

Thermo

EASY-nLC Series Troubleshooting and Maintenance Guide

(Software version 3.1)

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Release history: Revision A, March 2012; Revision B, January 2013. Revision B includes information about replacing a PLF pump with a PLU pump, information about upgrading the touch-screen software, and improved troubleshooting procedures.

Hardware versions: EASY-nLC II and EASY-nLC 1000 instruments

Software version: Touch-screen software version 3.1

For Research Use Only. Not for use in diagnostic procedures.



Regulatory Compliance

Thermo Fisher Scientific performs complete testing and evaluation of its products to ensure full compliance with applicable domestic and international regulations. When the instrument is delivered to you, it meets all pertinent electromagnetic compatibility (EMC) and safety standards as described in the Declaration of Conformity.

Changes that you make to the instrument might void compliance with one or more of these EMC and safety standards. Changes to the instrument include replacing a part or adding components, options, or peripherals not specifically authorized and qualified by Thermo Fisher Scientific. To ensure continued compliance with EMC and safety standards, replacement parts and additional components, options, and peripherals must be ordered from Thermo Fisher Scientific or one of its authorized representatives.

FCC Compliance Statement

THIS DEVICE COMPLIES WITH PART 15 OF THE FCC RULES. OPERATION IS SUBJECT TO THE FOLLOWING TWO CONDITIONS: (1) THIS DEVICE MAY NOT CAUSE HARMFUL INTERFERENCE, AND (2) THIS DEVICE MUST ACCEPT ANY INTERFERENCE RECEIVED, INCLUDING INTERFERENCE THAT MAY CAUSE UNDESIRED OPERATION.



CAUTION Read and understand the various precautionary notes, signs, and symbols contained inside this manual pertaining to the safe use and operation of this product before using the device.

Notice on Lifting and Handling of Thermo Scientific Instruments

For your safety, and in compliance with international regulations, the physical handling of this Thermo Fisher Scientific instrument **requires a team effort** to lift and/or move the instrument. This instrument is too heavy and/or bulky for one person alone to handle safely.



Notice on the Proper Use of Thermo Scientific Instruments

In compliance with international regulations: This instrument must be used in the manner specified by Thermo Fisher Scientific to ensure protections provided by the instrument are not impaired. Deviations from specified instructions on the proper use of the instrument include changes to the system and part replacement. Accordingly, order replacement parts from Thermo Fisher Scientific or one of its authorized representatives.

Notice on the Susceptibility to Electromagnetic Transmissions

Your instrument is designed to work in a controlled electromagnetic environment. Do not use radio frequency transmitters, such as mobile phones, in close proximity to the instrument.

For manufacturing location, see the label on the instrument.



Declaration of Conformity for the EASY-nLC II System

U.S. Safety and EMC (Electromagnetic Compliance) Standards

Safety

This instrument has been reviewed for compliance with standard ANSI/UL 3101-1, "Electrical Equipment for Laboratory Use; Part 1: General Requirements," 1st Edition.

EMC

This device complies with Part 15 of the FCC Rules. Operation is subject to the following two conditions:

1. This device may not cause harmful interference, and

2. This device must accept any interference received, including interference that may cause undesired operation.



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

Note

This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to Part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case users will be required to correct the interference at their own expense.

You must use shielded cables with this unit to ensure compliance with the Class A FCC limits.



Canadian Safety and EMC (Electromagnetic Compliance) Standards

Safety

This instrument has been reviewed for compliance with standard CAN/CSA-C22.2 No. 61010-1, Second edition - "Safety Requirements for Electrical Equipment for Measurement, Control, and Laboratory Use; Part 1: General Requirements."

EMC

This Class A digital apparatus meets all requirements of the Canadian Interference-Causing Equipment Regulations.

European Safety and EMC (Electromagnetic Compliance) Standards

Application of Council	2006/95/EEC "Low Voltage": Intertek Group plc
Directive(s)	89/336/EEC "Electromagnetic Compatibility": DELTA Denmark
Standard(s) to which conformity is declared	EN61010-1:2001, Second edition - "Safety Requirements for Electrical Equipment for Measurement, Control and Laboratory Use"
	EN61010-2-81 "Part 2-081: Particular requirements for automatic and semi-automatic laboratory equipment for analysis and other purposes"
	EN/(IEC) 61326-1:2006, A1(1998), A2(2001) and A3(2003) "EMC requirements for electrical equipment for measurement, control and laboratory use"
Manufacturer's Name	Proxeon Biosystems A/S
Manufacturer's Address	Edisonsvej 4, DK-5000 Odense, Denmark
Type of Equipment	Laboratory Instrumentation
Model Name	EASY-nLC [™] II
Model Numbers	LC110/LC111
Serial Number	LC-000100 and later
Year of Manufacture	2010-

I, the undersigned hereby declare that the equipment specified above conforms to the above Directive(s) and Standard(s).

Ole Vorm, Site Manager Aug

August 31, 2010



Declaration of Conformity for the EASY-nLC 1000 System

U.S. Safety and EMC (Electromagnetic Compliance) Standards

Safety

This instrument has been reviewed for compliance with standard UL 61010-1, "Electrical Equipment for Measurement, Control, and Laboratory Use; Part 1: General Requirements."

EMC

This device complies with Part 15 of the FCC Rules. Operation is subject to the following two conditions:

1. This device may not cause harmful interference, and

2. This device must accept any interference received, including interference that may cause undesired operation.



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

Note

This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to Part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case users will be required to correct the interference at their own expense.

You must use shielded cables with this unit to ensure compliance with the Class A FCC limits.

Canadian Safety and EMC (Electromagnetic Compliance) Standards

Safety

This instrument has been reviewed for compliance with standard CAN/CSA-C22.2 No. 61010-1, Second edition - "Safety Requirements for Electrical Equipment for Measurement, Control, and Laboratory Use; Part 1: General Requirements."

EMC

This Class A digital apparatus meets all requirements of the Canadian Interference-Causing Equipment Regulations.



European Safety and EMC (Electromagnetic Compliance) Standards

Application of Council	2004/108/EC "Electromagnetic Compatibility"
Directive(s)	2006/95/EC "Low Voltage Directive (LVD)"
Standard(s) to which conformity is declared	 IEC 61010-1: Second edition, 2001. Safety Requirements for Electrical Equipment for Measurement, Control and Laboratory Use. Also conformity with EN 61010-1:2001 UL 61010-1, second edition CAN/CSA-C22.2 No. 61010-1, Second Edition EN/(IEC) 61326-1:2006. Electrical equipment for measurement, control and laboratory use - EMC requirements - Part 1: General requirements EN/(IEC) 61000-3-2:2006+A1+A2. Electromagnetic compatibility (EMC) - Part 3-2: Limits - Limits for harmonic current emissions (equipment input current ≤16 A per phase) EN/(IEC) 61000-3-3:2008. Electromagnetic compatibility (EMC) - Part 3-3: Limits - Limitation of voltage changes, voltage fluctuations and flicker in public low-voltage supply systems, for equipment with rated current ≤16 A per phase and not subject to conditional connection FCC CFR 47 Part 15, Class A. Radiated Emissions and Conducted Emissions
Manufacturer's Name	Proxeon Biosystems A/S
M	Editor mil 4 DK 5000 Odana Danmala
Manufacturer's Address	Edisonsvej 4, DK-5000 Odense, Denmark
Type of Equipment	Laboratory Instrumentation
Model Name	EASY-nLC [™] 1000
Model Numbers	LC120
Serial Number	LC-010000 and higher
Year of Manufacture	2011–

I, the undersigned hereby declare that the equipment specified above conforms to the above Directive(s) and Standard(s).

Ole Vorm, Site Manager July 19, 2011



WEEE Compliance

This product is required to comply with the European Union's Waste Electrical & Electronic Equipment (WEEE) Directive 2002/96/EC. It is marked with the following symbol:



Thermo Fisher Scientific has contracted with one or more recycling or disposal companies in each European Union (EU) Member State, and these companies should dispose of or recycle this product. See www.thermoscientific.com/ rohsweee for further information on Thermo Fisher Scientific's compliance with these Directives and the recyclers in your country.

WEEE Konformität

Dieses Produkt muss die EU Waste Electrical & Electronic Equipment (WEEE) Richtlinie 2002/96/EC erfüllen. Das Produkt ist durch folgendes Symbol gekennzeichnet:



Thermo Fisher Scientific hat Vereinbarungen mit Verwertungs-/Entsorgungsfirmen in allen EU-Mitgliedsstaaten getroffen, damit dieses Produkt durch diese Firmen wiederverwertet oder entsorgt werden kann. Mehr Information über die Einhaltung dieser Anweisungen durch Thermo Fisher Scientific, über die Verwerter, und weitere Hinweise, die nützlich sind, um die Produkte zu identifizieren, die unter diese RoHS Anweisung fallen, finden sie unter www.thermoscientific.com/rohsweee.



Conformité DEEE

Ce produit doit être conforme à la directive européenne (2002/96/EC) des Déchets d'Equipements Electriques et Electroniques (DEEE). Il est marqué par le symbole suivant:



Thermo Fisher Scientific s'est associé avec une ou plusieurs compagnies de recyclage dans chaque état membre de l'union européenne et ce produit devrait être collecté ou recyclé par celles-ci. Davantage d'informations sur la conformité de Thermo Fisher Scientific à ces directives, les recycleurs dans votre pays et les informations sur les produits Thermo Fisher Scientific qui peuvent aider la détection des substances sujettes à la directive RoHS sont disponibles sur www.thermoscientific.com/rohsweee.

CAUTION Symbol	CAUTION	VORSICHT	ATTENTION	PRECAUCION	AVVERTENZA
	Electric Shock: This instrument uses high voltages that can cause personal injury. Before servicing, shut down the instrument and disconnect the instrument from line power. Keep the top cover on while operating the instrument. Do not remove protective covers from PCBs.	Elektroschock: In diesem Gerät werden Hochspannungen verwendet, die Verletzungen verursachen können. Vor Wartungsarbeiten muß das Gerät abgeschaltet und vom Netz getrennt werden. Betreiben Sie das Gerät nicht mit abgenommenem Deckel. Nehmen Sie die Schutzabdeckung von Leiterplatten nicht ab.	Choc électrique: L'instrument utilise des tensions capables d'infliger des blessures corporelles. L'instrument doit être arrêté et débranché de la source de courant avant tout intervention. Ne pas utiliser l'instrument sans son couvercle. Ne pas enlever les étuis protecteurs des cartes de circuits imprimés.	Descarga eléctrica: Este instrumento utiliza altas tensiones, capaces de producir lesiones personales. Antes de dar servicio de mantenimiento al instrumento, éste debera apagarse y desconectarse de la línea de alimentacion eléctrica. No opere el instrumento sin sus cubiertas exteriores quitadas. No remueva las cubiertas protectoras de las tarjetas de circuito impreso.	Shock da folgorazione. L'apparecchio è alimentato da corrente ad alta tensione che puo provocare lesioni fisiche. Prima di effettuare qualsiasi intervento di manutenzione occorre spegnere ed isolare l'apparecchio dalla linea elettrica. Non attivare lo strumento senza lo schermo superiore. Non togliere i coperchi a protezione dalle schede di circuito stampato (PCB).
	Chemical: This instrument might contain hazardous chemicals. Wear gloves when handling toxic, carcinogenic, mutagenic, or corrosive or irritant chemicals. Use approved containers and proper procedures to dispose waste oil.	Chemikalien: Dieses Gerät kann gefährliche Chemikalien enthalten. Tragen Sie Schutzhandschuhe beim Umgang mit toxischen, karzinogenen, mutagenen oder ätzenden/reizenden Chemikalien. Entsorgen Sie verbrauchtes Öl entsprechend den Vorschriften in den vorgeschriebenen Behältern.	Chimique: Des produits chimiques dangereux peuvent se trouver dans l'instrument. Portez des gants pour manipuler tous produits chimiques toxiques, cancérigènes, mutagènes, ou corrosifs/irritants. Utiliser des récipients et des procédures homologuées pour se débarrasser des déchets d'huile.	Química: El instrumento puede contener productos quimicos peligrosos. Utilice guantes al manejar productos quimicos tóxicos, carcinogenos, mutagenos o corrosivos/irritantes. Utilice recipientes y procedimientos aprobados para deshacerse del aceite usado.	Prodotti chimici. Possibile presenza di sostanze chimiche pericolose nell'apparecchio. Indossare dei guanti per maneggiare prodotti chimici tossici, cancerogeni, mutageni, o corrosivi/irritanti. Utilizzare contenitori aprovo e seguire la procedura indicata per lo smaltimento dei residui di olio.
	Heat: Before servicing the instrument, allow any heated components to cool.	Hitze: Warten Sie erhitzte Komponenten erst nachdem diese sich abgekühlt haben.	Haute Temperature: Permettre aux composants chauffés de refroidir avant tout intervention.	Altas temperaturas: Permita que los componentes se enfríen, ante de efectuar servicio de mantenimiento.	Calore. Attendere che i componenti riscaldati si raffreddino prima di effetturare l'intervento di manutenzione.
	Fire: Use care when operating the system in the presence of flammable gases.	Feuer: Beachten Sie die einschlägigen Vorsichtsmaßnahmen, wenn Sie das System in Gegenwart von entzündbaren Gasen betreiben.	Incendie: Agir avec précaution lors de l'utilisation du système en présence de gaz inflammables.	Fuego: Tenga cuidado al operar el sistema en presencia de gases inflamables.	Incendio. Adottare le dovute precauzioni quando si usa il sistema in presenza di gas infiammabili.
	Eye Hazard: Eye damage could occur from splattered chemicals or flying particles. Wear safety glasses when handling chemicals or servicing the instrument.	Verletzungsgefahr der Augen: Verspritzte Chemikalien oder kleine Partikel können Augenverletzungen verursachen. Tragen Sie beim Umgang mit Chemikalien oder bei der Wartung des Gerätes eine Schutzbrille.	Danger pour les yeux: Des projections chimiques, liquides, ou solides peuvent être dangereuses pour les yeux. Porter des lunettes de protection lors de toute manipulation de produit chimique ou pour toute intervention sur l'instrument.	Peligro par los ojos: Las salicaduras de productos químicos o particulas que salten bruscamente pueden causar lesiones en los ojos. Utilice anteojos protectores al manipular productos químicos o al darle servicio de mantenimiento al instrumento.	Pericolo per la vista. Gli schizzi di prodotti chimici o delle particelle presenti nell'aria potrebbero causare danni alla vista. Indossare occhiali protettivi quando si maneggiano prodotti chimici o si effettuano interventi di manutenzione sull'apparecchio.
	General Hazard: A hazard is present that is not included in the above categories. Also, this symbol appears on the instrument to refer the user to instructions in this manual.	Allgemeine Gefahr: Es besteht eine weitere Gefahr, die nicht in den vorstehenden Kategorien beschrieben ist. Dieses Symbol wird im Handbuch außerdem dazu verwendet, um den Benutzer auf Anweisungen hinzuweisen.	Danger général: Indique la présence d'un risque n'appartenant pas aux catégories citées plus haut. Ce symbole figure également sur l'instrument pour renvoyer l'utilisateur aux instructions du présent manuel.	Peligro general: Significa que existe un peligro no incluido en las categorias anteriores. Este simbolo también se utiliza en el instrumento par referir al usuario a las instrucciones contenidas en este manual.	Pericolo generico. Pericolo non compreso tra le precedenti categorie. Questo simbolo è utilizzato inoltre sull'apparecchio per segnalare all'utente di consultare le istruzioni descritte nel presente manuale.

When the safety of a procedure is questionable, contact your local Technical Support organization for Thermo Fisher Scientific San Jose Products.

Wenn Sie sich über die Sicherheit eines Verfahrens im unklaren sind, setzen Sie sich, bevor Sie fortfahren, mit Ihrer lokalen technischen Unterstützungsorganisation für Thermo Fisher Scientific San Jose Produkte in Verbindung.

Si la sûreté d'une procédure est incertaine, avant de continuer, contacter le plus proche Service Clientèle pour les produits de Thermo Fisher Scientific San Jose.

Cuando la certidumbre acerca de un procedimiento sea dudosa, antes de proseguir, pongase en contacto con la Oficina de Asistencia Tecnica local para los productos de Thermo Fisher Scientific San Jose.

Quando e in dubbio la misura di sicurezza per una procedura, prima di continuare, si prega di mettersi in contatto con il Servizio di Assistenza Tecnica locale per i prodotti di Thermo Fisher Scientific San Jose.

CAUTION Symbol	CAUTION	危険警告	危險警告
	Electric Shock: This instrument uses high voltages that can cause personal injury. Before servicing, shut down the instrument and disconnect the instrument from line power. Keep the top cover on while operating the instrument. Do not remove protective covers from PCBs.	電撃:この計測器は高電圧を使用し、人体に危害を与える可能性があります。 保守・修理は、必ず操業を停止し、電源を切ってから実施して下さい。上部カ バーを外したままで計測器を使用しないで下さい。プリント配線 板の保護カバーは外さないで下さい。	電擊:儀器設備使用會造成人身傷害的高伏電壓。在維修之前, 必須先關儀器設備並切除電源。務必要在頂蓋蓋上的情況下操作 儀器。請勿拆除PCB保護蓋。
	Chemical: This instrument might contain hazardous chemicals. Wear gloves when handling toxic, carcinogenic, mutagenic, or corrosive or irritant chemicals. Use approved containers and proper procedures to dispose waste oil.	化学物質:危険な化学物質が計測器中に存在している可能性があります。毒性、 発がん性、突然変異性、腐食・刺激性などのある薬品を取り扱う際は、手袋を 着用して下さい。廃油の処分には、規定の容器と手順を使用して下さい。	化學品:儀器設備中可能存在有危險性的化學物品。接觸毒性 致癌、誘變或腐蝕/刺激性化學品時,請配帶手套。處置廢油 時,請使用經過許可的容器和程序。
	Heat: Before servicing the instrument, allow any heated components to cool.	熱:熱くなった部品は冷えるのを待ってから保守 [↓] ・修理を行って下さい。	高溫:請先等高溫零件冷卻之後再進行維修。
	Fire: Use care when operating the system in the presence of flammable gases.	火災 :可燃性のガスが存在する場所でシステムを操作する場合は、充分な注意 を払って下さい。	火災:在有易燃氣體的場地操作該系統時,請務必小心謹慎。
	Eye Hazard: Eye damage could occur from splattered chemicals or flying particles. Wear safety glasses when handling chemicals or servicing the instrument.	眼に対する危険:化学物質や微粒子が飛散して眼を傷つける危険性がありま す。化学物質の取り扱い、あるいは計測器の保守・修理に際しては防護眼鏡を 着用して下さい。	眼睛傷害危險:飛濺的化學品或顆粒可能造成眼睛傷害。處理化 學品或維修儀器設備時請佩戴安全眼鏡。
	General Hazard: A hazard is present that is not included in the above categories. Also, this symbol appears on the instrument to refer the user to instructions in this manual.	ー 般的な危険: この標識は上記以外のタイプの危険が存在することを示しま す。また、計測器にこの標識がついている場合は、本マニュアル中の指示を参 照して下さい。	一般性危險:說明未包括在上述類別中的其他危險。此外,儀器 設備上使用這個標誌,以指示用戶本使用手册中的説明。
	When the safety of a procedure is questionable, contact your local Technical Support organization for Thermo Fisher Scientific San Jose Products.	安全を確保する手順がよくわからない時は、作業を一時中止し、お近く のサーモエレクトロンサンローゼプロダクトのテクニカールサポートセ ンターごご連絡ください。	如对安全程序有疑问,请在操作之前与当地的菲尼根技术服务中心联系。

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Preface

This guide describes the maintenance and troubleshooting procedures required to maintain the EASY-nLC[™] instrument in optimal working condition.

The hardware maintenance procedures are divided into two chapters: Routine Maintenance and Advanced Maintenance. The Routine Maintenance chapter contains common maintenance procedures that you can perform to keep the EASY-nLC instrument in optimal working condition. The Advanced Maintenance chapter contains hardware repair procedures that can only be performed by a Thermo Fisher Scientific field service engineer or a skilled technician with equivalent training.

For information about setting up the EASY-nLC instrument and running chromatographic separations, refer to the *EASY-nLC Series Getting Started Guide*.

Contents

- Related Documentation
- Downloading Manuals from the Customer Manuals Web Site
- Safety and Special Notices
- Contacting Us

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Related Documentation

In addition to this guide, Thermo Fisher Scientific provides the following documents for the EASY-nLC instrument:

- EASY-nLC Series User Guide for the Xcalibur 2.x Data System
- EASY-nLC Preinstallation Requirements Guide
- EASY-nLC Series Getting Started Guide

Downloading Manuals from the Customer Manuals Web Site

For Thermo Scientific software and hardware products, Thermo Fisher Scientific provides instructional guides as PDF files on its Customer Manuals Web site.

Note If you are using a Thermo Scientific application to control the EASY-nLC system as part of an LC/MS system, the EASY-nLC manuals are also available from the data system computer's Start menu.

- To download the EASY-nLC manuals from the Customer Manuals Web site
- 1. Go to mssupport.thermo.com.
- 2. On the Terms and Conditions page, click I Agree.
- 3. Click Customer Manuals in the left margin of the window.
- 4. On the right side of the window, click Search.

The Documents - Advanced Search page opens.

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The VIP (Virtual Instrument Partner) Program	Document Type: Part Number:	Manual	▼	
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- 5. Open a list of linked PDF documents for the EASY-nLC as follows:
 - a. In the Model list, select EASY-nLC and in the Document Type list, select Manual.
 - b. Click **Submit Search**.

- 6. On the Documents page, click the link to the right of Download to view the PDF.
- 7. Click the **Save a Copy** icon, [], browse to an appropriate folder, and then click **Save** to save a copy of the document.

Safety and Special Notices

Make sure to follow the safety practices presented in this guide, and observe the safety and special notices that appear in boxes.

Observe all written safety precautions during all phases of operation, service, and repair of this instrument. Failure to comply with these precautions or with specific warnings elsewhere in this manual violates safety standards of design, manufacture, and intended use of the instrument and might result in injury or loss of life.

The following two stickers appear on the EASY-nLC instrument:

This sticker warns you that the instrument includes a sharp needle and moving parts that are accessible to the operator. To prevent personal injury or damage to parts of the EASY-nLC instrument, take care when loading samples into the instrument's tray compartment.



This sticker alerts you to consult this manual for instructions on how to operate the instrument.



The safety and special notices in the documentation include the following:



CAUTION Highlights hazards to humans, property, or the environment. Each CAUTION notice is accompanied by an appropriate CAUTION symbol.



CAUTION Highlights electrical hazards to humans or property.



CAUTION Highlights a sharp object hazard to humans.



CAUTION Highlights an eye hazard to humans.



CAUTION Highlights a chemical hazard to humans, property, or the environment.



CAUTION Highlights lifting hazards.

IMPORTANT Highlights information necessary to prevent damage to software, loss of data, or invalid test results; or might contain information that is critical for optimal performance of the system.

Note Highlights information of general interest.

Tip Highlights helpful information that can make a task easier.

Contacting Us

There are several ways to contact Thermo Fisher Scientific for the information you need.

* To contact Technical Support for the EASY-nLC instrument

Web site	www.proxeon.com
E-mail	(North and South America)us.customer-support.analyze@thermofisher.com(Other continents)eu.techsupport.cmf@thermofisher.com
Address	Thermo Fisher Scientific Edisonvej 4 DK-5000 Odense C

Find software updates and utilities to download at mssupport.thermo.com.

* To order consumable and spare parts for the EASY-nLC instrument

For the EASY-nLC 1000 instrument, go to: www.proxeon.com/productrange/nano_lc_easy-nlc_1000/accessories_spares/index.html

For the EASY-nLC II instrument, go to: www.proxeon.com/productrange/nano_lc/accessories-spares/index.html

Introduction

The EASY-nLC instrument (see Figure 1) consists of a high-pressure dual pump chromatography system that provides split-free flows down to the low nanoLiter/min range, an automated sampling system, and a built-in computer with a touch-screen interface for instrument control and troubleshooting. The automated sampling system includes a tray compartment that can hold standard sample vials and microtiter plates, an XYZ robot that moves to the sample location, and a syringe pump that draws only the user-specified sample volume.

The EASY-nLC II instrument operates in the high-performance pressure range of 1 to 300 bar and the EASY-nLC 1000 instrument operates in the ultra-high-performance pressure range of 1 to 1000 bar.





To familiarize yourself with the EASY-nLC instrument and its instrument control software, review these topics.

Contents

- Hardware Components
- Touch-Screen Application
- Specifications

Note For information about setting up the EASY-nLC instrument, installing the columns, creating methods, and running batches, refer to the *EASY-nLC Series Getting Started Guide*.

Hardware Components

These topics describe the hardware components of the EASY-nLC instrument:

- Autosampler
- Solvent System Components Behind the Right Side Panel
- Computer and Autosampler Behind the Left Side Panel
- Back Panel

Autosampler

The autosampler module of the EASY-nLC instrument is located behind the tray compartment door. You can open the tray compartment door manually or by using the touch-screen controls.

With the use of a vial adapter plate or a microwell adapter plate, the tray compartment can hold sample vials, a 96-well microplate, or a 384-well microplate.

The robotic unit moves the *z*-axis needle holder along the *x* and *y* axes to the user-specified sample location during a run. When the autosampler needle reaches the sample location, it descends into the sample vial or microplate well.

IMPORTANT Because they can block the movement of the *z*-axis needle holder, avoid placing tall objects into the tray compartment.

The autosampler needle draws solvent from wash bottle W3 to perform a standard wash and ejects the solvent into wash bottle W4, which contains the needle wash insert. The autosampler can also perform a custom wash using the solvents from wash bottles W1–W3.

The waste beaker to the right of the tray compartment collects solvent that is delivered by pumps A and B when these pumps empty solvent to waste. This waste beaker also collects solvent delivered by pump A during the precolumn equilibration and sample loading steps (for a two-column setup).

The EASY-nLC Series includes two supported versions of the autosampler module:

- "ASC Model—Current" on page 3
- "ASA Model—Legacy EASY-nLC II Instruments" on page 4

Note The ASC model has an integrated cooling compartment, which ensures stable temperature conditions throughout the entire vial plate. By contrast, the ASA model is cooled from underneath only.

The following differences between the two autosampler models affect the routine calibration and maintenance procedures:

- The *z*-axis needle holders and the autosampler needles. This difference affects the autosampler needle replacement procedure.
- The location of the wash bottles. This difference affects the autosampler calibration procedure.
- The adapter plate holder. The adapter plate holder for the ASC model holds an additional six sample vials. This difference affects the autosampler calibration procedure and the maximum number of sample vials that you can run without reloading the autosampler.

ASC Model—Current

Figure 2 shows the ASC autosampler model that is installed in the current versions of the EASY-nLC II and EASY-nLC 1000 instruments.





Figure 3 shows the tray compartment for the ASC autosampler as viewed from above when the tray compartment door is open. The tray compartment of the ASC autosampler model holds a vial adapter or a well plate adapter, six additional vials, three wash solvent bottles, and a wash bottle with a wash insert for cleaning the autosampler needle.



Figure 3. Tray compartment for the ASC autosampler model (viewed from above)

Well plate (96 or 384) or vial (6 × 8) adapter Three positions for wash solvents

Waste bottle with wash insert for cleaning of the outside of the needle

Six extra vial positions for standards or regular samples

ASA Model—Legacy EASY-nLC II Instruments

Figure 4 shows the ASA autosampler model that is installed in earlier versions of the EASY-nLC II instrument.





Figure 5 shows the tray compartment for the ASA autosampler model as viewed from above when the tray compartment door is open. The tray compartment of the ASA autosampler model holds a vial adapter or a well plate adapter, three wash solvent bottles, and a wash bottle with a wash insert for cleaning the autosampler needle. The ASA autosampler model does not hold six additional vials.



Figure 5. Tray compartment for the ASA autosampler model (viewed from above)

Solvent System Components Behind the Right Side Panel

The EASY-nLC solvent delivery system is located behind the right side panel and consists of three syringe pumps, three pressure sensors, two flow sensors, four rotary valves, four check valves (two for each assembly), and a mixing Tee.

Note Two of the syringe pumps (pumps A and B) produce the binary mobile phase and the third syringe pump (pump S) draws the sample into the sample loop that is attached to one of the rotary valves (valve S).

These topics describe the hardware components behind the right side panel:

- View Behind the Right Side Panel
- Syringe Pumps
- Pressure Sensors
- Flow Sensors
- Check Valve Assemblies
- Six-Port Rotary Valves
- Autosampler Needle

View Behind the Right Side Panel

You can access the solvent system components by removing the instrument's right side panel, which is secured to the housing with three captive quarter-turn screws.

Figure 6 shows the EASY-nLC II solvent system.

Figure 6. Solvent system components behind the right panel of the EASY-nLC II instrument





Figure 7. Solvent system components behind the right panel of the EASY-nLC 1000 instrument



Syringe Pumps

The EASY-nLC instrument contains three syringe pumps. The binary liquid chromatography system consists of two high-pressure pumps: pumps A and B. The automated sampling system uses syringe pump S to draw sample from a sample vial or well and load it into the sample loop attached to valve S.

Figure 8 shows the EASY-nLC syringe pump components. The PLF model is installed in the EASY-nLC II instrument. The PLU model is installed in the EASY-nLC 1000 instrument and is supplied as spare part for both instruments.

Each syringe pump consists of these primary components: a stepper motor, a pump body, a pump head, a piston, a piston seal, and a backup ring. The PLU model also has an LED panel and a pump printed circuit board (PCB) cover. The piston seal is the only user-replaceable component.

The pump head holds up to 140 μ L of fluid. The backward movement of the piston draws solvent into the pump head and the forward movement of the piston pushes solvent into the attached high-pressure solvent line. The stepper motor controls the forward and backward movement of the piston within the pump head. The pump body provides piston alignment.

The piston seal allows the piston to move freely within the pump head. The piston seal is made of an extremely strong thermoplastic. The seal for the EASY-nLC 1000 pumps contains a wire spring that forces the inside flange of the seal against the piston to prevent leaks. The seals are not leak proof. In fact, wetting the surface of the pistons requires a small quantity of liquid, which acts as a lubricant to reduce wear on the piston seal. Through continued use and over time, the seal degrades and must be replaced. Allowing the pump to run dry and neglecting to rinse the pump head assemblies after pumping buffered eluents shorten the lifespan of the seals.

The backup ring holds the piston seal in place. It also provides better alignment of the piston.



Figure 8. Syringe pump components (showing the pump head removed from the pump body)

EASY-nLC II syringe pump (PLF model)

EASY-nLC 1000 syringe pump (PLU model)

The syringe pumps perform these functions.

Component	Function
Pumps A and B	Draw solvent from the solvent bottles on top of the instrument, empty solvent to the waste beaker, and deliver solvent to the mixing Tee.
Pump S	Draws solvent from the W3 bottle, draws sample from the specified sample location into the autosampler needle, and empties solvent to the W4 bottle in the autosampler tray compartment.

For maintenance purposes, the system tracks the volume pumped by each pump (see "Checking and Resetting the Device Usage Counters" on page 56).

The PLF model pumps installed in the EASY-nLC instrument have an error LED. The LED illuminates green when the instrument is turned on and the pump is functioning properly.

The LED panels for the PLU model pumps installed in the EASY-nLC 1000 instrument provide the following status information.

LED	States
Flow sensor for Pumps A and B	Illuminates blue—the instrument is turned on and the flow sensor is connected to the pump. Not illuminated—the instrument is not turned on or the flow sensor is not connected to the pump.
Power	Illuminates yellow—The instrument power is turned on. Not illuminated—The instrument power is turned off.
Error	Illuminates red—A pump error has occurred. Not illuminated—The pump is operating properly.

Pressure Sensors

For pumps A, B, and S, the three pressure sensors connect the solvent path between the pump outlet and the associated rotary valve.

Using the strain gauge principle to measure pressure accurately, the pressure sensors are flow-through, zero dead volume devices with a wetted path that contains 12 μ L of swept volume. The pressure sensors for the EASY-nLC II instrument have an operating range of 0 to 5000 psi (345 bar), and the pressure sensors for the EASY-nLC 1000 instrument have an operating range of 0 to 15 000 psi (1034 bar).

Flow Sensors

The two flow sensors provide feedback control for pumps A and B. The flow sensors are liquid mass flow meters with a CMOSens[™] patented technology. Each flow meter consists of a length of fused silica capillary, a sensor chip, a heating resistor, and two temperature sensors (see Figure 9).



Figure 9. Flow sensor schematic

The flow sensors determine the flow rate of solvents passing through the system by measuring the heat transfer through the fused silica capillary. Outside the capillary, a heating resistor on a thermally optimized membrane is maintained above ambient temperature. When liquid flows through the capillary, the temperature distribution upstream and downstream of the heating resistor is disturbed. The two temperature sensors measure this temperature asymmetry.

Because the temperature distribution depends on both the solvent flow rate and the solvent mixture, you must recalibrate the flow sensors whenever you change the solvent types for solvents A and B. For example, if you use methanol instead of acetonitrile for the solvent B mixture, run the script "Calibrate – Flow Sensors" on page 51.

The EASY-nLC 1000 instrument uses two flow sensors with a maximum measuring range of 5.0 μ L/min. The EASY-nLC II instrument uses two different flow sensors. The maximum measuring range is approximately 2.9 μ L/min for flow sensor A and 4.5 μ L/min for flow sensor B.

The wetted path for the EASY-nLC 1000 instrument includes external inline filters for the flow sensor inlets and outlets. When the inline filters become clogged, they must be replaced. For the EASY-nLC II instrument, the tubing that connects the valve to the flow sensor contains a built-in filter.

Check Valve Assemblies

When the corresponding high-pressure (6-port rotary) valves for pumps A or B are in position 1–2, the check valves perform these functions:

- The solvent-side check valve prevents backflow into the associated solvent inlet line when the associated pump is emptying solvent to the waste beaker.
- The waste-side check valve prevents backflow from the waste beaker when the associated pump is filling and drawing solvent from the associated solvent bottle.

Figure 10. Check valve connections



The check valves are replaceable. For more information about determining if the check valves are leaking, see "Identifying a Leaking Check Valve" on page 248.

Six-Port Rotary Valves

The three-position, six-port rotary valves consist of a driver, a rotor seal, and a stator (see Figure 11).

Driver Rotor seal Stator Screws that secure the stator to the driver

The six-port valves can be in three different positions: 1–2 position, 1–6 position, and centered (see Figure 12).



For maintenance purposes, the system tracks the number of valve shifts for each valve (see "Checking and Resetting the Device Usage Counters" on page 56).

Depending on the valve position, the valves perform the functions listed in Table 1.

Note The EASY-nLC solvent system contains three subsystems, one for each pump. A subsystem includes the pump, the pressure sensor, the solvent lines to and from the pressure sensor, and the rotary valve.

Figure 11. Valve components
Component	Function		
Valves A and B	Direct the solvent flow from syringe pump A and B, respectively.		
	1–2 position	Directs the solvent flow toward the check valve. When the valve is in the 1–2 position, the pump draws solvent from the solvent bottle on top of the instrument or empties solvent to the waste beaker.	
	1–6 position	Directs the solvent flow toward the mixing Tee.	
	Centered	• Prevents backflow into the solvent line upstream of the mixing Tee.	
		• Allows the back pressure for the solvent A and B lines to equalize before the gradient step of a sample run.	
		• Blocks the solvent path through the valve and to the subsystem for several maintenance scripts.	
Valve S	Directs the solv the mixing Tee.	rent flow from syringe pump S and the solvent flow from	
	1–2 position	The solvent flow bypasses the sample loop.	
	1–6 position	The solvent flow passes through the sample loop.	
	Centered	 Isolates the pump S subsystem for the Flush Air script. 	
		• Blocks the solvent path after valve S for the system leak test (Leaks script with the System check box selected).	
		• Shuts off the flow to the Column Out line and provides back pressure during the Flow Sensor Calibration script.	
Valve W	Directs the solvent flow through the venting Tee to the waste beaker or provides pressure venting to the system.		
	1–2 position	Directs the solvent flow toward the column by blocking the solvent path through valve W.	
	1–6 position	Directs the solvent flow from the venting Tee to the waste beaker.	
	Centered	Blocks the solvent flow through the valve to the waste beaker.	

Table 1.Valve functions

Autosampler Needle

During a sample run, the autosampler needle descends into the requested sample vial or well, and then pump S draws the requested sample volume into the needle tubing. The autosampler needle is a user-replaceable part. For information about replacing the autosampler needle, see "Replacing the Autosampler Needle" on page 84.

The autosampler needle for the ASC autosampler is a length of PEEKsil[™] tubing with a sleeve and a stainless steel fitting at one end and a PEEK[™] nut and plastic stop at the other end (see Figure 13). Fitting into a slot in the panel between the solvent system compartment and the tray compartment, the needle retainer properly positions the needle tubing.





Figure 14 shows autosampler needle for the ASA autosampler.

Figure 14. Autosampler needle for the ASA autosampler model (earlier EASY-nLC II instruments)



For both needles, the stainless steel fitting connects to port 1 of valve S. The tubing routes through a cutout in the panel that separates the solvent delivery system components from the autosampler compartment. The other end of the tubing slides into the *z*-axis holder and is secured with a PEEK nut.

Computer and Autosampler Behind the Left Side Panel

You can access the integrated computer and the autosampler needle fitting that connects to the needle guide by removing the left side panel of the instrument (see Figure 15). Three captive quarter-turn screws secure the left side panel to the instrument housing.

Figure 15. Internal features (behind the left side panel)



Back Panel

The system communication, contact closure, and power line connections are on the back panel of the instrument (see Figure 16 and Figure 17).





Figure 17. Back panel of the EASY-nLC instrument (with the latest computer box)



Table 2 lists the function of each back panel element.

Table 2.	Back pane	l elements
----------	-----------	------------

Element	Description
RS-232	For communication with add-on devices, for example a syringe drive
USB	Port for keyboard or mouse or removable USB storage device, two (2) provided
PS/2 (discontinued computer box only)	Input for connection of keyboard and mouse
LAN	For 10/100 MB/sec Ethernet connection
MONITOR	Output for connection of external display
P-BUS	For communication with add-on devices
IN/OUT pins	Contact closure (primarily for communication with the mass spectrometer)
I O	Power switch
Fuse holder below I O switch	 Replaceable fuse ratings: For 120 V, T 5 AL, 250 V For 230 V, T 2.5 AL, 250 V
P/N	Product order code
S/N	Instrument serial number
LAN MAC ADDRESS	MAC address of the embedded computer (Gives the EASY-nLC computer a unique network identifier on your local network.)

Touch-Screen Application

The EASY-nLC instrument includes an integrated computer with instrument control software that you access from the instrument monitor.

These topics provide an overview of the touch-screen application and describe how to log in to the EASY-nLC instrument as an administrator and how to close down the instrument:

- Description of the Touch-Screen Monitor
- Logging In to the EASY-nLC Instrument for Maintenance Tasks
- Closing Down the EASY-nLC Instrument

Description of the Touch-Screen Monitor

The instrument monitor is a transparent layer of glass that detects finger pressure (even while wearing gloves) and sends the corresponding commands to the computer (see Figure 18).



Figure 18. Touch-screen controls

Instrument control from this touch-screen application includes a method wizard that helps you create chromatographic methods, a batch editor that helps you create and run sample batches (injection sequences); maintenance scripts and record keeping features; and direct control of the valves, pumps, XYZ robot, and tray temperature.

Note This guide describes how to log in to the instrument as an administrator, how to use the maintenance scripts and record keeping features, and how to exit the touch-screen application in a controlled manner and turn off the instrument before performing maintenance procedures on the instrument's hardware components.

For information about the maintenance scripts and record keeping features, see Chapter 3, "Maintenance Scripts and Service Records." For information about creating methods and sample batches and running sample batches, refer to the EASY-nLC Series Getting Started Guide (for the touch-screen application).

The EASY-nLC touch-screen application contains five menu tabs at the top of the touch screen. When you press a tab at the top of the touch screen, a set of side tabs appears on the left side of the touch screen. Figure 19 shows the menu layout for the Home > Overview page.





Login and Exit button

During a session, the application tracks which page (side tab) you chose inside each menu (top tab) and shows that page when you next choose that menu.

The Thermo Scientific logo icon in the upper left corner and the Login button in the lower left corner are always available.

To view the version information for the application, press the Logo icon.

Logging In to the EASY-nLC Instrument for Maintenance Tasks

To run the maintenance scripts, use the direct controls, set up the system configuration, and so on, you must log in to the EASY-nLC instrument as an administrator.

* To log in as a user with administrative privileges

1. Press Login in the lower left corner of the touch screen (see Figure 19 on page 19).

The Login As User dialog box opens (see Figure 20).

Figure 20. Login As User dialog box

Login as	user		
User :	admin		•
Password :	****		
Acci	ept	Cancel	

- 2. In the User list, select a user with administrative rights.
- 3. In the Password box, enter the password for this user.

Tip The factory user name and password for the system administrator is admin.

4. Press Accept.

Login	as user	
User	admin	•
Passwoi	:d : *****	
A	ccept	Cancel

Figure 21. EASY-nLC login dialog box

Closing Down the EASY-nLC Instrument

Closing down the instrument in a controlled manner is important to allow all of the components to shut down in an orderly sequence. Using this controlled method saves important data so that the instrument starts with the correct information the next time you use it.

IMPORTANT If you turn off the power switch during normal operation, you risk damaging essential system components.

* To close down the EASY-nLC instrument and turn off the power

1. Press **Exit** (lower-left corner of the screen).

A confirmation dialog box opens (see Figure 22).

Figure 22. Confirmation dialog box that opens after pressing Exit

Administrator	
Administrator	Logout
	Change password
Stop Application Power Down	Cancel

2. Press Power Down.

The EASY-nLC application displays a white screen with a small progress bar. When the progress bar is filled out and the message appears indicating you can safely turn off the instrument, go to the next step.



3. After receiving the message that you can safely turn off the instrument, turn off the power switch on the back panel of the instrument.

Specifications

These topics describe the performance and technical specifications for the EASY-nLC syringe pumps and autosampler:

- Performance Specifications
- Technical Specifications

Performance Specifications

Table 3 lists the performance specifications for the EASY-nLC instruments.Table 3.Performance specification (Sheet 1 of 2)

ltem	Specification
Flow range (gradient)	20 to 2000 nL/min Recommended: 100 to 1000 nL/min
Flow while loading and equilibrating	Up to 25 µL/min
Pressure range	EASY-nLC II: 0 to 300 bar EASY-nLC 1000: 0 to 1000 bar
Retention time reproducibility (RT RSD)	Within the recommended flow range: 0.1 to 0.4% Outside the recommended flow range: typically better than 1%
Peak widths	Typically 3 to 5 seconds full width at half maximum (fwhm), using PepMap [™] nano-bore columns as supplied at delivery
Carryover	Typically < 0.05% Conditions: Injection of 100 fmol tryptic digests of BSA on Pepmap columns as supplied at delivery and by using the standard wash procedure for the autosampler
Autosampler sample pickup volume range	0.1 to 18 μL with the standard 20 μL loop 0.1 to 48 μL with a 50 μL loop
Injection reproducibility (injection RSD)	0.2% at 5 μL pickup 3.0% at 0.1 μL pickup
Injection linearity	0.9985 at 0.5 to 10 μL injection volume

Item	Specification
Autosampler formats	
ASC model	6 × 8 Vials + 6 Vials 1 × 96 well MTP + 6 Vials 1 × 384 well MTP + 6 Vials 2 × 48 PCR strips + 6 Vials 4 × 24 PCR strips + 6 Vials
ASA model (earlier versions of the EASY-nLC II instrument)	6 × 8 Vials 1 × 96 well MTP 1 × 384 well MTP 2 × 48 PCR strips 4 × 24 PCR strips
Autosampler cooling	20 °C below ambient, typical setting: 7 °C (45 °F)

Table 3.Performance specification (Sheet 2 of 2)

Technical Specifications

These tables list the technical specifications for the EASY-nLC instrument:

- General specifications, Table 4
- Physical specifications, Table 5
- Electrical specifications, Table 6
- Communication specifications, Table 7
- Hardware components, Table 8

Table 4. General specifications

ltem	Specification
Sound pressure level	< 70 dBA
Operating temperature	5 to 30 °C
Storage temperature	-25 to +60 °C
Humidity	20 to 80% RH, non-condensing
Sample viscosity	0.1 to 5 cP
Solvent A	Water with 0.1% formic acid
Solvent B	Acetonitrile with 0.1% formic acid
Note Use only LC/MS-grad	le solvents
Safety	According to IEC 61010

Table 5.Physical specifications

ltem	EASY-nLC II	EASY-nLC 1000
Width	35 cm (13.8 in.)	36 cm (14.2 in.)
Depth	38 cm (15.1 in.)	38 cm (15.1 in.)
Height	45 cm (17.7 in.)	45 cm (17.7 in.)
Weight	32 kg (70.5 lb)	35 kg (77 lb)
Weight in the shipping container	45 kg (99 lb)	45 kg (99 lb)

Table 6. Electrical specifications

Item	Specification
Power requirements	120 Vac, 50/60 Hz, 250 W 230 Vac, 50/60 Hz, 250 W
	For universal power supply (UPS) dimensioning, assume 250 W.
Fuses	For 120 Vac: one T 5 AL 250 V fuse (5 × 20 mm, IEC 60127) For 230 Vac: one T 2.5 AL 250 V fuse
	All fuses are UL Listed and CSA certified.

