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Multivariate morphometric analysis of dwarf honey bees, *Apis florea* Fabricius and *Apis andreniformis* Smith in North-east India

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Abstract

Aim: Morphometric study of dwarf honey bees had been undertaken from five physiographic zones of Northeast India.

Methodology: Bee samples were collected randomlyfrom various agro-ecosystems. 26 morphometric traits were measured under stereoscopic zoom microscope for morphometric study. Multivariate statistical analysis were used to investigate population variations and relationship between the groups.

Results: Two species of dwarf honey beesviz. *Apis florea* and *Apis andreniformis* were identified. The average body length of *A. florea* was 8.126±0.01 mm with average hamuli number of 12.150±0.07. In *A. andreniformis* the average body length and hamuli number were 6.940±0.08 mm and 10.467±0.03 respectively Cubital index in *A. andreniformis* was 5.214±0.03 while 2.491±0.07 in *A. florea*.

Interpretation: Principal component analysis accounted for 82.184% of the total morphological variation. *A. florea* formed two morphoclusters *viz.,* plain landraces including Brahmaputra valley and Barak valley and hill landraces including Arunachal Himalaya, Meghalaya Plateau and South eastern hill tract where they tend to cluster morphologically. Locally distributed species may be under future threat of extinction, due to changes in agricultural practices and climate change. The untapped native bee species are required to be documented properly in order to utilize the potentiality of these resources for the sustainable utilization and economic prosperity of the region.

Key words: Apis species, Dwarf honey bee, Morphometric analysis, North-east India, Physiographic zones

Exploration and morphometric studies of dwarf honey bee species from five physiographic zones of North east India

Two species of dwarf honey bees were identified	*Apis florea*	*Apis andreniformis*
Highest relative abundance	South-Eastern Hill Tract (1) and Arunachal Himalayas (1)	Brahmaputra valley (0.264)
Foraging source	69 plant species belonging to 29 families	43 plant species belonging to 13 families
The body length	7.943±0.02 mm to 8.294±0.01 mm	6.883±0.05 mm to 7.030±0.04 mm
Hamuli number	Ranged from 10 to 13	Ranged from 8 to 11
Worker bees	Reddish-brown (rufous) in appearance	Generally black in appearance
Cubital index	2.491±0.07	5.214±0.03

- Principal component analysis accounted for 82.184% of the total morphological variation among the dwarf honey bees from different physiographic zones
- The cluster and discriminant analysis revealed that *A. florea* formed two morphoclusters *viz.,* plain landraces including Brahmaputra valley and Barak valley and hill landraces including Arunachal Himalaya, Meghalaya Plateau and South eastern hill tract where they tend to cluster morphologically

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Introduction

Bees are among the most remarkable organisms on the planet. They are regarded as vital providers of ecosystem services and play an essential role in terrestrial ecosystems. Approximately, 17,000 species of bees have been identified, classified into 425 genera, 22 subfamilies and 6 families (Michener, 2000; Bingham, 1897). Based on the morphological traits, Ruttner (1995) categorized honey bees into four species: the European honey bee (*Apis mellifera*), the eastern honey bee (*Apis cerana*), the giant honey bee (*Apis dorsata*), and the dwarf honey bee (*Apis florea*). Dwarf honey bees are effective pollinators of numerous economically important crops and forest plants, and they have an extensive foraging range. (Wongsiri *et al.,* 2000). The red dwarf honey bee (*Apis florea*) is the most widespread species of dwarf honey bees, naturally found in the Indian subcontinent to the Malay Peninsula. The black dwarf honey bee (*Apis andreniformis*) is rarer and is distributed across regions from the Philippines to China and Myanmar (Oldroyd and Wongsiri, 2006; Hepburn and Radloff, 2011).

Together, *Apis florea* and *Apis andreniformis* form the subgenus *Micrapis,* representing the most primitive species within the genus *Apis*. Although both species live in a single exposed comb, they differ in terms of morphology, behavior and natural history. The workers of *Apis florea* typically have a reddish-brown (rufous) color whereas *Apis andreniformis* is darker, with the first segment of its abdomen being entirely black (Wu and Kuang, 1987). Both species are key pollinators in the semi-arid to tropical regions across various parts of Asia and Africa. They are highly effective in pollinating orchard and field crops, such as oilseeds, vegetables, fruits, and condiment plants. (Sihag, 2019). Northeast India is recognized as a hotspot for both faunal and floral biodiversity. There is a growing need for detailed studies on pollinator diversity, particularly regarding insect pollinators. Recently, many bee species have experienced significant declines in both abundance and distribution (Tahmasebi *et al.,* 2002; Cameron *et al.,* 2011).

The causes are not fully understood, but they are likely linked to pathogen spillover from commercial breeding practices, as well as changes in the agricultural methods and land use (Cameron *et al.,* 2011; Jacobson *et al.,* 2018). Additionally, climate change threatens numerous bee species globally, particularly those adapted to high-altitude environments, as suitable habitats continue to decrease (Hoiss *et al.,* 2012; Kerr *et al.,* 2015). Therefore, it is crucial to properly document the region's untapped native bee species to harness their potential for sustainable use and economic development. Morphometric studies are essential for both species identification and studying variations within populations. Morphometric analysis offers a quick, cost-effective and field-friendly method for identifying new species. It has proven to be a valuable tool for revealing both intraspecific and interspecific relationships (Amssalu *et al.,* 2004). To date, no research has focused on the morphometric characterization of dwarf honey bees across different altitudes in the North-east India. In light of the above, aim of this study was to assess the morphometric characteristics of dwarf honey bee species, comparing their characters across five physiographic zones of North-east India, while also studying their foraging sources and preferences among available floral resources under varying climatic conditions and altitudes.

Materials and Methods

Sample collection: North-east India is situated at 25.5736° N and 93.2473° E within the Indian subcontinent. A comprehensive study of dwarf honey bee species was conducted across five physiographic zones in the North-east India, namely the Arunachal Himalaya, Barak Valley, Brahmaputra Valley, Meghalaya Plateau, and the Southeastern Hill Tract. Each zone was further divided into various locations based on GPS coordinates (Table 1; Fig. 1). Bee samples were collected from plants while they were foraging for nectar or pollen using a nylon cloth sweeping hand net, and a few samples were obtained directly from bee colonies. The sampling method primarily involved random sampling across different agro-ecosystems. In addition to the collection, live photographs were taken with a Nikon D5200 camera equipped with various macrolenses. The collected samples were initially sorted in the field and then brought to the laboratory for further identification and analysis. The coordinates (latitude and longitude) and elevation of each sampling site, as well as information on the bee foraging sources, were recorded to gather ecological data on the species. After field collection, the specimens were dry-mounted on standard insect pins for identification, while the remaining specimens were preserved in 75% ethanol to prevent deformation. The relative abundance of the dwarf honey bee species was evaluated to determine how common or rare a species is in comparison to others at a specific location. It was calculated by dividing the number of species in one group by the total number of species in all groups. The result was then multiplied by 100 to express it as a percentage.

