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Cover: Muggar Crocodile basking on the banks of Savitri River at Mahad in Maharashtra, India. © Utkarsha M. Chavan.



## Foraging activity and breeding system of *Avicennia officinalis* L. (Avicenniaceae) in Kerala, India

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**Abstract:** Field studies were carried out to assess the foraging activity and the breeding system of *Avicennia officinalis* L. in Chettuwa, Thrissur, India during the two flowering seasons of 2018–2019. *A. officinalis*, also known as the Indian mangrove is a common mangrove occupying the river banks of the Kerala coast of southern India. The plant blooms massively during the onset of the first summer showers. The flowering to fruiting period lasted from April to July. Bagging experiments revealed that *A. officinalis* preferred a mixed breeding system though they had a low fruit set recorded in self-pollination experiments. A total of 15 species of foraging insects belonging to three orders, Hymenoptera, Diptera, and Lepidoptera, were observed. The three most abundant insect foragers were *Apis florea* Fabr., *Campsomeriella collaris* Fabr., and *Chrysomya megacephala* Fabr. Among these, *A. florea* showed a significantly high visitation rate followed by *C. collaris* and then *C. megacephala*. *C. collaris*, however, had the highest and most significant handling time for *Avicennia officinalis* than *A. florea*. The peak foraging activity was recorded from 1000h to 1100h and from 1500h to 1700h. These findings emphasize the importance of insect flower visitors in the breeding of *A. officinalis*, highlighting the need to maintain the plant-pollinator relationships for the protection of mangrove ecosystems.

**Keywords:** *Apis florea*, foraging behaviour, handling time, mangroves, pollinators, visitation rate.

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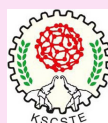
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## INTRODUCTION

Mangroves are shrubs seen in the coastal area of the tropics and the subtropics. They are well known for their irreplaceable role in protecting the coasts and for their large carbon pool. These salt-tolerant plants exhibit varied physiological and morphological adaptations that enable them to thrive in the extreme conditions of hypersalinity and tidal inundations. It is one of the important coastal ecosystems that provide numerous ecosystem services and carry out several ecological functions (UNEP 2014). This architecturally different and unique ecosystem is home to a variety of plant and animal species. Due to anthropogenic activities, however, there is a drastic decline in the mangroves around us. Nearly one-third of the mangrove forests have been lost over the past fifty years (Alongi 2002). According to the India state forest report by FSI (2019), the total area of mangroves in Kerala is about 9 km<sup>2</sup> which accounts for 0.18% of the total mangrove cover of India. *Avicennia officinalis*, belonging to the Avicenniaceae family is one of the major mangrove species occupying the coasts of Kerala. *Avicennia* species, in particular, are capable of surviving in the normal tidal range to hyper-saline conditions, thus they occupy diverse habitats (Raju et al. 2012). Hence, maintaining a highly productive ecosystem is essential to maintain the balance of nature.

Insects play a major role in many ecological processes especially pollination of mangroves. Pollination refers to the actual mechanism of transferring pollen from one flower to another (Tomlinson 1986). It is considered as one of the vital processes in plant–animal interaction. Pollinators play a crucial role in the breeding mechanisms of some mangrove plants (Pandit & Choudhury 2001). It is one of the important phenomenon in angiosperm reproduction to an extent and it is said to be coevolved and mutualistic (Huang & Giray 2012). Pollinators and their activity on mangroves of Kerala are less documented, however, in the eastern coasts, pollination biology of *Avicennia* species in the Coringa forest was documented by Raju et al. (2012). Studies on the reproductive strategies in *Aegiceras corniculatum* (L.) Blanco in the Gujarat coast was done by Pandey & Pandey (2014). The pollinators of mangroves from Sundarbans were carried out by Mitra et al. (2015). On the southern coast, Remadevi et al. (2019) documented the insect visitors of three mangrove species.

Understanding the breeding system of the plant is essential for any pollination-related studies. Mangroves exhibit both self-pollination and cross-pollination

(Kathiresan & Bingham 2001; Tomlinson 2016). Though *A. officinalis* exhibit both pollination mechanisms, self-pollination is unlikely due to protandry (Tomlinson 1986). Protandrous flowers have the male and female functions temporarily separated and the pollen is presented before the stigma becomes receptive (Imbert & Richards 1993). This study aims to determine the foragers and their foraging activity and the breeding system of *Avicennia officinalis* on the Kerala coast which might be helpful in protecting this species in this fragile ecosystem.

