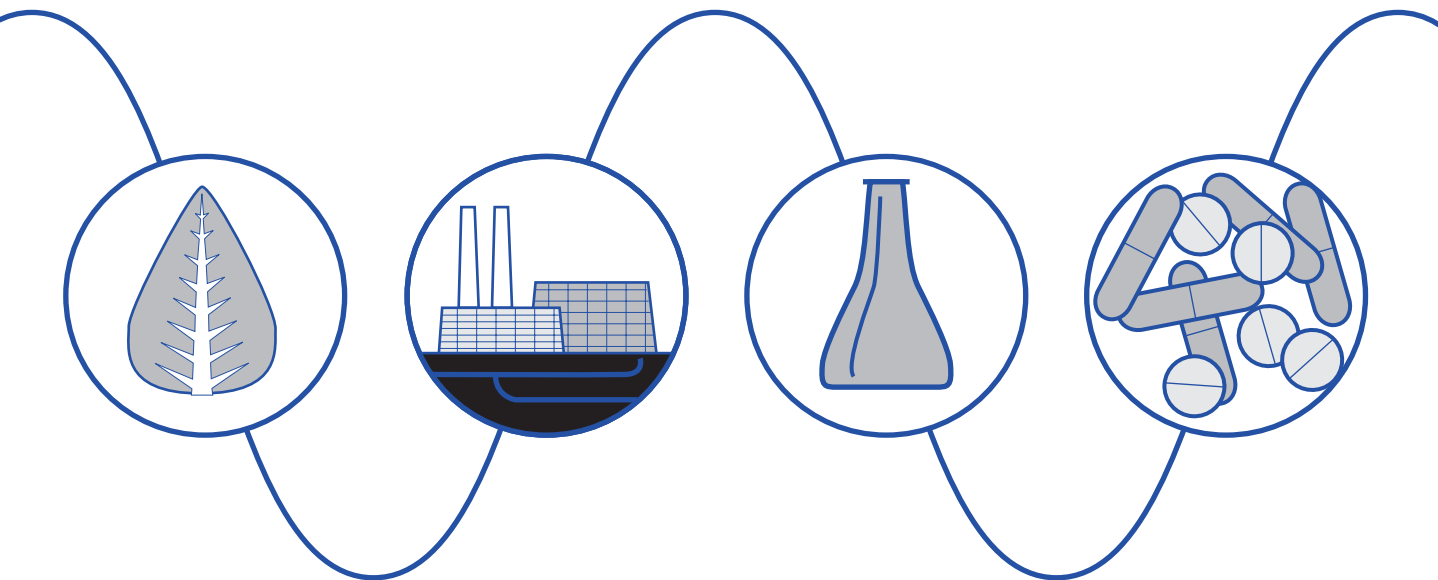


Waters 2488 Multichannel Absorbance Detector with MassLynx Control

Operator's Guide



Waters

34 Maple Street
Milford, MA 01757

71500248802, Revision A

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Note: When you use the instrument, follow generally accepted procedures for quality control and methods development.

If you observe a change in the retention of a particular compound, in the resolution between two compounds, or in peak shape, immediately determine the reason for the changes. Until you determine the cause of a change, do not rely on the separation results.

Note: The Installation Category (Overvoltage Category) for this instrument is Level II. The Level II Category pertains to equipment that receives its electrical power from a local level, such as an electrical wall outlet.



Atención: Changes or modifications to this unit not expressly approved by the party responsible for compliance could void the user's authority to operate the equipment.

Important : Toute modification sur cette unité n'ayant pas été expressément approuvée par l'autorité responsable de la conformité à la réglementation peut annuler le droit de l'utilisateur à exploiter l'équipement.

Achtung: Jedwede Änderungen oder Modifikationen an dem Gerät ohne die ausdrückliche Genehmigung der für die ordnungsgemäße Funktionstüchtigkeit verantwortlichen Personen kann zum Entzug der Bedienungsbezugnis des Systems führen.

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Atención: cualquier cambio o modificación realizado a esta unidad que no haya sido expresamente aprobado por la parte responsable del cumplimiento puede anular la autorización de la que goza el usuario para utilizar el equipo.



Caution: Use caution when working with any polymer tubing under pressure:

- Always wear eye protection when near pressurized polymer tubing.
- Extinguish all nearby flames.
- Do not use Tefzel tubing that has been severely stressed or kinked.
- Do not use Tefzel tubing with tetrahydrofuran (THF) or concentrated nitric or sulfuric acids.
- Be aware that methylene chloride and dimethyl sulfoxide cause Tefzel tubing to swell, which greatly reduces the rupture pressure of the tubing.

Attention : soyez très prudent en travaillant avec des tuyaux de polymères sous pression :

- Portez toujours des lunettes de protection quand vous vous trouvez à proximité de tuyaux de polymères.
- Eteignez toutes les flammes se trouvant à proximité.
- N'utilisez pas de tuyau de Tefzel fortement abîmé ou déformé.
- N'utilisez pas de tuyau de Tefzel avec de l'acide sulfurique ou nitrique, ou du tétrahydrofurane (THT).
- Sachez que le chlorure de méthylène et le sulfoxyde de diméthyle peuvent provoquer le gonflement des tuyaux de Tefzel, diminuant ainsi fortement leur pression de rupture.

Vorsicht: Bei der Arbeit mit Polymerschläuchen unter Druck ist besondere Vorsicht angebracht:

- In der Nähe von unter Druck stehenden Polymerschläuchen stets Schutzbrille tragen.
- Alle offenen Flammen in der Nähe löschen.
- Keine Tefzel-Schläuche verwenden, die stark geknickt oder überbeansprucht sind.
- Tefzel-Schläuche nicht für Tetrahydrofuran (THF) oder konzentrierte Salpeter- oder Schwefelsäure verwenden.
- Durch Methylenchlorid und Dimethylsulfoxid können Tefzel-Schläuche quellen; dadurch wird der Berstdruck des Schlauches erheblich reduziert.



Precauzione: prestare attenzione durante le operazioni con i tubi di polimero sotto pressione:

- Indossare sempre occhiali da lavoro protettivi nei pressi di tubi di polimero pressurizzati.
- Estinguere ogni fonte di ignizione circostante.
- Non utilizzare tubi Tefzel soggetti a sollecitazioni eccessive o incurvati.
- Non utilizzare tubi Tefzel contenenti tetraidrofurano (THF) o acido solforico o nitrico concentrato.
- Tenere presente che il cloruro di metilene e il dimetilsolfossido provocano rigonfiamento nei tubi Tefzel, che riducono notevolmente il limite di pressione di rottura dei tubi stessi.

Advertencia: manipular con precaución los tubos de polimero bajo presión:

- Protegerse siempre los ojos a proximidad de tubos de polimero bajo presión.
- Apagar todas las llamas que estén a proximidad.
- No utilizar tubos Tefzel que hayan sufrido tensiones extremas o hayan sido doblados.
- No utilizar tubos Tefzel con tetrahidrofurano o ácidos nítrico o sulfúrico concentrados.
- No olvidar que el cloruro de metileno y el óxido de azufre dimetilo inflan los tubos Tefzel lo que reduce en gran medida la presión de ruptura de los tubos.



Caution: The user shall be made aware that if the equipment is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.

Attention : L'utilisateur doit être informé que si le matériel est utilisé d'une façon non spécifiée par le fabricant, la protection assurée par le matériel risque d'être défectueuses.

Vorsicht: Der Benutzer wird darauf aufmerksam gemacht, dass bei unsachgemäßer Verwendung des Gerätes unter Umständen nicht ordnungsgemäß funktionieren.

Precauzione: l'utente deve essere al corrente del fatto che, se l'apparecchiatura viene usata in un modo specificato dal produttore, la protezione fornita dall'apparecchiatura potrà essere invalidata.

Advertencia: El usuario deberá saber que si el equipo se utiliza de forma distinta a la especificada por el fabricante, las medidas de protección del equipo podrían ser insuficientes.



Caution: To protect against fire hazard, replace fuses with those of the same type and rating.

Attention : Remplacez toujours les fusibles par d'autres du même type et de la même puissance afin d'éviter tout risque d'incendie.

Vorsicht: Zum Schutz gegen Feuergefahr die Sicherungen nur mit Sicherungen des gleichen Typs und Nennwertes ersetzen.

Precauzione: per una buona protezione contro i rischi di incendio, sostituire i fusibili con altri dello stesso tipo e amperaggio.

Precaución: sustituya los fusibles por otros del mismo tipo y características para evitar el riesgo de incendio.



Caution: To avoid possible electrical shock, power off the instrument and disconnect the power cord before servicing the instrument.








Attention : Afin d'éviter toute possibilité de commotion électrique, mettez hors tension l'instrument et débranchez le cordon d'alimentation de la prise avant d'effectuer la maintenance de l'instrument.

Vorsicht: Zur Vermeidung von Stromschlägen sollte das Gerät vor der Wartung abgeschaltet und vom Netz getrennt werden.



Precauzione: per evitare il rischio di scossa elettrica, spegnere lo strumento e scollegare il cavo di alimentazione prima di svolgere la manutenzione dello strumento.

Precaución: para evitar choques eléctricos, apague el instrumento y desenchufe el cable de alimentación antes de realizar cualquier reparación en el instrumento.

Commonly Used Symbols

	<p>Direct current</p> <p>Courant continu</p> <p>Gleichstrom</p> <p>Corrente continua</p> <p>Corriente continua</p>
	<p>Alternating current</p> <p>Courant alternatif</p> <p>Wechselstrom</p> <p>Corrente alternata</p> <p>Corriente alterna</p>
	<p>Protective conductor terminal</p> <p>Borne du conducteur de protection</p> <p>Schutzleiteranschluss</p> <p>Terminale di conduttore con protezione</p> <p>Borne del conductor de tierra</p>
	<p>Frame or chassis terminal</p> <p>Borne du cadre ou du châssis</p> <p>Rahmen- oder Chassisanschluss</p> <p>Terminale di struttura o telaio</p> <p>Borne de la estructura o del chasis</p>
	<p>Caution or refer to manual</p> <p>Attention ou reportez-vous au guide</p> <p>Vorsicht, oder lesen Sie das Handbuch</p> <p>Prestare attenzione o fare riferimento alla guida</p> <p>Actúe con precaución o consulte la guía</p>
	<p>Caution, hot surface or high temperature</p> <p>Attention, surface chaude ou température élevée</p> <p>Vorsicht, heiße Oberfläche oder hohe Temperatur</p> <p>Precauzione, superficie calda o elevata temperatura</p> <p>Precaución, superficie caliente o temperatura elevada</p>
	<p>Caution, risk of electric shock (high voltage)</p> <p>Attention, risque de commotion électrique (haute tension)</p> <p>Vorsicht, Elektroschockgefahr (Hochspannung)</p> <p>Precauzione, rischio di scossa elettrica (alta tensione)</p> <p>Precaución, peligro de descarga eléctrica (alta tensión)</p>

Commonly Used Symbols (Continued)

	<p>Caution, risk of needle-stick puncture Attention, risques de perforation de la taille d'une aiguille Vorsicht, Gefahr einer Spritzenpunktion Precauzione, rischio di puntura con ago Precaución, riesgo de punción con aguja</p>
	<p>Caution, ultraviolet light Attention, rayonnement ultraviolet Vorsicht, Ultraviolettes Licht Precauzione, luce ultravioletta Precaución, emisiones de luz ultravioleta</p>

2488 Multichannel Absorbance Detector Information

Intended Use

The Waters[®] 2488 Multichannel Absorbance Detector can be used for in-vitro diagnostic testing to analyze many compounds, including diagnostic indicators and therapeutically monitored compounds. When you develop methods, follow the "Protocol for the Adoption of Analytical Methods in the Clinical Chemistry Laboratory," *American Journal of Medical Technology*, 44, 1, pages 30–37 (1978). This protocol covers good operating procedures and techniques necessary to validate system and method performance.

Biological Hazard

When you analyze physiological fluids, take all necessary precautions and treat all specimens as potentially infectious. Precautions are outlined in "CDC Guidelines on Specimen Handling," *CDC – NIH Manual*, 1984.

Calibration

Follow acceptable methods of calibration with pure standards to calibrate methods. Use a minimum of five standards to generate a standard curve. The concentration range should cover the entire range of quality-control samples, typical specimens, and atypical specimens.

Quality Control

Routinely run three quality-control samples. Quality-control samples should represent subnormal, normal, and above-normal levels of a compound. Ensure that quality-control sample results are within an acceptable range, and evaluate precision from day to day and run to run. Data collected when quality-control samples are out of range may not be valid. Do not report this data until you ensure that chromatographic system performance is acceptable.

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Preface

The *Waters 2488 Multichannel Absorbance Detector with MassLynx Control Operator's Guide* describes the procedures for unpacking, installing, operating, verifying, maintaining, and troubleshooting the Waters® 2488 Multichannel Absorbance Detector. It also includes appendixes describing instrument specifications, spare parts, warranty and service information, and solvent considerations.

This guide is intended for use by anyone who installs, maintains, operates, or troubleshoots the Waters 2488 Multichannel Absorbance Detector. All personnel who use this guide should be familiar with HPLC terms and practices, and be capable of basic HPLC system operations such as making fluidic connections.

Organization

This guide contains the following:

[Chapter 1](#) summarizes the features of the Waters 2488 Detector and describes the theory and principles of operation.

[Chapter 2](#) describes how to install the Waters 2488 Detector, make fluidic and power connections, and connect the detector to other devices.

[Chapter 3](#) describes how to operate the Waters 2487 Detector from power-on to power-off, and use the operator interface.

[Chapter 4](#) describes how to clean and replace various parts of the Waters 2488 Detector.

[Chapter 5](#) describes the error messages, diagnostics, and recommended actions used in troubleshooting the Waters 2488 Detector.

[Appendix A](#) contains operational, optical, flow cell, and specifications for the Waters 2488 Detector.

[Appendix B](#) lists recommended and spare parts for the Waters 2488 Detector.

[Appendix C](#) provides information about the solvents used with Waters detectors.

Related Adobe Acrobat Reader Documentation

For detailed information about using Adobe® Acrobat® Reader, refer to the *Adobe Acrobat Reader Online Guide*. This Online Guide covers procedures such as viewing, navigating, and printing electronic documentation from Adobe Acrobat Reader.

Printing This Electronic Document

Adobe Acrobat Reader lets you easily print pages, page ranges, or the entire document by selecting **Print** from the File menu. For optimum print quantity, Waters recommends that you specify a PostScript® printer driver for your printer. Ideally, use a printer that supports 600 dpi print resolution.

Documentation Conventions

The following conventions may be used in this guide:

Convention	Usage
Purple	Purple text indicates user action such as keys to press, menu selections, and commands. For example, “Click Next to go to the next page.”
Italic	Italic indicates information that you supply such as variables. It also indicates emphasis and document titles. For example, “Replace <i>file_name</i> with the actual name of your file.”
Courier	Courier indicates examples of source code and system output. For example, “The <code>SVRMGR></code> prompt appears.”
Courier Bold	Courier bold indicates characters that you type or keys you press in examples of source code. For example, “At the <code>LSNRCTL></code> prompt, enter <code>set password oracle</code> to access Oracle.”
<u>Underlined blue</u>	Indicates hypertext cross-references to a specific chapter, section, subsection, or sidehead. Clicking this topic using the hand symbol automatically brings you to this topic within the document. Right-clicking and selecting Go Back from the shortcut menu returns you to the originating topic. For example: For verification procedures, see Section 3.3.2. Verifying the Detector .
Keys	The word <i>key</i> refers to a computer key on the keypad or keyboard. <i>Screen keys</i> refer to the keys on the instrument located immediately below the screen. For example, “The A/B screen key on the 2488 Detector displays the selected channel.”
...	Three periods indicate that more of the same type of item can optionally follow. For example, “You can store <i>filename1</i> , <i>filename2</i> , ... in each folder.”
>	A right arrow between menu options indicates you should choose each option in sequence. For example, “Select File > Exit ” means you should select File from the menu bar, then select Exit from the File menu.

Notes

Notes call out information that is helpful to the operator. For example:

Note: *Record your results before you proceed to the next step.*

Attentions

Attentions provide information about preventing possible damage to the system or equipment. For example:



Attention: *To avoid damaging the detector flow cell, do not touch the flow cell window.*

Cautions

Cautions provide information essential to the safety of the operator. For example:



Caution: *To avoid chemical or electrical hazards, always observe safe laboratory practices when operating the system.*



Caution: *To avoid possible electrical shock and injury, always turn off the detector and unplug the power cord before performing maintenance procedures.*



Caution: *To avoid possible burns, turn off the lamp at least 30 minutes before removing it for replacement or adjustment.*

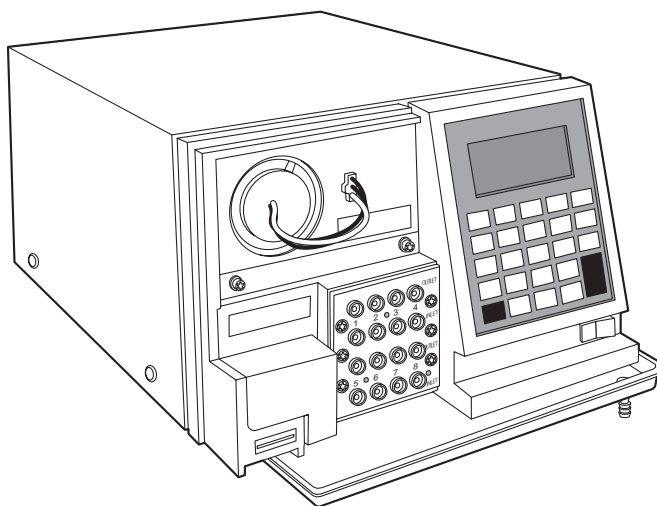
Chapter 1

2488 Detector Theory of Operation

This chapter introduces you to the Waters® 2488 Multichannel Absorbance Detector. It summarizes the 2488 Detector's features and describes the theory and principles of operation. See [Appendix A](#) for detector specifications, Appendix B for spare parts, [Appendix C](#) for information on high-performance liquid chromatography (HPLC) solvent considerations.

1.1 2488 Detector Description

The Waters 2488 Multichannel Absorbance Detector ([Figure 1-1](#)) is a tunable, ultraviolet/visible (UV/Vis) detector designed for multichannel (MUX) or parallel high-performance liquid chromatography mass spectroscopy applications (LC/MS).



TP01769

Figure 1-1 Waters 2488 Multichannel Absorbance Detector

This instrument functions as part of LC/MS systems based on the Micromass[®] MUX-Technology[™], which allows the Micromass LCT[™] and Waters ZQ[™] Mass Detectors to monitor four or eight separate electrospray inlets almost simultaneously.

Features

The Waters 2488 Multichannel Absorbance Detector operates from 190 to 700 nm. The 2488 Detector uses optics with a light-guided illumination system designed to allow up to eight flow cell absorbances to be measured simultaneously. The high optical throughput design provides sufficient energy to each flow cell to ensure excellent signal-to-noise performance. The narrow spectral band pass contributes to excellent linearity performance.

The Waters 2488 Multichannel Absorbance Detector has the following capabilities:

- **Single or dual wavelength** – Monitors absorbance at one or two wavelengths.
- **Wavelength verification on startup** – Ensures wavelength accuracy.
- **Automatic second order filter** – Automatically engaged for wavelengths 370 nm and greater and removed for wavelengths 369 nm or less.
- **Eight-channel flow cell configuration** – Allows you to select either four or eight channels to monitor.
- **Full diagnostic capability** – Supports built-in diagnostic tools to optimize functionality and performance.

1.2 Principles of Operation

To use the Waters 2488 Multichannel Absorbance Detector effectively, you should be familiar with the optical and electronic design of the detector and the theory and principles of operation.

This section describes the following parts and functions of the Waters 2488 Multichannel Absorbance Detector:

- Optics
- Wavelength verification and test
- Flow cell assembly
- Electronics

1.2.1 Waters 2488 Detector Optics

The Waters 2488 Multichannel Absorbance Detector optics are based on the Ebert tunable monochromator used in the 2487 Dual Channel Absorbance Detector. Optical

fibers convey the same single or dual wavelength to the multiple flow cells. The detector optics are modified for increased optical throughput and include the following:

- Deuterium (D_2) lamp
- Two mirrors: off-axis ellipsoidal mirror and spherical mirror
- Shutter, wavelength calibration filter, and second order filter
- Entrance slit
- Blazed, plane holographic diffraction grating
- Beam splitter
- Single reference photodiode and multiple sample photodiodes
- Waters multichannel flow cell with multiple light fibers (light fibers entrances are positioned at the exit slit location of the monochromator)

[Figure 1-2](#) shows a diagram of the 2488 Detector optics assembly light path and components.

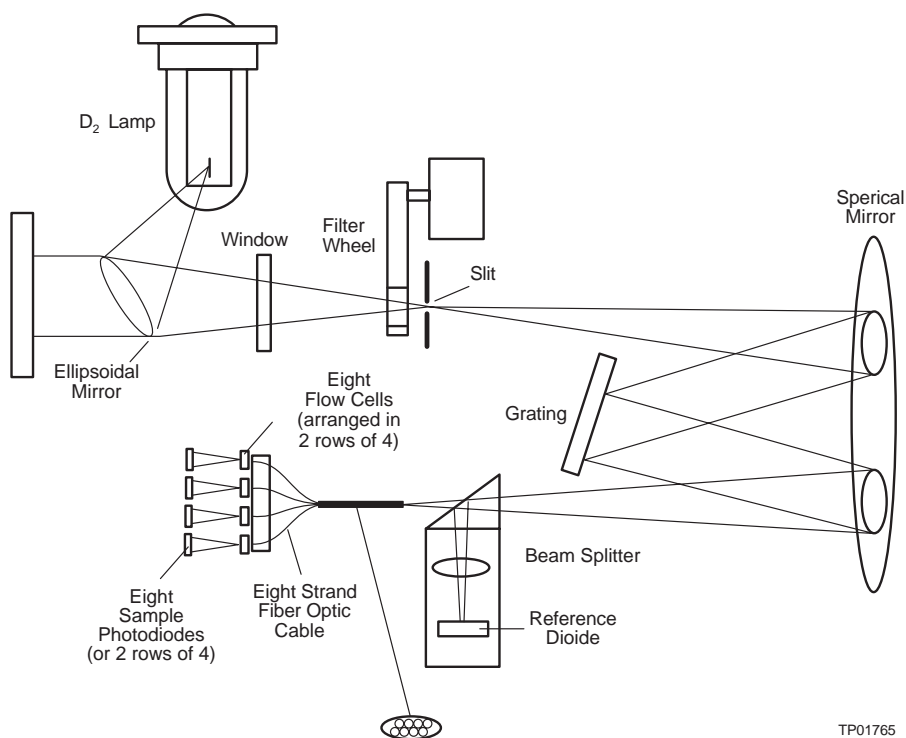


Figure 1-2 Waters 2488 Multichannel Absorbance Detector Optics Assembly

Optics Assembly Light Path

The Waters 2488 Multichannel Absorbance Detector provides an efficient design for high light throughput. It operates as follows:

1. The ellipsoidal mirror collects light from the lamp and focuses it through the filter wheel onto the entrance slit. The spherical mirror directs light toward the grating. A different portion of the spherical mirror focuses dispersed light of a particular wavelength, determined by the grating angle, onto the entrance of the light fiber bundle. Each light fiber transmits light to the entrance of one of the flow cells. Light exiting each flow cell is focused onto a separate photodiode for each channel.
2. The beam splitter, located just ahead of the flow cell assembly, diverts a portion of the light to a reference photodiode.
3. When you enter a new wavelength through the 2488 Detector's front panel (or through the MassLynx™ software), the detector rotates the grating to the appropriate position.
4. The preamplifier board integrates and digitizes the currents from all the photodiodes for processing by the signal processing electronics and sends the output to a computer.

