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continued on the back inside cover

Cover: Parasitic Hymenoptera: front—*Stilbum cyanarum* collected from Thirupathisaram, Kanyakumari District (© J. Alfred Daniel) | back—*Callipteroma sexguttata* collected from Paddy Breeding Station, Coimbatore District (© J. Alfred Daniel).

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COMMUNICATION

Camera trap survey of mammals in Cleopatra's Needle Critical Habitat in Puerto Princesa City, Palawan, Philippines

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Abstract: A camera trap survey was conducted in the recently protected Cleopatra's Needle Critical Habitat (CNCH) in Puerto Princesa City, Palawan, Philippines from February to May 2015 at 39 camera trap sites. A bait of common pig's blood was used at 36 sites, while the three remaining sites were surveyed without a bait and monitored a stream with a latrine site or mud bath with tracks. Seven native species were detected and three of these species were endemic to the island province. Species included: Common Palm Civet *Paradoxurus philippinensis*, Palawan Porcupine *Hystrix pumila*, Collared Mongoose *Urva semitorquata*, Palawan Stink Badger *Mydaus marchei*, Palawan Leopard Cat *Prionailurus bengalensis heaneyi*, Asian Small-clawed Otter *Aonyx cinereus*, and Malay Civet *Viverra tangalunga*. Analysis of the activity patterns of the three most commonly captured species revealed predominantly nocturnal activity for the Common Palm Civet, Palawan Porcupine, and Palawan Stink Badger. The Philippine Palm Civet showed occasional diurnal activity. The seven photo-captured species appeared most common, or were at the least recorded, below 750m. Five species (the Philippine Palm Civet, Palawan Porcupine, Collared Mongoose, Palawan Stink Badger, and Palawan Leopard Cat) were also recorded above 1000m. The CNCH supports two threatened species, the Palawan Porcupine and the Asian Small-clawed Otter, which are listed as Vulnerable by the IUCN, and the Collared Mongoose is listed as Near Threatened. The Palawan Leopard Cat is considered Vulnerable within the Philippines, although it has yet to be assessed by the IUCN. This documentation highlights the biodiversity significance within the newly protected critical habitat and the need to support ongoing conservation efforts within the critical habitat.

Keywords: Activity patterns, camera trap, carnivores, Felidae, Herpestidae, Hystricidae, Mephitidae, Mustelidae, Viverridae.

Abbreviations: CNCH—Cleopatra's Needle Critical Habitat | IUCN—International Union for the Conservation of Nature | SD—Secure Digital.

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Author contribution: PM—conceptualized study, collected and analyzed data, wrote final version of manuscript. SC—collected data, translated in the field. MR—collected and analyzed data. LC—supervised study, helped in the revision of the manuscript.

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INTRODUCTION

The seven carnivore species inhabiting Palawan, Philippines belong to five families: Viverridae, Herpestidae, Mephitidae, Felidae, and Mustelidae (Esselstyn et al. 2004). One porcupine species (family Hystricidae) also inhabits this island province (Esselstyn et al. 2004). Of these eight species, four are endemic to Palawan: Palawan Porcupine *Hystrix pumila* Günther, 1879; Palawan Stink Badger *Mydaus marchei* Huet, 1887; Palawan Leopard Cat *Prionailurus bengalensis heaneyi* Groves, 1997; and Palawan Bearcat *Arctictis binturong whitei* Allen, 1910. The remaining four species are indigenous: Common Palm Civet *Paradoxurus philippinensis* Jourdan, 1837; Collared Mongoose *Urva semitorquata* Gray, 1846; Malay Civet *Viverra tangalunga* Gray, 1832; and Asian Small-clawed Otter *Aonyx cinereus* Illiger, 1815. Researchers have reported the occurrence and morphology of these species in Palawan since the early 20th century (Allen 1910; Sanborn 1952; Rabor et al. 1986; Heaney et al. 1998; Esselstyn et al. 2004; Castro & Dolorosa 2006; Santiago-Flores et al. 2010; Manalo et al. 2016). Veron et al. (2015a & b) recently conducted molecular analyses of *U. semitorquata* and *P. philippinensis* in Palawan, resulting in some taxonomic changes.

Deforestation and mining are widespread in Palawan, evident in the 11% of forest loss between 2000 and 2005 and over 300 pending mining applications in 2008 (Mallari et al. 2011). Increased human immigration to Palawan has put greater stress on the land to sustain agriculture for the growing human population (Shivley & Martinez 2001). These mounting environmental pressures have been inadequately studied, but may have devastating effects on the habitats of the island's native species. We need ongoing ecological research to increase our understanding of how Palawan's wildlife will respond to the accumulating anthropogenic changes and how we can protect Palawan's wildlife.

In 2017, as part of a collaborative effort by the Centre for Sustainability PH, Inc., the City Government of Puerto Princesa, and the Palawan Council for Sustainable Development, the Cleopatra's Needle Critical Habitat (CNCH) in northeast Puerto Princesa City, Palawan was legally proclaimed, as per the Philippine Wildlife Act. This effort has safeguarded the native species occupying this forest from deforestation, while ensuring the rights of access for the Batak and Tagbanua indigenous people communities living within the critical habitat. Prior surveys of mammals within this newly-protected forest have been limited in sampling time and extent (Esselstyn et al. 2004; Marler et al. 2018).

In this study we aimed to document the eight target mammal species within the CNCH and observe patterns in the activity times of these species using baited and unbaited camera traps over a four-month period. The findings from this study contributed to the 2017 protection of the CNCH and will help guide future mammal research in this forest. We provide elevational occurrence and for the first time, activity patterns for several species of mammals in Palawan.

MATERIALS AND METHODS

Study Area

The CNCH is situated in Puerto Princesa City, Palawan, Philippines, approximately 50km north of the city proper (Figure 1b). Seven 'barangays' (Tagalog: smallest political districts in the Philippines) that comprise the 41,350-hectare critical habitat include: Binduyan, Concepcion, Langogan, New Pangangan, San Rafael, Tagabinet, and Tanabag. The centerpiece of the CNCH is Cleopatra's Needle Mountain (10.123^{0}N & 118.995^{0}E 1,593m; Figure 2). The CNCH is adjacent to the Puerto Princesa Subterranean River National Park, which extends to the west coast of the island. The CNCH is bordered by the Sulu Sea to the southeast. Major vegetation types in the CNCH include: lowland tropical/evergreen forest, lower montane forest, mossy forest, swamp forest, beach forest, and cultivated land for perennial and annual crops (Fernando et al. 2008). This study was conducted between February and May of 2015 in three political districts within the CNCH: Binduyan, Concepcion, and Tanabag. Study sites spanned lowland tropical forest (0–~900 m), lower montane forest (~900–~1100 m), and mossy forest (~1100–~1593 m) (Table 1).

METHODS

Camera trapping was conducted using Bushnell Trophy Cams (Model 119537C and Model 119436C). These trail cameras use a passive infrared (PIR) motion sensor, wherein cameras are triggered when heat passes within the detection cone of the infrared sensor. The cameras use built in infrared LED's to capture low light images and a color flash to capture brighter, daytime images. Model 119537C was capable of recording photographs or videos, while Model 119436C was capable of recording photographs followed by a video.

When camera model 119537C was used, it was set to

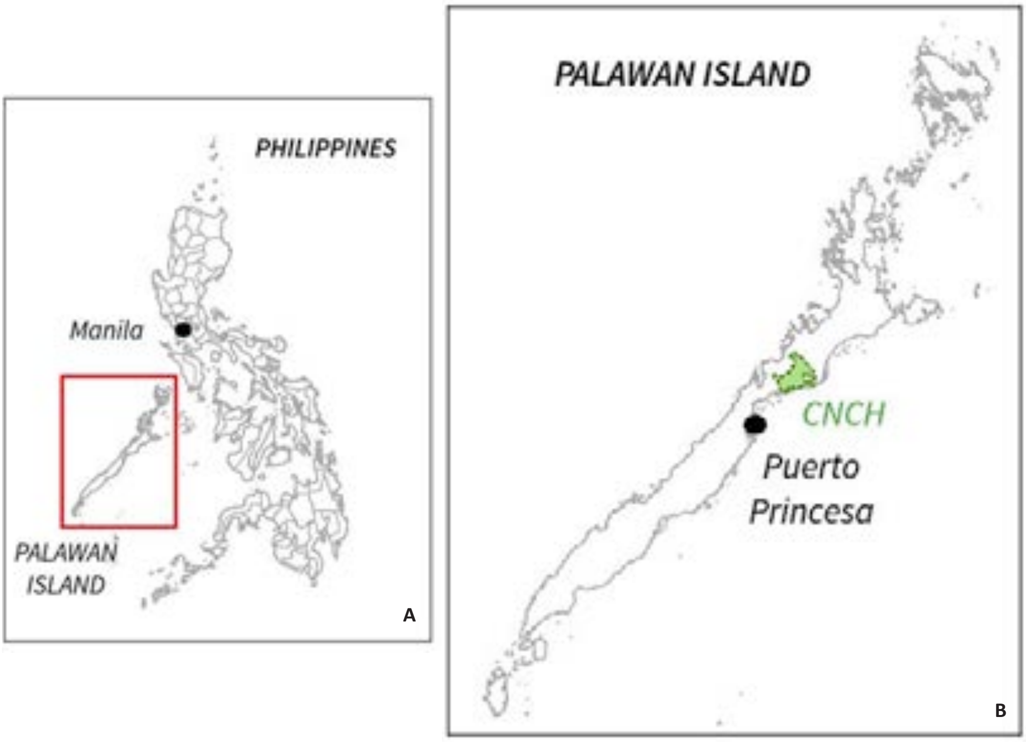

Figure 1. A—Palawan in the Philippines | B—Cleopatra's Needle Critical Habitat in Palawan. © Centre for Sustainability PH, Inc.

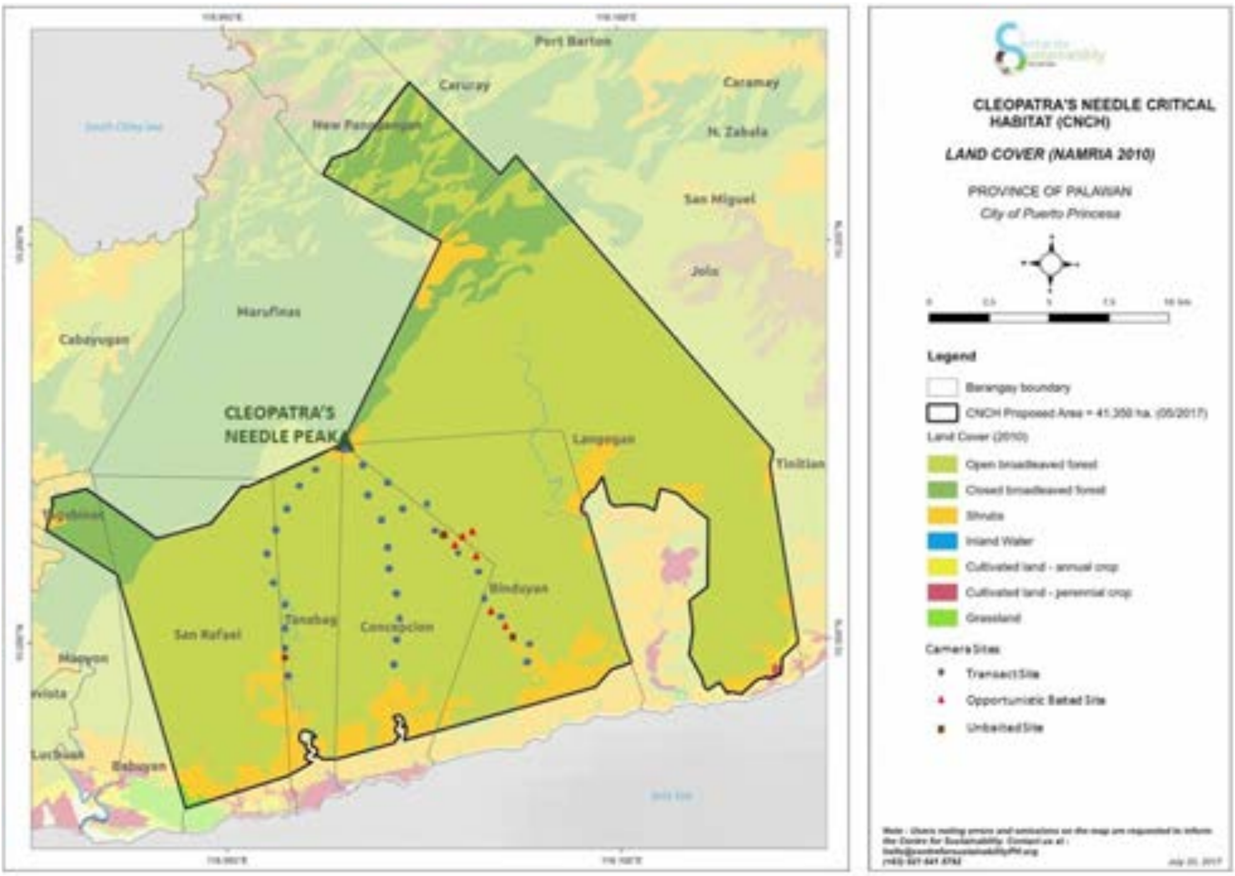

Figure 2. Map of the Cleopatra's Needle Critical Habitat with the locations of 39 camera trap sites. © Centre for Sustainability PH, Inc.

Table 1. Camera trap effort and elevational occurrence for the study sites in the Cleopatra's Needle Critical Habitat.

Study location	Camera trap effort		Trap-nights per elevation Unit (m)						
	Trap-nights	Hours	0–250	251–500	501–750	751–1000	1001–1250	1251–1500	1501–1593
Transect sites	769	8,456	129	280	96	144	96	0	24
Opportunistic baited sites	73	1,752	0	11	23	39	0	0	0
Unbaited sites	81	1,944	7	37	0	0	37	0	0
Total	923	12,152	136	328	119	183	133	0	24

take three photographs; when camera model 119436C was used, it was set to take three photographs followed by a 10-second video. A 32GB secure digital (SD) card was used in each camera. Cameras functioned for 24-hour cycles with a five-second trigger time between trigger events. Cameras were deployed at 39 locations throughout the critical habitat for a total of 12,152 trapping hours (Table 1). Thirty sites in three transects were referred to as the 'transect sites;' six sites set randomly were referred to as the 'opportunistic baited sites;' and three sites set near a stream or mud bath were referred to as the 'unbaited sites' (Figure 2).

For the transect sites, 10 cameras were set along each of the three 10-kilometer transects. Each transect extended from the southern border of the critical habitat towards the peak of Cleopatra's Needle Mountain (Figure 2), with one camera trap positioned at the peak. These sites employed the same methods used for the CNCH sites in Marler (2016). We followed regular trail routes created by hunters and by almaciga tree *Agathis philippinensis* Warb. resin collectors to reach pre-selected UTM coordinates (Gerber et al. 2010; Gerber et al. 2012). We created our own trails only when there were no existing trails. Camera trap sites were established near signs of animal presence (such as animal trails, droppings, or dig marks in the ground) at least 10m from the trail (O'Brien et al. 2003; Ancrenaz et al. 2012; Meek 2012). New coordinates were recorded using a Garmin etrex handheld GPS unit at each camera trap site. Camera traps were strapped to large trees 30–40 cm from the ground with 150ml of domestic pig's blood bait placed 2m in front of each camera trap (Thorn et al. 2009; Gerber et al. 2011; Meek 2012). This bait is likely to attract the carnivores in the forest, however, *H. pumila* is herbivorous and *A. binturong whitei* is mostly frugivorous, so they are unlikely to be strongly attracted to this bait. From the GPS coordinates recorded in the field, camera traps were ultimately spaced 1.05 km± 0.1 km (mean ± standard deviation) away from the next camera in each transect (O'Brien et al. 2003; Ancrenaz et al. 2012). These sites were surveyed for 21–39 nights.

Six opportunistic baited sites were surveyed east of the transect sites in Binduyan. Camera trap site selection was performed in the field at random, but the sites were similarly selected near signs of animal presence (O'Brien et al. 2003; Ancrenaz et al. 2012; Meek 2012). Once a site was selected, the coordinates were recorded, and the camera traps were strapped to large trees with a pig's blood bait, following the procedure for the transect sites. These sites were surveyed for 11–14 nights.

For the unbaited sites, two sites were selected near streams with *A. cinereus* spraints and one site was selected near a mud bath (1165 masl) created by Palawan Bearded Pigs *Sus ahoenobarbus* Huet, 1888 (Anito Dinampo and Pedro Mutin 2015 pers. comm.). The coordinates for these locations were recorded. Camera traps were strapped to large trees or sturdy logs at 40–90 cm above the ground and 1m away from the stream to best attain images of the stream or mud bath nearby. No bait was used in these locations. These sites were surveyed for 7–37 nights.

Upon retrieval of the camera traps in the field, SD cards were securely packed. The SD cards were observed for the presence of the eight target species and labeled accordingly from a computer in the lab. Photo-captures were recorded from the photographs and videos of the target species: if a species triggered the camera within a one-hour time frame, it was considered one photo-capture, regardless of the number of individuals in the image. The photo-captures were used to create time activity patterns for mammals with 4% or more of the target species photo-captures, following Sreekumar & Nameer (2018) who excluded carnivores under 4% of photo-captures from their time activity analysis. Photo-captures of the same species at the same camera-trap location within a one-hour time frame were considered one independent event; multiple species photo-captured in one image were each considered an independent event. We examined the independent events at various elevations across the camera trap array.

RESULTS

A total of 8,963 images and videos were recorded among 38 sites during the study period, as one camera within the transect sites malfunctioned. Seven of our target species were detected in 2,328 images and videos, with the following percentage of photo-captures: *P. philippinensis* (50%), *H. pumila* (42%), *M. marchei* (4%), *U. semitorquata* (3%), *P.b. heaneyi* (2%), *A. cinereus* (1%), and *V. tangalunga* (<1%) (Images 1–7, Table 2).

Time activity patterns were analyzed for *P. philippinensis*, *H. pumila* and *M. marchei*. We used 2,189 images for this analysis (Figure 3), which resulted in 318 independent time stamps. All three species were active throughout the night between 18.00–06.00 h. *Paradoxurus philippinensis*, however, showed activity as late as 10.00h in the morning and as early as 16.00h in the afternoon, with peak activity occurring near crepuscular hours between 04.00–06.00 h and 18.00–22.00 h. *Hystrix pumila* was active into crepuscular hours, with three peaks in activity occurring from 01.00–02.00 h, 19.00–20.00 h, and 22.00–23.00 h. *Mydaus marchei* was only found to be active at night, with stark peaks in activity between 00.00–01.00 h and 19.00–20.00 h.

Urva semitorquata was recorded eight times between 16:00 and 18:00 and three times during the day (06.54h, 09.33h and 14.41h). *Prionailurus bengalensis heaneyi* was recorded five times at night and twice after dawn (06.34h and 07.34h). *Aonyx cinereus* was recorded once at night, once at 16.51h, and twice after dawn (06.01h and 06.36h). *Viverra tangalunga* was recorded twice at 05.47h and 06.39h.

The independent events recorded for each species at various elevation ranges are found in Table 3. *Paradoxurus philippinensis* was found at every elevation range where we had camera traps and appeared most common between 250m and 1,000m. *Hystrix pumila* was documented up to 1,165m, but was more common below 1,000m. *Urva semitorquata* was photo-captured between 251m and 1,000m and twice at the peak of Cleopatra's Needle Mountain. *Mydaus marchei* was more readily found between zero and 750m, with one record at 1,233m. *Prionailurus bengalensis heaneyi* was found between zero and 1,250m, but was more common at elevations above 1,000m. *Aonyx cinereus* was recorded from the two camera trap sites by streams at 382m and 120m. *Viverra tangalunga* was recorded twice at 403m and 962m.

DISCUSSION

We recorded *P. philippinensis*, *H. pumila*, *M. marchei*, *V. tangalunga*, *U. semitorquata*, *P.b. heaneyi*, and *A. cinereus* using camera traps within the CNCH. The three most commonly photographed species, *P. philippinensis*, *H. pumila*, and *M. marchei*, exhibited predominantly nocturnal activity. The remaining species represented too small a percentage of the photo-captures to visualize activity patterns.

Three ecological studies (Allen 1910; Rabor 1986; Esselstyn et al. 2004) recorded *Arctictis binturong whitei* in Palawan. But in this study, we were unable to photo-capture this species probably because they are not attracted to pig's blood as bait. Our lack of photo-captures might also be due to our camera trap positioning on the forest floor. *Arctictis binturong whitei* is largely arboreal (Wemmer & Murtaugh 1981), hence arboreal positioned camera traps could increase the probability of photo-captures. Previous studies have recorded *A. binturong* using terrestrial camera traps in forests outside of the Philippines (Azlan & Lading 2006;

Table 2. Recorded mammals with their family, common, scientific, and local names with their corresponding IUCN status.

Family	Common name	Scientific name	Local name	IUCN Red List status
Hystricidae	Palawan Porcupine	*Hystrix pumila*	Durian	Vulnerable (Clayton 2018)
Mustelidae	Asian Small-clawed Otter	*Aonyx cinereus*	Dungon	Vulnerable (Wright et al. 2015)
Herpestidae	Collared Mongoose	*Urva semitorquata*		Near Threatened (Mathai et al. 2015)
Viverridae	Common Palm Civet	*Paradoxurus philippinensis*	Musang/ Alamid	Least Concern (Duckworth et al. 2016a)
	Malay Civet	*Viverra tangalunga*	Musang/ Tinggalong/ Tangalung	Least Concern (Duckworth et al. 2016b)
Mephitidae	Palawan Stink Badger	*Mydaus marchei*	Pantot	Least Concern (Widmann 2015)
Felidae	Palawan Leopard Cat	*Prionailurus bengalensis heaneyi*	Singgarong	Not Yet Assessed

Image 1. Camera trap image of the Common Palm Civet *Paradoxurus philippinensis*.

Image 2. Camera trap image of the Palawan Porcupine *Hystrix pumila*.

Image 3. Camera trap image of the Palawan Stink Badger *Mydaus marchei*.

Image 4. Camera trap image of two Collared Mongoose *Urva semitorquata* individuals.

Image 5. Camera trap image of the Palawan Leopard Cat *Prionailurus bengalensis heaneyi*.

Image 6. Camera trap image of five Asian Small-clawed Otter *Aonyx cinereus* individuals.

Mathai et al. 2010), but these detections were limited.

We photo-captured multiple individuals in one photograph for *H. pumila*, *U. semitorquata* and *A. cinereus*. Two and three *H. pumila* individuals were photo-captured in a single image. Two *U. semitorquata* individuals were photo-captured in a single image. Four and six *A. cinereus* individuals were photo-captured in a single image. Further data collection could help us determine average family size for these gregarious species within the CNCH.

The time activity patterns visualized for *P. philippinensis*, *H. pumila*, and *M. marchei* are similar throughout the night (Figure 4). Palawan lacks large mammals (Reis & Garong 2001), which could give the mammals in our study greater freedom to range without the danger of being preyed on. The CNCH carnivores may have less competition for resources that would otherwise be present in forests with larger predators. Further studies analyzing the time activity patterns of carnivores within the CNCH could prove mutually exclusive activity at specific hours by species with similar diets.

Species Accounts

Common Palm Civet *Paradoxurus philippinensis*: This civet is the most common carnivore in Palawan (Esselstyn et al. 2004), with widespread sightings in published surveys (Allen 1910; Sanborn 1952; Esselstyn et al. 2004; Marler et al. 2018) and the most photo-captures among mammals in this study. *Paradoxurus philippinensis*' primarily nocturnal activity contributes to other nocturnal observations for *P. hermaphroditus*, a close relative (Chetana & Ganesh 2007; Gray & Phan 2011). The occasional diurnal activity observed here was also reported by Mathai et al. (2010) for *P. hermaphroditus* in Borneo. We photo-captured this species across the elevation ranges, which mirrors this civet's common occurrence from sea level up to 2400m within the Philippines (Heaney et al. 2010).

Palawan Porcupine *Hystrix pumila*: This porcupine is endemic to the Palawan Faunal Region where it holds a Vulnerable listing in the IUCN Red List due to threats of habitat loss and hunting for the pet and bushmeat trade (Clayton 2018). *Hystrix pumila* is thought to be locally common, with several sightings during surveys in Palawan (Sanborn 1952; Heaney et al. 1998; Esselstyn et al. 2004; Manalo et al. 2016). This species had the highest record of independent events in our study, even though it was not attracted to our pig's blood bait due to its herbivorous diet. This implies that it is relatively common in the CNCH and bait is not required to obtain a large number of images. Our observations indicate that *H. pumila* is primarily nocturnal with some crepuscular activity, which is similar to the findings in Esselstyn et al. (2004) with reported activity for this species at dusk and night. Although *H. pumila* was documented up to 1,165m in our study, we found it was more common below 1000m. Previous accounts also indicate it is common from sea level to above several hundred meters

Image 7. Camera trap image of the Malay Civet *Viverra tangalunga*.

Table 3. Independent events for each recorded species at each elevational range within the Cleopatra's Needle Critical Habitat.

	Independent events by elevation (meters)							
Species	**0–250**	**251–500**	**501–750**	**751–1000**	**1001–1250**	**1251–1500**	**1501–1593**	**Total**
Common Palm Civet	6	74	22	16	7	0	4	129
Palawan Porcupine	18	47	44	21	4	0	0	134
Collared Mongoose	0	7	1	2	0	0	2	12
Palawan Stink Badger	2	9	5	0	1	0	0	17
Palawan Leopard Cat	1	1	0	1	4	0	0	7
Malay Civet	0	1	1	0	0	0	0	2
Asian Small-clawed Otter	12	8	0	0	0	0	0	20

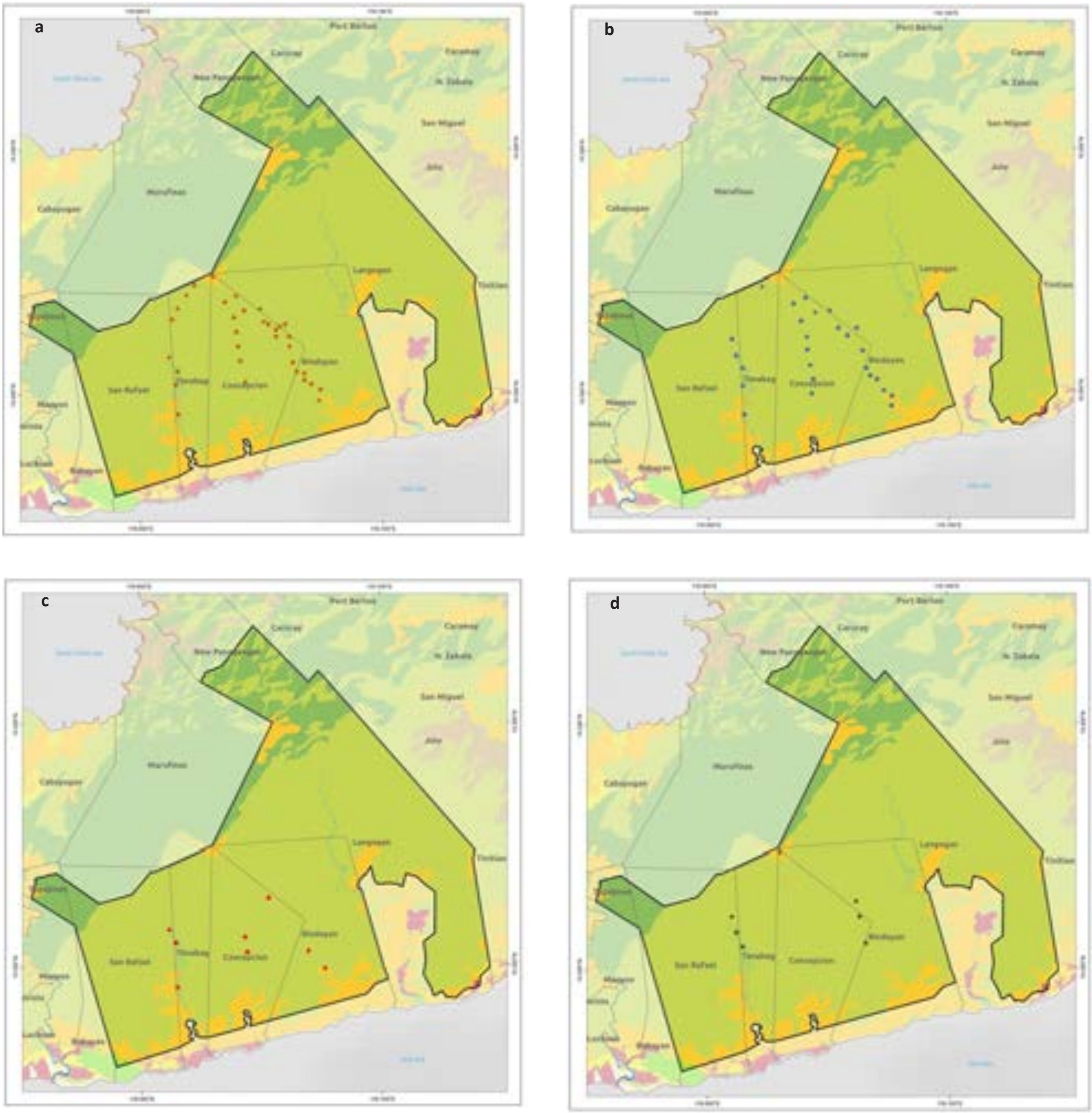

Figure 3. Occurrences of carnivores within the Cleopatra's Needle Critical Habitat: (a)—Common Palm Civet | (b)—Palawan Porcupine | (c)—Palawan Stink Badger | (d)—Collared Mongoose. ©Centre for Sustainability PH, Inc.

in elevation from disturbed to lowland forests (Heaney et al. 2010).

Palawan Stink Badger *Mydaus marchei*: This badger is endemic to and has a stable, widespread population in Palawan (Widmann 2015). This species has been reported in past surveys by sight and by smell (Sanborn 1952; Kruk 2000; Esselstyn et al 2004; Marler et al. 2018). *Mydaus marchei* is known to be nocturnal (Kruuk 2000) but has also been reported in the daytime (Grimwood 1976). Our findings support nocturnal activity for this species. *Mydaus marchei* is common in second growth and disturbed forests in Palawan (Heaney et al. 2010), suggesting it is primarily found in lowland areas where agriculture and land-modification occur. Our observations predominantly occurred in lowland tropical forest below 750m, with one sighting at 1233m.

Malay civet *Viverra tangalunga*: Scant information exists within the literature for *V. tangalunga* in Palawan (Allen 1910; Esselstyn et al. 2004), though there have been several sightings during surveys in other Philippine

Figure 3 continued. Occurrences of carnivores within the Cleopatra's Needle Critical Habitat: (e)—Palawan Leopard Cat | (f)—Asian Small-clawed Otter | (g)—Malay Civet. ©Centre for Sustainability PH, Inc.

islands (Rickart 1993; Heaney et al. 1999). Surveys of this species in Sulawesi, Malaysia and Borneo confirmed that it is primarily nocturnal with occasional daytime activity (Colón 2002; Jennings et al. 2005, 2010; Mathai et al 2010). Our two sightings occurred around dawn. This species is found from sea level to 1600m in the Philippines (Heaney et al. 2010), hence, our limited data contributes to this elevational range of occurrence.

Collared mongoose *Urva semitorquata*: *Urva semitorquata* is listed by the IUCN as Near Threatened due to habitat reduction from deforestation (Mathai et al. 2015); however, specific threats to the Palawan populations have yet to be assessed. This mongoose is only known to occur in Palawan and Busuanga islands in the Philippines (Heaney et al. 1998); few published sightings of 1 to 3 individuals per study exist in Palawan (Allen 1910; Sanborn 1952; Rabor et al. 1986). The diurnal observations reported in our study were consistent with diurnal observations of *U. semitorquata* in Borneo (Cheyne et al. 2010; Brodie & Giordano 2011).

Figure 4. Time activity patterns for three mammals in the Cleopatra's Needle Critical Habitat.

This species is known at low elevations in Palawan, near rivers (Heaney et al. 2010). We predominantly found this species between 250m and 1,000m, however, we photo-captured this species twice at the peak of Cleopatra's Needle. Further elevational studies for *U. semitorquata* could support an expansion of its known elevational range on Palawan.

Palawan Leopard Cat *Prionaillurus bengalensis heaneyi*: This leopard cat subspecies is only found in Palawan (Groves 1997) where it holds a Vulnerable listing within the Philippines (Department of Environment & Natural Resources 2017; Gonzalez et al. 2018). The subspecies has not yet been assessed by the IUCN. Published records of *P.b. heaneyi* are sparse (Allen 1910; Sanborn 1952; Rabor et al. 1986; Esselstyn et al 2004; Marler et al. 2018). Activity patterns for this subspecies do not exist, but activity pattern studies for *P. bengalensis* in Borneo and Thailand confirmed that the species is nocturnal (Grassman et al. 2005; Cheyne & Macdonald 2011; Lynam et al. 2013) with some crepuscular activity (Grassman et al. 2005). Saxena & Rajanshi (2014) also observed diurnal activity in India. Our photo-captures were at night and dawn. Leopard cats are found from 0m to 1,500m within the Philippines (Heaney et al. 2010). We similarly recorded this species from low to high elevations, with more photo-captures above 1,000m.

Asian Small-clawed Otter *Aonyx cinereus*: This otter is only found in Palawan within the Philippines. The IUCN lists this species as Vulnerable and the Department of Environment and Natural Resources (2017) lists this species as Endangered within the Philippines. This species has been reported by sight and by their droppings within Palawan (Esselstyn et al. 2004; Castro & Dolorosa 2006; Marler et al. 2018). *Aonyx cinereus* studies in Malaysia reveal nocturnal and crepuscular activity (Foster-Turley 1992). Our few sightings reflected this activity pattern with one sighting in the late afternoon. *Aonyx cinereus* is believed to occur in lower portions of rivers in Palawan (Heaney et al. 2010), which is reflected in our findings at our unbaited sites beside rivers.

CONCLUSION

Primary forest is at risk of being converted and lost as mining pressures (Mallari et al. 2011) and anthropogenic land modification (Shivley & Martinez 2001) increase in Palawan. This habitat loss coupled with hunting pressures for various species (Castro & Dolorosa 2006; Clayton 2018) and lack of proper environmental law enforcement (Castro & Dolorosa 2006) makes conservation work on the island a high priority. The seven species observed here appeared most common, or were at least recorded, below 750m in lowland tropical forest. This lowland area is prime location for land modification, such as agriculture and logging, and is thus a crucial area to protect. The protection of the CNCH in 2017 was monumental for Palawan's wildlife and indigenous communities. We need to support the ongoing research to understand the species within the CNCH and develop management strategies to ensure their survival.

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Habitat suitability modeling of Asian Elephant *Elephas maximus* (Mammalia: Proboscidea: Elephantidae) in Parsa National Park, Nepal and its buffer zone

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Abstract: Asian Elephants *Elephas maximus* in Nepal are known to have habitats and movement corridors in Parsa National Park (PNP) and its buffer zone (BZ), located east of Chitwan National Park. A study was conducted in this area to assess the suitability of PNP and BZ as elephant use areas, and to determine factors relevant to the presence of elephants in PNP. Field measurements were carried out in 67 plots for vegetation analysis. Boosted Regression Tree (BRT) analysis was used to examine the relationship of habitat suitability and variables including topography (slope, aspect, altitude), climate (precipitation, temperature), habitat preference, ground cover and crown cover. The results indicate that elephant habitat suitability is mainly determined by the dominant plant species, temperature, altitude, habitat preference and precipitation. Slope, ground cover, crown cover and substrate have lesser effects. Elephants were recorded up to 400m in the northeast and southeast aspects of the study area. Most suitable habitats were low slope forest dominated by *Acacia catechu* and *Myrsine semicerate* that received 300mm annual precipitation. The model emphasizes environmental suitability, and contributes to knowledge for conservation of elephants in PNP and BZ by delineating sites that require specific planning and management.

Keywords: Boosted Regression Tree, corridor, elephant habitat suitability, important value index, vegetation.

Nepali Abstract: नेपालमा पाइने जङ्गली हात्तीको वासस्थान र क्रियाकलाप विशेषत चितवन राष्ट्रिय निकुन्जको पुर्वी क्षेत्रमा पर्ने पर्सा राष्ट्रिय निकुन्ज, यसको मध्यवर्ती क्षेत्रमा रहेको छ । यस अनुसन्धानले जङ्गली हात्तीको वासस्थानको उपयुक्तता, हात्तीले प्रयोग गर्ने क्षेत्र र हात्तीको उपस्थितीका कारकको बारेमा अध्ययन गरेको छ । स्थलगत रुपमा ६७ वटा प्लटको स्थापना गरी उक्त प्लटभित्र रहेका वनस्पतीहरुको विश्लेषण गरिएको थियो । बुस्टेड रिग्रेसन ट्री विधि लागू गरेर हात्तीको वासस्थान उपयुक्तता र कारकहरु जसमा स्तलाकृति (उचाइ, भिरालो, मोहडा), जलवायु (वर्षा, तापक्रम), वासस्थान प्राथमिकता, जमीन आवरण, छत्र आवरण जस्ता विभिन्न कारकहरुको अध्ययन गरिएको थियो। अध्ययनको परिणामले हात्तीको वासस्थान मुख्यतया प्रमुख वनस्पती प्रजाती, तापक्रम, उचाइ, वासस्थान प्राथमिकता र वर्षा निर्धारण गर्छ भनेर जनाएको छ । भिरालोपन, जमीन आवरण, छत्र आवरण र अरु कारकहरुले हात्तीको बासस्थान निधारर्णमा कम असर गर्ने देखियो । अध्ययन गरिएको क्षेत्रबाट ४०० मि. उत्तरपुर्वी र दक्षिणपुर्वी मोहडासम्म हात्ती सकृय रहेको पाइयो । न्युन भिरालोपन भएका खयर (*Acacia catechu*) र कालीकाठ (*Myrsine semicerate*) प्रजातीले ढाकेको वनक्षेत्र जहा वार्षिक वर्षादर ३०० मिमि रहन्छ त्यस्तो क्षेत्र सबैभन्दा उपयुक्त बासस्थानको रुपमा पाइएको छ । उल्लेखित क्षेत्र तोकेर यस मोडलिंगले हात्तीको बासस्थानको वातावरणिय उपयुक्तता र पर्सा र यसको मध्यवर्ती क्षेत्रमा हात्तीको संरक्षणको लागी ज्ञान प्रदान गर्नतर्फ जोड दिएको छ, जुन कुरा हात्तीको वासस्थान व्यवस्थापन र योजनामा आवश्यक पर्छ ।

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INTRODUCTION

The Asian Elephant was recognized as an endangered species in 1975 after its inclusion in Appendix I of CITES (Convention on International Trade in Endangered Species of Wild Fauna and Flora) (Bisht 2002) and listed as "Endangered" on the IUCN Red List of Threatened Species (IUCN 2017). These elephants are found in a variety of habitats that include grasslands, tropical evergreen, moist deciduous, dry deciduous and dry thorn forests, as well as secondary forest, scrublands, and cultivated areas (Sukumar 2003). Armbruster & Lande (1993) stated that human encroachment of natural habitats is one of the most critical issues facing elephant conservation. In Asia, elephants have lost extensive habitat areas, and as a result, conflicts with people have increased (Santiapillai 1997).

In Nepal, elephants are distributed throughout the lowland Terai in four isolated populations ranging over 10,982km^2 of forest habitat (DNPWC 2008). The estimated number of resident wild elephants in Nepal is between 107 and 145 (DNPWC 2008; Pradhan et al. 2011). The eastern population has 7–15 resident animals and 100 migratory animals from India. In central Nepal, 20–25 elephants reside primarily in Parsa National Park (PNP) and Chitwan National Park (CNP). The western and far western populations consist of 60–80, and 15–20, wild elephants respectively (DNPWC 2008; Pradhan et al. 2011).

Habitat conservation is an important aspect of wildlife conservation, and habitat suitability analysis is an essential aspect of management of wild animals such as elephants. Habitat suitability modeling can predict the quality and suitability of habitats for given species based on predictor variables such as topography (aspect, slope, altitude), climate (temperature, precipitation) and other biotic and abiotic factors. Different methods of modeling are used to determine suitable habitats for elephants. The boosted regression trees (BRT) method is an ensemble tree-based species distribution modelling technique that iteratively grows small/ simple trees based on the residuals from all previous trees (Elith et al. 2008). BRT has proven useful for working with large datasets of environmental variables and observations (Elith et al. 2008). For example it has been used to identify determinants of above ground biomass (Adhikari et al. 2017) and fish species distribution (Elith et al. 2008; Trigal & Degerman 2015). BRT and geographic information have also proven to be effective in the assessment of habitat quality.

The present study aims: 1) to assess the suitable habitat of elephants, and 2) to determine which explanatory variables better explain elephant presence in PNP and buffer zone. This study has assessed habitat suitability in order to provide insights towards better management of elephant populations.

METHODS AND MATERIALS

Study area

The study was conducted in Parsa National Park (PNP) and its buffer zone (BZ), located in the sub-tropical zone of the southern part of Nepal. It has an area of 627km^2. In 1984, PNP was established to preserve the habitat of natural populations of Asian Elephant *Elephas maximus*, Tiger *Panthera tigris*, and Gaur *Bos gaurus* (Rimal et al. 2018). The BZ of PNP was declared in 2005, which covers an area of 285.17km^2 encompassing three districts and 11 village development committees (VDC) (Figure 1). The region experiences four different seasons: summer (April–June), rainy/monsoon (July–September), winter (October–December), and spring (January–March).

The forests of PNP consist of tropical and subtropical tree species. Sal *Shorea robusta* forests compose about 90% of the park's vegetation. The riverine forests are found along the banks of rivers entailing species like Sisso *Dalbergia sisoo*, Silk Cotton Tree *Bombax ceiba*, and Khair *Acacia catechu*. Grass including Siru *Imperata cylindrica* and Kans *Saccharum spontaneum* are in the park. PNP and BZ support various endangered animal species including wild Asian Elephant, Royal Bengal Tiger *Panthera tigris*, and Sloth Bear *Melursus ursinus*. Mammals including Blue Bull *Boselaphus tragocamelus*, Sambar *Rusa unicolor*, Hog Deer *Axis porcinus*, Barking Deer *Indian muntjac*, Rhesus Macaque *Macaca mulatta*, and Palm Civet *Paradoxurus hermaphrodites* are also found in the park. Anthropogenic pressures like sand extraction, shifting cultivation and domestic cattle grazing are high in PNP and BZ (CHEC Nepal 2012).

Quantitative data collection

Field work was conducted during the morning hours of May–June 2017. In reconnaissance, the habitats preferred by elephants were identified in consultation with local people. Questions concerned areas where elephants were frequently sighted, places where indirect signs of elephants were found, and availability of water. Reconnaissance field visits were made with the help of elephant rides, on foot and by vehicle, and areas were allocated into blocks according to habitat types. Sample plot centers were positioned using hand-held Garmin

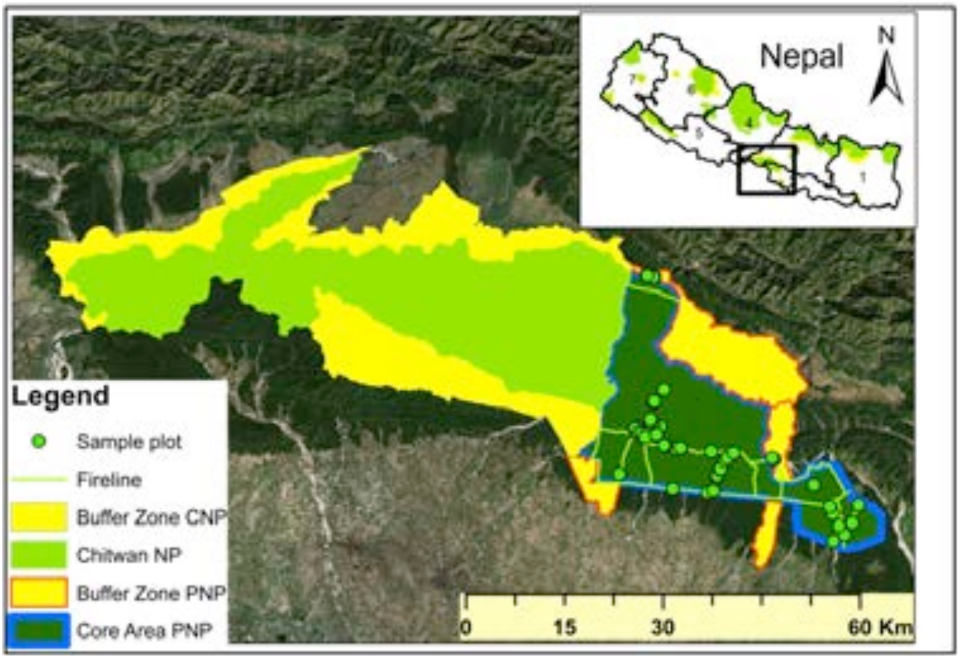

Figure 1. Location of the study area and sample plot distribution in Parsa National Park and buffer zone, Nepal.

global positioning system (GPS) with a 2–5 m accuracy.