## MATERIALS AND METHODS

### Frequency of visits by foragers

The study was carried out in Chettuva (Peringad village 10.5463°N 76.0641°E) (Figure 1), Thrissur District, Kerala the southern coast of India during 2018–2019. Flowering seasons were ascertained through regular field visits. Insect visitors were recorded during the daytime. For this, an area of a 3 sq. m. area was marked and 50 flowers were tagged and insects visiting them were observed from 0700 to 1700 h to understand their frequency of visits and peak foraging time. Anthesis were determined in prior, thus the time was selected. Insects were recorded visually, photographed, and collected for analysis and identification. Transparent bottles were used to collect the insects. Collected insects were preserved in 70% ethanol and identified using standard literature and with the help of experts. A bar chart was plotted to better interpret the frequency of insect visits.

### Breeding experiment

To understand the breeding pattern, mature floral buds of the same age from selected inflorescences on different individual plants were tagged and enclosed in paper bags. About 50 flowers were selected to study each mode of the breeding system. The flowers were bagged with fine mesh to observe the fruit set in spontaneous autogamy. To understand the manipulated autogamy, the stigmas of the flowers were hand-pollinated and then bagged. For geitonogamy, the emasculated flowers were hand-pollinated with pollen from another flower of the same plant and for xenogamy, the emasculated flowers were pollinated with pollen from the flower of a different plant of the same species and bagged and then observed till the fruit set. One set was left for open pollination, where flowers on a different plant were tagged and the fruit set was noted as given in Raju et al. (2012).

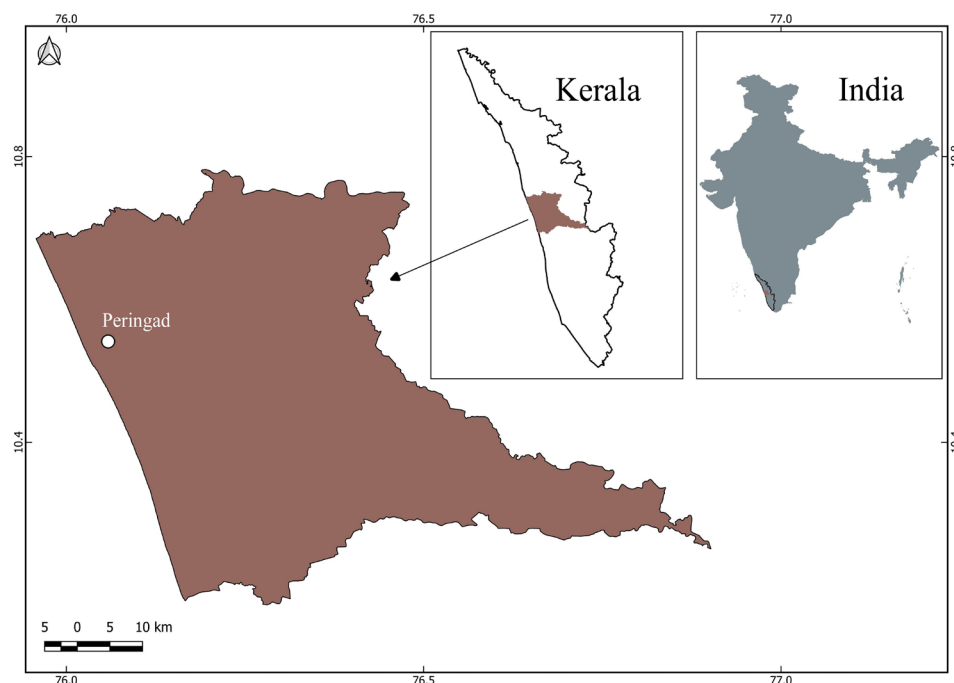


Figure 1. The study area (Peringad, Thrissur, Kerala).

#### Insect visitors and the number of flowers bloomed

The total number of insects visiting the bloomed flowers on a single branch of the plant in a minute was recorded to understand the relation between the insect visitors and the number of bloomed flowers. A total of 30 observations were recorded among the plants selected randomly.

