

# Thermo Scientific Dionex UltiMate 3000 Fluorescence Detectors

Sensitive detection with full UHPLC compatibility

Thermo Scientific™ Dionex™ UltiMate™ 3000 products are UHPLC compatible by design, establishing the new standard in conventional LC. Integrating hardware, software, and separation chemistry, we offer UHPLC to everyone, for all needs.

The Thermo Scientific Dionex UltiMate 3000 FLD-3000 is a high-sensitivity fluorescence detector series for UltiMate 3000 HPLC systems. It is available in Rapid Separation (RS) and Standard (SD) versions. The optics of the FLD-3000 series provide maximum stray-light suppression for best detection sensitivity. Operated with the Thermo Scientific™ Dionex™ Chromeleon™ Chromatography Data System (CDS) software, the detector provides automated qualification, various tools for method development, and instrument wellness monitoring for ease of use, maximum uptime, and the highest degree of regulatory compliance.



- Data collection at up to 200 Hz for optimal support of even the fastest UHPLC separations (FLD-3400RS).
- Standard detectors operate at up to 100 Hz data rate for optimum support of 62 MPa (9,000 psi) UltiMate 3000 standard systems.
- Lowest limits of detection with a Raman signal-to-noise ratio (S/N): > 550 ASTM (> 2100 using dark signal as noise reference).
- Unsurpassed reproducibility with active flow cell temperature control for stable fluorophore activity independent of changes in ambient temperature.
- Long-life xenon flash lamp for highest sensitivity and long-term operation without the need for frequent lamp changing.
- Optional second photomultiplier (PMT) for unique Dual-PMT operation, offering an extended wavelength range up to 900 nm without sacrificing sensitivity in the standard wavelength range.
- Two-dimensional (2D) or three-dimensional (3D) excitation, emission, or synchro scans to provide the highest degree of flexibility for method development or routine sample characterization.
- Innovative Variable Emission Filter for real-time compound-related sensitivity optimization (FLD-3400RS only).
- Large front-panel display for easy monitoring of the detector status.
- Two flow-cell sizes for easy optimization to application requirements: the 8  $\mu$ L flow cell is ideal for trace analysis, and the 2  $\mu$ L flow cell offers best peak resolution with narrow-bore HPLC and UHPLC columns.

FLD-3000 Series Features	FLD-3000 Series Benefits
Innovative optical design for maximum suppression of stray light	Allows unmatched sensitivity for lowest detection limits.
Unique fast-switching Variable Emission Filter for stray light reduction in front of the emission monochromator	Generates increased detection sensitivity by adapting the filter setting to the selected emission wavelength or by recording all light emitted with wavelengths higher than the selected cutoff filter in a single data channel (FLD-3400RS systems only).
Unique Dual-PMT option	Combines flexibility with sensitivity and maximizes the wavelength range up to 900 nm without sacrificing sensitivity in the UV-vis spectral range.
Data collection rate of up to 200 Hz in single-channel mode (FLD-3400RS)	Provides full support of any UHPLC application. Sufficient data points even for peak widths in the subsecond range, delivering superior quantitation accuracy in ultrafast separations.
Temperature-controlled flow cells	Supplies unsurpassed reproducibility in run-to-run and day-to-day routine operation, even at fluctuating column or ambient temperature.
High-intensity xenon flash lamp with smart control features	Provides significantly longer lamp life (> 15,000 h in long life mode) than conventional, continuous xenon arc lamps. In addition, the lamp only consumes 10% of the power of a conventional lamp.
2D and 3D scans of emission and excitation wavelengths	Allows fast identification of the optimum emission and excitation wavelength settings for your application.
8 $\mu$ L analytical flow cell 2 $\mu$ L micro flow cell	The 8 $\mu$ L analytical flow cell is the best choice for trace detection in combination with 2 mm to 4.6 mm i.d. UHPLC columns. The 2 $\mu$ L micro flow cell provides the best chromatographic efficiency and resolution with 2 mm i.d. columns. All relevant flow cell details are tracked by the detector and added to each sample.
Flow cell syringe injection kit	Cost-effective upgrade to introduce sample into the flow cell and perform an off-line measurement. Easy option to flush the flow cell when preparing for idle times.
Multiwavelength operation (FLD-3400RS systems only) on up to four channels with independent excitation and emission wavelengths	Allows detection of analytes with different fluorophores in a single analytical run.
Internal monitoring of relevant detector components, e.g. the lamp	Allows the user to plan ahead for required maintenance to ensure correct operation.
Qualification information is stored in the module	Avoids the use of unqualified equipment, even when the module is switched to another system.