1.2.2 Wavelength Verification and Test

The 2488 Detector deuterium arc lamp and the integral erbium filter show peaks and absorption bands in the transmission spectrum at known wavelengths. The erbium filter moves into the common light path ahead of the flow cell entrance slit. This allows the detector to locate three erbium absorption lines at these wavelengths:

- 256.7 nm (UV)
- 379.0 nm
- 521.5 nm

On startup, the detector verifies calibration, rather than recalibrates. It does this by comparing the locations of these spectral features with expected wavelengths based on stored data. When verification results vary from the stored calibration data by more than 1.0 nm, the detector displays a Wavelength Verification Failure message. (The calibration algorithm operates similarly, the sole difference occurring when actual data fails to match stored data. In that case, the algorithm replaces the stored data with the new data.)

The wavelength verification procedures use a grating homing sensor to establish an approximate home position. Once home is established, the detector locates and references the 656.1-nm peak in the deuterium lamp emission spectrum.

Verification tests require 5 minutes of lamp warmup time. Because verification occurs on startup, you should verify wavelength weekly on instruments that operate continuously. Do

this by switching the detector off and then on again. See [Section 3.3.3, Calibrating the Wavelength](#), for details.

You can manually calibrate the wavelength anytime. Manual calibrations produce new data that replace the previous calibration's. [Section 3.3.3](#) describes the calibration procedure.

When the detector recalibrates, it uses as its reference the first cell in the flow cell grouping you chose to use. When you choose flow cells 1–4 or 1–8, it calibrates using channel 1. When you select flow cells 5–8, it calibrates using channel 5. See [Section 5.2.2, Using the Diagnostics](#).

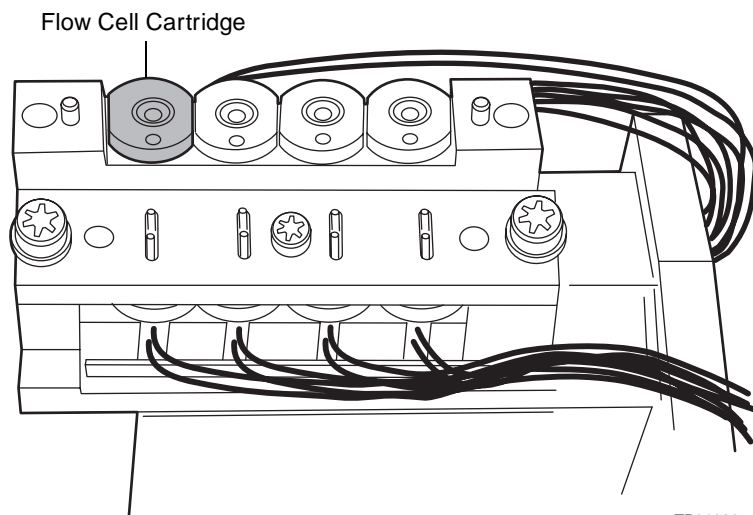
1.2.3 Waters 2488 Detector Flow Cell Assembly

The flow cells used in the 2488 Detector multichannel flow cell assembly ([Figure 1-3](#)) render the detector baseline insensitive to changes in mobile phase refractive index (RI). RI changes occur during gradient separations or result from temperature or pump-induced pressure fluctuations.

To achieve RI immunity, a combination of an aperture stop on the spherical mirror and a lens at the entrance of the flow cell prevent light rays from striking the internal walls of the flow cell. A second lens, at the exit of the flow cell, ensures that the light is collected by the detector. Compared to a conventional flow cell without lenses at the entrance and exit, the 2488 Detector maximizes the light transfer from the fiber through the flow cell to the photodiode.

In a conventional cell, a change in the RI can cause the light to bend and hit the wall of the flow cell. In the Waters multichannel TaperBeam™ flow cell assembly, the combination of the two lenses prevents light from hitting the cell walls.

The flow cells have a 4-mm path length and are designed as a cartridge assembly. The cartridge assembly can be easily removed and replaced.



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Figure 1-3 2488 Detector Flow Cell Assembly

Filtering Noise

The 2488 Detector provides two filters to minimize noise:

- **Hamming filter** – The default filter, which is a digital finite impulse response filter that creates the same amount of peak height degradation as the RC filter, and also enhances the filtering of high frequency noise.
- **RC filter** – The analog infinite impulse response filter, which is a first-order resistor-capacitor filter.

The behavior of the filter depends on the filter time constant that you select. The filter time constant adjusts the filter response time to achieve an optimal signal-to-noise ratio.

Lower time constant settings:

- Remove less baseline noise
- Produce narrow peaks with minimal peak distortion and time delay
- Make very small peaks harder to discriminate from baseline noise

Higher time constant settings:

- Greatly decrease baseline noise
- Shorten and broaden peaks

The default time constant of 1.0 second in the Hamming mode is satisfactory for most traditional applications. Lower time constant settings, such as 0.1 second, are recommended for fast, short column separations.

Note: For columns with an inner diameter of 2.1 mm or less, consider using the 0.3-second Hamming filter setting.

To calculate an appropriate time (filter) constant for special applications, use the formula:

$$TC = 0.2 \times PW$$

where:

TC = Time constant (filter) setting

PW = Peak width at half height of the narrowest peak

[Figure 1-4](#) shows the relationship between increased time constant and response times.

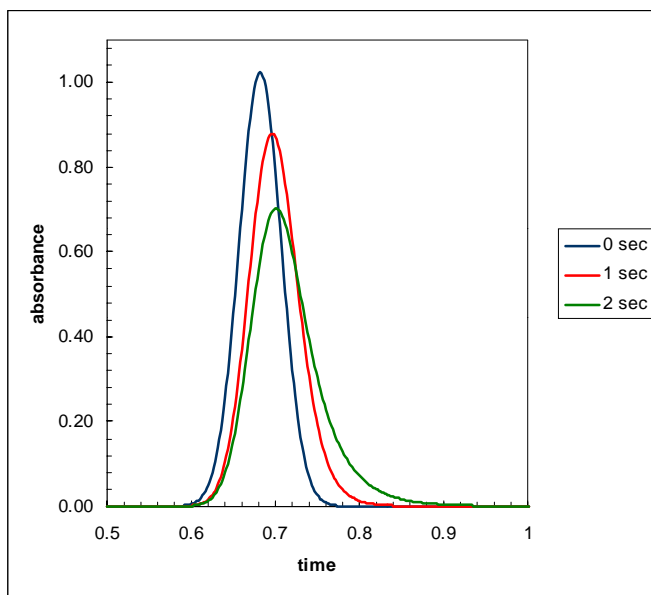


Figure 1-4 Time Constant (Filter Setting) Comparison

Note: Although the peak shape shows some distortion and the signal output is delayed with different time constants, the peak area remains the same.

1.2.4 Waters 2488 Detector Electronics

The 2488 Detector electronics control the following components:

- **Preamplifier board** – Collects and processes the analog input signals from the nine photodiodes (eight sample and one reference) to the microprocessor for further

signal conditioning. Sample and reference signals are integrated and A/D conversion is performed simultaneously. This ensures the best rejection of common mode noise in the reference and sample beams, leading to a very quiet baseline. All flow cells use the same reference signal.

- **Control board** – Receives inputs from the preamplifier board, keyboard, and external events, and provides control of the optics positioning subsystems and lamp power supply.
- **IEEE-488 interface** – IEEE-488 communication allows the 2488 Detector to communicate with the MassLynx software.
- **Keypad** – Allows you to configure, calibrate, and troubleshoot the 2488 Detector.
- **Lamp power supply** – Controlled by the CPU to provide stable deuterium lamp operation.
- **DC power supply** – Provides voltage for the analog and digital circuitry.

1.3 Operational Modes

The 2488 Detector operates in single or dual wavelength mode with the same one or two wavelengths across all eight flow cells.

1.3.1 Single Wavelength Mode

Single wavelength is the default mode of operation for the 2488 Detector. The 2488 Detector supports monitoring of a single wavelength from 190 to 700 nm, settable in 1-nm increments on channel A.

In single wavelength mode, the 2488 Detector automatically engages the second order filter for wavelengths 370 nm and above and removes it for wavelengths under 370 nm. The second order filter is an optical filter that blocks unwanted ultraviolet (UV) light produced by the diffraction grating that can interfere with absorbance detection.

1.3.2 Dual Wavelength Mode

In dual wavelength mode, the detector can monitor two wavelengths: one on Channel A and one on Channel B. All channels see the same wavelength, which alternates between A and B. The sampling frequency is reduced from 10 to 1 Hz, limiting use of this mode to more standard chromatography where peaks span at least 20 seconds to enable full characterization of a peak.

The 2488 Detector allows you to select any two wavelengths from 190 to 700 nm. Dual wavelength operation is accompanied by an increase in noise.



Attention: In dual wavelength mode:

- If both selected wavelengths are greater than 370 nm, the detector applies the second order filter to block unwanted UV light.
- If both selected wavelengths are less than or equal to 370 nm, the detector removes the second order filter.
- If the selected wavelengths bracket the 370-nm threshold, the detector does not apply the second order filter and issues a warning message that any data collected for the wavelength above 370-nm may contain inaccuracies because of possible UV light interference (second order effects).



Attention: To prevent acquisition of invalid data when operating the 2488 Detector in dual wavelength mode under the control of MassLynx software, you must select a data sampling rate of 1 point per second.

1.4 Data Selection Modes

1.4.1 Absorbance Data

The 2488 Detector primary data output is in absorbance. Absorbance can be monitored up to 4 AU. Linearity is specified to 2.5 AU.

When you press the Diagnostics key, you can monitor Sample or Reference Energy. The diagnostic displays the values for all eight flow cells regardless of the numbers configured for data acquisition sampling.

1.4.2 Sample Energy

Sample energy displays the sample photodiode outputs in nA/V (nano amperes).

1.4.3 Reference Energy

Reference energy displays the reference photodiode output in nA/V (nano amperes).

Chapter 2

Installing the 2488 Detector

2.1 Introduction

This chapter describes how to install the 2488 Detector and connect it to the electrical supplies and to other equipment in the system. The Waters 2488 Multichannel Absorbance Detector requires connections to electrical power and to sample and waste lines to operate in any standard laboratory environment. [Figure 2-1](#) shows the primary installation steps.

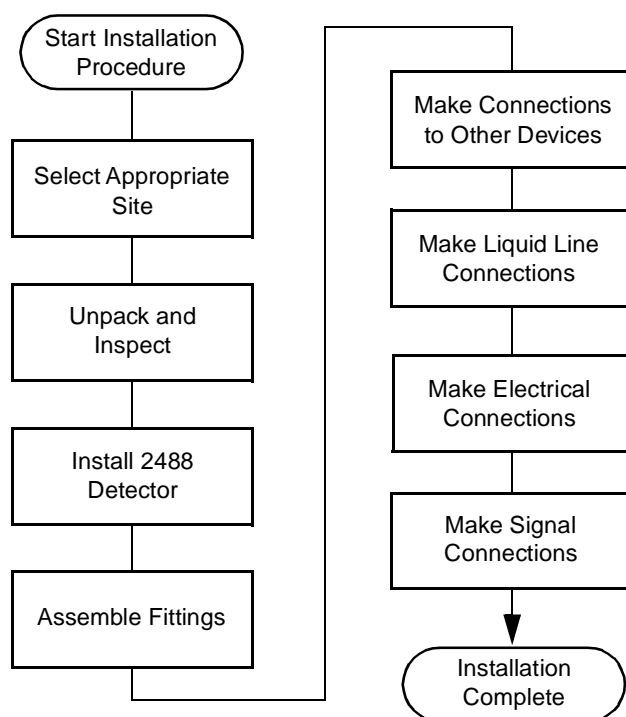
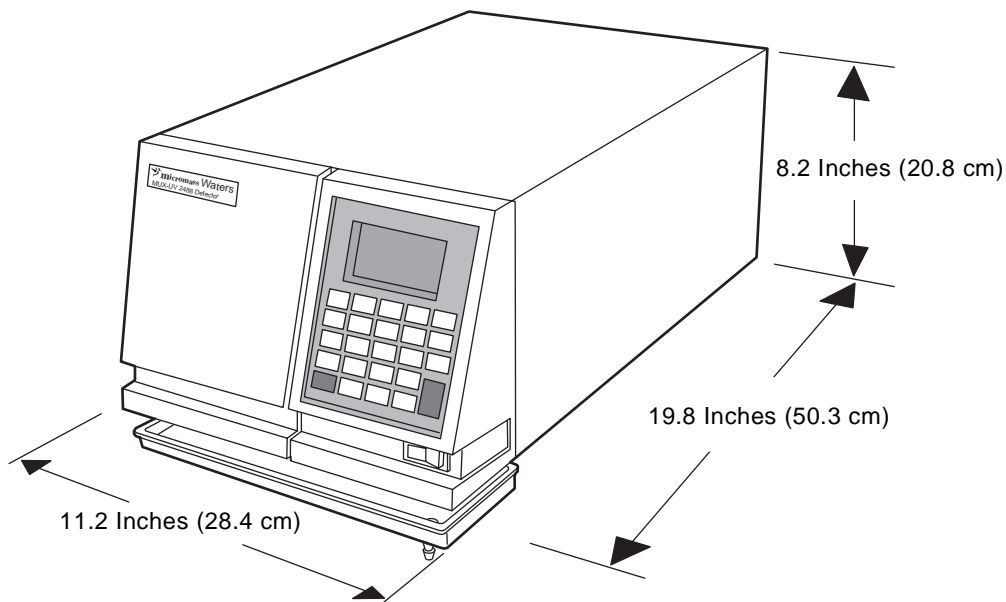


Figure 2-1 Major Installation Steps

After installing the 2488 Detector, verify its functions and keep the verified output on file. Verification ensures proper operation of the detector optics and electronics. For verification procedures, see [Section 3.3.2, Verifying the Detector](#).

[Figure 2-2](#) illustrates the size and the physical characteristics of the Waters 2488 Multichannel Absorbance Detector.



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Figure 2-2 2488 Detector Dimensions

Note: There is no access required to the instrument inside the top cover. Access is through the left front panel where the lamp housing and flow cell assembly are located.

2.2 Site Selection and Power Requirements

2.2.1 Site Selection

Install the Waters 2488 Multichannel Absorbance Detector in an area that meets the requirements in [Table 2-1](#).

Table 2-1 Installation Site Requirements

Parameter	Requirement
Operating temperature range	4 to 40 °C (39 to 104 °F)
Storage temperature range	–40 to 70 °C (–40 to 158 °F)
Operating relative humidity range	20 to 80%, noncondensing
Storage humidity range	20 to 80%, noncondensing
Bench space	5 inches (12.7 cm) clearance at rear
Vibration	Negligible
Static electricity	Negligible
Power	Grounded ac, 100/240 Vac, 50/60 Hz

You must mount the 2488 Detector on a level surface to allow proper function of the drip management system (drain tube which is supplied with the Startup Kit), which you can connect to a waste reservoir to divert solvent leaks from the flow cell.

2.2.2 Power Requirements

The 2488 Detector requires:

- Grounded alternating current (ac) power source.
- Minimal power transients and fluctuations.
- Line voltage of 100 to 240 Vac. Power consumption is 145 volt amps (VA).
- Requires 3.15 A fuse and operates in a nominal voltage range of 100 to 240 Vac.



Caution: To avoid electrical shock, power off the 2488 Detector and unplug the power cord before you replace the fuse.



Caution: To reduce the risk of fire hazard, always replace the fuse with those of the same type and rating.

2.3 Unpacking and Inspecting

The 2488 Detector is packed and shipped in one carton containing the:

- Waters 2488 Multichannel Absorbance Detector
- Startup Kit, which includes the *Waters 2488 Multichannel Absorbance Detector with MassLynx Control Operator's Guide*
- Certificate of structural validation
- Power cord
- Release notes

2.3.1 Unpacking

To unpack the 2488 Detector:

1. Unpack the contents of the shipping carton. As you unpack the carton, check the contents to make sure you have received all items.
2. Check the contents of the Startup Kit to ensure that you have received all the items.
3. Save the shipping carton for future transport or shipment.

2.3.2 Inspecting

If you see any damage or discrepancy when you inspect the contents of the carton, immediately contact the shipping agent and Waters Technical Service at 800 252-4752, *U.S. and Canadian customers only*. Other customers, call your local Waters subsidiary or your local Waters Technical Service Representative, or call Waters corporate headquarters for assistance in Milford, Massachusetts (U.S.A.).

If you are a Micromass customer, please contact Micromass at +44 161 946 2400, or +44 161 926 7200, or +44 161 435 4100 or call your local Micromass subsidiary.

Note: Make sure the instrument serial number, found on the nameplate on the rear panel or inside the left front panel of the 2488 Detector, corresponds to the number on the certificate of structural validation.

2.4 Assembling the Fittings

To assemble each fitting:

1. Slide the compression screw with ferrule over the tubing end ([Figure 2-3](#)).
2. Mount the ferrule with its taper end facing the end of the tubing ([Figure 2-3](#)).

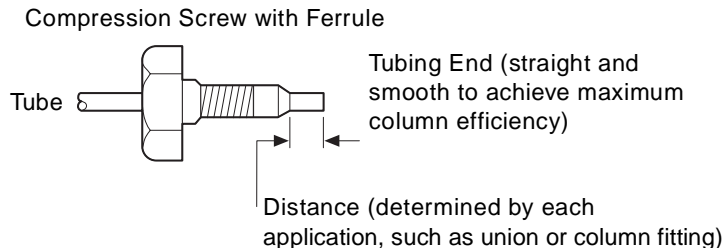


Figure 2-3 Ferrule and Compression Screw Assembly

Making Connections

To make connections at the column outlet and detector inlet, and at the detector outlet:

1. Seat each tubing end in the appropriate fitting.
2. Seat each ferrule by tightening the compression screw 1/2-turn past finger tight.

2.5 Connecting Liquid Lines

Before initial startup of the Waters 2488 Multichannel Absorbance Detector, you must complete the:

1. Liquid lines connections described in this section
2. Electrical connections described in [Section 2.8](#)



Caution: Observe safe laboratory practices when handling solvents. Refer to the Material Safety Data Sheets for the solvents in use.

3. Fitting assembly described in [Section 2.4](#)

You need to make the following liquid line connections on your detector unit:

- Column connections
- Drip management system connections

Before completing the column connections, Waters recommends you perform the verification procedures described in [Section 3.3.2, Verifying the Detector](#).

Waters recommends that you install a waste reservoir connected to the drain tube, located next to the rubber foot on the bottom-left front corner of the detector.



Attention: The 2488 Detector is shipped with flow cells that are pressure rated at 1000 psi. To prevent damage, do not connect any tubing or device that might cause back pressure to exceed the pressure rating of the tubing or flow cell.

Connecting Columns

Flow cells on the 2488 Detector are positioned in two banks, starting on the upper-left channels 1–4. The lower bank contains channels 5–8. The flow cell inlet is in the lower row of the bank and the flow cell outlet is the uppermost row of the bank ([Figure 2-4](#)).

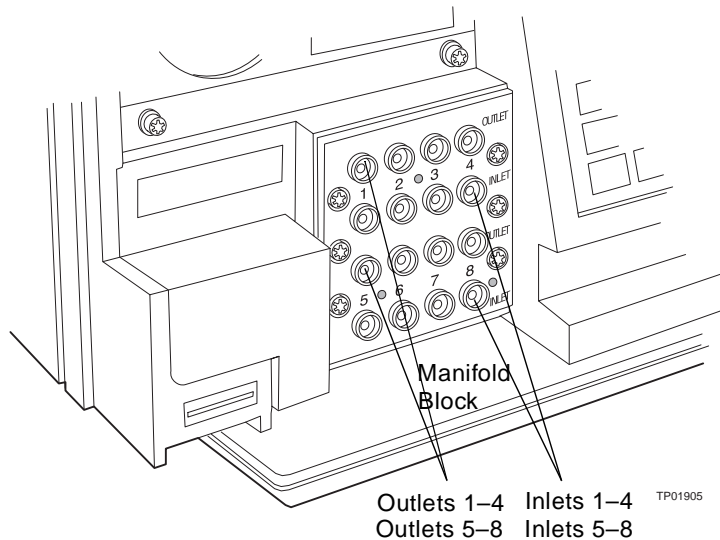


Figure 2-4 2488 Detector Liquid Line Connections

Note: If your system is a four-channel MUX, use either flow cells 1–4 (the upper bank of flow cells), or flow cells 5–8, (the lower bank of flow cells). Configuring your system this way minimizes any wavelength accuracy differences.

The 2488 Detector liquid line connections are in the flow cell manifold block to the right and front of the detector ([Figure 2-4](#)).

Required Materials

- PEEK 0.005-inch ID tubing
- Cutting tool

Making Inlet Tubing Connections

To make the inlet tubing connections:

1. Cut an appropriate length of PEEK 0.005-inch ID tubing for each channel from the length supplied in the Startup Kit.
2. Attach a PEEK compression fitting and ferrule (supplied in the 2488 Startup Kit) to either end.
3. Connect one end of each of the inlet tubing lengths to either the column or flow splitter outlet.
4. Connect the other end to the appropriate flow cell inlet according to the channel assigned.
5. Ensure the tubing is seated firmly, then tighten the compression screw.

To make the outlet tubing connections:

1. Cut an appropriate length of PEEK 0.005-inch ID tubing for each channel from the length supplied in the Startup Kit.
2. Attach a PEEK compression fitting and ferrule (supplied in the 2488 Startup Kit) to either end.
3. Connect the PEEK 0.005-inch ID tubing to the flow cell outlet for each channel and route to a waste container or the ZQ Mass Detector as appropriate.
4. Ensure the tubing is seated firmly, then tighten the compression screw.



Caution: *Observe safe laboratory practices when handling solvents. Refer to the Material Safety Data Sheets for the solvents in use.*

2.6 Connecting the Waste Line

The 2488 Detector waste line connection is located in the front-right corner of the drip tray.

Required Materials

- Tygon 2705 tubing
- 7/16-inch wrench
- Male connector, 1/8-inch NPT
- Waste container

Making the Waste Connection

To make the waste connection:

1. Screw the NPT fitting into the front-right corner of the drip tray.
2. Use a 7/16-inch wrench to secure the fitting to the drip tray. Do not overtighten.
3. Place the instrument or top of a mass spectrometer with the NPT fitting hanging over the edge.
4. Attach the Tygon 2705 tubing (supplied in the 2488 Startup Kit) to the NPT fitting and place the other end of the tubing into a waste container.

2.7 Connecting to Other Autosamplers

You can connect the 2488 Detector to other HPLC equipment. This section describes how to connect the 2488 Detector to an autosampler other than Gilson or CTC.

The autosampler signals the start of an injection through a contact closure signal on its Inject Start terminals. You can use this contact closure signal to command the 2488 Detector to perform an auto zero at the start of an injection.

Required Materials

When connecting cables to the terminals on the rear panel of the 2488 Detector, you need:

- Small flat-blade screwdriver
- Electrical insulation stripping tool

Connecting the Cables

To connect cables from other HPLC equipment to the A and B terminals on the rear panel of the 2488:

1. Remove terminal A or B ([Figure 2-7](#)).
2. Unscrew the connecting pin terminal: 1 and 2 for inject start, 10 and 11 for auto zero.
3. Using the stripping tool, strip the wire down about 1/8-inch from the end.
4. Insert the stripped wire into the appropriate connector: 1 and 2 for inject start, 10 and 11 for auto zero.
5. Tighten the screw until the wire is held securely in place.
6. Reinsert the terminal.
7. Press firmly to ensure that it is inserted fully.

2.8 Making Electrical Connections

To connect the 2488 Detector to the ac power supply:

1. Plug the receptacle end of the power cord into the ac input connector on the rear panel of the detector ([Figure 2-5](#)).

