ZETASIZER NANOSAMPLER BASIC GUIDE

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INTRODUCTION

This section covers the following information:

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About this manual

This manual covers the essential information necessary for ensuring safe and efficient operation of the NanoSampler system.

This manual covers the following instrument:

Instrument	Part number
NanoSampler	ZEN2000

The aims of this manual are to:

- Provide essential health and safety information, which all users must read, in addition to the requirements for the Zetasizer Nano instrument.
- Give information on the site requirements of the system.
- Describe the NanoSampler hardware components.
- Give a brief description of the measurement process.
- Set out various maintenance and troubleshooting procedures.
- Provide the specifications of the instrument.

Product documentation structure

This manual should be used in conjunction with the documentation for the Zetasizer Nano instrument. As a minimum all users of the NanoSampler must read the following documentation:

• Zetasizer Nano Basic Guide - the essentials required to get started, including health and safety. All users must read this manual before using the system.

How to use this manual

We recommend reading this manual fully before starting the first measurement.



Warning! The instrument and the samples to be measured may be hazardous if misused. Users must read the Health and Safety information before operating the system.

Help system

The Help system is Integrated with the Zetasizer software, and provides searchable, context-sensitive information covering all software features.

Access to the instrument

This manual refers to the various people who will have access to the instrument, as follows.

Malvern Instruments personnel

Malvern Instruments personnel (service engineers, representatives, etc.) have full access to the instrument and are the only people authorized to perform all service procedures that may require the removal of the covers.



Warning! Removal of the covers by unauthorized personnel will invalidate the warranty of the instrument. Unless advised within the content of this manual, only Malvern Instruments trained personnel are permitted to remove the main cover of any part.

Supervisor

The supervisor is responsible for the management and safety of the instrument and its operation, and for the training of operators. The supervisor can perform all maintenance routines identified in the Basic Guide.

Operator

An operator is a person trained in the use of the system. The operator can perform all user maintenance routines identified in the Basic Guide.



Warning! Under no circumstances should the supervisor or an operator remove the main cover of the instrument. Failure to follow these guidelines could result in exposure to hazardous voltages.

Assumed information

To make full use of this manual, the user should understand the following points.

Naming convention

The NanoSampler is referred to in full. The combination of the NanoSampler, instrument and computer is referred to as 'the system'.

Menu commands

Menu commands are always shown in bold text and shown in the form:

File-New-Measurement

This refers to selecting the **File** menu, then the **New** menu sub-item and finally the **Meas-urement** item.

Where to get help

This section gives information on the various channels in place to get help with your NanoSampler system.

Help desk

All queries about the system should be directed to your local Malvern Instruments representative, quoting the following information:

- **Model and serial number** of the instrument (usually located on the outside casing of the instrument).
- Software version (see Help-About in the software).
- Firmware version (the Helpdesk will inform you how to locate this information).

Contact the International Helpdesk if the local Malvern Instruments representative is not available:

Telephone: +44 (0) 1684 891800 or email: helpdesk@malvern.com.



Note: this help line is primarily English speaking.

Remote support

Malvern Instruments offers a remote support service, delivered by an Internet connection. Benefits include fast and efficient fault diagnosis, reducing downtime and costs.

Malvern website - www.malvern.com

The Malvern Instruments website offers a comprehensive range of particle characterization resources for use by customers 24 hours a day, seven days a week.

HEALTH & SAFETY

This section provides detailed health and safety information applicable to the NanoSampler instrument. All users of the system must read this information.

This section provides the following information:

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Electrical warnings and regulations	10
Power cords and power safety	10
Sample handling warnings	13
Storage and disposal	14
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General warnings and regulations



Warning! The instrument or the samples to be measured may be hazardous if misused. Read and fully understand this section before operating the system.

The instrument must only be stored or operated in environmental conditions conforming to the specification in the Site requirements section.



Warning! Use of the system in a manner not specified by Malvern Instruments Ltd. may impair the protection provided by the system.

Electrical warnings and regulations



Warning! The NanoSampler System contains high voltage components. Unless advised within the content of this manual, only Malvern Instruments trained personnel are permitted to remove the main covers from the instrument or connected devices.



Warning! This product must be connected to a protective earth. The metal parts of the optical unit are earthed using a protective earth connection.

If the instrument gets wet (for example, if sample or dispersant is spilt), switch it off and disconnect it from the mains power supply immediately. Carefully clean and dry the instrument before re-applying power.

The NanoSampler system components are mains powered devices and all power cables and electrical sockets should be treated accordingly. Do not place cables in positions where they are likely to become wet.

Power cords and power safety

The notes in this section indicate best practice and should be adhered to when connecting the Malvern instrument to the power supply.

Power cord set requirements

Power cord sets must meet the requirements of the country where the product is used. For further information on power cord set requirements, contact a Malvern Instruments representative.

General requirements

The requirements listed below are applicable to all countries:

- The power cord must be approved by an acceptable accredited agency responsible for evaluation in the country where the power cord set will be installed.
- The power cord set must have a minimum current capacity of 10A (7A in Japan only) and a nominal voltage rating of 125 or 250 volts AC, as required by each country's power system.
- The area of the wire must be a minimum of 0.75mm² or 18AWG, and the length of the cord must be less than 3m.
- The power cord must be routed so it is not likely to be walked on or pinched by items placed upon it or against it, or become wet. Particular attention must be paid to the plug, the electrical outlet, and the point where the cord exits the product.



Warning! Do not operate this product with a damaged power cord set. If the power cord set is damaged in any manner, replace it immediately.



Warning! Do not use the power cord received with this product on any other products.

Power safety information

The following notes indicate guidelines to follow when connecting the Malvern Instruments power supply using single and multiple extension leads, connection via AC Adapters and use of Uninterruptible Power Supplies (UPS).



Warning! To prevent electric shock, plug the instrument or accessory into correctly earthed electrical outlets. Never use the system without a properly connected protective earth conductor.

The power cord supplied is equipped with a grounding connection to ensure grounding integrity is maintained.

