



Survey of stingless bee (Hymenoptera: Apidae: Meliponini) population at few contrasting habitats of southern Karnataka, India

Shaguftha Heena, S Basavarajappa*

DOS in Zoology, University of Mysore, Manasagangotri, Mysore, Karnataka, India

Abstract

Stingless bees (Hymenoptera: Apidae: Meliponini) are the most diverse group of social insects live in perennial colonies. They bees are considered as one of the most effective pollinators and produce economically important hive products, which are being used by mankind all over the world. Hence, they are considered as one of the keystone species, their distribution and inventorying their population at different habitats is need of the day. During the present investigation, unexplored areas like contrasting habitats were selected at Mysore (11°45' to 12°40' N and longitude 75° 57' to 77°15' E at an elevation 750 msl) and Chamarajanagar (11°40' to 12°06' N and Longitude 76° 46' E at more than 780 msl elevation) districts to record the distribution of stingless bee population. Altogether, 199 stingless bee live nests were recorded at urban, sub-urban, rural and rocky-hill area of southern Karnataka. In urban areas were recorded more live nests compared to sub-urban, rural and rocky-hill areas. Statistical analysis revealed significant difference ($X^2 = 258.25$; $P > 0.001$) between the contrasting habitats and interestingly, 2017 to 2022 occurrence of stingless bee population between contrasting habitats indicated an increasing trend of stingless bee population. Further, nine different sites were preferred by stingless bees to establish nests at contrasting habitats and the external features such as shape of nest, entrance, nest tube length, nest entrance size, width of nest tube, nest tube colour, nesting elevation and nest opening orientation revealed significant differences between urban, sub-urban, rural and rocky-hill area of southern Karnataka. Furthermore, internal features such as brood cells, honey pots and pollen pots are located respectively at centre and peripheral portion of the nest. It is concluded that the pattern of nesting observed at urban, semi-urban, rural and rocky hilly areas in Mysore and Chamarajanagar districts indicated the existence of *Tetragonula* species in this part of the state. In this regard in depth investigations were made on morphological traits, cytological, enzymes and molecular level (such types of observations are published elsewhere) and confirmed the existence of *T. pagdeni* and *T. iridipennis* at contrasting habitats such as urban, semi-urban, rural and rocky hilly areas of southern Karnataka.

Keywords: Contrasting habitats, stingless bee population, southern Karnataka

Introduction

Stingless bees or 'dammar bees' (Hymenoptera: Apidae: Meliponini) are the most abundant and diverse group of social insects locally called 'Misri Jenu' and 'Nasaru Jenu' live in perennial colonies often composed with single queen (fertile female), few drones (males) and hundred to several thousands of worker bees (sterile females) by exhibiting specialized division of labour (Inoue *et al.*, 1984; Michener, 2000) ^[1,2]. They are 50 times more diverse than *Apis* species (Hymenoptera: Apidae) but are highly adaptable to various conditions and show great diversity in nest architecture and pattern of nest construction (Basavarajappa, 2010) ^[3]. The nests build by stingless bees are complex form and are species specific with unique characters that is often used to identify different species (Michener, 1974; Sakagami, 1982) ^[4,5]. They are generally construct in the pre-existing cavities including trees trunks, cracks in old wall, termite and ant mounds and in other human built substrates (Jongjitvimol and Wattanachaitingcharoen, 2007; Sheetal and Basavarajappa, 2009; Basavarajappa, 2010; Nayak *et al.*, 2013; Vijayakumar *et al.*, 2013) ^[6, 7, 3, 8, 9] using the wax secreted from the dorsal body of worker bees along with resin collected from various plants (Virkar *et al.*, 2014) ^[10]. Further, stingless bees are considered as one of the most effective pollinators and are known to pollinate as more than 60 crops (e.g. Coconut, Mango, Watermelon, Guava, Strawberry, Citrus, Avocado, Coffee, Cardamom, Tomato, Cucumber, Vegetables etc.) which are commercially