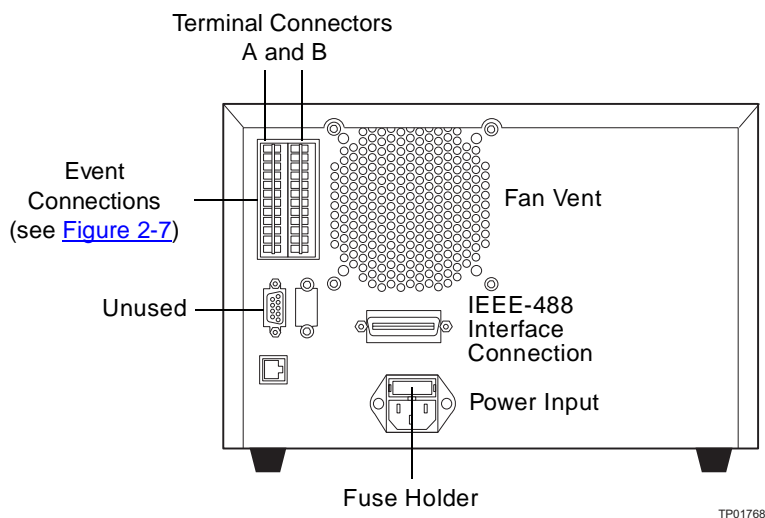


Figure 2-5 2488 Detector Rear Panel Electrical Connections

2. Plug the other end of the power cord into a properly grounded ac power source.

2488 Detector Rear Panel

The 2488 Detector connects to other system components through rear panel electrical connections ([Figure 2-5](#)).

Rear panel connections enable the following signals:

- **Event inputs** – There are three general-purpose TTL contact closures on the 2488 Detector A (Inputs) terminal that support the following functions:
 - Remote or inject start
 - Lamp on/off
 - Auto zero
- **IEEE-488 interface bus** – The IEEE-488 bus connection ([Figure 2-5](#)) allows remote control and direct data acquisition from MassLynx workstations.

2.9 Making Signal Connections

The rear panel of the 2488 Detector ([Figure 2-5](#)) provides an IEEE-488 communications port for operating the detector with external devices. The signal connections you need to make to your 2488 Detector depend on the types of instruments that make up your HPLC system.

This section describes the input/output (I/O) and digital signal connections that you can make from the two rear panel connectors and the IEEE-488 bus connector.

[Figure 2-6](#) shows an overview of the steps involved in making signal connections to the 2488 Detector.

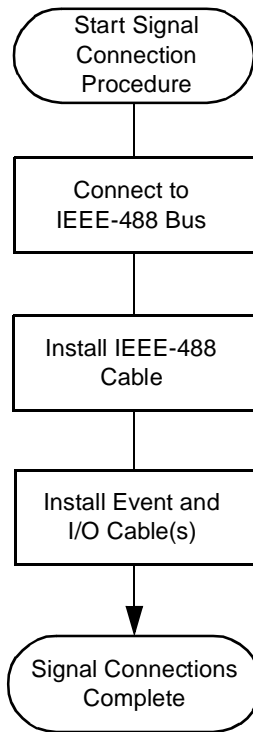


Figure 2-6 Overview of Making Signal Connections

2.9.1 Making I/O Signal Connections

The rear panel includes one removable connectors that hold the pins for the I/O signals, as shown in [Figure 2-7](#). These connectors are keyed so that you can insert them only one way.

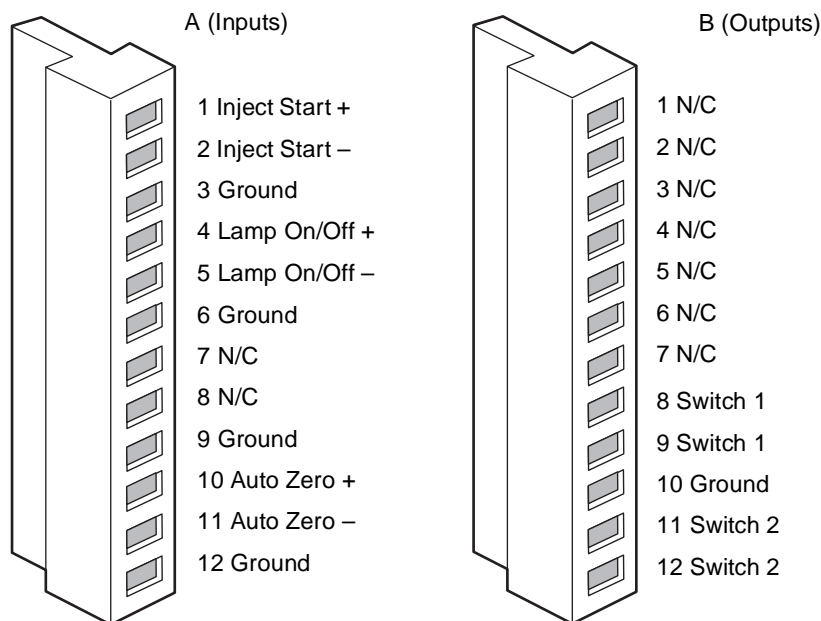


Figure 2-7 I/O Signal Inputs and Outputs

I/O Signals

[Table 2-2](#) describes each signal available on the I/O connectors. Refer to [Appendix A. 2488 Detector Specifications](#), for details on the electrical specifications for the signals.

Table 2-2 I/O Signals for the 2488 Detector

Signal	Description
Inject Start ^a	TTL contact closure. Configurable input to initiate sequencing of time-programmed events. Defines the start of a run (typically an injection) and resets and starts the run-time clock to 0.00 minutes. Initial conditions apply immediately.
Lamp On/Off ^a	Configurable input to allow an external device to turn the deuterium lamp off and on.
Auto Zero ^a	Configurable input to auto zero channels A and/or B.
Switch 1 (2)	Used to connect to another device. Can be controlled by threshold and timed events.
Switch 2 (2)	

a. Inject start, auto zero, and lamp inputs are configurable from the 2488 Detector Diagnostics screen by setting the appropriate parameter to High. See [“Configuring Inputs”](#) in [Section 3.2.6](#) for more detail.

2.9.2 Connecting the 2488 Detector to LCT-MUX or ZQ-MUX System Configuration

The 2488 Detector I/O under MassLynx control may require the following connections:

- Method start to a MassLynx, injector, or other device
- Auto zero on inject (optional)
- Turn lamp on and off (optional)

The Method Start signal must be configured. It is important to configure a “software trigger.” To configure the software trigger in MassLynx: in the Method Editor, click **Tools > Instrument Configure Menu > Triggers Tab**.

Method (Inject) Start Connections

You can use the Inject Start connections on an autosampler to connect to the Inject Start signals on the 2488 Detector to program the start of the active method.

To program an inject start, make the connections described in [Table 2-3](#) and illustrated in [Figure 2-8](#).

Table 2-3 Autosampler and 2488 Detector Inject Start Connections

2488 Detector (A Inputs)	Autosampler
Pin 1 Inject Start +	Inject Start + (any one of three paired with +)
Pin 2 Inject Start –	Inject Start – (any one of three paired with –)

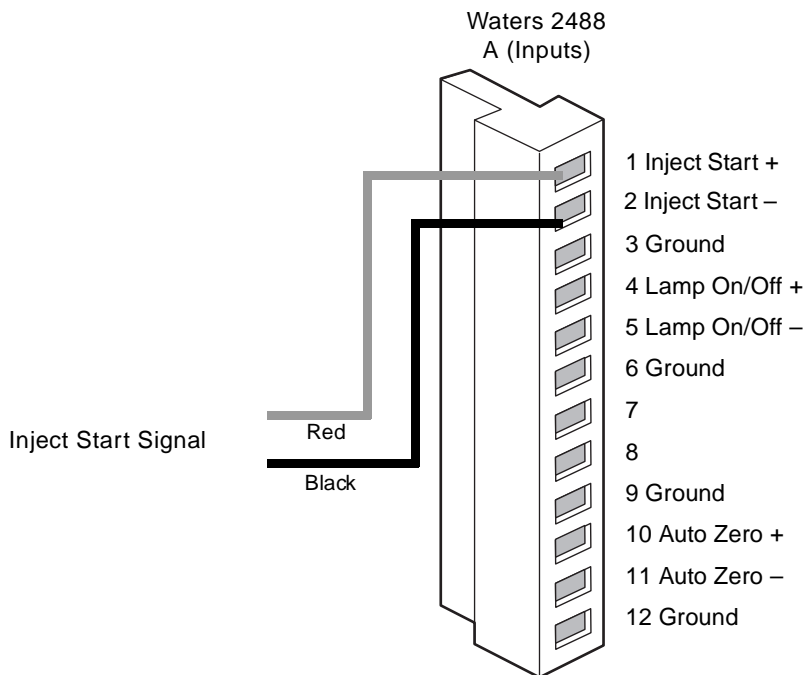


Figure 2-8 Waters 2488 Detector Inject Start Connections

Auto Zero Connections

To auto zero the 2488 Detector at the start of an injection, make the connections described in [Table 2-4](#) and illustrated in [Figure 2-9](#).

Table 2-4 Autosampler and 2488 Detector Auto Zero Connections

2488 Detector (A Inputs)	Autosampler
Pin 10 Auto Zero +	Auto Zero + (any one of three paired with -)
Pin 11 Auto Zero -	Auto Zero - (any one of three paired with +)

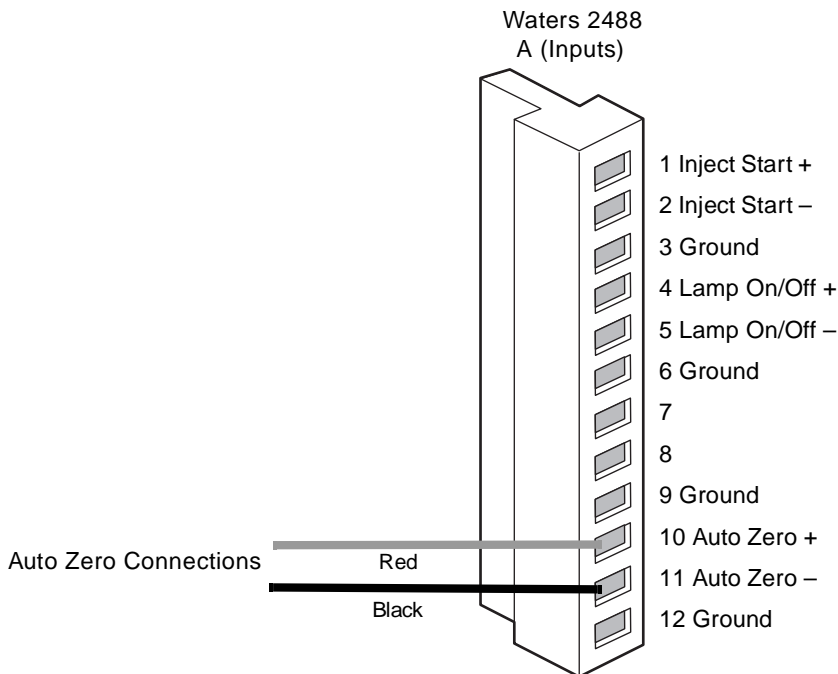


Figure 2-9 Auto Zero Connections to the 2488 Detector

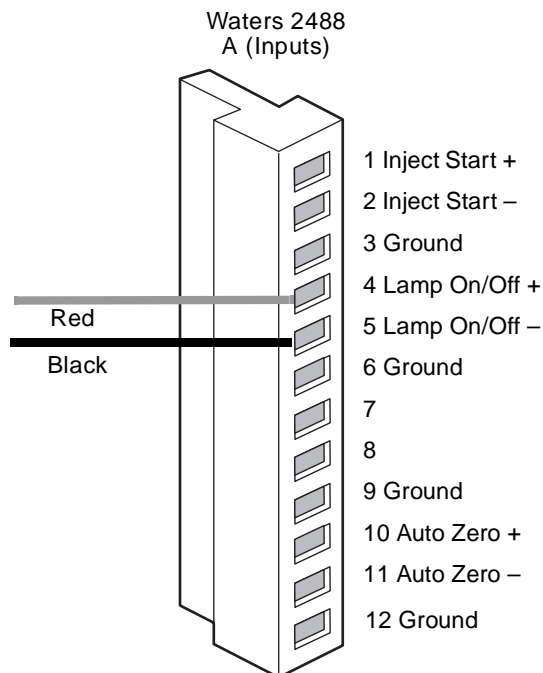
[Figure 2-10](#) illustrates the connections between the 2488 Detector and the autosampler. Use any available pair of Inject Start terminals on the autosampler.

Turning the 2488 Detector Lamp On or Off

After configuring the 2488 Detector lamp on/off signal, you can turn the lamp on or off by making the connections in [Table 2-5](#) and shown in [Figure 2-10](#) and using a keypad command ([Section 3.2](#)). Turning the lamp on or off is typically done in MassLynx software through IEEE access.

Table 2-5 2488 Detector Connections to Turn the Detector Lamp On or Off

2488 Detector (A Inputs)	Autosampler
Pin 4 Lamp On/Off +	Pin 1 Switch 1
Pin 5 Lamp On/Off –	Pin 2 Switch 2



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Figure 2-10 Connections to the 2488 Detector for Turning the Lamp On or Off

2.9.3 Making IEEE-488 Connections in MassLynx

The rear panel also includes one IEEE-488 interface connector for digital signal communications. Use the IEEE-488 interface connector to connect the 2488 Detector to a workstation with MassLynx software version 3.5 or higher. The IEEE-488 connector mates with a standard IEEE-488 cable.



Attention: To avoid possible damage to components, power off all instruments on the IEEE-488 control bus before you connect an IEEE-488 interface cable to an instrument.

Making IEEE-488 Connections in a MassLynx Workstation

When controlling the 2488 Detector from a MassLynx workstation, you can use the IEEE-488 interface to send and receive information from the data system. To do this:

1. Connect the single receptacle end of the IEEE-488 cable (supplied with the Waters Startup Kit) to your data system.

2. Connect the other end of the cable to the IEEE-488 connector on the 2488 Detector rear panel.



Attention: The maximum total cable length between IEEE-488 devices in a system is 65 feet (20 meters). The maximum recommended cable length between two IEEE-488 devices is 13 feet (4 meters). Longer total cable lengths can cause intermittent IEEE-488 communication failures.

3. Ensure all IEEE-488 cable screws are fastened finger tight ([Figure 2-11](#)).
4. Set a unique IEEE-488 address from 2 to 29 for the 2488 Detector. The default IEEE-488 address for the 2488 Detector is 4.

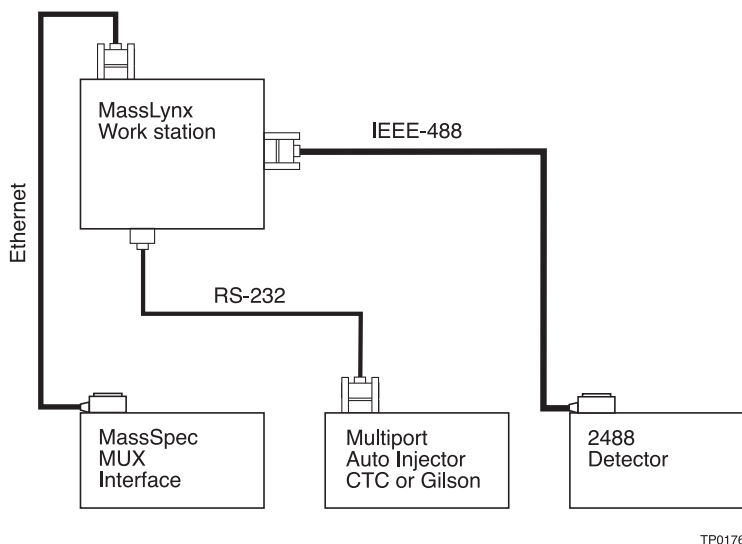


Figure 2-11 Connections to the 2488 Detector in a MassLynx System

Note: When connecting the 2488 Detector to a data system, all detector parameters not configurable by the data system, defer to local control parameters.

Chapter 3

Using the 2488 Detector

Having installed the 2488 Detector, you are ready to operate it.



Attention: Before pumping mobile phase or solvent through the flow cells, perform the procedures in [Section 3.3.2](#).

Using the Detector in a MUX-Based System

The 2488 Detector functions as part of a system based on MUX-Technology and MassLynx software.

To configure the instrument for a particular system:

- Set control parameters, observing the procedure in the system operator's guide.
- Set the detector's IEEE-488 address for remote operation ([Section 3.2.6](#)).

3

3.1 Starting Up the 2488 Detector

3.1.1 Initializing

Install the power supply cord, then switch on the detector, pressing the **On/Off** switch at the lower-right corner. The detector beeps three times as it runs a series of startup diagnostic tests. On successfully completing the tests, it displays results in the Startup Diagnostics screen ([Figure 3-1](#)).

STARTUP DIAGNOSTICS			
TPU	OK	ROM	OK
SCI	OK	RAM	OK
GPIB	OK	LCD	OK
QSPI	OK	CPU	OK

Figure 3-1 2488 Detector Startup Diagnostics Screen

After the Startup Diagnostics screen appears, the detector serially displays the following messages for about 5 minutes:

- Initializing grating
- Initializing system
- Lighting lamp
- Warmup time left: n minutes
- Homing optical filter
- Searching for 656 nm
- Optimizing system performance
- Finding calibration peaks
- Restoring last setup
- Completing initialization

When it finishes initializing, the detector displays the Absorbance Home screen ([Figure 3-2](#)). See [Section 3.2.3](#) for details about this and subsequent screens.

Note: Let the detector warm up for at least 30 minutes before you use it.

3.1.2 Diagnostics Failure

If one or more startup diagnostics fails, the detector beeps and displays an error message. For serious errors, it also displays “Error” in the Home screen’s Absorbance data fields ([Figure 3-2](#)).

[Section 5.1, Error Messages](#), for startup, calibration, operating error messages, and corrective procedures. [Section 5.3.2, Hardware Troubleshooting](#), describes hardware-related causes of startup failures and their corrective actions.

3.2 Using the Operator Interface

3.2.1 Using the Display

The 2488 Detector incorporates a graphic display and keypad operator interface. After startup diagnostics run successfully, the detector displays the Absorbance Home screen ([Figure 3-2](#)).

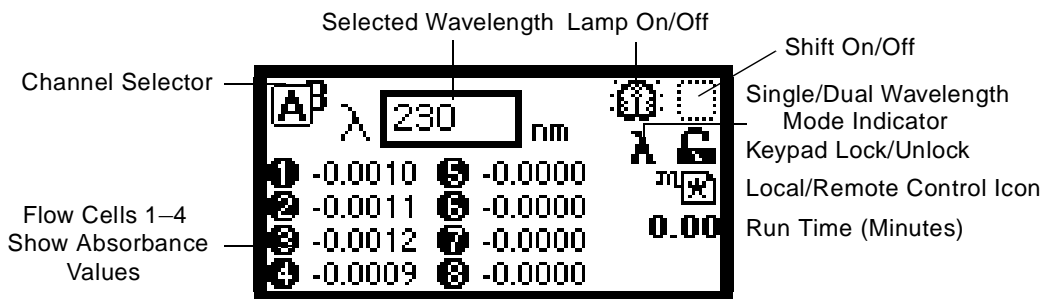


Figure 3-2 Home Screen Set to Monitor Flow Cells 1–8 at 230 nm

Note: The detector method retains the most recent injection's method parameters, updating them at the start of each run.

To redisplay the HOME screen, press **Home**. This screen shows factory-set defaults at the start of the detector's initial run. As the run proceeds, the screen reflects the newly acquired run data.

The Home screen displays absorbance values, in absorbance units (AUs), at one or two wavelengths for flow cells 1–4 (Figure 3-3), 5–8 (Figure 3-4), or 1–8 (Figure 3-2), depending on how you configure the detector (see Section 5.2.2).

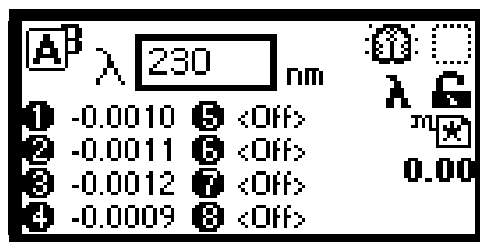


Figure 3-3 Home Screen Set to Monitor Flow Cells 1–4 at 230 nm

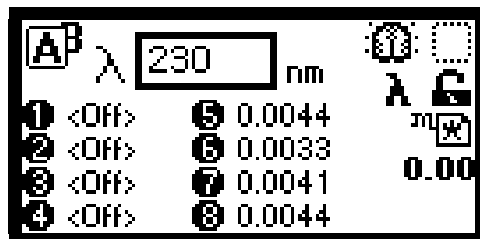


Figure 3-4 Home Screen Set to Monitor Flow Cells 5–8 at 230 nm

Note: Use the **A/B** key to toggle between Home screens for channels A and B.

Home Screen and Message Icons

Table 3-1 Home Screen Icons, Field Names, and Run Functions







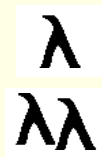










Icon or Field	Icon or Field Name	Function
	Wavelength	Selects the wavelength the detector will monitor for channel A or B. Note: <i>In single wavelength mode, channel B allows no manual control over wavelength selection.</i>
	Channel Selector	Selects a channel to monitor. The channel you select overlaps the unselected one. Displays ON A or ON B to indicate the channel a timed or threshold event is programmed for.
Numeric field	Absorbance	For flow cells 1–4, 5–8, or 1–8, this field displays current absorbance for the selected channel.
	Lamp On / Off	On = Lamp icon. Off = Lamp icon with a superimposed "X".
	Shift On / Off	 (Blank) = Shift off.  = Shift on.
	Single / Dual Wavelength	λ = Detector operating in single wavelength mode. λλ = Detector operating in dual wavelength mode.

Table 3-1 Home Screen Icons, Field Names, and Run Functions (*Continued*)

Icon or Field	Icon or Field Name	Function
 	Keypad Lock / Unlock	An open-padlock icon signifies unrestricted keypad entry for parameter changes. A closed-padlock icon signifies that you cannot change parameters.
	Sticky Diagnostics	Sticky diagnostic. See Section 5.2, User-Selected Diagnostics , for an explanation of sticky diagnostics.
 	Local Control / Remote Control	Remote/IEEE address = When the data system controls the detector via the IEEE-488 bus, a remote control icon contains the IEEE address for channel A. In dual wavelength mode, the IEEE address for channel B is the address displayed as +1.
Numerical field	Run Time (Minutes)	Displays the time elapsed since an inject start signal was received.
    		Message screen icons (from left): Error, Question, Warning, Information, and Standby.

Note: See [Table 3-3](#) for a list of ranges and defaults for function parameters.

3.2.2 Using the Keypad

The 2488 Detector's keypad includes 24 keys ([Figure 3-5](#)).

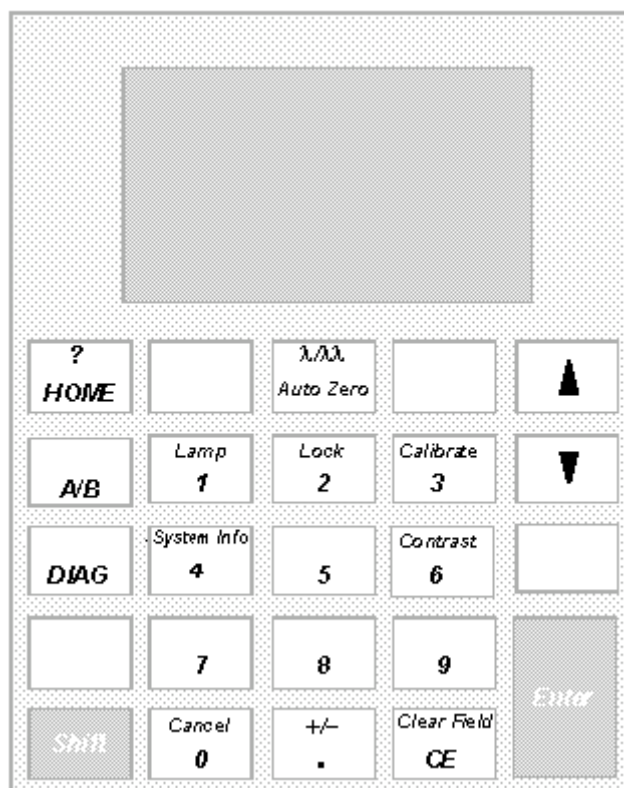


Figure 3-5 2488 Detector Keypad

Use the keypad to perform these tasks:

- Enter numerals (10 digits plus a decimal point)
- Enter, Shift, and Clear Entry
- Navigate
- Select channels
- Display Home, Diagnostics, and System Information screens
- Primary functions
- Secondary functions

For primary functions, press a primary function key, like Auto Zero or Diag.