Pasturage sources: *In-situ* observations were conducted while bees were foraging on plants to identify the foraging sources of nectar and/or pollen. A plant was considered a nectar source if the bees inserted their proboscis into the flower for nectar, and a pollen source if the corbiculae became loaded with pollen during foraging. When both behaviors were observed on the same plant, it was regarded as a source of both nectar and pollen. The types of plants observed included field crops, fruit crops, ornamental plants, vegetable crops, plantation crops, medicinal and aromatic plants, trees and weeds.

Species identification: The dwarf honey bee species were examined under Leica MZ-165 stereoscopic zoom dissection microscope with a DC-300 digital camera system and identified using standard taxonomic keys (Bingham, 1897; Michener, 2000; Hepburn and Hepburn, 2005). The most reliable characters adequate for identification of *A. florea* and *A. andreniformis* are: the "thumb" of the bifurcated basitarsus of the hindleg of drones of *A. florea* is much longer than that of *A. andreniformis* (Ruttner,

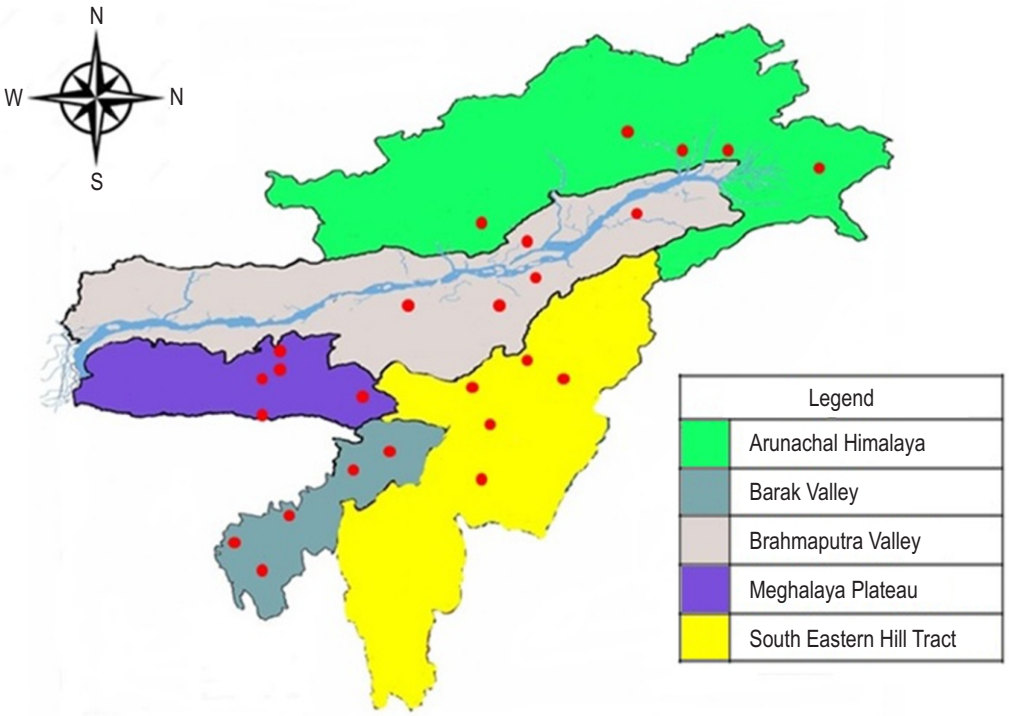

Fig. 1: Map depicting the sample collection locations for dwarf honey bees across different physiographic zones of North-east India.

Table 1: Physiographic regions of North-east India indicating sampling sites

Physiographic zone	Location	Latitude	Longitude	Altitude
Arunachal Himalaya	Pasighat	28.07°N	95.33°E	155 m
	Roing	28.14° N	95.84° E	390 m
	Basar	27.98° N	94.66° E	578 m
	Itanagar	27.1°N	93.62°E	750 m
	Hawai	27°53 N	96°48 E	1296 m
Barak valley	Agartala	23.83° N	91.26° E	12 m
	Karimganj	24.86° N	92.36° E	13m
	Udaipur	23.53°N	91.48°E	22 m
	Kailashahar	24.33° N	92.02° E	24 m
	Cachar	24.78° N	92.86° E	25m
Brahmaputra Valley	Jorhat	26.75°N	94.20° E	93m
	North Lakhimpur	27.24°N	94.11°E	96m
	Golaghat	26.59°N	93.75° E	98m
	Nagaon	26.57°N	93.00°E	70m
	Dibrugarh	27.47°N	94.92°E	110m
Meghalaya plateau	Umragnso	25.51°N	92.73° E	640m
	Umsning	25.75°N	91.89° E	782m
	Umiam	25.67°N	91.89° E	946m
	Sohra	25.28°N	91.73° E	1484m
	Sanmer	25.55N	91.84° E	1726m
South Eastern Hill Tract	Maibang	25.30° N	93.13° E	355m
	Medziphema	25.76°N	93.87° E	456m
	Imphal	24.81° N	93.90° E	786 m
	Haflong	25.16°N	93.01° E	966m
	Kohima	25.66°N	94.11° E	1445m

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Table 2: Relative abundance of dwarf honey bee populations across various physiographic regions of North-east India

Physiographic zones	Species	Total no. of individuals	Relative abundance (%)
Arunachal Himalayas	*Apis florea*	313	100
	Apis andreniformis	0	0
Barak valley	*Apis florea*	254	83.0
	Apis andreniformis	52	16.9
Brahmaputra Valley	*Apis florea*	363	73.6
	Apis andreniformis	130	26.4
Meghalaya Plateau	*Apis florea*	233	93.7
	Apis andreniformis	15	6.3
South Eastern Hill Tract	*Apis florea*	393	100
	Apis andreniformis	0	0

1988); differences in the structure of the endophallus (Lavrekhin, 1935; Wongsiri *et al.,* 1990; Koeniger, 1991); in worker bees, the jugal-vannal ratio of the hindwing of *A. florea* is greater (about 75) than that of *A. andreniformis* (about 65); and the cubital index of A. florea (about 3) is significantly less than in *A. andreniformis* (about 6). The marginal setae on the hind tibiae of *A. florea* are usually entirely white, those of *A. andreniformis* dark-brown to blackish in sclerotized individuals. Permeating the older literature is the idea that abdominal tergites 1 and 2 of *A. florea* are reddish and other segments at least partially reddish, while those of *A. andreniformis* are uniformly black (Hepburn *et al.,* 2005).

