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Dr. Pooja Singh

Ph.D., Consultant, National Bee Board, Ministry of Agriculture & Farmers Welfare, Govt. of India

Dr. MS Khan

Professor, GBPUAT, Pantnagar, Uttarakhand, India

Morphometric characterization of the stingless bees, *Tetragonula iridipennis* Smith (Hymenoptera: Apidae)

Dr. Pooja Singh and Dr. MS Khan

Abstract

Although, a good diversity of *Tetragonula iridipennis* is occurred in Northern Parts of the country, still very less studies have been conducted to explore their habitat and morphological features. Again, morphometry of drones of *T. iridipennis* is first time recorded in Temperate region/ Northern Parts of India. As by appearance, drone and worker bees look quite similar in appearance, the measurements of drone will help to distinguish them from worker bees. The studies will help in colonizing their feral colonies into hives and to design a suitable hives for their domestication.

Significant differences were found in various body parts of all three castes. Further, results showed an important characteristic distinguish feature in morphology/morphometry in three castes were that the drone has 10 segmented flagellum while worker and queen bees had 9 segmented flagellum. Another key difference was found in the shape and size (length and width) of the mandibles, which were bi-dentate in drone (0.40 ± 0.004 mm and 0.07 ± 0.001 mm) and tri-dentate in worker (0.533 ± 0.009 mm and 0.178 ± 0.003 mm) and queen bees (0.624 ± 0.049 mm and 0.154 ± 0.003 mm).

Keywords: *Tetragonula iridipennis*, penicillium, involucrum, cerumen

Introduction

Stingless bees are the smallest of the honey producing bees without venom apparatus and cannot sting. However, they do have well-developed mandibles by which they bite when an intruders disturbs the colony. Three distinct characters recognized for the identification of stingless bees viz., reduction of sting, presence of penicillium (a bunch of curved hairs on the outer corner of hind tibia) and reduction and weakness of wing venation. Stingless bees belong to the super family Apoidea, family Apidae and sub family Meliponinae. All Asian and African species of stingless bees belong to the tribe *Trigonini*. The various genera in this tribe include *Trigona*, *Plebeia*, *Tetragona* and *Nanotrigona* (Camargo *et al.*, 1988). Beekeeping with stingless bees is called meliponiculture (Crane, 1992).

Stingless bees, in a general way, build more complex nests than *A. mellifera* nests, although there are a great variety of forms, size and place of construction. In the construction of the brood comb, storage pots and involucrum, most species use cerumen, which is a mixture of wax and plant resin. Most stingless bees build their nests in empty trunks or in hollow branches. Some stingless bees construct underground nests using naturally abandoned ant nests and cavities under plant roots.

Different species are adapted to different tropical habitats but mostly they live at low altitudes. However, because of their predominance in the tropics, the biology of stingless bees has been far less explored in temperate regions (Sakagami *et al.*, 1983) [7]. As these bees are less explored in temperate regions, this become the objective of this study, which will help to find out the positional and opportunities of meliponiculture in temperate regions of India.

A total six species of *Trigona* are identified in India; these are *Trigona canifrons* Smith, *Trigona iridipennis* Smith, *Trigona atripes* Smith, *Trigona ventralis* Smith, *Trigona laeviceps* Smith and *Trigona ruficornis* Smith (Anonymous, 2011-13). The most common species of stingless bee found in India is *T. iridipennis*. *T. iridipennis* was redefined as the species belonging to India and Srilanka by Sakagami (1982) [6]. The generic name *Trigona* refers to their triangle shaped abdomen. The wings show iridescence hence the name iridipennis. They are sometimes called 'dammer bee' as they collect a kind of resin and mix it with wax called "cerumen" for constructing their nest along with wax produced from their body.

Corresponding Author:**Dr. Pooja Singh**

Ph.D., Consultant, National Bee Board, Ministry of Agriculture & Farmers Welfare, Govt. of India

The detailed study of morphology, habitation and diversity of *Tetragonula* spp. in India is still unexplored.

Material & Method

For the Present study samples were randomly collected from 8 hived colonies of the stingless bees, *Tetragonula iridipennis* Smith at Pantnagar, Uttarakhand (Table 1). Morphometric parameters of 50 eggs, 50 brood cells and 3 castes (50 adult workers, 50drones and 3 virgin queens) were studied under the stereoscopic binocular microscope and by taking accurate measurements with the help of digital Vernier Calipers and the differences were recorded and data was subjected to study the extent of variation within the population. The morphometry of drone genitalia and antennae of workers and drones were also studied by Scanning Electron Microscopy (SEM).