Nested quadrats of different size were purposively assigned in the study area (Figure 2). Total 67 plots were assigned and used to assess the status of tree, pole and regeneration condition. Quadrats of 10m × 10m were set in the study area to calculate the intensity for tree species. All plant species within each quadrat were identified and counted. For trees, trunk diameter at breast height (DBH; 1.3m) and height was measured. Quadrats of 5m × 5m were allocated randomly for shrubs. Herbs and regeneration were recorded from nesting sampling of 1m × 1m quadrate within the 5m × 5m quadrate.

Tree diameter, height, dominant species, crown cover and ground cover were measured, poles and regeneration were counted and in cases of grasses, clumps were counted within each quadrat. Plant species were identified by a local para-taxonomist, field guide and also based on literature related to plant identification in Nepal (Rimal et al. 2018). Leaves of unidentified tree species were brought to the faculty of forestry at Agriculture and Forestry University (AFU) and were identified.

To assess the habitat, important value index (IVI) and prominence value (PV) of vegetation available in the habitat range is crucial. The vegetation data collected in the field were used to calculate IVI, density, relative density, frequency, and relative frequency of the tree species by using equations 1–8 explained in Greig-Smith (1983). The IVI of a species signifies its dominance and

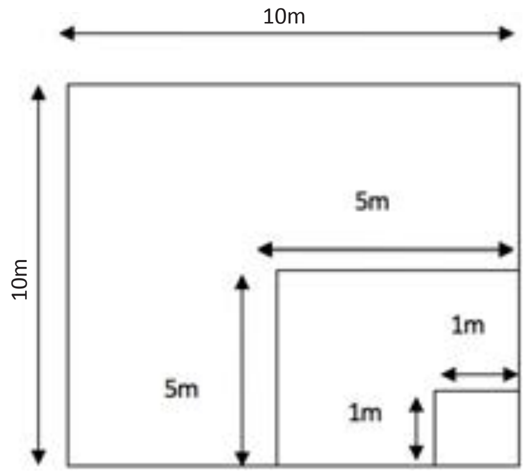

Figure 2. The layout of the quadrate used to assess the status of tree, pole and regeneration condition.

ecological success, its good power of regeneration and greater amplitude. The IVI was calculated by using three measures including relative frequency, relative density, and relative dominance. Vegetation data were calculated following the broad principle described by Mishra (1968) and Mueller-Dombois & Elienberg (1974). Basal area helps to determine the dominance and nature of the community and it refers to the actual ground covered by the stems. Density is generally used for large plants that have discrete individuals (Zobel et al. 1987). Frequency and relative frequency give an index on the spatial distribution of a species (Krebs 1978). To calculate the prominence value, the percentage cover of each species is estimated in each quadrate and recorded in classes as follows, for high coverage = > 50%, medium= 26–50% and low = 0–25%. Prominence value is used to calculate the availability of plants in the research sites (Jnawali 1995).

1. Density of species = (Total number of individuals of a species) / (Total number of quadrats sampled × area of a quadrate)

2. Relative density of species (RD) = (Total individuals of species) / (Total individual of all species)

3. Frequency of species = Number of plots in which a particular species occurs / Total number of plots sampled × 100

4. Relative frequency of species (RF) = Frequency value of a species / Total frequency of all species × 100

5. Relative dominance of species = Total basal area of a species / Total basal area of all species × 100

6. Basal area = $\pi d^2/4$

7. Important value index (IVI) = Relative density + Relative frequency + Relative dominance

8. PVX = MX (√FX)

(where, d = diameter at breast height (1.3 m) of tree, PVX = prominence value of species X; MX = mean percentage cover of species X; FX = Frequency of occurrence of species X)

Explanatory variables for modeling of habitat suitability distribution

A range of explanatory variables was derived from geospatial data sets for modeling habitat suitability. Table 1 presents the complete list of variables. The slope, aspects, and altitude were derived from the Japan Aerospace Exploration Agency (JAXA) digital elevation model (dem). Precipitation and temperature were downloaded from WorldClim data. Field measurements, dominant species, habitat preference, segment type, crown and ground cover and substrate conditions were derived. All topographic, climatic, and land use data available for the study area were resampled to 30m resolution and UTM 45N, WGS 84 projection system. For each absence and presence of GPS location, these variables were extracted. The correlation co-efficient between the explanatory variables and presence-absence data of elephant is shown in Appendix 1.

Statistical analysis

Boosted regression tree (BRT) (Elith et al. 2008) was used for examining the habitat preference area for elephant. BRT handles different types of predictor variables and accommodates missing data (Elith et al. 2008). Besides these, there is no need for prior data transformation or elimination of outliers. This is an advanced form of regression methods, which consists of two component—regression trees and boosting. BRT analysis was done using the 'dismo' package in R. The Bernoulli error distribution was used. Furthermore, the minimum predictive error was achieved when using a learning rate of 0.001, tree complexity (interaction depth) of 5, bag fraction of 0.75 and tolerance method "fixed". All predictor variables were used as BRT can handle multi-collinearity among variables.

RESULTS

Habitat assessment

IVI was calculated to find out the dominant tree species (Appendix 2) and prominence value was observed in the case of shrubs and herbs (Appendix 3 & 4). We calculated the species diversity of the study area for trees. Fifty-seven species were present in the quadrates; among them, 10 tree species were the dominant tree species present in the study area. In the study area, Sal (IVI–50.7753) was found most dominant and *Careya arborea* (IVI–5.2802) as least dominant. The species including *Mallotus philipinensis, Dillenia pentagaina, and Careya arborea* have the highest IVI among all, and they are the most preferred species of the elephant.

To determine the preferred habitat used by elephant, we calculated the PV of shrubs and herbs. Among 40 species, each of shrubs and herbs was present in the study area. Among the shrub species, *Eupatorium spp.* (PV–306.25) was the most abundant species and *Bauhinia vahilli* (PV–53.07) was the least abundant. Among the herb species, *Imperata cylindrica* (PV–317.66) was the most abundant and *Piper longum* (PV–29.48) the least abundant. The shrub species including *Eupatorium odoratum, Leea macrophylla, and Cleroden dronviscosum* and herb species including *Imperata cylindrica, Saccharum spontaneum,* and *Fritillaria camschatcensis* have the highest PV among all species found in the study area as well as, they are the most preferred species of the elephant in the study area.

Habitat suitability

The total deviance explained by the BRT model was 0.16. The correlation between different variables, including presence-absence, altitude, land cover of the plot, segment type, substrate condition, dominant species, ground cover, crown cover, habitat preference,

Table 1. Predictor variables used to model the habitat of Elephant.

Predictor variables	Format (Source)	Description
Temperature (× 10 °C) (1km × 1km)	Raster (WorldClim) 1*	The temperature of June was used
Precipitation (mm) (1km × 1km)	Raster (WorldClim) 1*	Precipitation of June was used
Slope (°) (30m × 30m)	Raster (Jaxa DEM) 2*	
Aspect (30m × 30m)	Raster (Jaxa DEM) 2*	
Altitude (30m × 30m)	Raster (Jaxa DEM) 2*	
Habitat preference	Field measurement	Species preferred by elephant, including *Mallotus philipinensis, Imperata cylindrica, Dillenia pentagina, Saccharum spontaneum, Careya arborea*, and *Pennisetum purpureum*
Dominant Species	Field measurement	Area dominated by species like *Acacia catechu* (AcC), *Bombax ceiba* (BoC), *Dillenia pentagaina (*DiP), *Albizzia procera (*AlP), *Lagerstroemia parviflora* (LaP), *Terminalia chebula* (TeC), *Trewia nudifolia* (TrN), and *Myrsine semicerata* (MyS)
Segment type	Field measurement	Divides the area into the segment by fire line, foot trail, pond, river, and railway
Crown and ground cover	Field measurement	Cover (%) of forest crown and ground
Substrate condition	Field measurement	The condition of the soil, including hard soil, soft soil, and leaf litter

1*—www.worldclim.org | 2*—www.global.jaxa.jp/press/2015/05/20150518_daichi.html.

precipitation, temperature, slope, and aspect is shown in Appendix 1. The relative influence of each predictor variables is shown in Figure 3. Each predictor variable has different relative contributions for the BRT model. Dominant species, temperature, and altitude have higher relative influence, whereas ground cover, crown cover and substrate condition have a lower relative influence.

Prioritized dependence plots visualize the effect of a single variable on the model response, holding all other variables constant. Model results vary the most with dominant species as seen in the first left plot (Figure 3). Dominant species (34%), temperature (15.3%), altitude (12.4%), habitat preference (11.1%) and precipitation (8.8 %) have the highest relative influence percentage and play a crucial role in the elephant distribution based on these plots. For more details on how they were calculated and model parameters used, see Sharma (2017).

On the basis of partial dependence plots, the elephant was more available at the altitude of 250–350 m with precipitation 310mm. The suitable habitat for the elephant was at the temperature of 28.5°C, a slope of 0–5° and in the northeastern and southeastern regions. Dominant species shows that *Acacia catechu* (AcC) and *Myrsine semicerata* (MyS) forest are more suitable for the elephant. Species including *Dillenia pentagaina* (DiP), *Saccharum spontaneum* (SaS), and *Pennnisetum purpureum* (PeP) are the most preferable species of the elephant. Elephants dwelled in forest dominated by *Mallotus philipinensis* followed by *Syzigiym cumini*. Thus areas having these species were the most suitable habitats.

The weighted mean of discrete data was not available, whereas the weighted mean of continuous data was altitude (264m), precipitation (310mm), temperature (28.6 °C), slope (5.6°) and aspect (190°). Elephants were found mostly around the fire line and river, at an altitude of 150–350 m with temperature around 28.6 °C, crown cover 40–70 and slope below 0–5° (Figure 4).

The correlation between elephant presence-absence and temperature is 0.24, that implies a slight positive relationship between them, the elephant is mostly found in increasing temperature (Appendix 1). Whereas there is almost no linear association between presence-absence, and slope, dominant species, land cover of the plot, crown cover and ground cover. The relationship of altitude with temperature is negative, i.e., 0.84, the temperature of the area increases with a decrease in altitude and vice versa.

The elephant distribution prediction map based on altitude, slope, aspect, precipitation, and temperature only using boosted regression tree model is presented in Appendix 5. Other predictor variables were based specifically on field data and their extrapolation to spatial scale was not possible.

DISCUSSION

The elephant population in Nepal is restricted to the Terai and Siwalik regions, where there have been large-scale conversion of forest and expansion of agricultural lands (Koirala et al. 2015). This has resulted

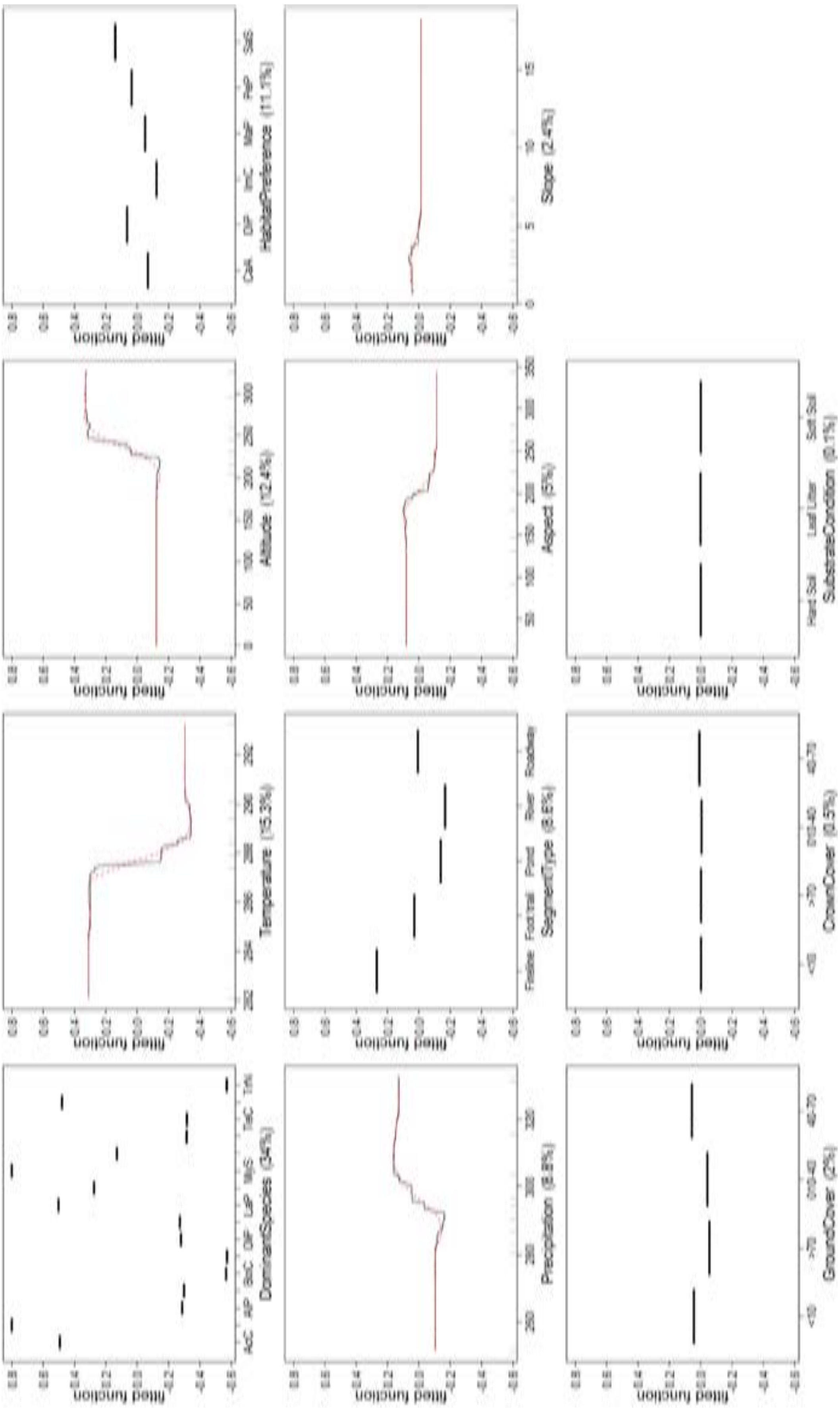

Figure 3. Single-variable partial dependence plots and smoothed response curves for the predictor variables, where Y-axis is the fitted function for the response variable (elephant). The relative influence is shown in parantheses.

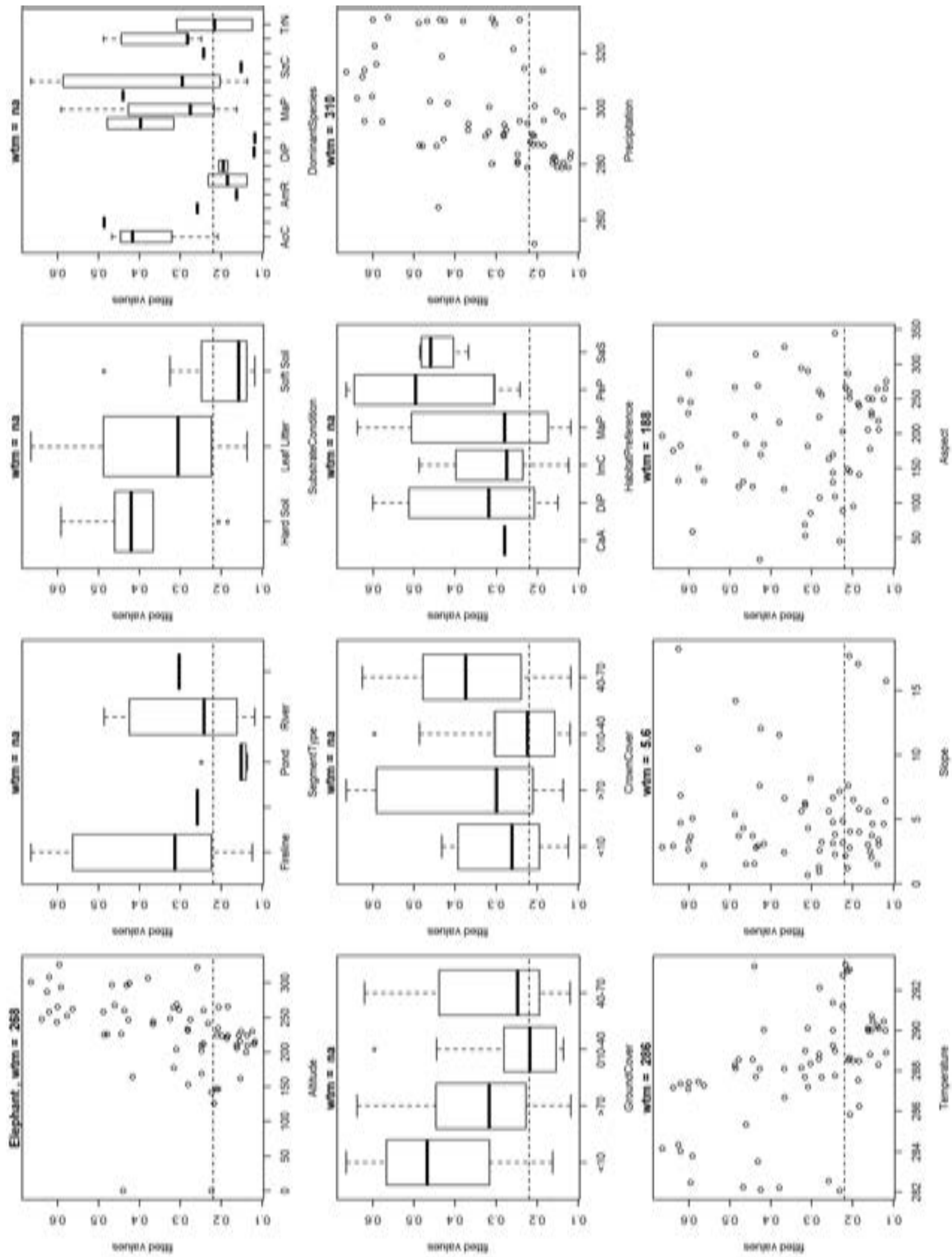

Figure 4. The weighted mean of all predictor variables.

in negative human-elephant interactions in many parts of Nepal. Movement of elephants outside the national park and wildlife reserve could have been the result of unsuitable habitat, reduced supply of food and water, and encroachment by human beings. Assessing habitat suitability of elephants assists in the preparation of sustainable management plans. PNP and BZ has been the habitat of elephant for long time, but habitat suitability studies are rare in this area. This research examines habitat suitability of the elephant in PNP and BZ based on different variables including dominant species, temperature, altitude, habitat preference, precipitation, segment type, aspect, slope, ground cover, crown cover and substrate condition.

Based on the BRT model, PNP and BZ are suitable habitats for elephants. We witnessed the outcome of parameters as per the physical, biological and climatic features of the area like slope, aspect, altitude, precipitation, temperature, habitat preference, crown cover and ground cover. The result shown by Koirala et al. (2016) posits that species like *Spatholobus parviflorus*, *Saccharum spontaneum*, *Shorea robusta*, *Mallotus phillippensis*, *Garuga pinnata*, *Litsea monopetala* contributed the highest proportion of diet for an elephant in the PNP. In consent with our result, similar species of trees, shrubs, and herbs have the highest IVI and PV and distributed in the lower part of the area. This result concludes that the study area is the most suitable for elephant to dwell. Our study revealed that the habitat is suitable in the Northeast and Southeast region of the study area, which is similar to the result of Shamsuddoha & Aziz (2015).

Rood et al. (2010) studies have found that the elephant's habitat use in a tropical forest is depicted by areas of high forest cover. Our analysis, however, found no marked relationship between ground cover, crown cover, and presence of elephants. Our findings revealed that in the study area, slope 0–5° and altitude 400m is suitable for elephants which are almost similar to the result of Areendran et al. (2011). In accordance with the studies of Douglas et al. (2006), Lin et al. (2008) and Ochieng (2015), suitable habitat for elephants was found to be limited by augmentation of both altitude and slope. There is no abundance of elephants' presence sign with the increment of the altitude and slope in PNP. In order to preserve their energy needs, Ntumii et al. (2005) mention that elephants avoid the height and steeply sloped area.

Variability in results might have occurred due to the differences in sampling methods, variance in forest condition, composition, and sampling area, etc. The research outcome was concluded based on only one season field work; however, taking all the results of four seasons might produce more effective result. Data of precipitation and temperature were extracted from Worldclim; the data taken from the nearest metrological station of Samara could be better with more accuracy. The outcomes from this study, linked to slope, and elevation are valid for PNP only, and cannot be generalized to the habitat of an elephant in other countries. Further research should focus on creating map of elephant distribution, habitat suitability, and threats to elephant from invasive species.

CONCLUSION

BRT was applied to assess elephant habitat suitability in PNP. In this study, we analyzed the distribution of elephant using a combination of biotic and abiotic environmental variables, including the topographic and climatic factors. The model emphasizes on environmental suitability and contributes to knowledge for conservation of elephant in PNP. It provides a basis for habitat analysis. Elephants were recorded up to 400m and in northeastern and southeastern aspects. Its presence could not be related to forest cover and substrate condition. The result from the modeling may become useful to plan and delineate areas for management of elephant. It presents scope to minimize HEC through precautionary measures.

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Author contribution: PS planned and conducted this research, HA and ST supervised this research. HA, PS and ST together worked on manuscript. HA and PS collected RS and GIS data. PS, RB and ST collected field data. AKR supervised PS during field data collection.

Appendix 1. Pearson correlation between different predictor variables and their scatter plots.

Appendix 2. Important Value Index (IVI) of dominance tree species

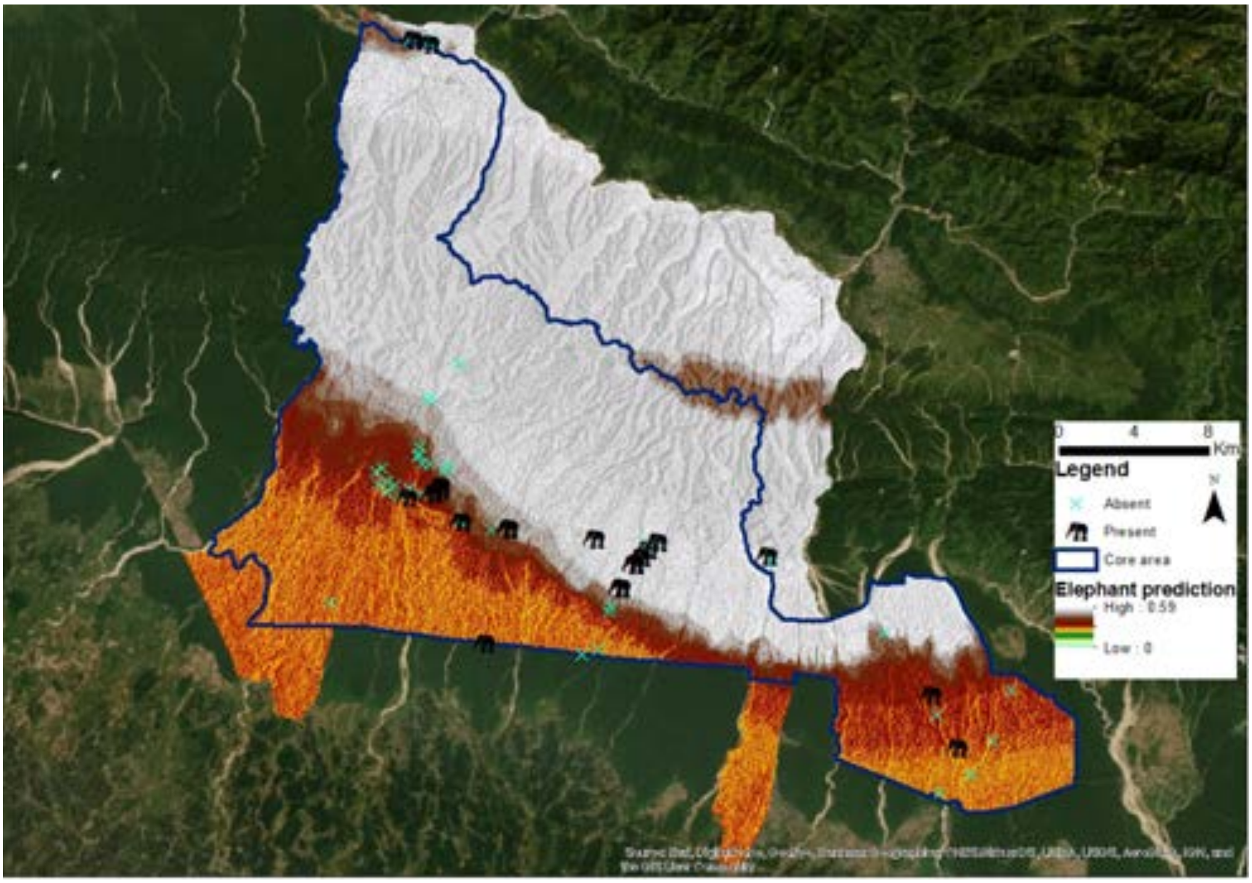
Scientific name	Local name	Relative frequency	Relative density	Relative dominance %	Important value index (IVI)
Shorea robusta	Sal	13.36	0.27	37.14	50.77
Mallotus philipinensis	Sindure	6.45	0.14	10.60	17.20
Terminalia tomentosa	Asna	4.60	0.04	8.22	12.87
Acacia catechu	Khair	4.60	0.11	5.57	10.29
Lagestromia parviflora	Bot dhayero	6.91	0.05	3.44	10.41
Dillenia pentagaina	Tatari	6.45	0.05	3.73	10.23
Adina cordifolia	Haldu	3.68	0.02	3.44	7.14
Garuga piñata	Dabdabe	5.06	0.03	1.97	7.08
Albigia procera	Setosiris	2.76	0.01	3.10	5.88
Careya arborea	Kumbe	2.76	0.01	2.50	5.28

Appendix 3. Prominence value of shrubs in the study area.

Scientific name	Local name	Number of individuals	Frequency	Mean cover of individual species (M_x)	Prominence value (PV_x)
Eupatorium odoratum	Setobanmara	188	50.72	43	306.25
Leea macrophylla	Galini	213	50.72	38	270.64
Clerodendron viscosum	Bhati	248	33.33	25	144.33
Murraya koenigii	Curry leaf	72	26.08	20	102.15
Fritillaria spp.	Thulobandhan	19	5.79	30	72.23
Lantana camera	Lantana	14	4.34	30	62.55
Asparagus racemosus	Kurilo	34	18.84	14	60.76
Agiratus conyzoides	Gande	111	5.79	25	60.19
Parthenium spp.	Parthenium	517	5.79	24	57.78
Bauhinia vahlli	Bhorla	20	8.69	18	53.07

Appendix 4. Prominence value of herbs in the study area.

Scientific name	Local name	Number of individuals	Frequency	Mean cover of individual species (M_x)	Prominence value (PV_x)
Imperata cylindrica	Siru	3516	42.02	49	**317.66**
Saccharum spontaneum	Kans	970	13.04	32	115.57
Fritillaria camschatcensis	Ban dhan	1035	18.84	20	86.81
Hemalthriya compressa	Ghodeydubo	699	10.14	25	79.62
Cynodon dactylon	Dubo	634	7.24	29	78.06
Digitarea spp.	Chitrebanso	382	10.14	23	73.25
Pennisetum purpureum	Elephant grass	163	4.34	22	45.87
Barlaria cristata	Kuro	151	7.24	16	43.07
Dendrobium spp.	Orchid	27	7.24	11	29.61
Piper longum	Pipla	18	8.69	10	**29.48**



Appendix 5. Elephant distribution prediction based on altitude, slope, aspect, precipitation, and temperature only using boosted regression tree model. Other predictor variables were based on field data and not available at the wall-to-wall spatial scale.

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COMMUNICATION

Current population status of the endangered Hog Deer *Axis porcinus* (Mammalia: Cetartiodactyla: Cervidae) in the Terai grasslands: a study following political unrest in Manas National Park, India

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Abstract: The Endangered Hog Deer *Axis porcinus* has experienced drastic population declines throughout its geographical range. There is limited knowledge of its current population status, particularly from northeastern India. In this study the population density of Hog Deer was assessed in Manas National Park, which was a deer stronghold prior to the armed conflict that lasted for almost two decades, resulting in depressed deer populations. With the cessation of conflict, efforts were invested by both government and conservation organisations for the recovery and conservation of charismatic fauna in the park. Studies on Hog Deer populations, however, were lacking and thus reliable information on current status is unavailable. Current population status and threats faced by Hog Deer were assessed to aid informed conservation decisions. Distance sampling techniques (line transects) were applied in the grassland habitat during the dry season of two consecutive years. The estimated Hog Deer density was 18.22±3.32 km^{-2}. The potential threats to Hog Deer identified in Manas include habitat loss, habitat degradation due to spread of invasive plant species, illegal hunting, and other anthropogenic disturbances. Our study suggests that the Hog Deer population, though reviving, needs immediate conservation attention.

Keywords: Armed conflict, geographical range, invasive species, population density.

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Author contribution: AS, BPL and SAH developed the concept and designed the framework. SAH provided valuable inputs in statistical analysis and manuscript writing. AS and BPL acquired the resources. AS collected data, performed statistical analysis and interpretation, manuscript writing and revisions. BPL and SAH supervised the project and contributed to the manuscript.

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INTRODUCTION

The Hog Deer *Axis porcinus*, historically distributed across South and Southeast Asia, underwent a drastic range-wide decline during the mid and late 20th Century (Brook et al. 2015; Timmins et al. 2015). Subsequently, *A. porcinus* was categorised as an Endangered species by the IUCN in 2008 (Timmins et al. 2015). Despite being an Endangered species, it is one of the least studied mammals and its range-wide decline was mostly overlooked (Brook et al. 2015). The southeastern Asian population is locally extinct in most countries, including China, Lao PDR and Vietnam (Ohtaishi & Gao 1990); the only wild populations remain in Cambodia and Myanmar (Brook et al. 2015; Lwin et al. 2016). In southern Asia, with a declining population trend, the Hog Deer is mostly confined to protected areas (Karanth & Nichols 2000; Biswas 2004; Odden et al. 2005). Timmins et al. (2015) recommended that estimating population abundance was important in assessing the conservation status of *A. porcinus*. Though deer population estimates are available for few well-managed protected areas, mostly in Nepal (Odden et al. 2005; Bhattarai & Kindlmann 2012; Lovari et al. 2015), data from other areas of southern Asia are lacking.

The Hog Deer (Image 1) is a grassland obligate (Dhungel & O'Gara 1991; Odden et al. 2005), primarily threatened by habitat degradation or loss and illegal hunting. It is an important prey for large carnivores (Stoen & Wegge 1996; Lovari et al. 2015), and thus plays a vital ecological role. India is one of the strongholds of *A. porcinus* populations in southern Asia, although historically it has received little attention and available information is mostly anecdotal (Biswas 2004). To implement rational conservation measures, reliable estimates of population abundance are fundamental,

Image 1. Hog Deer *Axis porcinus*,

and their lack can undermine the entire process (Lopez-Bao et al. 2018). Thus the current population status of *A. porcinus* was assessed and potential threats to the population in Manas National Park in Assam were documented.

Manas National Park (henceforth, Manas) in northeastern India harboured a population of approximately 10,000 Hog Deer until the 1980s (Tikader 1983), and armed conflict in the region from the mid-1980s to 2003 lowered the population density (Goswami & Ganesh 2014). The instability resulted in habitat degradation, destruction of park infrastructure and poaching/hunting in the absence of normal law and order. With the restoration of peace, conservation efforts were implemented to safeguard remaining wildlife populations (UNESCO 2005). The cessation of civil unrest facilitated access to Manas by various conservation organisations that work with management authorities to conserve wildlife and promote species recovery. Most of the management and conservation inputs have focussed on securing and conserving charismatic megafauna like One-horned Rhinoceros *Rhinoceros unicornis* and Bengal Tiger *Panthera tigris*, which are apex species in the ecosystem and iconic species for conservation. In comparison, lesser-known mammals like the Hog Deer have received little attention. With about 40% grassland habitat (Das 2018), Manas represents one of the last remnant patches in western Assam that can support grassland obligates such as One-horned Rhinoceros, Hog Deer, Hispid Hare *Caprolagus hispidus*, Pygmy Hog *Porcula salvania*, Bengal Florican *Houbaropsis bengalensis*, Swamp Deer *Rucervus duvaucelii*, Asiatic Water Buffalo *Bubulas arnee*, and others (Lahkar 2008). These grasslands are under threat from invasion by alien plant species, mostly by *Chromolaena odorata* and *Mikania micrantha* (Lahkar et al. 2011; Nath et al. 2019), agricultural encroachment, and cattle grazing (Sarma et al. 2008), which may have had an impact on Hog Deer population abundance.

It is evident that Hog Deer and their habitat in Manas deserve immediate conservation attention. Goswami & Ganesh (2014) attempted to estimate the population density of herbivores immediately after the cessation of the conflict, but their study had limited observations. The authors conducted line transect sampling on foot, which may have an influence on the detection probability (Wegge & Storaas 2009). This is the first intensive study from Assam that focussed on estimating the population density of Hog Deer. This provides an important insight regarding the current status of this threatened species and the need for management intervention for its long-term conservation.

MATERIALS AND METHODS

Study area

The study was conducted in Manas National Park (26.722^{0}N & 91.043^{0}E), which forms the core of the Manas Tiger Reserve in the northeastern Indian state of Assam (Figure 1). It lies along the foothills of the Himalaya,

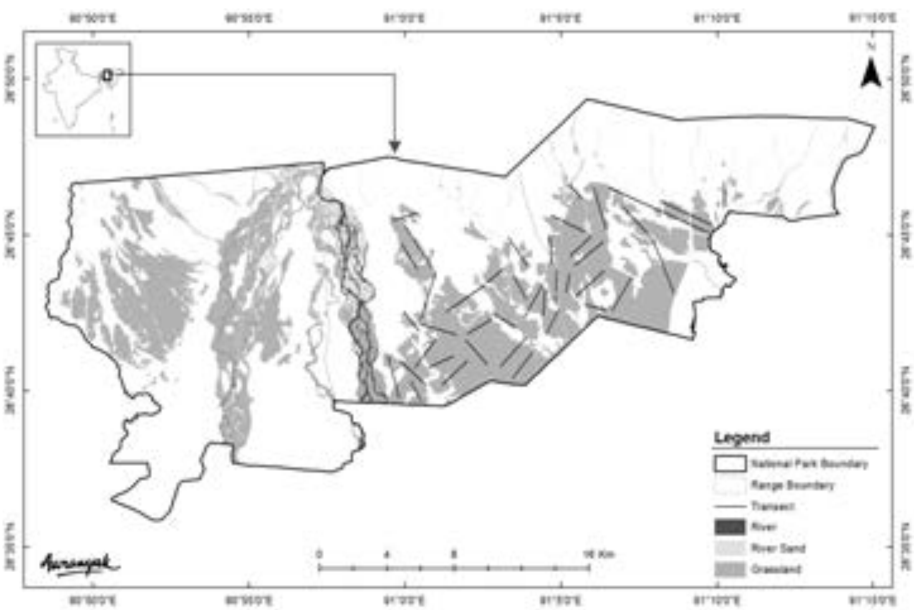

Figure 1. Manas National Park and lay-out of line transects in the grassland habitat in Manas. (Map Source: Aaranyak).

and is contiguous with the Royal Manas National Park of Bhutan to the north, bounded by villages to the south, by Daodhara and Batabari reserve forests to the east, and reserve forests to the west. The park comprises an area of 519km^2 (Sarma et al. 2008) and has a predominantly flat terrain. Broadly, the vegetation of Manas is classified as sub-Himalayan alluvial semi-evergreen forest, east Himalayan mixed moist and dry deciduous forests, the commonest type, and grasslands (Champion & Seth 1968). The grasslands are further classified into dry savannah grasslands and wet alluvial grasslands. These grasslands occur in seven major grass assemblages which harbour many threatened grassland obligates (Lahkar 2008). Manas harbours a rich faunal assemblage, with 60 species of mammals, around 470 avian species, and 42 species of herpetofauna. The climate of Manas is warm and humid, with rains from mid-March to October; most rain falls during the monsoon months from mid-May to September and November to February is relatively dry (Borthakur 1986).

In Manas, political stability was attained with the formation of Bodoland Territorial Autonomous Districts (BTAD) in 2003, and subsequently the conservation intervention gained momentum, such as with a Rhino restocking programme (Barman et al. 2014). Nevertheless, instances of occasional conflicts were prevalent in the western Range (Panbari) until 2016 (Lahkar et al. 2018). Therefore, the study was restricted in the central (Bansbari) and eastern (Bhuyanpara) administrative ranges of the park which had one such incident in 2014.

Field Survey

The population density of Hog Deer was derived through distance sampling (Buckland et al. 2004), which is established as a standard method and has been adopted widely to generate herbivore densities across various habitats in the tropical and temperate ecosystems in Asia (Varman & Sukumar 1995; Khan et al. 1996; Biswas & Sankar 2002; Jathanna et al. 2003; Wegge & Storass 2009; Wang 2010; Bhattarai & Kindlmann 2012; Goswami & Ganesh 2014; Lovari et al. 2015). The entire study area was overlaid with 2 x 2 km grid and stratified random sampling was adopted. Line transect surveys from elephant back (Wegge & Storaas 2009) were conducted in the grids with grassland cover as the species is a grassland obligate (Dhungel & O'Gara 1991; Odden et al. 2005) during the dry season of 2014–15 (henceforth, 2015) and 2015–16 (henceforth, 2016). A total of 75 transects were sampled, covering a total distance of 206.56km. Spatial replicates were used, as Hog Deer sightings were relatively low in Manas (Krishna et al. 2008) and transect lengths varied from 2 to 5 km. During the elephant transects, the Mahout (elephant driver) and one observer detected and counted the animals. For each detection the radial distance of the animal to the observer and sighting angle were measured using a range finder and a compass respectively.

Data Analysis

Initially, the encounter rate of Hog Deer per transect per year was compared to investigate whether there is any significant difference between them using a Z-test. As there was no significant difference between both years (z=0.05, P > 0.05, n1= 35, n2 =40), the data from two consecutive years were pooled to estimate the Hog Deer population density in the park using programme DISTANCE 7.1. Conventional distance sampling (CDS) approach in DISTANCE programme was used to derive Hog Deer density estimates (Buckland et al. 2001). Exploratory analyses were carried out to check for evasive movement before detection, heaping effect, and truncation of observation outliers (Buckland et al. 2001). The data were grouped into unequal distance bins, and chi-square goodness-of-fit values (the lowest) were considered to select the interval combination (Buckland et al. 2004; Zamboni et al. 2015). The data beyond the distance of 45m were truncated as they were outliers for better model fitting. The probability of detection was estimated using six models recommended by Buckland et al. (2001) combining probability density function (uniform, half normal and hazard-rate) with adjustments (cosines, simple and hermite polynomials). The models were selected based on the criterion of lowest AIC as generated by the program. The estimates were generated with standard error, the coefficient of variation and confidence intervals. Hog Deer density (D) was estimated, and approximate population size (N) was computed based on the size of the habitat area.

To derive the population structure and age-sex-ratio of Hog Deer, intensive surveys were conducted in the entire park and computed based on percentage sightings. Data were recorded both during the line transect sampling and opportunistic sightings over a period of two years on group size and composition. For each detection, the animals were classified into the following age-sex categories; fawn (1–12 months), yearlings (13–24 months) and adults (>24 months) based on Dhungel & O'Gara (1991) classification. Based on the sightings, adult male to adult female and doe to fawn ratio was calculated. The data from both the years were pooled as there was no significant difference between the adult male (z=0.49, P>0.05, n1=56, n2=68) and adult female (z=1.65, P>0.05, n1=56, n2=68) categories between the years. Furthermore, a significant difference between the percentage of adult male and female in a group was tested using z statistic. The percentage data was transformed using arcsine transformation and analysed using MS Excel.

RESULTS

A total of 202 sightings of Hog Deer were made along the 206.56km of transects during the two years sampling period. Of these, 56.20% of the sightings were from the central range and 43.80% from the eastern range. The overall density of Hog Deer in Manas was estimated to be 18.22 ± 3.32 km^{-2} (CV = 18.27%, 95% CI = 12.72–25.09). Based on comparisons of the lowest AIC values, the uniform key function with cosine adjustment best described the Hog Deer data (Figure 2). The result, with estimated density, percent coefficient of variation, 95 % confidence interval and AIC is summarised in Table 1. On extrapolating the population density of 18.22km^{-2} to the available grassland habitat in the park (194.57km^2, Das 2018), the population size of Hog Deer was estimated to be 3,545 ± 647.64 (CV = 18.27%, 95% CI = 2,475–5,077).

To understand the age structure of Hog Deer population, the percentage of different group types was calculated based on the number of animals detected during the line transect and other opportunistic sightings for both the years. In a few instances (4.59%), though, the sex of the animal could not be identified. The groups were classified as solitary-consisting of single animal, small (2–3 animals), medium (4–6 animals) and large (>6 animals) groups (modified from Biswas 2004). Most of the animals occurred solitary (50.79%), 36.50 % occurred in small groups, 10.31% in medium groups, and only 2.38% in large groups. The mean group size of Hog Deer is estimated to be 1.81 ± 0.11. The observed overall sex ratio in Manas, of adult male to adult female to fawn is 47.01:100:17.88. There is a significant difference between the adult male and female percentage in a group (z= 4.72, P<0.01, n1=n2=125).

DISCUSSION

Our study suggests that the current estimated Hog deer density in Manas differs substantially from that of the previous study which reported a density estimate of 4.59km^{-2} (Goswami & Ganesh 2014). One of the possible reasons is the difference in the line transect sampling method that the two studies have adopted. Sampling in grassland habitats on foot may influence the detection probability and underestimate the population abundances of species like Hog Deer (Wegge & Storaas

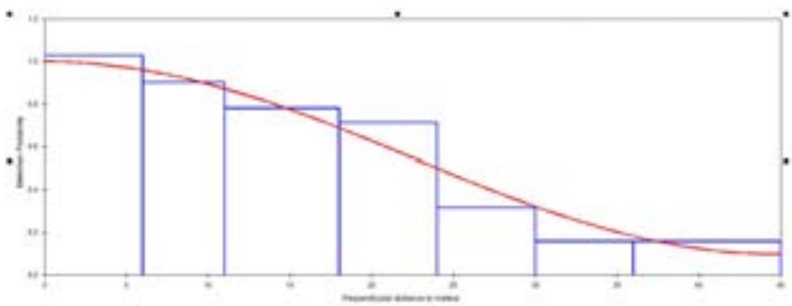

Figure 2. Distance data for Hog Deer *Axis porcinus* truncated at 45m, and fitted with the uniform cosine model.

Table 1. Summary of overall Hog Deer density estimate in six models as recommended by Buckland et al. (2001).

Parameter	Uniform + Cosine	Uniform + Simple polynomial	Half normal + Cosine	Half normal + Hermite polynomial	Hazard rate + Cosine	Hazard rate + Simple polynomial
Density km^{-2}	18.22	18.12	18.86	18.86	17.57	17.57
Percent Coefficient of variation	18.27	19.07	19.26	19.26	20.14	20.14
Upper CI	25.09	26.25	27.51	27.51	26.06	26.06
Lower CI	12.72	12.50	12.93	12.93	11.84	11.84
AIC	375	376.79	375.07	375.07	376.54	376.54

Table 2. Estimates of Hog Deer density across southern Asia.

