



Determination of Diphenhydramine Hydrochloride in Rabbit Plasma by LC-MS/MS and its Application to a Pharmacokinetic Study

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SUMMARY. A sensitive and selective liquid chromatography tandem mass spectrometry (LC-MS/MS) method for determination of diphenhydramine hydrochloride in rabbit plasma was developed and validated. After addition of triazolam as internal standard (IS), protein precipitation by acetonitrile was used as sample preparation. Chromatographic separation was achieved on a Zorbax SB-C18 (2.1 mm × 50 mm, 3.5 μm) column with acetonitrile-0.1 % formic acid as mobile phase with gradient elution. Electrospray ionization (ESI) source was applied and operated in positive ion mode; multiple reaction monitoring (MRM) mode was used to quantification using target fragment ions m/z 255.8 → 166.6 for diphenhydramine hydrochloride and m/z 342.9 → 308.0 for the IS. Calibration plots were linear over the range of 5-200 ng/mL for diphenhydramine hydrochloride in rabbit plasma. Lower limit of quantitation (LLOQ) for diphenhydramine hydrochloride was 5 ng/mL. RSD of intra-day and inter-day precisions were both less than 10 %. This developed method is successfully used in pharmacokinetic study of diphenhydramine hydrochloride in rabbit.

KEY WORDS: Diphenhydramine hydrochloride, Liquid chromatography-tandem mass spectrometry, Pharmacokinetics, Plasma.

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