Morphometric study: Ten specimens from each location were taken for morphometric studies. Various body parts like wing, legs, sterna, mouth parts (mandibles) etc., were dissected after boiling in 10% KOH solution for 2-3 min. The morphometric characters were selected based on the morphometric studies carried out by Ruttner, 1988; Amssalu *et al.,* 2004; Hepburn *et al.,* 2005. A total of 26 morphometric characters were measured after dissecting the bees and images were analyzed by a Leica MZ-165 stereoscopic zoom dissection microscope with a DC-300 digital camera system.

Statistical analyses: Multivariate statistical analysis, *i.e.,* Principal Component Analysis, Canonical Discriminant Analysis and Cluster Analysis were used to investigate population variations and relationship between the groups. All the statistical analyses were done by SPSS 20.0 statistical package.

Results and Discussion

In the current study, two species of dwarf honey bees, *Apis florea* Fabricius and *A. andreniformis* Smith, were identified. *A. florea* was found in all five physiographic zones whereas *A. andreniformis* was recorded in three zones: the Barak Valley, the Brahmaputra Valley and the Meghalaya Plateau. The relative abundance of *Apis florea* was found to be highest in the South-Eastern Hill Tract (1) and Arunachal Himalayas (1), followed by Meghalaya Plateau (0.937), Barak valley (0.830) and Brahmaputra valley (0.736) whereas the highest relative abundance of *Apis andreniformis* was recorded from Brahmaputra valley (0.264), followed by the Barak valley (0.169) and the lowest from the Meghalaya plateau (0.063) (Table 2). A brief description of the dwarf bee species is as follows:

***Apis florea* Fabricius (1787):** Worker beeswere reddish-brown (rufous) in appearance; basal two abdominal segments were black; the pubescent on head and thorax were generally white; pale yellow to white hairs on the hind tibia and basitarsus; narrow transverse band of silky white pile at the base of the 2nd to 5th abdominal segments; wings hyaline and iridescent. In drones, the "thumb" of the bifurcated basitarsus of the hind leg was long. The average body length of *A. florea* was 8.126±0.01 mm with cubital index of 2.491±0.07. The average hamuli number of 12.150±0.07 was recorded in *A. florea.*

***Apis andreniformis* Smith (1858):** Workers were generally black; contained few rufous workers; black hairs on the hind tibia and basitarsus; white pilose transverse bands at the base of the 2nd to 6th abdominal segments; wings hyaline were observed. In drones, the "thumb" of the bifurcated basitarsus of the hind leg was short. The average body length of *A. andreniformis* was 6.940±0.08 mm with cubital index of 5.214±0.03. The average hamuli number of 10.467±0.03was recorded in *A. andreniformis.*

Morphometric study of dwarf honey bees: Comparative morphometric study of dwarf honey bees revealed that the body length of *Apis florea* varied from 7.943±0.02 mm to 8.294±0.01 mm with an average of 8.126±0.01 mm, while the body length of *A. anderniformis* varied from 6.883±0.05 mm to 7.030±0.04 mm with an average body length of 6.940±0.08 mm (Table 3; Fig. 2 a,b). The current results closely align with those of Halling *et al.* (2001), who reported that the average body length of *A. florea* in India was 9 mm. Distance between ocille (0.223±0.01 mm), ocille ocular distance (0.345±0.09 mm), antenna length (2.584±0.01 mm) and mandible length (0.989±0.13 mm) were more in *A. florea* compared to *A. anderniformis,* where it was recorded to be 0.182±0.6 mm, 0.327±0.01 mm, 2.456±0.01 mm and 2.456±0.01 mm, respectively (Fig. 2 c, d). Thadsanee *et al.* (2004) reported that the mean antennal length of *Apis florea* from

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Table 3: Comparative morphometric study of dwarf honey bees from north-east India

