# <u>Thermo Scientific</u> Dionex UltiMate 3000 Variable Wavelength Detectors

Integrating hardware, software and separation chemistry, we offer UHPLC to everyone—for all needs—establishing the new standard in conventional LC.

The Thermo Scientific<sup>™</sup> Dionex<sup>™</sup> UltiMate<sup>™</sup> 3000 VWD-3000 is a variable wavelength detector (VWD) series for industry leading UV-vis detection. The forward optics design and wide range of available flow cells ensure optimal performance over a flow rate range of five orders of magnitude. Automated qualification, performance optimization, and instrument wellness monitoring deliver maximum uptime, simplify work-flow, and give you full confidence in your analytical results. The detector is available in a standard 100 Hz (VWD-3100) and a 200 Hz Rapid Separation version (VWD-3400RS) for the most challenging UHPLC applications.



# **High-Performance UV-Vis Detection**

- The VWD-3400RS variant provides data collection rates of up to 200 Hz for optimal support of today's and tomorrow's UHPLC separations.
- The VWD-3100 standard detector operates at up to 100 Hz data rate for optimum support of 62 MPa (9000 psi) Thermo Scientific<sup>™</sup> Dionex<sup>™</sup> UltiMate<sup>™</sup> 3000 Standard systems.
- Superior detection of trace analytes with low noise (< ±2.0 µAU) and drift (< 100 µAU/h).</li>
- The detector's large linearity range of up to 2.5 AU is ideal for applications with widely varying analyte concentrations.
- Up to four absorption channels (VWD-3400RS) and spectral scans support effective method development.

- Active temperature control of optics and electronics for data acquisition independent of ambient conditions.
- Front panel access for quick and easy lamps and flow cells changes.
- Automated qualification monitoring for full regulatory compliance.
- Large front panel display for monitoring the detector status even from a distance.
- Maximize uptime using predictive performance—based on monitoring the life cycle of detector lamps.
- The detector can be upgraded with the Thermo Scientific<sup>™</sup> Dionex<sup>™</sup> pH/Conductivity Monitor (PCM-3000) for accurate and precise pH- and conductivity monitoring.
- Unique 45 nL ultra-low dispersion UV monitor for dispersion-free UV detection in LC/MS.



VWD-3000 SERIES UV-VIS DETECTORS FEATURES AND HIGHLIGHTS	THEIR BENEFIT TO YOU
Outstanding linear calibration curves (correlation coefficient higher than 0.9997) over a wide absorbance range (up to 2.5 AU in the UV range).	High quantification accuracy over a wide concentration range enables the accurate detection and integration of very small peaks in the presence of high absorption peaks. No need to re-inject at a different dilution factor.
Outstanding baseline noise of less than $< \pm 2.0 \mu$ AU (230 nm, time constant 2 s, dry analytical flow cell).	Low limits of detection/quantification for superior accuracy in trace level analysis. Saves effort for sample enrichment and broadens applicability, e.g., in environmental analysis.
Data collection rates of up to 200 Hz.	Superior time-resolution for accurate integration—ready for today's and tomorrow's UHPLC detection challenges.
Dedicated reference wavelength photodiode and active lamp house temperature control minimize baseline drift to below 100 $\mu\text{AU/h}.$	Accurate peak integration for methods with long run times and substances eluting over a wide time range. High quantification accuracy even in laboratory environments without temperature control.
Wavelength range of 190–900 nm with combined use of deuterium and tungsten lamp on one optical axis.	Sensitive detection in the UV, visible, and near-infrared range.
Up to four absorption channels (VWD-3400RS) and scans across the entire wavelength range.	Easy identification of optimum absorbance wavelength without diode array technology.
Fast switching second-order filter for multiple wavelength operation in UV and visible range in the same chromatographic run.	Unlimited multiple wavelength detection over the complete spectral range for superior detection selectivity and sensitivity on all peaks in your chromatogram.
Tool-free front panel access to flow cell and lamps.	Easy access to detector fluidics and to wear parts.
Large flow cell portfolio for nano to semiprep flows (fused silica, stainless steel, or PEEK <sup>™</sup> )	Highly flexible for any individual application requirement.
Lamps and flow cells equipped with ID chips for identification (e.g., type and serial number) and lifespan monitoring.	History of wear parts is traceable even when the flow cell or lamp is placed in another VWD-3000 series module.
Thermo Scientific <sup>™</sup> Dionex <sup>™</sup> Chromeleon <sup>™</sup> software predicts the anticipated remaining lifespan for wear parts and automatically alerts the user when to change wear parts.	Enables users to plan ahead for required maintenance. Wear parts do not fail unexpectedly, which increases overall productivity.
Automated wavelength verification with internal holmium filter.	The detector ensures high wavelength accuracy and precision, as well as minimal manual effort for system qualification.
PCM-3000 upgrade for pH- and conductivity monitoring	The combination of UV-Vis detector and PCM-3000 is ideal for monitoring salt- or pH-gradient separations of monoclonal antibodies or other biomolecules.
Ultra-low dispersion UV monitoring for LC/MS applications	Obtain qualitative UV absorption data without sacrificing the chromatographic resolution of the quantitative measurement of the mass spectrometer.