For secondary functions, press the **Shift** key to invoke the function, like Lamp or Calibrate.

1. Press the **Shift** key and appropriate secondary key.
2. Enter information in parameter fields.
3. Press **Enter**.

Note: Most screens allow the **HOME** and **DIAG** functions anytime. Unlabeled keys do not function.

Selecting from Lists and Menus

For lists and menus numbered 1 to 9, enter the number that corresponds to your choice, then press **Enter**.

For the number 10, select **0**, then press **Enter**.

To go to the end of a list, select **•** (dot). For entries numbered 11 or 12, scroll to your choice, then press **Enter**.

Table 3-2 2488 Detector Keypad Functions

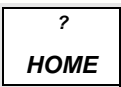
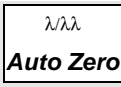


Key	Description	
	Unshifted (Primary Function)	Shifted (Secondary Function)
	HOME – Displays the Home screen.	? – Displays context-sensitive Help, where applicable.
	Auto Zero – Sets absorbance to 0 AU for each channel monitored. Enable or disable Auto Zero from MassLynx software. Note: This key has no effect when Auto Zero is disabled for both channels.	λλλ – Toggles between single and dual wavelength modes. The Home screen icon indicates the current mode.
 	▲ ▼ – Screens with edit, check, or choice fields mark the active field with a thickened border. <ul style="list-style-type: none"> • Use ▲ to activate fields above an active field, or to its left. • Use ▼ to activate fields below an active field, or to its right. On list screens, the keys also move a selection highlight up or down. Other screens, like the Display Contrast screen, indicate special uses for ▲ and ▼ .	

Table 3-2 2488 Detector Keypad Functions (Continued)

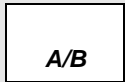



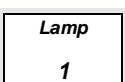
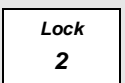
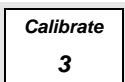
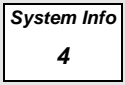
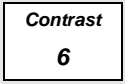
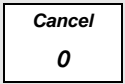
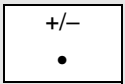
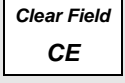

Key	Description	
	Unshifted (Primary Function)	Shifted (Secondary Function)
	A/B – Toggles between channels A and B when it appears in the upper-left corner of the display.	
	DIAG – Displays the diagnostic routines list.	
	Shift – Executes the secondary key functions marked in small type at the top of some keys. <i>Note: The shifted state is temporary. It resets following a single keystroke.</i>	
	0–9 – Numbered keys let you select an item from a numbered list. Simply press the key that corresponds to the item’s number. <i>Note: 0 = 10.</i> The numbered keys also enter a numeral in a current field.	0–9 – See descriptions for individual, shifted numeric keys.
	1 – See 0–9.	Lamp – Displays lamp use statistics for the deuterium lamp. Extinguishes the lamp. An icon on the Home screen indicates the lamp state.
	2 – See 0–9.	Lock – From the Home screen, enables or disables the keypad lock feature. Avoids inadvertently changing detector settings. An icon on the Home screen indicates the current lock state.
	3 – See 0–9.	Calibrate – Initiates the wavelength calibration routine.

Table 3-2 2488 Detector Keypad Functions (Continued)

Key	Description	
	Unshifted (Primary Function)	Shifted (Secondary Function)
	4 – See 0–9.	System Info – Displays the following system information: <ul style="list-style-type: none"> • Software version • IEEE address • Instrument serial number • Checksum
	6 – See 0–9.	Contrast – Lets you adjust contrast (viewing angle) in the liquid crystal display.
	0 – See 0–9.	Cancel – In some modes, quits a prompt without completing the current task. The word “Cancel” appears as a cue in the lower-right border of the message text, complementing this action.
	• – Inserts a decimal point. Places the cursor at the last entry in a list.	+/- – Reverses the sign of a number in an active field.
	CE – Clears an in-process edit, reintroducing a field's previous content. Sets the value of certain fields to a unique word. Thus, to change the voltage offset diagnostic to OFF, enter either a numeric offset value or press CE .	Clear Field – Removes the current entry field before you enter the desired values.
	Enter – Completes an edit field entry. Moves the active (highlighted) field, as when you press the Down arrow. Note: Does not do so immediately following a change to the Home screen wavelength setting. Acknowledges error messages and other prompts. In such cases, “Enter” appears as a cue in the lower-right border of the message text.	

3.2.3 Using the Operator Interface

To use the operator interface:

1. Press the **Enter** key or the ▲ and ▼ keys to navigate a screen's editable fields.

Note: An active field is highlighted by a thickened border.

2. Press **Enter** to advance the border highlight to the next editable field in a sequence. If you err, press **CE**. This reverses changes and returns you to the active entry field.
3. Press **Enter** again to display an active field's list. (A numeral at the right of an active field indicates the field contains a list of items.)

Note: When you know an item's number, you do not need to press Enter to display the item list. Instead, press the appropriate numbered key.

4. Select an item from the list, observing either of the following methods:
 - Press a numbered key that corresponds to the item's number.
 - Use ▲ and ▼ to find the listed item, and then press **Enter**.

Note: ▲ and ▼ do not increase or decrease numeric field values. For that function, use the numbered keys.

3.2.4 Initial Setup

To prepare the detector for operation:

1. Press **HOME**. The Home screen appears ([Figure 3-2](#)).
2. Press **λλ** to select the single or dual wavelength mode.
3. Let the detector warm up for 30 minutes.

3.2.5 Modify Instrument Method Parameters in MassLynx Software

To operate a system that includes the 2488 Detector, create an instrument method in the the Modify Instrument Method window ([Figure 3-6](#)). Three pages accept method parameters for your run: Wavelength A, Wavelength B, and Events.

Wavelength A or Wavelength B Page

Choose among the following parameters on the Wavelength A or Wavelength B page:

- **Single, Dual** – Click one or the other to operate in single or dual wavelength mode.
- **Wavelength** – Specifies the wavelength(s) you want to monitor. The range is 190 to 700 nm. The default is 254 nm.
- **Sampling Rate** – Specifies how many points the detector samples per second.

- **Data Mode** – Specifies a data mode. In single wavelength mode, the data mode is AbsorbanceA. For dual wavelength mode, it is AbsorbanceA (channel A) *and* AbsorbanceB (channel B).

Note: In dual mode, the sampling rate is always 1 ([Figure 3-7](#)).

- **Noise Filter Type** – Specifies a noise filter type.
- **Time Constant** – Adjusts the noise filter's time constant for optimum signal-to-noise performance without changing the sensitivity setting. The range is as follows:
 - Hamming: 0.1 to 5.0 (λ)
 - Hamming: 1 to 50 ($\lambda\lambda$)
 - RC: 1 to 99

Specify **0** to disable filtering. The default is 1.0. For details, see [Section 1.2.3](#).

- **Run Time** – Specifies the total run time, in minutes, for multichannel absorbance data.

Note: Always set timed events to operate within the method's run time.

- **(Auto Zero) On Wavelength Changes** – Automatically zeros the detector for each wavelength change.



Attention: This parameter is automatically selected. Except when you perform specific advanced operations, Micromass and Waters recommend it remain selected. If you clear this function, significant variances in measured absorbance can occur after each wavelength change.

- **(Auto Zero) On Inject Start or Auto Zero Event In or Auto Zero Key Press** – Selected by default, this parameter executes an auto zero each time it receives an inject start signal via contact closure, IEEE-448, or the keypad. Select the check box to disable this parameter from operating in either or both channels.
- **Description** – Enter comments on run features in this text field.

Note: For programming timed events, see [Section 3.3.6](#).

In [Figure 3-6](#), single wavelength mode is selected to monitor wavelength A. This mode lets you choose one of three sampling rates: 1, 2, or 5 points/second.

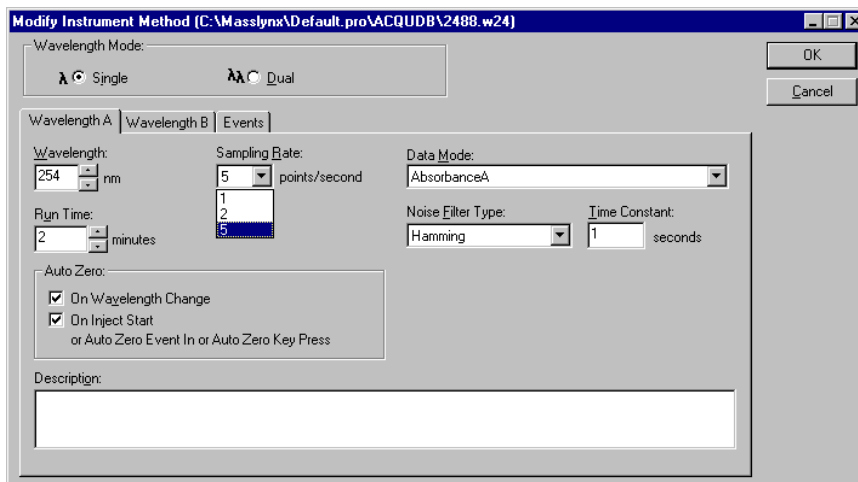


Figure 3-6 Modify Instrument Method Set for Single Wavelength Monitoring

In [Figure 3-7](#), dual wavelength mode is selected to monitor wavelength B. Unlike single wavelength mode, which offers a choice of four sampling rates, dual wavelength mode requires a 1 point/second sampling rate.

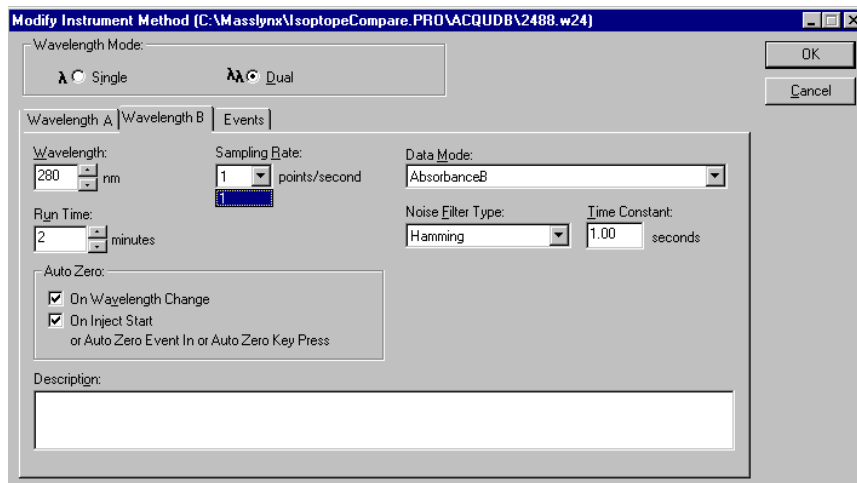


Figure 3-7 Modify Instrument Method Window Set for Dual Wavelength Monitoring

Events Page

Choose among the following parameters on the Events page ([Figure 3-8](#)) to change method parameters during a specified run time:

- **At Time** – Specifies how many minutes into a run the parameter change occurs.
- **Event** – Specifies the event being changed: Wavelength, Lamp, Time constant, or Switch 1 or 2.
- **Change to** – Specifies the event's new value. Values can be as follows:

Event	Value
Lamp	On or Off
Wavelength	190 to 700 nm
Switch 1 or 2	High, low, or pulse
Auto Zero	(no setting)
Time constant	"Wavelength A or Wavelength B Page"

- **Wavelength** – specifies the channel being changed: 1 for Wavelength A and 2 for Wavelength B.

[Figure 3-8](#) shows the Events page, accessed by clicking the Events tab in the Modify Instrument Method window. The page allows timed events viewing. In this case, the timed events are two wavelengths, one changing on each channel.

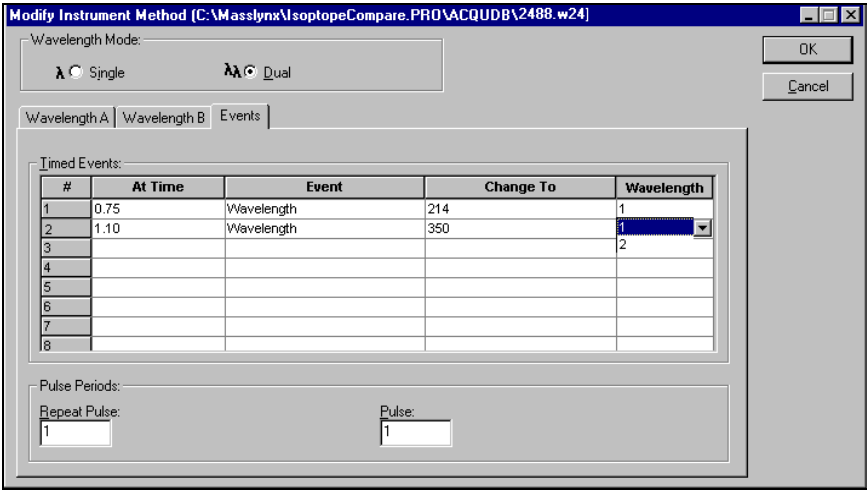


Figure 3-8 Modify Instrument Method Window's Events Page with Dual Mode

[Figure 3-9](#) again shows the Events page. Besides the two wavelengths in [Figure 3-8](#), it lists other timed events.

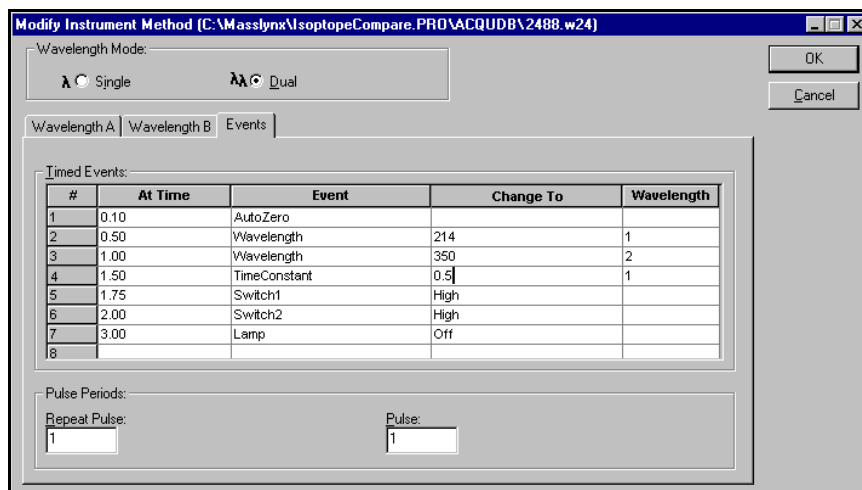


Figure 3-9 Modify Instrument Method Window's Events Page Showing Multiple Timed Events

[Figure 3-10](#) details the Events page drop-down list. This list of event choices appears when you click the Event column.

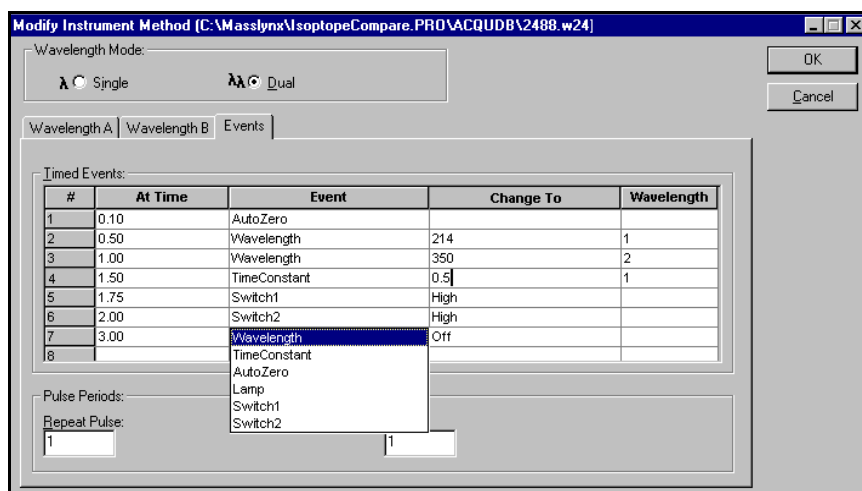


Figure 3-10 Modify Instrument Method Window's Events Page Showing Event List

3.2.6 Configuring the 2488 Detector

Configuration entails specifying settings for these items:

- IEEE address
- Inputs
- Pulses

Access the Configuration Menu Screen by pressing **DIAG**, then select **Configure**. The Configuration Menu screen appears ([Figure 3-11](#)).

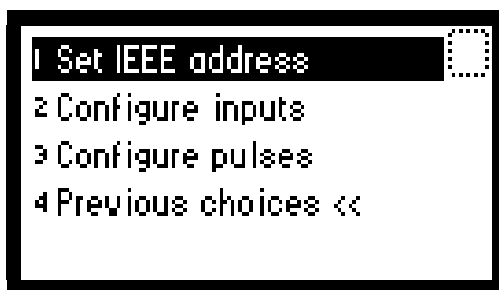


Figure 3-11 Configuration Menu Screen

Configuring the IEEE Address

The 2488 Detector's default address is IEEE-488, remote address 4.

To reconfigure the default:

1. Press **1** on the Configuration Menu screen. The first of three Configuration screens appears ([Figure 3-12](#)).

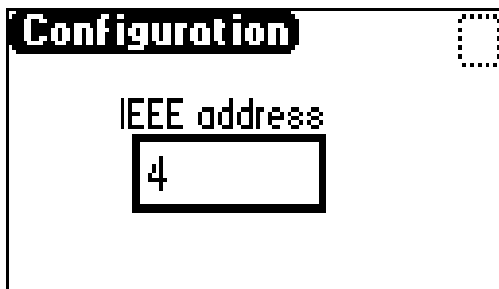


Figure 3-12 First Configuration Screen

2. Enter a numeral from 2 to 29 as the new IEEE-488 remote bus address.

Note: To remove the detector from IEEE control, clear the field by pressing **CE**.

Configuring Inputs

To specify switched input and output settings:

1. Press **2** on the Configuration Menu screen. The second Configuration screen appears ([Figure 3-13](#)).

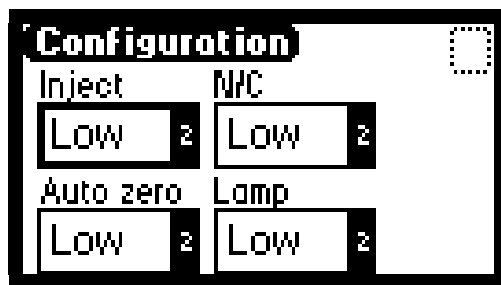


Figure 3-13 Second Configuration Screen

Three editable event fields—Inject, Auto zero, and Lamp—specify switched input and output settings for their events. (The fourth field, N/C, is not configured.)

2. Enter appropriate settings in the Inject, Auto zero, and Lamp fields. The following section describes the settings.

Settings

Inject – Specify the Inject input that signals a run's start. This event resets the run-time clock and immediately applies one of these initial method conditions:

- **High** – Start the run when contact closure changes from off (open) to on (closed).
- **Low** – Start the run when contact closure changes from on (closed) to off (open). This is the default.
- **Ignore** – Do not respond to Inject start input.

Press **Enter** or the **▲** and **▼** keys to select the appropriate entry. The default setting is Low.

Auto zero – Configure the auto-zero input as follows to automatically set absorbance readings on channel A and/or channel B to zero:

- **High** – Automatically set the channel to zero when contact closure changes from off (open) to on (closed).
- **Low** – Automatically set the channel to zero when contact closure changes from on (closed) to off (open). This is the default.
- **Ignore** – Do not respond to auto-zero input.

Determine the channel response via the auto-zero function (explained in [Table 3-3](#)), and press **Enter**, or the **▲** and **▼** keys, to select the appropriate entry. The default setting is Low.

- **N/C** – Not configured

Lamp – Configure the Lamp input level as follows to turn the deuterium lamp on or off via an external device:

- **High** – Turn lamp on when contact closure is on (closed).
- **Low** – Turn lamp on when contact closure is off (open).
- **Ignore** – Do not respond to Lamp input. This is the default setting.

Press **Enter**, or the **▲** and **▼** keys, to select the appropriate entry. The default setting is Ignore.

Note: To configure the flow cells you set for monitoring, see [Section 5.2.2](#).

Configuring Pulse Periods

To specify a single pulse (signal width), or activate a pulse or rectangular wave on SW1 or SW2, press **3** on the Configuration Menu screen. The third Configuration screen appears ([Figure 3-14](#)).

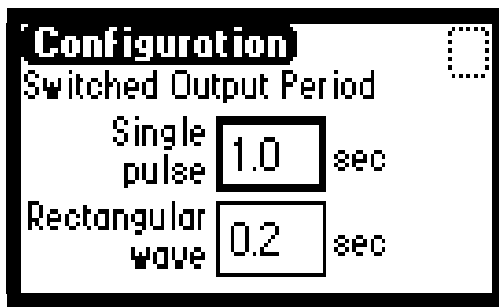


Figure 3-14 Third Configuration Screen

Single pulse – Specify the signal period, or single pulse width, in this field when programming SW1 or SW2 is to generate a pulse as a timed or threshold event. The range is 0.1 to 60 seconds.

Rectangular wave – Specify the signal period (the width of one pulse period in a rectangular wave or pulse train) in this field when programming SW1 or SW2.

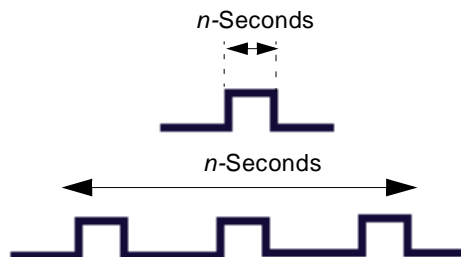


Figure 3-15 Setting the Pulse (Signal Width) with SW1 or SW2

Setting Display Contrast

The Contrast function lets you adjust screen contrast. Press **Contrast**. The Display Contrast screen appears ([Figure 3-16](#)).

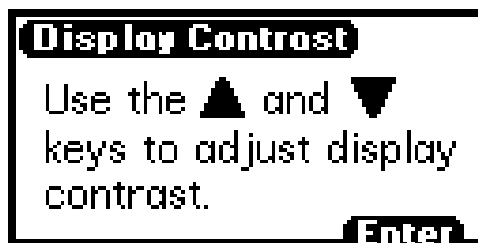


Figure 3-16 Display Contrast Screen

Use the ▲ and ▼ keys to adjust the contrast of the display.

Displaying System Info

Press **System Info** to display information about the detector, including the serial number, software version, and the IEEE address, if applicable ([Figure 3-17](#)).



Figure 3-17 System Info Screen

Using Help

Limited context-sensitive Help accompanies the 2488 Detector. Where Help accompanies a particular function, pressing **?** displays the Help screen ([Figure 3-18](#)). If no Help accompanies the function, pressing **?** yields no response.

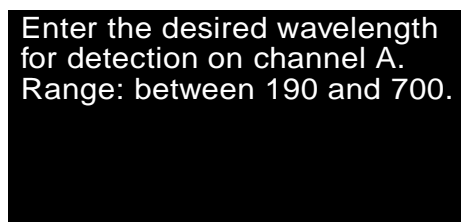


Figure 3-18 Help Screen

3.3 Operating the 2488 Detector

3.3.1 Overview of Detector Operation

Note: To prevent oxygen reabsorption, run the solvent degasser continuously, particularly when operating the detector at wavelengths below 230 nm.

Operating Modes

The detector monitors a range of 190 to 700 nm in either single or dual wavelength mode. On startup, it defaults to the mode selected when the instrument was last switched off.

Remote Control via MassLynx Software

As part of a Micromass MUX-Technology system under MassLynx control, the detector uses the IEEE-488 bus connector ([Section 2.9.3](#)).

To connect the detector to your HPLC system, see [Chapter 2](#).

The Home screen ([Figure 3-2](#)) displays the remote control icon and IEEE-488 address when MassLynx controls the detector.

