CHAPTER 2

Literature Review

2.1 Introduction

Stingless bees together with honey bees are eusocial insects. Stingless bees are pan-equatorial and can be found in most tropical or subtropical regions of the world such as Australia, Africa, Southeast Asia, meso-America and South America. Stingless bees belong to order Hymenoptera, family Apidae and tribe Meliponini (Michener, 2000). They are more than 500 described species in 32 genera with probably more than 100 species yet to be described (Michener, 2013). The greatest diversity of meliponines is found in the Neotropics of Central and South America with 400 described species. The remaining species are found in Africa and Indo-Australian regions (Cortopassi-Laurino *et al*., 2006; Michener, 2013; Pauly *et al*., 2013). Rasmussen (2008) described 32 species in Thailand contained in 10 genera which are shown in Table 2.1

Table 2.1 Stingless bee taxa in Thailand

Genus	Species
Geniotrigona	*Geniotrigona lacteifasciata* (Cameron, 1902)
	Geniotrigona thoracica (Smith, 1857)
Heterotrigona	*Heterotrigona itama* (Cockerell, 1918)
Homotrigona	*Homotrigona aliceae* (Cockerell, 1929)
	Homotrigona fimbriata (Smith, 1857)
	Homotrigona lutea (Bingham, 1877)
Lepidotrigona	*Lepidotrigona doipaensis* (Schwarz, 1939)
	Lepidotrigona flavibasis (Cockerell, 1929)
	Lepidotrigona nitidiventris (Smith, 1857)
	Lepidotrigona terminata (Smith, 1878)
	Lepidotrigona ventralis (Smith, 1857)

Table 2.1 (continued)

Genus	Species
Lisotrigona	*Lisotrigona cacciae* (Nurse, 1907)
	Lisotrigona furva (Engel, 2000)
Lophotrigona	*Lophotrigona canifrons* (Smith, 1857)
Pariotrigona	*Pariotrigona pendleburyi* (Schwarz, 1939)
Tetragonilla	*Tetragonilla atripes* (Smith, 1857)
	Tetragonilla collina (Smith, 1857)
Tetragonula	*Tetragonula fuscobalteata* (Cameron, 1908)
	Tetragonula geissleri (Cockerell, 1918)
	Tetragonula hirashimai (Sakagami, 1978)
	Tetragonula iridipennis (Smith, 1854)
	Tetragonula laeviceps (Smith, 1857)
	Tetragonula melina (Gribodo, 1893)
	Tetragonula pagdeni (Schwarz, 1939)
	Tetragonula pagdeniformis (Sakagami, 1978)
	Tetragonula reepeni (Friese, 1918)
	Tetragonula siridhornae (Michener & Boongird, 2004)
	Tetragonula testaceitarsis (Cameron, 1901)
	Tetragonula valdezi (Cockerell, 1918)
Tetrigona	*Tetrigona apicalis* (Smith, 1857)
	Tetrigona melanoleuca (Cockerell, 1929)
	Tetrigona peninsularis (Cockerell, 1927)

From: Rasmussen et al. (2008)

2.2 Life cycle of stingless bees

The life cycle of stingless bees is in numerous ways different from honey bees. In stingless bees, there can be two or more queens laying eggs in the same nest, although monogyny is the most common state (Bradbear, 2009). New queens are developed repeatedly, but most of them are not allowed to reach maturity. Some queens may live for 3-7 years and the replacement of the egg-laying queen does not happen every year. All stingless bees follow a mass-provisioning strategy in caring for their brood. The queen lays eggs in a specific way. First, a completed brood cell is half filled with honey and pollen by the workers. Then, one or more workers lay a trophic egg in the cell and the queen is encouraged to come up. Afterwards, the queen eats the worker egg and lays her own egg, and then continues to another cell. The workers close the cell by bending the upper collar of the cell against the center. The cell remains closed until the adult bee emerges. Stingless bee queens will lay between 10-100 cells with eggs a day, again according to the bee species. Following adult bee eclosion from brood cell, the cell is torn down, and the cerumen material is reused for building new cells. Fertile eggs from the queens develop into worker bees and queens. Drones come from unfertilized eggs from the queen, or from egg laying workers (Bradbear, 2009; Engels & Imperatriz-Fonseca, 1990).

Stingless bees multiply at the colony level by swarming. When a colony has reached a certain size and an appropriate new nest site is discovered, some worker bees will begin transporting building materials to the new location. More and more bees will fly to the new nest over the next few days, and eventually, a new queen from the old nest will transfer to the new nest and begin producing eggs (Bradbear, 2009; Wilson 1971).