Table 1. Eight flow cell variants to cover the full range from nano to semi-preparative applications.

Description	Cell Volume	Light Path Length (mm)	Maximum Pressure (MPa)
Standard (SST)	11 µL	10	12
Standard (PEEK)	11 µL	10	5
Semi-micro (SST)	2.5 µL	7	12
Semi-micro (PEEK)	2.5 µL	7	5
Ultra-low disperion UV Monitor	45 nL	10	30
Semi-preparative (PEEK)	0.7 µL	0.4	10
UZ-View <sup>™</sup> Nano (Fused Silica)	3 nL	10	20
UZ-View Cap (Fused Silica)	45 nL	10	20
UZ-View Micro (Fused Silica)	180 nL	10	20

### **Optical Design**

The VWD-3000 series uses a fast-turning grating monochromator placed in front of the flow cell (forward optics design). Shown in Figure 1, this design ensures that only a narrow band of wavelengths passes the flow cell, controlled by the optical slit. Unlike photodiode array detectors with polychromatic light passing through the flow cell, VWD-3000 detectors minimize all possible interferences and stray light effects in the flow cell. The result: excellent baseline noise performance and a wide linear and dynamic range. The baseline noise and drift are further reduced through the use of a reference diode.

### Ergonomic Design and Safety Features

The VWD-3000 is designed for maximum ease of use and safety. Flow cell connections and internal fluidics are physically separated from electronic parts. In the case of a leak, all liquid is safely drained to a leak sensor which can trigger a stop of the current sample queue or even a customized shut down program. UV and vis lamps are located behind a safety cover. Removing these lamps requires no tools, and is therefore equally easy and fast. All changes are automatically tracked by integrated ID chips for the cell and lamp. The relevant information is stored in the module firmware and moves with the detector if it is transferred to another LC system.

### **Meet Your Specific Requirements**

Select from nine different flow cells based on your column dimensions and applicationrelated requirements (Table 1). The VWD-3000 series offers a unique 3 nL cell for nano LC, a 45 nL cell for capillary LC, and a 180 nL cell for the lower end of micro LC (< 1 mm internal column diameter). These flow cells have a fused silica flow path and are fully biocompatible. The 2.5 µL semi-micro flow cell and the 11 µL analytical flow cells are available in stainless steel for optimal robustness and pressure resistance and in PEEK for applications requiring metalfree hardware. The unique 45 nL ultra-low dispersion monitor is designed to support LC/MS applications. Using this cell provides the best resolution of ultra-narrow peaks for both the absorption detection and a subsequent mass spectrometer. A dedicated semi-preparative flow cell with 0.4 mm optical light path (PEEK) completes the range of options.



