Original Article

Journal of Apiculture 32(4) : 359~362(2017)
DOI: 10.17519/apiculture.2017.11.32.4.359

Biometric Studies of the Stingless Bee *Tetragonula laeviceps* Complex (Apidae: Meliponini) from Northern Thailand

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(Received 1 October 2017; Revised 22 November 2017; Accepted 22 November 2017)

Abstract

Biometrical information for five colonies of the *Tetragonula laeviceps* complex from Chiang Mai province, Thailand, was collected. The examination was conducted from September to November 2016. The range of colony age was one to five years. Results reveal an average number of queens (2.0±0.7), workers (5,823.6±2,703.3), drones (349.8±701.0), brood cells (9,911.0±4,795.3), honey pots (433.0±312.4) and pollen pots (711.8±305.9). The average size of a honey pot was 6.1 ±1.0mm in width and 7.3±1.1mm in height. The average size of a pollen pot was 6.4±0.3mm in width and 7.7±0.4mm in height. The results show intraspecific variation within the species that is the most common stingless bee species used in Thai meliponiculture. These observations support the contention that *T. laeviceps* complex comprises a cryptic species.

Key words: *Tetragonula laeviceps*, Stingless bees, Colony biometrics, Cryptic species

INTRODUCTION

Considering the number of described stingless bee species in the Indo-Malayan/Australasian region to be 89 (Rasmussen 2008), the colony bionomics of only a relatively few species have been investigated. The number of meliponine species in Thailand is stated to be 32 (Rasmussen 2008). An early report of nest architecture and colony composition of *Tetragonula laeviceps* is that of Sakagami *et al*. (1983) who examined colonies in Sumatra (Indonesia). A more recent report is that of Chinh *et al*. (2004) from a study of *T. laeviceps* in Vietnam.

Tetragonula laeviceps is one of the most widely distributed and most commonly used stingless bee species in Thai meliponiculture (Chuttong *et al*., 2014). *Tetragonula laeviceps* is opportunistic in nest site selection often utilizing anthropogenic cavity structures which is a primary factor for its incorporation into stingless bee beekeeping. The taxonomic placement of *T. laeviceps* is in question and is considered to be a cryptic species complex (Michener 2007, Khamyotchai *et al*., 2015). The first taxonomic description of *T. laeviceps* is from Singapore (Smith, 1859). Due to the geographical breadth of the species and varying architectural features of nests, it can be assumed, as stated by Rasmussen (2008), species limits are uncertain.

The purpose of the work we report is to elucidate *T. laeviceps* complex colony bionomics as observed in northern Thailand with managed colonies.

MATERIALS AND METHODS

We obtained five colonies of *T. laeviceps* complex from local meliponiculturists in the northern Thai province of

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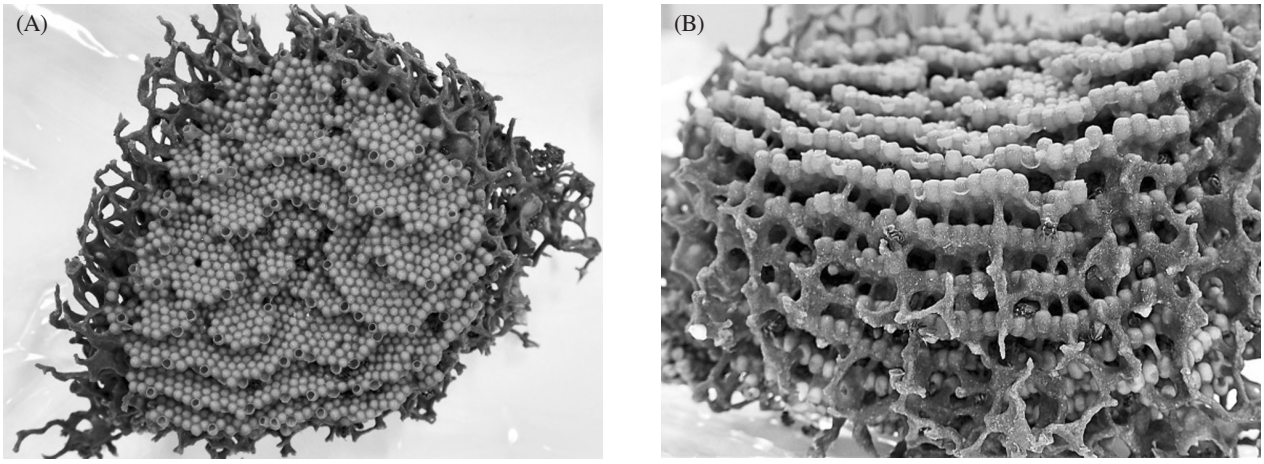

Fig. 1. Brood cells of *Tetragonula laeviceps* complex; top view (A), side view (B).

Table 1. Individual colony metrics

Colony No.	Number of					
	Workers	Males	Queens	Brood cells	Honey pots	Pollen pots
1	3,395	3	2	5,530	420	716
2	4,153	36	2	5,135	85	1,212
3	10,303	1,603	3	16,633	768	645
4	6,084	75	2	12,085	749	602
5	5,183	32	1	10,172	193	384
Average	5,823±2,703	349±701	2±0.71	9,911±4,795	443±312	712±306

Chiang Mai. The colonies varied in age from 1 to 5 years. While there was no standard unit size, our colonies were housed in 'hives' of *ca*. nine liters in volume. Colonies were euthanized by freezing prior to the careful dissection of colony matrices. We measured the following parameters: number of adult workers, males, and queens were counted under microscope, number of brood cells, pollen and honey pots were counted individually, and their respective volumes were measured by the digital caliper. Colony examinations took place during the late wet season of 2016 (September-November) at the Meliponini and Apini Research Laboratory, Postharvest Technology Research Center on the Chiang Mai University campus, Chiang Mai, Thailand.