#### Foraging behavior of insects using visitation rate and handling time

The foraging activity of insects was measured as the visitation rate and handling time, where the visitation rate can be defined as ‘the average number of flowers visited per unit time’ and handling time can be defined as ‘the average time spent on each visited flower’ (Herrera 1989). Individual insects were monitored continuously for a maximum of 2 min while they are actively foraging on the flowers of *A. officinalis*. Observations were made for 10 days during the massive flowering period each year. For, each time sequence of 2 min, the total time spent on flowers (TF—from landing to take off) and total observation time (TT—time in flowers plus time in flight between two consecutive flowers) were noted using separate stopwatches. The total number of flowers visited (NF) over the entire observation period was also recorded for each time sequence. From these observations, visitation rate (the average number of

flowers visited per unit time (NF/TT)), handling time (average time spent on each flower by an insect (TF/NF)), and flight time (average time spent in flight between two consecutive flower visits (TT-TF)/NF) were computed as given in Herrera (1989).

#### Statistical analysis

Statistical analyses were carried out using PAST 4.03 software. To analyse the relationship between the number of bloomed flowers and the number of insect visitors, correlation analysis was performed. Visitation rate (VR) and handling time (HT) were analysed using one-way ANOVA. This was carried out by taking the average value in each and was tested at a 5% significance level ( $P < 0.05$ ). Box plot was used for easy interpretation of the results.

## RESULTS AND DISCUSSION

Field observations were carried out to understand the breeding biology of *Avicennia officinalis*, and to identify major insect foragers, their visitation rate, and foraging activity on the Kerala coast. It was observed that flowering commenced in late March and extended up to July. Mature buds were seen during mid-April, and a peak in flowering was observed between late April and early May followed by June. There was a decline in

the flowering with the arrival of the monsoon and the flowering ceased by September, however, in previous studies, the flowering of *A. officinalis* on the Godavari Mangrove Forest, Andhra Pradesh, and on the Karnataka coast, both representing the East coast of India, were recorded in late summer to the end of August (Raju et al. 2012) and in July (Remadevi et al. 2019), respectively.

The flowers were fragrant, small, and sessile, 1–1.5 cm in length, yellow-orange, actinomorphic, and bisexual. Nearly 10–14 flowers were found in one unit of inflorescence and a bloomed flower lasted for six days. Stamens are four, epipetalous, filaments 2.5 mm long that are fused with corolla. Anthers are 1 mm long, basifixed alternating to petals. The ovary is superior and unilocular with four imperfect locules having one ovule each. The glabrous style has a tapering stigma that is bilobed.

Results from the bagging experiments revealed that cross-pollination resulted in better fruit set in *A. officinalis*, where the fruit formation was 70% for xenogamy, 64% for geitonogamy, 58% for open pollination, and 40% for manipulated autogamy (Table 1). In the current study, the rate of success of unmanipulated autogamy was 10%. These results were similar to the observation of Raju et al (2012). Manipulated autogamy indicated that the flowers were self-compatible, but the reduced reproductive success suggests that the pollination was vector-dependent. Raju et al. (2012) observed the role of insects like bees and flies in the pollination of *A. officinalis*. The successful formation of fruits in different modes of reproduction suggests that a flexible (or mixed) breeding system was entertained by the plant to promote outcrossing and genetic diversity through both self-pollination and cross-pollination (Primack et al. 1981; Reddi et al. 1995).

Fifteen insect species belonging to nine families and three orders (Hymenoptera, Diptera, and Lepidoptera) were identified as foragers (Table 2). Bees and flies visited the flowers in groups, whereas butterflies were found individually. Insects were observed probing in the upright position for collecting the pollen and nectar which allows the ventral part of the insects to touch the stamens and anthers. Thirteen insect species were reported as foragers in the Coringa forest in Andhra Pradesh (Raju et al. 2012). *Apis dorsata* Fabricius recorded in previous studies as one of the foragers on *A. officinalis* was not observed in the current study. On the other hand, *C. megacephala* was reported in Sunderbans and Karnataka (Raju et al. 2012; Remadevi et al. 2019; Chakraborti et al. 2019). Among the three orders, hymenopterans were found to be the most dominant