Figure 1. Schematic of the optical design of the VWD-3000 detector series.



Figure 2. The ergonomic internal front design: 1) lamp safety cover, 2) flow cell, 3) cell leak drainage, and 4) leak sensor.

# **Rapid Separation LC Detection**

Depending on the required accuracy and precision, a peak should consist of 20–30 data points to guarantee accurate determination of area and retention time. Already today, ultrafast chromatographic methods generate peak widths of less than 500 milliseconds and therefore the need for more than 50 Hz data collection rate. The Rapid Separation variable wavelength detector can not only fulfill this requirement; its maximum data collection rate of 200 Hz is even prepared to meet any future detection challenge.

### Full UHPLC Compatibility Beyond Data Collection Rate

The precise and fast-tuning monochromator is the VWD-3000 Series' backbone for full UHPLC compatibility. If your application requires a wavelength switch, only a marginal baseline resolution is required as it is always done in less than 250 milliseconds.

If you want to optimize the absorption wavelength of a UHPLC method, the detector design and Chromeleon software control are a powerful combination. Customizable Chromeleon trigger commands execute scans based on real peak detection, not just on an entered time. This assures that a scan is conducted at the right time, no matter how narrow the target peak is. Depending on the requirements, users can perform high-, medium-, or low-resolution scans. With the latter setting, scans can even be conducted during continuous-flow UHPLC separations.

For short-term use, the VWD-3400RS can also operate in a multiple wavelength mode. For any data point, the monochromator quickly changes between up to four wavelength channels. Even the simultaneous operation in the UV and vis range is possible at highest quality standards with the two fast switching second order filters for improved signal to noise. The detector can achieve a maximum data rate of up to 5 Hz which typically provides enough data for sub-5 min screening separations (Figure 5).



Figure 3. The VWD-3400RS is suited for any UHPLC application with up to 200 Hz data collection rate.



Figure 4. Multiple stop-flow scans in a single run cause shifting peak retention times. Superior Chromeleon software control executes scans where a peak is identified and can therefore easily handle varying retention times.



Figure 5. Up to four wavelengths can be recorded simultaneously with up to 5 Hz data collection rate, making the four-channel mode compatible with even fast chromatography.

# Superior Linearity and Minimal Noise

Figure 6 shows the chromatogram of a drug assay with detail of the baseline to reveal related impurities. The detector linearity is calculated from a calibration curve of the active ingredient, yielding an RSD of 0.27% and r<sup>2</sup> of 0.99999 up to a peak height of 3000 mAU. Outstanding baseline noise performance of the detector enables quantitative analysis of peaks in the lower µAU range. Figure 7 shows the detection of one of the impurity peaks from the pharmaceutical formulation at different dilutions. A signal-to-noise ratio greater than 15 is achieved even at the lowest concentration (approximately 200 µAU peak height). This enables quantification of impurities at 0.007% of the main peak, as confirmed by the calibration curve in Figure 7.

This demonstrates that the combination of excellent signal linearity and baseline noise of the VWD-3000 series easily supports drug assay and impurity profiling in a single run.



# Accurate pH and Conductivity Monitoring

The Thermo Scientific Dionex PCM-3000 is an upgrade of the VWD-3000 series for accurate on-line pH and conductivity monitoring. The combination of UV-vis with the PCM-3000 is ideal for pH gradient ion exchange chromatography of compounds such as monoclonal antibodies and charge variants, conductivity monitoring in gradient separations of biomolecules, or any other application where conductivity or pH monitoring is of interest. Figure 8. The PCM-3000 is a high performance, easy-to-use, and cost effective solution for pH and conductivity monitoring.