Characters	*Apis florea*						*Apis anderniformis*			
	Arunachal Himalayas (mm)±SD	Barak Valley (mm)±SD	Brahmaputra Valley (mm)±SD	Meghalaya Plateau (mm)±SD	South Eastern Hill Tract (mm)±SD	Mean (mm)±SD	Barak Valley (mm)±SD	Brahmaputra Valley (mm)±SD	Meghalaya Plateau (mm)±SD	Mean (mm)±SD
Body Length	8.191±0.12	7.970±0.02	7.943±0.02	8.294±0.01	8.234±0.14	8.126±0.01	6.883±0.05	6.908±0.05	7.030±0.04	6.940±0.08
Mesosoma Length	2.588±0.05	2.410±0.05	2.388±0.02	2.531±0.02	2.572±0.11	2.497±0.02	2.152±0.02	2.159±0.01	2.232±0.03	2.181±0.04
Metasoma Length	3.824±04	3.814±0.02	3.869±0.03	3.902±0.01	3.830±0.10	3.847±0.12	3.129±0.02	3.143±0.03	3.180±0.01	3.151±0.03
Head Length	1.779±0.05	1.745±0.02	1.686±0.05	1.861±0.02	1.833±0.11	1.780±0.01	1.602±0.01	1.606±0.01	1.618±0.01	1.609±0.01
Head Width	2.141±0.11	1.950±0.03	1.912±0.01	2.129±0.05	2.057±0.16	2.037±0.01	1.947±0.01	1.940±0.05	1.934±0.01	1.940±0.01
Distance Between Ocille	0.230±0.01	0.210±0.01	0.210±0.01	0.240±0.06	0.229±0.01	0.223±0.01	0.216±0.01	0.215±0.01	0.115±0.01	0.182±0.6
Ocille Ocular Distance	0.390±0.01	0.303±0.01	0.313±0.01	0.371±0.08	0.348±0.01	0.345±0.09	0.329±0.01	0.327±0.01	0.325±0.01	0.327±0.01
Antenna Length	2.611±0.02	2.530±0.02	2.545±0.02	2.634±0.08	2.602±0.05	2.584±0.01	2.459±0.05	2.455±0.02	2.453±0.01	2.456±0.01
Probosis Length	2.236±0.03	2.167±0.01	2.131±0.01	2.285±0.01	2.304±0.03	2.224±0.01	2.582±0.05	2.609±0.03	2.645±0.02	2.612±0.03
Mandible Length	1.010±0.03	0.985±0.02	0.978±0.01	0.987±0.03	0.988±0.02	0.989±0.13	0.928±0.01	0.929±0.01	0.930±0.01	0.929±0.01
Mandible Width	0.286±0.01	0.265±0.01	0.262±0.01	0.283±0.01	0.280±0.01	0.275±0.01	0.256±0.01	0.255±0.01	0.255±0.01	0.255±0.01
Fore Wing Length	5.689±0.11	5.490±0.01	5.608±0.07	6.292±0.01	6.027±0.20	5.821±0.01	5.734±0.03	5.748±0.02	5.766±0.01	5.749±0.01
Fore Wing Width	1.904±0.01	1.741±0.06	1.854±0.13	1.920±0.16	2.075±0.08	1.898±0.01	1.804±0.01	1.811±0.01	1.825±0.01	1.813±0.01
Cubital Index	2.379±0.04	2.548±0.06	2.455±0.01	2.562±0.04	2.514±0.03	2.491±0.07	5.239±0.01	5.223±0.04	5.181±0.02	5.214±0.03
Hind Wing Length	4.620±0.02	4.440±0.06	4.284±0.05	4.673±0.06	4.608±0.40	4.525±0.06	3.980±0.03	3.995±0.06	4.015±0.01	3.997±0.02
Hind Wing Width	1.405±0.01	1.370±0.07	1.379±0.02	1.416±0.06	1.397±0.06	1.393±0.01	1.325±0.0	1.325±0.01	1.326±0.01	1.325±0.01
Hamuli Number	12.440±0.51	11.700±0.02	11.250±0.20	12.960±0.04	12.400±0.06	12.150±0.07	10.200±0.06	10.400±0.03	10.800±0.02	10.467±0.03
Femur Length	1.393±0.01	1.307±0.33	1.305±0.01	1.386±0.01	1.364±0.04	1.351±0.0	1.238±0.01	1.240±0.01	1.244±0.09	1.241±0.03
Tibia Length	1.609±0.05	1.569±0.04	1.575±0.03	1.651±0.08	1.611±0.02	1.603±0.02	1.408±0.01	1.415±0.01	1.422±0.01	1.415±0.01
Meta-tarsus Length	0.990±0.01	0.972±0.01	0.977±0.01	0.992±0.03	0.988±0.02	0.983±0.10	0.933±0.02	0.934±0.02	0.935±0.01	0.934±0.01
3rd Sternite Length	1.390±0.01	1.344±0.01	1.342±0.01	1.387±0.08	1.374±0.03	1.367±0.01	1.291±0.02	1.292±0.06	1.294±0.01	1.292±0.02
3rd Sternite Width	2.245±0.05	2.237±0.05	2.088±0.05	2.258±0.02	2.218±0.08	2.209±0.01	2.172±0.12	2.164±0.01	2.157±0.02	2.164±0.01
Wax Plate Length	0.651±0.02	0.587±0.01	0.549±0.2	0.641±0.03	0.655±0.03	0.616±0.01	0.596±0.07	0.591±0.02	0.583±0.02	0.590±0.01
Wax Plate Width	1.098±0.03	1.069±0.01	1.074±0.01	1.121±0.05	1.087±0.06	1.089±0.05	1.067±0.04	1.064±0.01	1.063±0.04	1.065±0.02
Apical tergum length	0.730±0.01	0.725±0.01	0.714±0.01	0.733±0.02	0.729±0.02	0.726±0.05	0.719±0.01	0.718±0.01	0.718±0.02	0.718±0.03
Apical tergum width	1.054±0.03	1.016±0.01	1.010±0.01	1.064±0.02	1.042±0.03	1.037±0.02	1.024±0.01	1.023±0.01	1.023±0.01	1.023±0.03

Table 4: List of forage sources for dwarf honey bees from various physiographic zones of North-east India

Family	Scientific Name	Source	Flowering period	*Apis florea*	*Apis andreniformis*
Acanthaceae	*Barleria buxifolia*	Pollen-Nectar	October-January	Found	Not found
	Barleria prattensis	Pollen-Nectar	September-October	Found	Not found
	Barleria cristata	Pollen-Nectar	September-October	Found	Not found
Amaranthaceae	*Amaranthus gracilus*	Pollen	February-March	Found	Not found
	Amaaranthus spinosus	Pollen	September-October	Found	Found
	Celosia argentia	Pollen-Nectar	August-September	Found	Found
	Amaranthus viridis	Pollen-Nectar	Round the year	Found	Found
Anacardiaceae	*Mangifera indica*	Nectar	March-April	Found	Found
	Lannea coromadelica	Pollen	April- June	Found	Found
Apiceae	*Coriandrum sativum*	Pollen-Nectar	September-December	Found	Found
	Cocos nucifera	Pollen	January-December	Found	Not found
Asteraceae	*Aster* sp.	Pollen-Nectar	January-December	Found	Found
	Bidens biternata	Pollen-Nectar	July-February	Found	Not found
	Chrysanthemun indicum	Pollen-Nectar	January-December	Found	Found
	Cosmos caudatus	Pollen	November-December	Found	Found
	Guizotia abyssinica	Pollen-Nectar	September- November	Found	Found
	Zinnia elegars	Pollen-Nectar	Jul-Jan	Found	Found
Balsamnaceae	*Impatiens glandulifera*	Pollen-Nectar	July-September	Found	Not found
Bignoniaceae	*Delonix regia*	Pollen-Nectar	March-May	Found	Not found
Bombaceae	*Salmalia malabarica*	Nectar	February-March	Found	Not found
	Bombax ceiba	Pollen-Nectar	February-March	Found	Not found
Brassicaceae	*Brassica napus*	Nectar	Oct-November	Found	Found
Cannaceae	*Canna indica*	Pollen-Nectar	March-April	Found	Found
Caricaceae	*Carica papaya*	Pollen-Nectar	Whole year	Found	Found
Casuarinaceae	*Casuarina equisetifolia*	Pollen	February-July	Not found	Found
Combretaceae	*Terminalla arjuna*	Nectar,	May-June	Found	Found
Commelinaceae	*Commelina bengalensis*	Pollen	August- September	Found	Found
Compositae	*Helianthus annus*	Pollen	April-May	Found	Not found
	Ageratum conyzoides	Pollen	August-November	Found	Found
	Carthamus tenctoious	Pollen-Nectar	January-February	Found	Found
	Coreopsis grandiflora	Pollen-Nectar	July-September	Found	Found
	Cosmos sulphuresus	Pollen-Nectar	April-May	Found	Not found
	Tagetes erecta	Pollen-Nectar	January- March	Found	Found
Convolvulaceae	*Ipomoea palmate*	Pollen-Nectar	July-August	Found	Found
Cucurbitaceae	*Cucurbita maxima*	Pollen-Nectar	May-June	Found	Not found
	Benincosa hispida	Pollen-Nectar	June-August	Not found	Found
	Cucunis sativus	Pollen-Nectar	March-June	Found	Found
	Luffa acutangula	Pollen-Nectar	March-April	Found	Found
Dilleniaceae	*Dillenia indica*	Pollen-Nectar	June-July	Not found	Found
Fabaceae	*Arachis hypogaea*	Pollen	September-November	Found	Not found
	Cajan cajan	Nectar	December-March	Found	Not found
	Pisum sativum	Pollen	November-December	Found	Not found
	Vigna mungo	Pollen-Nectar	December-January	Found	Found
	Vigna radiate	Pollen-Nectar,	December-January	Found	Not found
	Lathyrus odoratus	Nectar-Pollen	January-April	Found	Found
Gramineae	*Zea mays*	Pollen	April-August	Found	Not found
	Elusine indica	Pollen	May-July	Found	Not found
Iridaceae	*Gladious* sp.	Pollen-Nectar	May-June	Not found	Found
Labiateae	*Ocinum sanctum*	Pollen-Nectar	August-September	Found	Found
	Leucas linifolia	Pollen	January-August	Found	Not found
Linaceae	*Linum usitatissimum*	Pollen	January-February	Found	Found
Malvaceae	*Hibiscus mutabilis*	Pollen-Nectar	February-March	Not found	Found
	Hibiscus rosa	Pollen-Nectar	February-May	Found	Not found
Meliaceae	*Azadirachta indica*	Nectar	April-May	Found	Found
Moraceae	*Artocarpus heterophyllus*	Pollen-Nectar	January-February	Found	Not found

