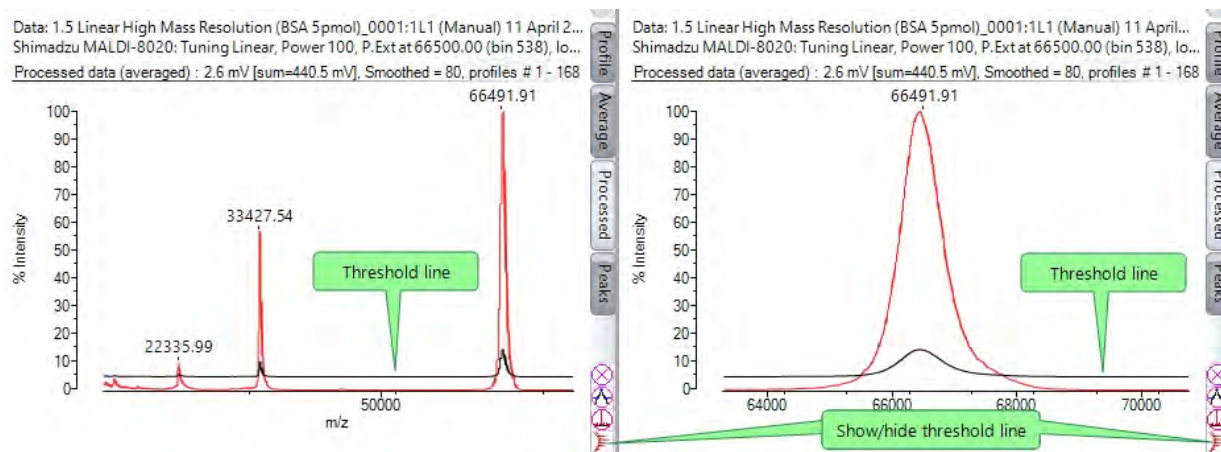


Use the **Threshold offset** field to define the height of the threshold. You need to determine the required offset on a spectrum-to-spectrum basis.

Adaptive threshold



This threshold type follows the signal noise; peak masses above are labelled, peaks below are ignored.



In the example above, the signal noise at the lower masses of the spectrum is very large and tapers off towards the higher masses. The threshold line is displayed in a darker colour.

Use the **Threshold offset** and **Response** fields to define the height and shape of the threshold. You will need to experiment with these values to find those that best fit your experiment.

Threshold offset and response

The **Threshold offset** field defines the height of the threshold above the baseline. Peaks above the threshold are included when the software determines the peaks, and peaks below are ignored. If you set the offset to 0.0mV, the threshold would track the baseline of the spectrum. Set this field so that it is at least above the signal noise.

Note: If you enter a low threshold value, you may cause the PC to operate slowly as the software attempts to label most of the detected peaks, including noise.

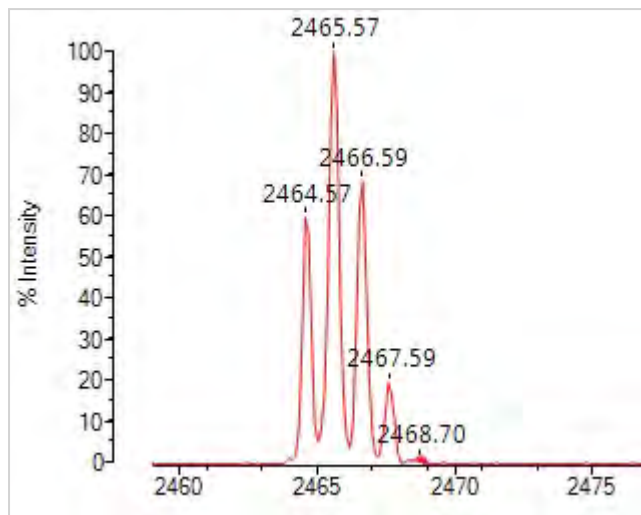
The **Response** field is only available for the Adaptive threshold peak method. This field allows the threshold to track the spectrum. If you set the **Threshold offset** to 0 mV and the **Response** to 1, the threshold tracks the baseline of the spectrum.

Increasing the **Threshold offset** and the **Response** values changes the shape of the adaptive threshold curve. You can set the noise to fall just below the curve, allowing the algorithm to identify peaks in the spectrum.

It is recommended that as a starting point you set the **Threshold offset** to 0.0 mV and the **Response** between 1mV and 2.5 mV.

Monoisotopic peak picking

This parameter is only applicable if isotopes are discernible. Generally, it is recommended that you use this parameter for experiments using peptides. Experiments with proteins does not require it. The monoisotopic mass is the only peak used in this process. It uses the lowest mass containing only the most abundant isotopes, for example, ^{12}C , ^{14}N , ^{16}O .



To access these parameters, select the **Monoisotopic peak picking** check box and then click the down arrow button to display the settings dialog. You can enter values directly into the data fields, or use the spin controls to adjust the currently entered value.

Minimum mass:	<input type="text" value="800"/>
Maximum mass:	<input type="text" value="3500"/>
Minimum isotopes:	<input type="text" value="1"/>
Maximum intensity variation:	<input type="text" value="50"/>
<input checked="" type="checkbox"/> Overlapping distributions	
Minimum peak percent:	<input type="text" value="40"/>

Minimum mass & Maximum mass

Enter the minimum and maximum mass values (in Daltons) to limit the range of masses to be considered when identifying the monoisotopic peak. The default values (800 and 3,500 Da) are usually sufficient for most peptide experiments.

Minimum isotopes

Enter the number of **additional** isotopes (n), that must contribute to a peak before it is to be considered as a candidate for Poisson modelling to determine a monoisotopic peak mass. As this field is "additional" the actual total number of isotopes is $n+1$.

Maximum intensity variation

Enter the tolerance parameter (%) that allows the isotope intensity to differ from its theoretical value by a specified percentage. Candidate isotopes which are outside this parameter are discarded.

Overlapping distributions

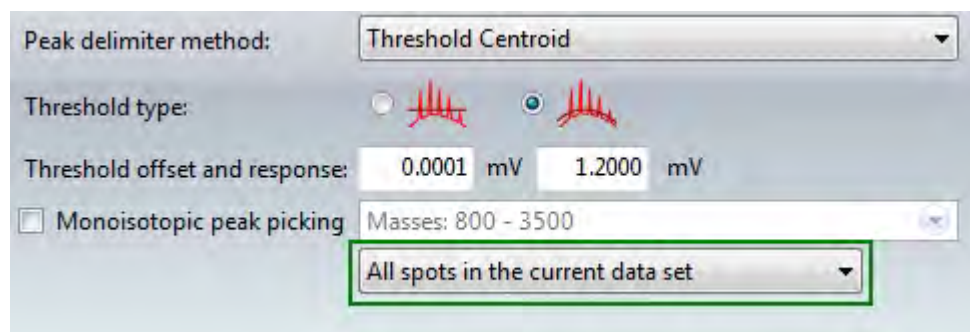
This is a check box, which if selected, permits the Poisson modelling to attempt to distinguish two overlapping isotopic clusters.

Reference: *Breen E J, et al. Automatic Poisson peak harvesting for high throughput protein identification. Electrophoresis 2000 June 21 (11):2243–2251.*

Minimum peak percent

Applies only if **Overlapping distributions** is selected. The algorithm identifies the isotopic masses associated with the dominant monoisotopic mass and subtracts this from the overlapping distribution. If the remaining masses do not constitute at least the specified percentage of the dominant contribution, they are discarded.

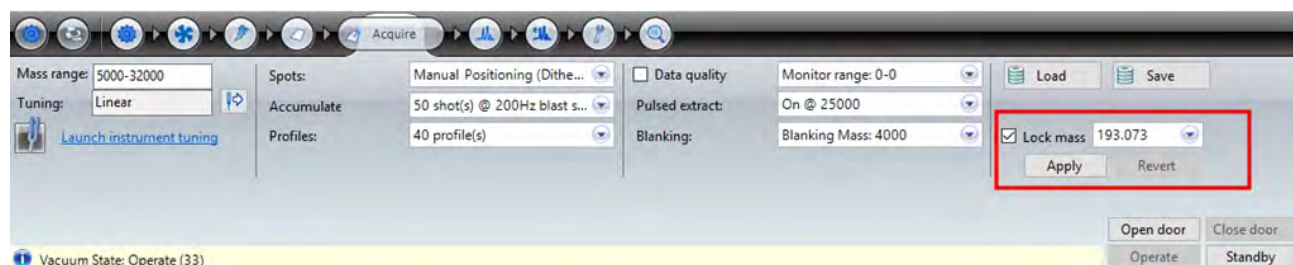
All spots in data sets



From the drop-down menu, select **All spots in the current data set** to apply the processing parameters you have set to the current acquisition, or **All spots in all loaded data sets** to apply the parameters to all loaded acquisitions.

Using Lockmass

The lock mass function is accessed from the Acquire tab and allows the user to specify a mass to correct mass drifts. This allows the user to compensate for systemic mass drifts between calibrations and allow the spectra to obtain higher mass accuracy. The specified lock mass value, accepted tolerance level and lock mass method are entered in the lock mass parameters. Lock mass is applied to the spectrum either automatically at the end of the acquisition or it can be applied manually to an already acquired spectrum.



Lock Mass Parameters

To set the lock mass parameters, perform the following:

1. Select the **Lock Mass** option.
2. Click the lock mass drop-down menu.
3. In the Lock Mass field, enter the mass value.
4. Enter the tolerance level value and select one of the following corresponding units from the drop-down menu:
 - Da - To apply the tolerance levels using Daltons.
 - mDa - To apply the tolerance levels using Millidaltons.
 - ppm - To apply the tolerance levels using part per millions.

NOTE: By default, if the tolerance level is left blank, a 1 Da value is applied.

5. Lock mass is a method of moving a peak of a known mass to its theoretical mass in the spectrum. Both of the provided lock mass methods do this, but they differ in how they scale the rest of the spectrum. From the Method drop-down menu, select one of the following correction methods:
 - MassCorrection - The mass correction method uses a proportional shift in the masses after the calibration is applied. That is, peaks that are further away from the lock mass peak will be shifted more and vice versa.
 - TimeCorrection - The time correction method uses a linear shift in time, before the calibration is applied. This correction will only provide good performance in the region close to lock mass peak (roughly within 10% of the mass), as the mass actually has a quadratic dependence on time, not linear.
6. To use the selected lock mass parameters, click **Apply**. The Lock Mass parameters are applied to all the displayed spectra in the Acquire tab when a peak is found within the tolerance range at the specified mass.
7. Click **Revert** to roll back the Lock Mass parameters and return the spectra to the previous calibration.

NOTE: When Lock Mass is applied post acquisition, the dataset must be (re)saved in order to retain the lock mass correction.

Improving Data quality

Data quality is used in conjunction with rastering (see [Setting acquisition parameters](#)). It allows you to define the criteria to filter which profiles are kept and which are discarded. The use of the data quality feature can often improve the quality of the final acquired spectra. However, throughput is typically reduced, as acquired profiles not meeting the required criteria are discarded.

NOTE: Data quality improvement are not available when using dithering.

To use this feature, you need to have prior knowledge of your data, usually from previous experiments where this feature was not used. You need to be able to anticipate what peaks you can expect and what criteria needs to be set to achieve your goals.

The screenshot shows the 'Common Settings' dialog box. The 'Filtering Criteria' section is checked and expanded, showing a table of criteria and their values. Below the table are three input fields for 'Minimum S/N %', 'Maximum consecutive failed profiles', and 'Minimum accepted profiles per point (%)'. A 'Settings...' button is also visible.

Criterion	Value
<input checked="" type="checkbox"/> Maximum Signal	Signal: 1
<input type="checkbox"/> Minimum Peaks in Segment	SegmentCount: 1; PeakCount: 3
<input type="checkbox"/> Minimum Resolution	Resolution: 5000
<input checked="" type="checkbox"/> Minimum Signal	Signal: 100
<input type="checkbox"/> Minimum Signal/Noise	SignalNoise: 1.5
<input type="checkbox"/> Min S/N Peaks in Segment	SignalNoise: 1.5; SegmentCount: 1

Minimum S/N %: 0
Maximum consecutive failed profiles: 2
Minimum accepted profiles per point (%): 30

To access these settings, select the **Data quality** check box on the **Acquire** tab.

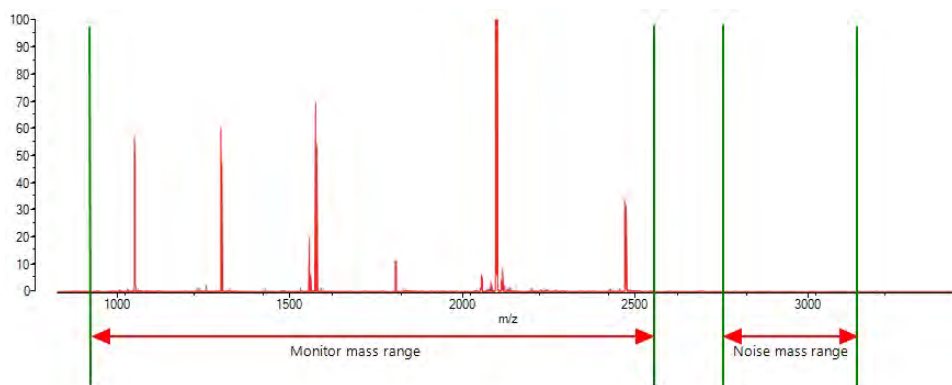
Common settings

Monitor mass range

Enter an upper and lower mass value in Daltons - only peaks detected in this range will be compared with the filtering criteria. You should ensure that this range covers all the peaks of interest.

Noise mass range

Enter an upper and lower value for the noise mass range. This defines an area outside of the monitor mass range which includes only "chemical" noise.

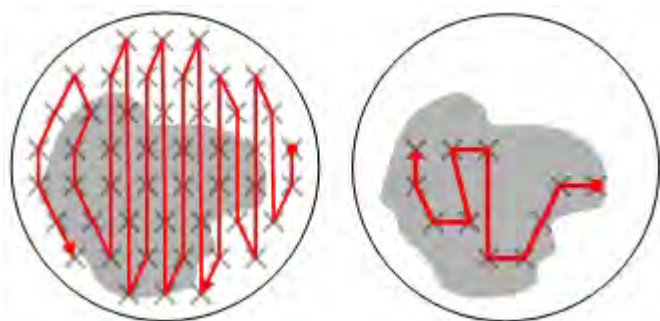


Acquisition strategy

From the drop-down list, select one of the two options below.

- **Find sweet spots** - typically used to increase acquisition speed or to ensure even coverage of the sample spot;
- **Exhaust raster spots** - typically used to improve data quality where the sample contains matrix or salt crystals on the surface.

Find sweet spots



1. The specified number of profiles (see [Setting acquisition parameters](#)) are distributed between the available raster points. The application attempts to acquire the required number of profiles (pass 1).
2. Each profile is assessed against the **Filtering criteria**. Only those which meet the criteria are accepted.
3. Raster positions which failed to produce acceptable profiles are discarded; the remaining/outstanding number of profiles required to complete the acquisition are distributed amongst the remaining raster positions (that gave acceptable spectra in the first pass) and a second pass of the sample is performed, with the laser only firing on the remaining raster positions (pass 2).
4. The profiles from the second pass are assessed against the filtering criteria. Raster positions are further filtered depending on whether they produced acceptable profiles.

This process continues until:

- there are no raster points remaining;
- the application has acquired the required number of profiles;
- the application has reached the maximum number of consecutive failures;
- the value in the field **Minimum accepted profiles per point (%)** is reached, see "Minimum accepted profiles per point (%)" on page 120.

Exhaust raster spots

NOTE: Using this method, throughput may be reduced.

1. The laser fires on the raster point.
2. Each profile is analysed against the **Filtering criteria**. However, if the sample contains matrix or salt crystals on the surface, the profiles will fail the criteria and will be discarded, and the acquisition moves to the next available spot.
3. As the surface of the spot is ionised, profiles of the sample are obtained.

The process continues until:

- The application has acquired the required number of profiles;
- The application has reached the maximum number of consecutive failures and all available raster points have been exhausted.

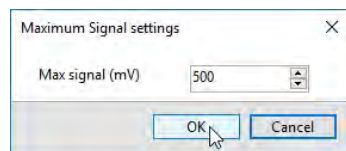
Filtering criteria

To use this feature, select the adjacent check box. You may need to experiment to determine the optimal value(s).

For each criterion:

1. Click to highlight the criterion and click **Settings**; a dialog box is displayed.
2. Enter the required value(s).
3. Click **OK**; the dialog box closes.
4. Select the required check box to enable/disable the criterion.

Maximum signal

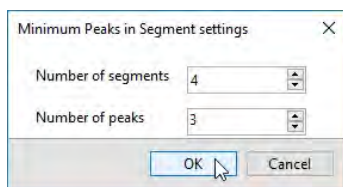


The signal of a displayed peak is shown in the spectrum header:

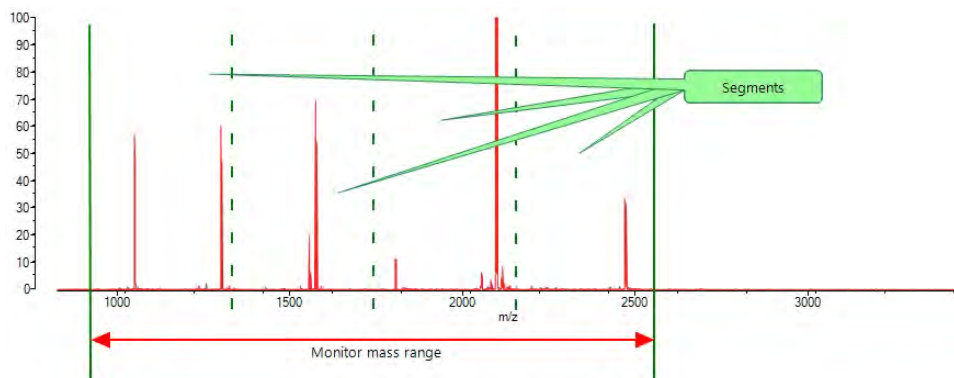
```
Data: 1.2 Linear Ultimate Peptide Resolution (Cal mix)_0002:1L1 (Manual) 11 April  
Shimadzu MALDI-8020: Tuning Linear, Power 35, P.Ext at 2466.00 (bin 103), Ion Ga  
Processed data (averaged) : 8.8 mV [sum=473.4 mV], Unsmoothed, profiles # 1 - 54
```

If any part of the processed signal (within the **Monitor mass range** field) exceeds the setting, the profile is rejected.

Minimum peaks in segment

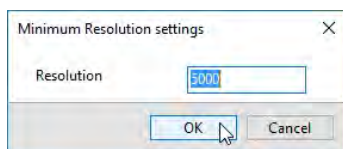


The application divides the mass range (within the **Monitor mass range** field) into the specified number of segments.

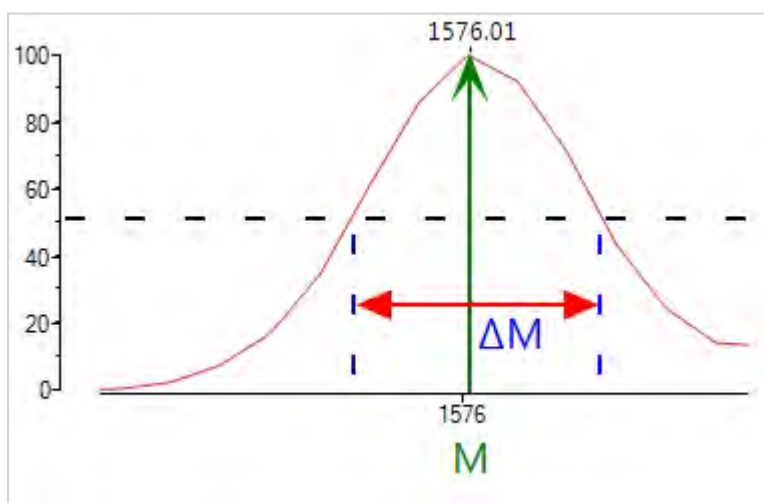


If the minimum number of labelled peaks within each segment is not achieved, the profile is rejected.

Minimum resolution



The resolution is a mathematical description of the shape of the peak:



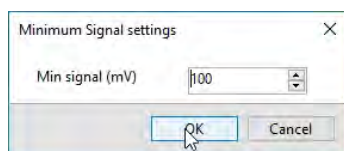
The formula is:

$$\text{Resolution} = M/\Delta M$$

where M = mass and ΔM (dMass) = mass range at 50% intensity of the peak.

If a peak (within the **Monitor mass range** field) does not exceed the setting, the profile is rejected.

Minimum signal



The signal of a displayed peak is shown in the spectrum:

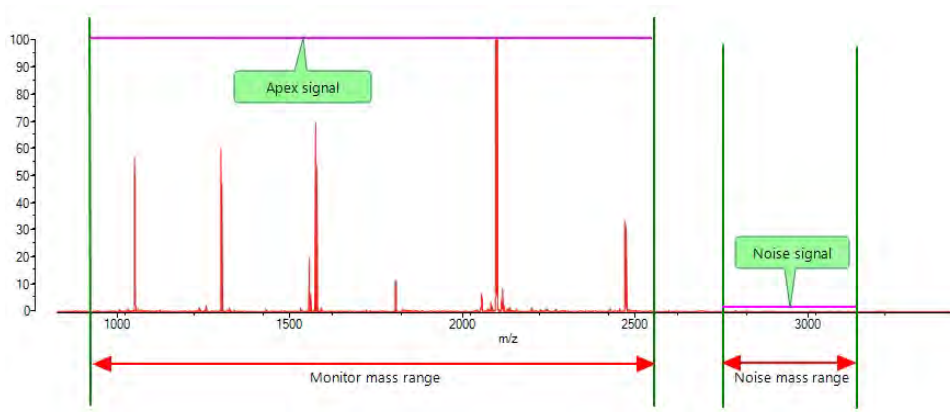
Data: 1.2 Linear Ultimate Peptide Resolution (Cal mix)_0002:1L1 (Manual) 11 April
 Shimadzu MALDI-8020; Tuning Linear, Power 35, P.Ext at 2466.00 (bin 103), Ion Ga
 Processed data (averaged) : 8.8 mV [sum=473.4 mV], Unsmoothed, profiles # 1 - 54

If any part of the processed signal (within the **Monitor mass range** field) does not exceed the setting, the profile is rejected.

Minimum signal/noise

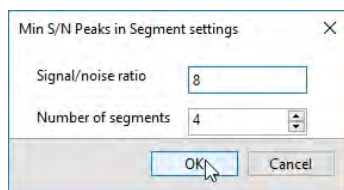
The signal-to-noise (S/N) is a ratio =

$$\frac{\text{Apex signal (within the **Monitor mass range** field)}}{\text{Noise signal (within the **Noise mass range** field)}}$$

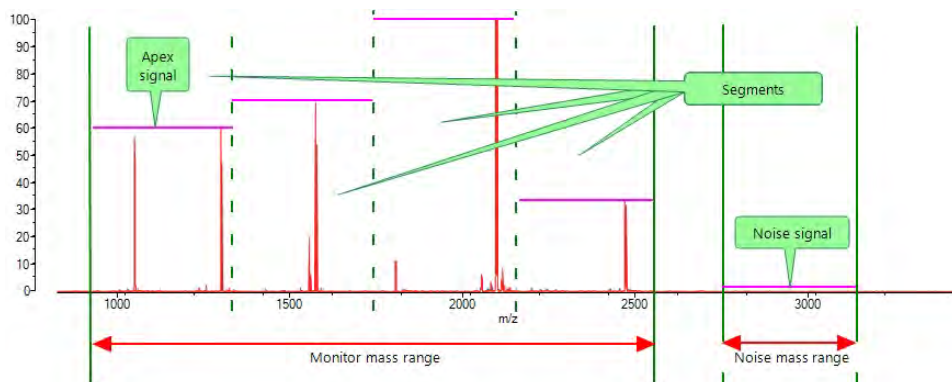


If the minimum ratio is not achieved, the profile is rejected.