In most species, mating between a new queen and drones takes place outside the nest (Hartfelder *et al*., 2006; Wilson, 1971), via swarms of flying males congregating outside of a colony possessing a virgin queen. Stingless bees differ from species to species in many aspects, one of which is the size of the worker bees; sizes range from two millimeters to slightly bigger than a honey bee. A stingless bee colony can contain from as few as a hundred to more than a hundred thousand bees (Bradbear, 2009).

2.3 Meliponiculture

In the Mesoamerican region there is a long tradition of stingless bee beekeeping compared to other regions of the world. This is because of the diversities of meliponines found in Tropical America (Obiols & Vasquez, 2013). Stingless bees have been known for their honey, pollen and wax (cerumen) production. Stingless bee beekeeping is known as meliponiculture. This activity, generally approached by traditional communities, has local characteristics according to regional differences in traditional knowledge and the targeted stingless bee species. This has been observed in many countries such as Mexico, Costa Rica, Guatemala, French Guiana, Venezuela, Argentina, Brazil, Africa and Australia (Aguilar *et al*., 2013; Ayala *et al*., 2013; Cortopassi-Laurino *et al*., 2006; Halcroft *et al*., 2013; Obiols & Vasquez 2013; Pauly *et al*., 2013; Pedro & Camargo 2013; Roig-Alsina & Gennari, 2013). Meliponiculture in Thailand can be described as an emerging stage, mostly practiced in Chanthaburi and Trat provinces the Southeast region of Thailand. Not all species can be managed which is strongly influenced by their nesting habitat. The most common species are the *T. laeviceps* and *T. pagdeni* species complexes which are opportunistic in selecting nest cavities and adaptable to human-made hives. During the past 20 years, the numbers of stingless bee beekeepers has been increasing. Approximately 700 meliponiculturists with approximately 5,000 hives support the commercial sale of stingless bee products, colonies and colonies managed for pollination (Chuttong *et al*., 2014). Even though 32 species are described in Thailand there are very few published researches on stingless bee and stingless bee honey.

Stingless bees store honey in pots, not in combs *as per* honey bees (Oddo *et al*., 2008). Although the amount of honey from a stingless bee colony cannot compete with the honey bee, the honey from stingless bees is valued more highly due to its relative scarcity. Honey produced by stingless bees is most often marketed as a natural medicinal remedy for a variety of ailments. This specific market niche makes the value of stingless bees honey often 20 times the value of *A. mellifera* honey (Vit *et al*., 1998). Abundant medical uses are attributed to Meliponini honey (Cortopassi-Laurino *et al*., 2006; Souza *et al*., 2006; Vit *et al*., 2004).

2.4 Stingless bee honey

Relative to honey from the western honey bee *A. mellifera,* there is a paucity of knowledge, lack of a historic research base and published research concerning stingless bee honey. The characterization of stingless bee honey has been the subject of only a few studies compared to the comprehensive documentation on *A. mellifera* honey. None of stingless bee honeys are included in the international standards for honey and it is not controlled by the food control authorities, in part due to the absence of official quality standards. The International Honey Commission is considering establishing quality standards for stingless bee honey (Codex, 2001; Souza *et al*., 2006).

In generally, honey is a very complex product composed of major compounds including monosaccharides, such as glucose and fructose, disaccharides, polysaccharides and minor components such as amino acids, enzymes, vitamins and minerals. The composition of honey is highly dependent on its floral source; however, certain external factors and processing also play a role. The quality of honey is mainly determined by its sensory, chemical, physical and microbiological characteristics (Iglesias *et al*., 2006). Honey has been used since ancient times mainly as a sweetening agent. Additionally, a wide and diverse range of therapeutic activities such as its antimicrobial and antioxidant properties, are known (Guerrini *et al*., 2009; Vit *et al*., 2004).

The composition of *A. mellifera* honey changes with time, which leads to darkening and loss of aroma and flavor (Sancho *et al*., 1992). The color of honey is one of the factors determining its price on the world market, and also its acceptability by consumers (Gonzales *et al*., 1999). The rate of darkening has been related to the composition of honey and of the storage temperature.

Castro-Vazquez, Diaz-Maroto, Gonzalez-Vinas, De La Fuente & Perez-Coello (2008) comment that the quality of the honey decreases with the time in storage and temperature exposure. Higher temperature exposures can have a significant effect on the chemical composition and organoleptic characteristics of *A. mellifera* honeys.

