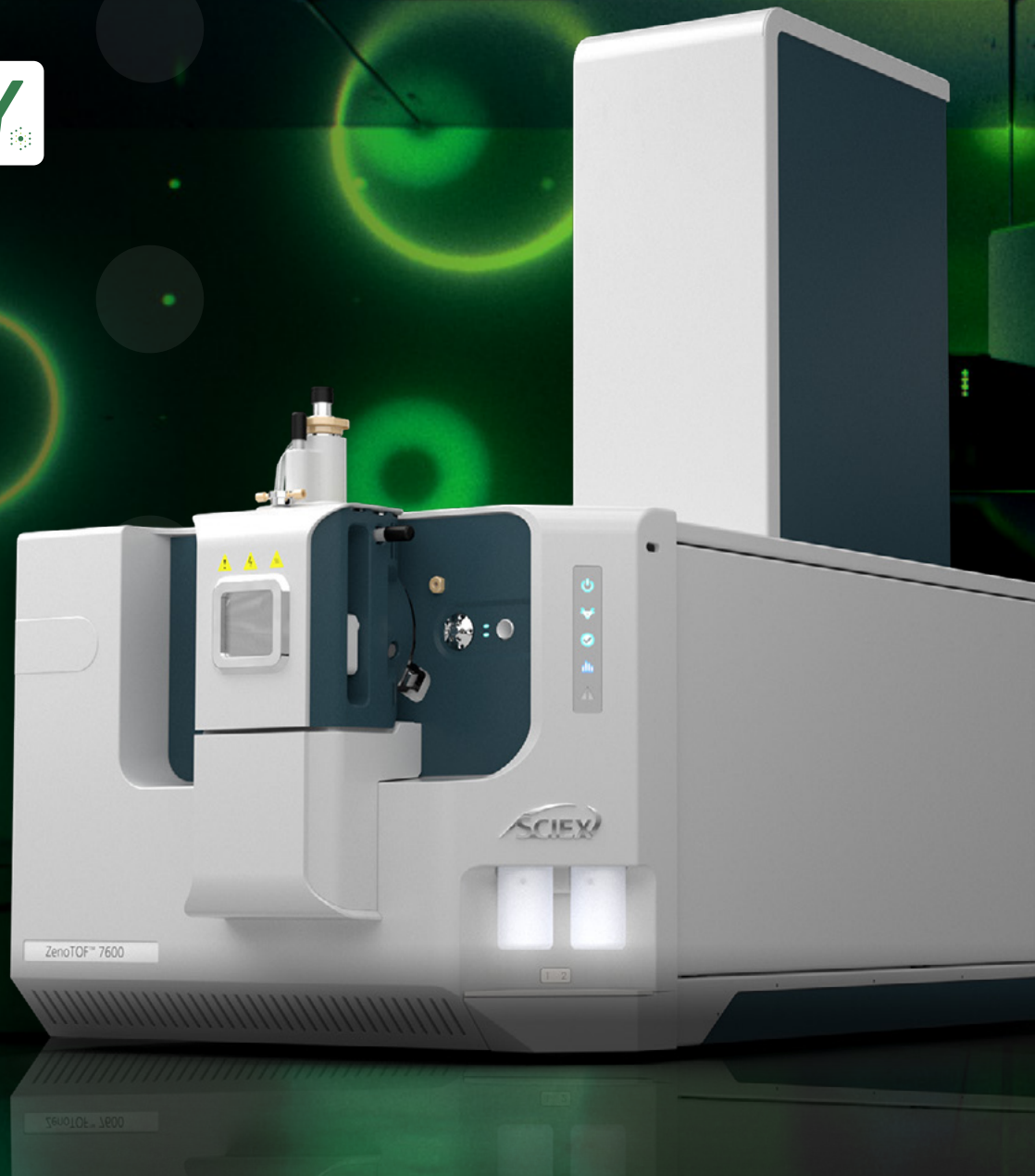
ZenoTOF 7600 system

BE ***EXTRAORDINARY***

The Zeno revolution is now...

Introducing the SCIEX ZenoTOF 7600 system



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ZenoTOF 7600 system

A powerful leap in proprietary innovation, this high-resolution accurate mass system combines the power of Zeno trap pulsing with EAD fragmentation technology (electron activated dissociation) to unlock sensitivity gains that reveal new, rare, or even previously undetected information on an everyday basis. Detect up to 20x more ions in every experiment and access a spectrum of tunable fragmentation techniques to uncover new perspectives for every molecule, in every experiment.

Electron activated dissociation

Fine tune the fragmentation energies specific to your molecules of interest, and acquire key MS/MS features for large molecules, peptides, lipids and small molecules.

Zeno trap pulsing

Gain more useful MS/MS information in every experiment, particularly on lower abundance species, with a 5-20x gain in MS/MS sensitivity coupled with either EAD or CID fragmentation.

Software

Powered by SCIEX OS mass spectrometry software, intuitive algorithms and automation enable you to make informed decisions quickly. Transform your LC-MS/MS workflows with remarkable quantitative usability, efficiency, and integrity that streamlines your entire lab.

Ion sources

Compatible with advanced source designs that minimize contamination and allow you to obtain fast, automated calibrations across flow rate ranges.

Flow rate flexibility

- Turbo V source
  - ESi 5µL/min to 3mL/min
  - APCI 200µL/min to 3mL/min
- Optiflow Turbo V
  - µflow 1µL/min to 200µL/min
  - nano 100nL/min to 1,000nL/min

Calibration delivery system

Designed with a new calibration solution that automates mass calibration, and ensures mass accuracy of the system is maintained throughout acquisition.

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The Zeno revolution is now...

Zeno trap and **EAD**. A powerful combination of unparalleled MS/MS sensitivity and a step-change in fragmentation technology. Together they provide the ability to acquire key MS/MS features needed to:

- Characterize large molecules including post-translational modifications
- Elucidate positional isomers on small molecules and lipids
- Identify and quantify proteins and peptides at unparalleled speed

- **Overcome QTOF MS/MS duty cycle deficiencies**
  >90% ions injected into the TOF
- **Sensitivity gains of up to 5-20x with Zeno trap pulsing**
  Identify and quantify low abundance species
- **Tunable fragmentation of all molecule types**
  Utilize controlled electron activated dissociation (EAD)
- **MS/MS scan rates of up to 133Hz**
  Improved DDA and high resolution MRM (MRMR^{HR})

Zeno MRM^{HR} cAMP

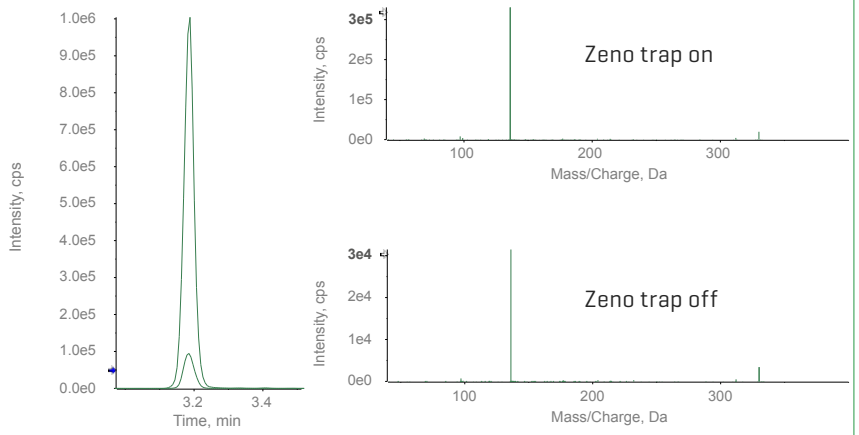

Figure 1. Impact of ions accumulated in the Zeno trap before being pulsed rapidly into the TOF both increases in sensitivity but also spectral quality.