Table 7. Communication specifications

ltem	Specification
Contact closure	3 outputs, 3 inputs, and 6 ground pins
IN circuit	TTL Level
OUT circuit	PhotoMOS [™] relay protected against high voltages. Continuous switch current from 0.8 to 5 V is 35 mA.
LAN	10/100 Mb/s BaseT Ethernet
USB	$2 \times \text{USB 1.1}$ for keyboard and mouse
RS-232	Reserved for hardware add-ons or high-level MS control by using serial communication
P-Bus	Reserved for instrument add-ons, using the internal EASY-nLC instrument protocol bus: 8-wire control and limited power at 9/24 V
PS/2 (discontinued computer box only)	Input for connection of keyboard or mouse
Monitor	Output for connection of external display

Table 8.Hardware components

ltem	Specification
Pumps	 140 μL volume (enough for a >10 h, 0–100% B, 300 nL/min gradient) 1 nL/min to 300 μL/min flow range External pressure sensor
Rotary valves	 VICI[™]/Valco[™] rotor/stator 6 ports 3 positions (1–6, 1–2, or CENTERED)
Autosampler	 Peltier-cooled. Capacity is maximum 20 °C (68 °F) below ambient temperature, measured at 60% RH. Expect less if you remove the side panels, use an adapter plate in the autosampler other than the standard plate, or both. Plate holder ejects through spring-mounted autosampler door. 4 glass bottles with plastic lids for waste or wash liquids

Maintenance Schedule

To familiarize yourself with the maintenance schedule for the EASY-nLC instrument, review this chapter.

Contents

- Daily Maintenance
- Weekly Maintenance
- Quarterly Maintenance
- Yearly Maintenance
- Field Service Repairs

Daily Maintenance

Thermo Scientific

Check the solvent levels, draw fresh solvent through the solvent system, and flush air out of the solvent system on a daily basis.

To check the solvent levels

1. Visually inspect solvent bottle A, solvent bottle B, and the autosampler bottle in position W3 (and the bottles in position W1 and W2 if used). Refill if necessary.

Tip You can also inspect the purity of the solvent to ensure no visible precipitates have formed that might lead to blockages in the pump line.

- 2. Visually inspect the autosampler wash bottle in position W4 and the plastic waste beaker in front of the solvent system compartment, and empty if necessary.
- To draw fresh solvent into the system and to flush air out of the system

Run the **Purge Solvent** scripts with two purge iterations, and run the **Flush Air** script until flush volumes fall below 10 μ L for the EASY-nLC II instrument or 12 μ L for the EASY-nLC 1000 instrument.

Weekly Maintenance

Because the vapor pressures of formic acid, water, and acetonitrile differ, the solvent composition changes over time. Refill the solvent bottles to maintain a consistent solvent composition.

Quarterly Maintenance

Perform the following procedures quarterly (every 3 months).

* To check the back pressure

To test for system blockages, run the **Back Pressure** script for both solvents (see "Test – Back Pressure" on page 48).

IMPORTANT The script evaluates the back pressure based on using 99.9% water for solvent A and 99.9% acetonitrile for solvent B.

* To check the cooler temperature

Check that the actual plate temperature matches its set point (readout on the Home > Overview page).



Figure 23. Cooler area of Autosampler dialog box

* To check the autosampler pickup function

Run the **Sample Pickup** script to check the sample pickup function (see "Test – Sample Pickup" on page 42).

* To check the EASY-nLC solvent system for leaks

Run the **Leaks** script for "A+B" and for "System," to check for and diagnose instrument leaks. For more information, see "Test – Leaks" on page 44 and "Running a System Leak Test" on page 218).

IMPORTANT Check the valve rotor shifts regularly. When the valve rotor shifts exceed 6000, Thermo Fisher Scientific recommends checking the system for leaks. From this point forward, perform leak tests every 1000 shifts and replace the rotor seal when the leak test fails.

To check the check valves

Run the **Purge Solvent** script (see "Prepare – Purge Solvent" on page 32) and do the following:

- When the pumps are ejecting solvent, make sure that solvent is exiting the A and B waste tubing into the waste beaker.
- When the pumps are refilling, make sure that no solvents are going into the pump through the waste tubing.

If you detect a leaking check valve, replace it.

Yearly Maintenance

Replace the solvent filters and check the flow sensor calibration once a year.

* To exchange low-pressure solvent filters and high-pressure inline filters

Thoroughly clean new solvent filters before use as these can be a contamination source.

To check the flow sensor calibration

Run the **Flow Sensor** calibration script with the **Inspection Only** parameter enabled (see "Calibrate – Flow Sensors" on page 51).

This checks that the flow sensor calibration is working within specifications.

Field Service Repairs

If any of these instrument components is irreparably damaged, contact your local Thermo Fisher Scientific field service engineer for repairs:

- Autosampler or autosampler cooler
- Rotary valve
- Pump (for both instruments) or pump PCB (for the EASY-nLC II instrument)
- Integrated computer
- Touch-screen monitor



CAUTION Chapter 5, "Field Service Maintenance," describes how to replace these instrument components. However, only a Thermo Fisher Scientific field service engineer can perform these procedures. Attempted repairs by untrained personnel might cause personal injury or irreparably damage the instrument.

Maintenance Scripts and Service Records

To familiarize yourself with the maintenance scripts and record keeping features provided by the EASY-nLC software, review this chapter.

Contents

- Maintenance Scripts
- Keeping Service Records
- Checking and Resetting the Device Usage Counters

Maintenance Scripts

The EASY-nLC touch-screen software includes a variety of built-in maintenance scripts that help you prepare the instrument for use and troubleshoot instrument problems.

To select a maintenance script

- 1. On the touch screen, press Maintenance > Scripts.
- 2. Select a category and then the script for the specific operation you want.

You can also schedule some of the scripts for execution using the job queue.

Each of the following topics combines the category with the actual script name.

Prepare category	Test category	Calibrate category
• Prepare – Purge Solvent	• Test – MS Connection	• Calibrate – Valve Tune
• Prepare – Flush Air	• Test – Sample Pickup	Calibrate – Flow Sensors
• Prepare – Precolumn Equilibration	• Test – Leaks	• Calibrate – Reset Pressure Sensor
• Prepare – Analytical Column Equilibration	• Test – Valve Check	• Calibrate – Direct Infusion
• Prepare – Isocratic Flow	• Test – Back Pressure	
	• Test – Autosampler Torqu	le
	• Test – Pump Torque	

Prepare – Purge Solvent

The Purge Solvent script fills the selected pump or pumps with solvent and then ejects it into the waste beaker. Use the Purge Solvent script when exchanging solvents, removing air from the solvent lines, or filling the pumps.

Figure 24 shows the parameters for the Purge Solvent script.

Figure 24.	Purge Solvent script parameters
------------	---------------------------------

Thermo SCIENTIFIC	Home Batch Setu	p Method Editor	Maintenance	Configuration
Scripts	Category:	Name:	6	
Log Book	Prepare	Purge solvent	No gra	apn
Support	Description Para	meters Output		
Devices				
	Parameter	▼ Value		
	Purge iterations	10		
	Purge pump A	×		
	Purge pump B	×		
	Purge pump S	×		
Job 00:00:21			No gra	aph 🚽
11 May 2011 11:36 admin Exit	Schedule	RT STOP		

* To run the Purge Solvent script

- 1. Open the Purge Solvent script as follows:
 - a. On the touch screen, press Maintenance > Scripts.
 - b. In the Category list, select **Prepare**.
 - c. In the Name list, select Purge Solvent.
- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. In the Purge Iterations box, enter the number of empty/fill cycles to be performed:
 - To refresh solvents on a daily basis, enter 2 iterations.
 - To exchange solvents, enter a minimum of **10** iterations.

Note Entering a value of zero (0) for purge iterations refills the pump.

- c. Select the check boxes for the appropriate pump or pumps.
- 3. Press Start.

Prepare – Flush Air

The Flush Air script removes air from inside the pump head by pressurizing the pump and then releasing that pressure into the flow path toward the waste beaker.

The pump refills, pressurizes, releases pressure, and empties for each iteration of the script. While the instrument executes the script, the software calculates the pressurization time dynamically based on measurements from previous iterations and measures the pumped volume required to reach 200 bar.

Note Because the solvents are slightly compressible and the pump itself expands, even an air-free pump allows some pumping before reaching 200 bar.

Depending on the solvents, a pumped volume less than 10 μ L for the PLF pump model or 12 μ L for the PLU pump model is acceptable.

Note Figure 8 on page 9 shows the PLF and PLU model pumps. While the PLU model pump is standard in EASY-nLC 1000 instruments and the PLF model pump is standard in most EASY-nLC II instruments, the PLU model pump is the replacement part for both instrument models.

Figure 25 shows the parameters for the Flush Air script.

Thermo SCIENTIFIC	Home Batch Seti	up Method Editor	Maintenance	Configuration	
Scripts	Category:	Name:	No area	ab D	
Log Book	Prepare -	Flush air 🗾	Lilo gia		
Support	Description Par	meters Output			
Devices	Description				
	Parameter	▼ Value			
	Flush pump A	×			
	Flush pump B	×			
	Flush pump S	×			
	Flush threshold [µl]	12.00			
Job 00:00:21			No graj	oh 🔹	
11 May 2011					
11.51					
admin	Schedule	ART STOP			
Exit					

Figure 25. Flush Air script parameters

✤ To run the Flush Air script

- 1. Open the Flush Air script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select **Prepare**.
 - c. In the Name list, select Flush Air.
- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. In the Flush Threshold [µL] box, enter the acceptable threshold for the pump model:
 - For the PLF pump (standard in most EASY-nLC II instruments), enter 10.
 - For the PLU pump (standard in the EASY-nLC 1000 instrument), enter 12.
- 3. Press Start.

The script ends after the first iteration that gives a flush volume below the set threshold.

Prepare – Precolumn Equilibration

The Precolumn Equilibration script equilibrates the precolumn with solvent from pump A.

Use this script to equilibrate or flush the precolumn and to determine a suitable flow rate for the sample loading step in your method.

Figure 26 shows the parameters for the Precolumn Equilibration script.

Figure 26. Precolumn Equilibration script parameters

I • No graph •
il
put
0.00
0.00
DP

* To run the Precolumn Equilibration script

- 1. Open the Precolumn Equilibration script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Prepare.
 - c. In the Name list, select **Precolumn Equilibration**.
- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. Enter the appropriate values:
 - In the Volume $[\mu L]$ box, enter the volume of solvent A to use to equilibrate the precolumn.

For best results, use at least 10 column volumes to equilibrate the precolumn.

- In the Flow $\left[\mu L/min\right]$ box, enter the flow rate for the precolumn equilibration step.

If the flow field is left empty, the pump operates at the set pressure.

• In the Max Pressure [bar] box, enter the maximum allowed pressure.

If the pressure field is left empty, the pump operates at the set flow (as long as it is below the instrument maximum pressure of 300 bar for the EASY-nLC II instrument or 1000 bar for the EASY-nLC 1000 instrument). If both a flow and a max pressure are specified, the pump flow is limited by whichever parameter is reached first.



CAUTION Check the maximum pressure rating for the precolumn. Running the instrument at pressures higher than the column's maximum pressure rating reduces the column lifespan.

3. Press Start.

Prepare – Analytical Column Equilibration

The Analytical Column Equilibration script equilibrates the analytical column with solvent from pump A.

Use this script to equilibrate or flush the analytical column, determine a suitable equilibration flow rate for the method, or initialize a new analytical column.

Figure 27 shows the parameters for the Analytical Column Equilibration script.

SCIENTIFIC	Home Batch Setu	p Method Editor	Maintenance	Configuration
Scripts	Category:	Jame:		
Log Book	Prepare	Analytical col e 🝷		apn 🥑
Support	Description Para	meters Output		
Devices	Description			
	Parameter	▼ Value		
	Volume [µl]	4.00		
	Flow [µl/min]			
	Max pressure [bar]	800.00		
Job 00:05:02 Sample 00:05:00 Gradient 00:00 2 May 2011 14:06 admin Exit	Schedule STA	RT STOP	No gra	aph

Figure 27. Analytical Column Equilibration script parameters

Note The Analytical Column Equilibration script sets the valves to these positions:

- Valve A: 1–6
- Valve B: Center
- Valve S: 1–2
- Valve W: 1–2

* To run the Analytical Column Equilibration script

- 1. Open the Analytical Col Equilibration script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select **Prepare**.
 - c. In the Name list, select **Analytical Col Equilibration** (abbreviated to fit in the space allotted).
- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. In the Volume $[\mu L]$ box, enter the volume of solvent A to use to equilibrate the analytical column.

For best results, use at least 10 column volumes to equilibrate the analytical column.

c. In the Flow box [μ L/min], enter the flow rate for the analytical column equilibration step.

If the flow field is left empty, the pump operates at the set pressure.

d. In the Max Pressure [bar] box, enter the maximum allowed pressure.

If the pressure field is left empty, the pump operates at the set flow (as long as it is below the instrument maximum pressure of 300 bar for the EASY-nLC II instrument or 1000 bar for the EASY-nLC 1000 instrument).

If both a flow and a maximum pressure are specified, the pump flow is limited by whichever parameter is reached first.



CAUTION Check the maximum pressure rating for the analytical column. Running the instrument at pressures higher than the maximum pressure rating for the column reduces the column lifespan.

3. Press Start.

Prepare – Isocratic Flow

The Isocratic Flow script runs solvents A and B at a fixed mixture ratio. The script uses the AFC system to accurately control the flow. Use this script to tune the mass spectrometer at a given B percentage or when cleaning the instrument, the columns, or both. Figure 28 shows the parameters for the Isocratic Flow script.

Thermo SCIENTIFIC	Home Batch Setur	Method Editor	Maintenance	Configuration
Scripts	Category: N	lame:	No ar	aph
Log Book	Prepare	Isocratic flow		
Support	Description Paran	neters Output		
Devices				
	Parameter	▼ Value		
	Volume [µl]	100.00		
	Flow [µl/min]	0.30	_	
	AB Mi× [%B]	30	No gra	aph 🔹
job 00:15:48	Run indefinitely	×		
11 May 2011 12:41 admin Exit	Schedule	RT STOP		

Figure 28. Isocratic Flow script parameters

* To run the Isocratic Flow script

- 1. Select the Isocratic Flow script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select **Prepare**.
 - c. In the Name list, select Isocratic Flow.
- 2. Set up the parameters (see Figure 28) for this script as follows:
 - a. Press the **Parameters** tab.
 - b. Enter the appropriate values:
 - In the Volume box [µL], enter the total volume to be delivered by the pumps.

Note If you select the Run Indefinitely check box, the software ignores this volume setting.

- In the Flow box [μ L/min], enter the flow rate to be used.
- In the AB Mix box [%B], enter the solvent composition as a percentage of solvent B.
- If you want the solvent flow to continue until you press Stop, select the **Run Indefinitely** check box.

3. Press Start.

When either pump runs out of solvent, both pumps refill. The refilling process takes approximately 2 minutes.

Test – MS Connection

The MS Connection script tests the contact closure function between the EASY-nLC instrument and the mass spectrometer.

Figure 29 shows the MS connection script.

Figure 29. MS Connection script

Thermo SCIENTIFIC	Home	Batch Setup	Method Editor	Maintenance	Configuration
Scripts	Category:	Nan	ne:		anh D
Log Book	Test	-) (M	S connection		
Support	Descrip	tion Parame	ters Output		
Devices Job 00:15:48	Tests th spectrom gradient	e connection to eter using gradi -done events	the mass ent-ready and	No gra	aph
11 May 2011 12:42 admin Exit	Schedu		т		

* To run the MS Connection script and test the contact closure connection

1. Connect the contact closure cable to the EASY-nLC instrument and the mass spectrometer.

Figure 30 shows the contact closure cable for an EASY-nLC instrument to be used with a Thermo Scientific mass spectrometer.

Figure 30. Thermo Fisher contact closure cable



Tip Use the Thermo Fisher contact closure cable (LC160) to make contact closure between the EASY-nLC instrument and a Thermo Scientific mass spectrometer. Connect the two-pin connector to the mass spectrometer's Start In pins. Connect the 12-pin connector to the back panel of the EASY-nLC instrument.

- 2. Check the contact closure settings for the LC/MS system as follows:
 - a. On the EASY-nLC touch screen, press Configuration > Connections.
 - b. For a Thermo Scientific mass spectrometer, make the following selections:
 - In the Instrument (cable no.) list, select Thermo Scientific (LC160).
 - In the Protocol list under Contact Closure Settings, select **One-way**.
 - In the State at Start list under Contact Closure Settings, select **Open**.
- 3. Turn on the mass spectrometer and set it up to wait for a contact closure signal.
 - For a third-party mass spectrometer, follow the instructions in the documentation provided with the mass spectrometer. Then, go to step 5.
 - For an LTQ Series mass spectrometer, go to step 4.
- 4. For an LTQ Series mass spectrometer, set up the mass spectrometer to wait for a contact closure signal as follows:
 - a. Open the Tune Plus window.
 - b. On the menu bar, choose **Control > Acquire Data**.

Figure 31 shows the Control menu of the Tune Plus window for an LTQ Series mass spectrometer.

Figure 31. Control menu



The Acquire Data dialog box opens (see Figure 32).

c. In the Start Mode area, select the **Contact Closure** option.

Figure 32. Acquire Data dialog box with the Contact Closure option selected

Acquire Data			? 💌
Fol <u>d</u> er:	C:\Thermo\Instruments\LTQ\data	Acquire Time	
<u>F</u> ile Name:	File	Ontinuously	<u>S</u> tart
Sample <u>N</u> ame:		© Sc <u>a</u> ns 10 🚔	Pause
Comment:		O Minut <u>e</u> s 1.00	
_	Use industry to the d	<u>G</u> o to Standby when Finished	Vie <u>w</u>
Instrument <u>M</u> ethod:	Use instrument method	Acquisition Status State: Idle	Ins <u>t</u> . Setup
	Start Mode	Time (min): 0.000	
	[mmediate		
	OK Cancel <u>H</u> elp		

d. Click Start.

The mass spectrometer remains in the Waiting for Contact Closure state until it receives a contact closure signal.

- 5. Send the contact closure signal from the EASY-nLC instrument to the MS detector as follows:
 - a. On the EASY-nLC touch screen, press Maintenance > Scripts.

The Maintenance Scripts page opens.

- b. In the Category list, select Test.
- c. In the Name list, select **MS Connection**.
- d. Press Start.

The EASY-nLC instrument sends a contact closure signal to the mass spectrometer. The contact closure signal triggers the mass spectrometer to start scanning. The Scan LED on the front panel of a Thermo Scientific mass spectrometer flashes blue when the contact closure signal is set up correctly.

Test – Sample Pickup

The Sample Pickup script tests the accuracy of the autosampler pickup function. During the script, the XYZ robotic arm moves to the specified sample location. Then pump S aspirates the specified volume from the vial or microplate well and ejects this volume to waste. To determine if the autosampler is aspirating the specified sample volume, weigh the sample both before and after you run the test script as described in "Checking Sample Pickup" on page 263.

Use this script to verify the calibration of the XYZ robotic arm and to determine the appropriate flow rate (aspiration rate by pump S) setting in the method (or Xcalibur instrument method) that you plan to use for your samples. The appropriate flow rate depends on the sample viscosity.

Figure 33 shows the parameters for the Sample Pickup script and the Position dialog box that opens when you press the value box for the Position parameter.

Thermo SCIENTIFIC	Home Batch Setu	o Method Editor	Maintenance	Configuration	
Scripts Log Book	Category: Test	Name:		lo graph	•
Support	Description Parar	neters Output			
Devices	Parameter	✓ Value	Position		
	Volume [µl]	10.00	Select posit	ion	
	Flow [µl/min]	20.00		0:Plate/vials	
	Position	0-A1		A	
				1	
Job 00:00:00			Accept		Cancel
6 Sep 2011 11:26					
admin Exit	Schedule	ART			

Figure 33. Sample Pickup script parameters

* To run the Sample Pickup script

1. Load the autosampler with the appropriate samples.

Tip For more information about running this script and testing the accuracy of the sample pickup function, see "Checking Sample Pickup" on page 263.

- 2. Open the Sample Pickup script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Test.
 - c. In the Name list, select **Sample Pickup**.
- 3. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. In the Volume box $[\mu L]$, enter the volume to be picked up.
 - c. In the Flow box [μ L/min], enter the flow rate that pump S uses to aspirate the sample. For aqueous samples enter 20 μ L/min.
 - d. In the Position box, specify the position of the sample in the autosampler. See the Position dialog box in Figure 33.
- 4. Press Start.

Test – Leaks

The Leaks script tests for possible leaks at the pump seal and in the flow path from the pump to its associated valve (high-pressure subsystems A or B). The Leaks script can also test for system leaks in the flow path downstream of valves A and B.

Use this script when you suspect leak problems in the pumps (pump piston seals), in the flow path between the pump outlet and the associated valve, or downstream of valves A and B. For information about running the system leak test, see "Running a System Leak Test" on page 218.

Note You cannot perform the leak test for pump S, as it does not operate under pressure during normal batch execution.

Figure 34 shows the parameters for the Leaks script

Thermo SCIENTIFIC	Home Batch S	etup Method Edito	Maintenance	Configuration
Scripts	Category:	Name:		
Log Book	Test	- Leaks		
Support	Description	arameters Output		
Devices			1	
	Parameter	✓ Value		
	Test	System 🔻		
		A		
		В	No a	aph
		A+B		
Job 00:15:48		System		
11 May 2011 12:46				
admin Exit	Schedule	START STOP		

Figure 34. Leaks script parameters

During the leak test of pump A or B, the following events occur.

Step	Event
1	The pump refills.
2	The associated valve centers, blocking the solvent flow through the valve.
3	The pump builds pressure to 280 bar for the EASY-nLC II instrument or 980 bar for the EASY-nLC 1000 instrument.
4	When the pressure stabilizes at 280 bar for the EASY-nLC II instrument or 980 bar for the EASY-nLC 1000 instrument, the pump calculates the flow loss based on the pump piston movement required to maintain this pressure. If the flow loss is less than 400 nL/min, the pump passes the leak test.

During a system leak test (Leaks script with System selected), the script pressurizes the flow paths shown with red dashed lines in Figure 35, monitors the flow sensors and pump piston movement, and identifies the location of a leak. Before you run a system leak test, you must connect the Column Out line to the Waste In line with the HPLC union.

Figure 35. Pressurized lines for a system leak test



✤ To run the Leaks script

- 1. Open the Leaks script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select **Test**.
 - c. In the Name list, select Leaks.
- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. To select pump A or B, both pumps, or the entire system flow path for the leak test, press the cell in the Value column and make your selection from the list:
 - To test the solvent path from pump A to valve A (upstream of valve A), select A.
 - To test the solvent path from pump B to valve B (upstream of valve B), select **B**.
 - To test the solvent path from pump A to valve A and from pump B to valve B (upstream of pumps A and B), select **A** + **B**.
 - To test the solvent path downstream of valves A and B, select **System**. Figure 35 on page 45 shows the pressurized solvent path for the system leak test.
- 3. For a system leak test, connect the Column Out line to the Waste In line using the union supplied in the accessory kit as shown in Figure 35 on page 45.

Instrument	Part number of the stainless steel union		
EASY-nLC II	SC600		
EASY-nLC 1000	SC900		

For information about troubleshooting system leaks, see "Troubleshooting the Results of the System Leak Test" on page 221 and "Troubleshooting a Pump that Fails the Leaks Script" on page 216.

Test – Valve Check

The Valve Check script performs a self-test for the selected valves of an EASY-nLC II instrument.

Use this script when valves begin to move slowly or cannot reach the required positions and after exchanging valve parts (rotor or stator).

IMPORTANT This script is only valid for EASY-nLC II valves with a serial number less than V-009999. Make sure to record any numbers that the script generates. If the overshoot is greater than 6, contact your local Thermo Fisher Scientific field service engineer.

Figure 36 shows the parameters for the Valve Check script.

SCIENTIFIC	Home Batch Set	up Method Editor	Maintenance	Configuration		
Scripts	Category:	Name:		anh a		
Log Book	Test.	Valve check 🔻				
Support	Description Par	ameters Output				
Devices]				
	Parameter	✓ Value				
	Include valve A	×				
	Include valve B	×				
	Include valve S					
	Include valve W		No gr	aph		
Job 00:15:48						
11 May 2011 12:47						
admin Exit	Schedule	ART				

Figure 36. Valve Check script parameters

To run the Valve Check script

- 1. Select the Valve Check script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select **Test**.
 - c. In the Name list, select Valve Check.
- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. Select the check boxes for the valves that you want to test.
- 3. Press Start.

Test – Back Pressure

The Back Pressure script determines instrument back pressure for solvent A and B. The script runs at a preset flow and measures the back pressure on the system.

IMPORTANT Before you perform the Back Pressure script, ensure that the solvent A bottle contains water and the solvent B bottle contains acetonitrile. This test is not valid for other solvents.

Use this script to test for a blockage in the solvent system. Before running the script, connect the Column Out line to the Waste In line as shown in Figure 35 on page 45.

Figure 37 shows the parameters for the Back Pressure script.

Thermo SCIENTIFIC	Home Batch Setu	up Method Editor	Maintenance	Configuration
Scripts	Category:	Name:		
Log Book	Test Back pressure		No gr	apn
Support	Description Para	meters Output		
Devices	Description			
	Parameter	▼ Value		
	Test solvent A	×		
	Test solvent B	×		
			No gr	aph
Job 00:15:48				
11 May 2011 12:50				
admin	Schedule ST	ART		

Figure 37. Back Pressure script parameters

✤ To run the Back Pressure script

- 1. Open the Back Pressure script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Test.
 - c. In the Name list, select Back Pressure.
- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. Test solvent A, solvent B, or both A and B by selecting the corresponding check box or check boxes.
- 3. Press Start.
Test – Autosampler Torque

The Autosampler Torque script measures the torque required to move the XYZ robot on each of its axes for the ASC autosampler.

IMPORTANT Do not run this script for the ASA autosampler. Contact your local Thermo Fisher Scientific representative before running this script.

Figure 38 shows the Description page of the Autosampler Torque script.

Figure 38. Autosampler Torque script description

SCIENTIFIC	Home Batch Setup	Method Editor	Maintenance	Configuration	
Scripts Log Book	Category: N. Test	ame: Autosampler torque 🝷		lo graph	•
Devices	Description Parama Tests the individual to the autosampler maniput	eters Output			
Job 00:01:02				No graph	•
24 aug 2010 17:50 admin Exit		RT STOP			

Test – Pump Torque

For the PLU pump in the EASY-nLC 1000 instrument, the Pump Torque script measures the actuator's ability to generate pressure at predefined torque levels.

IMPORTANT Do not use this script for the PLF pump in the EASY-nLC II instrument. Contact your local Thermo Fisher Scientific representative before running this script.

Figure 39 shows the parameters for the Pump Torque script.

Thermo SCIENTIFIC	Home Batch Setu	p Method Editor	Maintenance	Configuration
Scripts	Category:	Name:		
Log Book	Test 🔹	Pump torque	l log	apri
Support	Description Para	meters Output		
Devices				
	Parameter	✓ Value		
	Test pump A	×		
	Test pump B	×		
Job 00:15:48		PT STOP	Nogr	aph
Exit	Schedule			

Figure 39. Pump Torque script parameters

Calibrate – Valve Tune

The Valve Tune script automatically retunes the selected valve in the EASY-nLC II instrument.

IMPORTANT This script is only valid for an EASY-nLC II valve with a serial number below V–009999. Contact your local Thermo Fisher Scientific representative before running this script, as running this script incorrectly will compromise the performance of the instrument.

Calibrate – Flow Sensors

The Flow Sensors script calibrates the flow sensors for pumps A and B.

Use this script to do the following:

- Periodically check the accuracy of the flow sensor calibration for your maintenance records (select the Inspection Only check box).
- Calibrate the flow sensors when you change the type of solvent in solvent bottles A or B (clear the Inspection Only check box).

This calibration does not require any additional plumbing.

IMPORTANT Because the flow sensor calibration requires a stable operating temperature, ensure that the instrument's side panels are installed and that the instrument has been on for a minimum of one-half hour.

Figure 40 shows the parameters for the Flow Sensors script.

Figure 40. Flow Sensors script parameters

Thermo SCIENTIFIC	Home Batch Seti	up Method Editor	Maintenance	Configuration
Scripts	Category:	Name:		
Log Book	Calibrate 🔹	Flow sensors	No gr	apn
Support	Description Par	meters Output		
Devices		anicers output	ie.	
	Parameter	✓ Value		
	Include sensor A	×		
	Include sensor B	×		
	Inspection only			
	Exact inspection	۲		
	Fast inspection	0	No gr	aph 🚽
Job 00:15:48		· · · · · · · · · · · · · · · · · · ·		
11 May 2011 12:54				
admin Exit	Schedule	ART		

* To run the Flow Sensors script

- 1. Open the Flow Sensors script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select **Calibrate**.
 - c. In the Name list, select **Flow Sensors**.

- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. Select the check boxes for the flow sensors (A, B, or both) that you want to either inspect or calibrate and inspect.

Note To calibrate and inspect the flow sensors, this script performs a two-point calibration as follows:

- 1. Calibrates the 0 μ L/min point.
- 2. Inspects and evaluates this first calibration point.
- 3. Calibrates the 500 μ L/min point.
- 4. Inspects and evaluates this second calibration point.

When you select the Inspection Only check box, the script does not apply the new calibration. Selecting the Fast Inspection option instead of the Exact Inspection option reduces the duration of the inspection and evaluation steps from 16 minutes to 5 minutes, without reducing the accuracy of the calibration.

- c. Do one of the following:
 - To check the accuracy of the current calibration (only inspect the current calibration), select the **Inspection Only** check box.

Note When you select the Inspection Only check box, the program does not recalibrate the flow sensors.

- To calibrate and inspect the flow sensors (and apply the new calibration), clear the **Inspection Only** check box.
- d. Select the inspection duration as follows:
 - When you want an estimate of the flow sensor accuracy, select the **Inspection Only** check box and the **Fast Inspection** option.
 - When you are recalibrating the flow sensors, clear the **Inspection Only** check box and select the **Exact Inspection** option.
- 3. Press Start.

Calibrate – Reset Pressure Sensor

This script auto-zeroes the pressure sensor.

IMPORTANT Performing this script incorrectly will compromise the performance of the instrument. Contact your local Thermo Fisher Scientific representative before running this script.

Calibrate – Direct Infusion

Use the Direct Infusion script to analyze samples by direct infusion or to optimize the spray stability by infusing a known peptide.

In a direct infusion experiment, sample is introduced into the mass spectrometer without chromatographic separation by an analytical column. The EASY-nLC instrument picks up the sample volume from the specified location, and then pumps it directly into the ion source using the solvent flow from pump A.

Figure 41 shows the parameters for the Direct Infusion script.

Thermo SCIENTIFIC	Home Batch Setup	Method Editor	Maintenance Configuration
Scripts	Category: Na	ame:	No graph
Log Book	Calibrate	Direct infusion	Tio graph
Support	Description Param	eters Output	
Devices	Description		
	Parameter	✓ Value	
	Pickup volume [µl]	10.00	
	Pickup flow [µl/min]	20.00	
	Pickup position	0-A1	
	Infusion volume [µl]	10.00	
	Infusion flow [µl/min]	20.00	No graph
-	Wash	×	
Job 00:04:20		22	
17 Feb 2012 12:43			
admin Exit		STOP	

Figure 41. Direct Infusion script

***** To set up the system for a direct infusion experiment

1. Connect an emitter to the Column Out tubing.

Figure 42 shows an example of a direct infusion setup for an EASY-nLC 1000 instrument.

Figure 42. Setup for a direct infusion experiment



2. Mount the assembly onto your nanoflow ion source.

To run the Direct Infusion script

- 1. Select the Direct Infusion script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Calibrate.
 - c. In the Name list, select **Direct Infusion**.
- 2. Set up the parameters for this script as follows:
 - a. Press the **Parameters** tab.
 - b. In the Pickup Volume $[\mu L]$ box, enter the volume that you want the autosampler needle to pick up. The autosampler loads this volume into the sample loop.
 - c. In the Pickup Flow $[\mu L/min]$ box, enter the flow rate at which pump S aspirates the sample from the vial or well.
 - d. In the Pickup Position box, enter the vial or well position.
 - e. In Infusion Volume box $[\mu L]$, enter the volume that pump A delivers to the system to backflush the sample from the sample loop out through the Column Out line and into the mass spectrometer's ion source.
 - f. In the Infusion Flow $[\mu L/min]$ box, enter the flow rate at which pump A moves the sample into the mass spectrometer's ion source.
 - g. Select the Wash check box to add a Wash step after pump A has pumped the specified volume of solvent A through the system.

During the Wash step, pump S empties 100 μL of solvent A through the loop and needle to the waste bottle.

3. Press Start.

IMPORTANT The Direct Infusion script flushes the solvent lines before it ends. Stopping the script before it ends can leave sample solution in the flow path.

4. If you press Stop before the script ends, follow the instructions in message box that appears to make sure that the current sample is flushed from the system (see Figure 43).

Figure 43. Wash instructions that appear when you stop the Direct Infusion script

Attention	: Wash is enabled, but the script
was abort	ed!
Make sure	you wash loop & needle before
running f	urther infusions,
e.g. by s	pecifying zero for the pickup and
infusion	volumes.

Keeping Service Records

Use the Maintenance > Log Book page to electronically enter all the service actions that you perform on the instrument.

To record a service action

1. Press Maintenance > Log Book.

The Maintenance > Log Book page opens (see Figure 44).

Figure 44.	Log Book page	of the Maintenance	menu tab
------------	---------------	--------------------	----------

SCIENTIFIC	Home	Batch Set	up Method	Editor	Maintenance	Configu	ration		
Scripts Log Book Support Devices	Log recor	d contains (one	e term per line)		Log book Queue jol User Any	•	Time Fro F Time To:	m: irst record ast record Search)
	Tim	lestamp	Source		Su	Immary			
	05/08-201	10 17:52:49	admin	Batch: ad	min-20100805-1751				
	05/08-201	10 17:44:34	admin	Batch: ad	min-20100805-1742			1	
	19/07-201	10 10:41:58	admin	Batch: ad	min-20100719-1039				
Job 00:00:00	12/05-201	10 17:03:40	admin	Purge sol	vent (00:00:00)			~	
	29/04-201	10 10:29:34	admin	Purge sol	vent (00:00:00)				
23 aug 2010 12:37	29/04-201	10 10:28:42	Super	Purge sol	vent (00:00:00)				
admin	28/04-201	10 13:43:47	admin	Isocratic f	low (00:00:00)				▼
Exit	Enter	Log Entry						Details	

2. Press Enter Log Entry.

The Log Entry dialog box opens.

- 3. In the box at the top of the dialog box, enter a description of the service action.
- 4. In the Templates list, select a template.

The Templates list contains the following selections: Comment, New Part, Maintenance, Property Change, and Precolumn Changed by <User> on <Date>.

5. In the Components list, select a hardware component.

The Components list contains the following selections: None, Gradient, Valve B, Valve A, Valve S, Valve W, Analytical Column, Precolumn, Autosampler, Pump A, Pump B, and Pump S.

- 6. Do one of the following:
 - Press Add to add this entry and keep the Log Entry dialog box open.
 - Press Add and Close to add this entry and close the Log Entry dialog box.
 - Press Close to close the Log Entry dialog box without saving any entries.

To search the log

- 1. In the Log Book list, select **Queue Job** or **User**.
- 2. In the User List, select Any, Admin, or Guest.
- 3. In the Time From list, select a time.
- 4. In the Time To list, select a time.
- 5. Press Search.

The search results appear in the Search Result table.

Checking and Resetting the Device Usage Counters

The EASY-nLC system tracks the volume pumped by pumps A, B, and S and the number of valve and rotor shifts for valves A, B, and S.

To view the device usage values

- 1. Press Maintenance > Devices.
- 2. Do one the following:
 - To view the volume pumped by a pump, select the pump from the list of devices.
 - To view the number of valve and rotor shifts for a valve, select the valve from the list of devices.
- 3. Press the **Summary** tab.

The Summary view opens.

* To reset the volume pumped or reset the rotor shifts to zero

- 1. Open the appropriate Summary page.
- 2. Press Reset.

For a pump, the value in the Intermediate Volume box resets to 0. For a valve, the value in the Rotor Shifts box resets to 0.

Routine Maintenance

To maintain the EASY-nLC instrument, follow the maintenance procedures in this chapter.

Tip You can access most of the instrument components by removing either the left or right side panel of the instrument. Three captive quarter-turn screws secure each panel to the instrument housing.

- To remove a side panel from the housing, loosen (unlock) the screws from the housing by rotating them 90° counterclockwise with a #2 Phillips head screwdriver. When loosened, the screws rotate freely without constraint, but remain connected to the side panel.
- To secure a side panel to the housing, mount the panel. Then tighten the screws to the housing (lock) by rotating them 90° clockwise with a #2 Phillips head screwdriver.

For information about the suggested maintenance schedule and the built-in maintenance scripts and record keeping features provided by the EASY-nLC software, see Chapter 3, "Maintenance Scripts and Service Records."

Contents

- Maintaining a Clean Working Environment
- Replacing the Main Power Fuse
- Maintaining the Syringe Pumps
- Maintaining the Rotary Valves
- Replacing the Check Valves
- Replacing an Inline Filter for the EASY-nLC 1000 Instrument
- Using nanoViper Fittings
- Replacing the Autosampler Needle
- Replacing the Sample Loop
- Replacing a Pressure Sensor for the PLU Pump
- Replacing a Flow Sensor
- Replacing the Hard Drive
- Managing the Devices List

Maintaining a Clean Working Environment

Maintain a clean working environment for the EASY-nLC instrument.

When cleaning the outside of the EASY-nLC instrument, use a mild detergent and a clean cloth.

Replacing the Main Power Fuse

The EASY-nLC uses these fuses:

- For 120 Vac, the instrument uses one T 5 AL 250 V fuse (5 \times 20 mm, IEC 60127).
- For 230 Vac, the instrument uses one T 2.5 AL 250 V fuse.

Use only UL Listed and CSA-certified fuses, All fuses supplied with the instrument are UL Listed and CSA certified.



CAUTION Before removing the fuses, turn off the instrument and remove the power cable.

To replace the main power fuses

1. Move the EASY-nLC instrument to a benchtop where you can access the back panel.

Figure 45 shows the steps required to remove the fuse holder.

- 2. Turn off the power and pull out the power plug.
- 3. Place a flat-blade screwdriver into the slot in the top of the fuse holder, and then turn the screwdriver to loosen the fuse holder.
- 4. Pull the fuse holder out of the power entry module.

Figure 45. Replacing the main power fuse









Using a flat-blade screwdriver, loosen the fuse holder.

- 5. To install the appropriate fuse, do one of the following:
 - Insert the 5 A fuse into the slot that aligns with the white triangle at the end of the 110–120 V line power text.
 - Insert the 2.5 A fuse into the slot that aligns with the white triangle at the end of the 220-240 V text.
- 6. Make sure that the fuse ends align with the mounting brackets.

Note The fuse holder holds two fuses. One fuse is for a 220–240 V line power and the other is for a 110–120 V line power.

7. Insert the fuse holder into the power entry module in the orientation that corresponds to the operating power.

The triangle to the right of the voltage rating points toward the white mark at the bottom of the power entry module (see Figure 46).

Figure 46. Power entry module



Appropriate orientation for 220–240 line voltage

Maintaining the Syringe Pumps

Each pump contains a piston seal and a piston. Over time, the buffered solutions leave deposits on the pistons, and through constant contact with the moving pistons, the piston seals slowly deteriorate.

Note Most EASY-nLC II instruments contain PLF model pumps, which have been discontinued. If this pump is damaged, you must replace the PLF model pump with a PLU model pump.

Both the EASY-nLC 1000 instrument and new shipments of the EASY-nLC II instrument use PLU model pumps.

Figure 8 on page 9 shows the two pump models.

To replace a pump piston seal and clean the piston, follow these procedures:

- 1. "Retracting the Piston" on page 61
- 2. Depending on the pump model, follow one of these procedures:
 - "Replacing the Piston Seal and Cleaning the Piston in a PLF Pump" on page 62
 - "Replacing the Piston Seal in a PLU Pump" on page 67
- 3. "Priming the Pump" on page 71
- 4. "Resetting the Pump Usage Counter" on page 72
- 5. "Removing Air After Replacing a Piston Seal or a Pump" on page 73
- 6. Depending on whether you are performing maintenance on one of the chromatography pumps (pump A or B) or on the sample pump (pump S), follow one of these procedures:
 - For pumps A and B, run a pump leak test as described in "Running the Leaks Script after Replacing a Piston Seal or a Pump" on page 73.
 - For pump S, run a sample pickup test as described in "Test Sample Pickup" on page 42.

Retracting the Piston

Set the pump piston to the fully retracted position by running the Purge Solvent script with 0 iterations.

✤ To retract the pump piston

- 1. On the touch screen, press Maintenance > Scripts.
- 2. In the Category list, select Prepare.
- 3. In the Name list, select **Purge Solvent**.
- 4. Press the **Parameters** tab.
- 5. In the Purge Iterations box, enter **0** (see Figure 47).

Figure 47. Purge solvent script with 0 iterations

SCIENTIFIC	Home	Batch Setup	Method Editor	Maintenance	Configuration	
Scripts	Category:		Name:		Na wash	
Log Book	Prepare	-)	Purge solvent		Nu graph	
Support	Deccri	ntion Param	atare Output			
Devices	Descrip		eters Output			
	Pa	rameter	▼ Value			
	Purge ite	rations	C			
	Purge pu	mp A	×			
	Purge pu	mp B				
	Purge pu	mp S				
					No graph	-)
Job 00:00:00 2 May 2011 18:26 admin Exit	Sched		RT STOP			

- 6. Select the check box for the appropriate pump.
- 7. Press Start.

The piston moves backward until it reaches the 140 μ L position.

- 8. To replace the piston seal, go to the piston seal replacement procedure for your pump.
 - "Replacing the Piston Seal and Cleaning the Piston in a PLF Pump" on page 62

-or-

• "Replacing the Piston Seal in a PLU Pump" on page 67

Replacing the Piston Seal and Cleaning the Piston in a PLF Pump

Most EASY-nLC II instruments have PLF model pumps (see Figure 8 on page 9).

Note To replace the piston seal in a PLU pump, follow the instructions in "Replacing the Piston Seal in a PLU Pump" on page 67.

Replacing a piston seal in the PLF pump requires these tools and materials.

 #2 Phillips head screwdriver 13 mm open-ended wrench 1/4 in. open-ended wrench Pipette Piston seal and piston seal tool, P/N LC210 	Tools	Parts and materials
 13 mm open-ended wrench 1/4 in. open-ended wrench Pipette Piston seal and piston seal tool, P/N LC210 	• #2 Phillips head screwdriver	Powder-free safety gloves
 1/4 in. open-ended wrench Pipette Piston seal and piston seal tool, P/N LC210 	• 13 mm open-ended wrench	LC/MS-grade methanol
Pipette Piston seal and piston seal tool, P/N LC210	• 1/4 in. open-ended wrench	LC/MS-grade acetonitrile
	• Pipette	• Piston seal and piston seal tool, P/N LC210



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

* To replace the piston seal in a PLF model pump

- 1. If you have not already done so, set the piston to its fully retracted position as described in "Retracting the Piston" on page 61.
- 2. Set the corresponding valve to the **Center** position as described in "Using the Valve Controls" on page 204.
- 3. Close down the EASY-nLC instrument and turn off the power (see "Closing Down the EASY-nLC Instrument" on page 21).
- 4. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.

- 5. Remove the tubing connected to the pump head as follows:
 - If a PEEK fitting is connected to the pump head, use a 13 mm open-ended wrench, to remove it (see Figure 48).
 - **Figure 48.** Removing a PEEK fitting from the externally threaded pump head for a PLF model pump



PEEK fitting connected to an externally threaded pump head

• If a stainless steel fitting is connected to the pump head, use a 1/4 in. open-ended wrench to remove it.

Figure 49 shows a stainless steel fitting connected to an internally threaded pump head.

Figure 49. Internally threaded pump head for a PLF model pump



Stainless steel fitting connected to an internally threaded pump head 6. Using a 13 mm open-ended wrench, remove the pump head (see Figure 50).

