ABSTRACT

The MALDI TOFTOF™ mass spectrometer provides a flexible platform for the analysis of the fragmentation of deprotonated peptides. Previous work focusing on the fragmentation of protonated peptides has demonstrated that fragments observed in the TOFTOF™ derive from two distinct processes: first, metastable fragmentation of ions that are formed with high internal energy and hence dissociate on the time scale of detection; and second, collision induced dissociation (CID) promoted by high energy collisions (lab-frame kinetic energies ranging from 0.5 to 2.5 keV) between the peptide and a neutral gas. These distinct processes form separate categories of fragment ions that can be differentiated from each other by recording spectra under identical conditions with the exception of the presence (or absence) of gas in the collision cell. If there is no gas in the collision cell, then all fragments will be formed equally with high internal energy content, yet highly superior in quality to those observed using the post source decay (PSD) technique. If gas is added to the collision cell, the resultant spectra are a superposition of the ions formed both in the collision cell and the TOFTOF™ providing a background of expectation on standard peptides which can be extended to real systems where the nature of the peptide demands the use of negative ion mode.

RESULTS

A. Metastable Fragmentation - Negative Ions Versus Positive Ions

When ions are selected in the collision cell, the fragment ions will derive exclusively from metastable decay of "hot" ions and will be observed in low intensity under conditions mass spectra in which low energy fragmentation predominates. The work herein presented extends and complements the previously reported studies on metastable fragmentation of deprotonated peptides, a description of the fragmentation patterns of proteolytic fragments (e.g. y fragments) formed in metastable decay. Figure 1 shows the product ions observed when Glu1 fibrinopeptide B is selected as the collision cell. As is evident in Figure 1, the fragment ions are the consequence of fragment ions, each of which is a product ion series which will be observed. As has been mentioned elsewhere, the predominance of the y series in negative ion mode can lead to its utility in filling holes in the sequence of a protonated peptide.

B. Collision Induced Dissociation - Observation of Higher Energy Processes

When gas is added to the collision cell, a wealth of new fragment ions will result, resulting from the collision of the selected parent ion with a neutral gas molecule. As is evident in Figure 2, the fragment ions are the consequence of fragment ions, each of which is a product ion series which will be observed. As has been mentioned elsewhere, the predominance of the y series in negative ion mode can lead to its utility in filling holes in the sequence of a protonated peptide.

CONCLUSIONS

Laboratory frame collision energies are defined by the potential difference between the first acceleration source and the floating collision cell. To change the collision energy, the source conditions are kept constant and the potential of the collision cell is altered. The polarity of the spectrometer is under software control and can be rapidly switched between positive and negative ion modes.

REFERENCES