

Thermo

# **EASY-nLC Series**

### **Troubleshooting and Maintenance Guide**

Touch-screen application version 3.2

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

Software version: Touch-screen application version 3.2. For information about upgrading your instrument's touch-screen application, see "Downloading the Latest Firmware File" on page 315.

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### **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.

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.

For manufacturing location, see the label on the instrument.



### **WEEE Compliance**

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



Thermo Fisher Scientific is registered with B2B Compliance (B2Bcompliance.org.uk) in the UK and with the European Recycling Platform (ERP-recycling.org) in all other countries of the European Union and in Norway.

If this product is located in Europe and you want to participate in the Thermo Fisher Scientific Business-to-Business (B2B) Recycling Program, send an email request to weee.recycle@thermofisher.com with the following information:

- WEEE product class
- Name of the manufacturer or distributor (where you purchased the product)
- Number of product pieces, and the estimated total weight and volume
- Pick-up address and contact person (include contact information)
- Appropriate pick-up time
- Declaration of decontamination, stating that all hazardous fluids or material have been removed from the product

For additional information about the Restriction on Hazardous Substances (RoHS) Directive for the European Union, search for RoHS on the Thermo Fisher Scientific European language websites.

**IMPORTANT** This recycling program is **not** for biological hazard products or for products that have been medically contaminated. You must treat these types of products as biohazard waste and dispose of them in accordance with your local regulations.



### **Conformité DEEE**

Ce produit est conforme avec 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 sociétés de recyclage dans chaque état membre de l'Union Européenne et ce produit devrait être collecté ou recyclé par celle(s)-ci. Pour davantage d'informations, rendez-vous sur la page www.thermoscientific.fr/rohs.

### **WEEE Konformität**

Dieses Produkt entspricht der EU Waste Electrical & Electronic Equipment (WEEE) Richtlinie 2002/96/EC. Es ist mit dem folgenden 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. Weitere Informationen finden Sie unter www.thermoscientific.de/rohs.

#### CAUTION Symbol CAUTION



**Risk electric shock:** This instrument uses voltages that can cause electric shock and/or personal injury. Before servicing, shut down the instrument and disconnect it from line power. While operating the instrument, keep covers on. Do not remove the protective covers from the printed circuit board assemblies (PCBAs).



**Chemical hazard:** Wear gloves and other protective equipment, as appropriate, when handling toxic, carcinogenic, mutagenic, corrosive, or irritant chemicals. Use approved containers and proper procedures to dispose of waste oil and when handling wetted parts of the instrument.



Hot surface: Before touching, allow any heated components to cool.

**Flammable substances hazard:** Use care when operating the system in the presence of flammable substances.

**Risk of eye injury:** Eye injury could occur from splattered chemicals, airborne particles, or sharp objects. (Sharp objects that customers might install in the instrument include fused-silica tubing, the autosampler needle, and so on.) Wear safety glasses when handling chemicals or servicing the instrument.



**General hazard:** A hazard is present that is not included in the other categories. This symbol also appears on the instrument. For details about the hazard, refer to the instrument manual. When the safety of a procedure is questionable, contact Technical Support for Thermo Scientific San Jose products.

#### VORSICHT

**Stromschlaggefahr:** Dieses Gerät arbeitet mit Spannungen, die Stromschläge und/oder Personenverletzungen verursachen können. Vor Wartungsarbeiten muss das Gerät abgeschaltet und vom Netz getrennt werden. Betreiben Sie das Gerät nicht mit abgenommenen Abdeckungen. Nehmen Sie die Schutzabdeckungen von Leiterplatten nicht ab.

Gefahr durch Chemikalien: Tragen Sie beim Umgang mit toxischen, karzinogenen, mutagenen, ätzenden oder reizenden Chemikalien Schutzhandschuhe und weitere geeignete Schutzausrüstung. Verwenden Sie bei der Entsorgung von verbrauchtem Öl und beim Umgang mit medienberührenden Komponenten die vorgeschriebenen Behälter, und wenden Sie ordnungsgemäße Verfahren an.

Heiße Oberflächen: Lassen Sie heiße Komponenten vor der Berührung abkühlen.

Gefahr durch entzündbare Substanzen:

Beachten Sie die einschlägigen Vorsichtsmaßnahmen, wenn Sie das System in Gegenwart von entzündbaren Substanzen betreiben.

Augenverletzungsrisiko: Verspritzte Chemikalien, Schwebstoffpartikel oder scharfe Objekte können Augenverletzungen verursachen. (Scharfe Objekte, die Kunden möglicherweise im Gerät installieren, sind z. B. Quarzglas-Kapillaren, die Nadel des Autosamplers, usw.) Tragen Sie beim Umgang mit Chemikalien oder bei der Wartung des Gerätes eine Schutzbrille.

Allgemeine Gefahr: Es besteht eine weitere Gefahr, die nicht in den vorstehenden Kategorien beschrieben ist. Dieses Symbol wird auch auf dem Gerät angebracht. Einzelheiten zu dieser Gefahr finden Sie in den Gerätehandbüchern. Wenn Sie sich über die Sicherheit eines Verfahrens im Unklaren sind, setzen Sie sich, bevor Sie fortfahren, mit dem technischen Support für Thermo Scientific San Jose Produkte in Verbindung. **Peligro por sustancias inflamables:** Tenga mucho cuidado cuando utilice el sistema cerca de sustancias inflamables.

Riesgo de descargas eléctricas: Este instrumento

eléctricas v/o lesiones personales. Antes de revisar o

reparar el instrumento, apáquelo y desconéctelo de la

red eléctrica. Mantenga colocadas las cubiertas

mientras se utiliza el instrumento. No retire las

Peligro por sustancias químicas: Cuando

siempre recipientes homologados y siga los

Superficies calientes: Antes de tocar los

componentes calientes, espere a que se enfríen.

manipule sustancias químicas, tóxicas,

cubiertas protectoras del circuito impreso completo

carcinogénicas, mutágenas, corrosivas o irritantes,

utilice guantes y otro equipo de protección. Utilice

procedimientos adecuados cuando deseche aceite

residual o manipule partes moiadas del instrumento.

utiliza voltajes que pueden causar descargas

PRECAUCIÓN

(PCBA).

**Riesgo de lesiones oculares:** Las salpicaduras de sustancias químicas, las partículas flotantes en el aire y los objetos afilados pueden causar lesiones oculares. (Entre los objetos afilados que los clientes pueden instalar en el instrumento se encuentran tubos de sílice fundida, agujas del muestreador automático, etc.). Para manipular sustancias químicas o realizar tareas de mantenimiento, utilice gafas de seguridad.

Peligro general: Existen peligros que no se incluyen en las otras categorías. Este símbolo también aparece en el instrumento. Si desea obtener más información sobre estos peligros, consulte el manual del instrumento.

En caso de duda sobre la seguridad de un procedimiento, póngase en contacto con el personal de servicio técnico de los productos Thermo Scientífic San Jose.

#### MISE EN GARDE

**Risque de choc électrique :** l'instrument utilise des tensions susceptibles de provoquer une électrocution et/ou des blessures corporelles. Il doit être arrêté et débranché de la source de courant avant toute intervention. Ne pas utiliser l'instrument sans ses couvercles. Ne pas enlever les capots de protection des cartes à circuit imprimé (PCBA).

Danger lié aux produits chimiques : porter des gants et d'autres équipements de protection appropriés pour manipuler les produits chimiques toxiques, cancérigènes, mutagènes, corrosifs ou irritants. Utiliser des récipients homologués et des procédures adéquates pour la mise au rebut des huiles usagées et lors de la manipulation des pièces de l'instrument en contact avec l'eau.

**Surface chaude :** laisser refroidir les composants chauffés avant toute manipulation.

**Danger lié aux substances inflammables :** agir avec précaution lors de l'utilisation du système en présence de substances inflammables.

**Risque de lésion oculaire :** les projections chimiques, les particules en suspension dans l'air et les objets tranchants peuvent entraîner des lésions oculaires. (Les objets tranchants pouvant être installés par les clients dans l'instrument comprennent les tubes en silice fondue, les aiguilles du passeur automatique, etc.). Porter des lunettes de protection lors de toute manipulation de produit chimique ou intervention sur l'instrument.

**Danger d'ordre général :** indique la présence d'un risque n'appartenant pas aux catégories citées plus haut. Ce symbole figure également sur l'instrument. Pour plus de détails sur ce danger potentiel, se reporter au manuel de l'instrument.

Si la sûreté d'une procédure est incertaine, contacter l'assistance technique pour les produits Thermo Scientific San Jose.

Fla

AUTION Symbol	CAUTION	VORSICHT	PRECAUCIÓN	MISE EN GARDE
	<b>Laser hazard:</b> This instrument uses a laser that is capable of causing personal injury. This symbol also appears on the instrument. For details about the hazard, refer to the instrument manual.	<b>Gefahr durch Laserstrahlen:</b> Der in diesem Gerät verwendete Laser kann zu Verletzungen führen. Dieses Symbol wird auch auf dem Gerät angebracht. Einzelheiten zu dieser Gefahr finden Sie in den Gerätehandbüchern.	<b>Peligro por láser:</b> Este instrumento utiliza un láser que puede producir lesiones personales. Este símbolo también aparece en el instrumento. Si desea obtener más información sobre el peligro, consulte el manual del instrumento.	<b>Danger lié au laser :</b> l'instrument utilise un laser susceptible de provoquer des blessures corporelles. Ce symbole figure également sur l'instrument. Pour plus de détails sur ce danger potentiel, se reporter au manuel de l'instrument.
	<b>Ultra violet light hazard:</b> Do not look directly at the ultra-violet (UV) light or into the UV source. Exposure can cause eye damage. Wear UV eye protection.	<b>Gefahr durch UV-Licht:</b> Richten Sie Ihren Blick nicht direkt auf ultraviolettes Licht (UV-Licht) oder in die UV-Quelle. Dies kann zu Augenschäden führen. Tragen Sie eine UV-Schutzbrille.	<b>Peligro por luz ultravioleta:</b> No mire directamente a una luz ultravioleta (UV) ni a una fuente UV. La exposición puede causar daños oculares. Lleve protección ocular para UV.	<b>Danger lié aux rayons ultraviolets :</b> ne jamais regarder directement la lumière ultraviolette (UV) ou la source d'UV. Une exposition peut entraîner des lésions oculaires. Porter des protections oculaires anti-UV.
	<b>Sharp object:</b> Avoid physical contact with the object.	<b>Scharfes Objekt:</b> Vermeiden Sie den physischen Kontakt mit dem Objekt.	<b>Objeto puntiagudo:</b> Evite el contacto físico con el objeto.	<b>Objet tranchant :</b> éviter tout contact physique avec l'objet.
	Pinch point: Keep hands away from this area.	<b>Quetschgefahr:</b> Halten Sie Ihre Hände von diesem Bereich fern.	Puntos de pinzamiento: Mantenga las manos apartadas de esta área.	Risque de pincement : éloigner les mains de cette zone.
	<b>Heavy objects:</b> Never lift or move the instrument by yourself; you can suffer personal injury or damage the equipment. For specific lifting instructions, refer to the instrument manual.	Schweres Objekt: Bewegen und heben Sie das Gerät niemals allein an; dies kann zu Verletzungen oder zur Beschädigung des Geräts führen. Spezifische Anweisungen zum Anheben finden Sie im Gerätehandbuch.	<b>Objeto pesado:</b> Nunca levante ni mueva el instrumento por su cuenta, podría sufrir lesiones personales o dañar el equipo. Para obtener instrucciones específicas sobre levantamiento, consulte el manual del instrumento.	<b>Objet lourd :</b> ne jamais soulever ou déplacer l'instrument seul sous peine de blessure corporelle ou d'endommagement de l'instrument. Pour obtenir des instructions de levage spécifiques, se reporter au manuel de l'instrument.
A	<b>Trip obstacle:</b> Be aware of cords, hoses, or other objects located on the floor.	Stolpergefahr: Achten Sie auf Kabel, Schläuche und andere Objekte auf dem Fußboden.	Tropiezo con obstáculos: Tenga en cuenta los cables, mangueras u otros objetos colocados en el suelo.	<b>Risque de trébuchement :</b> faire attention aux câbles, tuyaux et autres objets situés sur le sol.
	When the safety of a procedure is questionable, contact Technical Support for Thermo 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 Scientific San Jose Produkte in Verbindung.	En caso de duda sobre la seguridad de un procedimiento, póngase en contacto con el personal de servicio técnico de los productos Thermo Scientífic San Jose.	Si la sûreté d'une procédure est incertaine, contacter l'assistance technique pour les produits Thermo Scientific San Jose.

CAUTION Symbol	CAUTION	<b>警告</b>	危险警告
	<b>Risk electric shock:</b> This instrument uses voltages that can cause electric shock and/or personal injury. Before servicing, shut down the instrument and disconnect it from line power. While operating the instrument, keep covers on. Do not remove the protective covers from the printed circuit board assemblies (PCBAs).	<b>感電の危険性</b> : この機器では、感電および/または身体傷害を引き起こ すおそれのある電圧を使用しています。整備点検の前には、機器の電 源を切り、電源コードを抜いてください。機器の作動中は、カバーを 付けたままにしてください。プリント基板アセンブリ (PCBA) から保護 カバーを取り外さないでください。	<b>触电危险:</b> 本仪器所用电压可能导致电击或人身伤害。进行维修服务前,务必关闭仪器电源并断开其电源连接。操作此仪器时,不要卸下顶盖。勿卸下印刷电路板组件 (PCBA)的保护盖。
	<b>Chemical hazard:</b> Wear gloves and other protective equipment, as appropriate, when handling toxic, carcinogenic, mutagenic, corrosive, or irritant chemicals. Use approved containers and proper procedures to dispose of waste oil and when handling wetted parts of the instrument.	<b>化学的危険性</b> : 毒性、発癌性、変異原性、腐食性、または刺激性のある 化学薬品を取り扱うときは、必要に応じて手袋などの保護具を着用し ます。廃油を処分したり、機器の接液部品を取り扱うときは、認可さ れた容器を使用し、適切な手順に従います。	<b>化学品危险:</b> 当处理毒性、致癌性、致突变性、腐蚀性或者刺激性化学品时,佩戴手套和其他保护性设备。当处理浸湿的仪器部件以及废油时,使用认可的容器和合适的步骤。
	<b>Hot surface:</b> Before touching, allow any heated components to cool.	<b>高温面</b> : 触れる前に、加熱した部品を冷ましてください。	<b>热表面:</b> 待高温部件冷却之后再进行维修。
	Flammable substances hazard: Use care when operating the system in the presence of flammable substances.	<b>可燃性物質の危険性</b> :可燃性物質があるところでシステムを作動させる 場合は十分注意してください。	<b>易燃物危险:</b> 在有易燃物质的场地操作该系统时,务必小心谨慎。
	<b>Risk of eye injury:</b> Eye injury could occur from splattered chemicals, airborne particles, or sharp objects. (Sharp objects that customers might install in the instrument include fused-silica tubing, the autosampler needle, and so on.) Wear safety glasses when handling chemicals or servicing the instrument.	<b>眼外傷の危険性</b> :飛散した化学薬品、浮遊粒子、または鋭利な物体に よって眼外傷を負うおそれがあります(機器に取り付けられる可能性が ある鋭利な物体は、ヒューズドシリカ、オートサンプラーニードルな どです)。化学薬品を取り扱ったり、機器を整備点検するときは、保護 メガネを着用します。	<b>眼睛伤害风险:</b> 眼睛受伤可能源自飞溅的化学品、空气中的颗粒, 或者锋利的物体。(安装在仪器内的锋利物体包括熔融石英管、 自动进样器的进样针等。)处理化学品或对仪器进行维修服务时, 务必戴上防护眼镜。
	<b>General hazard:</b> A hazard is present that is not included in the other categories. This symbol also appears on the instrument. For details about the hazard, refer to the instrument manual. When the safety of a procedure is questionable, contact Technical Support for Thermo Scientific San Jose products.	ー般的な危険性:それぞれのカテゴリーに当てはまらない危険がありま す。この標識記号は機器にも表示されています。この危険の詳細につい ては、機器のマニュアルを参照してください。 手順の安全性にご不明な点がある場合は、Thermo Scientific San Jose 製品の テクニカルサポートまでお問い合わせください。	<b>普通危险:</b> 未归入其他类别的危险。此符号也会在仪器上出现。有关此 危险的详细信息,参阅适当的仪器手册。若对任何步骤的安全事项有疑 问,联系 Thermo Scientific San Jose 产品的技术支持中心。

CAUTION Symbol	CAUTION	警告	危险警告
	<b>Laser hazard:</b> This instrument uses a laser that is capable of causing personal injury. This symbol also appears on the instrument. For details about the hazard, refer to the instrument manual.	<b>レーザー光線の危険性</b> : この機器では、身体傷害を引き起こすおそれ のあるレーザーを使用しています。この標識記号は機器にも表示され ています。この危険の詳細については、機器のマニュアルを参照して ください。	<b>激光危险:</b> 本仪器所用激光会导致人身伤害。此符号也会在仪器上出 现。有关此危险的详细信息,参阅适当的仪器手册。
	<b>Ultra violet light hazard:</b> Do not look directly at the ultra-violet (UV) light or into the UV source. Exposure can cause eye damage. Wear UV eye protection.	<b>紫外光の危険性</b> :紫外(UV)光またはUV光源を直接見ないでください。照 射によって眼損傷を引き起こすおそれがあります。UV保護メガネを着用 します。	<b>紫外光危险:</b> 不要直视紫外 (UV)光或者紫外光源。直视可能导致眼 睛伤害。佩戴紫外线防护眼镜。
	<b>Sharp object:</b> Avoid physical contact with the object.	<b>鋭利な物体</b> :物体との身体的接触を避けてください。	<b>锋利物体:</b> 避免直接接触锋利的物体。
	<b>Pinch point:</b> Keep hands away from this area.	<b>ピンチポイント</b> :この部分には手を挟まれないようにしてください。	<b>夹点:</b> 勿将手放在此部位。
	<b>Heavy objects:</b> Never lift or move the instrument by yourself; you can suffer personal injury or damage the equipment. For specific lifting instructions, refer to the instrument manual.	<b>重量物</b> :1 人で機器を持ち上げたり移動しないでください。身体傷害を 負ったり、機器を損傷するおそれがあります。具体的な持ち上げ方法 については、機器のマニュアルを参照してください。	<b>重物:</b> 切勿独自提起或移动本仪器;可能遭受人身伤害或损坏仪器。 有关具体的提起说明,参阅仪器手册。
A	<b>Trip obstacle:</b> Be aware of cords, hoses, or other objects located on the floor.	<b>作業の障害物</b> :床にあるコード、ホース、その他の物体に注意してく ださい。	<b>绊倒危险:</b> 注意地面上的线、管或其他物品。
	When the safety of a procedure is questionable, contact Technical Support for Thermo Scientific San Jose products.	手順の安全性にご不明な点がある場合は、Thermo Scientific San Jose 製品の テクニカルサポートまでお問い合わせください。	如对安全程序有疑问,联系 Thermo Scientific San Jose 产品的技术支持 中心。

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### **Preface**

This guide describes the maintenance and troubleshooting procedures required to maintain the EASY-nLC<sup>™</sup> 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
- Safety and Special Notices
- Contacting Us
- To suggest changes to the documentation or to the Help

Complete a brief survey about this document by clicking the button below. Thank you in advance for your help.