Advice on use of extension leads

Follow this advice when using single or multiple socket extension leads. These are also called trailing sockets.

- Ensure the lead is connected to a wall power outlet and not to another extension lead. The extension lead must be designed for grounding plugs and plugged into a grounded wall outlet.
- Ensure that the total ampere rating of the products being plugged into the extension lead does not exceed the ampere rating of the extension lead.
- Use caution when plugging a power cord into a multiple socket extension lead. Some extension leads may allow a plug to be inserted incorrectly.
- Incorrect insertion of the power plug could result in permanent damage to the instrument or accessory, as well as risk of electric shock and/or fire. Ensure that the ground connection (prong/pin) of the power cord plug is inserted into the mating ground contact of the extension lead

Advice on use of AC adapters



Warning! Do not use adapter plugs that bypass the grounding feature, or remove the grounding feature from the plug or adapter.

- Place the AC adapter in a ventilated area, such as a desk top or on the floor.
- The AC adapter may become hot during normal operation of the instrument or accessory. Use care when handling the adapter during or immediately after operation.
- Use only the Malvern-provided AC adapter approved for use with the instrument and/or accessory. Use of another AC adapter may cause a fire or explosion.

Advice on use of Uninterruptible Power Supplies (UPS)

To help protect the instrument and/or accessory from sudden, transient increases and decreases in electrical power, use a surge suppressor, line conditioner or UPS.

Sample handling warnings

- Always handle all substances in accordance with the COSHH (Control Of Substances Hazardous to Health) regulations (UK) or any local regulations concerning sample handling safety.
- Before using any substance, check the Material Safety Data Sheets for safe handling information.
- Use the instrument in a well ventilated room, or preferably within a fume cupboard, if the fumes from the sample or dispersant are toxic or noxious.
- Wear personal protective equipment as recommended by the Material Safety Data Sheets if toxic or hazardous samples are being handled, particularly during sample preparation and measurement.
- Wear protective gloves when handling hazardous materials, or those that cause skin infections or irritations.
- Do not smoke during measurement procedures, particularly where inflammable samples are used or stored.
- Do not eat or drink during measurement procedures, particularly where hazardous samples are used or stored.
- Take care when handling glass (e.g. microscope slides and beakers). Hazardous materials may enter a wound caused by broken glass.
- Always test a new sample or dispersant for chemical compatibility before use.
- After measuring hazardous samples, scrupulously clean the system to remove any contaminants before making another measurement.
- Always label samples for analysis using industry standard labelling, particularly if they are handled by a number of staff or stored for long periods. Clearly mark any operator hazard and associated safety precautions that are required for the handling of dangerous materials.
- Keep a record of all hazardous substances used in the system for protection of service and maintenance personnel.
- Always adopt responsible procedures for the disposal of waste samples. Most local laws forbid the disposal of many chemicals in such a manner as to allow their entry into the water system. The user is advised to seek local advice as to the means available for disposal of chemical wastes in the area of use. Refer to the Materials Safety Data Sheets.
- The surfaces of the system may be permanently damaged if samples are spilt on them. If a spillage does occur, disconnect the system from the power supply before scrupulously cleaning up the spillage.

Storage and disposal

Storage/shipment

If the NanoSampler needs to be stored for a long time, or if it must be shipped to a different location, proceed as follows:

- It is recommended that you thoroughly flush the system using a 5% methanol DI water solution. When the system has been reinstalled, in order to sterilize it, a 60% methanol DI water solution can be used, which should then be followed with a full flush using your standard transport fluid.
- 2. Disconnect and remove all tubing, except for the sample loop.
- 3. Switch off the NanoSampler.
- 4. In case of biohazard materials: remove and throw away the needle and other flow path materials, in accordance with an approved waste disposal program.
- 5. Use the original packaging materials to package the NanoSampler; also place the foam block in the tray location.
- 6. Dispatch the package; make sure any tax/import/export requirements are met.
- 7. Contact your local distributor if you need more information.

Disposal of Electrical & Electronic Equipment

When the need arises to dispose of the system, this should be done in a responsible manner. Follow these guidelines:

- Refer to local regulations on disposal of equipment; in Europe refer to the information below.
- Seek advice from the local Malvern Instruments representative for details.
- Decontaminate the instrument if hazardous materials have been used in it.

The following is applicable in the European Union and other European countries with separate collection systems.

This symbol on the product or on its packaging indicates that when the last user wishes to discard this product it must not be treated as general waste. Instead it shall be handed over to the appropriate facility for the recovery and recycling of electrical and electronic equipment.

By not discarding this product along with other household-type waste, the volume of waste sent to incinerators or landfills will be reduced and natural resources will be conserved.

For more detailed information about recycling of this product, please contact your local city office, your waste disposal service, or your Malvern Instruments representative.

Additional safety considerations

Adhere to the following guidelines when using the NanoSampler:

Operational safety

The NanoSampler houses a sharp needle that is used to pierce vial caps. This needle may cause cutting injuries if operating instructions are not followed.

Make sure that covers and doors are closed before starting the NanoSampler. If the doors are opened during a measurement the needle movement is slowed down automatically as a safety feature whilst the measurement continues.

Make sure that the power has been switched off while servicing any parts in the direct proximity of the pierce needle.

System operation and maintenance

To maintain the specified performance of the system, maintenance must be carried out as indicated in this manual.

Malvern Instruments offers service contracts and preventive maintenance services. Please contact your local dealer or the nearest sales office for more information.

Solvents

The solvents used may be flammable and toxic. The room in which the system is installed should be well ventilated to prevent solvent vapors causing a fire.

Use of open fire in the vicinity of this system must be strictly prohibited. Do not install the system in the same room with any other equipment that emits or could potentially emit sparks.

Sample containers (vials) should be sealed to minimize any risks related to solvent vapor.

Do not allow solvents to accumulate in the system. Perform periodic leak checks on supply lines.

Toxic hazard

When you use or analyze toxic fluids you need to take all possible precautions and treat all specimens as potentially toxic.