Min S/N peaks in segment



The application divides the mass range (within the **Monitor mass range** field) into the specified number of segments.



Within each segment, the highest intensity peak (Apex signal) are identified. The profile is accepted if the highest intensity peak in each segment has a signal/noise (S/N) ratio equal to, or greater than, the specified value.

The signal-to-noise (S/N) is a ratio=

$$\frac{\text{Apex signal (within the segment)}}{\text{Noise signal (within the **Noise mass range** field)}}$$

Minimum S/N %

The signal-to-noise (S/N) is a ratio=

$$\frac{\text{Apex signal (within the **Monitor mass range** field)}}{\text{Noise signal (within the **Noise mass range** field)}}$$

The application takes the first profile and calculates the spectrum quality, where:

$$\text{spectrum quality} = \frac{\text{S/N} \times \text{user setting}}{100}$$

The S/N values of subsequent spectra are compared to the spectrum quality value.

Example:

For a user setting of 20% and the first profile having a S/N value of 10, spectrum quality = $(10 \times 20)/100$
= 2

- Subsequent spectra that have S/N values of 2 or <2 are rejected;
- Subsequent spectra that have S/N values >2 are accepted.

The initial **Minimum S/N %** value is updated only after consecutive failures.

Maximum consecutive failed profiles

The value specified here determines how many profile failures are tolerated by the application before it moves on to a new position. This behaviour varies depending on the specified acquisition strategy as follows.

- If **Exhaust raster spots** is selected, the application will abandon the current raster point and move to the next raster point. If there are no more raster points defined, the application moves to the next sample spot;
- If **Find sweet spots** is selected, the application will abandon the current sample spot, and move to the next sample spot.

Minimum accepted profiles per point (%)

Only applicable if the **Acquisition strategy** is set to **Find sweet spots**. The application rejects raster points which have failed to provide a specified percentage of good profiles.

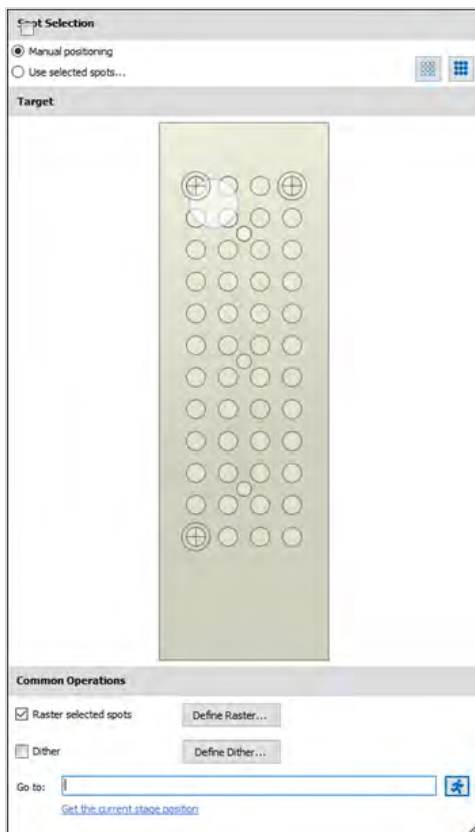
Intensity Mapping

Intensity mapping is a means to visualise the intensity distribution of an ion or range of mass across an area. This is a useful tool if you are looking for an answer to simple spatial distribution questions (e.g. sample homogeneity).

NOTE: When viewing an intensity map spectrum, the mass range must be less than 1000Da and all stored profiles have been selected during the acquisition.

1. Click the **Spots** drop-down arrow.

The Spot Selection window displays



2. Select the Raster selected spots option and click **Define Raster**.

The Define Raster window displays.

3. From the Raster type drop-down menu options, select **Regular Square**.
4. Select **Serpentine Raster** from the options in the Raster style drop-down menu.
5. Enter the raster width in the Width(μm) field.
6. To amend the height of the raster, enter the value in the Height(μm) field.

NOTE: When the raster height value is greater than 1000 (μm), the Number of raster points: field becomes active.

7. In the Number of raster points: field, specify how many raster points are required per sample.

NOTE: The value entered in this field will automatically update if the value in the Spacing between points (μm) field is amended.

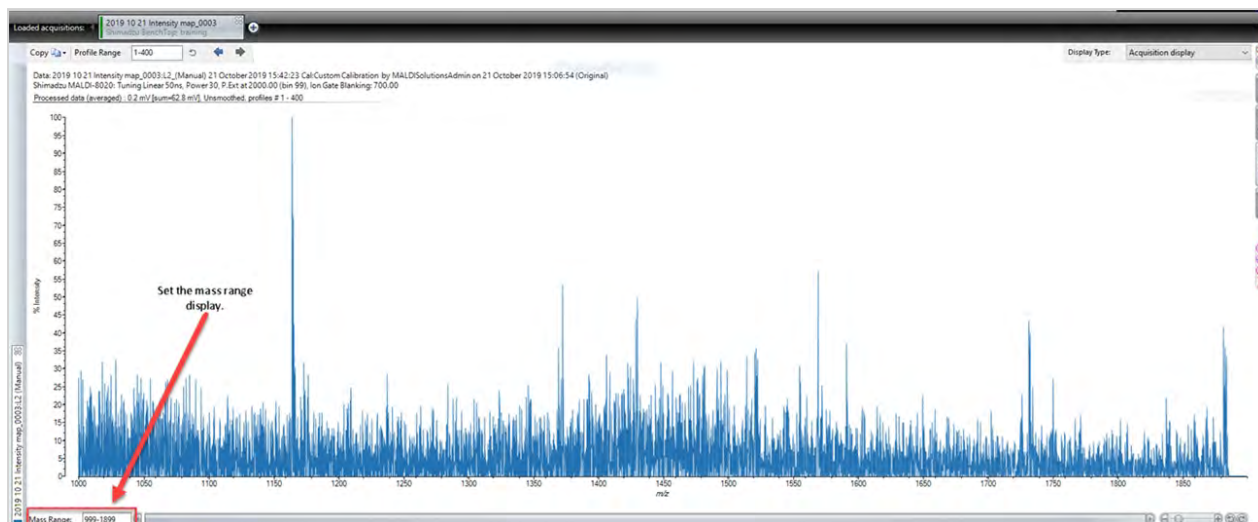
8. If required, enter an X-axis offset value in the Offset X(μm) field.
9. If required, enter an Y-axis offset value in the Offset Y(μm) field.
10. To amend the spacing between raster point, enter the value in the Spacing between points (μm) field.

NOTE: The value in the Spacing between points field is automatically calculated based on the values entered in the Width(μm) and Height(μm) fields. If the value is amended the number of raster points is automatically updated.

11. Click **OK**. and close the Spot Selection window.
12. Click the Profiles drop-down arrow.
13. Enter the number of profiles to be used in the Set number of profiles field.
14. Select **Store Individual Profiles**.
15. Do one of the following:
 - Click plus icon adjacent to the Loaded acquisition: label and load a previous acquisition and go to Step 16.
 - Click **Fire** to run the acquisition and go to Step 17.

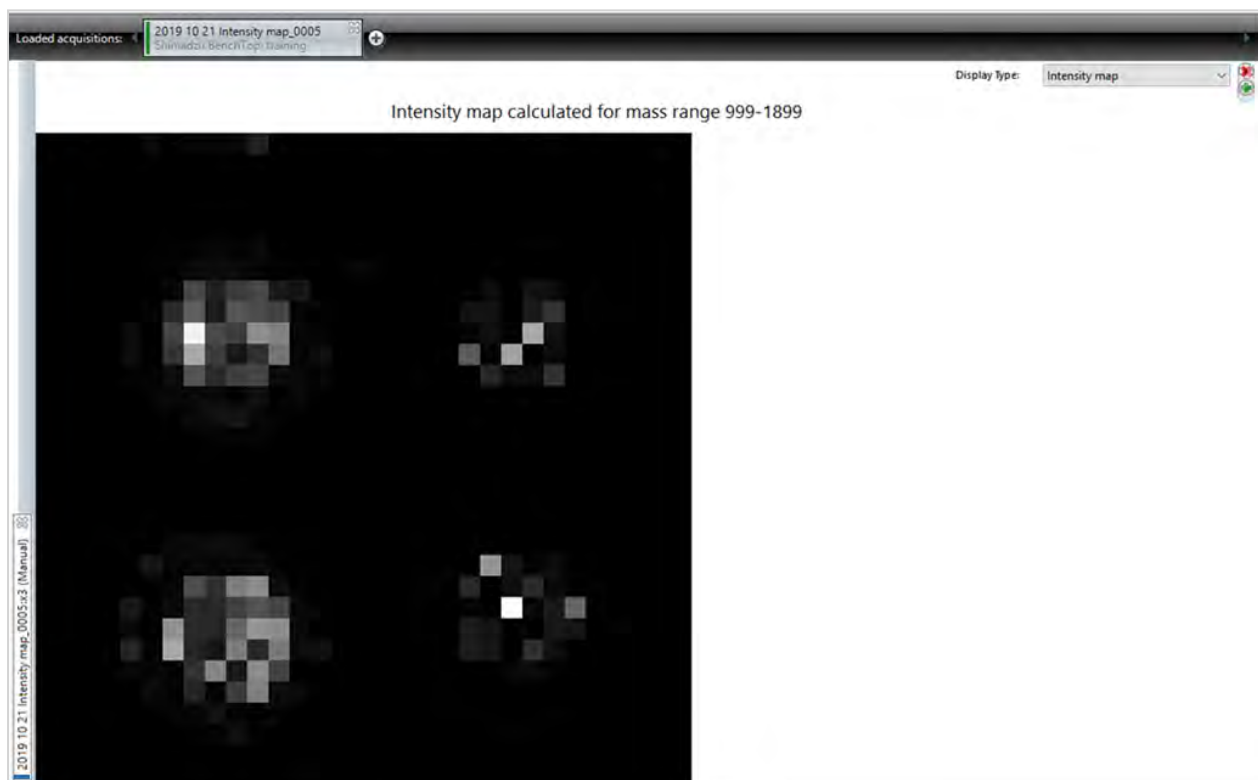
NOTE: For details on running acquiring data, see "Acquiring a spectrum" on page 66

16. Set the mass range value using the Mass Range field.



17. Select **Intensity map** from the Display Type drop-down menu.

The Intensity map displays.



NOTE: The intensity map above shows the intensity variation across samples. The two spots on the left are fresh and the two on the right have been previously analysed.

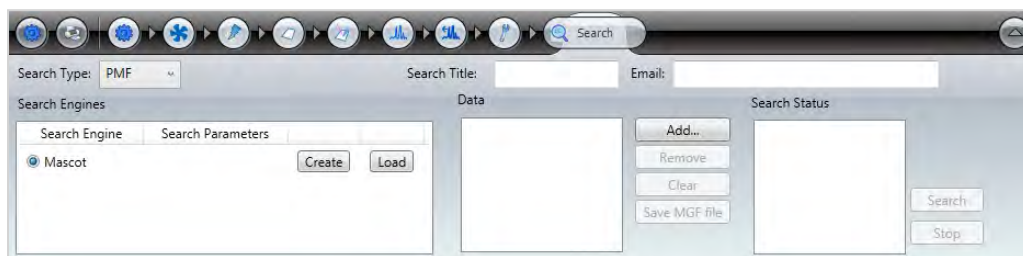
18. Click a spot on the Intensity map to display the profile associated with the selected pixel.

Searching with Mascot

You can search a database of proteins using the Mascot search engine to aid your analysis of PMF (Peptide Mass Fingerprint) or ISD (Ion Source Decay) spectrum. However, the results for PMF are for crude confirmation of a protein identification and are not suitable for publication.

To use this feature, you must have access to a Mascot server, or internet access to the Mascot web site.

Click the **Search** tab to display the search parameters:



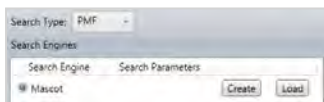
The Search feature requires you to either create a new Mascot search parameter set, or load an existing set before performing a search.

Loading a Mascot parameter set

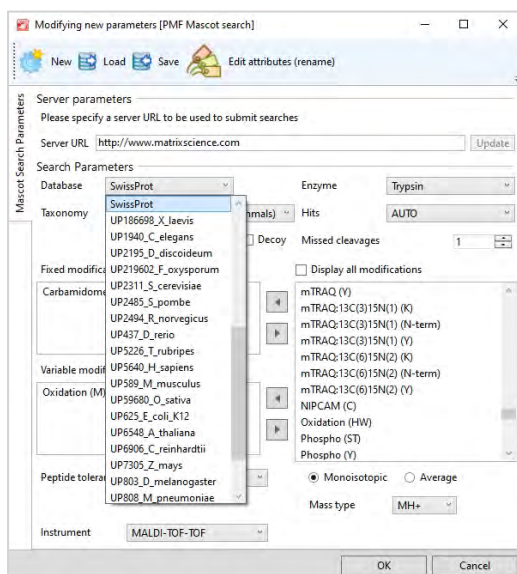
See [Loading & saving data](#).

Creating a Mascot parameter set

1. At the **Search Type** drop-down list, select either ISD or PMF.



2. Click the **Create** button.



3. Fill in the required **Mascot Search Parameters** fields and click **OK** (refer to the table below for an explanation of the available settings).

Field	Description
Server URL	The Matrix Science database URL is: http://www.matrixscience.com If you are using a local Mascot server, the URL is typically: http://<mascot server name>/mascot
Database	Select the required database from the drop-down list (usually SwissProt).
Enzyme	For ISD, select None. For PMF, select the required enzyme from the drop-down list.
Taxonomy	Select the required source of the protein.
Hits	Maximum number of hits displayed in a search results report.
Quantitation	Quantitation analysis only.
Decoy	Select to search the decoy database.
Missed cleavages	Generally, select 1.
Display all modifications	Expands the following modification fields to show all available modifications.
Fixed modifications and Variable modifications	The fields allow you to specify any modifications applied to the sample during preparation. Select the appropriate modification(s).
Peptide tolerance	Set the required tolerance and units.
Monoisotopic and Average	Select whether the spectral data is of monoisotopic (PMF) or average masses (ISD).
MSMS tolerance	Set the required tolerance and units. (ISD only)
Peptide charge	Generally, select +1 for positive ion mode analysis and -1 for negative ion mode analysis (negative ion mode available on MALDI-8030 only)
Instrument	Select MALDI-TOF-TOF
Mass type	For a PMF search in positive ion mode, select MH+. In negative ion mode using MALDI-8030 only, select M-H.
Error tolerant	Not normally selected.

4. For ISD, fill in the required **Additional Search Parameters** fields and click **OK**:

Modifying new parameters [ISD Mascot search]

New Load Save Edit attributes (rename)

Mascot Search Parameters

Server parameters
Please specify a server URL to be used to submit searches

Server URL

Search Parameters

Database Enzyme

Taxonomy Hits

Quantitation Decoy Missed cleavages

Additional Search Parameters

Fixed modifications

Variable modifications:

Peptide tolerance

MS/MS tolerance

Display all modifications

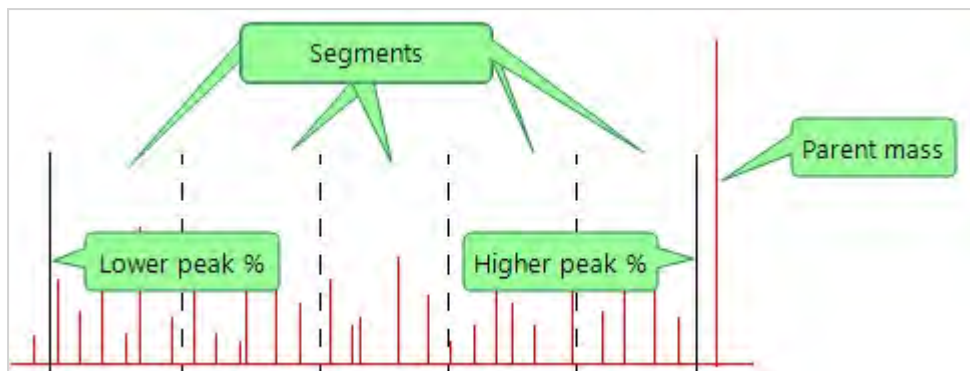
- Acetyl (K)
- Acetyl (N-term)
- Acetyl (Protein N-term)
- Amidated (C-term)
- Amidated (Protein C-term)
- Ammonia-loss (N-term C)
- Carbamidomethyl (N-term)
- Carbamyl (K)
- Carbamyl (N-term)
- Carboxymethyl (C)
- Cation:Na (C-term)

Monoisotopic Average

Peptide charge

Error tolerant

The **Additional peak parameters** define which fragment peaks are selected.



The **Lower peak %** and **Higher peak %** fields set the boundaries for selecting peaks for the fragment mass list. The limits are set as a percentage of the parent mass. Peaks outside the boundaries are ignored.

The **Max peaks** and **Segments** fields define the maximum number of peaks and their distribution. For example, if you set the maximum number of peaks to 40 and the number of segments to 5, the application will:

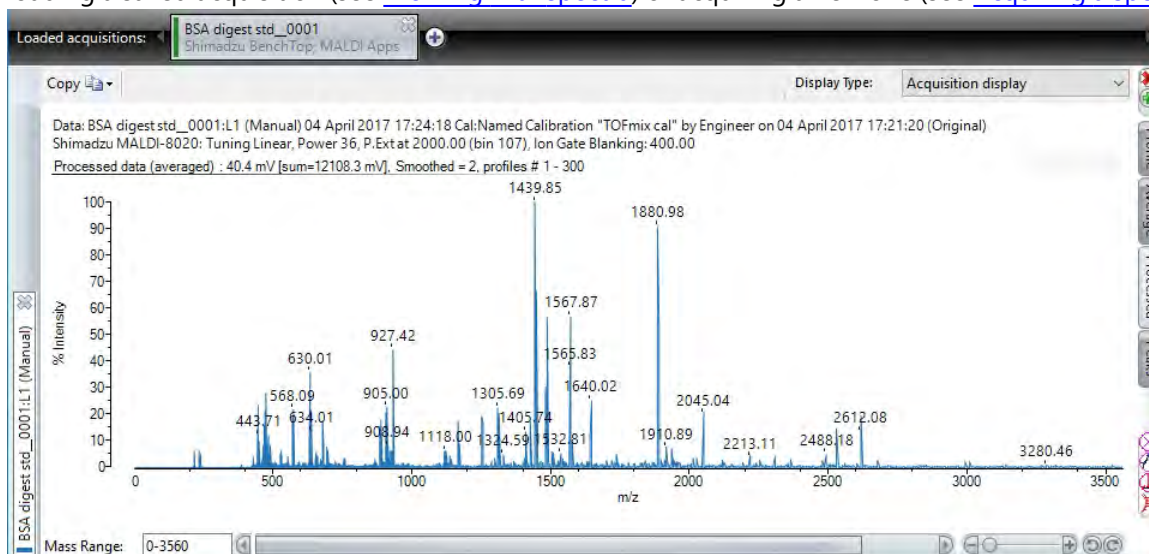
- Divide the spectrum into 5 equal segments;
- From each segment, pick the 8 (40 divided by 5) most intense peaks;
- Use the 40 peaks for the fragment mass list.

5. When the **Save data as** dialog is displayed, save the parameter set (see [Loading & saving data](#)).

Performing a Mascot search

Searching with PMF

1. Ensure that the spectrum you wish to use for the search is displayed on the **Loaded acquisitions** bar, either by loading a saved acquisition (see [Working with spectra](#)) or acquiring a new one (see [Acquiring a spectrum](#)).



2. On the **Search** tab, select **PMF** from the **Search Type** drop-down list.

3. In the **Search Title** and **Email** fields, enter the information you wish to appear at the top of the **Mascot Search Results** page.

4. Load a previously created Mascot parameter set (see [Loading & saving data](#)).
5. If required, make changes to the parameter set as described in [Creating a Mascot parameter search](#) and then click **Save**, or click **OK** if the current settings are appropriate.

In both cases, the name of the parameter set is displayed in the **Search parameters** column.

6. Enter the required **Minimum mass**, **Maximum mass** and **Maximum peaks** values, and then click **Fetch peaks** to display the available peak masses in the **Masses (Da)** list.

Note: You can select an entry in this list and click **Remove selected** to remove it, or click **Remove all** to clear the list. In the latter case, you must then click **Fetch peaks** again to repopulate the list.

7. Click **Search**.

The progress of the search operation is displayed in the **Search Status** box.

When Mascot has finished processing the data, you will see a browser window appear which contains the search results.

Mascot Search Results

User :
 Email : bill.stevens@stokar.co.uk
 Search title : BSA digest check
 MS data file : Automatically uploaded data
 Database : SwissProt 2017_06 (554860 sequences; 198649153 residues)
 Taxonomy : Mammalia (mammals) (66684 sequences)
 Timestamp : 4 Jul 2017 at 13:31:51 GMT
 Top Score : 75 for [ALBU_BOVIN](#), Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4

SwissProt Decoy	
Protein hits above identity threshold	1 0
Highest scoring protein hit	75 48

Mascot Score Histogram

Protein score is $-10 \cdot \log(P)$, where P is the probability that the observed match is a random event. Protein scores greater than 61 are significant ($p < 0.05$).

Concise Protein Summary Report

Format As: Concise Protein Summary [Help](#)
 Significance threshold $p <$ 0.05 Max. number of hits 5
 Preferred taxonomy All entries

[Re-Search All](#) [Search Unmatched](#)

1. [ALBU_BOVIN](#) Mass: 71244 Score: 75 Expect: 0.0021 Matches: 14
 Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4

Searching with ISD

- Follow steps 1 through 5 of the [Searching with PMF](#) procedure above, selecting **ISD** as the search type in step 2.
- Enter the required **Precursor mass** value, and then click **Fetch peaks** to display the available peak masses in the **Masses (Da)** list.

Note: You can select an entry in this list and click **Remove selected** to remove it, or click **Remove all** to clear the list. In the latter case, you must then click **Fetch peaks** again to repopulate the list.

- Optionally click **Save MGF file**, and then use the dialog to save the file to a suitable location on the computer.

Note: The MGF file (Mascot Generic Format) is a plain text (ASCII) file containing the displayed peak mass information.

4. Click **Search**.

The progress of the search operation is displayed in the **Search Status** box.

When Mascot has finished processing the data, you will see a browser window appear which contains the search results.

Mascot Search Results

User :
 Email : bill.stevens@stokor.co.uk
 Search title : BSA ISD check
 MS data file : Automatically uploaded data
 Database : SwissProt 2017_07 (255100 sequences: 198754136 residues)
 Taxonomy : Mammalia (mammals) (66712 sequences)
 Timestamp : 4 Jul 2017 at 09:30:48 GMT
 Protein hits :
 A1B2I BOVIN Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4
 A1B2I BAPHA Atypical chemokine receptor 1 OS=Pagio hamadryas GN=A1B2I PE=3 SV=1
 C0Q4 HUMAN Ubiquitin biosynthetic protein C0Q4 homolog, mitochondrial OS=Homo sapiens GN=C0Q4
 P0KX1 BOVIN Regulator of nucleic factor 3 alpha OS=Bos taurus GN=P0KX1 PE=1 SV=2
 C1PAC7 HUMAN Cilia- and flagella-associated protein 47 OS=Homo sapiens GN=C1PAC7 PE=2 SV=4

Mascot Score Histogram

Ions score is $-10 \cdot \log(P)$, where P is the probability that the observed match is a random event.
 Individual ions scores > 41 indicate peptides with significant homology.
 Individual ions scores > 60 indicate identity or extensive homology ($p < 0.05$).
 Protein scores are derived from ions scores as a non-probabilistic basis for ranking protein hits.