RESULTS AND DISCUSSION

From the summaries (Tables 1 and 2) there are considerable differences shown in the total number of workers per colony, with the most populous colony (#3) being the oldest examined colony (5 years of age) with 3 times more workers than the smallest population seen in colony #1 (2.5 years of age). Our average of 5,824 workers is 5 times larger than the biggest colony worker population reported by Sakagami *et al*. (1983) in Sumatra, but is comparable to an average worker population of 4,221 for colonies examined in Vietnam (Chinh *et al*., 2004).

The number of adult male bees was the statistic with the greatest variability, with a range of 3 males in a population of 3,400 adults (colony #1) to 1,603 males in a population of 11,909 adult bees, where the males comprised 13.5% of the adult population (colony #3). When compared with the work of Sakagami *et al*. (1983) who found male populations to range from 0 to 10% of the adult population, our results are very similar.

Four of the five examined colonies possessed more than one queen with colony #3 possessing 3 queens. Examination of the 4 multiple queen colonies revealed that in

Table 2. Average size and volume of honey pots, pollen pots and brood cells

Measures	Width (mm)	Height (mm)	Volume (mm^3)
Honey pots	6.06±0.96	7.30±1.05	140.29
Pollen pots	6.41±0.25	7.74±0.39	166.40
Brood cells	2.44±0.15	3.31±0.14	10.32

Table 3. Size of honey pots and brood cells in comparison to other *T. laeviceps* studies

	Size of honey pots (mm)		Size of brood cells	References
	Mean±SD	Range	Range	
Width (mm)	6.1±0.1	5.0-7.1	2.3-2.6	This study
	7.8±1.6	5.0-11.0	~3.0	Sakagami *et al*., 1983
	NR	5.0-7.0	NR	Chinh *et al*., 2004
Height (mm)	7.3±1.1	6.2-8.3	3.1-3.5	This study
	9.4±1.6	7.0-14.0	~3.9	Sakagami *et al*., 1983
	NR	12.0-15.0	4.5-5	Chinh *et al*., 2004

each instance only one queen was in a reproductive state exhibiting physiogastry, while the other queens (non-physiogastric) are assumed to be daughter virgin queens.

The average number of brood cells was 9,911 with an inter-colony range of 5,135 to 16,633. The brood cell to worker bee ratio ranged from 1.99 to 1.24 with an average of 1.69. The brood combs were in stacked layers, in what could be described as a spiral-like configuration with the layers being vertically separated by short pillars. Sakagami *et al*. (1983) characterizes the brood cells examined in Sumatra as amorphous clusters as does Chinh *et al*. (2004) for those examined in Vietnam. Such is not the case with the colonies we examined as shown in Fig. 1. Our results, for the size of individual brood cells (2.44mm in width by 3.31mm in height), is in contrast to the report of Chinh *et al*. (2004) which describes brood cells with a width of 2.4 and being *ca*. 4.5-5mm in height which makes them considerably greater in volume than those we observed in northern Thailand.

Our measures for honey pot height results in an average of 7.3mm, which differs rather sharply with the height given by Chinh *et al*. (2004) of 12-15mm.

Food reserves were calculated from the average volumes of honey and pollen pots multiplied by the average number of honey and pollen pots per colony. For our five colonies that calculation results in an average honey store of 62ml of honey and a pollen volume of 118ml. The colony with the most honey pots (768 pots) would hypothetically contain ~107ml of stored honey. The largest of 3 colonies examined by Sakagami *et al*. (1983) possessed 21ml of honey.

The results of our examination show some interesting differences from previous published research of *T. laeviceps* colony biometrics (Sakagami *et al*., 1983 and Chinh *et al*., 2004) most especially as concerns the size of honey pots (see Table 3). Additionally, the structure of the brood nests for all our examined colonies, as mentioned, was in a layered, spiral-like form, which is in contrast to an amorphous aggregation of brood pots reported by both Sakagami *et al*. (1983) and Chinh *et al*. (2004). The reported brood cell dimensions from these two previous reports also are in contrast to our findings (see Table 3).

Our observation of 4 of 5 colonies possessing multiple queens aligns with previous studies for *T. laeviceps* as well as other stingless bee species. In stingless bees, queens can be produced all year round. The number of queens depends on the species and condition of the colony. Productive colonies with sufficient food storage may possess multiple queens (Bradbear, 2009; Engles and Imperatriz-Fonseca, 1990). The report of Chinh *et al*. (2004) found the number of queen cells in *T. laeviceps* colonies ranged from zero to seven and for *Lisotrigona carpenteri* from none to fourteen.

Adult worker specimens from our five examined

colonies were taxonomically assigned to the *T. laeviceps* complex by Dr. H. Banziger who is arguably the preeminent meliponine taxonomist in SE Asia. His identification is based on Rasmussen and Michener (2010).

Our observations concerning brood nest architecture and brood pot and honey pot dimensions in northern Thailand differ when compared to earlier work examining *T. laeviceps* from more southern latitudes. This fortifies the hypothesis that this group is a cryptic species complex. Future work utilizing molecular genetics will go far in unraveling the taxonomic conundrum that *T. laeviceps* represents.

ACKNOWLEDGEMENTS

We sincerely thank Dr. H. Bänziger of Chiang Mai University for the bee identifications. We also thank the following students from Maejo University for their yeoman's service in colony dissections; Ms. Benjawan Chaiyatip, Ms. Janjira Yavilat and Mr. Panuphan Sangjaroen. We thank Assist. Prof. Yaowaluk Chanbang of the Postharvest Technology Research Center, Chiang Mai University for generously providing the laboratory facilities.

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