The Agilent StreamSelect LC/MS Solution: Increasing the Throughput of a Triple Quadrupole Mass Spectrometer

Technical Overview

Introduction

Liquid chromatography triple quadrupole mass spectrometry (LC/MS/MS) is ideally suited for the direct and rapid analysis of prepared biological samples. While analysis times can be shortened through appropriate LC method choices (gradient, flow rate, column packing, and so forth), a user is often interested in only a portion of the total data collected by an LC/MS system. Typically, there is time during each chromatographic separation where no compounds of interest are being analyzed by the mass spectrometer, leaving the instrument under-utilized for a long period of time.

The Agilent StreamSelect LC/MS Solution eliminates instrument idle time and maximizes productivity:

- Parallel HPLC streams running in staggered fashion provide up to twice the throughput of a traditional LC/MS system
- Seamless implementation using intuitive software - existing LC/MS methods can be applied with ease
- Excellent quantitative performance with proven and robust Agilent instrumentation

This technical overview illustrates the advantages of the StreamSelect LC/MS solution design for boosting lab productivity using innovative flow path design and software control to assure efficient and reliable operation. The software is capable of orchestrating the timing of all HPLC components and coordinating the analytical use of the mass spectrometer with minimal user interaction.

Agilent Technologies

Parallel system configuration for increased throughput

The standard Agilent StreamSelect LC/MS Solution (Figure 2) consists of the following components:

- Agilent 6400 Series Triple Quadrupole mass spectrometer
- Two Agilent 1260 Infinity Binary Pumps
- Two Agilent 1200 Infinity Thermostatted Column Compartments
- Stream Selection Valve (universal valve drive box with 8-position/9-port valve head)
- Agilent 1290 Infinity LC Injector HTC/HTS with a second injection port

These components comprise two parallel LC systems that are run in a staggered fashion, enabling the effluent streams to be directed to the mass spectrometer by the stream selection valve when appropriate (Figure 1).

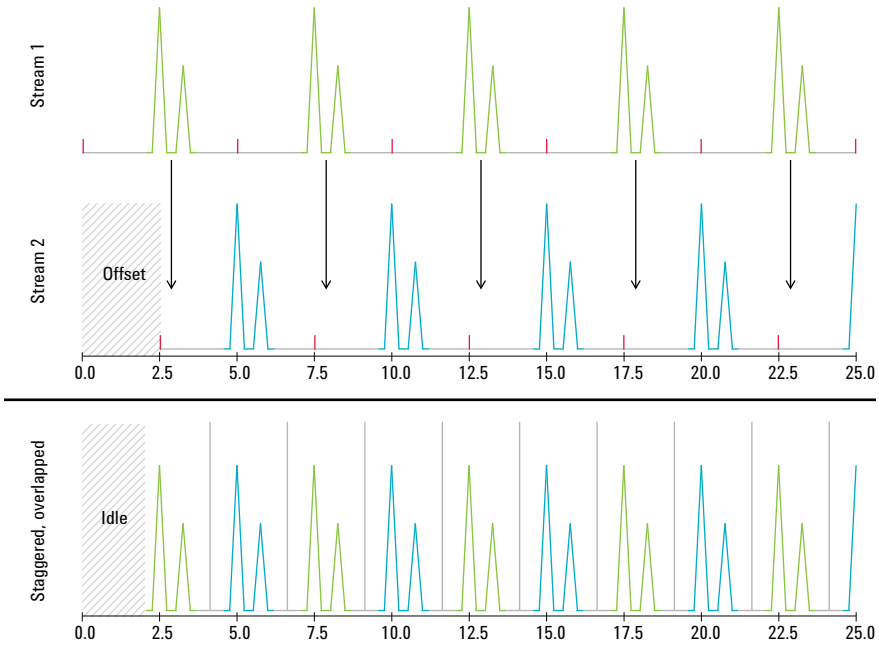

Figure 1. LC analysis of multiple samples is run on streams 1 and 2, with the analytes of interest eluting for only a portion of the total run time. The Agilent StreamSelect software orchestrates the staggered flow (stream 2 offset in run start time) to the mass spectrometer (bottom panel) through a stream selection valve. The effluent from each stream is analyzed by the mass spectrometer only when the analytes of interest are eluted.