Location	Habitat type	Density of Hog Deer (km^{-2})
Chitwan National Park, Nepal	Savanna grassland	15.5–19.1
Bardia National Park, Nepal	Floodplain grassland	77.3
Kaziranga National Park, India	Floodplain grassland	38.6
Sukhlaphanta Wildlife Reserve, Nepal	Grassland	4.1 (2010) and 11.6 (2011)
Keibul Lamjao National Park, India	Grassland/phumdis	2.51
[1]Manas National Park, India	Grassland	4.59
[2]Manas National Park, India	Grassland	18.22

Sources: Chitwan (Dhungel & O'Gara 1991), Bardia (Odden et al. 2005); Kaziranga (Karanth & Nichols 2000); Sukhlaphanta (Lovari et al. 2015); Keibul (Angom 2012); Manas 1 (Goswami & Ganesh 2014); Manas 2 (present study).

2009). Therefore, we conducted line transect surveys from elephant back following Wegge & Storaas (2009), which may have led to higher Hog Deer density estimate than the previous study. During the All India Tiger Monitoring exercise, the attempt to estimate prey density in Manas with an effort of 134km was hindered due to low number of observations (Jhala et al. 2015). The line transect sampling was conducted on foot, which might have resulted in lesser sighting records due to tall and dense vegetation. Therefore, sampling from elephant back in the grassland habitat in Manas is recommended for all the future population estimates of Hog Deer. In this study the estimated population size of Hog Deer is 3,545, considerably different from the previous estimate of 1,626. The sampling protocol to used derive this estimate was not clear. Nonetheless, our study finding indicates a possible recovery of the Hog Deer population over the years with the cessation of the conflict which can be attributed to enhanced protection and anti-poaching measures.

The Hog Deer population in Manas is female-biased. The sex ratio favouring the females is a characteristic of polygamous species (Dhungel & O'Gara 1991). Seidensticker (1976) reported a sex ratio of 51 males: 100 females: 24 fawns, whereas Mishra (1982) observed a ratio of 59 males: 100 females: 55 fawns. A similar sex ratio was also observed by Dhungel & O'Gara (1991) (56 males: 100 females). The mean group size of Hog Deer in Manas is similar to that of Chitwan (1.8, Dhungel & O'Gara 1991), but lower than reported in Jaldapara Wildlife Sanctuary (2.68, Biswas 1999). Hog Deer is primarily a solitary cervid (Odden & Wegge 2007), but congregates in small groups while feeding. During our study period we mostly documented Hog Deer singly or in small groups. Large groups comprising of more than six individuals were observed less frequently (12.69%). Biswas (1999) reported that 41% of animals were solitary, 56% occurred in small to large groups and only 3% occurred in very large groups (>10 animals) in Jaldapara. The largest congregation observed was of 33 animals, feeding on the fallen flowers of *Gmelina arborea* during the dry season in Manas.

Prior to the armed conflict, Manas harboured an abundant Hog Deer population of approximately 10,000 animals (Tikader 1983). The absence of empirical data on Hog Deer populations before and after the conflict limits our efforts to quantify the population change; interactions with experts who have worked in the area during the 1980s suggest that the population has declined sharply, more than 70% (Goutam Narayan pers. comm. December 2017). The local extinction of One-horned Rhinoceros (Talukdar 2003), depressed population of Swamp Deer (Das et al. 2009; Borah et al. 2013), Pygmy Hog (Bibhuti P. Lahkar pers. obs. 19.xii.2017) and Bengal Florican (Namita Brahma pers. comm. 19.xii.2017) due to the armed conflict (Lahkar et al. 2018), reflects that the grassland species declined drastically because of selective hunting by both opportunistic hunters and the anti-government forces (Goswami & Ganesh 2014). The possible drivers of Hog Deer decline are habitat degradation & reduction and illegal hunting. The grasslands which Hog Deer prefer have reduced in area over the last four decades (Sarma et al. 2008; Das 2018). The grassland patches such as 'Pahufield' area, 'Rhino camp' area and the grasslands particularly near the southern boundary of the park, mostly in the central range, which were prime Hog Deer habitats (Bibhuti P. Lahkar pers. obs. February 2002) are heavily infested with invasive plants such as *Chromolaena odorata* and *Mikania micrantha* (Nath et al. 2019). There is also livestock grazing pressure in the grassland (approximately 2000 cattle per day graze inside the park during the dry season, (Alolika Sinha pers. obs. 20.iii.2017) and can lead to severe competition for forage.

Trapping of Hog Deer for consumption using snares in the fringe village is not uncommon (Alolika Sinha pers. obs. 25.iii.2017). During the study period, four incidents of Hog Deer hunting were recorded in the fringe villages. This may underestimate hunting incidents, since many go unreported. We also found snares along the southern boundary of the park, which were possibly set-up to trap Hog Deer, other small mammals (e.g., hares), and birds. Another emerging threat to the species in Manas is attack by feral dogs. During the dry season, when the Hog Deer congregate to feed on *Gmelina arborea* flowers and fruits in the central range near an area called 'second gate', they are attacked by the feral dogs. We recorded six incidents over a period of two months (February-March 2016) where the feral dogs attacked and killed deer, although the dogs were not seen eating them. A multitude of factors like habitat degradation, occurrence of invasive plant species, and anthropogenic disturbances might affect the Hog Deer population in Manas. The influence of these various factors on Hog Deer population can be drawn more conclusively, upon long-term monitoring of its population and grassland habitat.

The present Hog Deer estimate, when compared with those from other areas in southern Asia revealed that Manas is an area of intermediate deer density (Table 2). Nevertheless, with the restoration of governance and administration, the management intervention improved substantially. A major step was the conversion of the former poachers/hunters into conservation volunteers and engaging them in regular patrolling of the park along with the forest personnel. Hog Deer are known to occur in high density in other well-protected areas (Table 2) (Karanth & Nichols 2000; Odden et al. 2005). The grasslands in Manas are one of the last remnant habitats in the eastern Terai (Lahkar 2008) and crucial for Hog Deer survival in the region (Biswas 2004). The scope of Hog Deer persistence beyond the National Park is limited due to scarcity of potential habitats and high anthropogenic pressure on these habitats.

Our study highlights the current population status of this threatened species, and we have documented potential threats to Hog Deer in Manas. This baseline population estimate will be useful to monitor future changes and conservation of Hog Deer in one of the high-value conservation landscapes. Manas is the most promising potential habitat for long-term survival of Hog Deer in western Assam, given that it is the best protected grassland habitat in the region. To this end, we suggest regular monitoring of Hog Deer populations and habitat improvement to document population recovery with the minimisation of the extant threats, and the formulation of future management strategies.

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A food spectrum analysis of three bufonid species (Anura: Bufonidae) from Uttarakhand region of the western Himalaya, India

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Abstract: The ecological diversity of insects and its predators like amphibians are important determinants in ecological balance. A total of 1,222 prey items in 84 specimens were examined to contribute the understanding of the diets of three *Duttaphrynus* species, viz., *himalayanus*, *melanostictus*, and *stomaticus* from Uttarakhand, the western Himalaya, India. Gut content analysis of three bufonids revealed acceptance of a wide range of terrestrial insects and other invertebrates as their food. The index of relative importance indicated that the most important preys were Formicidae, Coleoptera and Orthoptera. *Duttaphrynus melanostictus* had the broadest dietary niche breadth, followed by *D. himalaynus* and *D. stomaticus*. The wide prey spectrum well indicates that these species are the generalist and opportunist invertebrate feeder. Information pertaining to the food spectrum analysis contributes to understanding the ecological roles and used as a baseline data for future successful amphibian conservation and management programs in the Himalayan ecosystem.

Keywords: Bufonid, importance of relative index, Levin's measure, stomach flushing, western Himalaya.

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Author contribution: VNB & AKC performed the survey, data collection and finalized the manuscript. SS, GB, SB helped in survey and data collection. NL helped in data analysis at the time of manuscript revision. SNB supervised the overall study design and securing the fund.

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UTTARANCHAL
UNIVERSITY

INTRODUCTION

The family Bufonidae (Gray 1830) is one of the most species-rich families of anurans belonging to the class Amphibia. It is a large and geographically widespread taxon of neobatrachian frogs (Reig 1958; Lynch 1973; Duellman & Trueb 1986). It comprises more than 550 species in ca. 50 recognized genera geographically ubiquitous, only two of the remaining 32 genera have more than 10 species and all have relatively restricted geographic ranges (Frost 1985, 2011). Bufonidae comprises the true toad: they are best known for their thick, warty skin appearances and have prominent skin glands especially a pair of parotoid glands on the back of their heads. In the context of Uttarakhand, western Himalayan anuran fauna comprises three species of the family Bufonidae, namely, *Duttaphyrnus himalayanus* (Günther, 1864), *D. melanostictus* (Schneider, 1799), and *D. stomaticus* (Lutken, 1864).

Food is an important item for any living organism. The body requires the range of nutrition in organism's diet to keep all organs alive and in the correct balance. Diet is a also crucial part of the natural history of an animal, because not only does it reveal the source of the animal's energy for growth, reproduction and survival (Zug et al. 2001; Norval et al. 2014), but it also indicates part of the ecological roles such as food webs, resource portioning and ecological energetic. Anurans are thought to be opportunistic predators with their diets just reflecting the availability of food of appropriate size. Different studies suggest that food is a vital factor that explains the structure of anuran communities in different parts of the world (Duellman 1967; Inger & Colwell 1977; Duellman & Toft 1979; Toft 1980; Clcek & Mermer 2007). The stomach contents of many Bufonidae species have been examined in the past to determine their role in an ecosystem (Yu & Guo 2012; Sulieman et al. 2016).

Although the Uttarakhand region of the western Himalayan ecosystem embraces all types of amphibians on account of its varied climate, topographical, altitudinal and vegetational conditions, information about diets of amphibians is very scarce and the biology of most amphibians is poorly known from this region (Ray 1995; Bahuguna & Bhutia 2010). Therefore, the present work on a food spectrum analysis of three toad species fills the lacuna that would be helpful in understanding their feeding habitat and ecological role in Uttarakhand, the western Himalaya. Our analysis was aimed at (1) identifying and determining small invertebrate prey, (2) examining importance of the relative index of three toad species, (3) comparing the food spectrum and niche breadth among three toad species from its natural range.

MATERIALS AND METHODS

For the present study fieldwork was carried out in several localities, viz., Dayara (S1) (2,800m), Triyuginarayan (S2) (2,300m), Badhani tal (S3) (2,089m), Joshimath (S4) (2,240m), and Sem Mukhem (S5) (2,200m) (Fig. 1). Samples were studied in breeding seasons, i.e.,

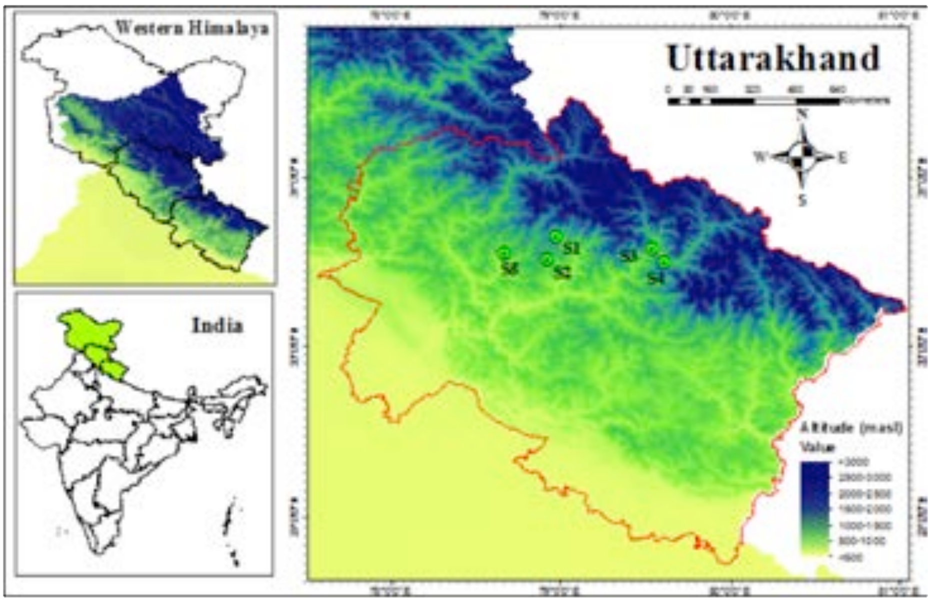

Figure 1. Location map of study area.

March–September from 2014–2017 at evening hours (18.00–23.00 h) in their natural habitats such as pools, ponds and in the vicinity of shaded mountain streams and so on. It was based on nocturnal visual encounter survey (Heyer et al. 2014).

Toads were collected manually in their habitats and stomach flushing was carried out immediately. Flushing was applied as soon as possible after capturing anurans, in order to precede digestion (Secor & Faulkner 2002; Sole et al. 2005). The subsequent immediate release of all specimens into their habitats ensured that the current activity of the treated specimens was not essentially disturbed by the stomach-flushing. The stomach contents were picked up with forceps and fixed in 70% ethanol in a vial. All contents were analyzed under a stereomicroscope (Olympus SZX 7). Identifications of food items were possible up to the order level with the exception of Hymenoptera, which was classified as Formicidae and non Formicidae and the rest of the items have been categorized as 'miscellaneous' (for broken materials) or unidentified (Gibb & Oseto 2006; Chowdhary et al. 2016). The food contents were then identified with the aid of keys provided by Ward & Whipple (1959). The food preferences of the three toad species were analyzed in terms of number, volume and frequency of occurrence. Prey's length and width were evaluated with a digital vernier caliper (Aerospace) to the nearest 0.1mm accuracy. Preserved items were measured and their volume (in mm^3) was calculated using the formula for ellipsoid bodies (Griffiths & Mylotte 1987).

$$V = \frac{4}{3}\pi\left(\frac{L}{2}\right)\left(\frac{W}{2}\right)$$

where, L=prey length, W=prey width

We obtained the frequency of occurrence of each prey categories in the diet dividing the number of stomachs which contained that category by the total number of stomach analyzed, with the exception of empty ones.

The index of relative importance (IRI) was employed as a measure that reduces bias in the description data of animal dietary items (Pinkas et al. 1971).

$$IRI = (N\% + V\%)\ F\%$$

Where N%=numeric percentage, V%=volumetric percentage, F=frequency of percentage

In order to compare the habitat trophic niche breadth the standardized Shannon-Weaver entropy index J' was used (Shannon & Weaver 1949).

$$J' = H'/\ln(n)$$

whereby,

$$H' = -\Sigma pi \ln(pi)$$

pi is the relative abundance of each prey categories, calculated as the proportion of prey items of given categories to the total number of prey items (n) in all compared species. To make H' index number more biological sense, it was converted into the effective number of species (ENS), which is the real biodiversity and allows to compare the biodiversity with the other community containing equally-common species of exp(H'), the ENS.

The niche breadth was obtained by Levins' standardized index (Krebs 1999), in which the value of Levins' measure (B) was first obtained by the following equation

$$B = 1/\Sigma pi^2$$

where, pi =fraction of item i in the diet

Levins' measure was then standardized on a scale of 0-1.0 by the following equation:

$$B_A = (B-1)/(n-1)$$

where, B_A corresponding to Levins' standardized niche breadth ranges from 0 (narrowest amplitude), when there is exclusive use of a single resource categories, to 1 (broadest amplitude), when all categories are equally used (Krebs 1999); the species is considered to have a wide niche breadth when $B_A \geq 0.5$.

RESULTS

The anurans used in this study, consisted of 84 specimens of three toad species. We recorded 1,222 prey items from 27 invertebrate categories (Table 1). Because toad samples were stomach-flushed within three hours after capture, few of the food materials were totally intact, most were partially digested. Parts with heavily sclerotised cuticle remained undigested so that heads, thorax, abdominal segments and single wings of arthropods allowed an identification of the item, at least to order level. Identified diet items belonging to the order Hymenoptera were categorized into Formicidae and non Formicidae. Mostly male *Bufo* specimens seem to stop feeding during courtship so some of them had an empty stomach (Table 1).

The most numerous prey taxon on the basis of number percent in the diet was Formicidae in all three toad species. The predominant food in terms of volume was Orthoptera in *D. himalayanus* and *D. melanostictus* while it was Lepidoptera in *D. stomaticus*. The index of relative importance (IRI) was maximum for Formicidae in the three toad species (Table 2; Fig. 2). Based on

Table 1. Prey details for all three bufonid species in studied sites of Uttarakhand, western Himalaya.

Total sample size	*Duttaphrynus himalayanus*	*Duttaphrynus melanostictus*	*Duttaphrynus stomaticus*
Individual with empty stomach	7	7	8
Total prey taxa present	24	25	19
Total no. of prey	376	322	524
Average no. of prey items/sample	22	13	20
Maximum no. of prey/sample	26	19	25
Terrestrial preys (%)	95.73	96.89	94.46
Aquatic preys (%)	4.26	3.10	5.53
Maximum length of prey items (mm)	26	26	22
Minimum length of prey item (mm)	9	4	2

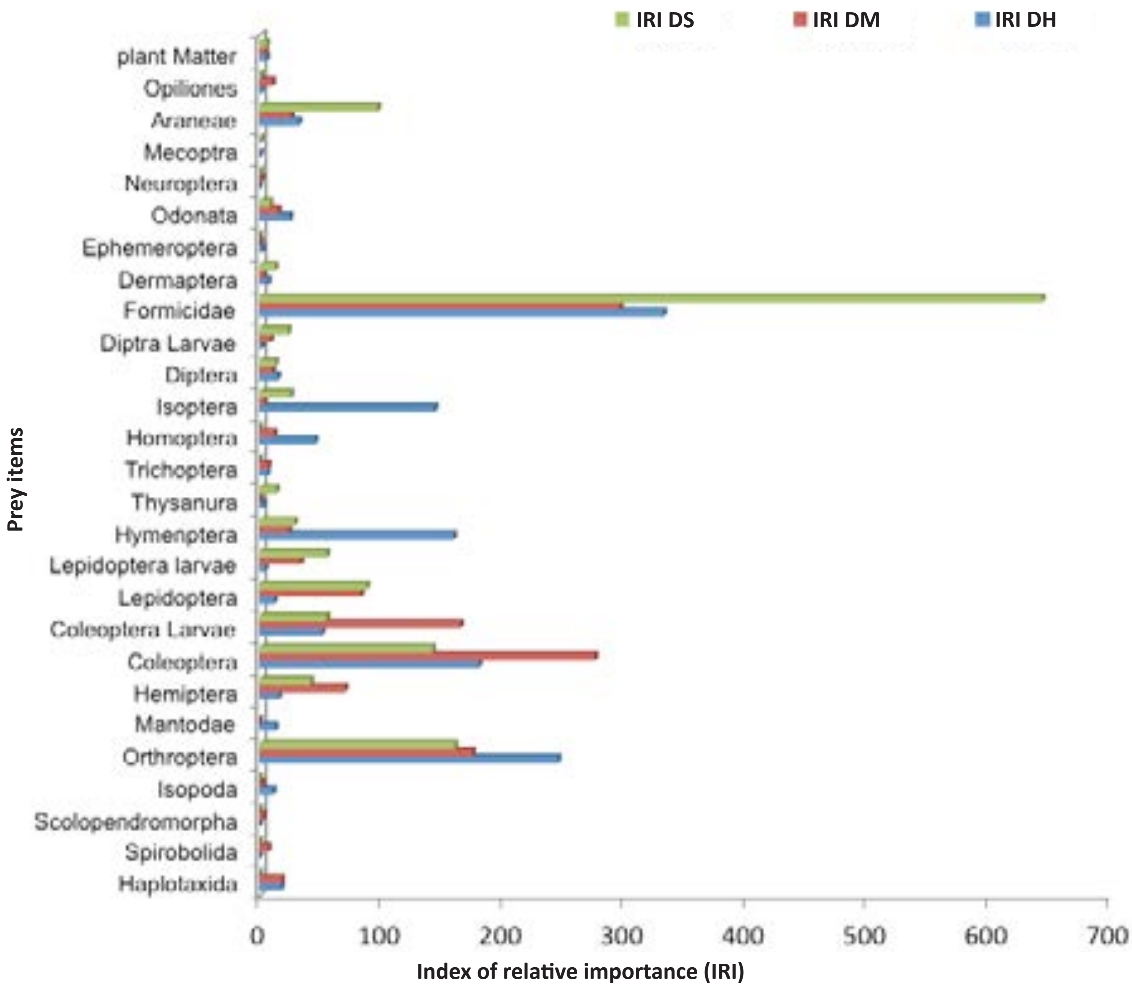

Figure 2. Index of relative importance (IRI) for prey items based on total diet contents of *D. himalayanus* (D.H.), *D. melanostictus* (D.M.) and *D. stomaticus* (D.S.) in Uttarakhand.

the Shannon-Wiener function, *D. melanostictus* had the highest prey diversity followed by *D. himalayanus* and *D. stomaticus* (Table 3). As for the niche breadth, *Duttaphrynus melanostictus* also had the broadest dietary niche breadth, followed by, *D. himalayanus* and *D. stomaticus*, in that order (Table 3).

Table 2. Shannon-Wiener function of niche breadth (H'), evenness measure (J'), Levin's measure of niche breath (B'), and standardized Levin's measure of niche breath (B_A) of prey items of studied bufonid species in Uttarakhand.

Species	Shannon-Wiener function		Levin's measure	
	H'(*)	J'	B	B_A
D. himalayanus	2.37 (10.69)	0.757	7.60	0.300
D. melanostictus	2.76 (15.79)	0.859	11.52	0.438
D. stomaticus	2.20 (9.02)	0.748	4.86	0.214

DISCUSSION

D. himalayanus is a large toad distributed in the high altitudinal region of the Himalaya, while *D. melanostictus* and *D. stomaticus* are found up to 2,500m but prefer lowland plains and agricultural as well as urban areas in Uttarakhand (Husain 2015). The inter-locality variations and similarities in the diets of these three toad species suggest that these are generalist predators that lack an apparent food preference, and that their diets are most likely dependent on what type of prey is available in inhabited areas, but prey diversity may vary among regions. As a result, *D. melanostictus* can be expected to have access to a greater variety of prey types. *D. melanostictus* was the only species that preyed upon all about the prey orders recorded and shown rich prey species biodiversity index by Shannon-Wiener measure of niche breadth (H'=2.76). In spite of this, due to the dominance of Formicidae in its diet, *D. stomaticus* has a lower prey diversity index (H'=2.20) than other toad species. *D. himalayanus* has intermediate value of prey diversity (H'=2.37) (Table 3). Toft (1980, 1981) stated that many species from the family Bufonidae are specialists, characterized by the preference of some arthropods (often Formicidae). Levins' measure of niche breadth does not allow for the possibility that resources vary in abundance. In many cases, ecologists should allow for the fact that some resources are very abundant and common, and other resources are uncommon or rare. Levin's measure of niche breadth (B_A) calculated for the three species of toads are less than 0.5 in our study which shows the opportunistic feeding behavior of the studied toad species. Study of Levin's measure of niche breadth (B_A) in *D. melanostictus* from southwestern Taiwan also showed resemblance (Norval et al. 2014).

Toad feeds exclusively on the ground on a wide variety of terrestrial food in which arthropods are dominant (Mercy 1999; Hirai & Matsui 2000; Kidera et al. 2008; Menin et al. 2015). Our study showed that arthropods and invertebrates including other prey groups are the main constituents of the diet. This study revealed consistency in the presence of a few dominant taxonomic groups of prey in these species, but differences in diversity of the occurrence of other prey items. This may be due to the fact that the diets of these toads are defined by prey availability more than by active choice. Previously, it had been reported that a higher frequency of prey and presence of different prey sizes in the stomachs of some toad species were due to the availability of prey in the habitat of the predator (Guix 1993; Sulieman et al. 2016).

Toads might be classified as an ant specialist and wide forager, this classification is justified by having slow moving locomotion, possessing toxins in the parotid glands, prefer small preys, and high frequency of ants founds per stomach (Ferreira & Teixeira 2009). Ants and several beetle groups are unpalatable to many predators due to formic acids and quinones, respectively (Zug & Zug 1979). Therefore, specialization on those preys might confer certain advantages. Predators specialized in eating unpalatable preys decrease food competition with other predators. In our study, Formicidae was the most common prey category consumed maximum in comparison to other prey categories. This is due to their abundance and wide range of habitats. Zug et al. (2001) and Damasceno (2005) also reported that ants are common and the basic food content of toads with low energy value due to a large amount of exoskeleton when compared to other insects such as larvae of some insects (e.g., caterpillars); however, the studied toad species readily feeds on arthropods, such as ants, beetles, millipedes and centipedes that contain noxious chemicals. Toads actually incorporate the noxious chemicals produced by such type of arthropods into their own defensive mechanisms (Daly 2007). Therefore, the kind of food spectrum is very important for the composition of the toad poison and its defensive activity also.

Observations of stomach content analysis of adult toads revealed that the diet composed of insects of the orders Coleoptera, Hymenoptera, Isoptera, Lepidoptera, Orthoptera, Hemiptera, and Diptera. Some of these are major pests of an agricultural crop of this region. Toads feed on these harmful pests and help in controlling them. Apart from insects, the diet also includes annelids, crustaceans and some plant materials. Plant matter such as stem of Doab Grass *Cynodon dactylon* was observed in the diet of *D. himalayanus* and plant seeds in *D. melanostictus* and *D. stomaticus*. Similar

Table 3. Dietary items of the D. himalayanus, D. melanostictus, and D. stomaticus with their respective absolute values and relative abundance (N and N%), frequency (F and F%), volume (V and V%) and Importance of relative index (IRI).

	Duttaphrynus himalayanus				*Duttaphrynus melanostictus*				*Duttaphrynus stomaticus*			
Prey Taxa	**N (%)**	**V (%)**	**F (%)**	**IRI**	**N (%)**	**V (%)**	**F (%)**	**IRI**	**N (%)**	**V (%)**	**F (%)**	**IRI**
Class: Clitellata												
Haplotaxida	4 (1.06)	209.34 (5.5)	3 (2.65)	17.42	9 (2.8)	226.08 (3.6)	4 (2.7)	17.43	0	0	0	0
Class: Diplopoda												
Spirobolida	0	0	0	0	5 (1.55)	117.75 (1.9)	3 (2.03)	6.99	0	0	0	0
Class: Chilopoda												
Scolopendromorpha	0	0	0	0	1 (0.31)	242.82 (3.9)	1 (0.68)	2.86	0	0	0	0
Class: Malacostraca												
Isopoda	9 (2.39)	66.98 (1.76)	3 (2.65)	11.02	3 (0.93)	32.15 (0.5)	2 (1.35)	1.96	0	0	0	0
Class: Insecta												
Orthoptera	10 (2.66)	1360 (35.74)	7 (6.19)	237.87	19 (5.9)	1360.67 (22)	9 (6.08)	169.38	18 (3.44)	736.85 (22.85)	11 (6.11)	160.61
Mantodea	5 (1.33)	84.78 (2.23)	4 (3.54)	12.59	0	0	0	0	0	0	0	0
Hemiptera	10 (2.66)	65.41 (1.72)	4 (3.54)	15.5	13 (4.04)	445.2 (7.2)	9 (6.08)	68.25	21 (4.01)	58.61 (1.82)	13 (7.22)	42.07
Coleoptera	48 (12.77)	267.94 (7.04)	10 (8.85)	175.32	46 (14.29)	183.16 (3)	23 (15.54)	268	43 (8.21)	169.56 (5.26)	19 (10.56)	142.11
Coleoptera larvae	21 (5.59)	94.2 (2.48)	7 (6.19)	49.96	31 (9.63)	267.94 (4.3)	17 (11.49)	160.27	29 (5.53)	50.24 (1.56)	14 (7.78)	55.16
Lepidoptera	6 (1.6)	66.98 (1.76)	4 (3.54)	11.89	17 (5.28)	602.88 (9.7)	8 (5.41)	81.12	13 (2.48)	942 (29.21)	5 (2.78)	88.02
Lepidoptera larvae	3 (0.8)	66.98 (1.76)	2 (1.77)	4.53	9 (2.8)	435.41 (7)	5 (3.38)	33.19	11 (2.1)	468.9 (14.54)	6 (3.33)	55.46
Hymenoptera	29 (7.71)	538.51 (14.15)	8 (7.08)	154.77	12 (3.73)	37.68 (0.6)	8 (5.41)	23.45	17 (3.24)	104.66 (3.25)	8 (4.44)	28.84
(Non Formicidae)												
Formicidae	101 (26.86)	47.1 (1.24)	13 (11.5)	323.25	61 (18.94)	83.73 (1.4)	21 (14.19)	287.91	221 (42.18)	75.36 (2.34)	26 (14.44)	642.95
Thysanura	4 (1.06)	18.84 (0.5)	2 (1.77)	2.75	1 (0.31)	37.68 (0.6)	1 (0.68)	0.62	9 (1.72)	32.94 (1.02)	9 (5)	13.69
Trichoptera	3 (0.8)	100.48 (2.64)	2 (1.77)	6.09	3 (0.93)	263.76 (4.3)	2 (1.35)	7.01	0	0	0	0
Homoptera	11 (2.93)	205.14 (5.39)	6 (5.31)	44.18	9 (2.8)	183.16 (3)	3 (2.03)	11.67	0	0	0	0
Isoptera	62 (16.49)	125.6 (3.3)	8 (7.08)	140.11	9 (2.8)	10.46 (0.2)	2 (1.35)	4.01	33 (6.3)	10.46 (0.32)	7 (3.89)	25.75
Diptera	9 (2.39)	32.96 (0.87)	5 (4.42)	14.41	11 (3.42)	28.26 (0.5)	4 (2.7)	10.48	14 (2.67)	8.63 (0.27)	8 (4.44)	13.06
Diptera larvae	2 (0.53)	18.84 (0.5)	2 (1.77)	1.81	14 (4.35)	6.28 (0.1)	3 (2.03)	9.02	22 (4.2)	2.09 (0.06)	10 (5.56)	23.68

Prey Taxa	*Duttaphrynus himalayanus*				*Duttaphrynus melanostictus*				*Duttaphrynus stomaticus*			
	N (%)	V (%)	F (%)	IRI	N (%)	V (%)	F (%)	IRI	N (%)	V (%)	F (%)	IRI
Dermaptera	4 (1.06)	61.23 (1.61)	3 (2.65)	7.09	13 (4.04)	56.52 (0.9)	1 (0.68)	3.35	13 (2.48)	14.13 (0.44)	8 (4.44)	12.97
Ephemeroptera	3 (0.8)	20.93 (0.55)	2 (1.77)	2.39	2 (0.62)	25.12 (0.4)	1 (0.68)	0.69	0	0	0	0
Odonata	6 (1.6)	200.96 (5.28)	4 (3.54)	24.36	1 (0.31)	1356 (21.9)	1 (0.68)	14.99	3 (0.57)	235.2 (7.29)	2 (1.11)	8.74
Neuroptera	0	0	0	0	2 (0.62)	14.13 (0.2)	2 (1.35)	1.15	2 (0.38)	18.84 (0.58)	1 (0.56)	0.54
Mecoptera	1 (0.27)	18.84 (0.5)	1 (0.88)	0.68	0	0	0	0	1 (0.19)	42.39 (1.31)	1 (0.56)	0.84
Class: Arachnida												
Araneae	19 (5.05)	32.37 (0.85)	6 (5.31)	31.33	16 (4.97)	23.55 (0.4)	7 (4.73)	25.3	37 (7.06)	28.26 (0.88)	22 (12.22)	97.01
Opiliones	1 (0.27)	58.61 (1.54)	1 (0.88)	1.6	7 (2.17)	58.61 (0.9)	5 (3.38)	10.53	4 (0.76)	5.23 (0.16)	2 (1.11)	1.03
Unidentified	0	14.13 (0.37)	3 (2.65)			33.49 (0.5)	4 (2.7)			216.66 (6.72)	4 (2.22)	
Plant matter	5 (1.33)	28.26 (0.74)	3 (2.65)	5.5	8 (2.48)	65.41 (1.1)	2 (1.35)	4.78	13 (2.48)	4.18 (0.13)	4 (2.22)	5.8
Total	376 (100)	3805.41 (100)	113 (100)	1296.423	322 (100)	6197.9 (100)	148 (100)	1224.4	524 (100)	3225.19 (100)	180 (100)	1418.36

observations for the intake of plant matter in Bufonidae were also made by Winston (1955) and Tyler (1958) as they had recorded the ingestion of the calyces of *Morinda lucida* by *D. regularis* and presence of the flowers of *Polygonum amphibium* and grass in the stomachs of *Rana esculanta*, respectively. Although the immediate most used explanation would imply accidental ingestion of vegetation while foraging for invertebrate preys, the idea that anurans may actually select plant matters as food items must be considered. According to Anderson et al. (1999) and Santos et al. (2004), plant contents may help in the elimination of intestinal parasites; provide roughage to assist in grinding up arthropod exoskeletons, and an additional source of water and nutrients.

CONCLUSION

The present findings indicate a high percentage of terrestrial food items found in three Bufonids reaffirms that *D. himalayanus, D. melanostictus*, and *D. stomaticus* are natural predator of various insect pests especially those which are considered as serious crop pests in this region. Diverse food items found in the bufonids' stomachs illustrate the ability to utilize a wide variety of prey taxa in the high altitude region of the western Himalaya also. Thus, they play a very important role in ecological balance as well as the economy of nature. This is the first unique report on feeding of these toads

Image 1. Some major diet items of *Duttaphrynus himalayanus.* © Vivekanand Bahuguna.

Image 2. Some major diet items of *Duttaphrynus melanostictus*. © Vivekanand Bahuguna.

Image 3. Some major diet items of *Duttaphrynus stomaticus*. © Vivekanand Bahuguna.

from Uttarakhand region of the western Himalaya. Information pertaining to the food spectrum analysis contributes to understanding the ecological roles in the ecosystem and used as a baseline data for future successful amphibian conservation and management programs in the Himalayan ecosystem.

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Moulting pattern and mortality during the final emergence of the Coromandel Marsh Dart Damselfly *Ceriagrion coromandelianum* (Zygoptera: Coenagrionidae) in central India

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Abstract: The final emergence of the Coromandel Marsh Dart Damselfly *Ceriagrion coromandelianum* was studied for 50 days (22 January–12 March, 2011) from the botanical garden of Hislop College, Nagpur, India, (a semi controlled site) where small underground cement tubs/tanks are used to grow macrophytes by the Botany department. In *C. coromandelianum* emergence is asynchronous, diurnal and occurs between 07.00h and 18.00h. Stage-I starts when the ultimate instar nymph of *C. coromandelianum* leaves the water body, searches for a suitable place and then begins to shudder its body to detach the trapped pharate from the nymphal exuvia. The pharate exerts pressure on the thoracic tergites to split the cuticle. Stage-II starts when the head and thorax of the pharate emerges out of the split exuvia. The pharate struggles to remove its trapped body from the nymphal exuvia. During Stage-III, the wings expand but are opaque; pigmentation of the body occurs simultaneously all over the body. Soon the whole body develops its species specific coloration while the expanding wings gain transparency, unfold and separate out and now the imago is ready for its maiden flight. Stages I, II, and III occupy 31.66%, 11.73%, and 56.60% of the total moulting period, respectively. A total of 243 emergences occurred during the observation period, 158 emergences occurred in tanks containing *Pistia stratiotes,* while 65 emergences in tubs containing *Nymphaea nouchali* indicating that *C. coromandelianum* prefers *P. stratiotes* over *N. nouchali* for oviposition. Twenty deaths were recorded during the present observation. Failure to moult (15%) and failure to emerge completely out of the exuvia (85%) were the two reasons for mortality.

Keywords: Dragonfly, emergence, exuvia, instar, metamorphosis, moulting, pharate.

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Author contribution: NRT and PRV contributed in field work and documentation of the oviposition behaviour. RJA set up the project and evaluated the findings.

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INTRODUCTION

In Odonata, moulting during the final emergence when the aerial imago is released from the exuvia of aquatic nymph is a fascinating event involving many different types of rhythmic movements. It is also a very vulnerable period since the helpless individual is exposed to various antagonistic factors of the environment. This process was documented by various workers (Tillyard 1917; Corbet 1957; Pajunen 1962; Trottier 1966; Ubukata 1981; Banks & Thompson 1985; Gribbin & Thompson 1990, 1991; Haslam, 2004; Andrew & Patankar 2010) and was evaluated by Corbet (1999) who divided it into four observable stages. Later, Andrew & Patankar (2010) modified this division and proposed only three stages taking into consideration the time-lag and attainment of morphological characters of the freshly moulted imago. Eda (1963), reported two major types of posture during emergence- horizontal emergence commonly found in Zygoptera and Gomphidae and the vertical, found in the remaining groups, though inverted emergence has also been reported in some species of Zygoptera (Rowe 1987). Mortality during emergence can be caused by three observable factors: failure to moult, failure to harden body/wings, and predation (Thompson 1991; Bennett & Mill 1993; Andrew 2010). It can range from 0% to 100% and is dependent upon factors like temperature, rain, wind, oxygen level, lack of suitable emergence support, overcrowding and predation (Corbet 1957, 1999; Pajunen 1962; Kurata 1974; Inoue 1979; Thompson et al. 1985; Gribbin & Thomas 1990; Bennett & Mill 1993; Jacob & Suhling 1999; Purse & Thompson 2003; Andrew 2010). Most studies on the final emergence in Odonata are confined to species of the sub-tropical and temperate regions, while only a few attempts have been undertaken to study this process in detail in the tropical region mostly covering only the anisopteran species (Mathavan & Pandian 1977; Andrew 2010, 2012; Andrew & Patankar 2010).

The zygopteran *Ceriagrion coromandelianum* (Fabricius, 1798) is a very common damselfly of the Indian subcontinent. The life history of this species was described in detail by Kumar (1980) and Sharma (2009). Kumar (1980) also described the larval morphology of all the instars. We have used this species to evaluate various aspects of odonate reproductive biology (Andrew et al. 2011a,b; Thaokar et al. 2018a,b). It is found almost throughout the year, ovipositing in various floating and submerged vegetations of small natural and man-made water bodies (Sharma 2009; Andrew et al. 2011a). The present paper describes the pattern and process of emergence of this damselfly with a note on mortality during this event.

MATERIAL AND METHODS

Site: The observation was carried out at the botanical garden of Hislop College, Nagpur, (21.147^{0}'N, 79.071^{0}E), India, a semi-controlled site, where small underground cement tubs are used to grow macrophytes by the Botany department. The tubs contain floating *Nymphaea nouchali, Lemna paucicostata* and submerged *Hydrilla verticillata* vegetation, while the cement tank contains only *Pistia stratiotes*. These are surrounded by bushes of flowering plants and post-noon, this area is under the shadow of the college building. *Ceriagrion coromandelianum* is found breeding all round the year at this site.

Mature F-0 larvae were collected from this site and kept in a glass tank partially filled with water along with floating vegetation. Natural conditions were maintained by keeping the containers near the large open windows. With the help of an aim-n-shoot Sony (DSC-W30) and Canon (G11) cameras, various stages of the process of moulting during metamorphosis were documented. All movements of the larva/emerging pharate were documented and an electronic stopwatch was used to record the time. Some of the emergences were directly recorded at the collection site. Daily collection of exuviae was undertaken at the study site from 22 January to 12 March, 2011 (Table 1). Details of the weather report of the city were procured from the website https://www.timeanddate.com.

RESULTS

The daily emergence of *Ceriagrion coromandelianum* was recorded for 50 days by collecting the exuviae from the water tubs of the above described site from 22 January to 12 March 2011. The tubs and tank were filled with floating *Pistia stratiotes* and *Nymphaea nouchali* which made a perfect substrate for the final emergence. A total of 243 emergences occurred during the observation period of 50 days at the study site (excluding the ones collected from the study site and reared in the laboratory), 158 emergences occurred in tanks containing *Pistia stratiotes* while 65 emergences in tanks containing *Nymphaea nouchali.* Two peaks of emergences were recorded, the first emergence on 14 February (19) and the second on 21 February (17) (Table 1). Fifty percent of the

Table 1. *Ceriagrion coromandelianum*: Number of emergence and mortality observed during the 50-day study period (mortality in parenthesis).

	Date	*Pistia stratiotes*	*Nymphaea nouchali*	Mortality	Total
1	22.i.2011	0	0	0	0
2	23.i.2011	2	0	0	2
3	24.i.2011	1	0	0	1
4	25.i.2011	2+(1)	0	1	3
5	26.i.2011	1	0	0	1
6	27.i.2011	3+(1)	0	1	4
7	28.i.2011	0	0	0	0
8	29.i.2011	0	1	0	1
9	30.i.2011	2	2+(1)	1	5
10	31.i.2011	1	0	0	1
11	01.ii.2011	0	1	0	1
12	02.ii.2011	0	0	0	0
13	03.ii.2011	1	2+(1)	1	4
14	04.ii.2011	0	0	0	0
15	05.ii.2011	3	1	0	4
16	06.ii.2011	5+(1)	1	1	7
17	07.ii.2011	2	0	0	2
18	08.ii.2011	2	0	0	2
19	09.ii.2011	1	2	0	3
20	10.ii.2011	1+(1)	0	1	2
21	11.ii.2011	2	0	0	2
22	12.ii.2011	4	0	0	4
23	13.ii.2011	12	4	0	16
24	14.ii.2011	16+(3)	3	3	22
25	15.ii.2011	6	3	0	9
26	16.ii.2011	2	3	0	5
27	17.ii.2011	4	1	0	5
28	18.ii.2011	3+(1)	2+(1)	2	7
29	19.ii.2011	6	0	0	6
30	20.ii.2011	12+(1)	5	1	18
31	21.ii.2011	8	5	0	13
32	22.ii.2011	10+(2)	1	2	13
33	23.ii.2011	4	5	0	9
34	24.ii.2011	5+(2)	1	2	8
35	25.ii.2011	5	3	0	8
36	26.ii.2011	0	0	0	0
37	27.ii.2011	2	4+(1)	1	7
38	28.ii.2011	4	1	0	5
39	01.iii.2011	5	2	0	7
40	02.iii.2011	0	0	0	0
41	03.iii.2011	0	0	0	0
42	04.iii.2011	2	2	0	4
43	05.iii.2011	5+(1)	1	1	7
44	06.iii.2011	4	2	0	6
45	07.iii.2011	1	2	0	3
46	08.iii.2011	3	0	0	3
47	09.iii.2011	5+(1)	3+(1)	2	10
48	10.iii.2011	1	0	0	1
49	11.iii.2011	0	0	0	0
50	12.iii.2011	0	0	0	0
	Total	**158+(15)**	**63+(5)**	**20**	**241**

total emergence of *C. coromandelianum* was observed by the 29th day (19 January 2011) (Fig. 1). The duration of the day was divided as morning (07.00–12.00 h), noon (12.00–16.00 h), and evening (16.00h–dusk). Emergence was not found during the pre-dusk and pre-dawn period. The number of emergence recorded were: morning 58 (23.86%), noon 166 (68.31%), and evening 19 (7.8%) (Table 2, Figure 2a). The highest number of emergence (22) was observed on 14 February 2011 (Max. temp. 34°C, min. temp 20°C, humidity 30% at noon) followed by 18 emergence on 20 February 2011 (Max. temp. 25°C, min. temp. 19°C, humidity 85% at noon) (Figure 2b). Depending upon the type of substrate *C. coromandelianum* can moult in both horizontal (on floating leaves) as well as vertical (on emerging stem) positions. Eleven complete events of metamorphosis leading to emergence of the pharate were observed and recorded. Moulting in *C. coromandelianum* is not time specific since this process occurs throughout the day between 07.00h and 18.00h (Table 3).

The following documentation describes one complete pattern of moulting during the final emergence of the damselfly, *C. coromandelianum* observed on 18 February 2011, which started at 12.55h and ended at 15.38h, (153 minutes) (Images 1–9). This process has been divided into three observable stages (Andrew & Patankar 2010).