Most research work on stingless bee honey has been done in Neotropics because there it is so rich in stingless bee species and historical usage. Camargo, Pedro, Moure, Urban, & Melo (2007) report 391 species in 32 genera found in Neotropics. In Brazil 236 species are recorded (Crane, 1992). In Venezuela and French Guiana more than 80 species of stingless bee are described and 18% have had their honey analyzed (Pauly *et al*., 2013; Pedro & Camargo, 2013) and 46 species of stingless bee are found in Mexico (Ayala *et al*., 2013). Former works of stingless bee honey physicochemical characteristics have been done in several countries in Central and South America. *Melipona favosa* is the most common species of stingless bee in Venezuela and the physicochemical analyses and sensory testing of this honey has been done (Bogdanov *et al*., 1996; Vit *et al*., 1994; Vit *et al*., 1998; Vit, 2013). Almeida-Muradian, Stramm, Horita, Barth, Silva & Estevinho (2013) compared *Tetragonisca angustula* honey physicochemical properties to *A. mellifera* honey from Brazil. Silva *et al.* (2006) also examined the physicochemical of *M. subnitida* honey from Brazil. In Columbia 8 species of stingless bee honey composition (moisture and carbohydrates) have been analyzed (Fuenmayor *et al*., 2013). Additionally, Fuenmayor *et al*. (2013); Torres, Garedew, Schmolz & Lamprecht (2004) also studied the physicochemical properties and antimicrobial activity of *Tetragonisca angustula* honey from Colombia. In Argentina *Tetragonisca angustula* and *Plebeia wittmanni* honey have been studied for their physicochemical properties and antibacterial activities (Sgariglia *et al*., 2010). Additionally the chemical and functional profiles of Ecuadorian stingless bee honey have been analyzed (Guerrini *et al*., 2009). Ferrufino & Vit (2013) evaluated the chemical composition and conducted microbiological analysis on 6 species of stingless bee honey from Bolivia. Ten species of Peruvian stingless bee honey have been analyzed for physicochemical, biochemical, antibacterial and antioxidant activity (Rodriguez-Malaver *et al*., 2009). In Guatemala, there are 32 species of stingless bees known to produce honey. The physicochemical, antimicrobial, sensory and pollen composition of 4 species of stingless bee honey have been analyzed (Dardon *et al*., 2013). In Costa Rica 6 species of stingless bee honey were studied regarding antimicrobial activity and microbiological safety (Zamora *et al*., 2014). A recent summary of stingless bee honey physicochemical composition of *ca.* 32 species from Central and South America is found in Vit, Pedro & Roubik (2013).

For Indo-Australian stingless bee honey physicochemical there is a scarcity of research. Previous research of stingless bee honey focused on the species of stingless bee involved in ‘commercial’ meliponiculture. Oddo *et al*. (2008) examined *Tetragonula carbonaria* honey from Australia compared to *A. mellifera* honey. This study found that stingless bee honey is higher in moisture content, electrical conductivity and free acidity; lower in enzyme activity and reducing sugars.

In Thailand some research work has been done with Thai stingless bee honey Sawattham, Vaithanomsat & Tadakittisarn (2009) revealed the compositions of *A. mellifera* honey and compared this analysis with three species of stingless bee honey (*T. pagdeni, T. laeviceps and L. terminata*). The results showed that the moisture content of stingless bee honey varied from 22.00 to 25.60% which was higher than those found in *A. mellifera* honey, while the total sugar (ºBrix) content of stingless bee honey was lower than honeys *A. mellifera* honey. The proportion of the mono-saccharides glucose and fructose of stingless bee honey was higher than found in *A. mellifera* honey and 15 amino acids were detected in higher contents in stingless bee honey than those from *A. mellifera* honey. Suntiparapop, Prapaipong & Chantawannakul (2012) studied the chemical and biological properties of *T. laeviceps* honey compared to *A. mellifera* honey and as others, found that *T. laeviceps* honey has a higher moisture content, higher acidity and lower pH.

The previous research states that stingless bee honey when compare to *A. mellifera* honey presents a higher moisture content, electrical conductivity and acidity, but is lower in diastase activity. The carbohydrate content in similar to *A. mellifera* but frequently lower in reducing sugars and often possessing more maltose. Organoleptically stingless bee honeys possess a sour-sweet (acidic) taste.