### **Related Documentation**

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

- EASY-nLC Series User Guide for the Xcalibur Data System
- EASY-nLC Preinstallation Requirements Guide
- EASY-nLC Series Getting Started Guide
- Safety and Regulatory Guide

The EASY-nLC instrument also ships with a printed copy of the *Safety and Regulatory Guide*. This guide contains important safety information about Thermo Scientific<sup>™</sup> liquid chromatography (LC) and mass spectrometry (MS) systems. Make sure that all lab personnel have read and have access to this document.

You can find the EASY-nLC Series manuals in the following locations:

- The USB flash drive provided in the EASY-nLC accessory kit
- The data system computer
- \* To view the product manuals on the data system computer

Choose Start > All Programs > Thermo Instruments > Manuals > LC Devices > Thermo.

#### \* To view the EASY-nLC Help

- From the EASY-nLC view of the Instrument Setup window, choose **Help** > **Thermo EASY-nLC Help** to open the Help to the Welcome topic.
- To view the Help topic for the current view, page, or dialog box, press the F1 key for information about setting parameters. Or, from the EASY-nLC view of the Instrument Setup window, choose **Help > Help On Current Item**.

### **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.



This guide includes the following cautions and special notices.



**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. You can use your smartphone to scan a QR code, which opens your email application or browser.

Contact us	<b>Customer Service and Sales</b>	Technical Support	
	(U.S.) 1 (800) 532-4752	(U.S.) 1 (800) 532-4752	
	(U.S.) 1 (561) 688-8731	(U.S.) 1 (561) 688-8736	
	us.customer-support.analyze @thermofisher.com	us.techsupport.analyze @thermofisher.com	
	<ul> <li>To find global contact information</li> <li>1. Go to www.thermoscientific.com.</li> </ul>	or customize your request	
<ol> <li>Click Contact Us, select the Using/Servicing a Product option, ar type the product name.</li> <li>Use the phone number, email address, or online form.</li> </ol>			
	<ul> <li>To find product support, knowledge bases, and resources</li> <li>Go to www.thermoscientific.com/support.</li> </ul>		
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Send a	n email message to Technical Publications	(techpubs-lcms@thermofisher.com).	
• Compl	ete a survey at www.surveymonkey.com/s	/PQM6P62.	
To order For the E www.pro	<b>consumable and spare parts for the EAS</b> CASY-nLC 1000 instrument, go to xeon.com/productrange/nano_lc_easy-nk	SY-nLC instrument c_1000/accessories_spares/index.html.	

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

### 1

### 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.

Figure 1. EASY-nLC II instrument on the left and EASY-nLC 1000 instrument on the right



To familiarize yourself with the EASY-nLC instrument and its touch-screen application, review these sections.

#### 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 sections 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—Installed in the EASY-nLC 1000 Instrument and Later Versions of the EASY-nLC II instrument" on page 3
- "ASA Model—Installed in Early Versions of the EASY-nLC II Instrument" 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 discontinued 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—Installed in the EASY-nLC 1000 Instrument and Later Versions of the EASY-nLC II instrument

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



Figure 2. ASC autosampler model installed in the current versions of the EASY-nLC 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—Installed in Early Versions of the EASY-nLC II Instrument

Figure 4 shows the ASA autosampler model that is installed in early 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 sections 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 Valve Pods
- 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  $\mu$ 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  $\mu$ 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<sup>™</sup> 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  $\mu$ L/min. The EASY-nLC II instrument uses two different flow sensors. The maximum measuring range is approximately 2.9  $\mu$ L/min for flow sensor A and 4.5  $\mu$ 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 266.

### **Six-Port Rotary Valve Pods**

The three-position, six-port rotary valve pods consist of a driver, a rotor seal, and a stator (see Figure 11). The rotary valve includes the valve pod and actuator.

Figure 11. Valve components



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.

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 backpressure 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 backpressure 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 94.

The autosampler needle for the ASC autosampler is a length of tubing with a nanoViper fitting at one end and a PEEK<sup>™</sup> nut and plastic stop at the other end (Figure 13). The needle retainer fits into a slot in the panel between the solvent system compartment and the tray compartment and properly positions the needle tubing. The nanoViper fitting connects to port 1 of valve S.



Figure 13. Autosampler needle (LC302 current version) for the ASC autosampler

The autosampler needle for the discontinued ASA autosampler is a length of fused-silica tubing (Figure 14). A stainless steel fitting secures one end of the tubing to port 1 of valve S, and a PEEK nut secures the other end of the tubing to the *z*-axis holder. The tubing routes through a cutout in the panel that separates the solvent delivery system components from the autosampler compartment.

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


## **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. Figure 16 shows the discontinued version of the instrument's computer box, and Figure 17 shows the version that is currently shipping (as of this release of the *EASY-nLC Troubleshooting and Maintenance Guide*).

Figure 16. Back panel of the EASY-nLC instrument (with the discontinued computer box)



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



Table 2 lists the function of each back panel element.

Table 2.	Back panel	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)
Ι O	Power switch
Fuse holder below I O switch	<ul> <li>Replaceable fuse ratings:</li> <li>For 120 V, T 5 AL, 250 V</li> <li>For 230 V, T 2.5 AL, 250 V</li> </ul>
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 a touchscreen instrument control application that you access from the instrument monitor.

These sections 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 when the operator is wearing gloves) and sends the corresponding commands to the computer (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** 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.

Figure 19. EASY-nLC touch screen version 3.2 with the menu layout on the Home > Overview page



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 :	
Accept	Cancel

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

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

4. Press Accept.

Figure 21. EASY-nLC login dialog box

Login as user	
User : admin	•
Password : *****	
Accept	Cancel

## **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** With the exception of an emergency shutdown, closing down the instrument in a controlled manner is important to allow all the components to shut down in an orderly sequence. If you turn off the power switch during normal operation, you risk damaging essential system components. Follow the procedure outlined here whenever possible.

### \* 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 sections 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)

Item	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 <sup>™</sup> 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 (installed in early versions of the discontinued 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	LC/MS-grade water with 0.1% formic acid	
Solvent B	LC/MS-grade acetonitrile with 0.1% formic acid	
<b>IMPORTANT</b> Use only UHPLC/MS-grade 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 <sup>™</sup> 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	<ul> <li>140 μL volume (enough for a &gt;10 h, 0–100% B, 300 nL/min gradient)</li> <li>1 nL/min to 300 μL/min flow range</li> <li>External pressure sensor</li> </ul>
Rotary valves	<ul> <li>VICI<sup>™</sup>/Valco<sup>™</sup> rotor/stator</li> <li>6 ports</li> <li>3 positions (1–6, 1–2, or CENTERED)</li> </ul>
Autosampler	<ul> <li>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.</li> <li>Plate holder ejects through spring-mounted autosampler door.</li> <li>4 glass bottles with plastic lids for waste or wash liquids</li> </ul>

# **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**

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



**CAUTION** Use only UHPLC/MS-grade solvents with the EASY-nLC instrument. Using HPLC-grade solvents and water from laboratory purification systems causes system blockages and poor spray stability.

### 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  $\mu$ L for the EASY-nLC II instrument or 12  $\mu$ 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 backpressure

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 backpressure 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 235).

**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 6, "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.

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# **Maintenance Scripts and Service Records**

For information about the maintenance scripts and record keeping features provided by the EASY-nLC touch-screen application, see these sections.

### Contents

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

## **Maintenance Scripts**

The EASY-nLC touch-screen application includes a variety of built-in maintenance scripts that help you prepare the instrument for use and troubleshoot instrument problems. These maintenance scripts are divided into three categories: Prepare, Test, and Calibrate. Each of the following sections 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 Torque
- Test Pump Torque

### 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.

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## 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
------------	---------------------------------

SCIENTIFIC	Home Batch Setup	Method Setup	aintenance Configuration
Scripts	Category:	Name:	Ne graph
Log Book	Prepare	Purge solvent	(Ind graph
Support	Description Parame	eters Output	
Devices	Parameter	- Value	
	Purge iterations	10	
	Purge pump A	×	
	Purge pump B	×	
	Purge pump S	×	
Job 00:00:00	Full purge		No graph
6 Mar 2016 18:46 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. Select the check boxes for the appropriate pump or pumps. If you select the Full Purge check box, the Purge Iterations parameter becomes unavailable.
  - c. 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.

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 script runs, it 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  $\mu$ L for the PLF pump model or 12  $\mu$ 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 Setup	Method Setup	Maintenance	Configuration	
Scripts	Category:	Name:		No graph	
Log Book	Prepare	Flush air 🗸			
Support	Description Parame	ters Output			
Devices	Description	Output	, ]		
	Parameter	▼ Value			
	Flush pump A	×			
	Flush pump B	×			
	Flush pump S	×			
	Flush threshold [µl]	12.00			
				No graph	-)
lob 00:00:00					
5 Mar 2015 19:24			-		
admin	Schedule	RT			
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  $[\mu L]$  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.

Thermo scientific	Home Batch Setur	Method Setup	laintenance Configuration
Scripts	Category:	Name:	No graph
Log Book	Prepare	Precolumn equili	
Support Devices	Description Param	eters Output	
	Parameter	▼Value	
	Volume [µl]	10.00	
	Flow [µl/min]		
	Max pressure [bar]	800.00	
			No graph
Job 00:00:00			
5 Mar 2015 19:31			
admin Exit	Schedule	ART STOP	

Figure 26. Precolumn Equilibration script parameters

### \* 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.

hermo	Home Batch	Setup Method Setur	Maintena	Configuration	
cripts	Category:	Name:			
og Book	Prepare	Analytical col eq	•	No graph	
upport	Description	aramotors Output			
evices	Description	arameters Output			
	Parameter	· · · · Value			
	Volume [µl]	4	.00		
	Flow [µl/min]				
	Max pressure [bar]	800	.00		
				No graph	
b 00:00:00					

Exit...

**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  $[\mu L/min],$  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 Setup	Method Setup	aintenance Configuration
Scripts Log Book Support	Category: Prepare	Vame: Isocratic flow	No graph
Devices	Parameter           Volume [µl]           Flow [µl/min]           0.0 Min [%D]	✓ Value 100.00 0.30	
Job 00:00:00	AB Mix [%B] Run indefinitely	30	No graph
5 Mar 2015 19:46 admin <b>Exit</b>	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 script ignores this volume setting.

• In the Flow box  $[\mu L/min]$ , 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.

SCIENTIFIC	Home Batch Setup Method Setup Maintenance Configuration
Scripts Log Book	Category: Name: Test MS connection
Devices	Description Parameters Output Tests the connection to the mass spectrometer using gradient-ready and gradient-done events
_Job00:00	No graph -
5 Mar 2015 19:57	
Exit	Schedule START STOP

Figure 29. MS Connection script

- \* 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 that connects to a Thermo Scientific mass spectrometer.



Figure 30. Thermo Fisher contact closure cable, part number LC160 (part name)

**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 12-pin connector to the back panel of the EASY-nLC instrument. For an LTQ<sup>™</sup> or TSQ<sup>™</sup> mass spectrometer, connect the two-pin connector to the mass spectrometer's Start In pins. For a TSQ Quantiva<sup>™</sup>, a TSQ Endura<sup>™</sup>, or an Orbitrap Fusion<sup>™</sup> mass spectrometer, modify the contact closure cable as described in the *EASY-nLC Series Getting Started Guide*.

- 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 either Thermo Scientific (LC160) or Thermo Scientific Orbitrap Fusion (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.





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			? <mark>- x -</mark>
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
<u>C</u> omment:		○ Minut <u>e</u> s	View
	Use instrument method	<u>G</u> o to Standby when Finished	
Instrument Method:		Acquisition Status	Ins <u>t</u> . Setup
	Start Mode	State: Idle Time (min): 0.000	
	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 281.

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.

SCIENTIFIC	Home Batch Setu	p Method Setup	laintenance	Configuration	
Scripts	Category:	Name:		o graph	
Log Book	Test	Sample pickup	<u> </u>	o graph	
Support	Description Paran	neters Output			
Devices			Position		
	Volume [µl]	▼ Value 10.00	Select positi	on	
	Flow [µl/min]	20.00	-	0:Plate/vials	
	Position	0-A1	-	A	
			-	1	
Job 00:00:00 6 Sep 2011 11:28 admin Exit	Schedule	ART STOP	Accept		Cancel

**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 281.

- 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 [ $\mu$ L/min], enter the flow rate that pump S uses to aspirate the sample. For aqueous samples enter 20  $\mu$ 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 235.

**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	Home Batch Setu	Method Setup	Maintenance	Configuration
Scripts	Category:	Name:		No graph
Log Book	Test 🔹	Leaks		ivo grapin
Support	Description Param	eters Output		
Devices				
	Parameter	▼Value		
	Test	System -		
		A		
		В		
		A+B		
		System		No graph
Job 00:00:00			·   `	
5 Mar 2015 20:12				
admin Exit	Schedule	ART 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 appropriate union (see Table 9 on page 46).

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 appropriate union for your instrument setup (Table 9).

Instrument	Part number	Description
EASY-nLC II	SC600	Stainless steel zero-dead-volume union for 1/32 in. OD tubing
EASY-nLC 1000	SC900	Viper union stainless steel zero-dead-volume union for 1/16 in. OD tubing
EASY-nLC 1000 with the modified Column Out and Waste In lines provided in the UHPLC Liquid Junction Kit	ES272	UHPLC fused-silica union

Table 9. Leak test unions

For information about troubleshooting system leaks, see "Troubleshooting the Results of the System Leak Test" on page 239 and "Troubleshooting a Pump That Fails the Leaks Script" on page 233.

## **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 below 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 Setu	Method Setup	Maintenance	Configuration	
Scripts	Category:	Name:			
Log Book	Test 🔹	Valve check		o graph	
Support	Description Para	meters Output			
Devices	Description	uncers Output	1		
	Parameter	✓ Value			
	Include valve A	×			
	Include valve B	×			
	Include valve S				
	Include valve W			o araph	
Job 00:15:48 11 May 2011 12:47 admin Exit	Schedule	ART STOP			

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 backpressure for solvent A and B. The script runs at a preset flow and measures the backpressure on the system.

**IMPORTANT** Before you perform the Back Pressure script, ensure that the solvent A bottle contains LC/MS-grade water and the solvent B bottle contains LC/MS-grade 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 Setup	Method Setup	Maintenance Configuration
Scripts	Category:	Name:	No graph
Log Book	Test	Back pressure	i vo graph
Support	Description Paramo	eters Output	
Devices			
	Parameter	▼Value	
	Test solvent A	×	
	Test solvent B	×	
			No graph
Job 00:00:00			
5 Mar 2015 20:23			
admin Exit	Schedule STA	STOP	

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.

Thermo SCIENTIFIC	Home Batch Setup Method Setup Maintenance Configuration
Scripts Log Book	Category: Name: Test Autosampler torque
Support Devices	Description Parameters Output Tests the individual torque required for the autosampler manipulator axes.
Job 00:01:02	No graph
24 aug 2010 17:60 admin <b>Exit</b>	Schedule START STOP

Figure 38. Autosampler Torque script description

## 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.

<b>Thermo</b>	Home Batch Setur	Method Setup	Maintenance	Configuration	
Scripts Log Book	Category: Test	Name: Pump torque		No graph	•
Support Devices	Description Param	eters Output			
	Parameter	▼ Value			
	Test pump A	×			
	Test pump B	×			
				No graph	•
Job 00:00:00					
<sup>5</sup> Mar 2015 20:32 admin <b>Exit</b>	Schedule	ART STOP			

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 30 minutes.

