Indian Journal of Entomology 86(1): 172-175 (2024) DoI. No.: 10.55446/IJE.2023.1651

MORPHOMETRICS OF INTRASPECIFIC POPULATIONS OF STINGLESS BEE *TETRAGONULA IRIDIPENNIS* SMITH FROM SOUTHERN TAMIL NADU

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ABSTRACT

Stingless bees also referred to as meliponine bees and morphological characteristics of these differ depending on are the species and environmental kinds. The current study explores morphology and morphometrics of *Tetragonula iridipennis* Smith in Tamil Nadu, India. The morphometrics of worker bees were compared over locations. The bees captured in the Kolli hills were significantly larger than those collected elsewhere. The mean value of head length (HL) (1.43 mm), head width (HW) (1.68 mm), antennal length (AL) (1.98 mm), hind leg length (HLL) (3.54 mm) and forewing length (FL) (3.84 mm) were recorded. The results revealed variance of four principal components (PC) reaching 82.4%, as (PC1) 38.58%, (PC2) 20.95%, (PC 3) 19.75% and (PC4) 8.10%. The characters of FL, HW, FW (forewing width), HLL, AL, OOD (ocello ocular distance), HTW (hind tibial width) and HL were the most discriminative in the principal component analysis.

Key words: Stingless bee, *Tetragonula iridipennis*, worker bees, morphometry, head length, hind leg length, forewing length, principal component analysis

Stingless bees are widely known as dammar bees and its native to eight described species in Indian subcontinent with the exception of higher elevations or the drier interior regions. (Rasmussen, 2013). *Tetragonula iridipennis* Smith has been identified as the dominant stingless bee species in Tamil Nadu (Swaminathan, 2000). Previous research on stingless bees has also been dealt with biology and foraging behaviour (Roopa, 2002), (Kuberappa et al., 2005) taxonomy and morphology (George, 1934; Sakagami 1978; Jobiraj and Narendran 2004; Danaraddi et al., 2012), biology and nesting behaviour (Danaraddi et al., 2009). The distribution of *T. irridipennis* has been documented in Kerala by Mohan and Devanesan (1999). One of the most extensively used classification systems for this taxa is morphometric taxonomy. Previously, stingless bees were classified based on their body size, number of hamuli, forewing, hindwing, and cephalic features. Stingless bees can be differentiated from corbiculate Apinae by combination of reduced fore wing venation with the presence of a jugal lobe on the hind wing (Rahman et al., 2013). Studies on the species diversity of *Tetragonula* in various regions of India are scarce. There is a lack of research findings regarding the diversity of stingless bee species in India, especially in southern districts of Tamil Nadu. In this research, the morphological and morphometric parameters of stingless bees were examined in different topographic regions of Southern Tamil Nadu.

MATERIALS AND METHODS

For the present study, twenty bee specimens were collected from 10 topographic locations viz., Alagar kovil 10.07’ N, 78.21’E, Madurai 9.96’ N, 78.20’ E, Megamalai 9.64’N, 77.40’E, Kadamalaigundu 9.53’ N, 77.29’ E, Kolli hills 11.24’N, 78.33’ E, Erumapatti 11.15’N, 78.29’ E, Sirumalai 10.19’ N, 77.99’E, Natham 10.50’ N, 78.10’E, Kodaikanal 10.29’N, 77.71’E and Batlagundu 10.16’N, 77.75’E. These specimens were preserved as dry and wet in 70% alcohol. The preserved specimens were treated with relaxing fluid (75% alcohol 106 ml, distilled water 98 ml, benzene 14 ml and ethyl acetate 38 ml.) for 2-4 hr. Using a Leica M 205 C stereozoom microscope preserved bees were dissected and fourteen important morphometric measurements were recorded viz. head length (HL), head width (HW), distance between two lateral ocelli (DBO), ocello-ocular distance (OOD), antennal length (AL), hind leg length (HLL), hind tibial length (HTL), hind tibial width (HTW), hind basitarsus length (HBL), hind basitarsus width (HBW), fore wing length (FL), fore wing width (FW), length of marginal cell (LMC) and number of hamuli in hind wing (NH). Some attributes were represented numerically, while morphometric characters of lengths and widths were measured in mm and a data matrix with 560 measurements was created. The morphological characters were measured as described by Rasmussen (2013). To detect population diversity within *T. iridipennis*, the data was subjected to ANOVA, factor analysis, principal components analysis, and discriminant function analysis using SPSS 21.0 statistical package.