important to mankind (Dos- Santos *et al.*, 2009; Del- Sarto *et al.*, 2005; Franco *et al.*, 2016; Rahman *et al.*, 2015) ^[11, 12, 13, 14] which constitutes around 30% of the food crops to human population (Slaa *et al.*, 2006) ^[15] around the world. Beside their pollination service for commercially important crops forest flora (Kerr *et al.*, 1963; Araujo *et al.*, 2004) ^[16, 17], their honey is rich with antioxidants, flavanoids, antioxidant, anti-inflammatory, anticancer, cardio protective properties and has high medicinal value (Rahman *et al.*, 2015) ^[14]. Stingless bee honey is commonly used as folk medicine for treating cold, cough and fever (Vijayakumar *et al.*, 2013) ^[9]. Moreover, stingless bee honey is used as one of the most effective natural products for wound healing, in the treatment of eye disorders, gastrointestinal tract diseases, neurological disorders and fertility disorders. As a result, stingless bee honey is 20 times costlier than the *Apis* species (Kumar *et al.*, 2012) ^[18]. Furthermore, propolis produced from the stingless bees exhibits notable antibacterial and antiviral properties which are used in pharmacological industries (Choudhari *et al.*, 2013) ^[19]. Since stingless bees are well known for their honey having high medicinal values and other economically important honey products such as pollen, wax and propolis, they are cultured in the name of 'Meliponiculture' at various parts of the world including in India to obtain the commercial products and to earn money (Nogueira – Neto, 1997) ^[20]. Various authors (Rasmussen and Camargo, 2008; Sheetal and Basavarajappa, 2009; Basavarajappa, 2010; Nayak *et al.*,

2013) [21, 7, 3, 8] have reported on different aspects of *Tetragonula* species at different parts of the world including in India.

Remarkably, the diversity of nesting architecture such as design of brood cells arrangement from horizontal combs to cluster forms (Roubik, 2006) [22] and divergence in choosing the substrates has been very well adapted by the stingless bees. The selection of nesting site is preferably determined by the availability of resources and protection from natural enemies (Roubik, 1989) [23]. Unlike *Apis* species, the nests of stingless bees are immobile and fixed. They are long lived having variety of nest entrances with different shapes and arrangement of brood cells and food storage containers (Roubik, 2006) [22]. Nesting habitats and nest architecture has showed greater variations not only among the genera but also among the species within the genus. Hence, the nesting attributes can be used as one of the tool for ecological, phylogenetic and taxonomic studies (Rasmussen and Camerago, 2008; Lima *et al.*, 2013) [21, 24].

Stingless bee colonies were recorded by Sheetal and Basavarajappa (2009) [7] in Manasagangotri Campus in Mysore city. Human interference on the nests of stingless bees at different habitats in Mysore district was studied by Basavarajappa (2010) [3]. Innumerable colonies which are present amidst Manasagangotri Campus have instigated to conduct in-depth research on stingless bees. Gopinatha and Basavarajappa (2023) [25] have investigated on the stingless bee population existed at south-eastern districts such as Bangalore Rural, Kolar and Doddaballapura districts. However, investigations on contrasting areas of Mysore Chamarajanagar districts of southern Karnataka are not available. Here, many beekeepers are culturing stingless bees in the name of 'Meliponiculture' to produce stingless bee honey. Reports on the distribution, diversity and nesting habitats of stingless bees is poor. Hence, the full diversity of stingless bees in southern Karnataka remains to be unknown (Basavarajappa, 2010) [3]. Hence, critical investigations are essential to explore the diversity and distribution of stingless bee population inhabited at different habitats of southern Karnataka. It would help identify any new species if any existed at different parts of southern Karnataka. On this line, in-depth investigations are conducted to update the knowledge on stingless bees in this part of the state.

Materials and Methods

Study area: To conduct present work, contrasting habitats of Mysore and Chamarajanagar district which experiences diversified agro-climatic conditions were selected in Mysore and Chamarajanagar districts (Figure 1). Mysore district lies at 11°45' to 12°40' N latitude and 75° 57' to 77 ° 15' E longitudes at an elevation 750 msl. In Mysore city, contrasting habitats such as Manasagangotri Campus (12° 18' 43"N latitude and 76° 38' 12"E longitude with an altitude 793 meters msl), Devaraj Market (12° 18' 38"N latitude and 76° 39' 07"E longitude with an altitude 773 meters msl), Ontikoppal Flowering Market (12° 19' 33"N latitude and 76° 37' 59"E longitude with an altitude 800 meters msl), Ramanuja Road (12° 17' 57"N latitude and 76° 39' 14"E longitude with an altitude 761 meters msl), rural area i.e., Hunsur (12° 18' 32"N latitude and 76° 16' 58"E longitude with an altitude 808 meter msl) in Mysore district and all these places experiences semi- malnad type of climate. In Chamarajanagar district lies at 11°40' to 12°06' N latitude and 76° 46' E longitude at an elevation from 778 to

857 meters msl (Kamath, 2001) [26]. Chamarajanagar district experiences dry climate compared to Mysore district, where sub-urban area (e.g. Begur - 11° 56' 16"N latitude and 76° 40' 16"E longitude with an altitude 778 meters msl) and rocky-hill area (e.g. Terakanambi - 11° 48' 54"N latitude and 76° 47' 30"E longitude with an altitude 857 meters msl), which experiences dry climate (Kamath, 2001) [26] were selected for the present investigation (Figure 1).