Always wear protective goggles and gloves when handling toxic samples.

Biological hazard

When you analyze biological fluids you need to take all possible precautions and treat all specimens as potentially infectious.

Always wear protective goggles and gloves when handling toxic or biologically infectious samples.

Waste disposal

Dispose of waste (samples, solvents, device) in accordance with a regulated waste disposal program.

Follow a regulated, approved waste disposal program and never dispose of solvents through the municipal sewage system.

SITE REQUIREMENTS

Several criteria must be satisfied at the installation site to ensure the effective and safe functioning of the NanoSampler. This section provides detailed information on these requirements.

The following topics are covered:

Environmental conditions	18
System requirements	19
Installation	21

Environmental conditions

Ensure that the following environmental conditions are satisfied. The installation site for the NanoSampler must be:

- Indoors the system is not intended for use outdoors.
- Well ventilated especially if noxious samples or dispersants are used.
- **Temperature stable** away from areas where the ambient air temperature could change, such as air-conditioning, fans or windows.
- Away from strong light sources for example, windows allowing in bright sunlight.
- Away from strong heat sources for example, radiators.

The bench that supports the NanoSampler must be:

- Horizontal
- Vibration free
- Capable of supporting the weight of the system.

Electrical considerations

• Avoid passing electrical cables through areas where liquids can be spilled.



Warning! Do not position the system so that power sockets are obstructed. They may need to be disconnected during an emergency.

System components weight

Component	Weight
NanoSampler	Approx. 19kg
Zetasizer Nano (ZSP)	19kg
Computer and Printer	See manufacturer's documentation

Operating environment

Operating environment	Indoor use only
Operating altitude	up to 2000m
Operating temperature	10-40°C
Operating humidity	20-80% RH

Transport and storage requirements

Store the unit in a dry place where no mold will form.

Transport and storage temperature	-25-60°C
Transport and storage humidity max.	85% RH
Transport and storage sound pressure level	LAeq <70dB

System requirements

The NanoSampler has the following space, power, airline and computer requirements.

Space required

Provide enough space to allow easy access to all components of the system. Allow 2500mm (width) x 650mm (depth) of bench space. This is to allow space for the instrument, and the computer including monitor. Allow at least 800mm above the bench surface for access to the cell area.

Dimensions of the main components of the system are given below.

Component	Width	Depth	Height
NanoSampler	300mm	510mm	360mm
Computer and printer	See manufacturer's documentation		

Power requirements



Warning! Only use the Power Supply Unit (PSU) supplied with the instrument. Failure to do so could impair operator safety. The mains power supply must be clean and filtered. If necessary, fit an Uninterruptible Power Supply (UPS) to remove any spikes or noise. The power requirement for each component of the NanoSampler system is given below.

Component	Power requirement	Power sockets required
NanoSampler	Mains supply:95 - 240 Vac +/-10%, 50-60Hz	1
External PSU	External PSU:100-240Vac 47/63Hz 3.2A, O/P: 40.8Vdc 3.18A	1
Computer	See manufacturer's doc- umentation	1
Computer Monitor	See manufacturer's doc- umentation	1

Computer specification and usage

Note: The NanoSampler requires the installation of Zetasizer 7.10 software or later.

Contact the Malvern Helpdesk or website for the recommended computer specification; otherwise consult the **Software Update Notification** document supplied on the software CD.

The NanoSampler data can be stored on a network but this is not recommended. Also, running other software at the same time may reduce its speed.

Installation

The NanoSampler will initially have been commissioned by Malvern trained personnel. This section indicates actions to undertake should you need to:

- Move the instrument
- Upgrade the computer

Moving the instrument

If it is necessary to move the system, please contact Malvern Instruments for guidance.



Warning! Removal of the covers by unauthorized personnel will invalidate the warranty of the instrument.



Note: If the original setup was performed as part of an IQ/OQ, moving the system invalidates this.



Note: Only install the system in a suitable environment as detailed in Environmental conditions in the Site Requirements.

If you need to lift the system, follow this procedure:

1. Lift the NanoSampler using both hands at the marked positions which are at the top of the instrument:



2. With both hands under the instrument, lift to its operating location. Keep the instrument upright.

Changing the computer

If the computer to be used with the instrument has to be changed, contact Malvern Instruments.

Upgrading the software

If an upgrade CD is provided, follow this procedure.



Note: If the original software is subsequently updated with a new version, any custom reports, parameter settings, SOPs, etc. will be preserved. However, back up this data first.

- 1. Remove the old version of the software using the Add/Remove Programs function in the Windows Control Panel.
- 2. Insert the new Zetasizer software CD into the CD drive.
- 3. If Autorun is enabled, the software will install automatically. (If it is not enabled, select **Start-Run-Setup.exe**). Then follow the on-screen instructions.

NANOSAMPLER HARDWARE

This section provides information on the typical hardware setup of a NanoSampler and gives more detail on the connections available.

The following topics are covered:

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System configuration

The NanoSampler is a versatile, compact sample delivery system that ensures highly precise and reproducible automated loading of samples into your Zetasizer Nano. With completely unattended operation the NanoSampler maximizes the productivity of your Zetasizer Nano and is ideally suited for laboratories where reproducibility and multivariate studies are important.

The following illustration shows the NanoSampler in a typical setup.



- 1. Zetasizer Nano
- 2. NanoSampler
- 3. Computer running Zetasizer software
- 4. Sample vials
- 5. Sample vial holders (not shown)
- 6. Solvent transport fluid bottle (not shown)

Front panel (closed/open)



- 1. Front panel handle
- 2. Observation window
- 3. Drainage ports



- 1. Front panel (open position)
- 2. Tubing guide
- 3. Wash liquid bottle (external bottle required for 96 vials)
- 4. Vial adapters holding sample vials

Interior (front)



The NanoSampler's **sampling compartment** houses the following parts:

- 1. Syringe
- 2. Needle arm
- 3. Injection valve
- 4. Valve leak bin
- 5. Sample compartment
- 6. Needle wash position / wash station
- 7. Wash liquid bottle

Back panel



The back panel of the instrument has the following items:

- 1. USB connector
- 2. Warning label
- 3. On/off switch
- 4. Fuse
- 5. Power connector

Plates and vials

The NanoSampler uses 2 x 48 well trays, each well of which holds a single 1.5mL vial.