Peptide Summary Report

Format As: Peptide Summary Help
 Significance threshold $p \leq 0.05$ Max number of hits 5
 Standard scoring ModPT scoring Ions score or expect cut-off 0 Show sub-sets 0
 Show pop-ups Suppress pop-ups Sort unassigned Decreasing Score Require bold red
 Preferred taxonomy All entries

Chapter 5 - Using SampleStation

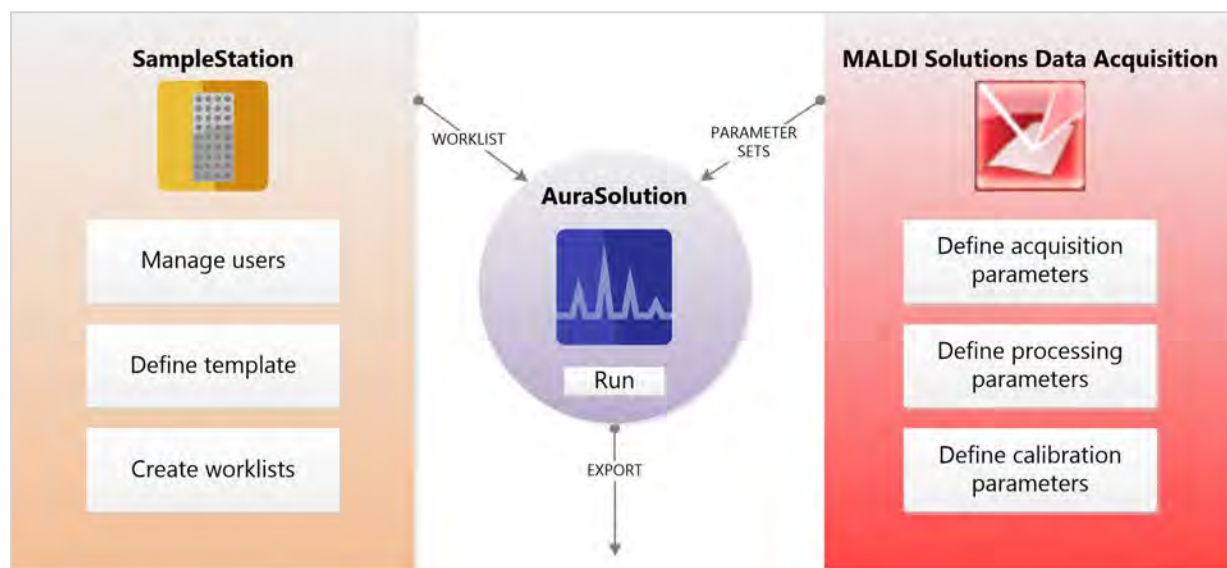
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Managing users	135
Managing roles & permissions	138
Defining the worklist template	142
Managing worklists	147

Introduction

SampleStation is an optional software module used in conjunction with *MALDI Solutions Data Acquisition* and *AuraSolution* to provide a workflow automation solution for your instrument. *SampleStation* is primarily used to define worklists which contain information on the target plates and samples to use when running automated acquisitions.

Note: *SampleStation* uses a different database for user accounts, which you need to set up separately before using the application.

The following diagram shows the relationship between the different software applications used in automation.



Note: *SampleStation* and *AuraSolution* are optional licensed software applications. These features may not be available on your instrument unless the correct software licence has been purchased. Please contact your local Shimadzu representative for details.

Starting the application

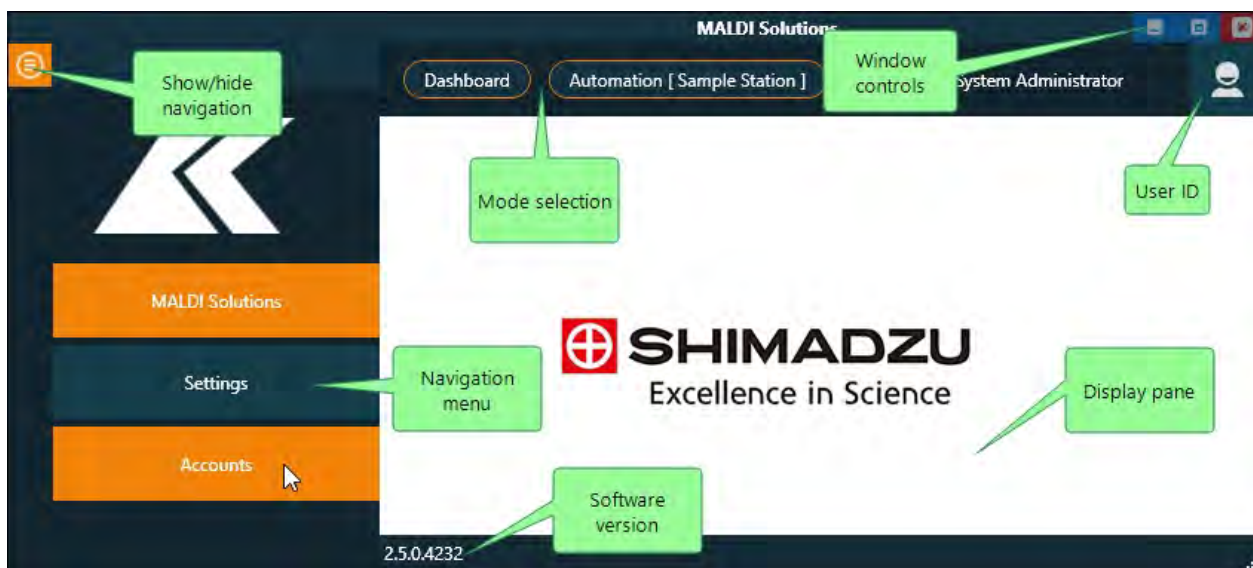
Logging in

1. On the **Start** menu, locate and select **Shimadzu Corporation > SampleStation**.



2. Enter the **Username** and **Password** supplied by your system administrator, and then click **Login**.

You will see the initial interface as shown below.

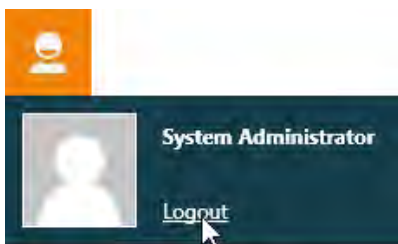


Notes:

- If you do not see the navigation menu after logging in, click the **Show/hide** button to display the menu.
- Use the window controls to maximise, minimise or close the application window.
- Once you have selected an option in the navigation menu, you can click the **Show/hide** button again to remove the menu, maximising the available space within the application window for the display pane.

Logging out

1. In the application window, click the **User ID** icon and then click **Logout**.



You will be returned to the **Login** dialog, where you can either log in as a different user, or use the window controls to close the application window.

Note: Closing the application window without first logging out will automatically log out the current user and then terminate the application.

Managing users

SampleStation uses a separate database to manage user access (i.e. it does not use the *MALDI Solutions Administration Console*), thus you must create separate user accounts within the application.

Adding a user

1. In *SampleStation*, click the **Show/hide** button to display the navigation menu, and then click **Accounts**.



Note: Once you have selected an option in the navigation menu, you can click the **Show/hide** button again to remove the menu.

2. In the mode selection bar, click **Manage Users**.



3. In the **Add User** dialog, enter the details for the new user account.

A screenshot of the 'Add User' dialog form. The form is organized into several sections: 'User Details' with fields for Username (darmstro), Email (dug.armstrong@kratos.co.uk), and Confirm Email (dug.armstrong@kratos.co.uk); 'Password security' with fields for Password and Confirm Password (both masked with dots); 'Personal Information' with fields for First Name (Dug), Last Name (Armstrong), Headline (Tech Writer), and Description; 'User system role' with a dropdown menu set to 'Administrator'; and 'Account Status' with an 'Account Active Status' checkbox. At the bottom, there are three buttons: 'New User', 'Save User' (highlighted with a mouse cursor), and 'Save user'.

Notes:

- In order to assign a role to a user, you must first create a role (see [Managing roles & permissions](#)) or use one of the built-in roles. Generally speaking, it is good practice only to assign one user the built-in **Administrator** role;
 - You must select the **Account Active Status** property in order to make the user account functional.
4. In the **Password security** section of the dialog, enter the same password in the **Password** and **Confirm Password** boxes.

Note: It is good practice to use alphanumeric passwords with at least one non-alphabetic (symbol) character and a minimum length of eight characters.

5. Click **Save User** to create the user account, or **New User** to clear the entered data.

The user account you have entered will appear in the **Users** list.

Username	First Name	Last Name	Email	Account Active	Role
admin	System	Administrator		<input checked="" type="checkbox"/>	Administrat
peterwhite	Peter	White	peterwhite@gmail.com	<input type="checkbox"/>	advanced u
johnny ions	Johnny	ions	jions@gmail.com	<input type="checkbox"/>	advanced u
johnwhite	John	White	johnwhite@gmail.com	<input type="checkbox"/>	advanced u
huang	CC	Huang	fxhcc@shimadzu.com.cn	<input type="checkbox"/>	Administrat
dugarmstrong	Dug	Armstrong	dug.armstrong@kratos.co.uk	<input checked="" type="checkbox"/>	Administrat

Amending a user

1. In the **Manage Users** dialog, select a user account from the **Users** list.
2. Make the required changes in the **Edit User** sections of the dialog (see [Adding a user](#)).
3. In the **Update Password** section of the dialog, enter the same password in the **Password** and **Confirm Password** boxes.

4. **Note:** It is good practice to use alphanumeric passwords with at least one non-alphabetic (symbol) character and a minimum length of eight characters.
5. Click **Save User** to update the user account.

Deactivating a user

Note: Once created, user accounts cannot be deleted. You can however deactivate a user account as follows:

1. In the **Manage Users** dialog, select a user account from the **Users** list.
2. In the **Edit User** section, deselect the **Account Active Status** property.
3. Click **Save User** to update the user account.

Deactivated user accounts may be reactivated at a later date by amending the account details and reselecting the **Account Active Status** property.

Managing roles & permissions

SampleStation uses a separate database to manage user access (i.e. it does not use the *MALDI Solutions Administration Console*), thus you must create separate roles within the application which you can then assign to individual user accounts (see [Managing users](#)).

Note that only users who have been assigned the **Edit Roles** permission may add or amend roles.

Adding a role

1. In *SampleStation*, click the **Show/hide** button to display the navigation menu, and then click **Accounts**.

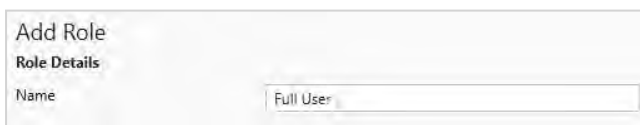


Note: Once you have selected an option in the navigation menu, you can click the **Show/hide** button again to remove the menu.

2. In the mode selection bar, click **Manage Roles & Permissions**.



3. In the **Manage Roles & permissions** dialog, enter the name of the new role in the **Add Role** section.

A screenshot of the "Add Role" dialog box. The dialog has a title bar that says "Add Role" and a subtitle "Role Details". Below the subtitle, there is a label "Name" followed by a text input field. The text "Full User" is entered into the input field.

4. Select the required permissions for the new role in the **Permissions** section.

Permissions

Application

View Solutions View Dashboard

Account

View Accounts Edit Users Edit Roles

Setting

View Settings Edit Automation Settings

Automation

View Automation View Sample Station Read Sample Worklist Edit Sample Worklist Edit Sample Worklist Export

[New Role](#) [Save Role](#)

NOTES:

- The **View Solutions** permission allows users to access MALDI Solutions functions (not currently used);
- The **View Dashboard** and **View Sample Station** permissions are not currently used.
- The **View Accounts** permission allows users to access the **Manage Users** and **Manage Roles & Permissions** functions.
- The **Edit Users** and **Edit Roles** permissions allow users to make changes to user accounts and roles respectively. This option is usually reserved for administrators.
- The **View Settings** permission allow users to access the **Settings** functions.
- The **Edit Automation Settings** permission allows users to makes changes to the defined target and custom fields data used in worklists.
- The **View Automation** permission allows users to access the **Automation** functions.
- The **Read Sample Worklist** permission allows users to load and view sample worklists.
- The **Edit Sample Worklist** permission allows users to create, edit or save sample worklists.
- The **Edit Sample Worklist Export** permission allows users to export a sample worklist as a CSV format file.

5. Click **Save Role** to create the role, or **New Role** to clear all selected permissions.

Amending a role

1. In *SampleStation*, click the **Show/hide** button to display the navigation menu, and then click **Accounts**.



Note: Once you have selected an option in the navigation menu, you can click the **Show/hide** button again to remove the menu.

2. In the mode selection bar, click **Manage Roles & Permissions**.



3. In the **Roles** list, click on the entry for the role you wish to amend.



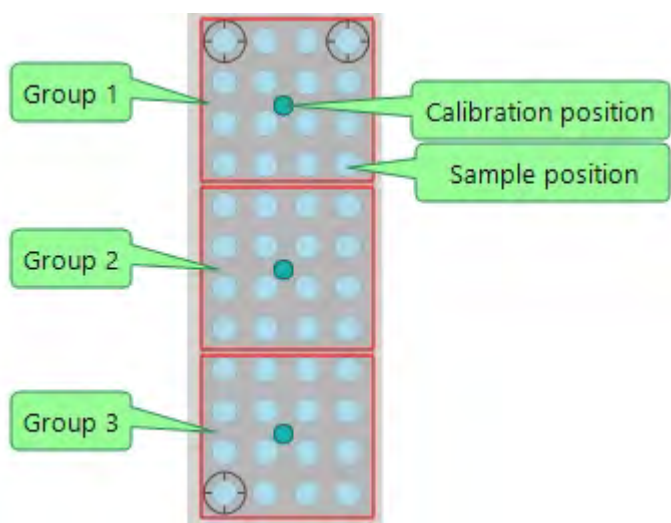
4. Make the changes you require to the role permissions (see [Adding a role](#)) and then click **Save Role**.

Defining the worklist template

The first time you use *SampleStation* with an instrument, you must define the template to be used when creating new worklists (see [Managing worklists](#)). The template defines the 'rules' which govern how users select samples when creating a worklist. This includes choosing the target plate layout, associating sample positions with specific calibration positions and defining custom data fields.

Notes:

- For the MALDI-8020 and MALDI-8030 instruments, only one target plate layout is supported - this comprises 48 defined sample positions plus three calibration positions. The following example illustrates the most common arrangement of groups - it is recommended that administrators use this layout except under special circumstances.



- A template group is different from a worklist group (see [Managing worklists](#)). A template group is used to associate a set of sample positions with a specific calibration position. A worklist group is a set of sample spots which share the same calibration position. When you select a sample spot for a worklist, the calibration position defined in the template group is used to add the correct calibration spot to the worklist group.
- Under normal circumstances you should only define three template groups, with one calibration position in each group. This will allow users to identify the correct calibration spot automatically when creating worklists.
- After you create the template, it will be used for all worklists on the instrument until you amend the template settings.
- Only users with the appropriate permissions can view, create or amend the template (see [Managing roles & permissions](#)). Normally, only the system administrator is expected to perform these functions.

Viewing the worklist template

1. In *SampleStation*, click the **Show/hide** button to display the navigation menu, and then click **Settings**.



Note: Once you have selected an option in the navigation menu, you can click the **Show/hide** button again to remove the menu.








The **Automation Settings** dialog will be displayed.

Required	Display Name	Default	Obsolete
<input checked="" type="checkbox"/>	Patient		<input checked="" type="checkbox"/>
<input checked="" type="checkbox"/>	Ward		<input checked="" type="checkbox"/>

Notes:

- If you have not previously defined a template, a banner will appear on this dialog displaying **No Sample Template**. In this case, you must create and save the template before you can create any worklists;
- On the MALDI-8020 and MALDI-8030 instruments, you cannot select a target type other than **1 x 48 2.8mm wells**;
- Hovering over a sample position in the slide graphic will display a pop-up containing the position ID number (e.g. **A4**);
- Click on the plus (+) control in the slide header to display a list of all defined groups;

- Click on the plus (+) control in the group header to display a list of all spots currently defined as group members;
- The slide graphic in the **Instrument Target** section uses the following symbols:

	Selected spot (regular)
	Unselected spot (regular)
	Group member (regular)
	Unselected spot (calibration)
	Selected spot (calibration)
	Group member (calibration)
	Spot already acquired

Creating the worklist template

1. Follow the procedure described in [Viewing the worklist template](#) above.
2. Click on the slide header and then click **Add Group**.
3. Click on the **Group 1** header and then click the plus (+) control to expand the member list.
4. Click on each spot in the slide graphic you wish to add to the group.

You will see the selected spot appear in the member list for the group. You may also click on the spot again to remove it from the member list if required.

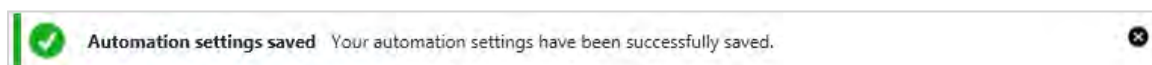
Note:

- Leaving unselected spots in a group will prevent users from accessing that spot in their worklist and is not recommended.
- You should include one calibration spot in each group.

Name	
Slide 1 - Worklist Template	
Name	
Group 1	
Spot Id	Spot Type
x1	Calibration
A1	Regular
A2	Regular
> A4	Regular
B2	Regular
B3	Regular
C3	Regular

- Continue to add spots as described above until the member list for the group is complete.
- Repeat steps 2 through 5, creating a further 2 groups.
- Add any custom fields you require (see [Adding custom fields](#)).
- Click **Save**.

You will see a message confirming the save operation.



Any new worklists created in the application will now use these template settings.

Amending the worklist template

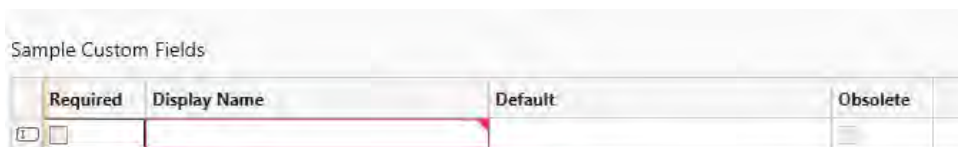
- Follow the procedure described in [Viewing the worklist template](#) above.
- Click on the plus (+) control in the slide header to display the available groups.
- Click on the plus (+) control in a group header to display the member list assigned to that group.
- Follow the procedure described in [Creating the worklist template](#) above to make any changes you require to the spot selection for the group.
- Repeat steps 3 and 4 if you need to amend any further groups.
- Follow the procedure described in [Adding custom fields](#) if you wish to make changes to the custom fields associated with the template.
- Click **Save**.

You will see a message confirming the save operation. Any new worklists created in the application will now use these amended template settings.

Adding custom fields

Notes:

- You may optionally add one or more custom fields to the template either during creation or when subsequently amending the template.
 - Once added, a custom field may not be deleted. However, you may mark a custom field as obsolete by selecting the **Obsolete** check box for the field. Obsolete custom fields will be ignored by the application when creating worklists.
1. Follow the procedure described in [Viewing the worklist template](#) above.
 2. In the **Sample Custom Fields** area of the **Automation Settings** dialog, click the first empty row in the custom fields list.



3. Click in the **Display Name** column, and then enter the title of the data field.
4. If you wish to make the field mandatory for data entry purposes, select the **Required** check box.
5. If you wish the field to be pre-filled with any default data, click in the **Default** column and then enter the required data.
6. Click **Save**.

You will see a message confirming the save operation. Any worklists created in the application will now use these custom fields to allow entry of sample description data.

Managing worklists

A worklist contains details of which samples are available for automation on a particular target plate. Worklists are used by the *AuraSolution* application to control the selection of samples during the automated acquisition process (see [Using AuraSolution](#)).

Notes:

- Worklists are associated with individual target plates using either a barcode number (for disposable plates) or a user-specified alphanumeric identifier (for reusable plates).
- You do not need to load a target plate into the instrument before creating a worklist for the plate.
- If you create a worklist for a reusable plate and then subsequently clean the plate, you must create a new worklist with a different identifier before you can reuse the plate for automation.
- Worklist groups are different from template groups (see [Defining the worklist template](#)). A worklist group is a set of sample spots which share the same calibration spot. When you select a sample spot for your worklist, the calibration position defined in the template group is used to automatically add the correct calibration spot to the worklist group.

Creating a worklist

1. In *SampleStation*, click the **Show/hide** button to display the navigation menu, and then click **MALDI Solutions**.



Note: Once you have selected an option in the navigation menu, you can click the **Show/hide** button again to remove the menu.

2. In the mode selection bar, click **Automation**.
The **Sample Worklist** dialog will be displayed.
3. Click **New**.
4. In the **New Sample Worklist** dialog, select the **Slide Type** from the drop-down list.
5. If you selected the **Disposable** slide type, select the **Worklist barcode** box and then either scan the slide barcode, to automatically populate the associated information, or enter the slide number (without hyphens).

New Sample Worklist

Slide Type: Disposable

Worklist barcode: DS162001095

Create Cancel

6. If you selected the **Reusable** slide type, enter a unique identifier in the **Worklist identifier** box.

New Sample Worklist

Slide Type: Reusable

Worklist identifier: SLIDEXY999

Create Cancel

Note: You should record the **Worklist identifier** separately for future reference.

7. Click **Create**.
8. If you selected the **Reusable** slide type, click **Yes** in the **Reusable Slide** dialog to confirm that the slide has been cleaned, or **No** to abort the procedure.

Reusable Slide

Has this reusable slide been cleaned and are all sample positions available?

Yes No

9. In the **Sample Worklist** dialog, enter a name for the slide in the **Slide Name** box.

Sample Worklist

New Load Clear Sample Group Save Export

	Slide Name	Barcode
	TestDA	DS162001095

10. Click the plus (+) control next to the slide name to display the group list for the new slide, which will initially be empty as shown below.