Zeno EAD IDA of Darunavir-O-glucuronide

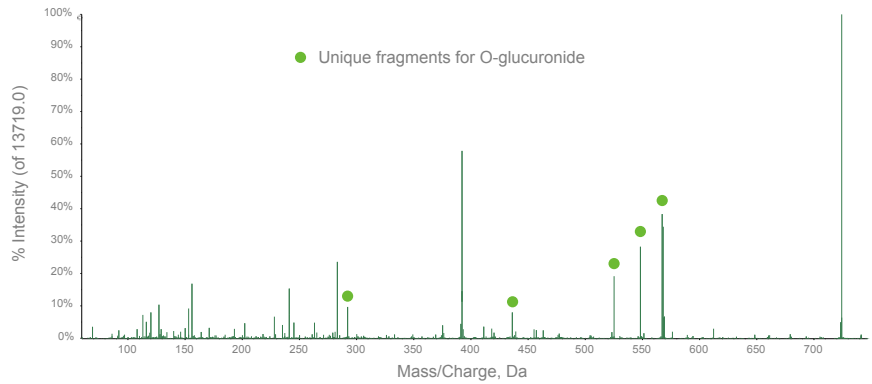

Figure 2. Impact of EAD for singly charged molecules. Here shown is Darunavir-O-glucuronide, a phase 2 metabolite which is very difficult to structurally elucidate via CID because the glucuronide motif is lost as a *neutral loss* under CID conditions. Utilizing EAD produces diagnostic fragment ions to determine the location of the glucuronide.

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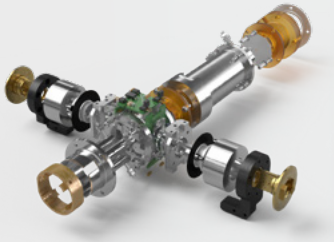
A new mindset in accurate mass LC-MS/MS technology

Driven by the power of Zeno trap coupled with EAD technology, this fragment-centric revolution unlocks sensitivity gains allowing you to uncover new information, for more certainty in your results, and to make better-informed decisions, faster.

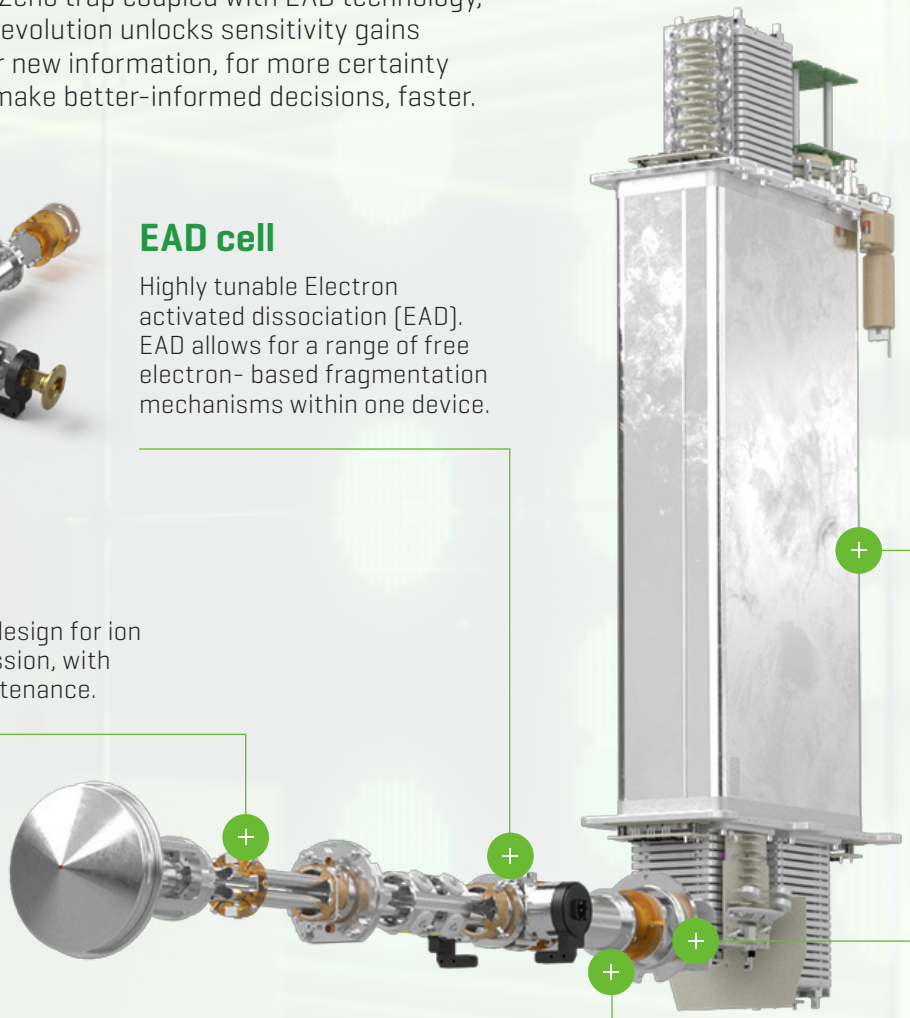


EAD cell

Highly tunable Electron activated dissociation (EAD). EAD allows for a range of free electron- based fragmentation mechanisms within one device.

QO Design

Improved ion optics design for ion capture and transmission, with easy access for maintenance.



TOF

N-optic TOF design provides optimal mass accuracy and resolution without compromising sensitivity.

- Heated TOF path & drone heaters
- Mass Range: 40 to 40 kDa in TOF
- Resolution: ≥ 42k at m/z 956
- MS/MS Speed: 133Hz
- Mass Accuracy: <2 ppm RMS Ext., <1 ppm RMS Int
- Positive & Negative LDR: >5 orders Inter-scan LDR

Zeno trap

Improved MS/MS duty cycle gain with 5-20x gain in MS/MS sensitivity coupled with either EAD or CID fragmentation.

LINAC collision cell

High drive frequency collision cell provides a better ion transmission, higher duty cycle, and improved resolution by focusing ions prior to entering TOF.