For recording measurements of brood cells, tooth prick sticks were placed on the existing brood comb in ten colonies. After 4 days, a batch of about 30-35 brood cells were formed on the tooth prick which were then carefully taken out from the brood combs. These tooth pricks along with the newly capped brood cells were kept in the position as it was in the brood comb, inside plastic petri dish containing slightly moist cotton layer with filter paper and immediately brought to the laboratory for further studies. After recording measurements on 50 brood cell dimensions, these cells were carefully opened from the top to expose the eggs and the food content. The eggs were taken out carefully with a soft horse hair needle for recording their length and width. The food content of individual brood cell was then measured with 10 μ l micro

capillaries soon after taking out the egg.

The adult worker bees were the forager bees and were collected from the entrance of the hived colonies in the glass vials which were later shifted to the insect killing jar before recording the morphometric data. The drone bees were collected with the help of insect net from the drone congregation area near the hived stingless bee colonies. The virgin queens were obtained from three queen cells formed in the hived colonies of the stingless bee. The queen cells were taken out from the brood comb carefully and kept in the petri dish in the laboratory (B.O.D. incubator at 27°C and 75% RH) till emergence of the queens. The measurement of various body parts of queen was recorded after 1 day after their emergence.

Various morphological parameters of *T. iridipennis* recorded and measured during the present investigations including the parameters given by Rasmussen (2013). The results were subjected to STPR-3 and STPR4 program for statistical analysis. The morphometric data were subjected to study the extent of variation in the population of *T. iridipennis* in Pantnagar.

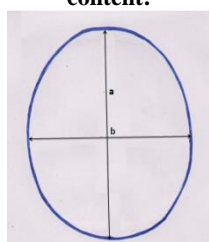
Measurements on following parameters were recorded (Rasmussen, 2013) and some more parameters were also added:

- Length and width of brood cell along with quantitative analysis of food content (Fig 1)
- Length and width of egg (Fig 2)
- Length of body of worker, drone and queen bee (Fig 3)
- Morphometry of cephalic (head) region and its appendages

Table 1: Particulars of the feral colonies of stingless bee, *T. iridipennis* Smith hived for experimentation at Pantnagar

S. N.	Shelter tree	Tree Trunk girth (ft)	Height from ground level (ft)	Length of colony (cm)
1.	<i>Cassia fistula</i>	3	3.5	46
2.	Pride of India (<i>Lagerstroemia indica</i>)	1.7	6.0	12
3.	Pride of India (<i>Lagerstroemia indica</i>)	2.0	4.2	16
4.	Pride of India (<i>Lagerstroemia indica</i>)	1.8	6.1	15
5.	Mango (<i>Mangifera indica</i>)	2.6	0.5	9
6.	Pride of India (<i>Lagerstroemia indica</i>)	1.6	4.5	42
7.	Gulmohar (<i>Poinciana regia</i>)	2.4	6.5	18
8.	Gulmohar (<i>Poinciana regia</i>)	2.8	7.2	52

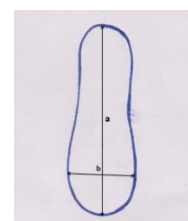
A. Length and width of brood cell along with quantitative analysis of food content:



a= Length of Brood cell
b= Diameter of Brood cell

Fig 1: Brood Cell

B. Length and width of egg:



a= Length of Egg
b= Width of Egg

Fig 2: Egg

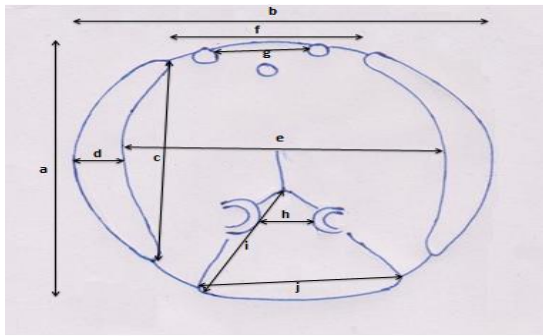
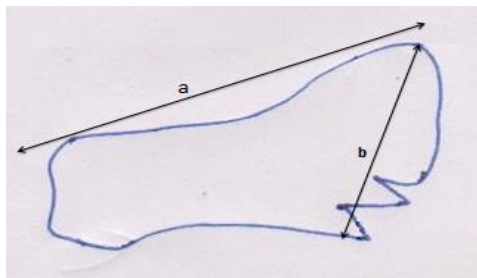


Fig 3: Head

- a= Length of Head
- b= Width of Head
- c= Length of Compound eye
- d= Width of Compound eye
- e= Distance b/w Compound eye (in front view)
- f= Distance b/w Compound eye (at dorsal view)
- g= Distance b/w dorsal ocelli
- h= Distance b/w antennal sockets
- i= Length of Clypeus
- j= Width of Clypeus



