

Thermo Scientific Vanquish UHPLC System

Best in UHPLC Column Thermostatting to Fit All Needs

Product Spotlight

The required technique to control the actual separation temperature is often underestimated and, in many cases, not well understood in UHPLC. Thermostatting has to be effective, but must not compromise the achievable column efficiency. On the other hand, it is very important to account for successful method transfer between different instruments. The Thermo Scientific™ Vanquish™ UHPLC system provides an entirely new and unique combination of active eluent pre-heating and advanced column thermostatting modes.

Combining Active Eluent Pre-Heating and Advanced Column Thermostatting

The active and real-time eluent pre-heater with a geometrical volume of 1 μL

- Negligible extra-column volume contribution even with 1 mm ID columns
- Highly effective and accurate control of the eluent temperature (up to 120 °C)
- Incoming eluent temperature setting independent of compartment temperature

The still air column thermostatting mode

- Minimum thermal mismatch by viscous heating under UHPLC conditions
- Best possible UHPLC efficiency and resolution

The forced air column thermostatting mode

- Fast thermal equilibration
- Best possible removal of frictional heat for reliable analyte retention
- Highest flexibility, e.g. for method transfer from other instruments



Eliminating Thermal Mismatch

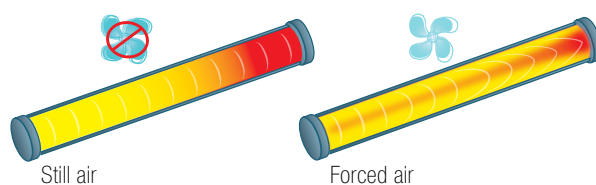
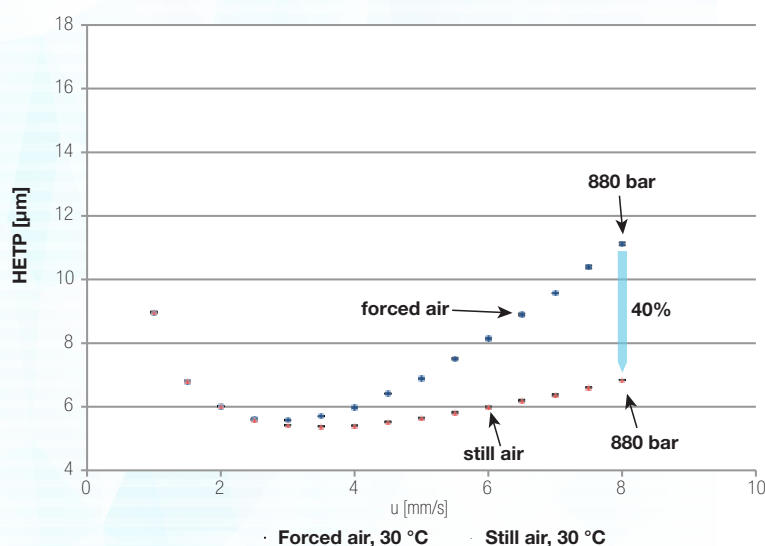
It is a matter of fact that small particle stationary phases generate significant heat by viscous friction of the mobile phase, especially under column backpressures at elevated linear velocities. A column thermostat that enables efficient removal of frictional heat avoids pronounced temperature increase inside the column due to this viscous friction. As the generated heat dissipates from the column center to the column wall, this heat removal induces a radial temperature gradient inside the column. In such a case, the column center is hotter than the column wall. Due to the cooperative effect of temperature induced lower retention and, secondarily, viscosity induced higher linear velocity, the analyte molecules move much faster in the column center than closer to the column wall where temperatures are lower. This causes severe peak distortion also known as thermal mismatch. If the exchange of heat between the interior of the column and the column compartment is suppressed, the frictional heat will stay inside the column. Hence, the overall temperature increases axially from column inlet to column outlet. At the same time, radial temperature gradients are eliminated and peak distortion through thermal mismatch is suppressed.

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Thermal Mismatch Under Viscous Heating Conditions

A Van Deemter curve provides information on the linear velocity for best separation efficiency. It also predicts the amount of efficiency loss when the analysis is accelerated by going beyond the optimum linear velocity (u_{opt}). High efficient, small particle stationary phases in UHPLC account for fast analyte diffusion into the stationary phase and back. Little efficiency loss occurs, even at very high linear velocities, for superfast analysis. However, any adverse effect from increased viscous heating at elevated linear velocities is not considered in this statement.



Conditions:

Column: Thermo Scientific™ Hypersil GOLD™, 1.9 µm, 2.1 x 100 mm
Mobile phase: Isocratic mode, 50% acetonitrile in water
Flow rate from 0.07 to 1.04 mL/min
Injection volume: 2.7 µL
Sample: Hexanophenone in mobile phase (5µg/mL)
Column and eluent pre-heater temperature 30°C
Column thermostating mode: Still air or forced air
UV-detection: 240 nm, 100 Hz, 0.05 s RT, slit and bandwidth 4 nm

Figure 1: Van Deemter curves of hexanophenone with either forced air or still air conditions.

