

# Reduced Solvent Consumption Mode Allows Continuous Nano LC Operation for One Month

## **INTRODUCTION**

In an effort to reduce operational cost and minimize waste production during nano LC analysis, Dionex has evaluated ways to decrease solvent consumption, using the UltiMate® 3000 nano LC system. This technical note describes the use of the UltiMate 3000's UltiFlow™ eluent delivery system in a reduced solvent consumption mode. The lower solvent consumption allows continuous operation for one month without solvent refill.

## **METHOD**

To operate the system in reduced solvent consumption mode, refer to the Materials table for the parts used. The standard nano (1:1000) splitter cartridge is replaced by a capillary (1:100) splitter cartridge. To ensure that the same flow is delivered on the column, the pump flow is automatically decreased by a factor of 10, which reduces solvent consumption. The extra delay time resulting from the reduced pump flow is partially ameliorated by replacing the pump outlet tubing with 130 µm i.d. PEEK™ tubing.

No changes in the instrumental control are required as the chip card on the splitter cartridge contains all necessary information. The user is concerned only with the desired column flow rate and applied gradient, and these are identical between normal and reduced solvent consumption mode.

The delay time is increased in reduced solvent consumption mode, requiring a slightly increased equilibration time (10–15 min) at the end of the run.

## RESULTS

Figure 1 shows an overlay of five consecutive cytochrome C digest injections; Table 1 shows the average retention time and %RSD for all observed peaks. Minor delays or dead volumes in a setup will cause peak 7b to elute as a shoulder of peak 7a. The baseline separation of these peaks (Figure 1 inlay) and retention time reproducibility (Table 1) show that the system performance is not affected by the reduced solvent consumption mode.

Longer gradients (120 min) and complex mixtures are more common in peptide mapping. With an increase in run time, the relative effect of longer equilibration time decreases. Figure 2 shows the excellent reproducibility of 10 consecutive separations of a tryptic digest of six proteins (PMD).

## SUMMARY

The new reduced solvent consumption mode of the UltiMate 3000 nano LC system provides high quality chromatography, reduced labor and solvent costs, and offers the following benefits:

- Up to a 10-fold reduction in solvent consumption can be achieved. The reduction depends on the splitter cartridge and the gradient length used.
- There is no visible decrease in performance when operating in reduced solvent consumption mode, only an increased delay time, and thus total run time.
- Up to one month of continuous operation is possible without solvent refill.

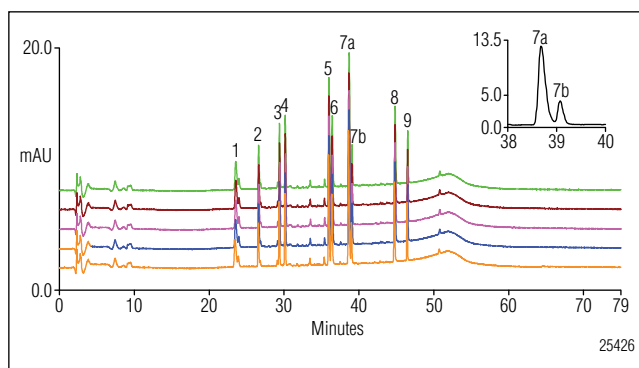


Figure 1. Overlay of five consecutive cytochrome C digest injections separated with the reduced solvent consumption mode.

**Table 1: Average Retention Times and RSD for All Detected Peaks from Figure 1**

Peak	1	2	3	4	5	6	7a	7b	8	9
$t_r$ (min)	23.5	23.6	29.4	30.1	36.0	36.4	38.6	39.0	44.8	46.5
RSD	0.17	0.12	0.08	0.08	0.07	0.09	0.09	0.09	0.10	0.08

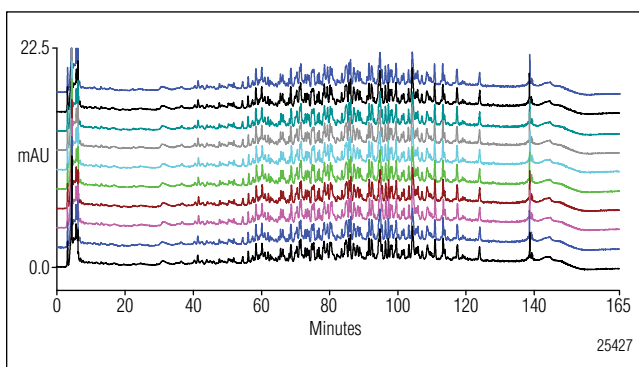


Figure 2. Overlay of 10 consecutive tryptic digest (PMD) separations.

## MATERIALS (SEE METHODS)

Description	Part Number
Capillary Pump to splitter, 130 $\mu$ m ID x 75 cm, PEEK, fittings .....	6720.0032
Biocompatible, Capillary Pump to splitter, 130 $\mu$ m ID x 75 cm PEEK, fittings .....	6721.0032
Splitter cartridge 1:100.....	6720.3160
Splitter cartridge 1:300.....	6720.3155
Splitter cartridge 1:1000.....	6720.3150
Biocompatible Splitter cartridge 1:100.....	6721.3160
Biocompatible Splitter cartridge 1:300.....	6721.3155
Biocompatible Splitter cartridge 1:1000.....	6721.3150

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### Dionex Corporation

1228 Titan Way  
P.O. Box 3603  
Sunnyvale, CA  
94088-3603  
(408) 737-0700

### North America

U.S. (847) 295-7500  
Canada (905) 844-9650

### South America

Brazil (55) 11 3731 5140

### Europe

Austria (43) 1 616 51 25 Benelux (31) 20 683 9768 (32) 3 353 4294  
Denmark (45) 36 36 90 90 France (33) 1 39 30 01 10 Germany (49) 6126 991 0  
Ireland (353) 1 644 0064 Italy (39) 02 51 62 1267 Sweden (46) 8 473 3380  
Switzerland (41) 62 205 9966 United Kingdom (44) 1276 691722

### Asia Pacific

Australia (61) 2 9420 5233 China (852) 2428 3282 India (91) 22 2764 2735  
Japan (81) 6 6885 1213 Korea (82) 2 2653 2580 Singapore (65) 6289 1190  
Taiwan (886) 2 8751 6655

www.dionex.com



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