Stage- I: At 12.55h the F–0 larva emerged out of water and climbed the floating leaf of *Pistia stratiotes*. It moved 4cm on the dry surface of the leaf, and rested. At 13.24h the larva began to shake the abdomen in the vertical plane. These movements were very slow and later it started moving it in the horizontal plane. This movement continued for 56sec. Then it started pushing the head and thorax against the leaf. The legs were spread while the posterior region of the abdomen was firmly pressed against the base. The larva moved the head sideways

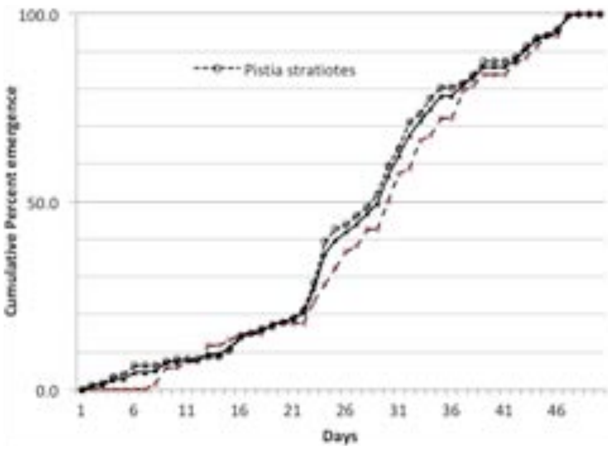


Figure 1. Cumulative percentage emergence of *Ceriagrion coromandelianum* during the 50-day study at Nagpur, India.

Images 1–6. Stages I and II of the final emergence of *Ceriagrion coromandelianum.* © Nilesh R. Thaokar.

and curved up the abdominal tip. It reset the grip of the fore and hind legs and raised the head and thorax. This movement continued interspaced with long intervals of motionless rest. It flexed the legs to elevate the anterior region of the body. At 14.03h, a split appeared along the cuticle of the dorsal region of thorax. This concluded Stage-I of moulting which took 68 minutes.

Stage- II: Within two minutes the head and thorax just elevated from the split exuvia without wriggling, leaving the exuvia on the leaf. The legs were straight sticking along the dorsal side of the body. At 14.07h, half the abdomen along with the head and thorax was outside the exuvia. The legs started flexing slowly. Initially only the forelegs exhibited movement but by 14.09h all the legs started moving and pawing the air without touching the substrate (leaf). The body of the pharate was still supported by the trapped abdomen. The thorax and abdomen formed an angle of 90 degree. The tiny compact wings lay parallel to the abdomen. The pharate now started making feeble movement of the legs trying to grip the leaf surface. As soon as it found a suitable grip for all the legs, the pharate smoothly extracted the remaining part of the abdomen from the exuvia without wriggling. It was 14.14h and the end of Stage-II. This stage took only 11 minutes.

Stage- III: The fore and mid legs of the pharate rested on the leaf while the hind legs now rested on the exuvia. It swayed the body forward and straightened the curved abdomen and swayed back to the original position without moving the legs. Slowly, the telescoped abdomen started expanding. Concomitantly, the wings also started stretching and by 14.42h, the wings were completely stretched but opaque white in colour and still stuck to each other. The pharate was motionless just re-adjusting the legs and re-gripping the leaf at regular intervals. The abdomen continued to expand and by 15.08h it was completely stretched and stiff. While the abdomen was expanding the pharate cleared the gut by forcefully expelling water (23 times) from the rectum at regular intervals. But for the eyes and a slight tinge of green on the thorax, the pharate was un-pigmented (at this stage, pink inter-segmental bands are observed in females which dissipate within a few minutes). Pigmentation of the body took place simultaneously all over the surface along with transparency of the wings and by 15.27h the freshly emerged imago became flight worthy. The imago now exhibited its characteristic species specific color patterning on the adult body. Stage-III took 74 minutes.

A comparative account of 11 complete metamorphoses on site and in the laboratory shows that on an average, the duration of Stage-I is 31.5 minutes (29.35%), Stage-II is 14 minutes (13.04%), and Stage-III is 61.83 (57.61%) in the laboratory. While on the site the average duration of

Table 2. ***Ceriagrion coromandelianum:*** **Number of emergence at different period of the day.**

	Date	Morning	Afternoon	Evening	Total
1	22.i.2011	0	0	0	0
2	23.i.2011	0	2	0	2
3	24.i.2011	0	1	0	1
4	25.i.2011	0	3	0	3
5	26.i.2011	0	1	0	1
6	27.i.2011	1	3	0	4
7	28.i.2011	0	0	0	0
8	29-.i.2011	0	1	0	1
9	30.i.2011	2	3	0	5
10	31.i.2011	0	1	0	1
11	01.ii.2011	0	1	0	1
12	02.ii.2011	0	0	0	0
13	03.ii.2011	2	2	0	4
14	04.ii.2011	0	0	0	0
15	05.ii.2011	1	2	1	4
16	06.ii.2011	2	4	1	7
17	07.ii.2011	0	2	0	2
18	08.ii.2011	0	2	0	2
19	09.ii.2011	1	2	0	3
20	10.ii.2011	0	2	0	2
21	11.ii.2011	1	1	0	2
22	12.ii.2011	1	2	1	4
23	13.ii.2011	3	11	2	16
24	14.ii.2011	6	13	3	22
25	15.ii.2011	2	6	1	9
26	16.ii.2011	1	4	0	5
27	17.ii.2011	1	3	1	5
28	18.ii.2011	2	5	0	7
29	19.ii.2011	3	3	0	6
30	20.ii.2011	4	11	3	18
31	21.ii.2011	4	8	1	13
32	22.ii.2011	5	7	1	13
33	23.ii.2011	1	8	0	9
34	24.ii.2011	3	5	0	8
35	25.ii.2011	3	4	1	8
36	26.ii.2011	0	0	0	0
37	27.ii.2011	2	5	0	7
38	28.ii.2011	2	3	0	5
39	01.iii.2011	2	4	1	7
40	02.iii.2011	0	0	0	0
41	03.iii.2011	0	0	0	0
42	04.iii.2011	0	4	0	4
43	05.iii.2011	1	6	0	7
44	06.iii.2011	1	5	0	6
45	07.iii.2011	0	3	0	3
46	08.iii.2011	0	3	0	3
47	09.iii.2011	1	8	1	10
48	10.iii.2011	0	1	0	1
49	11.iii.2011	0	0	0	0
50	12.iii.2011	0	0	0	0
	Total	**58**	**165**	**18**	**241**

Stage-I is 348 minutes (333.80%), Stage-II is 15 minutes (10.50 %), and in Stage-III it is 79.2 minutes (55.70%). Further, the average time to complete emergence is much higher on site (142.5 minutes) as compared to in the laboratory (107.33 minutes) (Tables 3, 4).

The mortality rate recorded during emergence was 8.2% (N= 20). Failure to moult (15%, Stage-I) and the failure to emerge out of the exuvia (85%, Stage-II) were the two reasons of mortality. During Stage-II, if the pharate is unable to extract the abdomen and wings from the exuvia within the optimal period, it results into a deformed imago (which may step out of the exuvia) with twisted, telescoped abdomen and crumpled, deformed wings ultimately leading to the death of the individual (Images 10–13).

Case of an unsuccessful emergence

On 18 February 2011, one larva was found out of water, preparing for the final emergence. By 12.48h a split was observed on the thorax and slowly the pharate extracted the head and thorax from the exuvia. Half the way it stopped and after a gap of three minutes it again started pulling itself out of the exuvia. At 12.58h, the wing buds along with the cuticle of the exuvia partly separated from the main body of the exuvia. The pharate struggled to pull out the wings from the exuvia wing bud case but with little success. Soon the body of the phatrate was completely out of the exuvia along with a major portion of the wings but the wing tips were still trapped. At 13.10h the part of the wings outside the exuvia started stretching and spreading and soon turned transparent, but the pharate could not release the trapped wing tips. By 14.13h although the complete body stretched but the wings lay trapped in the cuticle resulting in an adult with deformed wings (Images 14–19).

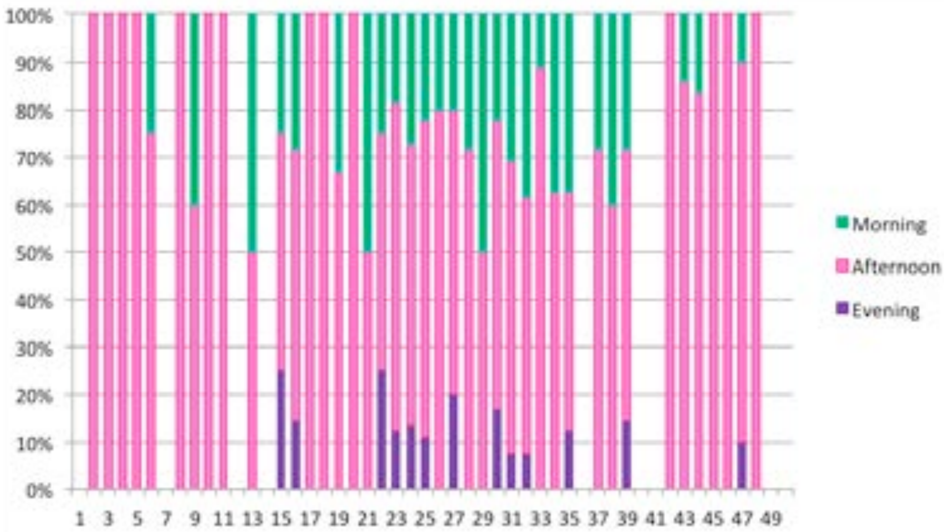


Figure 2a. Emergence of *Ceriagrion coromandelianum* at different periods of the day during the 50-day study at Nagpur, India.

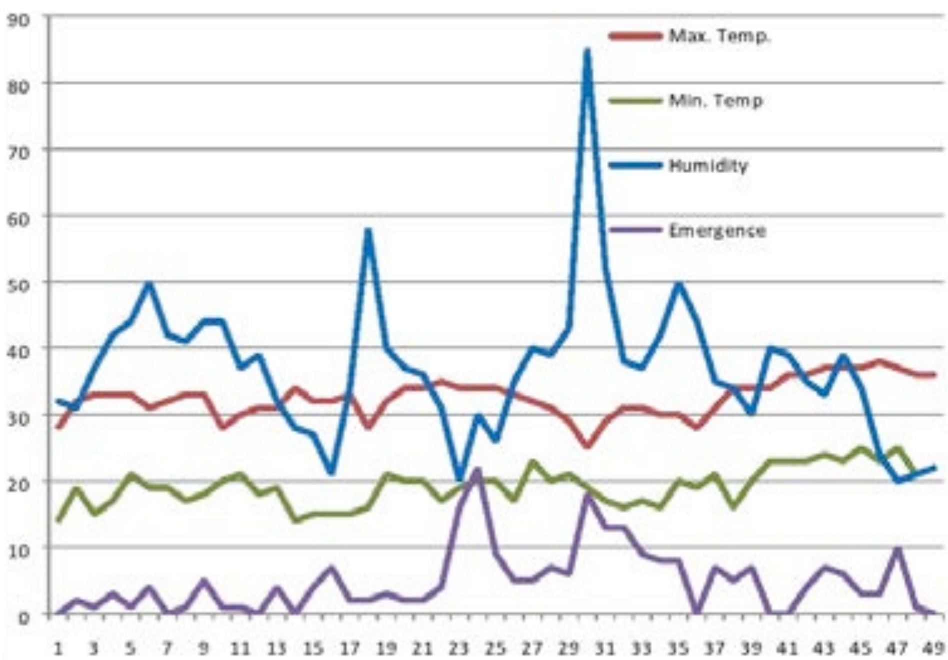


Figure 2b. Graphical representation of the daily weather (minimum temperature, maximum temperature, and humidity (at noon)) and emergence during the 50-day study at Nagpur, India.

DISCUSSION

There are two basic type of postures during emergence, the upright type where the larva completes its moulting at 0^o between body and exuvia as found in most Coenagrionidae, Gomphidae, Lestidae, Petaluridae, and the hanging type found in Aeshnidae, Calopterygidae, Corduliidae, and Libellulidae where the larva completes its moulting at an angle which ranges from 90 to 130 degrees and therefore it becomes necessary for the hanging type to climb on a vertical support substrate (Inoue 1964; Trottier 1966). Although horizontal emergence is common in Zygoptera and Gomphidae, inverted emergence occurs in the zygopteran *Xanthocnemis sinclairi* and some *Ischnura* spp. (Row 1987; Corbet 1999). Libellulidae mostly moult in a vertical position (Andrew 2010, 2012; Andrew & Patankar 2010). When the angle of emergence is manipulated, the larva tries

Table 3. *Ceriagrion coromandelianum*—duration and average timing (in minutes) of the three stages of the final emergence recorded in the laboratory.

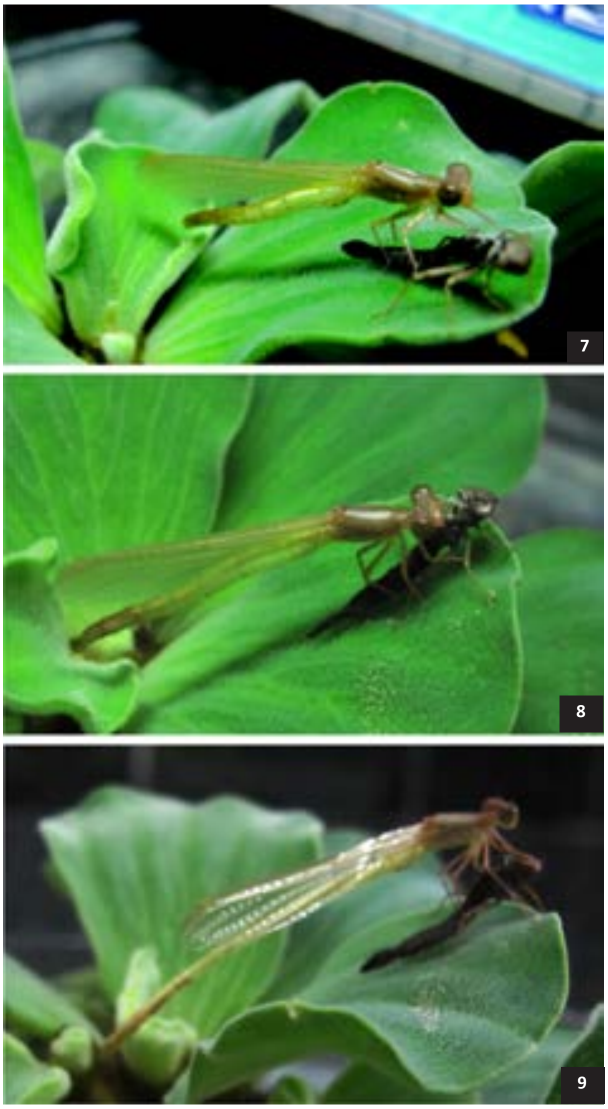
Time (h)	Stage I	Stage II	Stage III	Total
12.37–14.45	36	14	78	128
13.23–14.56	30	15	48	93
14.14–14.44	20	8	62	90
14.16–16.21	41	14	80	135
14.46–16.24	20	18	50	88
14.28–16.18	42	15	53	110
Total	**189** (29.35%)	**84** (13.04%)	**371** (57.61%)	**644**
Average	**31.5**	**14**	**61.83**	**107.33**

Table 4. *Ceriagrion coromandelianum*—duration and average timing (in minutes) of the three stages of the final emergence recorded on site.

Time (h)	Stage I	Stage II	Stage III	Total
12.43–15.08	65	10	70	145
12.56–15.06	47	11	72	130
13.06–15.47	46	13	102	161
14.10–16.30	35	25	80	140
14.23–16.38	47	16	72	135
Total	**240** (33.80%)	**75** (10.50%)	**396** (55.70%)	**711**
Average	**48**	**15**	**79.2**	**142.5**

to regain its original positioning by readjusting its body (*Calopteryx*, Heymer 1972) or by darting towards a vertical substrate (*P. flavescens*, Andrew & Patankar 2010). *C. coromandelianum* appears to be an opportunistic species and can moult in both horizontal as well as vertical position. In *C. coromandelianum,* the spreading of the wings is uniform as found in most libellulids but in most horizontal emergence the spreading starts from the base upwards (Eda 1963). Further in the hanging type of emergence, gravitational force plays an important role in setting the angle of the spreading wings with respect to the linear position of the body (Andrew & Patankar 2010), but in *C. coromandelianum* gravity does not have any influence as it exhibits both vertical and horizontal emergence. In *P. flavescens*, the pigmentation of the body starts from the thorax and terminal end of the abdomen (Andrew & Patankar 2010) but in *C. coromandelianum* pigmentation of the body takes place all over the surface, simultaneously.

Variation in the number of emergences during morning, afternoon and evening indicate that photoperiod and temperature have a direct bearing on the initiation of emergence in *C. coromandelianum.* Purse & Thompson (2003) reported that emergence in the damselfly *Coenagrion mercuriale* was positively correlated to the duration of sunlight of the previous day. Positive correlation was found between sunlight and daily emergence in *Lestes eurinus* (Lutz 1968). In *C. coromandelianum* too, maximum emergence is noticed during the afternoon period indicating a link between intensity of sunlight on emergence, but no statistically significant relationship could be established between daily temperature and humidity. Farkas et al. (2012) reported that in gomphid dragonflies, inter year variations found during emergence is due to annual fluctuation in the water temperature which may influence onset



Images 7–9. Stage III of the final emergence of *Ceriagrion coromandelianum.* © Nilesh R. Thaokar

Images 10–13. Incomplete metamorphosis during final emergence of *Ceriagrion coromandelianum.* Imago with twisted and telescoped abdomen and crumpled, warped wings (arrows). © Nilesh R. Thaokar

and synchrony of emergence. A comparative account of the time lag between the three stages of emergence between the anisopteran *P. flavescens* and zygopteran *C. coromandelianum* indicates that the major time of emergence is consumed in Stage-III for the stretching and spreading of the body and wings in both the groups.

Mortality during emergence is classified into three observable events: failure to moult, failure to expand & harden the wings, and predation. The first two are caused by factors such as low temperature, rain, wind, low oxygen level, lack of suitable emergence support and overcrowding (Corbet 1999). Lack of mass emergence results in little competition for support and eliminates overcrowding as a cause of mortality in *Onychogomphus uncatus* and *Orthetrum coerulescens* (Jakob & Suhlingg 1999) and a similar situation is also found in the observation. In the northern range margins of Britain, Purse & Thompson (2003) reported a low mortality rate of 4.9% including deformed individuals during the emergence of the damselfly *Coenagrion mercuriale.* In the damselfly *Pyrrhosoma nymphula* in Yorkshire, England the mortality during emergence ranged 3–5% and was mainly due to incomplete ecdysis, failure to expand wings and predation by spiders (Bennett & Mill 1993). In southern India, Mathavan & Pandian (1977) reported that the mortality rate of most libellulid dragonflies varied between 8% and 14% during emergence, whereas in central India the mortality rate of *P. flavescens* was 10.93% (Andrew 2010). In the Indian subcontinent predation rate is very less, and ranges from zero to 0.78% (Mathavan & Pandian 1977; Andrew 2010, 2012). Failure to moult at Stage-I indicates that there may be some endogenous (genetic) factors or injuries or dehydration which can be responsible for mortality at this stage, whereas failure to emerge out of the exuvia occurs in Stage-II and could be caused by loss of energy during moulting or difficulty in removing the trapped abdomen or wings from the exuvia (Jakob & Suhling 1999; Andrew & Patankar 2010). Strong winds are a major cause of mortality in Stage-III (Corbet 1999). In *P. flavescens*, 56% of the total mortality was found in Stage-III at an open drain in central India (Andrew 2010). In this study, we did not observe a single case of mortality at Stage-III probably because the site is well sheltered against strong winds by

Images 14–19. Unsuccessful emergence of *Ceriagrion coromandelianum* caused by wings trapped in the wing bud case of the exuvia. © Nilesh R. Thaokar

the surrounding building of the institution. In the present study, predation was not observed probably due to a lack of major predators at the semi controlled study site. Further, it couldn't be ignored that mortality will be more in natural habitats in and around areas where nesting density of predatory birds is high or where the pharate is more exposed to extreme physical factors (Corbet 1999). Jakob & Suhling (1999) found that the predatory rate during moulting in odonates is mostly less than one in most natural conditions. Nevertheless, we (Thaokar et al. 2018a,b) earlier reported that *C. coromandelianum* displays a refined hierarchy of preferences for oviposition and chooses floating leaves of *Nymphaea nouchali* over *Lemna paucicostata* and submerged *Hydrilla verticillata* but with the addition of *Pistia stratiotes* at the site, *C. coromandelianum* prefers *P. stratiotes* over *N. nouchali* for oviposition.

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COMMUNICATION

Diversity of parasitic Hymenoptera in three rice-growing tracts of Tamil Nadu, India

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Abstract: Parasitic hymenoptera play a vital role in rice ecosystems as biocontrol agents of pests. Surveys were conducted from August 2015 to January 2016 in three rice growing zones in Tamil Nadu: western zone, Cauvery Delta zone, and high rainfall zone. A total of 3,151 parasitic hymenoptera were collected, of which 1,349 were collected from high rainfall zone, 1,082 from western zone, and 720 from Cauvery Delta zone. Platygastridae, Ichneumonidae, and Braconidae were the most abundant families in all the three zones. The species diversity, richness, evenness as well as beta diversity were computed for all three zones via Simpson's, Shannon-Wiener and Margalef indices. The results showed the high rainfall zone to be the most diverse and the Cauvery Delta zone the least diverse, but with more evenness. Pairwise comparison of zones using Jaccard's index showed 75–79% species similarity.

Keywords: Cauvery Delta, diversity indices, high rainfall, parasitoids.

ஆய்வுச்சுருக்கம்: ஹைமினோப்டிரா ஒட்டுண்ணிகளானது நெல் செடியைத்தாக்கும் பூச்சிகளை கட்டுப்படுத்த மிகவும் உதவுகின்றது. தமிழ் நாட்டில் 2015 ஆம் ஆண்டு ஆகஸ்ட் மாதம் முதல் 2016 ஆம் ஆண்டு ஜனவரி மாதம் வரை ஹைமினோப்டிரா ஒட்டுண்ணிகளுக்கான கணக்கெடுப்பு மூன்று மண்டலங்களில் நடத்தப்பட்டது, அவையாவன மேற்கு மண்டலம், காவேரி டெல்டா மண்டலம் மற்றும் மழைமிகு மண்டலம். இந்த கணக்கெடுப்பின் மூலம் மொத்தம் 3151 ஒட்டுண்ணிகள் அகப்பட்டன. இவற்றுள் 1349 ஒட்டுண்ணிகள் மழைமிகு மண்டலத்திலிருந்தும், 1082 ஒட்டுண்ணிகள் மேற்கு மண்டலத்திலிருந்தும், 720 ஒட்டுண்ணிகள் காவேரி டெல்டா மண்டலத்திலிருந்தும் பிடிப்பட்டன. ஓட்டுண்ணி குடும்பங்களான பிளாட்டிகேஸ்டிரிடே, இக்கினிமோனிடே, பிரக்கோனிடே ஆகியவை அனைத்து மணிடலங்களிலும் இருந்து அதிக அளவில் பிடிப்பட்டன. பன்முகத்தன்மை குறியீடுகளான சிம்ப்ஸன்ஸ், ஷெணான் மற்றும் மார்கலெப் குறியீடுகள் கணக்கிடப்பட்டு மழைமிகு மண்டலத்திலேயே அதிக பல்லுயிர் பெருக்கம் இருப்பதாக கண்டறியப்பட்டது. மிக குறைந்த பல்லுயிர் பெருக்கம் காவேரி டெல்டா மண்டலத்தில் இருப்பதாக தெரியப்பட்டது.

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Author contribution: JAD involved in the collection of insects, segregation of collected insects up to family level, performed statistical analysis, and wrote the manuscript. KR involved in correction of the manuscript and he is the advisor of the whole study.

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INTRODUCTION

Rice fields harbor a rich and varied fauna compared to other agricultural areas (Heckman 1979; Fritz et al. 2011), which is dominated by arthropods. Communities of terrestrial arthropods in rice fields include pests and their predators and parasitoids (Heong et al. 1991). Fifty-thousand species of parasitic Hymenoptera have been described, and it is likely that this is a small percent of the total number of species (La-Salle & Gauld 1991). Parasitic Hymenoptera are more susceptible to extinction than phytophagous arthropods, and their loss can have devastating effects on ecological stability and community balance. Recently, biodiversity in agricultural land has received growing attention because it plays a significant role in agro-ecosystem function by keeping the pest populations under check (Jervis et al. 2007).

Most parasitic hymenoptera are keystone species, and their removal can result in a cascade effect (La-Salle & Gauld 1993). Utilization of parasitic Hymenoptera in insect pest management programs can bring high economic returns and support sustainable pest management. Wagge (1991) has pointed out that it is fundamentally important to conserve a large reservoir of parasitic Hymenoptera diversity. Given limited resources, it is necessary to identify groups of high priority for study, and parasitic Hymenoptera are one such group (La Salle & Gauld 1991). This study was conducted to evaluate the diversity of parasitic Hymenoptera in three different rice ecosystem zones.

MATERIALS AND METHODS

Sites of collection

The survey was carried out in rice fields during 2015–16 in three different agro-climatic zones of Tamil Nadu: western zone (District representation: Coimbatore at Paddy Breeding Station, Coimbatore, 427m, 11.007N, 76.937E), Cauvery Delta zone (District representation: Thiruvarur at Krishi Vigyan Kendra, Needamangalam, 26m, 10.774N, 79.412E), and high rainfall zone (District representation: Kanyakumari at Agricultural Research Station, Thirupathisaram, 17m, 8.207N, 77.445E) (Figure 1). Collections were made for 20 consecutive days in each zone to give equal weight and minimize chance variation in collections. In all three places conventional agronomic practices were followed. The time of sampling in each zone was decided by the rice growing season of the zone and the stage of the crop, i.e., 20 days in August–September 2015 in western zone, October–November 2015 in high rainfall zone, and December 2015–January 2016 in Cauvery Delta zone.

Methods of collection

Sweep nets, yellow pan traps at ground level, and yellow pan traps erected at canopy level were deployed continuously for 20 days.

Sweep Net

The net employed for collecting was similar to an ordinary insect net with 673mm mouth diameter and a 1,076mm long aluminum handle (Narendran 2001). The frame can be fitted to one end of a long handle that makes sweeping easy and effective. The net bag was made up of thin cotton cloth, 600mm in length with a rounded bottom. The top of the bag which fits around the frame was made of canvas folded over the frame and sewed in position. Sweeping of vegetation was as random as possible from ground level to the height of the crop. Sweeping was done in early morning and late evening hours for about half an hour per day, which involved 30 sweeps in total each day. One to-and-fro motion of the net was considered as one sweep.

Yellow pan traps kept at ground level

This trap was based on the principle that many insects are attracted to bright yellow colour. Yellow pan traps are shallow bright yellow trays 133 × 195 mm and 48mm deep (Noyes 1982). Twenty yellow pan traps were installed at ground level in each site on the bunds, half-filled with water containing a few drops of commercially available detergent to break the surface tension and a pinch of salt to reduce the rate of evaporation and prevent rotting of trapped insects. The spacing between traps was standardized at 1.5m. The traps were set for a period of 24h (Example: traps set at 10.00h on one day were serviced at 10.00h on the following day).

Yellow pan traps erected canopy level

Yellow pan traps were installed at the crop canopy by means of polyvinyl chloride pipes fitted below, with a screw attachment and were installed in 10 traps per zone in the same fashion as yellow pan traps kept at ground level.

Preservation and identification of the specimens

The parasitoids collected were preserved in 70% ethyl alcohol. The dried specimens were mounted on pointed triangular cards and studied under a Stemi (Zeiss) 2000-C and photographed under Leica M 205-A stereo zoom microscopes and identified up to the

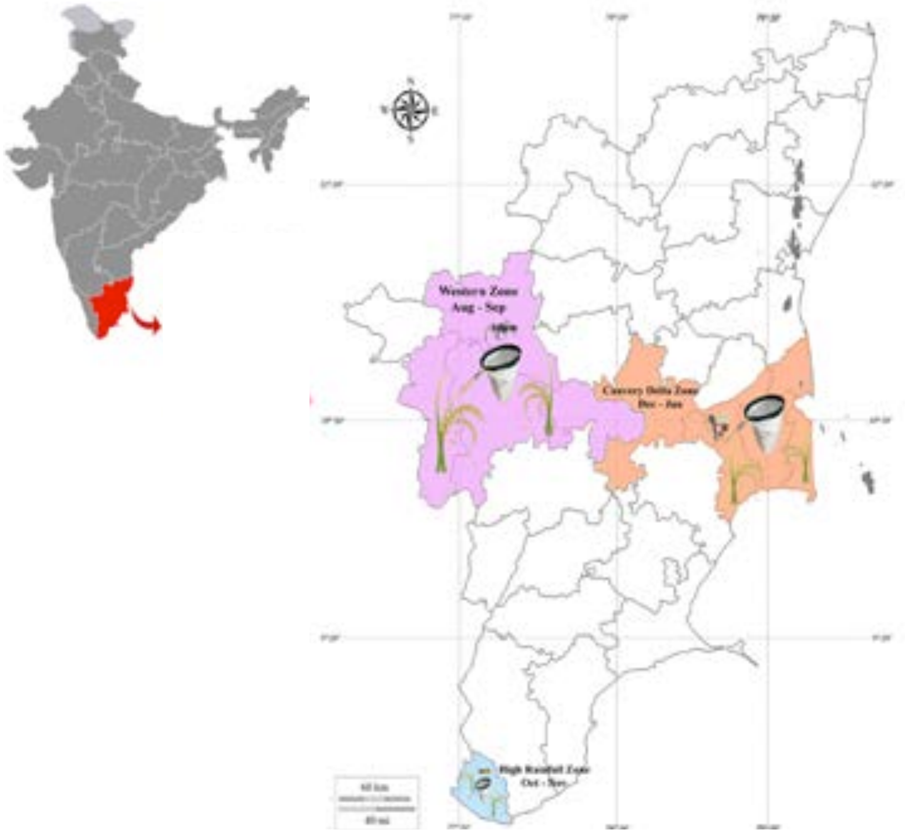

Figure 1. Three zones of collection.

family level through conventional taxonomic techniques following standard keys given by several authors like Narendran (1994), Jonathan (2006), Rajmohana (2006), Sureshan (2008) and "Universal Chalcidoidea Database" developed by Noyes (2017). Further, experts in particular groups of parasitic Hymenoptera were met in person for getting proper identity up to sub family/ genera/ species level wherever possible. Dr. Manickavasagam Sagadai, Sankararaman Hariharakrishnan, Dr. Gowri Prakash James, and Dr. Ayyamperumal Mani (in litt. 9 August 2016) of Annamalai University, Chidambaram, Tamil Nadu helped in identifying Chalcididae, Aphelinidae, Encyrtidae, Megaspilidae, and Dryinidae specimens. Ranjith Avunjikkattu Parambil (in litt. 6 September 2016) from the University of Calicut, Kerala helped in identifying Braconidae, Gasteruptiidae and in overall segregation of all the specimens. Dr. Rajmohana Keloth (in litt. 7 September 2016) from the Zoological Survey of India, Kozhikode, Kerala, helped in identifying Platygastridae, Diapriidae, Proctotrupidae, and Ceraphronidae specimens. Dr. Sureshan Pavittu M. and Dr. Raseena Farzana Vadakkethil Kuttyhassan (in litt. 24 October 2016) from the Zoological Survey of India, Kozhikode, Kerala, helped in identifying Pteromalidae and Torymidae specimes. Dr. Santhosh Shreevihar (in litt. 4 November 2016) from the Malabar Christian College, Kozhikode, Kerala helped in identifying Bethylidae and Eulophidae. Dr. Sudheer Kalathil (in litt. 22 November 2016) from Guruvayurappan College, Kozhikode, Kerala, helped in identifying Ichneumonidae specimens, Dr. P. Girish Kumar (in litt. 30 January 2017) from the Zoological Survey of India, Kozhikode, Kerala, helped in identifying Evaniidae, Eucharitidae, and Scoliidae specimens. Dr. Nikhil Kizhakiyal (in litt. 31 January 2017) from the Zoological Survey of India helped in identifying Eurytomidae specimens. Dr. Rameshkumar Anandan (in litt. 10 February 2017) from the Zoological Survey of India, Kolkata, West Bengal helped in identifying Mymaridae and a few Encyrtidae specimens. Dr. Poorani Janikiraman (in litt. 20 March 2017) from the National Research Centre for Banana, Trichy, Tamil Nadu, helped in identifying a few Eupelmidae specimens. Dr. Gary A.P. Gibson (in litt. 24 March 2017) from the Canadian National Collection of Insects, Arachnids, and Nematodes, Canada, helped in identifying a few Eupelmidae specimens by sending keys through mail. Dr. Arkady Lelej (in litt. 15 April 2017) from the Federal Scientific Center of the East Asia Terrestrial Biodiversity, Vladivostok, Russia, helped in identifying Mutillidae specimens through photographs. Dr. Matthew Buffington (in litt. 16 April 2017) from the United States Department of Agriculture, Washington, D.C. United States helped in identifying Figitidae specimens through photographs. Dr. Lynn Kimsey (in litt. 17 April 2017) from the Bohart Museum of Entomology, University of California, helped in identifying Chrysididae and Tiphiidae specimens through photographs. Nearly, 174 species of parasitoids were collected during the entire study period, however, some of the parasitoids were identified only up to the sub family/ generic level and only a few were identified up to the species level. Identified specimens are deposited at the Insect Biosystematics lab, Department of Agricultural Entomology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India.

Measurement of diversity

Relative Density

Relative density of the species was calculated by the formula, Relative Density (%) = (Number of individuals of one species / Number of individuals of all species) X 100.

Alpha Diversity

Alpha diversity of the zones was quantified using Simpson's diversity index (SDI), Shannon-Wiener index (H'), Margalef index (α) and Pielou's evenness index (E1).

Simpson's Index

Simpson's diversity index is a measure of diversity which takes into account the number of species present, as well as the relative abundance of each species. It is calculated using the formula, $D = \Sigma n (n-1)/ N(N-1)$; where n = total number of organisms of a particular species and N = total number of organisms of all species (Simpson 1949). Subtracting the value of Simpson's diversity index from 1, gives Simpson's Index of Diversity (SID). The value of the index ranges from 0 to 1, the greater the value the greater the sample diversity.

Shannon-Wiener Index

Shannon-Wiener index (H') is another diversity index and is given as follows: $H' = -\Sigma P_i \ln(P_i)$; where $P_i = S / N$, S = number of individuals of one species, N = total number of all individuals in the sample, ln = logarithm to base e (Shannon & Wiener 1949). The higher the value of H', the higher the diversity.

Margalef Index

Species richness was calculated for the three zones using the Margalef index which is given as Margalef index, $\alpha = (S - 1) / \ln (N)$; where S = total number of species, N = total number of individuals in the sample (Margalef 1958).

Pielou's Evenness Index

Species evenness was calculated using the Pielou's evenness index (E1). Pielou's Evenness Index, $E1=H'/\ln(S)$; where H' = Shannon-Wiener diversity index, S = total number of species in the sample (Pielou 1966). As species richness and evenness increase, diversity also increases (Magurran 1988).

Beta Diversity

Beta diversity is a measure of how different (or similar) ranges of habitats are in terms of the variety of species found in them. The most widely used index for assessment of beta diversity is Jaccard index (JI) (Jaccard 1912), which is calculated using the equation: JI (for two sites) = j / (a+b-j); where j = the number of species common to both sites A and B, a = the number of species in site A, and b = the number of species in site B. We assumed the data to be normally distributed and adopted parametric statistics for comparing the sites.

Statistical analysis

The statistical test ANOVA is also used for significant difference in the collections from three zones. The data on population number were transformed into X+0.5 square root before statistical analysis. The mean individuals caught from three different zones were analyzed by adopting randomized block design (RBD) to find least significant difference (LSD). Critical difference (CD) values were calculated at 5 per cent probability level. All these statistical analyses were done using Microsoft Excel 2016 version and Agres software version 3.01.

RESULTS AND DISCUSSION

Faunal survey of parasitic hymenoptera in rice ecosystems in western zone, Cauvery Delta zone and high rainfall zones of Tamil Nadu revealed that the family richness was maximum (25) in the high rainfall zone, followed by western zone (24), and minimum (19) in Cauvery Delta zone (Table 1). All the families of parasitic hymenoptera collected in the present study along with their presence and absence details were provided in Appendix 1. Apidae, Tiphiidae, and Gasteruptiidae were collected only from the western zone and Chrysididae, Mutiliidae, Megaspilidae, and Eucharitidae were

Table 1. Comparison of parasitoid families collected from three rice growing zones of Tamil Nadu.

Families	Zones						Total			
	Western		Cauvery delta		High rainfall					
	No.	%	No.	%	No.	%	No.	%	F	P
Apidae	1	0.1	0	0.0	0	0.0	1	0.0	1.00	0.37
Bethylidae	4	0.4	2	0.3	7	0.5	13	0.4	1.16	0.32
Dryinidae	2	0.2	5	0.7	1	0.1	8	0.3	0.98	0.37
Chrysididae	0	0.0	0	0.0	1	0.1	1	0.0	1.00	0.37
Mutillidae	0	0.0	0	0.0	3	0.2	3	0.1	1.87	0.16
Scoliidae	1	0.1	0	0.0	2	0.1	3	0.1	0.60	0.55
Tiphiidae	3	0.3	0	0.0	0	0.0	3	0.1	1.00	0.37
Ceraphronidae	15	1.4	11	1.5	41	3.0	67	2.1	5.33	0.00
Megaspilidae	0	0.0	0	0.0	1	0.1	1	0.0	1.00	0.37
Aphelinidae	8	0.7	1	0.1	6	0.4	15	0.5	2.32	0.10
Chalcididae	21	1.9	16	2.2	142	10.5	179	5.7	12.79	0.00
Encyrtidae	2	0.2	8	1.1	7	0.5	17	0.5	1.39	0.25
Eucharitidae	0	0.0	0	0.0	1	0.1	1	0.0	1.00	0.37
Eulophidae	41	3.8	23	3.2	97	7.2	161	5.1	6.89	0.00
Eupelmidae	20	1.8	19	2.6	42	3.1	81	2.6	1.60	0.21
Eurytomidae	31	2.9	19	2.6	67	5.0	117	3.7	2.74	0.07
Mymaridae	15	1.4	41	5.7	36	2.7	92	2.9	3.23	0.04
Pteromalidae	32	3.0	21	2.9	29	2.1	82	2.6	0.31	0.73
Torymidae	4	0.4	0	0.0	6	0.4	10	0.3	0.84	0.43
Trichogrammatidae	59	5.5	27	3.8	22	1.6	108	3.4	1.32	0.27
Figitidae	3	0.3	2	0.3	6	0.4	11	0.3	0.54	0.58
Diapriidae	44	4.1	21	2.9	54	4.0	119	3.8	1.45	0.24
Evaniidae	13	1.2	2	0.3	8	0.6	23	0.7	1.91	0.15
Gasteruptiidae	9	0.8	0	0.0	0	0.0	9	0.3	1.00	0.37
Braconidae	180	16.6	163	22.6	231	17.1	574	18.2	0.58	0.56
Ichneumonidae	218	20.1	159	22.1	227	16.8	604	19.2	0.67	0.51
Platygastridae	314	29.0	129	17.9	288	21.3	731	23.2	4.40	0.01
Proctotrupidae	42	3.9	51	7.1	24	1.8	117	3.7	1.08	0.34
Total No. collected	**1082**	-	**720**	-	**1349**	-	**3151**	-	-	
No. of families	**24**	-	**19**	-	**25**	-	**28**	-		

%—Relative Density | No.—Total number of individuals collected | F—Value | P—Value.

collected only from the high rainfall zone. Scoliidae and Torymidae were collected both form western and high rainfall zones, but not from the Cauvery Delta zone. In the study, a total of 1,349 individuals of parasitic Hymenoptera were collected from the high rainfall zone followed by the western zone (1,082), and the Cauvery Delta zone (720) (Figure 2). In all the three zones, Platygastridae, Ichneumonidae, and Braconidae were the most abundant.

Apart from that, Trichogrammatidae, Diapriidae, Proctotrupidae, Eulophidae, Pteromalidae, Eurytomidae, Chalcididae, Eupelmidae, Ceraphronide, Mymaridae and Evaniidae constituted 5.5, 4.1, 3.9, 3.8, 3.0, 2.9, 1.9, 1.8, 1.4, 1.4, and 1.2 per cent relative density, respectively, in the western zone. Other families, viz., Apidae, Bethylidae, Dryinidae, Scoliidae, Tiphiidae, Aphelinidae, Encyrtidae, Torymidae, Figitidae, and Gasteruptiidae were represented by less than 0.8 per cent.

In Cauvery Delta zone, surprisingly, Braconidae (22.6%) was found to be predominant followed by

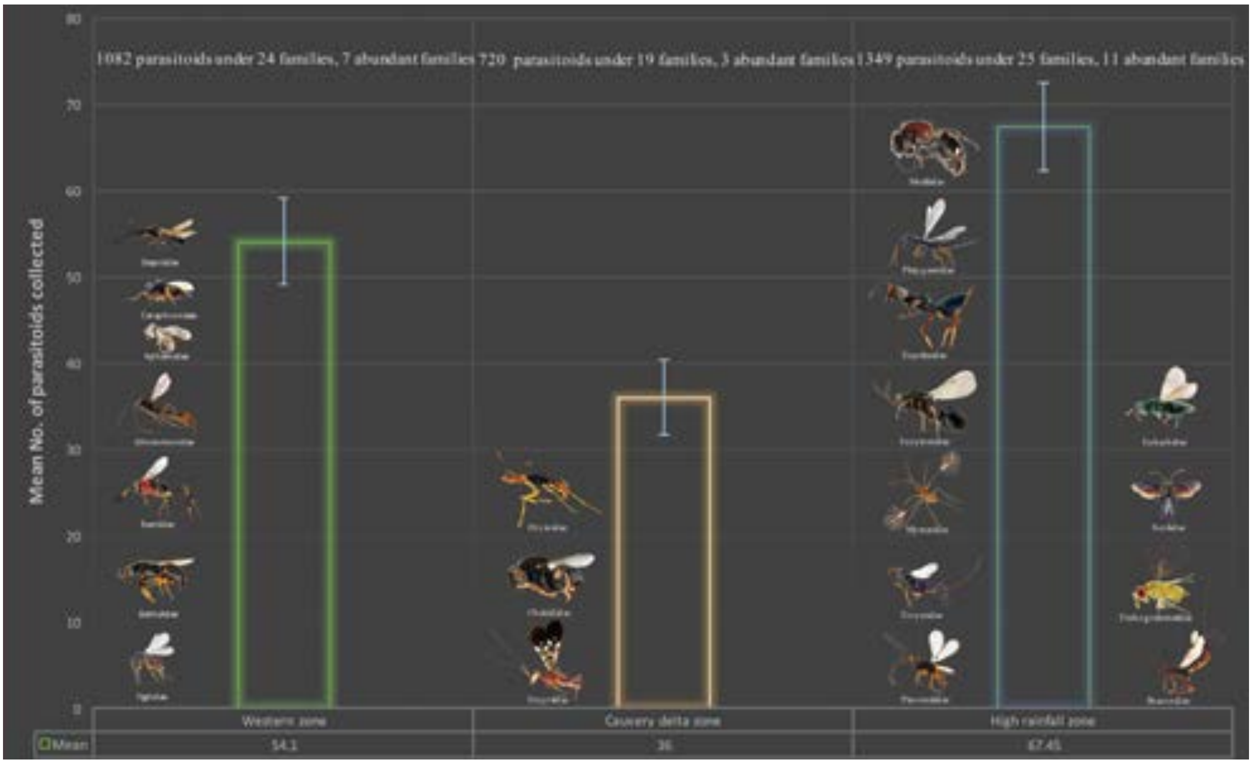

Figure 2. Parasitoids collected under three zones along with the families of abundance.

