Biology of the Indian Stingless Bee:

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By

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PREFACE

With evolution, both plant and animal kingdoms have developed into diverse species. Plants and animals interact in different ways, with the dominating type being mutual interactions among flowering plants and their pollinators. The most important pollinators are insects, including honeybees, solitary bees, and stingless bees. Through their pollination services, they help conserve plant diversity and optimise crop yield. Besides ecosystem services, some insects' nests or hive products (e.g., honeybees and stingless bees) are highly demanding and utilised from different perspectives.

Unfortunately, the population of pollinators, including honeybees and stingless bees, is declining worldwide. It may be due to climate change, habitat degradation, extensive monoculture, and the use of agrochemicals. The decline of pollinators negatively impacts biodiversity, crop yield, and, thereby, the economy of farmers. In recent years, beekeepers and researchers have started to manage bees (e.g., honeybees, orchid bees, mason bees, and stingless bees) to overcome the pollination deficit of crops. However, the management of non-native bees negatively influences the foraging behaviour of local pollinators and their survival. So, beekeepers and researchers are focusing on the management of native pollinators.

The Indian stingless bee *Tetragonula iridipennis* Smith, a unique species, is distributed in India, the Indomalayan realm, Nepal, and Sri Lanka. The species is widely distributed in India, from sea level to higher altitudes, including several states. The stingless bees forage on diverse plant species with significant floral fidelity. They are effective pollinators of many crops. However, little information about stingless bees is available, and the data deficit may render meliponiculture sustainable. Thus, comprehensive data about the Indian stingless bees is in high demand.

This book covers almost all aspects (e.g., distribution, morphology, colony organisation, nesting biology, foraging behaviour, threats and conservation strategies, bee products, pollination efficiency, etc.) of the stingless bee species. This information helps readers be aware of the stingless bee that can help conserve the bee species. It is hoped that the book will be useful to beekeepers, researchers, and students in acquiring knowledge about Indian stingless bees, which will help in the scientific management of stingless bees. The book will guide the beekeeper in extracting bee products from a hive. It will generate enough interest among scholars to inspire and guide future research on stingless bees.

Ujjwal Layek
Prakash Karmakar

CHAPTER 1

TAXONOMIC NOTES AND DISTRIBUTION OF INDIAN STINGLESS BEES

Stingless bee species in India

Rasmussen (2013) reported eight species of stingless bees from the Indian subcontinent. These were *Lepidotrigona arcifera* (Cockerell), *Lisotrigona cacciae* (Nurse), *Lisotrigona mohandasi* (Jobiraj & Narendran), *Tetragonula aff. laeviceps* (Smith), *Tetragonula bengalensis* (Cameron), *Tetragonula gressitti* (Sakagami), *Tetragonula iridipennis* (Smith), *Tetragonula praeterita* (Walker), and *Tetragonula ruficornis* (Smith). From North East, North, and South India, Rahaman et al. (2015) reported two genera and six different species of stingless bees: *Lepidotrigona arcifera*, *Tetragonula bengalensis*, *Tetragonula iridipennis*, *Tetragonula laeviceps*, *Tetragonula praeterita* and *Tetragonula ruficornis*. Viraktamath and Roy (2022) described five species of *Tetragonula*, namely, *Tetragonula vikrami* Viraktamath, (from Karnataka), *T. sumae* Viraktamath, (from Tamil Nadu), *T. ashishi* Viraktamath and Jagruti, (from Maharashtra), *T. shishirae* Viraktamath (from Rajasthan) and *T. shubhami* Viraktamath (from Chhattisgarh).

Systematic position of *Tetragonula iridipennis*

The Indian stingless bee (also called dammar bee), *Tetragonula iridipennis*, is a species belonging to the family Apidae, subfamily Apinae and tribe Meliponini. It was first described by Frederick Smith in 1854, who found the species on the island of Sri Lanka (Rasmussen 2013). Many older literature erroneously placed this species in the genus *Melipona*. Until recently, it was classified as *Trigona* and is still mistakenly referred to as *Trigona iridipennis*. The systematic position of the stingless bee is given below:

Domain: Eukaryota
Kingdom: Animalia
Phylum: Arthropoda
Class: Insecta
Order: Hymenoptera
Family: Apidae
Tribe: Meliponini
Genus: *Tetragonula*
Species: *T. iridipennis*

Distribution of Indian stingless bees

The stingless bees are native to the tropical and subtropical regions of the world. Stingless bees are divided into three monophyletic clades, and their distribution today largely corresponds to three major biogeographic regions- Neotropical, African, and Indo-Malayan-Australasian (Rasmussen and Cameron 2010). Indian stingless bee (*Tetragonula iridipennis*) is found in India, the Indomalayan realm, Nepal, and Sri Lanka. In India, the species is widely distributed, from sea level to a higher altitude, including several states like Jannu & Kashmir, Maharastra, Kerala, Tamil Nadu, and West Bengal (Layek and Karmakar 2018; Rahaman 2018; Balaji et al. 2023).

