

**OPTIMIZATION OF HEADSPACE SOLID PHASE
MICROEXTRACTION (HS-SPME) ON THE ANALYSIS OF
VOCS IN STINGLESS BEE HONEY (*Tetrigona apicalis*)**

ANIS NADIA BINTI HALIM

**BACHELOR OF SCIENCE (Hons.) CHEMISTRY
FACULTY OF APPLIED SCIENCES
UNIVERSITI TEKNOLOGI MARA**

JANUARY 2020

TABLE OF CONTENTS

	Page
ACKNOWLEDGEMENTS	iii
TABLE OF CONTENTS	iv
LIST OF TABLES	vi
LIST OF FIGURES	vii
LIST OF ABBREVIATIONS	viii
ABSTRACT	ix
ABSTRAK	x
CHAPTER 1 INTRODUCTION	
1.1 Background of the study	1
1.2 Problem statement	3
1.3 Objectives of study	4
1.4 Significant of study	5
CHAPTER 2 LITERATURE REVIEW	
2.1 Stingless bee honey	6
2.2 Volatile Organic Compounds (VOCs)	9
2.3 Gas chromatography–mass spectrometry (GC-MS)	10
2.4 Solid-Phase Microextraction (SPME)	10
2.4.1 Extraction mode of SPME	15
2.4.2 Optimization of SPME Extraction Conditions	16
a) Fiber coating selection	16
b) Optimization of extraction	18
2.4.3 Univariate analysis	19
2.4.4 Multivariate analysis	20

CHAPTER 3 METHODOLOGY

3.1	Overview of methodology	21
3.2	Chemical reagents	22
3.3	Honey sample	22
3.4	Fiber selection	22
3.5	Sample preparation	23
3.6	GC-MS conditions	23
3.7	Significant factors optimization	25
3.8	Experimental design	25

CHAPTER 4 RESULTS AND DISCUSSION

4.1	Selection of fiber	27
4.2	Extraction of VOCs	29
4.3	Modification of GC oven temperature programming	32
4.4	Optimization of significant factors	38

CHAPTER 5 CONCLUSION AND RECOMMENDATIONS

5.1	Conclusion	42
5.2	Recommendations	42

CITED REFERENCES	44
-------------------------	----

APPENDIX	56
-----------------	----

<i>CURRICULUM VITAE</i>	58
--------------------------------	----

LIST OF TABLES

Table	Caption	Page
2.4.1	Methods for the Analysis of Honey Samples	13
3.4.1	Guidelines of conditioning for SPME fiber coatings by Supelco.	23
3.8.1	Experimental design generated by Design-Expert	25
4.2.1	Extracted VOCs from stingless bee honey by different polarities of fibers using HS-SPME/GC-MS.	29
4.3.1	Extracted VOCs from stingless bee honey by different temperature program (a) without hold time and (b) with hold time using HS-SPME/GC-MS.	35
4.4.1	Level of significant factors in the Central Composite Design (CCD).	38
4.4.2	Central Composite Design (CCD) summary for HS-SPME of VOCs in stingless bee honey.	39

ABSTRACT

OPTIMIZATION OF HEADSPACE SOLID PHASE MICROEXTRACTION (HS-SPME) ON THE ANALYSIS OF VOCS IN STINGLESS BEE HONEY (*Tetrigona apicalis*)

Isolation of volatile organic compounds (VOCs) from stingless bee honey (*Tetrigona apicalis*) was done by using headspace solid phase microextraction (HS-SPME) separation with gas chromatography-mass spectrometry (GC-MS). Among three different types of SPME fibers examined, the mixed fiber coating, DVB/CAR/PDMS presented the maximum efficiency in extracting VOCs in which 22 compounds were identified and with the total peak area of 1.23×10^8 . Prior to the best extraction efficiency using the selected fiber, GC oven temperature programming with the use of hold time recorded a total numbers of 47 VOCs were detected compared to without the use of hold time. SPME significant factors was optimized with the use of multivariate analysis by employing Response Surface Methodology combine with Central Composite Design (RSM-CCD). The experimental design for RSM-CCD method was generated through Design-Expert version 12.0.3.0 (Stat Ease Software). From the result attained through CCD experiments, the value of coefficient of determination (R^2) for the total peak area of all of the compounds analysed was 0.8043. Extraction temperature, extraction time and salt addition at 60°C, 15 minutes and 45 %w/w respectively showed to be the best optimum conditions in extracting VOCs in stingless bee honey with desirability of 0.956.

Keywords: ANOVA, GC-MS, HS-SPME, *Tetrigona apicalis*, RSM-CCD, VOCs