Table 2. Diversity indices of parasitic hymenoptera from three rice growing zones of Tamil Nadu

Zones	Mean No. of parasitoids collected/day	Std. Error	SID	H´	a	E1	b %
Western	54.10 (7.21)^{b}	±4.95	0.85	0.98	3.29	0.30	W and C -79
Cauvery Delta	36.00 (5.79)^{c}	±4.31	0.83	0.97	2.73	0.33	C and H - 76
High rainfall	67.45 (8.10)^{a}	±5.14	0.87	1.02	3.33	0.31	H and W -75
S.E.D	0.41	-	-	-	-	-	-
CD (p=0.05)	0.84	-	-	-	-	-	-

Figures in parentheses are square root transformed values; In a column, means followed by a common letter(s) are not significantly different by LSD (p=0.05). SID—Simpson's Index of Diversity | H'—Shannon-Wiener Index | a—Margalef index | E1—Pielou's index | b—Beta diversity (Jaccard index). W—Western zone | C—Cauvery Delta zone | H—High rainfall zone.

Ichneumonidae (22.1%) and Platygastridae (17.9%), whereas in the other two zones, Platygastridae was predominant (21.2–29.0 %). Besides these three families, Proctorupidae, Mymaridae, Trichogrammatidae, Eulophidae, Diapriidae, Pteromalidae, Eurytomidae, Eupelmidae, and Chalcididae accounted for 7.1, 5.7, 3.8, 3.2, 2.9, 2.9, 2.6, 2.6 and 2.2 per cent relative densities, respectively. All the other families were represented by less than 1.5 per cent.

In the high rainfall zone, Chalcididae was the fourth most abundant family accounted for 10.5 per cent of total collections, followed by Eulophidae (7.2%), Eurytomidae (5.0%), Diapridae (4.0%), Eupelmidae (3.1%), Ceraphronidae (3.0 %), Mymaridae (2.7%), and Pteromalidae (2.1%). All other families were represented with less than that 1.6 per cent relative density.

A total of 3,151 individuals of parasitic hymenoptera were collected in the present study from the three rice-growing zones of Tamil Nadu. This constitutes 28 families under 11 super families, three super families under Aculeata and eight super families under Parasitica. Platygastridae accounts for 23.2 per cent (Table 1) which was the highest in the collection, followed by Ichneumonidae (19.2%) and Braconidae (18.2%) (Figure 3). These three families constitute more than half, i.e., 60.6 per cent of total collection. Chalcididae was the

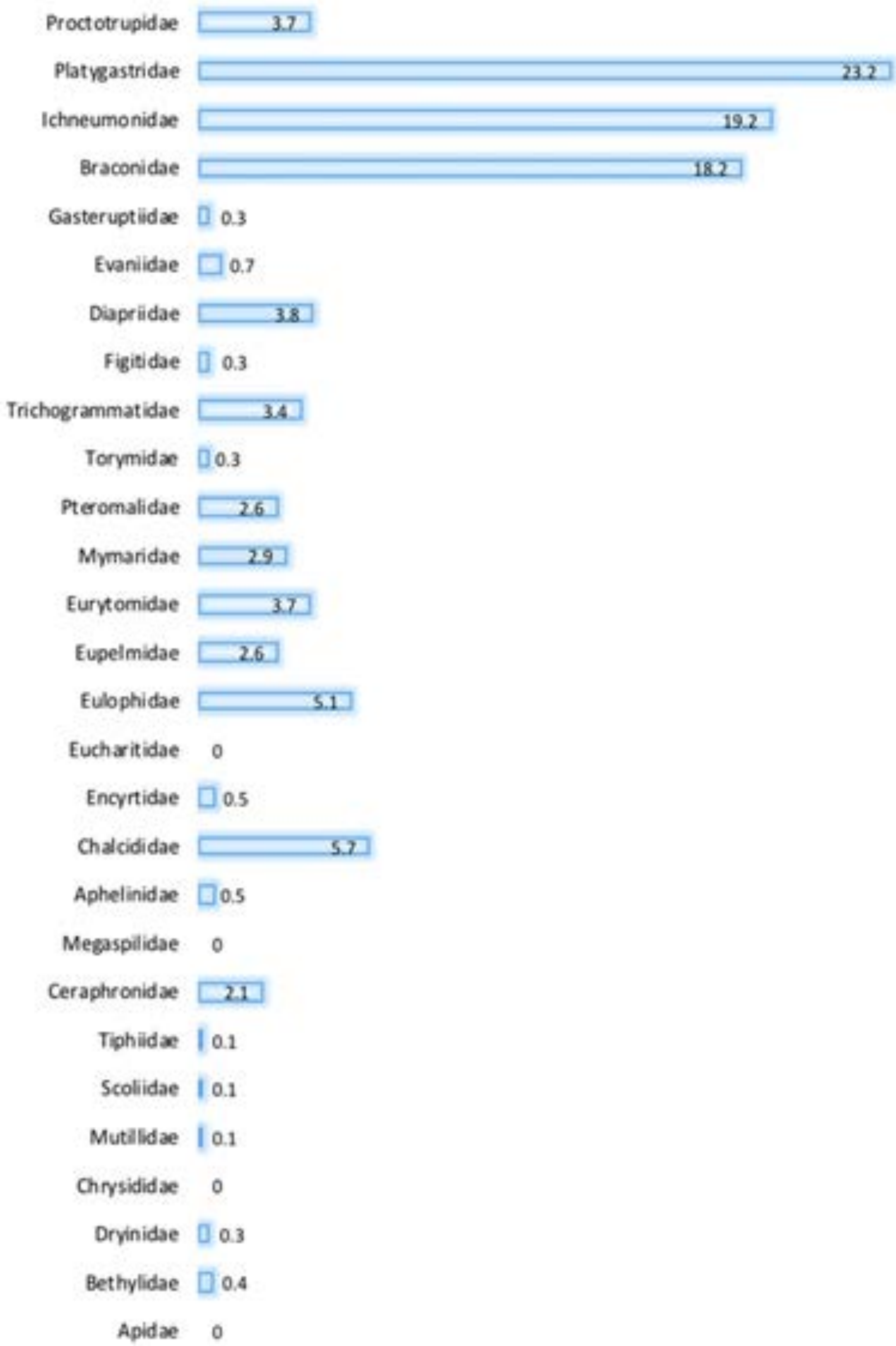

Figure 3. Relative densities of parasitic hymenoptera families from three zones of Tamil Nadu.

fourth most abundant family with 5.7 per cent relative density and Eulophidae constituted 5.3 per cent in the total collections. Diapriidae accounted for 3.8 per cent followed by Proctotrupidae and Eurytomidae with a relative density of 3.7 per cent each. Relative density of 3.4 per cent was constituted by Trichogrammatidae. Families such as Mymaridae, Pteromalidae, Eupelmidae, and Ceraphronidae accounted for 2.9, 2.6, 2.6 and 2.1 per cent, respectively (Figure 3). The other 15 families, viz., Apidae, Bethylidae, Dryinidae, Chrysididae, Mutillidae, Scolidae, Tiphiidae, Megaspilidae, Aphelinidae, Encyrtidae, Eucharitidae, Torymidae, Figitidae, Evaniidae, and Gasteruptiidae accounted for only 3.2 per cent of the total collections.

The ANOVA test results indicated that the P-value for Ceraphronidae, Chalcididae, Eulophidae, Mymaridae, and Platygastridae was less than 0.05, indicating significant difference between the zones for these five families. For all other families the P-value was greater than 0.05, which we consider to be non-significant. A mean of 67.45 ± 5.14 parasitoids per day was collected from high rainfall zone which is found to be statistically significant over other two zones. From the western zone, a mean of 54.10 ± 4.95 parasitoids were collected per day, while that in the Cauvery Delta zone was 36.00 ± 4.31 per day (Figure 2). From the Table 2, it is observed that the Simpson's diversity index ranges between 0.83 to 0.87. Though the index values are pretty much the same for all the three zones, it is the highest for the high rainfall zone (0.87), followed by the western zone (0.85), and the Cauvery Delta zone (0.83). The species composition among elevational zones can indicate how community structure changes with biotic and abiotic environmental pressures (Shmida & Wilson 1985; Condit et al. 2002). Studies on the effect of elevation on species diversity of taxa such as spiders (Sebastian et al. 2005), moths (Axmacher & Fiedler 2008), paper wasps (Kumar et al. 2008), and ants (Smith et al. 2014) reported that species diversity decreased with increase in altitude. According to Janzen (1976), however, diversity of parasitic Hymenoptera is not as proportionately reduced by elevation as in other insect groups, a fact that is in support of our results. A similar study conducted by Shweta & Rajmohana (2016) to assess the diversity of members belonging to the subfamily Scelioninae also declared that the elevation did not have any major effect on the overall diversity patterns. A similar trend was observed for the Shannon-Wiener index (H') and Margalef index (). From the values of Margalef index (a) for the three zones, it was observed that the high rainfall zone was very rich in species with a richness value of 3.33 followed by western zone (3.29) and Cauvery Delta zone (2.73). It is because of the fact that out of 28 families only 19 families were collected from this zone. The Pielou's evenness value (E1) for the sites clearly indicate that the Cauvery Delta zone showed maximum evenness pattern with evenness index value (0.33) followed by high rainfall zone (0.31) and western zone (0.30). The elevational diversity gradient (EDG) in ecology proposes that species richness tends to increase as elevation increases, up to a certain point creating a 'diversity bulge' at moderate elevations (McCain & Grytnes 2010). The elevation dealt with in this work ranged from 17–427 m which was not very high. So taking into account the scale and extent of elevation gradients, it can be said that species diversity and richness did not show any correlation, i.e., species diversity and richness were not proportional with that of elevation.

Altitudinal variation of parasitic Hymenoptera assemblages in an Australian subtropical rainforest was studied by Hall et al. (2015). To detect minute changes in species assemblages, species level sorting is found to give the best result (Grimbacher et al. 2008). The area under cultivation turns out to be a very important factor with respect to abundance and species density in rice fields (Wilby et al. 2006). The number of species in a habitat increases with increase in area (Gotelli & Graves 1996).

Comparison of species similarities using the Jaccard's index between the three sites, taken in pairs showed 79 per cent similarity between the western and Cauvery Delta zones and 76 per cent similarity between the high rainfall and Cauvery Delta zones, and 75 per cent similarity between the high rainfall and western zones.

CONCLUSION

This study reveals the diversity of Hymenoptera parasitoids of three different zones of rice ecosystems of Tamil Nadu, where the the high rainfall zone is the most diverse and the Cauvery Delta zone being the least. The reasons for the significant changes in diversity of parasitoids and their host insects are to be further studied so as to implement pest management strategies and to decide the right biological control tactics to manage pests. As very little is known of parasitic hymenoptera associated with rice ecosystem, this study attempted to enrich the information pertaining to hymenoptera parasitoids associated with rice ecosystems of Tamil Nadu. Thus, this study has generated baseline data which will be much useful for the taking up further in depth studies on Hymenoptera parasitoids of rice ecosystem.

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Appendix 1. List of parasitic hymenopteran families along with their presence and absence details in the three zones of study.

Families	Zones		
	Western	Cauvery Delta	High rainfall
Apidae	P	A	A
Bethylidae	P	P	P
Dryinidae	P	P	P
Chrysididae	A	A	P
Mutillidae	A	A	P
Scoliidae	P	A	P
Tiphiidae	P	A	A
Ceraphronidae	P	P	P
Megaspilidae	A	A	P
Aphelinidae	P	P	P
Chalcididae	P	P	P
Encyrtidae	P	P	P
Eucharitidae	A	A	P
Eulophidae	P	P	P
Eupelmidae	P	P	P
Eurytomidae	P	P	P
Mymaridae	P	P	P
Pteromalidae	P	P	P
Torymidae	P	A	P
Trichogrammatidae	P	P	P
Figitidae	P	P	P
Diapriidae	P	P	P
Evaniidae	P	P	P
Gasteruptiidae	P	A	A
Braconidae	P	P	P
Ichneumonidae	P	P	P
Platygastridae	P	P	P
Proctotrupidae	P	P	P

P—Prensent | A—Absent

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COMMUNICATION

Mapping octocoral (Anthozoa: Octocorallia) research in Asia, with particular reference to the Indian subcontinent: trends, challenges, and opportunities

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Abstract: Octocorallia (Cnidaria, Anthozoa) comprising over 3,600 nominal species within three orders, Alcyonacea, Helioporacea and Pennatulacea, is one of the most poorly known groups of marine invertebrates. Half of known octocoral species occur in the Indo-Pacific, but not much is understood about research efforts and outputs in this region, particularly in the Asian context. A review of the literature on Asian octocorals during a 40-year period from 1978 to 2018 revealed that most research was concentrated in particular regions/countries. An analysis of research originating from India indicated several issues, including low quality data and local taxonomic impediment. This paper examines the general trends and geographic disparity in Asian octocoral research over the past four decades, analyses the extent and source of such disparity by drawing parallels between India and the rest of Asia, and provides recommendations for improving octocoral studies in the region.

Keywords: India, Indian Ocean, marine invertebrates, sea fans, soft corals, taxonomy

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Author details: Ramvilas Ghosh is working on the taxonomic identity and subsequent evolution of gorgonians of the central Indian Ocean region. His research interests include octocoral taxonomy, phylogeny and conservation. Shalu Kannan is working on syngnathid fishes of India and her other interests include marine taxonomy, population genetics and conservation. Rajeev Raghavan is interested in interdisciplinary research that generates information to support conservation decision making in tropical aquatic ecosystems particularly in the Western Ghats Biodiversity Hotspot. His work cuts across multiple disciplines from taxonomy to evolutionary biogeography, fisheries management and conservation policies, and range from local to global scales. Kutty Ranjeet's interest includes understanding ecological dynamics and impacts on aquatic environments with focus on estuarine and nearshore communities. He also has experience in crustacean aquaculture and its biotechnology.

Author contribution: RG, RR and KR conceptualized and designed the work. RG and KS collected and analysed the data. RG, RR and KR wrote the manuscript.

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INTRODUCTION

Ocean life has been explored for millennia, with Aristotle's work of the 3rd Century BC on European marine biota being one of the earliest (Coll et al. 2010). Nevertheless, a large proportion of the world's marine biodiversity remains unknown (Mora et al. 2011). Knowledge of the extent and magnitude of this biodiversity (particularly lower microscopic forms) has been hindered by uneven sampling efforts and a shortfall in taxonomic expertise required for documentation (Wilson 2017). Octocorals (Cnidaria, Anthozoa), characterized by the presence of eight tentacles surrounding the mouth of the polyp, comprise a diverse group of marine organisms which includes blue corals, soft corals, sea fans and sea whips (gorgonians) and sea pens (Fabricius & Alderslade 2001). They are conspicuous members of coral reefs, often forming the frontiers (Steiner et al. 2018). They are also distributed over a broad range of bathymetry ranging from intertidal to the deep waters, and in some regions octocorals rival hard corals in biomass, abundance and diversity (Perez et al. 2016).

Octocorallia currently comprises over 3,649 nominal species within three orders, Alcyonacea, Helioporacea, and Pennatulacea (Daly et al. 2007; WoRMS 2019). They are however, one of the most poorly known groups of marine invertebrates, whose taxonomy is in a flux as a result of insufficient taxonomic expertise, high levels of homoplasy and lack of distinct diagnostic characters (except colony morphology and sclerite characteristics) that makes identification a complex affair (Perez et al. 2016). Further, missing/lost 'type material', inadequate species descriptions from the 19th and 20th century, and the likelihood of hundreds of undescribed species necessitate the reinforcement and acceleration of octocoral research, especially extensive taxonomic revisions for many alcyonacean genera (Daly et al. 2007). Despite their prominent worldwide diversity, only forty species have been assessed for the IUCN Red List of Threatened Species (IUCN 2019), highlighting the need for expanding and improving efforts for global and regional conservation prioritization (see examples Bramanti et al. 2009; Maldonado et al. 2013; Althaus et al. 2017).

Seventy per cent of known octocoral species occur in the Indo-Pacific (Perez et al. 2016), yet the region has been classified as 'data-poor' for octocorals (Bayer 1981). Though knowledge on taxonomy, diversity and distribution of octocorals in the larger Indo-Pacific region has improved substantially, many areas, e.g., the Indo-West Pacific (which is included in Asia), are still considered problematic when compared to the Mediterranean and Atlantic waters (Bayer 2002).

Based on this affirmation, we undertook a systematic review of the published literature on diversity and taxonomy of Asian octocorals (i.e., publications on octocorals reported from Asia published by both Asian and non-Asians) during a 40-year period between 1978 and 2018, to better understand the trends, status and regional inclinations of such studies. For example, despite having high levels of marine diversity (Tittensor et al. 2010) and two centuries of marine diversity inventories, comprehensive data on octocorals in and around the Indian subcontinent is extremely poor when compared to other groups of cnidaria (e.g., scleractinian corals and siphonophores; Venkataraman & Wafar 2005). In this background, we: (i) examine the general trends and geographic unevenness (if any) in Asian octocoral research, (ii) analyze the extent and source of such biases in the octocoral research arena by drawing parallels between India and the rest of Asia, and (iii) provide recommendations for improving octocoral diversity and taxonomic studies in the Indian region.

METHODS

Primary literature (concerning Asian octocorals) published during the period, 1978 to 2018 was extracted from Google Scholar™ using the following keywords: (octocorals OR Octocorallia OR Alcyonacea OR Helioporacea OR Gorgonacea OR Pennatulacea OR Stolonifera OR Telestacea OR Gorgonarian OR Gorgoniden OR Alcyonarien OR Octcorallien OR Penatulaceen OR 'soft corals' OR gorgonian OR 'sea pen' OR 'sea fan' OR 'sea whip') AND (Asia OR Japan OR Israel OR Iran OR Indonesia OR Vietnam etc.) AND (diversity OR distribution OR 'species description' OR taxonomy OR 'new species' OR 'new genus' OR 'new family'). More than 2,000 search results were manually screened to extract papers on 'diversity and taxonomy'. Based on the degree of relevance, individual papers were then eliminated by 'title' or 'abstract' alone, or by accessing the entire paper.

Similar boolean operators were used to extract papers on octocoral research in India, substituting the second and third set of keywords with India AND diversity OR distribution OR 'species description' OR taxonomy OR 'new species' OR 'new genus' OR 'new family' OR bioactivity OR pharmaceutical OR 'bioactive compounds' OR policy OR conservation OR ecology OR 'animal assemblage' OR 'animal association'; to understand the history and trend of Indian octocoral research. To ensure maximum inclusion of Indian papers, an explicit time scale was not specified, and careful cross-references were also made to consider unpublished proceedings, theses, library records etc. Only those publications supported by empirical field data (quantitative field surveys, voucher specimens, photographs) on any one of the following topics: diversity, distribution, taxonomic works such as revisions, species/generic description, nomenclatural acts and focused on Asia, or any study involving octocorals in the case of India, were included. Only peer-reviewed journal articles were considered, to maintain both consistency and quality of data. We recorded the year and country of publishing, author names (national and international separately), nationality of non-Indian authors and the type and name of the journal. In addition, we also assessed the octocoral 'diversity and taxonomic' publications from over 22 Asian maritime countries/islands excluding India for the past 40 years (1978–2018) to compare geographical trends.

For this paper, a 'taxonomic expert' is defined using a slight modification of the broader definition of Convention on Biological Diversity (CBD), as 'a person with good expertise and extensive knowledge on octocorals who is/was active for 10 years or more and/or has published more than one taxonomic paper during the last four decades' (Haas & Haüser 2005). A 'peer-reviewed publication' is defined as one published in a journal indexed in either the Web of Science™, SCOPUS, or Google Scholar™ but excluding 'predatory open-access journals' (Bohannon 2013).

RESULTS AND DISCUSSION

a. Forty years of octocoral studies (taxonomy and diversity) in Asia

The resulting list (n=205) indicates that nearly 40% (n=78) of the published literature on octocoral diversity in Asia originates from the Far East (Japan, Taiwan, Hong Kong, Russia, Korea, and China) (Figure 1), with Japan contributing the greatest share (n=29) of publications, and the highest number of newly described species (n=29; from 10 description papers). Nearly 85% of the new species descriptions were carried out by foreign researchers (non-Asians/researchers not from their home country; n=33) followed by the combination of national and foreign researchers (n=18). The top 10 scientists/taxonomic experts (Asian or otherwise) together account for over 80% of the total number of species descriptions from the Asian waters (Figure 2). Thus, apart from Japan, Israel and Iran, the contribution of Asian researchers to octocoral taxonomy during a 40-year period (1978–2018) is proportionally low, indicating a shortfall of local taxonomic expertise in the region. Most species descriptions were made from countries along the Red Sea and in West Asia (n=77), followed by the Far East including the seas of Japan, Taiwan and Hong Kong (n=46). The least number of species were described from southeastern Asia (n=30) despite this being a region of high endemism and biodiversity, and from south Asia (n=11, including India n=8).

From 1978 to 2018, India recorded over 65 publications on octocoral diversity and distribution (including occurrence and distribution reports, taxonomy, and new records); of which only 28 appeared in peer-reviewed journals. The rest include books/book chapters/reports (n=23), posters/pre-prints/conference papers (n=4) and predatory or dubious publications (n=10). Though the higher number of papers is a result of many studies from Japan or other Asian countries with high numbers of octocoral studies (Taiwan, Singapore, and Indonesia), issues such as poor-quality publications and ambiguous diversity assessments have impeded the progress of octocoral studies in India (also see sections below). Also, despite the large number of publications, only five dealt with new species descriptions. Foreign authors were involved in all the (currently valid) species descriptions (n=8) from three publications, while those species described by Indian authors (n=57) from two publications show no records in either Zoobank or in WoRMS.

Costello et al. (2013a) noted an overall increase in the number of taxonomists (for all taxa) in Asia, but the data on octocorals do not reflect this. Nevertheless, discovering and naming new octocoral species alone will not solve the issue of biodiversity assessment and estimation for this group, since the majority of octocoral genera need extensive taxonomic studies (i.e., re-descriptions and revisions) (Daly et al. 2007). A positive trend of increasing numbers of young researchers working on taxonomy and systematics of octocorals points to an encouraging future for this field of research (Williams 2018).

b. History and trends in Indian octocoral research

Octocoral research in India dates to the late 19th Century, followed by 100 years of mostly exploratory research that resulted in publications on taxonomy, diversity and distributions. A critical review of 193

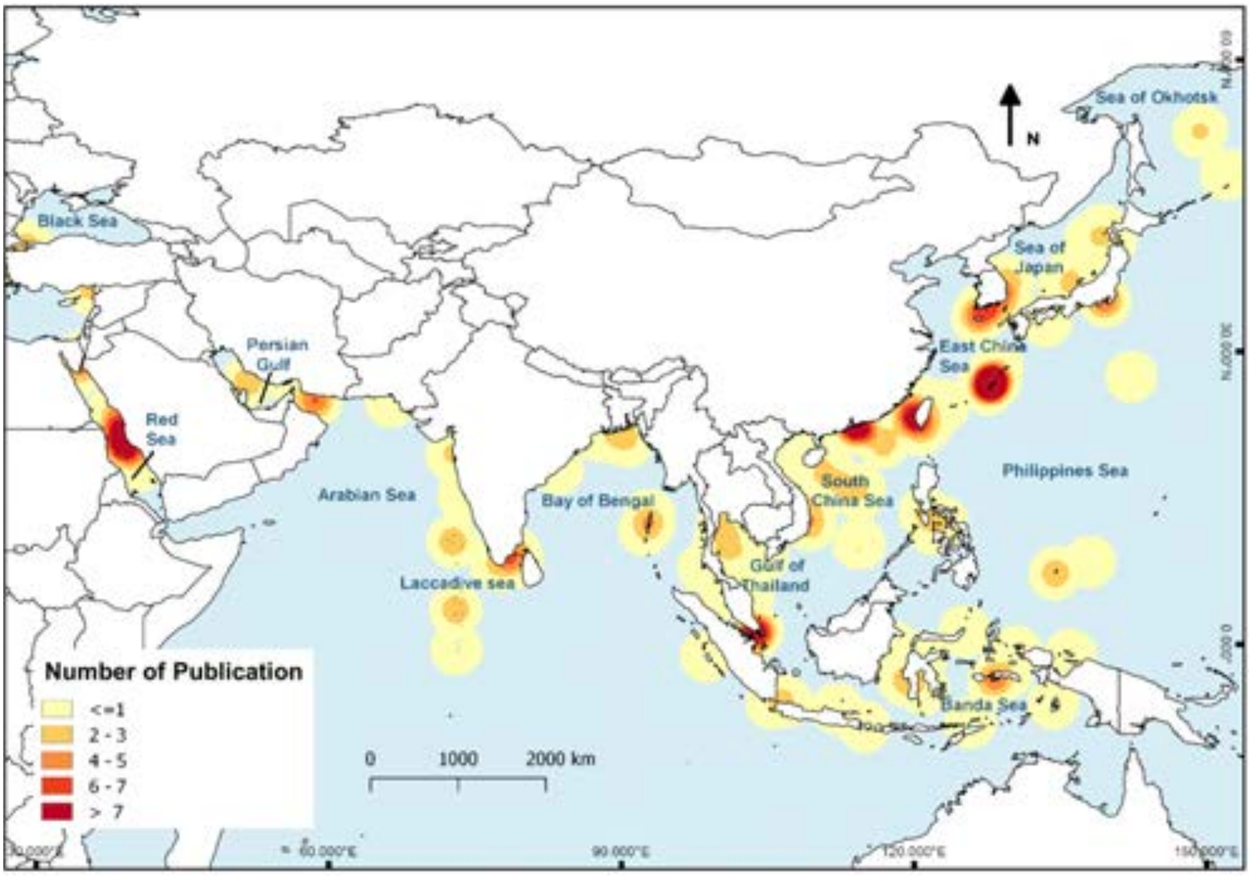

Figure 1. Geographical patterns of octocoral research in Asia from 1978 to 2018.

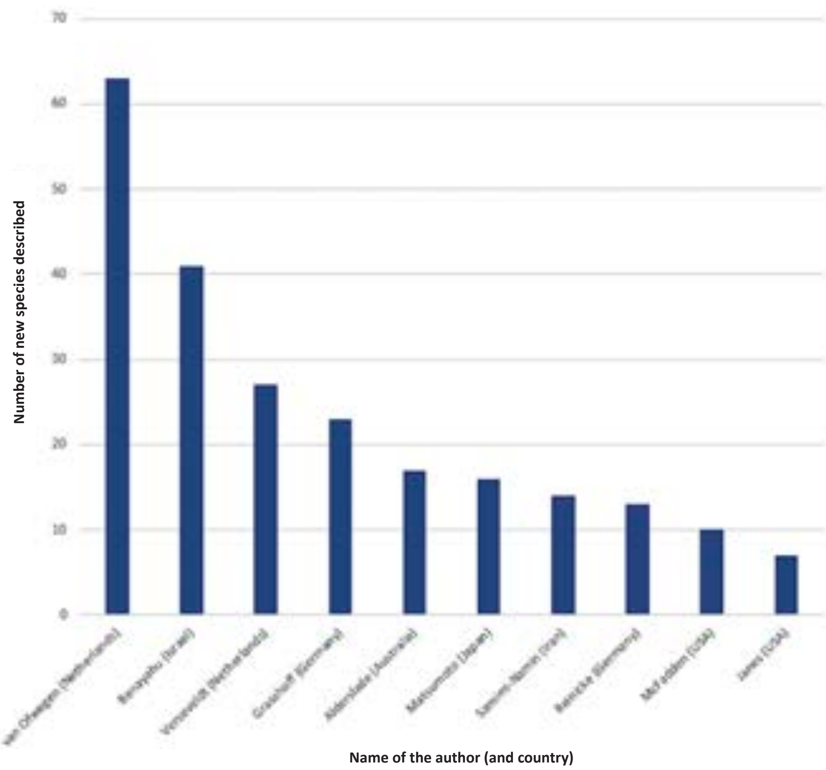

Figure 2. Contribution of top 10 authors in new species discoveries (Asian octocorals) from 1978 to 2018.

published and unpublished (e.g., theses, reports, newsletters, and posters) works on octocorals based on primary data/observations revealed that more than two-thirds have focused on 'taxonomy and diversity' and 'bioactivity' (Figure 3). Despite several publications on diversity and distribution of octocorals, taxonomic ambiguities and in several cases erroneous and unvalidated records of species have hampered the progress of octocoral research in India. Publications under the 'taxonomy and diversity' section are largely dominated by simple diversity and distribution (i.e., occurrence) studies, which in many cases are trivial and insignificant. Interestingly, the number of published 'taxonomic papers' (related to a taxonomic or nomenclatural act) is significantly less compared to those in 'taxonomy and diversity,' and the majority of such research from Indian waters was carried out by western researchers in the early 20th Century, as part of colonial natural history expeditions and investigations. Of this modest proportion of 'taxonomic papers', all but two involve foreign researchers, or a combination of both foreign and Indian researchers.

c. Publication trends in octocoral research in India – Quality vs Mediocrity

Good quality, peer-reviewed and publicly accessible biodiversity data can influence the reliability of communicating management and conservation policies and improve societal benefits (Costello et al. 2013b). Octocoral research in India has been scattered in several publication domains. While the majority of octocoral related publications are peer-reviewed (including those in journals), an equal number of mediocre publications in the form of grey literature and papers in predatory journals (as defined by Jeffrey Beall; see https://beallslist.weebly.com/) are a major concern for the advancement of octocoral research in India. The highest number of such publications have appeared recently (2000 to 2018), coinciding with the generally increased use of predatory journals by Indian scientists (see Raghavan et al. 2015). Since the year 2000, over 12 publications including those on diversity (checklist, distribution records), bioactivity and ecology have appeared in various predatory journals. Taxonomic research published after 1991 (except Williams & Vennam (2001)) has appeared mostly in predatory outlets or is in the form of mediocre publications circulated in single institutions/libraries, usually inaccessible to general public or academics, and in most cases containing invalid records. For instance, a monograph on gorgonians (Fernando 2011) has very limited circulation and most voucher specimens, including type material, is inaccessible to researchers (Ramvilas Ghosh pers. obs. 20.vii.2018), which contravenes the Recommendation 72F of the International Code of Zoological Nomenclature (ICZN). Similarly, a checklist on gorgonians by Kumar and Raghunathan in 2015, probably the only recent compilation of gorgonian fauna from India, has appeared in a predatory journal questioning the authenticity and quality of the data.

Scientific misconduct, in particular, plagiarism, has become a major menace in the Indian scientific and academic circles (see Raghavan et al. 2013; Amos 2014) and octocoral studies from the subcontinent are no different. "For example, it was noted that Rao & Devi's (2003) paper on the soft corals of the Andaman Islands is a blatant example of plagiarism. The authors describe over 50 species and illustrate 47 of these, each with a figure containing numerous drawings of sclerites,

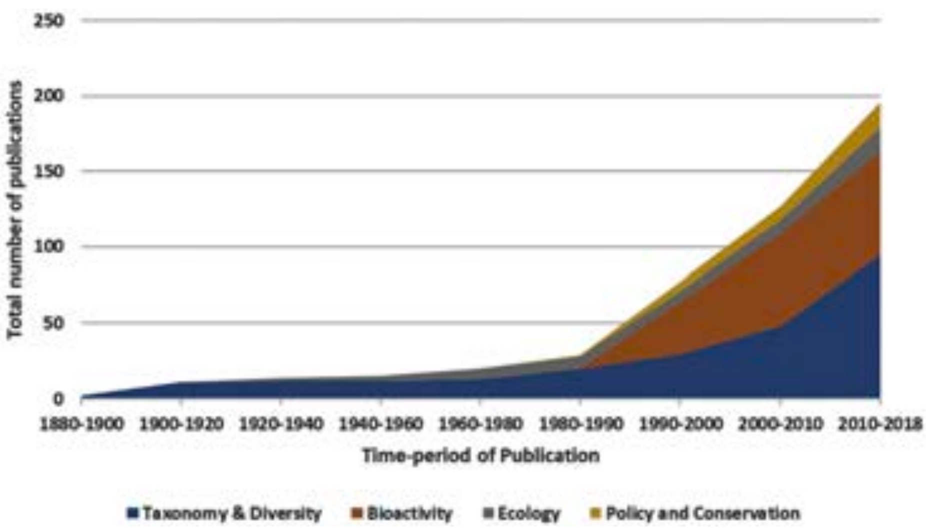

Figure 3. Historical shifts and bias associated with octocoral research in India.

with every single drawing hand-copied, with slight alterations, from the originals of Verseveldt (1980, 1982, 1983), primarily focused on his revisions of *Lobophytum*, *Sarcophyton* and *Sinularia*. Similarly, Rao & Devi's figures of *Lobophytum variatum* on page 34 are overtly copied from Verseveldt (1983)" (Phil Alderslade pers. comm. 09.i.2019).

d. Publication trends in octocoral research in India – Regional biases

Octocoral publications in India to date have been subject to regional inclinations, a trend that is similar to the whole of Asia. Much of the research focus has been on the southeastern coast (n=66), particularly in the Gulf of Mannar, and very little work has been carried out along the eastern coast of India (n=11). Between the island territories, higher numbers of publications have originated from the Andaman & Nicobar Islands (n=50), compared to Lakshadweep (n=16). The coastal and deeper waters off the southwestern, northwestern & eastern coasts, and the Lakshadweep Islands require extensive exploration and systematic taxonomic inventories to improve and contribute to the nation-wide understanding of octocoral diversity and distribution.

The absence of scientific institutions in some parts of the country (e.g., Lakshadweep) and the concentration of many institutions (both private and government) and museums in areas like the Gulf of Mannar could be the reason for the regional disparity in the studies of octocorals. But when considering cnidarian fauna in general, these regional disparities become very distinct for octocorals. For instance, comprehensive accounts on the cnidarian diversity are available for Scleractinia (Pillai 1991), Siphonophora (Daniel 1985), and Scyphomedusae (Chakrapany 1984). Also, inclusive data are available for hard corals (Scleractinia) from all major reef areas including mainland and the island territories (Venkataraman & Wafar 2005), so there must be another reason that octocorals have received less attention. Incidentally, scleractinian corals and coral dominated reefs receive much funding, and are considered of global significance due to their biodiversity and apparent vulnerability when compared to other marine ecosystems (Brooks et al. 2006).

We do not contend the fact that octocorals have never received scientific attention or research priority in India. In India, octocorals were heavily sought after for their bioactivity during the 1980s, however, most of the research was undertaken only to the level of extraction and chemical analysis, with no resulting industrial applications (Raveendran et al. 2011), which may have contributed (among other factors) to the current lack of interest in this fauna.

Even though regional accounts on the diversity of octocorals are available, most of them tend to be unreliable in terms of data-quality. "For instance, a paper on the octocorals from the Andaman & Nicobar Islands (Kumar et al. 2014) bases virtually all of the identifications on Grasshoff's (1999) monograph on the gorgonians of New Caledonia, which is a very popular book among Indian octocoral workers, as it has colored underwater images. But in the paper (Kumar et al. 2014), it is obvious that numerous colony pictures they present do not look like the actual species figured by Grasshoff (1999), and moreover as sclerites are an essential taxonomic character, the lack of illustrations renders it impossible for the readers to judge, and the authors to prove that the species are as claimed. Interestingly, since the publication of Grasshoff's (1999) monograph, many species previously considered to be endemic to New Caledonia have been recorded from the Andaman & Nicobar Islands - mostly dubious claims" (Phil Alderslade pers. comm. 09.i.2019).

e. The paradigm of species diversity, museums and specimens

In the context of taxonomic uncertainty, there is a high likelihood of underestimations or overestimations of Indian octocoral diversity especially in the case of gorgonians. Most gorgonian genera, and in particular *Junceella* and *Acanthomuricea* (reported from the 'Investigator' expedition) need considerable revision using an integrative approach and using modern molecular tools. With a limited amount of taxonomic expertise and capacity in India, the identity of many gorgonian species has been restricted to the generic level (Mary & Sluka 2014).

Author and date misnomers are yet another problem creating confusions in octocoral taxonomic data. For example, *Trimuricea reticulata* Gordon, 1926 mentioned in WoRMS (2019) and Global Biodiversity Information Facility (2019) should actually be *Trimuricea reticulata* (Thomson & Simpson, 1909) (see Samimi-Namin & van Ofwegen 2016). This type of outdated and obsolete information on species can be seen in several Indian checklists pertaining to octocorals. For example, both Venkataraman et al. (2004) and Thomas (1996) have used names for ellisellid genera (e.g., Gorgonella, Scirpearia) that have not been used by taxonomists for many decades. It is notable that regional checklists of octocorals from India (except Tudu et al. 2018 for sea pens) that contain outdated or erroneous records, are mostly published in poor-quality publications mostly without any rigorous peer-review, or in predatory journals.

Many specimens described from the Investigator expedition and currently housed in the invertebrate collections of the Zoological Survey of India (ZSI), Kolkata, "need re-examination and extensive re-evaluation (Phil Alderslade pers. comm. 09.i.2019). But there are enormous difficulties in accessing these specimens (see, for example, Samimi-Namin & van Ofwegen 2016), which reflects an appalling attitude of the regulating authorities. Issues regarding the difficulty, or even impossibility, of accessing these specimens has resonated around the global taxonomic community for numerous decades. "Indian biodiversity policies restrict the free exchange of specimens to overseas scientists and their institutions regardless of their reputation, and Indian scientists are also finding it increasingly difficult to access the museums of the Zoological Survey of India. Unless authorities change this dismaying situation and encourage international collaboration and allow Indian taxonomists the same kind of museum access that their overseas counterparts experience, genuine taxonomic research on Indian octocorals, and many other marine taxonomic groups, will continue to stagnate biodiversity documentation in India. This will also result in sub-standard and poorly compiled research reports as is occurring in the parallel case with scleractinian corals" (Phil Alderslade pers. comm. 09.i.2019).

Type material of many species collected from Indian waters by Indian and non-Indian expeditions (see examples in Verseveldt 1980; van Ofwegen 1990) are housed in foreign museums and accessing these types via loans is a 'kafkaesque' situation due to the National Biodiversity Authority (NBA) restrictions under the pretext of Biological Diversity Act (2002) and Biological Diversity Rules (2004) (see NBA, 2004). Thus, the lack of adequate taxonomic expertise, inaccessible types and voucher specimens at Indian museums and institutions, and expenses associated with visiting foreign museums where many types are housed, further delays the opportunity to rectify the many erroneous records in the Indian octocoral literature.

f. Opportunities and challenges

To a considerable extent, the issues pertaining to octocoral research discussed here can be solved through international and inter-institutional collaborations, a key strategy followed by countries like the United States of America which is a leader in global biodiversity documentation and research (Liu et al. 2011). As an example, in the case of octocoral-related taxonomic publications from Japan, Taiwan, Indonesia, and Israel (the largest contributors to such studies on Asian octocorals), international collaboration has not only enabled research results to be published in reputed journals, but also helped develop in-country capacity and taxonomic expertise supporting local researchers to document their octocoral diversity and independently publish their research results. Another critical impediment in many biodiversity-rich countries including India are the national regulations formulated under the pretext of the Convention on Biological Diversity (CBD), restricting biodiversity research of native scientists and discouraging international collaborations (Prathapan et al. 2018). As argued by Prathapan et al. (2018), there is no monopolistic situation in which a single country can identify all taxa, and none of the aims envisaged by CBD can be met unless scientists have access to the resources they wish to study and share with and involve the expertise of other countries. Similarly, Madhusudan et al. (2006) points out a distressing trend across India where researchers and scientists are refused entry into wildlife reserves (marine protected areas in this context), denying them opportunity to conduct scientific research that would actually inform the authorities what organisms inhabit these areas. Coupled with this, legislation like the Indian Wildlife Protection Act, 1972 (WPA 1972), prompts a poignant rhetoric in octocoral research. For example, in the case of gorgonians (protected under Schedule 1 of the Indian Wildlife Protection Act), the legislation has resulted in the restriction of sample collections and most exasperatingly the delay in getting research permissions to work on these taxa.

OPPORTUNITIES AND RECOMMENDATIONS

Given the above-mentioned issues and complexities in advancing the field of octocoral research in India, we suggest the following recommendations.

Convention on Biological Diversity (CBD), National legislations, and Research

Convention on Biological Diversity's Access and Benefit Sharing rules and the Nagoya Protocol (NP) obliges all committed parties including India, to develop necessary policies to foster equitable sharing of genetic resource and benefits arising from them (Buck & Hamilton 2011). But, despite several advancement in policies and management strategies, it is highly unlikely that the 'Aichi Biodiversity Targets' approved under the patronage of CBD can achieve much of an improvement in the state of biodiversity knowledge by 2020, particularly in the marine realm (Tittensor et al. 2014; Global Biodiversity Outlook 4 2014). The research community, undeniably the stakeholder most affected by the Nagoya Protocol, and CBD's Access and Benefit Sharing rules, are concerned by this state of affairs because Article 8(a) of Nagoya Protocol was formulated to 'promote and encourage research which contributes to the conservation and sustainable use of biological diversity, particularly in developing countries' (Buck & Hamilton 2011; CBD 2011). Therefore, to foster octocoral research in India, the restriction to the exchange of specimens for non-commercial, taxonomic and biodiversity research, arising due to national regimes under the misguided interpretation of CBD, should be objectively and urgently addressed. Perhaps placing a separate clause in the CBD accord to give special status to fundamental and non-commercial science, like taxonomy, for mutual exchange of data/specimens between institutions would allay the concerns of other stakeholders and reduce the complexity in undertaking biodiversity research (Prathapan et al. 2018). Similarly, national legislation like the Indian Wildlife Protection Act, 1972, and added amendments, which are meant to protect wild animals, should be made far less restrictive for octocorals as there is no viable commercial exploitation and the current situation hinders what little research is associated with them (e.g., all gorgonians). At present, the restriction limits sample collection and prohibits the exchange of specimens with foreign institutions and museums for the sake of taxonomic identification and archiving (WPA, 1972). We suggest, therefore, that the scheduled status of some octocorals especially in the case of gorgonians should be reconsidered, and improved conservation strategies like marine protected areas and 'no-take' zones be developed to protect this fauna, once research has been undertaken to determine where such areas would be best located.

Taxonomy and Quality Publication

As discussed in the relevant sections of this paper, octocoral research in India is beset by many mediocre publications which include works published in predatory journals. For instance, a recent checklist of octocorals in India (Kumar and others in 2018) was published in an outlet widely regarded as predatory, which perpetuates the trend of such unethical publishing practice among Indian researchers (see Raghavan et al. 2015; Patwardhan et al. 2018). These publishing companies masquerading under the pretext of an open access model continue to threaten science and science communication by narrowing the line between science and pseudoscience (Beall 2016). Since taxonomy and diversity research impacts national policies and influences other allied basic and applied research (Raghavan et al. 2014), flawed and mediocre publications pose serious impediment to India's international commitments like CBD's Aichi Targets. Because many mediocre publications in octocorals are from leading national research institutes like the Zoological Survey of India (ZSI), they reflect a bad image internationally which might result in blacklisting Indian taxonomists in general instead of just those deserving such a reputation. Scrapping Academic Performance Index (API) (Raghavan et al. 2015) and replacing traditional 'bibliometrics' with 'almetrics' to assess researcher's impact (Brown 2014) would render more popularity to science and reduce the unhealthy competition among researchers to publish more, causing some to resort to predatory or other sub-standard levels of publishing which lack peer review. Also, researchers and journals must avoid citing such dubious publications and thereby disavow unethical practice and unreliable research data. We also insist future octocoral taxonomist follow the modern trends in describing octocorals (Figure 4) and adhere to the rules of International Commission on Zoological Nomenclature (ICZN) (see Benayahu et al. 2017; Breedy & Guzman 2018).

Museums

Museums play a pivotal role in fundamental science like taxonomy and systematics through archiving and documenting specimens and manifesting a vast and irreplaceable resource for such studies (Brooke 2000). Many octocorals recorded as occurring in India have their type material housed in foreign museums. Physically accessing museum materials spread across the globe is not feasible in terms of money and time for a country like India. A realistic solution to overcome this issue is encouraging foreign collaboration, whereby researchers can gain experience and knowledge from international octocoral experts, benefitting both the researchers and octocoral science in India. At the same time, museums in India which house octocoral types (e.g., Zoological Survey of India) should change from being obstructive to acting as good advocates encouraging genuine requests to access specimens for verification and study. We also encourage these museums to digitally document and catalogue their octocoral specimens, both voucher specimens and types, and allow the information to be open to fair use for research nationally and internationally.