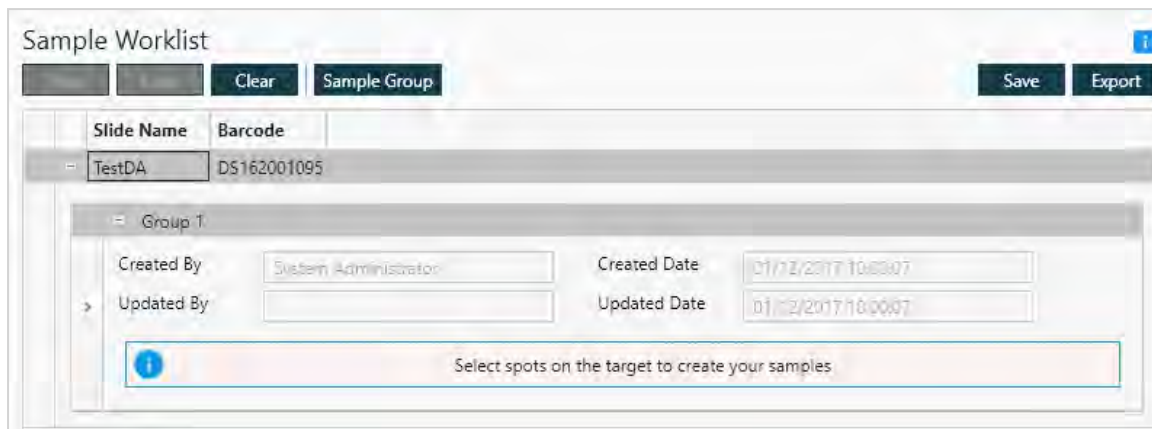
Sample Worklist

New Load Clear Sample Group Save Export

	Slide Name	Barcode
	TestDA	DS162001095






Add Sample Group

11. Click **Sample Group** to create an empty group as shown below.



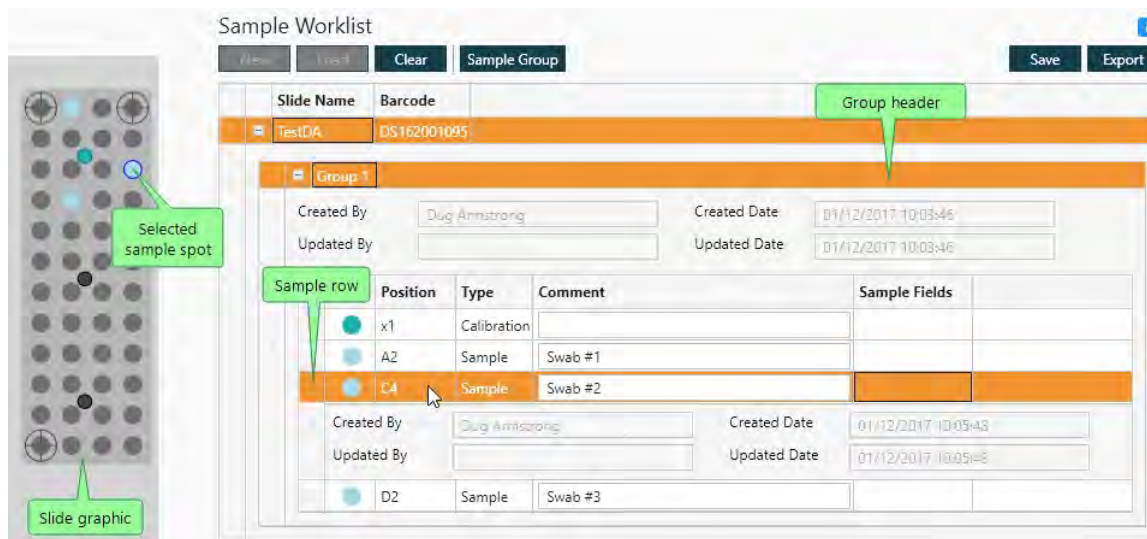
- On the slide graphic, click on a sample spot to add it to the currently selected group (clicking on it a second time will remove it).

Note: The slide graphic uses the following symbols.

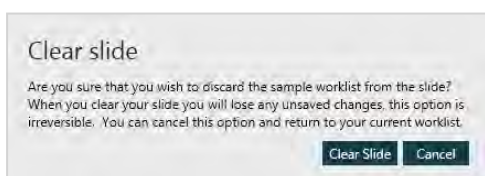
	Sample spot(member of current group)
	Sample spot (not included in current group)
	Sample spot (currently selected in group list)
	Calibration spot (not assigned to current group)
	Calibration spot (assigned to current group)

When you select the first sample spot for a group, the calibration spot associated with it (as defined in the template group - see [Defining the worklist template](#)) is automatically added to the group. This will then become the calibration spot for the whole group, regardless of which other sample spots you subsequently add to the group.

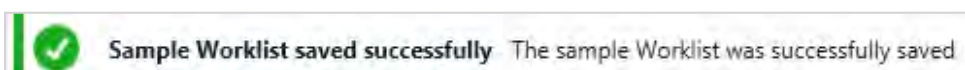
- Continue to add sample spots as described in step 12 until you have added all the spots you require. The group list will show a list of entries for all the spots you have selected.

**Notes:**

- Click on sample row in the group list to highlight the position on the slide graphic.
- Click on the group header to display all current group members.
- You may optionally enter descriptive text in the **Comment** column for each sample row in the list. Enter the text manually or, if contained within a barcode, by clicking on the field and using a barcode scanner.
- Any custom data entry fields that have been manually specified in the worklist template will appear in the **Sample Fields** column. Enter the information in the custom fields manually or populate the fields using a barcode scanner.
- If you wish to remove a group from the list, select the group header and then press the **Delete** key.
- If you wish to abandon the worklist entirely, click the **Clear** button . If you do so, the following dialog will be displayed:



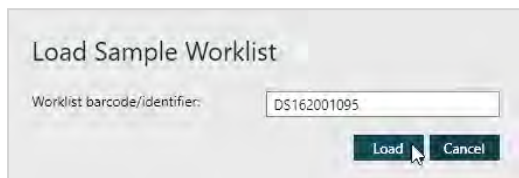
14. Click **Clear Slide** to abort the creation procedure, or **Cancel** to preserve the entered data. Optionally, repeat steps 11 through 13 if you want to add additional groups to the worklist.
15. Click **Save** to save the worklist to the database.
16. Confirmation of the save operation will be displayed in a banner at the top of the dialog.



Note: Once you have saved the worklist, it becomes uniquely associated with the target plate identifier you specified at the beginning of the creation procedure. The application will not allow you to create any further worklists using the same identifier, although the worklist can be amended at a later time.

Amending a worklist

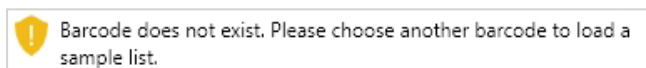
1. Follow steps 1 and 2 of the creation procedure above (see [Creating a worklist](#)).
2. In the **Sample Worklist** dialog, click **Load**.



3. In the **Load Sample Worklist** dialog, scan the target plate barcode or enter the identifier for the plate associated with the required worklist, and then click **Load**.

The **Sample Worklist** dialog will be populated with the requested worklist data.

Note: If you enter the identifier incorrectly, or no worklist exists with the specified identifier, the following error will be displayed.



In this case, either enter a valid identifier or click **Cancel** to abort.

4. Follow the procedure described in [Creating a worklist](#) above to make the required changes and save the amended worklist.

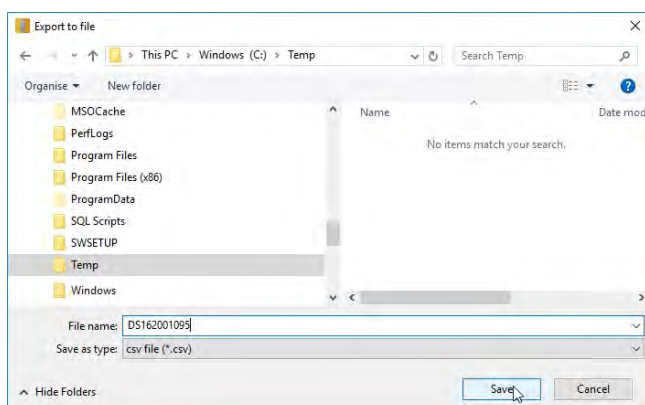
Exporting a worklist

You may export a worklist to a CSV format file for use in external programs (e.g. spreadsheet applications) as follows:

1. Load the required worklist by following steps 1 through 3 of the amend procedure above (see [Amending a worklist](#)).

Note: You may also export directly after creating a new worklist (see [Creating a worklist](#)).

2. On the **Sample Worklist** dialog, click **Export**.



3. Use the **Export to file** dialog to specify an output folder and file name, and then click **Save**.

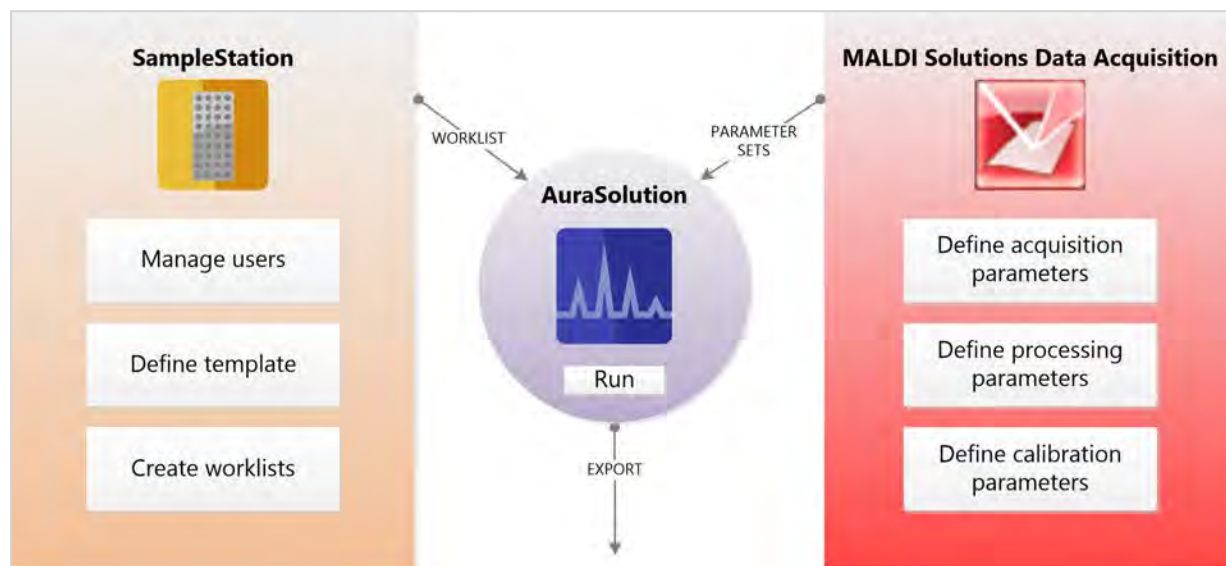
Chapter 6 - Using AuraSolution

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Introduction

AuraSolution is an optional software module used in conjunction with *MALDI Solutions Data Acquisition* and *SampleStation* to provide a workflow automation solution for your instrument. *AuraSolution* is used to run 'experiments', which comprise a series of acquisitions across multiple preselected samples with a minimum of intervention.

The following diagram shows the relationship between the different software applications used in automation.



Note: *SampleStation* and *AuraSolution* are optional licensed software applications. These features may not be available on your instrument unless the correct software licence has been purchased. Please contact your local Shimadzu representative for details.

Workflow overview

AuraSolution is the 'control' application used for automation, and as such is the final step in the automation workflow. Before running an experiment with *AuraSolution*, you need to complete the following tasks.

- Prepare a target slide with the appropriate samples (see [Working with target plates](#));
- Use the *SampleStation* application to create a worklist for the target plate (see [Using SampleStation](#));
- Use *MALDI Solutions Data Acquisition* to create the necessary parameter sets (see [Acquiring data](#)).

Once these tasks are complete, you can start *AuraSolution* and then run your experiment.

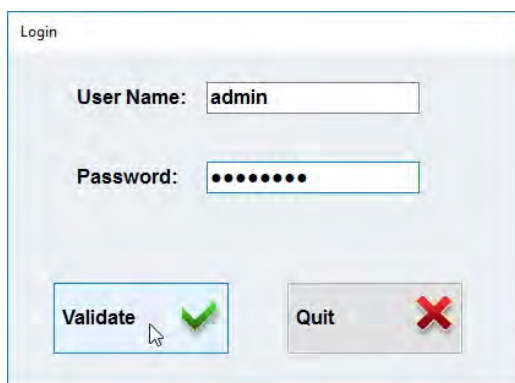
Starting the application

Note: The *AuraSolution* application uses the instrument database to control user access, therefore user accounts are controlled and administered with the Administration Console (see [Administration](#)). As a minimum, users who wish to use the application should have the **Basic**, **Instrument Control** and **Submit Experiments** permissions assigned to their accounts.

Logging into the application

Note: Before starting the application, you should close down *MALDI Solutions Data Acquisition* if it is currently running on the PC workstation.

1. On the **Start** menu, locate and select **Shimadzu Corporation > AuraSolution**.
2. Enter the **Username** and **Password** supplied by your system administrator, and then click **Validate**, or **Quit** to abandon the login procedure.



You will see a progress bar displayed while the application connects to the instrument, after which the main window opens.

Logging out

To close the application, use the window control (**X**) to close the application window and terminate the application. Alternatively, you can click the **Lock** button to perform the following actions.



- Click **Logout** to log out the current user;
- Click **Lock** to disable the user interface.

In the former case, you must log in again to continue working, while in the latter case you must re-enter your user name and password to continue.

Note: In order to conform to regulatory requirements, the application will automatically log out the current user after 5 minutes of inactivity.

Setting parameters

Before you are able to run experiments with *AuraSolution*, you must create and save parameter settings for acquiring, calibration and processing the data to the instrument database. The procedures for doing this can be found in the following topics.

- Acquisition parameters - see [Setting acquisition parameters](#);
- Calibration parameters -see [Calibration](#);
- Processing parameters - see [Processing data](#).

Loading parameters

Once you have created the parameter sets described above, you can load them into the application as follows.

1. On the *AuraSolution* toolbar, click **Parameters**.
2. In the parameters dialog, click **Load Parameters** and then select the desired acquisition parameters (see [Loading & saving data](#)).
3. Click **Load Calibration**, and then select the desired calibration parameters.
4. Click **Load Processing**, and then select the desired processing parameters.

The dialog will display the names of the parameter sets you specified for the Acquisition, Calibration and Processing parameters.



Note: These parameter sets will be used for all experiments you run with the application until you either select a different set or close the application.

Setting export options

You can set the behaviour of the export function as follows.

1. On the *AuraSolution* toolbar, click **Parameters**.
2. In the parameters dialog, select **Enable Export**.

3. Select the **ASCII** and/or **mzML** options in the **Format** column, and then click **Browse** to set a folder location for the exported file(s).

4. Select the type data to be exported using the following:

Note: A combination of the following options can be exported at any one time.

5. Select one of the following options under the **Select Export Peak Type** section of the dialog:

- **Continuous Processed Spectrum** - exports all data points processed using the specified processing parameters.
- **Continuous Averaged Spectrum** - exports all data points as unprocessed raw data.
- **Centroid Peaks** - exports peak mass data only.

Data collected during acquisitions will now be automatically exported to the folder location and file type(s) you specified above.

6. Select one of the following options under the **Select Data Intensity Type** section of the dialog:

- **Millivolts** - exports intensity in millivolt (mV) units which are the measured signals on the detector.
- **Counts** - exports intensity as counts which are an arbitrary, but higher value equivalent of mV.

7. Select one of the following options under the **Select Peak Intensity Type** section of the dialog:

- **Apex** - exports values from the apex of each peak which is the highest data point within the peak limit.
- **Area** - exports values as the area of each peak which is the sum of the intensities of all data points within the peak limit.

Note: The Area intensity is only active when Peak data (centroid) is selected.

The required data is exported to the specified location.

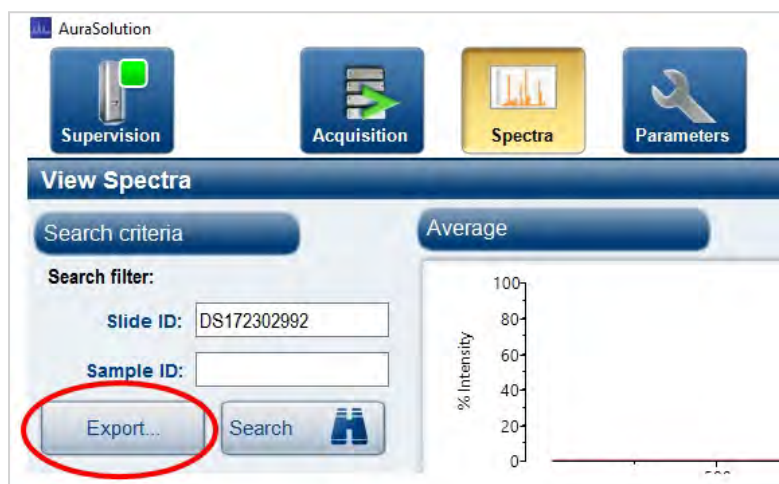
Export Button

Once the Export behaviour has been set, the **Export** button allows for data collected during acquisitions to be exported at any time - even after experiments have been completed.

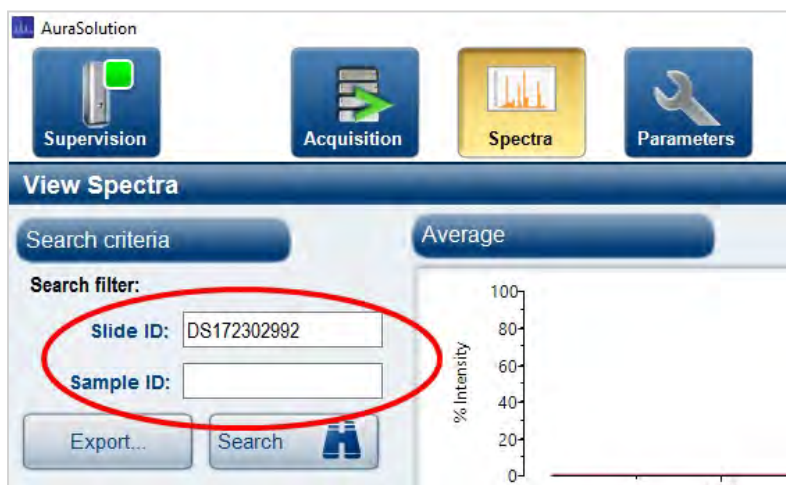
For example: You are able to re-export data or save data to export at a later time.

On the AuraSolution toolbar, click Spectra.

The **Export** button is shown circled below:




To Search for previous results to Export use either the **Slide ID**, or the **Sample ID**:



Exported results are available in your selected location.

Loading a target plate

Warnings & cautions

CAUTION	Damage to the instrument
	<p>Do not put wet samples or any other element such as fibre into the instrument, as moisture and particles within the vacuum system may cause damage to the internal components.</p>

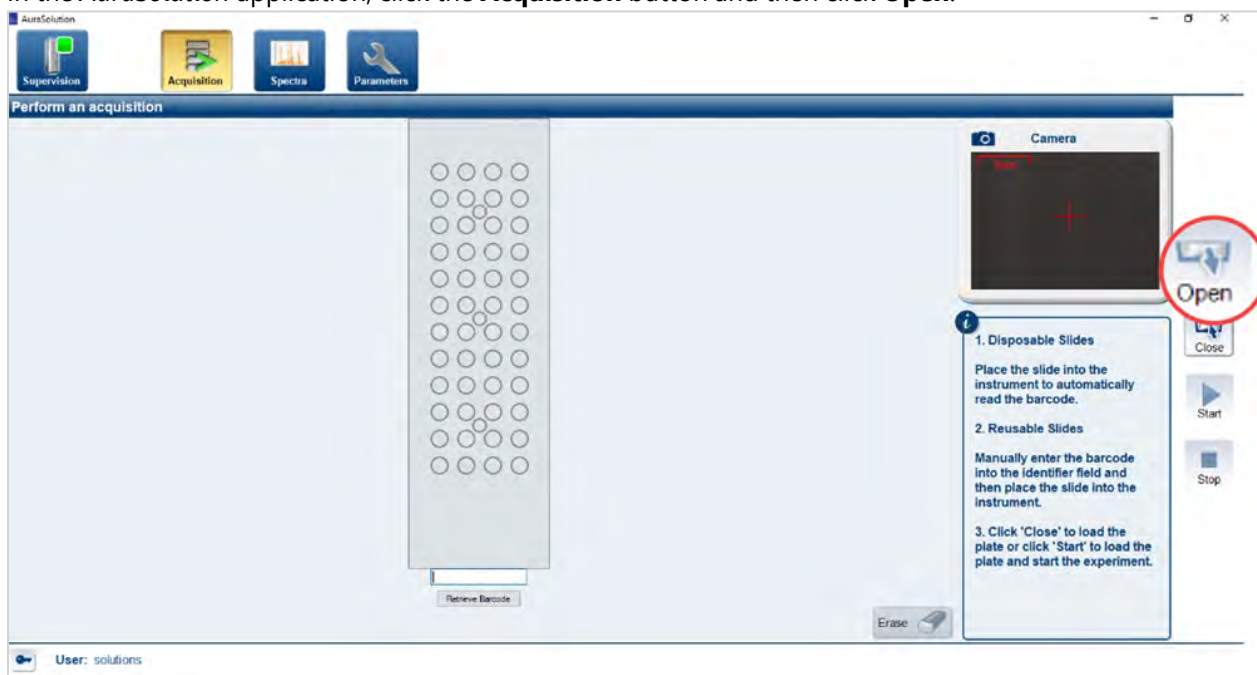
Before you begin

Before inserting a target plate, you must first create a sample worklist using the *SampleStation* application (see [Using SampleStation](#)). This will provide *AuraSolution* with details of which sample positions to use when acquiring data during an experiment.

For more information on target plate handling and sample preparation, see [Working with target plates](#). You should observe the recommended working practices and techniques in this section of the user guide before attempting the procedure described below.

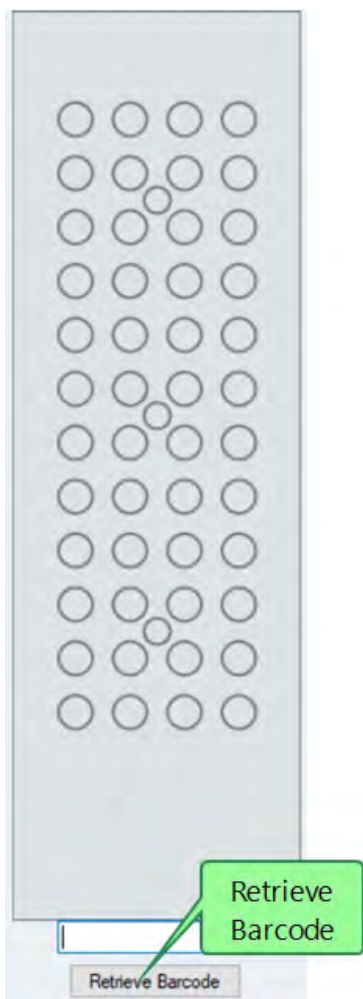
Loading a target plate

1. In the *AuraSolution* application, click the **Acquisition** button and then click **Open**.



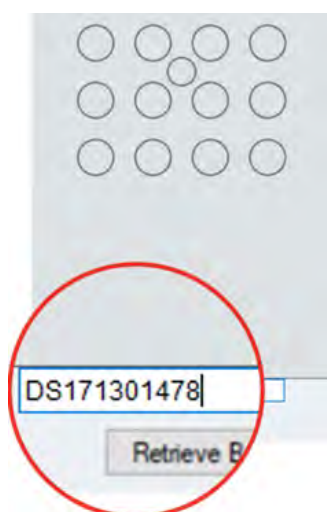
2. Wait until the status LED on the instrument front panel blinks **green** (this can take several minutes).
3. Follow steps 2 through 5 of the procedure for loading a target plate into the instrument (see [Loading and unloading target plates](#)).
4. If you loaded a reusable target plate, enter the worklist identifier you assigned to the plate when creating the sample worklist (see [Managing worklists](#)) directly into the box below the slide graphic, and then press **Enter**.

Note: If you loaded a disposable target plate, the bar code on the plate (which uniquely identifies it) will be read and entered into the box when the **Retrieve barcode** button is clicked:

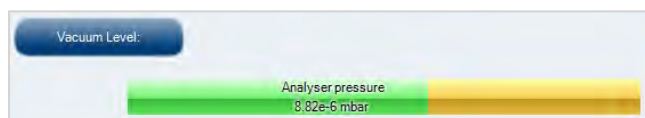


You must still however press the keyboard **Enter** key to register the slide with the application.

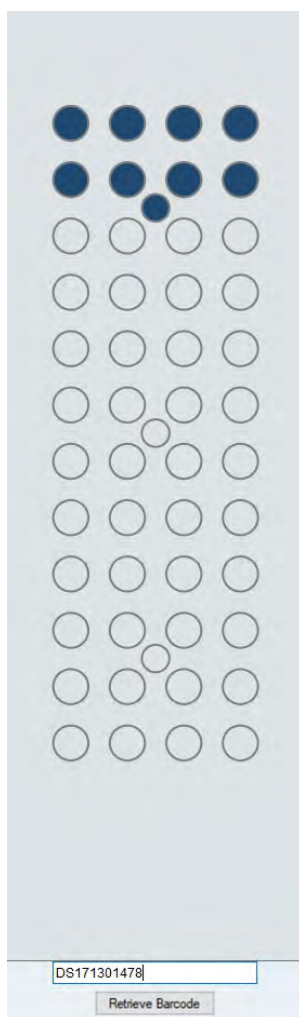
5. The bar code reader scans the bar code and displays the code. Reading the bar code may take a few seconds. An example successful read is shown below:



6. Click **Close**, and then wait for the **Vacuum Level** indicator to fully change from **orange** to **green** (this can take several minutes).