See [Section 2.9, Making Signal Connections](#), for more information on connecting the 2488 Detector to an external system.

3.3.2 Verifying the Detector

After installation, follow the procedures in this section to confirm that the detector operates properly.

Before You Begin

Micromass and Waters ship the 2488 Detector dry. You must therefore pump solvent through it before using it. First, flush the lines with filtered, degassed, HPLC-grade methanol. Then, barring miscibility problems, pump mobile phase at 1 mL/min for at least 15 minutes.

Note: *To ensure accurate verification, be sure to start up the 2488 Detector and follow steps 1 to 3 in this section and 1 to 4 in "Recording Sample and Reference Beam Energies" before pumping any mobile phase or solvent through the flow cell.*

1. Connect the detector to the data system. Refer to [Chapter 2](#) for details about connecting the detector to external devices.
2. Switch on the detector. The display shows initialization messages for about 5 minutes ([Section 3.1](#)).

After initializing, the Home screen appears ([Figure 3-2](#)).

3. Allow the detector to warm up for at least 30 minutes before using it.

If the startup verification diagnostics indicate a failure, note the error message to determine the appropriate corrective response, and consult [Chapter 5](#).

Recording Sample and Reference Beam Energies

Record the baseline sample and reference beam energies. Use these values to determine whether, in future runs, the following conditions affect results:

- Contaminated solvent
- Contaminated flow cell
- Defective lamp
- Air bubble in flow cell

To record sample and reference beam energies:

1. Set the wavelength to 230. (First, highlight the Home screen's λ field. Then, type **230**, and press **Enter**.)
2. Press **DIAG 2** and select **Sample & ref energy**. The Sample and Reference Energy Diagnostic screen appears ([Figure 3-19](#)).

A	Energy	λ	230	nm
Samples:	12.81	12.33		nA
Ref:	11.63	11.34		
	12.08	11.66		
124.89	nA	12.26	11.38	

Figure 3-19 Sample and Reference Energy Diagnostic Screen

3. Record the sample and reference energies for 230 nm for future reference.

Note: The Diagnostics screen displays values for all flow cells, regardless of which you selected to monitor.



Attention: Always run this procedure when you change the lamp. See [Section 4.6, Replacing the Lamp](#), for instructions on how to change the deuterium lamp.

4. Flush the flow cell with 30 to 60 mL of HPLC-grade methanol, setting the flow rate to 1 mL/min for at least 15 minutes.

Note: Depending on the flow cell configuration you choose (1–4, 1–8, or 5–8) use flow cells 1 or 5 to measure reference energy (see [Section 5.2.2](#)).

5. Verify peak response according to the verification test, below. If verification fails, repeat the test.

Peak Response Verification Test

Note: Before you begin the test, connect liquid tubing following instructions in [Section 2.5, Connecting Liquid Lines](#).

To test the detector's peak response:

1. Set the wavelength to 254 nm.
2. Press **Enter** to activate the sensitivity field.
3. Set the pump flow rate to **1.0 mL/min**.

4. Inject 1 μL of acetone.

The detector passes the test when the data system displays a peak.

3.3.3 Calibrating the Wavelength

Use the keypad's Calibrate key to recalibrate the detector during a run, or when calibration errors appear on startup.

Note: *You need not reboot the detector after a successful wavelength calibration.*

To manually calibrate the detector:

1. Press **Shift-Calibrate**. A prompt appears ([Figure 3-20](#)), reminding you to make sure flow cells are prepared.



Attention: *If flow cells are not clean, flush them with a colorless solvent — Micromass and Waters recommend HPLC-grade methanol or water — before proceeding.*

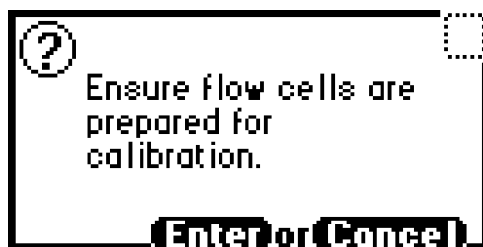


Figure 3-20 Wavelength Calibration Message

2. Press **Enter** to continue the calibration. (Press **Cancel** to return to the Absorbance Home screen without calibrating.)

When you press **Enter**, the detector displays a series of initialization messages. If the calibration succeeds, the detector beeps three times and displays the maximum error, in nanometers ([Figure 3-21](#)). This value constitutes the furthestmost calibration shift relative to the previous calibration.

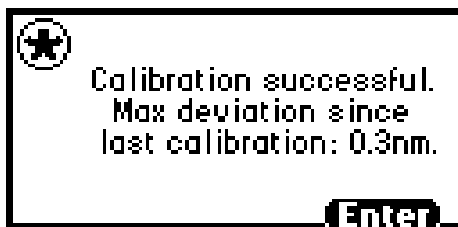


Figure 3-21 Successful Calibration Message

3. Press **Enter** to finish calibrating.

A “Calibration complete” message briefly appears. Other messages such as “Optimizing system performance” and “Restoring last setup” may appear before the Absorbance Home screen reappears.

Note: A successful calibration removes any error message the Absorbance Home screen previously displayed.

4. If the calibration fails, try again. If another failure results, switch off the detector, wait 5 seconds, and restart it. If the calibration fails yet again, consult [Chapter 5, Error Messages, Diagnostics, and Troubleshooting](#).

3.3.4 Operating the Detector in Single Wavelength Mode

The 2488 Detector is optimized for single wavelength operation, the default operating mode. To implement single wavelength mode:

1. If the detector is in dual wavelength mode, press $\lambda\lambda\lambda$ from the Home screen to toggle from dual to single mode. A momentary message indicates the detector is switching to single wavelength operation ([Figure 3-22](#)).

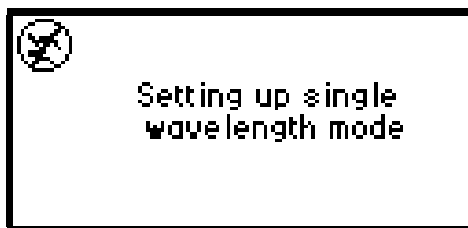


Figure 3-22 Wavelength Mode Change Message (Dual to Single Wavelength)

2. Enter the wavelength you want to monitor in the λ field, then press **Enter**. (You can also configure wavelength in the MassLynx Method screens.)

Note: The detector automatically applies the second-order filter for all wavelengths ≥ 370 nm.

3.3.5 Operating in Dual Wavelength Mode

Dual wavelength mode lets you select a different wavelength to monitor.

Changing from Single to Dual Wavelength Mode

To change from single to dual wavelength mode:

1. If the detector is in single wavelength mode, press λ/λ from the Absorbance Home screen to toggle from single to dual mode. A momentary message indicates the detector is switching to dual wavelength operation. The message "Setting up dual wavelength mode" appears ([Figure 3-23](#)).

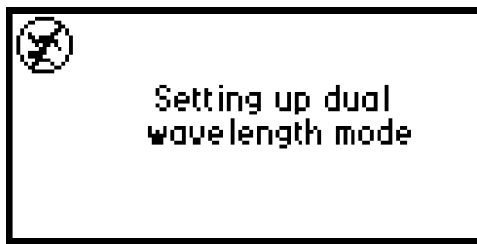


Figure 3-23 Wavelength Mode Change Message (Single to Dual Wavelength)

2. Enter the wavelength to monitor in the λ field, then press **Enter**.
3. Press **A/B** to switch channels, if necessary. The Absorbance Home screen for the previously inactive channel appears. (You can also configure wavelengths in the MassLynx Method screens.)

Note: For information about programming timed and threshold events, see [Section 3.3.6](#).

Blocking Second-Order Effects

The 2488 Detector applies a second-order filter when both selected wavelengths are greater than 370 nm (± 1 nm). However, if only one of the two wavelengths is greater than 370 nm (± 1 nm), the detector does not apply the filter. Instead, it displays a warning that data collected for the higher wavelength may be inaccurate due to possible UV light interference ([Figure 3-24](#)). These inaccuracies may evidence themselves as additional peaks and inaccurate peak areas. You should therefore select a wavelength pair that falls above or below the 370 nm threshold.



Figure 3-24 Warning Indicating Wavelengths Span 370 nm

3.3.6 Controlling Timed Events with MassLynx Software

When the MassLynx software controls the detector, the Home screen displays a remote control icon that contains the IEEE-488 address ([Figure 3-2](#)).

You can program timed events to the nearest 0.01 minute via the MassLynx software. As you enter timed events, each new entry is added to the end of the timed event list. If you enter an out-of-sequence time, relative to those you previously entered, the software orders it automatically. You can set the timed events shown in [Table 3-3](#).

Note: Always set timed events to operate within the method's run time.

Table 3-3 Timed Event Parameters

Number	Event	Units	Range or Default	Specify Channel
1.	wavelength	nm	190 to 700	yes
2.	time constant	seconds	0: Disable filter Hamming: λ 0.1 to 5.0, $\lambda\lambda$ 1 to 50 RC: 1 to 99 seconds	yes
3.	auto zero	does not apply	does not apply	yes
4.	lamp	1. off 2. on	off	no
5.	switch 1	1. high 2. low 3. pulse 4. rect wave	high	no

Table 3-3 Timed Event Parameters (Continued)

Number	Event	Units	Range or Default	Specify Channel
6.	switch 2	1. high 2. low 3. pulse 4. rect wave	high	no

3.3.7 Conserving Deuterium Lamp Life

To conserve lamp life without shutting down the detector, leave the instrument on, and use the Lamp key to switch off only the lamp.

When you switch off the lamp, the Home screen displays “Lamp off,” and an “X” superimposes the lamp icon. To manually switch the lamp on, press **Lamp**. As the lamp ignites, the Home screen displays the word “Igniting.” Once it has ignited, the Home screen displays the lamp icon. [Figure 3-26](#) illustrates the lamp-off and lamp-on sequence.

Use the Lamp key to:

- Switch the lamp off and on manually
- Display the lamp control screen ([Figure 3-25](#)), which indicates:
 - Time, in hours and minutes, the lamp has operated since the most recent startup
 - Total time, in hours and minutes, the lamp has operated since its installation
 - How many times the lamp has fired

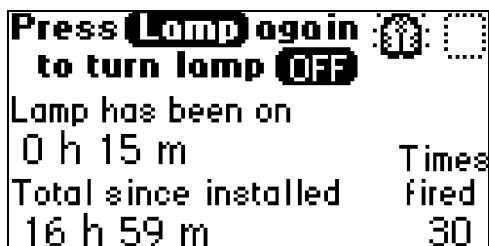


Figure 3-25 Lamp Control Screen

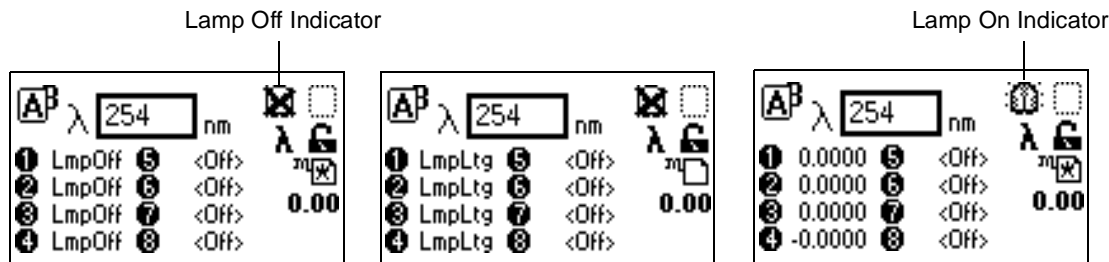


Figure 3-26 Lamp Lighting Sequence

You can further conserve lamp life by programming it to turn on and off automatically via a timed event. Select the **Timed events** option from the Method choice list. See [Section 3.3.6](#) and [Table 3-3](#) for details.

Alternatively, you can program the lamp through one of the external contact closures. See [Section 3.2.6](#) for details.

3.3.8 Shutting Down the 2488 Detector

Before you switch off the detector, remove any buffered mobile phase from the fluid path.

You can create a shutdown method that includes a timed event to turn the lamp off in the MassLynx Timed Events Method Editor screen.

Removing Buffered Mobile Phase



Attention: To avoid damaging the column, remove it before you perform the following procedure.

To remove mobile phase:

1. Replace the buffered mobile phase with 100% HPLC-quality water and flush the system for 10 minutes at 3 mL/min.
2. Replace the 100% water mobile phase with a solution of 90% methanol:10% water, and flush the system for 10 minutes at 3 mL/min.

Follow documented procedures for injector purging and pump priming.

Shutting Down the 2488 Detector Manually

Use the **On/Off** switch to shut down the detector.

Chapter 4

Maintenance Procedures

4.1 Introduction

Read the appropriate maintenance instructions before starting the procedures in this chapter. If you are uncertain about how to perform the procedures, call the Waters Technical Service Department to have a trained service representative perform the procedure.

Waters Technical Service

For problems with HPLC equipment, computer software, or hardware other than for the Waters 2488 Multichannel Absorbance Detector, consult the documentation for the applicable instrument or program.

If you encounter a problem with the 2488 Detector that you cannot troubleshoot, contact Waters Technical Service at 800 252-4752, *U.S. and Canadian customers only*. Other customers, call your local Waters subsidiary or your local Waters Technical Service Representative, or call Waters corporate headquarters for assistance in Milford, Massachusetts (U.S.A.).

If you are a Micromass customer, please contact Micromass at +44 161 946 2400, or +44 161 926 7200, or +44 161 435 4100 or call your local Micromass subsidiary.

Spare Parts

Stock the recommended spare parts to minimize downtime. Refer to [Appendix B, Spare Parts](#), for a list of recommended spare parts. Appendix B lists only those parts replaceable by the user. Parts not included in the list require replacement by a trained service representative.

Safety Precautions

When you perform the maintenance procedures in this chapter on your 2488 Detector, keep the following safety considerations in mind:



Caution: *To prevent possible electrical shock, never disconnect an electrical assembly (including all interface cables) while power is applied to the 2488 Detector.*



Caution: When you handle eluents, change tubing, or operate the 2488 Detector in general, always observe good laboratory practices. Know the physical and chemical properties of the eluents. Refer to the Material Safety Data Sheets for the eluents in use.



Caution: To avoid possible electrical shock, always turn power to the 2488 Detector off and disconnect the power cord before performing maintenance procedures.

Removing the Left Front Panel Cover

Note: You can use the left front panel cover to hold small parts when removing and replacing the 2488 Detector flow cell.

To remove the left panel front cover:

1. Holding onto the top of the cover, gently pull the bottom away from the unit.
2. Gently remove the top of the cover and store it nearby.

[Figure 4-1](#) illustrates the 2488 Detector with the left front panel cover removed.

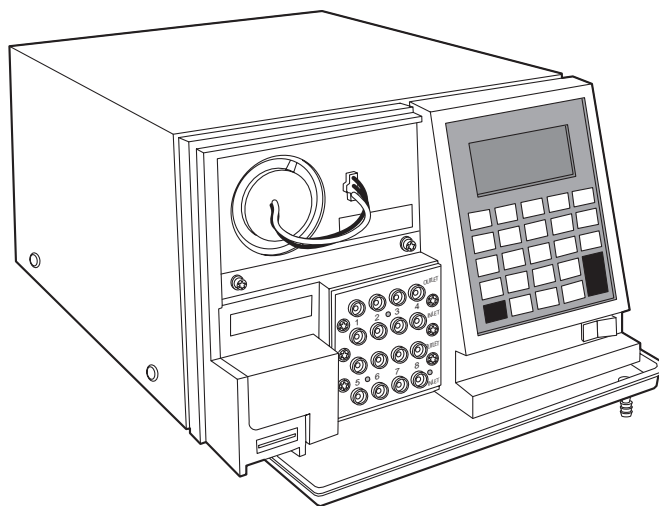


Figure 4-1 2488 Detector without Left Front Panel Cover

4.2 Routine Maintenance

The Waters 2488 Multichannel Absorbance Detector requires minimal routine maintenance. To achieve optimal performance:

1. Replace solvent reservoir filters in your HPLC system regularly.
2. Filter and degas solvents to prolong column life, reduce pressure fluctuations, and decrease baseline noise.
3. Flush buffered mobile phases out of the detector with HPLC-grade water followed by methanol/water 90:10 solution each time the detector is shut down. This process prevents:
 - Plugging of the solvent lines and flow cell
 - Damaging the components
 - Microbial growth

4.3 Inspecting, Cleaning, and Replacing Parts of the Flow Cells

A dirty flow cell can cause baseline noise, decreased sample energy levels, calibration failure, and other problems with operation of the 2488 Detector.



Attention: Do not clean or replace a broken lens. The optical components in a flow cell assembly are miniaturized and difficult to reassemble. Replace the flow cell cartridge.

4.3.1 Flushing the Flow Cells

Flush the flow cell after each shutdown, or when dirty.

Note: If the 2488 Detector is connected to another detector downstream, disconnect the 2488 outlet line.

To flush the flow cells:

1. Discontinue the eluent flow, and remove the column(s).
2. Attach tubing from the 2488 Detector to the injector outlet (from which the column has been removed). Flush the mobile phase from the detector with a miscible solvent and water (unless the mobile phase is miscible with water).
3. Flush the detector with HPLC-quality water to remove contaminants from the flow path.

4. Auto zero the detector at 230 nm.
5. Pump 6 M nitric acid through the flow cell to clean the internal pathways.
6. Flush again with HPLC-quality water until the solvent is neutral (or when absorbance at 230 nm is close to 0 and not varying).
7. Reattach the column to resume flow of the mobile phase. Use an intermediary solvent if you are using a mobile phase that is not miscible in water.



Attention: *If only four flow cells (4-way MUX) are used in a system and one flow cell fails, you can use the second bank of four flow cells. In the Select Flow cells diagnostic screen, select flow cells 5–8, and change the liquid lines to the other bank (see [Figure 5-4](#)).*

4.3.2 Removing the Flow Cell Assembly with Manifold and Optics Block

Before You Begin

Follow these precautions when disassembling and reassembling the flow cell assembly with manifold and optics block.

- Avoid scratching the optical block polished surfaces.
- Use a clean nonparticulating cloth or a similar clean surface in the work area where you are removing, rebuilding, and replacing the components of the flow cell assembly with manifold and optics block.

Required Materials

You need the following items to remove and replace the flow cell:

- Torx® 10 and 20 drivers
- M3 Allen wrench
- Small screwdriver

Save all parts after removal; you will need them when you complete your repairs.

Removing the Flow Cell Assembly

To remove the flow cell assembly:

1. Power down the 2488 Detector.
2. Disconnect all tubing attached to the front of the manifold and remove the bezel cover ([Figure 4-2](#)).
3. Use a Torx 10 driver to remove the two bezel cover screws ([Figure 4-2](#)), then lift the bezel cover straight up to remove.

4. Use a Torx 20 driver to remove the four screws attaching the manifold to the optics bench ([Figure 4-2](#)).

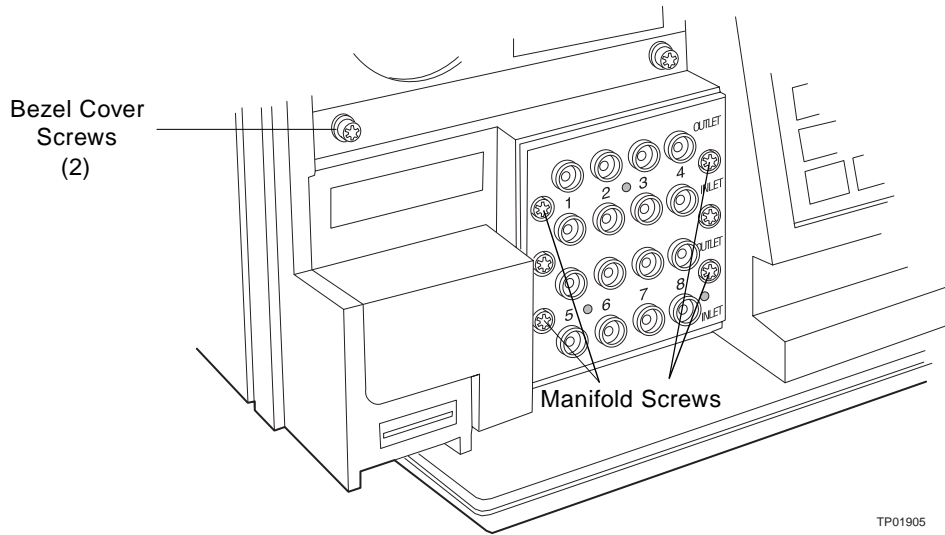
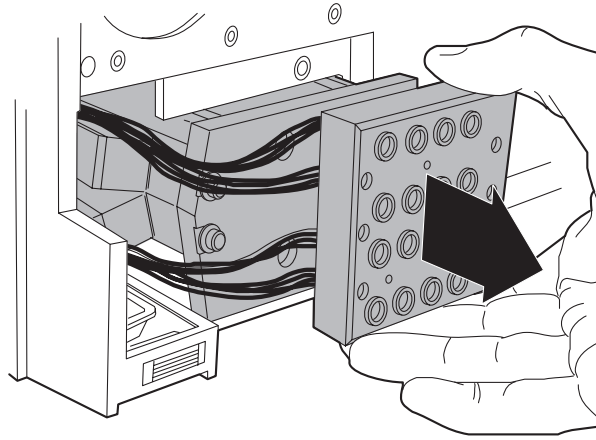


Figure 4-2 Bezel Cover and Manifold Screws

5. Grasp the manifold assembly and cautiously slide it forward ensuring that the wires do not catch on the chassis during removal. The wires are only long enough to allow you to detach the cables from the assembly. Do not strain the cables ([Figure 4-3](#)).

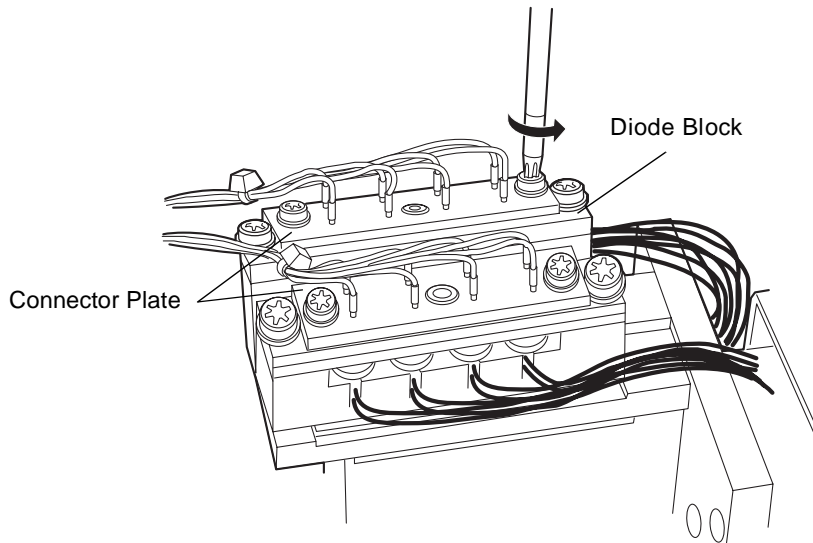
Note: If the assembly is difficult to remove, grasp the assembly and rock it back and forth until you can remove the assembly easily.



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Figure 4-3 Removing the Flow Cell Assembly with Manifold and Optics Block

6. Disconnect cables from the assembly by removing the two smaller screws with a Torx 10 driver ([Figure 4-4](#)).



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Figure 4-4 Disconnecting Cables

7. Use a small screw driver to pry the connector plate away from the diode block.

Note: *There are two screws per bank of channels.*

You can work on the optical flow assembly as a separate module.

4.3.3 Reassembling the Flow Cell Assembly with Manifold and Optics Block

Follow these precautions when disassembling and reassembling the flow cell assembly with manifold and optics block:

- Avoid scratching the optical block assembly polished surfaces.
- Use a clean nonparticulating cloth or a similar clean surface in the work area where you are removing and replacing components of the flow cell assembly with manifold and optical block.