# **LC/MS UV Monitoring**

The 45 nL ultra-low dispersion UV Monitor is designed for use with the UltiMate 3000 XRS system in combination with MS detection. In this combination, the intended use of the monitor is a qualitative characterisation while the mass spectrometer is used for quantification. Figure 9 demonstrates that this can be done in-line without loss of resolution. The overlay shows two isocratic UV chromatograms, one obtained with UV monitor before the detecting cell, one without. The plate count of all peaks remains constant.







Figure 7. Impurity profiling in a pharmaceutical formulation (from the same injection as shown in Figure 6).



Figure 9. Overlay of two isocratic UV chromatograms, one obtained with the 45 nL UV monitor before the detecting cell (blue), one without (black). The plate count of all peaks remains constant.

#### **Reliability and Ease of Use**

Reliability of scientific instruments mainly reflects a high quality standard of materials, excellent technical design, and strictly controlled manufacturing processes. However, instruments include wear parts that may fail at inopportune times. Figure 10 shows how Chromeleon software supports the user by predicting an estimated remaining lifetime of a detector lamp, based on monitoring characteristic criteria from the day of of installation. This combination of instrumentation and software significantly improves instrument uptime.

# Dionex AutoQ Equipment Qualification

Thermo Scientific<sup>™</sup> Dionex<sup>™</sup> AutoQ<sup>™</sup> Equipment Qualification routines turn tedious qualification work into a simple task. The Chromeleon CDS automatically operates the following test protocols:

- Installation Qualification
- Operational Qualification
- Performance Qualification

Upon completion, the software creates comprehensive reports, including passed/ failed checks and charts (Figure 11).

#### **Monitoring the Qualification Status**

After passing a qualification test, just hit the 'Update Qualification' button on the instrument panel and define the qualification interval (Figure 12). Chromeleon software ensures compliance by reminding you to requalify the instrument and prohibiting the use of it if the end of the grace period is reached.

Wellness: Detec	tor VWD-3400RS				
Instrument: Ultik	late 297				
manual char					
Detector Model	VwD-3400	Power On Time:	31.25	Serial No. Flowcell:	043352/006
Serial No.:	800250	Operating Hours:	7250	Serial No. Mainboard:	00000040FA54
Firmware Version:	3.14			Serial No. Optics:	0000003F1D6A
JV Lamp Details		UV Lamp	Operation Time	UV Lamp Relat	ive Intensity
Last Changed:	2012-08-31	Wellness	Gauge	Wellness Gauge	
Manufacturer:	Hamamatsu	🎽 🖂		a 📲 🖬 🖬 🖬 🖓	• <b>•••••</b> ••••••••••••••••••••••••••••••
Serial No:	AD2687				
Ignitions:	37	Warning Li	mit: 2400 hours	😂 Warning Limit:	60 %
Current Trials:	1	Limit	2800 hours	Limit	50 %
Failures:	0	VIS Lamp	Operation Time	VIS Lamp Rela	tive Intensity
		Wellness	Gauge	Wellness Gauge	
VIS Lamp Details Last Changed:	2012-08-31		0		
Manufacturer:	Fischer				
Serial No:	149648771	Warning Li	mit: 2400 hours	Warning Limit:	60 %
		Limit:	2800 hours	😌 Limit:	50 %

Figure 10. Wellness data and lifetime prediction of wear parts.



Figure 11. Detector linearity operational qualification report for the VWD-3000 Series.

Detector - Qualification		
Qualification: Detector VWD-3400RS		
Instrument: C-GEPM07_1		
Wellness Gauge		
× 1 · 1 · 1 · 1 · 1 · 1 · 1 · 1 · 1 · 1		
Last Qualified On:	2012-02-14	
Last Qualified By:	Pran	
Qualification Interval:	180 d	▲ ▼
Warning Period:	7 d	
Grace Period:	14 d	
	Updat	e Qualification
	<u></u>	

Figure 12. Chromeleon software monitors the module qualification status.