Figure 40 shows the parameters for the Flow Sensors script.

**Thermo** Maintenance Home Batch Setup Method Setup Configuration Name Category: Scripts No graph Calibrate Flow sensors Log Book Support Parameters Output Description Devices ▼ Value Parameter Include sensor A × Include sensor B × Inspection only Exact inspection ۲ No graph ast inspection Ο 00:00:00 Job 5 Mar 2015 20:36 admin START Schedule Exit...

Figure 40. Flow Sensors script parameters

#### ✤ 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  $\mu$ L/min point. Then inspects and evaluates this first calibration point.
- 2. Calibrates the 500  $\mu$ L/min point. Then 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.

SCIENTIFIC	Home Batch Setup	Method Setup	Maintenance	Configuration	
Scripts	Category: Na	ime:			
Log Book	Calibrate	Direct infusion		vo graph	<b>)</b>
Support	Description Param	atars Output			
Devices	Description	eters Output			
	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		Jo graph	-)
	Wash	×			
Job 00:04:20					
17 Feb 2012 12:43 admin Exit	Schedule	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  $\mu L$  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

Attentio was abor	n: Wash is enabled, but the script ted!
Make sur	e you wash loop & needle before
e.g. by infusion	specifying zero for the pickup and 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 Setu	p Method	Setup	Maintenance	Configur	ation	
Scripts Log Book Support Devices	Log record	d contains (one	term per line)		Log book Queue jot User Any		Time From: First Time To: Last	record
	Search	result	Source					
	05/08-201	0 17:52:49 a	admin	Batch: adı	5u min-20100805-1751	mmary		
	05/08-201	0 17:44:34 a	admin	Batch: adr	min-20100805-1742			
	19/07-201	0 10:41:58	admin	Batch: adr	min-20100719-1039			
Job 00:00:00	12/05-201	0 17:03:40	admin	Purge sol	vent (00:00:00)			
	29/04-201	0 10:29:34 a	admin	Purge sol	vent (00:00:00)			
23 aug 2010 12:37	29/04-201	0 10:28:42	Buper	Purge sol	vent (00:00:00)			
admin	28/04-201	0 13:43:47	admin	lsocratic f	INW (NN:NN:NN)			
Exit	Enter I	Log Entry						Details

#### 2. Press Enter Log Entry.

The Log Entry dialog box opens.

**Figure 45.** Log Entry dialog box

Log Entry		
ļ		
Template		Component
Comment	-)	None
Add & Close	Close	Add

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

The Template 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, Pump S, and other added devices such as the EASY-Spray<sup>™</sup> column heater.

- 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.

# **System Modifications**

The EASY-nLC 1000 instrument is designed to work with specific analytical columns. If you want to use analytical columns with bare fused-silica ends, you must modify the instrument as described in this chapter.

For information about installing columns with bare fused-silica ends, refer to the *EASY-nLC* Series Getting Started Guide.

#### Contents

- Ordering the UHPLC Liquid Junction Kit
- Installing the Modified Column Out and Waste In Lines

## **Ordering the UHPLC Liquid Junction Kit**

Use the following part number to order the UHPLC Liquid Junction Kit from Thermo Fisher Scientific: ES269. Table 10 lists the items in the UHPLC Liquid Junction Kit.

Table 10. UHPLC Liquid Junction Kit contents (Sheet 1 of 2)

Description	Part number
Column Out line with a nanoViper fitting at one end and bare fused-silica tubing at the other end	6041.5290
Waste In line with a nanoViper fitting on one end and bare fused-silica tubing at the other end	6041.5289
<ul> <li>UHPLC fused-silica union, which consists of the following:</li> <li>PEEK holder</li> <li>Two internally-threaded knurled nuts</li> <li>Two microferrules</li> <li>Stainless steel cartridge with coned ports and a</li> </ul>	ES272

280 µm thru-hole

**Note** The UHPLC fused-silica union has two functions:

- For a two-column setup, it connects the Column Out line to the precolumn inlet.
- For system leak testing and backpressure testing, it connects the Column Out line to the Waste In line. For information about leak testing, refer to the *EASY-nLC Series Troubleshooting and Maintenance Guide*.

#### Table 10. UHPLC Liquid Junction Kit contents (Sheet 2 of 2)



## Installing the Modified Column Out and Waste In Lines

This section describes how to install the modified Column Out and Waste In lines that are supplied in the UHPLC Liquid Junction Kit. For information about setting up the liquid junction, refer to the EASY-nLC Series Getting Started Guide.

Installing the modified Column Out and Waste In lines requires the following tools and materials.

Tools	Parts and materials
• #2 Phillips head screwdriver	• Liquid Junction Kit (P/N ES269)
• Flat-blade screwdriver	• Powder-free safety gloves

#### \* To install the modified Column Out and Waste In lines

- 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, make a quarter-turn to loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.

3. Using the black knurled tightening tool, remove the Column Out line from port 3 of valve S and the Waste In line from port 2 of valve W.

Figure 46 shows the Column Out line connection to valve S and the Waste In line connection to valve W.

Figure 46. Right panel removed from the EASY-nLC 1000 instrument



The Column Out and Waste In lines provided in the UHPLC Liquid Junction Kit have a nanoViper fitting at one end and bare fused-silica tubing at the other end.

- 4. Connect the Column Out line's nanoViper fitting to port 3 of valve S and the Waste In line's nanoViper fitting to port 2 of valve W. To avoid damage to the nanoViper fittings, take care to install these lines as described in "Using nanoViper Fittings" on page 93.
- 5. Leak test the system as described in "Running a System Leak Test" on page 235.

**Note** When running a system leak test, use the UHPLC fused-silica union to connect the Column Out line to the Waste In line.

6. Reconnect the right panel.

For information about installing analytical columns with bare fused-silica ends, refer to the *EASY-nLC Series Getting Started Guide*.

5

# **Routine Maintenance**

To maintain the EASY-nLC instrument, follow the 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. When loosened, the screws rotate freely without constraint, but remain connected to the side panel.

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

#### Contents

- Maintaining a Clean Working Environment
- Using Only LC/MS-Grade Solvents
- 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.

## **Using Only LC/MS-Grade Solvents**

Use only LC/MS-grade solvents with your EASY-nLC instrument. Using HPLC-grade solvents or water produced by a laboratory purification system can cause system blockages and poor spray stability.

To eliminate system blockages caused by contaminated solvents, Thermo Fisher Scientific recommends that you use only solvent blends ordered from Thermo Fisher Scientific in the solvent A and B bottles and the wash solvent bottle.

#### To order LC/MS grade solvents and solvent blends from Thermo Fisher Scientific

1. Click this icon.



Go to: www.FisherLCMS.com.

2. Click the **Solvents** tab.

## **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 48 shows the steps required to remove the fuse holder.

2. Turn off the instrument power and pull the power plug out of the power receptacle (Figure 47).

Figure 47. Power cable removed from the instrument's power receptacle



- 3. Remove the power fuse as follows (Figure 48):
  - a. 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.
  - b. Pull the fuse holder out of the power entry module.

**Figure 48.** Removing the fuse holder



- 4. 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.

5. 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.

6. 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 (Figure 49).





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.



**CAUTION** Overtightening the prebent stainless steel solvent lines damages their fittings. However, even without overtightening, these flow lines typically start to leak once they have been disconnected and reconnected more than three times.

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

- 1. "Retracting the Piston" on page 68
- 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 69
  - "Replacing the Piston Seal in a PLU Pump" on page 74
- 3. "Priming the Pump" on page 78
- 4. "Resetting the Pump Usage Counter" on page 79
- 5. "Removing Air After Replacing a Piston Seal or a Pump" on page 80
- 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 80.
  - 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 (Figure 50).

Figure 50. Purge Solvent script with 0 iterations

Thermo SCIENTIFIC	Home Batch Setup	Method Setup	laintenance	Configuration	
Scripts	Category: N	Name:	6	No graph	
Log Book	Prepare	Purge solvent		No graph	
Support	Description Parame				
Devices		Output			
	Parameter	▼ Value			
	Purge iterations	0			
	Purge pump A	×			
	Purge pump B				
	Purge pump S				
	Full purge			No graph	-
Job 00:00:00					
5 Mar 2015 20:52					
admin Exit	Schedule	RT STOP			

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

The piston moves backward until it reaches the 140 µL 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 69

-or-

• "Replacing the Piston Seal in a PLU Pump" on page 74

## **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 74.

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

Tools	Parts and materials
• #2 Phillips head screwdriver	Powder-free safety gloves
• 13 mm open-ended wrench	LC/MS-grade methanol
• 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 68.
- 2. Set the corresponding valve to the **Center** position as described in "Using the Valve Controls" on page 221.
- 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, make a quarter-turn to 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 (Figure 51).
    - Figure 51. 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 52 shows a stainless steel fitting connected to an internally threaded pump head.

Figure 52. 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 (Figure 53).

**Figure 53.** 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:  $300 \ \mu L/min.$
    - v. In the volume box, enter the dispense volume:  $140 \ \mu$ 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  $-300~\mu\text{L/min}$  and the dispense volume to  $140~\mu\text{L}.$
  - 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 54 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 55).



Figure 55. Using the piston seal tool to remove the worn piston seal (PLF model pump)

- 10. Install the new piston seal as follows:
  - a. Insert the guide tube into the pump head flange (Figure 56).

Figure 56. 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 (Figure 57).Figure 57. Piston seal mounted onto the piston seal tool (PLF model pump)



d. Insert the piston seal tool into the guide tool (Figure 58).

Figure 58. Inserting the piston seal tool into the guide (PLF model pump)



e. Push the piston seal tool into the guide until you feel resistance (Figure 59).Figure 59. 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 (Figure 60).

Figure 60. 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 78.

### **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
	• Pump-to-pressure sensor flow line, P/N LC512

#### 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 68.
- 2. Set the corresponding valve to the **Center** position as described in "Using the Valve Controls" on page 221.
- 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, make a quarter-turn to 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 (Figure 61).

Figure 61. 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 (Figure 62).

**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 62.** 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 (Figure 63).

**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 63. 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 (Figure 64).

Figure 64. 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 (Figure 65).

Figure 65. 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 (Figure 66).

Figure 66. Inserting the piston seal into the pump head (PLU model pump)



d. Gently pull the pump head off of the guide rods (Figure 67).

Figure 67. 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 (Figure 68).Figure 68. 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  $-300~\mu L/min$  and the volume to  $140~\mu L.$  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 51 on page 70), use a 13 mm open-ended wrench to tighten the fitting.
  - For a PLF pump with a stainless steel fitting (see Figure 52 on page 70) 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  $10~\mu\text{L}$  for the PLF pump or  $12~\mu\text{L}$  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 165. 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.

For an EASY-nLC 1000 instrument or the solvent B flow line of an EASY-nLC II instrument, you can easily damage the prebent stainless steel tubing that connects the pump to the pressure sensor by overtightening the fittings. If the script detects a leak, replace this flow line.

## **Maintaining the Rotary Valves**

The EASY-nLC instrument has four rotary valves labeled Valve A, Valve B, Valve S, and Valve W. Each valve contains two replaceable parts, a rotor seal and a stator.

Maintaining the valves requires these tools and materials. For information about the replacement parts, see the applicable procedure.

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 tissue and Q-tip swab
Magnifying glass	



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

To determine if a valve requires maintenance, do the following:

- For valves A and B, follow the instructions in "Troubleshooting a Pump That Fails the Leaks Script" on page 233.
- For valves S and W, follow the instructions in "Running a System Leak Test" on page 235.

**IMPORTANT** To determine whether the leak indicated by the Leaks script is caused by a leaking rotary valve, you must exclude these other possible causes of a Leaks script failure—excess air in the subsystem, a worn pump seal, a leak from the tubing that connects the pressure sensor to the pump, or a leak from the tubing that connects the pump to the valve.

With proper handling, you can typically disconnect and reconnect the prebent stainless steel tubing that connects the pressure sensor to the valve up to three times before this tubing begins to leak.

If you identify a valve as the source of a Leaks script failure, take the following action:

- 1. Clean the valve's rotor seal and stator, and check the rotor seal and stator for damage (see "Cleaning and Inspecting the Rotor Seal and Stator" on page 84).
- 2. Depending on whether these replaceable parts show visible damage, do the following:
  - If neither part shows visible damage, reassemble the valve and check for leaks (see "Returning the Instrument to Normal Operation after Disassembling a Rotary Valve" on page 88).
  - If only the rotor seal shows visible damage, replace the rotor seal (see "Replacing the Rotor Seal" on page 85). Reassemble the valve and check for leaks.
  - If the stator shows visible damage, replace the rotor seal and the stator ("Replacing the Rotor Seal and Stator" on page 86). Reassemble the valve and check for leaks.
- 3. If the leak persists for valve A or B of the EASY-nLC 1000 instrument or valve B of the EASY-nLC II instrument, replace the stainless steel tubing that connects the valve to the pressure sensor (LC513 for valve A or B or LC514 for valve S). Check for leaks.

## Disconnecting the Solvent Lines and Removing the Stator from the Valve

To perform maintenance on the valve, you must remove the stator from the valve (see Figure 11 on page 12). You do not need to remove all of the solvent lines from the valve to clean the valve components or replace the rotor seal.

#### \* To remove the stator from the valve for cleaning

- 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, make a quarter-turn to loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 3. To clean the valve components or replace a rotor seal, disconnect the following solvent lines:
  - For valve A or B, disconnect the low-pressure solvent line from port 2 and cap the end of the solvent line; for example, connect a union to the fitting and plug the union's free end with a blank fitting.



**CAUTION** If you leave the solvent line from the check valve connected to the stator, the stator continues to siphon solvent from the solvent bottle. If you leave the stator disconnected from the valve for a lengthy time period, the solvent stream builds up and spills onto other components in the instrument.

Figure 69 shows the solvent line connections for valve A or B of an EASY-nLC 1000 instrument.



Figure 69. Valve A or B solvent line connections (EASY-nLC 1000 instrument)

- For valve A, B, or S in an EASY-nLC 1000 instrument or valve B in an EASY-nLC II instrument, disconnect the stainless steel solvent line that connects the valve to the pressure sensor as follows:
  - i. Using a 1/4 in. open-ended wrench, completely loosen the fittings that secure the prebent stainless steel tubing to the valve and the pressure sensor.
  - ii. To remove the loosened fittings from their receiving ports, gently squeeze the bent portion of the prebent tubing (Figure 70). Gently squeezing the bent portion of the tubing automatically retracts the fittings from their receiving ports and prevents the damage that can occur by directly pulling the fittings out of the ports.



Figure 70. Gently squeezing the bent portion of the prebent stainless steel tubing



**CAUTION** To avoid damaging the stator or the rotor seal, follow these guidelines:

- 1. To prevent unbalanced screw tension, which can cause permanent distortion of the stator or rotor seal, loosen the two screws that secure the stator to the valve alternately in quarter-turn (90 degree) increments until all the load is removed.
- 2. Never rest the stator on its sealing surface.
- 3. To remove the stator from the valve, do the following:
  - a. Using a 9/64 in. L-hex wrench, loosen the two hex screws that secure the stator to the valve alternately in quarter-turn (90 degree) increments until all the load is removed.
  - b. Remove the loosened screws from the stator and set them aside.
  - c. Pull the stator away from the valve driver and do one of the following:
    - If you are not replacing the stator, carefully rest the stator with its outer surface facing down on top of the instrument or leave the stator suspended by the tubing.
    - If you are replacing the stator, set the damaged stator aside for disposal.

### **Cleaning and Inspecting the Rotor Seal and Stator**

To clean the sealing surfaces of the rotor seal and the stator, you do not need to remove all the tubing from the stator.

#### \* To clean and inspect the sealing surfaces of the rotor seal and stator

- 1. Disconnect the solvent lines and remove the stator as described in "Disconnecting the Solvent Lines and Removing the Stator from the Valve" on page 82.
- 2. Leave the rotor seal mounted to the valve driver.
- 3. Clean the mounted rotor seal with a lint-free tissue or Q-tip soaked in methanol.
- 4. With its sealing surface facing up, clean the stator's sealing surface with a lint-free tissue or Q-tip soaked in methanol (Figure 71).

Figure 71. Cleaning the stator's sealing surface



5. Using a magnifying glass, inspect the sealing surfaces of the rotor seal and the stator.

- 6. Do one of the following:
  - If only the rotor seal shows visible damage, go to the next procedure, "Replacing the Rotor Seal."
  - If the stator shows visible damage, go to "Replacing the Rotor Seal and Stator" on page 86.
  - If neither the stator nor the rotor seal shows visible damage, go to "Reconnecting the Stator and the Solvent Lines" on page 87.

### **Replacing the Rotor Seal**

The appropriate replacement rotor seal depends on the valve's serial number (Table 11).

**Table 11.** Replacement rotor seals

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

- 1. Remove the stator from the valve and clean its sealing surface as described in "Disconnecting the Solvent Lines and Removing the Stator from the Valve" on page 82 and "Cleaning and Inspecting the Rotor Seal and Stator" on page 84.
- 2. Gently pry the rotor seal away from the valve driver (Figure 72).

Figure 72. Rotor seal mounted on the valve driver



Rotor seal

3. With the rotor's sealing surface (engrave flow passages) facing away from the valve, carefully mount the new rotor seal onto the valve driver.

To reconnect the stator and the solvent lines, go to "Reconnecting the Stator and the Solvent Lines" on page 87.

## **Replacing the Rotor Seal and Stator**

The appropriate replacement stator depends on the valve's serial number.