Maximize productivity with online sample cleanup

The StreamSelect LC/MS Solution can also be configured with the option of online sample cleanup capability by adding the following components:

- Two Agilent 1260 Infinity Quaternary Pumps
- Two 2-position/6-port valves for the Agilent 1200 Infinity Thermostatted Column Compartment

The online sample cleanup configuration (Figure 3) functions in the same manner as the standard configuration (Figure 2), but also allows for more advanced chromatography. For example, samples can be loaded onto a trapping column where the analytes of interest are retained while salt and other matrix components are washed away by the first pump. This reduces the amount of matrix introduced into the mass spectrometer and automates sample preparation. Shortly before the analytes elute off of the trapping column, a valve is switched and the analytes are eluted onto an analytical column for further chromatographic separation using the second pump.

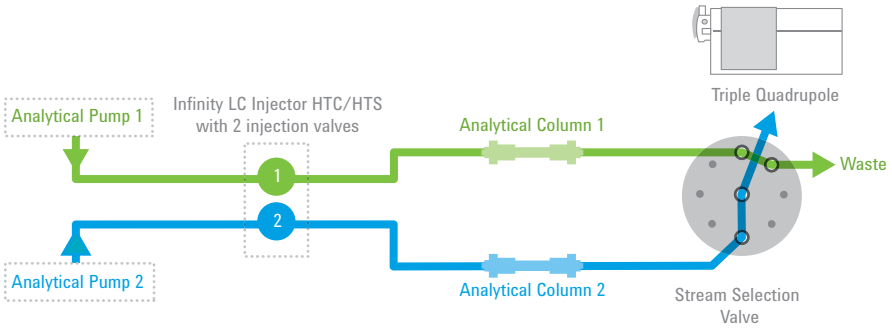

Figure 2. Agilent StreamSelect LC/MS Solution standard configuration, illustrating the use of two parallel LCs to feed the same triple quadrupole mass spectrometer.

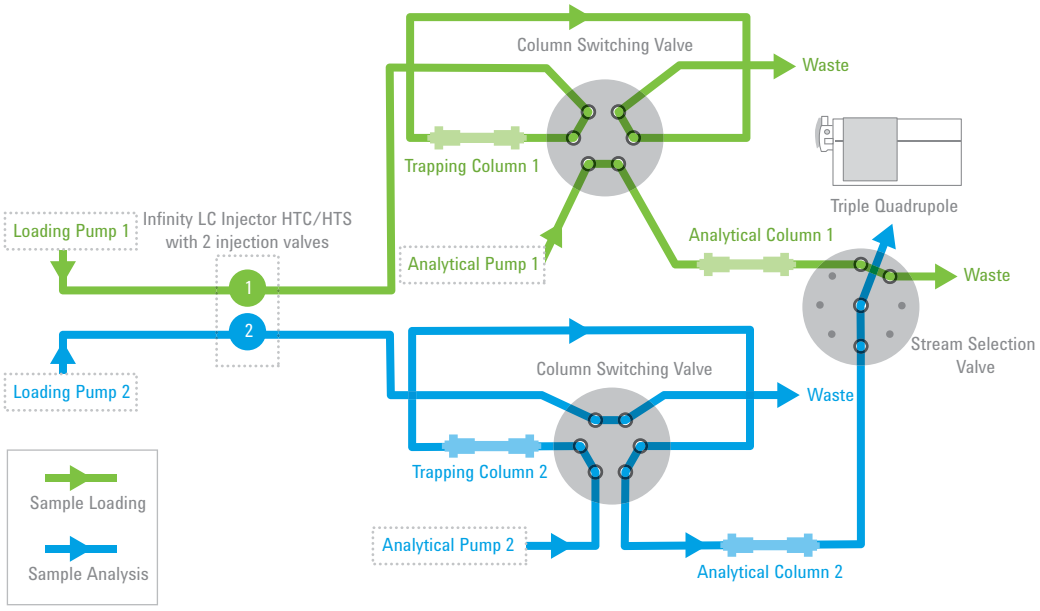

Figure 3. Agilent StreamSelect LC/MS Solution with online sample cleanup upstream of each of the two parallel analytical columns.

Intuitive software for effortless operation

Agilent MassHunter StreamSelect Software facilitates the easy and rapid creation of high throughput methods and can utilize existing LC/MS methods without need for redevelopment. After selecting an LC/MS method and defining a retention time window where analytes elute, the StreamSelect software automatically determines the necessary timings and coordinates all instrumentation. A single method is used to mirror run conditions on both streams. The Profile Editor saves application-specific settings to a profile, including templates for sample plate layouts, data file naming and system configuration (Figure 4). When loaded, a profile allows users to quickly start the instrument and submit batches.