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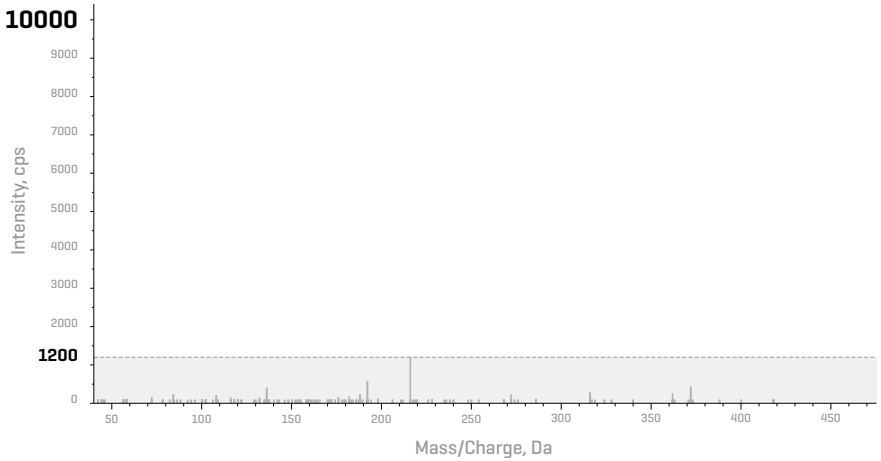
Zeno trap

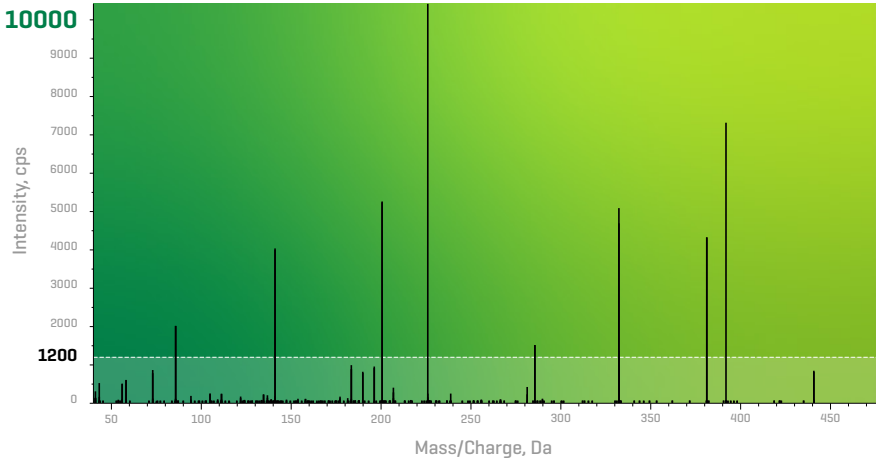
The next era of sensitivity for accurate mass

Ions are accumulated in the Zeno trap before being pulsed rapidly into the TOF, meaning up to 20x more ions can be detected. Consequently, each TOF experiment contains more useful MS/MS information, particularly on lower abundance species that were previously undetectable, introducing our customers to a new level of sensitivity.

Without Zeno trap pulsing



With Zeno trap pulsing



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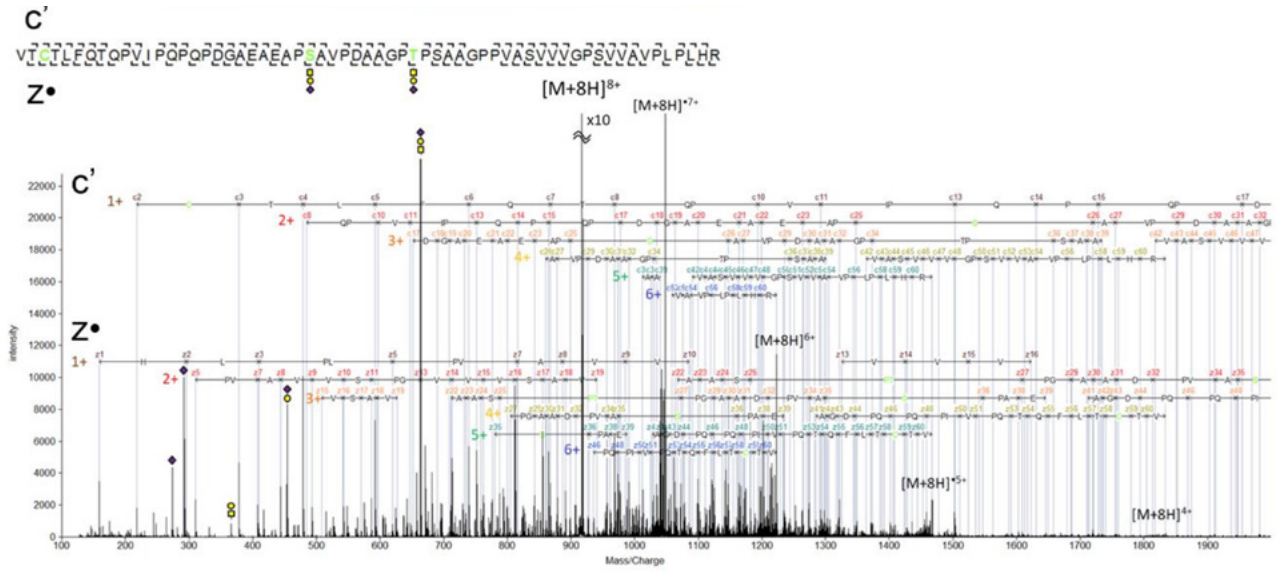
Electron activated dissociation (EAD)

A step change in fragmentation technology

The ability to tune electron kinetic energy extends the utility of the approach to all molecules type from singly charged small molecules to large multiply charged proteins. EAD allows for a range of reagent free electron- based fragmentation mechanisms within one device, and has the capability to fragment peptides whilst retaining critical MS/MS information for both identification and localisation of PTMs. Unlike other electron-based fragmentation techniques, EAD delivers reproducible, consistent data, even at fast scan speeds, compatible with UHPLC timeframes, delivering higher efficiency than ETD. In this instrument geometry coupling EAD with the Zeno trap allows for detection of very low abundant diagnostic fragment ion species leading to greater sequence coverage.

EAD fragmentation of an o-glycopeptide



Zeno pulsing

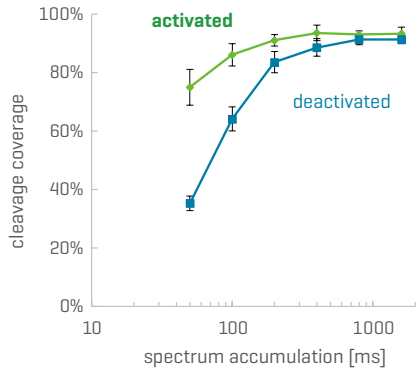


Figure 4. O-Glycosylation as shown requires comprehensive fragment coverage to confirm the glycan and also the site localization. Higher energy EAD (hot EAD) as shown for a complex O-glycopeptide shows near complete sequence coverage and both glycans localized in the centre portion of the peptide.

