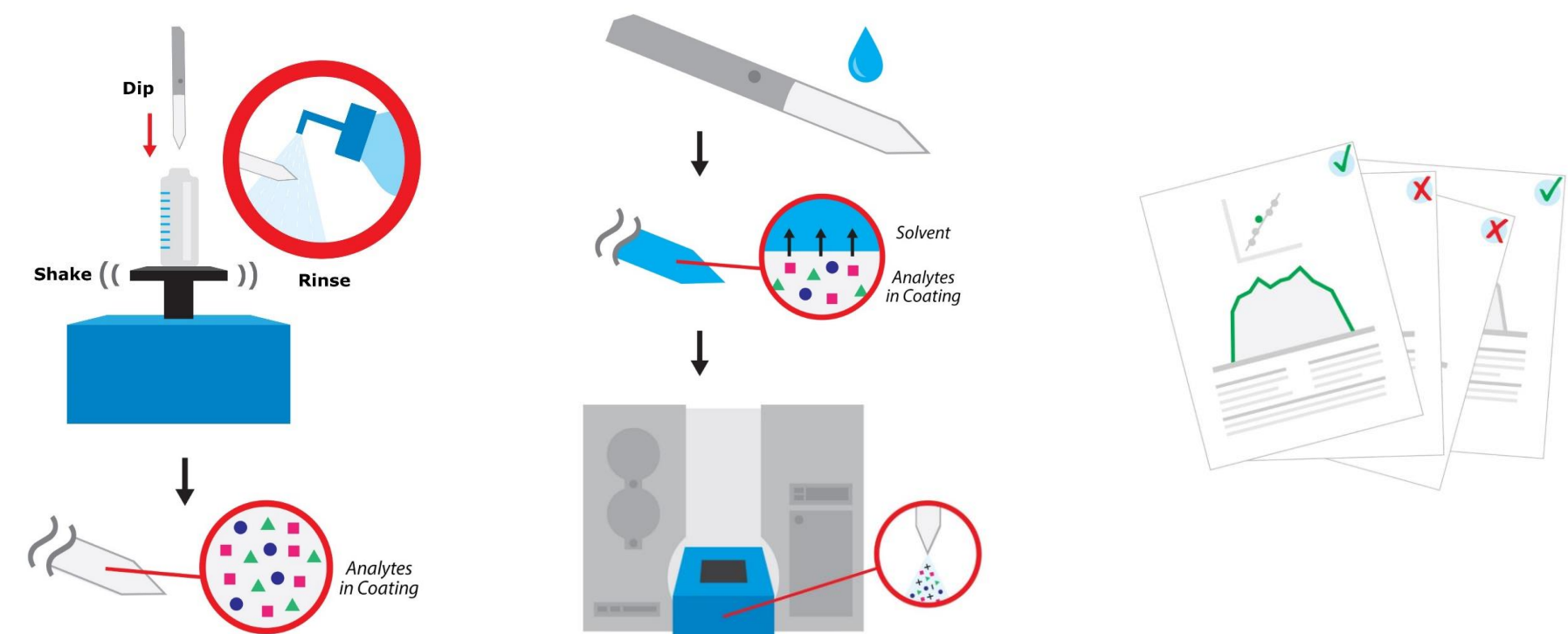


Building trust in direct-to-MS technologies: a fully automated and reliable interface for operation of Coated Blade Spray devices



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How does CBS work: the journey from the sample to the MS



Sample Preparation

- ✓ Blade is immersed in a vial/vessel containing the sample of interest.
- ✓ Quick agitation (<5 min) at 1000 rpm
- ✓ Then, the coating is rapidly rinsed with water (<10 s).

