

INTRODUCTION

Modern drug discovery involves high throughput screening processes on large numbers of samples. These samples are often produced as libraries using combinatorial chemistry. HPLC-MS is commonly used to confirm the presence and purity of potential target compounds. HPLC-MS has already proven to be an effective tool for characterizing pharmaceutical compounds, and currently is being optimized for speed in order to process the large numbers of samples that are created during the drug discovery process.

The microplate (96-, 384-, or 1536-well) is the ubiquitous standard for sample manipulation during drug discovery. Newer HPLC autosamplers provide the ability to inject directly from microplates, eliminating the need for any reformatting steps.

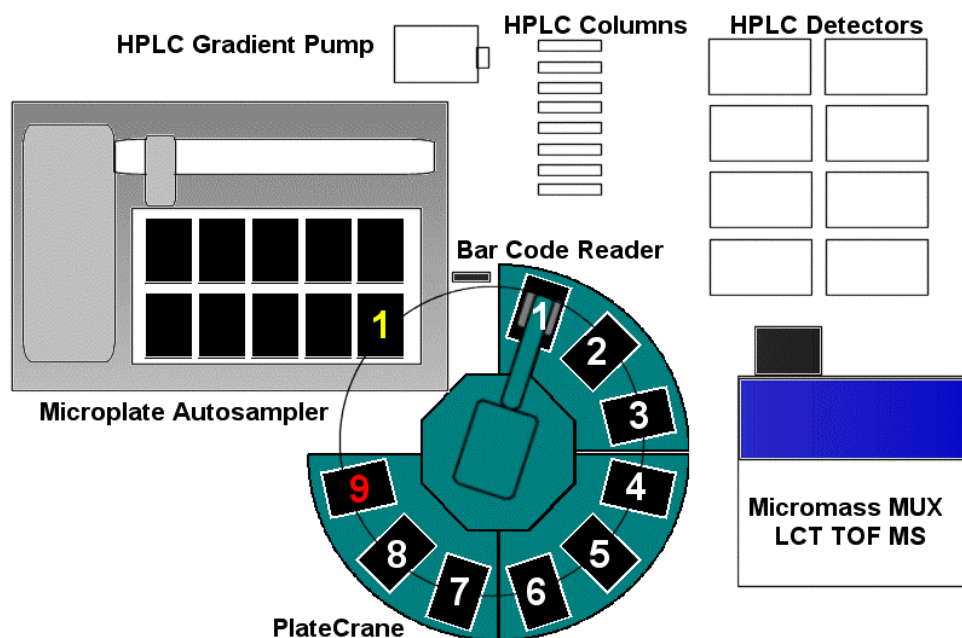
The inherent high speed capabilities of an MS system have been exploited by operating the system in a parallel fashion which allows a single MS to collect data from multiple HPLC systems simultaneously.

This bulletin describes MS/PlateLink from Hudson Control. MS/PlateLink provides the capability to build an automated robotic high throughput HPLC-MS workcell capable of parallel analysis of samples injected from microplates.

SYSTEM REQUIREMENTS

- **Hudson Control Group MS/PlateLink.** Software to track sample identification, perform database links, build mass spec input files, and oversee operation of the system. Includes Bar Code Reader.
- **Hudson Control Group PlateCrane** Pick and Place System to supply microplates from stacks to the autosampler.
- **Microplate HPLC Autosampler.** Autosampler configured with 8 injectors allows parallel injections to 8 HPLC systems.
- **HPLC Systems.** Up to 8 HPLC detection systems can be integrated into the workcell. HPLC detectors can include UV/VIS, Diode Array, Light Scattering, or other detection modes. Typically a single gradient pumping system is used with an 8-way splitter.
- **Micromass LCTTM LC-TOF-MS with MUX-technologyTM 8-channel ESI input.** Time of Flight MS platform with MUX electrospray interface which allows monitoring of data from 8 HPLC systems simultaneously. Includes **MassLynx** software.
- **Pentium Computer** to run MassLynx and MS/PlateLink software.. The computer will require one serial port for each module in the workstation. Standard PC's are easily configured with additional serial ports.

