Microwave-Assisted Extraction of Andrographolide, 14-Deoxy-11,12-didehydroandrographolide and Neoandrographolide from *Andrographis paniculata*: An Optimisation Study

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Abstract: **Background:** *Andrographis paniculata* is often used as functional foods owing to its broad range of pharmaceutical activities due to the presence of main terpene compounds such as andrographolide, 14-deoxy-11,12-didehydroandrographolide and neoandrographolide. The aforementioned compounds must be extracted from the plant material before it can be routinely used as a functional food ingredient.

**Objective:** This work presents an optimisation study of microwave-assisted extraction (MAE) of terpene from *Andrographis paniculata*. In this study, two factors at three-level central composite face centred design was employed to optimise the process variables, including the effect of microwave power (75-175 W) and ethanol concentration (20-85%) on extraction of terpene from *Andrographis paniculata*.

**Methods:** The experimental design was set based on initial screening on the effect of mesh size and solid solvent ratio. The present work was performed at fixed mesh size and solid solvent ratio, which corresponds to the highest yield established experimentally. Two most significant factors, i.e. microwave power and ethanol concentration was chosen and further studied for optimisation using a response surface methodology. The three-level, two-factor central composite design (CCD) has been employed to determine the effect of microwave power (15, 25 and 35 W/mL) and ethanol concentration (20, 52.5 and 85%) on the yield of terpene extraction. Identification and quantification of the terpene were performed via Ultra-Performance Liquid Chromatography- Photodiode Array (UPLC-PDA).

**Results:** The regression analysis showed good fit of the experimental data to the quadratic equation with coefficient of determination ($R^2$) value of 0.997. The optimum condition was found at microwave power of 140 W with 85% ethanol concentration, produced the highest yield of andrographolide (10.926 ± 0.053 mg/g DW), 14-deoxy-11,12-didehydroandrographolide (4.336 ± 0.215 mg/g DW) and neoandrographolide (5.698 ± 0.252 mg/g DW). The model developed through response surface modelling has a desirability of 97.5% and the error between the experiment and the model predictions ranged from 2.4 to 5.2%.

**Conclusion:** A successful optimisation of terpene extraction from *A. paniculata* via MAE was achieved at microwave power of 140 W and 85% ethanol. The model developed from the regression analysis is sufficiently accurate and can be used to predict the yield of terpene extraction from *A. paniculata*.

**Keywords:** 14-deoxy-11,12-didehydroandrographolide, *Andrographis paniculata*, andrographolide, microwave extraction, neoandrographolide, response surface methodology.

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