- a= Length of Mandible
- b= Diameter of Mandible

Fig 4- Mandible of worker and queen

- a= Length of Mandible
- b= Diameter of Mandible

Fig 5- Mandible of drone

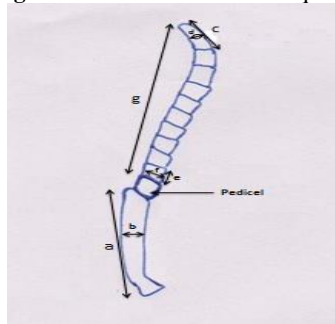
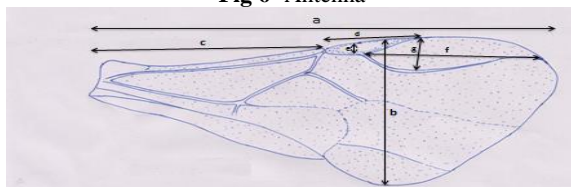


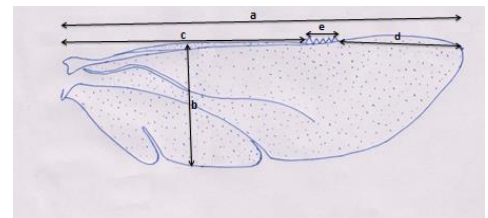
Fig 6- Antenna

- a= Length of Scape
- b= Diameter of Scape
- c= Length of 1st flagellomere
- d= Diameter of 1st flagellomere
- e= Length of last flagellomere
- f= Diameter of last flagellomere
- g= Length of flagellum



- a= Length of Fore wing
- b= Width of Fore wing
- c= Length of Pterostigma
- d= Length of Marginal cell
- e= Width of Marginal cell
- f= Length of Sub-marginal cell
- g= Width of Sub-marginal cell

Fig 7: Fore Wing



- a= Length of Hind wing
- b= Width of Hind wing
- c= Distance of Hamuli from wing base
- d= Distance of Hamuli from apical end
- e= Width of Hamuli

Fig 8: Hind Wing

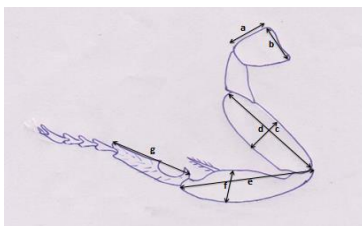


Fig 9: Fore LEG

- a= Length of Coxa
- b= Width of Coxa
- c= Length of Femur
- d= Width of Femur
- e= Length of Tibia
- f= Width of Tibia

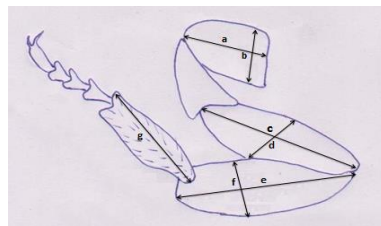


Fig 10: Middle LEG

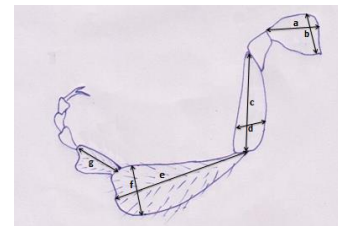


Fig 11: Hind LEG

g= Length of Basitarsus

Result & Discussion**Table 2:** Measurements (mm) of the various parameters examined in worker, drone and queen