OPERATION



The layout of a typical automated HPLC-MS workcell is shown here.

The PlateCrane is the original source of the samples to be analyzed. The samples are stored in microplates within stacks surrounding the PlateCrane. This configuration shows a standard PlateCrane configured with 9 stacks. The red stack #9 is left empty to accept used plates. White stacks #1-8 can be used to store plates to be analyzed. The maximum capacity of this configuration is $8 \times 30 = 240$ standard microplates. If required, larger capacities can be configured by using higher-capacity PlateCrane models such as the XL and XT.

The microplates are moved in front of the Bar Code Reader on the way to the HPLC autosampler, allowing the plate to be identified by the MS/PlateLink software in order to initiate operations.

MS/PlateLink is based on Hudson's Total Control for Windows (TCW) software. The Main Menu shown below is seen upon first launching the software. Prior to initially operating the system functions F4 and F5 are used to set up the microplate format to be used and to calibrate the PlateCrane.

Calibration is a single-time operation that “teaches” the PlateCrane the location of the stacks, the MS plate nest, and the HPLC autosampler deck position to be used.

PlateCrane System Setup

System Setup is a single-time operation that defines the PlateCrane model that is being used as well as the plate types.

simple to program, and includes the movements of the plates and the commands to initiate the bar code reader and Micromass MS.

METHOD EDITOR

Process Sequence for Method: **MASSPEC**

Esc

[illegible]

Available Process Steps:

[illegible]

Available Positions:

NONE	
Stack1	PlateCrane
Stack2	PlateCrane
Stack3	PlateCrane
Stack4	PlateCrane
Stack5	PlateCrane
TEST	Test
MS_Nest	MicroMass
Barcode	Barcode Scanner

Associated Instruments:

Add Step

Insert Step

Delete Step

Save Method As

Save Method

Change Method

Instrument Parameters

Current Method: MASSPEC

Step No.	Position:	Current Plate:
1	Stack1	
2	barcode2	
3	MSNest	
4	Stack5	

Selected Methods: MASSPEC

PlateCrane

System is PAUSED.

Terminate Run

Press <SPACE BAR> to UNPAUSE System

Esc Exit

Once a method has been written and stored, it is initiated by selecting F1 Run Method from the Main Menu. The Status Screen shown here will monitor the operations as the plates are moved from the PlateCrane to the HPLC-MS system for analysis.

