

Waters® 2700 Sample Manager Fraction Collection With A Difference

The Waters 2700 Sample Manager has powerful multiple function capabilities that are easy to setup and use. With the appropriate options installed, these functions allow users to inject concentrated extracts for semi-preparative separation, collect fractions from these injections, pool like fractions together, and inject those fractions for analytical analysis. The 2700 Sample Manager also has the ability to pre-treat these fractions before injection (serial dilute, add internal standards or derivitizing reagents. See Performance PerSPECTive WPP43).

In the following example, 6 injections of regular brewed coffee (concentrated ~ 5X) were subjected to a semi-preparative separation. Fractions (7 seconds each) were collected over several time intervals (between 1 and 4 minutes) in a 96•deep well plate to isolate the caffeine peak. The fractions from each semi-prep injection were pooled together and all 27 collected fractions were analyzed for caffeine using analytical conditions.

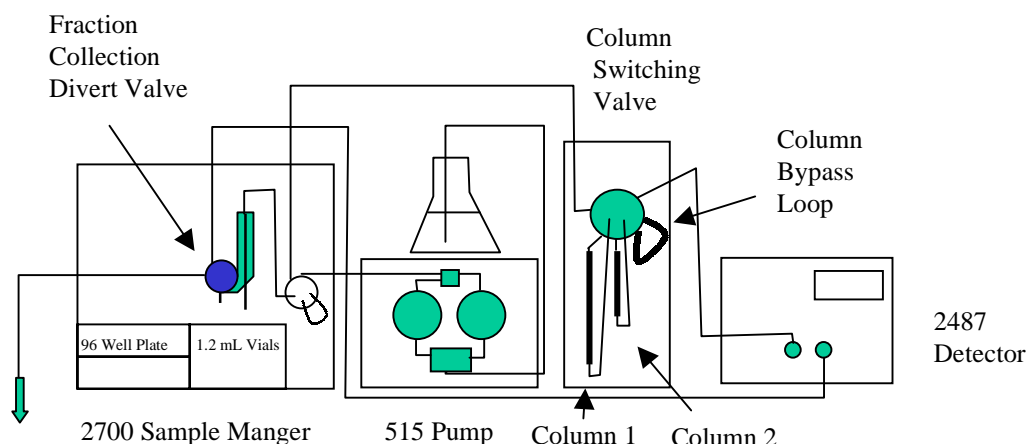


Figure 1 - Instrument Configuration

Conditions

Waters 2700 Sample Manager
 Waters 515 HPLC Pump
 Waters 2487 Detector: 271 nm
 Eluent: 35% Methanol, 65% Water
 Flow: 1 mL/minute
 Sample: Regular Brewed Coffee Concentrated ~5X
 Semi-Prep Column (#1): Symmetry® C₁₈, 4.6 x 150 mm (ambient temperature)
 Analytical Column (#2): Symmetry® C₁₈, 4.6 x 75 mm (ambient temperature)

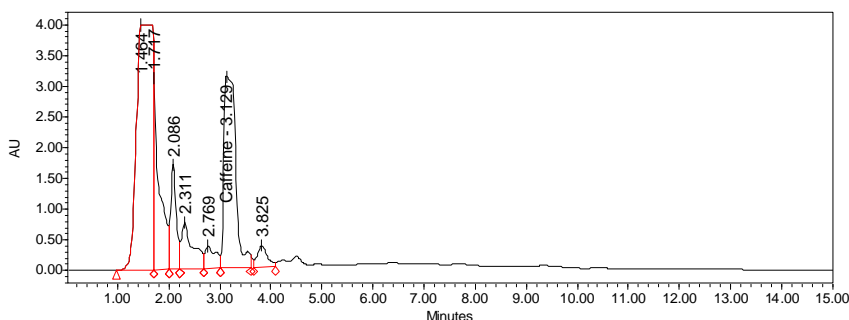


Figure 2 - Typical Semi-Preparative Chromatogram

The 2700 Sample Manager integrates injection and fraction collection in one instrument. It provides a seamless transition from semi-preparative separations and fraction collection to analytical injections.

Programming this type of multiple function analysis is quick and easy to do.