**Table 1. Results of the breeding experiments in *Avicennia officinalis*.**

Breeding system	No. of flowers pollinated	No. of Fruit set	Fruit set (%)
Autogamy	50	5	10%
Autogamy (manipulated)	50	20	40%
Geitonogamy	50	32	64%
Xenogamy	50	35	70%
Open-pollination	50	29	58%

**Table 2. Insect visitors of *Avicennia officinalis*.**

	Order	Family	Species
1.	Hymenoptera	Scolidae	<i>Campsomeriella collaris</i> Fabr.
2.		Apidae	<i>Apis florea</i> Fabricius
3.		Apidae	<i>Apis cerana indica</i> Fabr.
4.		Apidae	<i>Xylocopa</i> sp.
5.		Apidae	<i>Ceratina</i> sp.
6.		Vespidae	<i>Polistes</i> sp.
7.		Formicidae	<i>Oecophylla smaragdina</i> Fabr.
8.	Diptera	Calliphoridae	<i>Chysomya megacephala</i> Fabr.
9.		Calliphoridae	<i>Lucilia</i> sp.
10.		Sarcophagidae	<i>Parasarcophaga</i> sp.
11.		Syrphidae	<i>Eristalinus</i> sp.
12.		Dolichopodidae	<i>Dolichopus</i> sp.
13.	Lepidoptera	Nymphalidae	<i>Junonia atlites</i> L.
14.		Nymphalidae	<i>Danaus genutia</i> Cramer
15.		Nymphalidae	<i>Tirumala limniace</i> Cramer

forager group followed by dipterans and lepidopterans. In hymenopterans, Apidae were the major foragers and the most dominant pollinator species were *Apis florea* and *Campsomeriella collaris*. The greater preference for bees by *Avicennia* species was reported by Chakrabarty (1987) and Akter et al. (2020). This contrasts with the observation of Mitra et al. (2015) where the flowers of Avicenniaceae were found to be predominantly visited by dipterans with calliphorids as major visitors followed by sarcophagids, tabanids, tephritids, and drosophilids. Further, seven families of dipterans as predominant foragers of *A. officinalis* were recorded from the coast of Karnataka (Chatterjee et al. 2010).

Since hymenopterans have visited flowers more frequently than lepidopterans and dipterans, the foraging activities of two major hymenopteran species such as *C. collaris* and *A. florea* were considered for the detailed study. The foraging activity of both species showed a peak between 1000 and 1100 h in the forenoon

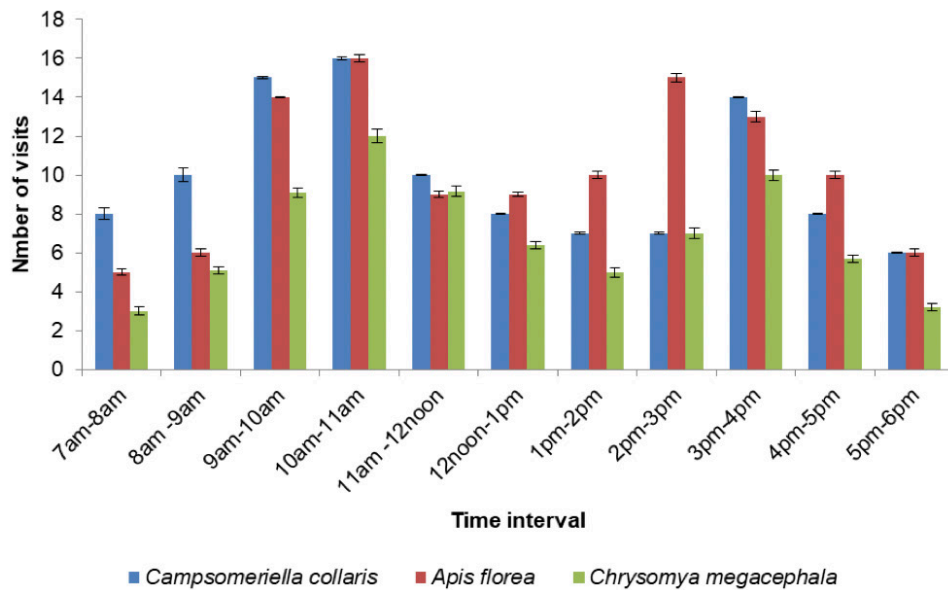


Figure 2. Hourly foraging activity of the three selected insects on *Avicennia officinalis*.