Table 12. Replacement stator

Instrument	Replacement part
EASY-nLC II valve	
• V-000100 to V-000999	Uncoated stator, P/N LC225
• V-001000 to V-019999	Coated stator, P/N LC226
EASY-nLC 1000 valve	
V-020000 and higher	UHPLC-optimized coated stator, P/N LC526

**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 and stator

- 1. Disconnect the stator from the valve as described in "Disconnecting the Solvent Lines and Removing the Stator from the Valve" on page 82.
- 2. Disconnect the remaining solvent lines and blind nuts from the valve. If you are replacing the stator in valve S, remove the sample loop.
  - Use a 1/4 in. open-ended wrench to remove blind nuts and solvent lines that are connected to the valve with stainless steel fittings.
  - Use a black nanoViper knurled nut to loosen and remove nanoViper fittings.
- 3. To remove the stator from the valve, do the following:
  - a. Using a 9/64 in. L-hex wrench, loosen the two hex screws that secure the stator to the valve alternately in quarter-turn (90 degree) increments until all the load is removed.
  - b. Remove the loosened screws from the stator and set them aside.
  - c. Set the damaged stator aside for disposal.
- 4. Gently pry the damaged rotor seal away from the valve driver and set the rotor seal aside for disposal.
- 5. With the rotor seal's sealing surface (engraved flow passage) facing away from the valve driver, carefully mount the new rotor seal onto the valve driver. For information about ordering the appropriate rotor seal, see Table 11 on page 85.

To reconnect the stator and the solvent lines, go to the next procedure, "Reconnecting the Stator and the Solvent Lines."
## **Reconnecting the Stator and the Solvent Lines**

After you clean or replace the valve components, you must reconnect the stator and solvent lines.

## \* To reconnect the stator and the solvent lines

1. With its sealing surface facing the valve, mount the cleaned or replacement stator to the valve driver.

**CAUTION** To avoid damaging the stator or the rotor seal, follow these guidelines:



- 1. To prevent unbalanced screw tension, which can cause permanent distortion of the stator or rotor seal, loosen the two screws that secure the stator to the valve alternately in quarter-turn increments until all the load is removed.
- 2. Never rest the stator on its sealing surface.
- 3. Insert the two hex screws that secure the stator to the valve and hand-tighten them until you feel resistance. Using a 9/64 in. L-hex wrench, tighten the screws alternately in quarter-turn (90 degree) increments until they are evenly torqued and the stator is flush against the valve body.
- 4. Reconnect the solvent lines and blind nuts:
  - To tighten the stainless steel fittings and blind nuts, use a 1/4 in. open-ended wrench. Take care to avoid overtightening the fittings.
  - To reconnect a swaged stainless steel fitting to its receiving port, press the tubing against the bottom of the port and tighten the fitting hand-tight. Then use a 1/4 in. open-ended wrench to tighten the fitting by an additional 1/4 to 1/2 turn.
  - To reconnect a nanoViper fitting, use a black nanoViper knurled nut to tighten the fitting fingertight. Take care not to overtighten these fingertight fittings.

To return the instrument to normal operation, go to the next procedure, "Returning the Instrument to Normal Operation after Disassembling a Rotary Valve."

## **Returning the Instrument to Normal Operation after Disassembling a Rotary Valve**

After performing valve maintenance, you must remove air from the system and check for leaks.

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

- 1. Turn on the EASY-nLC instrument and log in as an administrator or a super user.
- 2. 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.
- 3. Check the system or subsystem for leaks as follows:
  - If you removed the stator from 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.
- 4. If the Leaks script fails, do one of the following:
  - If you only cleaned the affected valve, replace the rotor seal as described in "Replacing the Rotor Seal" on page 85.
  - If you only replaced the rotor seal, replace the rotor seal and stator as described in "Replacing the Rotor Seal and Stator" on page 86.

**IMPORTANT** A Leaks script failure indicates one or more of the following problems:

- Excess air in the subsystem
- A leaking pump seal
- A leak from the tubing that connects the pressure sensor to the pump
- A leak from the tubing that connects the pump to the valve
- A leaking valve

To determine whether the valve is causing the Leaks script to fail, follow the instructions in "Troubleshooting a Pump That Fails the Leaks Script" on page 233 or "Running a System Leak Test" on page 235 for valves S and W.

With proper handling, you can typically disconnect and reconnect the prebent stainless steel tubing between the pressure sensor and the valve up to three times before this tubing begins to leak.

- 5. When the system is leak tight, 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. Go to the next procedure to reset the appropriate usage counter.

## \* To reset the usage counter for the valve

- 1. On the touch screen, press Maintenance > Devices.
- 2. Select the valve from the list of devices.
- 3. Press the **Summary** tab.
- 4. 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.

## **Replacing the Check Valves**

If a check valve is not functioning, replace it.

Replacing a check valve requires the following tools, materials, and replacement parts.

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, make a quarter turn to 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 (Figure 73).

Figure 73. Inline filter components (nut with an integrated tube, filter, and ferrule)



Replacing an inline filter requires the following tools and materials.

## Tools

- 5/16 in. open-ended wrench
- Parts and materials
  - 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, make a quarter-turn to 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 (Figure 74).

Replacing an Inline Filter for the EASY-nLC 1000 Instrument



6. Run the following scripts:

- Back Pressure script (see "Test Back Pressure" on page 48)
- Leaks script (see "Test Leaks" on page 44)

# **Using nanoViper Fittings**

For the EASY-nLC instrument, most of the plumbing connections are made with nanoViper fittings (Figure 75).

**Note** For more information about working with nanoViper fittings, see "Using nanoViper Fittings Quick Reference Guide" on page 341.





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 235.

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 <sup>a</sup>

 $^{a}$  For the current revision of the autosampler needle (LC302 revision B), a nanoViper fitting replaces the stainless steel nut.

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

- 1. Using a #2 Phillips head screwdriver, make a quarter-turn to loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.
- 2. Disconnect the autosampler needle from port 1 of valve S as follows:
  - If the autosampler needle has a stainless steel fitting (obsolete autosampler needle, part number LC302 revision A), use a 1/4 in. open-ended wrench to unscrew the fitting.
  - If the autosampler needle has a nanoViper fitting (autosampler needle, part number LC302 revision B), use the black knurled nut to unscrew the fitting.

Figure 76 and Figure 77 show the solvent line connections to valve S for the EASY-nLC II solvent system and the EASY-nLC 1000 solvent system, respectively.

Figure 76. Valve S solvent line connections for the EASY-nLC II instrument





## Figure 77. Valve S solvent line connections for the EASY-nLC 1000 instrument

3. For the discontinued version of the autosampler needle (LC302 revision A), remove the nut, ferrule, and sleeve from the needle tubing (Figure 78).

Figure 78. ASA autosampler needle 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 (Figure 79).

XYZ Robot-	
XTZ RODOL	
	Eject Plate Insert Plate
	6x8/standard vials Goto Calibration
	Goto Well
	Needle Lin Needle Down
-Wash	
V	v1 W2 W3 W4
(M	v1 W2 W3 W4
Cooler	v1 W2 W3 W4
Cooler	v1 W2 W3 W4
Cooler	v1 W2 W3 W4 7.0 ℃ Set Disable
Cooler	v1 W2 W3 W4 <b>7.0</b> ℃ Set Disable Temperature: <b>7.2</b> ℃

Figure 79. Autosampler direct control dialog box

- d. Press Goto.
- e. Go to step 7 on page 97.
- 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 80).

<b>Thermo</b>	Home	Batch Setup	Method Setup	Maintenance	Configura	tion
Scripts	Devices	4				
Log Book		Na	me	Туре		Add Device
Support	. Co	oled Autosampler (AS	SC) [Autosampler]	Autosampler		
Devices	EA	EASY-Spray		Column Heater		Remove Device
	EASY-nLC <sup>™</sup> [HPLC]			HPLC		Assign Slave
Job 00:00:00 5 Mar 2015 20:58 admin Exit	About Coole Plate	Properties d Autosampler New C x12/Abgene (AB-08 Copy To	Tools (ASC) [Autosamp Calibrate Current 200) [Uncalibrated] Delete	l <b>er]</b> : 8x12/Abgene (AB-080	0) Ma	als / Wash Calibrate anipulator Reset

Figure 80. 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, make a quarter-turn to 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 (Figure 81).

Figure 81. Removing the black needle holder from the slot in the metal plate



- 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 (Figure 82). Then carefully pull the autosampler needle upward and away from the holder.

Figure 82. 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 (Figure 83).

Figure 83. Holder on the back of the touch-screen monitor



- 5. Connect the needle to port 1 of valve S as follows:
  - For the ASA autosampler needle, do the following:
    - 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.
  - For the ASC autosampler needle, do the following:
    - a. Insert the nanoViper fitting into the receiving port. Then turn the screw clockwise until you feel resistance.
    - b. Using the black knurled nut, turn the screw clockwise to an angle between 0 and 45 degrees (1/8-turn).
- 6. Recalibrate the needle position as described in Chapter 8, "Calibrating the Autosampler's XYZ Robot."

7. Run two 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  $\mu$ L for the EASY-nLC II instrument or 12  $\mu$ L for the EASY-nLC 1000 instrument.

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, make a quarter-turn to 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 84 shows the sample loop connections for the EASY-nLC II instrument.

Figure 84. 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 343).
  - b. Disconnect the nanoViper fittings connected to ports 2 and 5 of valve S.

Figure 85 shows the sample loop connections for the EASY-nLC 1000 instrument.



Figure 85. 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 93. 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 Leaks" on page 240.

# **Replacing a Pressure Sensor for the PLU Pump**

This section 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 129.

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

Tools	Parts and materials	
• #2 Phillips head screwdriver	<ul> <li>Powder-free safety gloves</li> </ul>	

- Powder-tree satety gloves
- 1/4 in. open-ended wrench

• 2.5 mm L-hex wrench or ball driver

Pressure sensor, P/N LC502



**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 procedures:

- 1. "To remove a pressure sensor from the instrument" on page 102
- 2. "To install the new pressure sensor" on page 103

### 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 221.
- 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, make a quarter-turn to 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 86. 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 102, 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 102, 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," next procedure
- "Replacing a Flow Sensor in the EASY-nLC II Instrument" on page 108

## **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

• 8 mm open-ended wrench

- Powder-free safety glovesFlow 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 procedures:

- 1. "To remove an EASY-nLC 1000 flow sensor," next procedure
- 2. "To install an EASY-nLC 1000 flow sensor" on page 107
- 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, make a quarter-turn to 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 87 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 (Figure 87).
- 5. Remove the black cable from the flow sensor (Figure 88).

Figure 88. 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 (Figure 89).

Figure 89. Removing the screws attaching the flow sensor to the bracket



Location of screws that secure flow sensor A

7. Remove the flow sensor (Figure 90).

Figure 90. 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 89 on page 106) 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 88 on page 105).
- 4. Reinstall the inline filters on both ends of the flow sensor (see Figure 87 on page 105). Tighten the fittings with an 8 mm open-ended wrench.
- 5. Reconnect the nanoViper tubing to both ends of the flow sensor (see Figure 87 on page 105).
- 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.
- 8. 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 procedures:

- 1. "To remove an EASY-nLC II flow sensor," next procedure
- 2. "To install an EASY-nLC II flow sensor" on page 111

## 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, make a quarter-turn to 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 (Figure 91) that secure the flow sensor bracket to the instrument panel.



Figure 91. Removing the two screws that secure the flow sensor bracket to the panel



Remove the solvent lines from both ends of the flow sensor (Figure 92).
 Figure 92. Removing the solvent lines



5. Loosen the metal ring and remove the cable connected to the flow sensor (Figure 93).



**Figure 93.** Removing the cable to the flow sensor

Cable 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 (Figure 94).

Figure 94. Removing the screws that secure the flow sensor to the bracket



## 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.

- b. 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 94 on page 110).
- 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 93 on page 110).
- 3. Reconnect the solvent lines to both ends of the flow sensor (see Figure 92 on page 109).
- 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 91 on page 109).
- 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 (Figure 95).



Figure 95. 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	EASY-nLC II hard drive:
Torx T-10 wrench	• Compatible with obsolete computer box: P/N LC281
	• Compatible with current computer box: P/N LC286
	EASY-nLC 1000 hard drive:
	Compatible with obsolete computer box: P/N LC581
	• Compatible with current computer box: P/N LC586

To replace the hard drive, follow these procedures:

- 1. "To remove the hard drive," next procedure
- 2. "To install the hard drive" on page 115

## ✤ 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 (Figure 96).

Figure 96. 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 97.

Figure 97. 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 (Figure 98).

Figure 98. Removing the screws underneath the hard-drive drawer



5. Unplug the hard drive from the IDE cable (Figure 99).

Figure 99. Hard drive unplugged from the IDE cable



The 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 cable connector.

**Tip** If the IDE cable plug is difficult to access, remove the brass rod that holds the cable down (Figure 100). Use a Torx T-8 wrench to remove the screws. Place the rod back in the drawer afterward.

Figure 100. 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 98 on page 114).
- 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 96 on page 113).
- 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 startup procedure. Because the autosampler is not detected by the startup 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 8, "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 startup 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 startup 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.		
		_
Name	Туре	
Advion RePlay	Fluid Storage	
Autosampler (ASA)	Autosampler	
Autosampler (ASC)	Autosampler	
EASY-Spray	Column Heater	
EASY-nLC	HPLC	
New Era NE-1000	Syringe Drive	▼
Accept	Cance	

- 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.

# 6

# **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 Rotary Valve Pod
- 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 procedures:

- 1. "To remove the computer box," next procedure
- 2. "To install the new computer box" on page 124

## 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, make a quarter-turn to 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's LVDS cable (low voltage differential signaling) because it is easily damaged.

Figure 101 shows these cables in the EASY-nLC 1000 instrument and Figure 102 shows these cables in the EASY-nLC II instrument.



**Figure 101.** Cables connected to the computer box in the EASY-nLC 1000 instrument

Figure 102. 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 103.

Figure 103. Pressing the tab on the top of the plug of the 12 V cables



5. Using a Torx T-10 wrench, remove the three screws that secure the back panel to the instrument (see Figure 104).

**Note** In earlier versions of the 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 104. Screws that secure the back panel to the instrument housing



- 6. Using a Torx T-10 wrench, remove the two screws that secure the top panel to the instrument (see Figure 104).
- 7. Remove the computer box from the instrument as follows:
  - a. Using a Torx T-10 wrench, remove the bottom two screws that secure the computer box to the instrument (see Figure 105).
  - b. Raise the top panel by at least one inch.
c. Using a Torx T-10 wrench, remove the top two screws that secure the computer box to the instrument (see Figure 105).

Figure 105. Screws to remove from the computer box



d. Pull the computer box out of the EASY-nLC housing. Figure 106 shows an earlier version of the housing. In the current version, the top panel overlaps the computer box flange, so you must lift the top panel by about one inch before you can pull the computer box out of the housing.

Figure 106. Pulling out the computer box



e. Lower the top panel.

### To install the new computer box

- 1. Lift the top panel by about one inch.
- 2. Insert the new computer box into the EASY-nLC housing.
- 3. Reconnect all of the internal cables to the computer box.
  - Figure 101 on page 121 shows the connections to the computer box inside the EASY-nLC 1000 instrument.
  - Figure 102 on page 121 shows the connections to the computer box inside the EASY-nLC II instrument.
- 4. Using a Torx T-10 wrench and the four screws that you removed in step 7 on page 122, secure the computer box to the instrument (see Figure 105 on page 123).
- 5. Using a Torx T-10 wrench and the screws that you removed in step 5 on page 122, 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.

- 6. Using a Torx T-10 wrench and the screws that you removed in step 6 on page 122, secure the top panel to the instrument.
- 7. 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.
- 8. Turn on the EASY-nLC instrument, and log in as an administrator.
- 9. Add the autosampler to the Devices list (see "Managing the Devices List" on page 117).
- 10. Recalibrate the autosampler (see Chapter 8).

### **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
<ul><li>Torx T-10 wrench</li><li>2 mm L-hex wrench or straight wrench</li></ul>	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 procedures:

- 1. "To remove the monitor," next procedure
- 2. "To install the new monitor" on page 128

### ✤ 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 appliqué.
  - 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 107).
  - c. Go to step 5.

Figure 107. 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 108).



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

Figure 108. Removing the screws from the front of the monitor



- 6. Using a #2 Phillips head screwdriver, make a quarter-turn to 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 109).

Figure 109. 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 110 shows the white plastic needle guide on the back of the monitor.

Figure 110. White plastic needle guide



Carefully remove the monitor by pulling it straight forward (see Figure 111).
Figure 111. 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 110 on page 127 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 109 on page 126).
- 5. Using a 2 mm ball driver and the screws that you removed in step 5 on page 126, 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 125 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 126 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 102.



**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 procedures:

- 1. "To remove a pressure sensor that is connected to a PLF pump," next procedure
- 2. "To install a pressure sensor for a PLF pump" on page 133
- 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 221.
- 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, make a quarter-turn to 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 112).Figure 112. P-Bus communication cables connected to the pump PCB of the PLF pump



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

Figure 113. 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 114).

Figure 114. 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 115).

Figure 115. 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 116).



Figure 116. 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 117).





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 117).

### ✤ 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 129.
- 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 117 on page 132).
- 4. Reconnect the solvent lines to the pressure sensor (see Figure 116 on page 132).
- 5. Reconnect the pressure sensor communication cable, being careful not to put any stress on the cables (see Figure 115 on page 131).
- 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 113 on page 130).