Figure 50. Removing the pump head



Pump head (PLF model pump)

- 7. Clean the piston as follows:
 - a. Place the piston in the fully extended position as follows:
 - i. Turn on the instrument and log in as an administrator.
 - ii. Press Home > Overview.
 - iii. Press the pump icon for the pump you want to control. The Pump dialog box opens.
 - iv. In the flow rate box, enter the flow rate: $300\ \mu\text{L/min}.$
 - v. In the volume box, enter the dispense volume: 140μ L.
 - vi. Press Start.
 - b. Soak a lint-free tissue in methanol, and then squeeze out the excess solvent.
 - c. Clean the piston with the lint-free tissue soaked in methanol, and visually inspect the piston for any scratches. Take care to avoid solvent drips, as the pump printed circuit board can be damaged by exposure to solvents.



CAUTION The pump PCB is easily damaged by exposure to solvents.

- d. Return the piston to the fully retracted position as follows:
 - i. In the Pump dialog box, set the flow rate to $300~\mu L/min$ and the dispense volume to $140~\mu L.$
 - ii. Press Start.
- 8. When the piston is fully retracted, close down the EASY-nLC system and turn off the power (see "Closing Down the EASY-nLC Instrument" on page 21).

- 9. Remove the worn piston seal as follows:
 - a. To avoid contaminating the pump head, clean the piston seal tool and guide tool by soaking them in a beaker filled with 100% HPLC-grade methanol.

Figure 51 shows the piston seal tool.





b. Insert the piston seal tool into the worn piston seal and pull the piston seal out of the pump head (Figure 52).





- 10. Install the new piston seal as follows:
 - a. Insert the guide tube into the pump head flange (see Figure 53).

Figure 53. Guide tool inserted into the pump head flange (PLF model pump)



Guide tool

b. Using a pipette, fill the groove in the piston seal with methanol.

Tip Filling the piston seal groove with methanol before installing the seal reduces the time required for the Flush Air script to remove air from the pump.

c. Place the new seal with the groove facing upward on the piston seal tool (see Figure 54).

Figure 54. Piston seal mounted onto the piston seal tool (PLF model pump)



d. Insert the piston seal tool into the guide tool (see Figure 55).

Figure 55. Inserting the piston seal tool into the guide (PLF model pump)



e. Push the piston seal tool into the guide until you feel resistance (see Figure 56).Figure 56. Seating the piston seal (PLF model pump)



f. Remove the guide tube and the piston seal tool. and then check that the piston seal is properly seated (see Figure 57).

Figure 57. Piston seal properly seated in the pump head (PLF model pump)



- 11. Using a 13 mm open-ended wrench, reconnect the pump head to the pump body.
- 12. Go to "Priming the Pump" on page 71.

Replacing the Piston Seal in a PLU Pump

Replacing the piston seal in a PLU model pump requires these tools and materials.

Tools	Parts and materials
• #2 Phillips head screwdriver	Powder-free safety gloves
• 3 mm L-hex wrench	LC/MS-grade methanol
• 1/4 in. open-ended wrench	LC/MS-grade acetonitrile
• Pipette	• Piston seal and piston seal tool, P/N LC510

To replace the piston seal in a PLU model pump

- 1. If you have not already done so, set the piston to its fully retracted position as described in "Retracting the Piston" on page 61.
- 2. Set the corresponding valve to the **Center** position as described in "Using the Valve Controls" on page 204.
- 3. Close down the EASY-nLC instrument and turn off the power (see "Closing Down the EASY-nLC Instrument" on page 21).
- 4. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.

- 5. Remove the used piston seal as follows:
 - a. Using a 1/4 in. open-ended wrench, remove the stainless steel fitting connected to the pump head (see Figure 58).

Figure 58. Stainless steel fitting connected to the pump head (PLU model pump)



b. Using a 3 mm L-hex wrench, remove the two screws that secure the pump head to the pump body (see Figure 59).

IMPORTANT Do not move the piston if the pump head has been removed. Doing so can affect the piston alignment, which might cause the piston calibration to be lost and result in irreversible piston damage.

Figure 59. Using a 3 mm L-hex wrench to remove the pump head

Two screws that secure



c. Clean the exposed portion of the piston with a lint-free tissue soaked in HPLC-grade methanol and visually inspect the piston for scratches. Ensure that no solvent runs into the pump.



d. Using only your fingernails or a plastic tool, remove the old seal from the pump head (see Figure 60).

CAUTION Avoid using a metal tool to remove the piston seal from the pump head, as metal can scratch the inside of the pump head and generate leaks.

Figure 60. Worn piston seal installed in the pump head (PLU model pump)



- 6. Install the new piston seal as follows:
 - a. With the spring side of the piston seal facing upward, mount the new piston seal onto the piston seal tool (see Figure 61).

Figure 61. Piston seal mounted on the piston seal tool (PLU model pump)



Piston seal spring

b. Using a pipette, fill the rim of the piston seal with 100% LC/MS-grade methanol to remove the air from the piston seal spring (see Figure 62).

Figure 62. Filling the piston seal with methanol (PLU model pump)



c. Place the pump head on the guide rods and evenly press it all the way down against the piston seal tool to insert the seal into the pump head (see Figure 63).

Figure 63. Inserting the piston seal into the pump head (PLU model pump)



d. Gently pull the pump head off of the guide rods (see Figure 64).

Figure 64. Removing the pump head from the piston seal tool (PLU model pump)



- 7. Using the two screws and a 3 mm L-hex wrench, reconnect the pump head to the pump body. Do not connect the stainless steel tubing to the pump head.
- 8. Go to the next procedure, "Priming the Pump."

Priming the Pump

After you replace a piston seal, prime the pump with the appropriate solvent before you reconnect the solvent line to the pump head.

To prime the pump

- 1. Turn on the EASY-nLC instrument and log in as an administrator.
- 2. Set the piston to its fully extended position as follows:
 - a. On the touch screen, press **Home > Overview**.
 - b. Press the pump icon for the pump that you want to control.

The Pump *X* dialog box opens, where *X* identifies the pump (A, B, or S).

- c. Set the flow rate to $300\,\mu\text{L/min}$ and the volume to $140\,\mu\text{L}.$
- d. Press Start.
- 3. Fill the pump head with solvent as follows:
 - a. Pipette an aliquot of the appropriate solvent into the pump head (see Figure 65).Figure 65. Adding solvent to the pump head



CAUTION Be careful not to spill solvent on the PCBs.



- For the PLU pump model (standard in the EASY-nLC 1000 instrument and replacement part for both EASY-nLC instruments), the LED panel cover for each pump protects the PCB from accidental contact when the instrument's right panel is removed. However, the cover does not form a waterproof seal with the pump body so that the PCBs are exposed to solvent leaks from above.
- For the PLF pump model (EASY-nLC II instrument), the printed circuit boards are completely exposed.

b. In the Pump dialog box, set the flow rate to $-300~\mu\text{L/min}$ and the volume to $140~\mu\text{L}.$ Then press Start.

The piston moves down, drawing the solvent into the pump.

- c. Make sure the pump head is filled with solvent during the entire retraction of the piston.
- 4. Reconnect the solvent line to the pump head as follows:
 - For a PLF pump with a PEEK fitting (see Figure 48 on page 63), use a 13 mm open-ended wrench to tighten the fitting.
 - For a PLF pump with a stainless steel fitting (see Figure 49 on page 63) or a PLU pump, use a 1/4 in. open-ended wrench to tighten the fitting.
- 5. Mount the right side panel to the instrument housing. Then, with a #2 Phillips head screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.

After you reconnect the pump head to the pump body and the high-pressure line from the pressure sensor to the pump head, reset the pump usage counter, and then remove air from the system.

Note You reset the usage counters before removing air from the system because the air removal procedure requires the pump to go through 10 purge cycles.

Resetting the Pump Usage Counter

Reset the pump usage counter after you prime the pump.

- To reset the pump usage counter
- 1. On the touch screen, press Maintenance > Devices.
- 2. Select the pump from the list of devices.
- 3. Press the **Summary** tab.
- 4. Press Reset.

The value in the Intermediate Volume box resets to 0.

For more information, see "Checking and Resetting the Device Usage Counters" on page 56.

Removing Air After Replacing a Piston Seal or a Pump

To draw fresh solvent through the solvent system and to remove air from the solvent system components, including the pump heads, follow these procedures after you prime the pump and reset the usage counters.

* To draw fresh solvent into the system and remove air from the system

- 1. To draw fresh solvent into the system and remove air from the solvent lines, do the following:
 - a. On the touch screen, press Maintenance > Scripts.
 - b. In the Category list, select **Prepare**.
 - c. In the Name list, select **Purge Solvent** (see "Prepare Purge Solvent" on page 32).
 - d. Press the **Parameters** tab.
 - e. In the Purge Iterations box, enter **10**.
 - f. Select the check box for the appropriate pump.
 - g. Press Start and wait for the system to perform 10 purge cycles.
- 2. To remove air from the pump head, do the following:
 - a. In the Name list, select Flush Air (see "Prepare Flush Air" on page 33).
 - b. Press the **Parameters** tab.
 - c. Set the flush volume threshold to $10~\mu L$ for the PLF pump or $12~\mu L$ for the PLU pump.

Note Figure 8 on page 9 shows the PLU and PLF pump models.

d. Press **Start**, and wait for the script to finish.

Running the Leaks Script after Replacing a Piston Seal or a Pump

After you prime pump A or pump B, reset the usage counters, remove air from the system, and run a pump leak test.

Note For pump S, run a sample pickup test as described in "Test – Sample Pickup" on page 42.

For information about replacing a pump, see "Replacing a Pump" on page 150. Only a Thermo Fisher Scientific field service engineer can replace a damaged pump, as replacing the pump is an advanced maintenance procedure.

* To run a pump leak test on a replacement pump or a pump with a new piston seal

Follow the instructions in "Test – Leaks" on page 44. When the instrument is leak tight, the instrument is ready for use.

Maintaining the Rotary Valves

The EASY-nLC instrument has four rotary valves labeled Valve A, Valve B, Valve S, and Valve W. These valves contain the same internal components.

To determine if the valve requires maintenance, run the Leak test script as follows:

- For valves S and W, run the Leaks script on the system (System).
- For valves A and B, run the Leaks script on the respective subsystem (A or B).

To maintain the valves, follow these procedures:

- "Cleaning the Rotor Seal and Stator," on this page
- "Replacing the Rotor Seal" on page 77
- "Replacing the Stator" on page 78

Maintaining the valves by cleaning the rotor seal and stator or by replacing the rotor seal, the stator, or both parts requires these tools and materials.

Tools	Materials
• 9/64 in. hex wrench	Powder-free safety gloves
• 1/4 in. open-ended wrench	LC/MS-grade methanol
• #2 Phillips head screwdriver	Lint-free cloth



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

Cleaning the Rotor Seal and Stator

If a rotary valve is leaking, attempt to remove the leak by cleaning the rotor seal and the stator before taking the more costly step of replacing the rotor seal.

To clean the rotor seal and stator in a valve and return the instrument to normal operation, follow these steps:

- 1. "To remove the stator from the valve" on page 75
- 2. "To clean the rotor seal and the stator" on page 75
- 3. "To reassemble the cleaned valve and return the instrument to normal operation" on page 76

To remove the stator from the valve

- 1. Close down the EASY-nLC instrument, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 2. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 3. Using a 9/64 in. L-hex wrench, remove the two hex screws that secure the stator to the valve assembly, and then pull the stator away from the valve driver.
- 4. Depending on whether you are cleaning or replacing the rotor seal, go to the appropriate procedure:
 - "To clean the rotor seal and the stator," on this page
 - "To replace the rotor seal" on page 77

Note If you are cleaning the valve components or replacing only the rotor seal, you do not need to remove the tubing connected to the stator. If you are replacing the stator, you must remove all the solvent lines from the valve.

* To clean the rotor seal and the stator

- 1. If you have not already done so, remove the stator from the valve as described in the previous procedure, "To remove the stator from the valve."
- 2. Leave the rotor seal mounted to the valve driver (see Figure 66). Clean the mounted rotor seal with a lint-free tissue or Q-tip soaked in methanol.

Figure 66. Rotor seal mounted on the valve driver



3. Clean the stator with a lint-free tissue or Q-tip soaked in methanol (see Figure 67).

Figure 67. Cleaning the stator



* To reassemble the cleaned valve and return the instrument to normal operation

- 1. Mount the clean stator onto the valve driver.
- 2. Using a 9/64 in. L-hex wrench, tighten the two hex screws a little at a time by shifting from one screw to the other and back again until the screws are evenly torqued.
- 3. Mount the right side panel to the instrument housing. Then, with a #2 Phillips head screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.
- 4. Turn on the EASY-nLC instrument and log in as an administrator or a super user.
- 5. Go to the next procedure, "To remove air from the system and check for leaks after disassembling a valve."

* To remove air from the system and check for leaks after disassembling a valve

- 1. If you removed the stator from valves A or B, flush air from the system as follows:
 - a. Set up the Flush Air script as described in "Prepare Flush Air" on page 33.
 - b. Select the pump that is connected to the affected valve.
 - c. Set the flush volume threshold to $10~\mu L$ for the EASY-nLC II or $12~\mu L$ for the EASY-nLC 1000.
 - d. Press Start.
 - e. Wait for the script to finish.
- 2. Check the system or subsystem for leaks as follows:
 - If you removed the stator from in valve A or B, run the Leaks script for the affected valve (see "Test Leaks" on page 44).
 - If you replaced the rotor seal in valve S or W, run the Leaks script for the system.

When the system is leak tight, the instrument is ready for use.

Replacing the Rotor Seal

If cleaning the rotor seal and stator does not remove the system leak, replace the rotor seal. Replacing the rotor seal requires one of these parts.

Valve serial number	Replacement part
V-010000 and above	Rotor seal, P/N LC228
V-009999	Rotor seal, P/N LC224

Tip The About page of the Rotary Valve view on the Maintenance > Devices page lists the serial number of the selected valve.

To replace the rotor seal in a valve and return the instrument to normal operation, follow these steps:

- 1. "To remove the stator from the valve" on page 75
- 2. "To replace the rotor seal," on this page
- 3. "To reset the usage counter for the valve" on page 78
- 4. "To remove air from the system and check for leaks after disassembling a valve" on page 76

To replace the rotor seal

- 1. If you have not already done so, access the rotor seal by following "To remove the stator from the valve" on page 75.
- 2. Remove the rotor seal from the valve driver (see Figure 68).

Figure 68. Rotor seal mounted on the valve driver



- 3. Carefully mount the new rotor onto the valve driver, ensuring that the rotor sealing surface (engraved flow passage), is facing out.
- 4. If you have not already done so, clean the stator with a lint-free tissue or Q-tip soaked in methanol.
- 5. Mount the clean stator onto the valve driver.

- 6. Using a 9/64 in. L-hex wrench, tighten the two hex screws a little at a time by shifting from one screw to the other and back again until the screws are evenly torqued.
- 7. Mount the right side panel to the instrument housing. Then, with a #2 Phillips head screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.

* To reset the usage counter for the valve

- 1. Turn on the EASY-nLC instrument and log in to the system as an administrator.
- 2. Reset the usage counter for the valve as follows:
 - a. Press Maintenance > Devices.
 - b. Select the valve from the list of devices.
 - c. Press the **Summary** tab.
 - d. Press Reset.

The value in the Rotor Shifts box resets to 0.

For more information, see "Checking and Resetting the Device Usage Counters" on page 56.

3. To remove air from the system and check for leaks, follow "To remove air from the system and check for leaks after disassembling a valve" on page 76.

Replacing the Stator

Replacing the stator requires one of these parts.

Valve serial number	Replacement part
V-001000–V-099999	Coated stator, P/N LC226
V-000100–V-000999	Uncoated stator, P/N LC225

Tip The About page of the Rotary Valve view on the Maintenance > Devices page lists the serial number of the selected valve.

To replace the stator in a valve and return the instrument to normal operation, follow these steps:

- 1. "To remove the stator from the valve assembly" on page 79
- 2. "To install the new stator" on page 79
- "To remove air from the system and check for leaks after disassembling a valve" on page 76

* To remove the stator from the valve assembly

- 1. Close down the EASY-nLC system, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 2. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 3. Disconnect the solvent lines from the valve as follows:
 - Use a 1/4 in. open-ended wrench to remove stainless steel fittings.
 - Use the black nanoViper knurled nut to loosen and remove nanoViper fittings.
- 4. Using a 9/64 in. L-hex wrench, remove the two hex screws that secure the stator to the valve driver, and then pull the stator away from the valve driver.

To install the new stator

- 1. Mount the new stator onto the valve driver.
- 2. Using a 9/64 in. L-hex wrench, tighten the two hex screws a little at a time by shifting from one screw to the other and back again until the screws are evenly torqued.
- 3. Reconnect the solvent lines to the valve as follows:
 - For stainless steel fittings, use a 1/4 in. open-ended wrench to tighten the fittings.
 - For the nanoViper fittings in an EASY-nLC 1000 instrument, follow the instructions in "Using nanoViper Fittings" on page 83 to reconnect the nanoViper solvent lines to the valve ports. Take care not to overtighten these fingertight fittings.
- 4. To remove air from the system and check for leak, follow "To remove air from the system and check for leaks after disassembling a valve" on page 76.

Replacing the Check Valves

If a check valve is not functioning, replace it.

Replacing a check valve requires the following tools and materials.

Tools	Parts and materials
• 9/16 in. open-ended wrench	Powder-free safety gloves
• #2 Phillips head screwdriver	• Solvent-side check valve, P/N LC233
	• Waste-side check valve, P/N LC234



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

To replace a check valve

- 1. Close down the EASY-nLC system, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 2. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 3. Do the following:
 - For a solvent-side check valve, remove the solvent inlet line from the check valve, and then terminate the solvent line with a plug.
 - For a waste-side check valve, remove the waste line from the check valve.



CAUTION To prevent solvent from siphoning out of the solvent bottle on top of the instrument and onto system components, use a plug to terminate a solvent inlet line when you disconnect it from the solvent-side check valve.

The pump PCB is easily damaged by contact with solvents.

- 4. Do one of the following:
 - For a solvent-side check valve, use a 9/16 in. open-ended wrench to loosen the valve from the check valve assembly.
 - For the waste-side check valve, remove the check valve by turning it counterclockwise with your fingers.
- 5. Screw the new check valve onto the check valve assembly. Using a 9/16 in. open-ended wrench, slightly tighten the solvent-side check valve to the assembly.

- 6. Do one of the following:
 - For a solvent-side check valve, reconnect the solvent inlet line.
 - For a waste-side check valve, reconnect the waste line.
- 7. Mount the right side panel to the instrument housing. Then, with a #2 Phillips head screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.
- 8. Run the Purge Solvent script for the pump associated with the replaced check valve, and monitor the flow from the solvent bottle when the pump is filling and the flow to the waste beaker when the pump is emptying (see "Prepare Purge Solvent" on page 32).

Replacing an Inline Filter for the EASY-nLC 1000 Instrument

Inline filters are connected to both the inlet and outlet ports of the flow sensors. The inline filter consists of a stainless steel body (nut, filter, and tube) and a ferrule (see Figure 69).

Figure 69. Inline filter components (nut with integrated tube and filter and ferrule)



Replacing an inline filter requires the following tools and materials.

Tools	Parts and materials
• 5/16 in. open-ended wrench	Powder-free safety gloves
• #2 Phillips head screwdriver	Inline filter



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

To install an inline filter into the receiving port of a flow sensor

- 1. Close down the EASY-nLC system, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 2. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.

3. While holding the wide end of the ferrule toward the nut, slide the ferrule over the tube that extends from the threaded end of the nut.

IMPORTANT The inline filters have a swept volume (flow path volume) of less than one microliter.

To prevent leaks, take care to properly seat the ferrule in the receiving port. Once the ferrule is swaged onto the tubing, do not connect the fitting to a different receiving port.

- 4. Using a 5/16 in. open-ended wrench, tighten the nut.
- 5. Install the nanoViper solvent inlet line into the inline filter (see Figure 70).



Figure 70. Installed inline filters

- 6. Run the Back Pressure script and the Leak script on the system as described in these topics:
 - "Test Back Pressure" on page 48
 - "Test Leaks" on page 44

Using nanoViper Fittings

For the EASY-nLC instrument, most of the plumbing connections are made with nanoViper fittings (see Figure 71).

Note For more information about working with nanoViper fittings, see "Using nanoViper Fittings Quick Reference Guide" on page 323.





Even though nanoViper fittings can withstand UHPLC backpressures of up to ~1034 bar (~15 000 psi), they are fingertight fittings, which require only very small torques to seal. Therefore, you must follow the guidelines below to avoid damage by overtightening.

To use a nanoViper fitting

- 1. Insert the nanoViper fitting into the target port and slowly rotate the screw clockwise until you feel resistance.
- 2. Using the black knurled tightening tool, turn the screw clockwise to an angle between 0 and 45 degrees (1/8-turn).
- 3. Run the Leaks script for the system as described in "Running a System Leak Test" on page 218.

When the leak test ends, the system is at atmospheric pressure.

IMPORTANT To extend the lifetime of the nanoViper fittings, open and close connections at atmospheric system pressures only. Opening and closing connections at high system pressures can reduce the lifetime of the fitting system.

4. If the Leaks script fails because the new connection is not leak tight, use the black knurled tightening tool to turn the screw by as much as an additional 45 degrees. Do not turn the screw beyond an angle of 90 degrees from where you felt the initial resistance.

IMPORTANT To prevent damage to the sealing surface of the nanoViper fitting, take care not to overtighten the nanoViper fitting.

Replacing the Autosampler Needle

Replacing the autosampler needle requires these tools and materials.

Tools	Parts and materials
• 1/4 in. open-ended wrench	Powder-free safety gloves
• #2 Phillips head screwdriver	• ASA autosampler needle, P/N LC251
	• ASC autosampler needle, P/N LC302

* To remove the autosampler needle from an ASC or an ASA model autosampler

- 1. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 2. Using a 1/4 in. open-ended wrench, unscrew the nut that secures the autosampler needle to port 1 of valve S. Then remove the fitting from the port.

Figure 72 and Figure 73 show the solvent line connections to valve S for the EASY-nLC II solvent system and the EASY-nLC 1000 solvent system, respectively.



Figure 72. Valve S solvent line connections for the EASY-nLC II instrument




3. Remove the nut, ferrule, and sleeve from the needle tubing.

Figure 74. Autosampler tubing with a nut, ferrule, and sleeve at one end



- 4. Depending on the instrument model, move the *z*-axis needle holder to an accessible location within the tray compartment by doing the following:
 - For the ASA model, go to step 5.
 - For the ASC model, go to step 6.
- 5. For the ASA model, move the *z*-axis needle holder to position A1 as follows:
 - a. Press Home > Overview.
 - b. Press the Autosampler icon.

The Autosampler dialog box opens.

c. In the XYZ Robot area, select position A1 in the Well box (see Figure 75).

Figure 75. Autosampler direct control dialog box

Eject Plate Insert Plate 6x8/standard vials Goto Calibration
Eject Plate Insert Plate 6x8/standard vials Goto Calibration
6x8/standard vials Goto Calibration
6x8/standard vials Goto Calibration
Goto Well
Needle Up Needle Down
W2 W3 W4
7.0 °C Set Disable
Temperature: 7.2 ℃
Close

- d. Press Goto.
- e. Go to step 7 on page 86.
- 6. For the ASC model, move the z-axis needle holder to position W1 as follows:
 - a. Press Home > Overview.
 - b. Press the Autosampler icon.

The Autosampler dialog box opens.

c. In the XYZ Robot area, press **Go to Calibration**.

The Tools page of the Autosampler view on the Maintenance > Devices page opens (see Figure 76).

Thermo SCIENTIFIC	Home	Batch Setup	Method Editor	Maintenance	Configuration	
Scripts	Devices	5				
Log Book		Na	ime	Туре		Add Device
Support	Ct	Cooled Autosampler (ASC) [Autosampler]		Autosampler		
Devices	EASY-nLC™ [HPLC]			HPLC		Remove Device
	Piston Pump (flow feedback) (Pump A)		Piston Pump w/ flo	w fee	Assign Slave	
Job 00:00:00 6 May 2011 10:46 admin Exit	Cooled Autosampler (ASC) [Autosampler] Plate New Calibrate Current: 8x12/Abgene (AB-0800) 8x12/Abgene (AB-0800) Copy To Delete Use					

Figure 76. Tools page of the Autosampler view on the Maintenance > Devices page

d. In the Manipulator area, press **Reset**.

The *z*-axis needle holder moves to the W1 position.

7. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.

- 8. Depending on the instrument model, do the following:
 - For the ASC model, remove the black needle holder from the slot in the metal plate that separates the solvent system hardware from the autosampler compartment. Removing the fitting requires some gentle movement from both sides of the plate (see Figure 77).





- For the ASA model, remove the white plastic needle guide on the back of the touch-screen monitor.
- 9. Loosen the nut that is connected to the *z*-axis needle holder by turning it counterclockwise (see Figure 78). Then carefully pull the autosampler needle upward and away from the holder.

Figure 78. Autosampler needle connected to the *z*-axis needle holder

ASA model *z*-axis needle holder



ASC model z-axis needle holder



To install a new autosampler needle

- 1. Mount the new autosampler needle in the *z*-axis holder. Check that the small spring is situated between the PEEK nut and the plastic stop.
- 2. Do one of the following:
 - For the ASC autosampler, go to step step 3.
 - For the ASA autosampler, go to step step 4.
- 3. For the ASC autosampler, do the following:
 - a. Guide the end of the needle that connects to valve S through the large hole in the metal plate that separates the tray compartment from the solvent system compartment.
 - b. Install the black needle holder in the side plate.
- 4. For the ASA autosampler, slide the valve end of the tubing through the small plastic holder on the back of the touch-screen monitor (see Figure 79).

Figure 79. Holder on the back of the touch-screen monitor



- 5. Connect the needle to port 1 of valve S as follows:
 - a. Slide the provided blue sleeve and metal fittings onto the tubing.
 - b. To ensure a zero dead volume connection, hold the blue sleeve and the tubing firmly against the bottom of the valve port, and then tighten the fitting with a 1/4 in. open-ended wrench.
- 6. Recalibrate the needle position as described in Chapter 7, "Calibrating the Autosampler's XYZ Robot."
- 7. Run 2 iterations of the Purge Solvent script for Pump S.

For information about running the Purge Solvent script, see "Prepare – Purge Solvent" on page 32.

8. Run the Flush Air script for Pump S with a flush volume threshold of 10 μL for the EASY-nLC II system or 12 μL for the EASY-nLC 1000 system.

For information about running the Flush Air script, see "Prepare - Flush Air" on page 33.

Replacing the Sample Loop

Thermo Fisher Scientific provides sample loops in several sizes for the EASY-nLC instruments.

✤ To replace the sample loop

- 1. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 2. To remove the sample loop from the valve, do one of the following:
 - For the EASY-nLC II instrument, use a 1/4 in. open-ended wrench to loosen the fittings that secure the sample loop to ports 2 and 5 of valve S. Then remove the sample loop from the valve.

Figure 80 shows the sample loop connections for the EASY-nLC II instrument.

Figure 80. Sample loop connections for the EASY-nLC II instrument



- For the EASY-nLC 1000 instrument, do the following:
 - a. Return the system to atmospheric pressure (see "Returning the System to Atmospheric Pressure" on page 325).
 - b. Disconnect the nanoViper fittings connected to ports 2 and 5 of valve S.

Figure 81 shows the sample loop connections for the EASY-nLC 1000 instrument.

Figure 81. Sample loop connections for the EASY-nLC 1000 instrument



- 3. Connect the replacement sample loop to valve S as follows:
 - a. Insert the fittings at the end of the sample loop into ports 2 and 5 of valve S.
 - b. Depending on the instrument model, secure the connections as follows:
 - For the EASY-nLC II instrument, use a 1/4 in. open-ended wrench to tighten the fittings.
 - For the EASY-nLC 1000 instrument, secure the fittings to their receiving ports as described in "Using nanoViper Fittings" on page 83. Take care not to overtighten these fingertight fittings.
- 4. Check for leaks by running the system leak test as described in "Checking the Sample Loop Connections for a Leak" on page 222.

Replacing a Pressure Sensor for the PLU Pump

This topic describes how to replace a pressure sensor that is compatible with the PLU model pump (standard in EASY-nLC 1000 instruments).

Note For information about replacing a pressure sensor for a PLF model pump (standard in most EASY-nLC II instruments), see "Replacing a Pressure Sensor for a PLF Pump" on page 117.

Replacing the pressure sensor that is connected through solvent line tubing to a PLU model pump requires these tools and materials.

Parts and materials

Powder-free safety gloves

• Pressure sensor, P/N LC502

Tools

- #2 Phillips head screwdriver
- 1/4 in. open-ended wrench
- 2.5 mm L-hex wrench or ball driver



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

To replace a pressure sensor that is connected to a PLU pump, follow these steps:

- 1. "To remove a pressure sensor from the instrument" on page 91
- 2. "To install the new pressure sensor" on page 92

✤ To remove a pressure sensor from the instrument

- 1. Set the corresponding valve to the **Center** position as described in "Using the Valve Controls" on page 204.
- 2. Close down the EASY-nLC system, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 4. Using a 1/4 in. open-ended wrench, disconnect the stainless steel tubing attached to the inlet and outlet ports of the pressure sensor.
- 5. Using a 2.5 mm L-hex wrench or ball driver, remove the two screws that secure the pressure sensor L bracket to the instrument panel.
- 6. While holding the pressure sensor, use a 2 mm L-hex wrench to remove the two screws that secure the pressure sensor to the bracket.
- 7. Disconnect the pressure sensor cable from the pump.

Figure 82 shows the pressure sensor connections.





✤ To install the new pressure sensor

- 1. Using a 2 mm L-hex wrench and the screws that you removed in step 6 on page 91, secure the new pressure sensor to the bracket.
- 2. Using a 2.5 mm L-hex wrench or ball driver and the screws that you removed in step 5 on page 91, reconnect the pressure sensor bracket to the interior right panel of the instrument.
- 3. Reconnect the stainless steel solvent lines to the inlet and outlet ports of the pressure sensor. Using a 1/4 in. open-ended wrench, tighten the fittings.
- 4. Mount the right side panel to the instrument housing. Then, with a #2 Phillips head screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.
- 5. Reconnect the pressure sensor cable.
- 6. Run the Flush Air script as described in "Prepare Flush Air" on page 33.

Replacing a Flow Sensor

The procedure for replacing the flow sensor differs between the EASY-nLC 1000 system and the EASY-nLC II system.

Depending on the instrument model, follow one of these procedures to replace a flow sensor:

- "Replacing a Flow Sensor in the EASY-nLC 1000 Instrument," on this page
- "Replacing a Flow Sensor in the EASY-nLC II Instrument" on page 96

Replacing a Flow Sensor in the EASY-nLC 1000 Instrument

Replacing a flow sensor in the EASY-nLC 1000 system requires these tools and materials.

Tools	Parts and materials
• #2 Phillips head screwdriver	Powder-free safety gloves
• 8 mm open-ended wrench	• Flow sensor A/B, P/N LC540

• 2.5 mm hex wrench or ball driver



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

To replace a flow sensor in the EASY-nLC 1000 system, follow these steps:

- 1. "To remove an EASY-nLC 1000 flow sensor," on this page
- 2. "To install an EASY-nLC 1000 flow sensor" on page 95

To remove an EASY-nLC 1000 flow sensor

- 1. Close down the EASY-nLC system, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 2. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 3. Remove the nanoViper tubing and from the inlet and outlet of the flow sensor.

Figure 83 shows the flow sensors mounted behind the bracket. The flow direction is labeled on the bracket.



4. Using an 8 mm open-ended wrench, remove the inline filters from both ends of the flow sensor (see Figure 83).

Figure 83. Flow sensors

5. Remove the black cable from the flow sensor (see Figure 84).



Figure 84. Removing the cable from the flow sensor

6. Using an 2.5 mm hex wrench, remove the two screws that secure the flow sensor to the bracket (see Figure 85).

Figure 85. Removing the screws attaching the flow sensor to the bracket



Location of screws that secure flow sensor A

7. Remove the flow sensor (see Figure 86).

Figure 86. Removing the flow sensor



To install an EASY-nLC 1000 flow sensor

- 1. Insert the new flow sensor behind the bracket.
- 2. Insert the two screws (Figure 85 on page 94) that secure the flow sensor to the bracket, and tighten the screws with a 2.5 mm hex wrench.
- 3. Connect the black cable to the flow sensor (see Figure 84 on page 94).
- 4. Reinstall the inline filters on both ends of the flow sensor (see Figure 83 on page 93). Tighten the fittings with an 8 mm open-ended wrench.
- 5. Reconnect the nanoViper tubing to both ends of the flow sensor (see Figure 83 on page 93).
- 6. Replace the right side panel.
- 7. Turn on the EASY-nLC 1000 system, and wait one hour for the flow sensor to reach the correct operating temperature.
- Calibrate the new flow sensor by using the Flow Sensors script (see "Calibrate Flow Sensors" on page 51).

Replacing a Flow Sensor in the EASY-nLC II Instrument

Replacing a flow sensor in the EASY-nLC II instrument requires these tools and materials.

Tools	Parts and materials
• #2 Phillips head screwdriver	Powder-free safety gloves
• 2.5 mm hex wrench or ball	• Flow sensor A type SLG1430-150, P/N LC240
driver	• Flow sensor B type SLG1430-025, P/N LC241



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

To replace an EASY-nLC II flow sensor, follow these steps:

- 1. "To remove an EASY-nLC II flow sensor," on this page
- 2. "To install an EASY-nLC II flow sensor" on page 99

To remove an EASY-nLC II flow sensor

- 1. Close down the EASY-nLC system, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 2. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 3. Using a 2.5 mm hex wrench, remove the two screws (see Figure 87) that secure the flow sensor bracket to the instrument panel.



Figure 87. Removing the two screws that secure the flow sensor bracket to the panel



Remove the solvent lines from both ends of the flow sensor (see Figure 88).
 Figure 88. Removing the solvent lines



5. Loosen the metal ring and remove the cable connected to the flow sensor (see Figure 89).





connected to the flow sensor

6. Using a 2.5 mm hex wrench, remove the two screws that secure the flow sensor to the bracket (see Figure 90).





To install an EASY-nLC II flow sensor

- 1. Install the new flow sensor into the bracket as follows:
 - a. Align the new flow sensor with the mounting holes in the bracket.

IMPORTANT You must replace flow sensor A with type SLG1430-150 and flow sensor B with type SLG1430-025.

- Insert the two screws that you removed earlier in step 6 of "To remove an EASY-nLC II flow sensor," into the bracket (see Figure 90 on page 98).
- c. Using a 2.5 mm L-hex wrench, tighten the two screws to secure the flow sensor to the bracket.
- 2. Reconnect the cable to the flow sensor, and tighten the metal ring (see Figure 89 on page 98).
- 3. Reconnect the solvent lines to both ends of the flow sensor (see Figure 88 on page 97).
- 4. Remount the bracket to the panel as follows:
 - a. Align the bracket with the mounting holes in the panel.
 - b. Insert the screws that you removed from the bracket when you removed it from the panel.
 - c. Using a 2.5 mm L-hex wrench, tighten the two screws to secure the flow sensor bracket to the panel (see Figure 87 on page 97).
- 5. Mount the right side panel to the instrument housing. Then, with a #2 Phillips head screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.
- 6. Turn on the EASY-nLC II instrument.
- 7. Wait 30 minutes for the flow sensors to reach the proper operating temperature.
- Calibrate the new flow sensor by using the Flow Sensors script (see "Calibrate Flow Sensors" on page 51).

Replacing the Hard Drive

Use this procedure to replace the hard drive in either the EASY-nLC 1000 instrument or the EASY-nLC II instrument.

Thermo Fisher Scientific supplies several versions of the hard drive. Order the appropriate hard drive based on both the instrument model and the computer box where you are replacing the hard drive. The discontinued versions of the computer box have a PS/2 port (see Figure 91).



Figure 91. Comparison of computer ports between the obsolete and current computer boxes

Replacing the hard drive requires these tools and materials.

Tools	Replacement part
2.5 mm hex wrench	Hard drive:
Torx T-10 wrench	• EASY-nLC II instrument:
	 Compatible with obsolete computer box: P/N LC281
	 Compatible with current computer box: P/N LC286
	-10-
	• EASY-nLC 1000 instrument:
	 Compatible with obsolete computer box: P/N LC581
	 Compatible with current computer box: P/N LC586

To replace the hard drive, follow these steps:

- 1. "To remove the hard drive" on page 101
- 2. "To install the hard drive" on page 103

✤ To remove the hard drive

- 1. Close down the EASY-nLC instrument, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 2. Using a 2.5 mm hex wrench, loosen the two screws in the hard-drive drawer (see Figure 92).

Figure 92. Screws that secure the hard-drive drawer (obsolete computer box shown)



Two screws that secure the hard-drive drawer

3. Pull out the hard-drive drawer, as shown in Figure 93.

Figure 93. Pulling out the hard-drive drawer



4. Using a Torx T-10 wrench, remove the four screws underneath the hard-drive drawer that secure the hard drive to the drawer (see Figure 94).

Figure 94. Removing the screws underneath the hard-drive drawer



5. Unplug the hard drive from the IDE cable (see Figure 95).

Figure 95. Hard drive unplugged from the IDE cable





The flat (IDE) cable is connected to the hard disk. The text on the hard disk is upside down, and the pins on the hard drive fit in the same place on the flat cable connector.

Tip If the IDE cable plug is difficult to access, remove the brass rod that holds the cable down (see Figure 96). Use a Torx T-8 wrench to remove the screws. Place the rod back in the drawer afterward.

Figure 96. Removing the brass rod that holds the cable down



✤ To install the hard drive

- 1. Insert the new hard drive.
- 2. Connect the hard drive to the IDE cable.
- 3. Insert the four screws into the holes underneath the hard-drive drawer and then, using a Torx T-10 wrench, tighten the four screws to secure the hard drive to the drawer (see Figure 94 on page 102).
- 4. Close the hard-drive drawer.
- 5. Using a 2.5 mm L-hex wrench, insert and tighten the two screws that secure the drawer to the back panel (see Figure 92 on page 101).
- 6. Turn on the EASY-nLC instrument.
- 7. Press Vendor.
- 8. When prompted, enter the serial numbers for the instrument and the hard drive.

Note When you replace the hard drive, the pumps and valves are automatically detected during the boot procedure. Because the autosampler is not detected by the boot procedure, you must add it to the Device list. After you add the autosampler to the Device list, you must calibrate the plate formats that you plan to use and the wash bottle positions, as these calibrations were stored on the old hard drive.

- 9. Add the autosampler to the Devices list as follows:
 - a. Press Maintenance > Devices.
 - b. Press Add Device.

The Select a Device to Add dialog box opens.

- c. Locate the autosampler device and select it.
- d. Press **Accept** to add the autosampler device to the Devices list and close the Select a Device to Add dialog box.
- e. Verify that the autosampler device was added to the system by locating it in the Devices list.
- 10. Calibrate the autosampler as described in Chapter 7, "Calibrating the Autosampler's XYZ Robot."

Managing the Devices List

The Device list on the Maintenance >Devices page lists the current devices installed in the EASY-nLC instrument.

When you add or remove a device that is not automatically detected during the boot procedure, you must add the device to or remove the device from the Devices list on the Maintenance > Devices page. Devices that are not recognized during the boot procedure include the autosampler and the external RePlay device.

✤ To add a device to the Device list

- 1. Press Maintenance > Devices.
- 2. Press Add Device.

The Select a Device to Add dialog box opens.

Select a device to add.		
Туре		
Fluid Storage		
Autosampler		
Autosampler		
HPLC		
Syringe Drive		
Liquid Depositor	•	
	Type Fluid Storage Autosampler Autosampler HPLC Syringe Drive Liquid Depositor	

- 3. Locate the device in the list and select it.
- 4. Press **Accept** to add the autosampler device to the Devices list and close the Select a Device to Add dialog box.
- 5. Verify that the device was added to the system by locating it in the Devices list. Select the device in the Devices list, and review the information on the About page.

To remove a device from the Devices list

- 1. Press Maintenance > Devices.
- 2. Select the device in the Devices list.
- 3. Press Remove Device.

The Remove Device dialog box appears.

4. Press Accept to remove the device and return to the Maintenance > Devices page.

✤ To view information about a device

- 1. Press Maintenance > Devices.
- 2. Select the device in the Devices list.
- 3. Press the **About** tab.

The About view contains information about the serial numbers and firmware versions for the rotary valves, piston (syringe) pumps, autosampler, and HPLC.

5

Field Service Maintenance

This chapter provides procedures for use by Thermo Fisher Scientific field service engineers on replacing the subassemblies in the EASY-nLC instrument. Only Thermo Fisher Scientific field service engineers can perform these procedures.



CAUTION Only Thermo Fisher Scientific field service engineers can perform the procedures in this chapter.

Contents

- Replacing the Built-In Computer
- Replacing the Monitor
- Replacing a Pressure Sensor for a PLF Pump
- Replacing the ASC Autosampler
- Upgrading from an ASA Autosampler to an ASC Autosampler
- Replacing the Autosampler Cooler
- Replacing a Rotary Valve
- Replacing a Pump
- Replacing a Pump PCB

Replacing the Built-In Computer

The procedure for replacing the built-in computer is the same for the EASY-nLC 1000 and EASY-nLC II instruments, except where noted.

Replacing the computer box requires the following part and tools.

Tools	Parts
• #2 Phillips screwdriver	Computer box:
• Torx T-10 wrench	• EASY-nLC II instrument: P/N LC285
• Small flathead screwdriver	-or-
	• EASY-nLC 1000 instrument: P/N LC585

To replace the built-in computer, follow these steps:

- 1. "To remove the computer box," on this page
- 2. "To install the new computer box" on page 112

✤ To remove the computer box

- 1. Close down the EASY-nLC instrument (see "Closing Down the EASY-nLC Instrument" on page 21), and then unplug the power cable.
- 2. Disconnect all of the cables from the instrument's back panel.
- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the left side panel to the instrument housing. Then remove the panel.
- 4. Remove the internal cables connected to the computer box.

Note The cable connectors can be hard to loosen. You might need to wiggle the connectors carefully from side to side to loosen them.

Note Take care when you disconnect the monitor LVDS cable because it is easily damaged.

Figure 97 shows these cables in the EASY-nLC 1000 instrument and Figure 98 shows these cables in the EASY-nLC II instrument.



Figure 97. Cables connected to the computer box in the EASY-nLC 1000 instrument

Figure 98. Cables connected to the computer box in the EASY-nLC II instrument



Tip To remove the 12 V cables on the EASY-nLC II instrument, press the tab on the top of the plug, as shown in Figure 99.

Figure 99. Pressing the tab on the top of the plug of the 12 V cables



5. Using a Torx T-10 wrench, remove the screws that secure the back panel to the instrument (see Figure 100).

Note In older EASY-nLC instruments, 11 screws secured the back panel to the housing. In current EASY-nLC 1000 instruments, 3 screws secure the back panel to the housing.

Figure 100. Screws that secure the back panel to the instrument housing



6. Using a Torx T-10 wrench, remove the four screws that secure the computer box to the instrument (see Figure 101).



Figure 101. Screws to remove from the computer box

7. Pull the computer box out of the EASY-nLC housing, as shown in Figure 102.

Figure 102. Pulling out the computer box



✤ To install the new computer box

- 1. Insert the new computer box into the EASY-nLC housing.
- 2. Reconnect all of the internal cables to the computer box.
 - Figure 97 on page 109 shows the connections to the computer box inside the EASY-nLC 1000 instrument.
 - Figure 98 on page 109 shows the connections to the computer box inside the EASY-nLC II instrument.
- 3. Using a Torx T-10 wrench and the four screws that you removed in step 6 on page 111, secure the computer box to the instrument (see Figure 101 on page 111).
- 4. Using a Torx T-10 wrench and the screws that you removed in step 5 on page 110, secure the back panel to the instrument.

Tip Do not reconnect the left side panel to the instrument housing until after you recalibrate the autosampler.

- 5. Reconnect the cables to the back panel as follows:
 - Reconnect the power cable to the back panel and to a laboratory power outlet.
 - If the instrument was connected to a laboratory LAN port, reconnect the Ethernet cable.
 - If the instrument is part of a Thermo Scientific LC/MS system, reconnect the Ethernet cable to the back panel of the instrument, and make sure that the other end of the cable connects to the Ethernet switch for the data system hardware.
- 6. Turn on the EASY-nLC instrument, and log in as an administrator.
- 7. Add the autosampler to the Devices list (see "Managing the Devices List" on page 105).
- 8. Recalibrate the autosampler (see Chapter 7).

Replacing the Monitor

The procedure for replacing the monitor is the same for the EASY-nLC 1000 instrument and the EASY-nLC II instrument, except where noted.

Replacing the monitor requires the following part and tools.

