

Powerful spectroscopy for accurate analysis

# **Understanding UV-Vis**

### Why UV-Vis?

The power of UV-Vis lies in its simplicity. Ultraviolet-Visible (UV-Vis) spectrophotometry is literally as simple as "abc" since most analyses will follow Beer's Law: Absorbance = abc. "a" is unique to each molecule at a particular wavelength; "b" is the pathlength; and "c" is the concentration of the analyte.

### The significance of spectral bandwidth

Bandwidth is like a pupil in the human eye. The wider it is, the more light the instrument can see. But sometimes, when the bandwidth is too wide, the instrument may not see enough detail in your sample. Therefore, if you know that your samples have wide absorption peaks—large bandwidths are okay (and less expensive!), but if you want to look at narrow peaks—small bandwidths may be required. Check with our product specialist when in doubt.

#### **Cell material**

Even though glass looks perfectly transparent to our vision, it is not transparent to ultraviolet radiation (that is why you cannot get a suntan behind a closed window). For this reason "quartz" (aka "silica") cells need to be used to measure samples in the ultraviolet (UV) region; if you are always going to be measuring above ~325 nm, then "glass" (aka "optical glass") or plastic cells are okay and less costly to use.

### **Beam configurations**

Single beam spectrophotometers are simple and inexpensive. However, they are somewhat more prone to drift and should be "zeroed" once every hour. Dual-beam instruments offer better stability and can be used to monitor and subtract instrument and solvent variations, as well as provide better precision and stability for longer term measurements.

### Fixed wavelength measurement vs. scanning

UV-Vis measurements for quantitative analysis are typically performed at a single wavelength. A blank solution is measured to establish "100 %T" and then the sample is measured and the absorbance is calculated. If a calibrated method has been set up by measuring one or more standards the instrument may report concentration directly. UV-Visible spectroscopy is ideally suited for this kind of measurement. Some quantitative analyses require measurement at more than one wavelength, however, and these require a more sophisticated instrument equipped to move across the spectrum during the experiment or to measure the entire spectrum at once. These "scanning" instruments can also be used to find the location and size of absorbance peaks that can be used to identify analytes and the best wavelength at which to establish a quantitative measurement method for the analyte. All Thermo Scientific™ Spectrophotometers offer scanning capability with the exception of the Thermo Scientific™ NanoDrop™ Lite Plus instrument.

### Visible vs. UV-Visible spectrophotometers

A spectrophotometer equipped with a tungsten-halogen lamp and conventional glass optics can measure wavelengths no lower than 325 nm because the lamp does not emit and the optics do not transmit below this wavelength. These instruments are generally referred to as visible spectrophotometers even though they can measure slightly into the UV and near-infrared ranges. If all the measurements made in your laboratory are done at wavelengths between ~350 and 1100 nm then a visible spectrophotometer will be a good choice.

In practice, glass optics begin absorbing light at all wavelengths less than ~350 nm, so for UV measurements below 350 nm an instrument equipped with quartz optics and a lamp that emits in the UV region is recommended. Below 325 nm it is required. Even if your sample appears transparent it may absorb ultraviolet light. Many pharmaceuticals, organic chemicals, DNA, RNA and proteins absorb in the UV range. Established methods for quantifying their concentration in solution rely on UV-Visible spectrophotometers to perform the analysis.

### The significance of pathlength

Pathlength is the distance that the light travels through the sample. Doubling the pathlength will double the measured absorbance. A 10 mm pathlength is most commonly used for UV-Vis measurements. Special long-path cells with 20, 40, 50 or 100 mm pathlengths are available for applications where the absorbance of the sample is very low, or the sample solution is very dilute. For highly absorbing or more concentrated solutions, short-path cells of 5, 2 or 1 mm pathlengths can help to keep measured absorbance within the measurement range of the instrument.

#### Microvolume measurements

The NanoDrop Microvolume UV-Vis Spectrophotometers can quantify a wide variety of biomolecules using only 1–2  $\mu L$  of sample. Using surface tension, 1  $\mu L$  of sample forms a liquid column between two optical surfaces. This enables an automatic pathlength change from 1 mm to as low as 0.03 mm during each sample measurement and allows for a large dynamic range (2–27, 500 ng/ $\mu L$  for dsDNA) essentially eliminating the need to perform dilutions. Designed for life science labs, the NanoDrop instruments include preconfigured applications for dsDNA, ssDNA, RNA, labeled nucleic acids, Protein A280, Protein A205, Protein Colorimetric assays, labeled proteins, OD600, Kinetics, UV-Vis and Custom Methods.