RESULTS AND DISCUSSION

The characters measured in the head region included were head length (HL), head width (HW), distance between two lateral ocelli (DBO), ocello-ocular distance (OOD) and antennal length (AL) (Table 1). The head length (HL) was varied from 1.21 mm (Natham) to 1.46 mm (Kolli hills). Head width (HW) ranged between 1.54 mm (Madurai and Batlagundu) and 1.68 mm in (Kolli hills). These findings agree with those of Danaraddi (2007), who found that the width of the head including eyes in *T. iridipennis* ranged from 1.52 to 1.61 mm. Rasmussen (2013) reported that head length and width were 1.30 and 1.60 mm, respectively in *T. iridipennis*, 1.32 and 1.66 mm in *T. ruficornis*, 1.32 and 1.52 mm in *T. praeterita*, and 1.34 and 1.70 mm in *T. bengalensis*, while they were 1.1 and 1.24 mm in *T. iridipennis* respectively (Rahman et al., 2013). The distance between two lateral ocelli length (DBO) was 0.43 mm at Alagar Kovil location and 0.62 mm at Erumapatti. The ocello-ocular distance (OOD) was 0.20 mm in Alagar kovil, Natham and Batalagundu, while in Kolli hills, it was 0.29 mm. Tej et al. (2017) reported that the mean value of DBO ranged from 0.32 to 0.41 mm of two locations; and Efin et al. (2019) recorded a OOD value 0.28 mm which are comparable. The mean antennal length (AL) of the bees collected in Madurai and Kodaikanal was 1.66 and 1.98 mm in Kolli hills which is in line with the findings of Sharma et al. (2023) which was 1.89 mm.

The characters assessed in the thoracic region were hind leg length (HLL), hind tibial length (HTL), hind tibial width (HTW), hind basitarsus length (HBL), hind basitarsus width (HBW), fore wing length (FL), fore wing width (FW), length of marginal cell (LMC), and number of hamuli in hind wing (NH) (Table 1). The hind leg length (HLL) was greatest in the samples

