



Analytical and Semi-preparative Chromatography of Aromatic Compounds on a Single HPLC System

Application Note ENV0214

Introduction

A simple mixture of 4 compounds was used to evaluate the capacity of the Gilson **PLC 2050** to scale up a separation from analytical to semi-preparative HPLC. The analytical method was first developed on a LaChrom Elite™ analytical HPLC system and then transferred to the Gilson **PLC 2050** (Figure 1). The method is then scaled up 50-fold using the **PLC 2050** system to allow for higher throughput separations on a semi preparative HPLC column (25 mm I.D.).



Figure 1. PLC 2050 System from Gilson

Materials & Methods

LaChrom Elite (Hitachi) is equipped with a 10 mL/min quaternary gradient pumping system, Photodiode Array Detector (200-800 nm), and EZChrom Elite™ Chromatography Data Software.

PLC 2050 (Gilson) is equipped with a 50 ml/min quaternary gradient pumping system, UV/Vis detector (200-840nm), and Gilson Glider Prep Software.

Analytical Method is the same for all tests (see Table 1).

Column	Analytical: Merck Purospher® RP18e (250 mm x 4.6 mm I.D. 5µm with pre column) Preparative: Merck LiChrospher® 100 RP-18 (250mm x 25 mm I.D. 5µm)		
Sample	Mixture of hydroquinone, catechol and phenol at 0.5g/L and piperine at 0.01g/L.	Detection	UV/Vis detector at 280nm
Eluent A	phosphate buffer (0.02M, pH = 5.5 adjusted with NaOH 6N)	Eluent B	Methanol
Method	20% B	3 min	
	20% to 90% B	7 min	
	90% B	20 min	

Table 1. Experimental conditions used to generate the analytical and preparative chromatograms.



Results and Discussion

In this test, the profile of analytical separation (20 μ L injection) on the **PLC 2050** (Figure 2B) was similar to the chromatogram obtained using the LaChrom *Elite*TM HPLC system (Figure 2A). Furthermore, the use of a preparative column (1 ml injection) with the **PLC 2050** (Figure 2C) demonstrates clear separation of the compounds.

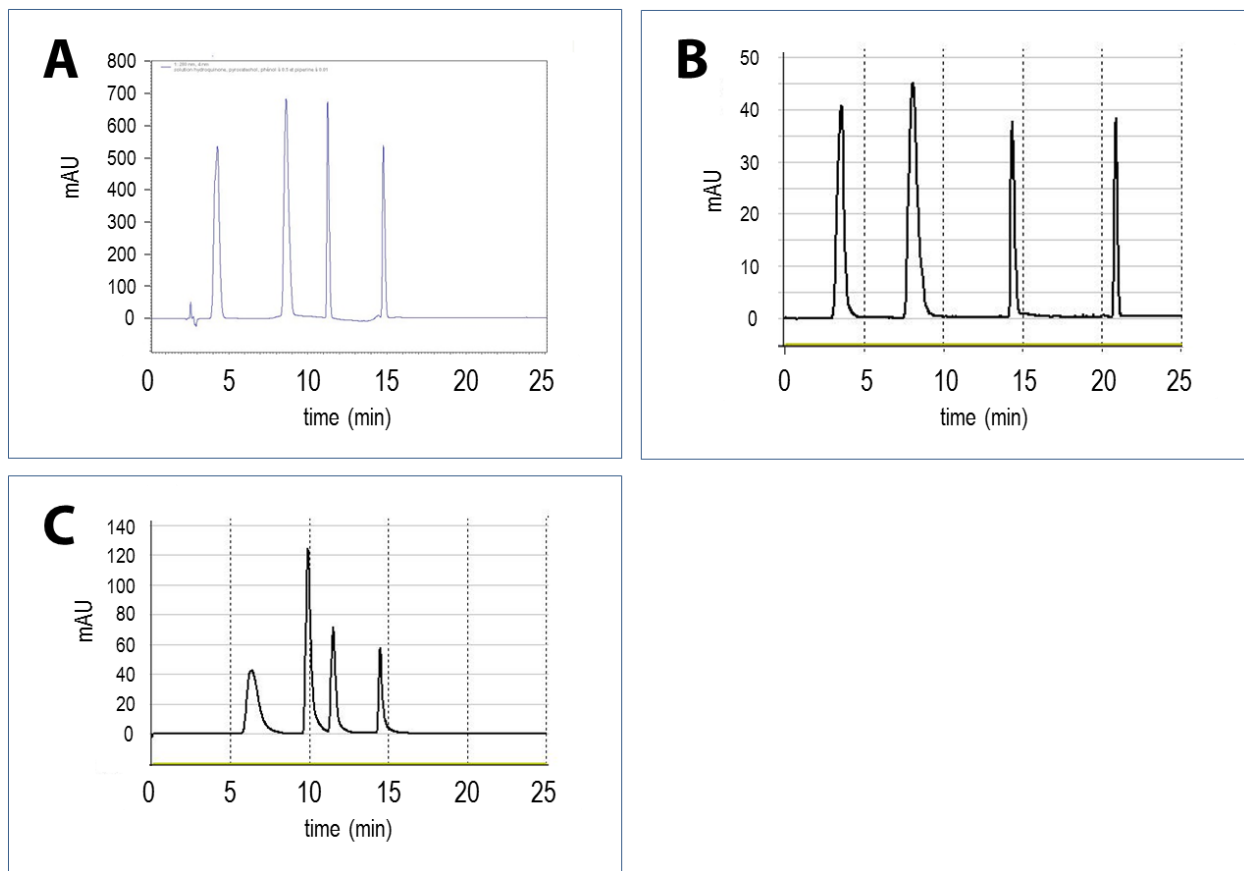


Figure 2. HPLC Chromatogram at 280 nm. Hydroquinone, pyrocatechol, phenol, and piperine were separated using A) an analytical column with a LaChrom *Elite*TM (Hitachi); B) a an analytical column with a Gilson PLC 2050 personal chromatography system; and C) a preparative column with a Gilson PLC 2050.

Summary

- The Gilson **PLC 2050** system allows for reproducible analytical injections on 4.6 mm I.D. columns with high efficiency.
- This application note also demonstrates scale up to a 25 mm I.D. column.
- It is also possible to use the Gilson PLC products to perform similar scale-up with preparative columns, up to 100 mm I.D. (results not shown).