product code:

see ordering information

Ultrospec 3300 pro and Ultrospec 4300 pro

Pharmacopoeia compliant UV/Visible Spectrophotometers

Description

The Ultrospec[™] 3300 pro is a stand-alone instrument with a high resolution graphics display and extensive built-in applications software that has a performance which fulfils the requirements of the Pharmacopoeia, Figure 1. The Ultrospec 4300 pro is a PC controlled instrument that is supplied complete with SWIFT II disk based applications software. They are ideal for use in the Development, Quality Control and Analytical laboratories of Biotech and Pharmaceutical companies engaging in drug development and research. Here, the requirements of high resolution scanning for assessment of impurities in sample preparations are fulfilled since the instruments have a bandwidth of less than 1.8 nm. Both instruments are available in a range of colours - classic, yellow, plum, apple - and have both exciting styling and design (registered design protection applied for) that place them at the forefront of their class.

The needs for Nucleic Acid Quantification and Protein Determination, essential techniques in genomic and proteomic



Figure 1. Ultrospec 3300 *pro* is a Pharmacopoeia compliant, stand-alone instrument with a high-resolution graphics display.

research, are well addressed by the Ultrospec 3300 *pro*, with stored methods and useful information pages. Customized methods in all application modes or equations involving a range of absorbances and factors can be entered and stored for use; a total of 50 user definable methods can be saved.

Features	Benefits
Pharmacopoeia compliant	Confidence that demanding requirements are met
GLP self test diagnostics	Prove performance of instrument at any time at no extra cost
Available in four colours: classic, yellow, plum, apple	Greater customer choice
High specification	< 1.8 nm bandwidth for high resolution scanning Maximum scan speed of 6200 nm/minute
Deuterium and tungsten lamps	Optimum performance across wavelength range
Qualification and performance verification logbook supplied as standard	Keep a log of key instrument performance parameters as a function of time
Stand alone (Ultrospec 3300 pro)	Discrete measurements without need of PC based control software
Built in applications software (Ultrospec 3300 pro)	No additional cost for software modules
Integral LCD with graphical user interface (Ultrospec 3300 <i>pro</i>)	Intuitive and easy to use Instantaneous display of graphics with data manipulation prior to printing
Spreadsheet interface software supplied as standard (Ultrospec 3300 <i>pro</i>)	Download to Microsoft [™] Excel at no extra cost for archiving of results and further manipulation of data
SWIFT II applications software supplied as standard (Ultrospec 4300 <i>pro</i>)	Full suite of application modules Automatic run log (audit trail)
PC only control on Ultrospec 4300 pro	Privacy of information on Windows [™] NT workstation for multiple users



Pharmacopoeia Compliant

Both Ultrospec 3300 *pro* and Ultrospec 4300 *pro* offer the user the confidence of using a UV/Visible spectrophotometer that is fully Pharmacopoeia compliant in terms of resolution, stray light, absorbance accuracy and wavelength accuracy.

The British Pharmacopoeia (A88 Appendix II B) states that to verify resolution, the spectrum of a 0.02 % (v/v) solution of toluene in hexane should be recorded; the ratio of the absorbance at the maximum (269 nm) and minimum (266 nm) should be at least 1.5, Figure 2. The European Pharmacopoeia (1984, V.6.19, 2nd Edition) states that to verify stray light, the absorbance of a 1.2 % w/v solution of potassium chloride with a pathlength of 1 cm should be more than 2.000 when compared with water as reference liquid. The European Pharmacopoeia also states that to verify wavelength accuracy, the absorption maxima of holmium perchlorate solution should be recorded. Each instrument is tested with the appropriate solutions described above to ensure that this is the case, and the results obtained are shipped with it as a final test certificate for 100 % user confidence.

Although the European Pharmacopoeia states that to verify absorbance accuracy, potassium dichromate in sulphuric acid solution should be used, we carry out absorbance accuracy tests using neutral density filter standards which are traceable to NPL or NAMAS. These results are also included with both instruments.

Good Laboratory Practise

Being able to check that an instrument is working to its published specification is an essential pre-requisite for GLP. Both instruments perform GLP self diagnostic tests for bandwidth, absorbance accuracy, wavelength accuracy and stray light and compares them with the values obtained during instrument manufacture (or last accredited engineer service), Figure 3. The instrument is supplied with a "Qualification and Performance Verification Logbook" which clearly explains and details the relevant Pharmacopoeia tests and provides a means of keeping an ongoing log of key instrument specification parameters as a function of time.



Figure 2. Spectrum of toluene in hexane (0.02 % v/v solution) obtained using Ultrospec 4300 pro and SWIFT II software. The ratio of the absorbance at the maximum (269 nm) and minimum (266 nm) should be at least 1.5 to fulfil the Pharmacopoeia; the results shown give a ratio of 1.722, proving that the instrument passes the BP test.