# Thermo Scientific instruments and software featured in this guide

- BioMate<sup>™</sup> 160 UV-Vis Spectrophotometer
- Evolution<sup>™</sup> One/One Plus UV-Vis Spectrophotometer
- Evolution Pro UV-Vis Spectrophotometer
- GENESYS<sup>™</sup> 30 Visible Spectrophotometer
- GENESYS 40 Visible/GENESYS 50 UV-Vis Spectrophotometer
- GENESYS 140 Visible/GENESYS 150 UV-Vis Spectrophotometer
- GENESYS 180 UV-Vis Spectrophotometer
- NanoDrop Eight Microvolume UV-Vis Spectrophotometer
- NanoDrop Ultra Microvolume UV-Vis Spectrophotometers and Fluorometers
- NanoDrop Lite Plus Microvolume UV Spectrophotometer
- SPECTRONIC<sup>™</sup> 200 Visible Spectrophotometer
- Insight<sup>™</sup> Pro Software
- VISION*lite*<sup>™</sup> Software

# Education, basic quality control

Instrument	SPECTRONIC 200	GENESYS 30	GENESYS 40/140
Catalog numbers	840-281700	840-277000	GENESYS 40: 840-297000 GENESYS 140: 840-308000
Spectrum		visible	
Spectral range	340–1100 nm	325–1	100 nm
Туре	Single	beam	Dual beam
Bandwidth	≤4.0 nm	5	nm
Lamp(s)		Tungsten-Halogen	
Detector	CCD	Silicon pl	hotodiode
Standard functions	A/%T/Concentration/Scan/multiwave- length/Quant (Up to 4 STDs)	A/%T/Concentration/Scan/Fixed with calcs/Quant (up to 6 STDs)/OD600	A/%T/Concentration/Scan/Fixed with calcs/Quant/Rate/0D600
User interface	On-board or computer control		On-board touchscreen tablet or computer control
Method/data storage	Quant methods to USB/All to computer	Methods On-board, Data to USB	On-board and Computer
Display	Color graphical LCD, $320 \times 240$ pixels, $7 \times 5$ cm, tiltable	5" diagonal, 32-bit color display, 800 × 480 pixels	7" color touchscreen, HD, 800 × 1280 pixels, fixed/tiltalbe
Printing	External printer	Snap-on printer accessory	Snap-on printer accessory, USB, network or Wi-Fi to page printers
Optional specialized functionality	Computer control with VISION <i>lite</i> . Adds kinetics, more sophisticated scanning and quant, plus PV capabilities		Solution color with VISION <i>lite</i> ColorCalc Software
Validation IQ/OQ	No		Yes
Capacity	single cell		Standard: single cell Optional: 8 or 4 cell (140 only)
Liquid samples – cuvettes		Rectangular or cylindrical to 100 mm	
Liquid samples – test tubes	13 mm × 100 mm	12–25 mm dia, up to 150 mm tall	12–25 mm dia, up to 150 mm tall (40 only) N/A (140 only)
Temperature control	N/A	Water thermostatted single cell holder	Water thermostatted single-cell holder (40 only) Peltier thermostatted cell holder (140 only)
Solid samples	N/A	Thin solid samples such as films and filters	
Warranty	1 year	2 year (plus 1 additional year with registration)	
Benefits and competitive advantage	Very simple operation  Auto-zero at all wavelengths will save time, reduce error  Live measurements every 2 seconds  Scan 400–900 nm in 10 seconds  Quant with 4 standards	Larger, removable sample compartment     Large, color screen     Familiar keypad controls     Export data files to USB     Print data to snap-on thermal printer	T'', HD color touchscreen  Easy-to-use user interface  Large sample compartment with front access simplifies sample handling  Network equipped – print or store data to a USB stick or a network  Go wireless – optional Wi-Fi adaptor

