Performance PerSPECtives

Waters® Column Heaters

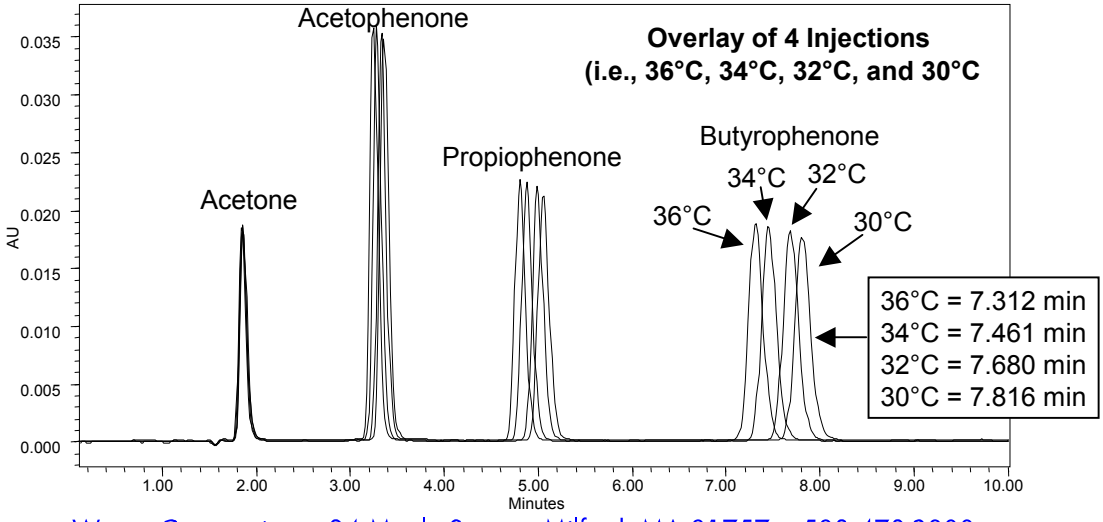
Benefits of Separation Temperature Control

Is Retention Time Variations Between Injections a Concern?
Several factors effect the ability to obtain reproducible HPLC separations. HPLC pump performance, sample and solvent preparation, and column performance are a few of the variables that can compromise the quality of collected data. Control and maintenance of column temperature throughout a series of analyses are an important, yet frequently overlooked, parameter that can affect retention time reproducibility. This Performance PerSPECtive shows how minor changes in column temperature (e.g., ± 2°C) significantly influence results obtained in a carefully controlled series of experiments.

Experimental Design and Results:
Table 1 and Figure 1 indicate the conditions used and results obtained when a reversed-phase, HPLC separation was performed at a series of precisely controlled temperatures. Significant shifts in retention times resulted when separation temperatures changed. **Do temperatures in your laboratory vary throughout the day? Can these temperature variations compromise the quality of results obtained?**