Tools	Part
Torx T-10 wrench2 mm L-hex wrench or straight wrench	Monitor (and adapter cable for computer boxes with a PS/2 keyboard connector as shown in Figure 16 on page 16), P/N LC282

To replace the monitor, follow these steps:

- 1. "To remove the monitor," on this page
- 2. "To install the new monitor" on page 116

To remove the monitor

- 1. Close down the EASY-nLC instrument (see "Closing Down the EASY-nLC Instrument" on page 21), and unplug the power cable.
- 2. Depending on the instrument model, do the following:
 - For the EASY-nLC II instrument, go to step 3 to remove the front panel.
 - For the EASY-nLC 1000 instrument, go to step 4 to remove the top-front panel.
- 3. To remove the front panel from the EASY-nLC II instrument, do the following:
 - a. Using a flat-edged tool, pry off the Thermo blue applique.
 - b. Using a Torx T-10 wrench, remove the 10 screws that secure the front plate to the EASY-nLC II instrument. Then remove the plate (see Figure 103).
 - c. Go to step 5.

Figure 103. Removing the front plate from the EASY-nLC II instrument



- 4. To remove the top-front panel from the EASY-nLC 1000 instrument, do the following:
 - a. Using a Torx T-10 wrench, remove the two screws that secure the top-front panel to the top back of the instrument.
 - b. Open the tray compartment door, and then use a Torx T-10 wrench to remove the screw that secures the top-front panel to the front of the instrument.
- 5. Using a 2 mm hex wrench, remove the four screws that secure the monitor to the instrument (see Figure 104).



CAUTION Use only hand tools with a hex tip to avoid damaging the screw heads.

Figure 104. Removing the screws from the front of the monitor



- 6. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the left side panel to the instrument housing. Then remove the panel.
- 7. Remove the USB, LVDS, and monitor cables connected to the computer box, being careful not to damage the LVDS plug (see Figure 105).

Figure 105. USB, LVDS, and monitor cables



8. If a white plastic needle guide is installed on the back of the monitor, remove it before removing the monitor.

Figure 106 shows the white plastic needle guide on the back of the monitor.

Figure 106. White plastic needle guide



Carefully remove the monitor by pulling it straight forward (see Figure 107).
 Figure 107. Removing the monitor



✤ To install the new monitor

- 1. Carefully insert the new monitor into the instrument housing.
- 2. If you removed the white plastic needle guide on the back of the monitor, reinstall it.

Figure 106 on page 115 shows the white plastic needle guide mounted to the back of the monitor.

- 3. If the computer box has a PS/2 keyboard connector on its back panel as shown in Figure 16 on page 16, connect the adapter cable that is supplied with the new monitor to the monitor cable.
- 4. Connect the USB, LVDS, and monitor cables to the computer box, being careful not to damage the LVDS plug (see Figure 105 on page 114).
- 5. Using a 2 mm ball driver and the screws that you removed in step 5 on page 114, secure the monitor to the instrument.
- 6. Depending on the instrument model, do the following:
 - For the EASY-nLC II instrument, use a Torx T-10 wrench and the 10 screws that you removed in step b on page 113 to secure the front plate to the housing.
 - For the EASY-nLC 1000 instrument, use a Torx T-10 wrench and the three screws that you removed in step 4 on page 114 to secure the top-front panel to the housing.
- 7. Mount the left side panel to the instrument housing. Then, with a #2 screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.
- 8. Plug in the power cable, and then turn on the EASY-nLC instrument.

Replacing a Pressure Sensor for a PLF Pump

The procedure for replacing a pressure sensor differs between EASY-nLC 1000 instruments with PLU pumps and EASY-nLC II instruments with PLF pumps.

Note Most EASY-nLC II instruments contain the PLF pump model.

The following procedure explains how to replace a pressure sensor that is connected to a PLF pump. For instructions about replacing a pressure sensor that is connected to a PLU pump, see "Replacing a Pressure Sensor for the PLU Pump" on page 90.



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

Replacing the pressure sensor that is connected to a PLF pump requires these items.

Tools	Parts and materials
• # 2 Phillips screwdriver	Powder-free gloves
• 2.0 mm L-hex wrench	• Pressure sensor, P/N LC202

- 2.5 mm L-hex wrench or ball driver
- 1/4 in. open-ended wrench

To replace a pressure sensor that is connected to a PLF pump, follow these steps:

- 1. "To remove a pressure sensor that is connected to a PLF pump," on this page
- 2. "To install a pressure sensor for a PLF pump" on page 121
- To remove a pressure sensor that is connected to a PLF pump
- 1. Set the corresponding valve to the **Center** position as described in "Using the Valve Controls" on page 204.
- 2. Close down the EASY-nLC II instrument (see "Closing Down the EASY-nLC Instrument" on page 21), and unplug the power cable.
- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.

4. Disconnect the P-Bus communication cables from the pump (see Figure 108).Figure 108. P-Bus communication cables connected to the pump PCB of the PLF pump



- P-Bus communication cables
- 5. Using a 2.5 mm L-hex wrench or ball driver, remove the four 2.5 mm hex screws that secure the PCB to the pump.

Figure 109 shows these screws. The lower right screw also connects the pressure sensor grounding cable to the PCB.

Figure 109. Four hex screws connecting the PCB to the PLF pump



Pressure sensor grounding cable

6. Gently pull the PCB toward you and rotate it to gain access to its back side (see Figure 110).

Figure 110. Turning the PCB around (PLF pump)

Pressure sensor communication cable connection to the back side of the PCB



7. Disconnect the pressure sensor communication cable, being careful not to put any stress on the cables (see Figure 111).

Figure 111. Disconnecting the pressure sensor communication cable (PLF pump)



Pressure sensor communication cable connection to the back side of the PCB 8. While holding the pressure sensor, use a 1/4 in. open-ended wrench to disconnect the solvent lines connected to the inlet and outlet of the pressure sensor (see Figure 112).



Figure 112. Disconnecting the solvent lines from the pressure sensor

9. Using a 2.5 mm L-hex wrench or ball driver, remove the two hex screws that secure the pressure sensor L bracket to the instrument panel (see Figure 113).





10. Using a 2 mm L-hex wrench or ball driver, unscrew the two hex screws that secure the pressure sensor to the L-bracket (see Figure 113).
* To install a pressure sensor for a PLF pump

- 1. Disconnect and remove the pressure sensor from the EASY-nLC II instrument as described in "To remove a pressure sensor that is connected to a PLF pump" on page 117.
- 2. Using a 2 mm L-hex wrench or ball driver, insert and tighten the two hex screws to secure the pressure sensor to the L-bracket.
- 3. Using a 2.5 mm L-hex wrench or ball driver, insert and tighten the two hex screws to secure the pressure sensor L-bracket to the instrument panel (see Figure 113 on page 120).
- 4. Reconnect the solvent lines to the pressure sensor (see Figure 112 on page 120).
- 5. Reconnect the pressure sensor communication cable, being careful not to put any stress on the cables (see Figure 111 on page 119).
- 6. Gently slide in the PCB.
- 7. Using a 2.5 mm L-hex wrench or ball driver, insert and tighten the four 2.5 mm hex screws to connect the PCB to the pump (see Figure 109 on page 118).

Note The screw in the lower right also connects the pressure sensor grounding cable to the PCB.

- 8. Reconnect the P-Bus communication cables to the pump (see Figure 108 on page 118).
- 9. Mount the right side panel to the instrument housing. Then, with a #2 screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.
- 10. Turn on the EASY-nLC II instrument and log in as an administrator (see "Logging In to the EASY-nLC Instrument for Maintenance Tasks" on page 20).
- To draw fresh solvent into the pump, purge the pump five times by using the Purge Solvent script on the Maintenance > Scripts page (see "Prepare – Purge Solvent" on page 32).
- 12. To remove air from the system, run the Flush Air script for the pump associated with the new pressure sensor (see "Prepare Flush Air" on page 33).

Replacing the ASC Autosampler

The procedure for replacing the ASC autosampler is the same for the EASY-nLC 1000 instrument and the EASY-nLC II instrument. However, if your EASY-nLC instrument has an installed ASA autosampler that is irreversibly damaged, you must upgrade to an ASC autosampler, because the ASA autosampler is no longer available. For instructions on upgrading to an ASC autosampler, see "Upgrading from an ASA Autosampler to an ASC Autosampler" on page 128.



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

Replacing the ASC autosampler requires these items.

Tools	Parts and materials
• #2 Phillips screwdriver	Powder-free gloves
• 3 mm L-hex wrench or ball driver	• 4 in. or 10 cm length tie wrap
• Torx T-10 wrench	• Autosampler (ASC), P/N LC301

• Cutting pliers

To replace the autosampler, follow these steps:

- 1. "To remove the ASC autosampler," on this page
- "To install the ACS autosampler into an instrument that previously included this model" on page 127

* To remove the ASC autosampler

- 1. Remove the old autosampler from the device list as follows:
 - a. Press Maintenance > Devices.
 - b. Select the autosampler in the Devices list.
 - c. Press Remove Device.

The Remove Device dialog box opens.

- d. Press Accept.
- 1. Close down the EASY-nLC instrument (see "Closing Down the EASY-nLC Instrument" on page 21). Then unplug the power cable.
- 2. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive quarter-turn screws that secure each side panel to the instrument housing. Then remove both panels.

- 3. Remove the computer box from the instrument as described in "To remove the computer box" on page 108.
- 4. Unscrew the autosampler needle fitting, and remove or pull up the autosampler needle from the needle holder.

Figure 114 shows the autosampler needle connected to the needle holder.

Figure 114. Needle connected to the needle holder



5. Unplug the bus cables on the left side of the instrument from the autosampler cooler and horizontal XYZ-axis PCB (see Figure 115).

Figure 115. Bus cables



6. Cut the tie wrap attaching the cables to the vertical part of the autosampler frame, as shown in Figure 116.

Figure 116. Cutting the strip attaching the cables to the autosampler frame



7. Using a Torx T-10 screwdriver, remove the screws that secure the back horizontal cable bar to the autosampler frame (see Figure 117).

Figure 117. Horizontal cable bar after removal of screws



8. Move the cable bar to the side (this is easier to do if you also remove the bus cable to valve B). See Figure 118.



Figure 118. Horizontal cable bar moved aside

- 9. Using a 3 mm hex wrench, remove the three screws that secure the autosampler to the bottom plate of the housing (see Figure 119, Figure 120, and Figure 121):
 - Remove the screw from the right side of the autosampler (see Figure 119).

Figure 119. Removing the screw from the right side of the autosampler



Remove the screw from the back left side of the autosampler (see Figure 120).
 Figure 120. Removing the screw from the back left of the autosampler



Remove the screw the screw on the front of the autosampler (see Figure 121).
 Figure 121. Removing the front screw from the autosampler



10. Remove the autosampler from the housing through the back of the instrument.

Figure 122 shows the housing with the autosampler partially removed.

Figure 122. Housing with autosampler partially removed



* To install the ACS autosampler into an instrument that previously included this model

- 1. Insert the new autosampler into the housing.
- 2. Using a 3 mm L-hex wrench and, if necessary, a needle-nosed pliers or tweezers, insert and tighten the screws that secure the autosampler to the bottom of the housing as follows:
 - Insert and tighten the screw on the back left side of the autosampler (see Figure 120 on page 126).
 - Insert and tighten the screw on the right side of the autosampler (see Figure 119 on page 125).
 - Insert and tighten the screw on the front of the autosampler (see Figure 121 on page 126).
- 3. Insert and tighten the screws that secure the back horizontal cable bar to the autosampler frame (see Figure 117 on page 124).
- 4. Install a new tie wrap to secure the cables to the vertical part of the autosampler frame.
- 5. Plug the bus cables into the autosampler cooler and horizontal XYZ-axis PCBs.
- 6. Reinstall the computer box as described in "To install the new computer box" on page 112.
- 7. Reinsert the autosampler needle into the holder and tighten the fitting.
- 8. Mount the right side panel to the instrument housing. Then, with a #2 screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.

Tip Reconnect the left side panel to the housing after you recalibrate the autosampler.

- 9. Reconnect the power cable.
- 10. Turn on the EASY-nLC instrument.
- 11. Add the ASC autosampler to the Devices list on the Maintenance > Devices page as follows:
 - a. Press Maintenance > Devices to open the Maintenance > Devices page.
 - b. Press Add Devices to open the Select the Device to Add dialog box.
 - c. Select the ASC autosampler from the list, and then press Accept.
 - d. Verify that the ASC autosampler has been added to the Devices list.
 - i. Select the ASC autosampler in the Devices list.
 - ii. Check the information on the About page.

For more information about adding devices to the Devices list, see "Managing the Devices List" on page 105.

12. Calibrate the autosampler as described in Chapter 7.

Upgrading from an ASA Autosampler to an ASC Autosampler

If your EASY-nLC II instrument has an ASA autosampler that is irreversibly damaged, you must upgrade to an ASC autosampler because the ASA autosampler is no longer available.



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

Upgrading the instrument by replacing an ASA autosampler with an ASC autosampler requires these items.

Fools .	Parts and materials
• 4 mm ballpoint hex wrench	Powder-free gloves
• 2 mm L-hex wrench or ball driver	• M5 ×16 MC hex screw
• 2.5 mm L-hex wrench or ball driver	• ASC autosampler, P/N LC301
• Torx T-10 wrench	
• #2 Phillips screwdriver	
• Tweezers	

To replace the ASA autosampler with an ASC autosampler, follow these steps:

- 1. "To remove the ASA autosampler from the EASY-nLC II instrument," on this page
- "To install the ASC autosampler into an instrument that previously held the ASA model" on page 130

* To remove the ASA autosampler from the EASY-nLC II instrument

1. Upgrade the touch-screen software to the latest version as described in "Downloading the Latest Firmware File" on page 296.

Note Read the release notes before you upgrade to ensure that you have the most recent version of the HPLC software.

- 2. Press Maintenance > Devices and remove the old autosampler from the device list.
- 3. Close down the EASY-nLC instrument (see "Closing Down the EASY-nLC Instrument" on page 21). Then unplug the power cable.
- 4. Remove the computer box as described in "To remove the computer box" on page 108.

Note Removing the computer box requires the removal of the left side panel and the back panel.

- 5. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 6. Remove the monitor as described in "To remove the monitor" on page 113.
- 7. Using a 2.5 L-hex wrench or ball driver, loosen valve W (see Figure 123).

Figure 123. Loosened valve W



- 8. Disconnect the two P-Bus communication cables from the cooler PCB and XYZ-axis PCB on the right side of the instrument (see Figure 129 on page 132).
- 9. Remove the ASA autosampler through the side of the housing.
- 10. Disconnect the cables that supply power to the fans, and remove the fan assembly at the back of the housing (see Figure 124).

Figure 124. Removing the fan assembly and cables



• To install the ASC autosampler into an instrument that previously held the ASA model

Note Because you are upgrading the instrument by adding an ASC autosampler, this upgrade procedure differs from the simple installation procedure on page 127.

1. As you insert the new autosampler into the housing, move the loosened valve W a little to create the required space, as shown in Figure 125.

Figure 125. Putting in the ASC autosampler



2. Align the holes in the autosampler plate with the three mounting holes in the bottom of the housing (see Figure 126).

Figure 126. Aligning the holes in the autosampler plate with the mounting holes



- 3. Reinstall the computer box into the EASY-nLC housing.
- 4. Reconnect all of the internal cables to the computer box.

Figure 98 on page 109 shows the connections to the computer box inside the EASY-nLC II instrument.

5. Insert the P-Bus terminator between the computer box and the P-Bus communication cables as shown in Figure 127. It does not matter which cable the terminator connects to.

Note Do not install the connector if the computer box is labeled "Terminated," as shown in Figure 128. This designation means that you do not have to install a separate terminator cable.

Figure 127. P-Bus connector inserted between the computer box and the P-Bus communication cables



P-Bus connector

Figure 128. Computer box with "Terminated" label



- 6. Using a Torx T-10 wrench and the four screws that you removed in step 6 on page 111, secure the computer box to the instrument (see Figure 101 on page 111).
- 7. Connect the P-Bus communication cables to the autosampler.

The longer cable connects to the cooler PCB. The shorter cable connects to the XYZ-axis PCB. Figure 129 shows these two cables.

Figure 129. P-Bus communication cables



Figure 130 shows the boards that the P-Bus communication cables connect to. Figure 130. Boards that the P-Bus communication cables connect to



8. Using a 2.5 mm hex wrench or ball driver, mount valve W.

- 9. Secure the autosampler to the housing as follows:
 - a. Using a pair of tweezers, place the $M5 \times 16$ hex screw in the back right mounting position, lifting up the autosampler in order to put the screw in place.

Figure 131, Figure 132, and Figure 133 demonstrate this procedure.

Figure 131. Lifting up the autosampler to position the screw



Figure 132. Placing the $M5 \times 16$ hex screw into the back right side of the autosampler frame



Figure 133. The M5 × 16 hex screw in place



b. Insert the screw into the back left side of the autosampler. Use a ball driver to tighten the screw (see Figure 134).

Figure 134. Securing the back left screw



c. Insert the screw into the front of the autosampler. Use a ball driver to tighten the screw (see Figure 135).

Figure 135. Securing the front screw



10. Place the air filter from the ASA autosampler onto the ASC cooler (see Figure 136).

Figure 136. Placing the air filter onto the cooler



Because the injection needles are different for the ASA and ASC autosamplers, you must install a needle arm that can be held in place underneath the mixing Tee holder.

11. Mount the aluminum needle arm under the mixing Tee holder (see Figure 137).

Figure 137. Aluminum needle arm



12. Place the mixing Tee in the holder. Use the aluminum needle arm when you attach the needle (see Figure 138).





13. Reinstall the monitor as described in "To install the new monitor" on page 116.

Tip Do not reconnect the left side panel to the instrument housing until after you recalibrate the autosampler.

14. Turn on the instrument, and then log in to the instrument as an administrator.

- 15. Add the ASC autosampler to the Devices list on the Maintenance > Devices page as follows:
 - a. Press Maintenance > Devices.

The Maintenance > Devices page opens.

b. Press Add Devices.

The Select the Device to Add dialog box opens.

- c. Select the ASC autosampler from the list, and then press Accept.
- d. Verify that the ASC autosampler has been added to the Devices list.

For more information about adding devices to the Devices list, see "Managing the Devices List" on page 105.

16. Calibrate the autosampler as described in Chapter 7, "Calibrating the Autosampler's XYZ Robot."

Replacing the Autosampler Cooler

The procedure for replacing the autosampler cooler is the same for the EASY-nLC 1000 and EASY-nLC II instruments, but it differs between an ASC cooler and an ASA cooler.

To replace the autosampler cooler, follow the appropriate procedure:

- "Replacing the ASC Cooler," on this page
- "Replacing the ASA Cooler" on page 140

Replacing the ASC Cooler

Replacing the ASC cooler requires these items.

Tools	Parts
• Torx T-10 wrench	ASC autosampler cooling module, P/N LC310
• 2.5 mm L-hex wrench	

To replace the ASC cooler, follow these steps:

- 1. "To remove the ASC cooler," on this page
- 2. "To install the new ASC cooler" on page 139

To remove the ASC cooler

- 1. Close down and turn off the EASY-nLC instrument (see "Closing Down the EASY-nLC Instrument" on page 21). Then disconnect the power cable.
- 2. Using a Torx T-10 wrench, remove the screws on the back cover panel, and remove the panel.
- 3. Access the PCB on the back of the cooler through the back of the instrument, and unplug the P-Bus communication cables, shown in Figure 139.

Figure 139. P-Bus communication cables



4. Using a 2.5 mm ball driver, remove the two 2.5 mm hex screws at the bottom of the cooler, shown in Figure 140.

Figure 140. Screws at the bottom of the cooler



5. Using a 2.5 mm ball driver, remove the 2.5 mm hex screw in the top center position of the cooler, as shown in Figure 141.

The screws do not fall out when loosened, because they are held in place by O-rings. **Figure 141.** Removing the top center screw



6. Pull out the cooler, as shown in Figure 142.

Figure 142. Pulling out the cooler



✤ To install the new ASC cooler

1. Insert the new cooler.

Be careful not to damage any of the cables during reassembly. Figure 143 shows a damaged cable.

Figure 143. Damaged cable



- 2. Using a 2.5 mm ball driver, insert and tighten the 2.5 mm hex screw in the top center position (see Figure 151 on page 143).
- 3. Using a 2.5 mm ball driver, insert and tighten the two 2.5 mm hex screws at the bottom of the cooler (see Figure 150 on page 143).
- 4. Plug the P-Bus communication cables into the PCB on the back of the cooler, accessing the PCB through the back of the instrument.
- 5. Align the back panel to the back of the instrument.
- 6. Using a Torx T-10 wrench, tighten the eleven screws that secure the back panel to the instrument.
- 7. Reconnect the power cable.
- 8. Turn on the EASY-nLC instrument.

Replacing the ASA Cooler

Replacing the ASA cooler in an EASY-nLC II instrument requires these items.

Tools	Parts
• 2 mm L-hex wrench or ball driver	ASA autosampler cooling module,
• 2.5 mm L-hex wrench or ball driver	P/N EXLC252
• 3 mm L-hex wrench	
• 5.5 mm open-ended wrench	
• #2 Phillips screwdriver	

To replace the ASA cooler, follow these steps:

- 1. "To remove the ASA cooler," on this page
- 2. "To install the ASA cooler" on page 145

* To remove the ASA cooler

- 1. Close down and turn off the EASY-nLC instrument (see "Closing Down the EASY-nLC Instrument" on page 21). Then disconnect the power cable.
- 2. Remove the autosampler door, shown in Figure 144, to avoid damaging it when you remove the hex screws underneath the adapter plate holder.

Figure 144. Autosampler door



- 3. Depending on the instrument model, do one of the following:
 - On the EASY-nLC II model LC100, use a 5.5 mm open-ended wrench to loosen the nuts underneath the door (see Figure 145).



CAUTION Be careful removing the lid because the spring on the door can snap back.

Figure 145. Nuts beneath the lid of the EASY-nLC II instrument



• On the EASY-nLC II model LC100-2, use a 2 mm L-hex wrench to loosen the screws on the front (see Figure 146).

Figure 146. Screws on the front of the EASY-nLC II instrument (model LC100-2)



4. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the left side panel to the instrument housing. Then remove the panel.

5. Using a flathead screwdriver or your fingernail, loosen the cap that secures the ribbon cable to the controller PCB, as shown in Figure 147.

Figure 147. Loosening the ribbon cap on the controller PCB



Figure 148 shows a close-up view of the cap that secures the cable to the PCB. Figure 148. Cap around the ribbon cable (close-up view)



6. Pull the ribbon cable out of the cap, as shown in Figure 149.

Figure 149. Pulling the ribbon out of the cap



7. Using a 2.5 mm L- hex wrench, remove the lid where the ribbon cable connects to the autosampler tray (see Figure 150).

Figure 150. Removing the lid



8. Using a 2 mm hex wrench, remove the screws in the small PCB underneath the lid.

Figure 151 shows the screw in the lower right corner, and Figure 152 shows the screw in the middle of the left side.

Figure 151. Loosening the screw in the lower right corner



Figure 152. Loosening the screw in the middle left side



9. Using a 3 mm L-hex wrench, loosen the four hex screws underneath the adapter plate holder, and remove the holder. Figure 153 shows the location of these screws.

Figure 153. Screws beneath the adapter plate holder



Lift and pull the adapter plate holder out of the autosampler, as shown in Figure 154.
 Figure 154. Pulling the adapter plate holder out of the autosampler



Note The sleeves around the holes on the bottom of the adapter plate holder can fall out. Figure 155 shows these sleeves.

Figure 155. Hole sleeves on the bottom of the adapter plate holder

Sleeve that has fallen out of the hole

✤ To install the ASA cooler

1. Align the arms of the autosampler bed, as shown in Figure 156.

Figure 156. Aligning the arms of the autosampler bed



Carefully place the new cooler in the autosampler bed, as shown in Figure 157.
 Figure 157. Placing the new cooler in the autosampler bed



3. Verify that the cables from the fan are routed in the groove (see Figure 158).

Figure 158. Verifying the placement of the fan cables



- 4. Using a 3 mm L-hex wrench, insert and tighten the four hex screws underneath the adapter plate holder. Figure 153 on page 144 shows the location of these screws.
- 5. Using a 2 mm L-hex wrench, tighten the two screws in the small PCB. Figure 151 on page 143 and Figure 152 on page 143 show the locations of these screws.
- 6. Using a 2.5 mm L-hex wrench, mount the lid where the ribbon cable connects to the autosampler cooler (see Figure 150 on page 143).
- 7. Insert the ribbon cable into the controller board (see Figure 147 on page 142 and Figure 149 on page 142).
- 8. Using a flat screwdriver or your fingernail, push the cap around the ribbon cable in place (see Figure 148 on page 142).
- 9. Reattach the autosampler door (see Figure 144 on page 140).
- 10. Verify that the autosampler moves smoothly in and out by moving it manually.
- 11. Reconnect the power cable.
- 12. Turn on the EASY-nLC II instrument, and check the cooler temperature from the Home > Overview page.
- 13. Before using the instrument, install the vial/plate adapter and perform an autosampler calibration. For instructions, see Chapter 7, "Calibrating the Autosampler's XYZ Robot."

Replacing a Rotary Valve

The procedure for replacing a rotary valve is the same for the EASY-nLC 1000 instrument and the EASY-nLC II instrument.



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

Replacing a high-pressure valve requires these items.

Tools	Parts and materials
• #2 Phillips screwdriver	Powder-free gloves
• 1/4 in. open-ended wrench	Switching valve:
• Torx T-10 wrench	– EASY-nLC 1000 instrument: P/N LC519
• 2.5 mm L-hex wrench	-or-
• Flathead screwdriver	– EASY-nLC II instrument: P/N LC218

To replace a rotary valve, follow these steps:

- 1. "To remove a rotary valve," on the page
- 2. "To install a new rotary valve" on page 149

To remove a rotary valve

- 1. Close down the EASY-nLC instrument (see "Closing Down the EASY-nLC Instrument" on page 21), and then unplug the power cable.
- 2. Remove the computer box as described in "To remove the computer box" on page 108.

Note Removing the computer box requires the removal of the left side panel and the back panel.

- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 4. From the right side of the instrument (solvent system compartment), remove the tubing from the stator on the valve that you are replacing. Be careful to avoid pulling any fused silica lines, as there is limited space around the valves.

Tip When replumbing the system, follow the solvent system schematics in Appendix C, "Consumables and Replacement Parts."

5. Using a 2.5 mm L-hex wrench, remove the four 2.5 mm hex screws that keep the valve in place inside this compartment.

As you remove the last hex screw, the valve might slip down into the autosampler compartment.

6. From the back of the instrument, disconnect the cables shown in Figure 159 from the valve to be exchanged.

Figure 159. Cables connected to valves



7. Pull out the valve, as shown in Figure 160.

Figure 160. Pulling out a valve



Note If there is a support collar between the body of the valve and the body of the EASY-nLC instrument, move it to the new valve.

* To install a new rotary valve

- 1. Using a small flathead screwdriver, set the rotary switch on the new valve of the PCB, shown in Figure 161, to the same address as the valve being replaced:
 - Valve A=1
 - Valve B=2
 - Valve S=3
 - Valve W=4

Figure 161. Valve rotary switch



- 2. Insert the new valve, making sure that the valve PCB faces down.
- 3. In the back of the instrument, connect the cables shown in Figure 159 on page 148 to the new valve.
- 4. Using a 2.5 mm L-hex wrench, insert and tighten the four hex screws that keep the valve in place inside the autosampler compartment.
- 5. In the solvent system compartment, restore the tubing and connections to the stator on the valve that was replaced.
- 6. Reinstall the computer box as described in step 1 through step 5 of "To install the new computer box" on page 112.
- 7. Mount the left and right side panels to the instrument housing. Then, with a #2 screwdriver, secure each panel by rotating the three captive screws a quarter-turn clockwise.
- 8. Run the Flush Air script as described in "To run the Flush Air script" on page 34:
 - Enter 12μ L for a PLU pump (standard in the EASY-nLC 1000 instrument).
 - Enter 10 µL for a PLF pump (standard in most EASY-nLC II instruments).
- 9. Run the Leaks script for the subsystem that you have worked on as described in "To run the Leaks script" on page 46.
 - For valve A, select **A**.
 - For valve B, select **B**.
 - For valves W or S, select **System**.

Replacing a Pump

The procedure for replacing a pump differs between EASY-nLC 1000 instruments that contain PLU model pumps and EASY-nLC II instruments that contain PLF model pumps.

Note Because Thermo Fisher Scientific no longer stocks the PLF model pump as a replacement part, you must replace both pump models with the PLU model pump.

To replace a pump, follow the appropriate procedure:

- "Replacing a PLU Pump," on this page
- "Replacing a PLF Pump with a PLU Pump" on page 154



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

Replacing a PLU Pump

Replacing a PLU model pump requires these tools and materials.

Tools	Parts and materials
• #2 Phillips screwdriver	Powder-free gloves
• 1/4 in. open-ended wrench	• PLU model pump, P/N LC501
• 2.5 mm hex wrench	
• 3 mm hex wrench	
Small flathead screwdriver	
• Pipette for priming the pump	

To replace a pump in the EASY-nLC instrument, follow these steps:

- 1. "To remove a PLU model pump from an EASY-nLC instrument" on page 151
- 2. "To install a PLU pump in an EASY-nLC instrument" on page 153

* To remove a PLU model pump from an EASY-nLC instrument

- 1. Set the valve for the pump to position 1–6 as follows:
 - a. Press **Home > Overview**.
 - b. Press the appropriate valve icon.
 - c. In the Valve dialog box, press Center.

Placing the valve in the Center position shuts off the solvent flow to the subsystem.

- 2. Close down and then turn off the EASY-nLC instrument.
- 3. Disconnect the power cable from the instrument's back panel.
- 4. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 5. Using a 1/4 in. open-ended wrench, disconnect the stainless steel tubing connected to the pump head, shown in Figure 162.

Figure 162. Fitting on the pump head



- 6. Depending on the instrument model, do one of the following:
 - For the EASY-nLC 1000 instrument, unplug the flow sensor cable (from pump A or B), the pressure sensor cable, and the P-Bus communication cables, shown in Figure 163.





• For the EASY-nLC II instrument, unplug the pressure sensor cable and the P-Bus communication cables from the pump.

The EASY-nLC II flow sensors do not have a flow sensor cable that connects to the pump.

7. Using a 3 mm ball driver or L-hex wrench, remove the four 3 mm hex screws that secure the pump to the instrument panel, and then remove the pump. Set the screws aside.

* To install a PLU pump in an EASY-nLC instrument

- 1. Connect the new pump as follows:
 - a. Using a 3 mm ball driver or L-hex wrench and the four 3 mm hex screws that you removed in step 7 on page 152, secure the new pump to the instrument panel.
 - b. Depending on the instrument model, do one of the following:
 - For the EASY-nLC 1000 instrument, reconnect the flow sensor cable, the pressure sensor cable, and the P-bus communication cables.
 - For the EASY-nLC II instrument, reconnect the pressure sensor cable and the P-bus communication cables.
- 2. Make sure that the address setting on the pump PCB is the same as the setting for the old PCB. Use a small flathead screwdriver to change the rotary switch address on the new pump PCB.



- 3. Reconnect the power cable to the instrument's back panel.
- 4. Turn on the EASY-nLC instrument, and log in as an administrator.
- 5. Prime the pump as described in "Priming the Pump" on page 71.
- 6. Reconnect the tubing to the pump head. Using a 1/4 in. open-ended wrench, tighten the fitting.
- 7. Draw fresh solvent into the solvent lines and remove air from the pump head as described in "Removing Air After Replacing a Piston Seal or a Pump" on page 73.
- 8. Depending on the pump you are replacing, do the following:
 - When replacing pump A or B, run the pump leak test, and then run the flow sensor calibration (see "Test Leaks" on page 44 and "Calibrate Flow Sensors" on page 51).
 - When replacing pump S, run the sample pickup test (see "Test Sample Pickup" on page 42).

Replacing a PLF Pump with a PLU Pump

You must replace an irreparably damaged PLF pump in an EASY-nLC II instrument with a PLU pump, as Thermo Fisher Scientific no longer stocks the PLF pump as a replacement part.

When you replace a PLF pump with a PLU pump, do the following:

- Replace the pressure sensor for the pump and the high-pressure tubing associated with the pump, as the PLU pump is not compatible with the pressure sensor used in the EASY-nLC II instrument.
- Upgrade the touch-screen software to version 3.1.4 or later, as the EASY-nLC II instrument only supports the PLU pump when running software version 3.1.4 or later.
- If you are replacing pump S in an EASY-nLC II instrument with an earlier version of the chassis, order and install the components in the Pump Mounting Kit so that the PLU pump can fit into the pump S position.

Replacing an EASY-nLC II pump (PLF model) with a PLU pump requires these items.

Tools	Parts and materials
• #2 Phillips screwdriver	Powder-free gloves
• 1/4 in. open-ended wrench	• Pipette
• 2 mm hex wrench or ball driver	PLU pump, P/N LC501
	Pressure sensor, P/N LC502
• 2.5 mm hex wrench or ball driver	• Tubing between the pump and the pressure sensor:
	 Pumps A and S: PEEK tubing, P/N LC212
• 13 mm open-ended wrench	– Pump B: prebent stainless steel tube, P/N LC215
• Small flathead screwdriver	• Tubing between the pressure sensor and the rotary
• 3 mm open-ended wrench	valve:
· ····· · · · · · · · · · · · · · · ·	 Pumps A and S: PEEK tubing, P/N LC213
	– Pump B: prebent stainless steel tube, P/N LC216

* To replace a PLF pump with a PLU pump in the EASY-nLC II instrument

1. Upgrade the touch-screen software to version 3.1 or later.

If the touch-screen software is older than version 2.7.8, you must first upgrade to version 2.7.8, and then upgrade to version 3.1.

For information about upgrading the software, refer to Appendix B of the *EASY-nLC Series Getting Started Guide* (for the touch-screen software). You can find the touch-screen software files in the user zone on <u>www.proxeon.com</u>.

- 2. Before you remove the right side panel, do the following:
 - a. Set the corresponding valve to the **Center** position as described in "Using the Valve Controls" on page 204.
 - b. Close down the EASY-nLC II instrument, and then turn off the power switch on the instrument's back panel (see "Closing Down the EASY-nLC Instrument" on page 21).
 - c. Disconnect the power cable from the instrument's back panel.
- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.

Figure 164 shows the solvent line connections from pump A to valve A, which are made with PEEK tubing. The solvent line connections from pump B to valve B are made with stainless steel tubing.

Figure 164. Pump connections in an EASY-nLC II instrument (PEEK tubing for pump A)



4. Disconnect the subsystem tubing between the pump head and the high-pressure valve as follows:

Note The PLF pump can have an internally or an externally threaded pump head. The latest version of the PLF pump has an internally threaded pump head.

- a. Disconnect the tubing from the pump head as follows:
 - If a PEEK fitting is connected to the pump head, use a 13 mm open-ended wrench to remove it (see Figure 165).

Figure 165. Externally threaded pump head for a PLF model pump



• If a stainless steel fitting is connected to the pump head (see Figure 166), use a 1/4 in. open-ended wrench to remove it.

Figure 166. Internally threaded pump head for a PLF model pump



Stainless steel fitting connected to an internally threaded pump head
b. While holding the pressure sensor, use a 1/4 in. open-ended wrench to disconnect the tubing connected to the pressure sensor's inlet and outlet ports (see Figure 167).

Solvent lines connected to the pressure sensor

Figure 167. Disconnecting the solvent lines from the pressure sensor

- c. Using a 1/4 in. open-ended wrench, disconnect the other end of the tubing that is connected to the subsystem valve as follows:
 - For valve S, disconnect the tubing from port 6.
 - For valve A or B, disconnect the tubing from port 1.
- 5. Disconnect the pressure sensor assembly and the damaged PLF pump from the instrument panel as follows:
 - a. Using a 2.5 mm L-hex wrench or ball driver, remove the two hex screws that secure the pressure sensor L bracket to the instrument panel (see Figure 168). Then set the two 2.5 mm hex screws aside for reuse when you reconnect the L bracket to the instrument panel.
 - b. Using a 2 mm L-hex wrench or ball driver, unscrew the two hex screws that secure the pressure sensor to the L-bracket (see Figure 168). Then set the L-bracket and the two 2 mm hex screws aside for reuse when you mount the new pressure sensor.

Figure 168. Pressure sensor L-bracket screws



c. Disconnect the P-Bus communication cables from the pump actuator (see Figure 169).

Figure 169. Disconnected P-Bus communication cable



- d. Using a 3 mm ball driver or L- hex wrench, remove the four screws that secure the pump to the instrument panel (see Figure 170).
- e. Set the four 3 mm screws aside for reuse in step 8a on page 159.

Figure 170. Removing the four screws that secure the pump to the instrument panel

3 mm driver



- 6. Install the new pressure sensor as follows:
 - a. Using a 2.0 mm L-hex wrench or ball driver and the two 2.0 mm hex screws that you set aside when you disconnected the original pressure sensor from the L-bracket, connect the new pressure sensor to the L-bracket.
 - b. Using a 2.5 mm L-hex wrench or ball driver and the two 2.5 mm hex screws that you set aside when you removed the L-bracket from the instrument panel, reconnect the L-bracket to the instrument panel.
- 7. If you are replacing pump S in an early version of the EASY-nLC II instrument, install the components supplied in the Pump Mounting Kit as described in "To install the bracket and adapter plate provided in the Pump Mounting Kit" on page 160. Then continue this installation at step 9.

- 8. Connect the new pump as follows:
 - a. Using a 3 mm ball driver or L-hex wrench and the four 3 mm hex screws that you set aside when you removed the damaged pump from the instrument, secure the new pump to the instrument panel.
 - b. Connect the pressure sensor cable from the new pressure sensor to the receptacle on the right side of the PLU pump.
 - c. Connect the P-Bus communication cables.
- 9. Check the address setting for the rotary switch. If necessary, use a small flathead screwdriver to change the rotary switch address on the new pump PCB.



• Pump S = 3

- 10. Install tubing from the pump to the pressure sensor and from the pressure sensor to the valve. Use PEEK tubing for subsystems A and S. Use stainless steel tubing for subsystem B.
- 11. Reconnect the power cable to the instrument's back panel, and turn on the EASY-nLC II instrument.
- 12. Prime the new pump as described in "Priming the Pump" on page 71.
- 13. Remove air from the system as described in "Removing Air After Replacing a Piston Seal or a Pump" on page 73.
- 14. Make sure that both the right and left instrument panels have been installed and that the instrument has been turned on for at least 30 minutes.
- 15. Run the flow sensor calibration for the new pump (see "Calibrate Flow Sensors" on page 51).
- 16. If you replaced pump S, run the Sample Pickup script (see "Test Sample Pickup" on page 42).

The instrument is now ready for use.

* To install the bracket and adapter plate provided in the Pump Mounting Kit

Note Some earlier versions of the EASY-nLC II instrument cannot accommodate a PLU pump in the pump S position without modification.

To provide space for the pump outlet tubing in these instruments, you must install the new flow sensor bracket and the pump adapter plate supplied in the Pump Mounting Kit. The new flow sensor bracket places the flow sensors in a higher position and the pump adapter plate places the pump in a lower position than their original positions on the instrument panel. This repositioning makes space for the tubing that is connected to the pump head.

- 1. If you have not already done so, follow step 1 on page 154 through step 6 on page 158 of the previous procedure, "To replace a PLF pump with a PLU pump in the EASY-nLC II instrument," to upgrade the touch-screen software, gain access to the solvent system compartment, remove the damaged pump, and replace the pressure sensor.
- 2. Disconnect the tubing from the inlet and outlet ports of the flow sensors.
- 3. Replace the flow sensor bracket with the new flow sensor bracket supplied in the Pump Mounting Kit as follows:
 - a. Using a 2.5 mm hex wrench, remove the two screws that secure the flow sensor bracket to the instrument panel (see Figure 171).
 - b. Set the two 2.5 mm screws aside for reuse when you secure the new flow sensor bracket.

Figure 171. Removing the two screws that secure the flow sensor bracket to the panel



2.5 mm hex wrench

- c. Using a 2.0 mm hex wrench or ball driver, remove the four screws that secure the two flow sensors to the flow sensor bracket (see Figure 172).
- d. Set the 2.0 mm hex screws aside for reuse when you secure the flow sensors to the new flow sensor bracket.

Figure 172 shows the four 2.0 mm hex screws that secure the flow sensors to the flow sensor bracket and the original flow sensor bracket. Figure 173 shows the flow sensors removed from the flow sensor bracket. The flow sensor cables are still connected to the instrument.

Figure 172. Screws that secure the flow sensors to the bracket



Figure 173. Flow sensors removed from the flow sensor bracket



e. Using a 2.5 mm hex wrench and the screws that you set aside when you removed the flow sensors from the original flow sensor bracket, connect the flow sensors to the new flow sensor bracket supplied in the Pump Mounting Kit.

Note The mounting holes in the new flow sensor bracket are lower than the mounting holes in the original flow sensor bracket. When you connect the new bracket to the instrument panel, it is positioned higher than the original bracket, allowing more space for the tubing that is connected to the pump head.

Figure 174. New flow sensor bracket (with lower mounting holes)



- 4. Using a 2.5 mm hex wrench and the screws that you set aside when you removed the original flow sensor bracket from the instrument panel, connect the new flow sensor bracket to the instrument panel.
- 5. Reconnect the tubing to the inlet and outlet ports of the flow sensors. Be careful when installing the fused silica line to flow sensor B, as the end of the tubing is fragile.



CAUTION Take care when you connect the fused-silica line to the outlet port of flow sensor B. The end of the tubing is fragile and easily damaged.

6. Using a 2.5 mm L-hex wrench or ball driver and the four screws provided in the Pump Mounting Kit, secure the pump adapter plate to the instrument panel.

Figure 175. Securing the pump adapter plate to the instrument panel



Pump adapter plate

- 7. Install the PLU pump in the pump S position as follows:
 - a. Using a 3 mm ball driver or L-hex wrench and the four 3 mm hex screws that you set aside when you removed the damaged pump from the instrument panel, secure the new pump to the pump adapter plate.
 - b. Connect the pressure sensor cable from the new pressure sensor to the receptacle on the right side of the PLU pump.
 - c. Connect the P-bus communication cables.
- 8. To complete the pump installation, start from step 9 on page 159 of the previous procedure, "To replace a PLF pump with a PLU pump in the EASY-nLC II instrument."

Replacing a Pump PCB

The procedure for replacing the pump PCB differs between the PLU model pump and the PLF model pump.

Note The PLU pump is standard in the EASY-nLC 1000 instrument. The PLF pump is standard in most EASY-nLC II instruments. Figure 8 on page 9 shows these two pump models.

To replace a pump PCB, follow the appropriate procedure:

- "Replacing a Pump PCB in the PLU Pump," on this page
- "Replacing a Pump PCB in the PLF Pump" on page 167

Replacing a Pump PCB in the PLU Pump

Replacing the pump PCB on the PLU model pump requires these items.

lools .	Parts		
• #2 Phillips screwdriver	Pump PCB, P/N LC574		

- 2.5 mm hex wrench
- Small flathead screwdriver

To replace the pump PCB on the PLU pump, follow these steps:

- 1. "To remove a pump PCB from the PLU model pump," on this page
- 2. "To replace a pump PCB on the PLU model pump" on page 166

To remove a pump PCB from the PLU model pump

- 1. Close down the EASY-nLC 1000 instrument as described in "Closing Down the EASY-nLC Instrument" on page 21.
- 2. Disconnect the power cable from the instrument's back panel.
- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 4. Disconnect the P-Bus communication cables, the flow sensor cable, and the pressure sensor cable from the pump.

Figure 176 on page 164 shows these cables connected to the pump.

5. Using a 2.5 mm L-hex wrench or ball driver, loosen the four captive screws on the PCB shield (see Figure 176). Then remove the PCB shield (see Figure 177).

Figure 176. Four PCB shield screws (PLU model pump)



The four captive screws remain in place (see Figure 177). **Figure 177.** Removing the PCB shield (PLU model pump)



- 6. Slide the PCB forward and away from the four mounting studs.
- 7. Disconnect the motor cable on the back of the pump PCB (see Figure 178).

Figure 178. Motor cable connected to the pump PCB (PLU model pump)



* To replace a pump PCB on the PLU model pump

- 1. Using a small flathead screwdriver, reset the address on the rotary switch on the pump PCB (see Figure 179) as follows:
 - Pump A =1
 - Pump B = 2
 - Pump S = 3

Figure 179. PCB rotary switch



- 2. Plug the motor cable into the back of the PCB (see Figure 178 on page 165).
- 3. Insert the new pump PCB onto the pump.
- 4. Install the PCB shield.
- 5. Using a 2.5 mm L-hex wrench or ball driver, tighten the four 2.5 mm hex screws in the PCB shield.
- 6. Reconnect the P-Bus communication cables, the flow sensor cable, and the pressure sensor cable (see Figure 176 on page 164) to the pump.
- 7. Mount the right side panel to the instrument housing. Then, with a #2 screwdriver, secure the panel by rotating the three captive screws a quarter-turn clockwise.
- 8. Reconnect the power cable to the back panel of the instrument.
- 9. Turn on the instrument.
- 10. Wait for 30 minutes to allow the temperature in the housing to stabilize.
- 11. Calibrate the flow sensor by following the automated script described in "Calibrate Flow Sensors" on page 51.