Methodology: Total four different contrasting habitats namely: urban (Manasagangotri Campus (MGC), Devaraj Market (DM), Ontikoppal Flowering Market (OFM), Ramanuja Road (RD), semi-urban (e.g. Begur in Chamarajanagar district), rural (e.g. Hunsur in Mysore district) and rocky-hill area (e.g. Terakanambi in Chamarajanagar district) were randomly selected. In each selected area, there are five sampling sites were earmarked and stingless bee nests were critically observed using visual count method (VCM) and All Out Search (AOS) Method as per Sheetal and Basavarajappa (2009) [7]. The environmental factors namely: temperature, humidity, rainfall and natural vegetation present around the selected contrasting habitats was collected to know their influence on stingless bee distribution. Observations on number of nests, nesting pattern, nest site preference, shape of the nest, nest entrance opening, colour of the nest, nest height from the ground level, nest projection length, nest entrance opening length and width, orientation of the nest entrance tube and number of worker bees guarded around the nest opening were recorded using standard methods. The collected data was systematically analyzed by following standard methods as per Saha (2009).

Results

Distribution of stingless bee nests: Table 1 shows the distribution of stingless bee nests at contrasting habitats of southern Karnataka. Altogether, 199 stingless bee live nests were recorded at urban, sub-urban, rural and rocky-hill area of southern Karnataka. In urban area i.e., Mysore city – Manasagangotri Campus (MGC), Devaraj Market (DM), Ontikoppal Flower Market (OFM) and Ramanuja Road (RD) areas, where on an average 73, 42, 22 and 13 live nests were recorded respectively (Table 1). However, sub-urban (Begur – Chamarajanagar district) area 15 live nests, rural (Hunsur – Mysore district) 29 live nests and rocky-hill area (Terakanambi – Chamarajanagar district) five live nests were recorded. Friedman Two way analysis of variance revealed significant difference ($X^2 = 258.25$; $P > 0.001$) between the contrasting habitats of southern Karnataka during 2017 to 2022. Further, year-wise, 195, 193, 199 and 209 live nests were recorded during 2017, 2018, 2019 and 2022 respectively (Table 1). Furthermore, per cent occurrence of stingless bee live nests at contrasting habitats of southern Karnataka is shown in Figure 2. Data from the Table 1 and clearly demonstrated the variation existed in relation to the occurrence of stingless bee live nests at contrasting habitats of southern Karnataka. Interestingly, there was an increasing trend of stingless bee nests from 2017 to 2022 was recorded in this part of the state.

External features of stingless bee nests: Nest sites: Table 2 shows the nest sites of stingless bees at contrasting habitats of southern Karnataka. Total nine different sites were preferred by stingless bees to establish nests at urban, sub-urban, rural and rocky-hill areas of southern Karnataka. At urban area and rural areas, stingless bees preferred to nest in the crevices of tree bark, stone wall steps, iron pipes, cement wall, natural or man-made crevices, stone wall, electric poles, ground (soil surface) and wooden door rims. At sub-urban area, live nests were recorded only on iron pipes, cement wall, crevices, stone wall and electric poles. However, at rocky-hill area, stingless bee nested at man-made establishments like cement and stone made walls. Number of live nests of stingless bees recorded all these sites are given in the Table 2. Moreover, Friedman two-way analysis of variance revealed significant difference ($X^2 = 217.375$; $P > 0.001$) between urban, sub-urban, rural and rocky-hill areas of Mysore and Chamarajanagar districts and it is clearly demonstrated the variation in selecting the nest sites at different contrasting habitats of southern Karnataka.