## Unmatched Performance Through Innovative Optical Design

The FLD-3000 series combines advanced optics with several unique features for extraordinary low stray light and superior S/N performance (Figure 1). The Variable Emission Filter ensures the highest possible sensitivity over the whole wavelength range. High-precision grating monochromators support fastest excitation and emission wavelength changes suitable also for multichannel operation (FLD-3400RS only). The Dual-PMT option increases the detection range up to 900 nm without the usual loss in UV sensitivity. The temperature-controlled flow cell increases the detection precision even under fluctuating ambient temperature.

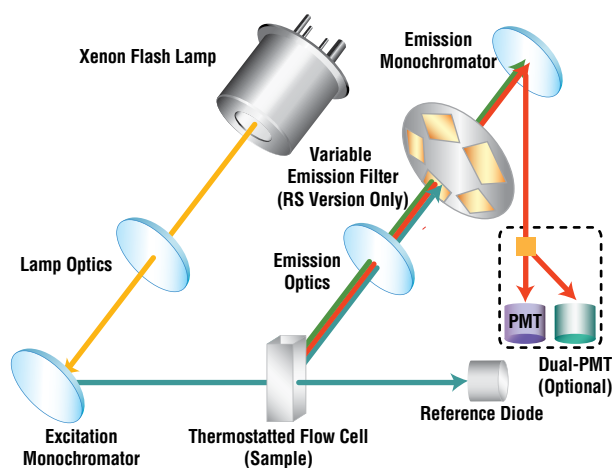


Figure 1. Schematic of the optical design of FLD-3000 series.

## A New Dimension of Stray Light Suppression

In fluorescence detection, undesired stray light significantly increases noise, and at the same time can give a false reading for the signal value. This can make calculation of the true S/N value difficult. The optics of the FLD-3000 series are highly optimized with regards to stray light suppression. This reduces noise, ensures a true reading for the signal value, and maximizes the S/N value across the entire wavelength range (Figure 2).

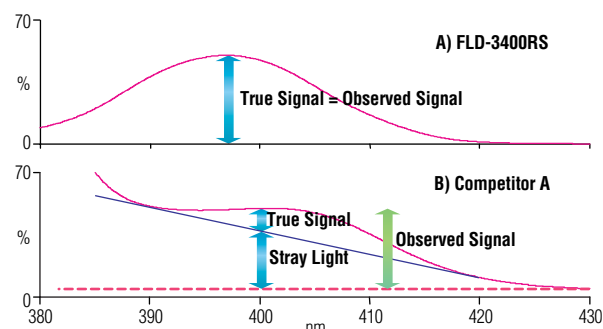


Figure 2. Raman band of water (Ex: 350 nm, Em: 397 nm): A) Raman peak clearly resolved with the FLD-3000 Series, and B) superimposed by a high level of stray light.

## Optimized Fluidics for Best UHPLC Compatibility

Both the micro and analytical flow cell variants are optimized for minimum dispersion and best peak symmetry. Figure 3 shows a separation of five polycyclic aromatic hydrocarbons (PAHs) on a Thermo Scientific™ Acclaim™ RSLC 2.1 mm × 50 mm, 3 μm column, detected with a 2 μL (micro) and an 8 μL (analytical) flow cell. Although all peaks elute within a time window of only 0.7 min and peak volumes are only 27 μL to 46 μL, even the analytical flow cell detects symmetric peaks and fully resolves them. The best S/N is obtained with the analytical flow cell. The micro flow cell achieves the best chromatographic resolution.

