Identification of Phosphorylation Sites by means of ESI hybrid RF/DC quadrupole-linear ion trap mass spectrometry

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ABSTRACT

Research was conducted on an Applied Biosystems/MDS Sciex Q TRAP™, a hybrid RF/DC quadrupole-linear ion trap mass spectrometer with ESI turbo spray source. QqQ Mass Ion trap. The instrument is based on a triple quadrupole ion path and is capable of all the conventional tandem quadrupole scans as well as several high sensitivity on-trap mass spectrometer scans using the first quadrupole as a pre-selected ion trap. LC-MS/MS experiments were performed using a ZORBAX 300SB C18, 2.1 mm × 150 mm column and a gradient from 5% to 95% acetonitrile + 0.1% formic acid in 2 minutes. To further improve candidate selectivity, the IDA feature also allows the use of highly specific scan types of triple quadrupole for the survey scan, such as precursor ion scan and neutral loss scan.

RESULTS

The phosphopeptides are identified using conventional precursor ion scan or neutral loss scan as the survey scan. A conformational scan in the positive mode with enhanced resolution scan is conducted for the charge state and precursor mass assignments. An important consequence is that these variables determine the optimized Collision Energy setting for MS/MS experiments. Consequently, low collision energies enhanced product ion spectra of hybrid RF/DC quadrupole-linear ion trap mass spectrometry in an automated single step LC-MS run can be applied in the neutral loss scan by identifying the facile loss of 80 Da and 98 Da in which are neutral loss of HPO and H3PO4.

REFERENCES


TRADEMARKS/LICENSING

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MATERIALS AND METHODS

Table 2 show a comparison of selectively of phosphopeptides detection for (a) EMS and (b) NL of 49 as the survey scan. It is apparent that the phosphopeptides detection and sequencing can be performed without polarity switching.

CONCLUSIONS

Phosphopeptides in a complex mixture are identified using conventional triple quadrupole neutral loss or precursor ion scan. The partial sequences and sites of phosphorylation of the identified phosphopeptides are convincingly built by low collision energies enhanced product ion spectra of Hybrid RF/DC quadrupole-linear ion trap mass spectrometry in an automated single step LC-MS run can be applied in the neutral loss scan by identifying the facile loss of 80 Da and 98 Da in which are neutral loss of HPO and H3PO4.