UNIVERSITI TEKNOLOGI MARA

DEVELOPMENT OF CHROMATOGRAPHIC FINGERPRINT USING ONLINE SOLID PHASE EXTRACTION-LIQUID CHROMATOGRAPHY (SPE-LC) AND CHEMOMETRIC APPROACH FOR QUALITY ASSESSMENT OF *Eurycoma longifolia*

NOR NASRIAH BINTI ZAINI

MSc

August 2020

AUTHOR'S DECLARATION

I declare that the work in this thesis was carried out in accordance with the regulations of Universiti Teknologi MARA. It is original and is the result on my own work, unless otherwise indicated or acknowledged as referenced work. This thesis has not been submitted to any other academic institution or non-academic institution for any other degree or qualification.

I, hereby, acknowledge that I have been supplied with the Academic Rules and Regulations for Post Graduate, Universiti Teknologi MARA, regulating the conduct of my study and research.

Name of Student	:	Nor Nasriah binti Zaini
Student's ID No.	:	2014445152
Programme	:	Master of Science (Chemistry) – AS756
Faculty	:	Faculty of Applied Sciences
Thesis Title	:	Development of Chromatographic Fingerprint Using Online Solid Phase Extraction-Liquid Chromatography (SPE-LC) and Chemometric Approach for Quality Assessment of *Eurycoma longifolia*

Signature of Student :

Date : August 2020

ABSTRACT

Eurycoma longifolia, locally known as Tongkat Ali is one of the popular herbs in Malaysia and has been reported to contain various chemically active compounds. The inconsistency in the composition of herbal products has always been a concern and thus, a reliable method to ensure the quality of *Eurycoma longifolia* is crucial. In this study, target compounds in *Eurycoma longifolia* were extracted using water by pressurised liquid extraction (PLE), identified and quantified by online solid phase extraction-liquid chromatography (SPE-LC). Optimization of PLE operating parameters conducted using the response surface method (RSM) gave an optimum extraction temperature of 100 °C and an extraction time of 30 minutes. For online SPE-LC with diode array detector (DAD), 2 columns, polystyrene divinyl benzene (PSDVB) and Waters C_{18} were used. The mobile phase compositions of water, methanol and acetonitrile and column switching time for online SPE-LC were optimized. The developed method was used to analyse *Eurycoma longifolia* roots from various sources and products from various manufacturers. Selected chemometric techniques: cluster analysis (CA), discriminant analysis (DA) and principal component analysis (PCA) were applied to the chromatographic dataset of 37 selected peaks. For root samples, CA revealed 3 clusters suggesting the classification of quality based on the amount of selected compounds. PCA analysis applied to roots and products clearly discriminated against the roots from products. CA gave 3 clusters of roots and products with a 100 % correlation coefficient by DA. The clustering of products in the same group with the roots may suggest that they were from the same source. Pattern matching analysis showed no significant difference ($p > 0.05$) between the roots and products within the same CA grouping with a percent similarity of 92.6 %. The developed online SPE-LC method provides a fast and efficient approach in obtaining a comprehensive fingerprint. The application of chemometrics to the fingerprint database could be a promising approach for quality assessment of herbal formulation.

ACKNOWLEDGEMENTS

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