Simple batch submission

Batch submission is a simple three-click process: identify a sample list (import file) in the Batch Submission window, specify sample location (plate assignment details), and click Finish (Figure 5). This simple process allows users to import sample lists in .csv, .txt (tab separated), .xls and .xlsx formats generated by LIS, LIMS, liquid handling robots or other sources. A batch queue displays the pending batches and their order. A large status monitor on the user interface is easily visible from a distance, allowing users to quickly monitor a system's status at a glance.

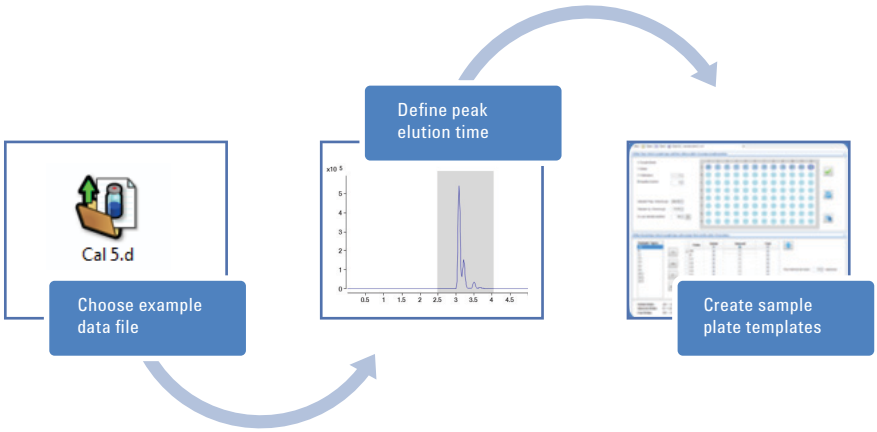

Figure 4. Using the Profile Editor for rapid method and profile setup.

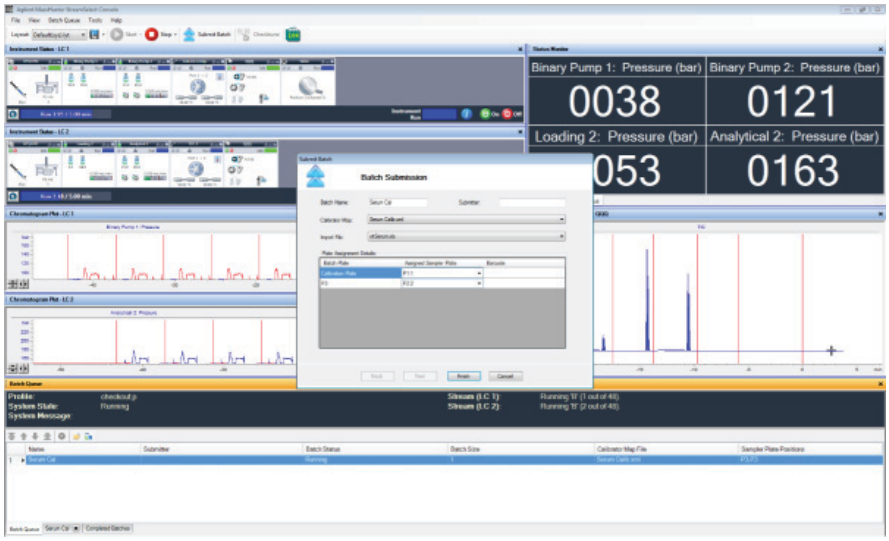

Figure 5. The user Interface of the Agilent MassHunter StreamSelect Software enables three-click batch submission and easy system monitoring.

Dynamic batch execution

The Calibrator Map function in StreamSelect software can be optionally enabled, allowing dynamic batch execution (Figure 6). Using this function, specialized sample types can be assigned to positions on the plate (double blank, blank, calibrator, QC), and open positions are available for unknown samples (Figure 6).

The Calibrator Map function can also be used to define sample bracketing, which allows the system to automatically coordinate the injection of system verification samples after a user-defined number of unknown samples. For example, an initial calibration curve (initial order) and an ending calibration check (end order) can be quickly defined (Figure 6). Run intervals for unknown samples can be defined, and system verification samples can be run in between intervals (interval order). Calibrator maps can be defined and saved as templates for quick and easy batch submission.

