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Simultaneous Determination of Four Cytochrome P450 Probe Drugs in Rat Plasma by a Simple Liquid Chromatography–Mass Spectrometry Method

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***SUMMARY*. A sensitive and simple liquid chromatography- mass spectrometry (LC-MS) method was developed and validated for determination of four cytochrome P450 probe drugs (phenacetin (CYP1A2), tolbutamide (CYP2C9), bupropion (CYP2B6) and omeprazole (CYP2C19) in rat plasma. Four cytochrome P450 probe drugs extracted from plasma samples by protein precipitation with acetonitrile and separation were carried out on Agilent Zorbax SB-C18 column (2.1 mm x 150 mm, 5 μm) at 30 °C, acetonitrile –0.1 % formic acid in water used as mobile phase with gradient elution. Electrospray ionization (ESI) source was applied and operated in positive ion mode, and selective ion monitoring (SIM) mode was used to quantify cytochrome P450 probe drugs. The method showed excellent intra-assay and inter-assay precision (relative standard deviation (RSD) and bias <15 %) for quality control (QC) samples spiked at a concentration of 10, 400, and 1600 ng/mL and the $r^2 > 0.99$ over the range investigated (5-2000 ng/mL). Lower limits of quantification (LLOQs) were estimated to be 5 ng/mL. The method was successfully applied to determinate cytochrome P450 probe drugs in a pharmacokinetic study.**

KEY WORDS: Bupropion, LC-MS, Omeprazole, Phenacetin, Rat plasma, Tolbutamide.

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