ESI-MS detection of mAb Tryptic Digest standard with a small-footprint ion trap mass spectrometer.

Overview

○The MT Explorer 30 (MTE30) redefines mass spectrometry with a small-footprint design, enabling high-performance analysis both in and outside of traditional lab settings. Its tandem MS capabilities provide rapid confirmatory detection for diverse applications, from forensic science to peptide analysis.

○ The MT Explorer 30 portable mass spectrometer effectively detected tryptic peptides derived from an mAb antibody sample, showcasing its capability for peptide analysis and by extension, protein analysis, highlighting the potential applications for analysis of protein digests, including antibody characterization, biopharmaceutical quality control, and point-of-care diagnostics.

Introduction

○ A tryptic digest of an antibody cleaves at lysine and arginine, producing peptide fragments that readily protonate during ionization. Peptide detection typically relies on electrospray ionization (ESI) coupled with a high-end mass spectrometer, but these systems are costly, require expertise, and are not field portable.

○ A miniature mass spectrometer with ESI capabilities provides an accessible alternative, enabling onsite peptide analysis with reduced resource requirements. Here, ESI-MS analysis of an mAb Tryptic Digest standard demonstrates effective peptide detection.

Methods

Sample Preparation:

mAb Tryptic Digest standard (Waters, USA) stock was diluted in a water and acetonitrile solution with 0.1% Formic Acid 10x for a concentration of 40µg/mL. ESI-MS analysis performed on MT Explorer 30 in positive ion mode:

Table 1: MT Explorer Parameters

General Parameters	
ESI Flow Rate:	0.8 µL/min
Spray voltage:	3 kV
Mass range:	100-1200
Injection Time:	10-20
Acquisition time:	1.5 min
Capillary temp:	210°C

Results and Discussion

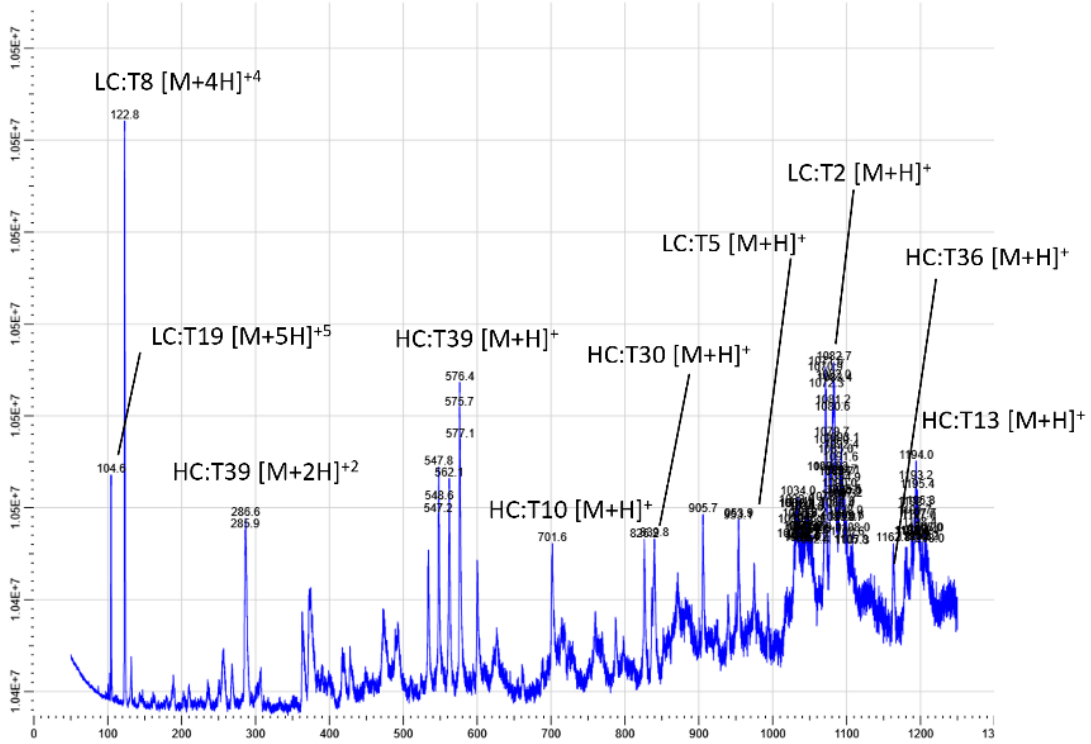

Fig. 2 ChromExplorer ESI-MS raw spectrum with peak annotations (full range 35-1250) NIST mAb Trypsin Digest.

a.
DIQMTQSPSTLSASVGDRVTITCSASSRVGYMHWY
QQKPGKAPKLLIYDTSKLASGVPSRFSGSGSGTEFT
LTISSLQPDDFATYYCFQGSGYPFTFGGGTKVEIKRT
VAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV
QWKVDNALQSGNSQESVTEQDSKDSTYSLSSTLTL
SKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

b.
pQVTLRESGPALVKPTQTLTLTCTFSGFSLSTAGMS
VGWIRQPPGKALEWLADIWWDDKKHYNPSLKDRLTI
SKDTSKNQVVLKVTNMDPADTATYYCARDMIFNFYF
DVWGQGTTVTVSSASTKGPSVFPLAPSSKSTSGGT
AALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQ
SSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKV
DKRVEPKSCDKTHTCPPCPAPELLGGPSVFLFPPKP
KDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVE
VHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKE
YKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSR
EEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENN
YKTTPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSC
SVMHEALHNHYTQKSLSLSPG

Fig.3 Visual representation of sequence detection by the MT Explorer 30. Blue shading represents the peptide sequences detected. a. Light Chain (LC) Sequence Coverage (singly, doubly, triply charged peptides – 65.7%) b. Heavy Chain (HC) Sequence Coverage - (singly, doubly, triply charged peptides – 42.3%)

- The MT Explorer 30 successfully detected ~50% of the total antibody sequence with minimal sample preparation or instrument optimization. This breakthrough provides a promising avenue for accessible, point-of-care and field proteomics.
- Further optimization and development of sample preparation methods, coupled with advancements in instrumentation, hold promise for expanding the applications of the MTE30 in protein analysis and biopharmaceutical research.