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Confident structural elucidation of xenobiotics

The ZenoTOF 7600 system brings a new level and depth of information to qualitative and quantitative workflows with electron activated dissociation. When combined with the Zeno trap comprehensive fragment coverage can be obtained to enable more confident structural elucidation.

The hardest challenge for CID is the fragmentation of phase 2 conjugated metabolites such as glucuronidation. EAD generates unique fragments which enables the differentiation of N- and O- Glucuronides which are otherwise lost during the CID fragmentation step and without the use of chemically synthesized standards, elucidation is rather challenging. With the EAD approach, 32 diagnostic fragments were detected this was possible because with the Zeno trap the signal is considerably enhanced. Therefore, the use of EAD is a step forward to improve productivity with fewer experiments needed.

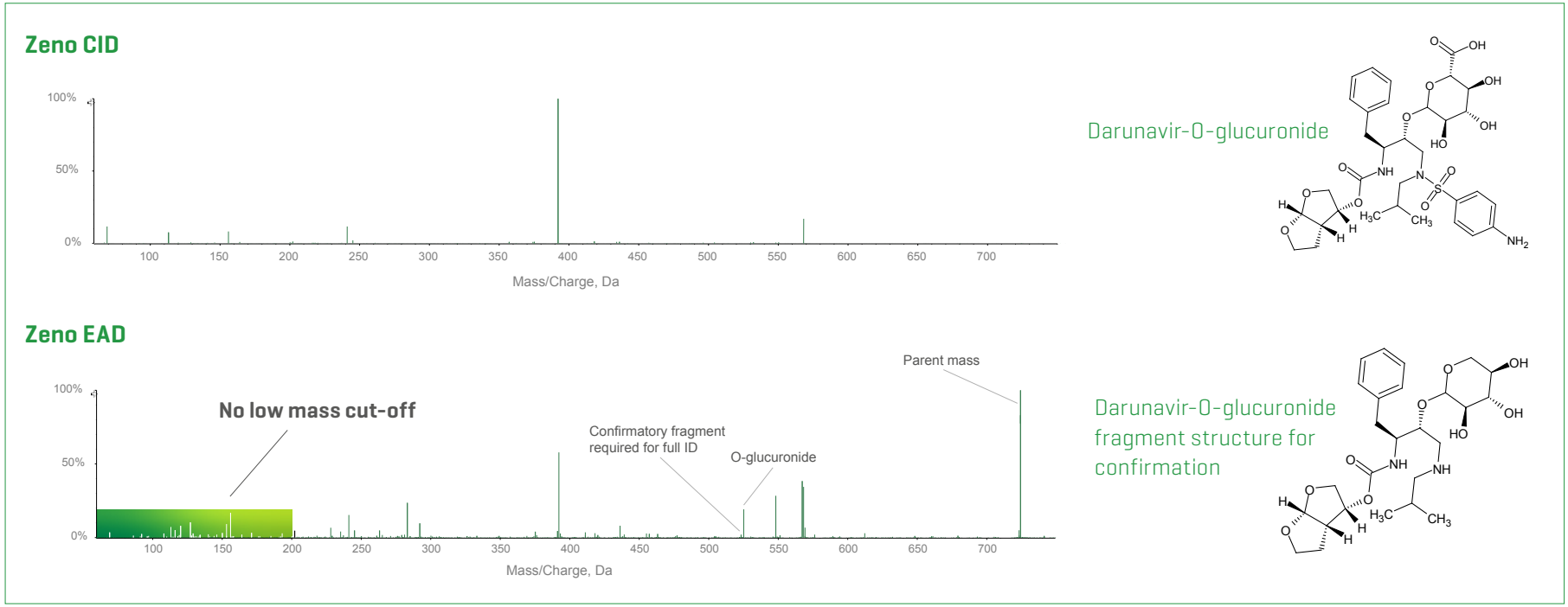

Figure 5. The EAD Spectrum. EAD spectrum which produces an important fragment ion with the glucuronide preserved which allows localization of the glucuronide modification. Also, low mass fragment ions are preserved by using a QTOF-based platform.

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Complete characterization of lipids in a single experiment

Lipids are a complex group of compounds with subtypes that share a similar high-level structure. For example, triglycerides consist of a glycerol group bonded to three long hydrocarbon chains with additional functional head groups attached in some cases. Small but meaningful differences between lipid species such as the location of a single double bond along the hydrocarbon chain can have important implications for health and disease. In one experiment, EAD provides all of the information for complete lipid characterization that normally requires multiple technologies and experiments.

Complete characterization of lipids involves the identification of:

- Head group
- Backbone
- Regio-isomerism
- Double bonds
- Cis/trans isomerism

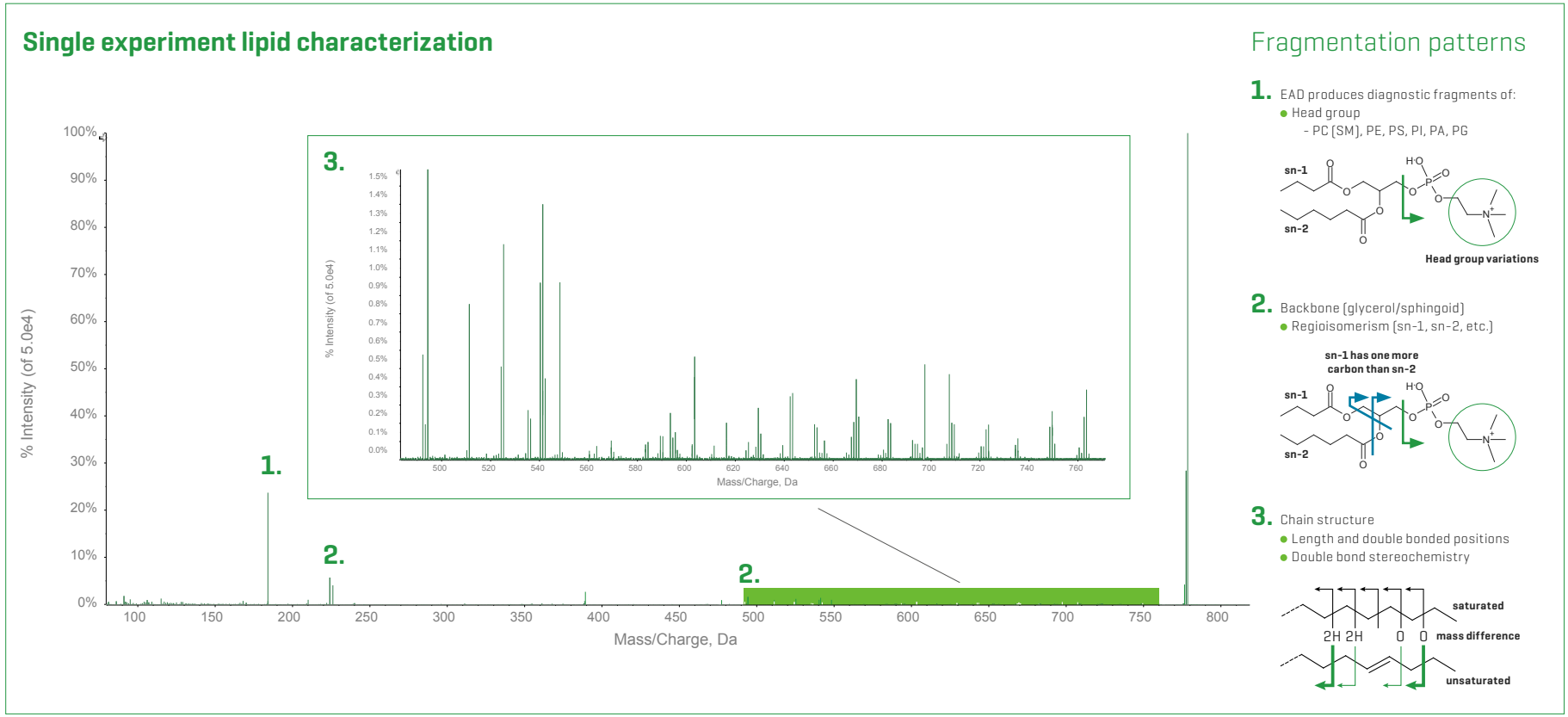

Figure 6. Single experiment lipid characterization. The complete de novo identification and characterization of the lipid PC 16:0 / 18:1(n-9:cis) in a single experiment.

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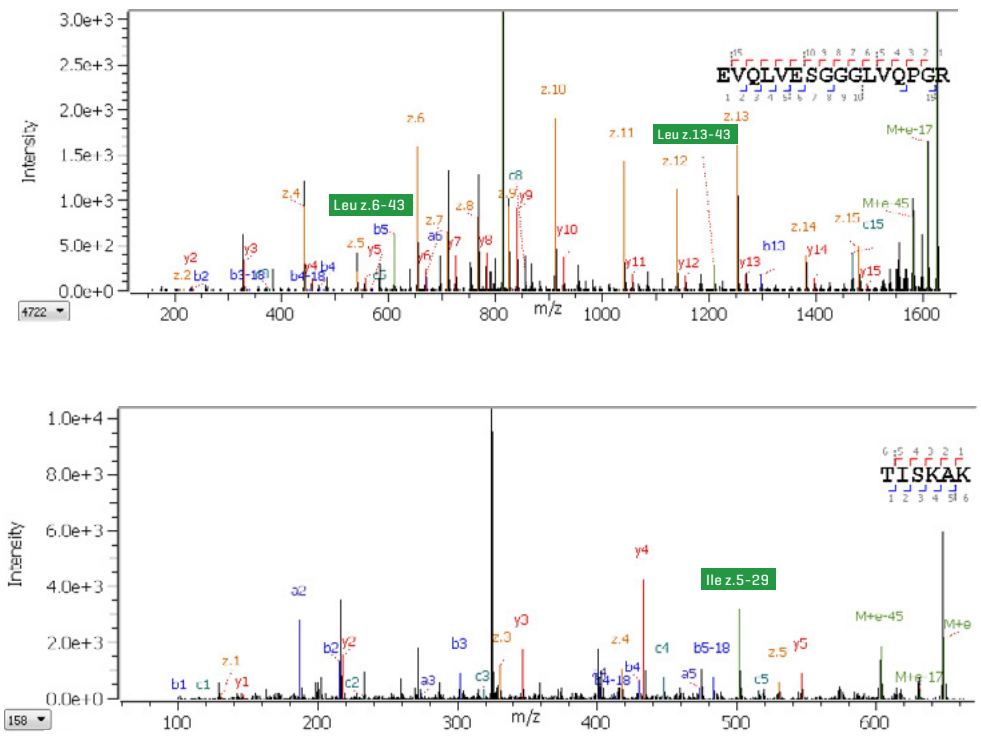
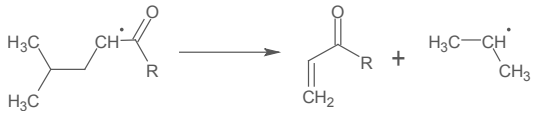
Differentiate between leucine and isoleucine in a single run

Some amino acids are identical in mass, such as aspartic acid/isoaspartic acid and leucine/isoleucine making their differentiation impossible using CID. Zeno EAD however, can identify these isomers from additional fragment ions that are produced. For example, with leucine/ isoleucine secondary w-ion fragments caused by further fragmentation of the backbone z-ion can be used for identification to rapidly confirm primary structure in the production of biopharmaceuticals.

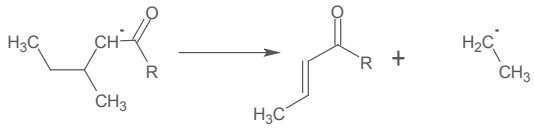
Differentiation of leucine and isoleucine using ZenoEAD



Leucine



Isoleucine



Key Benefit

Zeno EAD enables the differentiation of isomeric amino acids that otherwise cannot be differentiated using conventional low energy CID.

Figure 7. EAD clearly indicates the identity of two leucine residues within this peptide sequence through the loss of 43 Da from the z6 and z13 ions. At the bottom, loss of 29 Da from the z5 ion identifies an isoleucine within this peptide sequence.

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Straightforward localization of N-glycosylation

Glycopeptides are the most common critical quality attribute for protein therapeutics. Glycosylation can affect; protein folding, protein stability, solubility and cell adhesion and therefore needs to be fully characterized to ensure the safety of biologic drugs. CID fragmentation typically misses the specific fragment ion critical for localizing the attachment point of a glyco group on the peptide backbone. In contrast, EAD spectra are dominated by these fragment ions, making localization straightforward.

As the mass difference between fragment ions on both sides of the attachment point enable calculation of the glycan molecular weight, both EAD and CID can be complementary for glycopeptide analysis.

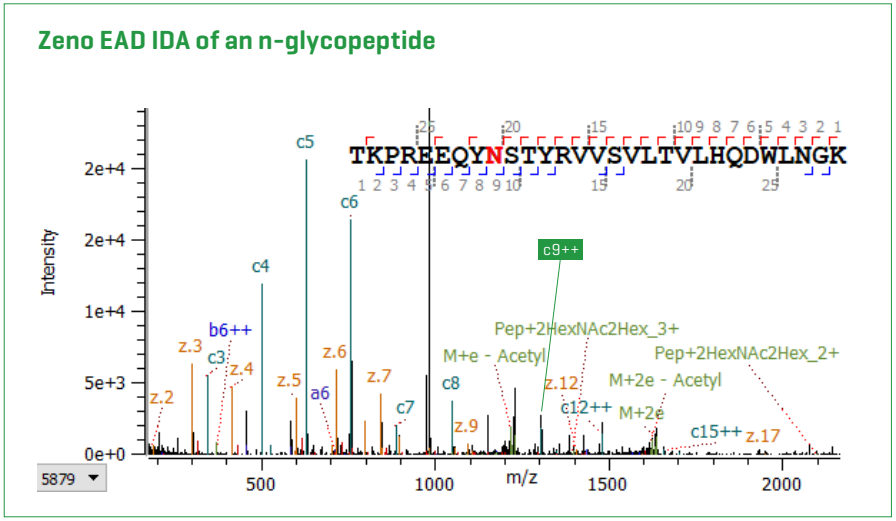

Figure 8. EAD provides both peptide sequence and glycan localization (c9++ ion).

