

EVOSEP ONE

*A Separation Tool Designed
for Clinical Omics*

EVOSEP





Evosep aims to improve quality of life and patient care by radically innovating protein based clinical diagnostics.

Clinical Proteomics must be Sensitive, Fast, and Robust

- More uptime with improved reliability and robustness
- Increased productivity with higher throughput and better duty cycle utilization
- Increased performance with better data quality



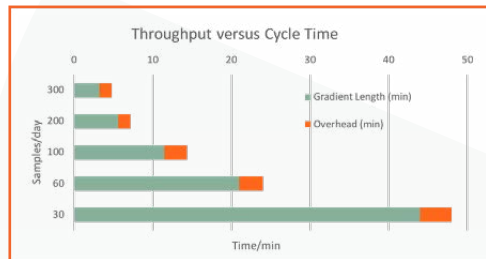
"Robust and fast workflows are indispensable for successfully implementing large-cohort clinical plasma proteomics studies",

Professor Matthias Mann,
Max-Planck Institute of Biochemistry,
Proteomics and Signal Transduction

Optimized standard methods

Throughput (samples/ day)	Cycle Time (min)	Gradient Length (min)	Flow rate ($\mu\text{l}/\text{min}$)	Column dimensions (length/ID/bead size)
300	4,8	3,2	4,0	4cm / 150 μm / 3 μm
200	7,2	5,6	2,0	4cm / 150 μm / 3 μm
100	14,4	11,5	1,5	8cm / 100 μm / 3 μm
60	24,0	21,0	1,0	8cm / 100 μm / 3 μm
30	48,0	44,0	0,5	15cm / 100 μm / 3 μm

High instrumentation efficiency



**Methods subject to change*

Disposable trap columns

- The sample is loaded and desalted offline on a C18 pipette tip, the Evotip
- The autosampler picks up the tip and places it in the injection port

Partial elution

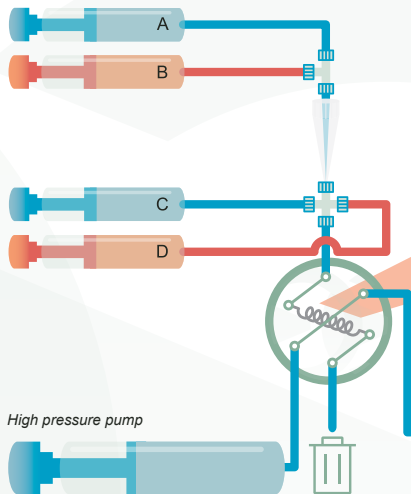
- Leaves contaminants on the Evotip disposable trap columns
- Elutes analytically relevant peptides
- Minimizes cross-contamination
- Extends column lifetime



Low maintenance components

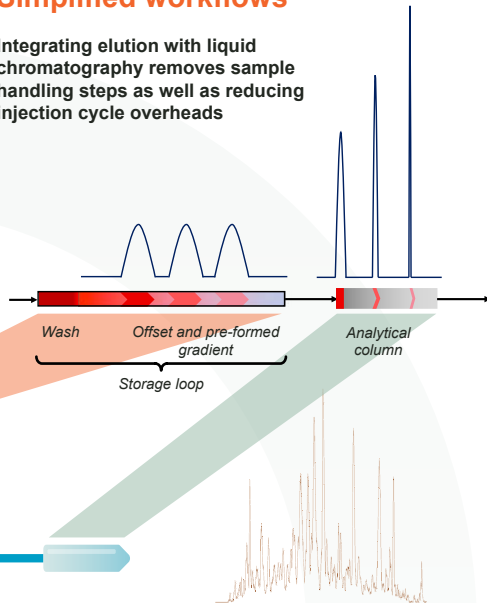
All the elution and gradient formation happen at low pressure, ensuring minimal wear and tear