Shape and colour of the nest entrance: Table 3 shows the different shaped nest entrances of stingless bees nest recorded at contrasting habitats of southern Karnataka. Total four different shaped nest entrances were recorded at urban, sub-urban, rural and rocky-hill areas. Oval shaped nest entrances were high and it was followed by irregular shaped entrance bearing nests. However, circular and tubular shaped nest entrances were almost same (Table 3). Moreover, Friedman two-way analysis of variance revealed significant difference ($X^2 = 101.5$; $P > 0.001$) between urban, sub-urban, rural and rocky-hill areas and demonstrated clearly difference between the shape of nest entrances of stingless bees at contrasting habitats in southern Karnataka. Further, the nest tube colours such as black, brown and brownish-black were recorded with stingless bee nests at urban, sub-urban, rural and rocky-hill areas of southern Karnataka. Amongst the colours, black coloured nest tubes were more predominant and it was followed by brownish-black and brown coloured nest tubes (Table 3). Friedman two-way analysis of variance revealed significant difference ($X^2 = 60.875$; $P > 0.001$) between urban, sub-urban, rural and rocky-hill areas demonstrated clearly the difference between the contrasting habitats at southern Karnataka.

Characters of nest tube entrance: Table 4 shows the nest tube length, width and nest entrance size, nesting elevation and orientation of stingless bee nest tubes at contrasting habitats of southern Karnataka. Total ten different sized nest tubes were recorded with stingless bee nests. However, stingless bees established different lengthen nest tubes at sub-urban, rural and rocky-hill areas. Friedman two-way analysis of variance revealed significant difference ($X^2 = 257.375$; $P > 0.001$) between urban, sub-urban, rural and rocky-hill areas and demonstrated clearly difference between the contrasting habitats at southern Karnataka (Table 4). Moreover, the nest tube width was also differed and totally ten different sized width bearing nest tubes were recorded at urban, sub-urban, rural and rocky-hill areas (Table 4). Amongst them, stingless bees established 0.4 to 0.6 cm width bearing nest tubes more and it was followed by 0.2 to 0.4 cm width bearing nest tubes compared to other different sized width bearing nest tubes at urban area, sub-

urban, rural and rocky-hill areas. Friedman two-way analysis of variance revealed significant difference ($X^2 = 182.875$; $P > 0.001$) between urban, sub-urban, rural and rocky-hill areas and demonstrated clearly the difference between the contrasting habitats of southern Karnataka (Table 4). Further, nest entrance size was ranged in between 0.0 to 2.0 cm (Table 4). The nests with 0.6 to 1.0 cm sized entrances were more and it was followed by the nest entrance opening size with 0.0 to 0.5 cm and 1.1 to 1.5cm sized entrance bearing nests. Friedman two-way analysis of variance revealed significant difference ($X^2 = 77.375$; $P > 0.001$) between urban, sub-urban, rural and rocky-hill area and demonstrated clearly the difference between the size of nest entrances at contrasting habitats in southern Karnataka (Table 4). Furthermore, the nests were found at different elevations at different contrasting habitats of southern Karnataka (Table 4). The nesting elevation was ranged in between 0.0 to 100 meters and above. Highest numbers of live nests were found at 31 to 40 meters height and it was followed by 21 to 30 meters, 41 to 50 meters and more than 100 meters height compared to other nests located below 20 meters height (Table 4). Friedman two-way analysis of variance revealed significant difference ($X^2 = 105.0$; $P > 0.001$) between urban, sub-urban, rural and rocky-hill areas and demonstrated clearly the difference existed between the nesting elevations chosen by the stingless bees while establishing the nests at southern Karnataka. However, nest tubes were established at different directions at contrasting habitats of southern Karnataka (Table 4). Total eight nest tube orientations namely: north, south, west, east, south-west, south-east, north-west and north east were recorded. Amongst the nest tube orientations, west side oriented nest tube bearing nests were more and it was followed by east and north-west, south-west, south-east and north oriented nest tube bearing nests compared to others (Table 4). Friedman two-way analysis of variance revealed significant difference ($X^2 = 92.7575$; $P > 0.001$) between urban, sub-urban, rural and rocky-hill areas demonstrated clearly the difference existed between the nest tube orientations with stingless bee nests at different contrasting habitats of southern Karnataka (Table 4).