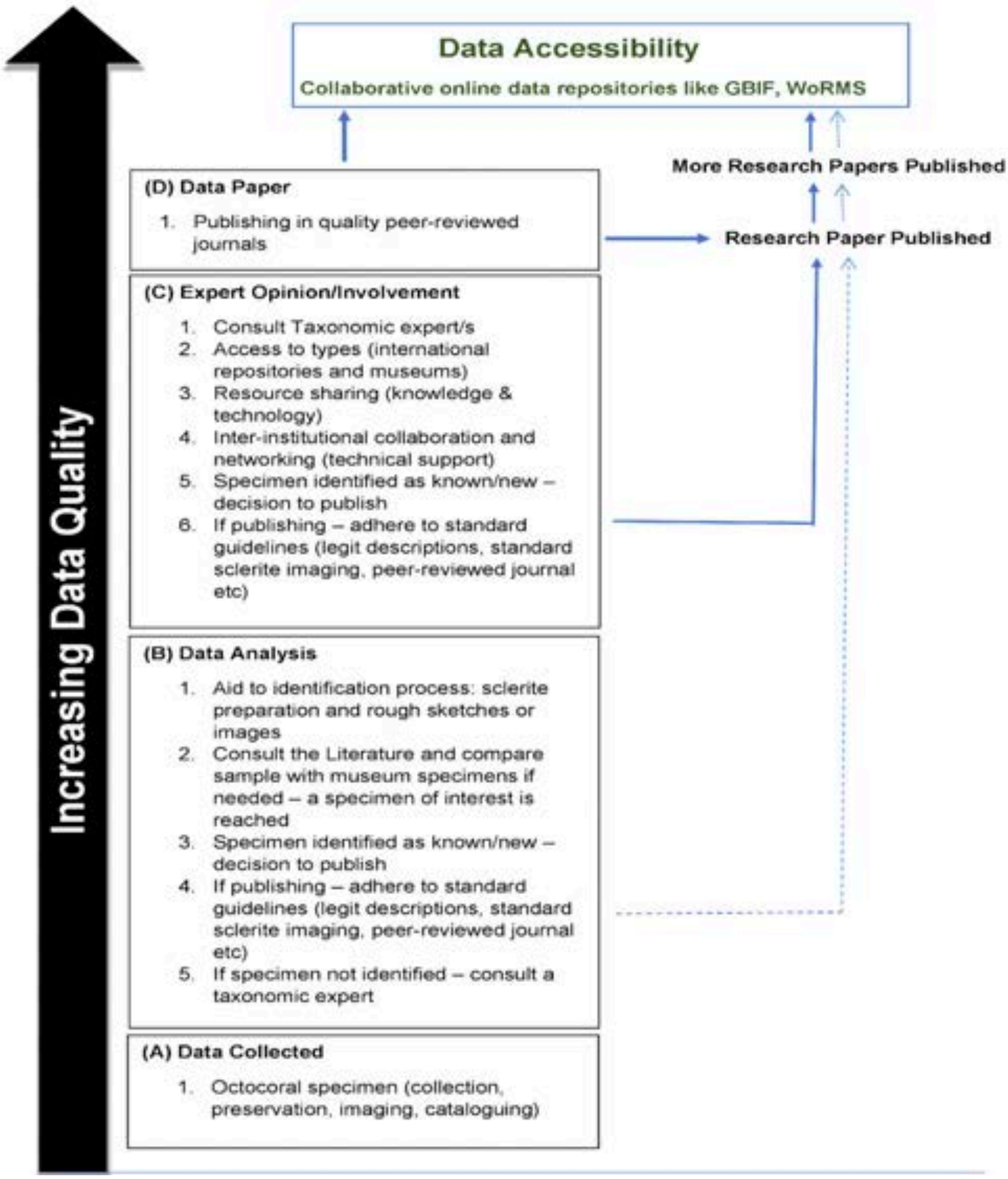

Figure 4. Conceptual model explaining the best practices to improve quality of octocoral diversity data and its accessibility. Adapted and modified from Costello et al. (2013b).

Bridging the biodiversity shortfalls

Based on available data it is evident that there exist huge gaps in data on octocorals, particularly related to the Linnaean (species diversity), Wallacean (geographic distribution) and Darwinian (evolution) shortfalls (see Hortal et al. 2015 for general discussion on biodiversity shortfalls). A consensus on the exact diversity and distribution range of many octocorals has not yet been reached as numerous records and data either remain incomplete or unreliable, particularly for the central Indian Ocean. Published literature also indicates that studies on evolution, phylogeny, biogeography, population genetics and abiotic tolerances of octocorals have not been undertaken as yet in countries such as India. For instance, the impasse in the case of the 'invasive snowflake coral' (*Carijoa riisei* Duchassaing & Michelotti, 1860) as a true invasive soft coral or a reestablished native species could be put to rest through genetic profiling (Patro et al. 2015). Such a study is currently being carried out by the authors of the present communication. Considering the expanse of the Indian subcontinent including the chains of islands and coral reefs, integrative taxonomic studies using morphology and molecular data (see Benayahu et al. 2018), research on evolution and phylogeny (see McFadden et al. 2017), including population genetics to study gene flow and connectivity (see Yesson et al. 2018) hold enormous potential. Not only are such studies inconspicuous in India, they are virtually non-existent because much of the basic biodiversity data (species diversity, abundance, and distribution) is unfortunately wanting, and will continue to be so unless there are significant changes to the culture and policies that are holding us back.

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Appendix 1.

	List of octocoral publications from Asia (1978–2018).
1	**Topçu, N.E. (2017).** Demographic structure of Gorgonian (Anthozoa, Octocorallia, Holaxonia) assemblages in the Bay of Saros. *KSÜ Journal of Natural Sciences* 20: 368–377.
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	List of relevant Indian publications on octocorals (1978–2018).
1	**Alderslade, P. & P. Shirwaiker (1991)**. New species of soft corals (Coelenterata: Octocorallia) from the Laccadive Archipelago. *Beagle: Records of the Museums and Art Galleries of the Northern Territory* 8:189–233.
2	**Bhagirathan, U., S.K. Panda, V.R. Madhu & B. Meenakumari (2008).** Occurrence of live Octocorals in the Trawling Grounds of Veraval Coast of Gujarat, Arabian Sea. *Turkish Journal of Fisheries and Aquatic Sciences* 8:369–372.
3	**Dhivya, P., V. Sachithanandam, P.M. Mohan (2012).** New record of *Carijoa riisei* at Wandoor–Mahatma Gandhi Marine National Park [MGMNP], Andaman and Nicobar Islands, India. *Indian Journal of Geo-Marine Sciences* 41: 212–214.
4	**George, A.M. (2008).** Abundance and community structure of reef building corals in Lakshadweep Islands, India. *Atoll Research Bulletin* 563:1–14
5	**George, R.M., N.K. Sanil & T.S. Naomi (2007).** A qualitative appraisal of the soft corals (Octocorallia: Alcyonacea) off Mandapam, South India. *Indian Journal of Fisheries* 54: 99–106.
6	**Jayasree, V., K.L. Bhat & A.H. Parulekar (1994).** *Sarcophyton andamanensis*, a new species of soft coral from Andaman Islands. *Journal of the Andaman Science Association* 10: 107–111.
7	**Jayasree, V., K.L. Bhatt & K.H. Parulekar (1996).** Occurrence and distribution of soft corals (Octocorallia:Alcyonacea) from the Andaman and Nicobar Islands. *Journal of the Bombay Natural History Society* 93: 202–209.
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9	**Kumar, J.S.Y., S. Geetha, V. Raghunathan & R. Sornaraj (2018).** A note on shallow water octocorallia from Nicobar Islands, India. *Indian Journal of Geo-Marine Sciences* 47: 60–66.
10	**Kumar, J.S.Y., S. Geetha, C. Raghunathan & R. Sornaraj (2018).** Diversity and distribution of gorgonians (Octocorallia) in the Long Island and it's adjacent areas in Middle Andaman, India. *Indian Journal of Geo-Marine Sciences* 47: 96–102
11	**Mary A.G. & S. Lazarus (2004).** Gorgonids off the southwest Coast of India. *Journal of the Marine Biological Association of India* 46: 32–43.
12	**Mary, A.G. & R.D. Sluka (2014).** Biodiversity and distribution of octocorals of Minicoy atoll, Lakshadweep. *Atoll Research Bulletin* 6: 1–17.
13	**Mary, A.G., R.D. Sluka & S. Lazarus (2011).** Octocoral diversity and distribution on the south–west Indian coast. *Marine Biodiversity Records* 4: 1–11.
14	**Rajendra, S., C. Raghunathan & K. Chandra (2017).** New record of *Sarcophyton cornispiculatum* Verseveldt, 1971 (Octocorallia: Alcyonacea: Alcyoniidae) in India, from the Andaman Islands. *European Journal of Zoological Research* 84: 167–171.
15	**Rajendra, S. C. Raghunathan & T. Mondal (2017).** New distribution records of three *Sarcophyton* species (Alcyonacea: Alcyoniidae) in Indian waters from Andaman Islands. *Journal of Threatened Taxa* 9: 10426–10432.
16	**Rajendra, S., C. Raghunathan, T. Mondal & K. Venkataraman (2017).** First report of soft coral *Sarcophyton birkelandi* Verseveldt, 1978 (Anthozoa: Alcyonacea) in Indian waters from Andaman Islands. *Journal of Threatened Taxa* 9: 10577–10580.
17	**Rajesh, S., K.D. Raj, G. Mathews, T. Sivaramakrishnan & J.K. Edward (2014).** Status of Alcyonacean corals along Tuticorin coast of Gulf of Mannar, Southeastern India. *Indian Journal of Geo-Marine Sciences* 43: 666–675.
18	**Thomas, P.A. & R.M. George (1987).** On five species of commercially important gorgonids new to Indian seas. *Indian Journal of Fisheries* 34: 20–27.
19	**Thomas, P.A., R.M. George & S. Lazarus (199**5). Distribution of gorgonids in the northeast coast of India with particular reference to *Heterogorgia flabellum* (Pallas). *Journal of the Marine Biological Association of India* 37: 134–142.
20	**Thomas, P.A., R.M. George, P.E.S. Manickam (1998).** Deep water Gorgonids collected aboard FORV SAGAR SAMPADA from the Southwest coast of India. *Journal of the Marine Biological Association of India* 40: 158–165.
21	**Thomas, P.A. & R.M. George (1986).** A systematic appraisal of the commercially important gorgonids of the Indian seas. *Journal of the Marine Biological Association of India* 28: 96–112.
22	**Tudu, P.C., D. Ray & A. Mohapatra (2018).** A Checklist of Indian Sea pen (Cnidaria: Anthozoa: Pennatulacea). *Indian Journal of Geo-Marine Sciences* 47: 1014–1017.
23	**van Ofwegen L.P. & J. Vennam (1991).** Notes on Octocorallia from the laccadives (SW India). *Zoologische Mededelingen* Leiden 65: 143–154.
24	**Veena, S. & P. Kaladharan (2012).** First record of *Cavernulina orientalis* (Thomson & Simpson, 1909) (Octocorallia: Pennatulacea: Veretillidae) from the Bay coast of Visakhapatnam, Andhra Pradesh. *Zootaxa*, 3204: 61–64.
25	**Veena, S. & P. Kaladharan (2013).** *Cavernularia obesa* from the bay coast of Visakhapatnam, Andhra Pradesh, India. *Marine Biodiversity Records* 6: e54.
26	**Vennam, J. & L.P. van Ofwegen (1996).** Soft corals (Coelenterata: Octocorallia: Alcyonacea) from the Laccadives (SW India), with a re–examination of *Sinularia gravis* Tixier–Durivault, 1970. *Zoologische Mededelingen,* Leiden 70:437–452
27	**Vibha, V.U., P.M. Mohan, M. Muruganantham & P. Nagarjuna (2015).** New record of *Cladiella pachyclados* (Klunzinger, 1877) at Port Blair, South Andaman, Andaman and Nicobar Islands, India. *Indian Journal of Geo-Marine Sciences* 44: 1787–1789.
28	**Williams, G.C. & J.S. Vennam (2001).** A revision of the Indo–West Pacific taxa of the gorgonian genus *Pseudopterogorgia* (Octocorallia: Gorgoniidae), with the description of a new species from western India. *Bulletin of the Biological Society of Washington* 10: 71–95.

Appendix 2.

	List of octocoral publications from India
1	**Kumaraguru, A.K., V.E. Joseph, M. Rajee & T. Blasubramanian (2008).** Palk Bay–Information and Bibliography, CAS in Marine Biology, Annamalai University, Parangipettai and Centre for Marine and Coastal Studies, Madurai Kamaraj University, Madurai, 227pp.
2	**Alderslade, P. & P. Shirwaiker (1991)**. New species of soft corals (Coelenterata: Octocorallia) from the Laccadive Archipelago. *Beagle: Records of the Museums and Art Galleries of the Northern Territory* 8: 189–233.
3	**Anjaneyulu ASR, Gowri PM, Murthy MVRK (1999).** New sesquiterpenoids from the soft coral *Sinularia intacta* of the Indian Ocean. *Journal of Natural Products* 62: 1600–1604.
4	**Anjaneyulu, A.S., V.L. Rao, V.G. Sastry & D.V. Rao (2008).** Trocheliophorin: A novel rearranged sesquiterpenoid from the Indian Ocean soft coral *Sarcophyton trocheliophorum*. *Journal of Asian Natural Products Research* 10: 597–601.
5	**Anjaneyulu, A.S.R. & K.V.S. Raju (1995).** Bioactive compounds of a new soft coral of the genus *Sinularia* of the Mandapam Coast. *Indian Journal of Chemistry - Section B Organic and Medicinal Chemistry* 34: 463–465.
6	**Anjaneyulu, A.S.R. & K.V.S. Raju (1995).** Secondary metabolites of a new soft coral of the genus *Sinularia* of the Mandapam coast. *Indian Journal of Chemistry - Section B Organic and Medicinal Chemistry* 34: 463–465.
7	**Anjaneyulu, A.S.R. & C.V.S. Prakash (1994).** Long chain Gamma-Lactone and Cyclopentanone derivatives from a sea pen of Virgularia genus of Indian Coast. *Indian Journal of Chemistry - Section B Organic and Medicinal Chemistry* 33: 55–58.
8	**Anjaneyulu, A.S.R. & C.V.S. Prakash (1995).** New sesqui and diterpenoids from the soft coral *Nephthea chabroli* of Indian coast. *Indian Journal of Chemistry - Section B Organic and Medicinal Chemistry* 34: 32–39.
9	**Anjaneyulu, A.S.R. & M.V.R.K. Murthy (2000).** Two new terpenoid derivatives from a new soft coral species of *Nephthea* genus of the Indian Ocean. *Indian Journal of Chemistry - Section B Organic and Medicinal Chemistry* 39: 42–56.
10	**Anjaneyulu, A.S.R. & N.S.K. Rao (1996).** Four new lobane diterpenoids from the soft coral *Lobophytum microlobulatum* of the Havellock island of the Andaman and Nicobar group of Islands. *Indian Journal of Chemistry* 35B: 1294–1303.
11	**Anjaneyulu, A.S.R., K.S. Sagar & G.V. Rao (1997).** New Cembranoid lactones from the Indian Ocean soft coral *Sinuiaria flexibilis. Journal of Natural Products* 60: 9–12.
12	**Anjaneyulu, A.S.R., C.V.S Prakash, U.V. Mallavadhani & K.V.S. Raju (1992).** Two new Sterols form a sea pen of *Virgularia* species of Indian Coast. *Journal of the Indian Chemical Society* 69: 150–152.
13	**Anjaneyulu, A.S.R., P.M. Gowri & M.V.R.K. Murthy (1998).** Dehydrosarcophytin, a new diterpenoid from the soft coral *Sacrophyton elegans* of the Indian Ocean. *Indian Journal of Chemistry - Section B Organic and Medicinal Chemistry* 38: 357–360.
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Appendix 3. Annotated list of gorgonians reported from Indian waters.

	Systematic position	Type locality	Current status as per WoRMS	Current distribution	EI	WI	A&N	LK
1	*Acanthogorgia breviflora* Whitelegge, 1897	Funafuti	Accepted	French Polynesia, Australia, New Caledonia, Tuvalu	0	0	1	0
2	*Acanthogorgia ceylonensis* Thomson & Henderson, 1905	Trincomalee	Accepted	Thailand, Oman, Indonesia, Sri Lanka	0	1	0	0
3	*Acanthogorgia muricata* Verrill, 1883	Barbados	Accepted	Indonesia, Barbados, Myanmar, Bahamas, Sri Lanka, Cape Guardafui	1	0	1	0
4	*Acanthogorgia spinosa* Hiles, 1899	Blanche bay (New Britain)	Accepted	New Caledonia, Indonesia, Myanmar, Australia, New Britain	0	0	1	0
5	*Acanthogorgia turgida* Nutting, 1911	Malay archipelago	Accepted	Indonesia	0	1	0	0
6	*Acanthomuricea arborea* (Thomson and Simson, 1909)	Arakan coast	No Records		1	0	0	0
7	*Annella mollis* (Nutting, 1910)	D'Aross	Accepted	Egypt, Japan, Palau, Philippines, Saudi Arabia, New Caledonia, Micronesia, Madagascar, Indonesia, Malaysia, Mauritius, Guam, Papua New Guniea, India, Northern Mariana Islands, Thailand, Fiji, El Salvador, Mayotte, Australia, Chinese Taipei, Marshall islands, American Samoa, Singapore	1	0	1	0
8	*Annella reticulata* (Ellis & Solander, 1786)	Philippines	Accepted	Philippines, New Caledonia, Australia, Saudi Arabia, Micronesia, Solomon Islands, Northern Mariana Islands, Palau, Indonesia, Mauritius, Papua New Guinea, Comoros, Tonga, Guam, Japan, Madagascar, Mayotte, Fiji, Thailand, Panama, Singapore, India	1	0	1	0
9	*Anthogorgia glomerata* Thomson & Simpson, 1909	Andamans	Accepted	Philippines, India	1	0	1	0
10	*Anthogorgia ochracea* Grasshoff, 1999	New Caledonia	Accepted	Vanuatu, New Caledonia	0	0	1	0
11	*Anthogorgia racemosa* Thomson & Simpson, 1909	Andamans	Accepted	India	1	0	1	0
12	*Anthogorgia verrilli* Thomson & Henderson, 1906	Andamans	Accepted	India	1	0	1	0
13	*Briareum hamrum* (Gohar, 1948)	Tumbatu, Zanzibar	Accepted	Israel, Oman, India	0	0	1	0
14	*Bebryce indica* Thomson, 1905	Gulf of Mannar	Accepted	Palau, Australia, Indonesia, Chinese Taipei, Sri Lanka,	1	0	0	0
15	*Bebryce sirene* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
16	*Bebryce studeri* Whitelegge, 1897	Funafuti	Accepted	French Polynesia, New Caledonia, Portugal, Indonesia, Philippines	1	0	1	0
17	*Bebryce thomsoni* Nutting, 1910	Kei islands (Indonesia)	Accepted	Indonesia	1	0	0	0
18	*Callogorgia versluysi* (Thomson, 1905)	Ceylon seas	Accepted	Palau, Sri Lanka, India	1	0	1	0
19	*Dichotella gemmacea* (Milne Edwards & Haime, 1857)	Red sea	Accepted	Australia, Philipines, Japan, New Caledonia, Fiji, Indonesia	0	0	1	0
20	*Discogorgia campanulifera* (Nutting, 1910)	Nusa Tenggara	Accepted	Indonesia	1	0	0	0
21	*Discogorgia squamata* (Nutting, 1910)	Indonesia?? *Placogorgia squamata??*	No records		1	0	0	0
22	*Echinogorgia complexa* Nutting, 1910	Papua	Accepted	Indonesia, India	1	0	0	0
23	*Echinogorgia macrospiculata* Thomson & Simpson, 1909	Andamans	Accepted	India	1	0	1	0
24	*Echinogorgia reticulata* (Esper, 1791)	Misrepresen-tation of date???	Accepted	Chinese Taipei, Australia, Japan, Pakistan, Indonesia, India, Mauritius, Amsterdam, Penguin channel	1	0	1	0
25	*Echinogorgia toombo* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	1	0	1	0
26	*Echinomuricea indica* Thomson & Simpson, 1909	Arakan coast	Accepted	Thailand, India	1	0	1	0
27	*Echinomuricea indomalaccensis* Ridley, 1884	Torres strait	Accepted	Australia, New Caledonia, Madagascar, Japan, Indonesia	1	0	1	0

	Systematic position	Type locality	Current status as per WoRMS	Current distribution	EI	WI	A&N	LK
28	*Ellisella andamanensis* (Nutting, 1910)	Maluku (Indonesia)	Accepted	Japan, New Caledonia, Fiji, Indonesia, India	1	0	1	0
29	*Ellisella azilia* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
30	*Ellisella cercidia* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
31	*Ellisella ceylonensis* (Simpson, 1910)	Galle (Sri Lanka)	Accepted	Sri Lanka, Indonesia, Japan, Papua New Guinea, Australia	1`	0	0	0
32	*Ellisella eustala* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
33	*Ellisella filiformis* (Toeplitz, 1889)	Probable misrepre-sentation of author	Accepted		1	0	0	0
34	*Ellisella maculata* (Studer, 1878)	Australia	Accepted	Australia, India, Indonesia	1	0	0	0
35	*Ellisella marisrubri* (Stiasny, 1938)	Red Sea	Accepted	Gulf of Suez (Red Sea)	0	0	1	0
36	*Ellisella nuctenea* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
37	*Euplexaura albida* Kükenthal, 1908	Australia	Accepted	Papua New Guinea, Australia	1	0	0	0
38	*Euplexaura amerea* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
39	*Euplexaura rhipidalis* Studer, 1895	Bintang Island	Accepted	New Caledonia, Japan, Indonesia, Carribbean sea, Burma	0	0	1	0
40	*Euplexaura thomsoni* Kükenthal, 1924		Accepted		1	0	0	0
41	*Guaiagorgia anas* Grasshoff & Alderslade, 1997	New Caledonia	Accepted	Australia, Papua New Guinea, New Caledonia, Indonesia	1	0	0	0
42	*Heliania spinescens* (Gray, 1859)	Philippines	Accepted	New Caledonia, Philippines, Fiji, Palau, Papua New Guinea, Vanuatu	1	0	0	0
43	*Heterogorgia flabellum* (Pallas, 1766)/*Psammogorgia flabellum* (Pallas, 1766)	Indian Ocean	Not Accepted	India	1	0	0	0
44	*Hicksonella princeps* Nutting, 1910	Sailus Besar	Accepted	Phillipines, Australia, Vanuatu, Malaysia, Japan, New Caledonia, Fiji, Indonesia,	0	0	1	0
45	*Isis hippuris* Linnaeus, 1758	North Sea	Accepted	Australia, Philippines, Papua New Guinea, Chinese Taipei, Malaysia, Japan, New Caledonia, Palau, Sri Lanka, Indonesia, India	0	0	1	0
46	*Junceella delicata* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
47	*Junceella eunicelloides* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
48	*Junceella juncea* (Pallas, 1766)	Indian Ocean	Accepted	Philippines, Australia, India, Micronesia, Northern Mariana Islands, Mauritius, Palau, Papua New Guinea, Vanuatu, Saudi Arabia, Comoros, Kiribati, Maldives, Singapore, China, New Caledonia, Japan , Myanmar, Madagascar, Indonesia, Vietnam, Sri Lanka, Taiwan	1	0	1	1
49	*Junceella miniacea* (Thompson & Henderson, 1906)	Andamans	No Records	Andamans	1	0	1	0
50	*Keroeides gracilis* Whitelegge, 1897	Funafuti	Accepted	New Caledonia, Australia, Nothern Mariana Islands, Philippines, Indonesia, India, Tuvalu, India	1	0	1	0
51	*Keroeides koreni* Wright & Studer, 1889	Japan	Accepted	Australia, Japan, Mayotte, Northern Mariana Islands, Kenya, Somalia, Marshall islands, Philippines, Indonesia, India, Japan, Sri Lanka	1	0	1	0
52	*Melithaea andamanensis* (van Ofwegen, 1987)	South Thailand	Accepted	Thailand, Andaman Sea	1	0	0	0
53	*Melithaea biserialis* (Kükenthal, 1908)	Red Sea	Accepted	Kenya, Madagascar	1	0	0	0
54	*Melithaea braueri* (Kükenthal, 1919)	Seychelles??	Accepted	Seychelles, Madagascar, India	0	0	1	0

	Systematic position	Type locality	Current status as per WoRMS	Current distribution	EI	WI	A&N	LK
55	*Melithaea caledonica* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
56	*Melithaea cinquemiglia* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
57	*Melithaea ochracea* (Linnaeus, 1758)		Accepted	New Caledonia, Papua New Guinea, Australia, Indonesia, Fiji, Japan, Palau,Philippines, Singapore, Malaysia, Sri Lanka, Vanuatu	0	0	1	0
58	*Melithaea ouvea* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
59	*Melithaea rubeola* (Wright & Studer, 1889)	Arafura Sea	Accepted	Philippines, Australia, Singapore, China, Malaysia, USA	1	0	1	0
60	*Melithaea squamata* (Nutting, 1911)	Timor	Accepted	Australia, Indonesia, Palau, Philippines	1	0	0	0
61	*Melithaea variabilis* (Hickson, 1905)	Maldives	Accepted	India, Mayotte, Marshall islands, French Southern Territories, British Indian Ocean Territories, Seychelles, Indonesia	0	0	1	1
62	*Menella flora* (Nutting, 1910)	Papua	Accepted	Indonesia, Egypt, India, China, New Guinea	1	0	1	0
63	*Menella indica* Gray, 1870	Back Bay (Bombay)	Accepted	Japan, India	0	0	1	0
64	*Menella kanisa* Grasshoff, 2000	Red Sea	Accepted	Eilat, Strait of Gubal, Sinai	0	0	1	0
65	*Menella kouare* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
66	*Menella praelonga* (Ridley, 1884)	Port Curtis	Accepted	Japan, Malaysia, Australia, Fiji	0	0	1	0
67	*Menella woodin* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
68	*Muricella complanata* Wright & Studer, 1889	Hyalonema Ground, Japan	Accepted	United States, Mosambique, Sri Lanka, Myanmar, India	1	0	1	0
69	*Muricella dubia* Nutting, 1910	Nusa Tenggara (Indonesia)	Accepted	Indonesia	0	1	0	0
70	*Muricella nitida* (Verrill, 1868)	Ebon island	Accepted	Japan	1	0	0	0
71	*Muricella paraplectana* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
72	*Muricella ramosa* Thomson & Henderson, 1905	Deep waters, GoM	Accepted	Australia, Thailand, South Africa, Japan, Oman, Indonesia, Sri Lanka, India	1	0	1	0
73	*Muricella rubra* Thomson, 1905	Ceylon seas	Accepted	Madagascar, Marshall islands, Mauritius, Sri Lanka, India	0	0	1	0
74	*Muricella umbraticoides* (Studer, 1878)	Gazelle (Australia)	Accepted	Indonesia, India	1	0	0	0
75	*Nicella carinata* Nutting, 1910	Duroa strait, Kei islands	Accepted	Australia, Palau, Japan, Philippines, Indonesia, Mauritius,	1	0	0	0
76	*Nicella dichotoma* (Gray, 1860)	Bombay	Accepted	Japan, Indonesia, Thailand, British Indian Ocean territory, India	1	0	1	0
77	*Nicella flabellata* (Whitelegge, 1897)	Funafuti	Accepted	New Caledonia, Thailand, India	1	0	1	0
78	*Nicella gemmacea* (Valenciennes, 1855)	Red Sea	No Records		1	0	0	0
79	*Nicella laevis* (Nutting, 1910)	Timer Island	No Records		1	0	0	0
80	*Nicella laxa* Whitelegge, 1897	Funafuti	Accepted	New Caledonia, Tuvalu	0	0	1	0
81	*Nicella magna Grasshoff, 1999*	New Caledonia	Accepted	New Caledonia, Vanuatu	1	0	0	0
82	*Nicella rubra* (Nutting, 1910)	Hyalonema Ground, Japan	No Records		1	0	0	0
83	*Paracis ceylonensis* (Thomson & Henderson, 1905)	Deep waters of Galle	Accepted	Thailand, Oman	1	0	0	0
84	*Paracis rigida* (Thomson & Simpson, 1909)	Andamans	Accepted		1	0	0	0
85	*Paracis spinosa* (Thomson & Henderson, 1906)	Andamans	Accepted		1	0	0	0
86	*Parisis fruticosa* Verrill, 1864	Sulu sea	Accepted	Australia, Palau, New Caledonia, India, Somalia, Thailand, Japan, Philippines, Niue, Madagascar, Indonesia, Mauritius, New Zealand, India	1	0	1	0

	Systematic position	Type locality	Current status as per WoRMS	Current distribution	EI	WI	A&N	LK
87	*Pseudopterogorgia australiensis* (Ridley, 1884)	Torres strait	Accepted	Australia	1	0	0	0
88	*Pseudopterogorgia formosa* (Nutting, 1910)	Nusa Tenggara (Indonesia)	Accepted	Indonesia	1	0	0	0
89	*Pseudopterogorgia fredericki* Williams & Vennam, 2001	St. Mary Isles, Bellikery, India	Accepted	India	1	0	0	0
90	*Pseudopterogorgia oppositipinna* (Ridley, 1888)	Mergui archipelago	Accepted	Australia	1	0	0	0
91	*Pseudopterogorgia rubrotincta* (Thomson & Henderson, 1905)	Indian Ocean	Accepted		1	0	0	0
92	*Pseudopterogorgia thomassini* (Tixier-Durivault, 1972)	Madagascar	Accepted	Madagascar	1	0	0	0
93	*Pterostenella plumatilis* (Milne Edwards and Haime, 1857)	Ceylon	Accepted	Philippines, Japan, Australia	1	0	0	0
94	*Rumphella aggregata* (Nutting, 1910)	Kei islands (Indonesia)	Accepted	Australia, Egypt, New Caledonia, Papua New Guinea, Vanuatu, Guam, Micronesia, Palau, Thailand, Indonesia, Yemen	0	0	1	0
95	*Rumphella torta* (Klunzinger, 1877)	Red Sea	No Records	India. Red sea	0	0	1	0
96	*Solenocaulon sterroclonium* Germanos, 1895	Ternate (Maluku)	Accepted	Australia, Indonesia	1	0	0	0
97	*Solenocaulon tortuosum* Gray, 1862	North Australia	Accepted	Australia, Singapore, Madagascar, Somalia, Maldives, India	1	0	0	0
98	*Subergorgia rubra* (Thomson, 1905)	Ceylon seas	Accepted	Thailand, Chinese Taipei, New Caledonia, Sri Lanka	1	0	1	0
99	*Subergorgia suberosa* (Pallas, 1766)	Sea of South Africa	Accepted	Philippines, Australia, Saudi Arabia, Chinese Taipei, Japan, Madagascar, Reunion, Micronesia, India, Palau, Japan, China, Malaysia, Mauritius, Papua New Guinea, Guam, Vanuatu, UAE, Indonesia, Thailand, Singapore, Marshall Islands, Fiji, New Caledonia, Tanzania, Christmas Islands	1	0	1	0
100	*Thesea flava* Nutting, 1910	Aru Islands	Accepted	Philippines, Indonesia, India	1	0	0	0
101	*Trimuricea caledonica* Grasshoff, 1999	New Caledonia	Accepted	New Caledonia	0	0	1	0
102	*Trimuricea reticulata* (Thomson & Simpson, 1909)	Trimuricea reticulata Gordon, 1926 - discrepancy in the author. GBIF shows both results but 0 occurrence for Trimuricea reticulata (Thomson & Simpson, 1909) - Refer Namin & Ofwegen 2016	Accepted	Myanmar, India	1	0	1	0
103	*Verrucella cerasina* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
104	*Verrucella corona* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
105	*Verrucella diadema* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	1	0	1	0
106	*Verrucella flexuosa* (Klunzinger, 1877)	Red Sea	Accepted	Japan, India	1	0	1	0
107	*Verrucella gubalensis* Grasshoff, 2000	Red Sea	Accepted		1	0	1	0
108	*Verrucella klunzingeri* Grasshoff, 2000	Eilat	Accepted	Red Sea	0	0	1	0
109	*Verrucella umbella* (Esper, 1798)	Foreign language..cant derive the type locality- Bay of Bengal??	Accepted	South Africa, Somalia, Thailand , Iran	1	0	1	0

	Systematic position	Type locality	Current status as per WoRMS	Current distribution	EI	WI	A&N	LK
110	*Verrucella umbraculum* (Ellis & Solander, 1786)	Batavia	Accepted	Papua New Guinea, Thailand, Singapore, Japan, Tanzania, Somalia, Pakistan, Philippines, South Africa, India	1	0	1	0
111	*Villogorgia ceylonensis* (Thomson & Henderson, 1905)	Deep waters of Galle	Accepted	Sri Lanka	1	0	1	0
112	*Villogorgia tenuis* (Nutting, 1908)	Hawaii	Accepted	United States of America, Somalia, Thailand	1	0	1	0
113	*Viminella crassa* (Grasshoff, 1999)	New Caledonia	Accepted	New Caledonia	0	0	1	0
114	*Viminella juncelloides* (Stiasny, 1938)	Red Sea	Accepted	India, Red Sea	0	0	1	0
115	*Acanthomuricea nagapatinamensis* Antony Fernando, 2011	Nagapattinam	No records	India	1	0	0	0
116	*Acanthogorgia cuddalorensis* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
117	*Acanthogorgia cylindricus* Antony Fernando, 2011	Cuddalore fish landing centre	No records	India	1	0	0	0
118	*Acanthogorgia delicata* Antony Fernando, 2011	Cuddalore fish landing centre	No records	India	1	0	0	0
119	*Acanthogorgia macrospiculata* Antony Fernando, 2011	Yerwadi beach	No records	India	1	0	0	0
120	*Acanthomuricea tuticorinensis* Antony Fernando, 2011	Tuticorin	No records	India	1	0	0	0
121	*Anthogorgia ramamoorthii* Antony Fernando, 2011	Cuddalore fish landing centre	No records	India	1	0	0	0
122	*Astrogorgia anastomosan* Antony Fernando, 2011	Pondiccherry	No records	India	1	0	0	0
123	*Astrogorgia bicolor* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
124	*Astrogorgia cuddalorensis* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
125	*Astrogorgia krusadaiensis* Antony Fernando, 2011	Vedalai	No records	India	1	0	0	0
126	*Astrogorgia macrosclera* Antony Fernando, 2011	Pondichery	No records	India	1	0	0	0
127	*Astrogorgia nagapainamensis* Antony Fernando, 2011	Nagapatina-mensis	No records	India	1	0	0	0
128	*Astrogorgia seshaiyaii* Antony Fernando, 2011	Vedalai	No records	India	1	0	0	0
129	*Astrogorgia sinensis* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
130	*Astrogorgia uvariensis* Antony Fernando, 2011	Uvari	No records	India	1	0	0	0
131	*Echinogorgia disimilis* Antony Fernando, 2011	Nagapatinam	No records	India	1	0	0	0
132	*Echinogorgia longispinosa* Antony Fernando, 2011	Pondicherry	No records	India	1	0	0	0
133	*Echinogorgia seshaiyaii* Antony Fernando, 2011	Pamban	No records	India	1	0	0	0
134	*Echinomuricea cuddalorensis* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
135	*Ellisella bayeri* Antony Fernando, 2011	Kanyakumari	No records	India	1	0	0	0
136	*Ellisella grasshoffi* Antony Fernando, 2011	Thiruchendur	No records	India	1	0	0	0
137	*Erythropodium pambanensis* Antony Fernando, 2011	Pamban	No records	India	1	0	0	0
138	*Euplexaura koothankuliensis* Antony Fernando, 2011	Koothankuli	No records	India	1	0	0	0
139	*Menella idinthakaraiensis* Antony Fernando, 2011	Idinthakarai	No records	India	1	0	0	0
140	*Nicella cuddlorensis* Antony Fernando, 2011		No records	India	1	0	0	0
141	*Nicella gracilis* Antony Fernando, 2011	Koothankuli	*Nicella gracilis* Cairns, 2007		1	0	0	0

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142	*Nicella rarus* Antony Fernando, 2011	bw Nagapattinam and Palayar	No records	India	1	0	0	0
143	*Paraplexaura mannarensis* Antony Fernando, 2011	Vedalai	No records	India	1	0	0	0
144	*Paraplexaura maxima* Antony Fernando, 2011	Pondicherry	No records	India	1	0	0	0
145	*Paraplexaura multiplanar* Antony Fernando, 2011	Vedalai	No records	India	1	0	0	0
146	*Paraplexaura platysclera* Antony Fernando, 2011		No records	India	1	0	0	0
147	*Pseudopterogorgia anastomosan* Antony Fernando, 2011	Vedalai, GoM	No records	India	1	0	0	0
148	*Pseudopterogorgia balasubramanii* Antony Fernando, 2011	Kothapatnam, Andhra	No records	India	1	0	0	0
149	*Pseudopterogorgia filiformis* Antony Fernando, 2011	Kodiakkarai, Palk Bay	No records	India	1	0	0	0
150	*Pseudopterogorgia flexibilis* Antony Fernando, 2011	Kodiakkarai, Palk Bay	No records	India	1	0	0	0
151	*Pseudopterogorgia kodiakaraiensis* Antony Fernando, 2011	Kodiakkarai, Palk Bay	No records	India	1	0	0	0
152	*Pseudopterogorgia kotapatnamensis* Antony Fernando, 2011	Kodiakkarai, Palk Bay	No records	India	1	0	0	0
153	*Pseudopterogorgia mandabamensis* Antony Fernando, 2011	Mandapam	No records	India	1	0	0	0
154	*Pseudopterogorgia mangalorensis* Antony Fernando, 2011	Surathkal	No records	India	0	1	0	0
155	*Pseudopterogorgia oliviae* Antony Fernando, 2011	Vedalai, GoM	No records	India	1	0	0	0
156	*Pseudopterogorgia pandiani* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
157	*Pseudopterogorgia philippi* Antony Fernando, 2011	Kodiakkarai, Palk Bay	No records	India	1	0	0	0
158	*Pseudopterogorgia rubra* Antony Fernando, 2011	Kodiakkarai, Palk Bay	No records	India	1	0	0	0
159	*Pseudopterogorgia undulata* Antony Fernando, 2011	Vedalai, GoM	No records	India	1	0	0	0
160	*Pseudopterogorgia vedalaiensis* Antony Fernando, 2011	Vedalai, GoM	No records	India	1	0	0	0
161	*Pseudopterogorgia williamsi* Antony Fernando, 2011	Vedalai, GoM	No records	India	1	0	0	0
162	*Trimuricea cuddalorensis* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
163	*Trimuricea indica* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
164	*Trimuricea longispinosa* Antony Fernando, 2011	Pondicherry	No records	India	1	0	0	0
165	*Trimuricea robusta* Antony Fernando, 2011	Uvari	No records	India	1	0	0	0
166	*Verrucella balasubramaniani* Antony Fernando, 2011	bw Cuddalore and Pondicherry	No records	India	1	0	0	0
167	*Verrucella bicolor* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
168	*Verrucella ixoboloides* Antony Fernando, 2011	bw Cuddalore and Pondicherry	No records	India	1	0	0	0
169	*Verrucella pambanensis* Antony Fernando, 2011	Pamban	No records	India	1	0	0	0
170	*Verrucella pinnata* Antony Fernando, 2011	Nagapattinam	No records	India	1	0	0	0
171	*Verrucella pondicheriensis* Antony Fernando, 2011	Pondicherry	No records	India	1	0	0	0

	Systematic position	Type locality	Current status as per WoRMS	Current distribution	EI	WI	A&N	LK
172	*Viminella dissimilis* Antony Fernando, 2011	Cuddalore	No records	India	1	0	0	0
173	*Acabaria cinquemiglia* Grasshoff, 1999 /*Melithaea cinquemiglia* (Grasshoff, 1999)	New Caledonia	Not accepted	New Caledonia, India	0	0	1	0
174	*Acabaria ouvea* Grasshoff, 1999/ *Melithaea ouvea* (Grasshoff, 1999)	New Caledonia	Not accepted	New Caledonia, India	0	0	1	0
175	*Acanella robusta* Thomson & Henderson, 1906	Indian Ocean	Accepted	India, Australia	0	0	1	0
176	*Astromuricea stellifera* Thomson & Crane, 1909	Kiu, Beyt Harbour	Accepted	India	0	0	1	0
177	*Bebryce mollis* Phillipi, 1842	Mediterranean Sea	Accepted	Italy, Morocco, Portugal, Spain, India, Maldives, Tunisia, Japan	0	0	1	0
178	*Cactogorgia alciformis* Thomson & Simpson, 1909	Andamans	No records	India, Andamans, Indonesia	0	0	0	1
179	*Calicogorgia tenuis* Thomson & Simpson, 1909	Andamans	Accepted	India	0	0	1	0
180	*Callogorgia indica* Versluys, 1906		Accepted	India	0	0	1	0
181	*Clathraria maldivenensis* van Ofwegen, 1987/*Melithaea maldivensis* (van Ofwegen, 1987)	Imma Island, Maldives	Not accepted	Maldives, India	0	0	0	1
182	*Echinogorgia flabellum* (Esper, 1791)	Maluku island	Accepted	Australia, Papua New Guinea, India	0	0	1	1
183	*Echinogorgia flora* (Nutting, 1910)/*Menella flora* (Nutting, 1910)				0	0	1	0
184	*Echinogorgia longispinosa*		No records		1	0	0	0
185	*Echinogorgia multispinosa* Thomson & Henderson, 1905	Ceylon seas	Accepted	Sri Lanka, India	0	0	1	0
186	*Echinogorgia ramulosa* (Gray, 1870)	Philippines	Accepted	Pakistan, India	0	0	1	0
187	*Echinomuricea andamanensis* Thomson & Simpson, 1909	Andamans	Accepted	India	0	0	1	0
188	*Echinomuricea indica* Thomson & Simpson, 1909	Arakan coast	Accepted	Thailand, India	0	0	1	0
189	*Echinomuricea ochracea* Thomson & Simpson, 1909	Indian Ocean	Accepted	India	0	0	1	0
190	*Echinomuricea splendens* Thomson & Simpson, 1909	Indian Ocean	Accepted	India	0	0	1	0
191	*Echinomuricea uliginosa* Thomson & Simpson, 1909	Lakshadweep	Accepted	Myanmar,India	0	0	1	0
192	*Echinomuricea uliginosa* Thomson & Simpson, 1909	Kalpeni Bank, Laccadives.	Accepted	India, Myanmar	0	0	1	1
193	*Echinomuricea uliginosa var. tenerior* Thomson & Simpson, 1909		No records		0	0	1	0
194	*Elasmogorgia flexilis* Hickson, 1905	Suvadiva	Accepted	Maldives, India	0	0	1	0
195	*Fanellia fraseri* (Hickson, 1915)	Gulf of Alaska	Accepted	United States, India	0	0	1	0
196	*Gorgonella rubra* (Thomson and Henderson, 1905)	Gulf of Mannar	No records	Indo-Australian	1	0	0	0
197	*Gorgonella umbella* (Esper, 1798)/Verrucella umbella (Esper, 1798)	Bay of Bengal	No records		1	0	0	0
198	*Gorgonella umbrculam* Ellis & Solander, 1786 /Verrucella umbraculum (Ellis & Solander, 1786)	Batavia	Not Accepted		1	0	1	0
199	*Keratoisis gracilis* (Thomson & Henderson, 1906)	Andamans	Accepted	Indo-Pacific	0	0	1	0
200	*Leptogorgia australiensis*(Ridley, 1884)/*Pseudopterogorgia australiensis* (Ridley, 1884)				1	0	0	0

	Systematic position	Type locality	Current status as per WoRMS	Current distribution	EI	WI	A&N	LK
201	*Lophogorgia lutkeni* Wright & Studer /*Leptogorgia lütkeni* (Wright & Studer, 1889)	Prince Edward Island	Not accepted	Prince Edward Island, Zanzibar, India	1	0	1	0
202	*Melithaea ornata* (Thomson & Simpson, 1909)	Andaman Sea	Accepted	No occurence in GBIF	0	0	1	0
203	*Melithaea philippinensis* (Wright & Studer, 1889)	Samboangan	Accepted	Indonesia, India	0	0	1	0
204	*Menacella gracilis* Thomson & Simpson, 1909	Andamans	Accepted	India	0	0	1	0
205	*Mopsella rubeola* (Wright & Studer, 1889)/*Melithaea rubeola* (Wright & Studer, 1889)	Arafura Sea	Not accepted	Australia, Philippines, Singapore, China, Indonesia, Malaysia, United States, India	1	0	1	0
206	*Muricella bengalensis* Thomson & Henderson, 1906	Bay of Bengal, Andamans	Accepted	India	0	0	1	0
207	*Muricella robusta* Thomson & Simpson, 1909	Andamans	Accepted	India	0	0	1	0
208	*Nicella pustulosa* (Thomson & Simpson, 1909)	Andamans	Accepted	India	0	0	1	0
209	*Nicella reticulata* Thomson & Simpson, 1909/Verrucella reticulata (Thomson & Simpson, 1909)	Indian Ocean	Not Accepted		0	0	0	1
210	*Paramuricea indica* Thomson & Henderson, 1906	Andamans	Accepted	India	0	0	1	0
211	*Perisceles ceylonensis (Thomson and Henderson)*		No records	Indian Ocean	1	0	0	0
212	*Placogorgia indica* Thomson & Henderson, 1906	Andamans	Accepted	India	0	0	1	0
213	*Placogorgia orientalis* Thomson & Henderson, 1906	Andamans	Accepted	India	0	0	1	0
214	*Plexauroides praelonga* (Ridley)		No records		1	0	0	0
215	*Plexauroides praelonga* (Ridley)		No records	Indo-Australian	1	0	0	0
216	*Plexauroides praelonga* var. cinerea (Ridley)		No records	Indo-Australian	1	0	0	0
217	*Scirpearia filliformia* Toeplitz		No records	Andamans, Australia	1	0	0	0
218	*Solenocaulon tortuosum* Gray, 1862	North Australia	Accepted	Singapore, Madagascar, Somalia, Australia, Maldives, India	1	0	1	0
219	*Subergorgia ornate* Whitelegge ??/ *Subergorgia ornata* Thomson and Simpson, 1909	Indian Ocean	Not accepted		0	0	0	1
220	*Subergorgia reticulata* Ellis & Solander, 1786/*Annella reticulata* (Ellis & Solander, 1786)	Philippines	Not accepted	Papua New Guinea, Comoros, Madagascar, Australia, Palau, Indonesia, Mauritius, Singapore, Guam, India, Northern Mariana Island	1	0	1	1
221	*Villogorgia rubra* Hiles, 1899	Ceylon seas	Accepted	Thailand, Indonesia, New Caledonia	0	0	1	0
222	*Wrightella braueri* Kükenthal, 1919 / *Melithaea braueri* (Kükenthal, 1919)	Seychelles??	Not accepted	Seychelles, Madagascar, India	0	0	1	0
223	*Pseudopterogorgia lutkeni* (Wright & Studer, 1889)		No records		1	0	0	0

EI—East coast of India | WI—West coast of India | A&N—Andaman & Nicobar Islands | LK—Lakshadweep Islands.