The distribution pattern is clumping to a random type (Figure 1.1). The minimum distance between the nearest two colonies is 5.6 m. The number of colonies per kilometre square area (i.e., density) varied from 0 to 18 (4.3 ± 3.46). Densities are comparatively higher in mixed vegetation (having many trees, shrubs, and herbs) and human habitats (having building structures and plants) than in agricultural field-dominated areas and deep forest areas (Table 1.1). In the Agricultural zone, they may quickly find their preferred nesting substrates, like building walls and tree trunks. In the deep forest area of Sal (*Shorea robusta* L.), nesting cavities may be unavailable. Also, they may not have diverse floral resources available. As their colonies are perennial, colony density does not vary by season, unlike Indian honeybees.

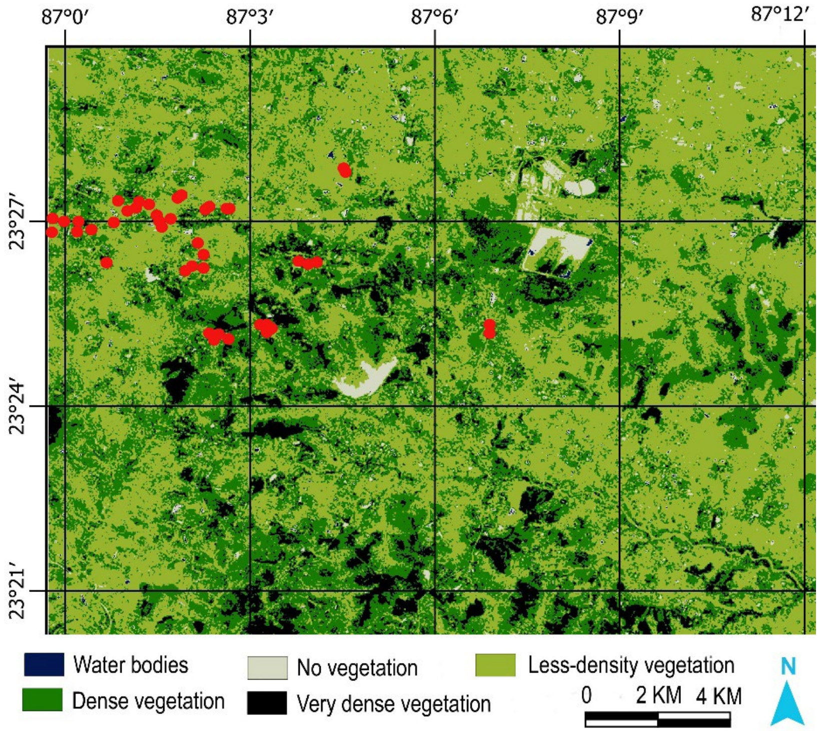

Figure 1.1. Map showing the clumping distribution of stingless bee colonies (red circles).

Table 1.1. Density (number of colonies/ m^2 area) of stingless bees in different landscapes.

Vegetation type	Density
Human settlement without vegetation	$0.2^d \pm 0.42$
Human settlement with sparse vegetation	$7.50^a \pm 3.31$
Mixed vegetation[1]	$4.90^b \pm 2.92$
Agricultural landscape	$0.40^d \pm 0.70$
Deep forest (tree dominated)	$1.10^c \pm 1.10$
Polluted area (factory dominated)	$0^e \pm 0$

[1]Mixed vegetation- moderate dense vegetation with a mixture of a few crop fields, trees, and bushy areas. Values are given in mean ± standard deviation. Different superscript letters (shown after the mean values) indicate significant differences (Dunn's post hoc test at 5%).

Factors influencing the distribution of stingless bee colonies

Stingless bee colonies are not homogeneously distributed throughout the landscape. In some zones (e.g., human settlement areas with sparse vegetation), their density remains higher than in others, like agricultural landscapes and human settlement areas without significant vegetation. Many factors are responsible for their unequal distribution:

- **Availability of nesting substrate:**
  The presence of suitable nesting substrates plays a pivotal role in determining the distribution and population density of stingless bee colonies. These bees are essential pollinators within tropical ecosystems and require specific nesting materials, such as hollow tree trunks, branches, or soil cavities, which vary by species. These substrates' characteristics, quality, and quantity in a particular region can significantly influence stingless bee colonies' concentration and spatial arrangement. In areas with appropriate nesting sites, bee populations are generally more dense and widespread. In contrast, regions where nesting substrates are limited due to deforestation, urban development, or agricultural growth experience a fragmented distribution of stingless bee colonies, often resulting in population declines. The competition for scarce nesting resources can intensify this challenge, leading to conflicts among species or compelling colonies to inhabit less-than-ideal sites, which may adversely affect their health and productivity. Furthermore, the presence of natural predators and environmental pressures, including climate change, can alter the suitability of nesting substrates, affecting the overall distribution of stingless bees. Gaining insight into the connection between the availability of nesting substrates and the distribution of stingless bees is vital for conservation initiatives, as it underscores the importance of preserving natural habitats and ensuring the availability of nesting resources to sustain healthy bee populations and the ecosystems they support.