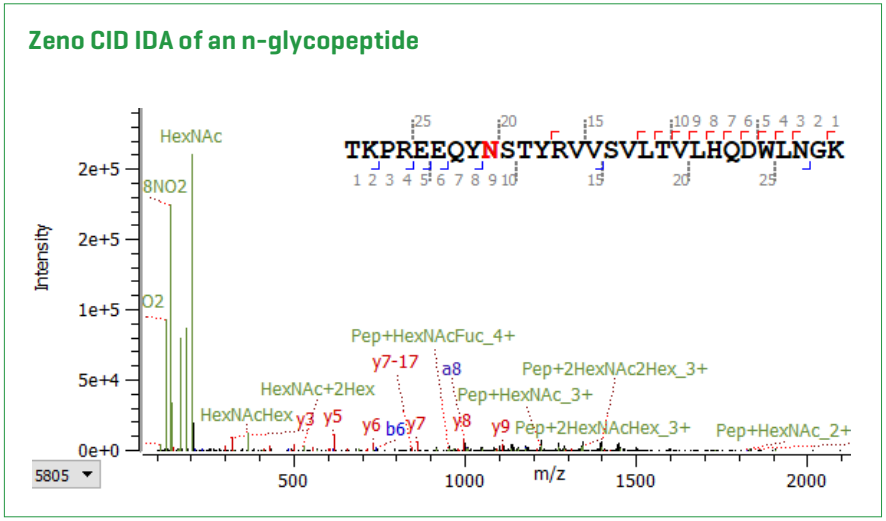

Figure 9. CID only shows glycan loss or peptide backbone information, not both in same MS/MS spectrum in a descriptive manner; localization is not possible.

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Over 40% more proteins identified using Zeno MS/MS

Utilizing high throughput micro flow methodologies, the ZenoTOF 7600 system breaks through the 3,000 protein groups in 21 minutes for the first time. Unique Zeno trap pulsing provides significant gains in peptide and protein identifications for proteomics experiments. Up to ~ 45% improvements in the number of protein IDs and up to a 145% increase in the number of peptide IDs, compared to previous TripleTOF technologies.

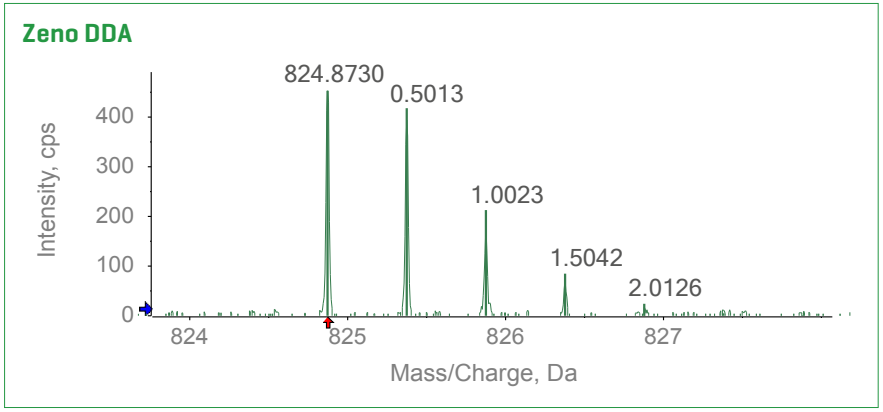

Figure 10. Impact of using Zeno trap for IDA. The TOF MS is shown for a peptide at 25 ng sample load which was triggered for the Zeno MS/MS. The isotopic fidelity is shown up to the M+4 isotope.

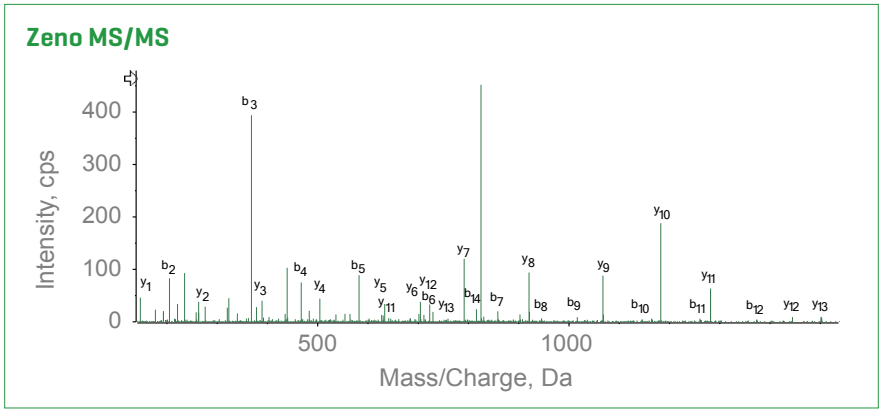

Figure 12. Impact of using Zeno trap for IDA. The Zeno CID MS/MS is shown for the precursor in figure 10. For a low abundant precursor a high quality MS/MS for identification is acquired with near complete sequence coverage and excellent signal to noise.

Peptide and protein gains

	Gradient duration (min)	1000 ng
Peptide Gains	10	72%
	45	145%
Protein Gains	10	41%
	45	46%

Figure 11. The impact of the Zeno trap on CID IDA(DDA) over previous platforms is shown. Significant improvements in both peptide and protein numbers are observed at short and medium gradient lengths.

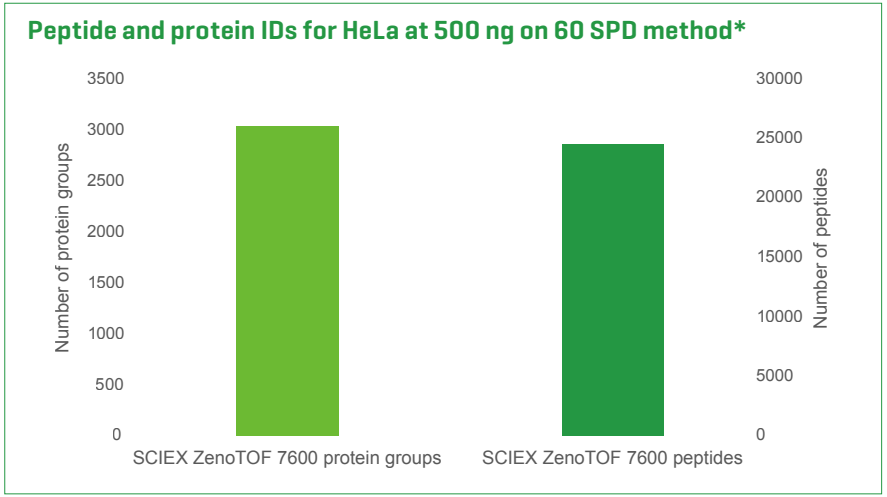

Figure 13. Routinely achieving high protein and peptide identifications at high throughput is difficult. The ZenoTOF 7600 system when coupled with a highly reproducible micro flow solution drives significant protein and peptide identifications breaking through >3,000 protein groups and >20,000 peptides using technical quadruplicates. * Evosep: Towards a Standardized Omics Platform with the 60 SPD https://www.evosep.com/wp-content/uploads/2020/06/AN-008-20-06_60SPD.pdf

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SWATH acquisition for higher throughput proteomics

The ZenoTOF 7600 unlocks unprecedented speed for SWATH acquisition with variable windows, with scan rates up to 133Hz (133 MS/MS per second). This enables quantitative flexibility to match the throughput demands of translational proteomics. Higher throughput proteomics requires accurate, precise and reproducible quantification of all analytes, at all concentrations.