Table continued

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Musaceae	*Musa* sp.	Pollen-Nectar	September- October	Found	Found
Mystaceae	*Psidium guajava*	Pollen-Nectar	March-April	Found	Found
	Callistemon linearis	Pollen-Nectar	March-April	Found	Not found
	Eucalyptus sp.	Pollen-Nectar	December-January	Found	Found
Nyctaginaceae	*Bougainvillea* sp.	Pollen,	April-May	Found	Not found
Oxalidaceae	*Averrhoa carambola*	Pollen	September-October	Found	Found
Pedaliaceae	*Sesamum indicum*	Pollen-Nectar	December-January	Found	Found
Polygonaceae	*Fagopyrus esculentum*	Pollen-Nectar	February-March	Found	Not found
Ranunculaceae	*Delphinium denudatum*	Pollen	March-April	Found	Not found
Rhamnaceae	*Zizyphus jujube*	Pollen-Nectar	August-September	Found	Not found
Rosaceae	*Prunus persica*	Pollen-Nectar	January-February	Found	Found
	Rosa sp.	Pollen-Nectar	January-December	Found	Found
Rutaceae	*Citrus sinensis*	Pollen-Nectar	April-May	Found	Not found
Sapindaceae	*Litchi chinensis*	Pollen-Nectar	February-March	Found	Not found
Solanaceae	*Solanum melongena*	Pollen-Nectar	June-September	Found	Not found
	Capsicum frutescens	Pollen	July-August	Found	Not found
	Lycopersion esculentum	Pollen	May-September	Found	Found
Verbenaceae	*Clerodendrum inerme*	Pollen-Nectar	March-April	Found	Found
Zingeberaceae	*Ellettaria cardamomum*	Nectar	July-November	Not found	Found
	Amomum sublatum	Nectar	May- September	Found	Not found

Thailand was 2.18±0.087 mm. The proboscis was recorded to be longer in *A. andreniformis* (2.612±0.03 mm) than *A. florea* (2.224±0.01 mm). The fore wing and hind wing length were recorded to be longer in *A. florea* (5.821±0.01 mm and 4.525±0.06 mm) compared to *A. andreniformis* (5.749±0.01 mm and 3.997±0.02 mm). The present findings are in conformity with Ruttner (1988) who recorded the forewing length of *A. florea* varied between 6.0 to 6.9 mm. Comparable results were reported by Makhoor and Ahmad (1998), who found the average forewing length and hindwing length of *A. florea* in the Jammu region to be 6.25 mm and 4.25 mm, respectively. The hamuli number in *A. florea* ranged between 10 to 13 with an average of 12.150±0.07, while in *A. andreniformis* it varied between 8 to 11 with an average of 10.467± 0.03 (Fig 2 g, h). The cubital index differed significantly in both the species. Cubital index in *A. andreniformis* was recorded to be 5.214±0.03, while 2.491±0.07 in *A. florea.* Wongsiri *et al.* (1990) found that the cubital index of *A. andreniformis* (6.37) was significantly higher than that of *A. florea* (2.86). They also noted that the number of hamuli in *A. florea* ranged from 10.5 to 13.2, while in *A. andreniformis* it ranged from 10.38 to 10.88. Patil *et al.* (2014) reported that the number of hamuli in *A. florea* ranged from 10.00 to 11.00, with an average of 10.71 ± 0.46. The femur and tibia were found to be longer in *A. florea* (1.351 ± 0.0 mm and 1.603 ± 0.02 mm) compared to *A. andreniformis* (1.241 ± 0.03 mm and 1.415 ± 0.01 mm). The lengths of the third sternite, wax plate, and apical tergum were quite similar in both species. Proboscis length is considered an indicator of geographical variation in some studies (Marghitas *et al.,* 2008). Additionally, body measurements may have correlations with honey yield. Szabo and Lekovich (1988) found significant positive correlations between honey production and both fore and hind wing area. Kolmes and Sam (1991) reported that honey production was strongly correlated with overall size, corbicular area, and wing measurements in honey bees. As a result, body characteristics can serve as indirect indicators of colony productivity or be used for selecting productive bees, as those with larger legs and wings tend to have greater flight power, allowing them to collect more pollen and nectar for brood rearing, thus supporting a larger colony population. Edriss *et al.* (2002) indicated that honey production could be enhanced by selecting forewing width. Similarly, Mostajeran *et al.* (2006) showed that honey production was associated with proboscis length, forewing length and width, hindwing length, leg length, femur length, tibia length, and metatarsus width. Therefore, there is a clear evidence that morphological body traits are important and correlated with the productive capabilities of the colony. Both *A. florea* and *A. andreniformis* construct nests consisting of a single exposed comb. The brood area is located below the supporting branch, while the honey storage area is positioned above and around the branch. The nests of *A. florea* are generally larger than those of *A. andreniformis*. The brood area of *A. andreniformis* has a height of 10.03 ± 3.29 cm and a width of 12.18 ± 3.62 cm, which is approximately 25% smaller in height and 16% smaller in width compared to the brood area of *A. florea,* which measures 12.00 ± 3.32 cm in height and 16.85 ± 5.28 cm in width (Wongsiri *et al.,* 1997). *A. florea* nests are typically found in more disturbed environments, such as urban and intensive agricultural areas. These bees often use small branches as a support for their nests, which are usually located in shaded spots. However, their nests are more exposed to sunlight, with the comb surface often exposed to direct sunlight for several hours each day. *A. florea* is also more likely to nest in a variety of locations, including high in tall trees, on building walls, or on building roofs. Their nesting height ranges from 1 to 15 m above the ground in dry evergreen forests. In contrast, *A. andreniformis* nests are typically found in undisturbed, mixed deciduous to evergreen forests, usually in dark and shady areas (20-35% sunlight), often near or over streams. These bees build their nests on small branches of