- **Availability of raw nesting materials:**
  The presence of raw nesting materials is crucial in determining the distribution and viability of stingless bee colonies. These bees depend on a range of natural substances, including resins, plant fibres, mud, and wax, for the construction and upkeep of their nests. The quantity and accessibility of these materials within an environment can influence

the locations where colonies choose to settle and their ability to thrive and expand. In areas with abundant raw nesting materials, stingless bees are likelier to flourish, resulting in greater densities of colonies. These materials are vital for the initial nest construction and ongoing maintenance and establishing protective barriers against predators and environmental threats. Conversely, regions with limited access to these resources may experience a decline in colony establishment as bees face challenges in building or maintaining their nests, potentially leading to a decrease in local populations. Human activities such as deforestation, agriculture, and urban development can further diminish the availability of raw nesting materials by transforming the landscape and reducing essential resources. This scarcity compels bees to travel greater distances to collect necessary materials, which can increase energy expenditure and negatively impact the overall health and productivity of the colony. Therefore, the distribution of stingless bee colonies is intricately connected to the availability of suitable nesting materials, underscoring the importance of conserving natural habitats and ensuring the availability of these resources for the sustainability of these vital pollinators.

- **Availability of food resources:**

Food resources are vital to stingless bee colonies' distribution and population density. These bees rely on a steady supply of nectar and pollen from flowering plants to nourish their colonies, support their larvae, and produce honey. The variety and quantity of floral resources in a specific area significantly determine the establishment and prosperity of stingless bee colonies. In regions abundant with flowering plants, particularly those that bloom year-round, stingless bees are more inclined to develop dense and stable populations, as they can readily access the essential resources needed for survival. In contrast, in areas where floral resources are limited, seasonal, or severely fragmented due to deforestation, agriculture, or urbanisation, the distribution of stingless bee colonies tends to be more dispersed and less concentrated. The bees may face difficulty locating sufficient food, resulting in longer foraging distances, which can diminish colony efficiency and survival rates. Additionally, competition for scarce floral resources can be fierce, both among different bee species and within the same species, potentially causing some colonies to be displaced. Climate change further complicates this scenario, as alterations in temperature and precipitation can disrupt the flowering patterns of plants, affecting food availability for stingless bees.

Consequently, preserving diverse, flower-rich habitats is crucial for sustaining healthy populations of stingless bees, as these environments provide the essential food resources that directly impact their distribution and ecological viability.

- **Pollution:**

Pollution profoundly affects the distribution and health of stingless bee colonies, presenting a significant threat to their survival and the ecosystems they sustain. Various forms of pollution, including air, water, and soil contamination, often stemming from industrial processes, agricultural practices, and urban development, can affect stingless bees directly and indirectly. The use of pesticides and herbicides in agriculture is particularly detrimental, as these chemicals can taint the bees' food sources—nectar and pollen—resulting in toxic exposure that compromises their foraging behaviour, reproductive success, and overall colony health. Additionally, airborne pollutants can hinder the bees' navigational abilities, as many species depend on chemical signals and pheromones for communication and orientation. This disorientation may lead to difficulty returning to their nests, consequently increasing mortality rates. Water pollution, caused by contaminating water sources with heavy metals or other hazardous substances, poses further risks when bees collect water for their colonies, introducing harmful elements into their living environment. Furthermore, soil pollution can diminish the quality of nesting sites, complicating searching for suitable locations for colony establishment. As pollution alters the availability and quality of environmental resources, it may compel stingless bees to migrate, decrease their population densities, or even result in local extinctions in areas with severe pollution. Therefore, the cumulative effects of pollution disrupt the natural distribution patterns of stingless bee colonies, underscoring the urgent need for environmental protections and sustainable practices to preserve these essential pollinators and the ecosystems they support.

CHAPTER 2

MORPHOMETRY OF INDIAN STINGLESS BEES

There are three casts of Indian stingless bees: workers (sterile females), queens (fertile females), and drones (fertile males). Workers and drones are similar in external morphology; drones are slightly darker than workers. The queen is much larger than workers and drones (Figure 2.1).

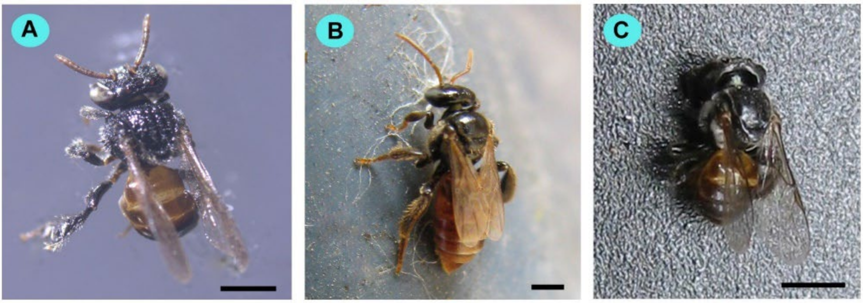

Figure 2.1. Different castes of stingless bees. (A) Worker, (B) queen, and (C) drone. Scale bars = 1 mm.

