

A Validated High-Performance Liquid Chromatography-UV Method for Determination of Malondialdehyde-Application to Study in Chronic Ciprofloxacin Treated Rats

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Abstract : Present work demonstrates the applicability of high-performance liquid chromatography (HPLC) with UV detection for the determination of malondialdehyde as malondialdehyde-thiobarbituric acid complex (MDA-TBA) in-vivo in rats. The HPLC-UV method for MDA-TBA was achieved by isocratic mode on a reverse-phase C18 column (250mm×4.6mm) at a flow rate of 1.0mLmin⁻¹ followed by UV detection at 278 nm. The chromatographic conditions were optimized by varying the concentration and pH followed by changes in percentage of organic phase optimal mobile phase consisted of mixture of water (0.2% Triethylamine pH adjusted to 2.3 by ortho-phosphoric acid) and acetonitrile in ratio (80:20 % v/v). The retention time of MDA-TBA complex was 3.7 min. The developed method was sensitive as limit of detection and quantification (LOD and LOQ) for MDA-TBA complex were (standard deviation and slope of calibration curve) 110 ng/ml and 363 ng/ml respectively. The method was linear for MDA spiked in plasma and subjected to derivatization at concentrations ranging from 100 to 1000 ng/ml. The precision of developed method measured in terms of relative standard deviations for intra-day and inter-day studies was 1.6-5.0% and 1.9-3.6% respectively. The HPLC method was applied for monitoring MDA levels in rats subjected to chronic treatment of ciprofloxacin (CFL) (5mg/kg/day) for 21 days. Results were compared by findings in control group rats. Mean peak areas of both study groups was subjected for statistical treatment to unpaired student t-test to find p-values. The p value was < 0.001 indicating significant results and suggesting increased MDA levels in rats subjected to chronic treatment of CFL of 21 days.

Keywords : MDA, TBA, ciprofloxacin, HPLC-UV

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