Reassembling the Flow Cell Assembly

To reassemble the flow cell assembly:

1. Line up sockets on the connector plate to the diode block leads. Slide the cable connector marked 5–8 on the bottom bank and 1–4 on the top bank and press the connector block into place.
2. Use a Torx 10 driver to replace the two screws to attach the connector plate to the diode block.
3. Grasp the manifold and slide it onto the optics assembly pins. Check the left side to ensure that the wires retract properly.
4. Use a Torx 20 driver to replace the four screws with lock washers evenly and attach the manifold to the optics bench.
5. Use a Torx 10 driver to replace the two screws and reattach the bezel cover.

4.3.4 Replacing the Flow Cell Cartridge



Attention: *To prevent contamination, particularly in the area of polished fiber optics blocks, use powder-free finger cots or gloves when disassembling, inspecting, cleaning, or replacing parts in the optical flow assembly.*

The 2488 Detector is shipped with eight flow cells, the top bank contains channels 1–4 and the bottom bank contains channels 5–8. If one flow cell is damaged, you can switch to the other flow cell bank; i.e., if flow cell 2 is damaged, you can switch to the bottom bank of flow cells containing channels 5–8.

Before you begin:

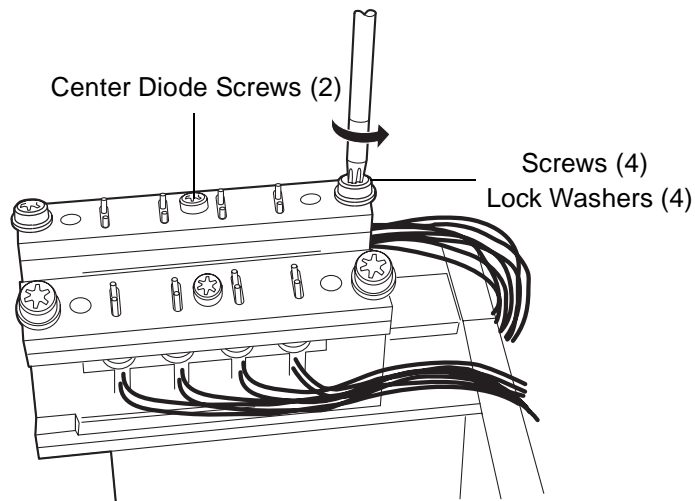
1. Unpack and inspect the new flow cell cartridge.
2. Flush and dry the flow cell that will be replaced ([Section 4.3.1](#)).

3. Remove the flow cell assembly with manifold and optics block ([Section 4.3.2](#)).
4. Identify which flow cell bank needs to be replaced: the top bank contains channels 1–4 or the bottom bank contains 5–8.

To replace the flow cell cartridge:

1. Use a Torx 20 driver to remove the two outer screws and the diode assembly ([Figure 4-5](#)).

Note: Do not remove the center screw ([Figure 4-5](#)) which holds the diode assembly together.



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Figure 4-5 Removing the Diode Assembly

2. Remove the flow cell cartridge from the flow cell block. Follow the tubes to the manifold block ([Figure 4-6](#)).

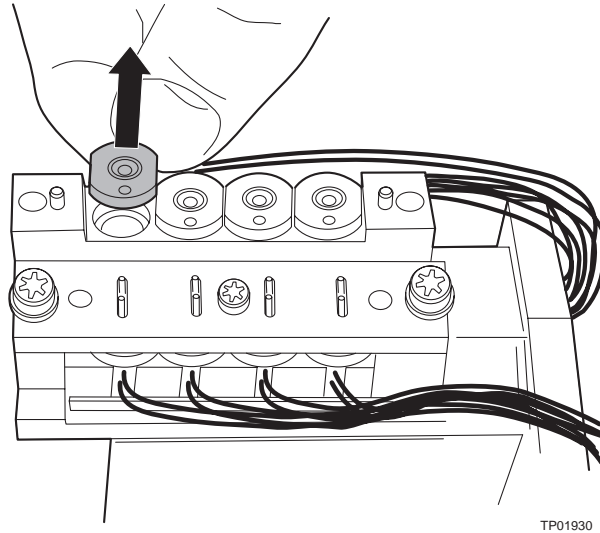


Figure 4-6 Removing the Flow Cell Cartridge from the Block

3. Unscrew the compression screw (located on the back side of the manifold) of the flow cell that is being replaced. You may be able to unscrew the compression screw without disconnecting the manifold from the flow cell assembly.

4. If the compression screw is not accessible, unscrew the three shoulder screws using the M3 Allen wrench ([Figure 4-7](#)).

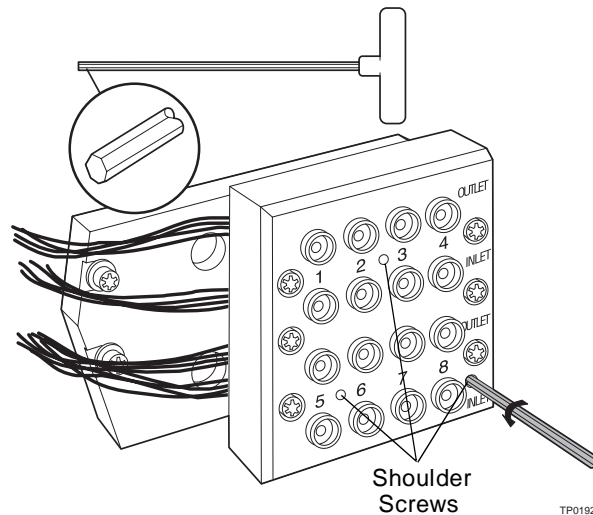


Figure 4-7 Detaching the Manifold from the Optical Assembly

Note: Within the manifold block, the manifold screws are concealed behind the manifold cover. On the back of each screw there is a compression spring.

4.3.5 Reassembling the Flow Cell Cartridge

To reassemble the flow cell cartridge:

1. Using the replaced flow cell as a bending template, bend the tubes of the new flow cell.
2. Slide the flow cell cartridge in the flow cell block.
3. Place the diode assembly over the flow cells and attach with the outer screws ([Figure 4-5](#)).

Note: Ensure the flow cell block is flush with the optical block and the screws are tightened firmly.

4. Connect the tubes to the manifold block.

Note: You must orient the tubes correctly: the tubing brazed on the top of the flow cell must connect to the union labeled "outlet".

5. Attach the manifold to the fiber optics block with the three (concealed heads) shoulder screws. You must replace the compression spring over the shoulder screws before you attach the manifold.
6. Reassemble the flow cell assembly with the manifold and optics block ([Section 4.3.3](#)).

4.4 Replacing the Union in the Manifold



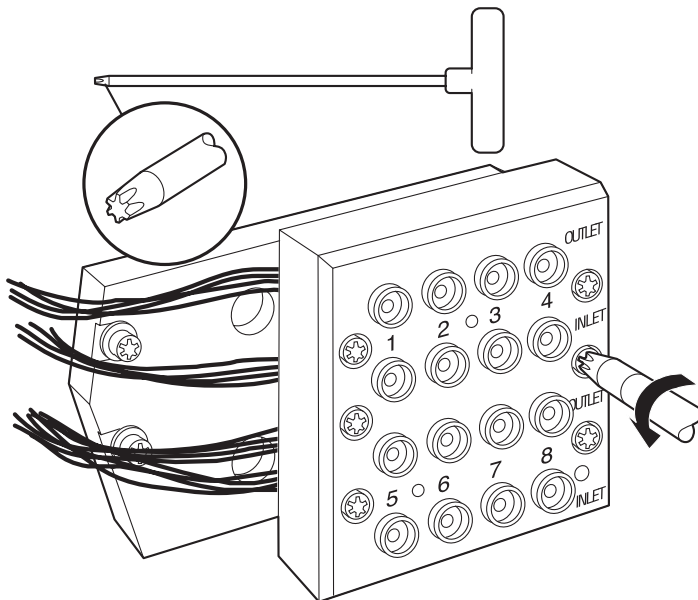
Attention: *The 2488 Detector is shipped with eight inlet and eight outlet unions. The top bank contains channels 1–4 and the bottom bank contains channels 5–8. If you experience a problem with one union and you are using only four channels, you can replumb the 2488 Detector to operate using the opposite bank of channels.*

To remove a union:

1. Flush and dry the flow cell for the union that you want to replace ([Section 4.3.1](#)).
2. Remove the flow cell assembly with the manifold and optics block ([Section 4.3.2](#)).
3. Unscrew the compression screw on the backside of the manifold that is connected to the union that you want to replace. You may be able to unscrew the compression screw without disconnecting the manifold from the flow cell assembly.
4. If the compression screw is not accessible, unscrew the three shoulder screws using the M3 Allen wrench. The screw heads are concealed behind the manifold cover in the manifold block ([Figure 4-7](#)).

Note: *On the back of each screw there is a compression spring.*

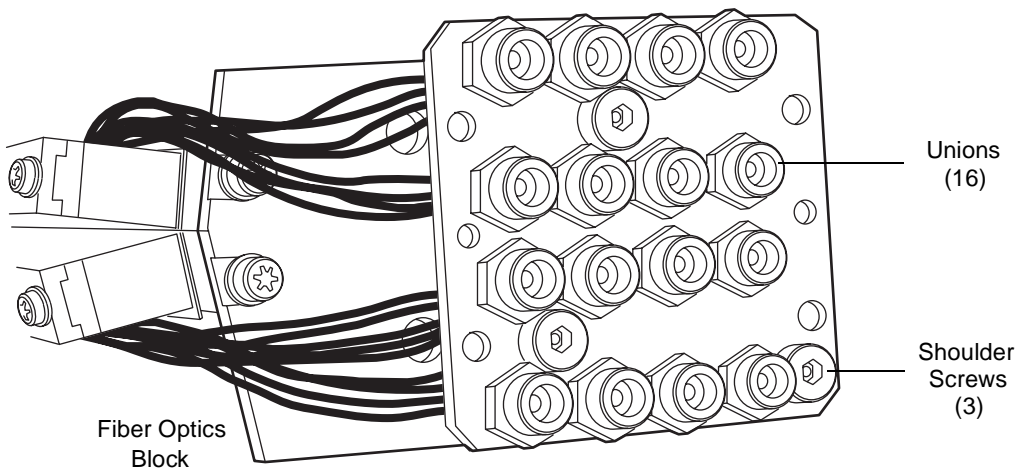
Use a Torx 20 driver to remove the two remaining visible screws ([Figure 4-8](#)).



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Figure 4-8 Removing the Manifold Cover

5. Remove the front cover of the manifold ([Figure 4-2](#)). Keep unions attached to the back of the manifold block ([Figure 4-9](#)).



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Figure 4-9 Manifold with the Cover Removed

6. If the three shoulder screws were unscrewed, reattach them to the fiber optics block and ensure that you add the compression spring to the back of each shoulder screw.
7. Align all unions with their proper holes and replace the front cover with two short screws and lock washers using a Torx 20 driver.
8. Reassemble the flow cell assembly with the manifold and optics block ([Section 4.3.3](#)).

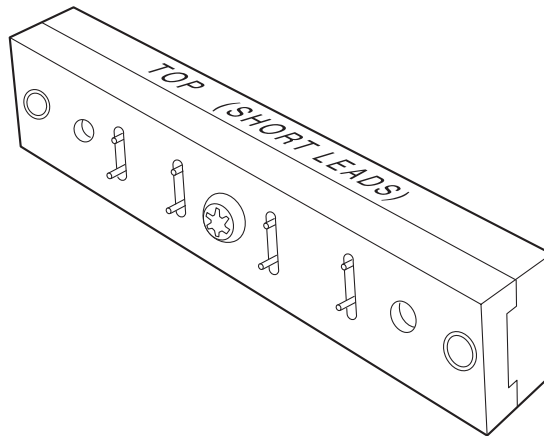
4.5 Replacing the Diode



Attention: To prevent contamination, particularly in the area of polished fiber optics blocks, use powder-free finger cots or gloves when disassembling, inspecting, cleaning, or replacing parts in the optical flow cell assembly.

To replace the diode:

1. Remove the flow assembly with the manifold and optics block ([Section 4.3.2](#)).
2. Using a Torx 20 driver, remove the two outer screws that hold the diode assembly ([Figure 4-5](#)).



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Figure 4-10 Diode Assembly

3. Replace diode assembly with the two outer screws using a Torx 20 driver.
4. See [Section 4.3.4](#) to reassemble the flow cell assembly with the manifold and optics block.

4.6 Replacing the Lamp



Attention: We recommend you change the 2488 Detector lamp when it repeatedly fails to ignite, or when the detector fails to calibrate. See [Table 5-1](#), [Table 5-2](#), and [Table 5-3](#).

This section describes the procedure for removing and replacing the 2488 Detector deuterium lamp.

Lamp Characteristics

The intensity of the deuterium source lamp varies by wavelength as shown in [Figure 4-11](#).

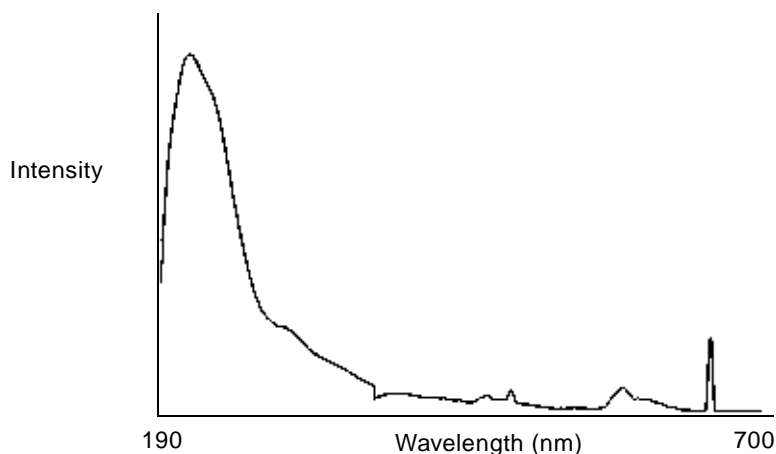


Figure 4-11 Deuterium Lamp Sample Beam Intensity Profile

Lamp Energy and Performance

As the lamps used in traditional absorbance detectors age, the signal-to-noise performance of the instrument degrades. Varying user requirements and inconsistent lamp behaviors make determining “useful” lamp life difficult.

The Waters 2488 Detector combines front-end electronics and lamp optimization software. The design compensates for aging lamps and resultant energy changes within the deuterium spectrum. This allows the 2488 Detector to operate at a consistently high signal-to-noise performance level. A second lamp (e.g. tungsten) is not needed due to longer, more visible wavelengths.

The 2488 Detector performs several self-diagnostic routines on startup, or when you press Calibrate. The lamp optimization diagnostic is one of them. After it verifies monochromator

calibration, the software evaluates energy levels in several characteristic regions of the spectrum. The integration time of the front-end electronics adjusts to maximize signal within these regions. This maintains a high signal-to noise ratio, which yields a clean signal.

Cycle the detector on and off, or press **Calibrate**, at least once a week to maintain instrument performance. Eventually, the lamp's signal deteriorates, and requires replacing.

Ultimately, you should judge the detector's performance according to your own needs. Noise and drift measurements are appropriate for evaluating performance and establishing acceptable sensitivity limits.

Waters guarantees 2000 hours of lamp life, or 1 year of operation, whichever comes first. The detectors on-board diagnostics allow you to record lamp usage and report its serial number. You can replace lamps at any time that is appropriate for their method.

When to Replace the Lamp



Attention: Always run the Change Lamp diagnostic ([Section 5.2.2, Using the Diagnostics](#), the "Using the Lamp, Display, and Keypad Diagnostics" discussion) after you install a new lamp.

Replace the lamp on your 2488 Detector when:

- It fails to ignite at startup
- The energy level in the lamp decreases in sensitivity to the point where the baseline is too noisy for your LC/MS application

Note: Always run the procedure in the discussion "Recording Sample and Reference Beam Energies" in [Section 3.3.2, Verifying the Detector](#), each time you change the 2488 Detector lamp.

Performance requirements and permitted tolerances vary across applications. If the lamp no longer provides an adequate signal-to-noise ratio for your application, replace it.

4.6.1 Removing the Lamp



Caution: To avoid possible eye damage from ultraviolet radiation exposure, turn off the unit and strictly adhere to the following procedures while changing the lamp.



Caution: Lamp housing becomes extremely hot during operation. To prevent possible burns to the skin, allow the lamp to cool for at least 30 to 60 minutes before removing.

To remove the lamp:

1. Turn off power to the 2488 Detector.
2. Allow the lamp to cool for at least 30 to 60 minutes.

Note: If you want the lamp to cool more quickly, turn off the lamp (see [Section 3.3.7](#)) and allow the unit fan to cool the lamp.

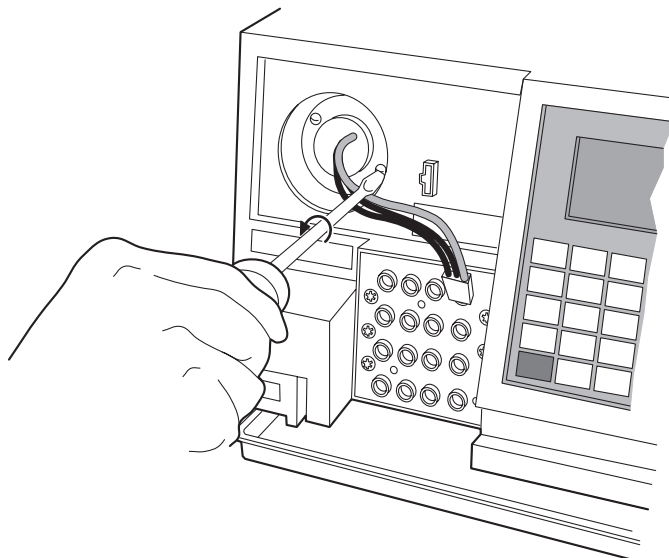
3. Remove the left front panel cover, as described in the discussion "[Removing the Left Front Panel Cover](#)" in [Section 4.1, Introduction](#).
4. Unplug the lamp ([Figure 4-12](#)).



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Figure 4-12 Lamp Assembly and Power Connector

5. Loosen the two captive screws in the lamp base ([Figure 4-13](#)).



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Figure 4-13 Loosening the Captive Screws at the Lamp Housing Base

6. Gently pull the lamp assembly out of the lamp housing ([Figure 4-14](#)).



Caution: Lamp gas is under slight negative pressure. To prevent shattering the glass, use care when disposing of the lamp.

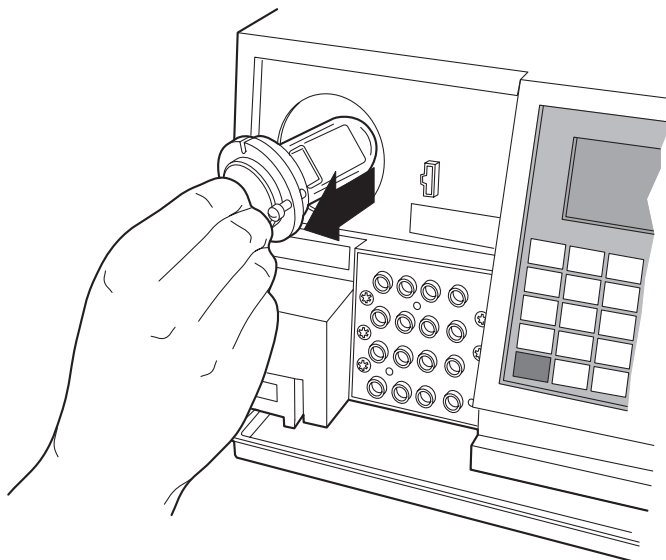


Figure 4-14 Removing the Lamp

4.6.2 Installing the New Lamp



Caution: To avoid possibly exposing your eyes to ultraviolet radiation, wear eye protection that filters ultraviolet light and keep the lamp in the housing during operation.



Attention: Do not touch the glass bulb on the new lamp. Dirt or fingerprints on the bulb affect detector operation. If the lamp needs cleaning, gently clean the bulb with ethanol and lens tissue. Do not use any abrasive tissue or apply excessive pressure.

Before you begin:

1. Unpack the lamp from its packing material. The new lamp may vary slightly from the lamp in [Figure 4-14](#).
2. Inspect the new lamp and lamp housing.
3. Record the serial number located on a label attached to the lamp connector wire using the procedure in [Section 4.6.3, Recording the New Lamp Serial Number](#).



Attention: Make sure power to the 2488 Detector is off.

To install the new lamp:

1. Position the lamp so the cut-out located on the lamp base plate is at the 1 o'clock position ([Figure 4-15](#)), in line with the alignment pin on the lamp housing. There is no additional alignment required.
2. Gently push the lamp forward until it bottoms into position.
3. Tighten the two captive screws.
4. Reconnect the lamp power connector.
5. When ready to resume operation of the 2488 Detector, turn the power to the unit on. Allow at least 5 minutes for the lamp to warm up before resuming operations.



Attention: When you change the lamp, always power off the detector. After installing a new lamp, always power on the detector and wait at least 5 minutes for the new lamp to warm up.

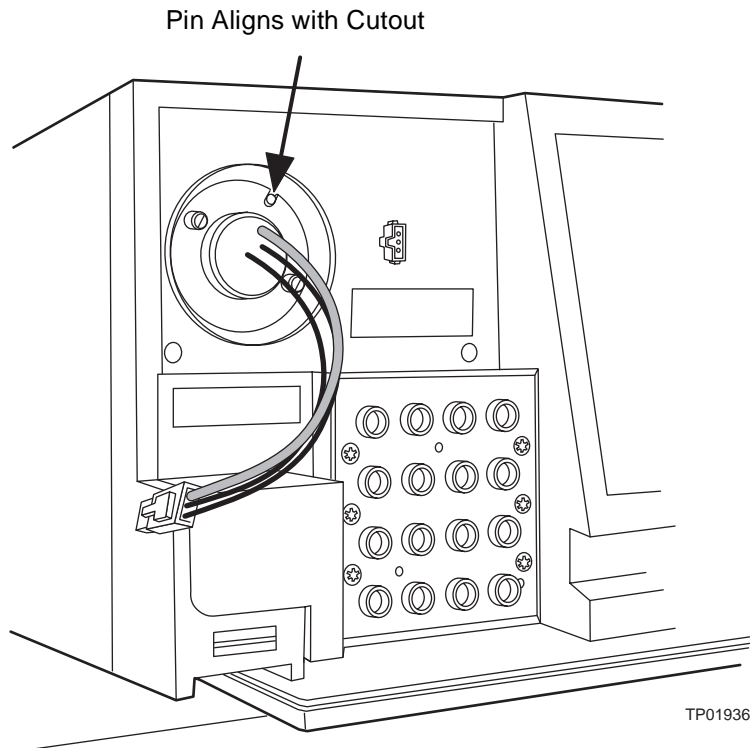


Figure 4-15 Aligning the Lamp

4.6.3 Recording the New Lamp Serial Number



Attention: Always run the Change Lamp diagnostic (see [Section 5.2.2, Using the Diagnostics](#), the "Using the Lamp, Display, and Keypad Diagnostics" discussion) after you install a new lamp.



Attention: If you do not record the new lamp serial number following the procedure in this section, the lamp warranty is voided.

The 2488 Detector software allows you to record and store the serial number and date of installation of a new lamp so that you can periodically check its age and its number of ignitions. To record the new lamp serial number:

1. Once the unit warms up, press the **DIAG** key.
2. Press **6, Lamp, display & keypad**.
3. Press **1, Change lamp**.

Note: Enter the 9-digit lamp serial number and not the lamp part number when performing this procedure.

4. Enter the 9-digit serial number of the new lamp in the active field ([Figure 4-16](#)). This field accepts numeric entries only.

Change Lamp

Serial number of new lamp:

Date installed:

Jan 1 Year

Figure 4-16 Change Lamp Screen

5. Press **Enter** to store the serial number and move to the Date installed field.
6. Select the month from the choice list.
7. Press **Enter** twice to update the month and move to the Day field.
8. Enter the number for the day of the month the lamp is installed, and press **Enter** to enter the date and move to the Year field.
9. Enter the year (last two digits only) and press **Enter**.
10. Press the **HOME** key. An "OK to store" message ([Figure 4-17](#)) appears.