System Operation

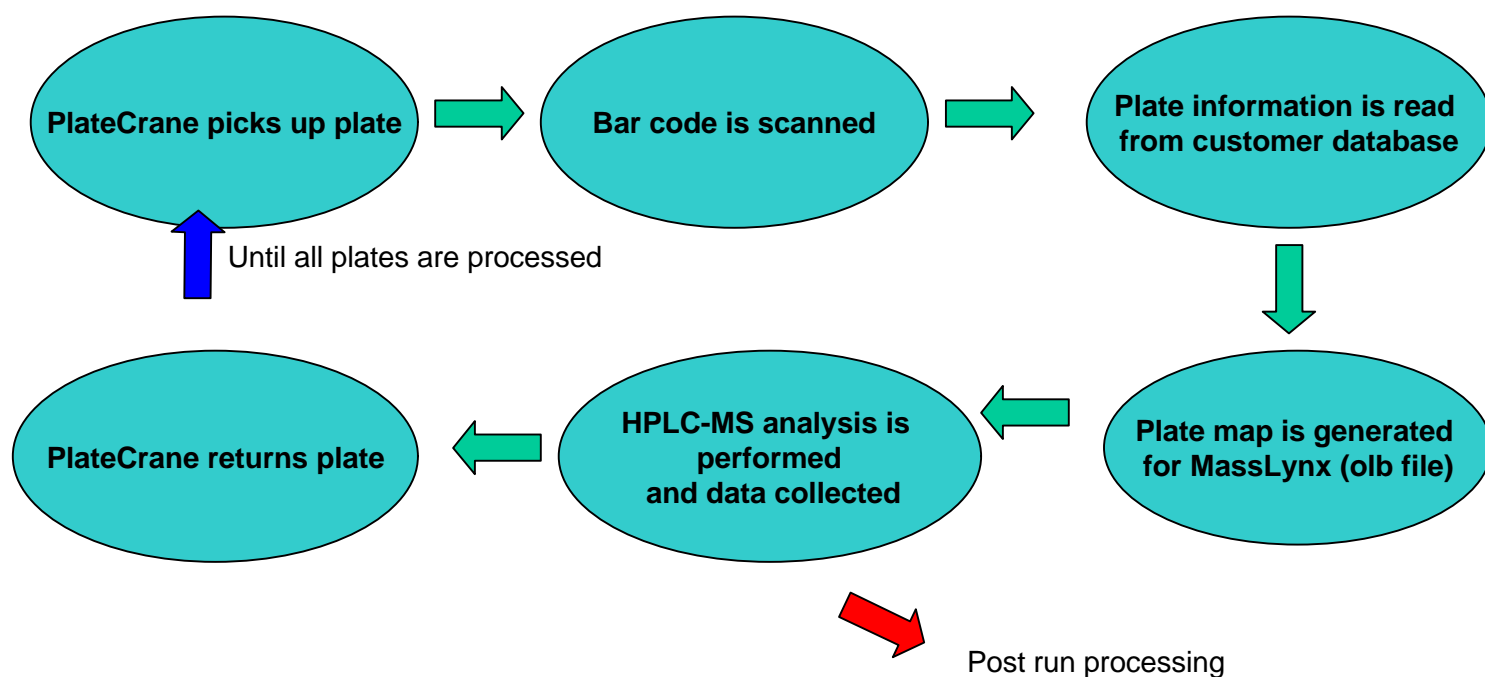
Hudson's TCW acts as supervisory control for the workcell. The PlateCrane, bar code reader, and MS are connected to the computer via their serial ports. The HPLC components are connected to the MS. The PlateCrane initiates operation of the system by retrieving a plate, performing a bar code read, placing the plate on the autosampler, and then initiating the MS to start. The MS in turn starts the HPLC system and performs data collection.

Hudson MS/PlateLink provides advanced software capabilities that allow the entire high throughput HPLC-MS operation to be automated AND seamlessly tracked and integrated with the end user's database. This is accomplished for each end user by the development of a custom link to their specific database, including Oracle, SQL, or any other standard database format. The user's database contains the critical information that is tied to the samples in each microplate, such as sample identification number, well positions of individual samples, sample origin information, and any other pertinent information that is necessary to track with each sample. As each microplate is retrieved from its stack for processing, its bar code label is read, and the software accesses the database to retrieve the sample information specific to that plate.

Once the plate has been identified and its information loaded from the database, MS/PlateLink creates an "analysis plate map" in a format that is recognizable by the MassLynx software. This is a text file with the extension ".olb". This file is placed within a specific subdirectory that the MassLynx software is continually scanning. When a new .olb file is detected, it is loaded by MassLynx and the HPLC-MS system is started.

The MassLynx software collects and stores the data from the MUX MS and the HPLC detectors. At the end of the run, the process is completed for all of the microplates that have been stored in the PlateCrane stacks.

The entire operation is diagrammatically illustrated below.



CONCLUSION

Hudson Control's longstanding experience with the development of integration software and hardware has been used to develop MS/PlateLink, a unique product that will tie together and automate a high throughput parallel HPLC-MS system.

REFERENCES

"System Aids Analysis of Compound Collections", S. Chatwin; D. Chow; E. Maliski; W. Talen; Y. Zhao; and D. Semin. Drug Discovery & Development, May 2001, 50-52. Describes a system using MS/PlateLink that has been implemented at Amgen.

"High Throughput Parallel LC/MS Analysis of Multiple Liquid Streams for Drug Discovery Applications using the 4-Way MUX-technology™ Electrospray Interface on LCT", A. Sage and K. Giles. Micromass® Application Note 241. Describes the operation of the HPLC-MS itself in more detail, including the plumbing configuration of the HPLC system and the MUX-MS operation, data collection, and data analysis.