Internal features of stingless bee nest: Table 5 shows the internal structure of stingless bee nest. The brood cells, honey pots and pollen pots are located respectively at centre and peripheral portion of the nest. Moreover, the brood cell colour was yellowish-brown, the honey pot colour was dark-brown and pollen pot colour was brown in colour (Table 5). Interestingly, the brood cells, honey pots and pollen pots were oval in shape with different size in terms of their length, width and diameter. Detailed measurements of brood cell, honey pot and pollen pot are given in Table 5. Data from the Table 5 clearly indicated the considerable difference with respect to the size of brood cell, honey pot and pollen cells of stingless bees. The pattern of nesting observed at urban, semi-urban, rural and rocky hilly areas in Mysore and Chamarajanagar districts indicated the existence of *Tetragonula* species in this part of the state. In this regard in depth investigations were made on morphological traits, cytological, is enzymes and molecular level and confirmed the existence of *T. pagdeni* and *T. iridipennis* at contrasting habitats such as urban, semi-urban, rural and rocky hilly areas of southern Karnataka. Such types of results are published elsewhere.

Discussion

Stingless bee population distributed differently at various contrasting habitats of southern Karnataka. During the present investigations, stingless bee population was recorded at all the contrasting habitats such as urban, sub-urban, rural and rocky-hill areas in Mysore and Chamarajanagar districts of southern Karnataka. Total 199 stingless bee live nests were recorded at urban, sub-urban, rural and rocky-hill areas. Surprisingly, urban area (e.g. Manasagangotri Campus (MGC), Devaraj Market (DM), Ontikoppal Flower Market (OFM) and Ramanuja Road (RD) in Mysore city) scored good number of live nests compared to sub-urban (Begur in Chamarajanagar district), rural (Hunsur in Mysore district) and rocky-hill area (Terakanambi in Chamarajanagar district) and hence indicated significant difference between the contrasting habitats. Moreover, the live nests density increased gradually from 2017 to 2022 and hence, stingless bee population density indicated an increasing trend in this part of the state. Moreover, stingless bees followed quite interesting nesting features. The external features of nests occurred at different contrasting areas revealed considerable variations. Total nine different sites such as crevices of tree bark, stone wall steps, iron pipes, cement wall; natural or man-made crevices, stone wall, electric poles, ground (soil surface) and wooden door rims were preferred to establish nests at urban, sub-urban, rural and rocky-hill areas. Surprisingly, all these nest sites were not uniformly distributed at different contrasting habitats and hence, per cent preference to establish the nests was site specific at urban, sub-urban, rural and rocky-hill areas and showed significant difference. Hence, nest establishment by stingless bees was site specific and it was based on the prevailed suitable conditions. Further, external features of nests such as shape and colour of nest entrance, nest tube length and width, nest entrance size, nesting elevation and nest opening orientations were differed considerably with nests established by stingless bees at urban, sub-urban, rural and rocky-hill areas. Interestingly, black, brown and brownish-black coloured nest tubes were built by stingless bees. Among them, black coloured nest tubes were more predominant compared to others. Perhaps, the colour of nests might be involved with the locally available soil, as stingless bees use little soil while establishing the nests. Total four types of nest tube entrance shapes were recorded. Of all, oval shaped nest entrance was predominant and it was followed by irregular shaped nest entrance compared to circular and tubular shaped nest entrances. Moreover, nest tube length and width established by stingless bees inhabited at urban, sub-urban, rural and rocky-hill areas indicated significant variations. Perhaps, stingless bees might have built different sized and shaped nest tubes to avoid predatory insect's entry into the nest. Moreover, the shape and size as well as the colour of the nests are important nesting morphological features, which may help mimic the surrounding nest site environment so as to overcome the predators/enemies problem. This strategy could help establish perennial colonies by stingless bees at different habitats.

Stingless bees nested at different elevations and it was ranged in between 0.0 to more than 100 meters. Interestingly, 31 to 40 meters elevation chosen by stingless bees as an ideal site for nest establishment and it was followed by 21 to 30 meters, 41 to 50 meters compared to