Table 1 and Figure 1: Effect of Temperature Variation on HPLC Separation

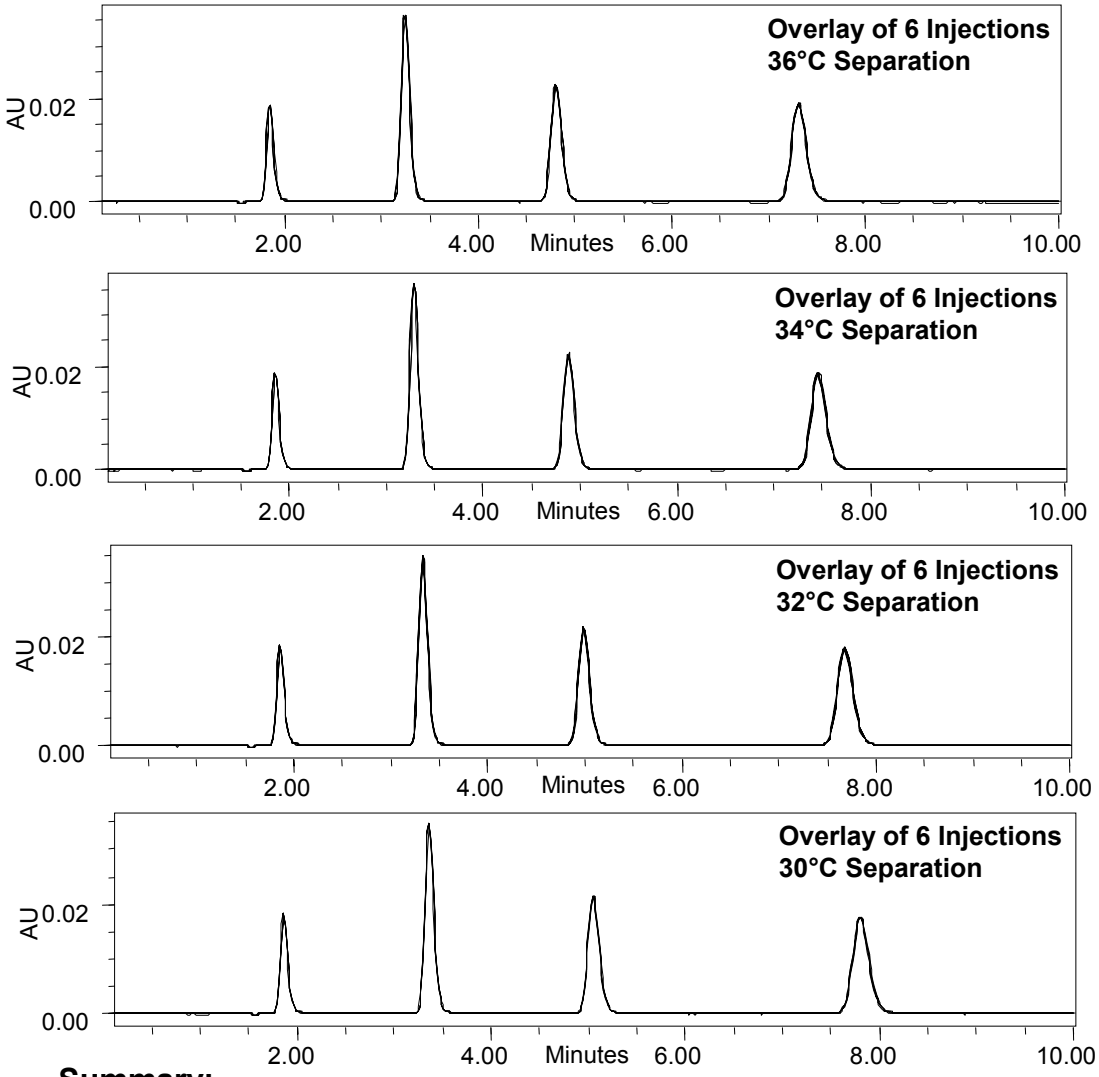
HPLC:	Waters Alliance® HPLC System with Column Heater
Detection:	Waters 2487 Detector at 254 nm at 5 pts/sec
Software:	Waters Millennium®32
Column:	Symmetry® C_{18}, 5 µm, 4.6 x 75 mm
Eluent A:	Water
Eluent B:	Methanol
Flow:	1.0 ml/min at 40% A / 60% B
Sample:	10 µL acetone (3.18 mg/mL), acetophenone (0.01 mg/mL), propiophenone (0.01 mg/mL), and butyrophenone (0.01 mg/mL)
Temp:	36°C, 34°C, 32°C, and 30°C



Waters Corporation 34 Maple Street Milford, MA 01757 508 478 2000

Column Temperature Control Improves Separation Reproducibility:
Controlling temperature variations within a laboratory can be difficult. Rather than controlling the laboratory environment, column temperature can be precisely controlled using an external column heater device. Compared to results obtained in Figure 1, Figure 2 shows how retention time reproducibility is significantly improved by precise column temperature control. (See Table 1).

Figure 2: Retention Time Reproducibility using Column Heater Device



Summary:

- Temperature variations in a laboratory can adversely affect the quality of HPLC collected data (e.g., retention time reproducibility, separation selectivity, peak shape, etc.).
- Waters offers a variety of column heater devices that effectively control separation temperatures using the Alliance System, Breeze™ System, or other solvent delivery technologies.
- Utilization of a Waters HPLC column heater device can significantly improve the quality of results obtained in laboratories where temperature control and maintenance are difficult.

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