If the plate has been correctly identified, you will see a graphical representation of the plate in the **Perform an acquisition** dialog. The graphic displays the identifier for the plate, with the sample spots and calibration spot (as defined in the sample worklist) highlighted in **blue**.



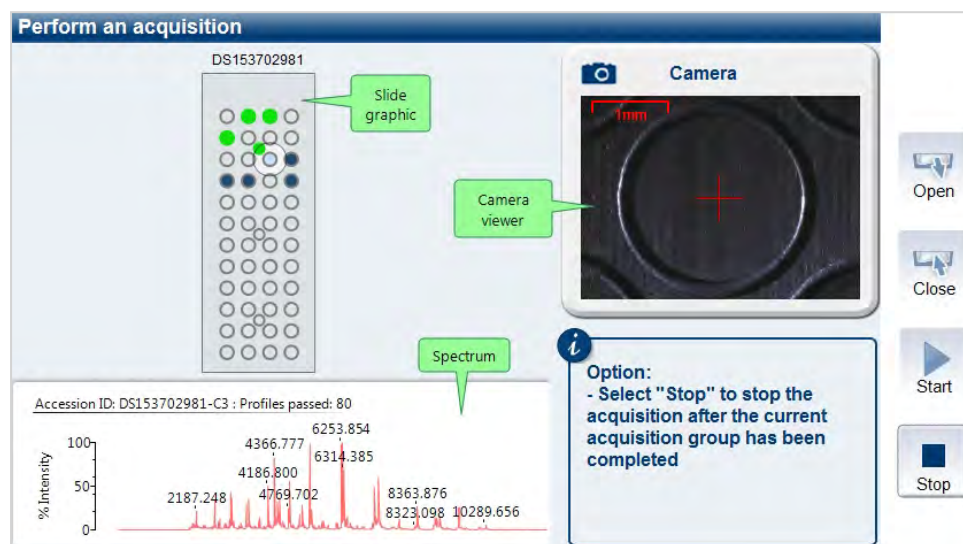
7. If the plate cannot be identified, click **Erase** and then either re-enter the identifier (for reusable plates) or unload the plate and repeat steps 1 through 5 to load a new plate.

Running experiments

An experiment is an automated series of acquisitions performed on a predefined set of samples. Before you begin, you must already have performed the procedures described in [Setting parameters](#) and [Loading a target plate](#).

1. In the *AuraSolution* application, click the **Acquisition** button.

The **Perform an acquisition** dialog is displayed.



2. To begin the experiment, click **Start**.
3. Wait for the experiment to end, or click **Stop** to terminate the experiment after the current acquisition group (as defined in the sample worklist) has been completed.

Notes:

- Progress is indicated on the slide graphic - the following symbols are used to indicate the spot status.

	No sample.
	Acquired during previous acquisition.
	Waiting for acquisition.
	Spectrum is being acquired.
	Spectrum has been acquired and passed quality checks.
	Spectrum has been acquired and passed quality checks but there is an error exporting data.
	Spectrum has been acquired and failed quality checks. Resulting spectrum peak list is available for exporting.
	Spectrum has been acquired and failed quality checks.
	Selected for re-acquisition.

- The spectrum window displays the spectral data for the current acquisition in real time;

- The camera viewer displays a video image of the instrument laser as it fires on the target plate;
- Each acquisition group is first calibrated using the designated calibration spot. If calibration is successful, a spectrum is then acquired for each defined spot in the group;
- If calibration fails, the acquisition group is ignored and the experiment proceeds to the next group;
- After each acquisition group is complete, a second acquisition of the calibration spot for the sample group is performed. If this fails, an error message is displayed.

Once the experiment has completed, you can analyse the results by right-clicking on a spot in the slide graphic, or clicking the **Spectra** button (see [Analysing results](#)). Note that right-clicking only displays spectral data for the current experiment. If you wish to view historical data, click the **Spectra** button.

Re-acquiring data

If you need to re-acquire data from the target plate after the experiment is complete (or you have manually stopped it), left-click on each spot you wish to re-acquire and then click **Start**. You can also re-acquire data from a previous experiment by re-loading the target plate associated with it.

Spectrum display

The spectrum displayed in the **Perform an acquisition** dialog shows intensity % (Y axis) versus m/z (X axis) with labels indicating the main peak masses. It also displays the **Accession ID** (target plate identifier and current sample position), and the number of profiles which passed the specified data quality parameters (see [Data quality](#)).

Note: If more than one spot shares the same **Accession ID**, right-clicking the spot always displays the spectrum for the last acquired spot.

Analysing results

The spectral data produced after running an experiment (see [Running experiments](#)) is stored in the instrument database and may be examined at any time by clicking the **Spectra** button, or right-clicking on an acquired spot in the **Perform an acquisition** dialog.

Note: If you right-click on a spot, you will only see data for the current experiment. Clicking the **Spectra** button allows you to view data for all historic experiments.

Viewing a spectrum

The **View Spectra** dialog displays the currently selected spectral data. The following views are displayed.



- **Average** - displays the average of all the spectra collected from the sample;
- **Processed Average** - displays the averaged data after applying smoothing, baseline subtraction, and peak detection to the data;
- **Peaks** - displays the centroid/apex mass peaks found in the processed data.

The screenshot shows the 'View Spectra' dialog with the following components:

- Search criteria:** Search filter with 'Slide ID: DS172302621' and 'Sample ID:' fields, and a 'Search' button.
- Results:** A table with columns 'Acquisition Time', 'Position', and 'User'. The row '08/02/18 10:19 J1 user1' is selected and highlighted in blue. A green callout 'Selected spectrum' points to this row.
- Details:** Profiles passed: 100, Profiles failed: 0, Spectrum Quality: Data Passed.
- Average:** A plot of % Intensity vs. mass-to-charge ratio showing several peaks. A green callout 'Toolbar' points to the top right corner.
- Processed Average:** A plot showing the same data with peaks identified and labeled with their mass-to-charge ratios: 4122.337, 6181.550, 6291.943, 12352.402, and 12561.028.
- Peaks:** A plot showing the identified peaks with their mass-to-charge ratios. A green callout 'Scroll bar' points to the bottom left, and another green callout 'Zoom control' points to the bottom right.

Notes:

- If more than one spectrum was produced for a given sample position, switch between them by selecting an entry in the **Results** box;
- In the **Results** box, the format of the acquisition date is always **DD/MM/YY**, regardless of the date format set by the operating system;
- To view data for a different sample position (or historical data for another slide), enter a **Slide ID** and/or **Sample ID** in the **Search filter** boxes and then click **Search**;

- The **Details** section of the dialog shows the number of profiles which passed the specified data quality parameters (see [Data quality](#));
- Use the scroll bar or scroll button on the toolbar  to move the spectrum right/left (when using the scroll button, click and drag directly in the spectrum window);
- Use the zoom control or zoom button on the toolbar  to enlarge a part of the spectrum (when using the zoom button, click in the spectrum window and drag out to select a zoom region).

Alarm reporting

The **Supervision** button on the *AuraSolution* toolbar acts as an indicator which will tell you if the instrument develops a fault while running an experiment. The button has two states.



Normal operation Fault detected

If the button indicates a fault with the instrument, click it for more details.

Supervision

System status: Pumping, load pumped

Analyser pressure: 2.55e-5 mbar

Load lock valve: Closed

Door: Closed

Source vent valve: Closed

Source backing valve: Open

Laser: 100

Source turbo: 1500 Hz

Backing pressure: 1.02e+0 mbar

List of Alarms

Description	Status
The instrument has reported an error.	Error
The start/stop operation on the acquisit...	Warning

Description: The start/stop operation on the acquisition has failed

Status:

Alarm date: 07/09/2017 11:48:25

Advice: If the problem continues please contact your administrator.

On the **Supervision** dialog, you can:

- View a representation of the current instrument status (see [Instrument status](#));
- Double-click on an entry in the **List of Alarms** box to see a description of the error.

If an instrument fault occurs, you should follow the advice given in the error description. If no advice is given, or you are unable to resolve the error, report the fault to your local Shimadzu service centre or distributor (see [Servicing](#)).

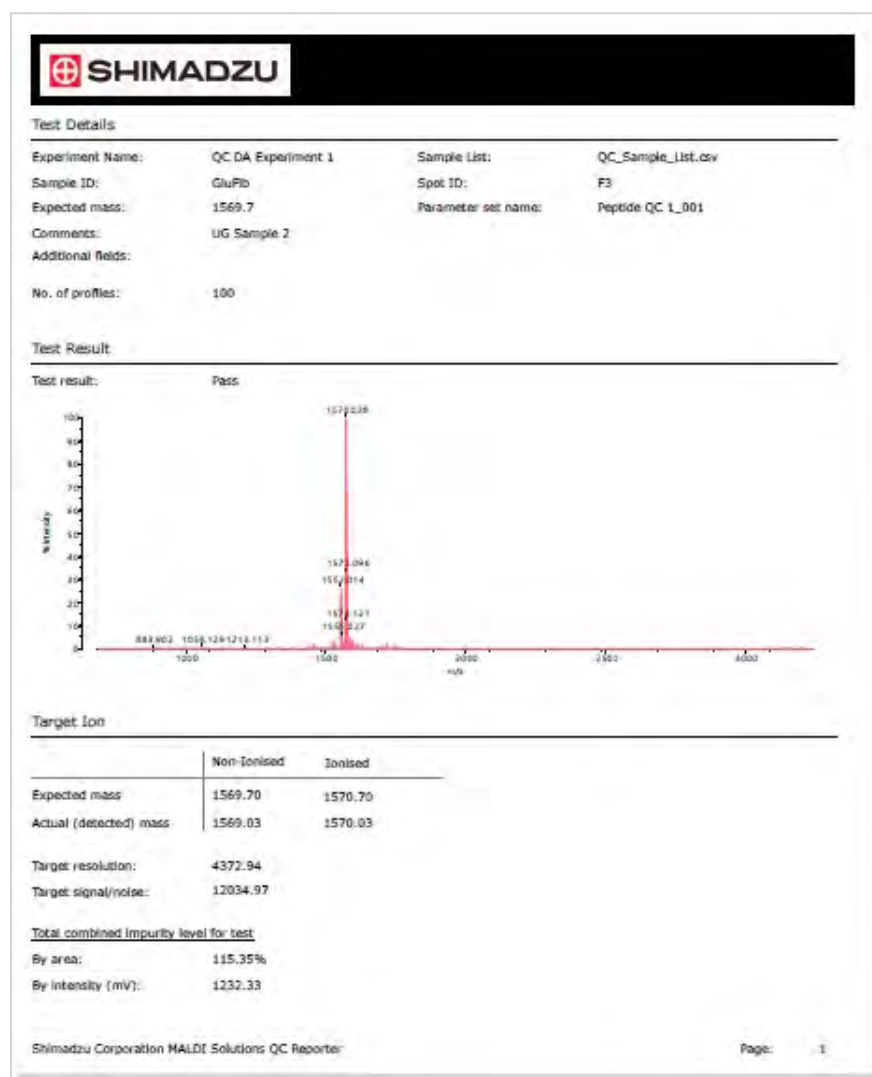
Note: You can also use the **Supervision** dialog to monitor the instrument status during normal operation.

Chapter 7 - Using QC Reporter

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Introduction

QC Reporter is an optional software application which is only compatible with MALDI-8020 and MALDI-8030 when using positive ion mode. However, *QC Reporter* does not support negative mode when using the MALDI-8030 instrument. *QC Reporter* allows you to perform quality control (QC) or quality assurance (QA) experiments on a set of samples. It is typically used for experiments with peptides, oligonucleotides or small molecules. *QC Reporter* works with the *MALDI Solutions Data Acquisition* application, so it is recommended that you are familiar with the latter before attempting to use *QC Reporter* (see [Acquiring data](#)).

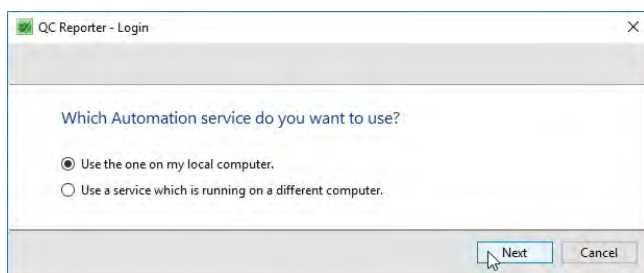


The quality of samples in an experiment is assessed in terms of the presence (or the absence) of the of the correct molecular weight, relative level of impurities and known adducts or contaminants within a preset tolerance. Individual samples are then passed (or failed) depending on their adherence to the specified criteria. The application allows you to view results during the experiment, and to produce reports in a customised format, as shown in the example above.

Note: *QC Reporter* is an optional licensed software application. These features may not be available on your instrument unless the correct software licence has been purchased. Please contact your local Shimadzu representative for details.

Starting the application

1. From the **Start** menu, locate and select **Shimadzu Corporation > QC Reporter**.

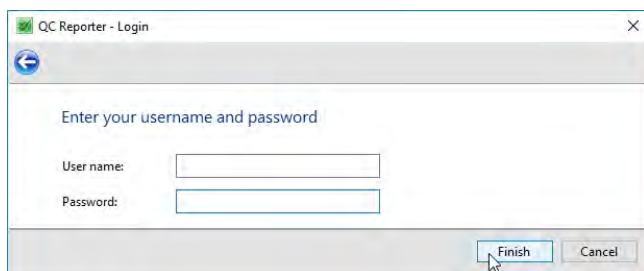


2. Select the automation service you wish to use and then click **Next**.

Notes:

- If you are using the PC workstation connected to the instrument, select **Use the one on my local computer**;
- If you wish to connect to a remote database (i.e. on another instrument) click **Connect directly to a known database**, however, this option not currently supported.

3. Enter your **User name** and **Password**, and then click **Finish**.



You will see the main menu of the application as shown below.



Note: Depending on your [user account privileges](#), you may not see all the options shown above.

Using parameter sets

When creating a new experiment, you must first create two QC parameter sets. These consist of the following elements.

Acquisition and criteria

This parameter set type contains the following elements.

- **QC Criteria** - these settings specify the conditions used to determine whether or not acquired sample data passes or fails the QC experiment;
- **Acquisition** - the settings used to acquire data from samples;
- **Data processing** - the settings used to process data acquired from samples.
- **Export** - the settings used to export the data.

Calibration

This parameter set type contains the following elements.

- **Calibration** - a list of reference masses used to calibrate the instrument during the experiment;
- **Acquisition** - the settings used to acquire data from calibrant samples;
- **Data processing** - the settings used to process data from calibrant samples.

With the exception of the **QC Criteria**, each of these elements can either be entered directly, or imported from a named parameter set created in the *MALDI Solutions Data Acquisition* application and then saved to the instrument database. The latter is the recommended approach, as some parameters can only be determined by experimentation depending on the types of samples you wish to analyse. Using saved parameter sets also allows you the opportunity to re-use parameter sets across different experiments and thus increase workflow efficiency.

Note: The application also requires a starting calibration, which is used as a reference point at the start of an experiment. This must be created in *MALDI Solutions Data Acquisition* and then saved to the instrument database before running the experiment.

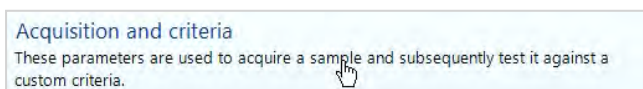
Creating a QC parameter set

Note: This procedure assumes you are creating a parameter set before creating or loading an experiment. If you are creating a parameter set after creating or loading an experiment, begin this procedure at step 2.

1. In *QC Reporter*, select **Create a new parameter set**.



2. In the **Select parameter set type** dialog, select **Acquisition and criteria**.



3. In the **Parameter Editor** dialog, use the arrow button  to expand the **QC Criteria** pane.

Select the criteria that constitutes a QC test. A QC test is marked as Pass if none of the Fail or Query criteria are met

Include in test	Criteria	Value	Measure	Fail	Query	Special Condit..
Absolute Criteria						
<input checked="" type="checkbox"/>	Mass tolerance is greater than	2	Da	<input checked="" type="radio"/>	<input type="radio"/>	<input type="radio"/>
<input type="checkbox"/>	S/N threshold is less than	50		<input checked="" type="radio"/>	<input type="radio"/>	
<input type="checkbox"/>	Minimum resolution	1000		<input checked="" type="radio"/>	<input type="radio"/>	
<input type="checkbox"/>	Sum of adducts is greater than	50	Apex(mV)	<input checked="" type="radio"/>	<input type="radio"/>	
<input type="checkbox"/>	Maximum impurity level	80	Apex(mV)	<input checked="" type="radio"/>	<input type="radio"/>	
Range Criteria						
<input type="checkbox"/>	S/N is within range			10	45	
<input type="checkbox"/>	Sum of adducts is within range		Apex(mV)	1	7	
<input type="checkbox"/>	Min > max acceptable impurity level		Apex(mV)	60	90	

Special Conditions - Optional additional specific criteria that may be applied

	Fail	Query
Mass NOT within tolerance but Adducts found:	<input checked="" type="radio"/>	<input type="radio"/>

Define the Mass calculation Type: Monoisotopic Average

Define Adducts... 0 selected
 Define Amino Acids... 0 selected
 Define Matrix Ions... 0 selected
 Define Impurities... 0 selected

4. Enter the criteria you wish to specify for your experiment.

Notes:


- The settings in this dialog define the pass/fail conditions which will be applied to each sample when the experiment is run;
- You can include or exclude a criterion by selecting or de-selecting the check box associated with it (the mass tolerance criterion is always selected).

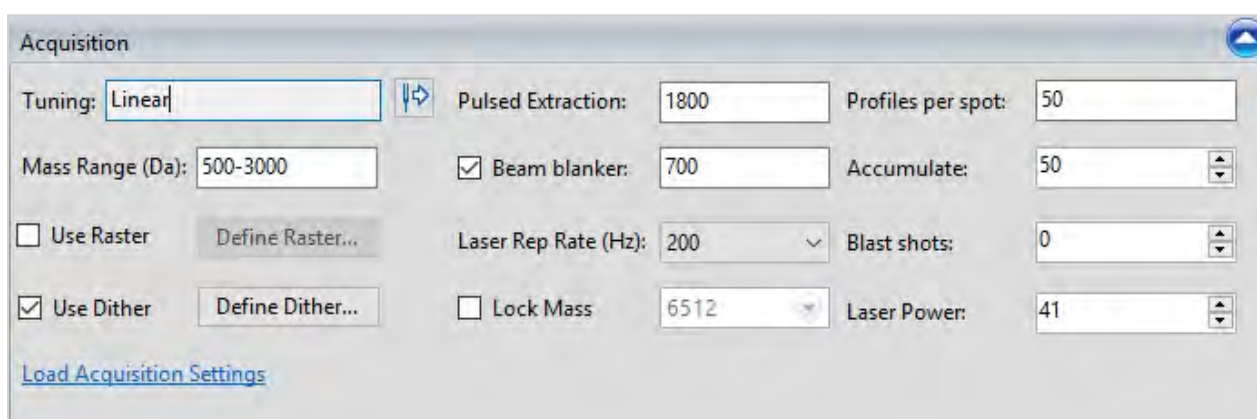
The criterion you can enter on this dialog are defined as follows.

Absolute Criteria	
Mass tolerance is greater than	<p>The allowable difference (using the selected units) between the expected mass and the observed mass. Deviations of more than the entered amount will trigger a Fail or Query result as selected with the accompanying radio buttons. Alternatively, you may select the Special Conditions button, in which case you can define one or more adducts whose presence in the sample changes the result criteria (see below).</p> <p>Note: Defining a tolerance using ppt or ppm units allows you to specify the tolerance as a proportion of the mass, e.g. a tolerance of 800 ppm results in a tolerance of ± 0.8 Da for a mass of 1000 Da.</p>

S/N threshold is less than	The amount of signal-to-noise ratio (SNR) allowable. If an SNR is observed below the entered range, the sample is failed or queried as indicated by the radio buttons.
Minimum resolution	The lowest acceptable mass resolution. If a resolution is returned that is less than this figure, the sample is failed or queried as indicated by the radio buttons.
Sum of adducts is greater than	A maximum figure for the allowable sum of adduct peaks, expressed in either mV or % area. If a larger figure is calculated, the sample is failed or queried as indicated by the radio buttons. Note: See below for details on specifying adducts.
Maximum impurity level	If the sum of all peaks (except the expected mass), expressed in either mV or % area, is greater than this value, the sample is failed or queried as indicated by the radio buttons.
Range Criteria	
S/N is within range	A minimum and maximum value for the acceptable SNR allowable. <ul style="list-style-type: none"> • If the value is below the range, the sample is failed; • If the value is within the range, the sample is queried; • If the value is above the range, the sample is passed.
Sum of adducts is within range	A minimum and maximum figure for the allowable sum of adduct peaks, expressed in either mV or % area. <ul style="list-style-type: none"> • If the value is below the range, the sample is passed; • If the value is within the range, the sample is queried; • If the value is above the range, the sample is failed.
Min > max acceptable impurity level	A minimum and maximum figure for the sum of all peaks (except the expected mass), expressed in either mV or % area. <ul style="list-style-type: none"> • If the value is below the range, the sample is passed; • If the value is within the range, the sample is queried; • If the value is above the range, the sample is failed.
Other	
Special conditions	An alternative criterion to use when the target mass may not be present as protonated species but as adduct. With this selection enabled, you can either generate a Query or a Fail result. In all cases, even if the target mass will not be found, the report will indicate the presence of any found adducts.
Define the mass calculation type	Choose to use either the Monoisotopic or Average method when detecting peaks (applies to the experiment as a whole).
Define Adducts	Click this button to define a list of adducts which will be used with the mass tolerance criterion (see Using the Compounds Browser).

Define Amino Acids	Click this button to define a list of amino acids whose loss will be identified on the relevant peak(s) displayed in the results report (see Using the Compounds Browser).
Define Matrix Ions	Click this button to define a list of matrix ions which will be identified on the associated peak(s) displayed in the results report (see Using the Compounds Browser).
Define Impurities	Click this button to define a list of impurities which will be included when the experiment considers impurity levels (see Using the Compounds Browser).

- Use the arrow button  to expand the **Acquisition** pane.
- On the **Acquisition** dialog, enter the acquisition parameters to be used for your experiment or click **Load Acquisition Settings** to load a parameter set you previously created in *MALDI Solutions Data Acquisition*.






The screenshot shows the 'Acquisition' dialog box with the following settings:

- Tuning: Linear
- Mass Range (Da): 500-3000
- Use Raster: (button: Define Raster...)
- Use Dither: (button: Define Dither...)
- Pulsed Extraction: 1800
- Beam blanker: 700
- Laser Rep Rate (Hz): 200
- Lock Mass: 6512
- Profiles per spot: 50
- Accumulate: 50
- Blast shots: 0
- Laser Power: 41

A [Load Acquisition Settings](#) link is visible at the bottom left.

Note: These acquisition settings will be used for acquiring data from each sample in the experiment. However, the QC Reporter does not support negative mode when using the MALDI-8030 instrument. For more information on how to use the settings, see [Setting acquisition parameters](#).