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SEM study of planktonic chlorophytes from the aquatic habitat of the Indian Sundarbans and their conservation status

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Abstract: Scanning electron microscopy (SEM) is the most modern technique for plankton research. The present paper deals with the taxonomy and morphology of some rare and endangered planktonic chlorophytes in relation to scanning electron microscopy. Water samples from the distinct water body of the Sundarbans have been concentrated and examined by scanning electron microscopy. A total of 45 species, of which 17 species of Scenedesmaceae, 10 species each of Hydrodictyaceae and Desmidiaceae, five species of Chlorococcaceae, two species of Selenastraceae and only one species of Chlorellaceae were recorded from the study site. Some species were recorded as new and rare from the study area. About 18 species including nine extremely rare, seven occasional, six frequent, four sporadic and one abundant was recorded in the present study. A detailed taxonomic description with line drawings is also included in the present communication.

Keywords: Conservation, morphology, plankton, scanning electron microscopy, Sundarban, taxonomy, water samples.

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Author contribution: Both GGS and RP conceived and designed the study concept, while GGS implemented monitoring and data collection in the field. GGS lead the writing of the manuscript with significant guidance and contribution from RP.

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INTRODUCTION

Planktonic chlorophytes play an important role in the aquatic ecosystem. They act as the primary producer of the aquatic food chain. Several reports have emphasized the importance of small planktonic chlorophytes in the aquatic habitat such as pond (Anand 1998), river, and sea (Vaulot et al. 2008). They also function as the progenitor of higher plants because of their photosynthetic pigments, starch as storage reserve food and chloroplast morphology (Lewis & McCourt 2004). They possess simple structural organization and reproduction. They also belong to unicellular and multicellular form.

The planktonic flora of the Indian Sundarbans was greatly diversified due to salinity gradient. Phytoplanktons play an important role in protecting the mangrove vegetation of the planet. In our earlier reports a few micro- and macro-chlorophytes were found from different habitats of the Indian Sundarbans (Satpati & Pal 2015, 2017). The habitats such as mud surface, tree bark, pneumatophore surface, stone surface, forest floor, water surface, and stilt root surface were surveyed for filamentous algae collection. A few reports were also available from other parts of the Sundarbans (Prain 1903; Naskar & Santra 1986; Sanyal & Bal 1986; Maity et al. 1987; Pal et al. 1988; Santra & Pal 1988; Santra et al. 1991; Mandal & Naskar 1994; Mukhopadhyay & Pal 2002). Changes in atmospheric temperature and global warming remarkably affect mangrove vegetation including micro- and macro-flora. The algal species are diminishing with changes in temperature and increased salinity. Micro- and pico-planktons are becoming rare and endangered and not functioning as primary producer of the ecosystem.

The present study was undertaken in order to identify some rare and endangered planktonic chlorophytes in different water bodies of the Indian Sundarbans. All taxa were documented on SEM and illustrations were also made for the same.

MATERIAL AND METHODS

Study area

The Sundarbans is the largest tiger inhabiting mangrove biosphere reserve in India and also a world heritage site designated by UNESCO. It is the largest chunk of mangrove ecosystem of the World encompassing many islands and rivers interconnected with creeks and canals. The deltaic appearance of the mangrove is formed by the confluence of three rivers, Ganges, Brahmaputra and Meghna in the Bay of Bengal. The major part of the Sundarbans (60%) lies in the Bangladesh and the remaining portion (40%) in India. The Hooghly River flows over India's state of West Bengal comprising mudflats, multiple tidal streams, open and closed mangrove systems. The Indian part of the Sundarbans is distinctive in terms of its vegetation, marine ecosystem and salinity. Continuous inundation of saline water into the fresh water ecosystem, greatly affect the floral diversity. The study area lies between 21.516–22.883 ^{0}N and 88.616–89.150 ^{0}E of the southeastern part of Bay of Bengal (Image 1).

Sampling site

The sampling site varies from fresh water to brackish water. A total number of 23 sites were studied in detail. All the sampling stations belong to the 24 Parganas (South and North) of the state of West Bengal, India. The name of the sampling sites and their physico-chemical parameters are given in Table 1.

Collection of samples

The phytoplanktons were collected from aquatic habitats during tidal action and also from the brackish water areas with the help of a truncated plankton net of 25µ mesh size. The samples thus collected were thoroughly washed with running tap water or saline water and then with double distilled water to remove soil particles and other debris. The sample material was then washed with phosphate buffer saline (PBS) 2–3 times and centrifuged at 8000rpm (Satpati & Pal 2017).

Measurement of Physicochemical Parameters

Physicochemical parameters like air and surface water temperature, pH, and salinity were recorded using digital thermometer (Eurolab ST9269B), pH meter (Eco testr) and Refractometer (Erma, Japan).

Preparation of voucher specimens

Samples were preserved in 4% (v/v) formalin and stored as voucher specimen in Calcutta University Herbarium (CUH) for further study.

Scanning Electron Microscopy (SEM)

One drop of washed material was put on a glass cover slip (Blue Star) and dried at 20°C. The samples were repeatedly washed with ethanol grade and dried at room temperature. After complete dehydration the cover slips were placed on carbon tape and put in Quorum (Q 150 TES) gold coater to coat the samples with gold. The scanning electron microscopic (SEM)

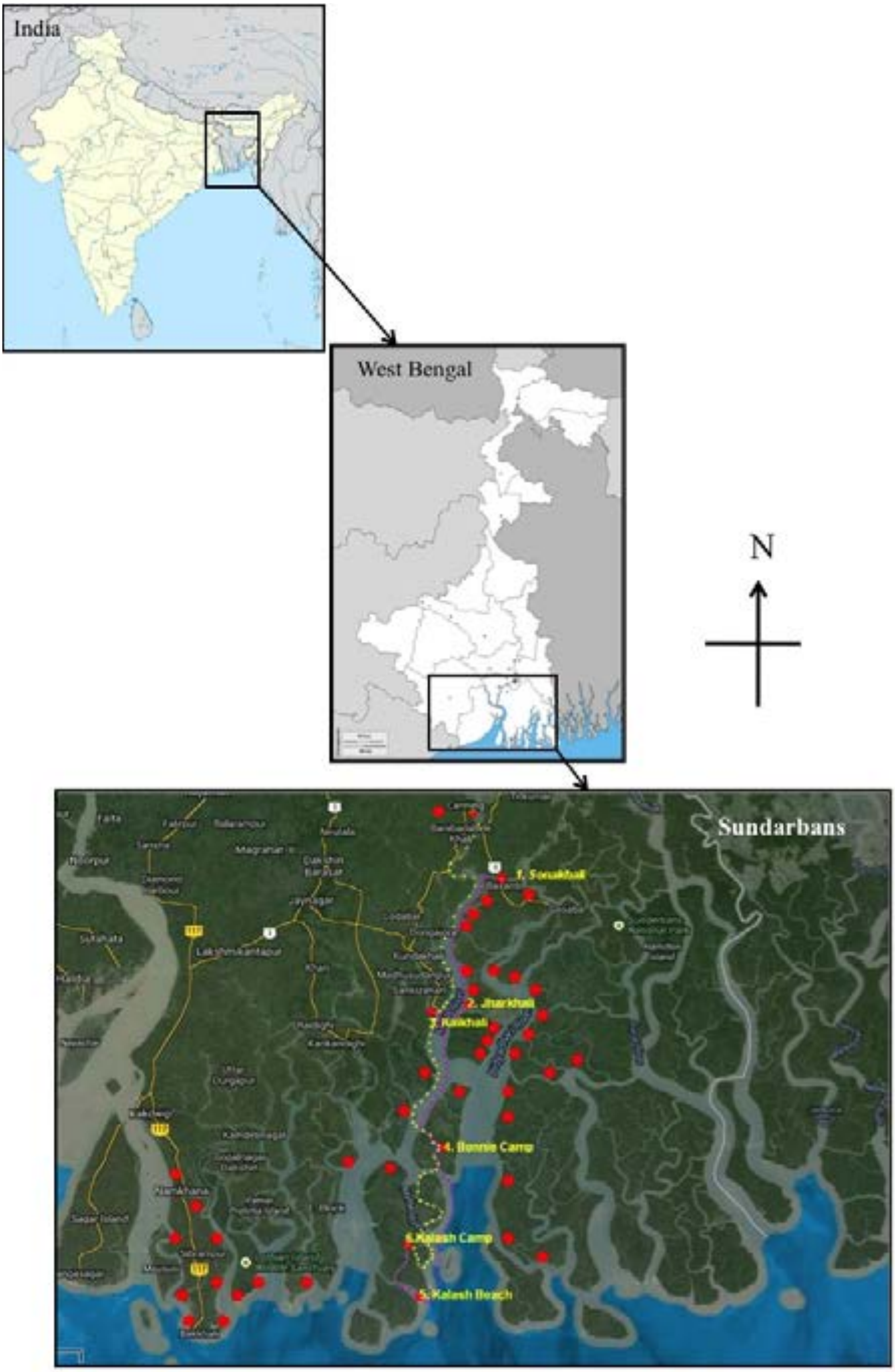

Image 1. Study area along different parts of Sundarbans (Google image, Red bullets indicating various sampling sites).

Table 1. Name of the sampling station and their physicochemical parameters.

	Sampling station	Physicochemical parameters			
		Air temperature (°C)	Surface water temperature (°C)	pH	Salinity (ppt)
1	Basanti	30.8	31.2	8.2	1.2
2	Jaigopalpur	31.3	31.7	7.8	1.5
3	Jharkhali	29.7	30.2	8.7	4.9
4	Rajbari	28.6	27.8	6.6	6.9
5	Malancha	27.4	26.5	6.2	7.4
6	Minakha	30.3	30.8	8.3	0.8
7	Canning	29.4	29.7	9.4	7.8
8	Bhagabatpur	27.6	28.2	9.6	3.4
9	Sandeshkhali	28.8	28	9.4	8.7
10	Namkhana	27.7	26.8	8.7	6.7
11	Fraserganj	28.6	28	8.6	11.6
12	Patibunia Island	27.6	27.4	9.8	12.3
13	Dabu	26.5	26.3	9.2	18.4
14	Hamanbere Island	27.5	26.8	9.6	12.5
15	Bakkhali	26.5	26	8.6	11.2
16	Sushni Island	30.5	31.7	8.4	22.1
17	Suryamoni Island	27.8	27.3	6.8	12.5
18	Kala Jungle	28.5	28.2	6.8	11.5
19	Morahero Island	27.5	27.2	9.3	12.2
20	Narayanitala	28.4	28.2	9.1	19.6
21	Cheramatla	30.2	31.2	8.7	19.6
22	Jammudwip	31.2	32	9.6	16.8
23	Aamarboni Island	28.8	28	9.2	15.2

images have been taken with Carl Zeiss EVO 18 (EDS 8100) microscope and Zeiss Inca Penta FETX 3 (Oxford instruments) attachment. Photographs were taken in different magnifications.

Camera Lucida drawing

The hand drawing was made under a compound microscope with the help of a prism and 0.1- and 0.2- Rotring isograph pen (Germany). The drawing was done on transparent tracing paper (A4 size). The cellular details of vegetative and reproductive parts of different species were outlined and scale measurement was given under 10X, 40X and 100X objectives with proper magnification. The cell length and breadth was measured with ocular lens as ocular division (O.D.) and standardized using stage micrometer.

Species identification

Identification of taxa was done using standard research articles and monographs (Smith 1950; Randhawa 1959; Prescott 1982; Anand 1998; Jaiswal & Tiwari 2003; Sen & Naskar 2003; Shukla et al. 2008; Bellinger & Sigee 2010; Das & Adhikary 2012; Tripathi et al. 2012; van Geest & Coesel 2012; Baruah et al. 2013; Keshri & Mallick 2013).

RESULTS

Taxonomic descriptions

A total of 45 species were identified and detailed descriptions of the species are enumerated below:

Family: Hydrodictyaceae

1. *Pseudopediastrum boryanum* (Turpin) E. Hegewald (Image 2A-B and Figure 1A)

Synonym: *Pediastrum boryanum* (Turpin) Meneghini

Basionym: *Helierella boryana* Turpin

[Prescott 1982; Day et al. 1995; Hu & Wei 2006; Kim & Kim 2012].

Coenobium entire; cells 5–6 sided with smooth or granular walls; peripheral cells with outer margins extended into two blunt-tipped processes, cells up to 14µ in diameter and 21µ long; 36 celled colony 85–90 µ wide. Cells are well ornamented with pores and wavy margins.

Occurrence: aquatic; voucher number: CUH/Al/MW-171.

2. *Pseudopediastrum boryanum* var. *perforatum* (Raciborski) Nitardy (Image 2C and Figure 1B)

Basionym: *P. boryanum* subsp. *perforatum* Raciborski

[Cambra-Sánchez et al. 1998; Kim & Kim 2012].

Coenobia are circular in outline and with well perforations. Coenobia composed of 4–32 cells. Incisions are wide and V-shaped. Each cell extended to two processes. Cell wall ultrastructure is very distinctly granular having honey comb like appearance. Diameter of the coenobia is 70–120 µ, cells 3–20 µ wide and 4–20 µ long.

Occurrence: aquatic; voucher number: CUH/Al/MW-172.

3. *Stauridium tetras* (Ehrenberg) Hegewald var. *apiculatum* (Fritsch) Keshri et Mallick comb. nov. (Image 2E and Figure 1C)

Synonym: *Pediastrum tetras* (Ehrenberg) Ralfs var. *apiculatum* Fritsch.

[Keshri & Mallick 2013].

Coenobia 4-celled, less rectangular with cells without intercellular spaces; marginal cells divided into two lobes with deep linear to cuneate incision on the outer side reaching the middle of the cell and are trapezoidal in shape; each lobe further divided into two lobes terminating in an apical nodular thickening; cells 5–15 µ in diameter and colony of four cells up to 14–28 µ in diameter. Cell wall ultrastructure varies being irregular net-like or warty.

Occurrence: aquatic; voucher number: CUH/Al/MW-199.

4. *Pediastrum obtusum* Lucks (Image 2F and Figure 1D)

[Prescott 1982; Kim & Kim 2012].

Coenobia nearly entire, with minute interstices formed by the retuse margins; Coenobia oblong to nearly star shaped. Coenobia 8–32 celled with deep narrow sinus forming two major lobes, lobes incised to form bluntly rounded lobules. The sinus outwardly closed due to the contact of two central lobules. The ultrastructure of cell wall shows dotted appearance having minute pores.

Cells are 5–10 µ in diameter and 6–12 µ in length. Coenobia 15–40 µ in diameter.

Occurrence: aquatic; voucher number: CUH/Al/MW-200.

5. *Pediastrum duplex* Meyen (Image 2G and Figure 1F)

Synonym: *P. napoleonis* Ralfs; *P. pertusum* Kützing; *P. duplex* var. *reticulatum* Lagerheim

[Bruhl & Biswas 1926; Prescott 1982; Day et al. 1995; Buchheim et al. 2005; Hu & Wei 2006; Kim & Kim 2012].

Coenobia 16-celled, arranged more or less compactly, semicircular in outline. The outer margin is smooth, concave and extended into two blunt tapering processes. Cells 10–15 µ in diameter and coenobia are 40–80 µ in diameter. The ultrastructure of cell wall is smooth with tiny pores.

Occurrence: aquatic; voucher number: CUH/Al/MW-201.

6. *Pediastrum araneosum* (Raciborski) Raciborski (Image 2H and Figure 1G)

Synonym: *P. angulosum* Ehrenberg ex Meneghini

[Prescott 1982; Day et al. 1995; John & Tsarenko 2002].

Coenobia entire with minute interstices. Central cells packed and peripheral cells with two minute lobes; margin concave between two lobes. Cell wall smooth and with reticulate ridges. The ultrastructure of cell wall shows tiny pores. Cells are 8–12 µ in diameter.

Occurrence: aquatic; voucher number: CUH/Al/MW-173.

7. *Pediastrum integrum* Nägeli (Image 2I and Figure 1E)

[Prescott 1982; McManus & Lewis 2005; Hu & Wei 2006; Tsarenko 2011; Kim & Kim 2012].

Coenobia 4, 8, 16 and 32 celled, without or little perforations. Cell wall reticulates with tiny granules. Shapes of the inner cells are similar to the peripheral cells. Outer margins of the peripheral cells with two truncate short processes. The tip of the processes is unequal. Coenobia 14–18 µ in diameter and cells are 4–8 µ in diameter.

Occurrence: aquatic; voucher number: CUH/Al/MW-174.

8. *Parapediastrum biradiatum* (Meyen) E. Hegewald (Image 2J and Figure 1H)

Basionym: *Pediastrum biradiatum* Meyen

[Prescott 1982; Menezes 2010; McManus & Lewis 2011; Tsarenko 2011].

Coenobia perforated, 16-celled; peripheral cells

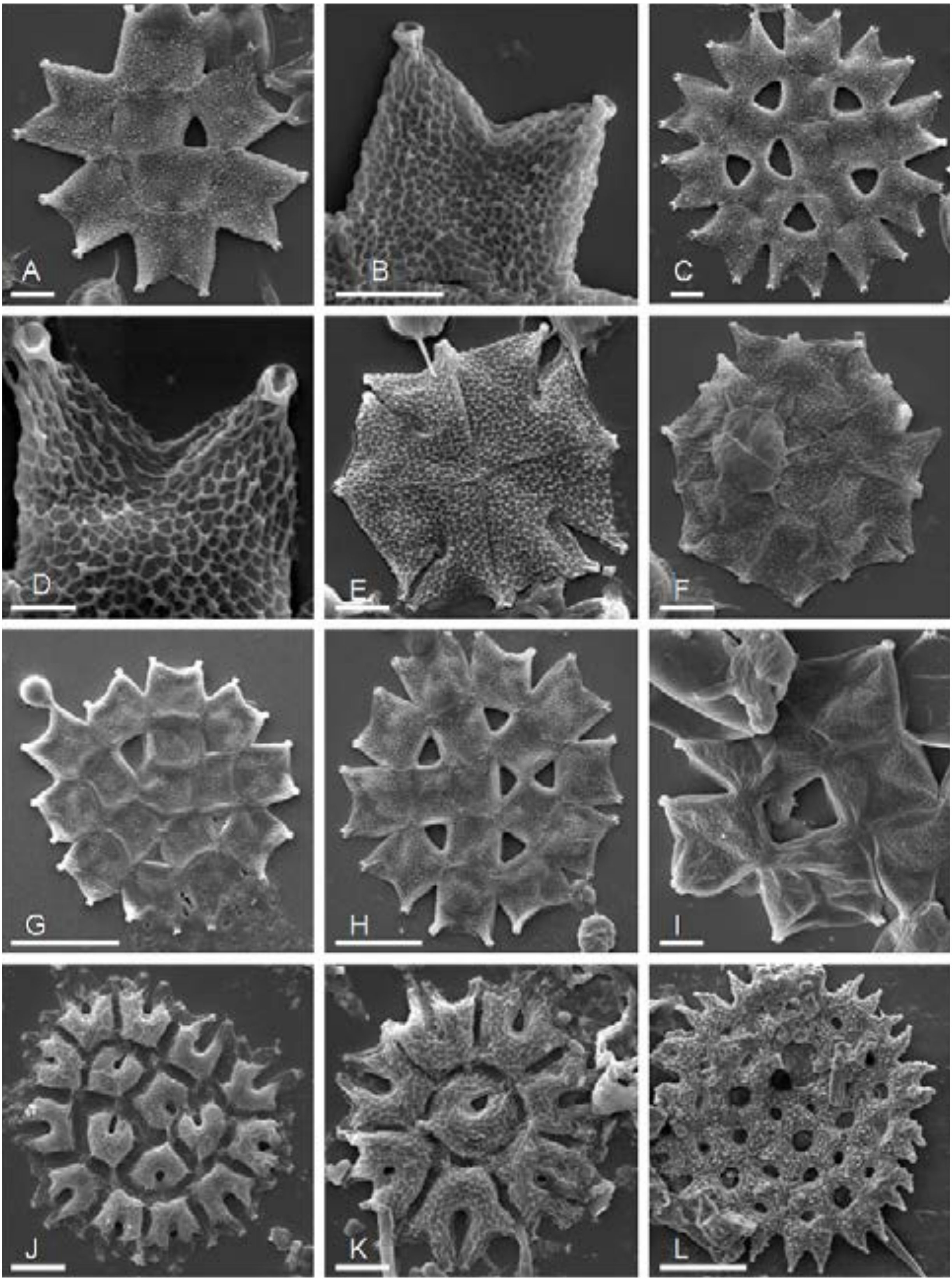

Image 2A–L. Scanning electron micrographs: A–B—*Pseudopediastrum boryanum* (×6.00KX and ×25.00KX) | C–D—*P. boryanum* var. *perforatum* (×6.00KX and ×25.00KX) | E—*Pediastrum tetras* var. *apiculatum* (×10.00KX) | F—*P. obtuosum* (×8.00KX) | G—*P. duplex* (×2.00KX) | H—*P. arneosum* (×4.25KX) | I—*P. integram* (×5.00KX) | J—*Parapediastrum biradiatum* (×7.50KX) | K—*Stauridium tetras* (×7.50KX) | L—*Pediastrum duplex* var. *duplex* (×1.75KX). Scale bar: A–B—3µ | C–L—2µ.

deeply bilobed, the lobes incised. Each cell is bilobed two times. Coenobia 12–16 µ in diameter; cells are 2–6 µ in diameter. The cell wall is reticulate and tiny pores are present on it.

Occurrence: aquatic; voucher number: CUH/Al/MW-175.

9. *Stauridium tetras* (Ehrenberg) E. Hegewald (Image 2K and Figure 1I)

Synonym: *Pediastrum tetras* (Ehrenberg) Ralfs; *Helierella renicarpa* Turpin; *Stauridium bicuspidatum* Corda

Basionym: *Micrasterias tetras* Ehrenberg

[Bruhl & Biswas 1926; Prescott 1982; Tsarenko 2011].

Coenobia oval or circular, 8-celled, marginal cells are deeply incised and form two lobes, each lobes truncate, generally further divided into two lobes and are trapezoidal in shape, inner cells 4–6 sided with a single linear or cuneate incision; cells 5–8 µ in diameter, eight celled colonies 12–18 µ in diameter. The ultrastructure of cells and coenobia shows presence of granules throughout. The cell surface is well wrinkled and folded.

Occurrence: aquatic; voucher number: CUH/Al/MW-81.

10. *Pediastrum duplex* (Meyen) var. *duplex* (Image 2L and Figure 1J)

[Prescott 1982; Anand 1998].

Coenobia circular, 40–70 µm in diameter; 16-32-64 celled. The peripheral cells are deeply incised to form V-shaped processes. The central and peripheral cells are of different sizes. The central cells are 5–8 µ in diameter and marginal cells are 8–12 µ in diameter. The central cells joined with each other and leave fine gaps within the coenobia. The ultrastructure of coenobia shows fine sculpture and granules throughout the cell wall.

Occurrence: aquatic; voucher number: CUH/Al/MW-202.

11. *Desmodesmus abundans* var. *brevicauda* G.M. Smith (Image 3A-B and Figure 1K–L)

[Menezes 2010; Tsarenko 2011; Tsarenko & John 2011; Gopalakrishnan et al. 2014].

Coenobium composed of four cells, cells smaller with relatively smaller spines. Cells are 2–4 µ in length and 1–2 µ in diameter. Spines fewer, 1–3 µ long. The ultrastructure of the cells shows smooth cell wall with slightly wavy margins.

Occurrence: aquatic; voucher number: CUH/Al/MW-187.

12. *Desmodesmus bicaudatus* (Dedusenko) P.M. Tsarenko (Image 3C and Figure 1M)

Basionym: *Scenedesmus bicaudatus* (Dedusenko)

[Tsarenko 2011; Tsarenko & John 2011].

Coenobium 2–4 celled, with linear or slightly alternate in arrangement, cells elongated, outer cells with a long curved spine at alternate poles; inner cells without spines, oval to cylindrical. Cells are 8–12 µ in length and 4–8 µ in width. The electron microscopic study revealed folded sculptured wall outside the cell with fine pores.

Occurrence: aquatic; voucher number: CUH/Al/MW-186.

13. *Desmodesmus serratus* (Corda) S.S. An, T. Friedl & E. Hegewald (Image 3D and Figure 2B)

Synonym: *Scenedesmus serratus* (Corda) Bohlin

Basionym: *Arthrodesmus serratus* Corda

[Prescott 1982; Fawley et al. 2011; Tsarenko 2011; Tsarenko & John 2011].

Coenobia composed of four ovate, oblong cells arranged in a single series; the outer and inner cells with longitudinal teeth; apices of all cells bearing 3–4 small teeth. Cells are 6-10 µ in length and 2–4 µ in width. The electron microscopic study revealed presence of beads like structure throughout the cell wall.

Occurrence: aquatic; voucher number: CUH/Al/MW-165.

14. *Desmodesmus armatus* (R. Chodat) E. Hegewald (Image 3E and Figure 2D)

Synonym: *Scenedesmus armatus* (Chodat) G.M. Smith

Basionym: *Scenedesmus hystrix* var. *armatus* R. Chodat

[Prescott 1982; Verschoor et al. 2004; Matusiak-Mikulin et al. 2006; Tsarenko 2011; Tsarenko & John 2011].

Coenobia composed of four cells and arranged in single series. Cells are elongated, ellipsoid; each cell with abundant uneven spines; each pole of the individual cell contains 3–6 uneven spines; each cell contains a longitudinal ridge. Cells are 4–8 µ in length and 3–5 µ in width. The sculptured cell wall with folded margin and granules are shown under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-166.

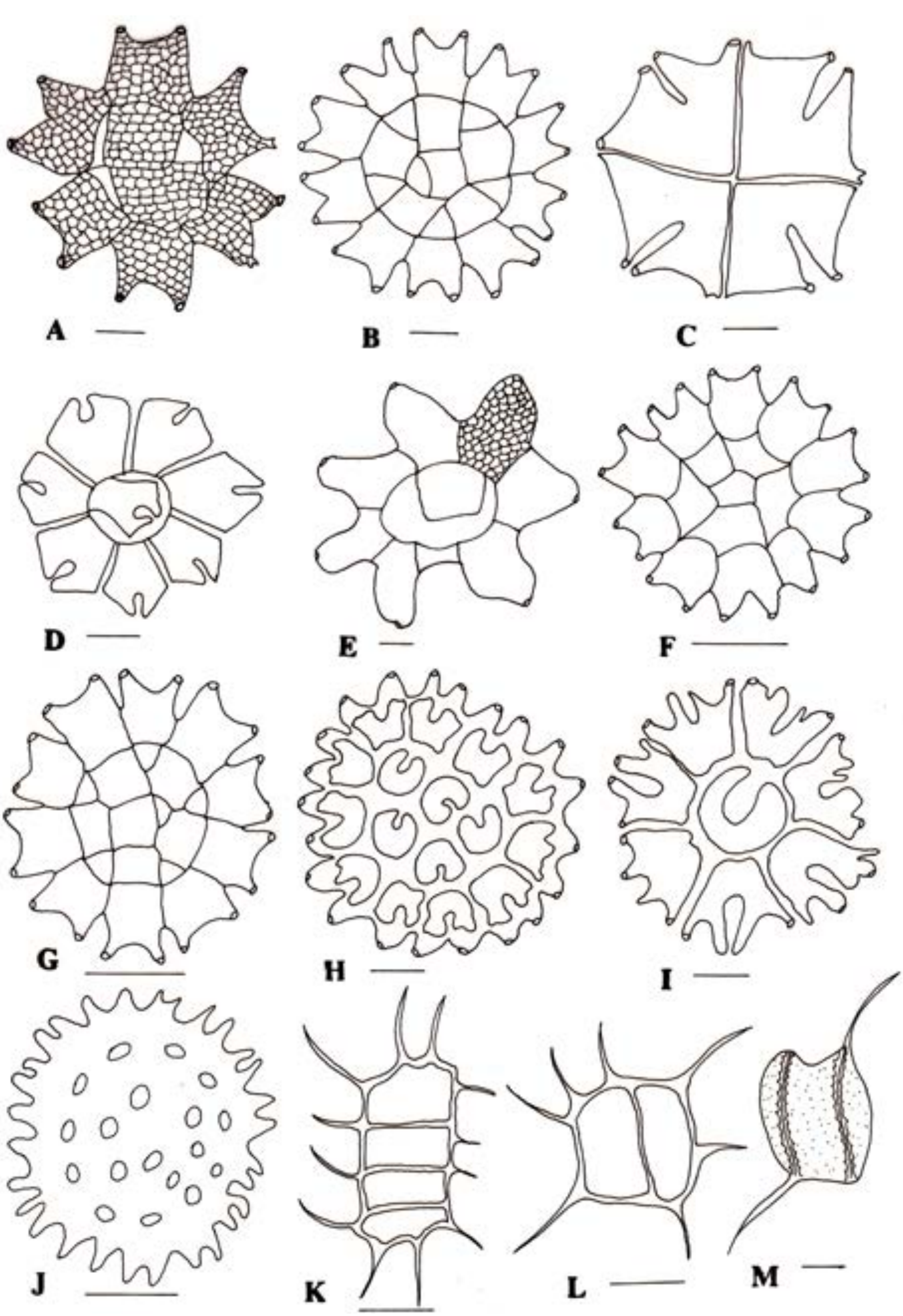

Figure 1A–M. Line drawings: A—*Pseudopediastrum boryanum* | B—*P. boryanum* var. *perforatum* | C—*Pediastrum tetras* var. *apiculatum* | D—*P. obtuosum* | E—*P. integram* | F—*P. duplex* | G—*P. arneosum*; |H—*Parapediastrum biradiatum* | I—*Stauridium tetras* | J—*Pediastrum duplex* var. *duplex* | K–L—*Desmodesmus abundans* var. *brevicauda* | M—*D. bicaudatus*. Scale bar: A, M—3µ | B–E, H–I, K–L—2µ | F, J—20µ | G—10µ.

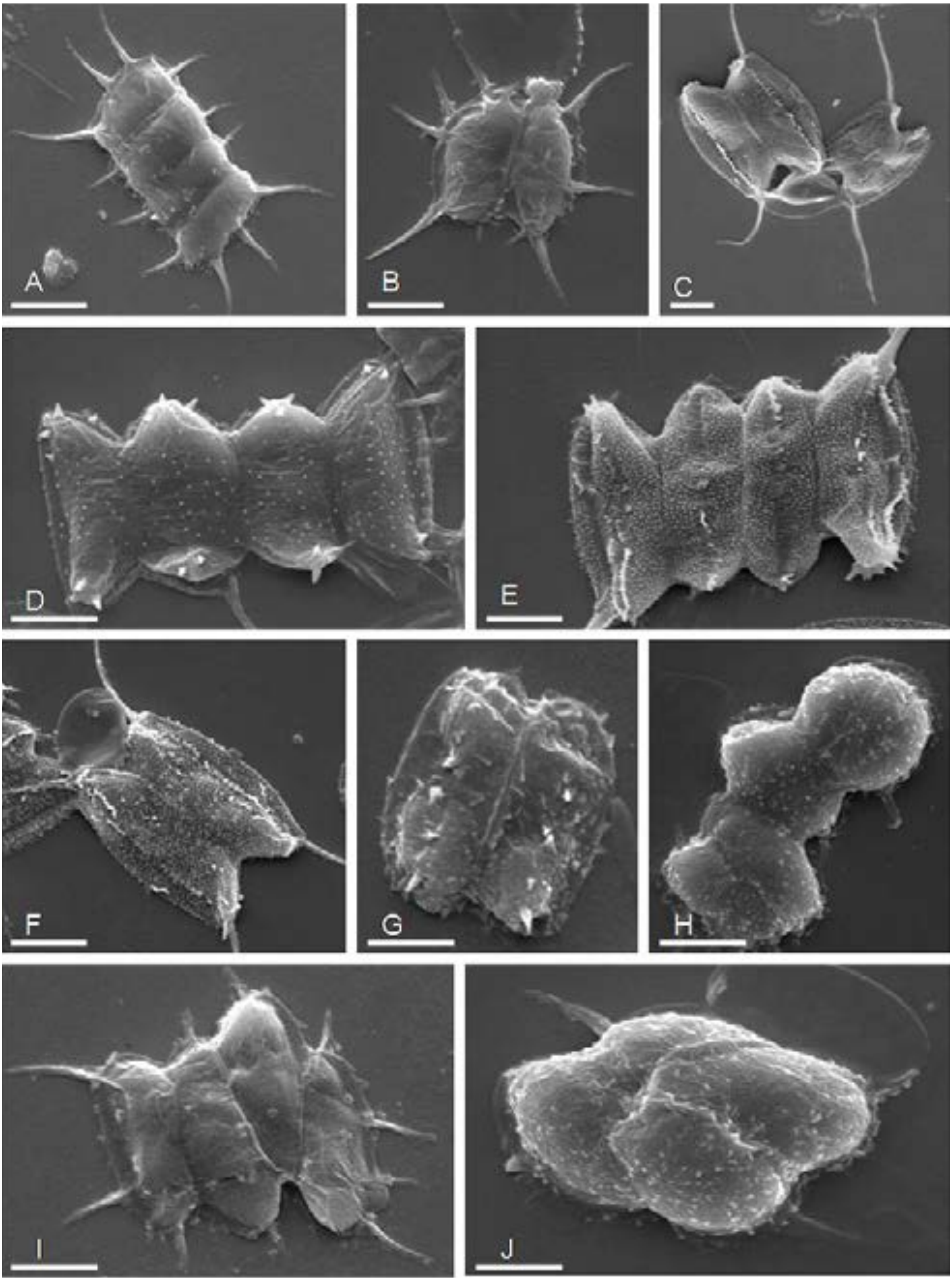

Image 3A–J. Scanning electron micrographs: A–B—*Desmodesmus abundans* var. *brevicauda* (×12.50KX and ×12.50KX) | C—*D. bicaudatus* (×5.00KX) | D—*D. serratus* (×15.00KX) | E—*D. armatus* (×12.50KX) | F—*Scenedesmus quadricauda* (×10.00KX) | G—*S. ellipticus* (×20.00KX) | H—*S. bijuga* (×17.50KX) | I—*Desmodesmus denticulatus* (×15.00KX) | J—*D. opoliensis* (×15.00KX). Scale bar: A–B—2µ | C—3µ | D–J—2µ.

15. *Scenedesmus quadricauda* (Turpin) de Brébisson (Image 3F and Figure 2A)

Basionym: *Achnanthes quadricauda* Turpin

[Bruhl & Biswas 1926; Prescott 1982; Day et al. 1995; Hu & Wei 2006].

Coenobia 2–4 celled, cylindrical, sometimes ellipsoid arranged in a single series. Cells are 6–12 µ long and 2–4 µ width; cells contain long spines at two opposite poles. Each cell contains a longitudinal ridge covered by small teeth. Under electron microscope the cell wall showed several granules and spines.

Occurrence: aquatic; voucher number: CUH/Al/MW-82.

16. *Scenedesmus ellipticus* Corda (Image 3G and Figure 2C)

Synonym: *S. ecornis* var. *flexuosus* Lemmermann; *S. linearis* Komárek

[John & Tsarenko 2002; Verschoor et al. 2004; Tsarenko 2011; Tsarenko & John 2011].

Coenobia 2-celled, arranged in a single row, cells cylindrical, bean shaped. Cells are 6–10 µ long and 2–4 µ width. Cell contains numerous small spines throughout the body. Under electron microscope the cell wall of each cell showed convoluted margins with numerous granules.

Occurrence: aquatic; voucher number: CUH/Al/MW-191.

17. *Scenedesmus bijuga* (Turpin) Lagerheim (Image 3H and Figure 2F)

Basionym: *Achnanthes bijuga* Turpin

[Prescott 1982; Cambra Sánchez et al. 1998].

Coenobia composed of four cells; cells alternately arranged; cells round to oblong to cylindrical, without teeth or spines; cells 2–6 µ in diameter and 3–8 µ long. Electron microscopic study revealed smooth cell wall with a few convoluted margins and granules.

Occurrence: aquatic; voucher number: CUH/Al/MW-83.

18. *Desmodesmus denticulatus* (Lagerheim) S.S. An, T. Friedl & E. Hegewald (Image 3I and Figure 2E)

Basionym: *Scenedesmus denticulatus* Lagerheim

[Prescott 1982; Day et al. 1995; Hu & Wei 2006; Tsarenko 2011; Tsarenko & John 2011].

Coenobia composed of 4 cells; cells alternately arranged in a single series; cells are ellipsoid to cylindrical, 4–6 µ long and 1–3 µ width; each cell with 1–4 small spines and teeth. Under electron microscope the cells showed wavy margins on the cell wall and a few apertures.

Occurrence: aquatic; voucher number: CUH/Al/MW-167.

19. *Desmodesmus opoliensis* (P.G. Richter) E. Hegewald (Image 3J and Figure 2G)

Basionym: *Scenedesmus opoliensis* P.G. Richter

[Prescott 1982; Hu & Wei 2006; Menezes 2010; Tsarenko 2011; Tsarenko & John 2011].

Coenobia composed of 2–4 celled arranged in a single series; cells 2–6 µ long and 1.5–3 µ width; cells with long spines at the apices. Cell wall is granulated and slightly folded as shown under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-168.

20. *Desmodesmus subspicatus* (Chodat) E. Hegewald & A. Schmidt (Image 4A and Figure 2H)

Basionym: *Scenedesmus subspicatus* Chodat

[Verschoor et al. 2004; Tsarenko 2011; Tsarenko & John 2011; Hilt (nee Korner) et al. 2012].

Coenobia 2-celled arranged in a single row; cells 6–8 µ in length and 2–3 µ width; apices of the cells contain 2–4 small teeth or spines. The cell surface showed numerous small teeth and granules under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-169.

21. *Desmodesmus brasiliensis* (Bohlin) E. Hegewald (Image 4B)

Basionym: *Scenedesmus brasiliensis* Bohlin

[Bruhl & Biswas 1926; Prescott 1982; Menezes 2010; Tsarenko 2011; Tsarenko & John 2011].

Coenobia composed of four cells arranged in a single row; cells 6–10 µ in length and 1–2 µ in width; apices of each cell with 1–4 small teeth and with a longitudinal median ridge extending between the apices of each cell. Median ridge of each cells are surrounded by folded margins. The outer two cells are covered with crown like structure shown under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-170.

22. *Comasiella arcuata var. platydisca* (G.M. Smith) E. Hegewald & M. Wolf (Image 4C and Figure 2J)

Synonym: *Tetrachlorella nephrocellularis* Komárek

Basionym: *S. arcuatus* var. *platydiscus* G.M. Smith

[Prescott 1982; Day et al. 1995; Menezes 2010].

Coenobia composed of eight cells arranged in a flat double series; no intercellular spaces between

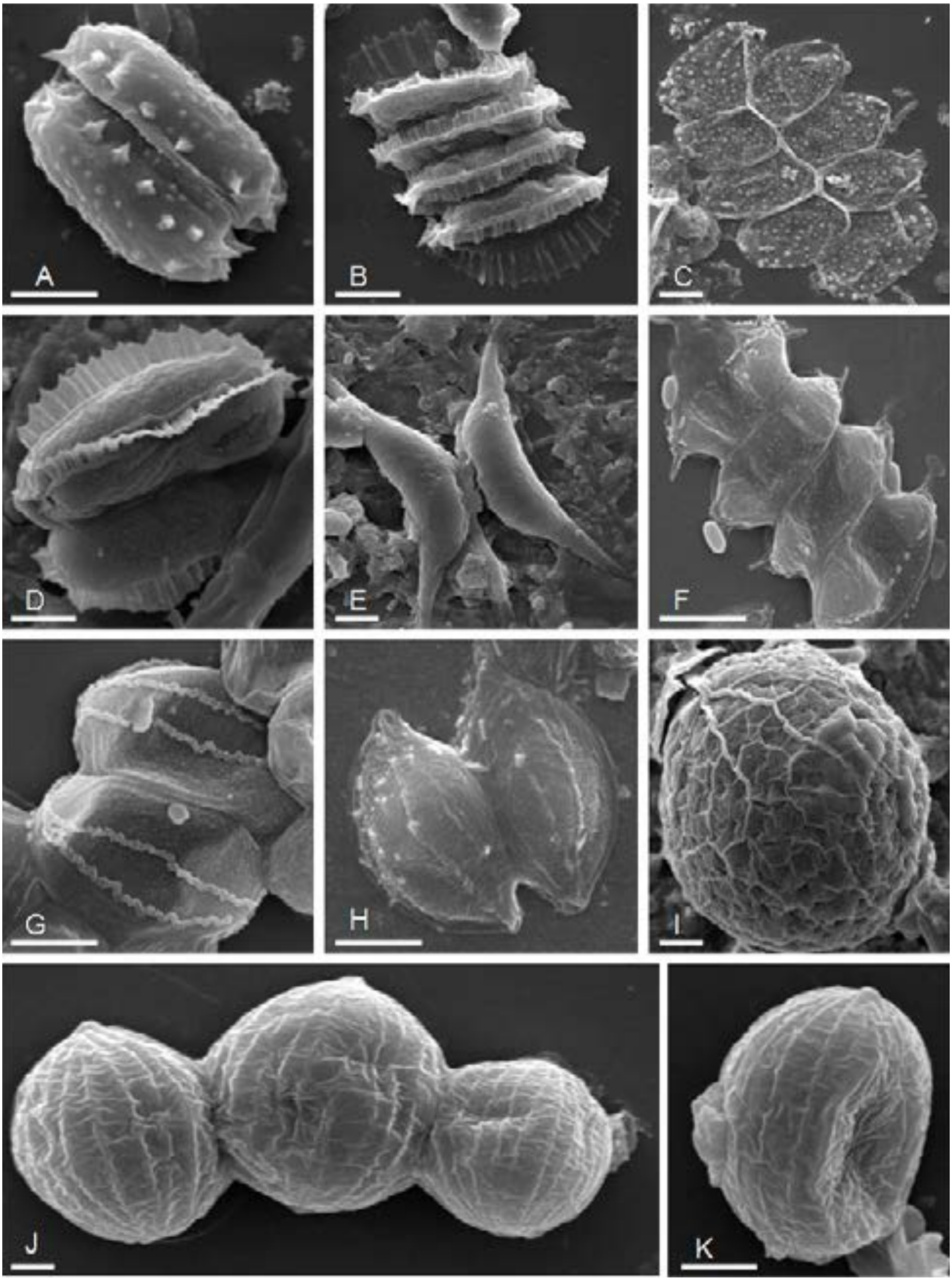

Image 4A–K. Scanning electron micrographs: A—*Desmodesmus subspicatus* (×20.00KX) | B—*D. brasiliensis* (×12.50KX) | C—*Comasiella arcuata* var. *platydisca* (×7.50KX) | D—*Scenedesmus acutiformis* (×15.00KX) | E—*Acutodesmus acuminatus* (×7.50KX) | F—*S. magnus* (×15.00KX) | G—*S. bijuga* var. *alternans* (×15.00KX) | H—*S. raciborskii* (×15.00KX) | I–K—*Chlorococcum infusionum* (×7.00KX, ×10.00KX and ×15.00KX). Scale bar: A–K—2µ.