External morphology of a worker bee

A. Head

Head length is 1.38 ± 0.21 mm (1.20–1.55 mm), and width is 1.71 ± 0.12 mm (ranged: 1.60–1.82 mm). Stingless bees have five eyes- two large compound eyes and three smaller simple eyes (also called ocelli). The median ocellus width is 82.63 ± 12.54 µm. The inter-ocellar distance is 176.57 ± 14.82 µm. The ocello-ocular distance is 284.61 ± 23.03 µm. The hexagonal cells in the compound eyes are about 18 µm in length and 16 µm in breadth (Figure 2.2). Inward of the compound eyes, there are many uniparous branched hairs about 25 µm long. The antennal length is 1.82 ± 0.08 (ranges: 1.65–1.81 mm), comprises of many segments. Each segment is about 60–80 µm in length. The antennal surfaces bear many unbranched hairs about 12 µm long (Figure 2.2). The proboscis is about 1.2 mm. The inner tube is about 850 µm.

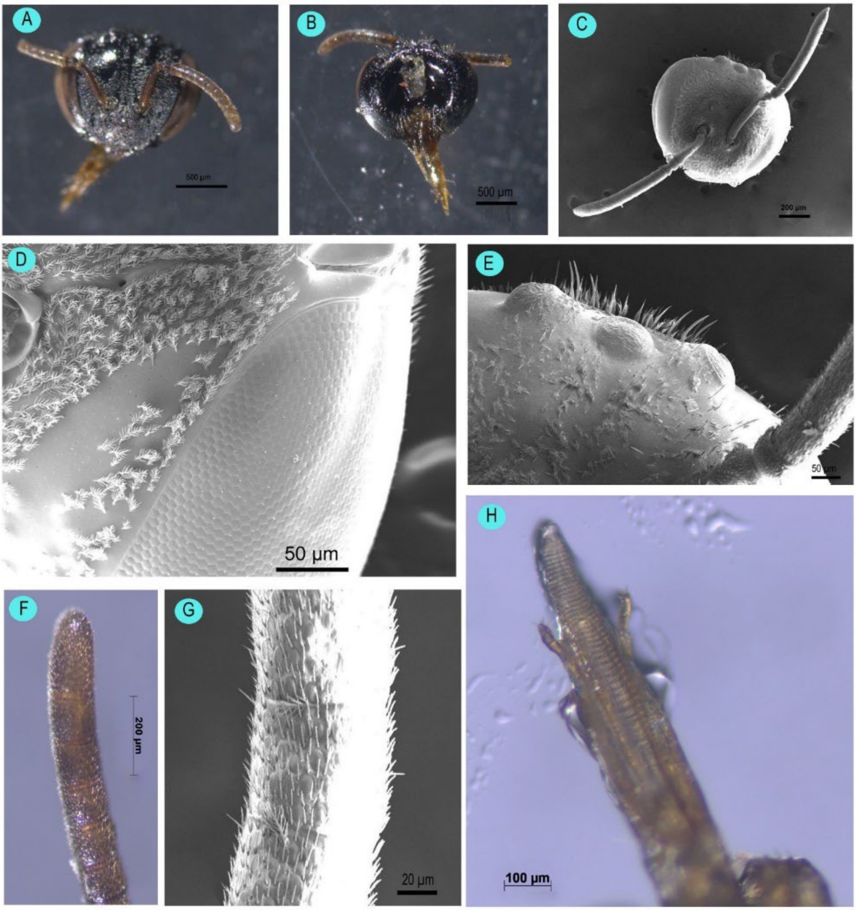

Figure 3.2. External morphology of different parts of the head. (A–C) head, (D) compound eye, (E) ocelli, (F–G) antenna, and (H) proboscis.

B. Thorax

The thorax is blackish, about 1.0 mm in length, and 1.2 mm in width. The dorsal surface bears many unbranched hairs about 80-100 µm long (Figure 2.3). The dorsal side bears two pairs of wings, and the ventral side has three pairs of legs.

