Trajan Scientific and Medical





Introduction

HDX has become an indispensable tool for protein binding studies in pharmaceutical development. For over 15 years the Automation team at Trajan (formally LEAP Technologies) have been automating HDX experiments, and is the industry leader in the field. Our HDX systems are helping researchers across the globe come to answers about their proteins faster than traditional methods.

Successful HDX experiments

Automation addresses the 4 key factors for successful HDX experiments:

- Accurate timing, accurate liquid transfer and accurate temperature control
- pH control
- Sustained 0°C environment
- In-line digestion

Hydrogen Deuterium Exchange is an experimental technique for obtaining information about changes in the tertiary structure of proteins under different physiological conditions.

The LEAP HDX platform is an advanced, automated scheduling and experimental workstation providing ease of use, high reproducibility and exceptional data quality for HDX experiments.

Labelling

HDX relies on accurate timing of the labelling step.

Quenching

The system will optimize the timing of the labelling step to ensure quenching is just in time for the LC-MS injection, using a chilled syringe.

H/D Exchange HD Structured regions Dynamic regions exchange slowly exchange rapidly **Quench & Digest** Digestion localizes the Quenching locks in deuterium

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A typical experiment might consist of a dozen time points in triplicate for each protein sample. The LEAP HDX system will schedule the experiments and perform all steps for each time point of every sample.

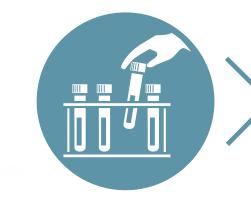
and unfolds the protein

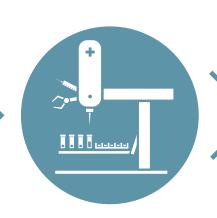
information

Long labelling experiments are automatically scheduled first to ensure the shortest possible instrument time.

Automated Hydrogen-Deuterium Exchange with optimized

consumables









Efficient system control and optimized experimental design

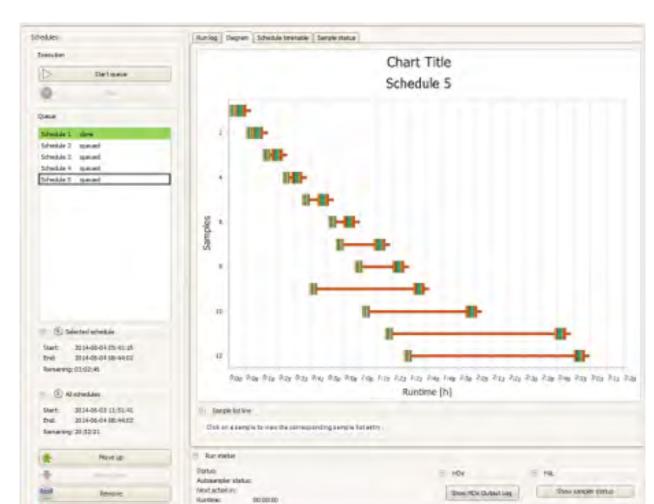
Our Chronos for HDX software now offers a streamlined interface for HDX users and automatically overlaps time points for maximum MS efficiency. Temperature control at all steps ensures reliable data. Sustained 0°C environment reduces back exchange, improves sample stability and overall reproducibility. Our most flexible automation platform yet offers the ability to run multiple experimental methods in the same sequence. Native data format compatibility with HDExaminer increases high confidence data and allows you to get to results faster.

ChronosHDX control software with HDExaminer data analysis

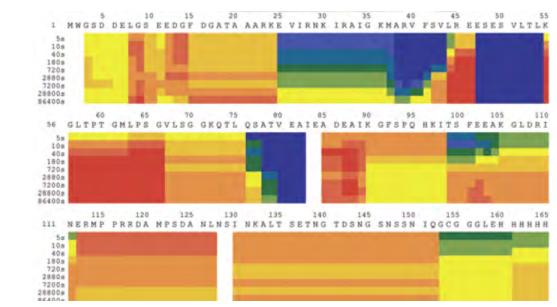
Meeting the challenge of producing the highest quality data sets.