Replacing a Pump PCB in the PLF Pump

Replacing a pump PCB (PLF model pump) in the EASY-nLC II instrument requires these items.

Tools	Parts
• #2 Phillips screwdriver	Pump PCB, P/N LC274
• 2.5 mm hex wrench	
Small flathead screwdriver	

To replace the PCB on a PLF pump, follow these steps:

- 1. To remove the pump PCB from the PLF model pump (EASY-nLC II instrument)
- 2. To install the pump PCB on a PLF pump (EASY-nLC II instrument)

To remove the pump PCB from the PLF model pump (EASY-nLC II instrument)

- 1. Close down the EASY-nLC II instrument as described in "Closing Down the EASY-nLC Instrument" on page 21.
- 2. Disconnect the power cable from the instrument's back panel.
- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 4. Disconnect the P-Bus communication cables (see Figure 169 on page 158) from the pump actuator.
- 5. Unscrew and remove the four 2.5 mm hex screws that connect the PCB to the pump.
- 6. Disconnect the pressure sensor grounding cable (yellow wire).
- 7. Gently slide the PCB outward and turn it around to gain access to its back side.
- 8. Disconnect the pressure sensor communication cable. Take care to avoid putting any stress on the cables.
- 9. Disconnect the motor cable. Again, take care to avoid putting any stress on the cables.
- 10. Remove the PCB.
- 11. Go to the next procedure "To install the pump PCB on a PLF pump (EASY-nLC II instrument)."

* To install the pump PCB on a PLF pump (EASY-nLC II instrument)

- 1. Remove the damaged PCB as described in the previous procedure, "To remove the pump PCB from the PLF model pump (EASY-nLC II instrument)" on page 167.
- 2. Using a small flathead screwdriver, reset the address on the rotary switch on the new pump PCB as follows:
 - Pump A =1
 - Pump B=2
 - Pump S=3
- 3. Reconnect the motor cable to the back of the PCB (see Figure 178 on page 165).
- 4. Mount the new PCB onto the pump.
- 5. Secure the PCB to the pump as follows:
 - Slide the four screws into the holes in the four corners of the PCB.
 - Slide the connector at the end of the pressure sensor grounding cable under the screw in the lower-right corner of the PCB.
 - Using a 2.5 mm hex wrench, tighten the four screws.

Figure 180. PLF pump with the pressure sensor cable connected





CAUTION To avoid damaging the PCB, make sure that the grounding connector does not touch any other components.

- 6. Reconnect the P-Bus communication cables.
- 7. Calibrate the flow sensor by following the automated script described in "Calibrate Flow Sensors" on page 51.

Troubleshooting

To troubleshoot problems with the EASY-nLC instrument, follow the troubleshooting tips and procedures in this chapter.

6

Contents

- Troubleshooting Tips
- Using the Direct Controls for Troubleshooting and Maintenance
- Troubleshooting a Pump that Fails the Flush Air Script
- Troubleshooting a Pump that Fails the Leaks Script
- Running a System Leak Test
- Troubleshooting the Results of the System Leak Test
- Identifying a Leaking Check Valve
- Troubleshooting a System Blockage
- Troubleshooting the Autosampler Aspiration and Calibration
- Checking Sample Pickup
- Troubleshooting Communication Problems
- Verifying that the LC/MS System Is Properly Grounded

Troubleshooting Tips

To troubleshoot problems with the EASY-nLC instrument, see these topics:

- "Autosampler Problems" on page 171
- "Contact Closure Problems" on page 172
- "Instrument Startup Problems" on page 172
- "Delayed Elution" on page 173
- "Excessive Duration or Higher Pressure for the Column Equilibration and Sample Loading Steps" on page 178
- "System Reaches Its Maximum Pressure During the Gradient" on page 180
- "Sample Signal Weak or Absent" on page 183
- "Slow or No Pressure Increase in Subsystem A or B" on page 187
- "Errors Reported by the Xcalibur Data System" on page 189
- "Carryover" on page 194
- "Spray Issues" on page 196
- "Chromatographic Performance" on page 197
- "Device Failures" on page 198
- "Miscellaneous" on page 202

Autosampler Problems

Table 9 lists troubleshooting tips for the autosampler.

Note For information about troubleshooting sample pickup, see "Troubleshooting the Autosampler Aspiration and Calibration" on page 261.

Table 9. Autosampler troubleshooting tips

Symptom	Possible causes	Action
The needle cannot	The needle tip is damaged or bent.	Do the following:
penetrate the plastic film on the microtiter		1. Check the software configuration for the adapter tray.
plate.		2. Eject the tray.
		3. Remove the left side panel and visually inspect the needle tip.
		4. Do one of the following:
		• If the needle tip is bent or damaged, replace it (see "Replacing the Autosampler Needle" on page 84).
		• If there is any residue on the needle tip, wipe the needle tip with a lint-free tissue soaked in LC/MS-grade ethanol.
XYZ robot error	Possible step loss on one of the motors because the movement of the autosampler's XYZ arm is blocked The error repeats without any visible reason.	Remove the obstacle and try again. You might need to calibrate the autosampler for more precise penetration of the microtiter plate or the wash bottles.
		For information about calibrating the autosampler, see Chapter 7, "Calibrating the Autosampler's XYZ Robot."
		Contact Thermo Fisher Scientific:
		• us.customer-support.analyze@thermofisher.com
		 eu.techsupport.cms@thermofisher.com
A server warning message about the plate	The plate format has been deleted from the system configuration. For information about deleting a plate format, p refer to the <i>EASY-nLC Series</i> <i>Getting Started Guide</i> .	If you want to run the batch, you must create the plate format again.
autosampler configuration pops up when you submit a batch.		For information about selecting, creating, and deleting plate formats, see "Managing Plate Formats" on page 274

Contact Closure Problems

Table 10 contains troubleshooting tips for contact closure problems.

Table 10.	Contact closure	troubleshooting tips
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Symptom	Possible causes	Action
The mass spectrometer is waiting for a contact closure signal.	Incorrect contact closure setup	If you are running two-way contact closure (with feedback from the mass spectrometer), set the contact closure Protocol setting to Two-way on the Configuration > Connections page.
		If the mass spectrometer is waiting for a signal, set the contact closure State setting to Open .
The EASY-nLC instrument is waiting for a ready signal from the mass spectrometer.		If you are running one-way contact closure (that is, the EASY-nLC instrument sends a start signal to the mass spectrometer, but feedback from the mass spectrometer is ignored), set the contact closure Protocol setting to One-way on the Configuration > Connections page.

Instrument Startup Problems

Table 11 contains troubleshooting tips for instrument startup problems.

Table 11. Startup troubleshooting tips

Symptom	Possible causes	Action
Dark monitor	Broken or open main power fuse	1. Turn on the power to the instrument.
		2. Remove the right side panel of the EASY-nLC instrument.
		3. If there is no light on the pumps, turn off the EASY-nLC instrument, replace the right side panel, and check the fuse.
	Defective monitor	Attach an external monitor to the VGA connector (labeled MONITOR) on the back panel of the instrument (see Figure 16 on page 16).
Startup bar is no longer proceeding.	Damaged instrument	Turn off the EASY-nLC instrument, wait 5 seconds before turning it on again, and then wait approximately 10 minutes for the restart process to finish. In some cases this recovers the system. Contact Thermo Fisher Scientific if the problem recurs:
		• us.customer-support.analyze@thermofisher.com
		• eu.techsupport.cms@thermofisher.com

Delayed Elution

The most common cause of delayed elution is a worn rotor seal in valve B, causing valve B to leak.

Table 12 lists the common causes of delayed elution. These topics show the effect of leaks and swept volume on the pressure and flow traces:

- "Leaks that Cause Delayed Elution" on page 174
- "Introduced Swept Volume" on page 176

Table 12. Common causes of delayed elution

Symptom	Possible cause	Action
Delayed elution	Leak in the solvent system	1. Run the Leaks script for the system (see "Test – Leaks" on page 44).
		2. Locate the leak by following "Troubleshooting the Results of the System Leak Test" on page 221.
	Incorrect tubing installed	1. Remove the right side panel of the instrument.
		2. Verify that the tubing matches the solvent system schematic.
		• For the EASY-nLC II instrument, see page 314.
		• For the EASY-nLC 1000 instrument, see page 316.
		3. Replace the incorrect tubing with the specified tubing.
	Swept volume introduced	Check the tubing connections.
	connections	In the EASY-nLC II instrument, you can inadvertently introduce swept volume (extra volume in the solvent path) by not seating the tubing against the bottom of the receiving port.

Leaks that Cause Delayed Elution

When there is a leak in the system upstream from flow sensor B, part of the solvent stream from pump A exits the mixing Tee toward flow sensor B, pushes solvent B back toward valve B, and partly fills the solvent line between the mixing Tee and flow sensor B during the column equilibration and sample loading steps (see Figure 181).

When the gradient step begins, instead of pushing solvent B into the mixing Tee, pump B initially pushes this small volume of solvent A, delaying the start of the actual gradient.

Figure 181. Negative flow of solvent B toward valve B during the column equilibration and sample loading steps



Figure 182 shows the flow and pressure traces for pump B for a system with a leak upstream of flow sensor B. The measured flow rate is negative rather than 0 nL/min during the column equilibration and sample loading steps.

Figure 182. Flow trace for pump B when there is a leak upstream of flow sensor B



column equilibration and sample loading steps

Introduced Swept Volume

Normally when solvent B (acetonitrile with 0.1% formic acid) reaches the column, the back pressure measured by pressure sensor A drops to less than one-half the amount measured for solvent A (water with 0.1% formic acid) alone. If the back pressure does not drop by more than 50 percent when solvent B reaches the column, the solvent system might contain additional swept volume introduced by poor tubing connections.

The additional swept volume increases the gradient delay volume so that the gradient reaches the column later.

Figure 183 shows the pressure dip that occurs when the gradient starts to reach the column. Figure 184 shows the pressure dip for the sample chromatographic method, but occurring two minutes later due to the added gradient delay volume.



Figure 183. Normal pressure dip for pressure sensor A as the gradient reaches the column

The pressure starts to drop at approximately 25 minutes as 100% solvent B reaches the column.



Figure 184. Pressure dip begins later when the system has introduced swept volume

Back pressure is proportional to the viscosity of the solvent mixture. If you are not running a water/acetonitrile gradient, the pressure profile of your gradient run will differ. Table 31 on page 311 lists the viscosities for two-solvent mobile phases consisting of a water/methanol mixture or a water/acetonitrile mixture.

reaches the column.

Excessive Duration or Higher Pressure for the Column Equilibration and Sample Loading Steps

Table 13 lists possible causes for the following:

- When the column equilibration and sample loading steps are controlled by a specified pressure, the duration of these steps is longer than expected.
- When these steps are controlled by a specified flow, the pressure is higher than expected during the column equilibration step, the sample loading step, or both steps.

During the sample loading and column equilibration steps, pump A delivers the user-specified volume of solvent A to the system at the user-specified flow rate or maximum pressure or the maximum system pressure. When the columns or the Column Out line is clogged, the system must lower the flow rate to maintain the pressure at the maximum pressure specified in the method. The lower flow rate increases the column equilibration time, the sample loading time, or both.

Table 13. Common causes of a longer than expected column equilibration time or sample loading time

Symptom	Possible cause	Action
When the method specifies a set flow rate for the column	Clogged column	Run the Back Pressure script for pump A (see "Test – Back Pressure" on page 48).
equilibration, sample loading, or both of these steps, the pressure is higher than usual.		If the test passes, one of the columns is probably clogged and must be replaced.
When the method specifies a	Clogged system	Run the Back Pressure script for pump A.
maximum pressure for one or both of these steps, the duration of these steps is longer.		If the test fails, go to "Troubleshooting a System Blockage" on page 249.

Figure 185 and Figure 186 show the flow rate and pressure traces for pump A for a normal run and a run with a partially clogged column, respectively. The flow rate for a run with a clogged column is lower than the flow rate for a normal run. The lower flow rate increases the duration of the column equilibration and sample loading steps.

Note The specifications for the precolumns used to acquire the data in Figure 185 and Figure 186 are as follows:

- 2 cm length
- 100 µm ID
- 5 µm particle size
- C18



Figure 185. Normal run





System Reaches Its Maximum Pressure During the Gradient

If the pressure trace shows maximum pressure during the gradient, review the possible causes in Table 14.

Table 14. Possible causes of maximum pressure spikes during a run

Symptom	Possible cause	Action
The system reaches its	Clogged column	Run the Back Pressure script for pumps A and B.
maximum pressure during a run.		If the test passes, one of the columns is probably clogged and needs to be replaced.
	Clogged system	Run the Back Pressure script for pumps A and B.
		If the test fails, go to "Troubleshooting a System Blockage" on page 249.
	Excessively high flow rate	Reduce the flow rate in the method.

When a column or the Column Out line is clogged, the back pressure can increase to the maximum system pressure for pump A, pump B, or pumps A and B during the gradient (see Figure 187).

Figure 187. Back pressure shown reaching the maximum system pressure for pumps A and B



When only the flow path between pump B and the mixing Tee has a blockage, the back pressure on pump B increases and differs from the back pressure on pump A.

Figure 188 shows the typical profile for a blockage in the solvent system between pump B and the mixing Tee.

Figure 188. Back pressure shown reaching the maximum pressure for pump B only



Figure 189 shows the flow path between pump B and the mixing Tee.

Figure 189. Solvent lines between pump B and the mixing Tee



No.	EASY-nLC II instrument	EASY-nLC 1000 instrument
1	Stainless steel tubing and fittings	Stainless steel tubing and fittings
2	Stainless steel tubing and fittings	Stainless steel tubing and fittings
3	PEEKsil tubing	nanoViper
	• Stainless steel fitting connected to port 6 of valve B	
	• PEEK fitting connected to the flow sensor inlet	
4	Fused-silica solvent line	nanoViper
	• PEEK fitting connected to the flow sensor outlet	
	PEEK fitting connected to the mixing Tee	

Sample Signal Weak or Absent

If the sample signal is missing or weak, review the troubleshooting tips in Table 15.

Table	15.	Possible	causes	of a	weak	or	absent	signa
lane	IJ.	LO22IDIG	Causes	UI d	WEak	UI	ansem	Signo

Symptom	Possible cause	Action
The sample signal is missing or weak.	Sample not aspirated (See Figure 190 on page 184.)	Run the Sample Pickup script. If the Sample Pickup script fails, see "Troubleshooting the Autosampler Aspiration and Calibration" on page 261.
	Damaged or incorrect column (See Figure 191 on page 185.)	Make sure that you have installed the correct column or columns.
		Check the back pressure on the columns by running the column equilibration scripts.
	Leak in the system	Run the Leaks test on the system.
	(See Figure 192 on page 186.)	If the test fails, see "Troubleshooting the Results of the System Leak Test" on page 221.
	Incorrect column configuration	Check the column configuration.
		1. Press Maintenance > Devices.
		 In the Devices list, select EASY-nLC (HPLC).
		3. Press the Properties tab.
		4. Clear the One Column Setup check box for a two-column setup or select this check box for a one-column setup.
		For more information, refer to the <i>EASY-nLC</i> Series Getting Started Guide.
	Incorrect solvent in W3 bottle	Make sure that the W3 bottle contains solvent A.
	Incorrect sample loop configuration	Check the loop configuration on the Properties page for the EASY-nLC HPLC device on the Maintenance > Devices page.
		Make sure that the configured loop volume matches the installed sample loop.

Weak or Absent Signal—Sample Not Aspirated

A pressure drop at the start of the sample loading step indicates that pump S has introduced air into the sample loop (see Figure 190).

When the method uses pressure to control the flow rate, the pressure should return to the specified level within approximately 4 seconds after the sample is loaded onto the column. If the pressure does not return to the specified level within approximately 4 seconds, pump S has probably introduced air into the system.



Figure 190. Typical pump A profile for a system with air in the sample loop

The measured flow rate

Weak or Absent Signal—Defective Precolumn

When compared to the Pump A graph for a normal run shown in Figure 185 on page 179, Figure 191 shows the reduced time for the precolumn equilibration and sample loading steps when the precolumn is damaged and has lost a significant portion of its packing material.

The flow rate for the column equilibration and sample loading steps is higher (>15 μ L/min) and the duration is shorter because the precolumn is exerting less back pressure.

Note The specifications for the precolumns used to acquire the data in Figure 185 and Figure 186 are as follows:

- 2 cm length
- 100 µm ID ٠
- 5 µm particle size ٠
- C18

Figure 191. Typical pump A profile for a damaged precolumn



Column equilibration step

Weak or Absent Signal—Leak Upstream of Flow Sensor B

Figure 192 shows a typical pump B profile for a system with a leak upstream of flow sensor B. Flow sensor B displays a negative flow during the column equilibration and sample loading steps.

If the gradient is short, a leak upstream of flow sensor B will cause a complete lack of signal because solvent B is not reaching the columns within the duration of the gradient.



Figure 192. Typical pump B profile for a leak upstream of flow sensor B

Slow or No Pressure Increase in Subsystem A or B

A lack of pressure or a slow increase in pressure in subsystem A or B is often caused by a worn piston seal, a worn rotor seal, or a leaking tubing connection.

Note A subsystem includes the pump, the solvent line that connects the pump to the pressure transducer, the solvent line that connects the pressure transducer to the valve, and the valve.

Table 16. Troubleshooting low back pressure in subsystems A or B

Symptom	Possible cause	Action
Back pressure is low in subsystem A or B.	Air in the pump	Run the Purge Solvent script for the affected pump with 2 iterations. Then, run the Flush Air script. If the Flush Air script fails, see "Troubleshooting a Pump that Fails the Flush Air Script" on page 212.
	Leak in the pump	Run the Leaks script for the affected pump. If the Leaks script fails, see "Troubleshooting a Pump that Fails the Leaks Script" on page 216.

Figure 193 shows the leak test profile for pump A. Even when the flow rate is set to the maximum allowable value, the pressure for pump A does not increase.



Figure 193. Leak test profile for pump A

Extremely low pressure

When subsystem A has a large leak, pump A can run out of solvent during a sample run, resulting in the server error message shown in Figure 194.

Figure 194. Server error message displayed when pump A runs out of solvent

Server	error message(s)
5/	/26/11 11:48 AM
	Issue code 11: Pump A stopped (out of solvent).
8	Reported by: Pump A

When subsystem B has a large leak or is filled with air, the gradient fails to start because the pressure cannot be increased to reach the starting conditions (see Figure 195).

Figure 195. Subsystem B leak



When pump B cannot generate sufficient pressure to start the gradient, the server error message shown in Figure 196 appears.

Figure 196. Server error message displayed when pump B cannot generate sufficient pressure to start the gradient

Server error message(s)				
	20/07/11 11:19 Issue code 87: Pump B unable to reach target pressure within 50 µl. Please check for air/leaks! Reported by: Business logic IFC 4.0 Close			

Errors Reported by the Xcalibur Data System

Table 17 lists the possible causes of the common errors reported by the Xcalibur data system.

Table 17. Possible causes of errors reported by the Xcalibur data system (Sheet 1 of 2)

Symptom	Possible cause	Action
An Acquisition Server failure prevents the sequence from running (see Figure 197 on page 190).	Lost connection between the EASY-nLC instrument and the external data system computer	Check the Ethernet connections between the EASY-nLC instrument and the data system computer.
		Exit the Xcalibur data system. Then, open the Thermo EASY-nLC Configuration dialog box and click Test Connection .
		If necessary, restart the EASY-nLC system.
The Acquisition Server displays the message that the EASY-nLC device has reported a failure during the Prepare For Run step.	Injection volume is greater than the loop size minus 2 µL (See Figure 198 on page 191.)	Check the loop configuration on the Properties page for the EASY-nLC HPLC device on the Maintenance > Devices page. Make sure that the configured loop volume matches the installed sample loop.
	Lowercase letter used in the sequence table for vial position (See Figure 199 on page 191.)	Change the letters to capitals.
Unable to submit samples. The following error message appears: Invalid Autosampler Vial Position	Incorrect plate configuration (See Figure 200 on page 192.)	Select the correct autosampler plate in the Thermo EASY-nLC Configuration dialog box.

 Table 17. Possible causes of errors reported by the Xcalibur data system (Sheet 2 of 2)

Symptom	Possible cause	Action
The mass spectrometer remains in the Waiting for Contact Closure state and data acquisition does not start (see Figure 201 on page 193).	Contact closure configured incorrectly	Press Configuration > Connections . Select the correct MS configuration and settings.
	Failing contact closure relay due to incorrect grounding	Check accordingly to "Verifying that the LC/MS System Is Properly Grounded" on page 268.

Figure 197 shows the Acquisition Server message that appears when the EASY-nLC instrument loses communication with the data system computer during a sequence run. The sequence pauses until you intervene.

Figure 197. Lost connection between the EASY-nLC instrument and the Acquisition Server



Figure 198 shows the Acquisition Server message that appears during the Prepare for Run step when the requested injection volume is greater than the configured loop size minus 2 μ L.

Figure 198. Incorrect injection volume



Figure 199 shows the Acquisition Server message that appears when you incorrectly use a lowercase letter to define the vial or well position. For the EASY-nLC device, you must use capital letters in the alphanumeric description of the vial or well position.

Figure 199. Incorrect vial position nomenclature

Incorrect use of a lowercase letter



Figure 200 shows the Home Page error message that appears when the sequence list contains a well plate position and the autosampler configuration is set up for the vial adapter plate format.

Figure 200. Incorrect vial position for the vial adapter plate format


Figure 201 shows that the mass spectrometer is waiting for a start signal from the EASY-nLC device before it starts data acquisition. The mass spectrometer remains in the Waiting for Contact Closure state until it receives the start signal. The EASY-nLC instrument sends the start signal when the run gradient step begins.



Figure 201. Waiting for Contact Closure error

Carryover

Table 18 lists the possible causes of carryover.

Table 18. Possible causes of carryover

Symptom	Possible cause	Action
Carryover from one run to the next	Incomplete elution of peptides from column	Add extra time to the end of the gradient or add a high-organic hold to the end of the gradient.
		To determine if the high-organic hold is long enough, make sure that you see a pressure drop during the hold period (see Figure 202 and Figure 203).
	Insufficient washing of needle and loop	Add extra wash steps to the method. Consider using an organic solvent for one of the custom wash solvents.
	Dead volumes in the column setup	Remake the connections in the column setup.

Figure 202 shows a gradient run with a high-organic hold at the end of the run that is too short for the column to see the high-organic solvent, as displayed by the lack of a pressure drop at the end of the run.



Figure 202. Gradient run without an adequate high-organic hold at the end of the run

The gradient reaches

Figure 203 shows the effect of a high-organic hold at the end of the gradient run on pressure. The pressure continues to drop until the column equilibrates to the high-organic content.

Figure 203. Gradient run with an adequate high-organic hold at the end of the run



The pressure begins to drop when the high-organic mobile phase reaches the column.

Spray Issues

Table 19 lists the possible causes of an unstable spray.

Table 19. Possible causes of an unstable spray

Symptom	Possible cause	Action
Unstable spray	Post-column outgassing	Degas the solvents.
	Poor connection in either the liquid junction or the DirectJunction	Check all of the connections.
	Incorrect MS settings or emitter position	Optimize the MS tune file for the desired flow rate and the emitter position.
	Dirty emitter	Clean the emitter with an organic solvent such as methanol, acetonitrile, or ethanol.
		Use only LC/MS-grade solvents.
	Poor high-voltage connection	Check all of the high-voltage connections. Reconnect as necessary.
	Air movement from an air conditioning vent	Deflect the air from the vent, or use a shield to protect the ion source.

Chromatographic Performance

Table 20 lists troubleshooting tips for common chromatography problems.

Table 20. Troubleshooting tips for chromatographic performance

Symptom	Possible cause	Action
Broad peaks	Dead volume between the	Check the connections.
		Replace the fittings if needed.
	Damaged column	Inspect the column, especially the outlet.
		Replace the column if it is damaged.
Poor retention time reproducibility	Temperature fluctuations in the	Monitor the laboratory temperature.
	laboratory	Stabilize the temperature by keeping the
		laboratory doors closed and by ensuring that
		the air conditioning system is kept constant
		at all times.
		Do not run the EASY-nLC instrument with
		the panels removed.
	Back flow on flow sensor B	Run the Leaks script for Pump A, Pump B, and System.
		Replace defective piston seals, rotor seals,
		and solvent lines as indicated.
	Spikes seen in flow sensor B	Use the scripts to check for a leak in the B
	during valve shifts	subsystem (Leak test Pumps A + B, and System).
		Replace defective seals, rotors, and solvent lines as needed.

Device Failures

Table 21 lists troubleshooting tips for devices failures.

 Table 21.
 Troubleshooting tips for device failures

Symptom		Possible cause	Action
The pump icon for a pump that is not functioning turns a pink color on the Home > Overview page. See Figure 204 on page 199.	? bar ? nl/min ?/140.0 μl	Communication failure to the pump. Could be the pump or the PC BUS cable.	Check that the cables are connected correctly.
The valve icon for a valve that is not functioning turns a pink color on the Home > Overview page. See Figure 205 on page 200.	A	Communication failure to the valve. Could be the valve or the PC BUS cable	Check that all of the cables are connected correctly.
The autosampler icon turns a pink color on the Home > Overview page. See Figure 206 on page 201.		Communication failure on the cooling unit. Could be the PC box, a damaged cable, or a loose connection.	Check that all of the cables are connected correctly.

A pump icon turns pink when the pump loses communication with the EASY-nLC computer. The pumps are serially connected, so if pump B loses communication, all of the pumps lose communication. Pump B is the first pump in the series.

Communication between the pumps and the embedded computer can be lost when the cables are loose, when the embedded computer fails, or when the pump PCB fails.

Figure 204 shows the pink pump icons on the Home > Overview page.



Figure 204. Pink pump icons on the Home > Overview page

A pink valve icon means that the communication to the unit has been lost. Be aware that the valves are serially connected, so a failing valve might cause other valve icons to turn pink as well.

Communication between the valves and the embedded computer can be lost when the cables are loose, when the embedded computer fails, or when the valve PCB fails.

Figure 205 shows the pink valve icons on the Home > Overview page.

Figure 205. Pink valve icons on the Home > Overview page



An autosampler icon turning pink means that the communication to the unit has been lost.

Figure 206 shows the pink autosampler icon on the Home > Overview page.

Figure 206. Pink autosampler icon on the Home > Overview page



Miscellaneous

For additional issues that might arise while using the EASY-nLC instrument, consult these miscellaneous troubleshooting tips (Table 22).

Table 22. Miscellaneous troubleshooting tips (Sheet 1 of 2)

Symptom	Possible causes	Action
Copying log files to a USB memory stick failed.	Memory stick in wrong format	Format the memory stick in FAT/FAT16 format.
	Memory stick not recognized by the instrument	Use one of the memory sticks supplied by Thermo Fisher Scientific, or try plugging the stick into the other USB port.
		Contact Thermo Fisher Scientific if neither action works: us.customer-support.analyze @thermofisher.com
		eu.techsupport.cms @thermofisher.com
Copying to the USB memory stick does not stop.	Too much data to be copied	Remove the memory stick from the EASY-nLC instrument and press Ignore when the error message appears.
		Contact Thermo Fisher Scientific for an
		instrument check-up:
		us.customer-support.analyze @thermofisher.com
		eu.techsupport.cms @thermofisher.com
Forgot the admin password.		If you are already logged in to the EASY-nLC instrument, Thermo Fisher Scientific can extract the password from the system in two ways:
		 If the network is set up for remote access, press Maintenance > Support and press Connect.
		-or-
		• Run a factory reset.
System is running slower and slower.	Memory filled with graph data from long gradient run	Press Home > Graph and disable the graphs by pressing No Graph in each graph window.
	System overload caused by other reasons	Restart the application or power down/power up from Maintenance > Scheduling .

Possible causes		
n position The valve' s dut calibration.	Check script (see "Test – Valve ge 47).	
	only valid for EASY-nLC II ves with a serial number less th	an
	ocal Thermo Fisher Scientific fi er if the reported overshoot is	eld
or SVG Program error in graphics) error release	ons you can continue your wor nore or OK. More serious error ht require restarting the HPLC	k
	the software quality, e-mail det	ails
	ipport.analyze	
	com	
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	com	
	the remote support server. See he EASY-nLC Instrument to the " on page 288.	ıe
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Table 22. Miscellaneous troubleshooting tips (Sheet 2 of 2)

Using the Direct Controls for Troubleshooting and Maintenance

You can access the direct controls for the instrument's hardware components by pressing the component icons on the Home > Overview page.

These topics describe how to use the direct controls:

- Using the Valve Controls
- Using the Pump Controls
- Using the Autosampler Controls

Using the Valve Controls

You can change the positions of the instrument's three-position valves from the Home > Overview page. For information about the valve positions, see "Six-Port Rotary Valves" on page 12.

To change the position of a valve

1. On the Home > Overview page, press the valve icon for the valve that you want to control.

The Valve dialog box opens (see Figure 207). The readback box lists the current valve position.

Figure 207. Valve dialog box



2. Press the button for the position that you want the valve to switch to: 1–2, **Center**, or 1–6.

The readback box displays the new position.

Using the Pump Controls

You can refill and empty the pumps from the Home > Overview page.

IMPORTANT Use the pump's direct controls for troubleshooting. To avoid contaminating the pumps, take care when you use these controls:

- For pump A or B, make sure that its corresponding valve is in position 1–2 before you refill the pump.
- For pump S, before you empty or fill the pump, use the autosampler's direct controls to insert the autosampler needle into the W4 or W3 bottle, respectively.

Tip To prepare the EASY-nLC instrument for operation, use the Prepare – Purge Solvent script to flush the solvent lines (see "Prepare – Purge Solvent" on page 32). The Purge Solvent script places the valves in the appropriate positions for filling and emptying pumps A and B and places the autosampler needle in the appropriate wash and waste bottles for filling and emptying pump S.

To work with the pump's direct controls, follow these procedures:

- Opening the Direct Controls for the Pumps
- Using the Direct Controls To Fill or Empty Pump A or B
- Using the Direct Controls To Fill or Empty Pump S

Opening the Direct Controls for the Pumps

Each pump is represented by a pump icon on the Home > Overview page.

✤ To open the pump dialog box

On the Home > Overview page, press the pump icon for the pump that you want to control.

Its dialog box opens (see Figure 208). The Position readback box displays the current pump position from 0 μ L (empty) to 140 μ L (full).





Note Because pump S does not have an associated flow sensor, the Pump S dialog box does not include a Real Flow readback box.

Using the Direct Controls To Fill or Empty Pump A or B

Before using the direct controls to fill or empty pump A or B, make sure that the valves are in the appropriate position. For information about controlling the position of the valves, see "Using the Valve Controls" on page 204.

IMPORTANT Before you fill pump A or B, make sure that the pump's associated valve is in position 1–2. You can contaminate the pump by attempting to fill the pump when its associated valve is in position 1–6.

* To fill pump A or B with solvent

- 1. On the Home > Overview page, check the position of the valve that controls the solvent flow direction for the pump of interest.
- 2. Make sure that the valve is set to position 1–2.

When valve A or B is in the 1-2 position, the corresponding pump draws solvent from the solvent bottle on top of the instrument (see Figure 209).



Figure 209. Position of the rotary valve when filling a pump

3. Press the pump icon for the pump you want to control.

The Pump dialog box opens (see Figure 208).

4. In the flow rate box, enter the filling flow rate: -0.01 to -300μ L/min.

For best results, enter a flow rate of $-100 \,\mu$ L/min to fill the pump.

- 5. In the volume box, enter the volume that you want pump A or B to draw.
- 6. Press Start.

The pump draws solvent until it draws the requested volume or reaches the full position, whichever occurs first.

* To empty pump A or B to waste

- 1. Check the position of the pump valve on the Home > Overview page.
- 2. Do one of the following:
 - To empty pump A or B directly to waste, make sure that its valve is in position 1–2.
 - To empty pump A or B to waste by way of valve W, place the pump valve in position 1–6 and valve W in position 1–6.

Valve	Position	Solvent flow	
A or B	1–2	Empties to the waste beaker.	
	1–6	Passes through valve S and into the Column Out line.	
S	1–2	Bypasses the sample loop.	
	1–6	Passes through the sample loop.	
W	1–2	Passes through the installed columns.	
	1–6	Exits the Column Out line to waste for a one column setup or passes through the precolumn and then exits the system to waste for a two-column setup.	

3. Press the pump icon for the pump you want to control.

The Pump dialog box opens (see Figure 208 on page 206).

4. In the flow rate box, enter the flow rate: **0.01** to **300** μ L/min.

When valve A or B is in position 1–6, the maximum system pressure limit constrains the maximum flow rate.

- 5. In the volume box, enter the volume that you want the pump to empty to waste.
- 6. Press Start.

The pump empties until it pumps the requested volume or reaches the empty position, whichever occurs first.

Using the Direct Controls To Fill or Empty Pump S

Before using the direct controls to fill or empty pump S, make sure that the autosampler needle is inserted into the appropriate wash or waste bottle.

IMPORTANT To avoid contaminating pump S, take care to place the autosampler needle in the appropriate wash bottle before filling the pump.

✤ To fill pump S

- 1. Make sure that a wash bottle with the appropriate solvent is installed in position W3.
- 2. Send the autosampler needle to the appropriate position as follows:
 - a. On the Home > Overview page, press the autosampler icon.

The Autosampler dialog box opens (see Figure 210 on page 210).

b. Press W3.

The autosampler needle moves to the W3 position.

c. Press Needle Down.

The autosampler needle descends into the W3 bottle.

- d. Press **Close** to close the Autosampler dialog box.
- 3. Draw solvent into pump S as follows:
 - a. Press the pump S icon.

The Pump S dialog box opens.

b. In the flow rate box, enter the filling flow rate: -0.01 to $-300~\mu\text{L/min}.$

For best results, enter a flow rate of -40μ L/min to fill pump S.

- c. In the volume box, enter the volume that you want pump S to draw.
- d. Press Start.

The pump draws solvent until it draws the requested volume or reaches the full position, whichever occurs first.

✤ To empty pump S to waste

- 1. Make sure that a wash bottle with a needle wash insert is installed in position W4.
- 2. Send the autosampler needle to the appropriate position as follows:
 - a. On the Home > Overview page, press the autosampler icon.

The Autosampler dialog box opens (see Figure 210 on page 210).

b. Press W4.

The autosampler needle moves to the W4 position.

c. Press Needle Down.

The autosampler needle descends into the W4 bottle.

- d. Press **Close** to close the Autosampler dialog box.
- 3. Dispense solvent from pump S as follows:
 - a. Press the pump S icon.

The Pump S dialog box opens.

- b. In the flow rate box, enter the flow rate: 0.01 to $300 \,\mu\text{L/min}.$
- c. In the volume box, enter the volume that you want the pump to empty to waste.
- d. Press Start.

The pump empties until it pumps the requested volume or reaches the empty position, whichever occurs first.

Using the Autosampler Controls

You can control the XYZ robot and the cooler temperature from the Home > Overview page.

To open the Autosampler dialog box

Press the autosampler icon on the Home > Overview page.

The Autosampler dialog box opens (see Figure 210).

Figure 210. Autosampler dialog box

Autosampler	
XYZ Robot	
8x12/Abgene (AB-0800) Goto Calibration	
Goto Well Needle Up Needle Down	Vial or well position box
Wash W1 W2 W3 W4	
Cooler	Temperature input box
7.0 ℃ Set Disable Temperature: 6.7 ℃	Temperature readback box
Close	

* To start the calibration procedure for the current plate format

1. Press Goto Calibration.

The Tools view for the autosampler on the Maintenance > Devices page opens.

2. To calibrate the XYZ robot for the plate format, follow the instructions in "Calibrating Plates" on page 280.

* To move the needle to a vial or well position

- 1. In the Vial or Well box, enter a vial or well position.
- 2. Press Goto.
- ✤ To move the needle to a wash bottle position

Press W1, W2, W3, or W4.

***** To change the cooler temperature

- 1. Enter a temperature in the temperature input box.
- 2. Press Set.
- 3. Monitor the temperature change in the temperature readback box.

Troubleshooting a Pump that Fails the Flush Air Script

If pump A, B, or S fails the Flush Air script and the Output page displays one of the following messages, a leak or blockage could be preventing solvent from entering the pump.

- Aborting unable to build pressure
- Aborting check solvent level

Note For information about the Flush Air script, see "Prepare – Flush Air" on page 33.

The following procedure systematically checks for a blockage in the check valve assembly, in the solvent line that connects the pressure sensor to the valve, and in the pump head.



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

* To determine why the pump has failed the Flush Air script

- 1. If you have not already done so, remove the right side panel of the instrument.
- 2. Depending on the pump, do one of the following:
 - For pumps A and B, go to step 3.
 - For pump S, go to step 4 on page 213.
- 3. For pumps A and B, determine whether the check valve assembly is functioning as follows:
 - a. Run the Purge script for the pump that has failed (see "Prepare Purge Solvent" on page 32).
 - b. Check that solvent is being aspirated and dispensed correctly through the Teflon[™] lines.
 - If the pump is drawing solvent during the fill portion of the purge cycle, the solvent-side check valve is working properly.
 - If the pump is dispensing solvent into the waste beaker during the empty portion of the purge cycle, the waste-side check valve is working properly.

Note If a check valve is not working properly, replace it as described in "Replacing the Check Valves" on page 80.

c. If the check valve assembly is functioning properly, go to step 4.

- 4. Check whether the pump can draw solvent through the tubing that connects the pressure sensor to the rotary valve as follows:
 - a. Using a 1/4 in. open-ended wrench, disconnect the solvent line from the rotary valve (see Figure 211).
 - For pumps A or B, disconnect the solvent line from port 1 of the valve.
 - For pump S, disconnect the solvent line from port 6 of the valve.

Figure 211. Rotary valve layout for the EASY-nLC 1000 instrument



- b. Place the free end of the solvent line into a small vial that contains the appropriate solvent. Use solvent A for pump A and solvent B for pump B.
- c. Run the Purge Solvent script and check that solvent is being aspirated from the vial into the pump.

For information about the Purge Solvent script, see "Prepare – Purge Solvent" on page 32.

- d. Depending on whether the pump aspirated the solvent, do one of the following:
 - If the pump does not aspirate solvent from the vial, go to step 5.
 - If the pump aspirates solvent from the vial, the problem is probably resolved. To verify that the system is working, go to step 4e.
- e. Reconnect all of the solvent lines, and then run the Flush Air script again.
- f. Depending on the results of the Flush Air script, do one of the following:
 - If the Flush Air script fails, go to step 6 on page 214.
 - If the Flush Air script passes, you have completed this troubleshooting procedure.

- 5. Check whether the tubing that connects the pressure sensor to the valve is blocked as follows:
 - a. Reconnect the solvent line to port 1 of the valve.
 - b. Using a 1/4 in. open-ended wrench, disconnect the fitting connected to the top of the pressure sensor.
 - c. Pipette solvent into the port on the top of the pressure sensor.

CAUTION Be careful not to spill solvent on the PCBs.



- For the PLU pump in either the EASY-nLC 1000 or EASY-nLC II instrument, the LED panel cover for each pump protects the PCB from accidental contact when the instrument's right side panel is removed. However, the cover does not form a waterproof seal with the pump body so that the PCBs are exposed to solvent leaks from above.
- For the PLF pump in the EASY-nLC II instrument, the PCBs are completely exposed.
- d. Run the Purge Solvent script and check that the pump is aspirating solvent through the pressure sensor.
- e. Depending on whether solvent is aspirated, do one of the following:
 - If the pump does not aspirate solvent, go to step 6.
 - If the pump aspirates solvent, the tubing that connected the pressure sensor to the valve is damaged. Do the following:
 - i. Using a 1/4 in. open-ended wrench, disconnect this tubing from the valve, and install a new solvent line.
 - ii. To confirm that installing new tubing fixes the problem, run two iterations of the Purge Solvent script. Then, run the Flush Air script with a threshold of 10 μ L for the PLF pump or 12 μ L for the PLU pump.
- 6. To determine if the problem is in the pump, do the following:
 - a. Reconnect the solvent line to the top of the pressure sensor and tighten the fitting.
 - b. Disconnect the solvent line from the pump head by doing one of the following:
 - If a PEEK fitting is connected to the pump head, use a 13 mm open-ended wrench, to disconnect it.
 - If a stainless steel fitting is connected to the pump head, use a 1/4 in. open-ended wrench to disconnect it.
 - c. Pipette the appropriate solvent into the port in the pump head (see Figure 65 on page 71).

- d. Run the Purge script and check that the pump is aspirating solvent from the pump head into the pump.
 - If the pump does not aspirate solvent from the pump head, the problem is in the pump.
 - If the pump does aspirate solvent from the pump head, replace the tubing that connects the pump to its pressure sensor.
- 7. If the problem is in the pump, verify that the piston is not dirty, broken, or scratched and replace the piston seal as described in "Maintaining the Syringe Pumps" on page 60.

Troubleshooting a Pump that Fails the Leaks Script

Follow this procedure to determine why the pump failed the Leaks script.

Note For information about running the Leaks script see "Test – Leaks" on page 44. In the Value column on the Parameters page of the Leaks script, select A to test pump A or B to test pump B.

This procedure systematically checks whether the following problems caused the Leaks script failure:

- Excess air in the system
- Tubing connections between the pump and the pressure sensor
- Tubing connections between pressure sensor and the valve
- A worn piston seal (in the pump head)
- A worn rotor seal (in the subsystem valve)



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

To troubleshoot a pump that has failed the Leaks script

- 1. Run the Flush Air script for the failed pump (see "Prepare Flush Air" on page 33). Enter the following value for the Flush Volume parameter:
 - For the PLF model pump, enter **10** µL.
 - For the PLU model pump, enter **12** µL.
- 2. Do one of the following:
 - If the Output page reports one of these messages, go to "Troubleshooting a Pump that Fails the Flush Air Script" on page 212.
 - Aborting unable to build pressure
 - Aborting check solvent level
 - If the script passes, go to step 3.
- 3. Run the Leaks script for the failed pump or both pumps (see "Test Leaks" on page 44).
 - If the Leaks script fails, the leak is either in the pump, the valve, or the connecting lines or fittings. Go to step 4.
 - If the Leaks script passes, excess air in the system was the source of the problem.

- 4. To determine whether the leak is between the pressure sensor and the valve or the pressure sensor and the pump, do the following:
 - a. Remove the fitting from the top of the relevant pressure sensor, and replace it with a blanking nut. Tighten the nut with a 1/4 in. open-ended wrench.
 - EASY-nLC II blanking nut: LC223
 - EASY-nLC 1000 blanking nut: LC523
 - b. Repeat the Test Leaks script.
 - c. Depending on the result of the Test Leaks script, do one of the following:
 - If the Test Leaks script passes, go to step 5, as the piston seal is leak tight and the leak is between the pressure sensor and the valve.
 - If the Test Leaks script fails, go to step 6, as the leak is between the pressure sensor and the pump.
- 5. To locate a leak between the pressure sensor and the valve, do the following:
 - a. Remove the blanking nut and replace the original fitting.
 - b. Manually check the fittings between the pressure sensor and the valve, and confirm that they are leak tight.

If the fittings are leak tight, the valve might be the source of the leak.

- c. If the valve is leaking, replace the worn rotor seal and clean or replace the stator in the affected valve (see "Maintaining the Rotary Valves" on page 74).
- d. Run the Test Leaks script for the relevant pump and confirm that you have fixed the leak.
- 6. To locate a leak between the pressure sensor and the pump, do the following:
 - a. Remove the blanking nut and replace the original fitting.
 - b. Confirm that the fittings between pump and the pressure sensor are leak tight.
 - c. Repeat the Test Leaks script.
 - d. If the script fails again, the cause might be a worn piston seal.
 - e. If you suspect that the leak is caused by a worn piston seal, go to "Maintaining the Syringe Pumps" on page 60.

Running a System Leak Test

To identify and locate a leak downstream from valves A and B, run the system leak test (Test – Leaks script with System selected), and then take appropriate action as described in Table 24 on page 221.

Table 23 lists the materials required to identify and locate leaks. Because this troubleshooting procedure requires you to handle the solvent lines, you must wear gloves and safety glasses.