Parameters	Measurements		
	Length (mm)		Width (mm)
Morphometry of Egg	0.929 ±0.067		0.35 ±0.031
Morphometry of Brood cells	Length (mm)	Width (mm)	Food content (µl) in brood cells
	3.602 ±0.080	2.628 ±0.100	
Length of body	Worker (mm) (n=50)	Drone (mm) (n=50)	Queen (mm) (n=3)
	3.85 ±0.124	3.62 ±0.048	7.05 ±0.825
Cephalic region and head appendages			
Length of head	1.308 ±0.027	1.327 ±0.052	1.38 ±0.02
Width of head	1.601 ±0.023	1.532 ±0.046	1.57 ±0.036
Length of compound eyes	0.975 ±0.038	1.092 ±0.042	1.10 ±0.187
Width of compound eye	0.389 ±0.025	0.430 ±0.041	0.254 ±0.025
Distance b/w compound eyes (in front view)	0.777 ±0.022	0.935 ±0.070	1.14 ±0.101
Distance of compound eyes at dorsal view	0.664 ±0.045	0.915 ±0.053	1.007 ±0.040
Distance between dorsal ocelli	0.144 ±0.015	0.290 ±0.045	0.294 ±0.015
Distance b/w antennal sockets	0.146 ±0.013	0.121 ±0.017	0.17 ±0.02
Length of Clypeus	0.569 ±0.035	0.614 ±0.050	0.50 ±0.02
Width of Clypeus	0.607 ±0.052	0.610 ±0.054	0.714 ±0.035
Length of mandible	0.533 ±0.040	0.40 ±0.022	0.624 ±0.085
Width of mandible	0.178 ±0.015	0.047 ±0.008	0.154 ±0.005
Length of scape	0.564 ±0.021	0.413 ±0.040	0.70 ±0.02
Diameter of scape	0.064 ±0.005	0.071 ±0.013	0.087 ±0.005
Length of 1 st flagellomere	0.068 ±0.004	0.112 ±0.016	0.124 ±0.006
Diameter of 1 st flagellomere	0.095 ±0.006	0.10 ±0.00	0.10 ±0.00
Length of last flagellomere	0.129 ±0.021	0.171 ±0.016	0.214 ±0.015
Diameter of last flagellomere	0.10 ±0.00	0.084 ±0.010	0.097 ±0.005
Length of flagellum	0.960 ±0.033	1.422 ±0.031	1.337 ±0.073
No. of segments in flagellum	9	10	9
Fore wing			
Length of Wing	3.557 ±0.079	3.451 ± 0.091	3.706 ±0.060
Width of Wing	1.295 ±0.031	1.337 ±0.052	1.194 ±0.075
Length of Pterostigma	0.578 ±0.040	0.610 ±0.050	0.47 ±0.065
Length of Marginal Cell	1.261 ±0.038	1.235 ±0.049	1.194 ±0.025
Width of Marginal Cell	0.243 ±0.023	0.272 ±0.026	0.184 ±0.011
Length of Sub-Marginal Cell	1.787 ±0.035	1.80 ±0.048	1.74 ±0.046
Width of Sub-marginal Cell	0.675 ±0.030	0.612 ±0.027	0.556 ±0.030
Hind wing			
Length of Wing	2.437 ±0.060	2.547 ±0.049	2.726 ±0.061
Width of Wing	0.673 ±0.041	0.692 ±0.027	0.73 ±0.026
Distance of hamuli from wing base	1.425 ±0.049	1.505 ±0.055	1.43 ±0.02
Distance of hamuli from apical end	0.729 ±0.020	0.718 ±0.029	0.706 ±0.015
Width of Hamuli	0.282 ±0.048	0.323 ±0.068	0.59 ±0.062
No. of Hamuli	5	5	5
Fore leg			
Length of Coxa	0.307 ±0.016	0.317 ±0.048	0.44 ±0.026
Width of Coxa	0.270 ±0.020	0.242 ±0.039	0.317 ± 0.015
Length of Femur	0.723 ±0.019	0.641 ±0.064	0.807 ±0.025
Width of Femur	0.174 ±0.015	0.145 ±0.033	0.257 ±0.015
Length of Tibia	0.659 ±0.031	0.651 ±0.062	0.746 ±0.045
Width of Tibia	0.172 ±0.009	0.149 ±0.033	0.21 ±0.02
Length of Basitarsus	0.401 ±0.043	0.388 ±0.050	0.504 ±0.032
Mid leg			
Length of Coxa	0.479 ±0.047	0.511 ±0.052	0.540 ±0.03
Width of Coxa	0.234 ±0.017	0.182 ±0.023	0.450 ±0.04
Length of Femur	0.815 ±0.042	0.787 ±0.074	1.034 ±0.032
Width of Femur	0.217 ±0.008	0.189 ±0.033	0.254 ±0.015
Length of Tibia	0.863 ±0.067	0.842 ±0.070	1.08 ±0.03
Width of Tibia	0.254 ±0.014	0.232 ±0.033	0.293 ± 0.041
Length of Basitarsus	0.485 ±0.037	0.503 ±0.033	0.517 ± 0.040
Hind leg			
Length of Coxa	0.481 ±0.055	0.467 ±0.047	0.580 ±0.07
Width of Coxa	0.392 ±0.029	0.351 ±0.026	0.513 ±0.025

Length of Femur	0.906 ±0.060	0.901 ±0.053	1.223 ±0.049
Width of Femur	0.232 ±0.018	0.178 ±0.026	0.306 ±0.045
Length of Tibia	1.359 ±0.085	1.362 ±0.117	1.527 ±0.058
Width of Tibia	0.510 ±0.026	0.453 ±0.047	0.476 ±0.025
Length of Basitarsus	0.530 ±0.055	0.467 ±0.034	0.664 ±0.047

All values are mean of fifty observations whereas for queen values are mean of three observations ± values are Standard Deviation (SD)

Egg

The eggs of *T. iridipennis* are whitish, translucent and cylindrical in shape having broader end at one side. The egg is laid in the center of provisioned brood cell. The broader side of egg is embedded about 1/3 of length in provision. The position of freshly laid egg is vertical inside the brood cell. The length and width of egg ranged from 0.83 to 1.05 mm (Mean: 0.929 ± 0.015 mm) and 0.30 to 0.41 mm (Mean: 0.35±0.007 mm). In an earlier study in Kerala, length and width of *T. iridipennis* egg were reported as 1.07 and 0.9 mm respectively (Anonymous, 2004).