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Table 5: Total variation among samples of dwarf honey bees from different Physiographic zones

Component	Initial Eigen values		Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings %		
Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
13.779	52.997	52.997	13.779	52.997	52.997	7.984	30.709	30.709
3.401	13.079	66.076	3.401	13.079	66.076	5.838	22.455	53.165
2.562	9.856	75.932	2.562	9.856	75.932	4.277	16.450	69.615
1.625	6.252	82.184	1.625	6.252	82.184	3.268	12.568	82.184
0.987	3.795	85.979						
0.821	3.159	89.138						
0.709	2.728	91.866						
0.602	2.316	94.182						
0.486	1.868	96.050						
0.301	1.158	97.208						
0.222	0.854	98.062						
0.187	0.721	98.783						
0.128	0.493	99.276						
0.076	0.294	99.570						
0.046	0.176	99.746						
0.032	0.124	99.869						
0.022	0.086	99.956						
0.011	0.044	99.999						
0.00	0.000	100.000						

shrubs, bamboos, bananas, or small trees, with a single, relatively small comb located between 0.7 and 9.0 m above the ground (Wongsiri *et al.,* 1997).

In the current study, 69 plant species from 29 families were identified as forage sources for *Apis florea,* while 43 plant species from 13 families were identified as forage sources for *Apis andreniformis* across different physiographic zones of North-east India (Table 4). Layek *et al.* (2014) examined the foraging behavior of *Apis florea* during winter and spring-summer in West Bengal, identifying 71 plant species from 32 families as forage sources. Previous studies have reported varying numbers of food plants for *Apis florea:* 119 species by Sharma and Gupta (1993), 140 species by Bhat *et al.* (1990), 43 species by Kuberappa *et al.* (1996), 172 species by Singh *et al.* (2002), and 260 species by Padmanabhan and Sujana (2007). The economic importance of *Apis andreniformis* is not well documented; however, it is likely that many plants within its range rely on this species for pollination (Rattanawannee and Chanchao, 2011).

Principal component analysis of dwarf honey bees was conducted using 26 morphometric parameters, with 19 of these parameters showing significant contribution to the variation in the bee population. The analysis grouped the components into four categories, each containing a common variable that is influenced by factors with an eigenvalue greater than one.The first component accounted 52.997% of the total variation and was primarily associated with antenna length, hind wing width, third sternite length, metasoma length, distance between ocelli, wax plate width, femur length, tibia length, apical tergum width, head width, and mandible width. The second component, which accounted for 13.079% of the variation, was influenced by the width of the third sternite, head length, apical tergum length, hamuli number, wax plate length, and hind wing length. The third component explained 9.856% of the total variation and was mainly linked to meta-tarsus length, body length, cubital index, mandible length, and mesosoma length. The fourth component accounted for 6.252% of the variation, with key associations to proboscis length, forewing width, forewing length, and the distance between the ocelli. Together, these four components accounted 82.184% of the total morphological variation among the dwarf honey bees from different physiographic zones. (Table 5).

Cluster and discriminant analyses indicated that *Apis florea* from five physiographic zones formed two distinct morphoclusters: the plain landraces, which included the Brahmaputra and Barak valleys and the hill landraces, which included the Arunachal Himalaya, Meghalaya Plateau, and Southeastern Hill Tract, where they tended to cluster morphologically (Fig. 3, 4). The dendrogram, constructed using the average linkage method based on sampling locations, revealed two main clusters, A and B. Cluster A consisted of *A. andreniformis* from the Brahmaputra Valley, Barak Valley and Meghalaya Plateau (Fig. 5). Cluster B was divided into two sub-clusters, B_1 and B_2. Sub-cluster B_1 was further split into two sub-clusters, B_{1a} and B_{1b}. Sub-cluster B_{1a} included *A. florea* from the Barak Valley, while sub-cluster B_{1b} included *A. florea* from the Brahmaputra Valley. Likewise, sub-cluster B_2 was divided into sub-clusters B_{2a} and B_{2b}. Sub-cluster B_{2a} included *A. florea* from

