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Quantification of Malondialdehyde in Human Urine by HPLC with DAD and Derivatization with 2,4-Dinitrophenylhydrazine

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Abstract

This study demonstrated that human urine can be derivatized in a simple manner and the proposed method can be suitable for the routine and noninvasive evaluation of oxidative stress. A rapid and sensitive method for the determination of malondialdehyde (MDA) concentrations in human urine has been developed using high-performance liquid chromatography. The MDA concentration in pooled human urine samples was determined after its conversion with 2,4-dinitrophenylhydrazine (DNPH) to a hydrazone (MDA-DNPH). The MDA-DNPH was separated on C18 column and quantified by diode array detection at 310 nm. The method was validated by the use of spiked human urine. The calibration curves of analyte showed good linearity ($r^2 > 0.999$) in the ranges. The limit of detection was estimated as 0.16 $\mu\text{g/mL}$. In addition, accuracy was from 104.0 to 112.7% and the precision was less than 3.2%. In this study, the developed method was applied to the analysis of real human urine samples. The level of MDA in urine of adults was $1.3 \pm 0.6 \mu\text{g/mg creatinine}$.

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