and between 1500 and 1630 h. The foraging activities were greatly dependent on resource availability and environmental parameters, especially temperature which majorly affects the activity (Mani 1982). In the study, both species favored high temperatures by increasing foraging activity to specific times of the day during the mid and late afternoon (Pandit & Choudhary 2001). On the Karnataka coast, nearly 23 insect species (Remadevi et al. 2019) and on the eastern coast, 14 insect species (Raju et al. 2012), and in Sunderbans, 23 insect species (Chakraborti et al. 2019) were reported foraging on *A. officinalis*. The foraging activities of other insects such as odonates were also noted though their role as foragers or pollinators has yet to be established (Panda et al. 2019). Moreover, the role of these insect visitors in maintaining a healthy mangrove ecosystem on the Kerala coast needs to be further elucidated. As the abundance of pollinators symbolizes a healthy environment, their periodic monitoring and assessment will be an effective tool in the environmental impact studies and management of any vulnerable ecosystem.

**Relation between flowers blooming and insect visitors**

Analyzing the relation between the number of flowers that bloomed and the number of insect visitors helps to understand pollinator behavior. The correlation study between the number of flowers that bloomed against the number of insect visitors showed a positive linear correlation ( $r = 0.88$ ,  $n = 20$ ,  $P < 0.05$ ; Figure 3) between the flower abundance and the number of insects visited. Similarly, a positive linear correlation

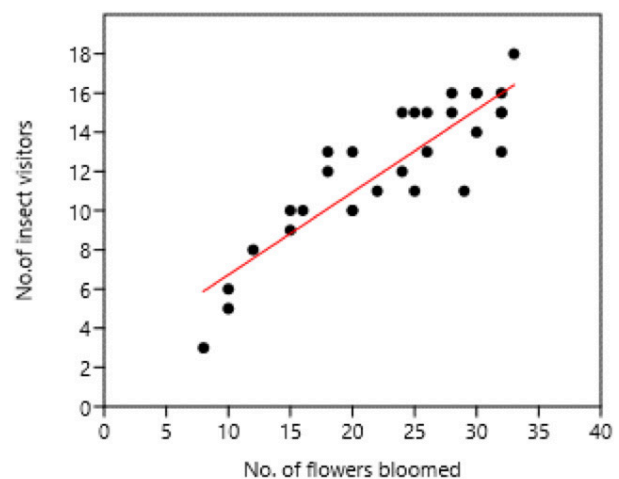


Figure 3. Correlation analysis ( $r$ ) between the number of flowers bloomed versus number of insect visitors ( $n = 30$ ). Values closer to 1 shows strong correlation and values closer to 0 indicate weak correlation.

was also observed between insect visitors and bloomed flowers in Sunderbans mangrove forests (Chakraborti et al. 2019).

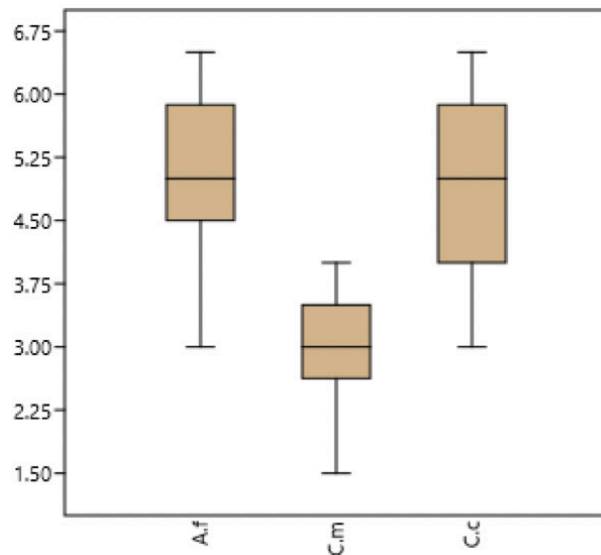
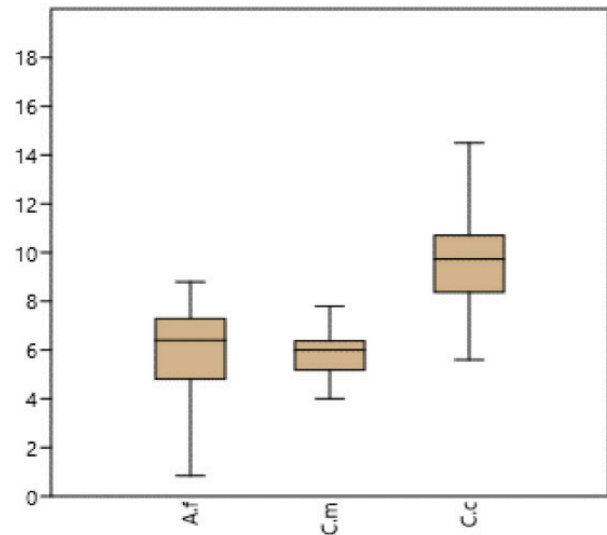
**Visitation rate and handling time**