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

Instrument	Union	Plug or blind fitting
EASY-nLC II	P/N SC600 Zero-dead-volume union for 1/32 in. OD tubing	
EASY-nLC 1000	P/N SC900	Thermo Scientific Dionex™ P/N 6040.2303
	HPLC zero-dead-volume union for 1/16 in. OD tubing	

Table 23. Materials required for running the system leak test and troubleshooting leaks

To run a system leak test, follow these steps:

- 1. "To prepare the instrument for a system leak test" on page 219
- 2. "To run a system leak test" on page 219

* To prepare the instrument for a system leak test

- 1. Run the Flush Air script for pump A as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Prepare. In the Name list, select Flush Air.
 - c. Press the **Parameters** tab.
 - d. In the Flush Threshold [µL] box, enter the following value:
 - For the PLF model pump (EASY-nLC II instrument), enter 10.
 - For the PLU model pump (EASY-nLC 1000 instrument), enter 12.

Figure 25 on page 33 shows the parameters for the Flush Air script.

e. Press Start.

The script starts and the Output page opens.

- 2. Depending on the results of the Flush Air script displayed on the Output page, do the following:
 - If the script passes, run a system leak test as described in "To run a system leak test" on page 219.
 - If one of these messages appears, go to "Troubleshooting a Pump that Fails the Flush Air Script" on page 212.
 - Aborting unable to build pressure
 - Aborting check solvent level



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

To run a system leak test

- 1. If you have not already done so, run the Flush Air script to prepare the instrument for a system leak test (see "To prepare the instrument for a system leak test" on page 219).
- 2. Connect the Column Out and Waste In lines with the appropriate union (see Table 23 on page 218).

For the EASY-nLC 1000 instrument, connect these lines to the union as follows:

a. Insert the nanoViper fittings on the ends of the Column Out and Waste In lines into the stainless steel union, and then slowly tighten the fittings until you feel resistance.



b. Tighten both fittings by an additional 45 degrees (1/8-turn). Do not tighten the fittings by more than a 1/4-turn (see "Using nanoViper Fittings" on page 83).

CAUTION Because the fittings seal against each other inside the union, do not tighten them by more than 90 degrees (1/4-turn). Overtightening the fittings can irreparably damage their sealing surfaces.

Figure 212 shows the connection between the Column Out and Waste In lines for the EASY-nLC 1000 instrument.

Figure 212. Column Out line connected to the Waste In line (EASY-nLC 1000 instrument)



IMPORTANT Follow the instructions in "Using nanoViper Fittings" on page 83 to ensure that the nanoViper fittings are securely connected to the union.

- 3. Open the Test Leaks script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Test.
 - c. In the Name list, select Leaks.
- 4. Set the parameters for the system leak test as follows:
 - a. Press the **Parameters** tab.
 - b. In the Value list, select **System**.
- 5. Press Start.

The script starts and the Output page opens. During the duration of this script, the EASY–nLC instrument pressurizes the flow paths highlighted with a dotted red line in Figure 35 on page 45, and it monitors the flow sensors and pump piston movements to identify and localize leaks. When the script ends, the Output page displays a message.

6. Depending on the message displayed on the Output page of the Test – Leaks script, take the appropriate action as listed in Table 24.

Troubleshooting the Results of the System Leak Test

Table 24 lists the actions required to troubleshoot the results of a system leak test.

Tip The "Using nanoViper Fittings Quick Reference Guide" on page 323 describes how to return the solvent lines in the flow path of the EASY-nLC 1000 instrument to atmospheric pressure. Print this reference guide to refer to while troubleshooting leaks coming from the nanoViper tubing connections.

Message	Action	
System tight. (Sensor A <100nl/min)	No action is required; however, the system leak test does not test for a a leak in valve B or pump B.	
Flow sensor A should be calibrated.	Rerun the system leak test. If the same message	
The 0 nL/min point is off by more than ±30 nL/min, so the software cannot determine leaks.	appears, run the Flow Sensor script to calibrate flow sensor A (see "Calibrate – Flow Sensors" on page 51).	
Flow sensor B should be calibrated.	Rerun the system leak test. If the same message	
The 0 nL/min point is off by more than ±30 nL/min, so the software cannot determine leaks.	appears, run the Flow Sensor script to calibrate flow sensor B (see "Calibrate – Flow Sensors" on page 51).	
Check valve S for leaks. (Flow sensor A >100 nL/min)	Go to "Checking Valve S for a Leak" on page 222.	
Check the sample loop for leaks.	Go to "Checking the Sample Loop Connections for a Leak" on page 222.	
Check the B solvent line (valve to	Go to "Locating a Leak in the Solvent Path from	
flow sensor) and valve B for leaks. (Flow sensor B ±30 nL/min)	Valve B to Flow Sensor B" on page 223.	
Check from pump A to	Go to "Locating a Leak in the Solvent Path from	
flow sensor A for leaks. (Pump A flow loss >1000 nL/min)	Pump A to Flow Sensor A" on page 224.	
Check from valve S to valve W,	Go to "Locating a Leak In or Between Valve S and	
including valve W, for leaks.	Valve W" on page 226.	
Check solvent lines between	Go to "Locating a Leak Between the Flow Sensors	
flow sensor A and valve S,	and Valve S ["] on page 234.	
flow sensor B, for leaks.		

Table 24. Output messages for a system leak test and actions to take when this test fails

Checking Valve S for a Leak

The system leak test returned the following message (see Table 24 on page 221):

Check valve S for leaks. (Flow sensor A >100 nL/min)

The most likely cause of this test result is a leak in the flow path between ports 3 and 4 of valve S.

* To fix the leaking valve

- 1. Clean the rotor seal and stator in valve S with methanol and a lint-free swab (see "Cleaning the Rotor Seal and Stator" on page 74).
- 2. Repeat the system leak test script and if it fails again, replace the rotor seal in valve S (see "Replacing the Rotor Seal" on page 77).
- 3. To return the EASY-nLC instrument to normal operation as part of an LC/MS system, secure the right side panel, reconnect the Column Out line to the column assembly, and then reconnect the Waste In line to the venting Tee.

Checking the Sample Loop Connections for a Leak

The system leak test returned the following message (see Table 24):

Check the sample loop for leaks.

The sample loop connections are leaking, or the rotor seal in valve S is worn.

* To fix the leaking sample loop connections or the leaking valve

- 1. Depending on the instrument model, do one of the following:
 - For an EASY-nLC II instrument, use a 1/4 in. open-ended wrench to tighten the sample loop fittings connected to ports 2 and 5 on valve S.
 - For an EASY-nLC 1000 instrument, return the instrument to atmospheric pressure as described in the "Using nanoViper Fittings Quick Reference Guide" on page 323. Then individually disconnect and reconnect the nanoViper fittings connected to ports 2 and 5 on valve S. Take care not to overtighten these fingertight fittings.
- 2. Rerun the system leak test (see "Running a System Leak Test" on page 218).
- 3. If the script still fails, clean the rotor seal and stator using methanol and a lint-free swab. Then rerun the system leak test as described in "Cleaning the Rotor Seal and Stator" on page 74).
- 4. If the problem persists, replace the rotor seal (see "Replacing the Rotor Seal" on page 77).
- 5. If the problem persists after replacing the rotor seal, replace the sample loop (see "Replacing the Sample Loop" on page 89).

Locating a Leak in the Solvent Path from Valve B to Flow Sensor B

The system leak test returned the following message (see Table 24 on page 221):

Check the B solvent line (valve B to flow sensor B) and valve B for leaks.

Flow sensor B has detected solvent flow toward valve B. This negative flow rate is caused by a leak in valve B, or in the connections to port 6 of valve B and the flow sensor inlet.

Note When the system is leak tight and valve B is in the Center position, the solvent flow through flow sensor B from pump A should be negligible. A negative flow rate of up to -30 nL/min is within the normal range. A higher negative flow rate indicates a leak that could affect the retention time of analytes.

Figure 213 shows a reading for flow sensor B of -60 nL/min. This means there is a leak in the solvent path from valve B to flow sensor B.

Figure 213. Solvent line between valve B and flow sensor B



To troubleshoot a leak in the solvent line connections between valve B and flow sensor B

- 1. Depending on the instrument model, do one of the following:
 - For the EASY-nLC II instrument, do the following:
 - Using a 1/4 in. open-ended wrench, tighten the stainless steel fitting connected to port 6 of valve B.
 - Hand tighten the PEEK fitting connected to the flow sensor inlet.
 - For the EASY-nLC 1000 instrument, return the solvent system upstream of valve S to atmospheric pressure as described in the "Using nanoViper Fittings Quick Reference Guide" on page 323. Then individually disconnect and reconnect the nanoViper fittings connected to port 6 on valve B and the flow sensor B inlet. Take care not to overtighten these fingertight fittings.

- 2. Rerun the system leak test.
 - If the system leak test still fails, go to "To troubleshoot a leak in valve B."
 - If the system leak test passes, return the system to normal operation.

To troubleshoot a leak in valve B

- 1. Clean the rotor and stator in valve B as described in "Cleaning the Rotor Seal and Stator" on page 74.
- 2. If necessary, replace the rotor seal as described in "Replacing the Rotor Seal" on page 77.

Locating a Leak in the Solvent Path from Pump A to Flow Sensor A

The system leak test returned the following message (see Table 24 on page 221):

Check from pump A to flow sensor A for leaks. (Pump A flow loss >1000 nL/min).

The cause of this test result is a leak between flow sensor A and the inside of pump A. The system leak test script has detected that the piston movement is 1000 nL/min more than the flow measured at flow sensor A.

This procedure checks whether the leak is in the solvent path between the valve and the flow sensor. If the leak in not between the valve and the flow sensor, this procedure directs you to the "Troubleshooting a Pump that Fails the Leaks Script" on page 216 where you determine if the leak is coming from the solvent path between the pump and the valve or from the pump itself.

Figure 214 shows the solvent path from pump A to flow sensor A.

Figure 214. Solvent path from pump A to flow sensor A



* To identify a leak in pump A or the solvent path between pump A and flow sensor A

- 1. Run the Leaks script for pump A only as follows:
 - a. Set the parameters for the Leaks script as follows:
 - i. If you exited the Leaks script on the Maintenance > Scripts page, reopen it.
 - ii. Press the **Parameters** tab.
 - iii. In the Value list, select **A** (pump A).
 - b. Press Start.
- 2. Depending on whether the leak test for pump A passes or fails, do the following:
 - If the script passes, the leak is in the solvent path from valve A to flow sensor A. Go to step 3.
 - If the script fails, the leak is in pump A. Go to "Troubleshooting a Pump that Fails the Leaks Script" on page 216.
- 3. Check the connections to port 6 of valve A and the inlet to flow sensor A as follows:
 - For the EASY-nLC II instrument, do the following:
 - Hand tighten the PEEK fitting connected to the flow sensor inlet.
 - Using a 1/4 in. open-ended wrench, tighten the stainless steel fitting connected to port 6 of valve A.
 - For the EASY-nLC 1000 instrument, return the flow path upstream of valve S to atmospheric pressure as described in the "Using nanoViper Fittings Quick Reference Guide" on page 323. Then individually disconnect and reconnect the nanoViper fittings connected to port 6 on valve A and the flow sensor A inlet. Take care not to overtighten these fingertight fittings.
- 4. Rerun the system leak test. Depending on whether the test fails or passes, do the following:
 - If the test fails, follow the instructions in "Maintaining the Rotary Valves" on page 74 to fix a leaking valve, which in this case is valve A. These instructions guide you through the process of cleaning the valve's rotor seal and stator, rerunning the system leak test, and then replacing the rotor seal if necessary.
 - If the test passes, the system is leak tight. You have completed this troubleshooting procedure. Return the EASY-nLC instrument to normal operation.

Locating a Leak In or Between Valve S and Valve W

If the system leak test returns the following message (see Table 24 on page 221), follow the workflow in Figure 215 and Figure 216 or the procedure on pages 228–234 to locate and remedy the leak.

Check from valve S to valve W, including valve W, for leaks.

Figure 215. Workflow for locating leaks in the solvent path between valves S and W or in these valves (Part 1)





Figure 216. Workflow for locating leaks in the solvent path between valves S and W or in these valves (Part 2)

Figure 217 shows the path between valves S and W for the EASY-nLC 1000 instrument.

Figure 217. Solvent path between valve S and valve W



To locate and remedy a leak in valve S or valve W or in the solvent path between these valves

- 1. Make sure that the connections to the leak test union are secure as follows:
 - a. Depending on the instrument model, do the following:
 - For the EASY-nLC II instrument, hand tighten the PEEK fittings.
 - For the EASY-nLC 1000 instrument, do the following:
 - i. Disconnect the nanoViper fittings from the union.
 - ii. Reinsert the nanoViper fittings into the stainless steel union. Then, slowly tighten the fittings until you feel resistance.
 - iii. Tighten both fittings by an additional 45 degrees (1/8-turn). Do not tighten the fittings by more than 90 degrees (1/4-turn).

For more information, see "Using nanoViper Fittings" on page 83.

Note The connections to the leak test union are one of the most common sources of leaks for the system leak test.

- b. For leak testing, set up and start the Analytical Column Equilibration script as follows:
 - i. On the Maintenance > Scripts page, select **Prepare** in the Category list, and then select **Analytical Col Equilibration** in the Name list.
 - ii. Press the **Parameters** tab.
 - iii. In the Volume $[\mu L]$ box, enter **140**. This volume should be sufficient to keep the solvent flow on during this troubleshooting procedure.
 - iv. Leave the Flow box $\left[\mu L/min\right]$ box empty so that pump A operates at the set pressure.
- v. In the Max Pressure [bar] box, enter **280** bar for the EASY-nLC II instrument or **980** bar for the EASY-nLC 1000 instrument.
- vi. Press Start.

The instrument places the valves in the following positions, and pump A begins pumping solvent through flow sensor A.

- Valve A: 1–6 Valve S: 1–2
- Valve B: Center Valve W: 1–2

Figure 218 shows the solvent system schematic on the Home > Overview page. Use this page to monitor the readback for flow sensor A and to access the valve controls. For information about changing the valve positions, see "Using the Valve Controls" on page 204.

Figure 218. Home > Overview page showing the readback for flow sensor A



- c. On the Home > Overview page, check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, you have remedied the leak and completed this troubleshooting procedure. Return the instrument to normal operation.
 - If the flow rate is >100 nL/min, go to the next step.

- 2. Make sure that the connection between the blind fitting and port 1 of valve W is leak tight as follows:
 - a. Using a 1/4 in. open-ended wrench, tighten the stainless steel blind fitting connected to port 1 of valve W.
 - b. With the Analytical Column Equilibration script running, monitor flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow is <100 nL/min, you have remedied the leak and completed this troubleshooting procedure. Return the instrument to normal operation.
 - If the flow is >100 nL/min, go to the next step.
- 3. Check if valve W is leaking as follows:
 - a. On the Home > Overview page, press the **valve W** icon and set the valve to the **Center** position.

Placing valve W in the Center position blocks the solvent flow into valve W and eliminates valve W as the possible source of the leak.

- b. With the Analytical Column Equilibration script running, monitor flow sensor A. Depending on the flow rate, do one of the following:
 - If the flow is <100 nL/min with valve W removed from the solvent path, valve W is the source of the system leak. To fix the leaking valve, follow the instructions in "Maintaining the Rotary Valves" on page 74 to clean the valve, replace the rotor, and if necessary, replace the stator. Then, rerun the system leak test.
 - If the flow is >100 nL/min, go to the next step.
- 4. Check for a leak-tight connection to port 2 of valve W as follows:
 - a. Depending on the instrument model, do the following:
 - For the EASY-nLC II instrument, use a 1/4 in. open-ended wrench to tighten the fitting to port 2 of valve W.
 - For the EASY-nLC 1000 instrument, do the following:
 - i. On the Home > Overview page, change the position of the following valves to return the system to atmospheric pressure:
 - Valve S: Center
 - Valve W: **1–6**
 - ii. Disconnect and then reconnect the nanoViper fitting from port 2 of valve W. Take care to avoid overtightening the fingertight fitting.
 - iii. On the Home > Overview page, reset valves A and S as follows:
 - Valve S: 1-2
 - Valve W: **1–2**

- b. With the Analytical Column Equilibration script running, monitor flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow is <100 nL/min, you have remedied the leak and completed this troubleshooting procedure. Return the instrument to normal operation.
 - If the flow is >100 nL/min, go to the next step.
- 5. Check the Waste In line as follows:
 - a. Depending on the instrument model, do the following:
 - For the EASY-nLC II instrument, disconnect the Waste In line from the leak test union. Then, block the free end of the union with a blind fitting.
 - For the EASY-nLC 1000 instrument, do the following:
 - i. On the Home > Overview page, change the position of the following valves to return the system to atmospheric pressure:
 - Valve S: Center
 - Valve W: 1-6
 - ii. Using the black knurled nut to loosen the nanoViper fitting, disconnect the Waste In line from the leak test union. Then, block the free end of the union with a blind fitting.
 - iii. On the Home > Overview page, reset valves S and W as follows:
 - Valve S: 1-2
 - Valve W: 1-2

Figure 219 shows the Waste In line disconnected from the leak test union and the free end of the union plugged with a blind fitting.



Figure 219. Setup for eliminating the Waste In line as the source of the system leak

- b. With the Analytical Column Equilibration script running, monitor flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow is <100 nL/min, install a new Waste In line. Then, reconnect the solvent lines and rerun the system leak test as described in "Running a System Leak Test" on page 218.

For ordering information, see Appendix C, "Consumables and Replacement Parts."

- If the flow is >100 nL/min, go to the next step.
- 6. Check for a leak-tight connection to port 3 of valve S as follows:
 - a. Depending on the instrument model, do the following:
 - For the EASY-nLC II instrument, use a 1/4 in. open-ended wrench to tighten the fitting to port 3 of valve S.
 - For the EASY-nLC 1000 instrument, do the following:
 - i. On the Home > Overview page, set **valve S** to the **Center** position to return the system to atmospheric pressure:
 - ii. Disconnect and then reconnect the Column Out line to port 3 of valve S. Take care to avoid overtightening the fingertight fitting.
 - iii. On the Home > Overview page, reset valve S to position 1-2.

- b. With the Analytical Column Equilibration script running, monitor flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow is <100 nL/min, reconnect the solvent lines and rerun the system leak test as described in "Running a System Leak Test" on page 218 to confirm that the system is leak tight.
 - If the flow is >100 nL/min, go to the next step.
- 7. Determine whether the leak is in the Column Out line or in valve S as follows:
 - a. Depending on the instrument model, do the following:
 - For the EASY-nLC II instrument, use a 1/4 in. open-ended wrench to disconnect the Column Out line from port 3 of valve S. Plug the port with a blind fitting. Then, use a 1/4 in. open-ended wrench to tighten the fitting.
 - For the EASY-nLC 1000 instrument, do the following:
 - i. On the Home > Overview page, set **valve S** to the **Center** position to return the system to atmospheric pressure:
 - ii. Using the black knurled nut, disconnect the Column Out line from port 3 of valve S. Then, block port 3 of valve S with a blind fitting.
 - iii. On the Home > Overview page, reset **valve S** to position **1–2**.

Figure 220 shows port 3 of valve S plugged with a blind fitting.

Figure 220. Port 3 of valve S plugged with a blind fitting



- b. With the Analytical Column Equilibration script running, monitor flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow is <100 nL/min, install a new Column Out line. Then, reconnect the solvent lines and rerun the system leak test as described in "Running a System Leak Test" on page 218.

For ordering information, see Appendix C, "Consumables and Replacement Parts."

• If the flow is >100 nL/min, remedy the leak in valve S as described in "Maintaining the Rotary Valves" on page 74. Then, rerun the system leak test as described in "Running a System Leak Test" on page 218.

Locating a Leak Between the Flow Sensors and Valve S

When one or more of the connections are leaking in the solvent path between the flow sensor outlets and the mixing Tee inlets, the system leak test returns this message (see Table 24 on page 221:

Check solvent lines between flow sensor A and valve S, including the mixing Tee and flow sensor B, for leaks.

As the flow sensors and mixing Tee differ for the EASY-nLC 1000 and EASY-nLC II instruments, follow the topic for your instrument to locate and remedy the leak:

- "EASY-nLC 1000: Locating a Leak Between the Flow Sensors and Valve S," on pages 235–245
- "EASY-nLC II: Locating a Leak Between the Flow Sensors and Valve S" on page 246

EASY-nLC 1000: Locating a Leak Between the Flow Sensors and Valve S

To determine whether the leak is coming from the connection to port 4 of valve S, the connection to the mixing Tee outlet, or the connections between the flow sensor outlets and the mixing Tee inlets, follow the workflow in Figure 221 or the procedure on pages 236-239.





To locate leaks between the flow sensors and valve S in the EASY-nLC 1000 instrument

- 1. To ensure a leak-tight connection to port 4 of valve S, do the following:
 - a. Disconnect and then reconnect the nanoViper fitting to port 4 of valve S.
 - b. From the Maintenance > Scripts page, start the Analytical Column Equilibration script with the maximum pressure set to **980** bar and the volume set to **140** μ L.

For information about setting up the Analytical Column Equilibration script, see step 1b on page 228.

When you start the Analytical Column Equilibration script, the instrument sets the rotary valves to these positions and begins pumping solvent through flow sensor A:

- Valve A: 1–6
- Valve B: Center
- Valve S: 1–2
- Valve W: 1–2
- c. On the Home > Overview page, check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, you have remedied the leak and completed this troubleshooting procedure. Return the instrument to normal operation.
 - If the flow rate is >100 nL/min, go to the next step.
- 2. To check the solvent line that connects the mixing Tee outlet to port 4 of valve S, do the following:
 - a. On the Home > Overview page, set valves A and W to the following positions to return the system to atmospheric pressure:
 - Valve A: Center
 - Valve W: 1–6

For information about changing the valve positions, see "Using the Valve Controls" on page 204.

b. Disconnect the nanoViper fitting from the mixing Tee outlet. Then, block the mixing Tee outlet with a blind fitting (see Figure 222).



Figure 222. Mixing Tee outlet plugged with a blind fitting

- c. On the Home > Overview page, reset these valve positions:
 - Valve A: 1–6
 - Valve W: 1–2

- d. On the Home > Overview page, check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, do the following:
 - i. Stop the Analytical Column Equilibration script and return the system to atmospheric pressure.
 - ii. Disconnect the damaged solvent line from port 4 of valve S.
 - iii. Install a new solvent line to connect the mixing Tee outlet to port 4 of valve S.

For ordering information, see Table 33 on page 316.

- iv. Rerun the system leak test (see "Running a System Leak Test" on page 218).
- If the flow rate is >100 nL/min, go to the next step.
- 3. To eliminate the solvent B channel as the leak source, do the following:
 - a. Set **valve A** to the **Center** position to return the system to atmospheric pressure while maintaining pressure on the front end of the system.
 - b. Disconnect the blind fitting from the mixing Tee outlet, and then reconnect the solvent line to the mixing Tee outlet.
 - c. Disconnect the solvent line from the mixing Tee inlet for the solvent B channel, and then block the inlet with a blind fitting.

Figure 223 shows the solvent line disconnected from the mixing Tee and a blind fitting in the inlet port of the mixing Tee for the solvent B channel.

d. Reset **valve A** to position **1–6**.



Figure 223. Mixing Tee inlet for the solvent B channel plugged with a blind fitting

- e. On the Home > Overview page, check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, go to "EASY-nLC 1000: Locating a Leak Between Flow Sensor B and the Mixing Tee" on page 243, as removing the solvent B channel from the flow path remedied the leak.
 - If the flow rate is >100 nL/min, the leak is in the solvent A channel, as removing the solvent B channel from the solvent path did not remedy the leak. Go to the next procedure, "EASY-nLC 1000: Locating a Leak Between Flow Sensor A and the Mixing Tee."

EASY-nLC 1000: Locating a Leak Between Flow Sensor A and the Mixing Tee

By following the workflow in Figure 221 on page 235 or the procedure on pages 236–239, you determined that the leaking connection is between flow sensor A and the mixing Tee. The system plumbing is set up as shown in Figure 223 on page 239, with the solvent channel B disconnected from the mixing Tee.

Follow the workflow in Figure 224 or the procedure on pages 241–242 to locate and remedy the leak.





To locate and remedy a leak between flow sensor A and the mixing Tee in the EASY-nLC 1000

- 1. To check if you must replace the solvent line that connects the outlet of flow sensor A to the mixing Tee, do the following:
 - a. Set **valve A** to the **Center** position.
 - b. Disconnect the solvent line from the inline filter that connects to the outlet of flow sensor A. Then, block the inline filter with a blind fitting.

Figure 225 shows the solvent path blocked at the outlet of flow sensor A.

c. Reset valve A to position 1–6.





- d. Check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, do the following:
 - i. Stop the Analytical Column Equilibration script.
 - ii. Disconnect the damaged solvent line from the mixing Tee's inlet for the solvent A channel.
 - iii. Install a new solvent line between the mixing Tee's inlet for the solvent A channel and the flow sensor's outlet.

For ordering information, see Table 33 on page 316.

- iv. Reconnect the solvent lines and rerun the system leak test (see "Running a System Leak Test" on page 218).
- If the flow rate is >100 nL/min, go to the next step.
- 2. To check the connection between the inline filter and the flow sensor A outlet, do the following:
 - a. Using a 5/16 in. open-ended wrench, tighten the inline filter to the outlet of flow sensor A.
 - b. Check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, do the following:
 - i. Stop the Analytical Column Equilibration script.
 - ii. Reconnect all of the solvent lines.
 - Rerun the system leak test (see "Running a System Leak Test" on page 218).
 - If the flow rate is >100 nL/min, do the following:
 - i. Stop the Analytical Column Equilibration script.
 - ii. Remove the damaged inline filter and install a new inline filter.
 - iii. Rerun the system leak test (see "Running a System Leak Test" on page 218).

EASY-nLC 1000: Locating a Leak Between Flow Sensor B and the Mixing Tee

By following the workflow in Figure 221 on page 235 or the procedure on pages 236–239, you determined that the leaking connection is between the flow sensor B outlet and the mixing Tee inlet. The system plumbing is set up as shown in Figure 223 on page 239, with the solvent channel B disconnected from the mixing Tee inlet.

Follow the workflow in Figure 226 or the procedure on pages 244–245 to locate and remedy the leak.

Figure 226. EASY-nLC 1000 workflow for locating a leak between flow sensor B and the mixing Tee



***** To locate a leak between flow sensor B and the mixing Tee in the EASY-nLC 1000

- 1. To ensure leak-tight connections for the solvent line that connects the outlet of flow sensor B to the mixing Tee inlet, do the following:
 - a. Set **valve A** to the **Center** position.
 - b. Set up the system plumbing as follows:
 - i. Remove the blind fitting from the mixing Tee's inlet for the solvent B channel.
 - ii. Reconnect the solvent line to the solvent B channel inlet of the mixing Tee.
 - iii. Redo the nanoViper connection to the mixing Tee's inlet for the solvent B channel.

The system plumbing now matches that for a system leak test.

- c. Reset valve A to position 1–6.
- d. Check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, you have completed this troubleshooting procedure. Return the instrument to normal operation.
 - If the flow rate is >100 nL/min, go to the next step.
- 2. Using a 5/16 in. open-ended wrench, tighten the inline filter to the outlet of flow sensor B.
- 3. To determine whether the solvent line or the inline filter is the leak source, do the following:
 - a. Set valve A to the Center position.
 - b. Disconnect the solvent line from the outlet of flow sensor B. Then, plug the free end of the solvent line with a leak test union and a blind fitting (see Figure 227).



Figure 227. Solvent line disconnected from the outlet of flow sensor B

- c. Reset **valve A** to position **1–6**.
- d. Check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, do the following:
 - i. Stop the Analytical Column Equilibration script.
 - ii. Remove the damaged inline filter from the outlet of flow sensor B and install a new inline filter.

For ordering information, see Table 35 on page 319.

- iii. Reconnect the system plumbing and rerun the system leak test (see "Running a System Leak Test" on page 218).
- If the flow rate is >100 nL/min, do the following:
 - i. Stop the Analytical Column Equilibration script.
 - ii. Install a new solvent line between the flow sensor B outlet and the mixing Tee inlet.

For ordering information, see Table 33 on page 316.

iii. Rerun the system leak test (see "Running a System Leak Test" on page 218).

EASY-nLC II: Locating a Leak Between the Flow Sensors and Valve S

Figure 228 on page 247 shows the solvent lines that connect the flow sensor outlets to the mixing Tee and the solvent line that connects the mixing Tee outlet to port 4 of valve S.

To systematically locate and remedy a leak in an EASY-nLC II instrument from the flow sensor outlets to port 4 of valve S

- 1. To ensure a leak-tight connection to port 4 of valve S, do the following:
 - a. Tighten the fitting in port 4 of valve S.
 - b. From the Maintenance > Scripts page, start the Analytical Column Equilibration script with the maximum pressure set to **280** bar and the volume set to **140** μ L.

For information about setting up the Analytical Column Equilibration script, see step 1b on page 228.

- c. On the Home > Overview page, check the readback for flow sensor A. Then, depending on the flow rate, do one of the following:
 - If the flow rate is <100 nL/min, you have remedied the leak and completed this troubleshooting procedure. Return the instrument to normal operation.
 - If the flow rate is >100 nL/min, go to the next step.
- 2. Hand tighten the following PEEK fittings:
 - Fittings connected to the mixing Tee
 - Fittings connected to the flow sensor outlets
- 3. With the Analytical Column Equilibration script running, monitor flow sensor A. Depending on the flow rate, do one of the following:
 - If the flow is <100 nL/min, tightening the solvent line connections remedied the leak, and you have completed this troubleshooting procedure. Return the instrument to normal operation.
 - If the flow is >100 nL/min, the leak is coming from one of the solvent lines. Go to the next step.

Note For information about ordering solvent lines for the EASY-nLC II instrument, see Table 32 on page 314.

- 4. Replace the solvent line that connects the mixing Tee outlet and port 4 of valve S. Restart the Analytical Column Equilibration script and monitor flow sensor A.
 - If the flow is <100 nL/min, you have completed this troubleshooting procedure.
 - If the flow is >100 nL/min, go to the next step.

- 5. Replace the solvent line that connects the outlet of flow sensor A to the mixing Tee inlet. Restart the Analytical Column Equilibration script and monitor flow sensor A.
 - If the flow is <100 nL/min, you have completed this troubleshooting procedure.
 - If the flow is >100 nL/min, go to the next step.
- 6. Replace the solvent line that connects the outlet of flow sensor B to the mixing Tee inlet. Restart the Analytical Column Equilibration script and monitor flow sensor A.
 - If the flow is <100 nL/min, you have completed this troubleshooting procedure.
 - If the flow is >100 nL/min, contact your local Thermo Fisher Scientific field service engineer.
- Figure 228. Solvent lines that connect the flow sensor outlets to the mixing Tee and the solvent line that connects the mixing Tee outlet to port 4 of valve S in the EASY-nLC II instrument



Identifying a Leaking Check Valve

The EASY-nLC instrument has four check valves. Each solvent inlet line connects to a check valve assembly with a solvent-side check valve and a waste-side check valve.



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

To determine if the waste-side check valve is leaking

- 1. Run the Purge Solvent script as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Prepare.
 - c. In the Name list, select **Purge Solvent**.
 - d. Press the **Parameters** tab.
 - e. Select the check box for the appropriate pump.
 - f. Press Start.
- 2. As the pump is filling, observe the waste line.

If you observe solvent moving toward the check valve assembly, the waste-side check valve is leaking.

3. If the waste-side check valve is leaking, replace it and repeat the Purge Solvent script.

* To determine if the solvent-side check valve is leaking

- 1. Start the Purge Solvent script.
- 2. As the pump is emptying, observe the solvent line.

If you observe solvent moving toward the solvent bottle from the check valve assembly, the solvent-side check valve is leaking.

3. If the solvent-side check valve is leaking, replace it and repeat the Purge Solvent script.

Troubleshooting a System Blockage

Use this troubleshooting procedure when the back pressure for a chromatographic method rises above the expected level or when the flow decreases below the set rate for the column equilibration or sample loading steps.

In most cases, an increase in back pressure is caused by a blocked column or a blocked Column Out line. The Column Out line has an ID of 30 μ m for the EASY-nLC II instrument and 20 μ m for the EASY-nLC 1000 instrument and is the smallest ID line that the sample passes through, from sample pickup to the columns.

Troubleshooting a system blockage requires these tools and materials.

Tools	Parts and materials	
1/4 in. open-ended wrench to loosen the stainless steel fittings	Powder-free gloves	
	• EASY-nLC II instrument: HPLC union, P/N SC600	
	• EASY-nLC 1000 instrument: HPLC union, P/N SC900	



CAUTION Wear powder-free gloves and safety glasses when handling parts of the LC system that come into contact with solvents.

When running the Back Pressure script, the system also checks for an unusually low back pressure. If the back pressure is unusually low, the system probably has a leak.

To troubleshoot a system blockage, follow these procedures:

- 1. "Running the Back Pressure Test for the A and B Solvent Paths" on page 250
- 2. Depending on the test results, replace the blocked components in the column assembly or follow one or both of these procedures:
 - "Troubleshooting a System Blockage when Test Solvent A Fails" on page 251
 - "Troubleshooting a System Blockage when Test Solvent B Fails" on page 257

Running the Back Pressure Test for the A and B Solvent Paths

The Back Pressure script determines instrument back pressure for solvent lines A and B. The script runs at a preset flow and measures the back pressure on the system.

IMPORTANT Before you perform the Back Pressure script, ensure that the solvent A bottle contains water and the solvent B bottle contains acetonitrile. This test is not valid for other solvents.

To run the back pressure script for both the A and B solvent paths

1. Remove the columns and connect the Column Out line to the Waste In line using the appropriate HPLC union.

Instrument	Union
EASY-nLC II	SC600
EASY-nLC 1000	SC900

- 2. Run the Back Pressure scripts for both Solvent A and Solvent B as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Test.
 - c. In the Name list, select **Back Pressure**.
 - d. Press the **Parameters** tab.
 - e. Select the Test Solvent A and Test Solvent B check boxes.
 - f. Press Start.
- 3. Depending on the test results, do one of the following:
 - If the Test Solvent A test fails and the following message appears, go to "Troubleshooting a System Blockage when Test Solvent A Fails" on page 251.
 - EASY-nLC II instrument—Pressure is xxx bar (exceeds 100 bar limit), test failed!
 - EASY-nLC 1000 instrument—Pressure is xxx bar (exceeds 175 bar limit), test failed!
 - If the Test Solvent B test fails and the following message appears, go to "Troubleshooting a System Blockage when Test Solvent B Fails" on page 257.

Pressure is xxx bar (exceeds 100 bar limit), test failed!

• If both Test Solvent A and Test Solvent B tests pass and the following message appears, inspect and if necessary replace the columns, the packed emitter, or both.

Pressure is xxx bar, test passed.

Troubleshooting a System Blockage when Test Solvent A Fails

Follow this procedure when the Back Pressure test for Test Solvent A fails (see "Running the Back Pressure Test for the A and B Solvent Paths" on page 250).

* To verify the system blockage

- 1. Run the Precolumn Equilibration script and check the back pressure as follows:
 - a. On the Maintenance > Scripts page, select **Prepare** in the Category list, and then select **Precolumn Equil** in the Name list.
 - b. Press the **Parameters** tab.
 - c. In the Volume [μ L] box, enter **140** μ L.
 - d. In the Flow [μ L/min] box, enter 2 μ L/min.
 - e. Leave the Max Pressure [bar] box blank.
 - f. Press Start.
 - g. Monitor the pump A pressure trace.
- 2. Depending on the back pressure, do the following:
 - If the back pressure is below the pressure threshold, the system blockage has been removed. To confirm that the back pressure is below the pressure threshold, rerun the Back Pressure script with the **Test Solvent A** check box selected.
 - If the back pressure is above the pressure threshold, keep the Precolumn Equil script running and go to "To locate the source of the blockage when the Test Solvent A test fails" on page 253.

Instrument	Pressure threshold	
EASY-nLC II	100 bar	
EASY-nLC 1000	175 bar	

Figure 229 shows the areas that the following procedure systematically checks for blockage.

Figure 229. Solvent path from pressure sensor A to the Waste In line



* To locate the source of the blockage when the Test Solvent A test fails

- 1. To determine if valve W is blocked, do the following:
 - a. Make sure that the Precolumn Equilibration script is set up and running as described in step 1 on page 251 and that the Column Out and Waste In lines are connected with a union.
 - b. Disconnect the Waste In line from port 2 of valve W (see Figure 229 on page 252).
 - c. Depending on the back pressure, do one of the following:
 - If the pressure is below the threshold, valve W is blocked. Go to "Maintaining the Rotary Valves" on page 74.
 - If the pressure is above the threshold, go to step 2.
- 2. To determine if the Waste In line is blocked, do the following:
 - a. Disconnect the Column Out line and the Waste In line from the union.



- b. Depending on the back pressure, do one of the following:
 - If the pressure is below the threshold, the Waste In line is blocked. Discontinue this troubleshooting procedure and replace the Waste In line with a new line.

Instrument	Pressure threshold	
EASY-nLC II	100 bar	
EASY-nLC 1000	125 bar	

- If the pressure is above the threshold, go to step 3.
- 3. To determine if the Column Out line is blocked, do the following:
 - a. Disconnect the Column Out line from port 3 of valve S.
 - b. Depending on the back pressure, do one of the following:
 - If pressure is below the threshold, the Column Out line is blocked. Discontinue this troubleshooting procedure and replace the Column Out line with a new line.

Instrument	Pressure threshold	
EASY-nLC II	100 bar	
EASY-nLC 1000	90 bar	

• If the pressure is above the threshold, reconnect the Column Out line, and then go to step 4.

- 4. To determine if valve S is partially or totally blocked, do the following:
 - a. Disconnect the mixing Tee line from port 4 of valve S.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is below the threshold, valve S is partially or totally blocked. Discontinue this troubleshooting procedure and go to "Maintaining the Rotary Valves" on page 74.

Instrument	Pressure threshold	
EASY-nLC II	100 bar	
EASY-nLC 1000	90 bar	

- If the pressure is above the threshold, reconnect the mixing Tee line to port 4 of valve S, and then go to step 5.
- 5. To determine if the line that connects valve S to the mixing Tee is blocked, do the following:
 - a. Disconnect the line from the mixing Tee.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is below the threshold, the line that connects the mixing Tee to valve S is blocked. Replace it.

Instrument	Pressure threshold	
EASY-nLC II	100 bar	
EASY-nLC 1000	65 bar	

- If the pressure is above the threshold, reconnect the line to the mixing Tee, and then go to step 6.
- 6. To determine if the mixing Tee is blocked, do the following:
 - a. Disconnect the line that exits flow sensor A from the mixing Tee.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is below the threshold, the mixing Tee is blocked. Clean the mixing Tee, or if necessary replace it.

Instrument	Pressure threshold	
EASY-nLC II	100 bar	
EASY-nLC 1000	65 bar	

• If the pressure is above the threshold, reconnect the line to the mixing Tee, and then go to step 7.

- 7. To determine if the line that connects the mixing Tee to flow sensor A is blocked, do the following:
 - a. Disconnect the line from the outlet of flow sensor A.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, the line that connects the mixing Tee to flow sensor A is blocked. Replace it with a new solvent line.
 - If the pressure is >20 bar, reconnect the line to flow sensor A, and then go to step 8 for an EASY-nLC 1000 instrument or step 9 for an EASY-nLC II instrument.
- 8. To determine if the inline filter connected to the flow sensor outlet of the EASY-nLC 1000 system is blocked, do the following:
 - a. Remove the inline filter from the flow sensor outlet.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, the inline filter is blocked. Replace it with a new filter.
 - If the pressure is >20 bar, reconnect the inline filter to flow sensor A, and then go to step 9.
- 9. To determine if flow sensor A is blocked, do the following:
 - a. Disconnect the solvent line from the flow sensor A inlet.
 - b. Depending on the back pressure for flow sensor A, do one of the following:
 - If the pressure is <20 bar, flow sensor A is blocked. Replace it with a new flow sensor as described in "Replacing a Flow Sensor" on page 92.
 - If the pressure is >20 bar, reconnect the line to flow sensor A, and then go to step 10 for an EASY-nLC 1000 system or step 11 for an EASY-nLC II system.
- 10. To determine if the inline filter connected to the flow sensor inlet of the EASY-nLC 1000 system is blocked, do the following:
 - a. Remove the inline filter from the flow sensor inlet.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, the inline filter is blocked. Replace it with a new inline filter.
 - If the pressure is >20 bar, reconnect the inline filter to flow sensor A, and then go to step 11.

- 11. To determine if the line that connects valve A to flow sensor A is blocked, do the following:
 - a. Disconnect the solvent line from port 6 of valve A.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, the line from valve A to flow sensor A is blocked. Replace it with a new solvent line.
 - If the pressure is >20 bar, reconnect the line to valve A, and then go to step 12.
- 12. To determine if valve A is blocked, do the following:
 - a. Disconnect the solvent line from port 1 of valve A.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, valve A is blocked. Inspect the valve and clean the stator. If the blockage persists, replace the rotor seal.
 - If the pressure is >20 bar, reconnect the line to valve A, and then go to step 13.
- 13. To determine if the line from pressure sensor A to valve A is blocked or if pressure sensor A is blocked, do the following:
 - a. Disconnect the solvent line from the outlet of pressure sensor A.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, the line that connects pressure sensor A to valve A is blocked. Replace it with a new solvent line.
 - If the pressure is >20 bar, pressure sensor A is blocked. Replace it with a new pressure sensor as described in "Replacing a Pressure Sensor for the PLU Pump" on page 90.

Troubleshooting a System Blockage when Test Solvent B Fails

Follow this procedure when the Back Pressure test for Test Solvent B fails (see "To run the back pressure script for both the A and B solvent paths" on page 250).

Figure 230 shows the areas that this procedure systematically checks for blockage.

Figure 230. Solvent system from pressure sensor B to the mixing Tee



* To troubleshoot a system blockage when only the Test Solvent B test fails

- 1. To determine if the blockage is in the mixing Tee, do the following:
 - a. Make sure that the Column Out line is connected to the Waste In line with a union.
 - b. Remove the solvent line from the mixing Tee that connects it to flow sensor B.
 - c. Repeat the Back Pressure script with the Test Solvent B check box selected.
 - d. Depending on the test results, do one of the following:
 - If Test solvent B passes, clean the mixing Tee and if necessary replace it.
 - If Test solvent B fails, go to step 2.
- 2. To determine if the blockage is in the solvent line that connects the B flow path to the mixing Tee, do the following:
 - a. Disconnect the solvent line from the outlet of flow sensor B.
 - b. Place valve B in position 1–6 as follows:
 - i. Press Home > Overview.
 - ii. Press the valve B icon.
 - iii. In the Valve B dialog box, press 1-6.
 - c. Set pump B to deliver 2 μ L/min as follows:
 - i. Press the pump B icon.
 - ii. In the flow rate box, type $2 \mu L/min$.
 - iii. Press Start.
 - iv. Leave the dialog box open.

For more information about the direct controls for the pump, see "Using the Pump Controls" on page 205.

- d. Monitor the Pressure readback box for pump B. Then, depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, the solvent line is blocked.
 - For the EASY-nLC II instrument, replace the solvent line with a new fused silica line. Then, to confirm that the blockage was in the discarded solvent line, repeat the Back Pressure script with the **Test Solvent B** check box selected.
 - For the EASY-nLC 1000 instrument, place valve B in position 1–2 to release the pressure. Remove the blocked nanoViper line and replace it with a new nanoViper line. Then, to confirm that the blockage was in the discarded solvent line, repeat the Back Pressure script with the Test Solvent B check box selected.
 - If the pressure is >20 bar, keep pump B running and go to step 3 for the EASY-nLC 1000 instrument or step 4 for the EASY-nLC II instrument.

- 3. For the EASY-nLC 1000 instrument, to determine if the inline filter connected to the flow sensor outlet is blocked, do the following:
 - a. Remove the inline filter from the flow sensor outlet.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, the inline filter is blocked. Replace it with a new filter.
 - If the pressure is >20 bar, reconnect the inline filter to flow sensor A, and then go to step 4.
- 4. To determine if flow sensor B is blocked, do the following:
 - a. Disconnect the solvent line from the inlet of flow sensor B, and then monitor the pressure.
 - b. Depending on the pressure, do one of the following:
 - If the pressure is <20 bar, flow sensor B is blocked. Replace it as described in "Replacing a Flow Sensor" on page 92.
 - If the pressure is >20 bar, keep pump B running and go to step 5 for the EASY-nLC 1000 instrument or step 6 for the EASY-nLC II instrument.
- 5. For the EASY-nLC 1000 instrument, to determine if the inline filter connected to the inlet of flow sensor B is blocked, do the following:
 - a. Remove the inline filter from the flow sensor outlet.
 - b. Depending on the back pressure, do one of the following:
 - If the pressure is <20 bar, the inline filter is blocked. Replace it with a new filter.
 - If the pressure is >20 bar, reconnect the inline filter to flow sensor A, and then go to step 6.
- 6. To determine if the line that connects valve B to flow sensor B is blocked, do the following:
 - a. Disconnect the line from port 6 of valve B, and then monitor the pressure.
 - b. Depending on the pressure, do one of the following:
 - If the pressure is below the threshold, the solvent line is blocked. Discontinue this troubleshooting procedure, and replace the solvent line that connects port 6 of valve B to the flow sensor inlet.