Table 1. Morphometrics of *T. iridipennis*

Locations/ Characters	Alagarkovil	Madurai	Sirumalai	Natham	Megamalai	Kadamalai gundu	Kodaikanal	Batlagundu	Kolli Hills	Eruma patti	Total mean
HL	1.39± 0.014^{b}	1.29± 0.014^{d}	1.22± 0.005^{e}	1.21± 0.010^{e}	1.31± 0.006^{cd}	1.30± 0.005^{cd}	1.39± 0.014^{b}	1.31± 0.019^{cd}	1.46± 0.014^{a}	1.33± 0.010^{c}	1.32±0.075
HW	1.59± 0.018^{d}	1.54 ± 0.01^{e}	1.60± 0.005^{cd}	1.63± 0.019^{b}	1.58± 0.008^{d}	1.58± 0.009^{d}	1.59± 0.013^{d}	1.54± 0.017^{e}	1.68± 0.009^{a}	1.62± 0.012^{bc}	1.59± 0.041
DBO	0.43± 0.014^{f}	0.51± 0.01^{e}	0.61± 0.019^{ab}	0.61± 0.019^{ab}	0.56± 0.000^{d}	0.51± 0.010^{e}	0.59± 0.000^{bc}	0.57± 0.009^{cd}	0.60± 0.008^{ab}	0.62± 0.005^{a}	0.56± 0.059
OOD	0.20± 0.00^{d}	0.22± 0.008^{c}	0.25± 0.000^{b}	0.20± 0.005^{d}	0.23± 0.005^{c}	0.25± 0.000^{b}	0.23± 0.005^{c}	0.20± 0.000^{d}	0.29± 0.008^{a}	0.23± 0.008^{c}	0.23± 0.028
AL	1.66± 0.046^{d}	1.73± 0.009^{c}	1.75± 0.000^{c}	1.64± 0.000^{d}	1.84± 0.008^{b}	1.76± 0.006^{c}	1.66± 0.014^{d}	1.67± 0.010^{d}	1.98± 0.005^{a}	1.73± 0.009^{c}	1.74± 0.102
HLL	3.50b± 0.041^{c}	3.23± 0.005^{e}	3.40± 0.005^{d}	3.50± 0.006^{c}	3.53± 0.008^{abc}	3.40± 0.009^{d}	3.43± 0.013^{d}	3.42± 0.028^{d}	3.54± 0.009^{a}	3.52± 0.009^{ab}	3.44± 0.092
HTL	1.41± 0.027^{b}	1.27± 0.008^{e}	1.28± 0.014^{e}	1.46± 0.006^{a}	1.41± 0.005^{b}	1.32± 0.009^{d}	1.41± 0.014^{b}	1.35± 0.000^{c}	1.36± 0.009^{c}	1.46± 0.012^{a}	1.37± 0.067
HTW	0.50± 0.014^{bc}	0.39± 0.000^{e}	0.52± 0.000^{a}	0.49± 0.005^{bc}	0.52± 0.014^{a}	0.45± 0.005^{d}	0.50± 0.008^{bc}	0.49± 0.005^{c}	0.51± 0.008^{ab}	0.46± 0.008^{d}	0.48± 0.039
HBL	0.52± 0.000^{a}	0.50± 0.005^{b}	0.52± 0.000^{a}	0.49± 0.005^{b}	0.50± 0.008^{b}	0.53± 0.000^{a}	0.52± 0.014^{a}	0.45± 0.005^{c}	0.47± 0.005^{c}	0.49± 0.005^{b}	0.50± 0.025
HBW	0.23± 0.000^{d}	0.25± 0.000^{bc}	0.25± 0.008^{bc}	0.25± 0.000^{bc}	0.28± 0.005^{a}	0.26± 0.008^{b}	0.23± 0.008^{d}	0.24± 0.005^{cd}	0.26± 0.008^{b}	0.28± 0.005^{a}	0.25± 0.017
FL	3.45± 0.087^{bc}	3.30± 0.027^{d}	3.45± 0.035^{bc}	3.40± 0.008^{c}	3.51± 0.048^{b}	3.38± 0.013^{c}	3.45± 0.017^{bc}	3.30± 0.008^{d}	3.84± 0.013^{a}	3.46± 0.005^{bc}	3.46± 0.147
FW	1.51± 0.000^{b}	1.33± 0.027^{f}	1.41± 0.036^{cde}	1.40± 0.014^{de}	1.41± 0.017^{cd}	1.36± 0.014^{ef}	1.51± 0.018^{b}	1.32± 0.027^{f}	1.56± 0.009^{a}	1.45± 0.005^{c}	1.43± 0.080
LMC	1.23± 0.027^{bcd}	1.18± 0.009^{de}	1.22± 0.009^{cd}	1.21± 0.027^{de}	1.25± 0.014^{ab}	1.20± 0.009^{de}	1.23± 0.008^{bcd}	1.19± 0.009^{de}	1.29± 0.009^{a}	1.23± 0.014^{bcd}	1.22± 0.032
NH	5± 0.000	5± 0.000	5± 0.000	5± 0.000	5± 0.000	5± 0.000	5± 0.000	5± 0.000	5± 0.000	5± 0.000	5± 0.000

Values expressed as mean (mm) ± standard deviation; Different superscript letters in same line denote significant differences ($p < 0.05$).