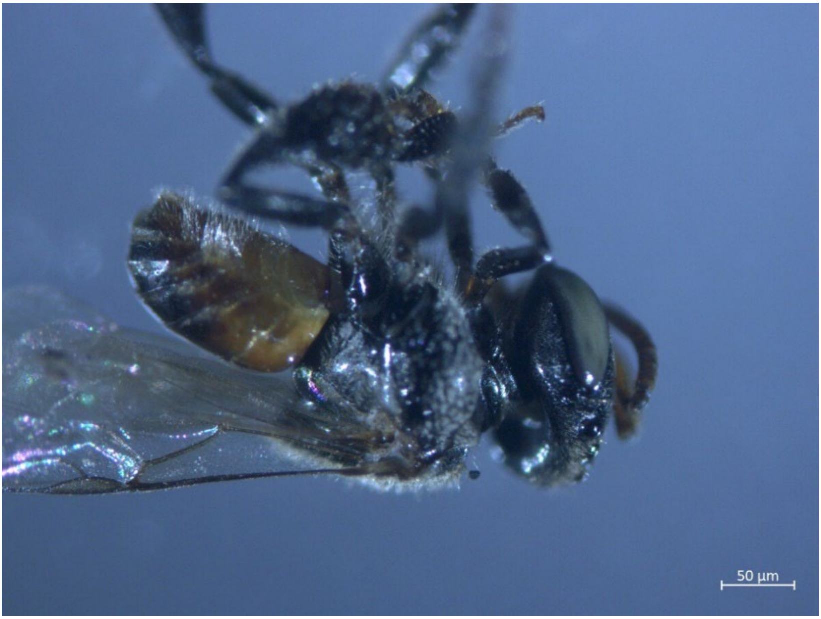

Figure 2.3. Thorax showing hairs.

Wings

Fore wings: about 2.5 mm in length and 1.1 mm in width at the distal broadest area and 300 µm. All over the surface bear minute (~20 µm) hairs (Figure 2.4).

Hind wings: about 1400 µm in length and 550 µm in width. All over the surface bear minute (~20 µm) hairs. The number of hamuli on the coastal margin is five.

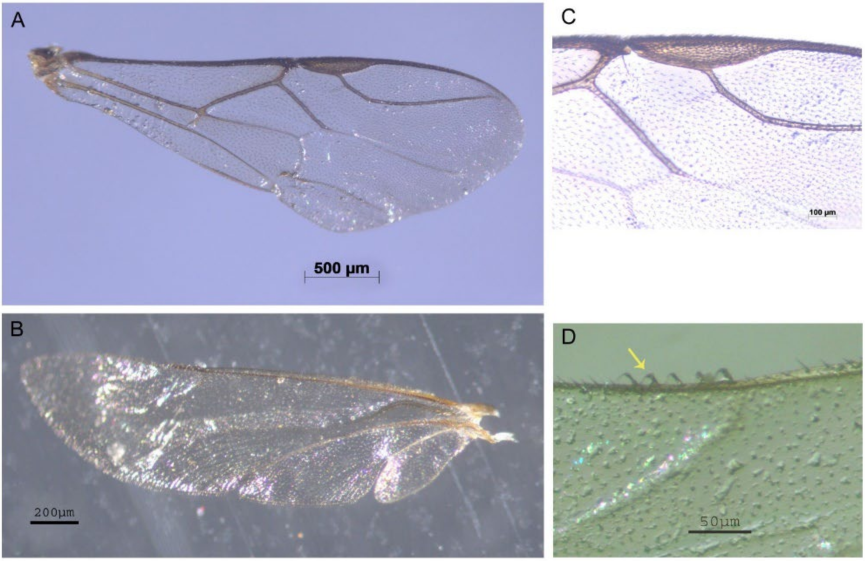

Figure 2.4. Wings of a worker bee. (A) Forewing, (B) hind wing, (C) a portion of the forewing, and (D) a portion of the hind wing showing the hamuli.

Legs

Forelegs: coxa is about 200 µm and bears hairs; trochanter is about 225 µm in length and 100 µm in width, deeply haired; femur is about 500 µm in length and 100 µm in width and bears hairs; tibia is about 400 µm in length and 80 µm in width, bear many hairs, have velum (a fan-like appendage) with 110 µm in length and 55 µm in width (Figure 2.5); tarsi are deeply haired and having antennae cleaner (a notch corresponding to the fan-like appendage); pretarsus with pointed claws.

Middle legs: These are almost similar to the forelegs but do not have fan-like appendages in the tibia and a notch in the tarsi.

Hind legs: coxa is about 250 µm and bears hairs; trochanter is about 250 µm in length and 200 µm in width, deeply haired; femur is about 550 µm in length and 100 µm in width and bears hairs; tibia is about 1000 µm in length, width varies from 700 µm at the lower region and 50 µm in the narrow upper part, bear many hairs, the posterior edge has many-branched hairs about 160 µm in length; tarsi are deeply haired, first segment flat and broad and later segments are narrow; pretarsus with pointed claws.