The system can be loaded with either one or two trays, each of which can contain any number of vials (up to 48) as required by your analysis.



Caution: only use vial holder plates supplied by Malvern Instruments. This is due to varying pitches of plates from different manufacturers, which could result in damage to the needle.

Using the hardware

Opening the door

To load samples into the NanoSampler, open the door using the following procedure:

1. Hold the door handle and gently pull upwards until in it is in a horizontal position:



2. Then slide the door into the instrument.





Note: before closing the front panel door withdraw the door fully by pulling it gently towards you.

Removing the cover

For easier access to the sampling area, remove the cover as described below:

1. Press the two black buttons on either side (top) of the NanoSampler simultaneously:



2. Then gently pull the cover towards you.



Loading plates

To load the plates ensure they are oriented as shown in the illustration and carefully insert into the NanoSampler being careful not to agitate the samples:



- 1. Tray 1
- 2. Tray 2
- 3. NanoSampler tray holder

USING THE SYSTEM

This section outlines the NanoSampler workflow and provides details on how to perform each step.

The topics covered in this section include:

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System and sample preparation	.35
NanoSampler feature key	.37
Software configuration	.38
Vial position files	. 47

Workflow overview

The flow-chart below details the process required to make a NanoSampler measurement.



Pre-measurement checklist

You need to make several checks before each run on the NanoSampler - more information on some of these steps is provided later in this section:

- 1. Prepare samples using 1.5mL vials and load into the NanoSampler.
- 2. Check that the Zetasizer Nano is switched on and that the **AutoSampler Features** key is active in the Zetasizer software.



Note: allow the NanoSampler to adopt ambient temperature for at least one hour before making a measurement.

- 3. Create an appropriate SOP and measurement file.
- 4. Ensure the **Quartz Flow Cell is in position and attached**. The flow cell should be inserted firmly into the Zetasizer cuvette holder, and should be clean. One pipe should connect the NanoSampler to the flow cell. The other pipe from the flow cell must be connected to a waste bottle.
- 5. Check both **Waste pipes** at the front of the unit both must discharge into a waste bottle.
- 6. Make sure the **Intake pipe** for transport and cleaning fluid is in the source bottle of the fluid, and also that the fluid is appropriate to your analysis. We recommend that you filter the transport fluid before beginning a NanoSampler measurement.



Note: for a full NanoSampler run of 96 vials with 3 aliquots analyzed for each vial, 2L of transport fluid and one 2L bottle each for the flow cell waste and the waste outlets is required.

System and sample preparation

Connection checks

Before switching on, ensure that the following connection checks have been carried out.

Electrical and communication connections

- 1. Connect the NanoSampler to the USB port with the cable provided.
- 2. Connect the power cable between the NanoSampler and the power outlet. Refer to Power cords and power safety on page 10 for more safety information.
- 3. Switch on the NanoSampler.
- 4. Start the Zetasizer software on your computer.

Fluid connections

- 1. Connect the drain tubing to the waste outlet.
- 2. Fill the wash solvent bottle inside the sampling compartment of the NanoSampler with distilled water and propanol (80/20 v/v%) or mobile phase.



Note: only water or organic solvents should be used. Do not buffer solutions as these may block and damage the system.

Sample preparation and loading

Take the following into account when handling samples:

- Standard vials are best filled by using a narrow-end pipette to allow air to escape when filling the vial.
- Do not fill vials/wells to the top. This prevents sample being drawn into the needle, risking cross-contamination of samples.
- Seals must be airtight to stop bubbles from forming and to prevent evaporation of volatile samples.
- Filtering the eluent with 0.2µm filter considerably reduces the risk of clogging. This also applies to samples; ensure you use the appropriate filter material for sample filtration.

Samples can be prepared under the same conditions (e.g. dispersant, concentration) as for a batch measurement. Ensure the transport/cleaning fluid used is appropriate to the samples. For example, if you are using an aqueous dispersant it is not advisable to use organic solvent as a cleaning fluid.

Filling vials



Note: replacement vials are available to order from Malvern Instruments.

Fill your sample vial, ensuring no air bubbles are present in the sample. A minimum sample volume of 750 μ l is required for one analysis; **1.6 mL is required for three replicate analyses**. Do not overfill the vial and ensure there is some headspace above the sample, as shown in the illustration - the level should be just above the 1.5mL marker line:





Note: Never fill the vial above the neck.

Loading vials

The NanoSampler contains two trays, each capable of holding 48 vials. The trays are marked A-F by row and 1-8 by column. The vials are analyzed in the order set by the playlist. When using the range sampling feature the playlist is populated in the order A1, A2, A3....A8, B1, B2, etc. with tray 1 analyzed before tray 2.

NanoSampler feature key

Before you can use the NanoSampler it must be activated by the installation of a feature key. To do this:

- 1. Select Tools-Options-Feature Keys.
- 2. From the feature key selection drop-down menu, select AutoSampler Features.
- 3. Click **Install licence key**. A message is displayed to confirm that the key has been installed correctly.
- 4. Click OK to close the Zetasizer Options window.
- 5. To confirm that the feature key is correctly installed, click the features button on the status bar.



6. A summary of the installed feature keys is displayed:

Feature	Licence Type	Notes
Research Features	None	
ER/ES Features (21 CFR Part 11)	None	
Microrheology Features	None	
Advanced Protein Features	None	
AutoSampler Features	Full	Licence code: b a
	Clos	e]

Software configuration

This section covers in detail the steps required to make a measurement once the instrument and sample have been prepared.

The NanoSampler is controlled by the SOP Playlist feature. The process involves setting up a new measurement file, appropriate SOP and SOP playlist.

