SHIMADZU
Excellence in Science

Application News

High Performance Liquid Chromatograph
Software for Efficient Method Development

Automatic Optimization of Gradient Conditions by AI Algorithm Using Integrated LC System

Shinichi Fujisaki

User Benefits

- The AI algorithm of LabSolutions™ MD can automatically optimize gradient conditions to greatly reduce labor of LC method development.
- Automatic optimization of gradient conditions can be applied not only to new method development, but also to existing method to efficiently improve resolution.

■ Introduction

In the typical LC method development, the process begins with "preparation" which includes mobile phase preparation, column installation, and creation of analysis schedules, then the analysis is started. After that, the acquired data is analyzed and "preparation" for the subsequent analysis is carried out, followed by starting the next analysis again. The method development progresses by repeating these processes, but in addition to the significant time required to repeatedly create analysis schedules, expertise in chromatography is necessary to explore optimal conditions based on data analysis. In other words, typical method development requires "human intervention". Therefore, eliminating human involvement and automating such method development processes would be desirable to improve labor efficiency. This article introduces an example of automatic optimization of gradient conditions that meet resolution criteria using LabSolutions MD (Technical Report C190-E309), a dedicated software for supporting method development. A mixture of seven small-molecule compounds was used as a model sample, and the optimization was performed by combining the AI algorithm in LabSolutions MD with the integrated LC system "i-Series" (Fig. 1).

Fig. 1 Integrated LC System "i-Series" (LC-2080C 3D)

■ Analytical Conditions and Target Compounds

The analytical conditions and target compounds are shown in Table 1. In this article, a mixture of seven small-molecule compounds was used as a model sample. Criteria were set for resolution and the elution time of the last peak, and LabSolutions MD automatically explored the gradient conditions to meet these criteria.

Table 1 Analytical Conditions and Target Compounds

System : LC-2080C 3D	
Sample : (1) Antipyrine, (2) Benzoic acid, (3) Salicylic acid, (4) Hydrocortisone, (5) Furosemide, (6) Naproxen, (7) Probenecid	
Mobile phase	
Pump A : 0.1% formic acid in water	
Pump B : Acetonitrile	
Column : Shim-pack Scepter™ C18-120 *1 (100 mm × 3.0 mmI.D., 1.9 µm)	
Analytical conditions	
B Conc.	: 20%(0 min)→X *2 %(3 min) →95%(3.01~4 min) →20%(4.01~8 min)
Column Temp.	: 40 °C
Flow rate	: 0.7 mL/min
Injection Vol.	: 5 µL
Detection	: 254 nm (STD cell)
Criteria of automatic optimization of gradient conditions	
Minimal resolution	: 3
Time of last eluting peak	: < 10 min

*1 P/N : 227-31013-03

*2 : X = 90, 91, 92, 93, 94 (5 patterns)

■ Automatic Optimization of Gradient Conditions

Fig. 2 shows the workflow of automatic optimization of gradient conditions using LabSolutions MD. This software has a unique AI algorithm to automatically explore gradient conditions that satisfy resolution criteria by alternately repeating "improvement of gradient conditions by AI (condition search)" and "analysis under improved conditions (correction analysis). For the criteria, "resolution" and "elution time of the last peak" can be set. In this article, automatic optimization of gradient conditions was applied to a mixture of seven small-molecule compounds to meet the criteria of resolution (> 3.0) and maximum elution time of 10 minutes for the last peak (Fig. 3), considering the reduction of analysis time.

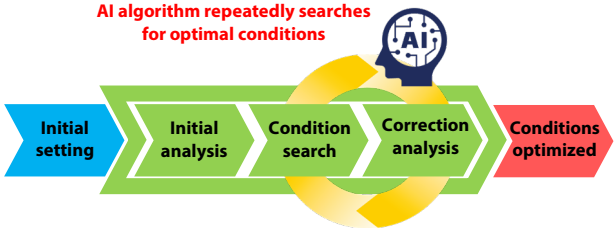


Fig. 2 Workflow for Automatic Optimization of Gradient Conditions by LabSolutions MD