Instrument	Pressure threshold	
EASY-nLC II	20 bar	
EASY-nLC 1000	10 bar	

• If the pressure is above this threshold, keep the pump running and go to step 7.

- 7. To determine if valve B is blocked, do the following:
 - a. Remove the solvent line from port 1 of valve B, and then monitor the pressure.
 - b. Depending on the pressure, do one of the following:
 - If the pressure is below the threshold, valve B is blocked. Go to "Maintaining the Rotary Valves" on page 74.

Instrument	Pressure threshold	
EASY-nLC II	20 bar	
EASY-nLC 1000	10 bar	

- If the pressure is above the threshold, keep the pump running and go to step 8.
- 8. To determine if either the line that connects valve B to pressure sensor B is blocked or pressure sensor B is blocked, do the following:
 - a. Remove the solvent line that connects valve B to pressure sensor B from pressure sensor B, and then monitor the pressure.
 - b. Depending on the pressure, do one of the following:
 - If the pressure is below the threshold, the solvent line is blocked. Replace it.

Instrument	Threshold pressure	
EASY-nLC II	20 bar	
EASY-nLC 1000	10 bar	

• If the pressure is >20 bar, pressure sensor B is blocked. Replace it as described in "Replacing a Pressure Sensor for the PLU Pump" on page 90.

Troubleshooting the Autosampler Aspiration and Calibration

Troubleshooting the autosampler aspiration requires these tools and materials:

- 1/4 in. open-ended wrench
- Powder-free safety gloves
- Sample vial filled with solvent A
- Microtiter plate (if you are using a microtiter plate format)

* To troubleshoot the autosampler aspiration of sample into the needle

- 1. Make sure that the W3 wash bottle contains sufficient solvent.
- 2. Use the Flush Air script to remove air from pump S as follows:
 - a. Press Maintenance > Scripts.
 - b. In the Category list, select Prepare.
 - c. In the Name list, select Flush Air.
 - d. Press the **Parameters** tab.
 - e. Select the Flush Pump S check box.
 - f. For Flush Threshold [μ L], enter **10** for the PLF model pump or **12** for the PLU model pump (see Figure 231).

Figure 231. Flush Air script

SCIENTIFIC	Home Batch Setup	Method Editor	Maintenance Configuration
Scripts Log Book Support	Category: Prepare Description Param	Name: Flush air • eters Output	Pump S — Desired flow — Pressure 1.0 250.0 T
Devices	Parameter Flush pump A Flush pump B Flush pump S Flush threshold (µI)	 ✓ Value □ □ ∞ 12.00 	200.0 graph
Job 00:00:00 6 May 2011 11:15 admin Exit	Schedule	RT STOP	

g. Press Start.

The script runs until the flush threshold reaches the specified value or one of these error messages appears on the Output page:

- Aborting unable to build pressure
- Aborting check solvent level
- 3. Depending on whether the Flush Air script passed or failed, do one of the following:
 - If the Flush Air script failed, leave the Maintenance > Scripts page open and go to step 4 to determine if the autosampler needle is blocked.
 - If the Flush Air script passed, leave the Maintenance > Scripts page open and go to step 5 to determine if the autosampler requires calibration.
- 4. To determine if the autosampler needle is blocked, do the following:
 - a. Using a 1/4 in. open-ended wrench, disconnect the PEEKsil autosampler needle from port 1 of valve S.
 - b. Disconnect the Teflon waste line from port 3 of valve W and connect it to port 1 of valve S.
 - c. Place the free end of the Teflon line in an HPLC vial filled with solvent A.
 - d. To open the Purge script, select **Purge** in the Name list.
 - e. Select the **Purge Pump S** check box.
 - f. Press Start.
 - g. Verify that pump S aspirates solvent from the HPLC vial.
 - h. Do one of the following:
 - If pump S can aspirate solvent through the Teflon waste tubing, the needle is blocked. Reconnect the Teflon waste tubing to port 3 of valve W and replace the autosampler needle (see "Replacing the Autosampler Needle" on page 84).
 - If pump S cannot aspirate solvent through the Teflon waste tubing, go to "Troubleshooting a Pump that Fails the Flush Air Script" on page 212.
- 5. To determine if the autosampler needle is going to the correct position and depth, go to "Checking Sample Pickup" on page 263.

Checking Sample Pickup

When you run the Sample Pickup script, the autosampler withdraws a specified volume from a specified position. To verify the volume withdrawn by the autosampler, you must place a known volume into the specified position, and then measure the remaining volume after you run the script. You can use a pipette or an analytical balance with microgram accuracy to determine the initial and final volumes.

* To check sample pickup from a microtiter plate or vial

- 1. Fill the well or vial with a 12 μ L of solvent A.
- 2. Weigh the microwell plate or vial.
- 3. Insert the tray into the tray compartment.
- 4. Press Maintenance > Scripts.
- 5. Run the Sample Pickup script with a pickup volume of 10 μ L and your default flow (see Figure 232).



Category:	Name:	Category:	Name:	Category:	Name:
Test	Sample pickup 🗣	Test	Sample pickup 🔻	Test	Sample pickup
Description Parameters Output		Description Parameters Output		Description	Parameters Output
Test autosampler sample pick up. Please fill plate well with a defined volume before running test. Max volume: Loop volume - 2µl. Max flow: 40.0 µl/min.		Parameter	✓ Value	'Sample pickup' Prime pump - fil	started
		Volume [µl]	10.00		
		Flow [µl/min]	20.00		
		Position	0-A1		
Schedule	ART STOP	Schedule	ART STOP	Schedule	START STOP

- 6. After the EASY-nLC application finishes the script, do the following:
 - a. Eject the tray.
 - b. Check that $2 \mu L$ is left in the well or vial by reweighing the microplate or vial or by checking the volume with a pipette.
 - c. If more than 2 μ L remains, recalibrate the autosampler, run the Flush Air script, and then rerun the sample pickup check.

For information about calibrating the autosampler, see Chapter 7. For information about the Flush Air script, see "Prepare – Flush Air" on page 33.

Troubleshooting Communication Problems

To troubleshoot communication problems, see these topics:

- "Network Access" on page 264
- "Network Connection Failures" on page 265
- "Testing the Network Connection to the EASY-nLC Computer" on page 266

Network Access

Table 25 contains troubleshooting tips for network access problems.

Table 25. Network access problems

Symptom	Possible causes	Action
System is not responding after pressing Save Configuration on the Network page under Configuration.	Length of completion time to update the network settings	As indicated, wait while the system changes the network settings.
Cannot access the EASY-nLC system through the network.	Changed network address	Check the network address of the EASY-nLC system on the Configuration > Network page.
	The stand-alone EASY-nLC instrument is not connected to a laboratory LAN port with intranet access.	Connect the stand-alone EASY-nLC instrument to a laboratory LAN port with intranet access.
	The LC/MS system is not connected to a laboratory LAN port with intranet access.	Connect the data system computer for the LC/MS system to a laboratory LAN port with intranet access.
Network Connection Failures

Table 26 lists the possible causes of a network connection failure. Figure 233 shows the Thermo EASY-nLC Configuration dialog box that you access from the Thermo Foundation Instrument Configuration application. When the EASY-nLC instrument cannot establish communication with the data system computer, the Connection Failed message appears when you click Test Connection.

Figure 233. Network connection failure

Thermo EASY-nLC Confi	guration		×	
EASY-nLC Network Configuration				
IP Address/Hostname:	10.32.134.79	The IP Address can be foun About box on the EASY-nLC	d in the Cinstrument	
User name:	admin	Run samples as this user (U exist on the EASY-nLC)	ser must	
Test Connection		Connection failed!		
LC device driver and ins	trument information:			
LC device driver : LCServer 1.1.1.1				
Autosampler Configurati	on	Solvents for LC Pur	mp	
Autosampler:				
Plate installed:	✓ Edit.			
Sample layout: Row fir	st Start pos: A1	B: acetonitrile		
ОК		[Cancel	

Symptom	Possible cause	Action
The connection between the EASY-nLC instrument and the virtual interface for the Xcalibur data system fails.	The network connection has been unexpectedly disabled.	Test the network connection as described in "Testing the Network Connection to the EASY-nLC Computer" on page 266. Close down the EASY-nLC instrument as described in "Closing Down the EASY-nLC Instrument" on page 21, and then turn off the power. Restart the Xcalibur data system computer. Turn on the EASY-nLC instrument and log in as an administrator. Retest the connection.
	An antivirus program is blocking port 6666.	Test the connection. Make sure that the data system computer is not connected to the Internet, and then uninstall the antivirus program.
	The network firewall is blocking port 6666.	Test the connection. Remove the firewall.
	Connection between the data system computer and the EASY-nLC instrument has been lost.	Test the connection.

Table 26. Possible causes of a network connection failure

Testing the Network Connection to the EASY-nLC Computer

If the connection between the EASY-nLC instrument and the data system computer fails when you click Test Connection in the Thermo EASY-nLC Configuration dialog box of the Foundation Instrument Configuration window, follow this procedure.

To test the network connection between the data system computer and the EASY-nLC instrument

1. From the Windows Start menu, choose Run.

The Run dialog box opens.

- 2. In the Open box, type **cmd**.
- 3. Press OK.

The Command Prompt window opens.

4. Type telnet IP address for the EASY-nLC system 6666 (see Figure 234).

Figure 234. Command Prompt window with the default IP address for the EASY-nLC instrument



- 5. Press the ENTER key.
 - If the connection is set up correctly, a login prompt appears in the Telnet window (see Figure 235).
 - If the connection is not set up correctly, the following message appears:

Could not open connection to host.

Figure 235. Telnet window with login prompt

🛤 Telnet 172.16.0.103	- 🗆 🗙
Connected from: 172.16.0.101[172.16.0.101:4172] Guessed Locale: en	^
Negotiated Terminal Info: ANSI,80,25 Kernel Core messageCenter Login to use system login:_	
	-
4	► //

Verifying that the LC/MS System Is Properly Grounded

When you use the EASY-nLC instrument as an inlet to an MS detector, the LC/MS system might require two or more fourplex outlets. The interconnected electrical outlets must have a common point to one ground connector. Connecting the hardware to external grounds at different potentials can create a ground loop that causes noise and interference and can damage the contact closure relays on the instrument's back panel.

IMPORTANT The EASY-nLC instrument, the MS or LC detector, and the (optional) data system hardware must have a common ground for several reasons:

- Improper grounding or no grounding can result in shock and fire hazards in case of instrument malfunction.
- Connecting the hardware to external grounds at different potentials can create a ground loop that causes noise and interference.
- Improper grounding or no grounding can damage the contact closure relays on the instrument's back panel. Fixing damaged relays requires the replacement of the instrument's built-in computer.



CAUTION Electrical safety regulations require that the EASY-nLC instrument be grounded to prevent electrical shock and a fire hazard in case of malfunction.

* To verify a common ground between the EASY-nLC instrument and external hardware

- 1. Connect the EASY-nLC instrument to line power, but do not turn on the instrument.
- 2. Using a multimeter, measure the resistance between the EASY-nLC chassis and the external hardware chassis (for example, the mass spectrometer chassis or the data system computer) as follows:
 - a. Connect one probe to an unpainted surface on the EASY-nLC chassis and connect the other probe to an unpainted surface on the external hardware chassis (see Figure 236).

Figure 236. Resistance measurement



Multimeter set up to take a resistance measurement

b. Ensure that the resistance measurement is from 0 to 10 ohms.

Calibrating the Autosampler's XYZ Robot

To calibrate the autosampler's XYZ robot, follow the instructions in this chapter.

Contents

- Replacing the Autosampler Adapter Plate
- Managing Plate Formats
- Preparing the Autosampler for the Calibration Routines
- Calibrating Plates
- Calibrating the Wash Bottle and Extra Vial Positions

For the ASC autosampler, the calibration covers the following areas of the autosampler.



Microtiter plate or vial plate

Wash solvent bottles



For the legacy ASA autosampler, the calibration covers the following areas of the autosampler.

Calibrate the autosampler in the following situations:

- When you want to use a new type of vial or microwell plate.
- When you would like to replace the needle.
- For problems with sample pickup; for example, the needle is not placed in the center of the well, or not all of the liquid is picked up from one or more of the vials.
- For problems with the wash/waste bottles; for example, the needle is not placed in the center of the bottle.

Replacing the Autosampler Adapter Plate

The EASY-nLC system ships with the vial adapter plate installed. The microtiter adapter plate comes in the accessory kit. To load microtiter (microwell) plates into the tray compartment, you must replace the vial adapter plate with the microtiter adapter plate.

The vial adapter plate holds 48 vials + 6 extra vials. The microtiter adapter plate holds one 96-well plate or one 384-well plate.

Follow the procedure for the autosampler model in your EASY-nLC system to switch between the two adapter plates as necessary.

- "Replacing the Adapter Plate in the ASC Model Autosampler" on page 271
- "Replacing the Adapter Plate in the ASA Model Autosampler" on page 272

Replacing the Adapter Plate in the ASC Model Autosampler

The ASC model is the current autosampler model for both the EASY-nLC II and the EASY-nLC 1000 systems.

* To replace the adapter plate in the ASC model autosampler

1. Press **Home > Overview**.

The Overview page of the Home menu opens.

2. Make sure that the area in front of the tray compartment is clear, and then press **Eject Plate**.

The tray compartment opens. Figure 237 shows the vial adapter plate mounted to the tray holder. The four studs on the bottom side of the plate fit snugly into the mounting holes on the tray holder.

Figure 237. Open tray compartment



3. Remove the current adapter plate by pulling it up and away from the tray holder (see Figure 238).

Figure 238. Removing the vial adapter plate before installing the microwell plate adapter plate



4. Install the other adapter plate in the autosampler.

Replacing the Adapter Plate in the ASA Model Autosampler

The ASA model autosampler is the original autosampler installed in the EASY-nLC II system.

* To replace the adapter plate in the ASA autosampler

- 1. Eject the tray as follows:
 - a. Press Home > Overview.
 - b. Press Eject Plate.

Figure 239 shows an ejected tray.

Figure 239. Tray compartment of the ASA model autosampler



2. Using the 2.5 mm hex wrench provided with your system, remove the screws that secure the plate to the tray.

The adapter plate is mounted with 4 or 6 hex screws as shown in Figure 240.

Figure 240. Screws that secure the adapter plate to the tray



3. Place the new adapter plate on the tray (see Figure 241).

Figure 241. Tray for the ASA model autosampler



- 4. Fasten the screws in the following balanced order:
 - a. Screw in the upper right corner.
 - b. Screw in the lower left corner.
 - c. Screw in the upper left corner.
 - d. Screw in the lower right corner.
 - e. Screws in the middle.

Note Mounting the screws in a balanced order can avoid distortion of the adapter plate.

The autosampler is now ready for new plate formats. For information about selecting a plate format and creating a new plate format, see "Managing Plate Formats" on page 274. After you create a new plate format, perform the calibration routines described in this chapter.

Managing Plate Formats

When the EASY-nLC system is initially installed, the application includes plate formats for vials and microtiter plates

Follow the appropriate procedure to use an existing plate format, create your own plate format, or delete a plate format that you no longer need.

- "Selecting a Plate Format," on this page
- "Creating a New Plate Format" on page 275
- "Deleting a Plate Format" on page 277

Selecting a Plate Format

* To select the plate format that you want to use

- 1. Open the Tools view for the autosampler as follows:
 - a. Press Maintenance > Devices.
 - b. In the Devices list, select the autosampler device.
 - c. Press the **Tools** tab.

Figure 242 shows the Tools view for the autosampler.

SCIENTIFIC	Home	Batch Setup	Method Editor	Maintenance	Configuration	
Scripts	Devices					
Log Book		Na	me	Туре		Add Device
Support	Co	oled Autosampler (A	SC) [Autosampler]	Autosampler		
Devices	EA	SY-nLC™ [HPLC]		HPLC		Remove Device
	Pis	ton Pump (flow feed)	back) [Pump A]	Piston Pump w/ flo	w fee 🔻	Assign Slave
	About	About Properties Tools				
	Cooled Autosampler (ASC) [Autosampler]					
Job 00:00:00	Plate	New	Calibrate Current	: Vials	Vials / V	Vash Calibrate
	62	k8 Vials			Manipul	ator
23 aug 2010 12:38 admin		Сору То	Dele e	Use		Reset
Exit						
Copy To Calibrate Selected plate format button button				nat		

Figure 242. Tools view for the autosampler

- 2. Select a plate format from the Plate list.
- 3. Press Use.

Tip If you are using a Thermo Scientific software application to control your liquid chromatograph/mass spectrometer (LC/MS) system, the plate selection for the EASY-nLC system in your Thermo Scientific instrument control software must match the touch-screen application selection.

Creating a New Plate Format

To create a new plate format

- 1. Open the Tools view for the autosampler as follows:
 - a. Press Maintenance > Devices.
 - b. In the Devices list, select the autosampler device.
 - c. Press the Tools tab.

Figure 242 shows the Tools view for the autosampler.

- 2. Do one of the following:
 - To create a new plate format, go to step 3.
 - To copy an existing plate format (copies the format selection), go to step 4.
- 3. Create a new plate format as follows:
 - a. Press New.

The Create New Plate Format dialog box opens (Figure 243).

Figure 243.	Create	New	Plate	Format	dialog	box
-------------	--------	-----	-------	--------	--------	-----

Create New Plate Format	
Create new plate format	
Format: 16x24	
Name: HT MTP	
Accept	Cancel

- b. In the Format list, select a plate format: 6×8 (vials), 8×12 (96-well plates), or 16×24 (384-well plates).
- c. In the Name box, enter a name for the new plate format.
- d. Press Accept.

The new plate name appears in the plate list with the text [uncalibrated] appended on the right.

- e. Go to "Calibrating Plates" on page 280 to calibrate the vial or well positions.
- 4. Copy an existing plate format as follows:
 - a. Select an existing plate format from the Plate list.
 - b. Press Copy To.

The Copy Plate Format dialog box opens.

- c. In the Name box, type a name for the new plate format.
- d. Press Accept.
- e. Go to "Calibrating Plates" on page 280 to calibrate the vial or well positions.

Deleting a Plate Format

✤ To delete a plate format

- 1. Open the Tools view for the autosampler on the Devices page of the Maintenance menu (see step 1 on page 275).
- 2. Select a plate format from the Plate list.

Note You cannot delete the plate in use. When you select the plate in use (highlighted in bold text), the Delete button becomes unavailable.

3. Press Delete.

The Delete Plate Data dialog box opens.

4. Press Accept.

Preparing the Autosampler for the Calibration Routines

To prepare the autosampler for the calibration routines, you must remove the left side panel from the instrument and load the appropriate adapter plate, vials or microtiter plate, wash bottle (W4) with insert, and (optional) extra vial into the tray compartment.



CAUTION Calibrating the autosampler requires removal of the side plate and visual inspection of the autosampler needle holder. Because the autosampler compartment contains moving parts and sharp needles, make sure to keep hands clear when operating the autosampler during calibration.

* To prepare the autosampler for the plate calibration routine

- 1. Install the appropriate adapter plate.
- 2. To calibrate the vial or well positions on a plate, do one of the following:
 - To calibrate the 6×8 vial positions, remove the vial caps from two vials and load them into the adapter plate in positions A1 and F8.
 - To calibrate a microwell plate, remove the mat or cover from the microwell plate and load the microwell plate onto the adapter plate.
- 3. Open the Tools view for the autosampler as follows:
 - a. Press Maintenance > Devices.
 - b. In the Devices list, select the autosampler.
 - c. Press the **Tools** tab.
- 4. In the Plate area, do the following:
 - a. Select the appropriate plate format from the list, or create a new plate format (To create and select plate formats, see "Managing Plate Formats" on page 274).
 - b. Press Use.
- 5. Remove the left side panel so that you can watch the movement of the needle.

To calibrate the plate, go to the next topic "Calibrating Plates."

* To prepare the autosampler for the V1 and W4 calibration routine

1. Place an empty vial in position V1.

The V1 position is not available for the microtiter plate formates.

- 2. Prepare the bottle for position W4 (used for ejecting waste and cleaning the outside of the injection needle).
- 3. For information about preparing the W4 wash bottle, refer to the *EASY-nLC Series Getting Started Guide*.

4. Install the wash bottle in position W4.

IMPORTANT For the autosampler to determine the appropriate depth for the needle, the wash bottle in position W4 must contain the needle wash insert.

5. Remove the left side panel so that you can watch the movement of the needle.

To calibrate the wash bottle and extra vial positions, go to "Calibrating the Wash Bottle and Extra Vial Positions" on page 285.

Calibrating Plates

For information about selecting or creating a plate format, see "Managing Plate Formats" on page 274.

IMPORTANT When calibrating the XYZ robot for vial or microwell plates, make sure to remove the vial caps or plate covers. Keeping the caps or covers on makes the needle susceptible to bending.

During a sample run, the *z*-axis needle holder moves to the specified sample position along the *xy*-plane. When the *z*-axis needle holder reaches the specified location, the needle descends along the *z*-axis into the sample vial or well.

Calibrating plates requires determining the *xyz* positions for the top left and bottom right vial or well. Table 27 lists the top left and bottom right positions for the three plate formats.

Plate format	Top left position	Bottom right position
6 × 8	A1	F8
8×12	A1	H12
16 × 24	A1	P24

Table 27. Top left and bottom right positions on the plate formats

To calibrate plates

- 1. If you have not already done so, prepare the autosampler for the plate calibration routine (see "To prepare the autosampler for the plate calibration routine" on page 278).
- 2. Open the Autosampler Plate Calibration dialog box as follows:
 - a. Press Maintenance > Devices.
 - b. In the Devices list, select the autosampler.
 - c. Press the **Tools** tab.

The Tools view opens (see Figure 244).

Figure 244. Tools view for the autosampler

About Properties Tools		
Cooled Autosampler (ASC) [Autosampler]		
Plate	Vials / Wash	
New Calibrate Current: Vials	Calibrate	
6x8 Vials		
Copy To Delete Use	Reset	

d. In the Plate area of the Tools view for the autosampler, press **Calibrate** (see Figure 244 on page 280).

The Autosampler Plate Calibration dialog box opens.

Figure 245 shows the dialog box at the beginning of the plate calibration routine. The top left and bottom right positions are highlighted in green. The position readbacks for the *xyz* axes are unpopulated, - / [-].



Figure 245. Plate view in the Autosampler Plate Calibration dialog box

3. Press the green vial or well in the upper left corner of the plate view (1).

The needle moves to the current saved coordinate. The saved coordinates for the x and y axis appear on the left side of the slash, the needle does not descend into the vial or well, and the *z*-axis value on the left side of the slash remains at 0.

The values on the left side of the slash change as you adjust the needle position.

- 4. Center the needle above the vial or well as follows:
 - Press the Needle In/Out (3) buttons to adjust the *x*-axis position.
 - Press the Needle Left/Right (4) buttons to adjust the *y*-axis position.



- 5. Using the step buttons in the Needle Up/Down area (5), adjust the needle height as follows:
 - a. Using the big step button $(\downarrow\downarrow)$, lower the needle until it reaches a level of 2–3 mm (0.08–0.12 in.) above the vial or well.
 - b. Using the small step button (\downarrow) , slowly lower the needle until it reaches the bottom of the vial or well.

Tip For the ASC autosampler, the O-ring at the top of the needle holder rises a little when the needle reaches the bottom of the vial or well (see Figure 246).

Figure 246. O-ring pushed slightly above the ASC needle holder



Observe when the O-ring moves up a bit, or gently hold needle tubing as you press the step button.

Tip For the ASA autosampler, do one of the following to determine the bottom position of the vial or well:

- To feel when the needle reaches the bottom of the vial or well, hold the needle gently as you press the down arrow on the touch-screen interface.
- To see when the needle reaches the bottom of the vial or well, draw a mark on the needle and observe the needle mark as you press the down arrow on the touch-screen interface. When the needle mark moves up a bit, the needle has reached the bottom of the vial. Figure 247 shows the ASA needle holder and needle.





Use a black marker pen to make a line on top of the PEEKsil tubing.

c. Press the small step button (1) once to raise the needle slightly off the bottom of the vial or well.

The appropriate needle height is one small step higher than the bottom of the vial or well.

6. When you are satisfied with the current *xyz*-coordinate for the vial or well, press **Capture**.

The color of the vial or well on the plate graphic changes from green to red and the position readbacks for the *xyz* axes are unpopulated, - / [-] (see Figure 248).

Autosampler Pla	ate Calibration	
Calibrate Plate HT	мтр	Needle Up/Down
<u>†</u> †		
<u>↑</u>		<u>↑</u>
- / [-]		- / [-]
Ļ		Ļ
++	W1 W2 W3 W4	++
Reset	- Needle Left/Right ← - / [-] → →	Capture
	Save Eject Close	

Figure 248. Plate calibration captured for the A1 position

7. Press the green vial or well in the lower right corner of the plate view.

The needle moves to the current saved coordinate. The saved coordinates for the x and y axis appear on the left side of the slash, the needle does not descend into the vial or well, and the *z*-axis value on the left side of the slash remains at 0.

- 8. Repeat step 4 through step 6 on that vial or well.
- 9. Press Save and then press Close to close the calibration dialog box.
- 10. Replace vial caps and plate covers as necessary.
- 11. Go to the next procedure to calibrate the W4 bottle position or reinstall the EASY-nLC instrument's left side panel.

IMPORTANT To maintain a stable temperature inside the tray compartment, the left side panel must be installed.

Calibrating the Wash Bottle and Extra Vial Positions

In addition to holding the sample trays, the EASY-nLC tray compartment holds four wash bottles (W1–W4) and six extra vials (V1–V6).

- * To calibrate the W4 wash bottle position, the extra vial positions, or both
- 1. If you have not already done so, prepare the autosampler for the extra vial and wash bottle calibration as described in "To prepare the autosampler for the V1 and W4 calibration routine" on page 278.
- 2. Open the Autosampler Vials/Wash Calibration dialog box as follows:
 - a. Press Maintenance > Devices.
 - b. In the Devices list, select the autosampler.
 - c. Press the **Tools** tab below the Devices list.
 - d. In the Vials/Wash area, press Calibrate.

The Autosampler Vials/Wash Calibration dialog box opens (see Figure 249).

3. Press the bottle container in W4 (1) (see Figure 249).

The needle moves to the current saved coordinate. The saved coordinates for the x and y axis appear on the left side of the slash, the needle does not descend into the bottle, and the *z*-axis value on the left side of the slash remains at 0.

Figure 249. Vials/Wash view in the Autosampler Vials/Wash Calibration dialog box



- 4. Center the needle above the bottle as follows:
 - Press the Needle In/Out (2) buttons to adjust the *x*-axis position.
 - Press the Needle Left/Right (3) buttons to adjust the *y*-axis position.



- 5. Using the step buttons in the Needle Up/Down area (5), adjust the needle height as follows:
 - a. Using the big step button $(\downarrow\downarrow)$, lower the needle until it reaches a level of 2–3 mm (0.08–0.12 in.) above the bottle.
 - b. Using the small step button (\downarrow) , slowly let the needle enter the hole and lower the needle until it reaches the bottom of the wash insert in the bottle.

Note The O-ring at the top of the needle holder rises a little when the needle reaches the bottom of the well.

c. Press the large step button $(\uparrow\uparrow)$ twice to raise the needle off the bottom of the wash bottle insert.

Two large steps higher than the bottom of the wash bottle insert is the appropriate needle height.

Note The ASA autosampler has a limit of 850 steps on the z axis (vertical). This limit is set to avoid bent needles. You might be able to go for more than 850 steps during calibration, but the needle does not go more than 850 steps during normal work.

- 6. Press **Capture** when you are satisfied with the current *xyz*-coordinate for position W4. The color of the bottle container number changes from green to red.
- 7. (Optional) To calibrate the extra vial positions for the 6 × 8 plate format, do the following:
 - a. Press V1 vial (5) to select it.
 - b. Repeat step 4 through step 6 for vial V1.
- 8. Press Save and then press Close to close the calibration dialog box.

Remote Support

The EASY-nLC instrument includes a remote support feature that enables Thermo Fisher Scientific Technical Support to diagnose and troubleshoot your EASY-nLC system remotely while the instrument is connected to the support server. When you connect the instrument to the remote server, you can also install the latest firmware file for the touch-screen application.

Technical Support cannot initiate the communication link between your EASY-nLC instrument and the support server. You must initiate this communication link from your EASY-nLC instrument.

To connect the EASY-nLC instrument to the support server or download system files to a removable storage device, follow these procedures.

Contents

- Connecting the EASY-nLC Instrument to the Support Server
- Saving System Files on a USB Removable Storage Device
- Downloading the Latest Firmware File

IMPORTANT To establish communication with the support server, you must first connect the EASY-nLC instrument directly to your local network, and then open the communication link for remote support through the touch-screen software. Thermo Fisher Scientific Technical Support cannot initiate this communication link.

The EASY-nLC instrument uses Secure Shell (SSH[™]), a network protocol for secure data communication, to connect to the support server, which uses the Linux[™] operating system.

To use the remote support feature, the firewalls for your local network must allow outgoing TCP/IP traffic from the EASY-nLC system to the support server at IP address 195.41.108.93 port 22.

When you establish the communication link with the support server, the support server triggers the file compression utility on the EASY-nLC instrument to build a zip file with the appropriate log files, and then retrieves the zip file from the EASY-nLC instrument. Meanwhile, the EASY-nLC instrument sends an e-mail with your supplied message and contact information to Thermo Fisher Scientific Technical Support.

After you send a message through the remote support feature, a member of the Thermo Fisher Scientific Technical Support team will contact you.

8

Connecting the EASY-nLC Instrument to the Support Server

Use this procedure to establish a communication link between the EASY-nLC instrument's computer and the support server.

* To open a communication link between the EASY-nLC system and the support server

1. Using a shielded Ethernet cable, connect the LAN port on the back panel of the EASY-nLC system (see Figure 250) directly to a laboratory LAN port that provides access to the Internet.

To use the remote support feature, the firewalls for your local network must allow outgoing TCP/IP traffic from the EASY-nLC system to the support server at IP address 195.41.108.93 port 22.

Figure 250. Ethernet connection between the EASY-nLC system and the laboratory LAN port



2. If you have not already set up the support server configuration, set it up as follows:

a. Press **Configuration > Network**.

The Network page of the Configuration menu opens (Figure 251).

Figure 251. Network page of the Configuration menu

Thermo SCIENTIFIC	Home Batch Setup Method Editor Maintenance Configuration
Users	LAN
Connections	Profile: Internet Configuration: O Dynamic O Fixed
Network	IP: 10.32.134.69 Subnet: 255.255.255.0
Data	Gateway: 10 . 32 . 134 . 254 DNS: 10 . 32 . 135 . 28
	Name: Ic-009808 Domain:
	Apply
	Support server
Job 00:00:35 Sample 00:00:24	IP: 195 . 41 . 108 . 93 Default IP
5 Mar 2012 16:27	Apply
admin Exit	

- b. In the LAN area, do the following:
 - i. Select **Internet** in the Profile list.
 - ii. Press Apply.
- c. In the Support Server area, do the following:
 - i. Enter the following IP address: **195.41.108.93**.
 - ii. Press Apply.

- 3. Send a support request message and a zip file containing the appropriate log files to Technical Support as follows:
 - a. Press Maintenance > Support.

The Support page of the Maintenance menu appears (see Figure 252).

Figure 252. Support page of the Maintenance menu

SCIENTIFIC	Home Batch Setup Method Editor Maintenance Configuration
Scripts	Connect to EASY-nLC support:
Log Book	Remote Port Number: 2000 Connect Disconnect Status: Not Connected
Support Devices	Message window (can be used to chat with support when connected):
	Log file copy:
	● To home directory 〇 To USB removable storage
	Copy Log Files Status: Idle
	Contact:
Job 00:00:00	Thermo Fisher Scientific easysupport@proxeon.com Edisonsvej 4 http://www.proxeon.com/hplc DK-5000 Odense C Phone: +45 6557 2300 Denmark Fax: +45 6557 2301
30 Aug 2010 14:14	
admin	
Exit	

b. In the Connect to EASY-nLC Support area, press Connect.

Depending on whether the instrument has the latest firmware file installed, one of these dialog boxes appears:

- If the instrument has the latest firmware file installed, the Message to Support Team dialog box appears (see Figure 253).
- If the instrument does not have the latest firmware file installed, the Upgrade is Available dialog box appears (see Figure 256 on page 296).
- c. If the Upgrade is Available dialog box appears, do one of the following:
 - Upgrade the software as described in "Downloading the Latest Firmware File" on page 296.

-or-

• Click **Skip**.

The Message to Support Team dialog box appears.

d. Select a support region, provide valid contact information, and describe the problem that you are experiencing.

Message to su	pport team
Support region:	
Name:	
Email address:	
Phone number:	
Enter message:	
Accept	Cancel

Figure 253. Message to Support Team dialog box

Note When you place the cursor in the Phone Number box, the program prompts you for a country code and phone number.

Tip When you place the cursor in the Email Address box, the program prompts you to enter an address in this format: *name@company*.com.

To enter the @ symbol, press **Shift**. The numbers row changes to the symbols row. Then press @.

e. Press Accept.

After you enter the requested information, the Status box on the right side of the Support page should change from Not Connected to Connected.

Status:	Connected!
---------	------------

- f. Depending on the connection status, do one of the following:
 - If the status changes from Not Connected to Connected, go to step 3g.
 - If the connection fails, see "To troubleshoot a failed connection to the support server" on page 292.

When the connection fails, the SSH Connection Error dialog box might appear with this message, "Not able to determine connection status. See message.log for details."

The message.log file is in the admin\systemLog folder on the EASY-nLC system. For information about accessing the admin folder from another computer on your local network, refer to the *EASY-nLC Series Getting Started Guide*.

While the message.log file might provide useful information to a member of Thermo Scientific Technical Support, users should follow the suggested troubleshooting procedure, "To troubleshoot a failed connection to the support server" on page 292.

g. After the status changes to Connected, allow enough time for the EASY-nLC system to create the zip file in the admin folder and for the support server to retrieve the zip file. Then go to step 4.

After you send a message with the appropriate contact information through the remote support feature, a member of the Thermo Fisher Scientific Technical Support team will contact you.

- 4. To close the communication link, press Disconnect.
- 5. To return the system to normal operation, do the following:
 - a. Reconnect the Ethernet cables as appropriate.

For information about connecting the EASY-nLC system to a data system computer, refer to the *EASY-nLC Series Getting Started Guide*.

- b. In the LAN area of the Configuration > Network page, do the following:
 - i. Select **LAN** in the Profile list.
 - ii. Press Apply.

* To troubleshoot a failed connection to the support server

- 1. Check the support server configuration (see step 2 on page 289).
- 2. Verify that the Ethernet cable is securely connected to both the LAN port on the back panel of the EASY-nLC system and the laboratory LAN port.
- 3. Verify that the Ethernet cable is working.
- 4. Ask your IT administrator if the network firewalls allow outgoing traffic to port 22 through an SSH connection.

Saving System Files on a USB Removable Storage Device

The EASY-nLC system includes network capability. Through the network you can back up your system, export and import batches and methods, analyze your system by examining the log files, and perform many other tasks. However, when you are not connected to the network or the network is broken, or you do not have a computer with network access that is near the EASY-nLC system, you can use the copy function to copy all the system files on the system to a USB removable storage device, such as a USB flash drive or memory stick.

The EASY-nLC system ships with a USB flash drive for your convenience. But you can use any USB storage device formatted with the file system FAT/FAT16 and at least 128 MB of free space.

✤ To copy system files to a USB storage device

1. Press Maintenance > Support.

The Support page of the Maintenance menu opens (see Figure 252 on page 290).

2. Insert the USB storage device into the connector panel on the back of the EASY-nLC instrument. Choose between one of two USB ports on the upper left side of the panel (see Figure 254).

Figure 254. USB port location on the back panel of the EASY-nLC instrument



3. In the Log File Copy area, select the To USB Removable Storage option.

Note When you select the To Home Directory option, the EASY-nLC system creates zip files in the admin folder.

For information about accessing the admin folder from another computer on your local network, refer to the *EASY-nLC Series Getting Started Guide*.

4. Press Copy Log Files.

When the copying is finished, the following confirmation appears.

Сору То USB		
1	Copied successfully! It is now safe to remove the USB storage.	
	Close	

- 5. Press Close and remove the USB storage device from the connector panel.
- 6. Insert the USB storage device into your computer.

Five zipped files appear on the USB storage device (see Figure 255 and Table 28 on page 295).

Figure 255. Windows XP view of the files on the USB memory stick



File name	Description
batchlog.zip	Contains one folder for each batch you have run with information on samples, methods, and EASY-nLC device performance.
etc.zip	Contains system configuration files.
export.zip	Contains exported batches and methods per user and system backup.
logfiles.zip	Contains system log files with information on overall system performance.
maintlog.zip	Contains one folder for each maintenance script you have run with information on EASY-nLC device performance.

If you have problems with your system, forward the files to Thermo Fisher Scientific Technical Support for assistance.

Downloading the Latest Firmware File

If the EASY-nLC instrument does not have the latest version of the touch-screen application, connect the instrument to the remote server and install the appropriate firmware files as described in the following procedure.

✤ To install the latest firmware file

- 1. If the instrument is not already connected to the remote server, connect the instrument as follows:
 - a. From the touch screen, press Maintenance > Support.

The Support page of the Maintenance menu appears.

b. In the Connect to EASY-nLC Support area, press Connect.

If the EASY-nLC instrument does not have the latest version of the touch-screen application, the Upgrade is Available dialog box appears.

Figure 256 shows the message for an instrument with firmware version 2.4.

Figure 256. Firmware upgrade message

\simeq An upgrade is available	×
The installed software can be upgraded to version "2.5#3032	2".
Note: the current version can be displayed by pressing the logo in the upper left corner of the application. You can find the Software Release Notes by entering the "EASY-nLC User Zone" from http://www.proxeon.com.	
Would you like to download an installable firmware file?	
A dialog will be shown when the transfers of the instrument log files and optional firmware file are completed.	
Download Skip	

2. Click Download.

When the file transfer is complete, the Transfer of Log and Firmware Files Completed dialog box appears.

Figure 257 shows the transfer information for an instrument with firmware version 2.4.

Figure 257. Information for upgrading from a previous version

	×
Transfers completed: The log files have been transferred to the remote support serv and the firmware file has been downloaded to the instruments firmware folder.	er
To upgrade the application software you should now shut down and power-cycle the instrument. When the instrument restarts double-tap the 'Upgrade' button on the desktop.	n
Note: There are 4 further upgrades to download and install after this one. After the downloaded upgrade has been installed you should connect to the Remote Support Server again to fetch the next upgrade.	d
Ok	

- 3. Click OK.
- 4. To begin the firmware upgrade, do the following:
 - a. Press **Exit** in the lower-left corner of the touch screen.

A confirmation dialog box opens (see Figure 22 on page 21).

b. Press Power Down.

The EASY-nLC application displays a white screen with a small progress bar. When the progress bar is completely filled, a message appears indicating you can safely turn off the instrument.

- c. Turn off the instrument, and then turn it back on.
- d. When the Upgrade button appears, double-tap it.
- 5. If the complete upgrade requires more firmware files, reconnect to the server and install the upgrade files as described in step 1 through step 4 of this procedure.

Table 29 lists the software versions that you must install to upgrade the software to the current version.

 Table 29.
 Software installation matrix

Current version	Install versions
2.4 #2923	2.5 #3032, 2.7.8.1, 2.8.11.1, and 3.1.4.1
2.5 #3032	2.7.8.1, 2.8.11.1, and 3.1.4.1
2.7.8.1	2.8.11.1 and 3.1.4.1
2.8.11.1	3.1.4.1
3.1.4.1	current as of January 2013

Returning the EASY-nLC Instrument

If it is necessary to return the EASY-nLC system to the factory, follow the instructions in this chapter.

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Contents

- Transport Instructions
- Declaration of Contamination

Transport Instructions

- To prepare the instrument for transport
- 1. Press Home > Overview.
- 2. Press Eject Plate.
- 3. Remove the plates/tubes.
- 4. Remove the waste beaker and the wash/waste bottles.
- 5. Press Insert Plate.
- 6. Close down the system (see "Closing Down the EASY-nLC Instrument" on page 21).
- 7. Remove the cables on the back side.

Tip For the original ASA autosampler only: Secure the autosampler by using the transport collars (see Figure 258) before shipping the instrument. (This is not necessary on the latest ASC model.)



Figure 258. (ASA autosampler only) Transport collar location

 Insert two transport collars after closing down the instrument.

- 8. Fill out the Declaration of Contamination of Equipment in "Declaration of Contamination" on page 301.
- 9. Place the EASY-nLC instrument in the original shipping container.
Declaration of Contamination

To return, service, or repair the EASY-nLC instrument, you must complete and submit a "Declaration of Contamination of Equipment" to Thermo Fisher Scientific with the returned equipment. Qualified personnel must review the declaration. Contact Thermo Fisher Scientific for additional copies of this form or if you have any questions regarding the contents of this declaration.

Description of equipment

Equipment type/model:		Serial No.:	
Date of receiving/purchasing equipr	nent:		
Reason for return:	[] Maintenance	[] Repair	[] End of test
	[]		
Describe symptoms and problems: _			
Equipment condition			
Has the equipment been used?		[] Yes	[] No
Has the equipment been exposed to	potentially harmfu	l substances?	
		[] Yes	[] No
If yes, attach list of all known harmfu precautions associated with the subs	ıl substances includ tances.	ling chemical name	and symbol, and
Were any of the harmful substances			
Radioactive?		[] Yes	[] No
• Toxic?		[] Yes	[] No
 Corrosive? 		[] Yes	[] No
 Explosive? 		[] Yes	[] No
Has the equipment been properly de	econtaminated and	/or cleaned before l	being returned?
		[] Yes	[] No
Legally binding declaration			
I hereby declare that the information judge any contamination level.	n supplied on this f	orm is accurate and	l sufficient to

Name:		
Job title:	Organization:	
Address:		
Telephone:	Fax:	E-mail:
Signature:		

Error Codes

Table 30 lists the possible error codes.

Table 30. Error codes (Sheet 1 of 8)

No.	Issue code name	Description	Cause	Solution
1	ERROR_IO_CLOSING_ FILE	Failed closing log file	File system error	Internal filesystem error - Replace the hard drive.
2	ERROR_IO_WRITING_ TO_FILE	IOException	I/O error occurred	Internal filesystem error - Replace the hard drive.
3	ERROR_IO_NO_FILE_ OPEN	Log was closed trying to log the following message:	Filesystem error	Internal filesystem error - Replace hard drive
4	ERROR_IO_CREATE_ FILE	Failed creating log file 'LOG_FILE_NAME'	I/O error occurred	Internal filesystem error - Replace the hard drive.
5	ERROR_IO_CREATE_ FOLDER	Failed creating log folder 'LOG_FOLDER_NAM E'	I/O error occurred	Internal filesystem error - Replace the hard drive.
6	ERROR_IO_FILE_ EXISTS	Failed creating log 'LOG_FILE_NAME AND_PATH' (file already exists)	Filesystem error	Internal filesystem error - Replace the hard drive.
7	ERROR_IO_FILE_CAN NOT_WRITE	Failed creating log "LOG_FILE_NAME_ AND_PATH' (file not writable)	Filesystem error	Internal filesystem error - Replace the hard drive.
8	ERROR_BATCH_BEIN G_EDITED	It appears the current batch is being edited, please close or reschedule it.	[see issue description]	Close or reschedule the batch.
9	ERROR_DEVICE_ ACCESS_FAILED	Failed accessing device DEVICE_ID	Missing the hardware component driver	Add the device to the list on the Maintenance > Devices page.

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Table 30. Error codes (Sheet 2 of 8)

No.	Issue code name	Description	Cause	Solution
10	ERROR_PREP_ GRADIENT_ PRESSURE_BUILD_ FAILED	Pump B unable to reach 90% A pressure within 30 µL. Please check for air/leaks!	Pump unable to reach set pressure during prep gradient	Check that solvent bottle is not empty and check for air and leaks in the system.
11	ERROR_PUMP_ STOPPED_EMPTY	stopped (out of solvent).	Out of solvent	Refill the solvent reservoir bottle. Check for air and leaks in the system.
12	ERROR_PUMP_ STOPPED_ UNEXPECTEDLY	stopped unexpectedly (unknown cause).	Pump hardware issue	Check the pump functionality.
26	ERROR_QUEUE_ ELEMENT_BEING_ EDITED	Current queue element is being edited; stopping.	Trying to execute a job currently being edited	Close or reschedule the batch.
27	ERROR_PBUS_IN_ SERVICE_MODE	P-BUS interface in service mode.	Trying to initialize the RS485 interface while serial port is in the service mode	Check that the RS485 switch on PC is in the ON position.
28	ERROR_PBUS_ INTERFACE_INIT	Initialization of P-BUS interface failed	[see issue description]	Troubleshoot P-BUS communication - Check that the cables are properly connected.
29	ERROR_PBUS_ Send_bytes	Error writing to interface COMM_PORT	[see issue description]	Troubleshoot P-BUS communication - Check that the cables are properly connected.
30	ERROR_PBUS_ READ_BYTES	Error reading from interface COMM_PORT	[see issue description]	Troubleshoot P-BUS communication - Check that the cables are properly connected.
31	ERROR_SENSIRION_ Factor_parsing	Failed parsing flow sensor factor from INFO_COMMAND	Could not parse some string to a double	Cycle EASY nLC power.
32	ERROR_SENSIRION_ FACTOR_NOT_ FOUND	Flow sensor factor not found in info output. Factor unknown.	Error getting flow sensor scaling factor	Cycle EASY nLC power.