Defining SOPs for the NanoSampler

The SOP creation step is the same as for a standard size measurement on the Zetasizer. See the Zetasizer User Manual for more details. For a NanoSampler measurement, ensure:

- The measurement type selected is Size.
- The cell specified is the Quartz Flow Cell, ZEN 0023.

The process for creating SOPs is covered in detail in the Help system and the **Zetasizer Nano User Manual**.

Creating the SOP Playlist

Through the SOP Player you can use the NanoSampler to :

- Run an SOP on an individual well.
- Run an SOP on a range of wells.

To start the SOP player, choose:

• Measure-SOP Player

From here you can build up a playlist of actions to direct the NanoSampler. Each action is explained below, together with the process for building up an SOP Playlist.

SOP Player - New Playlist				_ 0 🔀
Accessory	Eject 🕜 Help	•		
Player Log sheet				
Action	Actions Properties			
	Standard Actions			
	Run SOP	Run macro	Pause playlist	Set temperature
	Titrator Actions			(); ();
	Go to pH	Inject titrant		
	Autosampler Actions			(Q)
	B Run SOP	₽ Wash	🗟 Initial purge	
📙 🖾 🕶 🐻 🖉 🕷 👘 🐚 🗙 Sequence i	runs:			
Configure your playlist and press Start when ready				

The Autosampler Actions are all grouped together:

- Run SOP
- Wash
- Initial purge

Wash

The wash step pumps transport fluid through the Quartz Flow Cell in order to remove sample. One wash cycle is equivalent to pumping 1000 µL through the Flow Cell.

• To add a wash step, left click on the **Wash** icon. The number of wash cycles in the wash step is shown under the **Properties** tab (3 by default). To change this enter the number of wash cycles required.

Initial Purge

The initial purge flushes the sample and buffer loops with transport fluid to ensure that no air bubbles are present within the system.



Note: always perform an initial purge when the NanoSampler has not been operational for more than a day.

- Click the Initial Purge icon to add it to the playlist actions.
- Click the Actions tab to add the next action to the Autosampler Action sequence.

Run SOP

The **Run SOP** action loads the relevant SOP for analyzing your sample and directs the NanoSampler to the relevant sampling positions.

• Click **Run SOP**, set up the parameters and then locate the SOP file and click **Open**.

General setup

There are three ways to run an NanoSampler measurement:

- Single Vial specify a single vial on which to run the SOP.
- Range specify a complete range of adjacent vials on which to run the SOP.
- Load from file specify any pattern of vials on which to run the SOP.



The following options are common to all running methods:

- **Perform wash between vials/before or after sequence/between aliquots** the wash sequence flushes the flow cell and relevant tubing (the needle itself is washed after any sample load step and is not adjustable). The number of wash cycles will be as specified in the default settings unless manually edited. The more wash cycles used, the more fluid is required in the system.
- Aliquots per vial number of samples taken from each vial (up to a maximum of 3). The analysis is run on each aliquot.
- Append vial location to sample name(s) this is useful when later identifying the source of the measurement for example, if activated, the measurement of tray 1, well A01 will ensure the records for that vial are named "....(1A01)" etc.
- **Default wash cycles** the default number of wash cycles is set to 3. This can be changed, and will be applied to ALL wash steps in the sequence. Wash cycles can be specified to run between different vials, and also at the start and end of the sequence if required.

For each aliquot to be measured two commands are added to the playlist – **Load Sample** (which contains the location parameters and the missing vial action), and below it, the name of the SOP to be used.

Click Load Sample then the Properties tab to view or edit these parameters.

🚰 SOP Player - New Playlist		- • •
Accessory Start Stop	Eject 🕐 Hel	p
Player Log sheet		
Action	Actions Properties	
Initial Autosampler purge	▲ Sample Load Proper	ties
Initial Autosampler purge	Tray ID	1
Initial Autosampler purge	Row ID	A
Load Sample (1A01)	Column ID	1
≌ip test-autosam1 ((1A01))	Missing vial action	Skip to next load step
	Tray ID	
	The tray which contains the	e vial to sample from
🛅 🚵 = 🖬 🗶 🐰 🐚 🛅 🗙 Seq	uence runs: 1	
Configure your playlist and press Start when ready.		
].;;

The position of the vial to be analyzed is identified by Tray, Row, and Column (the default starting position is Tray 1, Row A, Column 1). To edit these settings click the relevant property and enter the correct number/letter.

To change the **Sample Name** click on the SOP icon and enter the correct title in the **Properties** tab. This will be displayed in the 'Sample Name' column of the measurement file.

To complete the playlist sequence, a wash step can be added at the end to remove the sample from the flow cell and clean the system ready for the next sample vial.

This sequence of steps can be repeated to build up a playlist for a sequence of vials.

Single vial

The single position option is shown with no first and last position. Any vial in either tray can be selected for the analysis.

- 1. From the Naming and positioning section, select Single Vial.
- 2. Enter the **Position** of the vial using the **Tray**, **Row** and **Column** selection options. By default, the position is Tray 1, Row A, Column 1.

Options	
Defau	t wash cycles 3 🚔
🔽 Per	form wash between vials
🔽 Per	form wash before sequence
Per	form wash after sequence
Aliquol	s per vial 1 🚔
E Per	form wash between aliquots
	and dellaration is comely accord?
	INFOLD STATE CALLER FOR THE SALE AND A STATE OF ST
<u>е</u> он	sena viandeation to sample name(s)
Naming a	and positioning
Naming a	and positioning e Vial
Naming a	and positioning e Vial Range Load from file
Naming a Singl Position Tray	nd positioning e Vial () Range () Load from file) Tray 1
Naming a Singl Position Tray	and positioning e Vial Range Load from file Tray 1 Tray 2
Naming a Singl Position Tray	and positioning e Vial Range Load from file Tray 1 Tray 2
Naming a Singl Position Tray Row	Ind positioning e Vial Range Load from file Tray 1 Tray 2 A v
Naming a Singl Position Tray Row	Ind positioning e Vial Range Load from file Tray 1 Tray 2 A -

3. Click OK, then select the SOP to finalize the selection and complete the playlist.

Range

Using this method you must take samples from a contiguous range of wells, which can be from either tray or span both trays. For example, you could start and begin sampling only Tray 2 and take samples from only half of the wells if required.

