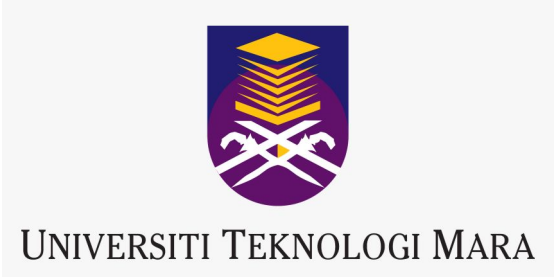

IDENTIFICATION OF HONEY VOLATILE COMPOUNDS BY GAS CHROMATOGRAPHY-MASS SPECTROMETRY (GC-MS) AND ITS BIOLOGICAL ACTIVITY

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ABSTRACT

IDENTIFICATION OF HONEY VOLATILE COMPOUNDS BY GAS CHROMATOGRAPHY-MASS SPECTROMETRY (GC-MS) AND ITS BIOLOGICAL ACTIVITY

The issue regarding the fate of honey products is emerging widely in the market. To differentiate between fate honey and original honey, the chemical and biological profiles of stingless bee honey are essential to fraud and to guarantee the authenticity of the honey. Thus, this study aimed to identify the volatile chemical compounds of stingless bee honey using Gas Chromatography-Mass Spectroscopy (GC-MS). In addition, this study assessed the total phenolic and flavonoid content, which were determined spectrophotometrically. Biological activities, such as the antioxidant and antimicrobial activity of the *Trigona itama* sample, were also assayed using 2,2-Diphenyl-1-picrylhydrazyl (DPPH) and minimum inhibitory and bactericidal concentrations assays. The GC spectral data revealed that the major chemical classes found in honey are ether and phenol. One of the major compounds is butylated hydroxytoluene. Besides, 7,15-dihydroxydehydroadiabetic acid and benzene-ethanamine are reported for the first time. The honey sample recorded high total phenolic content of 92.97±1.2 mg GAE/g extract and total flavonoid content of 98.77±0.85 mg QUE/g extract. In addition, the honey samples also exhibited strong antioxidant activities with IC_{50} of 4.62 µg/mL, comparable with the positive control, quercetin (IC_{50} = 1.59 µg/mL). The stingless bee honey also demonstrated moderate antimicrobial activity against all the tested bacteria, namely *Staphylococcus aureus* (SA), *Streptococcus pyogenes* (SP), *Escherichia coli* (EC), and *Pseudomonas aeruginosa* (PA). The findings from this study indicated that stingless bee (*Trigona itama*) honey could be a strong antioxidant agent and a potent inhibitor of all tested bacteria, SA, SP, EC, and PA. The data obtained from this study will serve as basic information for further product development and increase the commercialization potential.

CHAPTER 1

INTRODUCTION

1.1 Background of the study

Honey contains a lot of antioxidants. Acting as a natural antioxidant against reactive oxygen species, it can aid in preventing cell damage or harm. Antioxidant qualities differed between types of honey from different countries and geographical regions. The chemical composition of honey, notably the existence of phenolic and flavonoid molecules, minimizes oxidative processes or free radicals within food systems and human health. The amount and arrangement of hydroxyl groups in the molecules of interest determine their potential as an antioxidant.

The antimicrobial effects of honey have long been recognized in traditional medicine. The chemical composition of honey, which includes hydrogen peroxide as well as other non-peroxide compounds, is directly responsible for its healing properties. Despite actuality, cerumen, a substance made by stingless bees, also has antioxidant qualities (Giampieri *et al*., 2012). The polar extract of cerumen has been demonstrated to decrease linoleic acid catabolism, demonstrating a powerful antioxidant action that can prevent lipid peroxidation and protect cell membrane integrity. Furthermore, based on employing a human erythrocyte model, it was demonstrated that the ethanol extract of cerumen has antioxidant capabilities that lower the

CHAPTER 2

LITERATURE REVIEW

2.1 Chemical profiles

2.1.1 Chemical compounds in stingless bee honey

Honey contains about 200 substances which consist mainly of sugars, water, and other substances such as proteins, organic acid, a variety of vitamins, minerals, pigments, and phenolic compounds (Escuredo and Seijo 2019: da Silva *et al.,* 2016). Volatile compounds in honey come from various sources, including the transference of volatile compounds from the plant. Honeybees, on the other hand, have the ability to generate or transform plant elements into other volatile compounds. More than 400 different chemical compounds have been identified in the volatile fraction of honey, some of which are used as commercial honey markers, such as 3,9-epoxy-1-mentadieno (**1**) and cis-rose (**2**), which have been proposed as markers for lemon honey; diketones, sulphur compounds, and alkanes are characteristic of eucalyptus honey; and hexanal and heptanal are the main compounds in the aroma of lavender honey. Sugars, proteins, amino acids, vitamins, organic acids, phenolic compounds, and volatile compounds are among the chemical substances found in honey (da Silva *et al*., 2013). Honey can transform and degrade into new products such as furans, amino acids, alcohols, new phenolic compounds, and new volatile compounds as a result of processes such as the specific enzymes