Low pressure pumps



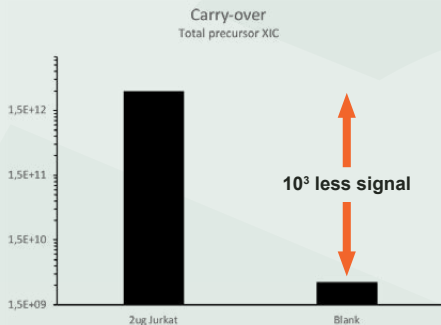
Simplified workflows

Integrating elution with liquid chromatography removes sample handling steps as well as reducing injection cycle overheads

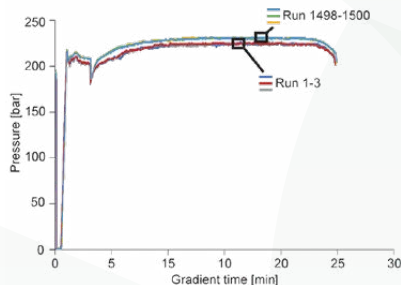
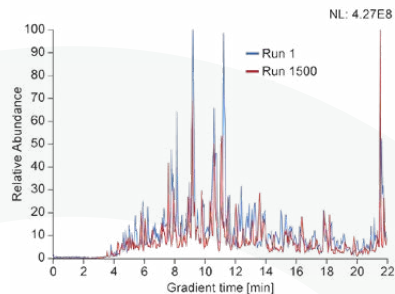


Robust throughput

- 10x reduction in carry-over
- 1,000 of runs on each analytical column
- 30,000 sample service interval

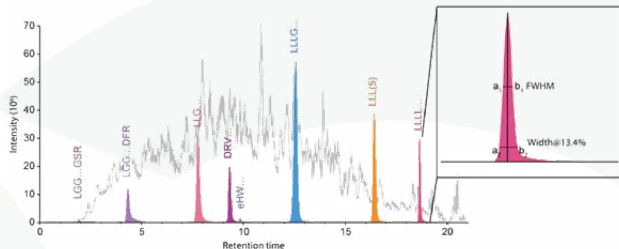


Data courtesy: Dr. Jacob Jaffe, Shawn Egri, and Dr. Steven A. Carr,
The Broad Institute, Cambridge, MA



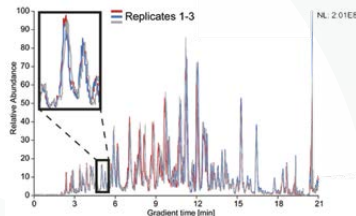
1500 injections of 1ug HeLa on the same column

Chromatographic performance

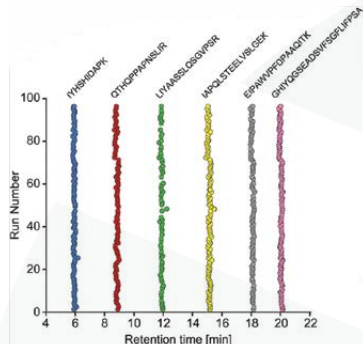


- Intelligent pump control allows efficient use of acquisition window
- Peptide mix in HeLa background used to calculate peak data
- Overlaid runs show good reproducibility in complex standard

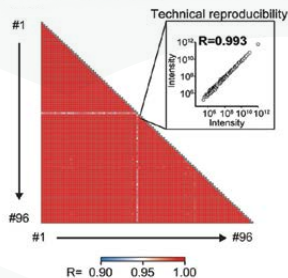
Throughput	FWHM	Peak capacity
Samples/day	[sec]	4σ / FWMM
300	1.5	35 / 72
200	2.2	41 / 97
100	4.0	63 / 129
60	6.8	79 / 161
30	15.2	111 / 216



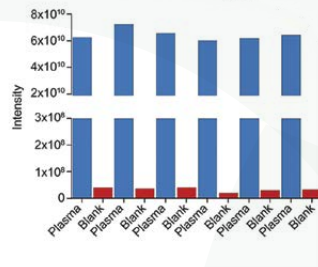
Reproducible plasma performance



Retention time stability of selected peptides over 96 runs



Pearson correlation matrix comparing the 96 runs



Low carry-over between runs (<0.1%)