1. From the Naming and positioning section, select Range.



2. Specify both the **First position** (where the sampling commences) and the **Last position** (where the sampling ends).

€

Note: using this method, vials must be placed in the plate in sequence - it is not possible to sample from non-contiguous vials using this function - instead the **Load from file** option should be used.

Vials are analyzed by row, then column. Ensure the location of your vials in the trays corresponds to that entered in the SOP Player. Sampling begins with the tray and vial indicated as the **First position** and completes on the tray and vial **Last position**. For example if you only wished to sample from Tray 1, and had loaded it with 10 vials in this pattern (A1-B2):



The software would be configured as shown:

Run SOP Parameters	×
Options	-
Default wash cycles	3 🌩
📝 Perform wash betwe	en vials
Perform wash before	sequence
Perform wash after set	equence
Aliquots per vial	1
Reform wash betwee	en aliquote
Append vial location	to sample name(s)
Naming and positioning	
💿 Single Vial 🛛 💿 Ra	ange 💿 Load from file
First Position	Last Position
Tray 💿 Tray 1	Tray 💿 Tray 1
🔘 Tray 2	🔘 Tray 2
Row 🖌 👻	Row B 👻
Column 1 🚖	Column 2 🚖
	UK Cancel

- 3. Click **OK** to confirm the settings.
- 4. The system now prompts you to locate the SOP you want to use for this measurement (this must be a size SOP) do this and click **Open** to confirm your selection. The playlist now automatically propagates with each step required to sample all the vials specified.

SOP Player - test2.sopl	
Accessory Start Stop Accessory	
Player Log sheet	
Action Action Actions Properties	
Load Sample [1A01] Load Sample [1A01 #2] E Load Sample [1A01 #2] E	
2%) test-sudoram1 (Test Sample (1402 #1)) ■ Load Sample (1402) © test-sudoram (Test Sample (1402 #1))	
Coad Sample (1402) Coad Sample (1402 #2) Coad Sample (1402 #2) Coad Sample (1402 #2) Coad Sample (1402 #2)	
constantial (1etc sample (LAC2 +3)) Load Sample (LAC3	
*** total social (Tet 3 social	
D Load Sample (1A04) C ter-success (1A04 #1) C ter-success (1A04 #1) C ter-success (1A04 #1)	
Configue your playfeit and press Stati when ready	
T: 25.0°C / Pos: 4.20mm / Attn: 8	

Each individual aliquot is automatically assigned a unique identifier based on the Tray position and aliquot number. The playlist can be reviewed and edited if required.

Missing vial action

 Click the cog icon on the Autosampler Actions bar to display the Autosampler settings window:

Autosampler settings	- • •
Missing vial action Skip to next vial Abort playlist	
ОК	Cancel

The system can be set so that any wells that do not contain a vial are dealt with in a consistent way. This default setting is then used on all new playlists that are created.

- Skip to next load step the system passes the well and tries the next available well in the playlist.
- **Abort playlist** the playlist is stopped. Any data already recorded is retained in the measurement file.

Note: You can override this on a per-vial basis by clicking the Load Sample action for the vial you need to amend and then modifying the **Missing vial action** in the **Properties** tab.

Starting the SOP Playlist

- 1. Click the green **Start** button on the SOP Player.
- 2. Choose a location to save the SOP Playlist, which can be saved for subsequent repeat runs. We recommended that you save the Playlist in the same folder as the SOPs.



- 3. After clicking **Save** the sequence will begin.
- 4. Each measurement is now added to the measurement file and incrementally numbered in the order the samples were taken. If the option to add the vial location to the sample name was selected, the name will be labeled appropriately.

Whilst the measurements are taking place, it is possible to monitor the progress of the measurement on the SOP Player window using, for example the Multi-view, Intensity PSD or Log sheet tabs.

Vial position files

Normally you set a range of wells from which to sample, based on a defined start and end point and capturing all wells that fall into that range. However, to offer greater flexibility, for example if you wanted to sample every other column in the tray, with the others empty, the NanoSampler system caters for this with vial position files. These are simple tab-delimited text files that define your sampling "map" and can be generated in any text editor, for example Notepad, or by using Microsoft Excel. In addition to defining the sampling schedule, the names entered into the text file are used in the measurement file.

Using Notepad to create position files:

The file should be configured as shown in the image below:

Vial Locations Ful	ll.txt - Notepad							
<u>File Edit Format</u>	<u>V</u> iew <u>H</u> elp							
Sample A1 Sample B1 Sample C1 Sample D1 Sample E1 Sample F1	Sample A2 Sample B2 Sample C2 Sample D2 Sample E2 Sample F2	Sample A3 Sample B3 Sample C3 Sample D3 Sample B3 Sample F3	Sample A4 Sample B4 Sample C4 Sample D4 Sample E4 Sample F4	Sample A5 Sample B5 Sample C5 Sample D5 Sample E5 Sample F5	Sample A6 Sample B6 Sample C6 Sample D6 Sample E6 Sample F6	Sample A7 Sample B7 Sample C7 Sample D7 Sample D7 Sample F7 Sample F7	Sample A8 Sample B8 Sample C8 Sample D8 Sample B8 Sample F8	•

Tabs are used to separate measurement vials. If a measurement is not required from the vial, no text should be entered (i.e. the presence of text tells the system to measure that position).

