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# Determination of Saikosaponin B1 and Saikosaponin B3 in Rat Plasma by UPLC-MS/MS and its Application to a Pharmacokinetic Study

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***SUMMARY*. A sensitive and selective UPLC-MS/MS method was developed and validated for the determination of saikosaponin B1 and saikosaponin B3 in rat plasma. Saikosaponin F was used as internal standard (IS). Chromatographic separation was performed on a UPLC BEH (2.1 × 50 mm, 1.7 $\mu$m) column with a mobile phase consisting of acetonitrile and water (containing 0.1% formic acid). The gradient elution flow rate was 0.4 mL/min and the elution time was 4 min. Nitrogen was used as desolvation gas (800 L/h) and cone gas (50 L/h). The MRM transitions were m/z 825.4→617.5 for saikosaponin B1, m/z 857.6→649.6 for saikosaponin B3, and m/z 973.7→101.0 for IS, respectively. Acetonitrile precipitation was adopted to remove protein in rat plasma before UPLC-MS/MS analysis. Calibration curves of both saikosaponin B1 and saikosaponin B3 were linear (r > 0.995) over the concentration range of 5-5000 ng/mL with the lowest limit of quantitation (LLOQ) of 5 ng/mL. The intra-day and inter-day precisions of saikosaponin B1 represented as RSD were less than 14 and 10%, respectively. The intra-day and inter-day precisions of saikosaponin B3 were less than14 and 11%, respectively. The accuracy of both compounds were over the range of 94 and 109.6%. This method was confirmed to be fast, sensitive and specific and successfully applied to a pharmacokinetic study of saikosaponin B1 and saikosaponin B2 in rats after sublingual intravenous administration.**

*RESUMEN*. Se desarrolló y validó un método de UPLC-MS/MS sensible y selectivo para la determinación de saikosaponina B1 y saikosaponina B3 en plasma de rata. Saikosaponina F se usó como estándar interno (IS). La separación cromatográfica se realizó en una columna UPLC BEH (2,1 × 50 mm, 1,7 $\mu$m) con una fase móvil que consiste en acetonitrilo y agua (que contiene 0,1% de ácido fórmico). El caudal de elución en gradiente fue de 0,4 mL/min y el tiempo de elución fue de 4 min. Se usó nitrógeno como gas de desolvatación (800 L/h) y gas de cono (50 L/h). Las transiciones de MRM fueron m/z 825.4→617.5 para saikosaponina B1, m/z 857.6→649.6 para saikosaponina B3 y m/z 973.7→101.0 para IS, respectivamente. Se adoptó la precipitación con acetonitrilo para eliminar la proteína en plasma de rata antes del análisis por UPLC-MS/MS. Las curvas de calibración de saikosaponina B1 y saikosaponina B3 fueron lineales (r > 0.995) en el rango de concentración de 5-5000 ng/mL con el límite más bajo de cuantificación (LLOQ) de 5 ng/mL. Las precisiones intradía e interdía de saikosaponina B1 representada como RSD fueron menores del 14% y del 10%, respectivamente. Las precisiones intradía e interdía de saikosaponina B3 fueron menores del 14 y del 11%. La precisión de ambos compuestos estuvo en el rango de 94% y 109.6%. Se confirmó que este método era rápido, sensible y específico y se aplicó con éxito a un estudio farmacocinético de saikosaponina B1 y saikosaponina B2 en ratas después de la administración sublingual intravenosa.

*KEY WORDS:* pharmacokinetics, saikosaponin B1, saikosaponin B2, UPLC-MS/MS.

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