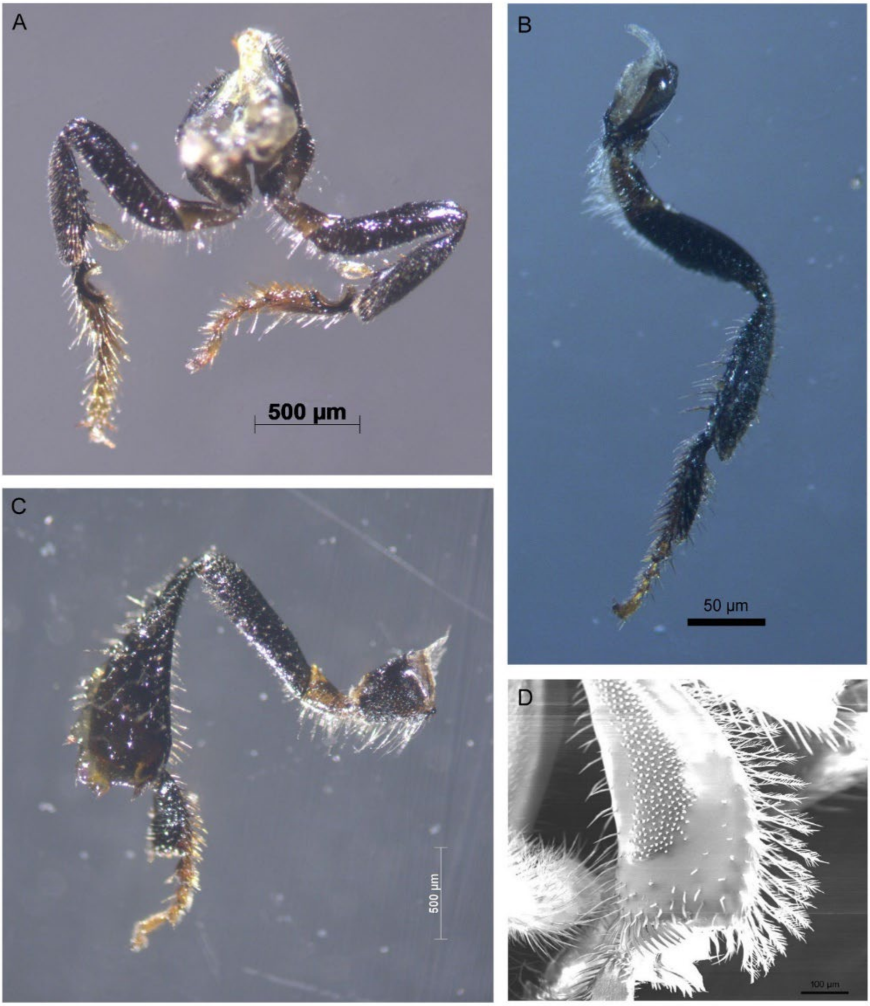

Figure 2.5. Legs of a worker bee. (A) Forelegs, (B) middle leg, (C) hind leg, and (D) corbicula.

C. Abdomen

The abdomen is about 1200 µm in length and 900 µm in width. The distal end is nearly blunt. The abdomen part is amber in colour, with a blackish zone on the dorsal surface at the distal end. Dorsal scales overlap each other and are sparsely haired (Figure 2.6). Ventral scales are also overlapping and much-haired (hairs are unbranched and about 150 µm long).

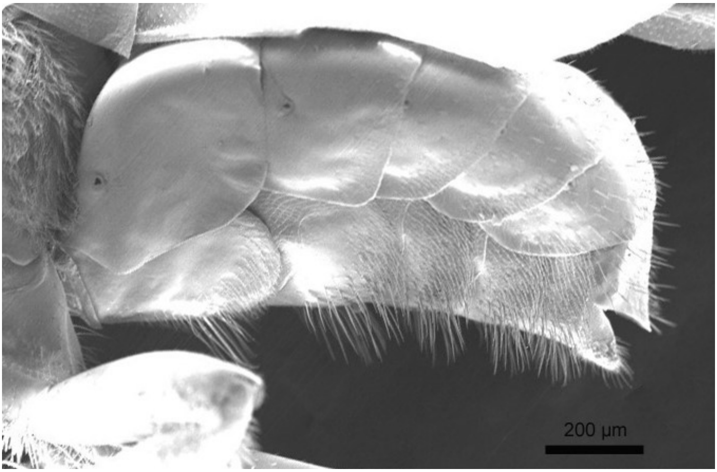

Figure 2.6. SEM image of the abdomen of a stingless bee.

External morphology of a queen

The body size of the queen is larger than that of the workers and about 7.6 mm long. The head parts are similar to that of a worker bee. The queen thorax is identical to the worker, but the hind legs do not have pollen baskets (Figure 2.7). The abdomen is longer, somewhat taper, and pointed (Figure 2.7).

Figure 2.7. (A) A queen stingless bee, (B) abdomen, (C) a part of foreleg showing velum, and (D) a part of hind leg showing ill-developed corbicula.

External morphology of a drone

The drones' bodies are similar in size to the workers' and about 5.1 mm long. Their head parts are similar to those of a worker bee. The drone's thorax is identical to the workers, but the hind legs do not have welldeveloped pollen baskets. The abdomen is shorter, somewhat blunt, and darker than the workers.

