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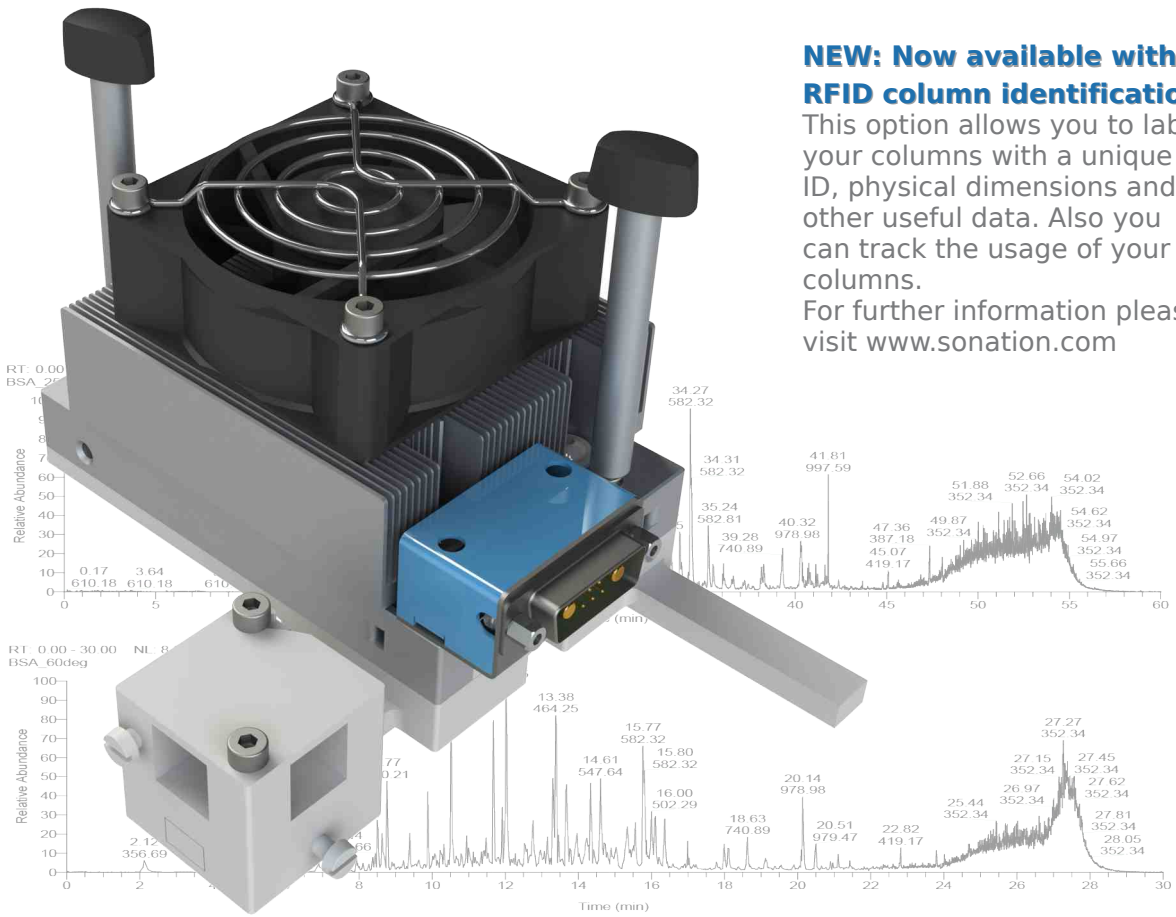
Column oven for Nano-ESI

Sonation column oven PRSO-V1

Developed by scientists of the Max-Planck-Institute of Biochemistry in Munich-Martinsried, the column oven regulates the temperature of the column in Nano-ESI applications.
Due to a constant column temperature significantly better results can be achieved and with higher temperatures faster wash out cycles and longer columns can be realized.



