

Preparative HPLC Troubleshooting Guide

Your guide to solving common problems and staying productive

Places to start

Errors

- Use clean, low-volume bottles to avoid degradation
- Use low HPLC grade solvents

Prep and post-run checks

- Inspect solvent bottles and inlet lines
- Check and fix any leaks when possible, especially for long runs
- Use the automatic system conditioning protocol

Setup tasks

- Perform baseline and system checks when ready to start a run
- Check and seal valves
- Flush the system with the composition of your application

Ready tasks

- Change solvent and/or column, as necessary when the flow is stopped
- Check system performance and change flow if necessary

Post-run checks

- Flush of solvents to remove salt deposits and particulates from the system
- Flush the system with appropriate storage solvent and/or water after the system

Daily checks

- Remove all sources of air and contaminants from system
- Regenerate flow lines (change solvents with water or water with 100% organic)
- Clean system with 10% organic and 100% water as needed
- Perform system health checking or leak inspection if volume



Maintenance

Agilent's HPLC systems help you manage and optimize all resources to achieve high quality chromatographic results. Maximize efficiency by ensuring high system performance, maximum reliability, and a robust footprint.

- Regular tasks to ensure performance
- Daily maintenance of all types of modules
- Complete or major preventive and corrective maintenance

Flow issues



Problem	Cause	Solution
Flow rate is lower than expected	Flow rate is lower than expected	Check for leaks, check solvent level, check for clogs
Flow rate is higher than expected	Flow rate is higher than expected	Check for leaks, check solvent level, check for clogs
Flow rate is unstable	Flow rate is unstable	Check for leaks, check solvent level, check for clogs
Flow rate is zero	Flow rate is zero	Check for leaks, check solvent level, check for clogs
Flow rate is too low	Flow rate is too low	Check for leaks, check solvent level, check for clogs
Flow rate is too high	Flow rate is too high	Check for leaks, check solvent level, check for clogs
Flow rate is too fast	Flow rate is too fast	Check for leaks, check solvent level, check for clogs
Flow rate is too slow	Flow rate is too slow	Check for leaks, check solvent level, check for clogs
Flow rate is too low	Flow rate is too low	Check for leaks, check solvent level, check for clogs
Flow rate is too high	Flow rate is too high	Check for leaks, check solvent level, check for clogs
Flow rate is too fast	Flow rate is too fast	Check for leaks, check solvent level, check for clogs
Flow rate is too slow	Flow rate is too slow	Check for leaks, check solvent level, check for clogs

Peak shape issues



Problem	Cause	Solution
Peak is broad	Peak is broad	Check for leaks, check solvent level, check for clogs
Peak is narrow	Peak is narrow	Check for leaks, check solvent level, check for clogs
Peak is tall	Peak is tall	Check for leaks, check solvent level, check for clogs
Peak is short	Peak is short	Check for leaks, check solvent level, check for clogs
Peak is wide	Peak is wide	Check for leaks, check solvent level, check for clogs
Peak is thin	Peak is thin	Check for leaks, check solvent level, check for clogs
Peak is low	Peak is low	Check for leaks, check solvent level, check for clogs
Peak is high	Peak is high	Check for leaks, check solvent level, check for clogs
Peak is flat	Peak is flat	Check for leaks, check solvent level, check for clogs
Peak is sharp	Peak is sharp	Check for leaks, check solvent level, check for clogs
Peak is blurry	Peak is blurry	Check for leaks, check solvent level, check for clogs
Peak is noisy	Peak is noisy	Check for leaks, check solvent level, check for clogs
Peak is clean	Peak is clean	Check for leaks, check solvent level, check for clogs

Peak resolution issues



Problem	Cause	Solution
Peaks are overlapping	Peaks are overlapping	Check for leaks, check solvent level, check for clogs
Peaks are separated	Peaks are separated	Check for leaks, check solvent level, check for clogs
Peaks are close	Peaks are close	Check for leaks, check solvent level, check for clogs
Peaks are far	Peaks are far	Check for leaks, check solvent level, check for clogs
Peaks are together	Peaks are together	Check for leaks, check solvent level, check for clogs
Peaks are apart	Peaks are apart	Check for leaks, check solvent level, check for clogs
Peaks are mixed	Peaks are mixed	Check for leaks, check solvent level, check for clogs
Peaks are clear	Peaks are clear	Check for leaks, check solvent level, check for clogs
Peaks are messy	Peaks are messy	Check for leaks, check solvent level, check for clogs
Peaks are neat	Peaks are neat	Check for leaks, check solvent level, check for clogs
Peaks are organized	Peaks are organized	Check for leaks, check solvent level, check for clogs
Peaks are chaotic	Peaks are chaotic	Check for leaks, check solvent level, check for clogs
Peaks are calm	Peaks are calm	Check for leaks, check solvent level, check for clogs
Peaks are wild	Peaks are wild	Check for leaks, check solvent level, check for clogs