Brood cells and quantitative analysis of food content

The brood cells of *T. iridipennis* were constructed in clusters (or like bunch of grapes) on pillars raised with resinous material on the bottom of the hive. Each brood cell was jointed to adjacent brood cell via 2 to 3 external connections. The newly constructed brood cells were dark brown which later become lighter in colour. The brood cell was destroyed and thrown out from the colony after emergence of adult bee from it. In this way, these bees use each brood cell only for a single time.

The length and width of brood cells ranged from 3.49 to 3.77 mm (Mean: 3.602±0.017 mm) and 2.48 to 2.82 mm (Mean: 2.628±0.022 mm), respectively. Freshly capped brood cells were carefully opened individually from top and food content of each cell was measured with the help of microcapillaris (10µl). The quantity of the food content (mixture of pollen and nectar) in brood cell ranged from 4.50 to 7.60µl (Mean: 6.045±0.235µl).

Length of body (Worker, Drone and Queen)

The body length of the worker bees and the drone bees ranged from 3.62 to 4.03 mm (Mean: 3.85±0.028 mm) and 3.54 to 3.70 mm (Mean: 3.62±0.011 mm), respectively. The body length of 3 newly emerged queen bees ranged from 6.39 to 7.98 mm.

Cephalic region and head appendages

The length of head is more in queen (1.38±0.027 mm)

followed by drone (1.327±0.052 mm) and worker (1.308±0.02 mm), respectively while the width of head of worker (1.601±0.023 mm) and queen (1.57±0.036mm) was found similar statistically. Drone had significantly less width of head (1.532±0.046 mm) in comparison to worker and queen.

Length of compound eyes was found non-significant in all three castes: worker (0.975±0.038mm), drone (1.092±0.042mm) and queen (1.1±0.187mm), while the width of compound eyes was more in drone (0.43±0.041mm), followed by worker (0.389±0.025mm) and queen (0.254±0.025mm), respectively.

Distance between compound eyes (in front view) was insignificant between queen (1.14±0.101mm) and drone (0.935±0.070mm) which were significantly higher than worker (0.777±0.022mm). Similarly, distance of compound eyes at dorsal view was again found insignificant between queen (1.007±0.040mm) and drone (0.915±0.053mm) but significantly less in worker (0.664±0.045mm).

Distance between dorsal ocelli was also found similar in queen (0.294±0.015mm) and drone (0.29±0.045mm). This distance was significantly higher than in worker (0.144±0.015mm). Distance between antennal sockets was found maximum in queen (0.17±0.02mm) followed by worker (0.146±0.013mm) and drone (0.121±0.017mm).

The length of clypeus differed significantly and was maximum in drone (0.614±0.050mm) followed by worker (0.569±0.035mm) and queen (0.50±0.02mm) while the width of clypeus was found insignificant in worker (0.607±0.052 mm) and drone (0.61±0.054mm) but significantly differed with queen (0.714±0.035mm).

The major difference was observed was that the mandibles of drone are bidentate while these are tridentate in worker and queen (Plate-1). The length and width of mandible was significantly differed in all three castes. Length of mandible was maximum in queen (0.624±0.085mm) followed by worker (0.533±0.040mm) and drone (0.40±0.022mm), while the width of mandible was maximum in worker (0.178±0.015mm) followed by queen (0.154±0.005mm) and drone (0.047±0.008mm).

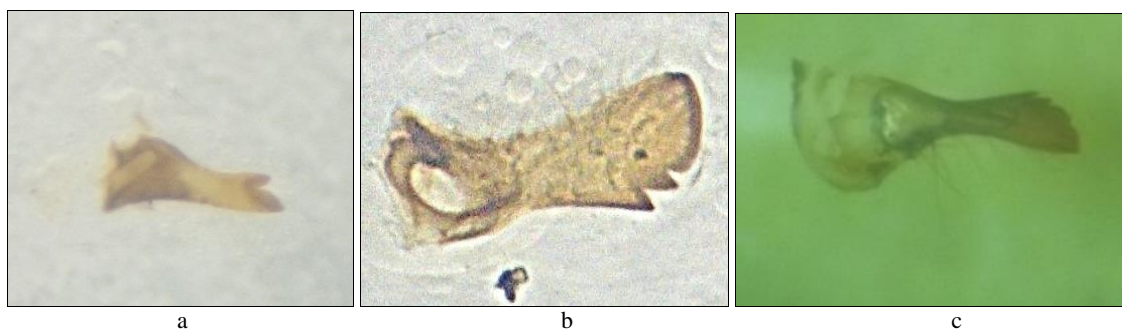


Plate 1: Mandible of *Tetragonula iridipennis* Smith a: drone (bidentate), b & c: worker and queen (tridentate)

Length and diameter of scape were significantly different in all three castes and both were maximum in queen. The length of scape of queen was maximum (0.70±0.02mm) followed by

worker (0.564±0.021mm) and drone (0.413±0.040mm). The diameter of scape was maximum in queen (0.087±0.005mm) followed by drone (0.071±0.013mm) and worker

(0.064 ± 0.005 mm).