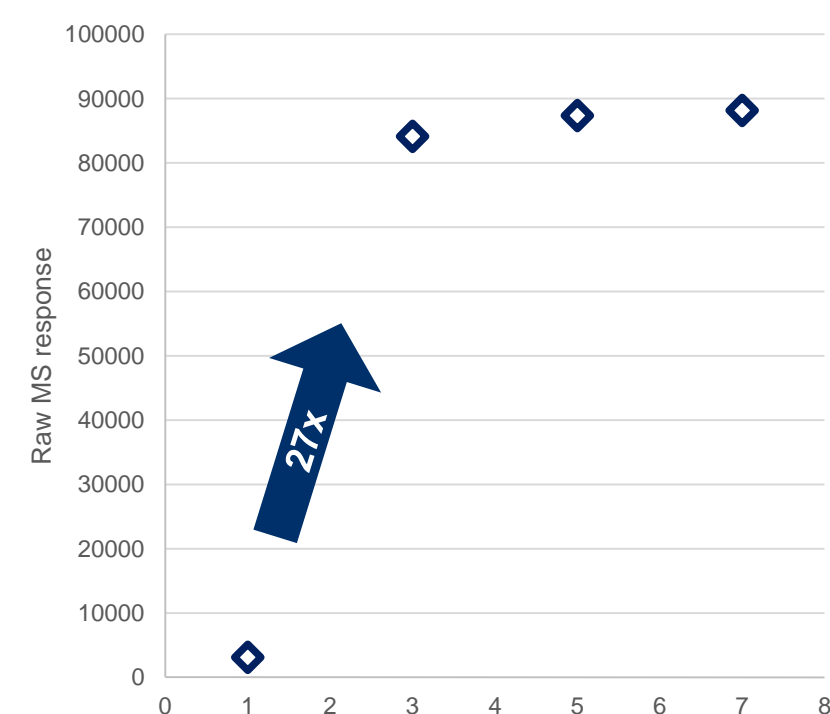
Elution/Ionization

- ✓ CBS device is placed in the front-end of the MS system for analysis.
- ✓ A droplet of solvent is added to the coated area of the device and,
- ✓ High-voltage is applied to the non-coated area of the blade to generate an electrospray from the tip of the blade.

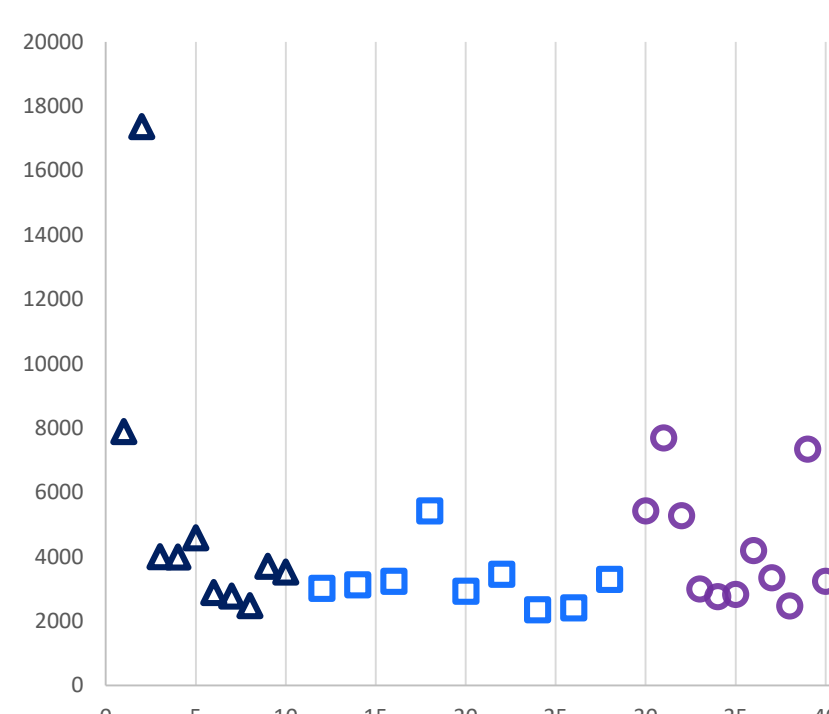
Quantitative Analysis

- ✓ Ratio of analyte signal (area under the curve) to the one of the internal standard is calculated.
- ✓ Preferably, isotopic label of the analyte of interest is used for correcting potential variability in the instrumental response.