other elevations. Stingless bees are weak fliers unlike *Apis* species, travel within 200 to 400 meters circumferences to forage and collect nesting material so as to establish the nests nearer to the ground. Few nests were found at rocky-hill area where, the habitat itself was more than 100 meters height and hence, stingless bee population surviving such habitat indicated higher nesting elevation. Further, stingless bees build their nests by establishing differently oriented nest tubes. The nest tubes were oriented towards north, south, west, east, south-west, south-east, north-west and north east directions at urban, sub-urban, rural and rocky-hill areas of southern Karnataka. Similar type of observations was reported by Basavarajappa (2010) ^[3], Gopinatha and Basavarajappa (2023) ^[25] at different parts of Karnataka. Amongst the nest tube orientations, west side oriented nest tube bearing nests were more and it was followed by east and north-west oriented nest tube bearing nests compared to others. Statistical analysis of all the morphological characters such as shape, colour of nest tube entrance, nest tube length and width, nest entrance size, nesting elevation and nest opening orientations indicated significant differences between urban, sub-urban, rural and rocky-hill areas of southern Karnataka. Similar type of observations was made by Gopinatha and Basavarajappa (2023) ^[25] at south-eastern Karnataka. Thus, stingless bee nests physical features were uneven, differed considerably and built depending on the requirement at their preferred nest sites in southern Karnataka. Furthermore, internal structures of nests namely: brood cells, honey pots and pollen pots, brood cell colour, honey pot colour and pollen pot colour were not much differed between the nests established at different contrasting areas such as urban, sub-urban, rural and rocky-hill areas of southern Karnataka. However, the brood cells, honey pots and pollen pots shape and sizes considerably differed. So, by considering external features and internal features of the nest attributes of stingless bee population inhabited at urban, sub-urban, rural and rocky-hill areas of southern Karnataka indicated the existence of two different *Tetragonula* species in this part of the state. After critical assessment of morphological features, cytological and molecular studies of stingless bee population inhabited at urban, sub-urban, rural and rocky-hill areas it was confirmed the existence of *Tetragonula pagdeni* and *T. iridipennis* which are not evenly distributed at urban, sub-urban, rural and rocky-hill areas. Results of such observations will be published elsewhere.

Thus, stingless bees adapt elite strategies (Nayak *et al.*, 2013) ^[8] to establish their colonies spatially at human modified habitats (Sheetal and Basavarajappa, 2009) ^[7] and natural ecosystems. They follow certain modifications while establishing their nests to overcome the prevailed challenges at contrasting habitats in the nature (Karthick *et al.*, 2018) ^[28]. By establishing different coloured nest tube, big or small or moderate sized nest tube entrance, selecting specific height for building nests, differently oriented nest tubes are certain devices followed while building their nests at human modified habitats (Basavarajappa, 2010) ^[3] and natural ecosystems (Gopinatha and Basavarajappa, 2023) ^[25]. Our observations are on par with the observations of Michener (2000) ^[2], Rajitparinya *et al.* (2000) ^[29], Raakhee and Devanesan (2000) ^[30], Swaminathan (2000) ^[31], Batista *et al.* (2003) ^[32],

Michener and Boongird (2004) [33], Jobiraj and Narendran (2004) [34], Sung *et al.* (2004) [35], Del- Sarto *et al.* (2005) [12], Klaskasikorn *et al.* (2005), Slaa *et al.* (2006) [15], Rasmussen and Cameron (2007) [37], Jongjitvimol and Wattanachaiyingcharoen (2007) [6], Cortopassi-Laurino *et al.* (2007) [38], Dos-Santos *et al.* (2009) [11], Danaraddi and Shashidar (2009) [39], Sheetal and Basavarajappa (2009) [7], Raju *et al.* (2009) [40], Basavarajappa (2010) [3], Rasmussen and Cameron (2010) [41], Danaraddi *et al.* (2012) [42], Kumar *et al.* (2012) [18], Salim *et al.* (2012) [43], Nayak *et al.* (2013) [8], Silva *et al.* (2013) [44], Rasmussen (2013) [45], Vijayakumar *et al.* (2013) [9], Ayala *et al.* (2012) [46], (Viraktamath *et al.* (2013) [47], Camargo and Pedro (2013) [48], Pedro (2014) [49], Francoy *et al.* (2016) [13], Rahman *et al.* (2015) [14], Makkar *et al.* (2018) [50], Nayak and Prakash (2018) [51], Shanas and Faseeh (2019) [52], Viraktamath and Shishira (2020) [53] and Viraktamath and Roy (2022) [54], who have studied different stingless bee species at various agro-climatic conditions at different parts of the world including India.

Conclusion

Stingless bee species (e.g. *Tetragonula pagdeni* and *T. iridipennis*) adapt elite strategies to establish their colonies spatially at human inhabited areas, natural and rocky-hill areas, where human interferences are less. They prefer various natural and man-made structures to establish their colonies. While establishing the colonies, they show various modifications such as nest entrance chamber colour, nest entrance shape, length and width etc. Moreover, the nests are established at different heights. They establish differently oriented nest entrance tube to explore the available resources such as nectar, pollen and resin to the colony. During the present investigation, it was suspected the existence of two stingless bee species at contrasting habitats of southern Karnataka. After doing physical features of the nest, morphological traits of the stingless bee workers, cytological and molecular studies (such type of observations are published elsewhere) it was confirmed that *Tetragonula iridipennis* and *Tetragonula pagdeni* are distributed at different contrasting areas of Mysore and Chamarajanagar districts of southern Karnataka.