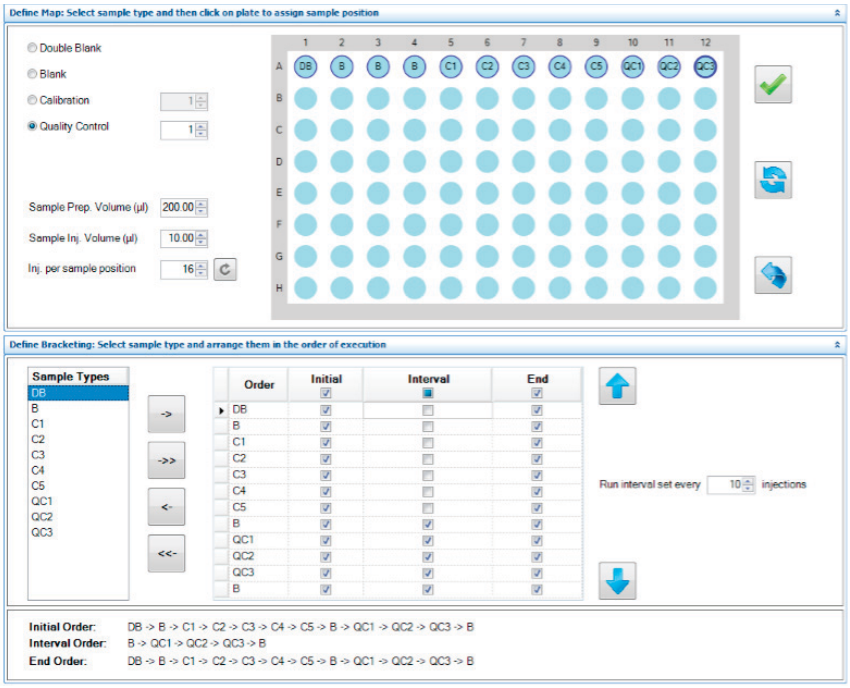

Figure 6. The Calibrator Map function enables dynamic batch execution.

Smart recovery from LC errors

MassHunter StreamSelect software orchestrates the communication between all LC and MS components of a StreamSelect LC/MS system. The software keeps track of each component's state, ensuring that a sample is run on a stream only when all components of that stream are ready. This ensures robust and reliable sample handling and eliminates the possibility of mislabeling data files.

A key feature of the software is the ability to automatically divert all samples to the available LC system in the event that one LC becomes unavailable (out of solvent, a leak, over pressure, and so forth). This feature assures continued productivity without losing any samples (Figure 7), and is available irrespective of the use of a static work list or the dynamic Calibrator Map function. However, the Calibrator Map assures that redundant analyses on specialized sample types are not performed (blanks, calibrators and QC).

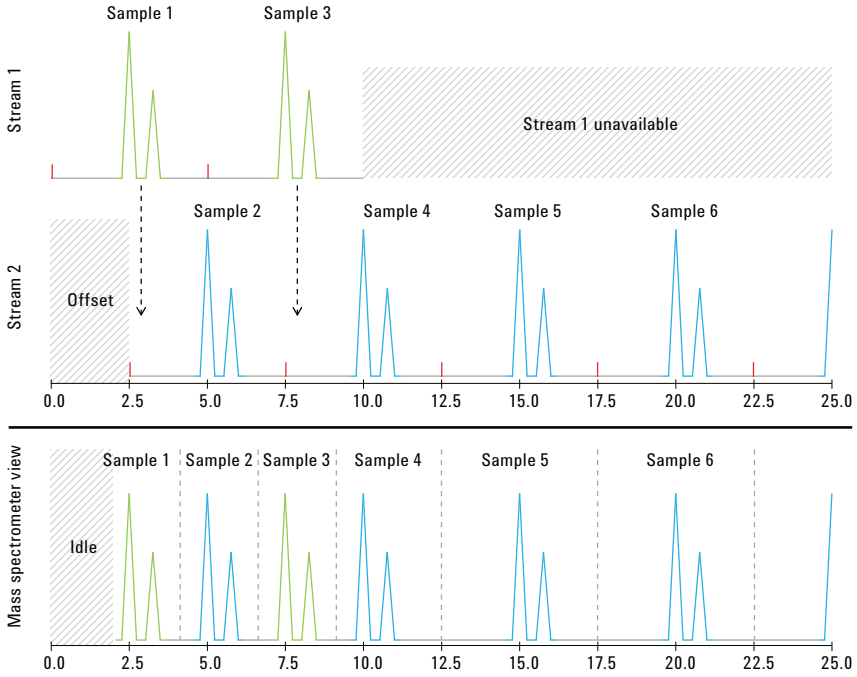

Figure 7. Schematic illustration of the ability of StreamSelect Software to automatically divert the sample stream to the functional LC in the event of an error in the other instrument. In this example, the LC running Stream 1 fails after Sample 3 is analyzed. Sample 5 and all further samples are then seamlessly re-assigned for analysis on Stream 2, assuring continued productivity.