Length of 1st flagellomere was insignificant in queen and drone with more length in queen (0.124 ± 0.006 mm) followed by drone (0.112 ± 0.016 mm) which were statistically higher than worker (0.068 ± 0.004 mm). Diameter of 1st flagellomere was almost similar in queen (0.10 ± 0.00 mm), drone (0.10 mm ± 0.00) and worker (0.095 ± 0.006 mm).

Length of last flagellomere was significantly differ in all three castes with maximum length in queen (0.214 ± 0.015 mm) followed by drone (0.171 ± 0.016 mm) and worker

(0.129 ± 0.021 mm), while the diameter of last flagellomere was insignificantly differ in all three castes with slightly more in worker (0.10 ± 0.00 mm) followed by queen (0.097 ± 0.005 mm) and drone (0.084 ± 0.010 mm).

Length of flagellum was found insignificantly differ in drone (1.422 ± 0.031 mm) and queen (1.337 ± 0.073 mm). The worker had significantly lesser length (0.96 ± 0.033 mm) as compared to drone and queen. Worker and queen had 9 segments in flagellum (Plate-2a) while drone had 10 segments in flagellum (Plate-2b).

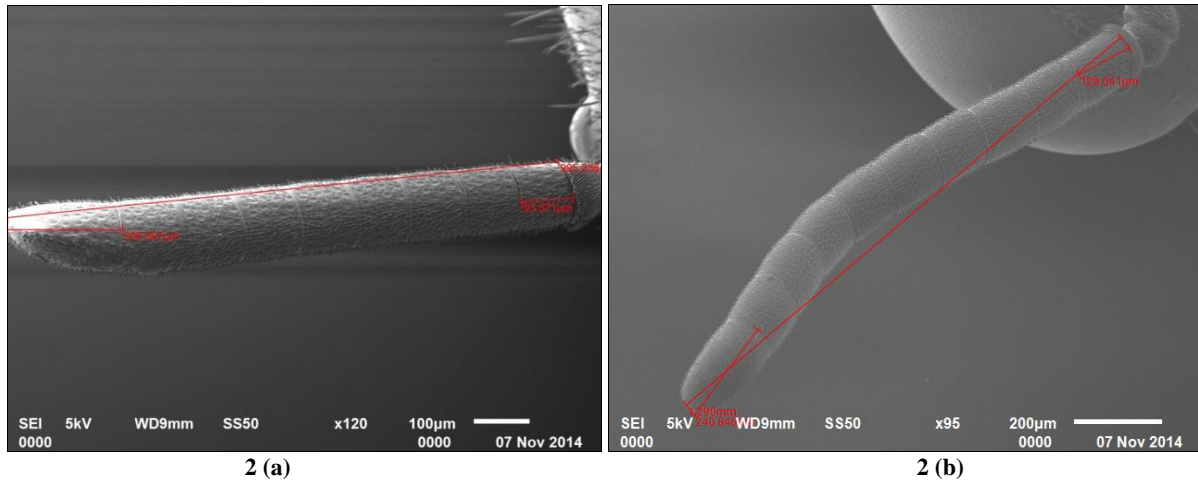


Fig 2: a) Antennae of stingless bee, *T. iridipennis* Smith worker bee and

b) drone bee

Thoracic appendages

Fore wing

Length and width of fore wing, both were insignificantly differed in worker and drone. Length of fore wing is maximum in queen (3.706 ± 0.060 mm) followed by worker (3.557 ± 0.079 mm) and drone (3.451 ± 0.091 mm). The width of fore wing is maximum in drone (1.337 ± 0.052 mm) followed by worker (1.295 ± 0.031 mm) and queen (1.194 ± 0.075 mm). Length of pterostigma was insignificantly differed in drone and worker which was maximum in drone (0.61 ± 0.050 mm) followed by worker (0.578 ± 0.040 mm) and queen (0.47 ± 0.065 mm).