Table 1: Distribution of stingless bee nests at contrasting habitats of southern Karnataka

Year	Districts of southern Karnataka							Total
	Mysore				Chamarajanagar			
	Urban area		Rural		Sub-urban area		Rocky- hill area	
	MGC	DM	OFM	RD	Hunsur	Begur	TK	
2017	70	39	24	17	32	10	3	195
2018	74	35	25	12	29	13	5	193
2019	75	46	12	10	31	19	6	199
2022	73	48	27	13	24	18	6	209
Mean	73	42	22	13	29	15	5	199
X2	258.25*							-

Note: each value is a mean of three observations. MGC: Manasagangotri Campus; DM: Devaraj Market; OFM: Ontikoppal Flower Market; RD: Ramanuja Road and TK: Terakanambi.* Value is significant at 0.001% level.

Table 2: Nest sites of stingless bees recorded at contrasting habitats of southern Karnataka

Sl. No.	Nest site	Districts of southern Karnataka							Total
		Mysore				Chamarajanagar			
		Urban area		Rural		Sub-urban area		Rocky- hill area	
		MGC	DM	OFM	RD	Hunsur	Begur	TK	
1.	Tree bark	1	-	1	2	2	-	-	6
2.	Stone steps	9	8	-	1	2	-	-	20
3.	Iron pipes	16	10	2	1	3	2	-	34
4.	Cement wall	22	13	6	3	5	1	2	52
5.	Crevices	4	2	-	2	1	6	-	15
6.	Stone wall	15	4	9	1	-	3	3	35
7.	Electric pole	5	5	4	1	5	3	-	33
8.	Ground	1	-	-	1	7	-	-	9
9.	Wooden Door Rim	-	-	-	1	4	-	-	5
Total		73	42	22	13	29	15	5	199
X2		217.375*							-

Note: each value is a mean of three observations. MGC: Manasagangotri Campus; DM: Devaraj Market; OFM: Ontikoppal Flower Market; RD: Ramanuja Road and TK: Terakanambi.* Value is significant at 0.001% level.

Table 3: Shape and colour of nests of stingless bees recorded at contrasting habitats of southern Karnataka

Districts	Contrasting Habitats	Nest shape				Colour of nest entrance tube		
		Oval	Circular	Irregular	Tubular	Black	Brown	Brownish- Black
Mysore	MGC	36	15	14	8	58	11	4
	DM	14	2	22	4	38	4	-
	OFM	15	1	5	1	18	2	2
	RD	1	3	7	3	1	3	7
	Rural (Hunsur)	18	2	5	4	10	1	4
Chamarajanagar	Sub-urban (Begur)	12	-	1	2	10	1	4
	Rocky-hill (Terakanambi)	3	-	1	1	4	1	-

Total	99	23	55	22	139	27	33
X2	101.5*				60.875*		

Note: each value is a mean of three observations. MGC: Manasagangotri Campus; DM: Devaraj Market; OFM: Ontikoppal Flower Market and RD: Ramanuja Road. * Value is significant at 0.001% level.

Table 4: Few nest tube entrance characters of stingless bees recorded at contrasting habitats of southern Karnataka

District	Contrasting Habitats	Nest Tube			Nest	
		Length in range (cm)	Entrance size (cm)	Entrance width (cm)	Elevation (in meter)	Orientation
Mysore	MGC	0.0- 2.0	0.0-2.0	0.0-1.2 & 1.4-2.0	0-80 & Above 100	N, S, W, E, SW, SE, NW & NE
	DM	0.0-2.0	0.0-2.0	0.0-1.2 & 1.4-2.0	0-70 & Above 100	N, W, E, SW, SE & NW
	OFM	0.0-1.6	0.0-1.5	0.0-1.2 & 1.4-1.6	0-80 & Above 100	W, E, SW, & NW
	RD	0.0-1.2	0.6-1.5	0.0-1.8	21-40 & 51-80	N, S, W, E, SW, SE, NW & NE
	Rural (Hunsur)	0.0-2.0	0.0-2.0	0.0-1.0 & 1.4-1.8	0-60 & 81-90	N, S, W, E, SW & NW
Chamarajanagar	Sub-urban (Begur)	0.21-2.0	0.0-2.0	0.0-0.6	0-40 & 71-Above 100	N, S, W, E, SW, SE, NW & NE
	Rocky-hill (Terakanambi)	0.41-1.0, 1.21-1.4 & 1.61-1.8	0.0-0.5 & 1.6-2.0	0.6-1.0	91-Above 100	W, SW, SE & NE
X2		257.375*	77.375*	182.875*	105.0*	192.75*

