

A 3D molecular model of a protein, likely myoglobin, is shown in the upper half of the page. The protein is rendered in a vibrant orange-red color with a textured, almost crystalline surface. It has a complex, folded structure with several deep pockets and loops. The background is a dark blue gradient with some lighter blue and yellow highlights, suggesting a microscopic or laboratory environment.

## DIRECT ANALYSIS OF INTACT PROTEINS USING THE ADVION TOUCH EXPRESS™ OPEN PORT SAMPLING INTERFACE (OPSI) WITH THE ADVION expression<sup>L</sup> COMPACT MASS SPECTROMETER

### APPLICATION NOTE

**Mass Spec:** expression CMS  
**Sampling:** Touch Express OPSI

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In this application note, the Advion Touch Express Open Port Sampling Interface (OPSI) is coupled with the Advion expression<sup>L</sup> Compact Mass Spectrometer (CMS) to demonstrate fast, benchtop analysis of intact proteins – 2  $\mu$ L of myoglobin (horse) as well as 2  $\mu$ L cytochrome C (horse).

## INTRODUCTION

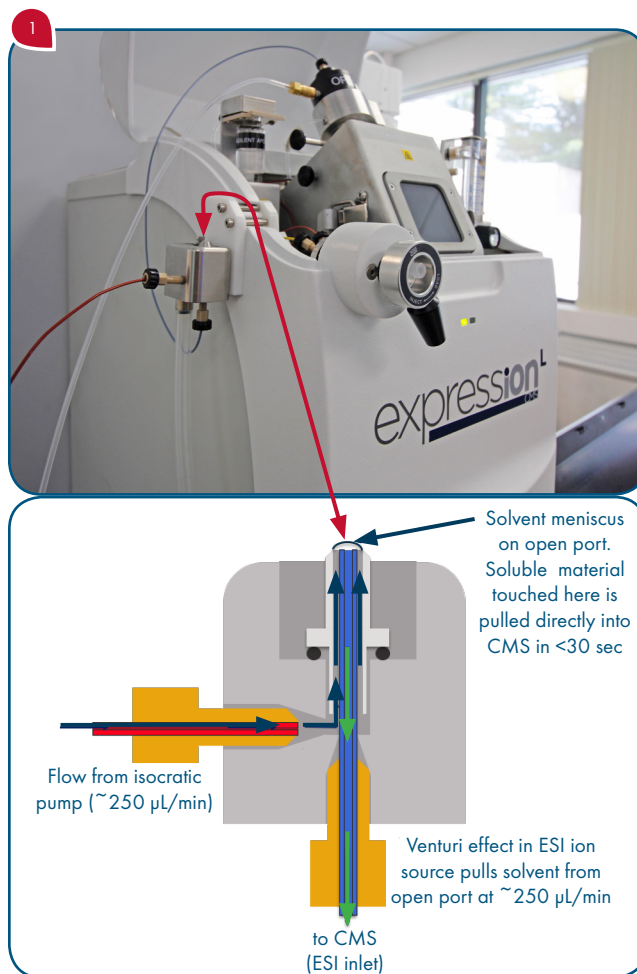
The Touch Express OPSI is a new sampling device for mass spec analysis using a technique developed by Gary Van Berkel and Vilmos Kertesz of Oak Ridge National Laboratory<sup>[1]</sup>. This fast, versatile sample inlet paired with the Advion expression CMS provides simple molecular weight determination of surfaces, solids and liquids in < 30 sec.

The OPSI incorporates an open port of continuous, low-volume solvent, flowing directly into the electrospray ion source of the mass spectrometer. Solvent is delivered to the open port sampling area through the inner of a pair of concentric tubes. The solvent forms a meniscus at the open port before being drawn down the outer tube into the ion source of the mass spectrometer under the Venturi effect of the nebulization gas in the ion source. Analysis is carried out by simply touching a sample to this open port.

In this application note, the OPSI is coupled with the CMS to demonstrate fast, benchtop analysis of intact proteins – 2  $\mu$ L of myoglobin (horse) as well as 2  $\mu$ L cytochrome C (horse).