### Featured accessories for SPECTRONIC 200



100 mm Rectangular Cell Holder 840-250500



Test Tube Holder 840-250700

#### Featured accessories for GENESYS 30/40



Test-Tube Holder 840-277500



Tall Test-Tube Cap 840-277600



Long Path Rectangular Cell Holder 840-277700



5-100 mm Path Cylindrical Cell Holder 840-277800



Film/Filter Holder 840-278100



Thermostat
Rectangular Cell Holder
(no baseplate)
335079-000

#### Featured accessories for GENESYS 140



8 Position Carousel for 10 mm Pathlength Cells 840-303400



Peltier Thermostatted Single Cell Holder 840-306600



Integrated Sipper System 840-306900



4 × 10–50 mm Rectangular Cell Holder

840-314700



Long Path
Rectangular Cell
Holder
840-303800



5-100 mm Path Cylindrical Cell Holder 840-303100



Film/Filter Holder 840-303200

## Routine QC and research

Instrument	GENESYS 50/150	GENESYS 180	
Catalog numbers	GENESYS 50: 840-298000 GENESYS 150: 840-300000	840-309000	
Spectrum	UV-V	risible	
Spectral range	190–1	100 nm	
Туре	Dual beam Double beam		
Bandwidth	2.0 nm		
Lamp(s)	Xenon		
Detector	Dual Silicon Photodiodes		
Standard functions	A/%T/concentration/scan/rate/Quant, fixed with equations		
User interface	On-board touchscreen or computer control		
Method/data storage	On-board and Computer		
Display	7" color touchscreen, HD, 800 × 1280 pixels, tiltable (GEN 150/180 only)		
Printing	USB, network and wireless printing to page printers. Snap-on thermal printer option.		
Optional specialized functionality	Solution color with VISION lite ColorCalc Software Enymatic analysis with VISION lite EnzLab Software Wine and juice analysis with VISION lite Wine Software		
Validation IQ/OQ	Yes		
Capacity	Single Cell (standard), Multi-cell up to 8 (optional—GENESYS 150 only)	Single Cell and Multi-cell up to 8 (standard)	
Liquid samples	Up to 100 r	nm cuvettes	
Temperature control	Single cell Peltier thermostatted cell holder (GEN 150/180 only)		
Solid samples	Thin solid samples su	uch as films and filters	
Warranty	2 years (plus 1 addition	nal year with registration)	
Benefits and competitive advantage	Tiltable 7" (GENESYS 150 only), HD color touchscreen tablet to avoid glare  Compatible with automated cell changers, Peltier, sipper and fiber optic probe accessory options (GENESYS 150 only)  Network equipped — print or store data to a USB stick or a network	Double-beam optics     Ideal solution when a reference beam is required, such as for kinetics	
		Make measurements outside the sample compartment and without cuvettes using fiber optic coupler and probe	
			Go wireless – optional Wi-Fi adaptor

### Featured accessories for GENESYS 40/50



**Test Tube Holder** 840-277500



**Tall Test Tube Cap** 840-277600



**Long Path Rectangular Cell Holder** 840-277700



**Printer** 840-278000



**Long Path Cylindrical Cell Holder** 840-277800



Film Filter Holder 840-278100

### Featured accessories for GENESYS 140/150/180



8 Position Carousel for 10 mm Pathlength Cells 840-303400



Wi-Fi Adaptor for **GENESYS 40-180** 840-309900



**Long Path Rectangular Cell Holder** 840-303800



Film/Filter Holder 840-278100



5-100 mm Path **Cylindrical Cell Holder** 840-303100



Printer 840-278000



Integrated Sipper System 840-306900



4 × 10-50 mm Rectangular Cell Holder 840-314700

# Advanced QC, research and pharma

Instrument	Evolution One	Evolution One Plus	Evolution Pro
Catalog Numbers	840-341400	840-341500	840-340200
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Spectrum		UV-Visible	
Spectral range		190–1100 nm	
Туре		Double beam	
Bandwidth	1.0 nm	1.0 nm, 2.0 nm & AFBG	0.5, 1.0, 1.5, 2.0 & 4.0 nm
Lamp(s)		Xenon	
Detector	Dual silicon photodiodes		
Standard functions	A/%T/Quant/fixed wavelength with advanced calculations/scan/rate/performance verification		
User interface	Computer control		
Method/data storage	Computer control		
Printing	Windows® compatible printers		
Optional specialized functionality	21 CFR Part 11, customized Analyzer, color, biochemical methods including DNA melting, autosampler		
Validation IQ/OQ	Yes		
Capacity		Multi-cell (up to 8)	
Liquid samples	Up to	100 mm cuvettes; Up to 40 mm tubes and	l vials
Temperature control	Peltier accessories and a range of thermostatted cell holders with liquid recirculators		
Solid samples	Thin solid samples	Thin and thick solids, powders, reflecting surfaces, films, DRA, SRA	Thin and thick solids, specular surfaces and films, SRA
Warranty		2 years	
Benefits and competitive advantage	<ul> <li>Automated performance verification with CVC accessory</li> <li>CUE software scripting capability to simplify complex assays with the touch of a button</li> <li>Powered by Insight Pro Software</li> <li>Smart Accessories™ with unique serial number recorded for comprehensive event logs and audit trails</li> <li>Specialized, easy-to-use accessories for increased productivity</li> <li>Xenon lamp requires no warm-up for instant measurements and has 3-year warranty</li> <li>0.5 nm bandwidth for highest resolution data (Evolution Pro only)</li> <li>Application Focused Beam Geometry (AFBG) matches hardware to your application (Evolution One Plus only)</li> <li>Double-beam configuration (Large sample compartment and 21 cm beam separation option available with Evolution Pro only)</li> </ul>		