Among the three major pollinators, *A. florea* has the highest visitation rate ( $5 \pm 0.20$  flowers/min) followed by *C. collaris* ( $4.95 \pm 0.25$  flowers/min) and *C. megacephala* ( $3.07 \pm 0.15$  flowers/min) (Figure 4). *C. collaris* has the highest handling time ( $9.59 \pm 0.42$ ) followed by *A. florea* ( $6.05 \pm 0.4$ ) and *C. megacephala* ( $5.85 \pm 0.23$ ) (Figure 5). A significant difference was noted in the visitation rate

**Table 3. One-way ANOVA of insect visitors on *Avicennia officinalis* for their visitation rate (VR) and handling time (HT).**

Test for equal means					
Visitation rate					
	Sum of squares	df	Mean square	F	P <0.05
Between groups	48.1583	2	24.0792	30.11	0.0001*
Within groups	45.5875	57	0.799781		
Total	93.7458	59	24.0792		
Handling time					
Between groups	175.396	2	87.6982	32.73	0.00003*
Within groups	152.741	57	2.67966		
Total	328.137	59			

\*Significant at P &lt;0.05.

**Figure 4. Box-plot analysis for visitation rate (VR) of three species of insects on *A. officinalis*, *Apis florea* (A.f), *Chrysomya megacephala* (C.m), *Campsomeriella collaris* (C.c).****Figure 5. Box plot analysis for handling time (HT) of three species of insects on *A. officinalis*, *Apis florea* (A.f), *Chrysomya megacephala* (C.m), and *Campsomeriella collaris* (C.c).**

and handling time between these three insect visitors on *A. officinalis* (Table 3). The significant difference is seen in the visitation rate of *A. florea* & *C. megacephala* (P-value 0.0002) and that between *C. collaris* and *C. megacephala* (P-value 0.0003) (Figure 4). It was also found that the handling time for hymenopterans was greater than that of the dipteran giving us an inference that hymenopterans could be a potential pollinator for *A. officinalis*. Visitation rates may depend upon floral characters and have insect-specific variations. Different insects respond differently to the same plant due to these reasons (Primack & Inouye 1993; Pandit & Choudhury 2001).

Mangroves are one of the most threatened

ecosystems all over the world (Alongi 2002). From the conservation point of view, this study gives us insights into the importance of insect visitors and pollinators in the fragile ecosystem of the Kerala coasts of India. More studies, however, are to be carried out to understand the different ecological requirements of various species in this ecosystem and to implement better conservation measures.

## CONCLUSION

The common mangrove *Avicennia officinalis* bloomed during mid-summer from April to July on



the Kerala coast of southern India. The three most abundant insect foragers on *Avicennia officinalis* found in this study were *Campsomeriella collaris* (Fabr.), *Apis florea* (Fabr.), and *Chrysomya megacephala* (Fabr.). Bagging experiments showed that an effective fruit set was more prominent in cross-pollinated flowers than self-pollination. *Apis florea* was an efficient forager as they showed a higher visitation rate and low handling time than *Campsomeriella collaris*. Understanding the foraging behavior help in implementing necessary measures at the habitat level for the protection of the pollinators which in turn helps in ecosystem conservation. Therefore, it is essential to protect these insect species for the long-term conservation of the mangrove ecosystem.

