

CHAPTER I

INTRODUCTION

Honey is a thick, sweet liquid produced by bees from the nectar of flowers (Al-Waili, 2004). The ancient Egyptians observed that honey not only delicious food, but also pointed out its use as a healing substance. Honey has a unique nutritive composition including carbohydrates, water and more than 180 substances including amino acids, vitamins, minerals and enzymes (Ghoshdastidar & Chabrabarti, 1992; Weston & Brocklebank, 1999). Honey is a product that carry spores of yeast, mold and bacteria, which may persist in it (Al Hindi, 2005). Honey can be used as an ingredient in food or in nutraceutical, cosmetic and as a medicine. The origin, chemical and microbiological composition play an important role on the quality and characteristic of honey (El-Sherbiny & Rizk, 1979).

Honey is able to inhibit the growth of microorganisms such as viruses, spoilage fungi, pathogenic bacteria and yeasts. The antibacterial properties of some type of honey were well reported, especially against Gram-positive bacteria (Bogdanov, 1997; Molan, 1997; Tan *et al.*, 2009; Rozaini *et al.*, 2004). Both bactericidal and bacteriostatic effects of honey have been reported for many strains especially pathogenic bacteria as reported by Bogdanov *et al.* (2008). *Burkholderia cepacia* that causes chronic granulomatous disease and pulmonary infections such as wound infections and urinary tract infections in hospitalized patients was sensitive to Manuka honey at low concentrations. It is suggested

that clinical applications for treating *B. cepacia* with honey including honey aerosols as an assistant to antibiotic topically and therapy for wounds and infections (Cooper, *et al.*, 2000a & b). Honey contains cenicamic acid and some flavonoids which have been approved for antibacterial applications (Rahman *et al.*, 2010).

Some of the published papers related to the antimicrobial activity of honey as follows: isolation and identification of phenolic acids in Malaysian honey with antibacterial properties (Aljadi & Yusoff, 2003); antibacterial activity of local Malaysian honey (Tumin *et al.*, 2005); the antibacterial properties of Malaysian Tualang honey against wound and enteric microorganisms (Tan *et al.*, 2009); antibacterial activity of propolis and honey against *Staphylococcus aureus* and *Escherichia coli* (Rahman *et al.*, 2010); antibacterial properties of Tualang honey and its effect in burn wound management: a comparative study (Nasir *et al.*, 2010); Aween *et al.*, (2012a & b) isolated *Lactobacillus acidophilus* from Malaysian, Libyan, New Zealand and Saudi Arabia honey and showed good inhibition activity against Gram-positive and Gram-negative pathogenic bacteria.

Currently, work on Libyan honey was reported by Aween *et al.* (2012a & b) with the title “Evaluation of antibacterial activity of *Lactobacillus acidophilus* strains isolated from honey and “Antibacterial activity of *Lactobacillus acidophilus* strains isolated from honey marketed in Malaysia against selected multiple antibiotic resistant (MAR) Gram-positive bacteria”.

Peptides isolated from sources such as milk, plants, trees, propolis, bee pollen and bees are reported to have antimicrobial activities (James *et al.*, 1996; Douglas *et al.*, 2003;

Broekaerta *et al.*, 2010; Mukesh, 2011; Vriens *et al.*, 2014; Fadaei, 2012; Wilsona *et al.*, 2012; Hayes *et al.*, 2006). Since amino acids are reported presence in honey, it may be possible that honey also may contain peptides that could function as antimicrobial agents.

Therefore, the aims of this study were to elucidate the antibacterial activity of diluted honey available in Malaysia namely, Tualang (Malaysia), Manuka (New Zealand), Kharroob (Libya), Alseder (Libya) and Acacia (Malaysia) and, to ascertain the possible contribution of proteins or peptides in the antibacterial activity of honey.

The objectives of this study were:

1. To determine the antibacterial activity of honey samples against pathogenic bacteria using disc diffusion method, well method, plate count assay, micro-titer plates and nanophotometer assay.
2. To characterize the antibacterial activity of honey in different conditions of pH, heat and enzymatic treatments.
3. To detect, isolate and purify proteins from honey samples.
4. To characterize the isolated proteins from honey using SDS-PAGE, RP-HPLC and Gel filtration (Sephadex column) assay.
5. To identify the antibacterial peptides fractions from Sephadex chromatography using LC MS-MS.