Identification of Bis-aryl hydrazine crosslinked peptides by Ultraviolet Photodissociation Mass Spectrometry

Myles W. Gardner, Jennifer S. Brodbelt

The University of Texas at Austin, Department of Chemical and Biochemistry, Austin, TX 78712

Overview

Purpose

To identify bis-aryl hydrazine crosslinked peptides from unmodified and dead-end modified peptides by using tandem mass spectrometry and photodissociation techniques.

Methods

Unmodified and bis-aryl hydrazine crosslinked peptides were generated from a melittin (Ac-AKAAAAR + Ac) complex and irradiated with ultraviolet light (Figure 1). The resulting spectra were analyzed with ThermoFinnigan’s Xcalibur software. Crosslinked peptides were identified by comparing their mass spectra with those of known compounds.

Results

Screening of Bis-aryl Hydrazine Crosslinked Peptides using UV-MS

Several model peptides were used to test 100 mM 5PF and subsequently treated with either irradiation or photolysis. After UV treatment, no ions were detected in the mass spectra of these peptides. However, after photolysis, several ions were detected, including those corresponding to the crosslinked peptides. These results suggest that UV-MS is a sensitive technique for detecting bis-aryl hydrazine crosslinked peptides.

Charge-Dependence on UV-MS and UV-Photodissociation

Some bis-aryl hydrazine crosslinked peptides were detected in the UV-MS spectrum, but the intensity of the peaks was much lower than that of the unmodified peptides. This suggests that the charge state of the peptides may play a role in the detectability of the crosslinked peptides. Further experiments are needed to understand the charge dependence of UV-MS.

Conclusions

UV-MS and UV-photodissociation are promising techniques for detecting bis-aryl hydrazine crosslinked peptides. These techniques may be useful for the identification of crosslinked peptides in complex biological samples.

References