NEW: Now available with RFID column identification
This option allows you to label your columns with a unique ID, physical dimensions and other useful data. Also you can track the usage of your columns.
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- **Longer columns can be used with standard nano HPLC's**
- **Columns can be packed with smaller beads for better separation**
- **Sample loading times can be shortened**
- **Increase efficiency of the column wash out by using higher temperature and higher flowrates during the wash, controlled by the Thermo Xcalibur software**
- **Better reproducibility of chromatograms independent of room temperature and fluctuations from air conditioning systems**

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The Sonation column oven allows for shortening or lengthening of gradients without loss of sensitivity

Dr. Richard Scheltema (MPI of biochemistry Martinsried, Germany)

With high per analysis costs on liquid chromatography / mass spectrometry platforms (average write-off of €300 a day for a typical LC/MS-MS platform[1] and the incredible sequencing speed of the current generation of mass spectrometers, it can be beneficial to do the same LC/MS-MS experiment in half the time. The Sonation column oven offers a solution to stabilize the temperature around the analytical column, improving the reproducibility of experiments by reducing time shifts of analytes due to temperature and/or pressure fluctuations. An additional benefit of the column oven is that by increasing the temperature on the analytical column the back pressure on the LC is lowered allowing for use of higher flow-rates. Here we demonstrate a reduction of 50% of the analysis time on a tryptic digest of Bovine Serum Albumin (BSA) protein by increasing the temperature from 25°C (based on ambient temperature around the mass spectrometer) to 60°C and the flow-rate from 250 nl/min to 450 nl/min, while still maintaining a median pressure of 460 bar.

In Figure 1 the difference in flow-rate at the same pressure level is displayed for the two runs (standard run at 60 min; reduced run at 30 min). The increase of

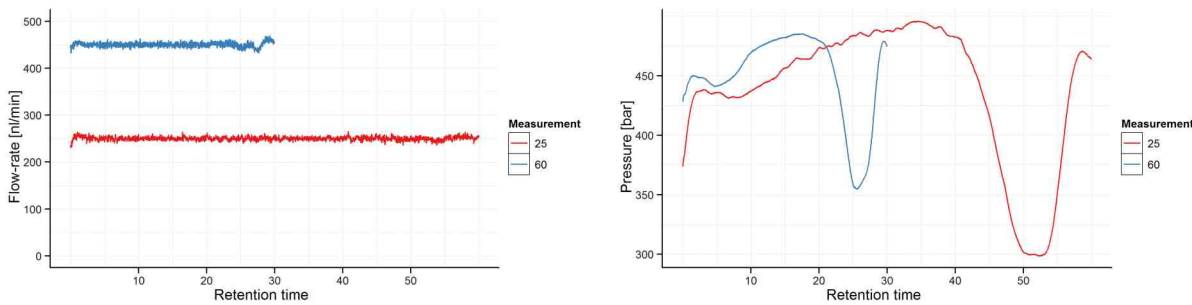

Figure 1: Flow-rate can be increased almost two-fold with an increase in temperature from 25°C to 60°C while keeping the pump pressure relatively stable.

