THE USE OF ION-ION CHEMISTRY IN AN ELECTROSPRAY ION TRAP MASS SPECTROMETER FOR THE DETECTION AND IDENTIFICATION OF CHEMICAL/BIOLOGICAL THREATS

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In this presentation we discuss recent progress in the development of ion/ion reaction ion trap mass spectrometry for the detection of biological warfare threats. The key to this detection technology is the rapid separation and identification of protein biomarkers from bacteria, viruses, and toxins using a novel ion chemistry approach. Electrospray is widely accepted as a method for ionizing large proteins from solution. The technique is characterized by the formation of a distribution of multiply charged ionic species. While the MS/MS dissociation of these ions is facilitated by the multiple charge states, determination of the molecular weight (especially in the presence of a complex matrix) is problematic. The ability to reduce the ion charge state in the mass analyzer using ion-ion chemistry allows the determination of molecular weight and the assignment of charge state to the various ion signals. When combined with protein sequence information from the MS/MS analysis, the technique allows the rapid identification of these proteins (and thereby the host organism if the protein is unique) using existing protein databases. The combination of the ion storage capabilities in an ion trap with the ion selection and charge state manipulation allows the isolation of target proteins within the ion trap analyzer, thus minimizing the need for extensive "front-end" sample cleanup or pre-analysis separations. In addition, since this approach can use existing databases to identify proteins, it offers the exciting prospect for the *a priori* identification of unknowns of biological origin.

To demonstrate the broad applicability of this technique to biological threats, data will be presented on the analysis of proteins from viruses, toxins and bacteria. A survey analysis of crude protein extracts from E. Coli, and the simulant Erwinia herbicola are presented, along with data generated from ovalbumin. Additionally, we demonstrate that the viral coat protein of (the Dugway Proving Ground) simulant bacteriophage MS2 can be analyzed with minimal sample preparation directly from E. Coli lysates. By performing tandem mass spectrometry and subsequent ion/ion reactions on a series of MS2 precursor ions derived from electrospray ionization, sequential fragmentation is obtained where the mass differences can be correlated with residual amino acid mass to generate a section of primary sequence information called a “sequence tag”. Sequence tags can subsequently be used to search protein databases to unequivocally identify the protein of interest. We discuss the advantages and disadvantages of current database search routines, and describe the development of a new search algorithm designed to work exclusively from data derived from fragmentation rules of intact proteins.

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