**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 112 on page 130).
- 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 140.



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

Deute and materials

Replacing the ASC autosampler requires these items.

#### Tools

10015	
• #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 procedures:

- 1. "To remove the ASC autosampler," next procedure
- "To install the ACS autosampler into an instrument that previously included this model" on page 139

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

- 4. Remove the computer box from the instrument as described in "To remove the computer box" on page 120.
- 5. Unscrew the autosampler needle fitting, and remove or pull up the autosampler needle from the needle holder.

Figure 118 shows the autosampler needle connected to the needle holder.

Figure 118. Needle connected to the needle holder



6. Unplug the bus cables on the left side of the instrument from the autosampler cooler and horizontal XYZ-axis PCB (see Figure 119).

Figure 119. Bus cables



7. Cut the tie wrap attaching the cables to the vertical part of the autosampler frame, as shown in Figure 120.

Figure 120. Cutting the strip attaching the cables to the autosampler frame



8. Using a Torx T-10 screwdriver, remove the screws that secure the back horizontal cable bar to the autosampler frame (see Figure 121).

Figure 121. Horizontal cable bar after removal of screws



9. Move the cable bar to the side (this is easier to do if you also remove the bus cable to valve B). See Figure 122.



Figure 122. Horizontal cable bar moved aside

- 10. Using a 3 mm hex wrench, remove the three screws that secure the autosampler to the bottom plate of the housing (see Figure 123, Figure 124, and Figure 125):
  - Remove the screw from the right side of the autosampler (see Figure 123).

Figure 123. Removing the screw from the right side of the autosampler



Remove the screw from the back left side of the autosampler (see Figure 124).
Figure 124. Removing the screw from the back left of the autosampler



Remove the screw the screw on the front of the autosampler (see Figure 125).
Figure 125. Removing the front screw from the autosampler



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

Figure 126 shows the housing with the autosampler partially removed.

Figure 126. 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 124 on page 138).
  - Insert and tighten the screw on the right side of the autosampler (see Figure 123 on page 137).
  - Insert and tighten the screw on the front of the autosampler (see Figure 125 on page 138).
- 3. Insert and tighten the screws that secure the back horizontal cable bar to the autosampler frame (see Figure 121 on page 136).
- 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 124.
- 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 117.

12. Calibrate the autosampler as described in Chapter 8.

# 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.

Tools	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 procedures:

- 1. "To remove the ASA autosampler from the EASY-nLC II instrument," next procedure
- "To install the ASC autosampler into an instrument that previously held the ASA model" on page 142

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

1. Upgrade the touch-screen application to the latest version as described in "Downloading the Latest Firmware File" on page 315.

**Note** To understand the effect of installing the latest touch-screen software, read the release notes provided in the EASY-nLC User Zone before you upgrade.

- 2. Press Maintenance > Devices and remove the old autosampler from the device list.
- 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 120.

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

- 5. Using a #2 Phillips head screwdriver, make a quarter-turn to 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 125.
- 7. Using a 2.5 L-hex wrench or ball driver, loosen valve W (see Figure 127).

Figure 127. 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 133 on page 144).
- 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 128).

Figure 128. 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 139.

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 129.

Figure 129. 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 130).

Figure 130. 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 102 on page 121 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 131. 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 132. This designation means that you do not have to install a separate terminator cable.

Figure 131. P-Bus connector inserted between the computer box and the P-Bus communication cables



P-Bus connector

Figure 132. Computer box with "Terminated" label



- 6. Using a Torx T-10 wrench and the four screws that you removed in step 7 on page 122, secure the computer box to the instrument (see Figure 105 on page 123).
- 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 133 shows these two cables.

Figure 133. P-Bus communication cables



Figure 134 shows the boards that the P-Bus communication cables connect to. Figure 134. 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 135, Figure 136, and Figure 137 demonstrate this procedure.

Figure 135. Lifting up the autosampler to position the screw



Figure 136. Placing the  $M5 \times 16$  hex screw into the back right side of the autosampler frame



Figure 137. 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 138).

Figure 138. 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 139).

Figure 139. Securing the front screw



10. Place the air filter from the ASA autosampler onto the ASC cooler (see Figure 140).

Figure 140. 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 141).

Figure 141. Aluminum needle arm



12. Place the mixing Tee in the holder. Use the aluminum needle arm when you attach the needle (see Figure 142).



13. Reinstall the monitor as described in "To install the new monitor" on page 128.

**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 117.

16. Calibrate the autosampler as described in Chapter 8, "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," next procedure
- "Replacing the ASA Cooler" on page 152

### **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 procedures:

- 1. "To remove the ASC cooler," next procedure
- 2. "To install the new ASC cooler" on page 151

### 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 143.

Figure 143. 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 144.

Figure 144. 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 145.

The screws do not fall out when loosened, because they are held in place by O-rings. **Figure 145.** Removing the top center screw



6. Pull out the cooler, as shown in Figure 146.

Figure 146. 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 147 shows a damaged cable.

Figure 147. 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 155 on page 155).
- 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 154 on page 155).
- 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, P/N EXLC252
• 2.5 mm L-hex wrench or ball driver	
• 3 mm L-hex wrench	
• 5.5 mm open-ended wrench	
• #2 Phillips screwdriver	

To replace the ASA cooler, follow these procedures:

- 1. "To remove the ASA cooler," next procedure
- 2. "To install the ASA cooler" on page 157

### \* 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 148, to avoid damaging it when you remove the hex screws underneath the adapter plate holder.

Figure 148. 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 149).



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

Figure 149. 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 150).

Figure 150. Screws on the front of the EASY-nLC II instrument (model LC100-2)



4. Using a #2 Phillips head screwdriver, make a quarter-turn to 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 151.

Figure 151. Loosening the ribbon cap on the controller PCB



Figure 152 shows a close-up view of the cap that secures the cable to the PCB. **Figure 152.** Cap around the ribbon cable (close-up view)



6. Pull the ribbon cable out of the cap, as shown in Figure 153.

Figure 153. 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 154).

Figure 154. Removing the lid



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

Figure 155 shows the screw in the lower right corner, and Figure 156 shows the screw in the middle of the left side.

Figure 155. Loosening the screw in the lower right corner



Figure 156. 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 157 shows the location of these screws.

Figure 157. Screws beneath the adapter plate holder



Lift and pull the adapter plate holder out of the autosampler, as shown in Figure 158.
Figure 158. 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 159 shows these sleeves.

Figure 159. 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 160.

Figure 160. Aligning the arms of the autosampler bed



Carefully place the new cooler in the autosampler bed, as shown in Figure 161.
Figure 161. Placing the new cooler in the autosampler bed



3. Verify that the cables from the fan are routed in the groove (see Figure 162).

Figure 162. 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 157 on page 156 shows the location of these screws.
- 5. Using a 2 mm L-hex wrench, tighten the two screws in the small PCB. Figure 155 on page 155 and Figure 156 on page 155 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 154 on page 155).
- 7. Insert the ribbon cable into the controller board (see Figure 151 on page 154 and Figure 153 on page 154).
- 8. Using a flat screwdriver or your fingernail, push the cap around the ribbon cable in place (see Figure 152 on page 154).
- 9. Reattach the autosampler door (see Figure 148 on page 152).
- 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 8, "Calibrating the Autosampler's XYZ Robot."
## **Replacing a Rotary Valve**

A rotary valve consists of the valve pod and actuator. The procedure for replacing a rotary valve is the same for the EASY-nLC 1000 instrument and the EASY-nLC II instrument. Replace the rotary valve if its motor fails.



**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	• Valve (valve pod and actuator):
• Torx T-10 wrench	EASY-nLC 1000 instrument: P/N LC519
• 2.5 mm L-hex wrench	EASY-nLC II instrument: P/N LC218
• Flathead screwdriver	

To replace a rotary valve, follow these procedures:

- 1. "To remove a rotary valve," next procedure
- 2. "To install a new rotary valve" on page 161

#### 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 120.
- 3. Using a #2 Phillips head screwdriver, make a quarter-turn to 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. From the back of the instrument, disconnect the cables shown in Figure 163 from the valve to be exchanged.

Figure 163. Cables connected to valves



- 6. 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, hold the valve to prevent it from slipping down into the autosampler compartment.
- 7. Pull out the valve, as shown in Figure 164.

Figure 164. 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 PCB of the new valve, shown in Figure 165, to the same address as the valve being replaced:
  - Valve A=1
  - Valve B=2
  - Valve S=3
  - Valve W=4

Figure 165. 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 163 on page 160 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 replacement valve.
- 6. Reinstall the computer box as described in step 2 through step 7 of "To install the new computer box" on page 124.
- 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  $12 \mu$ L for a PLU pump (standard in the EASY-nLC 1000 instrument).
  - Enter  $10 \ \mu$ L 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 Rotary Valve Pod**

The valve pod includes the stator and rotor. It does not include the actuator.

To replace a rotary valve pod, you must remove the computer box from the instrument to access the rotary valve, and you must remove the valve from the instrument to remove the valve pod from the actuator.

Replacing a rotary valve requires these items.

Tools	Parts and materials
• #2 Phillips screwdriver	Powder-free gloves
• 1/4 in. open-ended wrench	Rotary valve pod:
• Torx T-10 wrench	Valve serial number V-020000 and higher:
• 2.5 mm L-hex wrench	P/N LC353
• Flathead screwdriver	Valve serial number V-010000 to V-019999: P/N LC352
• 9/64 in. hex driver	

- To remove the rotary value and disconnect the value pod from the actuator
- 1. Remove the rotary valve from the instrument as described in "To remove a rotary valve" on page 159.
- 2. Using a 9/64 in. hex driver, loosen the screw that secures the valve pod to the actuator (Figure 166). Then pull the valve away from the actuator.

Figure 166. Screw that secures the valve pod to the actuator



Screw that secures the valve pod to the actuator

#### \* To install a new valve pod and reinstall the valve

1. Insert a new valve pod into the actuator and rotate it until port 4 of the valve stator faces the PCB side of the pod (Figure 167).



#### Figure 167. Correct position of the valve stator

- 2. Using a a 9/64 in. hex driver, secure the valve pod to the motor by tightening the side screw.
- 3. Reinstall the valve as described in "To install a new rotary valve" on page 161.
- 4. Record the current number of rotor shifts for the valve that you just removed.
- 5. Reset the usage counters for the valve (rotor shifts) as described in "Checking and Resetting the Device Usage Counters" on page 56.
- 6. Record the valve replacement in the Log Book as follows:
  - a. From the touch-screen application, press Maintenance > Log Book.

The Maintenance > Log Book page opens.

b. Press Enter Log Entry.

The Log Entry dialog box opens.

- c. Make the following selections and entries:
  - In the Template list, select New Part.

The text "New Part" appears in the Log Entry description box (Figure 168).

- In the Component list, select the respective valve.
- In the description box, enter the number of valve shifts for the valve when you replaced the valve pod (see step 4 of this procedure).

Log Entry		
New Part New valve pod ir shifts reached 10	nstalled when the n ) 000.	umber of valves
Template New Part	•	Component None
Add & Close	Close	Add

Figure 168. Log Entry dialog box with an example entry

- 7. Run the Flush Air script as described in "To run the Flush Air script" on page 34:
  - Enter  $12 \ \mu$ L for a PLU pump (standard in the EASY-nLC 1000 instrument).
  - Enter  $10 \ \mu L$  for a PLF pump (standard in most EASY-nLC II instruments).
- 8. 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," next procedure
- "Replacing a PLF Pump with a PLU Pump" on page 169



**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 procedures:

- 1. "To remove a PLU model pump from an EASY-nLC instrument," next procedure
- 2. "To install a PLU pump in an EASY-nLC instrument" on page 168

## **\*** 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, make a quarter-turn to 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 169.

Figure 169. 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 170.

Figure 170. Flow sensor cable, pressure sensor cable, and P-Bus communication cables for an EASY-nLC 1000 instrument



• 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 167, 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.
  - Pump A =1
    Pump B = 2
    Pump S = 3
- 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 78.
- 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 80.
- 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 application to version 3.1.4 or later, as the EASY-nLC II instrument only supports the PLU pump when running 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	PLU pump, P/N LC501	
ball driver	Pressure sensor, P/N LC502	
• 2.5 mm hex wrench or	• Tubing between the pump and the pressure sensor:	
ball driver	<ul> <li>Pumps A and S: PEEK tubing, P/N LC212</li> </ul>	
• 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:	
	<ul> <li>Pumps A and S: PEEK tubing, P/N LC213</li> </ul>	
	– 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 application to version 3.1 or later.

If the touch-screen application is earlier 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 firmware, refer to Appendix B of the *EASY-nLC Series Getting Started Guide* (for the touch-screen application). You can find the touch-screen firmware files in the user zone on www.proxeon.com.

- 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 221.
  - 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, make a quarter-turn to loosen the three captive screws that secure the right side panel to the instrument housing. Then remove the panel.

Figure 171 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 171. 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 172).

Figure 172. Externally threaded pump head for a PLF model pump



• If a stainless steel fitting is connected to the pump head (see Figure 173), use a 1/4 in. open-ended wrench to remove it.

Figure 173. 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 174).

Figure 174. 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 175). 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 175). Then set the L-bracket and the two 2 mm hex screws aside for reuse when you mount the new pressure sensor.

#### Figure 175. Pressure sensor L-bracket screws



c. Disconnect the P-Bus communication cables from the pump actuator (see Figure 176).

Figure 176. 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 177).
- e. Set the four 3 mm screws aside for reuse in step 8a on page 174.

Figure 177. 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 EASY-nLC II 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 175. 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.

ID

• Pump A =1



- 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 78.
- 13. Remove air from the system as described in "Removing Air After Replacing a Piston Seal or a Pump" on page 80.
- 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 169 through step 6 on page 173 of the previous procedure, "To replace a PLF pump with a PLU pump in the EASY-nLC II instrument," to upgrade the touch-screen application, 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 178).
  - b. Set the two 2.5 mm screws aside for reuse when you secure the new flow sensor bracket.

Figure 178. 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 179).
- d. Set the 2.0 mm hex screws aside for reuse when you secure the flow sensors to the new flow sensor bracket.

Figure 179 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 180 shows the flow sensors removed from the flow sensor bracket. The flow sensor cables are still connected to the instrument.

Figure 179. Screws that secure the flow sensors to the bracket



Figure 180. 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 181. 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 182. 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 174 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," next procedure
- "Replacing a Pump PCB in the PLF Pump" on page 182

## **Replacing a Pump PCB in the PLU Pump**

Replacing the pump PCB on the PLU model pump requires these items.

Fools	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 procedures:

- 1. "To remove a pump PCB from the PLU model pump," next procedure
- 2. "To replace a pump PCB on the PLU model pump" on page 181

#### \* 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, make a quarter-turn to 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 183 on page 179 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 183). Then remove the PCB shield (see Figure 184).

**Figure 183.** Four PCB shield screws (PLU model pump)



The four captive screws remain in place (see Figure 184). **Figure 184.** 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 185).

Figure 185. 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 186) as follows:
  - Pump A =1
  - Pump B = 2
  - Pump S = 3

Figure 186. PCB rotary switch



- 2. Plug the motor cable into the back of the PCB (see Figure 185 on page 180).
- 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 183 on page 179) 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 procedures:

- 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, make a quarter-turn to 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 176 on page 173) 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 182.
- 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 185 on page 180).
- 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 187. 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.

# 7

# **Troubleshooting**

To troubleshoot problems with the EASY-nLC instrument, follow the troubleshooting tips and procedures in this chapter.

#### 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
- 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 pages:

- "Autosampler Problems" on page 187
- "Contact Closure Problems" on page 188
- "Instrument Startup Problems" on page 188
- "Delayed Elution" on page 189
- "Excessive Duration or Higher Pressure for the Column Equilibration and Sample Loading Steps" on page 194
- "System Reaches Its Maximum Pressure During the Gradient" on page 196
- "Sample Signal Weak or Absent" on page 199
- "Slow or No Pressure Increase in Subsystem A or B" on page 203
- "Errors Reported by the Xcalibur Data System" on page 206
- "Carryover" on page 211
- "Spray Issues" on page 213
- "Chromatographic Performance" on page 214
- "Device Failures" on page 215
- "Miscellaneous" on page 219

## **Autosampler Problems**

Table 13 lists troubleshooting tips for the autosampler.

**Note** For information about troubleshooting sample pickup, see "Troubleshooting the Autosampler Aspiration" on page 279.

Table 13. Autosampler troubleshooting tips

Symptom	Possible causes	Action
The needle cannot penetrate the plastic film on the microtiter plate.	The needle tip is damaged or bent.	Do the following:
		<ol> <li>Check the configuration settings for the adapter tray on the Tools page for the autosampler device, which you can access from the Maintenance &gt; Devices page.</li> </ol>
		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 94).
		• 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	Something is blocking the movement of the autosampler's XYZ arm and causing a step loss on one of the motors.	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 8, "Calibrating the Autosampler's XYZ Robot."
	The error repeats without any visible reason.	Contact Thermo Fisher Scientific:
		<ul> <li>us.customer-support.analyze@thermofisher.com</li> </ul>
		<ul> <li>eu.techsupport.cms@thermofisher.com</li> </ul>
A server warning message about the plate	The plate format has been deleted from the system	If you want to run the batch, you must create the plate format again.
tormat for the autosamplerconfiguration. For information about deleting a plate format, refer to the EASY-nLC Serieswhen you submit a batch.Getting Started Guide.		For information about selecting, creating, and deleting plate formats, see "Managing Plate Formats" on page 292.