Total CBS solution minimizes/eliminates instrumental carry-over



Blank signals collected after intercalated injections of samples containing 5000 ng/mL of MTX. As can be seen, there is an increment of 27 times between injection number 1 and injection number 2



Blank signals obtained before (blue triangles), during (light blue squares), and after (purple circles) injection of 10 intercalated highly concentrated samples of methotrexate at 5000 ng/mL

Statement of the Problem

Although several direct-to-MS technologies have been developed over the past 3 decades, their massive acceptance has been halted by reduced performance in at least one of the following areas: reproducibility, sensitivity/selectivity, throughput, or instrumental carry-over. Herein we present the first fully automated MS interface for one at a time reliable operation of Coated Blade Spray (CBS) devices. This work not only focuses on demonstrating the screening and quantitation capabilities of CBS-MS/MS towards a set of target analytes of clinical relevance, but also in building trust in this sans-chromatography technology that aims to eventually substitute tests used in the clinical laboratory such as immunoassay and liquid chromatography.

Methods & Materials

CBS coated with HLB particles were manufactured by Restek Corporation. Instrumental analysis via tandem mass spectrometry were performed on either a Thermo TSQ Altis or a SCIEX 4500. A fully automated interface for CBS compatible with Thermo and SCIEX instruments was also developed by Restek Corporation. The interface offers the following features:

I/O communication between interface and MS: starts sample acquisition (similar to LC autosampler) and waits for ready signal from MS