Peak detection issues



Problem	Cause	Solution
Peak is not detected	Peak is not detected	Check for leaks, check solvent level, check for clogs
Peak is detected	Peak is detected	Check for leaks, check solvent level, check for clogs
Peak is missed	Peak is missed	Check for leaks, check solvent level, check for clogs
Peak is caught	Peak is caught	Check for leaks, check solvent level, check for clogs
Peak is ignored	Peak is ignored	Check for leaks, check solvent level, check for clogs
Peak is noticed	Peak is noticed	Check for leaks, check solvent level, check for clogs
Peak is overlooked	Peak is overlooked	Check for leaks, check solvent level, check for clogs
Peak is recognized	Peak is recognized	Check for leaks, check solvent level, check for clogs
Peak is forgotten	Peak is forgotten	Check for leaks, check solvent level, check for clogs
Peak is remembered	Peak is remembered	Check for leaks, check solvent level, check for clogs
Peak is unremembered	Peak is unremembered	Check for leaks, check solvent level, check for clogs
Peak is remembered	Peak is remembered	Check for leaks, check solvent level, check for clogs
Peak is unremembered	Peak is unremembered	Check for leaks, check solvent level, check for clogs

Peak quality issues



Peak quality issues



Peak quality issues



Peak quality issues



Peak quality issues



Problem	Cause	Solution
Peak is broad	Peak is broad	Check for leaks, check solvent level, check for clogs
Peak is narrow	Peak is narrow	Check for leaks, check solvent level, check for clogs
Peak is tall	Peak is tall	Check for leaks, check solvent level, check for clogs
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Ed Bowker Staff, Staff Bowker, Ed



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Flavoprotein Protocols Steven K. Chapman, Graeme A. Reid, 2008-02-03 As a scientist with an interest in proteins you will at some time in your career isolate an enzyme that turns out to be yellow or perhaps you already have. Alternatively you may identify a polypeptide sequence that is related to known flavin containing proteins. This may or may not be your first encounter with flavoproteins. However even if you are an old hand in the field you may not have exploited the full range of experimental approaches applicable to the study of flavoproteins. We hope that *Flavoprotein Protocols* will encourage you to do so. In this volume we have sought to bring together a range of experimental methods of value to researchers with an interest in flavoproteins whether or not these researchers have experience in this area. A broad range of techniques from the everyday to the more specialized is described by scientists who are experts in their fields and who have extensive practical experience with flavoproteins. The wide range of approaches from wet chemistry to dry computation has as a consequence demanded a range of formats. Where appropriate particularly for analytical methods the protocol described is laid out in easy to follow steps. In other cases e.g. the more advanced spectroscopies and computational methods it is far more apt to describe the general approach and relevance of the methods. We hope this wide ranging approach will sow the seeds of many future collaborations between laboratories and further our knowledge and understanding of how flavoproteins work. *Journal of Chromatography*, 2003

Handbook of Food Science, Technology, and Engineering - 4 Volume Set Y. H. Hui, Frank Sherkat, 2005-12-19 Advances in food science technology and engineering are occurring at such a rapid rate that obtaining current detailed information is challenging at best. While almost everyone engaged in these disciplines has accumulated a vast variety of data over time an organized comprehensive resource containing this data would be invaluable to have. *The Environmental Toxicology and Chemistry*, 2003

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High Performance Liquid Chromatography in Phytochemical Analysis Monika Waksmundzka-Hajnos, Joseph Sherma, 2010-11-08 The powerful efficient technique of high performance liquid chromatography HPLC is essential to the standardization of plant based drugs identification of plant material and creation of new herbal medicines. Filling the void in this critical area *High Performance Liquid Chromatography in Phytochemical Analysis* is the first book to give a complete overview. *Aviation Week & Space Technology*, 1999

XVIII Simpósio de Plantas Medicinais do Brasil, 2005 F&S Index United States Annual, 1998

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Forthcoming Books Rose Army, 1998-04 Getting Started in HPLC Lloyd R. Snyder, 1985

Troubleshooting HPLC Systems Lloyd R. Snyder, *Troubleshooting HPLC Systems* Paul C. Sadek, 2000 A complete up to date guide to the use maintenance and troubleshooting of HPLC systems. The last twenty five years have seen a dramatic rise in the use of High Performance Liquid Chromatography HPLC in laboratories worldwide. **Troubleshooting HPLC**

Systems provides analysts as well as laboratory technicians and managers with a readily accessible and immensely useful guide to the new generation of HPLC equipment and techniques With an emphasis on effective troubleshooting of HPLC systems this lab companion covers system configuration and functions problem solving procedures maintenance and HPLC basics It then walks chromatographers investigating the source of a malfunction through each system component from solvents and reservoirs to sample preparation to columns and detectors Special features of Troubleshooting HPLC Systems include A detailed review of HPLC instrumentation and accessories The role of operating parameters as indicators of system performance Step by step troubleshooting protocols for each system component How to set up a preventive maintenance program for HPLC systems An overview of the categories of HPLC separations A compilation of HPLC terms and definitions Tables and charts detailing solvents properties

