# EXTRACTION OF SELECTED BIOACTIVE COMPOUNDS FROM *AVERRHOA BILIMBI* FRUITS AND THEIR THERMAL STABILITY DURING SPRAY DRYING

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DOCTOR OF PHILOSOPHY

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We hereby declare that we have checked this thesis and in our opinion, this thesis is adequate in terms of scope and quality for the award of the degree of Doctor of Philosophy (Bio-process Engineering).

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## STUDENT'S DECLARATION

I hereby declare that the work in this thesis is my own except for the quotations and summaries which have been duly acknowledged. The thesis has not been accepted for any degree and is not concurrently submitted for award of other degree.

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NORLIA MUHAMAD

Thesis submitted in fulfillment of the requirements
for the award of the degree of
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Faculty of Chemical and Natural Resources Engineering

UNIVERSITI MALAYSIA PAHANG

JULY 2017

## DEDICATION

*In the name of Allah, The Most Gracious and The Most Merciful*

*I humbly dedicated this thesis to…*

*my dear husband; Ahmad Fakaruddin Md. Salleh*
*my lovely sons; Ahmad Haikal and Ahmad Haziq*
*my special encouraging parents; Mak and Abah*
*my remarkable family members,*
*my friends,*
*who gave me everlasting inspiration,*
*never ending encouragements,*
*priceless support*
*and pray without ceasing..*

*Thank you very much*

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## LIST OF SYMBOLS

| | |
|---|---|
| % | Percent |
| °C | Degree celcius |
| $E_a$ | Arrhenius activation energy |
| $k$ | Reaction rate constant |

## LIST OF ABBREVIATIONS

| | |
|---|---|
| DE | Dextrose equivalent |
| DW | Dry weight |
| FESEM | Field emission scanning electron microscopy |
| GA | Arabic gum |
| GAE | Gallic acid equivalent |
| HPLC | High performance liquid chromatography |
| LCMS-Q-TOF | Liquid chromatography mass spectrometry coupled to quadruple time-of-flight |
| AE | Microwave-assisted extraction |
| MD | Maltodextrin |
| ME | Maceration |
| OFAT | One-factor-at-a-time |
| RSM | Response surface methodology |
| SE | Soxhlet extraction |
| SFE | Supercritical fluid extraction |
| UAE | Ultrasonic-assisted extraction |
| UHPLC | Ultra-high performance liquid chromatography |
| UV | Ultra violet |
| WPI | Whey protein isolate |