Using Microsoft Excel to create position files:

The file should be configured as shown in the image below:

	A	В	С	D	E	F	G	Н
1	Sample A1	Sample A2	Sample A3	Sample A4	Sample A5	Sample A6	Sample A7	Sample A8
2	Sample B1	Sample B2	Sample B3	Sample B4	Sample B5	Sample B6	Sample B7	Sample B8
3	Sample C1	Sample C2	Sample C3	Sample C4	Sample C5	Sample C6	Sample C7	Sample C8
4	Sample D1	Sample D2	Sample D3	Sample D4	Sample D5	Sample D6	Sample D7	Sample D8
5	Sample E1	Sample E2	Sample E3	Sample E4	Sample E5	Sample E6	Sample E7	Sample E8
6	Sample F1	Sample F2	Sample F3	Sample F4	Sample F5	Sample F6	Sample F7	Sample F8

If no measurement is required from a vial, ensure the cell is left empty.

When the file has been created, save in the Text (Tab Delimited).txt format.

Note: when viewing a vial position file in Excel the labeling in Excel of rows and columns must be ignored - columns are actually rows. For example, H1 in Excel is actually the vial position A8. See the Excel image for clarification.

Tip: empty rows at the beginning of an Excel file (i.e. columns in the vial position) are deleted by Excel when you save a TXT file. There are two workarounds: either color the empty cells or select both the full and empty cells, and then choose **Data-Group Rows**. The empty cells are then incorporated into the TXT file correctly when you export to TXT, enabling the system to recognize their "empty" status correctly.

MAINTENANCE & TROUBLESHOOTING

The instrument is designed to minimize supervisor/operator maintenance. This topic explains the routine user maintenance procedures that can be performed.



Warning! Unless advised within the content of this manual, only Malvern Instruments trained personnel are permitted to remove the main covers from the instrument or connected devices.

This section covers the following procedures:

Maintenance	50
Troubleshooting	.54

Maintenance

This section provides details of maintenance procedures required to keep your NanoSampler operating correctly. For some of the procedures described it may be necessary to remove the cover - see: Using the hardware on page 28.



Note: do not disconnect the NanoSampler from the power source for any of the maintenance procedures described in this section. This is to allow software control of the system.



Note: only use Malvern spare parts to replace the items detailed in this section.

Cleaning the NanoSampler

Clean the outside of the unit using a damp cloth with non-aggressive cleaning liquid. Other items that may need periodic cleaning include:

- **Valve leak bin**: a special leak bin is installed underneath the injection valve. You can clean this bin with a damp cloth with non-aggressive cleaning liquid.
- **Sample tray**: if sample has been spilled on the sample tray, clean the tray with a damp cloth and non-aggressive cleaning liquid.
- **Drain tubing**: regularly flush the drain tubing to prevent clogging and to ensure that liquids and condensate are disposed of.

Tubing



Note: a tubing spares kit is available to order from Malvern Instruments.

The NanoSampler tubing covered in this section is user-replaceable. All other tubing must be assumed to be only replaceable by your Malvern service engineer - contact Malvern Instruments if in doubt.



Note: use only ferrules and nuts that are supplied by Malvern Instruments.

The tubing is compressed into the valve by the ferrule to ensure that the connection is leak-tight. When making tubing connections:

- 1. Slide the nut over the tubing.
- 2. Slide the ferrule over the tubing, with the narrow end towards the end that will be connected to the valve. Push the tubing through the ferrule until 3-4 mm of the tubing projects from the narrow end of the ferrule.
- 3. Insert the end of the tubing in the valve connection until it touches the back of the connection.
- 4. Hold the tubing in this position and screw the nut into the connection on the valve.
- 5. Fasten the nut finger-tight.

Replacing cell transport fluid tube

As part of your regular maintenance routine, or whenever you suspect that the tubing may have become blocked, this can be replaced as follows:

• Undo the retaining connector from the top tube, where it connects to the right side of the valve at the top of the syringe. Remove the tube. Replacement is a reversal of the removal process.



Note: ensure that only parts supplied by Malvern Instruments are used.



Waste tubing

The following connections for disposal of waste liquids are made:

- **General waste**: connect the drain tubing (in the installation kit of the NanoSampler) to the left-hand drain hose connector (see System configuration on page 25). Place the other end in a waste bottle on the floor. This is used to drain all liquid dispensed to the wash position. Sample liquid that is not injected is also removed through this tubing.
- **Condensation water and leakage drain**: leaked solvents and condensation (from cooling) are drained through the right-hand hose connector (**see System configuration on page 24**).

Ensure that none of the drain or waste tubes are twisted as this might obstruct the flow path.

Replacing fuses

If you need to replace the fuses, ensure that you install fuses of the same type and rating. Fuses are located in the fusebox at the back of the NanoSampler (**see System con-figuration on page 24**). Fuses of the following types are installed in the NanoSampler:

• Fuse type: 2 x 2.5AT, 250V, 1500A



Warning: switch off and disconnect the NanoSampler from its power source by removing the mains power cord before replacing fuses.

Troubleshooting

The following table provides details on potential problems and how to resolve them:

Problem	Possible Cause	Remedy
Unexpected or "flat line" meas- urements	Blocked tubing	Run full purge on system. Ensure washes are built into the SOP playlist.
(as above)	Insufficient fluid in wash bottle, unit is sampling air only	Refill bottle
(as above)	Broken tubing	Replace tubing
Flashing LED on front of unit	Error	Power off and restart instrument and software
Sample leakage	Poor joint sealing on tubing	Ensure all connectors are hand tight, replace cracked or visibly degraded connectors.
Play list will not start, user notified by software	Communications loss: either dis- connected USB cable or NanoSampler unplugged during measurement	Check if USB cable between NanoSampler and PC is dis- connected . Check power supply connections.
During measurement: SOP Player stages skipped	(as above)	(as above)
Power failure mid-measurement. NanoSampler stops.	Power supply issues	Restart system, check error mes- sages in log sheet. Flush system, restart measurement, ensuring a wash cycle is first.

Error codes

The following errors may occur with the NanoSampler. These are automatically generated by the system and written to the log file, which can be opened in any text editor:

C:\Program Files (x86)\Malvern Instruments\Zetasizer Software\Autosampler.log



Note: the errors listed here are those for which a remedy can be performed by the user. In the case of other errors, please contact Malvern support.