Note: each value is a mean of three observations. MGC: Manasagangotri Campus; DM: Devaraj Market; OFM: Ontikoppal Flower Market and RD: Ramanuja Road. N: North, S: South; W: West; E: East; SW: South-west; SE: South-east; NW:North-west; NE: North-east.* Value is significant at 0.001% level

Table 5: Internal nest parameters of stingless bees nest

Internal structure of stingless bee nest				
Sl. No	Parameters	Brood cell	Honey pot	Pollen pot
1.	Location	Center	Periphery	Periphery
2.	Colour	Yellowish Brown	Dark Brown	Brown
3.	Shape	Oval	Oval	Oval
4.	Length	0.34 ± 0.06 cm	1.22 ± 0.26 cm	1.18 ± 0.06 cm
5.	Width	0.25 ± 0.05 cm	1.01 ± 0.32 cm	0.65 ± 0.07 cm
6.	Diameter	0.82 ± 0.09 cm	1.79 ± 0.09 cm	2.13 ± 0.12 cm
7.	Honey collected per honey pot		0.109 ± 0.01 ml	
8.	Honey Colour		Light Yellow	

Note: Each value is a mean of 20 observations.

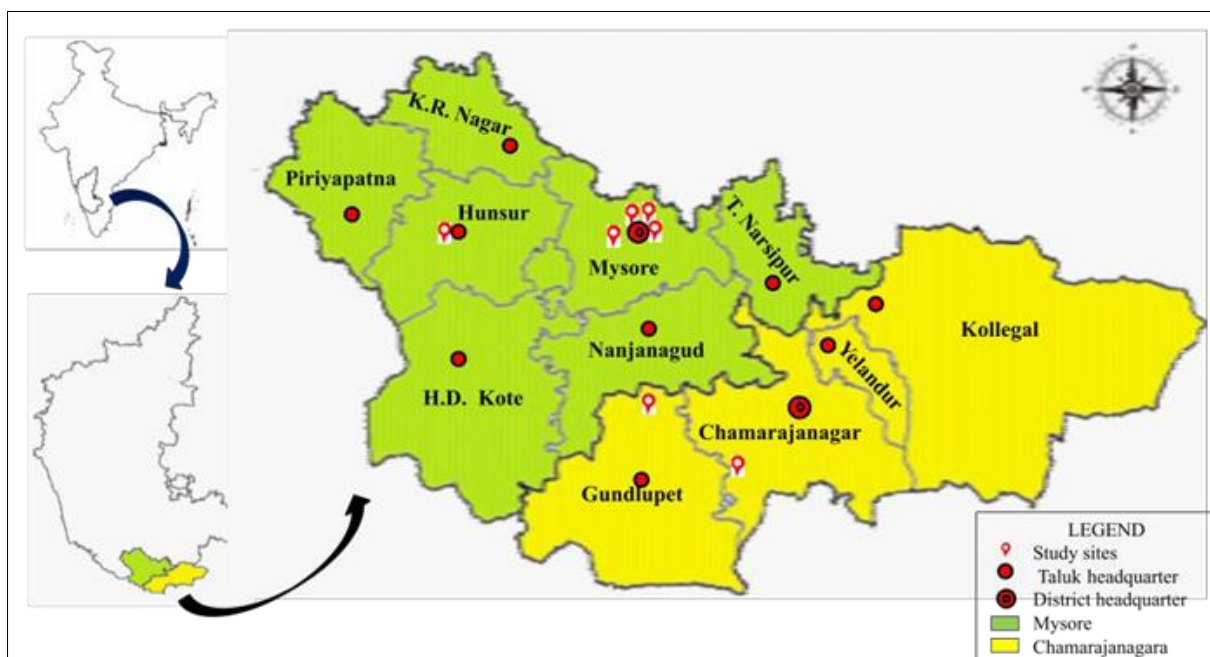


Fig 1: Map showing the study areas in Mysore and Chamarajanagar districts of southern Karnataka



Fig 2: Internal view of nest with stingless bees

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