FREE FATTY ACIDS DETERMINATION IN HUMAN PLASMA OF HEALTHY AND DIABETIC SUBJECTS BY LIQUID CHROMATOGRAPHY-HIGH RESOLUTION MASS SPECTROMETRY

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Free fatty acids (FFAs) constitute minor components of human plasma that may bind to cell-surface GPCR receptors, exerting diverse effects. For instance, the alterations in their levels have been linked to type 2 diabetes (T2D) and obesity [1]. The presence of 2- and 3-hydroxy FAs has also been reported in human plasma in minor quantities, however, the existence of other saturated hydroxy fatty acids (SHFAs) in plasma, has not been reported in literature to date. Previously, we demonstrated that a series of SHFAs (also present in dairy products), exhibit antiproliferative activity *in vitro* against different human cancer cell lines and suppress β-cell apoptosis [2].
In this work, we present a liquid chromatography-high resolution mass spectrometry (LC-HRMS) method for the rapid determination of a variety of oxidized and common FFAs in human plasma, in a 10-min single run, avoiding any sample pre-treatment. LC-MS/MS measurements were performed with an ABSciex Triple TOF 4600 combined with a micro-LC Eksigent and an autosampler. Electrospray ionization in negative mode was used for the MS experiments. Halo C18 2.7 μm, 90 Å, 0.5×50 mm^2 (Eksigent) was used as the column and the mobile phase consisted of a gradient (A: acetonitrile/0.01% formic acid/isopropanol 80/20 v/v; B: H_2O/0.01% formic acid). In our study, 35 common FFAs and 39 oxidized (hydroxy and oxo) saturated FFAs, were detected and quantified in plasma samples of 28 healthy subjects, 29 T2D patients and 14 T1D patients. Hydroxystearic acids (HSAs) and hydroxypalmitic acids (HPAs) with a hydroxyl group at positions higher than 2 and 3, namely 7HSA, 8HSA, 11HPA, and 16HPA and one oxo fatty acid, 6OSA, were quantified in human plasma for the first time. Interestingly, alterations in the levels of medium-chain FFAs (C6:0 to C10:0) were observed between the control group and diabetic patients.

References

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