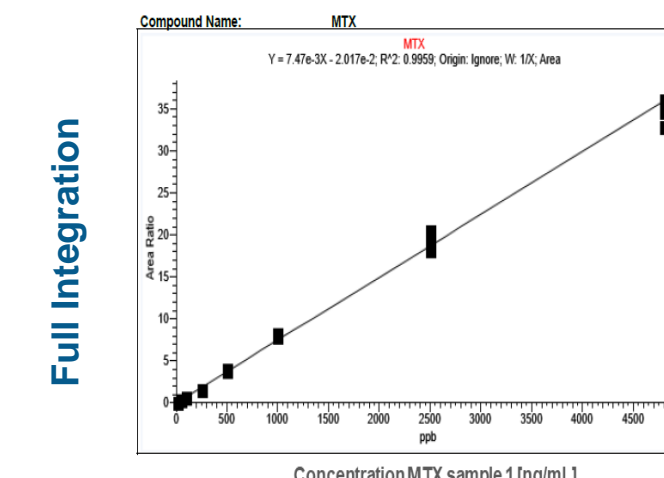
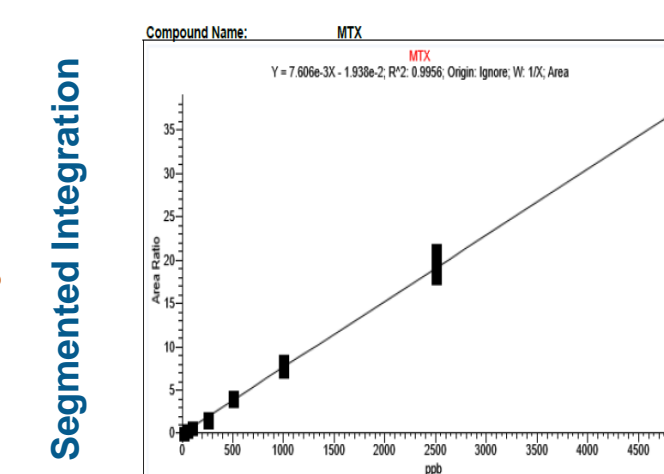
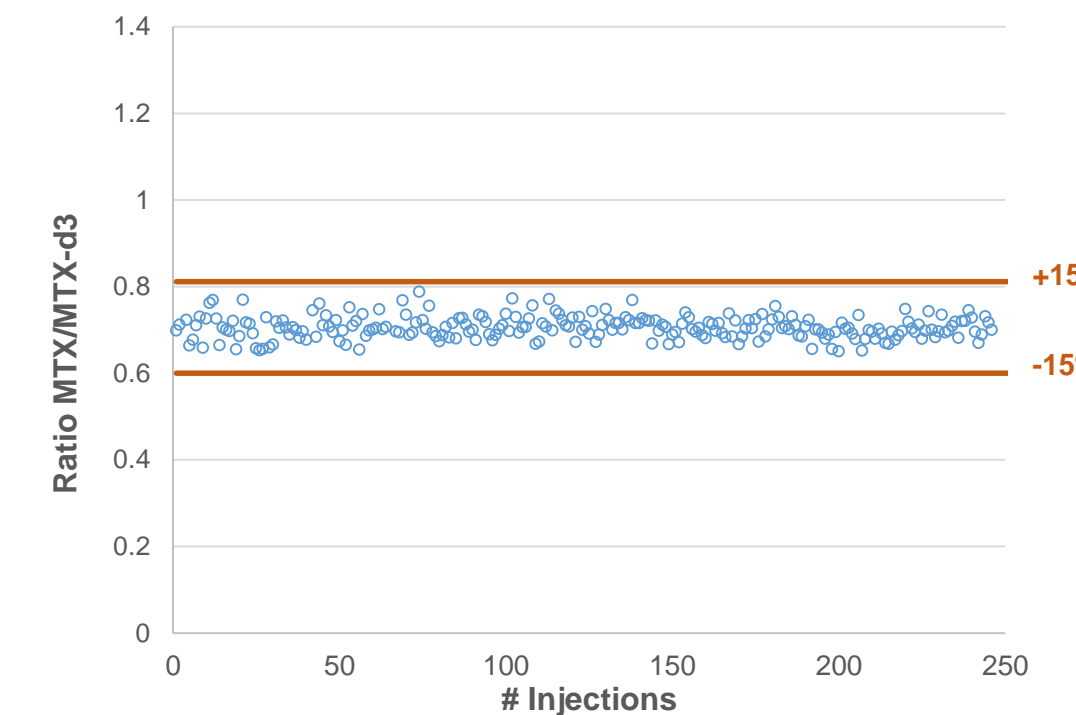
Interface mounting consistent w/ manufacturer specifications. Safety enclosure: positive pressure and interlocked with HV



Automated and timed workflow: CBS accurate positioning, coating wetting, MS start trigger, timing elution, timing high voltage application, and reset for new acquisition. Injections were made every 20s

