QUALITY IMPROVEMENT OF MANUKA HONEY THROUGH THE APPLICATION OF HIGH PRESSURE PROCESSING

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A thesis submitted in fulfilment of the requirements for the degree of

Doctor of Philosophy in Chemical and Materials Engineering

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ABSTRACT

The quality of honey is known to be compromised when it goes through thermal processing due to its negative impact on the unstable and thermolabile honey components which originated from the nectar and bees themselves. This present work is undertaken to access the use of an emerging food preservation technique known as "High Pressure Processing" for treating honey, as an alternative to the conventional thermal processing. In this thesis, honey quality has been addressed by measuring the effects of high pressure processing parameters (pressure, time and temperature) on nutritional properties of honey, namely total phenolic content and antioxidant activity. Honey samples, contained in small pouches, were subjected to different pressures (200-600 MPa) at close to ambient temperatures (25-33°C) for different holding times (10 to 30 min). Thermal processing (49-70°C) was also carried out for comparison purpose. Results demonstrated that high pressure processing operated at 600 MPa for 10 min has capability to increase significantly the total phenolic content and antioxidant activity by 47% and 30%, respectively. Besides, the result showed that high pressure processing can maintain the natural colour of honey which relates directly to consumer perception, while retaining its shear-thinning behaviour and viscosity with no significant changes ($p > 0.05$). High pressure processing can also control hydroxymethylfurfural (HMF) concentration in honey during process within the standard limit, 16.93 to 18.76 mg/kg (which is below than the maximum allowed limit of 40 mg/kg). This work also reveals that high pressure processing can enhance antibacterial activity of Manuka honey significantly. It shows an increase in the percentage inhibition of *Staphylococcus epidermidis* from 64.15 ± 5.86% to 84.34 ± 7.62% when honey was subjected to 600 MPa. Storage studies for one year at room temperature (25°C) demonstrated that high pressure-treated samples have a good retention to the physicochemical, nutritional and rheological properties of honey throughout storage, which confirms that the positive effect of high pressure on honey is not a temporary effect. Whereas, an insight study on the safety part showed that the *Saccharomyces cerevisiae* cell varied linearly with ° Brix, indicating that food compressibility has a significant role in the microbial inactivation.

Dedicated to my parents and family for their endless love

"There are no shortcuts to any place worth going."

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Praised be to God the Almighty for infinite blessings

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To all friends (special to Mr Alifdalino), I would not have survived all the stresses and worries, if not for the beautiful moments you gave me over the past three years. Last but not least, the most special thank goes to my beloved mom, Gayah Sulaiman and dad, Mohd Fauzi Abdul Razak for their unconditional and endless love, and also for being there in times of need. Both of you have showered me with full of love, understanding, support and continuous pray. Also thank you to my beloved brothers and sisters, all my family members and not forgotten, Mr Rasdizarapi Salhudin for their endless support and encouragement throughout my study. I love all of you very much.

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NOMENCLATURE

AA	= antioxidant Activity
ANOVA	= analysis of variance
AU	= absorbance Unit
Abs	= absorbance
$Abs_{control}$	= absorbance reading of the control
Abs_{sample}	= absorbance reading of the sample
A_{660nm}	= absorbance at 660 nm
BPF	= brown pigment formation
CFU	= colony forming unit
C*p*	= specific heat capacity
DHA	= dihydroxyacetone
DNA	= deoxyribonucleic acid
DPPH	= 2,2-diphenyl-1-picrylhydrazyl
D-value	= decimal reduction time at specific temperature
FAO	= food and agriculture organization of the United Nations
GAE	= gallic acid equivalent
g	= gram
HHP	= high hydrostatic pressure
HMF	= hydroxymethylfurfural
HPP	= high pressure processing
Hz	= hertz
hrs	= hours
in	= inches
j	= joule
$K_4Fe\ (CN)_6.3H_2O$	= potassium hexacyanoferrate in water

L = litre

MGO = methylglyoxal

MIC_{95} = minimum inhibitory concentration of at least 95% inhibition

MPa = megapascal

MWCO = molecular weight cut off

min = min

mL = mililitre

mS = milisiemen

N = number of microorganism

NaCl = sodium chloride

Na_2CO_3 = sodium carbonate

N_o = initial number of microorganisms

nm = nanometers (wavelength)

OH = hydroxyl molecules

P = pressure

pH = decadic logarithm of acid dissociation

psi = pounds-force per square inch

R^2 = linear regression coefficient/ coefficient of determination

r = correlation coefficient

rpm = revolutions per minute (measure of the frequency of a rotation)

S = entropy

SD = standard deviation

S.cerevisiae = *Saccharomyces cerevisiae*

S. epidermidis = *Staphylococcus epidermidis*

T = temperature

TCD = total colour difference

TPC = total phenolic content

T_{avg} = average temperature

T_{set} = setting temperature

t = time

UF = ultrafiltration

UHP = ultra high pressure

UV = ultraviolet

V = volume

YPD = yeasts peptone dextrose

$Zn(CH_3COO)_2.2H_2O$ = zinc acetate in water

Z_p- value = Pressure required for one log reduction in the *D*-value.

ºC = degree centigrade

< = less than

≥ = greater than or equal to

± = plus minus

% = percentage

w/v = weight/volume

µ = micro

β = thermal expansion coefficient

W = watt

*L**, *a**, *b** = colour parameters: *L** from 0: black to 100: white; *a** from –80: green to +80: red; *b** from –80: blue to +80: yellow.

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Nature of contribution by PhD candidate	Contribute to the idea, do all experimental works and do writing task.
Extent of contribution by PhD candidate (%)	80%

CO-AUTHORS

Name	Nature of Contribution
Prof Mohammed Farid	As a main supervisor, contribute to the main idea, make a review and check the whole paper from beginning untill the paper get accepted.
Dr Filipa Silva	As a co-supervisor, give ideas on experimental works and assist in review and check the whole paper

Certification by Co-Authors

The undersigned hereby certify that:

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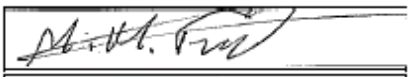
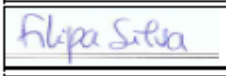
Prof Mohammed Farid		3/12/2014
Dr Filipa Silva		3/12/2014
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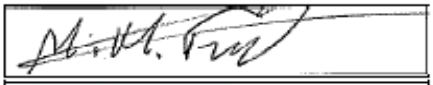
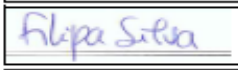
Prof Mohammed Farid		3/12/2014
Dr Filipa Silva		3/12/2014
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Chapter 5:

N.A. Fauzi & M.M.Farid. High Pressure Processing (HPP) of Manuka honey: Brown pigment formation, improvement of antibacterial activity and hydroxymethylfurfural content

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Please indicate the chapter/section/pages of this thesis that are extracted from a co-authored work and give the title and publication details or details of submission of the co-authored work. Chapter 6 High Pressure Processed Manuka Honey: Change in Nutritional and Rheological Properties over One Year Storage Contents submitted to journal publication and under review

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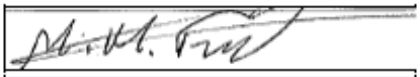
Prof Mohammed Farid		Click here
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Please indicate the chapter/section/pages of this thesis that are extracted from a co-authored work and give the title and publication details or details of submission of the co-authored work. Chapter 7 An insight on the relationship between food compressibility and microbial inactivation during high pressure processing Contents submitted to journal publication and under review

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CHAPTER 1

Introduction

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