## **Contact Closure Problems**

Table 14 contains troubleshooting tips for contact closure problems.

Table 14.         Contact closure troubleshootir	ng 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 <b>Two-way</b> on the Configuration > Connections page.
		If the mass spectrometer is waiting for a signal, set the contact closure State setting to <b>Open</b> .
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 <b>One-way</b> on the Configuration > Connections page.

## **Instrument Startup Problems**

Table 15 contains troubleshooting tips for instrument startup problems.

## Table 15. Startup troubleshooting tips

Symptom	Possible causes	Action
Dark monitor	Broken or open main	1. Turn on the power to the instrument.
	power fuse	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**

Table 16 lists the common causes of delayed elution. The most common cause of delayed elution is a worn rotor seal in valve B, causing valve B to leak.

These sections show the effect of leaks and swept volume on the pressure and flow traces:

- "Leaks that Cause Delayed Elution," next section
- "Introduced Swept Volume" on page 192

## Table 16. 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 239.
	Incorrect tubing installed	1. Remove the right side panel of the instrument.
Swept volume introduced by incorrect tubing connections		2. Verify that the tubing matches the solvent system schematic.
		• For the EASY-nLC II instrument, see page 332.
		• For the EASY-nLC 1000 instrument, see page 334.
		3. Replace the incorrect tubing with the specified tubing.
	Swept volume introduced	Check the tubing 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 188).

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 188. Negative flow of solvent B toward valve B during the column equilibration and sample loading steps



Figure 189 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 189. 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 backpressure 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 backpressure 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 190 shows the pressure dip that occurs when the gradient starts to reach the column. Figure 191 shows the pressure dip for the sample chromatographic method, but occurring two minutes later due to the added gradient delay volume.



Figure 190. 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 191. Pressure dip begins later when the system has introduced swept volume

100% solvent B reaches the column.

Backpressure 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 35 on page 329 lists the viscosities for two-solvent mobile phases consisting of a water/methanol mixture or a water/acetonitrile mixture.

# Excessive Duration or Higher Pressure for the Column Equilibration and Sample Loading Steps

Table 17 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 17. Common causes of a longer than expected column equilibration time or sample loading time

Possible cause	Action
Clogged column	Run the Back Pressure script for pump A (see "Test – Back Pressure" on page 48).
	If the test passes, one of the columns is probably clogged and must be replaced.
Clogged system	Run the Back Pressure script for pump A.
	If the test fails, go to "Troubleshooting a System Blockage" on page 267.
	Possible cause Clogged column Clogged system

Figure 192 and Figure 193 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 192 and Figure 193 are as follows:

- 2 cm length
- 100 µm ID
- 5 µm particle size
- C18


Figure 192. 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 18.

Table 18. Possible causes of maximum pressure spikes during a run

Symptom	Possible cause	Action
The system reaches its maximum pressure during a run.	Clogged column	Run the Back Pressure script for pumps A and B.
		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 267.
	Excessively high flow rate	Reduce the flow rate in the method.

When a column or the Column Out line is clogged, the backpressure can increase to the maximum system pressure for pump A, pump B, or pumps A and B during the gradient (see Figure 194).

Figure 194. Backpressure 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 backpressure on pump B increases and differs from the backpressure on pump A.

Figure 195 shows the typical profile for a blockage in the solvent system between pump B and the mixing Tee.

Figure 195. Backpressure shown reaching the maximum pressure for pump B only



Figure 196 shows the flow path between pump B and the mixing Tee.

Figure 196. 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 19.

Table	19.	Possible	causes	of a	weak	or	absent	signal
			00.0000			۰.		0.90.

Symptom	Possible cause	Action
The sample signal is missing or weak.	Sample not aspirated (See Figure 197 on page 200.)	Run the Sample Pickup script. If the Sample Pickup script fails, see "Troubleshooting the Autosampler Aspiration" on page 279.
	Damaged or incorrect column (See Figure 198 on page 201.)	Make sure that you have installed the correct column or columns.
		Check the backpressure on the columns by running the column equilibration scripts.
	Leak in the system	Run the Leaks test on the system.
	(See Figure 199 on page 202.)	If the test fails, see "Troubleshooting the Results of the System Leak Test" on page 239.
	Incorrect column configuration	Check the column configuration.
		1. Press Maintenance > Devices.
		<ol> <li>In the Devices list, select EASY-nLC (HPLC).</li> </ol>
		3. Press the <b>Properties</b> tab.
		4. Clear the <b>One Column Setup</b> 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 197).

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 197. Typical pump A profile for a system with air in the sample loop

#### Weak or Absent Signal—Defective Precolumn

When compared to the Pump A graph for a normal run shown in Figure 192 on page 195, Figure 198 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  $\mu$ L/min) and the duration is shorter because the precolumn is exerting less backpressure.

**Note** The specifications for the precolumns used to acquire the data in Figure 192 and Figure 193 are as follows:

- 2 cm length
- 100 µm ID
- 5 µm particle size
- C18



The measured flow rate is > 15  $\mu$ L/min.



### Weak or Absent Signal—Leak Upstream of Flow Sensor B

Figure 199 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 199. Typical pump B profile for a leak upstream of flow sensor B

sample loading steps

## 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 20. Troubleshooting low backpressure in subsystems A or B

Symptom	Possible cause	Action
Backpressure 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 229.
	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 233.

Figure 200 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 200. 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 201.

Figure 201. 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).
$\mathbf{\overline{O}}$	Reported by: Pump A
	Close

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 202).

Figure 202. Subsystem B leak



When pump B cannot generate sufficient pressure to start the gradient, the server error message shown in Figure 203 appears.

Figure 203. Server error message displayed when pump B cannot generate sufficient pressure to start the gradient

Ser	ver error message(s)
8	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 21 lists the possible causes of the common errors reported by the Xcalibur data system.



Symptom	Possible cause	Action
An Acquisition Server failure prevents the sequence from running (see Figure 204 on page 207).	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 <b>Test Connection</b> .
		If necessary, restart the EASY-nLC instrument.
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 205 on page 208.)	Check the loop configuration on the Properties page for the EASY-nLC HPLC device on the Maintenance > Devices page.
riepare for Run step.		Make sure that the configured loop volume matches the installed sample loop.
	Lowercase letter used in the sequence table for vial position	Change the letters to capitals.
	(See Figure 206 on page 208.)	
Unable to submit samples. The following error message appears:	Incorrect plate configuration (See Figure 207 on page 209.)	Select the correct autosampler plate in the Thermo EASY-nLC Configuration
The mass spectrometer remains in	Contact closure configured	Brees Configuration > Connections
the Waiting for Contact Closure	incorrectly	riess Configuration > Confiections.
state and data acquisition does not start (see Figure 208 on page 210).		Select the correct MS configuration and settings.
	Failing contact closure relay due to incorrect grounding	Check whether the LC/MS system is properly grounded as described in "Verifying that the LC/MS System Is Properly Grounded" on page 285.

Figure 204 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 204. Lost connection between the EASY-nLC instrument and the Acquisition Server



Figure 205 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  $\mu$ L.

Figure 205. Incorrect injection volume



Figure 206 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 206. Incorrect vial position nomenclature

Incorrect use of a lowercase letter



Figure 207 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 207. Incorrect vial position for the vial adapter plate format



Figure 208 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 208. Waiting for Contact Closure error

## Carryover

Table 22 lists the possible causes of carryover.

Table 22. 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 209 and Figure 210).
	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 209 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 209. Gradient run without an adequate high-organic hold at the end of the run

Figure 210 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 210. 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 23 lists the possible causes of an unstable spray.

**Table 23.** 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 24 lists troubleshooting tips for common chromatography problems.

Table 24	Troubleshooting	tins for	chromatographic	nerformance
10010 24.	noubleanooting	101	omornatographic	periornance

Symptom	Possible cause	Action
Broad peaks	Dead volume between the	Check the connections.
	continuit and childer	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 during valve shifts	Use the scripts to check for a leak in the B subsystem (Leak test Pumps A + B, and System).
		Replace defective seals, rotors, and solvent lines as needed.

## **Device Failures**

Table 25 lists troubleshooting tips for devices failures.

Table 25.         Troubleshooting tips for device failu	res
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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 211 on page 216.	? 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 212 on page 217.	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 213 on page 218.		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 211 shows the pink pump icons on the Home > Overview page.

**Figure 211.** 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 212 shows the pink valve icons on the Home > Overview page.

Thermo Home Batch Setup Method Setup Maintenance Configuration 0.2 bar 0.0 nl/min 140.0/140.0 µl LSP disabled Overview Active 
 Pending
 Finished 
 Cancelled

 Idle flow disabled Graphs 000 -0.7 nl/min 000 -0.7 bar 0.0 nl/min 140.0/140.0 µl Queue 7.0 °C Solve S -0.6 bar 0.0 nl/min 140.0/140.0 µl %B C:4117,700 i 00000 20.0 µl В Refill A B Initialize system Pickup sample Load sample Equilibrate precolumn Prepare gradient Equilibrate analytical column Run gradient Autosampler wash + refill S Job 00:00:00 Eject / Insert tray Sample Job 31 Oct 2011 08:49 admin START Next Exit...

Figure 212. Pink valve icons on the Home > Overview page

An autosampler icon turning pink means that the communication to the unit has been lost.

Figure 213 shows the pink autosampler icon on the Home > Overview page.

Figure 213. 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 26).

Table 26. 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
		<ul> <li>eu.techsupport.cms@thermofisher.com</li> </ul>
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 <b>Ignore</b> 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:
		<ul> <li>If the network is set up for remote access, press Maintenance &gt; Support and press Connect.</li> </ul>
		0r
		• Run a factory reset.
System is running slower and slower.	Memory filled with graph data from long gradient run	Press <b>Home &gt; Graph</b> and disable the graphs by pressing <b>No Graph</b> in each graph window.
	System overload caused by other reasons	Restart the application or power down/power up from <b>Maintenance &gt; Scheduling</b> .

Symptom	Possible causes	Action
Valve – unknown position	The valve's duty cycle requires calibration.	Run the Valve Check script (see "Test – Valve Check" on page 47).
		This script is only valid for EASY-nLC II instrument valves with a serial number below V-009999.
		Contact your local Thermo Fisher Scientific field service engineer if the reported overshoot is greater than 6.
Unhandled error or SVG (scalable vector graphics) error	Program error in the software release	In most situations you can continue your work by pressing Ignore or OK. More serious error situations might require restarting the HPLC.
		To improve on the software quality, email details to the following:
		• us.customer-support.analyze@thermofisher.com
		• eu.techsupport.cms@thermofisher.com
		Or, connect to the remote support server. See "Connecting the EASY-nLC Instrument to the Support Server" on page 306.

 Table 26.
 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.

To use the direct controls, follow these procedures:

- 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 Valve Pods" 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 214). The readback box lists the current valve position.

#### Figure 214. 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 215). The Position readback box displays the current pump position from 0  $\mu$ L (empty) to 140  $\mu$ 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 221.

**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 216).



Figure 216. 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 215).

4. 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  $-100 \ \mu$ 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 215 on page 223).

4. In the flow rate box, enter the flow rate: **0.01** to **300**  $\mu$ 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 217 on page 227).

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  $-40 \mu$ 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 217 on page 227).

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 217).

**Figure 217.** Autosampler dialog box

Autosampler	
XYZ Robot Eject Plate Insert Plate Goto Calibration Goto Calibration	
Goto Well Needle Up Needle Down	Vial or well position box
Wash W1 W2 W3 W4	Turrentur installer
Cooler	Iemperature 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 297.

#### ✤ 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 230.
- 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<sup>™</sup> 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 89.

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 218).
    - 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 218. 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 231.
  - 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  $\mu$ L for the PLF pump or 12  $\mu$ 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 68 on page 78).

- 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 67.

# **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  $12 \mu L$ .
- 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 229.
    - 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 81).
- 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 67.

# **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 28 on page 239.

Table 27 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™
	Viper union	P/N 6040.2303
	Zero-dead-volume union for 1/16 in. OD tubing	
EASY-nLC 1000	P/N ES272	IDEX P/N P-116
with fused-silica Column Out and Waste In lines	UHPLC fused-silica union	Blind microferrule
	Non-conductive microunion with stainless steel cartridge and microferrules for 360 um OD	
	fused-silica tubing	

**Table 27.** Materials required for running the system leak test and troubleshooting leaks

To run a system leak test, follow these procedures:

- 1. To prepare the instrument for a system leak test
- 2. To run a system leak test

#### **\*** 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  $[\mu L]$  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 the next procedure, "To run a system leak test."
  - If one of these messages appears, go to "Troubleshooting a Pump That Fails the Flush Air Script" on page 229.
    - 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 the previous procedure To prepare the instrument for a system leak test).
- 2. Connect the Column Out and Waste In lines with the appropriate union (see Table 27 on page 235).

**Note** The EASY-nLC 1000 instrument ships with Column Out and Waste In lines that have nanoViper fittings at both ends. However, if you have modified the instrument to use columns that have bare fused-silica ends, these solvent lines do not have fittings on their free ends.

For an EASY-nLC 1000 instrument with the standard nanoViper Column Out and Waste In lines, do the following:

- a. Insert the nanoViper fittings on the ends of the Column Out and Waste In lines into the Viper union.
- b. Alternate tightening each fitting in small increments until you feel resistance. Then 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 93).



**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 219 shows the connection between the standard Column Out and Waste In lines for the EASY-nLC 1000 instrument. The Viper union makes a zero-dead-volume connection between the two nanoViper fittings.

Figure 219. Column Out line connected to the Waste In line (EASY-nLC 1000 instrument)



Figure 220 shows an internal view of the Viper union with nanoViper fittings connected to both ports. The two nanoViper fittings seal against each other in the center of the union, and a portion of the last thread on each nanoViper fitting is visible.

Figure 220. Viper union with nanoViper fittings connected to both ports (internal view)



**IMPORTANT** Follow the instructions in "Using nanoViper Fittings" on page 93 to ensure that the nanoViper fittings are securely connected to the union.

For an EASY-nLC 1000 instrument with modified solvent lines, connect the fused-silica Column Out and Waste In lines with the UHPLC fused-silica union (P/N ES272) that is supplied in the UHPLC Liquid Junction Kit as follows:

a. Insert the stainless steel cartridge into the PEEK holder.

Figure 221 shows an enlarged cross section of the UHPLC fused-silica union connected to the solvent lines.

Figure 221. UHPLC fused-silica union (enlarged cross section)



- b. Slide a female nut and a microferrule onto the free ends of both the Column Out tubing and the Waste In tubing.
- c. Insert the ferrules into the ends of the PEEK holder and hand tighten the nuts.

Figure 222 shows the solvent lines connected to the UHPLC fused-silica union.

Figure 222. UHPLC fused-silica union connected to the Column Out and Waste In lines



- 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 28.

# **Troubleshooting the Results of the System Leak Test**

Table 28 lists the actions required to troubleshoot the results of a system leak test.

**Tip** The "Using nanoViper Fittings Quick Reference Guide" on page 341 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.

<b>Table 28.</b> Utiput messages for a system leak test and actions to take when this test
--

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 appears, run the Flow Sensor script to calibrate flow sensor A (see "Calibrate – Flow Sensors" on page 51).	
The 0 nL/min point is off by more than $\pm 30$ nL/min, so the script cannot determine leaks.		
Flow sensor B should be calibrated.	Rerun the system leak test. If the same message appears, run the Flow Sensor script to calibrate flow sensor B (see "Calibrate – Flow Sensors" on page 51).	
The 0 nL/min point is off by more than $\pm 30$ nL/min, so the script cannot determine leaks.		
Check valve S for leaks. (Flow sensor A >100 nL/min)	Go to "Checking Valve S for Leaks" on page 240.	
Check the sample loop for leaks.	Go to "Checking the Sample Loop Connections for Leaks" on page 240.	
Check the B solvent line (valve to flow sensor) and valve B for leaks. (Flow sensor B ±30 nL/min)	Go to "Locating Leaks in the Solvent Path from Valve B to Flow Sensor B" on page 241.	
Check from pump A to flow sensor A for leaks. (Pump A flow loss >1000 nL/min)	Go to "Locating Leaks in the Solvent Path from Pump A to Flow Sensor A" on page 242.	
Check from valve S to valve W, including valve W, for leaks.	Go to "Locating Leaks In or Between Valve S and Valve W" on page 244.	
Check solvent lines between flow sensor A and valve S, including the mixing Tee and flow sensor B, for leaks.	Go to "Locating Leaks Between the Flow Sensors and Valve S" on page 252.	

## **Checking Valve S for Leaks**

The system leak test returned the following message (see Table 28 on page 239):

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 and Inspecting the Rotor Seal and Stator" on page 84).
- 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 85).
- 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 Leaks**

The system leak test returned the following message (see Table 28):

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 341. 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 235).
- 3. If the script still fails, replace the rotor seal (see "Replacing the Rotor Seal" on page 85).
- 4. If the problem persists after replacing the rotor seal, replace the sample loop (see "Replacing the Sample Loop" on page 100).

## Locating Leaks in the Solvent Path from Valve B to Flow Sensor B

The system leak test returned the following message (see Table 28 on page 239):

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 223 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 223. 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 341. 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 and Inspecting the Rotor Seal and Stator" on page 84.
- 2. If necessary, replace the rotor seal as described in "Replacing the Rotor Seal" on page 85.