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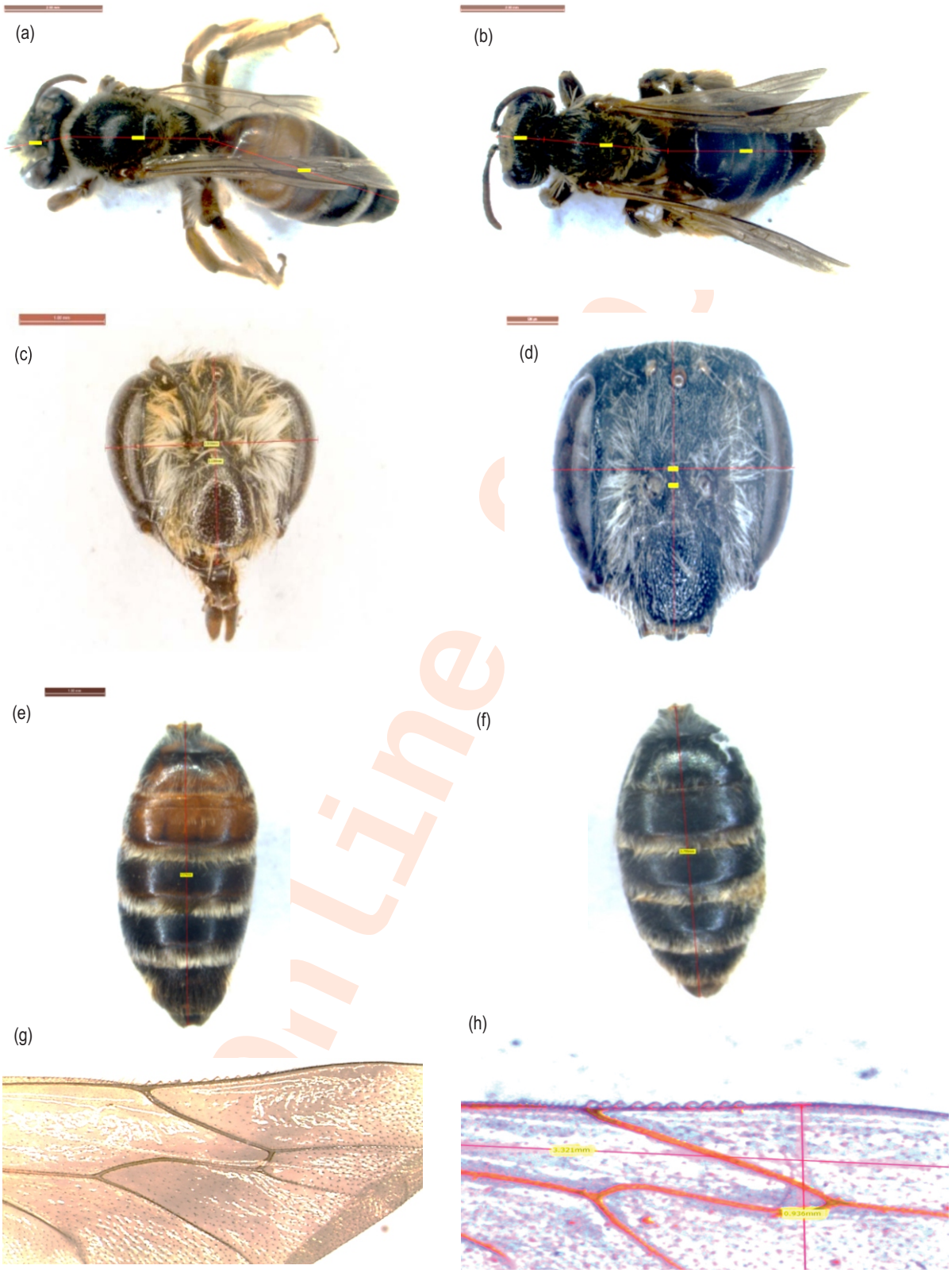

Fig 2: (a) *Apis florea* worker bee; (b) *Apis andreniformis* worker bee; (c) Head of *Apis florea* worker bee; (d) Head of *Apis andreniformis* worker bee; (e) *Metasoma* of *Apis florea* worker bee; (f) Metasoma of *Apis andreniformis* worker bee; (g) Hamuli of *Apis florea* worker bee; (h) Hamuli of *Apis andreniformis* worker bee.

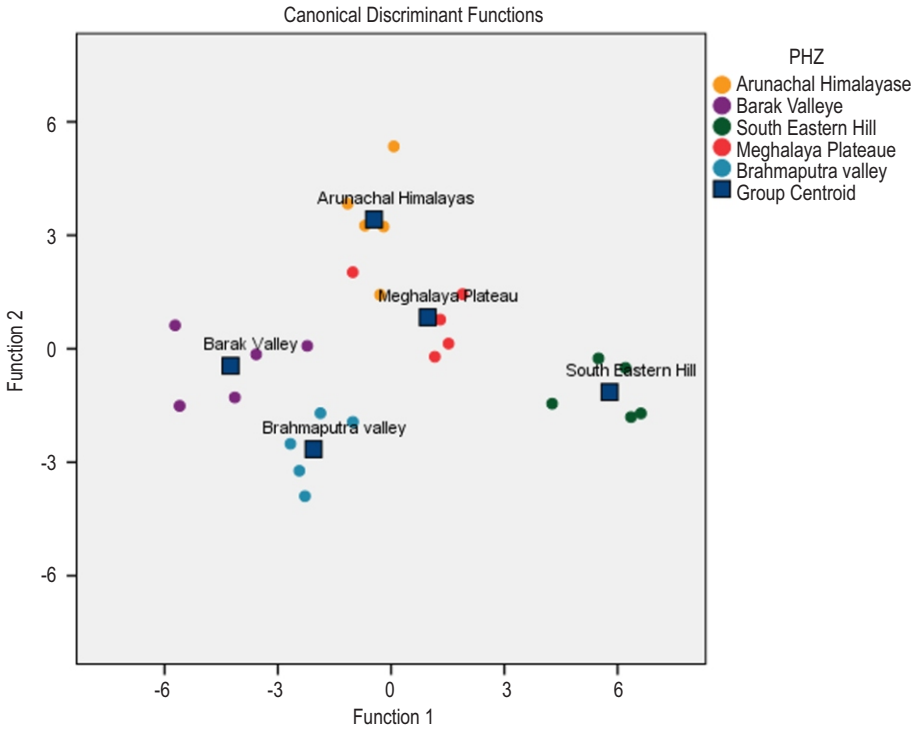

Fig. 3: Scatter plot generated from Canonical Discriminant Analysis showing clusters of *Apis florea* across various physiographic zones of North-east India.

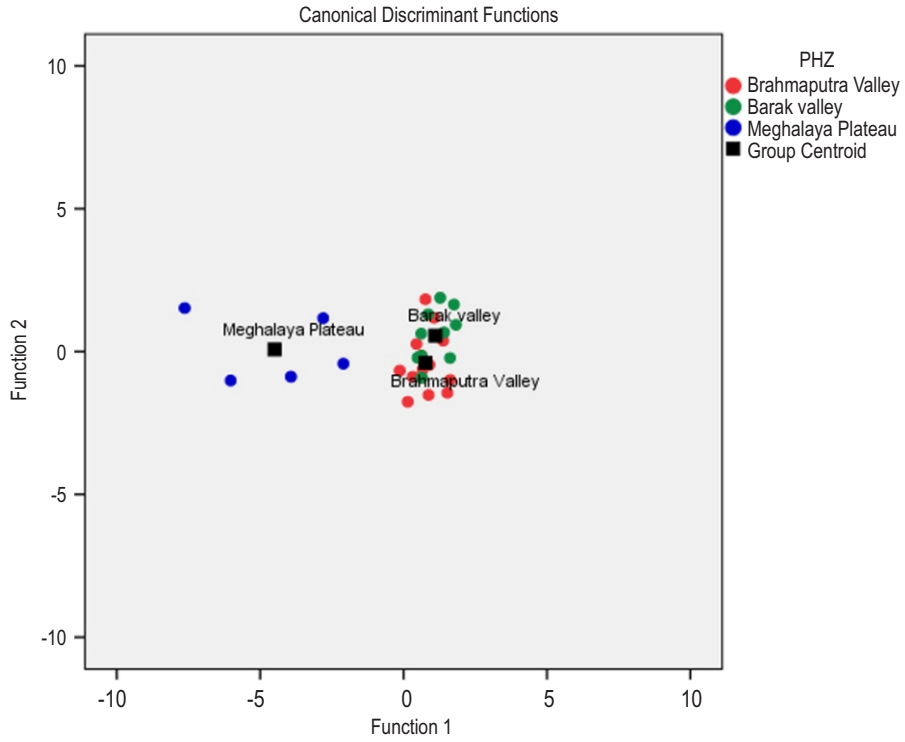

Fig. 4: Scatter plot generated from Canonical Discriminant Analysis displaying clusters of *Apis andreniformis* across various physiographic regions of North-east India.