Tray unit

Error Code	Description	Action
ERROR 294	Home sensor not reached.	Check for obstructions on tray move- ment (slide the tray back and forth)
ERROR 295	Tray position check error.	Check for visible obstructions in tray area
ERROR 296	Home sensor not de-activated.	Check if the transport foam is removed from tray compartment
ERROR 298	Tray position is unknown.	Restart instrument

Needle unit

Error Code	Description	Action
ERROR 303	Horizontal: needle position is unknown.	Restart instrument
ERROR 304	Horizontal: home sensor not reached.	Check movement for any obstruction
ERROR 306	Horizontal: home sensor not de-activated.	Check movement for any obstruction
ERROR 308	Needle position check error.	Check for obstruction in horizontal movement
ERROR 312	Vertical: needle position is unknown.	Restart instrument
ERROR 313	Vertical: home sensor not reached.	Check movement for any obstruction
ERROR 315	Vertical: home sensor not de-activated.	Check movement for any obstruction

Syringe dispenser unit

Error Code	Description	Action
ERROR 324	Syringe valve did not find destination pos- ition.	Check belt and pulleys
ERROR 330	Syringe home sensor not reached.	Check belt and pulleys
ERROR 331	Syringe home sensor not de-activated.	Check belt and pulleys
ERROR 334	Syringe position is unknown.	Restart instrument
ERROR 335	Syringe rotation error.	Check for flowpath restriction by per- forming a wash

Electronics

Error Code	Description	Action
ERROR 280	EEPROM write error.	Restart instrument
ERROR 282	EEPROM error in settings.	Restart instrument
ERROR 283	EEPROM error in adjustments.	Restart instrument
ERROR 284	EEPROM error in log counter.	Restart instrument
ERROR 290	Error occurred during initialization	Restart instrument

APPENDIX

This section contains the following information:

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Specifications

General

Installation category	11
Pollution degree	2

Physical Dimensions

Dimensions	300 mm (w) x 510 mm (d) x 360 mm (h)
Weight	19 kg



Note: For physical dimensions of the Zetasizer Nano, please refer to the Zetasizer Nano Basic Guide.

Electrical

Power requirements	95 - 240 Vac +/-10%, 50-60Hz
Power consumption	200VA
Fuses	2 x 2.5AT, 250V, 1500A
International Standard	IEC60127-2, UL recognized

Communication

Communication ports	RS232 standard, USB
Max. length for comms cables	3m

Sampling

Minimum sample volume single aliquot	750μL
Sample volume for subsequent aliquots	420µL
Maximum aliquots per vial	3
Cross contamination	<0.05%
Syringe volume	500µL
Needle volume	60µL

Sample capacity	2 racks of 48 1.5mL vials
Maximum viscosity	6.44cP
Vial/Plate	dimensions (incl. cap) Max. plate/vial height: 47 mm (incl. septa or cap)
Loop volume (sample / buffer)	1000µL
Vial detection	Missing vial/well plate detection by sensor
Switching time injection valve	Electrically < 100ms
Piercing needle precision	±0.6mm
Wash solvent	Integrated wash solvent bottle (full 96 wells requires external bottle of 2L
Wetted parts in flow path	SS316, PTFE, EFTE, PEEK, Glass

Chemical compatibility

Components of the NanoSampler that may come into contact with the sample are manufactured from materials that are considered to give the widest protection from chemical attack. However, it is important to check that any sample used is chemically compatible with the materials mentioned.



Note: The paint on the housing of the NanoSampler is not compatible with Chloroform.

Regulatory statements

FCC Notice (US only)

The Federal Communications Commission (FCC) mark on this product signifies conformance to FCC regulations relating to Radio Frequency Devices. These have been satisfied by testing the product against, and being found to be compliant with:

FCC CFR 47 Part 15:October 2011.Class A digital device.

The device complies with part 15 of the FCC Rules. Operation is subject to the following two conditions:

- 1. This device may not cause harmful interference, and
- 2. This device must accept any interference received, including interference that may cause undesired operation.

Note: This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 of the FCC rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at his own expense.

Note: Changes or modifications not expressly approved by Malvern Instruments Limited could void the user's authority to operate the equipment.

CE Declaration of Conformity

The CE badge on this product signifies conformance to the following European Directives:

- EMC directive 2004/108/EC {BS EN 61326-1: 2006}
- Low Voltage Directive 2006/95/EC {BS EN 61010-1: 2010}

- Low Voltage Directive 2006/95/EC {BS EN 61010-2-081:+A1}
- Machinery directive 2006/42/EC
- WEEE directive 2012/19/EC Waste Electrical and Electronic Equipment Directive
- RoHS directive; 2011/65/EC Restriction of Hazardous Substances Directive

Canadian Regulatory Information

(This applies to Canada Only.)

This digital apparatus does not exceed the Class A limits for radio noise emissions from digital apparatus set out in the Radio Interference Regulations of the Canadian Department of Communications.

Note that Canadian Department of Communications (DOC) regulations provide, that changes or modifications not expressly approved by Malvern Instruments Limited could void your authority to operate this equipment.

This Class A digital apparatus complies with Canadian ICES-003.

Cet appareil numérique de la classe A est conforme à la norme NMB-003 du Canada.

VCCI acceptance (Japan only)

The Voluntary Control Council for Interference (VCCI) mark on this product signifies compliance to Japanese EMC regulations as specified by VCCI.

> この装置は、情報処理装置等電波障害自主規制協議会(VCCI)の基準 に基づくクラスA情報技術装置です。この装置を家庭環境で使用すると電波 妨害を引き起こすことがあります。この場合には使用者が適切な対策を講ず るよう要求されることがあります。

Translation:

This is a Class A product based on the standard of the Voluntary Control Council for Interference by Information Technology Equipment (VCCI). If this equipment is used in a domestic environment, radio disturbance may occur, in which case the user may be required to take corrective actions.