CHAPTER 3

NESTING BIOLOGY OF INDIAN STINGLESS BEES

Nesting substrate

Most researchers use visual-based methods to investigate the nesting substrates for stingless bees. The Indian stingless bees (*Tetragonula iridipennis*) are cavity-nesting species. Their substrate selection may influenced by available empty spaces. They use cavities of tree trunks, building walls, rock crevices, and ridges of agricultural fields (Layek and Karmakar 2018). Other than these, they also colonise inside electric boxes, pipes, and termitariums. Sheltering trees were diverse like *Albizia lebbeck*, *Borassus flabellifer*, *Butea monosperma*, *Ficus benghalensis*, *Ficus religiosa*, *Holoptelea integrifolia*, *Madhuca longifolia* and *Phoenix sylvestris*. Their nested walls are made of mud, mud and stones, mud and bricks, and cement and bricks (Table 3.1; Figure 3.1).

Table 3.1. Nesting substrate for Indian stingless bee.

Nesting substrate	Frequency of occurrence	Source
▪ Building wall		
Made up of mud	+++	Layek and Karmakar 2018; Saaivignesh et al. 2023
Made up of mud and brick	+	Layek and Karmakar 2018
Made up of mud and stone	+++	Layek and Karmakar 2018
Made up of cement and brick	+	Layek and Karmakar 2018
▪ Tree trunk		
Albizia lebbeck	+	Layek and Karmakar 2018
Borassus flabellifer	+	Layek and Karmakar 2018
Butea monosperma	+++	Layek and Karmakar 2018
Ficus benghalensis	++	Layek and Karmakar 2018
Ficus religiosa	++	Layek and Karmakar 2018
Holoptelea integrifolia	+	Layek and Karmakar 2018
Modhuca longifolia	+	Layek and Karmakar 2018
Morus sp.	+++	Choudhary et al. 2021
Phoenix sylvestris	+	Layek and Karmakar 2018
Tectona grandis	+	Own observation
▪ Rock crevices	+	Own observation
▪ Ridge of agricultural field	+	Layek and Karmakar 2018
▪ Pipes		
Iron pipes	+	Saaivignesh et al. 2023
Electrical pipes	+	Nayak Pavithra et al. 2013; Saaivignesh et al. 2023
Water pipes	+	Nayak Pavithra et al. 2013
▪ Electric box	+	Own observation

Note: +++ very frequent, ++ moderate frequent, + less frequent

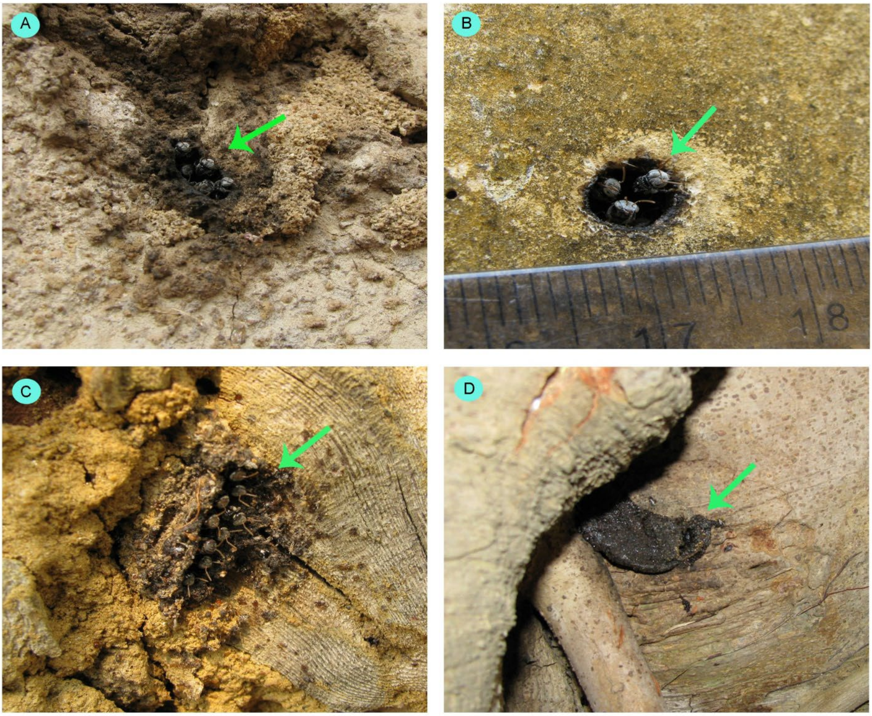

Figure 3.1. Wild colonies of Indian stingless bees. (A) Mud wall, (B) cement wall, (C) tree trunk of *Butea monosperma*, and (D) tree trunk of *Ficus benghalensis*.

Nest aggregation

The number of stingless bee colonies per site may vary from one to many. However, most sites have a single bee colony, followed by two, three, and more. Higher aggregation can be found only in a few sites. Colony aggregation is more common in walls than other nesting substrates. In the case of aggregated colonies, the minimum distance between the two nearest colonies reported by Choudhary et al. (2021) is about 12.5 cm. However, the very close (2–5 cm distance) situation of nest entrances of the stingless bee colonies is also noticeable (Figure 3.2). Aggregation of colonies may be due to the rapid division of parental colonies into sister colonies, scarcity of nesting substrate, and availability of food resources.