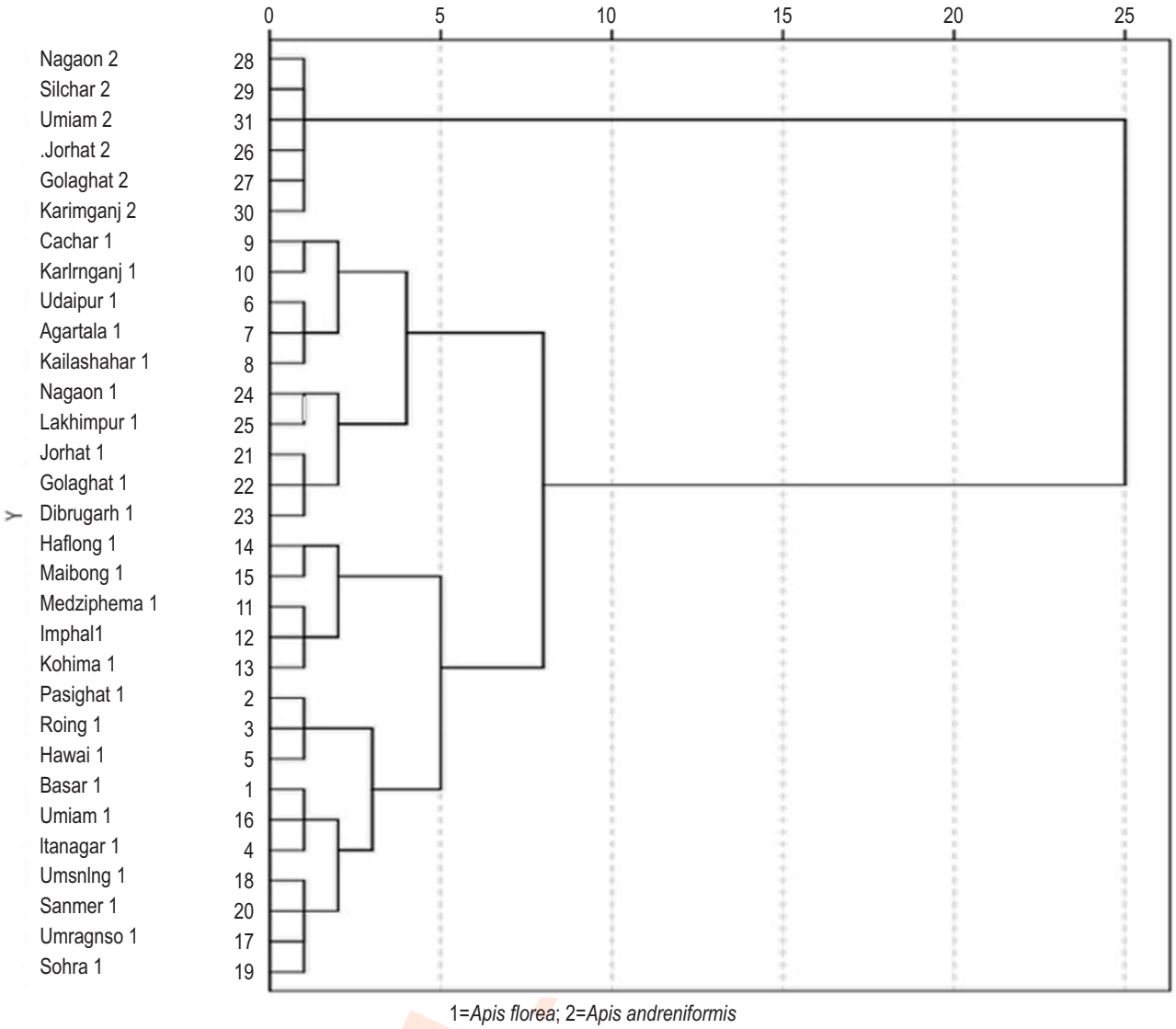

Fig. 5: Dendrogram illustrating the relationships between dwarf honey bee populations from different physiographic zones of North-east India.

the South-eastern Hill Tract, while sub-cluster B_{2b} comprised *A. florea* from the Arunachal Himalaya and Meghalaya Plateau. Singh *et al.* (1990) identified three biometric groups in the Eastern Himalayan region: Manipuri bees, Brahmaputra Valley bees, and Himalayan bees. Hepburn *et al.* (2001) also found morphoclusters linked to physiographic differences, which contributed to partial temporal reproductive isolation associated with altitude. Rattanawannee *et al.* (2007) studied the morphometric and genetic variation of small dwarf honeybee *Apis andreniformis* in Thailand and reported that factor and cluster analyses of 20 key morphological traits grouped all sampled *A. andreniformis* from Thailand and Malaysia into a single cluster. Populations of *A. florea* and *A. andreniformis* appear to be steadily declining, primarily due to habitat loss, excessive and indiscriminate pesticide use, and forest fires (Potts *et al.,* 2010). Therefore, it is crucial to find effective ways to protect these species. Both are under significant environmental stress, yet they play a vital role in pollinating both cultivated and wild plants. They are essential for agricultural productivity and for maintaining natural ecosystems and their biodiversity.

Locally adapted strains, subspecies, and ecotypes of honey bees generally experience lower losses than non-native bees. As a result, conserving these native strains as genetic resources for breeding disease- and stress-resistant lines is critical. Additionally, a comprehensive understanding of the origin and distribution of these bee species is vital to uncovering how and when these adaptations developed. Studying the evolutionary relationships of these bees would lay the groundwork for behavioral studies within an evolutionary context,

helping to explain the origin of complex social behaviors, such as the use of dance and sound to communicate the location of food or shelter. Furthermore, a global phylogenetic analysis would provide insights into divergence times and the ancestral biogeographic distributions of major bee groups.

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