Fig. 3 Criteria Setting for Automatic Optimization

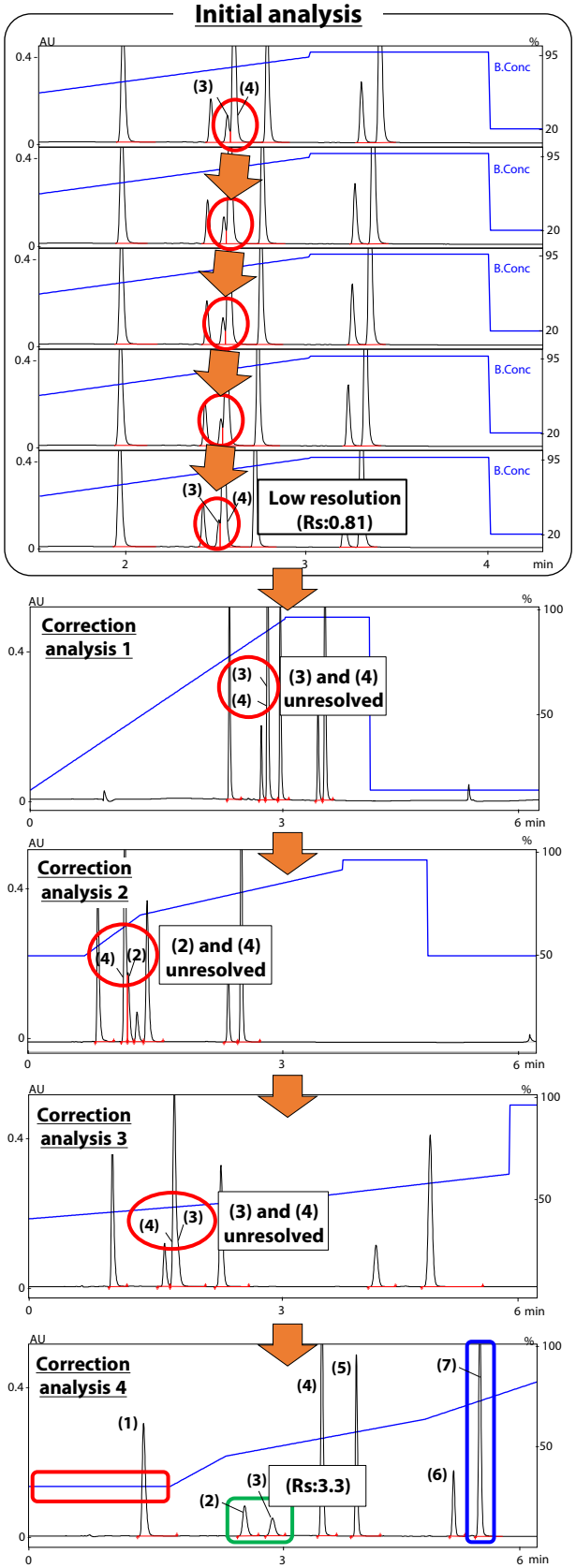


Fig. 4 Result of Automatic Optimization of Gradient Conditions
* blue line shows gradient conditions
* Peak numbers correspond to those listed in Table 1

The result of automatic optimization of gradient conditions is shown in Fig. 4. After the initial analyses were performed with five different gradient patterns (B Conc. in Table 1), the separation of seven small-molecule compounds was automatically optimized through four correction analyses. The result of the initial analyses showed that peaks (3) and (4) were not sufficiently resolved (red circles in the initial analyses in Fig. 4). Through repeated correction analyses by the AI algorithm, the gradient conditions that met the criteria were automatically explored (correction analysis 4). In correction analysis 4, the resolution criteria (> 3.0) was achieved (green box) by introducing an isocratic elution (red box). In addition, the gradient slope was automatically adjusted, and the retention time of the last peak (blue box) met the criteria of being under 10 minutes. This demonstrates that the AI algorithm enables users, regardless of their level of chromatography experience, to easily explore gradient conditions that satisfy the defined criteria.

■ Conclusion

Automatic optimization of gradient conditions using the AI algorithm in LabSolutions MD was applied to a mixture of seven small-molecule compounds in combination with i-Series. As a result, gradient conditions that met the criteria (minimum resolution > 3.0 and elution time of the last peak < 10 minutes) were successfully explored. In method development, optimizing gradient conditions typically requires human intervention at various steps, such as creating analysis schedules and interpreting data. However, the use of an AI algorithm significantly reduces manual effort and enables automation of the entire workflow.

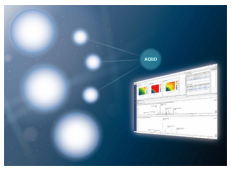
01-00933A-EN
First Edition: Sep. 2025
Revision A: Oct. 2025

SHIMADZU
Shimadzu Corporation
www.shimadzu.com/an/

› Please fill out the survey

Related Products Some products may be updated to newer models.

› i-Series
High Performance Liquid Chromatograph



› Method Development System
Automatic Optimization of Gradient Conditions with···

Related Solutions

› Pharmaceutical and Biopharmaceutical

› Small Molecule Pharmaceutical

› Price Inquiry

› Product Inquiry

› Technical Service / Support Inquiry

› Other Inquiry