- Use the arrow button  to expand the **Data Processing** pane.
- On the **Data Processing** dialog, enter the processing parameters to be used for your experiment or click **Load Data Processing Settings** to load a parameter set you previously created in *MALDI Solutions Data Acquisition*.

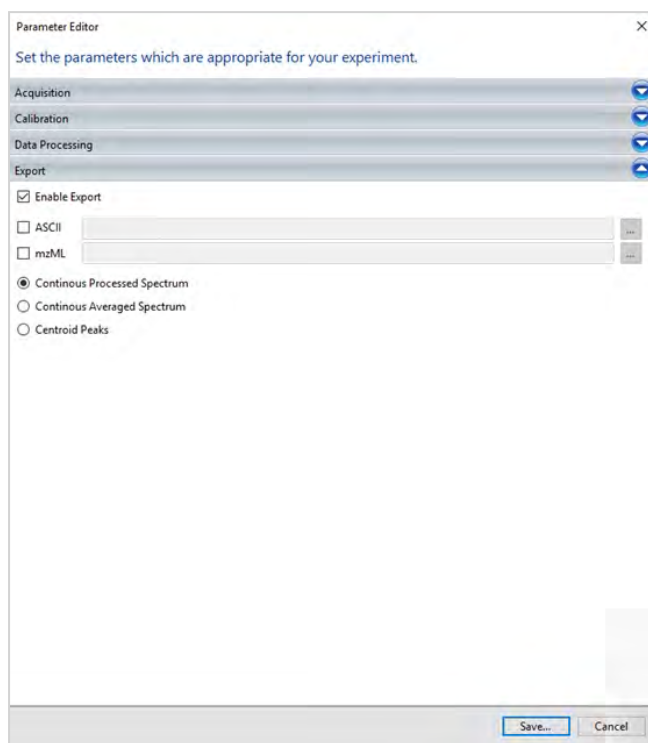
<input checked="" type="checkbox"/>	Subtract baseline using filter width:	<input type="text" value="0"/>
<input checked="" type="checkbox"/>	Smooth data using filter width:	<input type="text" value="10"/>
Peak delimiter method:		<input type="text" value="Threshold"/>
Threshold type:		<input checked="" type="radio"/>  <input type="radio"/> 
Threshold offset:		<input type="text" value="1.00"/> mV
Threshold response:		<input type="text" value="1.00"/>
Peak location method:		<input type="text" value="Apex"/>
Minimum peak width:		<input type="text" value="10"/>
<input type="checkbox"/>	Monoisotopic peak picking	
	Minimum mass:	<input type="text" value="800"/>
	Maximum mass:	<input type="text" value="1000"/>
	Minimum isotopes:	<input type="text" value="2"/>
	Maximum intensity variation:	<input type="text" value="50"/>
<input checked="" type="checkbox"/>	Overlapping distributions	
	Minimum peak percent:	<input type="text" value="40"/>

[Load Data Processing Settings](#)

Note: These settings will be used for processing data from each sample in the experiment. For more information on how to use the settings, see [Processing data](#).

9. If the data is to be exported, do the following. Alternatively, if the data is not required, go to [step 14](#).

10. Navigate to the Export tab.



11. Select Enable Export to export the sample data on-the-fly whilst the QC Experiment is running.
12. Specify either ASCII or mzML separately or select both and when prompted, specify the location where the file is to be exported to.
13. Select one of the following options:
 - Continuous Processed Spectrum.
 - Continuous Averaged Spectrum.
 - Centroid Peaks.

The data will be exported to the specified location.


14. Click **Save**.

You will be prompted to assign attributes and a name to the QC saved parameter set (see [Loading & saving data](#)).

Note: If the same Acquisition or Calibration parameter set is saved with the same name, the software does not overwrite the new parameter set to the previous one, but it creates a duplicate to which a numerical extension is automatically added. For example, if a parameter set is edited and saved multiple times as QC Exp, it will be saved each time as QC Exp_001, QC Exp_002 etc.

15. Repeat steps 1 and 2 this time selecting **Calibration** in the **Select parameter set type** dialog.

Note: If you are creating a parameter set after creating or loading an experiment, repeat step 2 only.

16. Use the arrow button  to expand the **Calibration** pane.

Mass	Formula	Abundance	Description
1046.54		Monoisotopic	Angiotensin2
1296.69		Monoisotopic	Angiotensin1
1570.68		Monoisotopic	Glu-1-fibrino
1800.94		Monoisotopic	Acetyl renin
2093.09		Monoisotopic	ACTH 1-17

Tolerance: 10 Da

Compounds

Load References

- Enter the list of reference masses to be used for your experiment using one of the following methods.
 - Click **Load References** to load a previously saved list of references (see [Loading & saving data](#)).
 - Click **Compounds** to select entries based on their chemical names (see [Using the Compounds Browser](#)).
 - Enter the list of masses directly using the on-screen form by typing the expected m/z values into the **Mass** column and selecting the correct **Abundance** type from the drop-down list. You can press **Enter** to add a new row to the list, **Delete** to remove an entry, or click on a column header to sort the list by the selected column value.
- Repeat steps 5 through 8 to set the acquisition and data processing parameters you wish to use when acquiring calibrant samples. Repeat steps 9 to 13 if you wish to export the calibrant data on-the-fly.

Note: These may be different from those used to acquire and process data from the regular samples.
- Click **Save**.

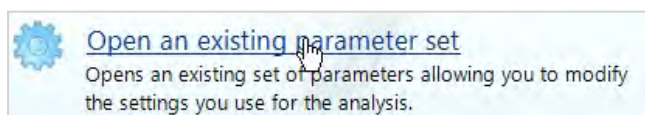
You will be prompted to assign attributes and a name to the QC saved parameter set (see [Loading & saving data](#)) before returning to the main application menu.

Note: If the same Acquisition or Calibration parameter set is saved with the same name, the software does not overwrite the new parameter set to the previous one, but it creates a duplicate to which a numerical extension is automatically added. For example, if a parameter set is edited and saved multiple times as QC Exp, it will be saved each time as QC Exp_001, QC Exp_002 etc.

Editing a QC parameter set

Note: This procedure assumes you are editing a parameter set before creating or loading an experiment. If you are editing a parameter set after creating or loading an experiment, begin this procedure at step 2.

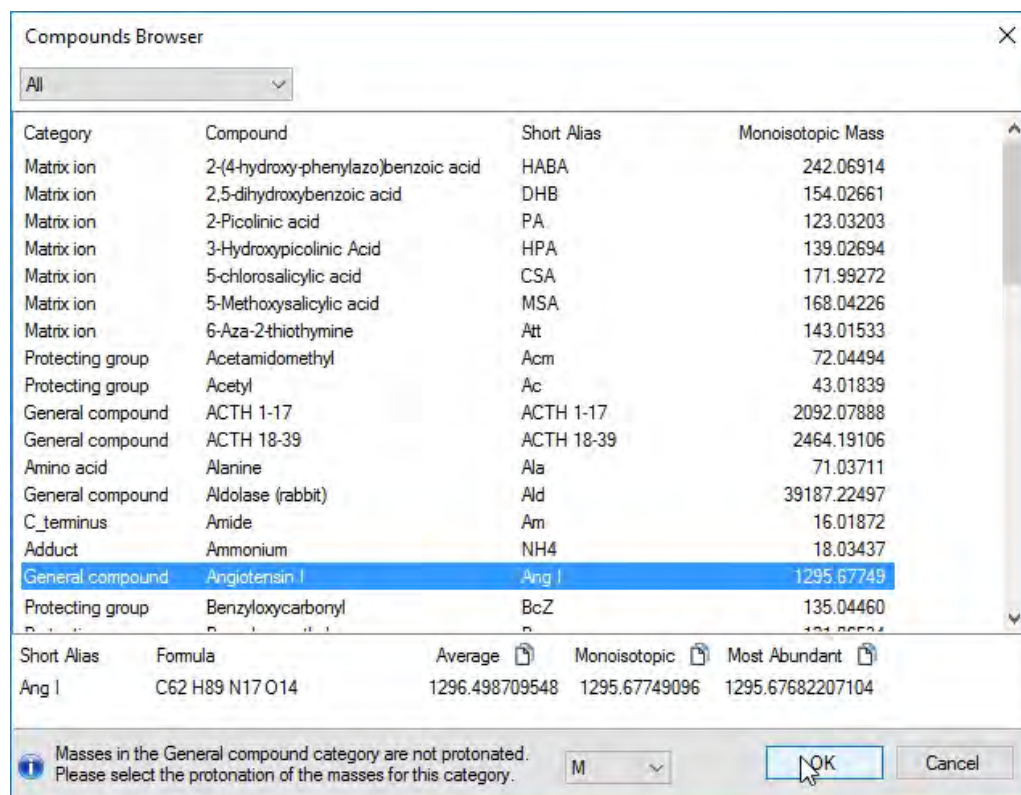
- In *QC Reporter*, select **Open an existing parameter set**.



- Locate and select the desired parameter set by using the browser controls (see [Loading & saving data](#)).
- Follow the steps described above (see [Creating a QC parameter set](#)) to make changes to the loaded parameters.

Using the Compounds Browser

The **Compounds Browser** is a utility feature which allows you to select entries from a database of known compounds to be entered into the parent dialog when creating or editing parameter sets.



To select a compound, click on a list entry and then click **OK**, or **Cancel** to abort the compound selection.

Notes:

- You can re-order the list of compounds by clicking on a column header to sort the list by the selected column entries;
- Use the drop-down list to show only entries in a particular category (this will be done automatically if you clicked a specific button while creating or editing parameters. For example, if you clicked the **Define Adducts** button you will only see adduct compounds in the list;
- Compounds in the **General compound** category require you to specify a protonation type before clicking **OK**.

Creating a sample list

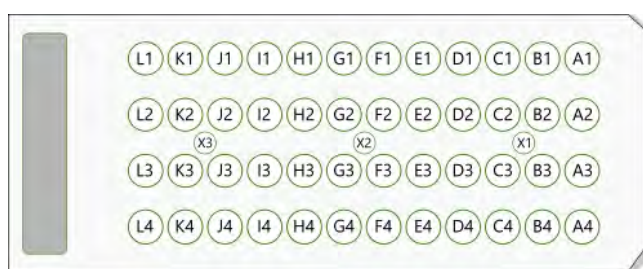
Experiments in *QC Reporter* use sample lists to define the spot positions on a loaded target plate containing samples and calibrants, the target mass each sample is expected to conform to, and the parameter set(s) to use during acquisition.

Sample lists are created in an external spreadsheet application or text editor, and use the industry-standard CSV (Comma Separated Value) format. Within the sample list file, the following data items are expected.

Column Label	Data Type	Description
Spot ID*	alphanumeric	The sample position (or spot) containing the sample, e.g. A2
Target mass*	numeric	Expected mass of the sample e.g. 1045.5 Note: The target mass specified should be in the neutral (non-ionised) form, as the software will calculate the protonated species automatically when producing the results.
Sample ID	alphanumeric	Identifies the sample by name e.g. Angio II
Comments	alphanumeric	Explanatory comment related to the sample e.g. Subject 12
Parameter set*	alphanumeric	The name of the saved parameter set to use when acquiring data for the sample e.g. QC Exp_001 . For samples, this will be an Acquisition and criteria parameter set. For calibration spots, this will be a Calibration parameter set. Note: Names should be entered exactly as they are saved in the instrument database, or the experiment will not run. Note: When specifying the parameter set in the QC sample list, if there are multiple versions of the same parameter set, it is necessary to specify the correct numerical extension, else the parameter set will not be found or the wrong version may be used.
Additional	alphanumeric	Any other text for user information only (not displayed or used in the application).

Notes:

- Entries marked with an asterisk (*) are considered mandatory;
- The first row of the sample list should contain the column labels as represented above;
- When entering the **Spot ID**, use the following diagram as a guide.



Creating a sample list with a spreadsheet

The example below shows how the sample list should be structured in a spreadsheet application (e.g. **Microsoft Excel**).

	A	B	C	D	E
1	Spot ID	Target mass	Sample ID	Comments	Parameter set
2	x2		cal	Cal	Cal_002
3	H1	1569.7	sample	sample	Peptides_005
4	H2	1569.7	sample	sample	Peptides_005
5	H3	1569.7	sample	sample	Peptides_005
6	G1	1569.7	sample	sample	Peptides_005
7	G2	1569.7	sample	sample	Peptides_005

Note: When saving from a spreadsheet, always select the **Save As** (or equivalent) menu option and then select **CSV** as the target format, saving the file with the **.csv** extension, e.g. **sample_list.csv**

Once saved, you can check that the output format is correct by loading it into a plain text editor and making sure it matches the structure shown below.

Creating a sample list using a text editor

The example below shows how the sample list should be structured in a text editor application (e.g. **Microsoft Windows Notepad**).

```
samp_list.csv - Notepad
File Edit Format View Help
Spot ID,Target mass,Sample ID,Comments,Parameter set,Additional
x1,0,Cal TOFMix,UG Sample Cal,Peptide Cal QC_001,Standard calibration
F2,1569.7,Glu Fib,UG Sample #1,Peptide QC 1_001,Main Sample
F3,1569.7,Glu Fib,UG Sample #2,Peptide QC 1_001,Control
G2,1560.7,TOFMix,UG Sample #3,Peptide QC 1_001,Main Sample
G3,1560.7,TOFMix,UG Sample #4,Peptide QC 1_001,Control
```

Note: When saving from a text editor, always save the file with the **.csv** extension, e.g. **sample_list.csv**

Running experiments

Before running an experiment using *QC Reporter*, you must first have performed the following prerequisite tasks.

- Prepared a set of samples on a suitable target plate (see [Working with samples](#));
- Created and saved a sample list which describes the sample arrangement on the target plate (see [Creating a sample list](#));
- Created and saved a calibration and acquisition parameter set, and a calibration file to use as a starting calibration for the instrument (see [Using parameter sets](#)).

It is also recommended that you do the following.

- Create and save one or more named acquisition parameter sets suitable for acquiring data from your samples;
- Create and save one or more named data processing parameter sets suitable for processing the data acquired from your samples;
- Create and save a named reference list of the masses you expect to detect during the experiment.

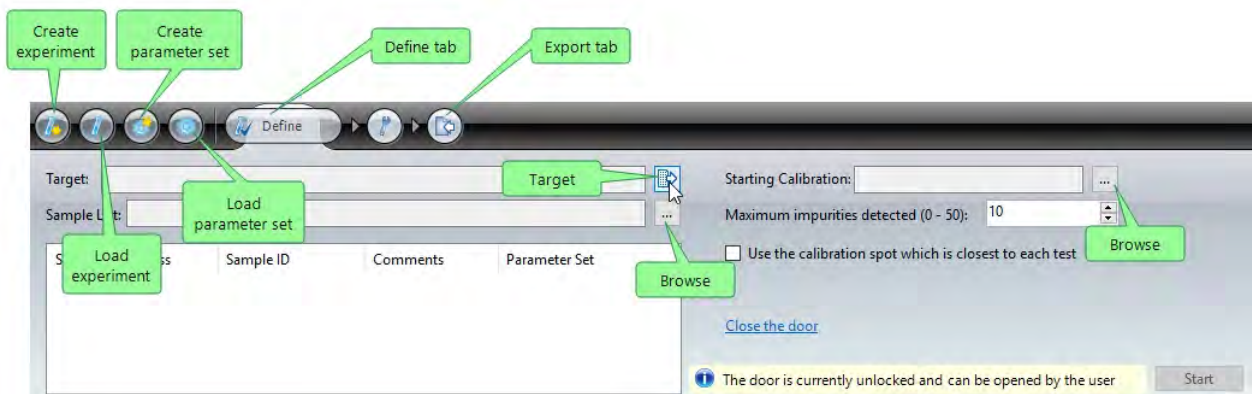
Note: Although you can set these parameters within *QC Reporter*, it is advised that you create them independently in *MALDI Solutions Data Acquisition*. See [Using parameter sets](#) for details.

Creating an experiment

1. In *QC Reporter*, select **Create a new experiment**.



The main application window will be displayed.



2. On the **Define** tab, click the **Target** icon and then select the type of target plate you wish to use.

Note: On the MALDI-8020 and MALDI-8030 instruments, you can only select the **1x48 2.8 mm wells** plate type.

3. In the **Sample List** box, click the browse icon (...) and then select the sample list file you created previously (see [Creating a sample list](#)).

The sample list will be populated with the entries contained in the selected file and then displayed on screen.

Spot ID	Mass	Sample ID	Comments	Parameter Set
x2	0	Cal TOFMix	UG Sample Cal	Peptide Cal QC_001
F2	1569.7	Glu Fib	UG Sample #1	Peptide QC 1_001
F3	1569.7	Glu Fib	UG Sample #2	Peptide QC 1_001
G2	1560.7	TOFMix	UG Sample #3	Peptide QC 1_001
G3	1560.7	TOFMix	UG Sample #4	Peptide QC 1_001

The arrangement of sample spots will also be displayed on the slide graphic in the main window.



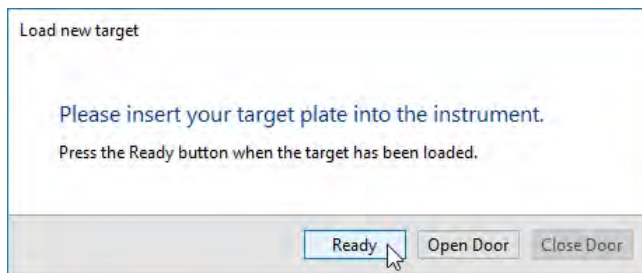
- In the **Starting Calibration** box, click the browse icon (...) and then select the starting calibration file created previously in MALDI Solutions Data Acquisitions (see [Loading & saving data](#)).
- If required, in the **Maximum impurities detected** box, use the spin control to set the maximum number of impurities which will be displayed in the final report.

Note: An impurity is a peak which the application cannot identify. If (for example) you set a maximum of three peaks and five are detected, the three peaks with the highest intensity will be displayed on the report.
- If required, select the **Use the calibration spot which is closest to each test** option.

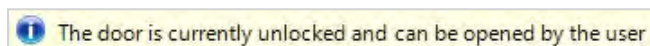
Note: This option will override the spot order described in the sample list.
- To change the settings for any of the parameter sets specified in the sample list, click the **Load parameter set** button, and then follow the procedure described in [Using parameter sets](#). The sample list .csv file should be updated with the new parameter set name/extension and re-loaded.

Note: Any parameter sets you create must be identified by name in the sample list, otherwise the experiment will not run.
- If you wish to clear the experiment definition you are currently working on and start afresh, click the **Create experiment** button.

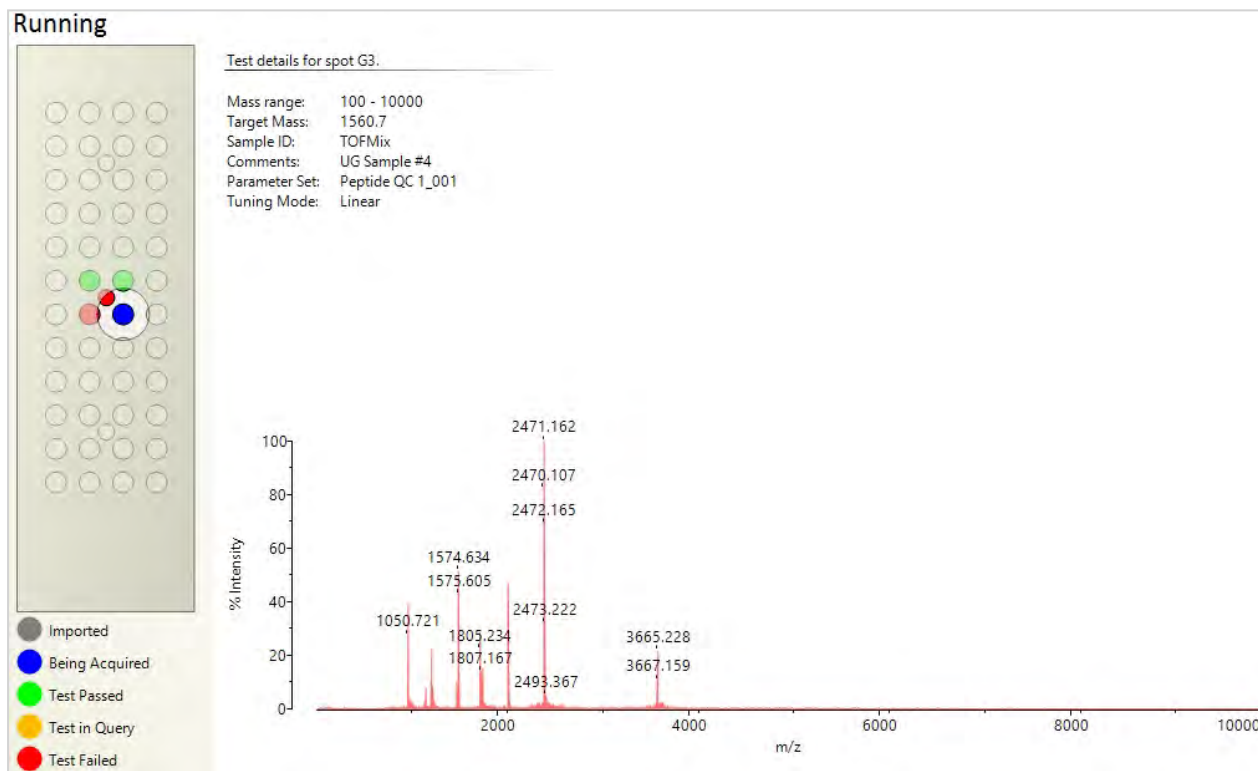
Caution: Doing this will lose all data you have entered in the application dialog.
- If you are satisfied that all experimental parameters are in order, click **Start**.
- In the **Save Data As** dialog, enter a name and any other attributes you require for the experiment (see [Loading & saving data](#)) and then click **Save**.
- When the **Load new target** dialog appears, load the target plate into the instrument (see [Loading and unloading target plates](#)) and then click **Ready**.



Note: If the door is closed, first click the **Open Door** button and then wait for the status message shown below to appear in the main application window. After loading the target in the instrument and manually closing the user door, click the **Close Door** button, then click **Ready**. If the target plate is already in the instrument, click **Ready** button directly.



- The experiment will now run, displaying a summary of the results in the main application window as it progresses.

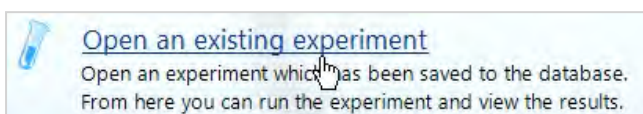


Notes:

- While the experiment is running, the camera viewer displays a real-time view of the laser as it fires at each sample (see [Using the camera viewer](#));
- Spots are colour-coded according to the on-screen key to indicate their current status;
- You can view the spectral data being acquired for each spot as the experiment progresses;
- To examine the results of the experiment, see [Viewing results](#).

Viewing an experiment

1. In *QC Reporter*, select **Open an existing experiment**.



2. Use the browser controls to navigate to the desired experiment, and then click **Load**.

The main application window will be displayed with the current experiment settings displayed.

Note: You can also load an existing experiment from the main application window by clicking the **Load experiment** button. In this case, any settings already displayed in the main window will be overwritten with the saved data.

3. Edit the experiment settings as described in **Creating an experiment** above.

Note: In order to save your changes, you must run the experiment. This will prompt you to save the experiment, overwriting the instrument database entry for the experiment with the new settings unless you specify an alternative name when saving.

Viewing results

After running an experiment, you can view the results either within the application, or through a generated report which can be customised with your company/business logo and exported to one or more report files using the industry-standard PDF file format.

Viewing individual test results

1. In the *QC Reporter* **View experiment results** pane, click on a highlighted spot on the slide graphic, or double-click on an entry in the results list corresponding to the sample you wish to examine.