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### New records of pteridophytes in Mount Matutum Protected Landscape, South Central Mindanao, Philippines with notes on its economic value and conservation status

– Christine Dawn Galope-Obemio, Inocencio E. Buot Jr. & Maria Celeste Banaticla-Hilario, Pp. 22039–22057

### Some threatened woody plant species recorded from forests over limestone of the Philippines

– Inocencio E. Buot Jr., Marne G. Origenes, Ren Divien R. Obeña, Elaine Loreen C. Villanueva & Marjorie D. delos Angeles, Pp. 22058–22079

### Status of mangrove forest in Timaco Mangrove Swamp, Cotabato City, Philippines

– Cherie Cano-Mangaoang, Zandra Caderon Amino & Baingan Brahim Mastur, Pp. 22080–22085

### A comparative analysis of the past and present occurrences of some species of *Paphiopedilum* (Orchidaceae) in northeastern India using MaxEnt and GeoCAT

– Debonina Dutta & Aparajita De, Pp. 22086–22097

### Foraging activity and breeding system of *Avicennia officinalis* L. (Avicenniaceae) in Kerala, India

– K. Vinaya & C.F. Binoy, Pp. 22098–22104

### Diversity patterns and seasonality of hawkmoths (Lepidoptera: Sphingidae) from northern Western Ghats of Maharashtra, India

– Aditi Sunil Shere-Kharwar, Sujata M. Magdum, G.D. Khedkar & Supriya Singh Gupta, Pp. 22105–22117

### Population trends of Mugger Crocodile and human-crocodile interactions along the Savitri River at Mahad, Maharashtra, India

– Utkarsha Manish Chavan & Manoj Ramakant Borkar, Pp. 22118–22132

### Paresis as a limiting factor in the reproductive efficiency of a nesting colony of *Lepidochelys olivacea* (Eschscholtz, 1829) in La Escobilla beach, Oaxaca, Mexico

– Alejandra Buenrostro-Silva, Jesús García-Grajales, Petra Sánchez-Nava & María de Lourdes Ruíz-Gómez, Pp. 22133–22138

### Notes on the nesting and foraging behaviours of the Common Coot *Fulica atra* in the wetlands of Viluppuram District, Tamil Nadu, India

– M. Pandian, Pp. 22139–22147

### Population abundance and threats to Black-headed Ibis *Threskiornis melanocephalus* and Red-naped Ibis *Pseudibis papillosa* at study sites in Jhajjar district, Haryana, India

– Anjali & Sarita Rana, Pp. 22148–22155

### Crop raiding and livestock predation by wildlife in Khaptad National Park, Nepal

– Ashish Bashyal, Shyam Sharma, Narayan Koirala, Nischal Shrestha, Nischit Aryal, Bhupendra Prasad Yadav & Sandeep Shrestha, Pp. 22156–22163

## Review

### An annotated checklist of odonates of Amboli-Chaukul-Parpoli region showing new records for the Maharashtra State, India with updated state checklist

– Dattaprasad Sawant, Hemant Ogale & Rakesh Mahadev Deulkar, Pp. 22164–22178

## Short Communications

### The new addition of Blue Pimpernel of Primulaceae to the state flora of Assam, India

– Sushmita Kalita, Barnali Das & Namita Nath, Pp. 22179–22183

### A new species of genus *Neocerura* Matsumura, 1929 (Notodontidae: Lepidoptera) from India

– Amritpal Singh Kaleka & Rishi Kumar, Pp. 22184–22189

### Rediscovery of an interesting preying mantis *Deiphobella laticeps* (Mantodea: Rivetiniidae) from Maharashtra, India

– Gauri Sathaye, Sachin Ranade & Hemant V. Ghate, Pp. 22190–22194

### Camera trapping records confirm the presence of the elusive Spotted Linsang *Prionodon pardicolor* (Mammalia: Carnivora: Prionodontidae) in Murlen National Park (Mizoram, India)

– Amit Kumar Bal & Anthony J. Giordano, Pp. 22195–22200

## Notes

### First sighting record of the Orange-breasted Green-Pigeon *Treron bicinctus* (Aves: Columbiformes: Columbidae) from Chittaranjan, West Bengal, India

– Shahbaz Ahmed Khan, Nazneen Zehra & Jamal Ahmad Khan, Pp. 22201–22202

## Book Reviews

### Decoding a group of winged migrants!

– Review by Priyanka Iyer, Pp. 22203–22204

### First steps of citizen science programs in India

– Review by Aishwarya S. Kumar & Lakshmi Nair, Pp. 22205–22206

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