## Locating Leaks in the Solvent Path from Pump A to Flow Sensor A

The system leak test returned the following message (see Table 28 on page 239):

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 233 where you determine if the leak is coming from the solvent path between the pump and the valve or from the pump itself.

Figure 224 shows the solvent path from pump A to flow sensor A.

Figure 224. 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 233.
- 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 341. 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 81 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 Leaks In or Between Valve S and Valve W

If the system leak test returns the following message, follow the workflow in Figure 225 and Figure 226 or the procedure on pages 246–252 to locate and remedy the leak.

Check from valve S to valve W, including valve W, for leaks.

Figure 225. Workflow for locating leaks in the solvent path between valves S and W or in these valves (Part 1)





Figure 226. Workflow for locating leaks in the solvent path between valves S and W or in these valves (Part 2)

Figure 227 shows the path between valves S and W for the EASY-nLC 1000 instrument.

Figure 227. 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. Tighten the test union connections as follows:
  - 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 93.

**Note** The connections to the leak test union are one of the most common sources of leaks for the system leak test.

- 2. To leak test the connections to the test union, do the following:
  - a. 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 228 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 221.

Figure 228. Home > Overview page showing the readback for flow sensor A



- b. 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.

- 3. 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.
- 4. 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 81 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.
- 5. 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.
- 6. 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 229 shows the Waste In line disconnected from the leak test union and the free end of the union plugged with a blind fitting.



#### Figure 229. 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 235.

For ordering information, see Appendix C, "Consumables and Replacement Parts."

- If the flow is >100 nL/min, go to the next step.
- 7. 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 235 to confirm that the system is leak tight.
  - If the flow is >100 nL/min, go to the next step.
- 8. 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 230 shows port 3 of valve S plugged with a blind fitting.

Figure 230. 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 235.

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 81. Then, rerun the system leak test as described in "Running a System Leak Test" on page 235.

## Locating Leaks 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 28 on page 239:

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 section for your instrument to locate and remedy the leak:

- "EASY-nLC 1000: Locating a Leak Between the Flow Sensors and Valve S," on pages 253–263
- "EASY-nLC II: Locating a Leak Between the Flow Sensors and Valve S" on page 264

#### 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 231 or the procedure on pages 254–257.

Figure 231. EASY-nLC 1000 workflow for locating leaks between the flow sensors and valve S



#### 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**  $\mu$ L.

For information about setting up the Analytical Column Equilibration script, see step 22 on page 246.

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 221.

b. Disconnect the nanoViper fitting from the mixing Tee outlet. Then, block the mixing Tee outlet with a blind fitting (see Figure 232).



#### Figure 232. 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 37 on page 334.

- iv. Rerun the system leak test (see "Running a System Leak Test" on page 235).
- 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 233 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 233. 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 261, 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 231 on page 253 or the procedure on pages 254–257, 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 233 on page 257, with the solvent channel B disconnected from the mixing Tee.

Follow the workflow in Figure 234 or the procedure on pages 259–260 to locate and remedy the leak.

Figure 234. EASY-nLC 1000 workflow for locating a leak between flow sensor A and the mixing Tee (Part 1)



# 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 235 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 37 on page 334.

- iv. Reconnect the solvent lines and rerun the system leak test (see "Running a System Leak Test" on page 235).
- 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.
      - iii. Rerun the system leak test (see "Running a System Leak Test" on page 235).
    - 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 235).

#### EASY-nLC 1000: Locating a Leak Between Flow Sensor B and the Mixing Tee

By following the workflow in Figure 231 on page 253 or the procedure on pages 254–257, 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 233 on page 257, with the solvent channel B disconnected from the mixing Tee inlet.

Follow the workflow in Figure 236 or the procedure on pages 262–263 to locate and remedy the leak.

Figure 236. 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 237).



Figure 237. 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 39 on page 337.

- iii. Reconnect the system plumbing and rerun the system leak test (see "Running a System Leak Test" on page 235).
- 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 37 on page 334.

iii. Rerun the system leak test (see "Running a System Leak Test" on page 235).

#### EASY-nLC II: Locating a Leak Between the Flow Sensors and Valve S

Figure 238 on page 265 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**  $\mu$ L.

For information about setting up the Analytical Column Equilibration script, see step 22 on page 246.

- 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 36 on page 332.

- 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 238. 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 backpressure 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 backpressure is caused by a blocked column or a blocked Column Out line. The Column Out line has an ID of 30  $\mu$ m for the EASY-nLC II instrument and 20  $\mu$ 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	Powder-free gloves
to loosen the stainless steel	• EASY-nLC II instrument: HPLC union, P/N SC600
intiligs	• EASY-nLC 1000 instrument: Viper 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 backpressure. If the backpressure 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," next procedure
- 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 269
  - "Troubleshooting a System Blockage when Test Solvent B Fails" on page 275

## **Running the Back Pressure Test for the A and B Solvent Paths**

The Back Pressure script determines instrument backpressure for solvent lines A and B. The script runs at a preset flow and measures the backpressure 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 (Viper union)

- 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 269.
    - 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 275.

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 268).

#### \* To verify the system blockage

- 1. Run the Precolumn Equilibration script and check the backpressure 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  $[\mu L]$  box, enter **140**  $\mu L$ .
  - d. In the Flow [ $\mu$ L/min] box, enter 2  $\mu$ L/min.
  - e. Leave the Max Pressure [bar] box blank.
  - f. Press Start.
  - g. Monitor the pump A pressure trace.
- 2. Depending on the backpressure, do the following:
  - If the backpressure is below the pressure threshold, the system blockage has been removed. To confirm that the backpressure is below the pressure threshold, rerun the Back Pressure script with the **Test Solvent A** check box selected.
  - If the backpressure 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 271.

Instrument	Pressure threshold
EASY-nLC II	100 bar
EASY-nLC 1000	175 bar

Figure 239 shows the areas that the following procedure systematically checks for blockage.

Figure 239. 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 269 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 239 on page 270).
  - c. Depending on the backpressure, do one of the following:
    - If the pressure is below the threshold, valve W is blocked. Go to "Maintaining the Rotary Valves" on page 81.
    - 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 backpressure, 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 backpressure, 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 backpressure, 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 81.

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 backpressure, 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 backpressure, 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 backpressure, 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 backpressure, 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 backpressure 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 104.
    - 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 backpressure, 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 backpressure, 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 backpressure, 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 backpressure, 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 102.

### **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 268).

Figure 240 shows the areas that this procedure systematically checks for blockage.

Figure 240. 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  $\mu$ 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 222.

- d. Monitor the Pressure readback box for pump B. Then, depending on the backpressure, 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 backpressure, 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 104.
    - 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 backpressure, 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 81.

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	Pressure threshold
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 102.

## **Troubleshooting the Autosampler Aspiration**

10015	Parts and materials
1/4 in. open-ended wrench to loosen the stainless steel fittings	<ul> <li>Powder-free gloves</li> <li>Sample vial filled with solvent A</li> <li>Microtiter plate (if you are using a microtiter plate format)</li> </ul>

Troubleshooting the autosampler aspiration requires these tools and materials:

#### \* 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 [ $\mu$ L], enter **10** for the PLF model pump or **12** for the PLU model pump (see Figure 241).

#### Figure 241. Flush Air script

SCIENTIFIC	Home Batch Setu	p Method Setup	Maintenance Configuration	
Scripts Log Book Support	Category: Prepare	Name: Flush air 🔹	Pump S — Desired flow — Pressure 1.0 250.0 250.0	1
	Parameter         Flush pump A         Flush pump B         Flush pump S         Flush threshold [µl]	<ul> <li>✓ Value</li> <li>□</li> <li>□</li> <li>∞</li> <li>12.00</li> </ul>	2000 r 2000 r 2000 r 2000 r 1500 r 1500 r 1500 r 500 0.0 r 500 r	
Job 00:00:00 6 May 2011 11:15 admin Exit	Schedule ST.	ART 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 94).
    - If pump S cannot aspirate solvent through the Teflon waste tubing, go to "Troubleshooting a Pump That Fails the Flush Air Script" on page 229.
- 5. To determine if the autosampler needle is going to the correct position and depth, go to the next procedure, "Checking Sample Pickup."

## **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  $\mu$ 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  $\mu$ L and your default flow (see Figure 242).

Figure 242. Sample Pickup script under the Maintenance menu tab

Category:	Name:	Category:	Name:	Category:	Name:
Test	Sample pickup	Test	Sample pickup 👻	Test	Sample pickup
Description Para	ameters Output	Description Paran	neters Output	Description P	arameters Output
Test autosampler sam fill plate well with	nple pick up. Please n a defined volume	Parameter	▼Value	'Sample pickup' s Prime pump - fill	tarted ing
before running test. Max volume: Loop vol	ume - 2µ1.	Volume [µl]	10.00		
Max flow: 40.0 µl/mi	.n.	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  $\mu$ 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 8. For information about the Flush Air script, see "Prepare – Flush Air" on page 33.

## **Troubleshooting Communication Problems**

To troubleshoot communication problems, see these sections:

- Network Access
- Network Connection Failures
- Testing the Network Connection to the EASY-nLC Computer

### **Network Access**

Table 29 contains troubleshooting tips for network access problems.

Table 29. 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 30 lists the possible causes of a network connection failure. Figure 243 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 243. Network connection failure

Thermo EASY-nLC Confi	guration	
EASY-nLC Network Conf	iguration	The TD Address on the found in the
IP Address/Hostname:	10.32.134.79	About box on the EASY-nLC instrument
User name:	admin	Run samples as this user (User must exist on the EASY-nLC)
Test Connection		Connection failed!
LC device driver and in	CServer 2.0.0.0	
Autosampler Configurati	on	Solvents for LC Pump
Autosampler:		A: water
Plate installed:	▼ Edit.	
Sample layout: Row fit	st Start pos: A1	B: acetonitrile
ОК		Cancel

Table 30. Possible causes of a network connection failure

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 284. 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.

## **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 244).

Figure 244. Command Prompt window with the default IP address for the EASY-nLC instrument

🔤 C:\WINDOWS\system32\cmd.exe	- 🗆 ×
Microsoft Windows XP [Version 5.1.2600] (C) Copyright 1985-2001 Microsoft Corp.	<u> </u>
C:\Documents and Settings\lab>telnet 172.16.0.103 6666	
•	▼ ▶ //

- 5. Press the ENTER key.
  - If the connection is set up correctly, a login prompt appears in the Telnet window (see Figure 245).
  - If the connection is not set up correctly, the following message appears:

Could not open connection to host.

Figure 245. Telnet window with login prompt



## 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:
  - Connect one probe to an unpainted surface on the EASY-nLC chassis and a. connect the other probe to an unpainted surface on the external hardware chassis (see Figure 246).

Figure 246. 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

8

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 instrument 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," next procedure
- "Replacing the Adapter Plate in the ASA Model Autosampler" on page 290

### **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/Insert Tray**.

The tray compartment opens. Figure 247 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 247. Open tray compartment



3. Remove the current adapter plate by pulling it up and away from the tray holder (see Figure 248).

Figure 248. 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/Insert Tray.

Figure 249 shows an ejected tray.

Figure 249. 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 250.

Figure 250. Screws that secure the adapter plate to the tray



3. Place the new adapter plate on the tray (see Figure 251).

Figure 251. 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 the next procedure "Managing Plate Formats." After you create a new plate format, perform the calibration routines described in this chapter.

## **Managing Plate Formats**

When the EASY-nLC instrument is initially installed, the touch-screen 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," next procedure
- "Creating a New Plate Format" on page 293
- "Deleting a Plate Format" on page 295

### **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 252 shows the Tools view for the autosampler.

Thermo SCIENTIFIC	Home	Batch Setup	Method Setup	Maintenance	Configuration	
Scripts	Devices					
Log Book		Na	me	Туре		Add Device
Support	Co	oled Autosampler (A	3C) [Autosampler]	Autosampler		
Devices	EAS	SY-Spray		Column Heater		Remove Device
	EAS	SY-nLC™ [HPLC]		HPLC		Assign Slave
	About	Properties	Tools			
	Coole	d Autosampler	(ASC) [Autosamp	oler]		
Job 00:00:00	Plate	New	Calibrate	:Vials		/ash Calibrate
	6×	8 Vials			Manipul	Reset
23 aug 2010 12:38 admin		Copy To	Dele e	Use		
Exit						
		Copy To button	Calibrate button	Sele	cted plate form	nat

Figure 252. 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 application or data system to control your liquid chromatograph/mass spectrometer (LC/MS) system, the plate selection for the EASY-nLC instrument in the EASY-nLC Configuration dialog box (Foundation Instrument Configuration) 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 252 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 253).

Figure 253.	Create	New F	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:  $6 \times 8$  (vials),  $8 \times 12$  (96-well plates), or  $16 \times 24$  (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 297 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 297 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 293).
- 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  $6 \times 8$  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 292).
  - b. Press Use.
- 5. Remove the left side panel so that you can watch the movement of the needle.

To calibrate the plate, see "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 303.

## **Calibrating Plates**

For information about selecting or creating a plate format, see "Managing Plate Formats" on page 292.

**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 31 lists the top left and bottom right positions for the three plate formats.

Table 31. Top left and bottom right positions on the plate formats

Plate format	Top left position	Bottom right position
6 × 8	A1	F8
8×12	A1	H12
16×24	A1	P24

#### ✤ 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 296).
- 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 254).

Figure 254. Tools view for the autosampler

About	Properties	Tools		
Cooled	Autosampler	(ASC) [Au	tosampler]	
Plate				Vials / Wash
	New	Calibrate	Current: Vials	Calibrate
6x8	Vials		•	Reset
Co	ру То	Delete	Use	

d. In the Plate area of the Tools view for the autosampler, press **Calibrate** (see Figure 254 on page 298).

The Autosampler Plate Calibration dialog box opens.

Figure 255 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 readouts for the *xyz* axes are unpopulated, - / [-].

Autosampler Plate Clibration

Figure 255. 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 256).

**Figure 256.** 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 discontinued 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 257 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 readouts for the *xyz* axes are unpopulated, - / [-] (see Figure 258).

Autosampler Pla	te Calibration
Calibrate Plate HT Needle In/Out	MTP
- 7 [-]	
Reset	Needle Left/Right     ←     - / [-]     →     ⇒     Capture
	Save Eject Close

Figure 258. 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 296.
- 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 259).

3. Press the bottle container in W4 (1) (see Figure 259).

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 259. 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.

# 9

## **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

## **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.

**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 application. Thermo Fisher Scientific Technical Support cannot initiate this communication link.

The EASY-nLC instrument uses Secure Shell (SSH<sup>™</sup>), a network protocol for secure data communication, to connect to the support server, which uses the Linux<sup>™</sup> 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 46.226.216.237, 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 email 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.

#### **\*** 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 260) 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 46.226.216.237, port 22.



Figure 260. 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 261).

Figure 261. Network page of the Configuration menu with a fixed IP address for the MS LAN

<b>Thermo</b>	Home Batch Setup Method Setup Maintenance Configuration				
Users	LAN				
Connections	Profile: MS LAN   Configuration: O Dynamic   Fixed				
Network Time Data	IP: 172 . 16 . 0 . 103 Subnet: 255 . 255 . 0 . 0 Gateway: 0 . 0 . 0 . 0 DNS: 0 . 0 . 0 . 0 Name: LC120 Domain:				
	Apply				
Job 00:00:00	Support server           IP:         46         226         216         Default IP				
8 Apr 2015 15:07	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: **46.226.216.237**.
  - 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 262).

Figure 262. Support page of the Maintenance menu

SCIENTIFIC	Home Batch Setup Method Setup 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): Show Clear				
	Log file copy:				
	● To home directory ○ To USB removable storage				
	Copy Log Files Status: Idle				
	Contact:				
Job 00:00:00 30 Aug 2010 14:14 admin Exit	For technical support, go to "Contact Us" on http://www.thermoscientific.com or go to http://www.proxeon.com/support				

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 263).
- If the instrument does not have the latest firmware file installed, the Upgrade is Available dialog box appears (see Figure 266 on page 315).
- c. If the Upgrade is Available dialog box appears, do one of the following:
  - Upgrade the firmware as described in "Downloading the Latest Firmware File" on page 315.

-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 263. 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 the next procedure, "To troubleshoot a failed connection to the support server."

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."

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 **MS 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 308).
- 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 262 on page 309).

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 264).

Figure 264. 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.

Cop	ογ Το 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 265 and Table 32). **Figure 265.** Files on the USB memory stick

Compute	rr ► USB20FD (G:) ►		Q
Organize 🔻 Share wit	h ▼ Burn New folder 8	≣ ▼	
<ul> <li>▲ ★ Favorites</li> <li>※ Recent Places</li> <li>※ Desktop</li> <li>※ Downloads</li> <li>▷ Libraries</li> <li>▲ ● Computer</li> <li>▷ ▲ OSDisk (C:)</li> <li>▷ ▲ Local Disk (D:)</li> <li>▷ ▲ USB20FD (G:)</li> <li>▷ ▲ Network</li> </ul>	<ul> <li>Name</li> <li>user_admin.zip</li> <li>user_guest.zip</li> <li>etc.zip</li> <li>logfiles.zip</li> <li>maintlog.zip</li> </ul>	•	Select a file to preview.
5 items			

File name	Description
user_admin.zip	Contains a batch log and one folder for each batch that admin users have run with information on samples, methods, and EASY-nLC device performance.
user_guest.zip	Contains a batch log and one folder for each batch that guest users have run with information on samples, methods, and EASY-nLC device performance.
etc.zip	Contains system configuration files.
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.