Sample Group	# of Inj's	μL Sample	μL Rinse	Millen. Method Set	Runtime, min	HPLC Column	Pre-treatment	FC
1 COFFEE_CON(6)	20	450	Semi_Prep_Coffee	15	Semi Prep		Yes	
2 WASH	2	20	450	Analytical_Caff	1	Bypass		
3 COFFEE_FRAC1	20	450	Analytical_Caff	5	Analytical			
4								
5								
6								
7								
8								
9								
10								
11								
12								

Start [▶] Pause [||] Stop [■] Position: Operational Status: Idle Samp.#: / Inj.#: 4

Figure 3 - Run Window

Zone	Name	Start (min)	Ret. (min)	End (min)	Slice (sec)	# Tubes
1		0.983		1.900	7	8
2		1.900		2.000	7	1
3		2.000		2.217	7	2
4		2.217		2.400	7	2
5		2.400		3.017	7	6
6	Caffeine	3.017		3.483	7	4
7		3.667		4.083	7	4
8					0	0
9					0	0
10					0	0
11					0	0
12					0	0

Total: 27
Collection Group: COFFEE_FRACS
Num Tubes: 32
Min Vol (μL): 2000
Pump Flow (mL/min): 1
Detector Based: ☐
Distribute Proportionately: ☐
Inquire Millennium [Inquire Millennium] Cancel [Cancel] OK [OK]

Figure 4 - FC Setup Window

Line 1 of the run window (Figure 3) automatically directs the 2700 Sample Manager to perform 6 semi-preparative injections with fraction collection and pool them together. Selecting fraction collection (FC = Yes), opens the fraction collection setup window (Figure 4). In this example, multiple time windows were collected. These time windows were imported directly from a Millennium^{®32} software report using the Inquire Millennium button in the fraction collection setup window. Individual imported collection zones can be selected and the start/end times can be edited. The 2700 Sample Manager can collect up to 12 time windows and over 2000 individual fractions (using high density 384• well plates).

Line 2 of the run window switches the column selection valve to a bypass loop and performs 2 wash injections to lower carryover. The 2700 Sample Manager has very low carryover characteristics (See Performance PerSPECTive WPP44), however, when dealing with concentrated semi-preparative extracts, this short wash step ensures minimum cross contamination when performing the analytical analysis.

Line 3 of the run window switches the column selection valve to the analytical column and injects all of the collected fractions.

In this example, data was collected and analyzed in Millennium³² software. Sample chromatograms are shown in Figure 2 (semi-prep) and Figure 5 (analytical).

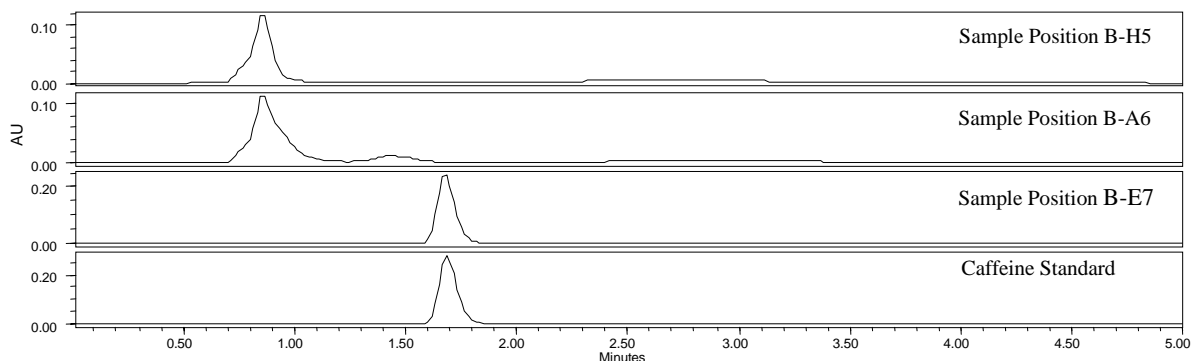


Figure 5 - Typical Analytical Chromatograms

Options

Diverter Valve Assembly - Part Number: WAT272006

Column Switching Valve, 2 Columns - Part Number: WAT050045

Column Switching Valve, 6 Columns - Part Number: WATPR5001-104-03