Figure 4-17 OK to Store Message

11. Press **Enter** to enter the serial number and date of installation of the new lamp or press **Cancel** to cancel the entry.
12. When a confirmation message ([Figure 4-18](#)) appears, press **Enter** to continue.



Figure 4-18 Lamp Serial Number Confirmation Message

13. Perform a manual wavelength calibration (see [Section 3.3.3, Calibrating the Wavelength](#)).



Attention: To run the verification procedures with the new lamp installed, recalibrate the detector or recycle power to the 2488 Detector after replacing the lamp.

4.7 Replacing Fuses

The fuse holder is located on the rear panel of the detector ([Figure 4-19](#)). Two fuses are shipped in place with the 2488 Detector, rated as indicated in [Section 2.2.2, Power Requirements](#).

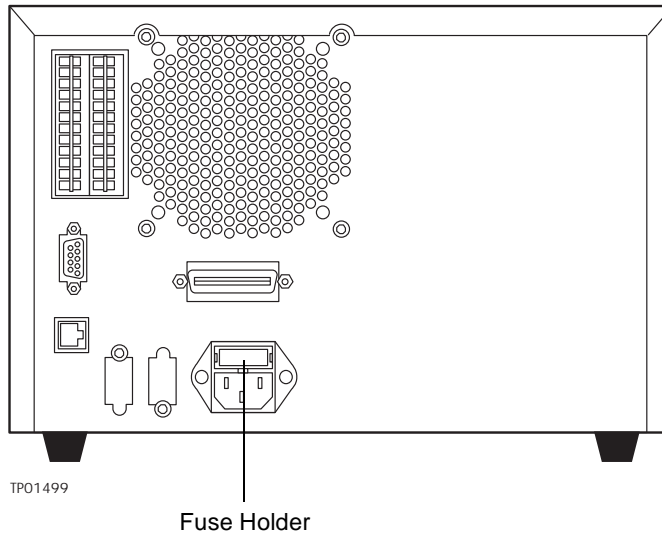


Figure 4-19 2488 Detector Rear Panel Fuse Holder



Caution: To avoid possible electrical shock, power down and unplug the instrument before checking the fuses. For continued protection against fire hazard, replace fuses only with those of the same type and rating.

Identifying Faulty Fuses

Suspect a faulty fuse if:

- The 2488 Detector fails to power up.
- The display is blank.
- The fan does not turn on.

Replacing Rear Panel Fuses

To replace a suspected blown or faulty fuse:

1. Unplug the power cord from the rear of the 2488 Detector.
2. Insert your fingers into the fuse holder slots on the rear panel of the detector. Exerting a minimum pressure, pull on the spring-loaded fuse holder and remove it from the rear panel ([Figure 4-20](#)).

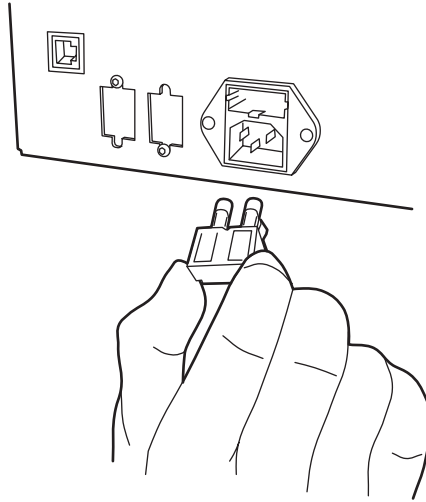


Figure 4-20 Removing and Replacing the Rear Panel Fuses and Fuse Holder

3. Remove and discard the old fuse.
4. Make sure that the new fuse is properly rated for your requirements.
5. Insert the new fuse into the fuse holder.
6. Insert the fuse holder into the receptacle and gently push until it locks into position on the detector rear panel.
7. Connect the power cord the rear panel of the 2488 Detector.
8. Power on the 2488 Detector.

Chapter 5

Error Messages, Diagnostics, and Troubleshooting

The Waters 2488 Multichannel Absorbance Detector provides both user and service diagnostics to troubleshoot system problems.

- **Error messages** – Describe power-up, calibration, and other error messages along with recommended actions for correcting the errors.
- **Diagnostics** – Describe user diagnostics for troubleshooting and configuring the detector.

5.1 Error Messages

5.1.1 Startup Error Messages

Startup diagnostics are executed automatically when power to the 2488 Detector is turned on. These diagnostics verify the proper operation of the detector electronics. If one or more of the internal startup diagnostics checks fails, the detector:

- Beeps
- Displays an error message
- For serious errors, displays the word “Error” instead of the run-time absorbance on the Absorbance Home screen

Note: *To prevent errors at startup, be sure the flow cell contains degassed transparent solvent (methanol or water).*

The tables in this section are organized as follows:

- Messages requiring you to perform corrective action. These include messages encountered at startup, and during calibration or operation.
- Messages requiring you to recycle power, then contact Technical Service personnel if the error persists (see [“Waters Technical Service”](#) in [Section 4.1](#)). Most of these errors are encountered on startup.

[Table 5-1](#) provides startup, calibration, and operating error messages, descriptions, and recommended actions you can take to correct the problem.

Refer to [Section 1.2.2, Wavelength Verification and Test](#), [Section 3.3.2, Verifying the Detector](#), and [Section 3.3.3, Calibrating the Wavelength](#), for verification and calibration information and procedures.

Table 5-1 Startup, Calibration, and Operating Error Messages

Error Message	Description	Corrective Action
Wavelengths span 370 nm: Order filter not in use	<p>In dual wavelength mode:</p> <ul style="list-style-type: none"> • If both selected wavelengths >370 nm, the detector applies the second order filter to block unwanted UV light. • If both selected wavelengths <370 nm, the detector removes the second order filter. • If the selected wavelengths bracket the 370-nm threshold, the detector does not apply the second order filter and issues a warning message that any data collected for wavelengths above 370 nm may contain inaccuracies because of possible UV light interference (second order effects). 	Select wavelengths that are both above or both below 370 nm.
Lamp failure	Lamp indicates Off when it should be On.	<p>Check the lamp icon.</p> <p>Recycle power to the 2488 Detector.</p> <p>Replace the lamp.</p>
Lamp lighting failure	The lamp failed to ignite.	<p>Recycle power to the 2488 Detector.</p> <p>Check lamp power connection.</p> <p>Replace lamp.</p>

Table 5-1 Startup, Calibration, and Operating Error Messages (*Continued*)

Error Message	Description	Corrective Action
Lamp external input conflict	A timed event or action from the unit's front panel attempts to change the lamp state in conflict with enabled lamp input contact closure.	Check the contact closure status. Check timed events. Recycle power to the 2488 Detector.
Calibration not found	Stored calibration data not valid.	Perform manual calibration procedure.
Peak not found: Erbium <i>n</i> nm	Erbium filter calibration range does not contain a local maximum.	Flush the flow cell.
Peak not found: 656 nm Deuterium	Unit sensors cannot determine 656 nm peak.	Flush the flow cell.
Calibration unsuccessful: Peak out of range <i>n.nn</i> nm	Results of a calibration operation differ by more than 1.0 nm. Unit uses previously stored calibration points.	Flush flow cell.
Calibration differs: <i>n</i> nm	At startup, the unit performs a complete verification which includes remeasurement of all calibration points. New calibration points are compared to stored information from the most recent manual calibration. If any one of the points differs by more than 1.0 nm, the detector displays this message.	Recycle power to the 2488 Detector. Perform manual calibration. Contact Technical Service.

5.1.2 Error Messages Preventing Operation

During initialization, calibration, and operation, the 2488 Detector may display the word "Error" on the Home screen. An error of this type is usually fatal, preventing further operation of the detector and suspending the Absorbance Home display ([Figure 5-1](#)). Absorbance output is halted.

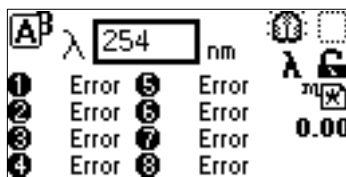


Figure 5-1 Fatal Error Display on the Absorbance Home Screen

When you encounter a fatal error, ensure that:

- There are no bubbles or highly absorptive mobile phase in the flow cell.
- The flow cell is clean.

Recycle power to the 2488 Detector. If the fatal error persists, contact Waters Technical Service.

[Table 5-2](#) lists the error messages that require you to contact Waters Technical Service if recycling power does not correct the error.

Table 5-2 Possibly Fatal Error Messages

Error Message	Description	Corrective Action
Electronic A/D failure	Lamp optimization is adjusted at the minimum level.	Recycle power.
	Acquisition via A/D converters is interrupt-driven. If interrupt is too long, problem with data acquisition is indicated.	Recycle power.
Configuration not found	Stored configuration data is invalid.	Recycle power to the 2488 Detector. This action removes the error.
Lamp data not found	Stored lamp data is invalid.	Recycle power to the 2488 Detector. This action removes the error.

Table 5-2 Possibly Fatal Error Messages (*Continued*)

Error Message	Description	Corrective Action
Grating initialization failure: Backlash too high	Backlash is the difference between the forward and reverse peak positions of deuterium at 656 nm. If this difference is greater than 1 step, the unit displays this message.	Recycle power to the 2488 Detector. Contact Technical Service.
Electronic A/D failure	Data acquisition via A/D converters is interrupt-driven. If interrupt is too long, problem with data acquisition is indicated.	Recycle power to the 2488 Detector. Contact Technical Service.
System cannot respond	Error occurs while unit is positioning next wavelength or changing modes. Occurs during initialization or calibration.	Recycle power to the 2488 Detector. Contact Technical Service.
Filter initialization failure: No reference energy	Unit sensors cannot find any light energy before homing the optical filter.	Recycle power to the 2488 Detector. Contact Technical Service.
Filter initialization failure: No filters found	Unit sensors observe transition to dark before homing the optical filter.	Recycle power to the 2488 Detector. Contact Technical Service.
Filter initialization failure: No response	Unit sensors cannot identify any dark regions.	Recycle power to the 2488 Detector. Contact Technical Service.
Filter initialization failure: Shutter position	Unit sensors cannot find the shutter position.	Recycle power to the 2488 Detector. Contact Technical Service.
Filter initialization failure: Order filter position	Unit sensors cannot find the order filter position.	Recycle power to the 2488 Detector. Contact Technical Service.
Filter initialization failure: Erbium position	Unit sensors cannot find erbium filter position.	Recycle power to the 2488 Detector. Contact Technical Service.
Grating initialization failure: No home sensor	Search for the home sensor failed.	Recycle power to the 2488 Detector. Contact Technical Service.

Table 5-2 Possibly Fatal Error Messages (*Continued*)

Error Message	Description	Corrective Action
System not calibrated.	The calibration read from nonvolatile memory is not valid.	Recycle power to the 2488 Detector. Perform manual calibration. Contact Technical Service.
Communication failure: Reference A/D	A/D communication test failed.	Recycle power to the 2488 Detector. Contact Technical Service.
Communication failure: Sample A/D	A/D communication test failed.	Recycle power to the 2488 Detector. Contact Technical Service.
Dark current reference too low.	The dark energy level is below threshold level (0).	Recycle power to the 2488 Detector. Contact Technical Service.
Dark current reference too high.	The dark energy level is above threshold level (1,000,000).	Recycle power to the 2488 Detector. Contact Technical Service.
Dark current sample too low.	The dark energy level is below threshold level (0).	Recycle power to the 2488 Detector. Contact Technical Service.
Dark current sample too high.	The dark energy level is above threshold level (1,000,000).	Recycle power to the 2488 Detector. Contact Technical Service.

5.2 User-Selected Diagnostics

5.2.1 Overview

Diagnostics allow you to troubleshoot the detector and verify the detector electronics and optics. To perform user-selectable diagnostics:

1. Press the **DIAG** key on the 2488 Detector front panel. The detector displays the Diagnostics choice list ([Figure 5-2](#)).

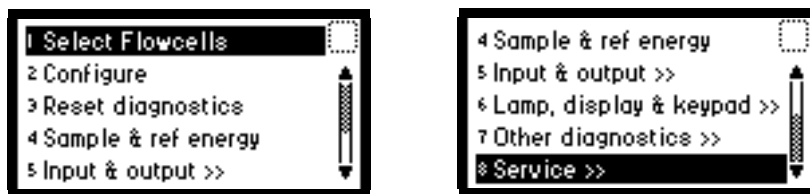


Figure 5-2 Diagnostics Choice List

To access a specific diagnostic test, move the ▲ OR ▼ arrow key to the diagnostic you want to run and press **Enter**, or press a number from **1** to **8** corresponding to the diagnostic number on the detector keypad. Choices that display other choices are indicated by >>.



Sticky diagnostics remain in effect until you disable them. A sticky diagnostic is a visual reminder that you have enabled certain diagnostics which may affect the detector's normal functioning. When a sticky diagnostic is active, the 2488 Detector Absorbance Home screen displays a wrench icon ([Figure 5-3](#)).

- You can disable a specific sticky diagnostic by resetting it to the default settings.
- You can disable all active sticky diagnostics by pressing **DIAG 3, Reset diagnostics**.

If no sticky diagnostics are active, the wrench icon is removed from the Absorbance screen. When you power off the 2488 Detector, all sticky diagnostics are disabled.

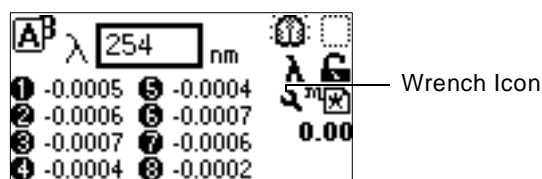


Figure 5-3 Absorbance Home Screen with Sticky Diagnostic Active

You can select the optical filter override sticky diagnostic.

Note: When you select sticky diagnostics, results are affected. To make a manual optical filter change, select **3 Reset diagnostics** from the Diagnostics choice list, or reset power to the 2488 Detector.

[Table 5-3](#) lists the 2488 Detector diagnostics by their number on the choice list, followed by a brief description. See [Section 5.2.2, Using the Diagnostics](#), for more information.

Table 5-3 2488 Detector Diagnostics

Diagnostic	Description
1 Select Flowcells	Select the flow cell bank (1–4, 1–8, or 5–8) for analysis.
2 Configure	Set the IEEE address and configure the terminal block contact closures and switches.
3 Reset diagnostics	Reset all diagnostics to defaults. Remove sticky diagnostics and wrench icon.
4 Sample & ref energy	View sample and reference energy (displayed in nanoAmps) on channel A or B.
5 Input & output	Diagnostics to control four contact closure inputs and two switch outputs.

Table 5-3 2488 Detector Diagnostics (*Continued*)

Diagnostic	Description
6 Lamp, display & keypad>>	List of diagnostics used to test lamp, display, and keypad functions: <ol style="list-style-type: none"> 1. Change lamp 2. Test keypad 3. Test display 4. Previous choices <<
7 Other diagnostics >>	Override the default filter setting: <ol style="list-style-type: none"> 1. Optical filter override 2. Previous choices <<
8 Service	Provide diagnostics used by Technical Service personnel.

5.2.2 Using the Diagnostics

The 2488 Detector employs both user-selectable and service diagnostics. You access user diagnostics by pressing the **DIAG** key. Only qualified Waters service personnel can access service diagnostics.

Contacting Technical Service

If, after running the user diagnostics applicable to your problem and troubleshooting the detector (see [Section 5.3, Troubleshooting](#)), you are unable to correct an error condition, contact Technical Service at 800 252-4752, *U.S. and Canadian customers only*. Other customers, call your local subsidiary or your local Waters Technical Service Representative, or call Waters corporate headquarters for assistance in Milford, Massachusetts (U.S.A.).

If you are a Micromass customer, contact Micromass at +44 161 946 2400, or +44 161 926 7200, or +44 161 435 4100 or call your local Micromass subsidiary.

Using the Select Flow Cells Diagnostic

Use the Select Flow Cells diagnostic ([Figure 5-4](#)) to specify a flow cell bank for analysis. When you select a bank that differs from the current setting, the wavelength is automatically calibrated.

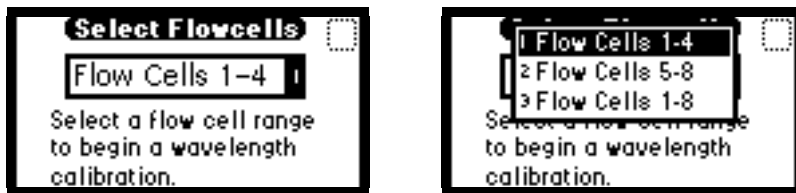


Figure 5-4 Select Flow Cells Diagnostic

To use the Select Flow Cells diagnostic:

1. Press **DIAG 1**.
2. To select a bank, press **Enter**. The second Select Flow Cells screen appears.
3. Scroll up or down to select the flow bank you want to analyze and press **Enter**.
4. The Wavelength Calibration screen appears and automatically calibrates the wavelength. Press **Enter**.

The flow cell bank selection determines which flow cell (1–5) is used for initialization and calibration: for flow cell bank 1–8 or 1–4, use flow cell 1 as the calibrating flow cell, and for bank 5–8, use flow cell 5 as the calibrating flow cell.

Note: In the event that flow cells (1–4) fail in a four-channel system, the second bank of four flow cells (5–8) serves as a spare flow cell set.

Using the Input & Output Diagnostics

You use the Input & Output diagnostics to monitor contact closures and toggle event switches.

To perform any of the Input & Output diagnostics, press **DIAG 5, Input & output**. A choice list appears ([Figure 5-5](#)).

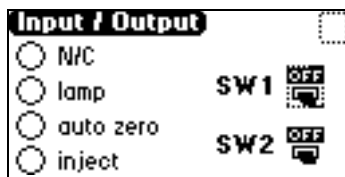


Figure 5-5 Input and Output Diagnostic Choice List

Monitoring Contact Closures and Setting Switches

The Input & Output diagnostics allow real-time monitoring of the state of the contact closure inputs. A solid (filled in) circle indicates the contact closure is closed (ON = High). An open (empty) circle indicates the contact closure is open (OFF = Low).

Outputs (SW1 and SW2) will be available for future use.

Using the Sample and Reference Energy Diagnostics

You use the Sample and Reference Energy diagnostics to examine noise fluctuations and compare them with the AU time trace. The current sample and reference energy readings are displayed in nanoAmps (nA). See [Figure 5-6](#).

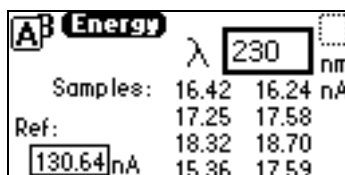


Figure 5-6 Sample and Reference Energy Diagnostics

To use the Sample and Reference Energy diagnostic:

1. Press **DIAG 4**.
2. To change the wavelength, enter a new wavelength number.
3. Press **Enter**. When the new wavelength shifts to the left, the corresponding sample and reference energies for each flow cell are displayed for each flow cell.
4. If you are operating the detector in dual wavelength mode, press the **A/B** key to view sample and reference energy on the other wavelength for each flow cell.

Using the Lamp, Display, and Keypad Diagnostics

To access the lamp, display, and keypad diagnostic tests, press **DIAG 6**.

Using the Change Lamp Diagnostic



Attention: Make sure power to the 2488 Detector is off before you replace the lamp.

1. From the Lamp, display & keypad choice list, press **1**, **Change lamp** to display the Change Lamp diagnostic ([Figure 5-7](#)).

The screen displays the title "Change Lamp" in a black box. Below it is the label "Serial number of new lamp:" followed by a large rectangular input field. Underneath is the label "Date installed:" followed by two input fields. The first field contains "Jan" and the second field contains "1". To the right of these fields is the label "Year" followed by another input field.

Figure 5-7 Change Lamp Diagnostic

2. Enter the 9-digit serial number and date of installation for the new lamp. Press **Enter** after each entry.

Use this function whenever you change the lamp to enter a new serial number and installation date. See [Section 4.6.2, Installing the New Lamp](#), and [Section 4.6.3, Recording the New Lamp Serial Number](#), for full details of the lamp replacement procedure.



Attention: If you do not record the new lamp serial number following the procedure in [Section 4.6.2, Installing the New Lamp](#), and [Section 4.6.3, Recording the New Lamp Serial Number](#), the lamp warranty is voided. The date of the previous lamp installation remains in the 2488 Detector memory.

Using the Test Keypad Diagnostic

1. From the Lamp, display & keypad choice list, press **2**, **Test keypad** to run the keypad test. A bitmap representation of the keypad is displayed ([Figure 5-8](#)).

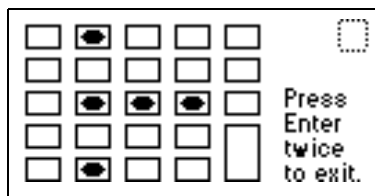


Figure 5-8 Keypad Diagnostic

2. Press any key to begin the test, then press each key until you have tried all of them. If the keypad is operating properly, each key location is filled in, then cleared with another press of the key. If any key does not respond when pressed, contact your Waters service representative.

Note: You must press **Enter** twice to exit the keypad diagnostic.

Using the Test Display Diagnostic

1. From the Lamp, display & keypad choice list, press **3, Test display** to run the display test.

When you press 3, the display fills from top to bottom and from right to left, then returns to the Lamp, display & keypad choice list. If the display does not completely fill either horizontally or vertically, contact your Waters service representative.

2. From the Lamp, display & keypad choice list, press **4** to return to the diagnostics choice list.

Using the Other 2488 Detector Diagnostics

The user diagnostic screen ([Figure 5-9](#)) provides an additional diagnostic feature: you can override the optical filter.

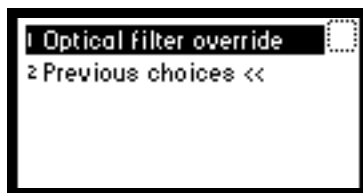


Figure 5-9 User Diagnostic

Overriding the Optical Filter



1. From the Other diagnostics choice list, press **1 Optical filter override** to manually override the detector's automatic filter choice. The Optical Filter Override diagnostic is displayed ([Figure 5-10](#)).

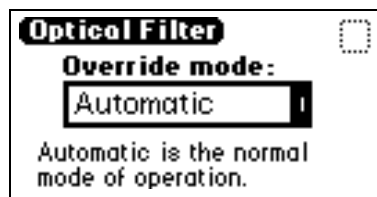


Figure 5-10 Optical Filter Override Diagnostic

2. Press **Enter** to display these filter choices:

Automatic	1
Second Order	2
None	3
Erbium	4
Shutter	5

The 2488 Detector normally operates with the filter in the Automatic position. This is a sticky diagnostic.

Note: When you select sticky diagnostics, results are affected. To make a manual optical filter change, select **3 Reset diagnostics** from the Diagnostics choice list, or reset power to the 2488 Detector.

3. Press the number corresponding to your filter choice or leave the default filter choice (Automatic) on.
4. To turn this diagnostic off, press **DIAG 3** or select **Automatic** from the choice list.

5.2.3 Service Diagnostics

Only qualified service personnel can access the 2488 Detector service diagnostics.

5.3 Troubleshooting

5.3.1 Introduction

This section provides some causes of errors and recommended troubleshooting actions. Keep in mind that the source of apparent detector problems may lie within the chromatography or your other instruments, as well as the 2488 Detector.

Most detector problems are relatively easy to correct. If you are unable to correct a problem or a failed condition, contact Waters Technical Service at 800 252-4752, *U.S. and Canadian customers only*. Other customers, call your local Waters subsidiary or your local Waters Technical Service Representative, or call Waters corporate headquarters for assistance in Milford, Massachusetts (U.S.A.).

If you are a Micromass customer, contact Micromass at +44 161 946 2400, or +44 161 926 7200, or +44 161 435 4100, or call your local Micromass subsidiary.