The length and width of marginal cell was significantly differed in all three castes which was maximum in worker (1.261 ± 0.038 mm) followed by drone (1.235 ± 0.049 mm) and queen (1.194 ± 0.025 mm), while the width of marginal cell was maximum in drone (0.272 ± 0.026 mm) followed by worker (0.243 ± 0.023 mm) and queen (0.184 ± 0.011 mm). Length of sub marginal cell was insignificantly differ in all three caste and was slight more in drone (1.80 ± 0.048 mm) followed by worker (1.787 ± 0.035 mm) and queen (1.74 ± 0.046 mm), while width of sub marginal cell in worker (0.675 ± 0.030 mm) and drone (0.612 ± 0.027 mm) were similar but significantly more than queen (0.556 ± 0.030 mm).

Hind wing

The length and width of hind wing was significantly different in all three castes and was maximum in queen and minimum in worker. The maximum length in queen hind wing (2.726 ± 0.061 mm) was followed by drone (2.547 ± 0.049 mm) and worker (2.437 ± 0.060 mm), respectively and maximum width of queen hind wing (0.73 ± 0.026 mm) was followed by drone (0.692 ± 0.027 mm) and worker (0.673 ± 0.041 mm), respectively.

Distance of hamuli from wing base were similar in worker

(1.425 ± 0.049 mm) and queen (1.43 ± 0.02 mm) and was significantly less than in drone (1.505 ± 0.055 mm). Distance of hamuli from apical end were insignificantly differ in worker (0.729 ± 0.020 mm) and drone (0.718 ± 0.029 mm) but was significantly higher than in queen (0.706 ± 0.015 mm). The width of hamuli in three castes differed significantly which was maximum in queen (0.59 ± 0.062 mm) followed by drone (0.323 ± 0.068 mm) and worker (0.282 ± 0.048 mm), respectively.

Fore leg

Length of coxa in fore leg was insignificantly differ in worker and drone. Maximum length of coxa was present in queen (0.44 ± 0.026 mm) followed by drone (0.317 ± 0.048 mm) and worker (0.307 ± 0.016 mm), respectively. Width of coxa was significantly different in all three castes. It was again maximum in queen (0.317 ± 0.015 mm) followed by worker (0.27 ± 0.020 mm) and drone (0.242 ± 0.039 mm), respectively. Queen has maximum length and width of coxa and drone has minimum length and width of coxa.

Length and width of femur was significantly different among three castes. The corresponding values were again maximum in queen (0.807 ± 0.025 mm and 0.257 ± 0.015 mm) and minimum in drone (0.641 ± 0.064 mm and 0.145 ± 0.033 mm), respectively. The worker has intermediate length and width of coxa (0.723 ± 0.019 mm and 0.174 ± 0.015 mm).

Length of tibia was maximum again in queen (0.746 ± 0.045 mm) followed by worker (0.659 ± 0.031 mm) and drone (0.651 ± 0.062 mm), respectively. The width of tibia was again maximum in queen (0.21 ± 0.02 mm) followed by worker (0.172 ± 0.009 mm) and drone (0.149 ± 0.033 mm). Queen had maximum length and width of tibia among three castes. The length of basitarsus was also significantly higher in queen (0.504 ± 0.032 mm) than worker (0.401 ± 0.043 mm) and drone (0.388 ± 0.050 mm), respectively.

Middle leg

Length of coxa was maximum in queen ($0.54\pm 0.03\text{mm}$) followed by drone ($0.511\pm 0.052\text{mm}$) and worker ($0.479\pm 0.047\text{mm}$), respectively. Width of coxa was also maximum in queen ($0.45\pm 0.04\text{mm}$) followed by worker ($0.234\pm 0.017\text{mm}$) and drone ($0.182\pm 0.023\text{mm}$), respectively. Queen had maximum length and width of coxa.

Length and width of femur in middle leg was maximum in queen ($1.034\pm 0.032\text{mm}$ and $0.254\pm 0.015\text{mm}$) followed by worker ($0.815\pm 0.042\text{mm}$ and $0.217\pm 0.008\text{mm}$) and drone ($0.787\pm 0.074\text{mm}$ and $0.189\pm 0.033\text{mm}$), respectively. The length and width of tibia was also maximum in queen ($1.08\pm 0.03\text{mm}$ and $0.293\pm 0.041\text{mm}$) followed by worker ($0.863\pm 0.067\text{mm}$ and $0.254\pm 0.014\text{mm}$) and drone ($0.842\pm 0.070\text{mm}$ and $0.232\pm 0.033\text{mm}$), respectively. The length of basitarsus was statistically similar in queen ($0.517\pm 0.040\text{mm}$) and drone ($0.503\pm 0.033\text{mm}$) higher than in worker ($0.485\pm 0.037\text{mm}$).