## METHODS

At a concentration of 1 mg/mL in 10 mM ammonium acetate, myoglobin (horse) and cytochrome C (horse) were used as examples and deposited directly at the open port by pipette tip. Analysis is carried out with positive-ion ESI using acetonitrile with 0.1% formic acid as the carrier solvent at a flow rate of 250  $\mu$ L/min.



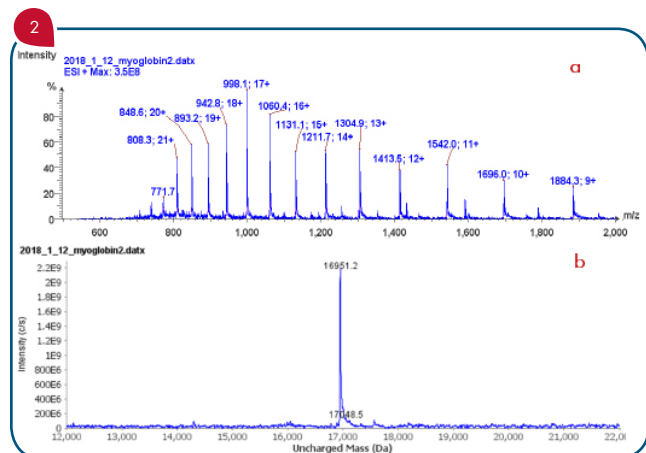
**Figure 1:** Experimental setup and schematic of CMS and OPSI.

## RESULTS AND DISCUSSION

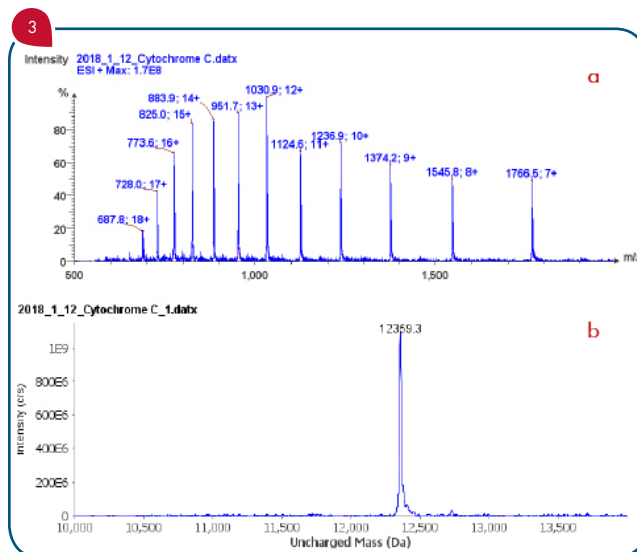
Following sample analysis, a charge deconvolution feature within Advion Data Express software automatically calculates the uncharged protein mass from the characteristic multiply-charged spectrum envelope obtained from ESI of biomolecules. The full mass spectra and deconvolved, uncharged mass for myoglobin and cytochrome C are shown in Figures 3 and 4, respectively.

A total of 14 multiply charged ions of myoglobin were detected with charge state ranging from +9 to +22 (Figure 3A). The uncharged mass of myoglobin is calculated to be 16,951.2 (Figure 3B), within 1 Da of the theoretical mass, 16,950.5.

Similarly, for cytochrome C, a total of 12 multiply charged ions were detected with charges ranging from +7 to +18 in the  $m/z$  range from 700 to 1,800 Da (Figure 4A). The uncharged mass of cytochrome C is calculated to be 12,359.3 (Figure 4B), within 1 Da of the theoretical mass of 12,360.0.



**Figure 2:** (A) Mass spectra and (B) deconvolved, uncharged mass for myoglobin.



**Figure 3:** (A) Mass spectra and (B) deconvolved, uncharged mass for cytochrome C.

## CONCLUSION

The Advion Touch Express OPSI provides:

- Simple, robust operation – no mechanical parts
- Fast analysis – results in < 30 seconds
- Direct sampling – just touch and assay
- No carryover or contamination – the port is continuously self-cleaning

### REFERENCES

[1] Van Berkel G., Vilmos K.; An open port sampling interface for liquid introduction atmospheric pressure ionization mass spectrometry; Rapid Comm. Mass Spec., 2015 29(19) 1749-1756