Case study

Whisper Zoom combines ease-of-use and simplicity for routine, high-sensitivity workflows

- Whisper[™] Zoom delivers sensitivity for highly versatile applications
- Robust methods enable high-throughput analysis for inter-laboratory studies
- Short learning curve facilitates rapid method implementation

Transition of high-sensitivity workflows to relevant applications

High-sensitivity proteomics is critical for advancing clinically relevant fields like single-cell proteomics, spatial proteomics, immunopeptidomics, and the study of low-abundance, post-translational modifications (PTMs). While mass spectrometry (MS) enables the analysis of minute sample amounts, its true potential is only realized when integrated with high-performance liquid chromatography (LC) methods that offer sensitivity, robustness, scalability, and user-friendliness. Whisper Zoom methods, powered by Evotip technology, are designed to meet these requirements, enabling efficient sample handling in high-throughput workflows. The simplicity of using Whisper Zoom methods helps both experienced researchers and non-experts. Meet three of our customers, for whom Whisper Zoom has become routine analytical technology. They have different analytical needs depending on their application, whether it is phosphoproteomics, single cell proteomics, or the analysis of laser micro-dissected cancer cells, they all share the need for reliable and simple solutions to drive their research. Learn from their experiences with the Whisper Zoom technology and see for yourself that cutting-edge proteomics can indeed be simple.

> "This was the first time I've ever seen the installation of new technology work so smoothly. From day one, we were already connected and collecting single-cell data."

Dr. Pierre Sabatier, Researcher at Uppsala University

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Achieving 100x more efficiency in phosphoproteomics

The newly developed Whisper Zoom methods are designed to deliver enhanced sensitivity, while maximizing recovery and achieving superior chromatographic performance. This advancement enables high-sensitivity proteomics experiments to be applied across a broad spectrum of research areas.

Pierre Sabatier, researcher at Uppsala University and former member of the Olsen lab at University of Copenhagen, noted that Whisper Zoom has reduced input material requirements for phosphopeptide enrichment workflows by up to 100-fold. This enabled him to perform deep phosphoproteome analysis on donor-derived pancreatic islet cell clusters, hereby advancing diabetes research.

> "All my collaborators, many of whom are biologists or pharmacologists, are amazed. They're thrilled because we no longer need to constantly debate whether to reduce the number of replicates by pooling material due to limited amount of sample."

Dr. Pierre Sabatier, Researcher at Uppsala University



Christoph Krisp, an Applications Development Scientist at Bruker, highlights that "The lower flow rates of Whisper Zoom not only boost signal through better ionization efficiency but also sharpen peaks, reducing ion suppression". This improvement in sensitivity and chromatographic separation is essential for high-sensitivity workflows like immunopeptidomics and differentiating splice variants.

Jongmin Woo, a scientist at Johns Hopkins University, reports that the use of Whisper Zoom 40 SPD has resulted in a 200% increase in peptide identifications compared to the standard 30 SPD method. This significant boost in sensitivity is allowing him to study the heterogeneity of patient-derived, laser micro-dissected cancer cells, with the goal of advancing cancer detection and treatment strategies.

Consistent and robust inter-laboratory analyses

High-sensitivity workflows become irrelevant in clinical applications if it sacrifices robustness and reproducibility. The Whisper Zoom methods are specifically developed to address these challenges by utilizing minimal-length flow paths, which significantly enhance both robustness and reproducibility.

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Pierre Sabatier highlights that this feature is particularly advantageous for efficiently processing valuable clinical samples, where limited material often restricts the possibility of repeat analyses. In clinical research, the ability to produce consistent and reliable results is paramount, especially when working with scarce samples. The enhanced robustness of the Whisper Zoom methods ensures that researchers can trust the integrity of their findings, leading to more confident interpretations and better clinical decisions.

The Evotip, a single use and disposable peptide trap column designed to minimize carryover, is essential for ensuring the robustness of low-flow methods such as Whisper Zoom. Jongmin Woo emphasizes that the convenience of Evotips, which can be discarded after loading peptide samples, significantly enhance instrument performance. By minimizing the risk of column clogging, Evotips play a critical role in maintaining long-term reliability and delivering consistent results.