Ultrospec 3300 pro UV/Vis Spectrophotometer					
Lab name					
Instrument		Ultrospec 3300pro			
Serial number		81844			
Software		4195 V1.0, Slave V	1.0		
Last serviced		20/11/0009:52			
Instrument stat	Instrument state at calibration				
GLP Calibrate	d	20/11/00 09:52			
Calibration		Full UV/Visible			
	656nm (1.3 - 1.8nm)	1.7nm	PASS		
Wavelength (6	556.1nm)	656.0	PASS		
Absorbance a	-				
220nm (1.763	,	1.772 1.649	PASS		
	340nm (1.633-1.665A) :		PASS PASS		
500nm (1.477-1.491A) :		1.484	PA33		
Stray light at	25 % T) .	0.013%	PASS		
220 nm (<0.025 %T) :			.,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,		
UV lamp:		85 % of original energy			
	Vis lamp: 95 % of original energy				
Current instrum					
Accessory Eight Position Cell Cha		0			
UV lamp:	installed 15/11/00 11				
	baseline in use	20/11/00	09:53		
	baseline stored	20/11/00	09:53		
Vis lamp:	installed 15/11/00 11	,	00 53		
	baseline in use baseline stored	20/11/00 20/11/00	09:53 09:53		
	Daseinie Storeu	20/11/00	09:00		

Figure 3. Ultrospec 3300 pro GLP self diagnostics print out

Ultrospec 3300 pro display and keypad

A high resolution, ¹⁴ VGA size LCD graphics display provides the user with set up parameters and experimental results in either English, German, French, Spanish, Italian or Russian. The information bar at the bottom of the display contains user prompts and instrument status. The Ultrospec 3300 *pro* has a wide range of software options which are presented on the LCD in an index card format and accessed by pressing the function key for set up and utilities and the mode key for use in experiments. Text entry is very easy, since the keypad has an alphanumeric function at the appropriate menu options, and letter entry is in the same manner as when using a mobile phone.

Utilities and set up accessed by pressing function key (GLP off)

Accessory	Printer	Display	Set up
Identify accessory	Select printer driver	Change contrast	User
			Lamps
			Baseline
			Clock
			Service

Set up

On the User page, user and laboratory names entered are printed out as a header at the top of experimental results together with the instrument description, Figure 5; this can be an asset register number. The choice of having a GLP print out at the start of each working day when the instrument is switched on is available here also. Output, whether to printer for hard copy, computer using the spreadsheet interface software, or both, is also selected here.









Operating modes accessed by pressing mode key

Basic	Methods	Nucleic	Protein	Applications
Absorbance	Save	DNA	Bradford	Wavelength Scanning
Transmission	Recall	RNA	Lowry	Multi Wavelength / Equation Entry
Concentration	Clear	Oligo	Biuret	Reaction Kinetics
		Scan	BCA	Standard Curve
		Tm Calculation	UV Methods	Substrate Concentration
		Information	Information	

Nucleic Acid Quantification

Stored routines for DNA, RNA and oligonucleotide samples ensure that the quantification and purity check of nucleic acids is both rapid and easy. Volumes as low as 3µl and 7µl can be accommodated using the capillary and the ultra micro-volume cell and the appropriate cell holder, respectively; in addition, the UViMicro UV transmitting disposable cell (volume 20 - 2000 µl, standard 15mm optical height) is compatible. Dilution factor is accounted for automatically, Figure 6. The integrity of a nucleic acid preparation can also be investigated using the nucleic acid 200 - 350 nm scan routine. Both A260 multiplied by the specific quantification factor and the A260/A280 absorbance ratio are reported.

The Tm or melting temperature characterizes the stability of a nucleic acid hybrid formed between an oligonucleotide and its complementary strand and is essential information for design of effective Polymerase Chain Reaction (PCR) and sequencing primers and oligonucleotide hybridisation probes for Southern, Northern and dot blot analyses. Tm can be calculated theoretically on the Ultrospec 3300 *pro* by entering the oligonucleotide base sequence, the oligonucleotide concentration and the salt concentration. Molecular weight, conversion factor (μ g/ml) and concentration in pmol/ μ l, useful for sequencing, are also calculated, Figure 7.

Ultrospec 3300 *pro* also contains useful information for selecting the appropriate cell to use, formulae for mass to moles conversion and the codon dictionary, Figure 8.