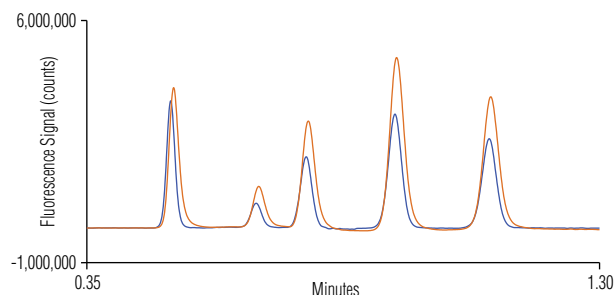
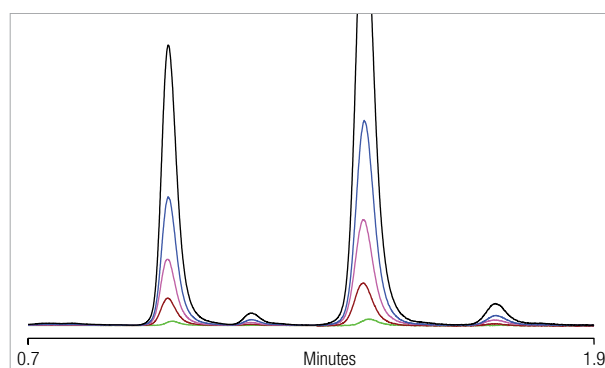


Figure 3. Sub-1.2 min separation of five PAHs on a 2.1 × 50 mm, 3 μm column using an analytical (orange) and a micro (blue) flow cell. The analytical flow cell resolves all peaks and achieves the best S/N. The micro flow cell provides the best chromatographic resolution.

## RSLC Technology for Any Application

The FLD-3400RS is the technology leader in fluorescence detection and ideally matches the performance and flexibility of the UltiMate 3000 RS platform. Data rates up to 200 Hz, very high sensitivity, tunable emission filters, two flow-cell variants, different lamp operation modes, and detection at up to 900 nm without compromise provide more possibilities than any other fluorescence detector on the market. Run existing applications or easily develop new ultrasensitive, ultrafast applications with sub-2 min run time (Figure 4).



Standard	G2	G1	B2	B1
1	155.0	513.0	157.0	550.5
2	77.5	265.5	78.5	275.3
3	38.8	128.3	39.3	137.7
4	15.5	51.3	15.7	55.1
5	3.1	10.3	3.1	11.0

Figure 4. Overlay of five aflatoxin standard separations using an Acclaim RSLC C18 2.1 × 100 mm, 2.2 μm column (B2, B1, G2, G1 in order of elution). Table concentrations in ppb.

## Shortest Run Times Even with Excitation and Emission Switching

For the best detection sensitivity, trace compounds must be analyzed with optimum excitation and emission wavelengths. In a separation of different fluorescing analytes, it becomes necessary to switch the wavelengths between the peaks. UHPLC conditions shorten the available time window for the required grating movements significantly. The FLD-3000 series is designed with fast-turning grating monochromators which reach the new setting in less than 250 ms. Figure 5 indicates the total switching time of 350 ms under real-life conditions, including response time and software operation delay. This time includes a sensitivity change and a response time delay. This switching time is compared against a third-party detector operated under identical conditions. Another manufacturer's instrument requires 6.1 s to reach the new settings and is therefore not suitable to support UHPLC separations. In contrast, the FLD-3000 detectors are ideally suited to support very complex UHPLC separations. Figure 6 shows a 5.5 min separation of 17 fluorescing PAHs (with eight simultaneous changes of wavelength, sensitivity, and filter).

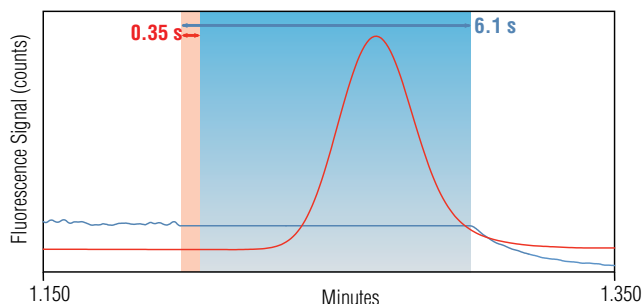


Figure 5. Data acquisition with a FLD-3000 series detector (red) and a third-party detector (blue). The FLD-3000 series detector easily completes the wavelength and sensitivity change before the peak. The third-party detector is ready after the peak has already left the flow cell.