HPLC Marvin C. McMaster,2006-12-18 This Second Edition of the classic handbook details how to set up an HPLC system that capitalizes on the latest innovations It covers new techniques in high temperature micro flow and ultra fast chromatography the linking of an HPLC to a mass spectrometer and more Complete appendices and supplementary material online this guide has everything chromatographers need to know to confidently separate identify purify and quantify compounds

Maintaining and Troubleshooting HPLC Systems Dennis J. Runser,1981-05-06 Provides users of HPLC equipment with a comprehensive text for troubleshooting and maintaining HPLC systems Describes how the chromatographer can maintain the HPLC system in operating condition what to look for and do to prevent and solve HPLC problems and what can and should be done before calling a service representative Organized into chapters which basically represent the typical components of the HPLC system with each chapter describing a basic element of the HPLC system in terms of maintenance and solving system problems Arranged as a guide and working manual to help the chromatographer reduce instrument downtime allowing for more efficiency and cost effectiveness in the HPLC laboratory

High-Temperature Liquid Chromatography Thorsten Teutenberg,2010-06-03 High temperature liquid chromatography has attracted much interest in recent years but has not yet recognized its full potential in the chromatographic community There is a widespread reluctance in industry to use temperature to speed up the separation process influence the selectivity of a separation or implement novel detection techniques However the technology has now matured and could revolutionize chromatography as we see it today Better equipment such as heating systems able to generate faster heating rates is becoming more readily available Also columns based on silica gel which can withstand higher temperatures for an extended period are now being introduced Nevertheless further technological and methodical efforts are needed to establish the method in a regulated environment like the pharmaceutical industry This is the only text to cover all the practical aspects as well as the underlying theoretical principles of setting up an HPLC system for high temperature operation It is not intended solely for academics but will also benefit the researcher interested in more practical considerations The author is a recognized expert and has conducted several studies with partners from industry to validate

the method Many real examples from these studies have been included in the book The aim is to support practitioners in the creation of their own protocols without the need to rely solely on trial and error The book starts with a brief definition of high temperature liquid chromatography before going on to cover system set up the heating system mobile phase considerations suitable stationary phases method development using temperature programming analyte stability and special hyphenation techniques using superheated water as a mobile phase In each chapter experimental data is used to illustrate the main statements and the advantages over conventional HPLC are evaluated The book concludes with a critical outlook on further developments and applications underlining the necessary advances needed to make high temperature HPLC more robust

Manual for HPLC Ramu BANDAMEEDI, Saibaba SV, Chittela BABU, 2018-02 This text is intended to be a guide for both the novice to preparative HPLC and as an aid to the chemical engineer planning to introduce this black art into the industrial environment The first question to ask is What is preparative To many the isolation of a few grams of an extremely potent molecule may be considered as largescale In some instances 50 g of a vaccine will supply the annual market for a particular disease state In more traditional drug therapies a few tonne may be more typical The second question to be answered is What is HPLC This abbreviation is often derived from the term High Performance Liquid Chromatography though the term High Pressure Liquid Chromatography is often preferred since high performance can also be achieved at low pressure Just to confuse the issue is this the pressure created by the resistance to liquid flow through the column or the pressure at which the column is packed

MAINTAINING AND TROUBLESHOOTING HPLC SYSTEM : A USER'S GUIDE. Dennis J. Runser, 1981

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could be a trigger so. Latest News / Checking In: - Lisa Merrill - MS, RD, CDE, LLC Asking for some prayers and positive healing vibes as he undergoes OPEN HEART SURGERY on OCT 10. Surgeon is replacing a valve and repairs to 2 others and some ... Abstinent Eating - Lisa Merrill - MS, RD, CDE, LLC Lisa Merrill - MS, RD, CDE, LLC. Registered Dietitian, Master of Science in ... Lisa Merrill - MS, RD, CDE, LLC. UB Associates.Design & Developed by VW Themes. Handouts - Lisa Merrill - MS, RD, CDE, LLC Lisa Merrill - MS, RD, CDE, LLC. Registered Dietitian, Master of Science in ... Lisa Merrill - MS, RD, CDE, LLC. UB Associates.Design & Developed by VW Themes. Sample Plans for Eating : r/OvereatersAnonymous I worked with a dietitian named Lisa Merrill who understands OA (Google her if you're interested) and she helped me develop a fairly expansive ... Lisa Merrill - Senior Researcher - American Institutes for ... President of the Americas at Unblu Inc. Boston, MA · Lisa M. VP of Business Development at Goldmine Leads, AI strategist. Tampa, FL. Tips for abstinent travel Read and write on program literature everyday to keep the program close. (If you have space in your luggage, prior to departure, have OA friends write you notes ... Lisa Merrill - Graduate Student Lisa Merrill. --Doctoral Candidate in Public Health, Epidemiology. Graduate, Online & Professional Studies at UMass Lowell ...