Protein Determination

The Ultrospec 3300 *pro* has stored routines for the popular Bradford, Lowry, Biuret and BCA protein determination methods, Figure 9, and users can modify the protocols to suit their needs. Micro protein assay versions of these protocols where absorbance values are lower, are easily carried out on the Ultrospec 3300 pro by increasing the integration (or measurement) time to minimise the effects of instrument electronic noise. The instrument also has stored routines for some UV protocols; Christian Warburg for protein impurity in nucleic acid preparations, and empirical methods at 280 / 205 nm and 225 / 215 nm. Ultrospec 3300 *pro* also contains useful information for factors relating absorbance to concentration for some common proteins and look up tables for amino acids.







Figure 7. Tm calculation for a primer



Figure 8. Information page



Figure 9. Protein determination using Bradford method

Wavelength Scanning

With a wavelength range of 190 - 1100 nm at step intervals of 0.1 - 1.0 nm, a maximum scanning speed of 6200nm/minute, an absorbance range of - 3.000 to + 3.000, and a bandwidth of < 1.8 nm, the Ultrospec 3300 *pro* is perfect for scanning, Figure 10. The instrument has a precision encoder embodied in the software to ensure highly accurate and reproducible wavelength setting with a range of scanning speeds at different step intervals. In addition, the 1st, 2nd and 4th derivative of a spectrum is only a key press away, as is a peak identification routine.

Scan speed,	Step Interval, nm			
nm / minute	1.0	0.5	0.2	0.1
Fast	6200	3500	3000	1550
Medium	3500	2300	1550	880
Slow	2500	1450	700	405

Multi Wavelength / Equation Entry

Many quality control and development processes require the use of absorbance values in equations to determine a meaningful parameter. The Ultrospec 3300 *pro* is able to measure absorbances at specified wavelengths and then use these values in a user defined equation, Figure 11; two separate equations can be used in one method, which can of course be saved for instant recall. Thus post measurement calculations can be done automatically, Figure 12. This powerful facility enables customisation of the instrument to suit individual user needs and the flexibility to change them as required.

Reaction Kinetics

The analysis of change in absorbance with time, particularly for NAD/NADH assays at 340 nm, is one of the most powerful applications of the UV/Visible spectrophotometer. Ultrospec 3300 *pro* allows the entry of lag time, reaction time and a slope conversion factor; assays can be carried out in serial or parallel and a customised method can, of course, be saved for future use. The progress of the reaction is monitored graphically, with a display of weighted average slope, Figure 13. Temperature control can be achieved with our range of either water or Peltier heated accessories.



Figure 10. Typical wavelength scan



Figure 11. Multi Wavelength Equation Entry



Figure 12. Results calculated automatically



Figure 13. Typical enzyme kinetics assay

Standard Curve

The graphical display of the Ultrospec 3300 pro enables easy visualisation of a plot of the absorbances of a series of standards (with replicates), Figures 14 and 15; the curve fitting methods of linear regression, linear interpolation and quadratic spline are all available. Samples are then measured against the apprpriate standard curve to give quantitative results that, if required, can be printed out. With common protein determinations already available as standard routines on the Ultrospec 3300 *pro*, there is ample method storage space (up to 50) for the busiest of laboratories. Micro assays, where absorbance values can be very low, can be performed if the integration (measurement) time is increased (up to 5 seconds).

Substrate Concentration

With the common use of reagent diagnostic kits in many clinical and food analysis laboratories, the Ultrospec 3300 *pro* is ideal for the measurement of absorbance change (e.g. NAD/NADH assays at 340 nm) over a specified time for solutions of different concentrations, Figure 16. A calibration curve is shown on the display, and the linear regression, linear interpolation and quadratic spline curve fitting methods can be applied prior to the measurement of samples. Once again, the method storage facility is useful for those analyses that need to be done on a routine basis.

Method Storage

Fifty methods can be stored and retrieved from any combination of the Wavelength Scanning, Enzyme Kinetics, Standard Curve, Substrate Concentration and Multi Wavelength modes to supplement those already available on Ultrospec 3300 *pro* for Nucleic Acid Quantification and Protein Determination, Figure 17. Titles are set using an alphanumeric input facility.



Figure 14. Typical Standard Curve experiment



Figure 15. Data relating to a linear regression are displayed



Figure 16. Set up for Substrate Concentration experiment

	10 11 - 20	21 - 30	31 - 40 41 - 50
1	UV 215,225 UV215	22/06/99	6 Multiwave 22/06/99 Multi-wave test
2	Wavescan Wavescan test	22/06/99	7 Kinetics 22/06/99 Kinetic test
3	Abs Ratio Abs Ratio test	22/06/99	8 Std Curve 22/06/99 Std Curve test
4	Abs Diff Abs Diff test	22/06/99	9 Substrate 22/06/99 Substrate test
5	3 Pt Net 3pt Net test	22/06/99	0 Multiwave 22/06/99 Cobat

Figure 17. Method Storage facility

Output to printer

The Ultrospec 3300 *pro* prints to a range of popular printers with the Centronics interface, enabling a printed record of results, and GLP print outs, to be kept for filing. There are built in printer drivers enabling the output of graphics to generic printer types; Seiko DPU-414, Epson FX and similar, Epson Stylus and similar and HP DeskJet and LaserJet. Figure 18 shows the instrument with a printer stand and the Seiko DPU-414 thermal printer; this combination will be welcomed in those demanding laboratory environments which require high performance but where bench space is at a premium.