HDX experiments require time-sensitive operation. Therefore, in traditional methods, an operator has to attend throughout the course of HDX experiments. However, time-controlled automation enables unattended operation using intelligent scheduling with Chronos software.

Trajan uses ChronosHDX scheduling software to automate the precise labelling and quenching of samples, as well as injection and digestion of proteins on the LC-MS analysis system.



- Precise on-exchange data point timing.
- Reproducible experimental conditions.
- Multi-valve control for trapping and elution of digested peptides to the LCMS system.
- Throughput optimization with elimination of timing conflicts.
- Simpler graphic user interface.
- Advanced method editing capabilities.
- Integrated with HDExaminer data processing from Sierra Analytics.



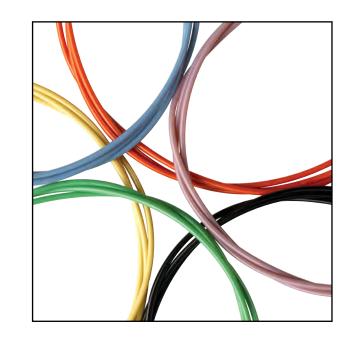


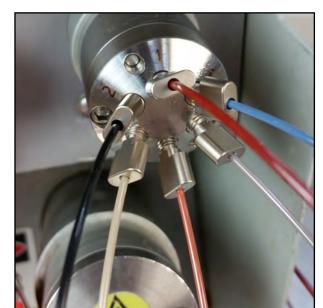
Optimized HDX fluidics

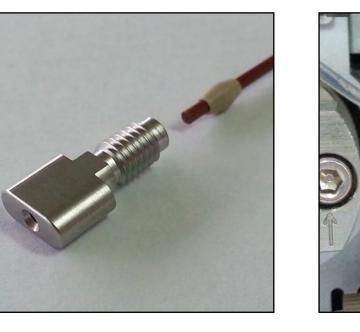
2018 Introduction of optimized HDX fluidics that enables full fluidic path backflushing, extending column life and maintaining consistent retention times to deliver higher quality data.

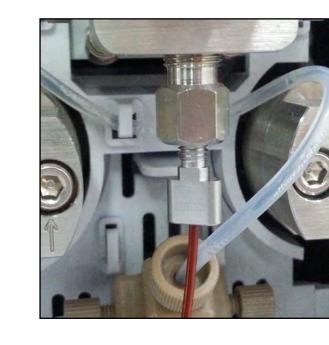
The new fluidics pathway is built upon UHPLC rated valves and column hardware (21 kpsi), with flow paths minimized to reduce swept volumes.

Trajan manufactured HDX specific pre-cut high pressure lines and fittings, designed to improve reliability and enhance performance. All fluidic components are designed to offer consistently reproducible results.









ProDx consumables designed for HDX



Columns and fluidics consumables specifically designed to optimize all aspects of the HDX user experience.

ProDx Protease column

Trajan has developed the ProDx Protease column specifically for use with the LEAP HDX system. The ProDx solution (ProDx Protease column with the updated valve design) improves workflow efficiencies and reduces instrument

The ProDx Protease column has optimized performance at 0°C, which is the ideal temperature for performing HDX reactions, without the need for separate temperature controls, which elevates back exchange. The ProDx column performs at lower backpressures with reduced carryover, reduced chance of contamination and minimizing the potential for overpressure leakage.

The ProDx column performs with lower carryover and increased desalting efficiency. When used in combination with the new valve system it can be backflushed. Improved protease coupling in the ProDx range reduces carryover from non-specific binding. Packed in a bioinert glass lined SS 2.1 mm x 30 mm column ensures the ProDx Protease column withstands high pressure digestion practices, making it compatible with other systems.

