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Simultaneous estimation of artesunate, dihydroartemisinin and quercetin in rat plasma by a simple sensitive UFLC-MS/MS method

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Objective: A novel, accurate, precise and rapid LC-MS/MS method has been developed for the simultaneous estimation of artesunate (ART), dihydroartemisinin (DHA) and quercetin (QRT) in rat plasma.

Methods: Agilent zorbax C18 column (3 μ m, 50 mm×4.6 mm) was used a stationary phase and mobile phase constitutes 0.1% formic acid (90 volumes) and methanol (10 volumes), at a flow rate of 0.4mL•min–¹ and injection volume of 10 μ L. Artemisinin (ART) was used as internal standard. For collision induced dissociation experiments argon gas was employed. Mass detection was performed by electrospray ionization tandem mass spectrometry via multiple reaction monitoring (MRM) in positive mode except for quercetin where negative ionization was used. Plasma samples were subjected to solid phase extraction using methanol as eluent to extract ART, DHA and QRT.

Results: The MRM transitions of ART, DHA, QRT and ATM were found to be $407.2 \rightarrow 261.0$, $307.1 \rightarrow 261.0$, $301.1 \rightarrow 151.0$ and $283.2 \rightarrow 265.05$, respectively. Collision energies used were -17, -12, 22 and -15ev for ART, DHA, QRT and ATM, respectively. The extraction recoveries of ART, DHA and QRT were found to be 80.5-88.69%, 76.7-81.52% and 89.76-94.89%, respectively. The lower limit of quantification (LLOQ) was 1 ng·mL⁻¹ for ART and DHA whereas for QRT it was 50 Ng.mL⁻¹.

Conclusion: The developed LC-MS/MS method is simple, sensitive and rapid. Moreover, the proposed method can be successfully applied for the simultaneous analysis of ART, DHA and QRT in rat plasma for the pharmacokinetics studies