# Connection to Other Systems for Data Recording

A two-channel analog output card is available as an option for the VWD-3000 series. The installation of it is plug-and-play (Figure 13). With no need to dismantle the HPLC system, the card easily slides into the VWD from the back of the module. Both analog outputs provide a resolution of 20 bits and the analog output voltages are updated with a data rate of 50 Hz for UHPLC compatibility even when connected to external data recording devices.



Figure 13. Using standard tools, the optional D/A converter can be installed at the instrument's rear panel in just a few minutes.

SPECIFICATIONS		
Data collection rate:	Single wavelength up to 100 Hz (VWD-3100) Single wavelength up to 200 Hz (VWD-3400RS) Multiple wavelength up to 5 Hz (VWD-3400RS)	
Noise, single wavelength:	$<\pm2.0~\mu$ AU (typical) at 230 nm, time constant 2 s, dry analytical flow cell $<\pm3.5~\mu$ AU (typical $<\pm2.5~\mu$ AU) at 254 nm, time constant 1 s, dry analytical flow cell	
Noise, multiple wavelength (VWD-3400RS):	$<\pm10$ µAU (typical $<\pm7$ µAU) at 254 nm and 280 nm, 2 s time constant, dry analytical flow cell	
Drift:	$< 10^{\text{-4}}$ AU/h at 254 nm and dry analytical flow cell	
Linearity:	< 5% RSD (typical $< 3%$ RSD) at 2.5 AU caffeine, wavelength: 272 nm based on ASTM	
Light source:	Deuterium lamp, tungsten lamp (Tungsten lamp optional on VWD-3100) Temperature control for both lamps	
Wavelength range:	190–900 nm The tungsten lamp is recommended for wavelengths $> 600$ nm	
Wavelength accuracy:	±1 nm (over detector lifetime)	
Wavelength repeatability:	±0.1 nm	
Optical bandwidth:	6 nm at 254 nm	
I/O interfaces:	Four digital inputs, four digital outputs	
Analog output:	Two analog outputs available as an option via DAC plug-in module Software selectable: absorbance, 20-bit resolution, 0–1 V (full range) and 0–10 V (full range with adjustable mAU ranges).	
Safety feature:	Power-up diagnostics of optics, cooling fans, motors, and electronics leak sensor	
Power consumption:	85–260 V AC, 50/60 Hz, max. 150 W Wide range (automatic voltage selection)	
Wetted parts:	PEEK, quartz glass, stainless steel	
Dimensions (h $\times$ w $\times$ d):	$16 \times 42 \times 51$ cm (6.3 × 16.5 × 20 in.)	

# **Ordering Information**

In the U.S., call (800) 346-6390 or contact the Thermo Fisher Scientific Regional Office nearest you. Outside the U.S., order through your local Thermo Fisher Scientific office or distributor. Refer to the following part numbers:

Detectors	Part Number
VWD-3100 Variable Wavelength Detector (without flow cell)	5074.0005
VWD-3400RS Rapid Separation Variable Wavelength Detector (without flow cell)	5074.0010
Accessories	Part Number
Analytical flow cell, 11 µL, stainless steel	6074.0250
Analytical flow cell, 11 µL, PEEK	6074.0200
Semi-micro flow cell, 2.5 µL, stainless steel	6074.0360
Semi-micro flow cell, 2.5 µL, PEEK	6074.0300
Ultra-low disperion UV Monitor, 45 nL	6074.0285
Semi-preparative flow cell, 0.7 µL, PEEK	6074.0320
Nano flow cell, UZ-View, 3 nL	6074.0270
Capillary flow cell, UZ-View, 45 nL	6074.0280
Micro flow cell, UZ-View, 180 nL	6074.0290
Deuterium lamp	6074.1110
Tungsten lamp	6074.2000
Analog output card	6074.0305
PCM-3000 pH and Conductivity Monitor for VWD-3000 Series (Shipped with pH and conductivity flow cells and a pH electrode)	6082.2005

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