Data courtesy: Dr. Philipp Geyer, Max Planck Inst., Martinsried

Fast and sensitive HCP screening

- Bottom-up mass spectrometry protocols are established for analyzing host cell proteins (HCPs)
- Detecting low abundance HCPs confidently and with high throughput is now possible with the Evosep One paired with a trapped ion mobility separation TOF instrument



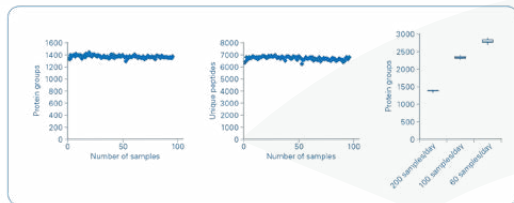
61 HCPs identified in 21 minutes from 25 µg NISTmAb (according to Huang et al., Anal. Chem. 2017, 89, 5436-5444) equivalent to approximately 1 µg load.

- 3 times faster*
- 2 times more sensitive*

**than 1h industry standard*

Data courtesy: FN-06 flash note; Bruker Daltonik GmbH, Germany

Protein identification with short gradients



- Stable protein and peptide ID rates over 96 injections with the 200 samples/day methods
- Examples of identification rates possible with different methods.

All measurements were performed on a Bruker Daltonics timsTOF Pro with 50 ng of HeLa digest and at least 15 replicates.

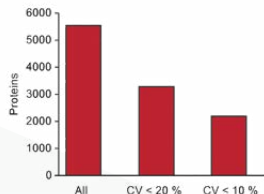
Data courtesy: LCMS-141 app note, Bruker Daltonik GmbH, Germany

Fast generation of DIA libraries

Spectral library of mammalian cell line proteomes made from 46 fractions of HeLa digests.

Total time	Gradient time	Peptides	Proteins
18.4h	16.1h	132,850	9,918

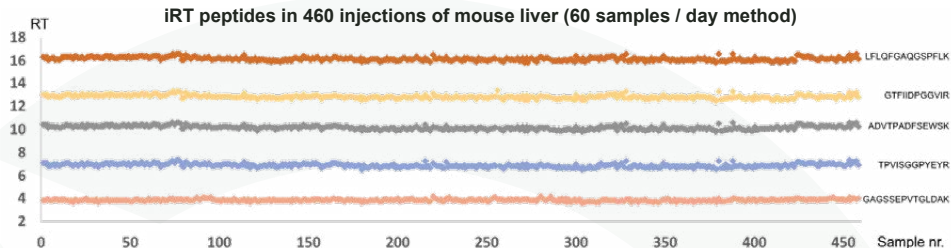
Example ID and quantitation performance with 60 samples/day method



MS1 120,000 res.
MS2 48 windows 1 Da overlap 15,000 res., 22 ms IT

Data courtesy: Prof. Jesper Olsen and Dorte Bekker-Jensen, Novo Nordisk Center for Proteome Research, Copenhagen on a Thermo Scientific Q Exactive HF-X

Large clinical proteomics sample set

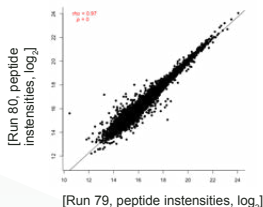


Enables large cohort studies in a fast and robust manner with

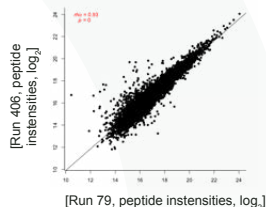
- Stable retention times
- High data consistency

across hundreds of injections

Close Technical Replicates



Distant Technical Replicates



Data courtesy: Dr. Ben Collins, Dr. Evan Williams and Prof. Ruedi Aebersold, ETH Zürich on a Sciex TripleTOF 6600

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Making clinical proteomics
100 times more robust
and 10 times faster

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