A RAPID AND SENSITIVE LC-ESI-MS/MS METHOD FOR DETECTION AND QUANTITATION OF METHYLPREDNISOLONE AND METHYLPREDNISOLONE ACETATE IN RAT PLASMA

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A rapid, sensitive and specific liquid chromatography-electrospray-tandem mass spectrometric (LC-ESI-MS/MS) method for the simultaneous detection and quantitation of methylprednisolone acetate (MPA) and methylprednisolone (MP) in rat plasma, using a triple-stage quadrupole, has been developed and validated.
MP-D_2 was used as internal standard (IS) and acetonitrile was added to plasma samples for protein precipitation. After extraction with dichloromethane, the analytes were separated on a C-12 reversed-phase column by isocratic elution (6 min at a flow rate 0.2 mL min^{-1}) with water containing 0.01% formic acid (A) and acetonitrile (B) (50:50, v/v). Quantitation was performed in positive-ion Multiple Reaction Monitoring (MRM) mode by applying the following precursor-to-product ion transitions: MPA m/z 417 → 135 + 161 + 253; MP m/z 375 →135 + 161 + 253; IS m/z 377 → 135 + 161 + 253. The method, validated over the concentration range 6-600 ng mL^{-1}, has been shown to meet the current requirements of bioanalytical validation, providing satisfactory results in terms of linearity, recovery, intra-day and inter-day precision and accuracy. The lower limit of quantitation (LLOQ) was 6 ng mL^{-1} for both the analytes (0.080 pmol and 0.072 pmol injected for MP and MPA, respectively). The method was successfully applied to monitor the plasma levels of MPA and MP following intra-articular (IA) injections of a low MPA (Depo-Medrol ®) dose in rats.

References

Di Francesco, V. Frerichs, J. Donnelly, C.Hagler, J.Hochreiter, K. M. Tornatore, J. Chromatogr. B 859 (2007) 42–51
R.L. Taylor, S. K. Grebe, R.J. Singh, Clin. Chem. 50 (2004) 2345–2352
Zhang, R. Mehvar, J. Pharm. Sci. 90 (2001) 2078-2087
Mehvar, R O. Dann, D. A Hoganson, J. Pharm. Biomed. Anal. 22 (2000) 1015-1022