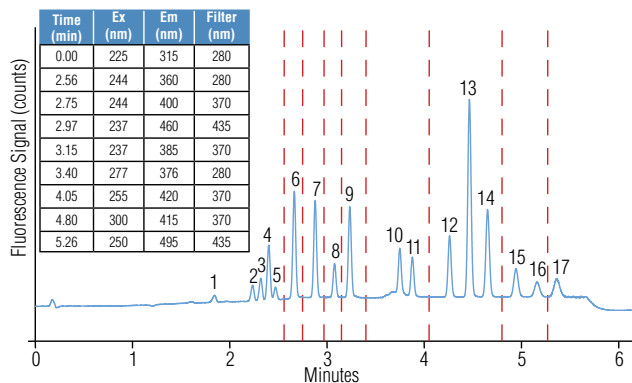


Figure 6. Fluorescence detection of 17 PAHs within 5 min using a Nucleodur C18 PAH 3 × 100 mm, 3 μm column at a flow of 2 mL/min. The detection is made possible by the fast-moving filters and monochromators of the FLD-3400RS.

## Unsurpassed Reproducibility

Fluorescence is sensitive to temperature changes. Changing temperatures will yield different emission intensities for equal fluorophore concentrations. To minimize this effect, the flow cells of the FLD-3000 series are temperature controlled. This significantly improves the precision in run-to-run and day-to-day routine operation, even at fluctuating ambient temperatures (Figure 7).

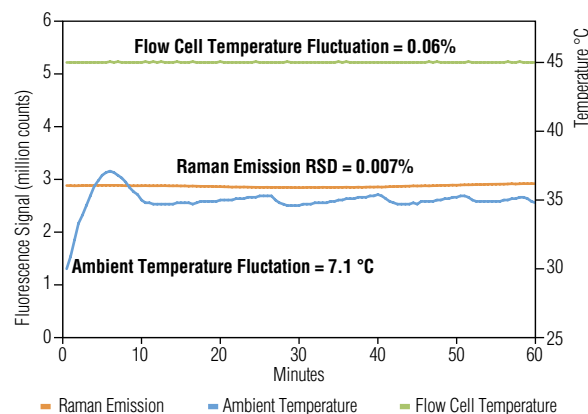


Figure 7. The thermostatted flow cell guarantees reproducible results even under varying ambient temperatures. Overlay of Raman signal intensity (orange), flow cell temperature (green) and ambient temperature (blue).

## Features for Maximum Uptime

FLD-3000 detectors use a xenon flash lamp. This lamp features three different flash frequency modes (LongLife, Standard, HighPower) for optimizing the sensitivity or lamp lifetime. Operating the lamp in LongLife mode leads to lifetimes of >15,000 h, several times more than a conventional, continuous xenon arc lamp used in other manufacturers' detectors. Conventional lamps also require a significant warm-up period and must remain switched on even if no fluorescence signal is acquired. Therefore, the real data acquisition time of a conventional xenon lamp can be significantly lower than its specified lifetime. The FLD-3000 series flash lamp is thermally optimized for warm-up within seconds. This means the lamp is used only when it is needed—during data acquisition.

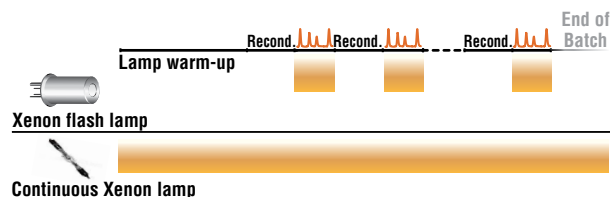


Figure 8. Lamp handling for maximum lifetime. The xenon flash lamp does not require long warm-up times and is only on during data acquisition. For minimal lamp wear, it can even be switched off when no peaks elute, e.g., during reconditioning times. Continuous xenon lamps are less flexible and require long-term temperature stabilization.

### Smart Use of Lamp Frequency Settings

The lamp frequency can be varied even during a chromatographic separation. Combining the lamp frequencies in a smart way provides optimum S/N and increases lamp lifetime at the same time. During dwell times and equilibration, the lamp flash frequency can be reduced (LongLife, 20 Hz). While peaks are eluting, a high-lamp-flash frequency (e.g., HighPower, 300 Hz) reduces baseline noise and, therefore, results in optimum S/N. In the Figure 9 example, the reduction of the flash frequency during idle times increases the lamp lifetime by 100%.