View experiment results

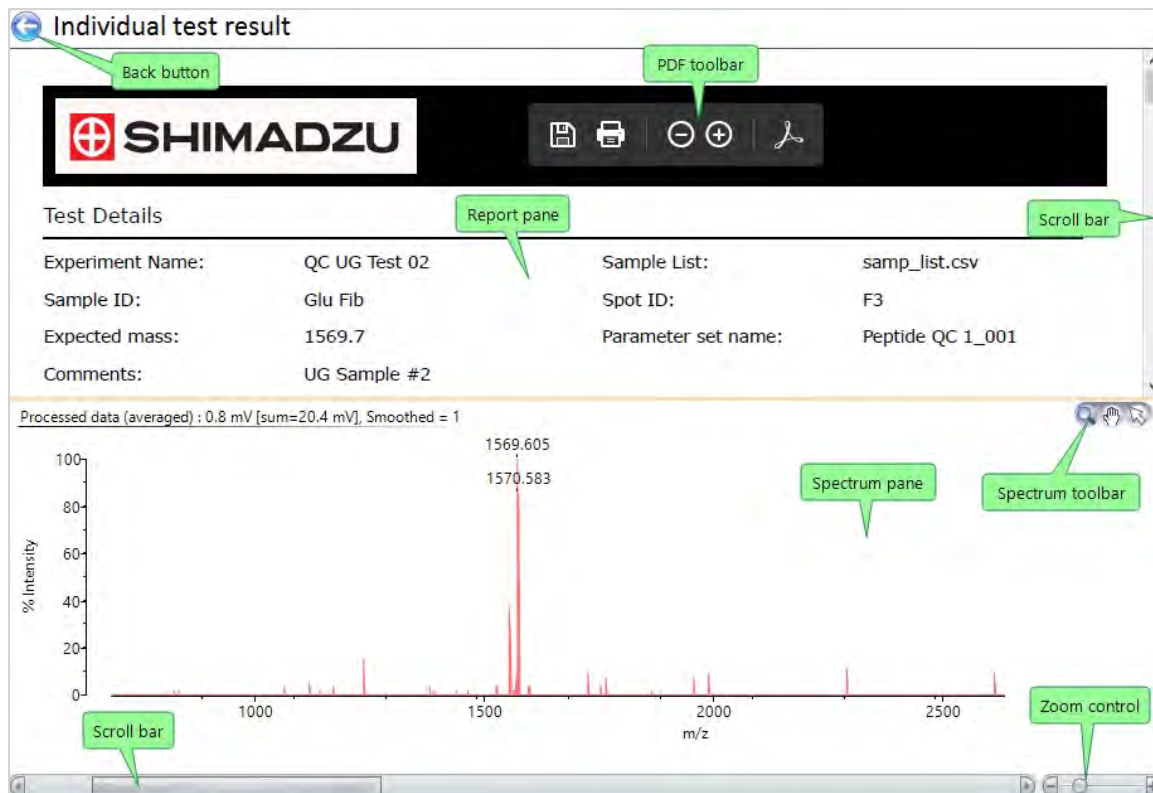
Select a test from the list below (or the target on the left) to display the results of the test in greater detail.

Spot ID	Sample ID	Parameter Set	Expected Mass	Actual Mass	Comments	Test Result
x2	Cal TOFMix	Peptide Cal QC_001		Not found	UG Sample ...	Pass
G2	TOFMix	Peptide QC 1_001	1560.7	Not found	UG Sample 3	Fail
F2	GluFib	Peptide QC 1_001	1569.7	1570.19	UG Sample 1	Pass
F3	GluFib	Peptide QC 1_001	1569.7	1569.03	UG Sample 2	Pass
G3	TOFMix	Peptide QC 1_001	1560.7	Not found	UG Sample 4	Fail

● Imported
● Being Acquired
● Test Passed
● Test in Query
● Test Failed

[View summary report](#) [View entire report](#)

The **Individual test results** pane displays the acquired spectrum for the selected sample, and a scrollable window containing a preview of the generated report.



In the **Individual test results** pane, the following controls are available.

- Scroll bars for each pane allow you to move the report preview up/down, and the spectrum left/right within their respective panes;
- The zoom control allows you to enlarge or reduce the displayed spectrum;
- The spectrum toolbar provides additional viewing tools as follows;

	Peak tool - click and drag over a spectrum to zoom, for example, to examine peaks.
	Drag tool - click and drag to scroll the spectrum.
	Move peak label tool - click a peak mass label and drag it to the required location.

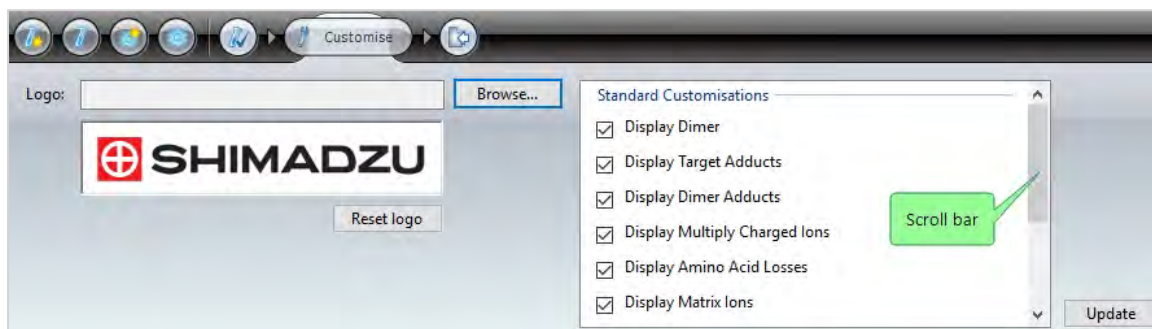
- The PDF toolbar appears when you hover over the report pane, allowing you to save, print or zoom the displayed report as required;
- Click the **Back** button to return to the **View experiment results** pane.

Viewing reports

In the **View experiment results** pane, click the **View summary report** or **View entire report** links to see an on-screen preview of either report format.

Customising reports

1. In the *QC Reporter* main application window, select the **Customise** tab.



Note: Use the scroll bar to navigate to each of the following customisation lists.

2. In the **Standard Customisations** list, deselect any display items you wish to exclude from the report.

Note: This will affect all report types.

3. In the **Individual Report Customisations** list, use the colour picker to select an alternative banner colour if required.
4. In the **Summary Report Customisations** list, use the colour picker to select an alternative banner colour if required.
5. If you want to change the default logo graphic, click Browse and then use the file browser to select an alternative logo graphic.

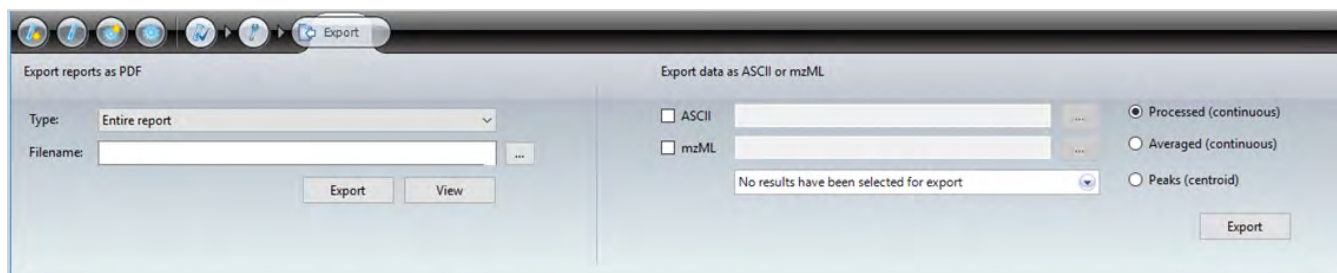
Notes:

- The image size should be 280 x 50 pixels - you can use other sizes but they may appear distorted on the report;
- The image may be in PNG, GIF, BMP or JPG format;
Note: With PNG format files, a transparent background may be used.
- You can click reset logo to revert to the default logo graphic.

6. Click **Update** to apply the changes to the report format.

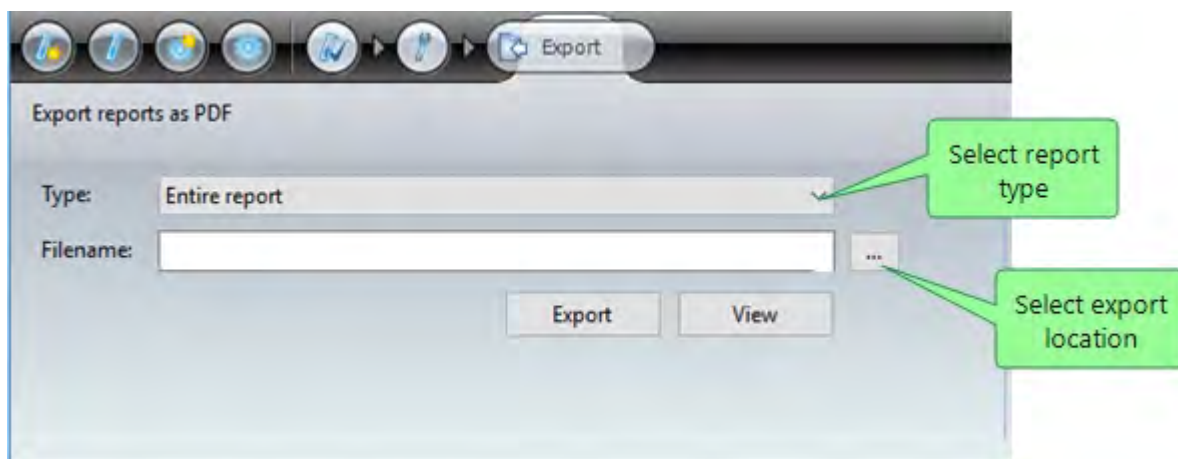
Exporting Reports and Data

The Export tab within QC Reporter is split into two sections. You can either export reports in a PDF format or export the data used in either ASCII or mzML format.

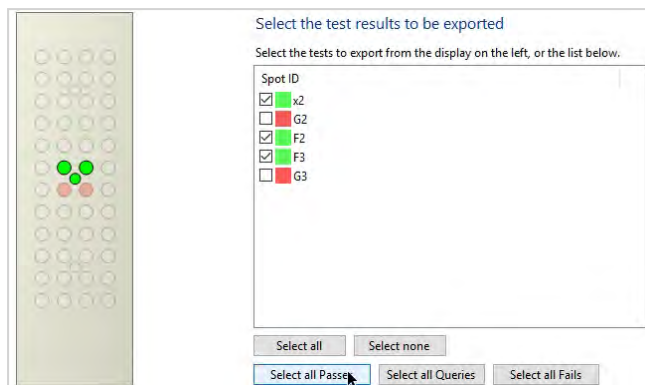


Exporting Reports

1. In the *QC Reporter* main application window, select the **Export** tab.



2. In the **Type** drop-down list, select the type of export from the following options.
 - **Entire report** - all results are combined into a single file;
 - **Summary report** - all results are summarised in a single file;
 - **Specific test reports into a single file;**
 - **Specific test reports each into a separate file.**
3. If you selected a single file export type, click the browse button (...) and then specify a file and path for the report file.
4. If you selected the separate files export type, click the browse button (...) and then specify a folder name for the report files. Each file name corresponds to the sample ID. For example, A1.
5. If you selected either of the specific report types, click the **Select results** button and then use the selection dialog to specify the sample spots you wish to generate reports for.

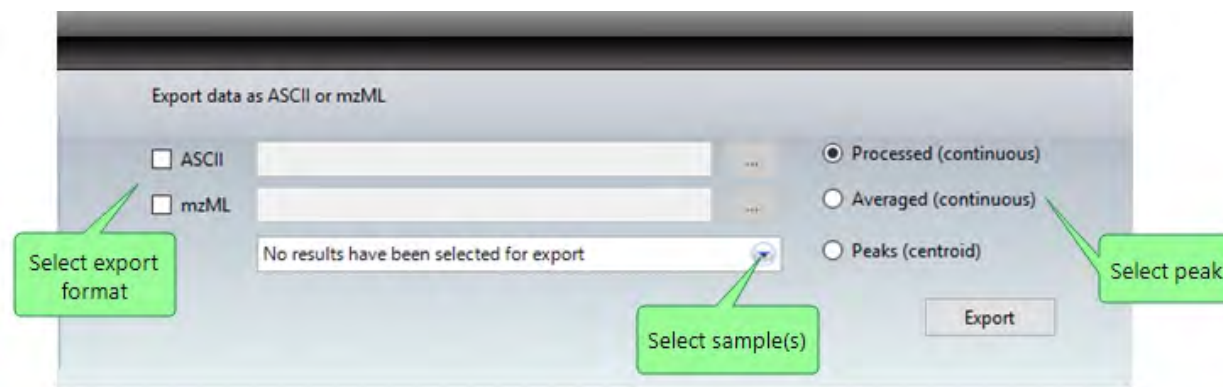


Note: Click outside this dialog to return to the **Export** tab.

- Click **Export**, and then wait from the displayed progress bar to complete.

Exporting Data

- In the QC Reporter main application window, click the Export tab.



- Select either **ASCII** or **mzML** format, or both and click the browse button (...) to specify a file and path for the report file.
- Choose the export peak type by selecting the corresponding radio button.
- Click the **Select results** button and then from the selection dialog specify the sample spots to be exported.
- Click **Export** and then wait for the confirmation message dialog.

Chapter 8 - Maintenance

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Introduction

Over the lifetime of the instrument, you may be required to perform certain tasks in order to keep the equipment functioning properly. The procedures in this section describe how to ensure your instrument is performing efficiently over time, and how to address any issues which might arise during normal usage.



Making backups

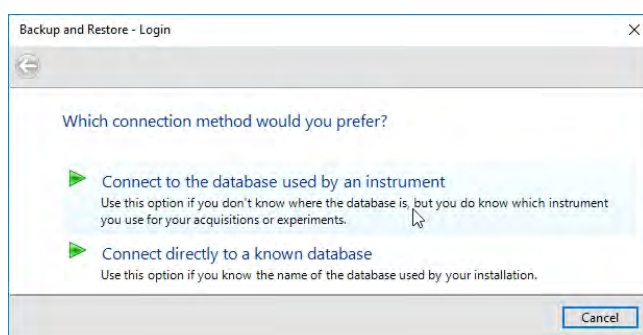
The *MALDI Solutions Data Acquisition* application (and other related programs) use a database for storing all data associated with the operation of the instrument. This database is based on the SQL Server format, and may be backed up or restored independently of the application software. This process should be managed by your regular IT process for system backups, and is outside the scope of this guide. There are however some general points to note regarding external database administration.

- Third-party tools such as Microsoft SQL Server Management Studio should not be used to perform any operations on the instrument database. Use *MALDI Solutions Administration Console* (see [Administration](#)) to change user permissions, data attributes or database entries.
- During installation, the maximum memory that SQL server is allowed to utilise is set to half the total system memory.
- The installation media contains a sample backup script (**Database Scripts => Backup.bat**). This can be used to provide a starting point for the design of any external backup procedure. The batch file (by default) makes scheduled copies of the main database and log files to a separate location on the local PC workstation which can then be made subject to any network backup procedures implemented at your site. It is recommended that any backup regime incorporates an element of off-site storage in order to minimise the risk of losing data.
- The **Administrator** user created during installation is assigned the **Kratos_Basic** and **Kratos_Advanced** user roles. These roles allow the user to perform any operation within the MALDI Solutions product suite. The user also holds the **securityadmin**, **db_owner** and **processadmin** roles.
- When new users are created in *MALDI Solutions Administration Console*, they are given a basic level of access through the **Kratos_Basic** role. If a user account is subsequently elevated to an administrator level, the user will also be assigned the additional roles described above. Newly created users will have the **CHECK_POLICY** property disabled. This means that the passwords entered through the Administration Console will not have to conform to any corporate password policy. Additionally, password ageing will be disabled, as this is not supported by any of the MALDI Solutions products.

In addition to the system-level backups described above, users can backup and restore selected data items with the *MALDI Solutions Backup and Restore* application as described below.

Starting the application

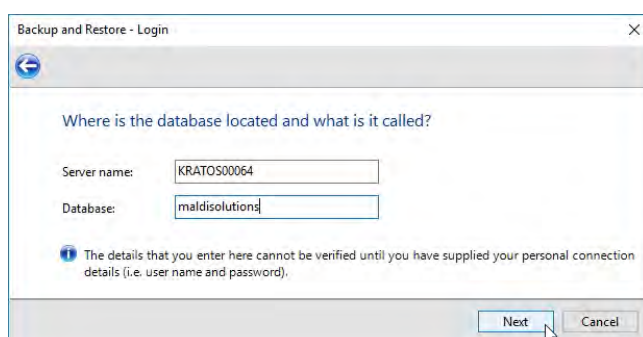
1. From the **Start** menu, locate and select **Shimadzu Corporation > Backup and Restore**.



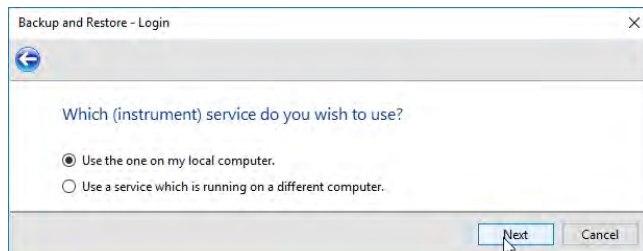
2. If you wish to use the default instrument database, click **Connect to the database used by an instrument** and then proceed to step 6.
3. If you are wish to connect to a remote database (i.e. on another instrument) click **Connect directly to a known database**.
4. Enter the **Server name** and **Database** name, and then click **Next**.

Note:

- The **Server name** should be the network name of the PC workstation on the remote instrument.
- The **Database** is always **maldisolutions** unless you have a specific requirement to use a non-standard database.

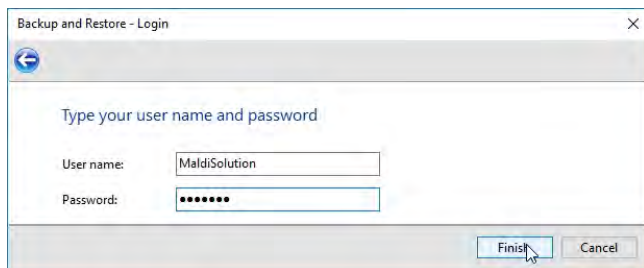


5. Continue with the login procedure from step 7.
6. Select the instrument service you want to use and then click **Next**.

**Note:**

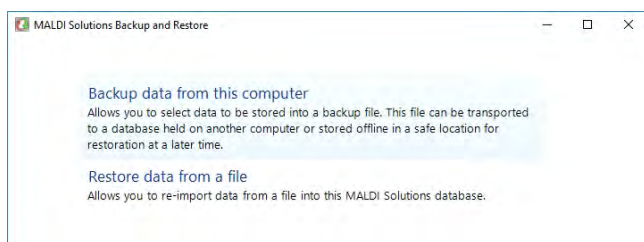
- If you are using the PC workstation connected to the instrument, select **Use the one on my local computer**.
- If you are logging on from a remote computer, select the **Use a service which is running on a different computer** and then specify the network name of the PC workstation connected to the instrument.
- If you chose to use a different database in step 1, the instrument service running on the computer which hosts that database will be used automatically and you will not see this dialog.

7. Enter your **User name** and **Password**, and then click **Finish**.



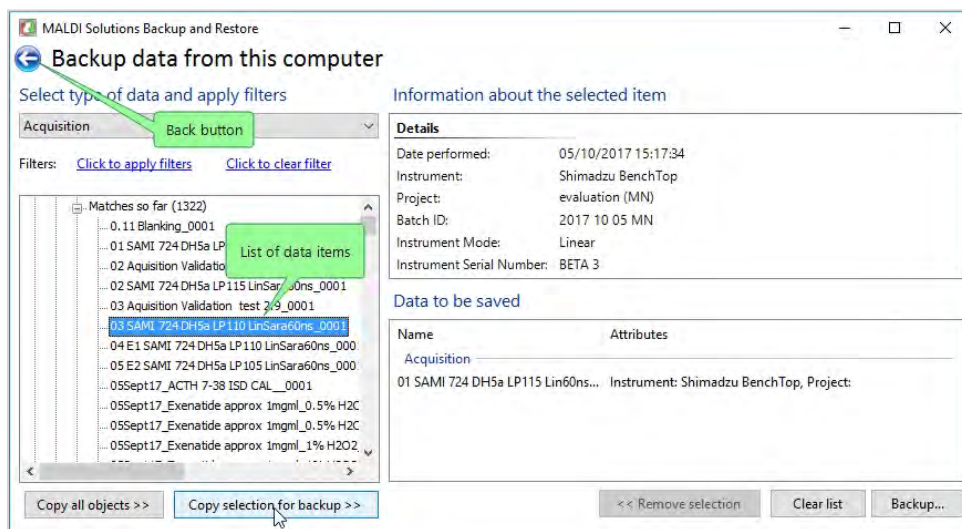
You will see the main menu of the application as shown below.

Note: If you chose to use a different database in step 1, the application will first attempt to validate the database details you entered in step 4, and will terminate if it is unable to connect using the details you entered.



Making a backup

1. In *MALDI Solutions Backup and Restore*, click **Backup data from this computer**.



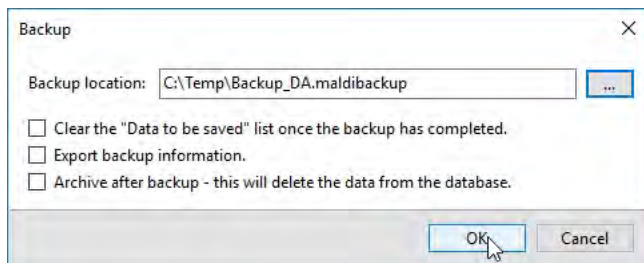
2. In the list of data items, select one or more items in the list and then click **Copy selection for backup >>**.

The selected items will be added to the **Data to be saved** pane.

Notes:

- Use the **Click to apply filters** link to reduce the number of data items shown in the list.
- You can select multiple data items in the list by using the Shift and Ctrl keys when clicking.
- To select all data items in the list, click **Copy all objects >>**.

- To remove an item from the **Data to be saved** pane, select the item and then click << **Remove selection**.
 - To remove all items from the **Data to be saved** pane, click **Clear list**.
3. Once you have selected all the data items you wish to backup, click **Backup ...**



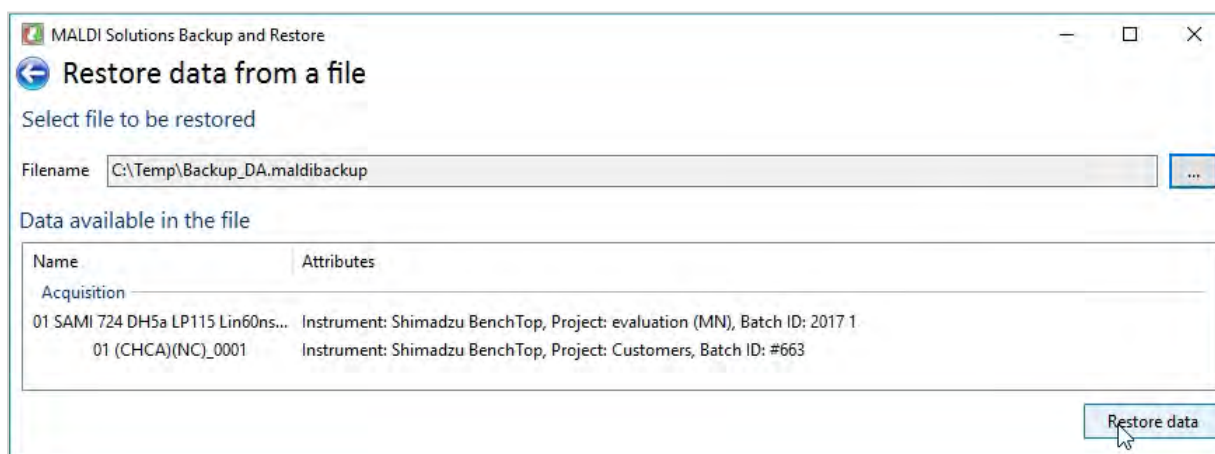
4. In the **Backup location** box, click ... to select a suitable file name and folder location for the backup file, and then click **OK** to begin the backup.