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# ABSTRAK

*A. bilimbi* (ver. Nama Belimbing Buluh) mengandungi banyak sebatian bioaktif berguna yang membawa kepada pelbagai aktiviti seperti antioksidan, anti-diabetes dan anti-batuk kering. Analisis terhadap ekstrak *A. bilimbi* menggunakan UHPLC-QTOF-MS, analisis kualitatif dan pengenalpastian menggunakan Pangkalan Data Metlin menunjukkan kehadiran tiga sebatian; asid nicotinic, asid pantothenic dan catechin. Berdasarkan pengesahan melalui spektrometri jisim dan spectrum UV, asid nicotinic, asid pantothenic dan catechin boleh digunakan sebagai penanda *A. bilimbi*. Pemisahan komponen bioaktif daripada bahan-bahan tumbuhan biasanya dilakukan melalui proses pengekstrakan. Hasil komponen bioaktif dalam ekstrak itu bergantung kepada pelarut yang digunakan, kaedah pengekstrakan dan keadaan proses pengekstrakan digunakan. Dalam penyelidikan ini, pengekstrakan menggunakan ultrasonik telah digunakan. Kadar hasil pengekstrakan tertinggi sebanyak 0.28 mg/g DW asid nicotinic, 1.00 mg/g DW pantothenic asid dan 0.22 mg/g catechin DW dari *A. bilimbi* diperolehi pada suhu 30 °C, kuasa sonication daripada 0.070 W/ml, masa 30 min, nisbah pelarut kepada pepejal 8 ml/g dan kepekatan metanol 40%. Komponen bioaktif diketahui mengalami proses degradasi apabila terdedah kepada suhu yang tinggi dalam tempoh masa yang panjang, ia sering berlaku semasa proses pengekstrakan dan penghasilan serbuk. Proses tersebut sering dicapai pada suhu sedikit tinggi. Oleh itu, kehilangan komponen bioaktif kerana degradasi perlu difahami sebelum kaedah kawalan yang boleh dibuat. Hasil kajian menunjukkan degradasi asid nicotinic, asid pantothenic dan catechin mengikuti model kinetik tertib pertama. Asid pantothenic menunjukkan pemalar kadar degradasi yang paling rendah menunjukkan proses degradasi yang paling perlahan., diikuti oleh asid nicotinic dan catechin untuk semua suhu dikaji. Haba tenaga pengaktifan degradasi asid nicotinic, asid pantothenic dan catechin adalah 43.85 kJ/mol, 58.86 kJ/mol dan 21.27 kJ/mol. Asid pantothenic mempunyai tenaga pengaktifan yang paling tinggi menunjukkan bahawa sebatian tersebut adalah lebih sensitif kepada perubahan suhu. Pemikrokapsulan boleh memberikan halangan fizikal di antara bahagian teras dan komponen luar produk, seterusnya akan memberi perlindungan haba yang lebih baik semasa proses pengeringan semburan. Pemikrokapsulan komponen bioaktif daripada *A. bilimbi* oleh pengeringan semburan menggunakan maltodekstrin menghasilkan kandungan asid nicotinic, asid pantothenic dan catechin yang tinggi. Keputusan yang diperolehi daripada kajian ini menunjukkan bahawa keadaan operasi yang terbaik untuk pengeringan semburan ekstrak *A. bilimbi* dengan pengekalan tertinggi asid nicotinic, asid pantothenic dan catechin adalah dengan menggunakan 10% maltodekstrin DE18 pada suhu udara masuk 140 °C.

## ABSTRACT

*A. bilimbi* (ver. name Belimbing Buluh) contains many useful bioactive compounds leading to various activities such as antioxidant, anti-diabetic and anti-tuberculosis. Analysis of the *A. bilimbi* extracts using UHPLC-QTOF-MS, qualitative analysis and identification using Metlin Database shows the presence of three compounds; nicotinic acid, pantothenic acid and catechin. Following the mass spectrometry and UV spectra confirmation, the nicotinic acid, pantothenic acid and catechin can be used as a marker of *A. bilimbi*. Recovery of bioactive compounds from the plant materials are normally performed via extraction. The yield of bioactive compound in the extract is dependent on the solvent used, extraction method and condition. In this work, ultrasonic assisted extraction was studied. The highest extraction yields of 0.28 mg/g DW nicotinic acid, 1.00 mg/g DW pantothenic acid and 0.22 mg/g DW catechin from *A. bilimbi* were obtained at temperature of 30 °C, sonication power of 0.070 W/ml, time of 30 min, solvent to solid ratio of 8 ml/g and methanol concentration of 40%. Bioactive compounds are known to suffer from a degradation process when exposed to high temperature over a long period, which is often the case during the extraction and powder making process. The recovery process is often accomplished at slightly elevated temperatures, thus it is vital to understand the loss of bioactive compounds due to thermal degradation before the mitigating method can be formulated. The results showed nicotinic acid, pantothenic acid and catechin degradation followed the first-order kinetics model. Pantothenic acid showed the lowest degradation rate constant, followed by nicotinic acid and catechin for all studied temperature, indicating a slowest degradation. The thermal degradation activation energy of nicotinic acid, pantothenic acid and catechin were 43.85 kJ/mol, 58.86 kJ/mol, and 21.27 kJ/mol, respectively. Pantothenic acid has the highest activation energy which implies that the compound is more sensitive to temperature change. Microencapsulation can provide a physical barrier between the core compound and the other components of the product which would then provide a better heat protection during spray drying. Microencapsulation of bioactive compounds from *A. bilimbi* by spray drying using maltodextrin resulted in a high retention of nicotinic acid, pantothenic acid and catechin. Results obtained from this work demonstrated that the best operating conditions for spray drying of *A. bilimbi* extract whereby the highest retention of nicotinic acid, pantothenic acid and catechin are 10% maltodextrin DE18 and air inlet temperature 140 °C.

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