### Featured accessories for Evolution One Spectrophotometers



8-Cell Peltier System 699-131200



Single Cell Peltier System

Available soon



Combination Filter, Vial & Square 10 mm Cell Holder 840-211800



100 mm Rectangular Cell Holder



Smart Thermostatted Rotary 7-Cell Changer 840-214900



Smart Thermostatted Linear 8-Cell Changer 840-219000



Standard Fiber
Optic Dip Probe with
10 mm Tip
840-305000



Temperature Probe Hub and Temperature Probes 840-214600



ISA-220 Integrating Sphere 222-269400



Powder Cell Holder 840-289300

## Life science

Instrument	GENESYS 30	BioMate 160	Evolution One
Catalog Numbers	840-277000	840-301000	840-341400
		THE SECOND	
Spectrum	Visible	UV-Visible	
Spectral range	325-1100 nm	190–1100 nm	
Туре	Single Beam	Dual Beam	Double Beam
Bandwidth	5.0 nm	2.0 nm	1.0 nm
Lamp(s)	Tungsten-Halogen	Xei	non
Detector	Silicon Photodiode	Dual Silicon Photodiodes	
Standard functions	A/%T/concentration/scan/OD600/ fixed with eqns/Quant	A/%T/concentration/scan/0D600/ fixed with eqns/Quant/rate/DNA & protein methods	A/%T/concentration/scan/rate/ temperature kinetics/DNA meltin
User interface	On-board keyboard and screen or computer	On-board touchscreen, tablet or computer Control	Computer control
Method/data storage	On-board: Methods USB: Method and data Computer: Method and data	On-board: Method and data Computer: Method and data	Computer control
Display	5" diagonal, 32-bit color display, 800 × 480 pixels	7" color touchscreen, HD, 800 × 1280 pixels, tiltable	N/A
Printing	Optional GENESYS printer only	Optional GENESYS printer, page printers by USB, network or Wi-Fi (with optional transmitter)	Windows printers
Optional specialized functionality	Optional accessory for measuring test tubes and nephelo flasks	Optional: Beckman cell holder, 50 µL disposable cell holder, 8 position cell holder (not thermostatted), Peltier thermostatted single cell holder (20–60 °C)	Temperature ramping with Peltie thermostatted cell holders Advanced measurement automation with thermostatted multiple cell holders
Validation IQ/OQ	No	Yes	Yes
Capacity	Single cell	Single Cell (standard), multi-cell up to 8 (optional)	
Liquid samples	Up to 100 mm cuvettes; Up to 25 mm test tubes	Up to 100 mm cuvettes	
Temperature control	Optional water thermostatted single cell holder	Optional Peltier thermostatted single cell holder (20–60 °C)	Peltier accessories and a range of thermostatted cell holders with liquid recirculators
Warranty	2 year (plus 1 additional year with registration)		2 years
Benefits and competitive advantage	Easy to learn, easy to use interface with 5" high resolution color screen and tactile rubber keypad     Large sample compartment with front access simplifies sample handling     Best choice for OD600 measurements	Tiltable 7", high-resolution touch-screen tablet to avoid glare  Compatible with automated cell changers, Peltier, sipper and fiber optic probe accessory options  Network equipped – print or store data to a USB stick or a network  Go wireless – optional Wi-Fi adaptor	Double-beam optics Ideal solution when a reference beam is required, such as for kinetics Widest range of accessory option Best option for temperature control Temperature kinetics and DNA melting with single or 8-cel