Download to spreadsheet

The ability to download directly to Microsoft Excel from the Ultrospec 3300 *pro* is a very powerful feature. Results can easily be archived in this common format or exported to other compatible applications for presentation or further manipulation. Data is downloaded via a serial interface into a macro that has to be loaded onto the PC; both the cable and the macro aresupplied as standard with the instrument. Figure 19 shows results that have been output to Microsoft Excel and graphed.

Control by SWIFT II software

Although Ultrospec 3300 *pro* is a stand-alone instrument that can be used for discrete measurements on the laboratory bench, it can be controlled from a PC using SWIFT II software. SWIFT II is a suite of applications software written for the Windows 95 environment, but also Windows 98 and NT compatible, for Wavelength Scanning, Enzyme Kinetics, Time Drive, Quantification, Multi Wavelength and Fraction Analysis applications, and provides extensive post run manipulations on data acquired using the spectrophotometer.

SWIFT II software is supplied with the Ultrospec 4300 pro. Figure 20 shows Ultrospec 4300 pro connected to, and controlled by, SWIFT II software. SWIFT II software is described in detail in a companion data file.



Figure 18. Ultrospec 3300 pro with printer and printer stand



Figure 19. Download of a wavelength scan to Microsoft Excel



Technical specifications

Wavelength range	190-1100 nm, in 0.1nm steps
Scanning speed	6200 nm/minute maximum, 1 nm step
Monochromator	Concave grating with 1200 lines/mm
Wavelength calibration	automatic upon switch-on
Spectral bandwidth	< 1.8nm
Wavelength accuracy	± 0.7nm
Wavelength reproducibility	± 0.2nm
Light sources	tungsten halogen and deuterium arc
Detector	single solid state silicon photodiode
Photometric range	- 3.000 to + 3.000 A, 0.01 to 99999 concentration units, 0.1 to 200 %T
Photometric linearity	\pm 0.5% or \pm 0.003 A to 3.000 A at 546nm, whichever is the greater
Photometric reproducibility	within 0.5% of absorbance value to 3.000A (at 546nm)
Stray light	typically <0.025%T at 220nm using Nal, <0.025%T at 340nm using NaNO ₂
Stability warm up	\pm 0.001A/h at 340 nm at 0A after
Noise	\pm 0.001A near 0A and \pm 0.002A near 2A at 546 nm
Sample compartment size	140 x 220 x 80 mm
Dimensions	500 x 360 x 190 mm
Weight	13kg
Power requirements	90-265 V AC, 50/60Hz, 150 VA

Specifications are measured after the instrument has warmed up at constant ambient temperature and are typical of a production unit. As part of our policy of continuous product development we reserve the right to alter specifications without notice. We supply support agreements that help you to fulfil demands of regulatory guidelines concerning GLP/GMP. These include calibration and certification using filters traceable to international standards by certificated engineers using calibrated test tools. The choice of agreement apart from break down coverage can include both preventative maintenance and certification. The manufacturer of these products designs and manufactures in accordance with an ISO 9001 approved quality system. The products are CE compliant.

For further information, visit us at:



Figure 21. The Ultrospec *pro* range is available in a choice of four colours

Ordering Information

Ultrospec 3300 pro UV/Visible Spectrophotometer

(includes Spreadsheet Interface Software, Serial cable and Qualification Logbook)

Classic	80-2112-33
Yellow	80-2112-34
Plum	80-2112-35
Apple	80-2112-36

Ultrospec 4300 pro UV/Visible Spectrophotometer

(includes SWIFT II Applications Software, Serial cable and Qualification logbook)

Classic	80-2112-43
Yellow	80-2112-44
Plum	80-2112-45
Apple	80-2112-46
Companion products	
Seiko DPU-414 thermal printer	Enquire
Printer stand	80-2106-60
SWIFT II Method Applications Software	80-2108-31
Personal Computer	Enquire
UViMicro disposable cells (supplied in packs of 100)	80-2110-94
Companion literature	
SWIFT II data file	18-1140-40
Accessories data file	18-1140-41

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