of Kolli hills (3.54 mm) and 3.23 mm in Madurai. Similarly, the highest hind tibial length (HTL) (1.46 mm) was found in Natham and Erumapatti samples and lowest in Madurai (1.27 mm). The Hind tibial width (HTW) varied from 0.39 mm in Maduari to 0.51 mm in Kolli hills samples. The mean length of hind basitarsus (HBL) was the lowest in Batlagundu samples (0.45) mm and highest in Kadamalaigundu (0.53 mm). Hind basitarsus width (HBW) ranged around 0.23 mm in Alagar kovil and Kodaikanal samples and 0.28 mm in Megamalai and Eruma patti samples. These findings are in accordance with those of Trianto and Purwanto (2020) who reported the length and width of hind tibia of 1.74 and 0.54 mm and length and width of hind basitarsus of 0.55 and 0.33 mm in *T. iridipennis.* The fore wing length and width was highest in Kolli hills as 3.84 and 1.56 mm, respectively. The fore wing length of 3.30 mm was observed in Madurai and Batlagundu samples and the lowest forewing width was recorded in Batlagundu samples (1.32 mm); while the forewing length is longer compared as to the samples of *T. iridipennis* (3.2 to 3.9 mm) as measured by Vijayakumar and Jeyaraaj (2014). The length of marginal cell was the highest in Kolli hills (1.29 mm) and lowest in Madurai (1.18 mm) samples and these findings are similar to the description of Viraktamath and Roy (2022). Five hamuli were recorded in all of the bees collected from and no difference was observed in the number of hamuli, which is supported by the findings of Devanesan et al. (2003) and Kuberappa et al. (2005). The work of *T. iridipennis* described in this study are in accordance with previous workers. Four factors with high eigen values were obtained from the sample means. Analysis of variance of all the 14 morphometrics had significant statistical difference between groups ($p < 0.05$). The total variance of the four principal components reached 82.4 % and the variance of four PCA viz. PC1 (38.58 %), PC2 (20.95 %), PC 3 (19.75 %), and PC4 (8.10 %) were observed (Table 2). The FL, HW, FW, HLL, AL, OOD, HTW and HL were the most discriminative characters found in the principal component analysis. These characters aid in the formation of separate clusters and this is seen from the length of the resulting line (Fig. 1). From this study, new records of stingless bee have been set for different topographic levels of Tamil Nadu, with respect to their body sizes as adaptation for foraging and collecting floral resources.

ACKNOWLEDGEMENTS

The authors thank the Department of Agricultural Entomology, Agricultural College and Research Institute, Madurai. TNAU for providing facilities.

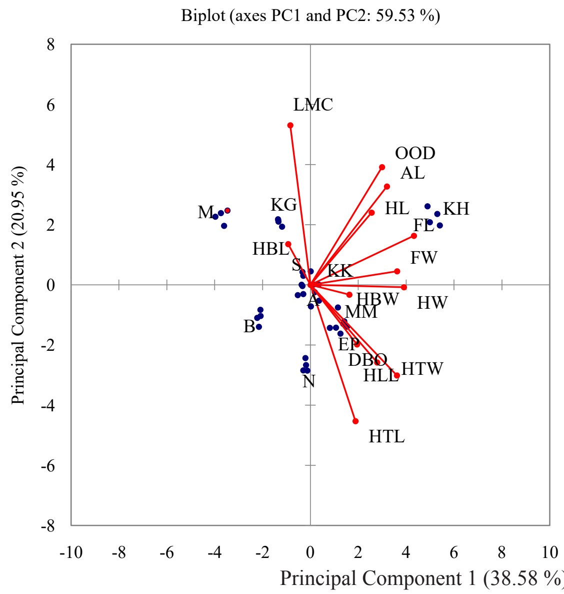

Fig. 1. Discriminative characters and clusters-PCA analysis of *T. iridipennis*; The first letter and the shortest form of clusters denotes collected locations

Table 2. PCA analysis of morphometrics of *T. iridipennis*

Principal Component	Eigen value	Variability (%)	Cumulative %
1	5.016	38.583	38.583
2	2.724	20.950	59.534
3	1.918	14.753	74.287
4	1.053	8.103	82.390
5	0.863	6.635	89.024
6	0.695	5.343	94.368
7	0.298	2.292	96.659
8	0.213	1.641	98.301
9	0.182	1.401	97.441
10	0.116	0.893	98.334
11	0.095	0.734	99.068
12	0.083	0.635	99.704
13	0.039	0.296	100

FINANCIAL SUPPORT

The first author expresses his gratitude to the Tamil Nadu Government for provision of full-time Ph.D. scholarship as financial assistance.

AUTHOR CONTRIBUTION STATEMENT

The study was conceptualized and designed by J Jayaraj and M Shanthi. K Balaji carried out the experiments and prepared the manuscript. S

Vellaikumar, C Rajamanickam, N Chitra and K Suresh assisted with the sample collection and data analysis. The article was read and approved by all the authors.

CONFLICT OF INTEREST

No conflict of interest.

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(Manuscript Received: February, 2023; Revised: September, 2023;
Accepted: September, 2023; Online Published: September, 2023)
Online First in www.entosocindia.org and indianentomology.org Ref. No. e23994