When You Contact Waters

To expedite your request for service, have the following information available when you call Waters Technical Service:

- 2488 Detector serial number
- Problem symptom(s)
- Operating wavelength(s)
- Configuration, flow cell channels, and IEEE address
- Flow rate
- Filter setting
- Type of column
- Operating pressure
- Solvent(s)
- System configuration (other components)

Diagnostics

The 2488 Detector performs some user-selected diagnostics to help you troubleshoot basic system problems. Refer to [Section 5.2.2, Using the Diagnostics](#), for diagnostic descriptions and instructions on how to use them.

Error messages that may appear as you start up or operate the 2488 Detector and suggested corrective actions are described in [Table 5-1](#) and [Table 5-2](#).

Power Surges

Power surges, line spikes, and transient energy sources can adversely affect detector operations. Be sure that the electrical supply used for the detector is properly grounded and free from any of these conditions.

5.3.2 Hardware Troubleshooting

Contains general hardware troubleshooting for the 2488 Detector.

Table 5-4 General System Troubleshooting

Symptom	Possible Cause	Corrective Action
Detector inoperative	Fuse blown	Check that the front panel display is operational; if not, replace the ac rear panel fuse.
	No power at outlet	Check outlet by connecting another electrical unit known to be in working order and see if it operates.
Front panel display fails to illuminate	Broken electrical connection	Contact Technical Service.
	Fuse blown	Check and, if necessary, replace fuse(s).
	Bad LCD or control board	Contact Technical Service.
Front panel displays odd characters	Faulty EPROMs Bad LCD control board	Contact Technical Service.
IEEE problems	Incorrect IEEE address	Set the correct IEEE address.
	Bad IEEE cable with correct IEEE address(es) set (2-29) (external)	Check the IEEE address. Replace the IEEE cable.
Keypad not functioning	Keypad defective	Recycle power to the 2488 Detector and run the keypad diagnostic. Contact Technical Service.
Deuterium lamp does not light	Faulty lamp	Replace the lamp.
	Lamp not plugged in	Plug in the lamp connector.
	Bad lamp power supply board	Contact Waters Technical Service.
	Lamp switch off	Check the rear panel connections (may be programmed to turn the lamp off) or timed event within a method (may be programmed to turn the lamp off).

Table 5-4 General System Troubleshooting (*Continued*)

Symptom	Possible Cause	Corrective Action
No sample and reference energy	Lamp burned out	Attempt to reignite using the Lamp key. Replace the lamp.
	Lamp off	Check the lamp icon. Run the Sample & ref energy diagnostic.
Calibration or energy error on startup	UV absorber in flow cell	Flush the flow cell. Perform manual calibration.
Peak out of range error at startup	Mobile phase absorbance too high; air bubbles	Be sure the flow cell mobile phase does not absorb above 250 nm. Ensure there are no air bubbles in the flow cell. Recalibrate the 2488 Detector. If problems persist, contact Technical Service.
Flat-topped peaks	Insufficient dynamic range. Check the Sample and Reference Energy of mobile phase. If Sample is less than 1/100th of Reference, there is a high background absorbance of the mobile phase.	1. Change wavelength. 2. Change mobile phase. 3. Reduce sample concentration.
No absorbance response during data acquisition.	Flow cell bank selection does not match flow cells used.	Make sure flow cell bank selection matches flow cells used in liquid line connections.

Appendix A

2488 Detector Specifications

This appendix lists operating specifications for the Waters® 2488 Multichannel Absorbance Detector, as shipped:

- [Table A-1](#) lists operational specifications.¹
- [Table A-2](#) lists optical specifications.
- [Table A-3](#) lists flow cell specifications

Table A-1 Operational Specifications

Item	Specification
Wavelength range	190 to 700 nm
Bandwidth	5 nm
Wavelength accuracy	±3 nm
Wavelength repeatability	±0.1 nm
Noise, single wavelength mode ^a	< $\pm 1.0 \times 10^{-5}$ AU, dry cell, 235 nm, 1.0 second filter time constant (TC)
Linearity ^a	5% at 2.5 AU, propyl paraben, 257 nm
Drift, single wavelength mode	< $\pm 1.5 \times 10^{-4}$ AU per 15 min. dry flow cell at 235 nm, 1.0-second filter time constant
Filter setting range	Single wavelength: 0.1 to 5.0 seconds, Hamming (default) 1 to 99 seconds, RC Dual wavelength: 1 to 50 seconds, Hamming (default) 1 to 99 seconds, RC
Optical Component Specifications	
Lamp source	Deuterium arc lamp
Flow cell	Either 4 or 8 channels, 4-mm path length, low dispersion
Path length	4 mm (standard analytical)
Cell volume	3 µL (standard analytical)

1. Specifications are derived from instrument performance after a 1-hour warmup time.

Table A-1 Operational Specifications (*Continued*)

Item	Specification
Cell pressure	1000 psi max
Materials	316 stainless steel, fused silica, Teflon®
Environmental Specifications	
Operating temperature	4 to 40 °C (39 to 104 °F)
Operating humidity	20 to 80%, noncondensing
Shipping and storage temperature	–40 to +70 °C (–40 to +158 °F)
Shipping and storage humidity	20 to 80%, noncondensing
Electrical Specifications	
Line frequency	50 to 60 Hz
Line voltage	100 to 240 Vac
Fuse ratings	Two fuses: F 3.15 A 250 V FAST BLO, 5 x 20 mm (IEC)
Power consumption	145 VA (nominal)
Two event outputs	Type: Contact closure Voltage: ± 30 V Current: 1 A
Four event inputs	Input voltage: ± 30 V maximum 100 ms minimum period
Dimensions	
Height	8.2 inches (20.8 cm)
Length	19.8 inches (50.3 cm)
Width	11.2 inches (28.4 cm)
Weight	20.5 pounds (9.3 kg)

a. ASTM Standard E1657-94

Table A-2 Optical Specifications

Monochromator	Ebert Configuration
Grating	Plane holographic 1800 grooves/mm
Optical bandwidth	5 nm
Lamp power	30 W

Table A-3 Flow Cell Specifications

Cell	Volume (μl)	Path Length (mm)	Sample Tube Internal Diameter (Inches)		Pressure Rating
			In/Out		psi/bar
Analytical Cell	3	4	0.005	0.005	1000/70

Appendix B

Spare Parts

[Table B-1](#) lists the spare parts for the Waters 2488 Multichannel Absorbance Detector. These spare parts are user-replaceable.

Table B-1 Spare Parts

Item	Part Number
Fuses	
Fuse Holder	WAT055426
Fuse, 3.15 A (240 V, 5 x 20 mm)	700001800
Optical Parts and Assemblies	
2487/2488 Lamp Preventive Maintenance Kit	WAS081142
Photodiode Block containing four photodiodes and cover	700001803
Union, 0.031 x 0.063	700001789
Flow cell assembly with manifold	700001786
Waters Startup Kit, 2488 Detector	200000142
Event cable	WAT020321
Cable, IEEE-488, 3.3 feet	WAT087198
Fuse, 3.15 A (240 V, 5 x 20 mm)	WAT163-16
Connector plug, 12 pin	WAT270868
Comp screw, one piece, 1/16	700001790
Tubing, PEEK, 0.06 x 0.005 x 50 feet	700001794
Tubing, PEEK, 0.06 x 0.010 x 50 feet	700001795
Tubing, 0.375 OD x 0.250 ID, Tygon 2705	700001792
Screwdriver, Slotted, Pocket Clip Style	700001824
Torx Key, T10	WAT410164
Torx Key, T20	WAT410165
Allen Key, M3	700001825

Table B-1 Spare Parts (*Continued*)

Item	Part Number
General Parts	
Drip tray, 2488	700001792
Connector male, 1/8 NPT, 1/4 inch tube	700001826
<i>Waters Multichannel Absorbance Detector with MassLynx Control Operator's Guide</i>	71500248802

Appendix C

Solvent Considerations



Caution: To avoid chemical hazards, always observe safe laboratory practices when operating your system.

C.1 Introduction

Clean Solvents

Clean solvents provide:

- Reproducible results
- Operation with minimal instrument maintenance

A dirty solvent can cause:

- Baseline noise and drift
- Blockage of the solvent filters with particulate matter

Solvent Quality

Use HPLC-grade solvents to ensure the best possible results. Filter through 0.45- μm filters before use. Solvents distilled in glass generally maintain their purity from lot to lot; use them to ensure the best possible results.

Preparation Checklist

The following solvent preparation guidelines help to ensure stable baselines and good resolution:

- Filter solvents with a 0.20- μm filter.
- Degas and/or sparge the solvent.
- Stir the solvent.
- Keep in a place free from drafts and shock.

Water

Use water only from a high-quality water purification system. If the water system does not provide filtered water, filter it through a 0.45- μ m membrane filter before use.

Buffers

When you use buffers, dissolve salts first, adjust the pH, then filter to remove insoluble material.

Tetrahydrofuran (THF)

When using unstabilized THF, ensure that your solvent is fresh. Previously opened bottles of THF contain peroxide contaminants, which cause baseline drift.



Caution: THF contaminants (peroxides) are potentially explosive if concentrated or taken to dryness.

C

C.2 Solvent Miscibility

Before you change solvents, refer to [Table C-1](#) to determine the miscibility of the solvents to be used. When you change solvents, be aware that:

- Changes involving two miscible solvents may be made directly. Changes involving two solvents that are not totally miscible (for example, from chloroform to water), require an intermediate solvent (such as isopropanol).
- Temperature affects solvent miscibility. If you are running a high-temperature application, consider the effect of the higher temperature on solvent solubility.
- Buffers dissolved in water may precipitate when mixed with organic solvents.

When you switch from a strong buffer to an organic solvent, flush the buffer out of the system with distilled water before you add the organic solvent.

Table C-1 Solvent Miscibility

Polarity Index	Solvent	Viscosity CP, 20 °C	Boiling Point °C (1 atm)	Miscibility Number (M)	λ Cutoff (nm)
-0.3	n-decane	0.92	174.1	29	—
-0.4	Isooctane	0.50	99.2	29	210
0.0	n-hexane	0.313	68.7	29	—

Table C-1 Solvent Miscibility (Continued)

Polarity Index	Solvent	Viscosity CP, 20 °C	Boiling Point °C (1 atm)	Miscibility Number (M)	λ Cutoff (nm)
0.0	Cyclohexane	0.98	80.7	28	210
1.7	Butyl ether	0.70	142.2	26	—
1.8	Triethylamine	0.38	89.5	26	—
2.2	Isopropyl ether	0.33	68.3	—	220
2.3	Toluene	0.59	100.6	23	285
2.4	p-xylene	0.70	138.0	24	290
3.0	Benzene	0.65	80.1	21	280
3.3	Benzyl ether	5.33	288.3	—	—
3.4	Methylene chloride	0.44	39.8	20	245
3.7	Ethylene chloride	0.79	83.5	20	—
3.9	n-Butyl alcohol	3.00	117.7	—	—
4.2	Tetrahydrofuran	0.55	66.0	17	220
4.3	Ethyl acetate	0.47	77.1	19	260
4.3	1-propanol	2.30	97.2	15	210
4.3	2-propanol	2.35	117.7	15	—
4.4	Methyl acetate	0.45	56.3	15, 17	260
4.5	Methyl ethyl ketone	0.43	80.0	17	330
4.5	Cyclohexanone	2.24	155.7	28	210
4.5	Nitrobenzene	2.03	210.8	14, 20	—
4.6	Benzonitrile	1.22	191.1	15, 19	—
4.8	Dioxane	1.54	101.3	17	220
5.2	Ethanol	1.20	78.3	14	210
5.3	Pyridine	0.94	115.3	16	305
5.3	Nitroethane	0.68	114.0	—	—
5.4	Acetone	0.32	56.3	15, 17	330
5.5	Benzyl alcohol	5.80	205.5	13	—
5.7	Methoxyethanol	1.72	124.6	13	—
6.2	Acetonitrile	0.37	81.6	11, 17	190
6.2	Acetic acid	1.26	117.9	14	—
6.4	Dimethylformamide	0.90	153.0	12	—

Table C-1 Solvent Miscibility (Continued)

Polarity Index	Solvent	Viscosity CP, 20 °C	Boiling Point °C (1 atm)	Miscibility Number (M)	λ Cutoff (nm)
6.5	Dimethylsulfoxide	2.24	189.0	9	—
6.6	Methanol	0.60	64.7	12	210
7.3	Formamide	3.76	210.5	3	—
9.0	Water	1.00	100.0	—	—

How to Use Miscibility Numbers

Use miscibility numbers (M-numbers) to predict the miscibility of a liquid with a standard solvent ([Table C-1](#)).

To predict the miscibility of two liquids, subtract the smaller M-number value from the larger M-number value.

- If the difference between the two M-numbers is 15 or less, the two liquids are miscible in all proportions at 15 °C.
- A difference of 16 indicates a critical solution temperature from 25 to 75 °C, with 50 °C as the optimal temperature.
- If the difference is 17 or greater, the liquids are immiscible or their critical solution temperature is above 75 °C.

Some solvents prove immiscible with solvents at both ends of the lipophilicity scale. These solvents receive a dual M-number:

- The first number, always lower than 16, indicates the degree of miscibility with highly lipophilic solvents.
- The second number applies to the opposite end of the scale. A large difference between these two numbers indicates a limited range of miscibility.

For example, some fluorocarbons are immiscible with all the standard solvents and have M-numbers of 0, 32. Two liquids with dual M-numbers are usually miscible with each other.

A liquid is classified in the M-number system by testing for miscibility with a sequence of standard solvents. A correction term of 15 units is then either added or subtracted from the cutoff point for miscibility.

C.3 Buffered Solvents

When using a buffer, use a good quality reagent and filter it through a 0.45- μ m filter.

Do not leave the buffer stored in the system after use. Flush all fluidic pathways with HPLC-quality water before shutting the system down and leave distilled water in the system (flush with 90:10 90% HPLC-quality water to 10% methanol for shutdowns scheduled to be more than one day). Use a minimum of 15 mL for sparge-equipped units, and a minimum of 45 mL for inline vacuum degasser-equipped units.

C.4 Head Height

Position the solvent reservoirs at a level above the HPLC equipment or on top of the pump or detector (with adequate spill protection).

C.5 Solvent Viscosity

Generally, viscosity is not important when you are operating with a single solvent or under low pressure. However when you are running a gradient, the viscosity changes that occur as the solvents are mixed in different proportions can result in pressure changes during the run. For example, a 1:1 mixture of water and methanol produces twice the pressure of either water or methanol alone.

If the extent to which the pressure changes will affect the analysis is not known, monitor the pressure during the run using the Chart Out terminal.

C.6 Mobile Phase Solvent Degassing

Mobile phase difficulties account for 70% or more of all liquid chromatographic problems. Using degassed solvents is important, especially at wavelengths below 220 nm.

Degassing provides:

- Stable baselines and enhanced sensitivity
- Reproducible retention times for eluting peaks
- Reproducible injection volumes for quantitation
- Stable pump operation

This section presents information on the solubility of gases, solvent degassing methods, and solvent degassing considerations.

C.6.1 Gas Solubility

Only a finite amount of gas can be dissolved in a given volume of liquid. This amount depends on:

- The chemical affinity of the gas for the liquid
- The temperature of the liquid
- The pressure applied to the liquid

Changes in the composition, temperature, or pressure of the mobile phase can all lead to outgassing.

Effects of Intermolecular Forces

Nonpolar gases (N_2 , O_2 , CO_2 , and He) are more soluble in nonpolar solvents than in polar solvents. Generally, a gas is most soluble in a solvent with intermolecular attractive forces similar to those in the gas (like dissolves like).

Effects of Temperature

Temperature affects the solubility of gases. If the heat of solution is exothermic, the solubility of the gas decreases when you heat the solvent. If the heat of solution is endothermic, the solubility increases when you heat the solvent. For example, the solubility of He in H_2O decreases with an increase in temperature, but the solubility of He in benzene increases with an increase in temperature.

Effects of Partial Pressure

The mass of gas dissolved in a given volume of solvent is proportional to the partial pressure of the gas in the vapor phase of the solvent. If you decrease the partial pressure of the gas, the amount of that gas in solution also decreases.

C.6.2 Solvent Degassing Methods

This section describes the solvent degassing techniques that will help you to attain a stable baseline. Degassing your solvent also improves reproducibility and pump performance.

You can use either of the following methods to degas solvents:

- Sparging with helium
- Vacuum degassing

Sparging

Sparging removes gases from solution by displacing dissolved gases in the solvent with a less soluble gas, usually helium. Well-sparged solvent improves pump performance. Helium sparging brings the solvent to a state of equilibrium, which may be maintained by slow sparging or by keeping a blanket of helium over the solvent. Blanketing inhibits reabsorption of atmospheric gases.

Sparging may change the composition of mixed solvents.

Vacuum Degassing

The inline vacuum degasser operates on the principle of Henry's Law to remove dissolved gases from the solvent. Henry's Law states that the mole fraction of a gas dissolved in liquid is proportional to the partial pressure of that gas in the vapor phase above the liquid. If the partial pressure of a gas on the surface of the liquid is reduced, for example, by evacuation, then a proportional amount of that gas comes out of solution.

Vacuum degassing may change the composition of mixed solvents.

C.6.3 Solvent Degassing Considerations

Select the most efficient degassing operation for your application. To remove dissolved gas quickly, consider the following.

Sparging

Helium sparging gives stable baselines and prevents reabsorption of atmospheric gases. Use this method to retard oxidation when you are using THF or other peroxide-forming solvents.

Vacuum Degassing

The longer the solvent is exposed to the vacuum, the more dissolved gases are removed. Two factors affect the amount of time the solvent is exposed to the vacuum:

- **Flow rate** – At low flow rates, most of the dissolved gas is removed as the solvent passes through the vacuum chamber. At higher flow rates, lesser amounts of gas per unit volume of solvent are removed.
- **Surface area of the degassing membrane** – The length of the degassing membrane is fixed in each vacuum chamber. To increase the length of membrane, you can connect two or more vacuum chambers in series.

The inline degasser is available as an option or factory-installed in the Waters 2690 Separations Module, XE model.

C.7 Wavelength Selection

This section includes UV cutoff ranges for:

- Common solvents
- Common mixed mobile phases
- Chromophores

UV Cutoffs for Common Solvents

[Table C-2](#) shows the UV cutoff (the wavelength at which the absorbance of the solvent is equal to 1 AU) for some common chromatographic solvents. Operating at a wavelength near or below the cutoff increases baseline noise due to the absorbance of the solvent.

Table C-2 UV Cutoff Wavelengths for Common Chromatographic Solvents

Solvent	UV Cutoff (nm)	Solvent	UV Cutoff (nm)
1-Nitropropane	380	Ethylene glycol	210
2-Butoxyethanol	220	Isooctane	215
Acetone	330	Isopropanol	205
Acetonitrile	190	Isopropyl chloride	225
Amyl alcohol	210	Isopropyl ether	220
Amyl chloride	225	Methanol	205
Benzene	280	Methyl acetate	260
Carbon disulfide	380	Methyl ethyl ketone	330
Carbon tetrachloride	265	Methyl isobutyl ketone	334
Chloroform	245	Methylene chloride	233
Cyclohexane	200	<i>n</i> -Pentane	190
Cyclopentane	200	<i>n</i> -Propanol	210
Diethyl amine	275	<i>n</i> -Propyl chloride	225
Dioxane	215	Nitromethane	380
Ethanol	210	Petroleum ether	210
Ethyl acetate	256	Pyridine	330
Ethyl ether	220	Tetrahydrofuran	230
Ethyl sulfide	290	Toluene	285
Ethylene dichloride	230	Xylene	290

Mixed Mobile Phases

[Table C-3](#) contains approximate wavelength cutoffs for some other solvents, buffers, detergents, and mobile phases. The solvent concentrations represented are those most commonly used. If you want to use a different concentration, you can determine approximate absorbance using Beer's Law, since absorbance is proportional to concentration.

Table C-3 Wavelength Cutoffs for Different Mobile Phases

Mobile Phase	UV Cutoff (nm)	Mobile Phase	UV Cutoff (nm)
Acetic acid, 1%	230	Sodium chloride, 1 M	207
Ammonium acetate, 10 mM	205	Sodium citrate, 10 mM	225
Ammonium bicarbonate, 10 mM	190	Sodium dodecyl sulfate	190
BRIJ 35, 0.1%	190	Sodium formate, 10 mM	200
CHAPS, 0.1%	215	Triethyl amine, 1%	235
Diammonium phosphate, 50 mM	205	Trifluoroacetic acid, 0.1%	190
EDTA, disodium, 1 mM	190	TRIS HCl, 20 mM, pH 7.0, pH 8.0	202, 212
HEPES, 10 mM, pH 7.6	225	Triton-X™ 100, 0.1%	240
Hydrochloric acid, 0.1%	190	Waters PIC® Reagent A, 1 vial/liter	200
MES, 10 mM, pH 6.0	215	Waters PIC Reagent B-6, 1 vial/liter	225
Potassium phosphate, monobasic, 10 mM	190	Waters PIC Reagent B-6, low UV, 1 vial/liter	190
Potassium phosphate, dibasic, 10 mM	190		
Sodium acetate, 10 mM	205	Waters PIC Reagent D-4, 1 vial/liter	190

Wavelength Selection for Chromophore Detection

Certain functional groups found in most compounds absorb light selectively. These groups, known as chromophores, and their behavior can be used to categorize the detection of sample molecules.

[Table C-4](#)¹ lists some common chromophores, and their detection wavelengths (λ_{\max}), as well as the molar absorptivity (ϵ_{\max}) of each group. Use this information as a guide to select the optimal operating wavelength for a particular analysis. Because of the diversity

possible within a given sample, scanning over a range of wavelengths may be necessary to determine the best wavelength for a particular analysis.

Table C-4 Electronic Absorption Bands of Representative Chromophores

Chromophore	Chemical Configuration	λ_{max} (nm)	ϵ_{max} (L/m/cm)	λ_{max} (nm)	ϵ_{max} (L/m/cm)
Ether	—O—	185	1000		
Thioether	—S—	194	4600	215	1600
Amine	—NH ₂	195	2800		
Thiol	—SH	195	1400		
Disulfide	—S—S—	194	5500	255	400
Bromide	—Br	208	300		
Iodide	—I	260	400		
Nitrile	—C \equiv N	160	—		
Acetylide	—C \equiv C—	175–180	6000		
Sulfone	—SO ₂ —	180	—		
Oxime	—NOH	190	5000		
Azido	>C=N—	190	5000		
Ethylene	—C=C—	190	8000		
Ketone	>C=O	195	1000	270–285	18–30
Thioketone	>C=S	205	strong		
Esters	—COOR	205	50		
Aldehyde	—CHO	210	strong	280–300	11–18
Carboxyl	—COOH	200-210	50–70		
Sulfoxide	>S—O	210	1500		
Nitro	—NO ₂	210	strong		
Nitrile	—ONO	220-230	1000–2000	300–400	10
Azo	—N=N—	285-400	3–25		
Nitroso	—N=O	302	100		
Nitrate	—ONO ₂	270 (shoulder)	12		

1. Willard, H. H. and others. *Instrumental Methods of Analysis*, 6th ed. Litton Educational Publishing, Inc., 1981. Reprinted by permission of Wadsworth Publishing Co., Belmont, California, 94002.

Table C-4 Electronic Absorption Bands of Representative Chromophores (*Continued*)

Chromophore	Chemical Configuration	λ_{max} (nm)	ϵ_{max} (L/m/cm)	λ_{max} (nm)	ϵ_{max} (L/m/cm)
Allene	$\text{—(C=C)}_2\text{—}$ (acyclic)	210–230	21,000		
Allene	$\text{—(C=C)}_3\text{—}$	260	35,000		
Allene	$\text{—(C=C)}_4\text{—}$	300	52,000		
Allene	$\text{—(C=C)}_5\text{—}$	330	118,000		
Allene	$\text{—(C=C)}_2\text{—}$ (alicyclic)	230–260	3000–8000		
Ethylenic/ Acetylenic	$\text{C=C—C}\equiv\text{C}$	219	6,500		
Ethylenic/Amido	C=C—C=N	220	23,000		
Ethylenic/ Carbonyl	C=C—C=O	210–250	10,000– 20,000		
Ethylenic/Nitro	C=C—NO_2	229	9,500		

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