

Testing isotopic labeling with [$^{13}\text{C}_6$]glucose as a method of advanced glycation sites identification.

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Rok wydania

2012

Czasopismo

Analytical Biochemistry

Numer woluminu

431

Strony

57-65

DOI

10.1016/j.ab.2012.08.026

Kolekcja

Naukowa

Język

Angielski

Typ publikacji

Artykuł

Streszczenie

The Maillard reaction occurring between reducing sugars and reactive amino groups of biomolecules leads to the formation of a heterogeneous mixture of compounds: early, intermediate, and advanced glycation end products (AGEs). These compounds could be markers of certain diseases and of the premature aging process. Detection of Amadori products can be performed by various methods, including MS/MS techniques and affinity chromatography on immobilized boronic acid. However, the diversity of the structures of AGEs makes detection of these compounds more difficult. The aim of this study was to test a new method of AGE identification based on isotope (^{13}C) labeling. The model protein (hen egg lysozyme) was modified with an equimolar mixture of [$^{12}\text{C}(6)$]glucose and [$^{13}\text{C}(6)$]glucose and then subjected to reduction of the disulfide bridges followed by tryptic hydrolysis. The digest obtained was analyzed by LC-MS. The glycation products were identified on the basis of characteristic isotopic patterns resulting from the use of isotopically labeled glucose. This method allowed identification of 38 early Maillard reaction products and five different structures of the end glycation products. This isotopic labeling technique combined with LC-MS is a sensitive method for identification of advanced glycation end products even if their chemical structure is unknown.

Słowa kluczowe

Nonenzymatic glycation, Amadori products, Advanced glycation end products, Isotopic labeling, mass spectrometry

Adres publiczny

<https://doi.org/10.1016/j.ab.2012.08.026>