Reliable and reproducible results

When using a multistream system, it is critical that each LC system produces equivalent results. An existing LC/MS method for the analysis of 25-OH vitamin D_2 and D_3 was implemented using the Agilent StreamSelect LC/MS Solution to demonstrate the excellent reproducibility of quantitation between two LC systems (Figure 8).

Retention time and peak area reproducibility is another important consideration for these types of systems. Figure 9 demonstrates the exceptional retention time and peak area reproducibility observed on the StreamSelect LC/MS system, using an LC/MS method for the analysis of clozapine.

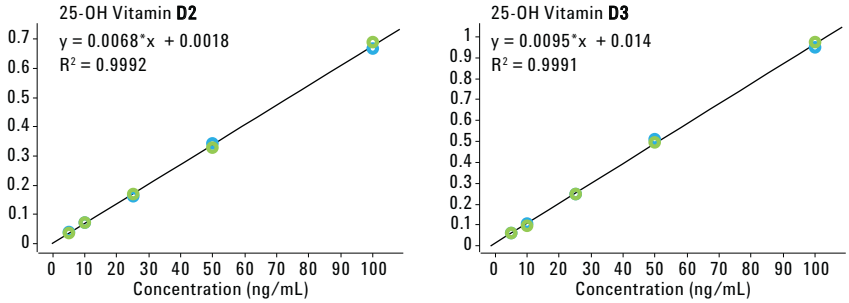

Figure 8. Equivalent quantitation for 25-OH vitamin D2 and D3 on two LC systems run in parallel: LC1 (stream 1) - green circles; LC2 (stream 2) - blue circles.

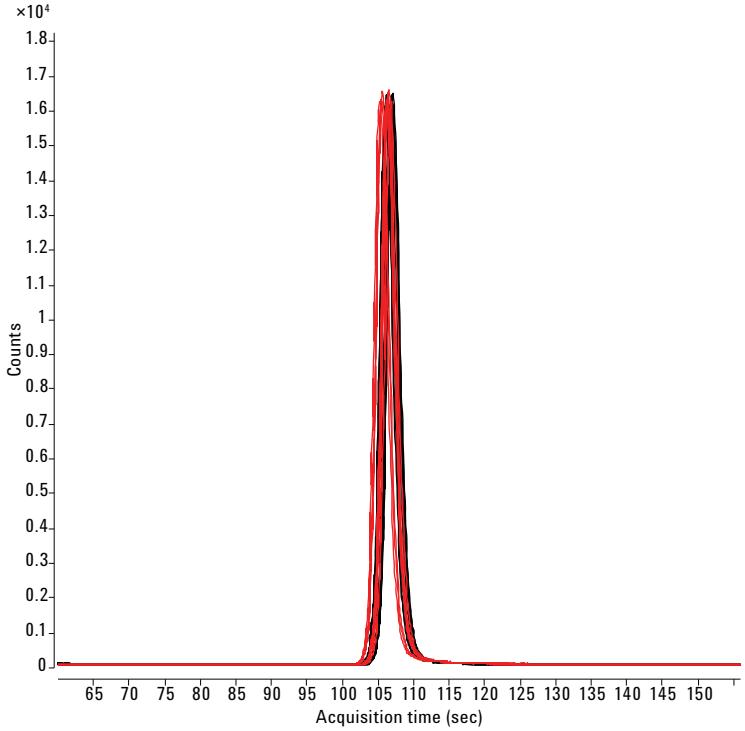

Figure 9. Injection and analysis reproducibility between LC systems: black – LC 1; red – LC 2 (N = 22).

Conclusion

The Agilent StreamSelect LC/MS Solution meets the productivity demands of high throughput labs by providing unmatched throughput, reliability and robust performance with a single triple quadrupole mass spectrometer. Sensitivity, linearity, and reproducibility are equivalent to conventional single LC systems. StreamSelect software enables rapid implementation of existing LC/MS methods without requiring any changes, and batch submission is a simple three-click process. The dynamic software control also protects against data loss if one LC should become unavailable, and prevents redundant sample analysis in the same instance.

www.agilent.com/chem/streamselect

Published in the USA, October 10, 2013
5991-3274EN

Agilent Technologies