### Combining Sensitivity with Flexibility

PMTs are commonly used for fluorescence detection due to their significantly higher sensitivity compared to photodiodes. However, PMTs provide a limited wavelength range. A typical PMT supports an emission wavelength range up to 600 nm. To overcome this limitation, the Dionex FLD-3000 detectors can be ordered with, or field upgraded to, a Dual-PMT configuration. This unique option adds a second red-sensitive PMT to extend the wavelength range to 900 nm without sacrificing the sensitivity of the UV-vis region (Figure 10). Even the PMT selection can be changed during a chromatographic run to make sure each analyte is detected at highest S/N. Figure 11 provides an example of a dye separation. In addition to the switching of the excitation and the emission wavelengths, the detecting PMT changes whenever beneficial.

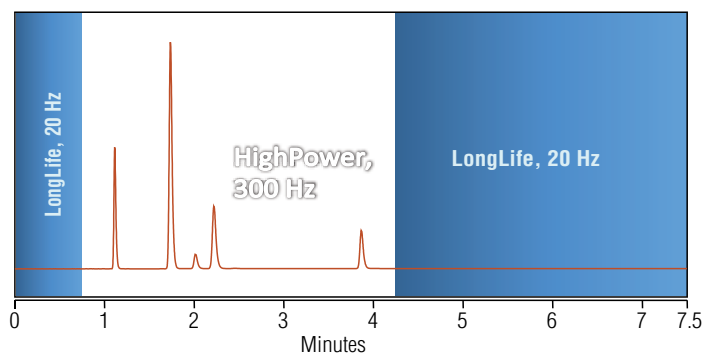


Figure 9. Alternating lamp flash frequencies during a run combines best S/N with an increased lamp lifetime. In this example, reducing the number of lamp flashes during idle times increases lamp lifetime by 100%.

### The Ultimate in Flexible Method Development

The FLD-3000 series provide various tools for effective development of selective methods for fluorescent analytes.

- 2D or 3D Excitation, Emission, or Synchro Scans: Perform a scan at a defined time of the chromatogram or simply use the 3D scanning functionality. This mode repeatedly acquires spectra during the entire separation and allows reviewing the data with familiar DAD-like spectral tools (Figure 12).
- Unique Variable Emission Filter: This automatically picks the best suitable emission filter for maximum stray light reduction (FLD-3400RS only).
- Zero-Order Mode: When the fluorescence characteristics of an analyte are unknown, the zero-order mode of the emission monochromator can be used to optimize the separation. It sets the emission grating to reflect the complete emission spectrum on the PMT.
- Multichannel Operation: Simultaneous operation of up to four different excitation and emission wavelengths are possible (FLD-3400RS only).

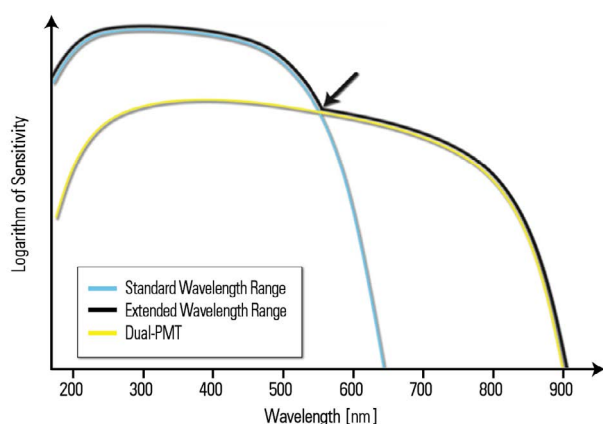


Figure 10. The Dual-PMT option of the Dionex FLD-3000 series is a unique solution for providing optimum sensitivity across all wavelengths.