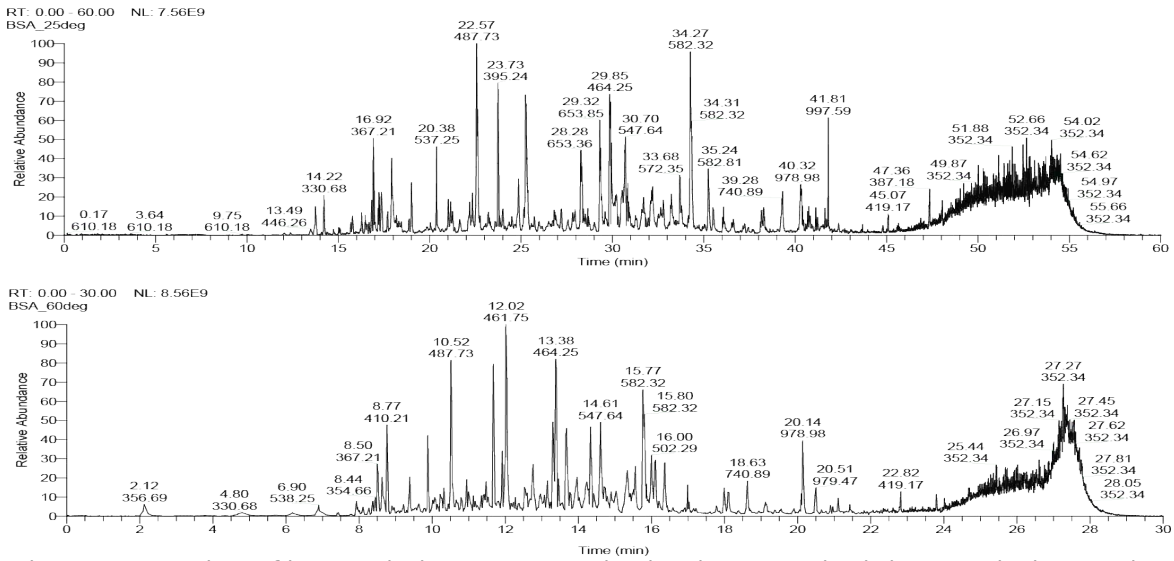

Figure 2: Comparison of base peak chromatograms, showing that separation is intact at the increased analysis speed.

35°C is enough to reduce the back-pressure to the same level for almost double the flow-rate. A basepeak chromatogram of the runs is displayed in Figure 2, from which it is clear that the separation between the peaks is still maintained, even though the sample is almost two-fold more diluted for the higher flow-rate, without loss of intensity for the base peaks. The required increase in intensity is achieved by the two-fold decrease in analysis time, which drives the percent mobile phase more quickly to a higher level. Additional sharpening is achieved by the elevated temperature on the analytical column. The combined effect is shown

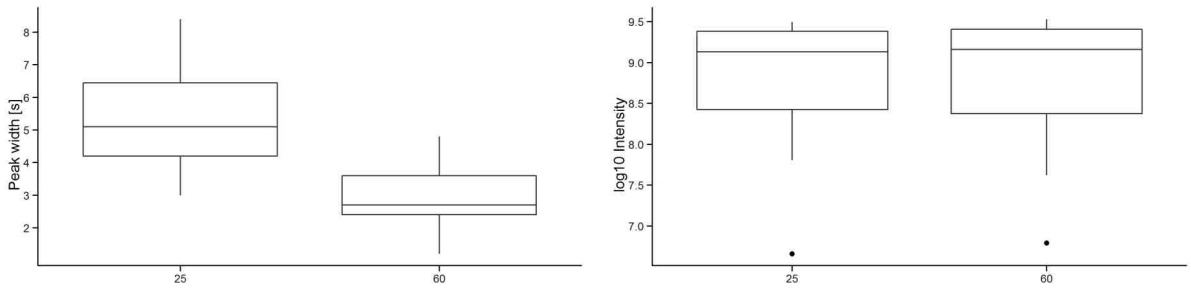

Figure 3: Reduced peak widths at a stable intensity per peak of a selection of tryptic BSA peptides.

for a selection of tryptic BSA peptides in Figure 3. The peak width at half height is reduced by almost 50%, driving the intensity for the peaks up to the same level in the higher dilution of the increased flow-rate. Note that the peak-width of the BSA peptides has been reduced to 3s, requiring the high scan speed of the modern mass spectrometry equipment.

Material and methods

Sample preparation

BSA standard (Sigma-Aldrich) was solubilized in a mixture of 6M urea and 2M thio-urea, reduced with DTT and alkylated with IAA. The proteins were digested with LysC for 3 hr and Trypsin for 12 hr at room temperature. A total of 5 pmol of the final peptide mixture was loaded on a stage tip and desalted.

LC-MS/MS

An Easy-nLC nano-flow ultra-high performance liquid chromatograph (Thermo Fisher Scientific) was coupled online to a Q Exactive (Thermo Fisher Scientific). Prior to analysis 0.5 pmol of the peptide mixture was introduced on a 20 cm fused silica analytical column (75 µm ID, spray nozzle about 5 µm ID) packed with C18 material (Reprosil-Pur C18-AQ, 3 µm particle size, Dr. Maisch GmbH).

Reference

1. SprayQc: A Real-Time LC-MS/MS Quality Monitoring System To Maximize Uptime Using Off the Shelf Components; Scheltema RA, Mann M.; J Proteome Res. 2012

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