Notes:

- You can optionally clear the **Data to be saved** list automatically after the backup is complete by selecting this option in the dialog.
 - Select **Archive after backup** to automatically delete the data you are backing up from the live instrument database.
 - Select **Export backup information** to create a CSV format file containing information about the backup (useful for audit purposes).
5. Wait until the progress bar indicates that the backup is complete, and then click **OK**.

Restore data

1. In *MALDI Solutions Backup and Restore*, click **Restore data from a file**.

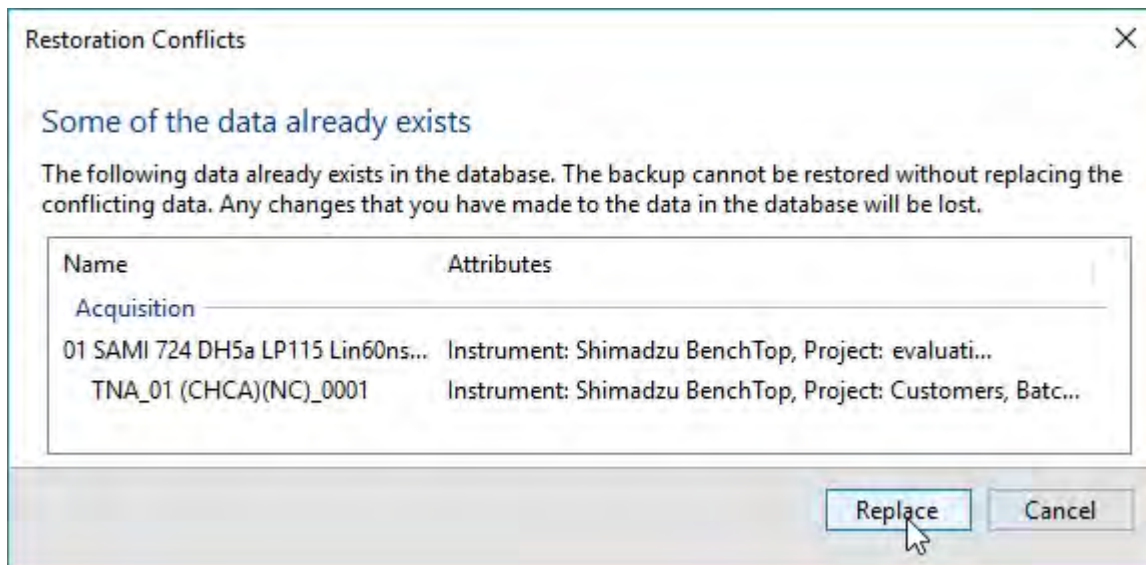


2. In the **Filename** box, type the full path and file name of the data file you wish to restore, or use the ... button to browse to the file's location.

A summary of the data contained in the selected file will be displayed in the **Data available in the file** pane.

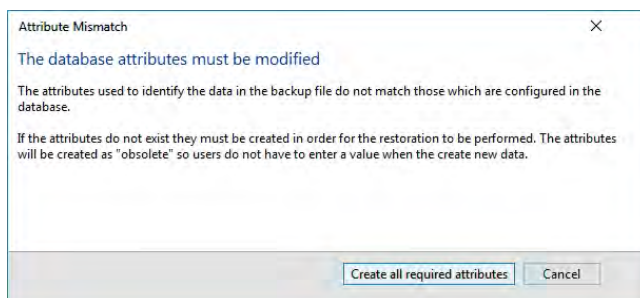
3. Click **Restore data**.

- If the data you are attempting to restore already exists in the database, the **Restoration Conflicts** dialog is displayed.



Click **Replace** to overwrite the data in the database with the file contents, or **Cancel** to abort the restore operation.

- If the data you are attempting to restore has different attributes to that in the database, the **Attribute Mismatch** dialog is displayed.



Click **Create all required attributes** to create the required attributes in the database, or **Cancel** to abort the restore operation.

- Wait until the progress bar indicates that the restore operation is complete, and then click **OK**.

Troubleshooting

This section lists the most common issues you may encounter and how to solve them. If you are unable to resolve an issue with your instrument, or no corrective action is listed below, please refer the problem to your local Shimadzu service centre or distributor (see [Servicing](#)).

Instrument status

If you suspect that the instrument is faulty, you should open the *MALDI Solutions Current Instrument Status* application (see [Instrument status](#)) to determine the current operating condition of the instrument. This will often give you a good indication of where the fault may have occurred.

Instrument problems

Problem	Cause / corrective action
The front panel LEDs do not light when the power is turned ON.	Check if the power cable is correctly connected and if 100 to 240 V power is being supplied to the instrument. See Instrument layout & connections .
PC cannot communicate with the instrument.	Check that the USB 3.0 cable from the instrument is connected to a USB 3.0 port on the PC. See Instrument layout & connections .
In <i>MALDI Solutions Data Acquisition</i> , the Operate button is not active.	<p>Open the <i>MALDI Solutions Current Instrument Status</i> application (see Instrument status) and monitor the Analyser pressure.</p> <ul style="list-style-type: none"> • If pressure is greater than 5.0e-6 mbar, the instrument will not switch to Operate mode and the high voltage supply will not switch on. • If pressure is less than 5.0e-6 mbar, high voltage supplies may be faulty.
In <i>MALDI Solutions Data Acquisition</i> , the Fire button is not active.	<p>Open the <i>MALDI Solutions Current Instrument Status</i> application (see Instrument status) and monitor the System status.</p> <ul style="list-style-type: none"> • If the System status is Standby, the laser will not fire. Click the Operate button to place the instrument in Ready/Operate mode. • If the System status is Operate, the laser may require servicing.

Problem	Cause / corrective action
After loading a target plate, instrument takes more than 5 minutes before the Operate button is active.	If your samples are wet, the instrument will take longer to achieve operating vacuum. Ensure samples are dry before loading the target plate, and use powder-free chemically resistant gloves when handling target plates.
	Check that: <ul style="list-style-type: none"> • The sample door is fully closed. • There is no dust on the O-ring door seal. If there is dust, wipe with a clean lint-free cloth. • The O-ring door seal is not damaged.
	If the instrument has been switched off for a long period of time, operating pressure within the vacuum system may take several hours to achieve.
Source turbo does not reach full speed (1500 Hz).	This may indicate a fault with one or more of the following components: <ul style="list-style-type: none"> • Diaphragm pump; • Turbo pump; • Inlet filter. There may also be a leak in the vacuum system.
The front door cannot be closed.	Ensure the target plate is properly inserted into the front slot (see Loading & unloading target plates).
	The O-ring door seal has come loose.

Data acquisition problems

Problem	Cause / corrective action
The ion signal is unstable or low.	The laser power is too low for the samples. Increase the laser power to get a good signal.
	The laser power is degraded. Increase the laser power to get the same ion signal as before.
	The sample quality is poor. Use freshly prepared samples.
	The Source plate is contaminated. See Cleaning for instructions on how to clean the source plate.
	The detector is degraded.
	High-voltage supply is degraded.

Problem	Cause / corrective action
The base line is too high.	Detector signal cable is damaged.
	The detector is degraded.
	High-voltage supply is degraded.

QC testing

In order to keep your instrument performing optimally, and to detect any deterioration in performance, you should perform the following QC tests on a monthly basis and record the results.

Notes:

- Only adequately trained persons should attempt these procedures.
- The samples used in the tests are supplied in the starter kit supplied with a new instrument (see [Consumables](#)).
- The table below shows the suggested format for recording the results of these tests.

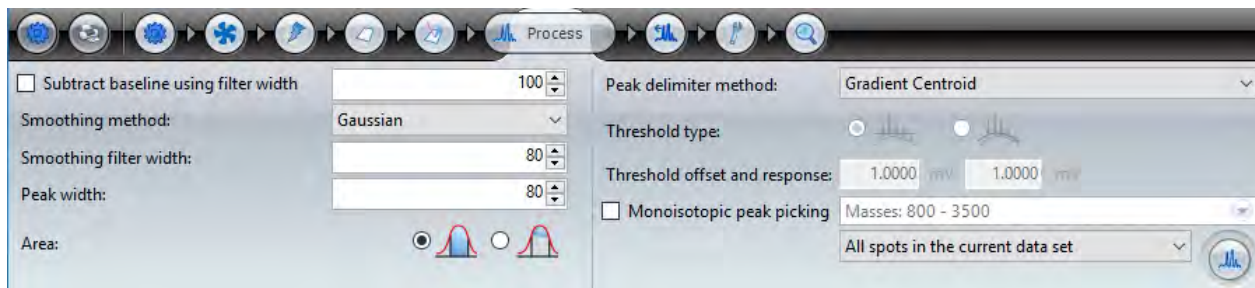
Test	Specification	Period 1	Period 2	Period 3	Period 4
High mass	Sum amplitude >500 mV	PASS			
	Resolution >50	PASS			
	Signal-to-noise >5:1	PASS			
Mass resolution	Sum amplitude >500 mV	PASS			
	Resolution >4,000	PASS			
	Signal-to-noise >5:1	PASS			

High mass test

1. Prepare a fresh target plate spotted with the following samples:
 - Test sample: 500 fmol/ μ L BSA in 70:30 0.1% TFA/acetonitrile;
 - Matrix: sinapinic acid, 20 mg/mL in 50:50 acetonitrile/0.1% TFA;
 - Spotting: 0.5 μ L sample/0.5 μ L matrix.
2. Load the target plate with the sample into the instrument (see [Loading & unloading target plates](#)).
3. In *MALDI Solutions Data Acquisition*, select the **Acquire** tab and then set the following parameters.



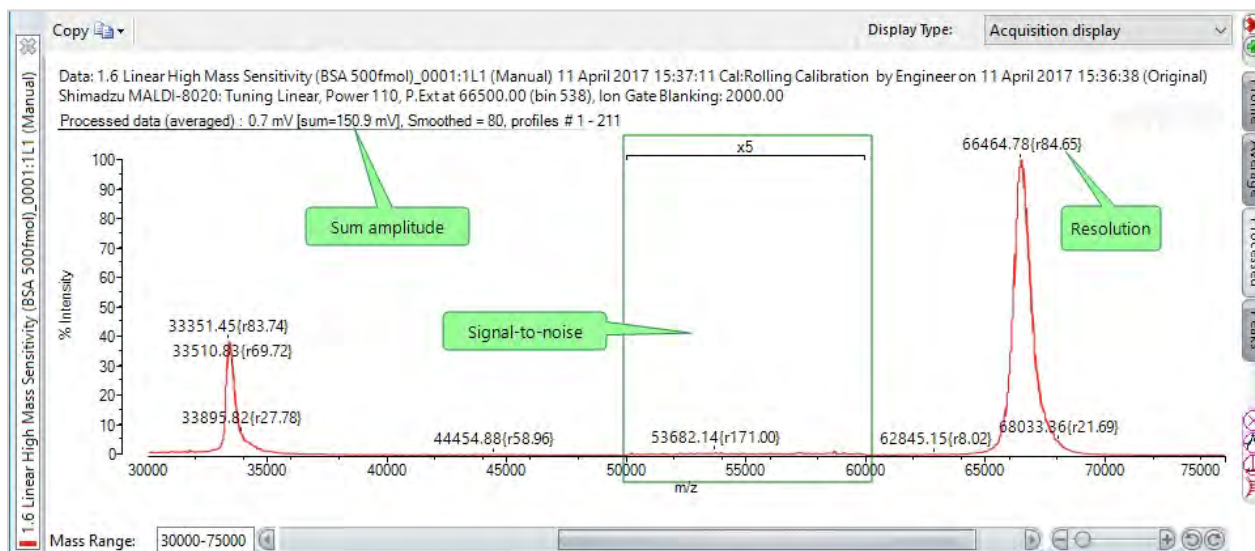
4. Select the **Process** tab and then set the following parameters.



5. Return to the **Acquire** tab and set the **Blanking** parameter to **Off**.
6. Position the camera crosshairs on the correct sample well (see [Using the camera viewer](#)).
7. Click **Fire**, and observe the acquired spectrum.
8. Adjust the laser power so that a clear peak is visible at 225.08 Da, and then click **Stop**.
9. Increase the laser power setting by 30 to 50, and set the **Blanking** parameter to 2000 Da.
10. Click **Fire**, and observe the acquired spectrum.

Note: In order for the test to pass, the following results should be obtained.

- Sum amplitude > 500 mV;
- Resolution > 50;
- Signal-to-noise ratio > 5:1 (50,000 to 60,000 Da).

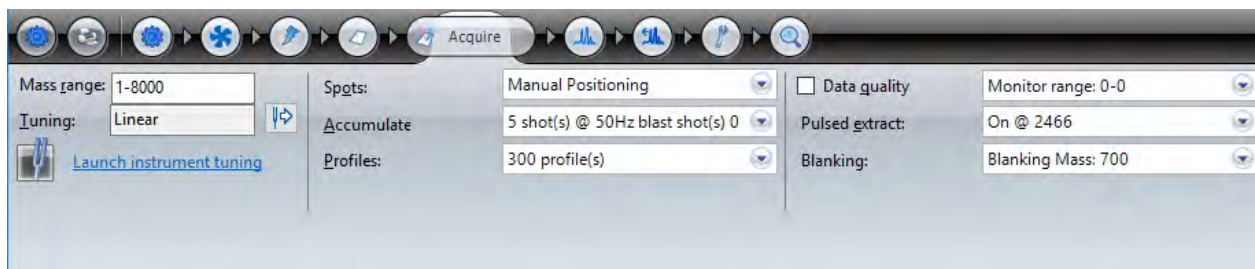


11. Record the results (optionally using the suggested format shown above).

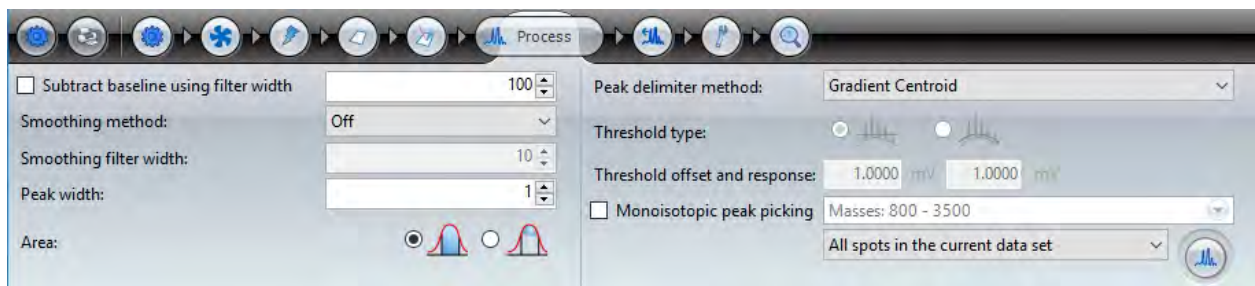
Mass resolution test

1. Prepare a fresh target plate with the following sample:
 - Test sample: 1 pmol/ μ L ACTH 18-39 in 70:30 0.1% TFA/acetonitrile;
 - Matrix: alpha-cyano-hydroxy-4-cinnamic acid, 10 mg/mL in 50:50 acetonitrile/0.1% TFA;
 - Spotting: 0.5 μ L sample/0.5 μ L matrix.

2. Load the target plate with the sample into the instrument (see [Loading & unloading target plates](#)).
3. In *MALDI Solutions Data Acquisition*, select the **Acquire** tab and then set the following parameters.



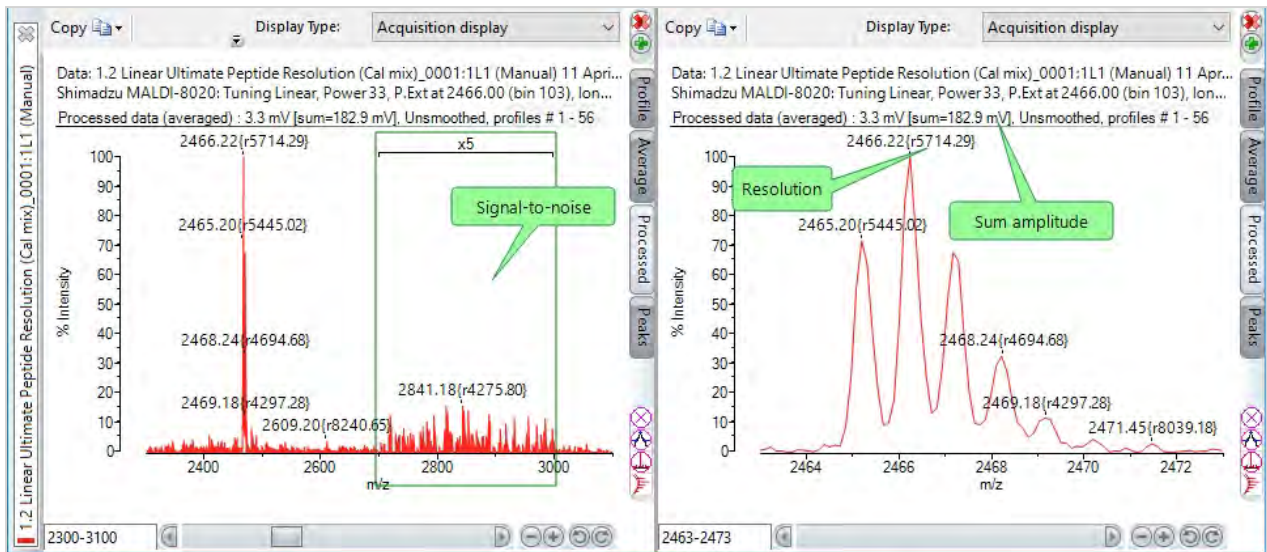
4. Select the **Process** tab and then set the following parameters.



5. Return to the **Acquire** tab and set the **Blanking** parameter to **Off**.
6. Position the camera crosshairs on the correct sample well (see [Using the camera viewer](#)).
7. Click **Fire**, and observe the acquired spectrum.
8. Adjust the laser power so that a clear peak is visible at 190.05 Da.
9. Click **Stop**.
10. Increase the laser power setting by 30 to 50, and set the **Blanking** parameter to 700 Da.
11. Click **Fire**, and observe the acquired spectrum.

In order for the test to pass, the following results should be obtained.

- Sum amplitude > 100 mV;
- Resolution > 4000;
- Signal-to-noise ratio > 5:1 (2700 to 3000 Da).



- Record the results (optionally using the suggested format shown above).

Servicing

Due to hazardous voltages and a Class 3B laser, only trained service engineers are permitted to carry out preventative maintenance and servicing of your instrument.

Preventative maintenance

Preventative maintenance servicing ensures that the instrument continues to provide trouble-free operation. We recommend that the instrument is serviced at least once a year. After the one-year warranty period, service contracts are available to provide the appropriate level of support. Contact your local service centre for details.

Fault reporting

Any faults with your instrument should be reported to your local Shimadzu service centre or distributor. When reporting faults, please try to include as much information as you can about the fault and the conditions under which it was encountered. As a minimum, please provide the following details:

- The instrument serial number (printed on the rating plate fitted to the rear of the instrument);
- Details of any error messages;
- A description of the symptoms of the fault.

Cleaning

The following procedures may be used to perform basic cleaning of the instrument. Periodic cleaning ensures that your instrument continues to perform optimally during its lifetime and produces the best possible results during operation.

Cleaning the instrument

When cleaning the instrument covers or PC workstation monitor, please note the following:

- DO NOT use cleaners containing alcohol (such as screen wipes), which corrode plastic.
- DO NOT use acid or alkaline solvents, or abrasive creams.

Covers

Use a soft damp, or antistatic, cloth to clean the covers of the instrument. Alternatively, you can use diluted washing-up liquid.

Monitor

Periodically clean the screen with a soft, dry cloth; DO NOT use tissue paper, etc. as these will damage the screen.

The frame of the monitor can be cleaned with a cloth lightly moistened with diluted washing-up liquid followed by a wiping with a soft, dry cloth.

Cleaning the cooling fans

Use a vacuum cleaner or soft dust brush to remove accumulated dust from the exterior of the two inlet fans on the rear of the instrument.



Source cleaning

Over time, contamination from your samples and matrix can build up on the source plate forming an insulating layer that will adversely affect the operation of the instrument. The MALDI-8020 and MALDI-8030 instruments includes a source cleaning feature which uses the built-in laser to remove contamination from the source plate.

Note: Only users with the appropriate permissions can access this feature (see [Managing users](#)).

1. In *MALDI Solutions Data Acquisition*, select the **Target** tab and then click **Clean Source**.
2. In the confirmation dialog, click **OK**.

Notes:

- You can monitor the process in the [camera viewer](#) as the laser fires at the source in a predefined pattern.
 - The laser cleaning process takes approximately 10 minutes to complete.
3. When the completion message dialog appears, click **OK** to finish.

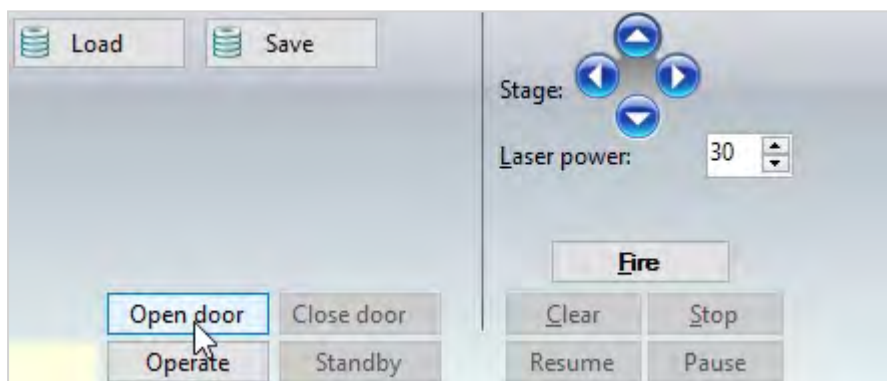
Replacing the door seal

The interior face of the sample door is fitted with an O-ring seal which maintains the integrity of the vacuum inside the sample chamber. If the seal becomes damaged or excessively dirty, the vacuum system will be unable to reach the correct pressure and target plate loading may be compromised.

A replacement door seal is included in the starter kit supplied with your instrument (see [Consumables](#)). If you suspect that the existing seal is damaged or contaminated, replace it using the following procedure.

Note: DO NOT attempt to clean the seal with any solvent as this will change the properties of the seal.

1. In *MALDI Solutions Data Acquisition*, select the **Acquire** tab and then click **Open Door**.



- When the front panel LED blinks **green**, pull back gently from the top of the door to open.



- Wearing powder-free gloves, lift the old seal out of the groove in the sample door and dispose of it safely.



Note: Do not use any metal tools near the sample door interior as scratches may occur.

- Clean the groove using a lint-free cloth moistened with HPLC-grade acetone.
- Fit the new seal into the groove, ensuring it is pressed firmly into position all around the sample door interior.
- Test the operation of the sample door by loading a target plate (see [Loading & unloading target plates](#)).

Consumables

The following parts are available as consumable items for the instrument. To order additional supplies of these items, please contact your local Shimadzu service centre or distributor.

Description	Part number
Fleximass-DS polymeric target slides (Pack of 16)	TO-430R00
Fleximass-SR48 (Pack of 4)*	TO-431R00
O-ring (ID61 CS3.5)*	224-00127

Note: Items marked with an asterisk (*) above are included in the starter kit supplied with a new instrument.



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