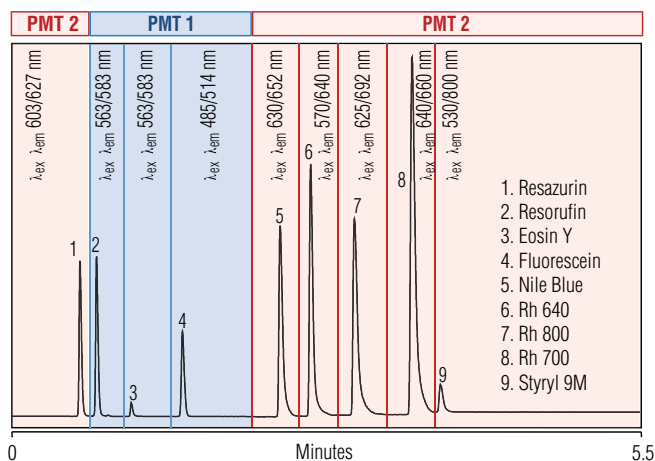


Figure 11. UHPLC analysis of dyes with a Dual-PMT detector configuration, which can change the measuring PMT during a chromatographic run. This ensures that any analyte achieves optimum S/N across the entire wavelength range up to 900 nm.

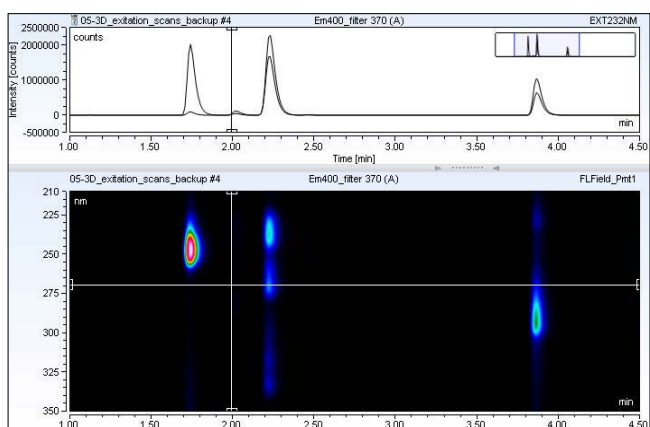


Figure 12. Excitation, emission, and synchro 3D scans acquire spectral data during the entire chromatographic run. From a single run, optimum detection wavelengths can be extracted by interfacing with an easy-to-use, DAD-like 3D field console.

## Take Control of All Functions—with Ease

Using Operational Simplicity™ as its guiding principle, the latest generation of Chromeleon 7 software takes users from samples to results in the shortest time possible. This software suite ensures users are utilizing the FLD-3000 detectors effectively and with ease, despite its various options and functions. In addition to the intuitive instrument control shown in Figure 13, the software's main new processing features include the Cobra™ peak detection algorithm for fast, accurate peak detection, and the SmartPeaks™ integration assistant for easy handling of unresolved peaks. Users can also automate processes with eWorkflows, a new tool that automates all chromatography workflows via an intuitive and easy-to-use interface.

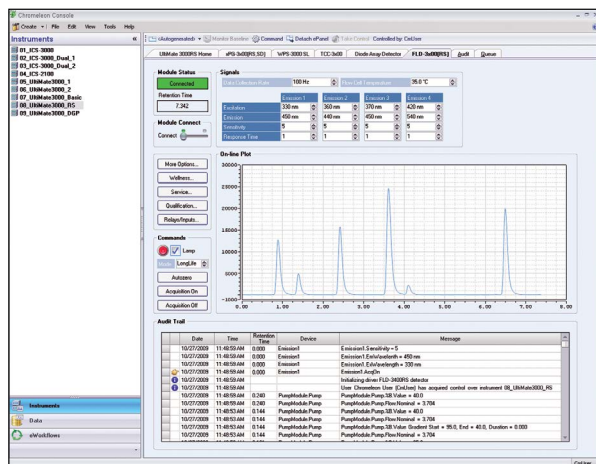


Figure 13. Intuitive ePanels provide direct access to all main instrument control features and easy access to advanced options.

## Thermo Scientific Dionex AutoQ Equipment Qualification

Thermo Scientific™ Dionex™ AutoQ™ Instrument 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 test results and related charts.

## Method Transfer Made Easy

Conventional LC methods can be made much more productive by using short UHPLC columns packed with smaller particles. The UltiMate 3000 systems fully support any UHPLC column format; the only remaining challenge is to define the right application parameters. Chromeleon 7.1 helps in the process of transferring a method to a different column format. It features an embedded Method Transfer Wizard that combines chromatographic results already available in the CDS, with some user input on the planned column to create a new instrument method (Figure 15).