The slope of a Van Deemter curve at elevated linear velocity (C-term) indicates efficiency drop that can have a significant contribution from thermal mismatch due to viscous heating. Solely the Vanquish system provides two complementary thermostating modes – still air and forced air – which elucidate this effect. Figure 1 shows Van Deemter curves of hexanophenone recorded with increasing viscous heating effect by accelerating the linear velocity. At the most elevated analysis speed of 8 mm/s, the slope in the C-term region is more than three times higher with forced air mode, and the efficiency is by 40% lower. This pronounced effect of efficiency loss is caused by the viscous heat removal through forced air. Imagine that effect at 1500 bar instead of the 900 bar backpressure observed during this experiment. The still air mode is advantageous if best achievable efficiency at very high analysis speed is wanted. Moreover, the still air mode accounts for a 10% better efficiency and 20% faster analysis speed under optimum conditions at a linear velocity of 3 mm/s.

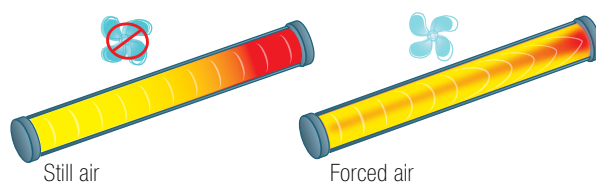
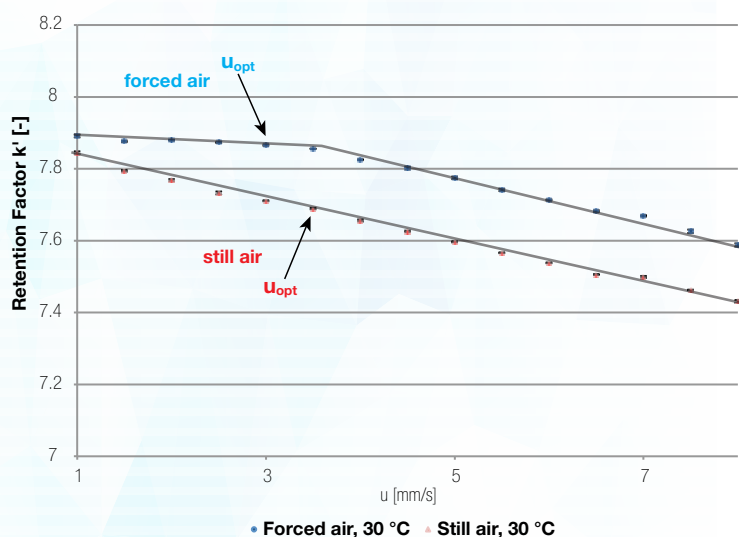
When viscous heating is expected, one might suggest that the ideal method of column thermostating is the still air mode. But is it always true?

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Retention Control Under Viscous Heating Conditions

From the same experiment, we plotted data that are normally not taken into account in Van Deemter experiments. Figure 2 shows the retention factor of hexanophenone with increasing viscous heating effect by accelerating the linear velocity for the two different thermostating modes of the Vanquish UHPLC.



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Mobile phase: Isocratic mode, 50% acetonitrile in water
Flow rate from 0.07 to 1.04 mL/min
Injection volume: 2.7 μL
Sample: Hexanophenone in mobile phase (5 $\mu\text{g/mL}$)
Column and eluent pre-heater temperature 30 $^{\circ}\text{C}$
Column thermostating mode: Still air or forced air
UV-detection: 240 nm, 100 Hz, 0.05 s RT, slit and bandwidth 4 nm

Figure 2: Retention factors of hexanophenone with either forced air or still air conditions.

These results show another indicative difference between the two thermostating modes provided by the Vanquish UHPLC. With still air, viscous heat is not removed. The retention factor does not stay constant throughout the experiments, but decreases with linear velocity even at the low flow rate range. In order to determine the linear velocity for best efficiency, the user has to accept that the retention factor cannot be kept constant. Using the forced air mode, however, the retention remains constant up to a linear velocity slightly higher than the optimum.

Retention factor changes can be very critical for certain applications as selectivity and resolution are dependent on analyte retention. For method transfer from HPLC to UHPLC, between two UHPLC systems with a different column oven, or when method speed-up is applied especially in UHPLC. Chromatographic results may change when the frictional heat contribution is not addressed adequately.

The forced air mode is advantageous if best achievable retention factor reproducibility between HPLC and UHPLC is the goal. This can be very useful as long as the efficiency loss due to frictional heat does not compromise peak resolution.

The Vanquish UHPLC system column thermostat supports the choice between retention control or efficiency optimization, providing a solution for all requirements.

[Learn more at thermoscientific.com/vanquish](https://thermoscientific.com/vanquish)