Table 32.	Zipped files	created	by pressing	Copy Log	Files
-----------	--------------	---------	-------------	----------	-------

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 266 shows the message for an instrument with firmware version 2.4.

Figure 266. Firmware upgrade message

⊻ An upgrade is available	• ×
The installed software can be upgraded to version "2.5#30	032".
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 instrume log files and optional firmware file are completed.	ent
Download Skip	

2. Click Download.

When the file transfer is complete, the Transfer of Log and Firmware Files Completed dialog box appears.

Figure 267 shows the transfer information for an instrument with firmware version 2.4.

Figure 267. Information for upgrading from a previous version

	• × •
Transfers completed: The log files have been transferred to the remote support s and the firmware file has been downloaded to the instrumer firmware folder.	erver nts
To upgrade the application software you should now shut d and power-cycle the instrument. When the instrument restarts double-tap the 'Upgrade' butto on the desktop.	own on
Note: There are 4 further upgrades to download and install after this one. After the downloaded upgrade has been installed you sho connect to the Remote Support Server again to fetch the next upgrade.	r ould
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 33 lists the firmware versions that you must install to upgrade the firmware to the current version.

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 or 3.1.6.1	3.2.0.2
3.2.0.2	current as of April 2015

Table 33. Firmware installation matrix



## **Returning the EASY-nLC Instrument**

If it is necessary to return the EASY-nLC instrument to the factory, follow the instructions in this chapter.

#### Contents

- Transport Instructions
- Declaration of Contamination

### **Transport Instructions**

Before packaging the instrument for transport, take care to remove all solvents from the instrument.

- **\*** To prepare the instrument for transport
- 1. Press Home > Overview.
- 2. Press Eject/Insert Tray.
- 3. Remove the plates/tubes.
- 4. Remove the waste beaker and the wash/waste bottles.
- 5. Press Eject/Insert Tray.
- 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 discontinued ASA autosampler only: Secure the autosampler by using the transport collars (see Figure 268) before shipping the instrument. (This is not necessary on the ASC model that is current as of this revision of the *EASY-nLC Series Troubleshooting and Maintenance Guide*.)



Figure 268. (ASA autosampler only) Transport collar location

- 8. Fill out the Declaration of Contamination of Equipment as described in the next section "Declaration of Contamination."
- 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	ment:		-
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 harmforprecautions associated with the subs	ul substances incluc tances.	ling chemical name	e and symbol, and
Were any of the harmful substances			
Radioactive?		[] Yes	[ ] No
• Toxic?		[] Yes	[ ] No
Corrosive?		[] Yes	[ ] No
<ul> <li>Explosive?</li> </ul>		[] Yes	[ ] No
Has the equipment been properly de	econtaminated and	/or cleaned before	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	d sufficient to

Name:			
Job title:	Organizatio	on:	
Address:			
Telephone:	Fax:	E-mail:	
Signature:			

## **Error Codes**

Table 34 lists the possible error codes that might appear.

Table 34. 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	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 the hard drive.
4	ERROR_IO_CREATE_ FILE	Failed creating log file 'LOG_FILE_NAME'	I/O error	Internal filesystem error - Replace the hard drive.
5	ERROR_IO_CREATE_ FOLDER	Failed creating log folder 'LOG_FOLDER_NAM E'	I/O error	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.

Α –

Table 34. 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.	Turn off the EASY-nLC instrument, and then turn it back on.
32	ERROR_SENSIRION_ FACTOR_NOT_ FOUND	Flow sensor factor not found in info output. Factor unknown.	Error getting flow sensor scaling factor	Turn off the EASY-nLC instrument, and then turn it back on.

#### Table 34. 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	Turn off the EASY-nLC instrument, and then turn it back on.
34	ERROR_SENSIRION_ NO_DATA	No data received from flow sensor for <i>X</i> seconds	[See issue description.]	Turn off the EASY-nLC instrument, and then turn it back on. 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	Turn off the EASY-nLC instrument, and then turn it back on.
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.]	Turn off the EASY-nLC instrument, and then turn it back on. If the problem persists, replace the flow sensor.
				See "Replacing a Flow Sensor" on page 104.
38	ERROR_SENSIRION_ ECHO_MISMATCH	Flow sensor echo "ECHO" does not match command	Flow sensor fails to "talk back."	Rare error - Connect the EASY-nLC system to remote support.
		COMMAND .		See "Connecting the EASY-nLC Instrument to the Support Server" on page 306.
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.

#### Table 34. Error codes (Sheet 4 of 8)

No.	Issue code name	Description	Cause	Solution
40	ERROR_SENSIRION_ IN_IDLE_MODE	Cannot calibrate sensor in idle mode.	Flow sensor in idle mode during calibration	Turn off the EASY-nLC instrument, and then turn it back on. Check the flow sensor functionality.
41	ERROR_SENSIRION_ CALIBRATION	Cannot store flows larger than 9999 nL/min.	Abnormal value trying to be written to the flow sensor	Turn off the EASY-nLC instrument, and then turn it back on. Check flow sensor functionality.
42	ERROR_RS232_INIT	Initialization of interface "PORT_NAME" failed	Access to interface failed	Turn off the EASY-nLC instrument, and then turn it back on.
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 the touch-screen application. See Chapter 8, "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 the touch-screen application. See Chapter 8, "Calibrating the Autosampler's XYZ Robot."

#### Table 34. Error codes (Sheet 5 of 8)

No.	Issue code name	Description	Cause	Solution
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 the touch-screen application. See Chapter 8, "Calibrating the Autosampler's XYZ Robot."
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 touch-screen application and the EASY-nLC Configuration dialog box (Foundation Instrument Configuration). 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 34. 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. Turn off the EASY-nLC instrument, and then turn it back on. 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 9).

#### Table 34. 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 149.
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	Turn off the EASY-nLC instrument, and then turn it back on.
85	IC_STATUS_FLAG_ WARNING		Monitor reported flag(s) raised (log file only).	Follow the error message instructions.
86	IC_STATUS_FLAG _SEVERE		Monitor reported flag(s) raised (in user interface, current run stopped).	Follow the error message instructions.
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 239.
88	IC_STATUS_FLAG_ ERROR		Monitor reported flag(s) raised (in user interface).	Follow the error message instructions.

Table 34. 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 does not 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 239.

## **Mobile Phase Viscosity**

Table 35 lists the viscosities for two-solvent mobile phases consisting of a water/methanol mixture or a water/acetonitrile mixture.

Б

	η <sub>25</sub> (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
Reference: Herman, D. P.; Billiet, A. H	H.; de Galan, L. <i>J.</i>	of Chromatogr., <b>1989</b> , 1, 463.

Table 35. Mobile-phase viscosity at 25 °C for reversed-phase gradients

## **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 appendix.

#### 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 websites:

• For the EASY-nLC 1000 instrument

www.proxeon.com/productrange/nano\_IC\_easy-nlc\_1000/accessories\_spares

• For the EASY-nLC II instrument

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 bottles and lids
- All flow lines including Tee-pieces and fittings (nuts, ferrules, sleeves, and valve stops)
- All sample vials, microtiter plates, and associated lids and mats
- Solvent filters, inline filters, and all associated filter holders
- Valve rotors and stators
   Pump p

- Autosampler needle
- Check valves
- Columns
- Fuses
- Pump piston seals

## Solvent System Schematic for the EASY-nLC II Instrument

Table 36 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 269.

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 it does not include the mixing Tee itself.

Table 36. 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 Valve B to flow sensor B	PEEKsil, 50 µm ID, 25 cm length	LC222
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	ASA model, PEEKsil, 150 µm ID, 55 cm length	LC251
		ASC model, nanoViper, 150 μm ID, 55 cm length	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 Port 2 of valve B to check valve B	Teflon, 500 µm ID, 15 cm length	Kit, 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 Tubing from check valve B to the waste beaker	Teflon, 500 µm ID, 39 cm length	N/A
14	Tubing from valve W to the waste beaker	Teflon, 500 µm ID, 27 cm length	LC263



Figure 269. 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 37 lists the tubing requirements and identifies the tubing components shown in Figure 270. 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 37. 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	nanoViper, 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





## **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 38 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 38. Common replacement parts for the EASY-nLC II instrument

### **Common Replacement Parts for the EASY-nLC 1000 Instrument**

Table 39 lists the annual consumption of common replacements parts for the EASY-nLC 1000 instrument.

Table 39. 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 Chapter 5, "Routine Maintenance."

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 345 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 backpressures 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

#### Connecting nanoViper Fittings

#### 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 235).

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.



## 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
- Autosampler needle connected to port 1 of Valve S

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 Viper 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	Parts and 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
	Pump-to-pressure sensor flow line for an EASY-nLC 1000
	instrument, LC512

#### **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  $\mu L$  position.

- 2. Exit the EASY-nLC 1000 system and turn off the power.
- 3. Using a #2 Phillips head screwdriver, make a quarter-turn to 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  $\mu$ L (empty) to 140  $\mu$ L (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

	300 µl/min
140.0 µI	Stop Start
Pressure:	<b>53.0</b> Bar
Position:	<b>27.75</b> 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 345).
- 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 345.
  - 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. If the script detects a leak, replace the stainless steel tubing that connects the pump to the pressure sensor.
- 14. . 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	Parts and 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  $\mu L$  position.
- 2. Exit the EASY-nLC II system and turn off the power.
- 3. Using a #2 Phillips head screwdriver, make a quarter-turn to 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 or the rotor seal and the stator when you detect a leak or blockage in the valve.

**IMPORTANT** To determine whether the leak indicated by the Leaks script is caused by a leaking rotary valve, you must exclude these other possible causes of a Leaks script failure:

- Excess air in the subsystem
- A leak from the tubing that connects the pump to the pressure sensor
- A leak from the tubing that connects the pressure sensor to the valve
- A worn pump seal

#### Contents

- Accessing the Rotary Valves
- Replacing the Rotor Seal
- Replacing the Rotor Seal and the Stator
- Returning the Instrument to Normal Operation



**CAUTION** Wear powder-free safety gloves when working with solvents and handling the wetted components of the instrument.

Replacing a piston seal or stator requires these tools, materials, and replaceable parts.

#### Tools

#### Materials

- 9/64 in. hex wrench
- LC/MS-grade methanol
- 1/4 in. open-ended wrench
- Powder-free safety glovesLint-free tissues and Q-tips
- #2 Phillips head screwdriver

#### **Replaceable parts**

- Rotor seal
  - Valves with serial numbers V-010000 and higher, P/N LC228
  - EASY-nLC II valves with serial numbers V-009999 and lower, P/N LC224
- Stator
  - EASY-nLC II stator for valves with serial numbers V-001000 to V-019999, P/N LC226
  - EASY-nLC 1000 stator for valves with serial numbers V-020000 and higher, P/N LC526
- Prebent stainless steel tubing, P/N LC513

#### **Accessing the Rotary Valves**

- 1. Close down the EASY-nLC system as follows:
  - a. On the lower-left corner of the touch screen, press **Exit**. A confirmation dialog box opens.

Administrator		Logout
		Change password
Stop Application	Power Down	Cancel

b. Press Power Down.

A progress bar appears. When the progress bar is completely filled, a message appears indicating you can safely turn off the instrument.

- 2. Turn the instrument's power switch to the Off position. The power switch is on the instrument's back panel.
- 3. Using a #2 Phillips screwdriver, make a quarter-turn to loosen the three captive screws that secure the panel to the right side of the EASY-nLC instrument, and then remove the panel.

### **Replacing the Rotor Seal**

- 1. Access the rotary valves (see procedure above).
- 2. Disconnect the following solvent lines:
  - For valve A or B, disconnect the low-pressure solvent line from port 2 (Figure 1). Cap the end of the solvent line with a union and blank fitting.

Figure 1. Valve A or B solvent line connections (EASY-nLC 1000)



- -Stainless steel tubing connecting the valve to the pressure sensor
- For valve A, B, or S in an EASY-nLC 1000 instrument or valve B in an EASY-nLC II instrument, disconnect the stainless steel tubing that connects the valve to the pressure sensor as follows:
  - i. Using a 1/4 in. open-ended wrench, completely loosen the two fittings that secure the tubing to the valve and the pressure sensor.
  - ii. To remove the loosened fittings from their receiving ports, gently squeeze the bent portion of the prebent tubing.





**CAUTION** To avoid damaging the stator and rotor seal, follow these guidelines:

- To prevent unbalanced screw tension, alternate loosening the two screws that secure the stator to the valve in quarter-turn increments until all the load is removed.
- Never rest the stator on its sealing surface.
- 3. Using a 9/64 in. L-hex wrench, alternate loosening the two hex screws that secure the stator to the valve in quarter-turn increments until all the load is removed. Remove the loosened screws from the stator and set them aside for reuse. Pull the stator away from the valve driver and carefully rest the stator, with its outer surface face down, on the top of the instrument or leave the stator suspended by the tubing.
- 4. Gently pry the rotor seal away from the valve driver.
- 5. Clean the stator's sealing surface with a lint-free tissue or Q-tip soaked in methanol (Figure 2). Verify that its sealing surface is not damaged. If the stator shows visible damage, replace it.

Figure 2. Cleaning the stator's sealing surface



- 6. Carefully mount the new rotor onto the valve driver, ensuring that the rotor seal's sealing surface (with engraved flow passages), is facing out.
- 7. Mount the stator onto the valve driver.
- Insert the two hex screws that secure the stator to the valve and hand-tighten them until you feel resistance. Using a 9/64 in. L-hex wrench, alternate tightening the screws in quarter-turn (90-degree) increments until they are evenly torqued and the stator is flush against the valve body.
- 9. Reconnect the solvent lines. To reconnect a swaged stainless steel fitting to its receiving port, hand-tighten the fitting as you press the tubing against the bottom of the port, and then use a 1/4 in. open-ended wrench to tighten the fitting by an additional 1/4 to 1/2 turn.
- 10. Go to "Returning the Instrument to Normal Operation."

#### **Replacing the Rotor Seal and the Stator**

- 1. Access the rotary valves. See "Accessing the Rotary Valves," on the previous page.
- 2. Disconnect the solvent lines as described in step 2 of "Replacing the Rotor Seal."
- 3. Disconnect the remaining solvent lines from the valve. Use a 1/4 in. open-ended wrench to loosen stainless steel fittings. Use a black nanoViper knurled nut to loosen nanoViper fittings. For valve S, remove the sample loop from the valve.
- 4. Remove the damaged stator as described in step 3 of "Replacing the Rotor Seal." Set the stator aside for disposal.

- 5. Gently pry the damaged rotor seal away from the valve driver.
- 6. Carefully mount the new rotor seal onto the valve driver, ensuring that the rotor seal's sealing surface (with engraved flow passages), faces out.
- 7. Mount the new stator onto the valve driver.
- 8. Secure the stator to the valve driver as described in step 8 of "Replacing the Rotor Seal."
- 9. Reconnect the solvent lines as described in step 9 of "Replacing the Rotor Seal." Use a black nanoViper knurled nut to tighten the nanoViper fittings. Take care to avoid overtightening these fingertight fittings.
- 10. Go to "Returning the Instrument to Normal Operation."

## **Returning the Instrument to Normal Operation**

- 1. Turn on the EASY-nLC instrument and log in 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.

- 3. 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** and wait for the script to finish.
- 4. 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.

To run a system leak test, you must connect the Column Out line to the Waste In line with the appropriate union.

5. Reconnect the right side panel to the instrument. Tighten the three captive screws with a #2 Phillips screwdriver.

Running a system leak test requires a leak test union.

#### Leak test union

EASY-nLC II: Zero-dead-volume (ZDV) union for 1/32 in. OD tubing, P/N SC600

EASY-nLC 1000: Viper (ZDV) union for 1/16 in. OD tubing, P/N SC900

EASY-nLC 1000 with fused-silica Column Out and Waste In lines: UHPLC fused-silica union for 360  $\mu m$  OD fused-silica tubing, P/N ES272

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

#### **Removing the Damaged Autosampler Needle**

#### To remove the autosampler needle

- 1. Using a #2 Phillips screwdriver, make a quarter-turn to loosen the three captive screws that secure the panel to the right side the EASY-nLC instrument, and then remove the panel.
- 2. Disconnect the autosampler needle from port 1 of valve S as follows:
  - If the autosampler needle has a stainless steel fitting, use a 1/4 in. open-ended wrench to unscrew the fitting.
  - If the autosampler needle has a nanoViper fitting, use the black knurled nut to unscrew the fitting.

Figure 1. Valve S solvent line connections for the EASY-nLC II system







3. For the ASA or obsolete ASC autosampler needle, remove the nut, ferrule, and sleeve from the needle tubing.

Figure 3. ASA 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 **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

bled	Autosampler	(ASC) [Autosampler]	
te			Vials / Wash
	New	Calibrate Current: 8x12/Abgene (AB-0800)	Calibrate
-			
8x1	2/Abgene (AB-0	800)	Manipulator
_			Reset
	- 1	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



- 5. Connect the needle to port 1 of valve S as follows:
  - For the ASA autosampler needle, do the following:
    - i. Slide the provided blue sleeve and metal fittings onto the tubing.
    - ii. 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.
  - For the ASC autosampler needle, do the following:
    - i. Insert the nanoViper fitting into the receiving port. Then turn the screw clockwise until you feel resistance.
    - ii. Using the black knurled nut, turn the screw clockwise to an angle between 0 and 45 degrees (1/8-turn).
- 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 two 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  $\mu$ L for the EASY-nLC II system or 12  $\mu$ L for the EASY-nLC 1000 system.

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