Hind leg

Length and width of coxa was maximum in queen ($0.58\pm 0.07\text{mm}$ and $0.513\pm 0.025\text{mm}$) followed by worker ($0.481\pm 0.055\text{mm}$ and $0.392\pm 0.029\text{mm}$) and drone ($0.467\pm 0.047\text{mm}$ and $0.351\pm 0.026\text{mm}$), respectively. The length and width of femur was also maximum in queen ($1.223\pm 0.049\text{mm}$ and $0.306\pm 0.045\text{mm}$) followed by worker

($0.906\pm 0.060\text{mm}$ and $0.232\pm 0.018\text{mm}$) and drone ($0.901\pm 0.053\text{mm}$ and $0.178\pm 0.026\text{mm}$), respectively. There was insignificant difference in length of femur in worker and drone.

The length of tibia was maximum in queen ($1.527\pm 0.058\text{mm}$) followed by drone ($1.362\pm 0.117\text{mm}$) and worker ($1.359\pm 0.085\text{mm}$), respectively but the difference in length of tibia of drone and worker was insignificant. The width of tibia was maximum in worker ($0.51\pm 0.026\text{mm}$) followed by queen ($0.476\pm 0.025\text{mm}$) and drone ($0.453\pm 0.047\text{mm}$), respectively. The length of basitarsus was maximum in queen ($0.664\pm 0.047\text{mm}$) followed by worker ($0.53\pm 0.055\text{mm}$) and drone ($0.467\pm 0.034\text{mm}$), respectively.

Drone genitalia

Morphometry of drone genitalia was described as per description of genital apparatus given by Sakagami (1978) [6] for genus *Tetragonula*. The gonocoxite (Stipes), gonostylus (Volsella) and Penis valve (Sagitta) were dark brown and sclerotized in appearance. The penis was whitish translucent structure with bulbous base. The length and width (at base) of gonocoxite was $322.049\ \mu\text{m}$ and $344.072\ \mu\text{m}$, respectively at $\times 75$. The distance between both gonostylus in expanded stage was $1.117\ \text{mm}$. The distance between both Penis valve in expanded stage was $1.216\ \text{mm}$. Length of penis $513.387\ \mu\text{m}$ in expanded stage (Plate-3).

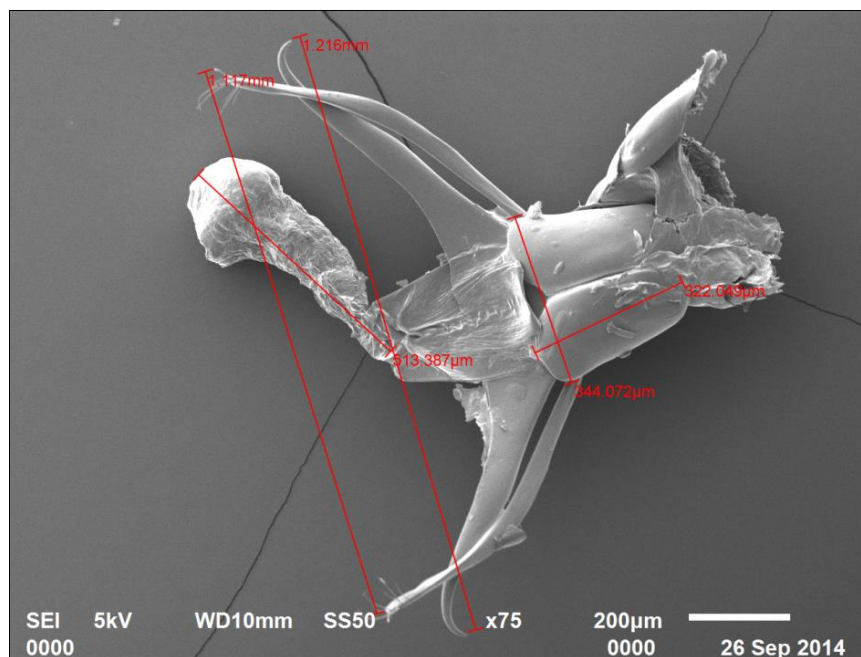


Plate 3: Genital apparatus (Genitalia) of drone of stingless bee, *T. iridipennis* Smith- SEM image

Conclusion

In the present study the major characteristics distinguish feature in morphology/morphometry of three castes were that the drone has 10 segmented flagellum while worker and queen bees had 9 segmented flagellum. Another key difference was found in the shape and size of the mandibles, which were bi-dentate in drone ($0.40\pm 0.004\ \text{mm}$ and $0.07\pm 0.001\ \text{mm}$) and tri-dentate in worker ($0.533\pm 0.009\ \text{mm}$ and $0.178\pm 0.003\ \text{mm}$) and queen bees ($0.624\pm 0.049\ \text{mm}$ and $0.154\pm 0.003\ \text{mm}$). These two are unique features which would help in easy and instantaneous identification of drones from workers.

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