### Featured accessories for GENESYS 30



**Test Tube Holder** 840-277500



**Tall Test Tube Cap** 840-277600



**GENESYS Printer Accessory** 840-278000

#### Featured accessories for BioMate 160



**Beckman Style Cell** Holder 840-299000



**8 Position Carousel** for 10 mm **Pathlength Cells** 840-303400



Microcell Holder 840-303300



**Peltier Thermostatted** Single-Cell Holder



840-306600



**Printer Accessory** 840-278000

### **Featured accessories for Evolution One**



8-Cell Peltier System 699-131200



Single Cell Peltier **System** Available soon



**Temperature Probe Hub and Temperature Probes** 840-214600

## Life science

2110 20101100				
Instrument	NanoDrop Eight	NanoDrop Ultra/Ultra <sup>c</sup> / Ultra FL/Ultra <sup>c</sup> FL	NanoDrop Lite Plus	
Catalog Numbers	NDE-GL	Uitra: NDULTRAGL Uitra <sup>c</sup> : NDULTRACGL Uitra FL: NDULTRAFLGL Uitra <sup>c</sup> FL: NDULTRACFLGL	No Printer: NDLPLUSGL Printer: NDLPLUSPRGL	
Spectrum	UV-V	UV-Visible		
Spectral range	190-850 nm (scans)	190-850 nm (scans)	Fixed: 230 nm, 260 nm, 280 nm	
Bandwidth	≤1.8 nm			
Lamp(s)	Xenon			
Detector	2048-element linear silicon CCD array			
Standard functions	DNA, RNA, protein A280 and A205, colorimetric protein assays, proteins and labels	DNA, RNA, protein A280 and A205, proteins and labels. Ultra <sup>c</sup> and Ultra <sup>c</sup> FL Only: Kinetics Ultra FL and Ultra <sup>c</sup> FL Only: DNA Fluorescence and RNA Fluorescence	DNA, RNA and purified A280 protein measurements	
User interface	Computer software	Built-in touchscreen or computer software	Built-in touchscreen	
Method/data storage	Computer	On-board, Wi-Fi, PC and USB transferable	On-board and USB transferable	
Display	N/A	10.1" color touchscreen, tiltable	LCD touchscreen	
Printing	Computer	Print from on-board control via Wi-Fi, Ethernet or USB connection	Optional label printer	
Optional specialized functionality	Software for 21 CFR Part 11 compliance	- SciVault 2 Software for 21 CFR Part 11 compliance - Acclaro Pro Software for exceptional accuracy	N/A	
Validation IQ/OQ	Yes		No	
Capacity	Microvolume (up to 8 samples at a time)	Microvolume and cuvette	Microvolume	
Liquid samples	1–2 μL	1–2 µL samples and 10 mm cuvette (Ultra <sup>c</sup> and Ultra <sup>c</sup> FL)	1–2 µL	
Temperature control	N/A	Cuvette at 37 °C (Ultrac and Ultrac FL)	N/A	
Warranty	1 year	2 years	1 year	
Benefits and advantage	High-sample throughput (measures 8 samples simultaneously)     1-2 uL of sample     Sample contaminant identification     Differentiates mammalian DNA and RNA     Optional 21 CFR Part 11 software     Data output for LIMS integration	Stand-alone instrument with on-board control  10.1" high resolution touchscreen interface  Contaminant identification  Differentiates mammalian, plant, and bacterial DNA and RNA  Integrated qPCR recipe calculator  LIMS API Integration  Only 1–2 µL of sample  Computer, Wi-Fi, USB, Cloud Data Transfer	Most popular methods including dsDNA, RNA, and Protein A280     Purity ratios A260/A280 & A260/A230     Last 1000 measurements saved locally; export data to a USB device     Optional printer available for freezer compatible labels	
		Guided troubleshooting		

· Diagnostic Scheduler

# Featured accessories and software for NanoDrop Ultra Spectrophotometers and Fluorometers



SciVault 2 Software
NDU-SCIVAULT2



DYMO 550 Printer
DYMO-PRNTR-ND



PV-1 Performance Verification Solution CHEM-PV-1



NanoDrop Ultra
Instrument Dust Cover
NDU-INST-COVER



Cuvette Qualification Filter Kit for NanoDrop Instruments

840-257300

### Featured accessories and software for NanoDrop Eight



SciVault™ Software 840-371100



PV-8 Performance Verification Kit

### Other accessories



PR-1 Reconditioning Kit for NanoDrop Instruments CHEM-PR1-KIT



NanoDrop Lite Plus
Printer

NDLP-PRNTR-MOD





Learn more at thermofisher.com/uv-vis

thermo scientific