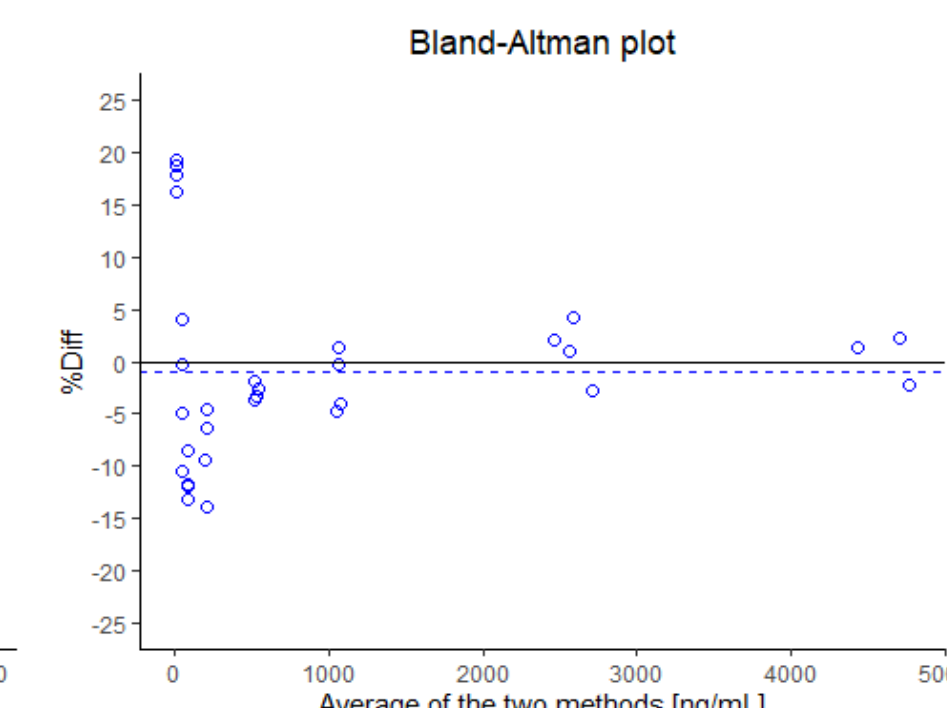
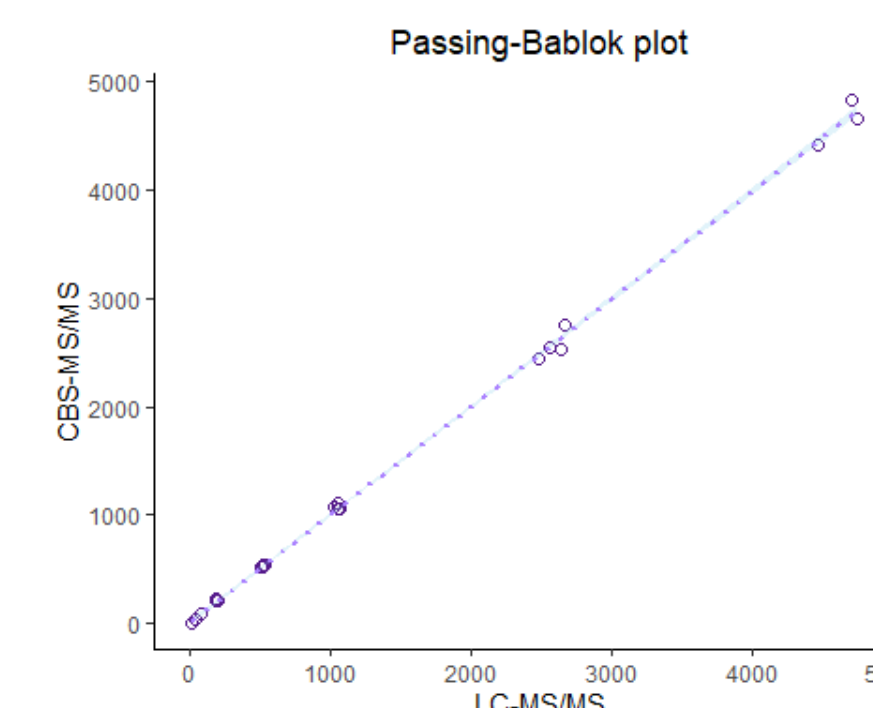
Automated inlet cleaning system between each run to ensure no chemical memory effects or inlet contamination

Coated Blade Spray is as Reliable as LC-MS/MS approach



250 consecutive injections (CV < 3.7% with IS correction) can be completed by an analyst standing in front of the MS in less than 2h and without having to replace/clean the mass spectrometer inlet (left). No statistical differences were observed when comparing full versus segmented area integration (right).

Coated Blade Spray delivers a comparable selectivity to LC-MS/MS



	EST	LCI	UCI	Criteria
Intercept	3.950441	-2.4370767	11.995562	Include 0 (pass)
Slope	1.019019	0.9853865	1.033811	Include 1 (pass)

Excellent agreement between CBS-MS/MS and the confirmatory test via LC-MS/MS (i.e. compared via Passing-Bablok regression) was achieved for MTX when extracted from serum samples in a range between 5-1000 ppm