Table 30. Error codes (Sheet 3 of 8)

No.	Issue code name	Description	Cause	Solution
33	ERROR_ SENSIRION_ INIT	Port COM_PORT does not support reading/Error getting input stream from flow sensor port COM_PORT	Error getting input from flow sensor	Cycle EASY nLC power.
34	ERROR_SENSIRION_ NO_DATA	No data received from flow sensor for <i>X</i> seconds	[see issue description]	Cycle EASY nLC power / Check the flow sensor functionality.
35	ERROR_SENSIRION_ IN_MEASURE_MODE	Flow sensor in measurement mode. Stop measurements before sending further commands	Trying to start a flow sensor that is already started	Cycle EASY nLC power.
36	ERROR_SENSIRION_ IO	IO Error while communicating with flow sensor	[see issue description]	Check the flow sensor communication/cable.
37	ERROR_SENSIRION_ TIMEOUT	No response from flow sensor (timeout)	[see issue description]	Cycle EASY nLC power - If the problem persists, replace the flow sensor. See "Replacing a Flow
				Sensor" on page 92.
38	ERROR_SENSIRION_ ECHO_MISMATCH	Flow sensor echo "ECHO" does not match command "COMMAND"	Flow sensor fails to "talk back"	Rare error - Connect the EASY-nLC system to remote support.
				See "Connecting the EASY-nLC Instrument to the Support Server" on page 288.
39	ERROR_SENSIRION_ BAD_RESPONSE	Flow sensor error: ERROR_MESSAGE/Flo w sensor did not respond with OK, but did not report an error either	Flow sensor failing, but does not report it	Rare error - Connect the EASY-nLC system to remote support.
40	ERROR_SENSIRION_ IN_IDLE_MODE	Cannot calibrate sensor in idle mode	Flow sensor in idle mode during calibration	Cycle EASY nLC power. Check the flow sensor functionality.

Table 30. Error codes (Sheet 4 of 8)

No.	Issue code name	Description	Cause	Solution
41	ERROR_SENSIRION_ Calibration	Cannot store flows larger than 9999 nL/min	Abnormal value trying to be written to the flow sensor	Cycle EASY nLC power. Check flow sensor functionality
42	ERROR_RS232_INIT	Initialization of interface "PORT_NAME" failed	Access to interface failed	Cycle EASY nLC power.
54	ERROR_RS485_ Command_verify	COMPONENT_ID COMMAND failed: ERROR_MESSAGE	Error occurred when sending COMMAND	Troubleshoot P-BUS communication/hardware.
55	ERROR_ MANIPULATOR_ UNKNOWN_RACK_ SHELF	Unknown rack shelf RACK_SHELF_VALUE	Unknown rack shelf position supplied to driver	Verify that the sample position is correct.
56	ERROR_ MANIPULATOR_ UNKNOWN_RACK_ POS_ID	Unknown rack position id RACK_POSITION_ID_ VALUE	Could not move to bad rack position	Verify that the sample position is correct.
57	ERROR_ MANIPULATOR_SET_ GRID_CONFIG L	Grid configuration change failed (AUTOSAMPLER_ RETURN_CODE)	Error uploading autosampler plate calibration (0 = asrOK, 1 = asrBusy, 2 = asrBadLocation, 3 = asrBadCoordinate, 4 = asrEjected, 5 = asrAxisError, 6 = asrChecksumError)	Calibrated position not recognized by software. See Chapter 7, "Calibrating the Autosampler's XYZ Robot."
58	ERROR_ MANIPULATOR_SET_ WASH_CONFIG	Wash configuration change failed (AUTOSAMPLER_ RETURN_CODE)	Error while saving autosampler wash calibration (see code 57 for asr codes)	Calibrated position not recognized by software. See Chapter 7, "Calibrating the Autosampler's XYZ Robot."
59	ERROR_ MANIPULATOR_SET_ VIAL_CONFIG	Vial configuration change failed (AUTOSAMPLER_RET URN_CODE)	Error while saving autosampler vial calibration (see code 57 for asr codes)	Calibrated position not recognized by software. See Chapter 7, "Calibrating the Autosampler's XYZ Robot."

Table 30. Error codes (Sheet 5 of 8)

No.	Issue code name	Description	Cause	Solution
60	ERROR_ MANIPULATOR_TRAY _EJECT	tray eject failed (AUTOSAMPLER_RET URN_CODE)	see code 57 for asr codes	Verify that the correct plate is configured in the EASY-nLC software and the Xcalibur VI. Check that no objects in the tray compartment are obstructing the XYZ robot movement. Reset manipulator and run the Torque script for the ASC model autosampler.
61	ERROR_ MANIPULATOR_TRAY _LOAD	tray load failed (AUTOSAMPLER_RET URN_CODE)	see code 57 for asr codes	(see above)
62	ERROR_ MANIPULATOR_GRID _ADDRESS	grid address failed (AUTOSAMPLER_RET URN_CODE)	Could not move to sample position (see code 57 for asr codes)	(see above)
63	ERROR_ MANIPULATOR_ WASH_ADDRESS	wash address failed (AUTOSAMPLER_RET URN_CODE)	Could not move to wash position (see code 57 for asr codes)	(see above)
64	ERROR_ MANIPULATOR_VIAL_ ADDRESS	vial address failed (AUTOSAMPLER_RET URN_CODE)	Could not move to vial position (see code 57 for asr codes)	(see above)
65	ERROR_ MANIPULATOR_ NEEDLE_DOWN	needle down failed (AUTOSAMPLER_RET URN_CODE)	Could not move needle down (see code 57 for asr codes)	(see above)
66	ERROR_ MANIPULATOR_ NEEDLE_UP	needle up failed (AUTOSAMPLER_ RETURN_CODE)	Could not move needle up (see code 57 for asr codes)	(see above)

Table 30. Error codes (Sheet 6 of 8)

No.	Issue code name	Description	Cause	Solution
69	ERROR_VALVE_SET_ POSITION	COMPONENT_ID setPosition failed: Unrecognized position POSITION/ COMPONENT_ID setPosition failed: Problems switching to position POSITION/ COMPONENT_ID needed ATTEMPTS attempts switching to position POSITION	Valve unable to switch position	Clean the rotor seal and the stator. Replace the rotor seal if necessary. If the problem persists, do the following: • For valve serial number < V-009800, run the Valve Tune script. • For valve serial number > V-009800, replace the valve.
70	ERROR_PARALLEL_ PORT_WRITE		Contact closure port IO error	Contact Thermo Fisher Scientific.
71	ERROR_PARALLEL_ PORT_NOT_ INITIALIZED		Contact closure port IO error	Contact Thermo Fisher Scientific.
72	ERROR_PARALLEL_ PORT_MASS_SPEC_ BUSY		Contact closure port IO error	Contact Thermo Fisher Scientific.
73	ERROR_PARALLEL_ PORT_MASS_SPEC_ TIMEOUT		Contact closure port IO error	Contact Thermo Fisher Scientific.
74	ERROR_PARALLEL_ PORT_INIT		Contact closure port IO error	Contact Thermo Fisher Scientific.
75	ERROR_PARALLEL_ PORT_WRONG_PORT		Contact closure port IO error	Contact Thermo Fisher Scientific.
77	ERROR_DEVICE_LOW _24_VOLT_SUPPLY	DEVICE: Low voltage (VALUE V)	[see issue description]	Faulty 24V supply - Connect to remote support
78	ERROR_ MANIPULATOR_INIT	DEVICE: Init failed (not ready within 1 minute)	[see issue description]	Autosampler hardware issue. Cycle EASY-nLC power. If the problem persists, check the P-bus cables to the autosampler.
79	ERROR_DEVICE_SELF _TEST	DEVICE: Status bit(s) set after self test: 0xBITS_IN_HEX	Error returned from hardware component firmware	Connect the system to the remote support server (see Chapter 8).

Table 30. Error codes (Sheet 7 of 8)

No.	Issue code name	Description	Cause	Solution
80	ERROR_DEVICE_IN_ BOOT_MODE	DEVICE: Device in boot mode after device reset	Error returned from hardware component firmware because a PCB is damaged.	Contact Thermo Fisher Scientific.
81	ERROR_PELTIER_ SPURIOUS_ TEMPERATURE	DEVICE: Spurious temperature (TEMPERATURE_IN_ DEGREES_CELSIUS)	Cooler sensor defective	Replace the cooler. See "Replacing the Autosampler Cooler" on page 137.
82	ERROR_DEVICE_LOW _CURRENT	DEVICE: Low current consumption (CURRENT_VALUE A)	Peltier element defective	Replace the cooler.
83	ERROR_VALVE_ SPURIOUS_ANGLE	DEVICE: Spurious angle in position POSITION (ANGLE)	[see issue description]	For valve serial number < V-009800, run the Valve Tune script.
84	ERROR_APPLICATION _STATE	HISTORY_TRACE	Application state empty or illegal	Cycle EASY nLC power.
85	IC_STATUS_FLAG_ WARNING		Monitor reported flag(s) raised (log file only).	Act according to the error message.
86	IC_STATUS_FLAG _SEVERE		Monitor reported flag(s) raised (in user interface, current run stopped).	Act according to the error message.
87	IC_LC_CONDITION_ UNSATISFIED	Waiting for some condition during run unsuccessful.	Failed reaching a specific flow, pressure, etc. within a specified volume/period of time.	Troubleshoot the LC system for leaks and air. See "Troubleshooting the Results of the System Leak Test" on page 221.
88	IC_STATUS_FLAG_ ERROR		Monitor reported flag(s) raised (in user interface).	Act according to the error message.

Table 30. Error codes (Sheet 8 of 8)

No.	Issue code name	Description	Cause	Solution
89	IC_VALVE_MULTIPLE_ ATTEMPTS	Needed 2 attempts to switch to position 1-6	A switching valve needed multiple attempts to reach the target position.	Clean the rotor seal and the stator. Replace the rotor seal if necessary. If the problem persists, do the following: • For valve serial number < V-009800, run the Valve Tune script. • For valve serial number > V-009800, replace the valve.
90	IC_DEVICE_ UNSUPPORTED_ OPERATION		A device type operation was attempted on a device instance that doesn't support it.	Contact Thermo Fisher Scientific.
92	IC_BUSINESSLOGIC_ REFILL_DURING_ GRADIENT	Gradient interrupted by pump refill warning	Pump solvent volume too small for next gradient step.	No action is necessary. This is just a warning that the pumps were refilled during gradient processing.
93	IC_SCRIPT_AUTO_ ABORT	Maintenance script self-abort	Probably because a pump ran out of solvent while building/maintaining pressure	Check for air and leaks in the subsystem. See "Troubleshooting the Results of the System Leak Test" on page 221.

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Mobile Phase Viscosity

Table 31 lists the viscosities for two-solvent mobile phases consisting of a water/methanol mixture or a water/acetonitrile mixture.

	ղ ₂₅ (cP)	
Mobile phase (%v organic/water)	Methanol	Acetonitrile
0	0.89	0.89
10	1.18	1.01
20	1.40	0.98
30	1.56	0.98
40	1.62	0.89
50	1.62	0.82
60	1.54	0.72
70	1.36	0.59
80	1.12	0.52
90	0.84	0.46
100	0.56	0.35
Defense D.D. Hannes A.H. Dilling		and of Character and has

Table 31. Mobile-phase viscosity at 25 °C for reversed-phase gradients

Reference: D.P. Herman, A.H. Billiet, and L. de Galan, *Journal of Chromatography*, 463 (1989) 1

Consumables and Replacement Parts

To order consumables and replacement parts for the EASY-nLC system, see the ordering information and solvent system schematics in this chapter.

Contents

- Ordering Information
- Solvent System Schematic for the EASY-nLC II Instrument
- Solvent System Schematic for the EASY-nLC 1000 Instrument
- Common Replacement Parts

Ordering Information

To obtain ordering information for the EASY-nLC instrument's spare parts and accessories, go to the following Web sites:

• For the EASY-nLC 1000 instrument, go to:

http://www.proxeon.com/productrange/nano_lC_easy-nlc_1000/accessories_spares

• For the EASY-nLC II instrument, go to:

http://www.proxeon.com/productrange/nano_lC/accessories_spares

These EASY-nLC parts are consumable parts and not covered by the normal first year Limited Warranty offered by Thermo Fisher Scientific, or any other service contract agreement containing extended warranty coverage.

- All flow lines including Tee-pieces and fittings (nuts, ferrules, sleeves, and valve stops)
- Check valves
- · Solvent filters, inline filters, and all associated filter holders
- Pump piston seals
- Valve rotors and stators
- All bottles and lids
- All sample vials, microtiter plates, and associated lids and mats
- Columns
- Fuses
- Autosampler needle

Solvent System Schematic for the EASY-nLC II Instrument

Table 32 lists the tubing requirements for the EASY-nLC II instrument. The numbers in the ID column correspond to the numbers in the solvent system schematic shown in Figure 259.

Kit LC230 includes the Teflon tubing and the check valve assembly required to replace the low-pressure solvent system for one pump. Kit LC243 includes a complete set of flow lines to the mixing Tee, but does not include the mixing Tee itself.

Table 32. Tubing requirements for the EASY-nLC II system

No.	Connections	Tubing	Part number
1	Pump A outlet to pressure sensor A inlet	PEEK, 300 µm ID, 15 cm length	LC212
	Pump S outlet to pressure sensor S inlet		
	Pressure sensor A outlet to port 1 of valve A		
	Pressure sensor S outlet or port 2 of valve S		
2	Pump B outlet to pressure sensor B inlet	stainless steel, prebent	LC215
3	Pressure sensor B outlet to port 1 of Valve B	stainless steel, prebent	LC216
4	Valve A to flow sensor A	PEEKsil, 50 µm ID, 25 cm length	LC222
	Valve B to flow sensor B		
5	Flow sensor A to mixing Tee	PEEKsil, 20 µm ID, 21 cm length	Kit LC243
6	Flow sensor B to mixing Tee	Fused silica, 10 µm ID, 15 cm length	
7	Mixing Tee to valve S	PEEKsil, 30 µm ID, 13 cm length	
8	Autosampler needle connected to port 1 of valve S	alve S PEEKsil, 150 μm ID, 55 cm length	
		ASA model	LC251
		ASC model	LC302
9	Column Out tubing connected to port 3 of valve S	PEEKsil, 30 µm ID, 37 cm length	LC260
10	Waste In line, venting Tee to port 2 of valve W	PEEKsil, 75 µm ID, 50 cm length	LC262
11	Port 2 of valve A to check valve A	Teflon, 500 µm ID, 15 cm length	Kit,
	Port 2 of valve B to check valve B		Low-pressure
12	Tubing from check valve A to solvent bottle A Tubing from check valve B to solvent bottle B	Teflon, 500 µm ID, 37 cm length	solvent lines LC230
13	Tubing from check valve A to the waste beaker	Teflon, 500 µm ID, 39 cm length	
	Tubing from check valve B to the waste beaker	-	
14	Tubing from valve W to the waste beaker	Teflon, 500 µm ID, 27 cm length	LC263



Figure 259. Solvent system schematic for the EASY-nLC II system

Solvent System Schematic for the EASY-nLC 1000 Instrument

The pump and valve compartment behind the right panel of the EASY-nLC instrument contains all the flow lines between the components. In addition to pumps and valves, the solvent delivery and waste system includes several filters and four check valves. The two check valves on each solvent line ensure that solvent is drawn from the solvent bottles and ejected to waste without switching valve positions.

Table 33 lists the tubing requirements and identifies the tubing components shown in Figure 260. The Low-Pressure Solvent Line Kit (P/N LC230) contains the Tees, check valves, and low-pressure solvent lines to connect one pump (A or B) to the waste system and to its corresponding solvent bottle and six-port rotary valve. The EASY-nLC 1000 Flow Line Kit (P/N LC570) contains all of these flow lines. The stainless steel tubing is prebent.

Table 33. Tubing requirements for the EASY-nLC 1000 system

No.	Connections	Tubing	Part number
1	Pump outlet to pressure sensor inlet	Stainless steel, 250 µm ID, 150 mm length	LC512
2	Pressure sensor outlet to valve A or B	Stainless steel, 250 µm ID, 150 mm length	LC513
3	Pressure sensor outlet to valve S	Stainless steel, 250 µm ID, 150 mm length	LC514
4	Mixing Tee to valve S, valve A to flow sensor A, valve B to flow sensor B, flow sensor A to mixing Tee	nanoViper, 20 μm ID, 350 mm length	LC522
5	Flow sensor B to mixing Tee	nanoViper, 10 µm ID, 180 mm length	LC543
6	Autosampler needle connected to port 1 of valve S	PEEKsil [™] , 150 µm ID, 550 mm length	LC302
7	Column Out tubing connected to port 3 of valve S	nanoViper, 20 µm ID, 550 mm length	LC560
8	Waste In line, venting Tee to port 2 of valve W	nanoViper, 75 μm ID, 550 mm length	LC562
9	Sample loop, 20 µL	nanoViper, 250 µm ID, 410 mm length	LC472
D	Port 2 of valve A to check valve A Port 2 of valve B to check valve B	Teflon™, 500 µm ID, 150 mm length	kit LC230
E	Tubing (2) from check valves to solvent bottles	Teflon, 500 µm ID, 390 mm length	kit LC230
F	Tubing (2) from check valves to waste beaker	Teflon, 500 µm ID, 390 mm length	kit LC230
G	Tubing from valve W to waste beaker	Teflon, 500 µm ID, 330 mm length	LC263



Figure 260. Solvent system schematic for the EASY-nLC 1000 system

Common Replacement Parts

For proper maintenance of your EASY-nLC instrument, Thermo Fisher Scientific recommends that you maintain an inventory of replacement parts.

- Common Replacement Parts for the EASY-nLC II Instrument
- Common Replacement Parts for the EASY-nLC 1000 Instrument

Common Replacement Parts for the EASY-nLC II Instrument

Table 34 lists the annual consumption of common replacements parts for the EASY-nLC II instrument.

Description	Part number	Quantity
Pump Piston Seal Replacement Kit (contains four piston seals and the piston seal tool)	LC210	4 piston seals (1 per pump) + 1 extra
Valve rotor seal for valve serial numbers V-009999 and below (contains one rotor seal)	LC224	8 rotor seals (2 per valve)
Valve rotor seal for valve serial numbers V-010000 and above (contains one rotor seal)	LC228	8 rotor seals (2 per valve)
Column Out solvent line	LC260	2
Waste In solvent line	LC262	1
Autosampler needle, ASA model	LC251	1
Autosampler needle, ASC model	LC302	2
Valve to flow sensor line (contains two flow sensor lines)	LC222	2
Replacement filter discs (contains two 10 µm filter discs)	LC232	2 filter discs (1 per solvent bottle)

Table 34. Common replacement parts for the EASY-nLC II instrument

Common Replacement Parts for the EASY-nLC 1000 Instrument

Table 35 lists the annual consumption of common replacements parts for the EASY-nLC 1000 instrument.

Table 35. Common replacement parts for the EASY-nLC 1000 instrument

Description	Part number	Quantity
Pump Piston Seal Replacement Kit (contains four spring-energized piston seals and the piston seal tool)	LC510	4 piston seals (1 per pump) + 1 extra
Valve rotor seal (contains one rotor seal)	LC228	8 rotor seals (2 per valve)
Column Out solvent line	LC560	2
Waste In solvent line	LC562	1
Autosampler needle, ASC model	LC302	2
Flow sensor filters (contains four flow sensor filters)	LC542	4 flow sensor filters (2 per flow sensor)

Quick Reference Guides for Routine Maintenance

This appendix contains a set of quick reference guides for the routine maintenance procedures that you can perform to keep the EASY-nLC system in optimal working condition. These guides are the condensed versions of the procedures in the Routine Maintenance chapter.

If you have replaced a PLF pump in the EASY-nLC II instrument with a PLU pump, follow the instructions in "Maintaining the PLU Pump Quick Reference Guide" on page 327 to maintain the new PLU pump.

Contents

- Using nanoViper Fittings Quick Reference Guide
- Maintaining the PLU Pump Quick Reference Guide
- Maintaining the PLF Pump Quick Reference Guide
- Maintaining the Rotary Valves Quick Reference Guide
- Replacing a Damaged Autosampler Needle

Using nanoViper Fittings Quick Reference Guide

For the EASY-nLC 1000 instrument, most of the plumbing connections use nanoViper fittings (see Figure 1). Figure 1. nanoViper fitting



Even though nanoViper fittings can withstand UHPLC back pressures of up to ~ 1034 bar (~ 15000 psi), they are fingertight fittings, which require only very small torques to seal. To avoid damage by overtightening, follow the instructions in this guide.

Contents

- Connecting nanoViper Fittings
- nanoViper Tubing Connections
- Returning the System to Atmospheric Pressure

To connect a nanoViper fitting to a receiving port

- 1. Insert the nanoViper fitting into the receiving port. Then turn the screw clockwise until you feel resistance.
- 2. Using the black knurled nut, turn the screw clockwise to an angle between 0 and 45 degrees (1/8-turn).
- 3. Run the system leak test (see "Running a System Leak Test" on page 218).

When the leak test ends, the system is at atmospheric pressure.

IMPORTANT To extend the lifetime of the nanoViper fittings, open and close connections at atmospheric system pressures only. Opening and closing connections at high system pressures can reduce the lifetime of the fitting system.

4. If the Leaks script fails because the new connection is not leak tight, use the black knurled nut to turn the screw up to an additional 45 degrees. Do not turn the screw beyond an angle of 90 degrees from where you felt the initial resistance.

IMPORTANT To prevent damage to the sealing surface of the nanoViper fitting, take care not to overtighten the fingertight nanoViper fitting.



Connecting nanoViper Fittings

nanoViper Tubing Connections

Figure 2 shows the nanoViper tubing connections in the EASY-nLC 1000 instrument.

- Port 6 of valve A to flow sensor A inlet
- Port 6 of valve B to flow sensor B inlet
- Flow sensor A outlet to mixing Tee inlet
- Flow sensor B outlet to mixing Tee inlet
- Mixing Tee outlet to port 4 of valve S
- Sample loop connected to ports 2 and 5 of valve S
- Column Out line connected to port 3 of valve S and the HPLC union
- Waste In line connected to port 2 of valve W and the HPLC union or the venting Tee

Figure 2. nanoViper tubing connections in the EASY-nLC 1000 instrument



Returning the System to Atmospheric Pressure

Upstream of Valve S Solvent Lines Before you disconnect a nanoViper fitting from its receiving port, always return the solvent line to atmospheric pressure. The high-pressure solvent lines in the EASY-nLC solvent system can be divided into two categories based on their position relative to valve S.

- Upstream of Valve S Solvent Lines
- Downstream of Valve S Solvent Lines

The following solvent lines (numbered from left to right in Figure 3) are in the flow path upstream of valve S.

#	Solvent line	#	Solvent line
1	Port 6 of valve A to flow sensor A inlet	4	Flow sensor B outlet to mixing Tee inlet
2	Port 6 of valve B to flow sensor B inlet	5	Mixing Tee outlet to port 4 of valve S
3	Flow sensor A outlet to mixing Tee inlet	6	Sample loop connected to ports 2 and 5 of valve S

* To return the solvent lines that are upstream of valve S to atmospheric pressure

- 1. On the touch screen, press Home > Overview.
- 2. Using the valve controls, set the valves to the following positions.

Valve	Position	Effect
A and B	Center	Maintains the pressure between the pump A and valve A during the Analytical Column Equilibration script.
S and W	1–6	Vents the system pressure downstream of valve A.

Figure 3 shows the flow path when valves A and B are Centered and valves S and W are in position 1–6.

Figure 3. Solvent lines that are downstream of valves A and B and upstream of valve S, numbered 1–6



Downstream of Valve S Solvent Lines

The following solvent line connections are in the flow path downstream of valve S:

- Column Out tubing connected to port 3 of valve S and the HPLC union (P/N SC900)
 - Waste In tubing connected to port 2 of valve W and the HPLC union or the venting Tee

* To return the solvent lines that are downstream of valve S to atmospheric pressure

- 1. On the touch screen, press Home > Overview.
- 2. Using the valve controls, set the valves to the following positions.

Valve	Position	Effect
S	Center	Maintains the system pressure upstream of valve S.
W	1–6	Vents the system pressure downstream of valve S.

Figure 4 shows the Column Out and Waste In solvent lines that are downstream of valve S.

Figure 4. Solvent lines that are downstream of valve S



Maintaining the PLU Pump Quick Reference Guide

Replace the piston seal when you detect a leak in the pump.

Contents

- Removing a Used Piston Seal
- Installing a New Piston Seal

Replacing a piston seal requires these tools and materials.

Tools	Materials
3 mm hex wrench	Pipette
Piston seal tool	Powder-free safety gloves
1/4 in. open-ended wrench	LC/MS-grade methanol
#2 Phillips head screwdriver	LC/MS-grade acetonitrile
	Piston seal, P/N LC510

Removing a Used Piston Seal

- 1. Set the pump piston to the fully retracted position as follows:
 - a. On the touch screen, press Maintenance > Scripts.
 - b. In the Category list, select Prepare.
 - c. In the Name list, select Purge Solvent.
 - d. Press the **Parameters** tab.
 - e. Press **Purge Iterations** and enter **0**.
 - Figure 1. Purge Solvent script set to 0 iterations



f. Select the check box for the appropriate pump.

g. Press Start.

The piston moves downward to the 140 μL position.

- 2. Exit the EASY-nLC 1000 system and turn off the power.
- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument. Then remove the panel.



CAUTION Wear powder-free gloves when handling the wetted components of the LC system.

- 4. Remove the used piston seal as follows:
 - a. Using a 1/4 in. open-ended wrench, remove the stainless steel tubing connected to the pump head.

Figure 2. Stainless steel fitting removed from the pump head



b. Using a 3 mm hex wrench, remove the two screws that secure the pump head to the pump body.

Figure 3. Pump head removed from the pump body



- c. Clean the visible portion of the piston with a lint-free tissue soaked in LC/MS-grade methanol and visually inspect the piston for scratches. Ensure that no solvent runs into the pump.
- d. Remove the old seal from the pump head with great care. Do not use any metal tools, as they can scratch the inside of the pump head and generate leaks.



Installing a New Piston Seal

- 1. With the spring side of the piston seal spring facing away from the piston seal tool, mount the new piston seal onto the piston seal tool.
- 2. Using a pipette, remove the air from the piston seal spring by carefully filling the rim of the piston seal with methanol.

Figure 4. Filling the piston seal with methanol



3. Place the pump house on the guide rods and evenly press it all the way down to insert the seal and then gently remove the house when the seal is in position.

Figure 5. Inserting the piston seal into the pump head



- 4. Reassemble the pump head on the pump using the two screws and a 3 mm hex wrench. Do not connect the stainless steel tubing to the pump head.
- 5. Turn on the EASY-nLC system and log in as an administrator.
- 6. Fill the pump head with the appropriate solvent as follows:
 - a. Press Home > Overview.
 - b. Press the pump icon for the pump that you want to control. The Position readback displays the current pump position

from 0 µL (empty) to 140 µL (full).

c. Set the flow rate to $300~\mu\text{L/min}$ and the volume to $140~\mu\text{L}.$ Then press Start.

The piston moves up to the top of the pump head. **Figure 6.** Pump dialog box

unip A	
	300 µl/min
140.0 µI	Stop Star
Pressure:	53.0 Bar
Position:	27.75 µl
Real flow:	332.6 nl/min

d. Pipette the appropriate solvent into the pump head.Figure 7. Adding solvent to the pump head



e. Set flow rate to $-300~\mu\text{L/min}$ and the volume to $140~\mu\text{L}.$ Then press Start.

The piston moves down, drawing solvent into the pump head. Make sure the pump head is filled with solvent during the entire retraction of the piston.



CAUTION Be careful not to spill solvent onto the printed circuit board behind the LED panel.

- 7. Using a 1/4 in. open-ended wrench, reconnect the tubing to the pump head (see Figure 2 on page 327).
- 8. Reinstall the right side panel.
- 9. Reset the usage counter for the pump as follows:
 - a. Press Maintenance > Devices.
 - b. Select the pump from the list of devices.
 - c. Press the **Summary** tab.
 - d. Press Reset.

The value in the Intermediate Volume box resets to 0.

- 10. To draw fresh solvent through the solvent lines and to remove air, do the following:
 - a. Open the Parameters page for the Purge Solvent script as described in step 1 on page 327.
 - b. In the Purge Iterations box, enter 10.
 - c. Select the check box for the appropriate pump.
 - d. Press **Start** and wait for the system to perform 10 purge cycles.
- 11. To remove air from the pump head, do the following:
 - a. In the Name category on the Maintenance > Scripts page, select Flush Air.
 - b. Press the Parameters tab.
 - c. In the Flush Threshold $[\mu L]$ box, enter 12.
 - d. Press **Start**.
 - e. Wait for the script to finish.
- 12. Run the Leaks script for the pump with the new piston seal
 - a. On the Maintenance > Scripts page, select **Test** in the Category list, and then select **Leaks** in the Name list.
 - b. Press the Parameters tab.
 - c. Select the check box for the pump with the new seal.
 - d. Press Start.
- 13. . When the instrument is leak tight, it is ready for use.

Maintaining the PLF Pump Quick Reference Guide

Replace the piston seal when you detect a leak in the pump.

Contents

- 1. Removing the Pump Head and Cleaning the Piston
- 2. Removing the Worn Piston Seal
- 3. Installing a New Piston Seal
- 4. Priming the Pump
- 5. Preparing the Instrument for Operation



CAUTION Wear powder-free safety gloves when working with solvents and handling the wetted components of the instrument.

Replacing a piston seal requires these tools and materials.

Tools	Materials
3 mm hex wrench	Pipette
Piston seal tool	Powder-free safety gloves
1/4 in. open-ended wrench	LC/MS-grade methanol
#2 Phillips head screwdriver	LC/MS-grade acetonitrile
	Piston seal, P/N LC210 (includes the piston seal tool)

Removing the Pump Head and Cleaning the Piston

- 1. Set the pump piston to the fully retracted position as follows:
 - a. On the Maintenance > Scripts page, Select Prepare in the Category list and then select Purge Solvent in the Name list.
 Press the Parameters tab. In the Purge Iterations box, enter 0.
 Select the check box for the appropriate pump.
 - b. Press Start. The piston moves down to the 140 μL position.
- 2. Exit the EASY-nLC II system and turn off the power.
- 3. Using a #2 Phillips head screwdriver, with a quarter-turn loosen the three captive screws that secure the right side panel to the instrument. Then remove the panel.
- 4. Remove the tubing connected to the pump head as follows:
 - If a PEEK fitting is connected to the pump head, use a 13 mm open-ended wrench, to remove it.

Figure 1. PEEK fitting on the externally threaded pump head



 PEEK fitting connected to an externally threaded pump head • If a stainless steel fitting is connected to the pump head, use a 1/4 in. open-ended wrench to remove it.

Figure 2. Stainless steel fitting on the internally threaded pump head



Stainless steel fitting connected to an internally threaded pump head

Using a 13 mm open-ended wrench, remove the pump head.
 Figure 3. Removing the pump head



- 6. Place the piston in the fully extended position as follows:
 - a. Turn on the instrument and log in as an administrator.
 - b. Press **Home > Overview**, and then press the pump icon for the pump you want to control. The Pump dialog box opens.
 - c. Set the flow rate to $300~\mu\text{L/min}$ and the volume to $140~\mu\text{L}.$
 - d. Press Start.
- 7. Clean the piston with the lint-free tissue soaked in methanol, and visually inspect the piston for any scratches.



CAUTION Take care to avoid solvent drips onto the pump PCB.

- 8. Return the piston to the fully retracted position as follows:
 - a. In the Pump dialog box, set the flow rate to $300~\mu L/min$ and the dispense volume to $140~\mu L.$
 - b. Press Start.
- 9. When the piston is fully retracted, close down the EASY-nLC system and turn off the power.



Removing the Worn Piston Seal

Clean the piston seal tool in a beaker filled with 100% methanol.
 Figure 4. Two-component piston seal tool for the PLF model pump



2. Insert the piston seal tool into the worn piston seal and pull the piston seal out of the pump head.

Figure 5. Using the piston seal tool to remove the worn piston seal



Worn piston seal

Installing a New Piston Seal

Insert the guide tube into the pump head flange.
 Figure 6. Guide tool inserted into the pump head flange



Guide tool

- 2. Using a pipette, fill the groove in the piston seal with methanol.
- 3. Place the new seal with the groove facing up on the piston seal tool.

Figure 7. Piston seal mounted onto the piston seal tool



Insert the piston seal tool into the guide tool.
 Figure 8. Inserting the piston seal tool into the guide



- 5. Push the piston seal tool into the guide until you feel resistance.
- 6. Remove the guide tube and the piston seal tool, and then check that the piston seal is properly seated.
- 7. Using a 13 mm open-ended wrench, reconnect the pump head to the pump body. Do not connect the stainless steel tubing to the pump head.

Priming the Pump

- 1. Turn on the EASY-nLC instrument and log in as an administrator.
- 2. Fill the pump head with the appropriate solvent as follows:
 - a. Press **Home > Overview**.
 - b. Press the pump icon for the pump that you want to control.
 - c. Set the flow rate to $300~\mu L/min$ and the volume to $140~\mu L.$ Then press Start.

The piston moves upward to the top of the pump head.

- d. Pipette the appropriate solvent into the pump head.
- e. Set the flow rate to $-300~\mu\text{L/min}$ and the volume to $140~\mu\text{L}.$ Then press Start.

The piston moves down, drawing solvent into the pump head. Make sure the pump head is filled with solvent during the entire retraction of the piston.



CAUTION Be careful not to spill solvent onto the printed circuit board behind the LED panel.

3. Using a 1/4 in. open-ended wrench, reconnect the tubing to the pump head.

Preparing the Instrument for Operation

- 1. Reinstall the right panel.
- 2. Reset the usage counter for the pump as follows:
 - a. Press Maintenance > Devices.
 - b. Select the pump from the list of devices.
 - c. Press the **Summary** tab.
 - d. Press **Reset**.

The value in the Intermediate Volume box resets to 0.

- 3. To draw fresh solvent through the solvent lines and to remove air, do the following:
 - a. Open the Parameter page for the Purge Solvent script.
 - b. In the Purge Iterations box, enter 10.
 - c. Select the check box for the appropriate pump.
 - d. Press **Start** and wait for the system to perform 10 purge cycles.
- 4. To flush air from the pump head, do the following:
 - a. In the Name category on the Maintenance > Scripts page, select Flush Air.
 - b. Press the Parameters tab.
 - c. In the Flush Threshold $\left[\mu L\right]$ box, enter 10.
 - d. Press **Start**.
 - e. Wait for the script to finish.
- 5. Run the Leaks script for the pump as follows:
 - a. On the Maintenance > Scripts page, select **Test** in the Category list, and then select **Leaks** in the Name list.
 - b. Press the Parameters tab.
 - c. Select the check box for the pump with the new seal.
 - d. Press Start.
- 6. When the instrument is leak tight, it is ready for use.

Maintaining the Rotary Valves Quick Reference Guide

Replace the rotor seal when you detect a leak or blockage in the valve.

Contents

- Replacing a Rotor Seal
- Replacing the Stator



CAUTION Wear powder-free safety gloves when working with solvents and handling the wetted components of the instrument.

Replacing a piston seal requires these tools and materials.

Tools	Materials
9/64 in. hex wrench1/4 in. open-ended wrench	 Rotor seal, P/N LC228 (EASY-nLC system valves with serial numbers V-010000 and above)
• #2 Phillips head screwdriver	• Rotor seal, P/N LC224 (EASY-nLC II system valves with serial numbers below V-009999)
	 LC/MS-grade methanol
	Powder-free safety gloves

Replacing a Rotor Seal

- 1. Close down the EASY-nLC system, and then turn off the power to the instrument.
- 2. Using a #2 Phillips screwdriver, with a quarter-turn loosen the three captive screws that secure the panel to the right side of the EASY-nLC instrument, and then remove the panel.
- 3. Using a 9/64 in. L-hex wrench, remove the two hex screws that secure the stator to the valve assembly, and then pull the stator away from the valve driver.
- 4. Remove the rotor seal from the valve driver.

Figure 1. Rotor seal mounted on the valve driver



5. Clean the stator with a lint-free tissue or Q-tip soaked in methanol.

Figure 2. Cleaning the stator



- 6. Carefully mount the new rotor onto the valve driver, ensuring that the rotor sealing surface (engraved flow passage), is facing out.
- 7. Mount the stator onto the valve driver.
- 8. Using a 9/64 in. L-hex wrench, tighten the two hex screws a little at a time by shifting from one screw to the other and back again until the screws are evenly torqued.
- 9. Reconnect the right side panel.
- 10. Turn on the EASY-nLC instrument and log in as an administrator.
- 11. Reset the usage counter for the valve as follows:
 - a. Press Maintenance > Devices.
 - b. Select the valve from the list of devices.
 - c. Press the **Summary** tab.
 - d. Press **Reset**.
 - The value in the Rotor Shifts box resets to 0.
- 12. If you replaced the rotor seal in Valve A or B, flush air from the system as follows:
 - a. On the Maintenance > Scripts page, select **Prepare** in the Category list. Then select **Flush Air** in the Name list.
 - b. Select the pump that is connected to the affected valve.
 - c. Set the flush volume threshold to $10~\mu L$ for the EASY-nLC II or $12~\mu L$ for the EASY-nLC 1000.
 - d. Press Start.
 - e. Wait for the script to finish.
- 13. Run the Leaks script as follows:
 - a. Select **Test** in the Category list. Then select **Leaks** in the Name list.
 - b. Do one of the following:
 - If you replaced the rotor seal in Valve A or B, run the Leaks script for the affected valve.
 - If you replaced the rotor seal in Valve S or W, run the Leaks script for the system.

When the system is leak tight, the instrument is ready for use.



Replacing the Stator

- Close down the EASY-nLC system, and then turn off the power to the instrument (see "Closing Down the EASY-nLC Instrument" on page 21).
- 2. Using a #2 Phillips screwdriver, loosen the three quarter-turn screws that secure the panel to the right side the EASY-nLC instrument, and then remove the panel.
- 3. Disconnect the solvent lines from the valve as follows:
 - Use a 1/4 in. open-ended wrench to remove stainless steel fittings.
 - Use the black nanoViper knurled nut to remove nanoViper fittings.
- 4. Using a 9/64 in. L-hex wrench, remove the two hex screws that secure the stator to the valve driver, and then pull the stator away from the valve driver.
- 5. Mount the new stator onto the valve driver.
- 6. Using a 9/64 in. L-hex wrench, tighten the two hex screws a little at a time by shifting from one screw to the other and back again until the screws are evenly torqued.
- 7. Reconnect the solvent lines to the valve as follows:
 - Use a 1/4 in. open-ended wrench to tighten stainless steel fittings.
 - Use the black nanoViper knurled nut to tighten nanoViper fittings.
- 8. Reconnect the right side panel to the instrument. Tighten the three screws with a #2 Phillips screwdriver.
- 9. Turn on the EASY-nLC instrument and log in as an administrator.
- 10. If you replaced the stator in Valve A or B, run the Flush Air script for the valve with the replacement stator.
- 11. If you replaced the stator on Valve A or B, run the Leaks script for the valve with the replacement stator. If you replaced the stator on Valve S or W, run the Leaks script for the system.

Replacing a Damaged Autosampler Needle

Replace the autosampler needle when it is bent or clogged.

Contents

- Removing the Damaged Autosampler Needle
- Installing a New Autosampler Needle



CAUTION Wear powder-free safety gloves when working with solvents and handling the wetted components of the instrument.

Replacing a damaged needle requires these tools and materials.

Tools	Materials
• 1/4 in. open-ended wrench	Powder-free safety gloves
• #2 Phillips head screwdriver	• ASA autosampler needle, P/N LC251
	 ASC autosampler needle, P/N LC302

Removing the Damaged Autosampler Needle

To remove the autosampler needle

- 1. Using a #2 Phillips screwdriver, with a quarter-turn loosen the three captive screws that secure the panel to the right side the EASY-nLC instrument, and then remove the panel.
- 2. Using a 1/4 in. open-ended wrench, unscrew the nut that secures the autosampler needle to port 1 of valve S. Then remove the fitting from the port.

Figure 1. Valve S solvent line connections for the EASY-nLC II system



Figure 2. Valve S solvent line connections for the EASY-nLC 1000 system



Remove the nut, ferrule, and sleeve from the needle tubing.
 Figure 3. Autosampler tubing with a nut, ferrule, and sleeve at one end



- 4. Move the *z*-axis needle holder to an accessible location within the tray compartment by doing one of the following:
 - For the ASA model, go to step 5.
 - For the ASC model, go to step 6.
- 5. For the ASA model, move the *z*-axis needle holder to position A1 as follows:
 - a. Press Home > Overview.
 - b. Press the autosampler icon.
 - c. The Autosampler dialog box opens.

d. In the XYZ Robot area, select position $\ensuremath{A1}$ in the Well box.

Figure 4. Autosampler direct control dialog box



e. Press Goto.

f. Go to step 7.



- 6. For the ASC model, move the *z*-axis needle holder to position W1 as follows:
 - a. Press Home > Overview.
 - b. Press the autosampler icon.
 - The Autosampler dialog box opens.
 - c. In the XYZ Robot area, press Go to Calibration.

The Tools page of the Autosampler view on the Maintenance > Devices page opens.

Figure 5. Tools page of the Autosampler view on the Maintenance > Devices page

huoc	Properties	Tools	
oled	Autosampler	(ASC) [Autosampler]	Male (Wash
a.c		alberta Courset and State	Calibrate
		Current an 2469	Manipulator
8x1	2/Abgene (AB-0	00)	Reset
Co	ру То	Delete	
-			

d. In the Manipulator area, press **Reset**.

The *z*-axis needle holder moves to the W1 position.

- 7. Using a #2 Phillips screwdriver, loosen the three quarter-turn screws that secure the panel to the left side of the EASY-nLC instrument, and then remove the panel.
- 8. Do one of the following:
 - For the ASC model, remove the black needle holder from the slot in the panel that separates the solvent system hardware from the autosampler compartment. Removing the fitting requires some gentle movement from both sides of the plate. **Figure 6.** Removing the black needle holder from the slot in the panel



- For the ASA model, remove the white plastic needle guide on the back of the touch-screen monitor.
- 9. Loosen the nut that is connected to the *z*-axis needle holder by turning it counterclockwise. Then carefully pull the autosampler needle upward and away from the holder.

Figure 7. Autosampler needle connected to the z-axis needle holder

ASA z-axis needle holder

ASC z-axis needle holder





Installing a New Autosampler Needle

To install an new autosampler needle

- 1. Mount the new autosampler needle in the *z*-axis holder. Check that the small spring is situated between the PEEK nut and the plastic stop.
- 2. Do one of the following:
 - For the ASC autosampler, go to step step 3.
 - For the ASA autosampler, go to step step 4.
- 3. For the ASC autosampler, do the following:
 - a. Guide the end of the needle that connects to Valve S through the large hole in the metal plate that separates the tray compartment from the solvent system compartment.
 - b. Install the black needle holder in the side plate.
- 4. For the ASA autosampler, slide the valve end of the tubing through the small plastic holder on the back of the touch-screen monitor.

Figure 8. Holder on the back of the touch-screen monitor



Holder

- 5. Connect the needle to port 1 of Valve S as follows:
 - a. Slide the provided blue sleeve and metal fittings onto the tubing.
 - b. To ensure a zero dead volume connection, hold the blue sleeve and the tubing firmly against the bottom of the valve port, and then tighten the fitting with a 1/4 in. open-ended wrench.
- 6. Recalibrate the needle position.
- 7. Press Maintenance > Scripts.
- 8. In the Category list, select Prepare.
- 9. In the Name list, select **Purge Solvent**. Then run 2 iterations of the Purge Solvent script for Pump S.
- 10. In the Name list, select Flush Air. Then run the Flush Air script for Pump S with a flush volume threshold of 10 μ L for the EASY-nLC II system or 12 μ L for the EASY-nLC 1000 system.

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