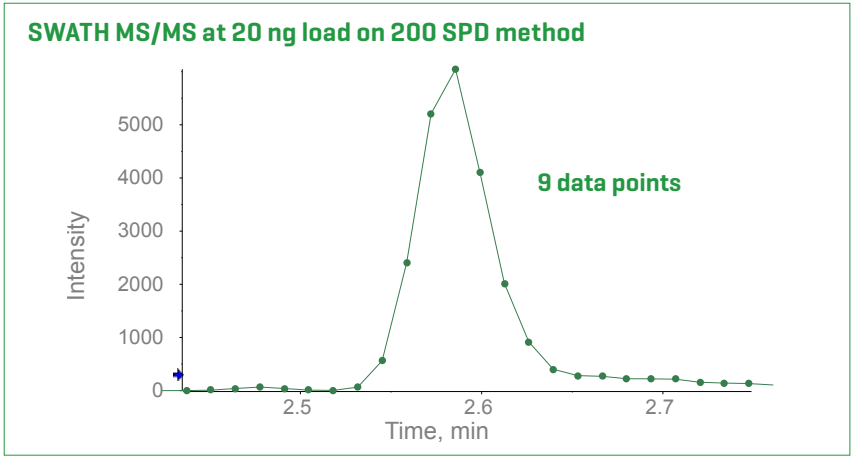

Figure 14. The step to 5 minute gradients at 200 samples per day for quantification requires high acquisition rates for MS/MS to maintain the number of data points at all concentrations. Shown is 20 ng of a K562 digest for the peptide YYVTYYDAPGHR.

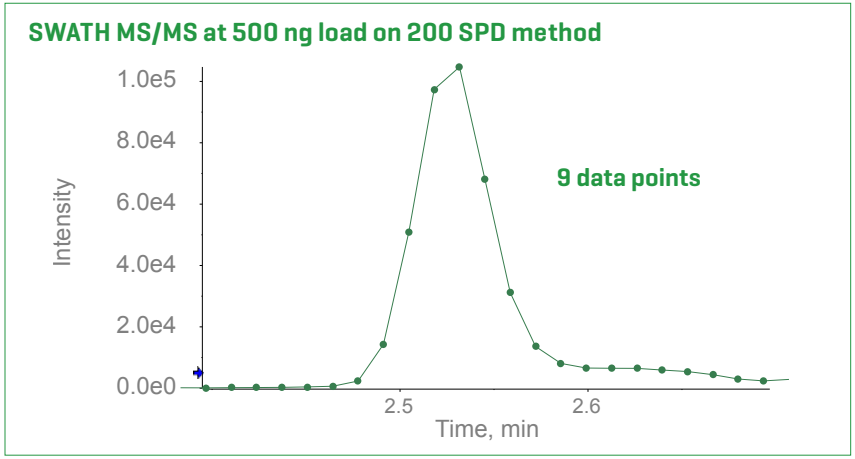

Figure 15. Continuation from figure 14, is a high load at 500ng of a K562 digest with the peptide YYVTYYDAPGHR extracted.

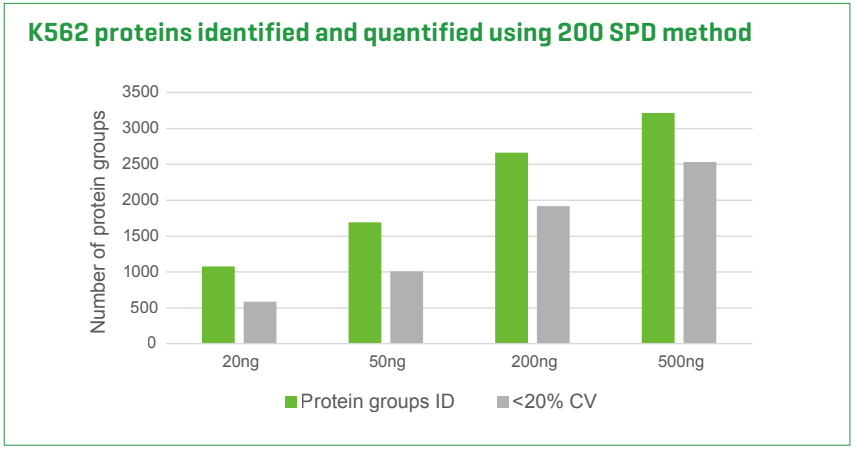

Figure 16. The ZenoTOF 7600 system shows excellent protein group coverage and 20% CV cutoff quantified proteins in just 5 minutes utilizing the 200 SPD method.

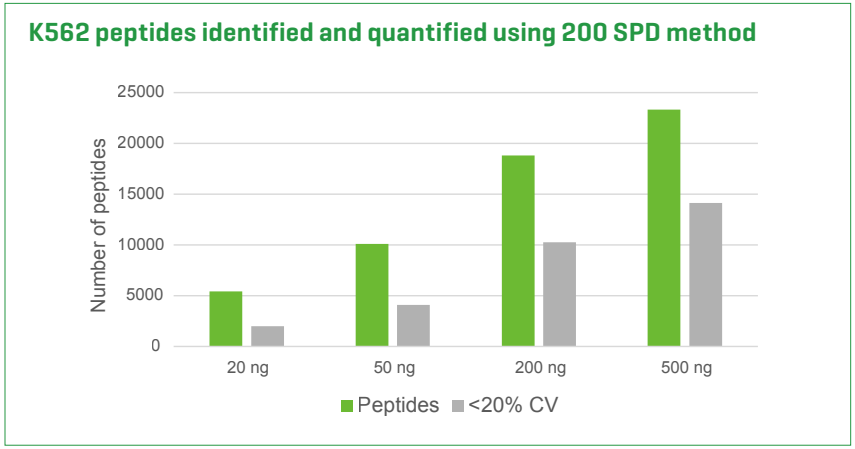

Figure 17. The ZenoTOF 7600 system shows excellent peptide coverage and 20% CV cutoff quantified peptides in just 5 minutes utilizing the 200 SPD method.