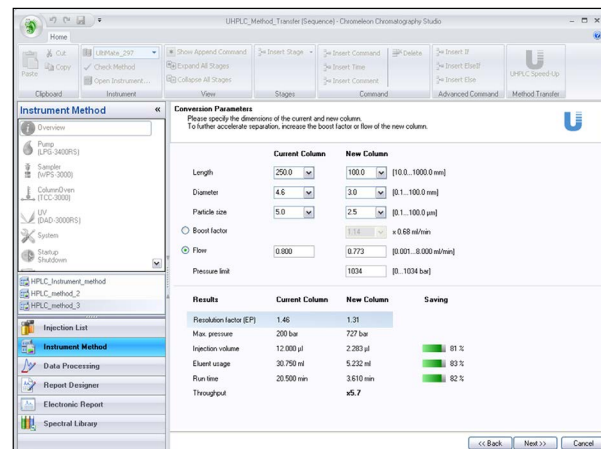


Figure 15. Transfer of conventional LC methods is exceptionally easy with the Method Transfer Wizard embedded into Chromeleon 7.1.

Based on the prediction of the chromatographic performance, the user can decide to further speed up the method or calculate for a different column. The Wizard also predicts how much time and solvent the new method will save.

## KEY SPECIFICATIONS

Light Source:	Xenon Flash Lamp				
Lamp Pulse Frequency:	HighPower (~300 Hz); Standard (~100 Hz); LongLife (~20 Hz)				
Data Collection Rate:	Single wavelength: up to 200 Hz (FLD-3400RS under Chromeleon 7.1 or later) Multiple wavelength: up to 4 Hz (FLD-3400RS) Single wavelength: up to 100 Hz (FLD-3100)				
Spectrum Scanning Modes:	2D or 3D excitation, emission or synchro scans (3D scans under Chromeleon 7.1 or later)				
Wavelength Range:	Variant	Ex Min	Ex Max	Em Min	Em Max
	FLD-3100	200	630	265	650
	FLD-3400RS	200	630	220	650
	FLD-3100 with Dual-PMT	200	880	265	900
	FLD-3400RS with Dual-PMT	200	880	220	900
Spectral Bandwidth:	Excitation: 20 nm Emission: 20 nm				
Emission Filter:	Variable, 5 positions (FLD-3400RS) Fixed, 280 nm (FLD-3100)				
No. of Fluorescence Channels:	Up to 4 (FLD-3400RS) 1 (FLD-3100)				
Excitation/Emission Wavelength Switching Time:	< 250 ms				
Wavelength Accuracy:	±2 nm				
Wavelength Repeatability:	±0.2 nm				
Sensitivity:	Raman S/N: > 550 ASTM over the entire lifetime of the lamp (> 2100 using dark signal as noise reference)				
Flow Cell Thermostatting:	Ambient + 15 °C to 50 °C				
Flow Cells:	Analytical flow cell volume: 8 µL 20 bar (290 psi) maximum cell pressure Micro flow cell volume: 2 µL 40 bar (580 psi) maximum cell pressure				
PC Connection:	All functions controllable through USB				
GLP Features:	Automatic Equipment Qualification (Dionex AutoQ), System Wellness, and Qualification Monitoring with Chromeleon CDS, cell i.d.-chip				
Dimensions (h × w × d):	16 × 42 × 51 cm (6.3 × 16.5 × 20 in.)				
Weight:	18 kg				

## 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.

<b>Detector</b>	<b>Part Number</b>
Dionex FLD-3100 Fluorescence Detector (without flow cell)	5078.0010
Dionex FLD-3100 Fluorescence Detector with Dual-PMT (without flow cell)	5078.0015
Dionex FLD-3400RS Rapid Separation Fluorescence Detector (without flow cell)	5078.0020
Dionex FLD-3400RS Rapid Separation Fluorescence Detector with Dual-PMT (without flow cell)	5078.0025
<b>Accessories</b>	<b>Part Number</b>
Analytical Flow Cell, 8 µL, stainless steel	6078.4230
Micro Flow Cell, 2 µL, stainless steel	6078.4330
Dual-PMT Option	6078.5360
Flow Cell Syringe Injection Kit	6078.4200

## [www.thermofisher.com/dionex](http://www.thermofisher.com/dionex)

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<b>Brazil</b> +55 11 3731 5140	<b>India</b> +91 22 6742 9494	<b>Singapore</b> +65 6289 1190	
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