

Equipment Operation Procedure

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Title:	Use and Ma	intenance of the NanoDrop
Version:	v2	
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SOP History		
Number	Date	Reason for Change
v1	01/02/2013	Original
V2	01/02/2018	Update

1.0 Purpose –

The purpose of this SOP is to outline the principles of the routine use and maintenance of the NanoDrop in Laboratory 248 at the St Andrews School of Medicine (SASoM).

2.0 Scope -

This SOP applies to routine use and maintenance of the NanoDrop within the SASoM.

3.0 Responsibilities

It is the responsibility of all users of the NanoDrop within the SASoM to comply with this SOP.

4.0 Procedure –

Principles of Operation

The NanoDrop can be used to measure the following:

- Nucleic acid concentration and purity of nucleic acid samples <15,000 ng/µL (dsDNA) without dilution
- General UV-Vis spectrophotometry
- Purified protein analysis (A280)
- Fluorescent dye corporation for Microarray and Proteins & Labels applications
- Expanded spectrum measurement and quantitation of fluorescent dye-labeled proteins, conjugates, and metalloproteins
- BCA Assay analysis of protein



- Bradford Assay analysis of protein
- Lowry Assay analysis of protein
- Pierce 660nm Assay analysis of protein
- Microbial cell culture measurements
- Kinetic methods

Operation:

On the laptop double-click the NanoDrop software icon and select the software application of interest from the right pane.

NANODROP 2000/2000c						
Group	Classic 💌					
Nucleic Acid	Protein A280	Kinetics Editor				
Micro Array	Proteins & Labels	Method Editor				
UV-Vis	Protein BCA	- Default Method				
Cell Cultures	Protein Bradford					
	Protein Lowry					
	Protein Pierce 660 nm					

Follow the prompts for instrument initialisation.

Select Add to report prior to a measurement to save the sample data to a workbook.

Establish a blank using the appropriate buffer.

Raise the sampling arm and pipette 2ul of the blank onto the lower measurement pedestal. Lower the sampling arm and click Blank. The blank solution is generally the same buffer that the molecule of interest is suspended or dissolved in.

Raise the sample arm and wipe away the blank from the measurement pedestals using a dry, lint free laboratory wipe.

Simple wiping prevents sample carryover in subsequent measurements for samples varying by more than 1000 fold in concentration.

Enter the sample ID in the appropriate field. Pipette 1.5µl of sample, lower sample arm and click Measure.

It is generally recommended that an aliquot of the blanking buffer be measured as if it were a sample.

Although it is not necessary to blank between each sample, it is recommended that a new blank be taken every 30 minutes when measuring many samples.



A final cleaning of both measurement surfaces with distilled water is recommended after the last sample measurement.

Software features:

The NanoDrop 2000c software interface is divided into a left pane and a right pane. Task bars and Action buttons are located in the left pane while the right pane acquisition pages display the sample spectra.



Task Bars

Home	displays the main menu with the available selection of applications
Measure	(specific application)- active application screen
My Data	manages data archiving and retrieval
Reports	exportable user configurable report associated with current data set
Diagnostics	accesses the Intensity and Calibration checks
Options	includes tabs for account management and report printing options
Action Icons	
Measure	initiates the measurement of a sample
Print Screen	prints a copy of the spectrum and associated sample data to the default printer
Blank	initiates the measurement of the buffer or carrier liquid in which the sample is suspended
Re-Blank	establishes a new reference (blank) and recalculates the absorbance spectrum for the most recent sample and displays the revised spectrum on the screen
Optional Sele	ections
Add to Repor	used to indicate sample data that should be added to the current report
Overlay spec	tra selection of this feature displays multiple spectra at a time
Small sample	e vol 0.5ul volume capability used for concentrated samples. Nucleic acid and

0.5ul volume capability used for concentrated samples. Nucleic acid and A280 applications only (examples >150 ng/ul dsDNA or >4.5 mg/ml BSA)

Use cuvette

Menu Bar

File used to open and close workbooks Help electronic Help files- may be accessed directly from many software screens

select to enable cuvette measurements

Sample data is stored in workbooks at user-specified locations. Workbooks may be accessed through the My Data task bar in the left pane.

The laptop has been networked and data can be saved directly onto the Systems Patholgy server (<u>\\medshares.st-andrews.ac.uk</u>) or stored locally in the short term.



5.0 Personnel protection -

Howie coat must be worn at all times.

Gloves as specified in the appropriate COSHH RA

6.0 Spillages -

Always clean up any spills immediately after use.

Only you know what you have spilt and are aware of that chemicals hazard

Mop up spills with paper towels. Wash the site of spillage with water $\underline{\&}$ detergent.

7.0 General maintenance -

Apply 3-5ul of dH₂0 onto the bottom pedestal. Never use a squirt bottle to apply de-ionized water or any other liquid to the surface of the instrument.

Lower the upper pedestal arm to form a liquid column; let it sit for approximately 2-3 minutes.

Wipe away the water from both the upper and lower pedestal with a dry, lint-free lab wipe.

Additional cleaning: Substitute 0.5 MHCI for the dH₂O in the procedure above when proteins have dried on the pedestal.

Decontamination: Use a sanitizing solution, such as a 0.5% solution of sodium hypochlorite (1:10 dilution of common commercial bleach solution, freshly prepared), to decontaminate the measurement pedestals. Follow with $3-5\mu$ of dH₂0.

The instrument should be subject to a Yearly visit by engineer to calibrate the NanoDrop.

8.0 Training -

All users have to be trained before using the instrument by a designated person.

9.0 Related documents -

- 10.1 Equipment manual
- 10.2 Equipment Maintenance Log
- 10.3 Equipment Maintenance Information sheet
- 10.4 Risk assessments RA/GEN/027



10.0 Approval and sign off -

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