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Zeno MRM^{HR} unlocks new levels of sensitivity

Zeno trap pulsing on demand gives the ability to detect lower abundance ions at the same times as those in abundance, redefining the limits of quantification achievable with accurate mass. MRM^{HR} is a targeted approach that delivers high sensitivity and selectivity for screening and targeted quantification.

Small molecule

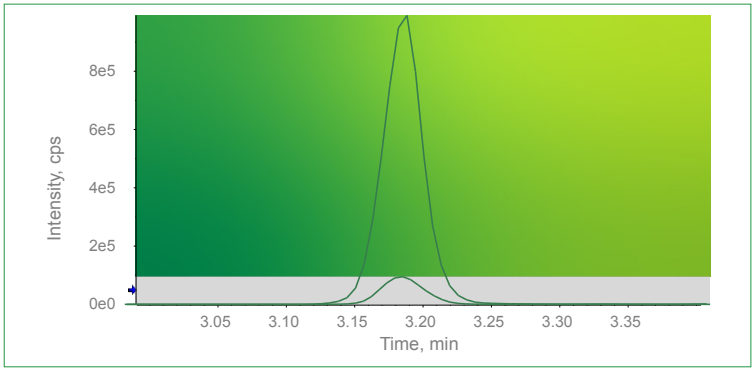

Figure 18. Targeted metabolite quantification - Significant sensitivity gains in MS/MS. Comparison of extraction ion chromatograms (XICs) for cAMP fragments obtained from MS/MS collect with Zeno trap on vs. Zeno trap off. Signal/noise ratio improved ~12.5 fold when using the Zeno trap.

Peptide quantification

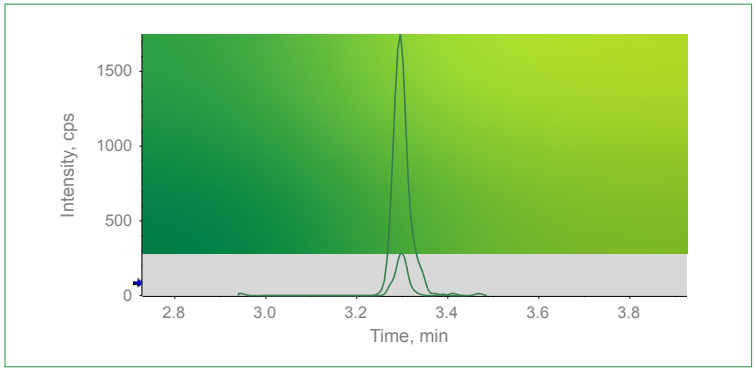

Figure 19. Targeted peptide quantification - Significant sensitivity gains in MS/MS. Comparison of extraction ion chromatograms (XICs) for LILTLTHGTAVC[CAM]TR fragments obtained from MS/MS collected with Zeno trap on vs. Zeno trap off. Signal/noise ratio improved ~8 fold when using the Zeno trap.

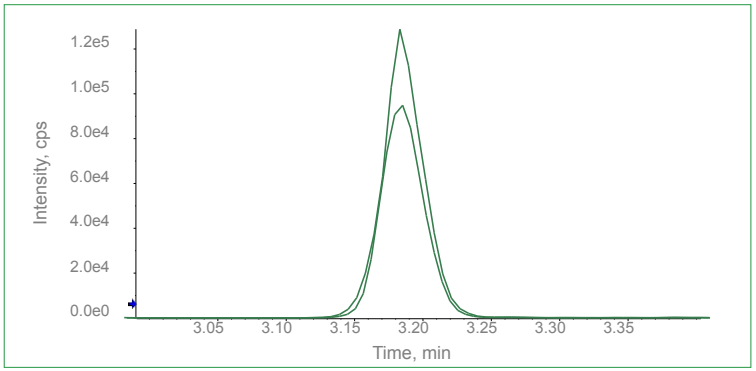

Figure 20. Better sensitivity in MS/MS with 10x less sample. Comparison of extraction ion chromatograms for cAMP fragments obtained from MS/MS collect with Zeno trap on (0.2 µL injection) vs. Zeno trap off (2 µL injection).

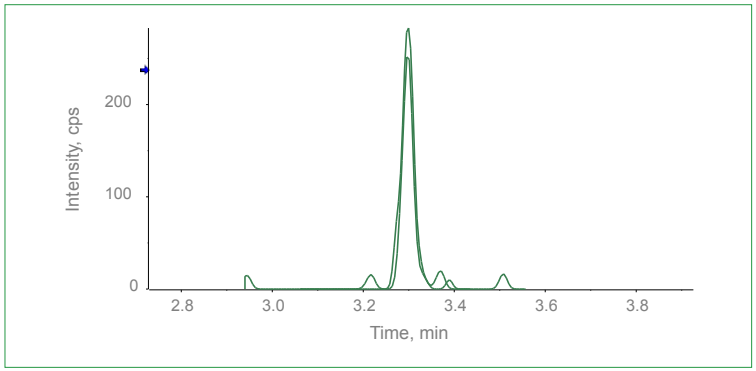

Figure 21. Better sensitivity in MS/MS with 8x lower concentration. Comparison of XICs for LILTLTHGTAVC[CAM]TR fragments obtained from MS/MS collect with Zeno trap on vs. Zeno trap off. Zeno trap off at 6.18ng/mL and Zeno trap on at 0.757 ng/mL.

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Software that powers the modern laboratory

The SCIEX ZenoTOF 7600 system is powered by SCIEX OS software. Fully integrated software that acquires, processes and reports your accurate mass data. Since it's launch SCIEX OS has continued to evolve it's functionality to meet the ever expanding challenges and opportunities of analytical science.

Consistency, accuracy and connectivity are present throughout all workspaces of SCIEX OS. Powerful algorithms and automation that will enable you to cut through complexity, straight to insight. SCIEX OS streamlines your lab delivering high-quality, actionable data for a wide range of applications,

- One platform for acquisition, processing and data management
- Simple user interface, modular design tiles
- Method development becomes routine and easily transferable
- Customizable for specific workflow requirements

"In new innovations the software is key, as the software enables the creation of results. Software that integrates data acquisition, processing, interrogation and reporting gives added value in terms of efficiency with which these results can be generated."

LIEVE DILLEN
Senior Principal Scientist for Assay Development and Analytical Support, Development Bioanalysis group, Janssen R&D, Belgium

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Delivering insight

Software is the vital connector between technology and insights that will drive discovery. Whether you are characterizing potentially complex proteins, routinely screening, or quantifying modalities in complex matrices they each require advanced data processing technologies to interrogate data and deliver actionable insight. The SCIEX ZenoTOF 7600 system is powered by a suite of software tools that will make these discoveries within your existing data pipeline.

SCIEX OS software empowers you with more than just processing. Take advantage of automated functionality, library management and audit trails for compliance. All the tools you need for any LC-MS/MS workflow, from routine to the most complex, in a single location.

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Genedata solutions in silico

Spectronaut™ Pulsar

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