Figure 3.2. Two closely situated nest entrances of Indian stingless bees.

Nest height from the ground level

The heights of the nests from ground level may reach beyond 5 m, with most of the heights being 2–3 m in West Bengal (Layek and Karmakar 2018). Sometimes, stingless bees construct nests very close to ground level (Figure 3.3), especially at sites remote from human settlement. Choudhary et al. (2021) documented that the minimum distance of the nest from the ground in the North-Western plains (Ludhiana) is about 0.75 m. Multiple factors may govern their nesting height, such as flight range, availability of cavities, surrounding vegetation, and disturbance. Stingless bees prefer to construct nests comparatively at lower elevations than wild honeybees (Layek et al. 2023c). The cavity-nesting colonies of stingless bees are more protected than open-nesting colonies of honeybees, even at lower elevations.

Figure 3.3. A wild colony of Indian stingless bees situated very close to the ground level.

Nest orientation

The orientation of the entrance for wild stingless bee colonies can be divided into eight levels: north, south, east, west, northeast, southeast, northwest, and southwest. The different orientations of the nest entrance can be recorded using a GPS inbuilt service and a regular orienteering compass. The nest entrances are oriented in all directions: east, west, north, south, northeast, southwest, northwest, and southeast. Frequencies of entrance orientation vary from region to region. In West Bengal, most surveyed colonies have entrances in the west and north directions (Layek and Karmakar 2018). In Karnataka, higher percentages of nest entrances are recorded in north and north-east directions (Nayak Pavithra et al. 2013). In the north-western plains of India, most colonies preferred making a nest entrance facing east and west (Choudhary et al. 2021).

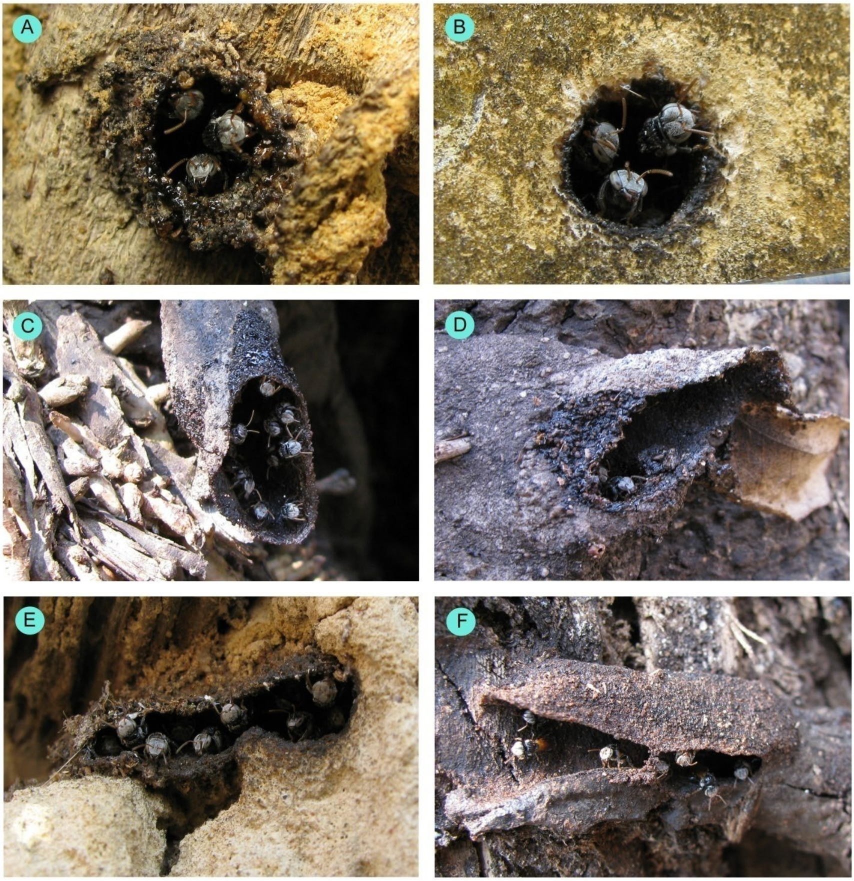

Figure 3.4. Different shapes of nest entrance. (A–B) Circular, (C–D) oval, (E) slit-like, and (F) irregular.

Nest structure

Characteristics of nest entrances can be used to identify species of stingless bees. A few colonies (especially those in a tree trunk) have an external tunnel of 15–78 mm long. The opening of nest entrances is varied in shape, like circular, oval, slit, and irregular (Figure 3.4). The entrance size may be small (about 5×4 mm^2), medium, or large (20×11 mm^2). The colour of the nest entrance may merge with the substratum or sometimes