Implementing the full range of ProDx (ProDx Protease column, trap and the new valve system) ensure reduced carryover and the ability to backflush to remove frit loaded or undigested intact material. This overcomes traditional issues with non-selective binding and poor reproducibility.

Clean columns give consistent retention times and ultimately better quality data – the ProDx solution providing reproducible outcomes every time.

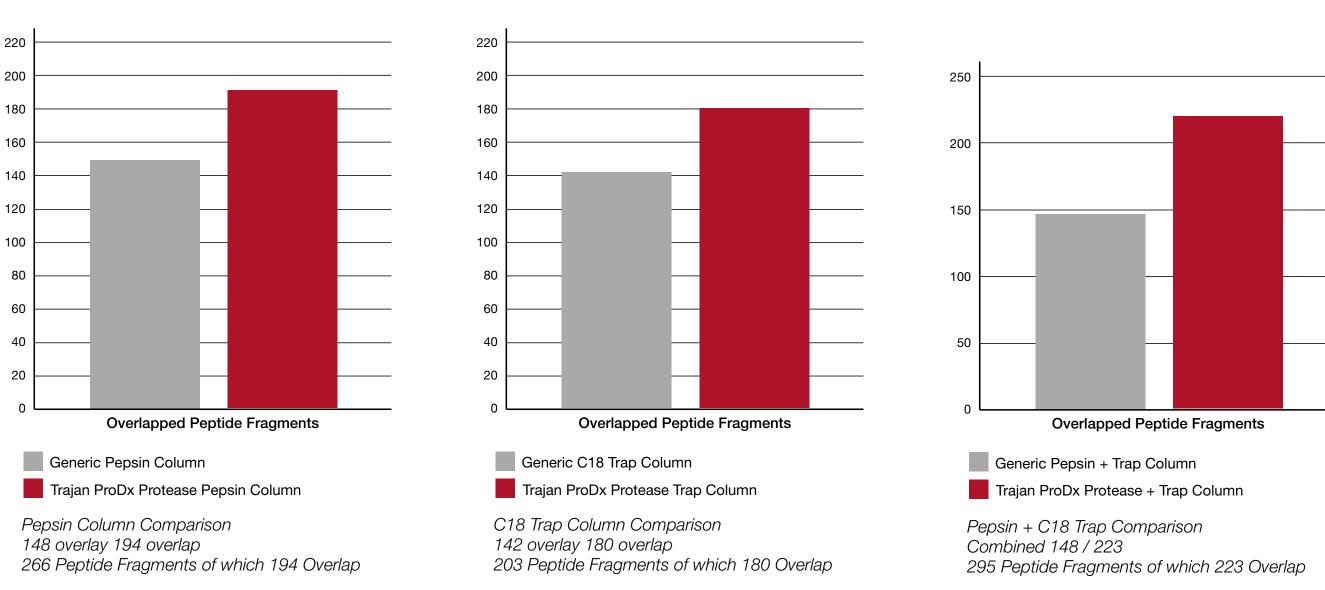
ProDx Trap columns

While many columns used for HDX trapping are not designed for use with low temperature HDX systems, ProDx Trap columns are designed to operate at 0°C and above, with consistent recovery, and demonstrate reduced back pressure at standard conditions of 200 µL/min, 100% Aqueous and 0°C, lowering instances of leakage and need for troubleshooting.

ProDx trap columns are pre-configured with high pressure rated lines for ease of use and elimination of issues with high pressure leakage that commonly occurs through relying on a pre or guard column to the system with multiple connections. ProDx Trap Columns are designed with end frits that are sized to prevent rapid blocking by intact proteins and aggregates.

To ensure confidence in your data, combine the ProDx Protease and Trap columns – the demonstrated solution for your HDX work.

More overlapping fragments = More high confidence data



Contact automation@trajanscimed.com for further information

