

Selective ESI-MS detection of carbonyl containing compounds by aminoxyacetic acid immobilized on a resin.

Autorzy

Monika Kijewska
Tomasz Koch
Mateusz Waliczek
Andrzej Konieczny
Piotr Stefanowicz
Zbigniew Szewczuk

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There are numerous examples of bioactive compounds containing carbonyl groups including modified proteins with oxidation of side chain of amino acid residues to aldehyde/ketone groups which are frequently considered as markers of oxidative stress. The carbonyl unit can be also distinguished as a substructure in many illegal drugs including anabolic steroids as well as cations derivatives. Based on chemoselective formation of oximes by solid phase immobilized hydroxylamine derivatives we proposed the protocol for derivatization and selective detection of carbonylated compounds in human serum albumin hydrolysate as a complex peptide mixture and of testosterone in urine samples. This allowed for the removal of the matrix and the qualitative and quantitative analysis of the derivatized analyte by LC-MS/MS (or LC-MRM). Herein we report the preparation and chemical characterization of a novel, ChemMatrix - based resin functionalized with aminoxyacetic acid (AOA). The hydroxylamine moiety in this resin is combined with a peptide linker (GRG) containing an arginine residue to enhance the ionization efficiency. Application of an isotopically labeled carbonylated peptide ((H-Leu-Val-Thr(O)-Asp-Leu-Thr-Lys [$^{13}\text{C}_6$, $^{15}\text{N}_2$]-OH and testosterone- d_3 allowed us to carry out quantitative analyses of detected compounds. Our method is general and may be applied for analysis of carbonylated compounds in biological samples. Our method based on application of functionalized resin allowed to quantify the level of free testosterone in small sample (0.5 mL) of urine, while the non-derivatized testosterone from urine sample was not detected during direct LC-MRM analysis.

Słowa kluczowe

Aminoxyacetic acid, Selective detection, ChemMatrix rink resin, Oxime, Carbonylated peptides, Derivative of testosterone, LC-MS analysis

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