> "The consistency and standardization of the system makes it easy to compare performance across different locations. Retention times are always spot on because we use the same defined workflows, the same column type, and the same gradients, making the system highly comparable and reliable for troubleshooting, even across different locations."

Dr. Christoph Krisp, Application Development Specialist at Bruker Daltonics



Moreover, the use of Evotips allows researchers to focus on generating high-quality data without the worry of carryover effects from previous samples. This is especially crucial in high-throughput environments, where consistent performance is necessary for drawing accurate conclusions from complex datasets.

Another unique feature of the Evosep One is the standardization of Whisper Zoom methods, which enables true inter-laboratory analysis, facilitating collaboration and ensuring that methodologies are consistent and reliable regardless of the lab setting. This standardization not only enhances workflow efficiency but also broadens the applicability of proteomics research across various scientific disciplines. Christoph Krisp emphasizes this capability is a significant advantage for conducting application development across multiple Bruker laboratories.

Scalable solutions unlock single-cell heterogeneity

The Whisper Zoom methods are not only characterized by their sensitivity and robustness but also their scalability, with the capacity to process up to 120 samples per day. This is crucial for processing large cohort studies, that are essential in achieving robust statistical power to draw valid conclusions. Christoph explains, "Scalability is crucial for large cohort studies where robust statistical power is needed. While analyzing one cell at a time for hundreds of cells is valuable, higher throughput is necessary to avoid spending months on the instrument".

Christoph is particularly impressed by the short overhead times associated with the Whisper Zoom methods. Even at low flow rates, the system requires only a couple of minutes for sample loading and column equilibration. This efficiency is facilitated by a distinctive feature of the Evosep One: the pre-formed gradient, which streamlines the workflow and minimizes idle time.

To further enhance the throughput of the Whisper Zoom setup, Christoph has integrated TMT-based multiplexing for SCP preparation with the Whisper Zoom 120 SPD method and the fast-scanning, highly sensitive timsTOF Ultra 2 mass spectrometer (Bruker). This innovative



configuration enables the processing of over 1,000 cells in a single day, effectively minimizing batch effects typically associated with TMT-based workflows. Furthermore, it ensures reproducible proteome depth across analyses, which is vital for extracting meaningful biological insights from large-scale, single-cell experiments. By combining these advanced techniques, researchers can achieve unprecedented levels of throughput and precision in proteomics, paving the way for significant advancements in biological research.

This combination not only enhances workflow efficiency but also significantly elevates the quality and reliability of the data generated, enabling scientists to make more informed decisions in their studies and contribute to a deeper understanding of complex biological systems.

"We used multiplexing at 120 SPD, running over 1,000 samples in a day. Although the number of identified proteins decreased slightly due to moving from DIA to DDA and the nature of multiplexing, we still identified around 1,500 proteins per cell or single-cell equivalent. The throughput and statistical power were impressive." Christoph Krisp explains.

Ease-of-use enables adoption from day one

The ability to quickly and seamlessly adopt new technologies, such as the simple and user-friendly Whisper Zoom methods, empowers researchers to concentrate on generating valuable data rather than troubleshooting. For Jongmin and his colleagues at Johns Hopkins University, this is a key factor in choosing the Evosep One over other conventional liquid chromatography systems.

Pierre has also experienced firsthand the advantages of Whisper Zoom's simplicity and the robustness of the instrument. Since implementing



the Whisper Zoom methods in the Olsen lab seven months ago, they have successfully processed nearly 10,000 low-input samples, demonstrating the method's efficiency in handling demanding workloads.

This enhanced stability not only streamlines daily workflows but also contributes to more reliable data output. By reducing the complexities associated with traditional systems, Whisper Zoom allows researchers to maximize their productivity and focus on their core scientific inquiries, ultimately accelerating the pace of discovery in their respective fields. *The combination of user-friendly features and consistent performance makes Whisper Zoom an invaluable asset in the modern research environment.*

> "Even though we have conventional chromatography systems available, people choose the Evosep One because it has a shorter learning curve."

Dr. Jongmin Woo, Researcher Analyst at Johns Hopkins University



The Evosep One is for General Laboratory Use.