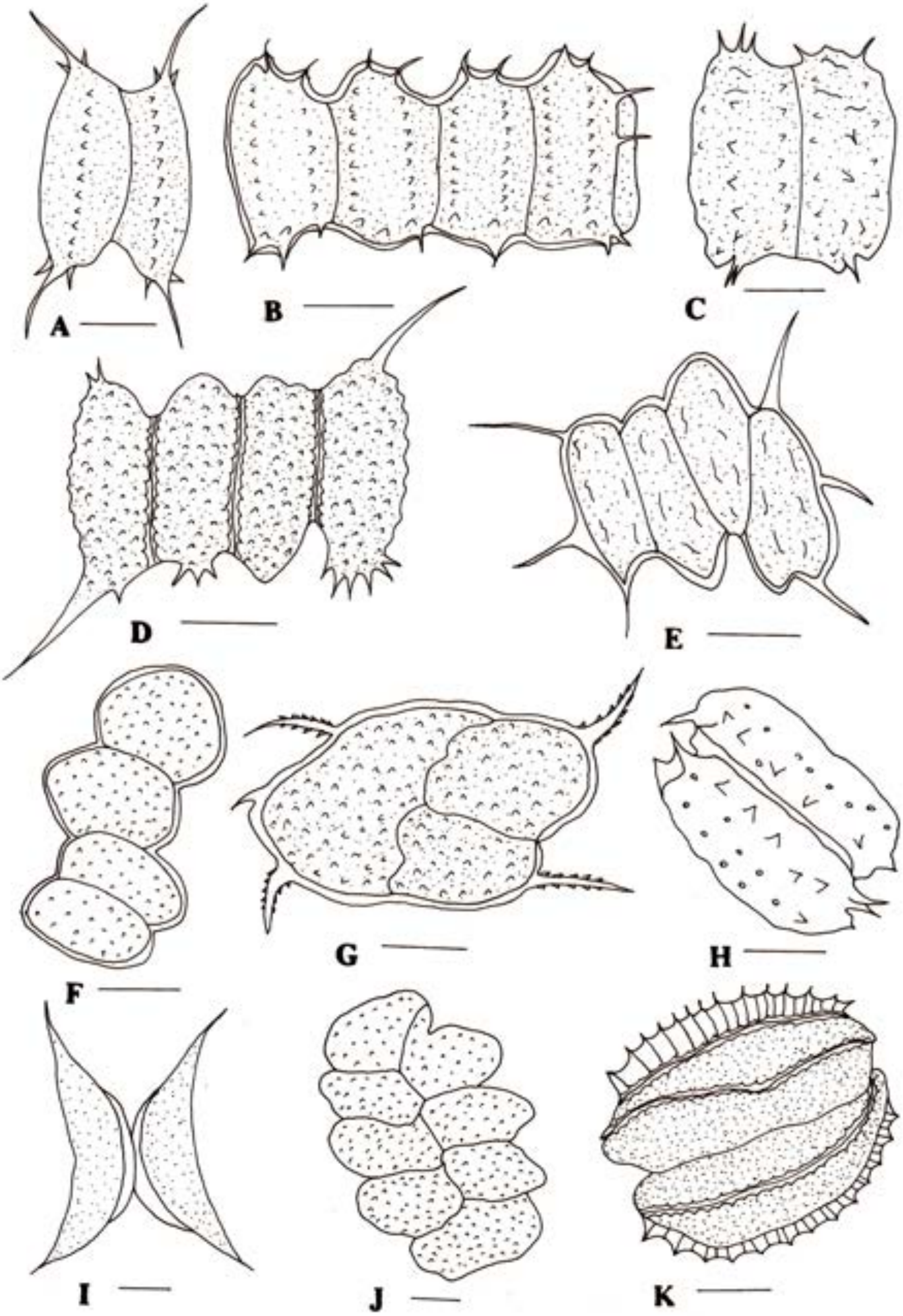

Figure 2A–K. Line drawings: A—*Scenedesmus quadricauda* | B—*S. serratus* | C—*S. ellipticus* | D—*Desmodesmus armatus* | E—*D. denticulatus* | F—*Scenedesmus bijuga* | G—*Desmodesmus opoliensis* | H—*D. subspicatus* | I—*Acutodesmus acuminatus* | J—*Comasiella arcuata* var. *platydisca* | K—*Scenedesmus acutiformis*. Scale bar: A–K—2µ.

the joining of cells; cells are 6–10 µ long and 3–6 µ width. Numerous small teeth and granules are found throughout the cell surface of each cell under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-196.

23. *Scenedesmus acutiformis* Schröder (Image 4D and Figure 2K)

Synonym: *Acutodesmus acutiformis* (Schröder) Tsarenko & D.M. John

[Prescott 1982; Day et al. 1995; Cambra Sánchez et al. 1998; John & Tsarenko, 2002; Verschoor et al. 2004; Hu & Wei 2006].

Coenobia of 2-celled arranged in a single row; cells 10–16 µ long and 6-8 µ broad; each cell having 2–3 facial longitudinal ridges covered by folded margins. The crown-like folded cell wall with smooth surface was shown under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-192.

24. *Acutodesmus acuminatus* (Lagerheim) Tsarenko (Image 4E and Figure 2I)

Synonym: *Scenedesmus acuminatus* (Lagerheim) Chodat

Basionym: *Selenastrum acuminatum* Lagerheim

[Bruhl & Biswas 1926; Prescott 1982; Tsarenko 2011; Tsarenko & John 2011].

Coenobia composed of two cells arranged in a curved series; cells strongly lunate with sharply pointed apices; cells 12–20 µ long and 2–4 µ width; the concave faces of the cells directed outward. The smooth cell wall is observed under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-195.

25. *Scenedesmus magnus* Meyen (Image 4F and Figure 3A)

Synonym: *Desmodesmus magnus* (Meyen) Tsarenko; *Scenedesmus longus* Meyen

[Prescott 1982; John & Tsarenko 2002].

Coenobia composed of four cells arranged in a single row; cells cylindrical 2–6 µ long and 1–3 µ width; apices of both inner and outer end of each cell with 1–2 sharp spines. Cells are compactly arranged and contain a median ridge. The convoluted cell wall with tiny pores was observed under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-193.

26. *Scenedesmus bijuga* var. *alternans* (Reinsch) Hansgirg (Image 4G and Figure 3B)

[Prescott 1982; Caraus 2002].

Coenobia 2-celled arranged in a single row; cells oval 6–10 µ long and 4–6 µ width. Under electron microscope the ridges were shown well and cell wall covered with tiny pores.

Occurrence: aquatic; voucher number-:CUH/Al/MW-198.

27. *Scenedesmus raciborskii* Woloszynska (Image 4H and Figure 3C)

Synonym: *Acutodesmus raciborskii* (Woloszynska) Tsarenko & D.M. John; *Scenedesmus incrassatulus* var. *mononae* G.M. Smith

[Prescott 1982; John & Tsarenko 2002; Tsarenko 2011].

Coenobia 2-celled arranged in a single row; cells elliptical or spindle shaped 6–8 µ long and 2–4 µ width; the cells are swollen in the middle and tapered at the two ends. Fine ridges and folds were found on the cell wall under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-194.

Family: Chlorellaceae

28. *Chlorella vulgaris* Beyerinck [Beijerinck] (Image 5A)

Synonym: *C. pyrenoidosa* var. *duplex* (Kützing) West; *Pleurococcus beijerinckii* Artari

[Shihira & Krauss 1965; Prescott 1982; Krientez et al. 2004; Rindi & Guiry 2004].

Unicellular, green, free floating planktonic, single or aggregated form, cells small spherical, single prominent, cup shaped parietal chloroplast, cells 4–8 µ in diameter.

Occurrence: aquatic; voucher number: CUH/Al/MW-80.

Family: Chlorococcaceae

29. *Chlorococcum infusionum* (Schrank) Meneghini (Image 4I-K)

Synonym: *Cystococcus humicola* Nägeli; *Lepra infusionum* Schrank; *Chlorococcum humicola* (Nägeli) Rabenhorst

Basionym: *Lepraria infusionum* Schrank

[Smith 1950; Prescott 1982; Chrétiennot-Dinet 1990; John & Tsarenko 2011].

Free living, unicellular, green, cells are solitary or sometimes in colonial form; striking variation in size shows between various cells when the alga grows in an expanded stratum, young cells are thin walled and spherical or somewhat compressed, old cells have thick walls that are often irregular in outline, chloroplasts of young cells are parietal massive cups, completely filling the cell except for a small hyaline region at one side, they contain one pyrenoid, as a cell increases in size, the chloroplast usually becomes diffuse and contains several pyrenoids, young cells are 50–125 µ in diameter and mature cells are 120–210 µ in diameter. Under electron microscope several groves and ridges were found on the cell surface. Many tiny pores were also observed on the cell walls.

Occurrence: aquatic, endozoic; Voucher number: CUH/Al/MW-190.

30. *Tetraëdron caudatum* (Corda) Hansgirg (Image 5B and Figure 3D)

Synonym: *Polyedrium pentagonum* Reinsch; *Tetraëdron caudatum* var. *punctatum* Lagerheim

Basionym: *Asteriscium caudatum* Corda

[Hindák 1980; Prescott 1982; Cambra Sánchez et al. 1998; Hu & Wei 2006; Tsarenko 2011; Tsarenko & John 2011].

Cells flat, irregular, 5-sided, the angles rounded and tipped with a short, sharp spine; the sides between the angles concave; margins of the cells were narrowly and deeply incised; cells 6–12 µ in diameter. Granulated cell wall with honey comb like pores were found under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-176.

31. *Tetraëdron minimum* (A. Braun) Hansgirg (Image 5C-D and Figure 3E–G)

Synonym: *T. platyisthmum* (W. Archer) G.S. West; *T. quadratum* (Reinsch) Hansgirg

Basionym: *Polyedrium minimum* A. Braun

[Hindák 1980; Prescott 1982; Andreyeva 1998; Hu & Wei 2006; Tsarenko & John 2011].

Cell flat, tetragonal, the angles rounded and without spines and processes, sometimes very minute process were found on each angles; cell margin concave; cells 8–16 µ in diameter. Various apertures and undulating margins were observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-177.

32. *Tetraëdron trigonum* (Nägeli) Hansgirg (Image 5E and Figure 3H–I)

Basionym: *Polyedrium trigonum* Nägeli

[Prescott 1982; Day et al. 1995; Hu & Wei 2006].

Cell triangular, the angles narrower and tapering at each corner, each angle was terminated to a small spine or processes, each arm of the triangle is straight, margins convex; cells 12–20 µ in diameter. Smooth and wavy cell surface was observed under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-178.

33. *Tetraëdron gracile* (Reinsch) Hansgirg (Image 5F and Figure 3J)

Synonym: *T*. *trigonum var. gracile* (Reinsch) DeToni

Basionym: *Polyedrium gracile* Reinsch

[Prescott 1982; Day et al. 1995; Hu & Wei 2006].

Cell triangular, the angles narrower and more curved like starfish, the angles tapering and terminated to spines; cells 10–22 µ in diameter; the arms of triangle are not straight and form V-shaped structure. The electron micrograph showed wrinkled margins and a triangular ridge on the cell surface.

Occurrence: aquatic; voucher number: CUH/Al/MW-179.

Family: Selenastraceae

34. *Selenastrum gracile* Reinsch (Image 5G-H and Figure 4A)

[Prescott 1982; Hindák 1988; Day et al. 1995; John & Tsarenko 2002; Hu & Wei 2006; Tsarenko 2011; Tsarenko & John 2011; Das & Keshri 2012].

Cells are found in colonies; cells are sickle shaped and in irregular arrangement; cells 2–8 µ in diameter; apices of the cells are sharply pointed. Electron micrograph showed folded and wrinkled cell surface.

Occurrence: aquatic; voucher number: CUH/Al/MW-180.

35. *Selenastrum bibraianum* Reinsch (Image 5I and Figure 4B)

Basionym: *Kirchneriella bibraiana* (Reinsch) E. G. Williams; *Ankistrodesmus bibraianus* (Reinsch) Korshikov

[Prescott 1982; Hindák 1988; Day et al. 1995; John & Tsarenko 2002; Hu & Wei 2006; Tsarenko 2011; Tsarenko & John 2011; Das & Keshri 2012].

Cells are found in colonies; cells lunate to sickle shaped; the apices of the cells are not sharply pointed; cells 12–20 µ long and 2–6 µ width. Small teeth-like projections on the cell surface were observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-181.

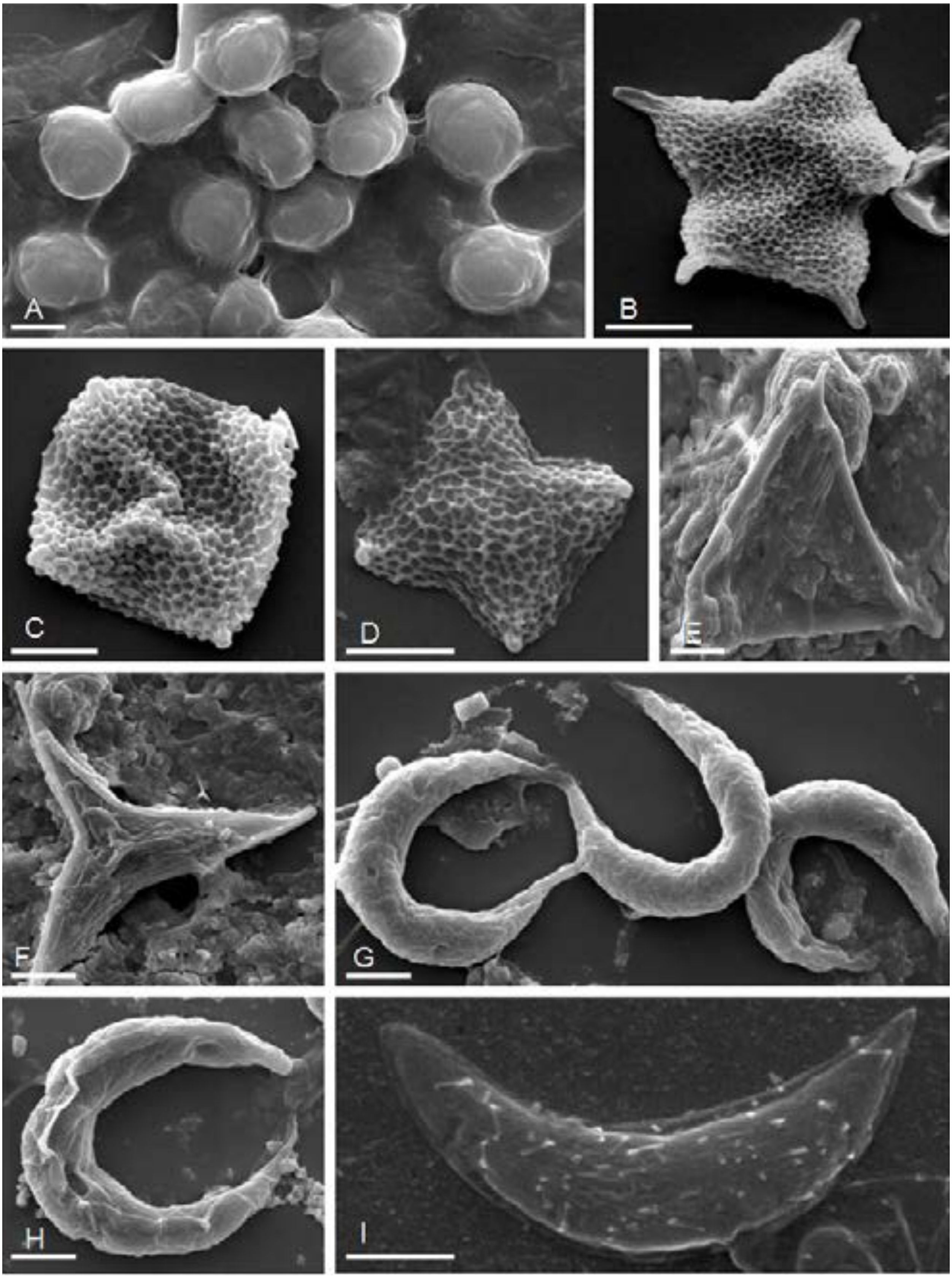

Image 5A–I. Scanning electron micrographs: A—*Chlorella vulgaris* (×10.00KX) | B—*Tetraedron caudatum* (×12.50KX) | C–D—*T. minimum* (×15.00KX and ×20.00KX) | E—*T. trigonum* (×7.50KX) | F—*T. gracile* (×10.00KX) | G–H—*Selenastrum gracile* (×10.00KX) | I—*S. bibraianum* (×20.00KX). Scale bar: A–I—2µ.

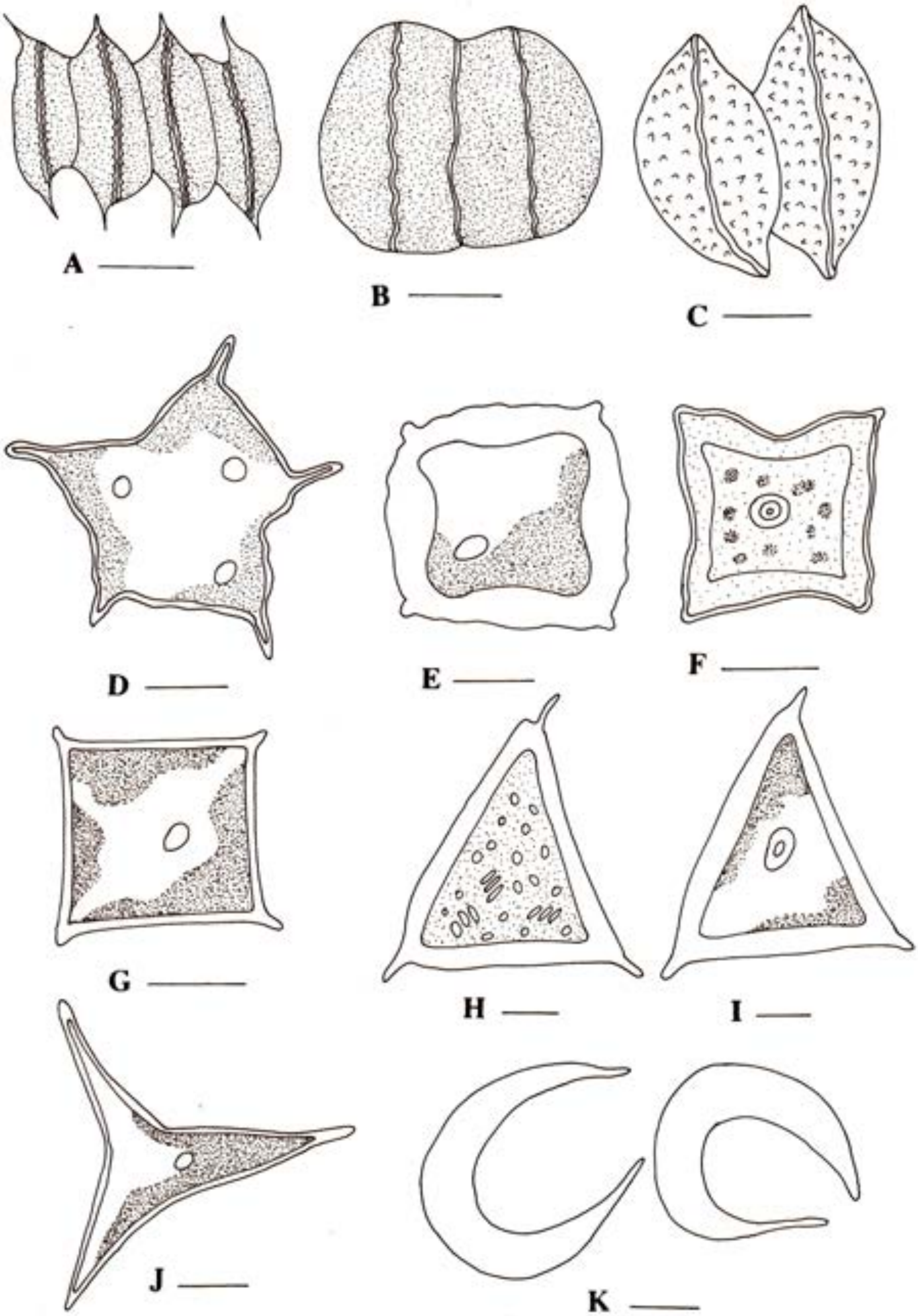

Figure 3A–K. Line drawings: A—*Scenedesmus magnus* | B—*S. bijuga* var. *alternans* | C—*S. incrassatulus* var. *monorae* | D—*Tetraedron caudatum* | E–G—*T. minimum* | H–I—*T. trigonum* | J—*T. gracile* | K—*Selenastrum gracile*. Scale bar: A–K—2µ.

Family: Desmidiaceae

36. *Euastrum denticulatum* F. Gay (Image 6A and Figure 4C)

Synonym: *E. denticulatum* var. granulatum West; *E. amoenum* F. Gay

[Ruzicka 1981; Day et al. 1995; Kouwets 1999; Wei 2003; Martello 2004; Coesel & Meesters 2007; Brook et al. 2011].

Cell solitary, green, longer than broad, small spine like projections are found on the surface of the cells; sinus narrow and linear; cells 18–26 µ long and 13–17 µ broad. The convoluted cell surface with wrinkled margins was observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-182.

37. *Euastrum dubium* Nägeli (Image 6B and Figure 4D)

Synonym: *E. dubium* var. *triquetrum* Nägeli

[Ruzicka 1981; West & West 1905; Kouwets 1999; Wei 2003; Hu & Wei 2006; Coesel & Meesters 2007; Brook et al. 2011].

Cell solitary, green, semi-cells trapezi-form, basal angles broadly rounded, four very small spines like projections or processes were shown at each corner of the cell; the margins were denticulate; cells 18–22 µ long and 12–16 µ broad. Ornamented cells with wavy margin were observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-183.

38. *Teilingia wallichii* (D.L. Jacobsen) Bourrelly (Image 6C and Figure 4H)

Basionym: *Sphaerozosma wallichii* J. Jacobsen

[Day et al. 1995; Kouwets 1999].

Colonies thread like attached by apices into long filaments or cells attached by spherical apical processes; apical processes of cells are very short; individual cells are 'X' shaped; cells 2–8 µ in diameter. Under electron microscope, smooth cell walls with minute processes were observed.

Occurrence: aquatic; voucher number: CUH/Al/MW-184.

39. *Staurastrum pantanale* K.R.S. Santos, C.F. da Silva Malone, C. Leite Sant'Anna & C.E. de Matos Bicudo (Image 6D and Figure 4E)

[Santos et al. 2013].

Cells 3-radiate, 18–24 µ long, 8–12 µ broad with processes of 8–10 µ long; isthmus 4–8 µ wide; median constriction deep; sinus acute, angular; margins deeply crenate. Cell wall provided with minute acute granules in concentric series on the processes, observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-185.

40. *Staurastrum johnsonii* West & G.S. West (Image 6E and Figure 4F)

Synonym: *S. leptocladum* L.N. Johnson

[Kouwets 1999; Coesel & Meesters 2013].

Cells 3-radiate, composed of two halves called semi-cells; cells 20–24 µ long and 10–14 µ wide; the processes 10–16 µ long with small spine like projections. Cell wall with crenate margins and acute granules were studied under the scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-159.

41. *Staurastrum simonyi* var. *semicirculare* Coesel (Image 6F and Figure 4K)

[Coesel & Meesters 2007; Coesel & Meesters 2013].

Cell triangular, 14–18 µ long and 8–12 µ wide; cell wall smooth, small minute apertures were present throughout the surface, observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-160.

42. *Staurastrum oxyacanthum* W. Archer (Image 6G and Figure 4G)

[Kouwets 1999; Coesel & Meesters 2007; Brook et al. 2011; Coesel & Meesters 2013].

Cells 3-radiate, composed of two halves of semi-cells, cells 30–44 µ long and 20–30 µ broad and isthmus 8–12 µ in diameter; the processes are deeply incised, 20–30 µ long; cell margin dentate with spine like projections; each spine is bifurcated to for two daughter spines. Several spines and wavy margins were observed under electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-161.

43. *Cosmarium dubium* Borge (Image 6H and Figure 4I)

[Day et al. 1995].

Cell solitary, green, 30–50 µ long and 20–25 µ broad; isthmus 8–10 µ; the connection between two semi-cells is smooth; the cell wall is well ornamented with small rounded projections, observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-162.

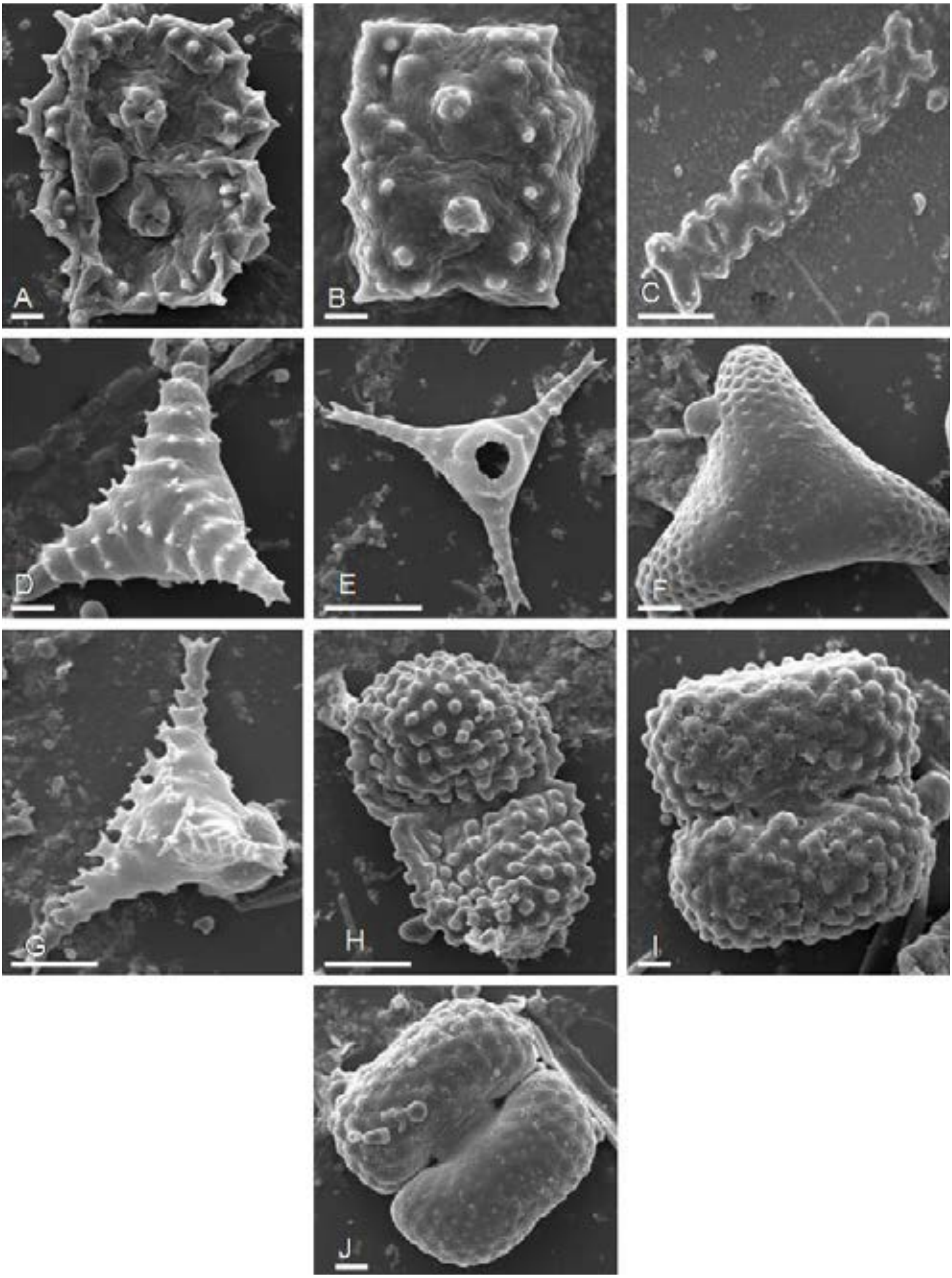

Image 6A–J. Scanning electron micrographs: A—*Euastrum denticulatum* (×5.00KX) | B—*E. dubium* (×7.50KX) | C—*Teilingia wallichii* (×3.50KX) | D—*Staurastrum pantanale* (×9.00KX) | E—*S. johnsonii* (×4.50KX) | F—*S. simonyi* var. *semicircularae* (×6.00KX) | G—*S. oxyacanthum* (×4.00KX) | H—*Cosmarium dubium* (×3.25KX) | I—*C. punctatum* (×5.00KX) | J—*C. reniforme* (×5.00KX). Scale bar: A–B, D, F, I–J—2µ | C, E, G–H—10µ.

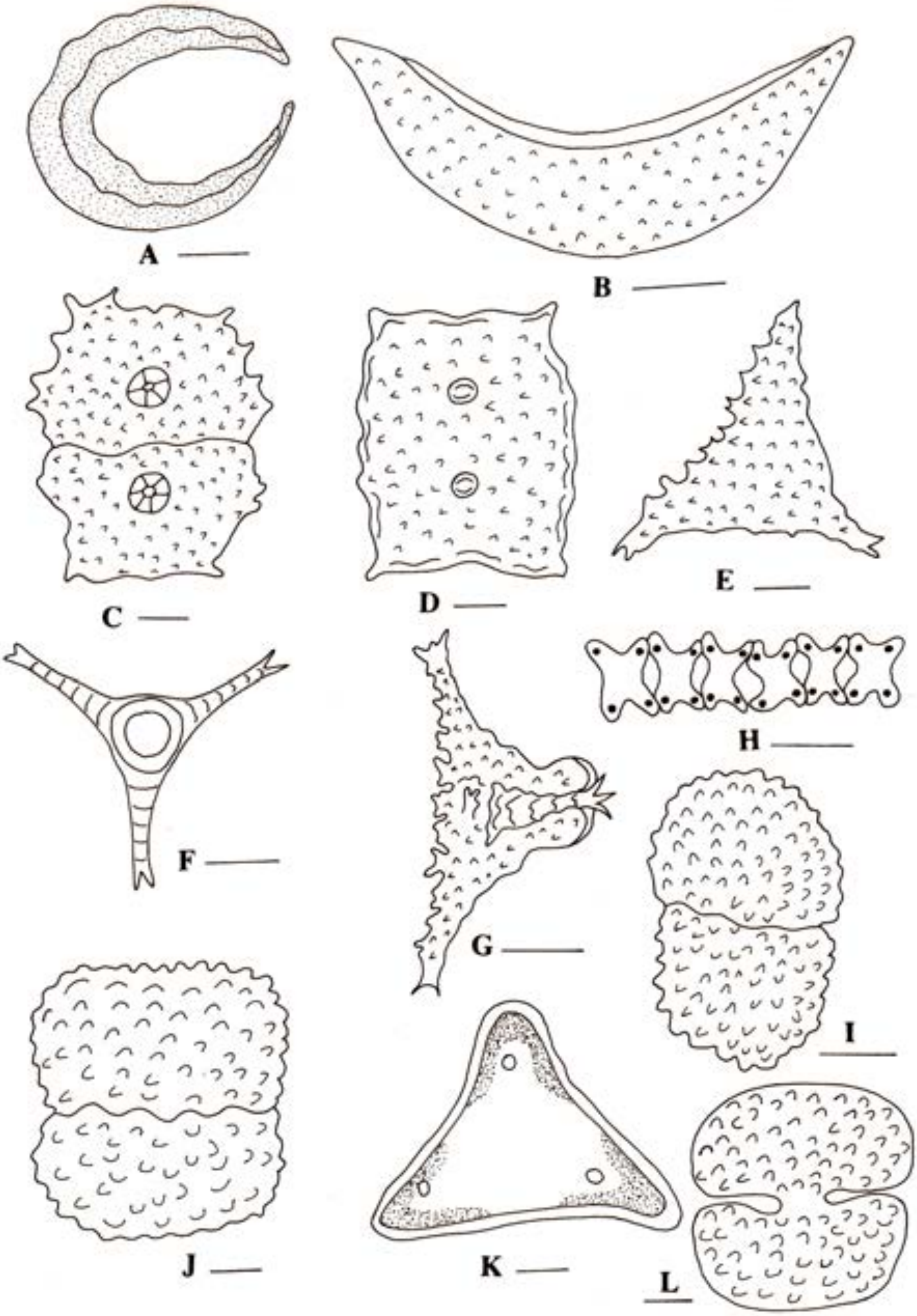

Figure 4A–L. Line drawings: A—*Selenastrum gracile* | B—*S. bibraianum* | C—*Euastrum denticulatum* | D—*E. dubium* | E—*Staurastrum pantanale* | F—*S. johnsonii* | G—*S. oxyacanthum* | H—*Teilingia wallichii* | I—*Cosmarium dubium* | J—*C. punctatum* | K—*Staurastrum simonyi* var. *semicircularae* | L—*C. reniforme*. Scale bar: A–E, J–L—2µ | F–I—10µ.

44. *Cosmarium punctulatum* Brébisson (Image 6I and Figure 4J)

Synonym: *C. punctulatum* var. *granulusculum* (Roy & Bissett) West & West

[West & West 1908; Day et al. 1995; Kouwets 1999; Hu & Wei 2006; Martello 2006; Brook et al. 2011].

Cell solitary, green, 20–26 µ long and 18–22 µ broad; isthmus 4–8 µ; cells 'dumble' shaped. Cell wall rough, well ornamented with small rounded processes observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-163.

45. *Cosmarium reniforme* (Ralfs) W. Archer (Image 6J and Figure 4L)

Basionym: *C. margaritiferum* var. *reniforme* Ralfs

[West &d West 1908; Day et al. 1995; Kouwets 1999; Hu & Wei 2006; Coesel & Meesters 2007; Brook et al. 2011].

Cell solitary, green, 20–24 µ long and 12–16 µ broad; isthmus 6–8 µ; cells 'dumble' shaped. Cell wall not so rough, well ornamented with small rounded globular projections, observed under scanning electron microscope.

Occurrence: aquatic; voucher number: CUH/Al/MW-164.

DISCUSSION

The present investigation reveals that the Indian Sundarbans exhibit rare green planktonic diversity. A few studies on planktonic chlorophytes and diatoms were found in the Gangetic belt and Bhagirathi-Hooghly estuary of the southern coast (Mukhopadhyay & Pal 2002; Chowdhury & Pal 2008). Mukhopadhyay & Pal (2002) have reported nine species of chlorophytes and five species of rhodophytes from the estuarine and coastal region of Bay of Bengal. A detailed systematic account of 19 genera and 32 species of diatoms has been done so far from the coastal belt (Chowdhury & Pal 2008). A new planktonic diatom, *Cocconeis gracilariensis* was investigated from the brackish water ecosystem of the Indian Sundarbans as epiphytic on *Gracilaria* sp. (Satpati et al. 2017). Continuous inundation of marine water in the freshwater ecosystem is the major problem for diminishing these planktonic chlorophytes. A total number of 46 taxa belonging to six groups have been reported from the Sundarban estuarine ecosystem (Manna et al. 2010). They have reported two green algal taxa *Chlorella* and *Dunaliella* in association with cyanobacteria and diatom assemblages. The conservation of these planktonic chlorophytes is suggested to protect the primary food chain of aquatic ecosystem. A few strains were maintained in the laboratory in isolated condition with accession number. Planktonic Eugenophytes were also reported and conserved from different brackish water habitats of the Indian Sundarbans (Satpati & Pal 2017). A total of 41 species of euglenoids were reported in our previous study (Satpati & Pal 2017). Some work was available on planktonic diatoms from the Sundarbans ecoregion (Choudhury & Bhadury 2014; Satpati et al. 2017). Most of the work was conducted in the Bangladesh region of the Sundarbans mangrove (Aziz & Rahman 2011). Based on previous literature, Sarkar (2011) has reported 166 species of phytoplanktons from estuarine ecosystem and associated brackish water wetlands of the Indian Sundarbans. Most of the work has been done on cyanobacteria, diatoms and filamentous chlorophytes. In this study the present group highlighted on rare green planktonic chlorophytes which were not reported in earlier studies.

Riverine fresh water run-off and tidal influx of marine water are two antagonistic hydrological processes resulting in dynamic changes in phytoplanktons and their community structure. The continuous anthropogenic perturbations, nutrient overload, increasing human population density, globalization and economic development causes vulnerability of phytoplanktons in estuarine ecosystems (Roshith et al. 2018). Temporal succession of phytoplankton assemblages of Sundarbans' mangrove was reported in a tidal creek system of the Sundarbans mangrove (Bhattacharjee et al. 2013). The phytoplankton assemblage depends on the physicochemical parameters of water and nutrient availability. The tropical and sub-tropical coastal ecosystems of the World serve as a great carbon sink due to the presence of mangroves and phytoplanktons. The biogeochemistry of carbon regulated by the key functions of genes present in the phytoplanktons reveals to illustrate their diversity (Bhattacharjee et al. 2013). The availability of phytoplanktons and mangroves helps to maintain the aquatic food chains of the coastal environments. Roshith et al. (2018) have reported the most updated information on the green phytoplanktons of Hooghly-Matla estuary. They have reported about 44 species of green chlorophytes of which 32 belong to Chlorophyceae, 11 belong to Trebouxiophyceae and 1 to Prasinophyceae. The Indian part is still less explored and more work is needed to investigate the different areas of the Sundarbans. The detailed morphological study of

Table 2. Name of the identified taxa, their latitude- longitude and conservation status.

	Name of the taxa	Latitude & Longitude (N, E)	Conservation status
1	*Pseudopediastrum boryanum*	21.893, 88.956	Rare
2	*Pseudopediastrum boryanum* var. *perforatum*	21.893, 88.956	Rare
3	*Stauridium tetras* var. *apiculatum*	22.328, 88.820	Rare
4	*Pediastrum obtusum*	22.328, 88.820	Occasional
5	*Pediastrum duplex*	22.328, 88.820	Frequent
6	*Pediastrum araneosum*	21.893, 88.956	Occasional
7	*Pediastrum integrum*	21.893, 88.956	Rare
8	*Parapediastrum biradiatum*	21.893, 88.956	Rare
9	*Stauridium tetras*	21.893, 88.956	Sporadic
10	*Pediastrum duplex* var. *duplex*	22.328, 88.820	Sporadic
11	*Desmodesmus abundans* var. *brevicauda*	22.328, 88.820	Rare
12	*Desmodesmus bicaudatus*	22.328, 88.820	Occasional
13	*Desmodesmus serratus*	22.246, 88.819	Extremely rare
14	*Desmodesmus armatus*	22.246, 88.819	Rare
15	*Scenedesmus quadricauda*	22.246, 88.819	Abundant
16	*Scenedesmus ellipticus*	22.327, 88.818	Frequent
17	*Scenedesmus bijuga*	22.246, 88.819	Frequent
18	*Desmodesmus denticulatus*	22.246, 88.819	Occasional
19	*Desmodesmus opoliensis*	22.246, 88.819	Extremely rare
20	*Desmodesmus subspicatus*	22.246, 88.819	Rare
21	*Desmodesmus brasiliensis*	22.246, 88.819	Rare
22	*Comasiella arcuata var. platydisca*	22.327, 88.818	Extremely rare
23	*Scenedesmus acutiformis*	22.327, 88.818	Frequent
24	*Acutodesmus acuminatus*	22.327, 88.818	Frequent
25	*Scenedesmus magnus*	22.327, 88.818	Rare
26	*Scenedesmus bijuga* var. *alternans*	22.246, 88.819	Rare
27	*Scenedesmus raciborskii*	22.327, 88.818	Extremely rare
28	*Chlorella vulgaris*	22.328, 88.820	Sporadic
29	*Chlorococcum infusionum*	22.055, 88.731	Sporadic
30	*Tetraëdron caudatum*	22.055, 88.731	Occasional
31	*Tetraëdron minimum*	22.055, 88.731	Rare
32	*Tetraëdron trigonum*	22.055, 88.731	Rare
33	*Tetraëdron gracile*	22.055, 88.731	Occasional
34	*Selenastrum gracile*	22.055, 88.731	Frequent
35	*Selenastrum bibraianum*	22.055, 88.731	Occasional
36	*Euastrum denticulatum*	22.055, 88.731	Rare
37	*Euastrum dubium*	22.055, 88.731	Extremely rare
38	*Teilingia wallichii*	22.055, 88.731	Extremely rare
39	*Staurastrum pantanale*	22.055, 88.731	Extremely rare
40	*Staurastrum johnsonii*	22.246, 88.820	Rare
41	*Staurastrum simonyi* var. *semicirculare*	22.246, 88.820	Extremely rare
42	*Staurastrum oxyacanthum*	22.246, 88.820	Extremely rare
43	*Cosmarium dubium*	22.246, 88.820	Rare
44	*Cosmarium punctulatum*	22.246, 88.820	Rare
45	*Cosmarium reniforme*	22.246, 88.820	Rare

phytoplanktons is lacking from the Indian Sundarbans to understand their role in primary productivity.

Conservation Status

In the present study about 18 species including nine extremely rare, seven occasional, six frequent, four sporadic and one abundant were recorded. The most abundant species in the mangrove ecosystem was *Scenedesmus quadricauda*. On the basis of sampling, *Stauridium tetras*, *Pediastrum duplex* var. *duplex*, *Chlorella vulgaris*, and *Chlorococcum infusionum* were found to be sporadic (Table 2). The species recorded as frequent were *Pediastrum duplex*, *Scenedesmus ellipticus*, *Scenedesmus bijuga*, *Scenedesmus acutiformis*, *Acutodesmus acuminatus*, and *Selenastrum gracile*. The details about the conservation status of the species including latitude and longitude are given in Table 2.

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Is cultivation of *Saussurea costus* (Asterales: Asteraceae) sustaining its conservation?

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Abstract: *Saussurea costus* (Falc.) Lipsch., (Asteraceae) known in English as Costus, is a threatened Himalayan medicinal plant listed on CITES (2014) Appendix I, Schedule VI of the Wildlife Protection Act (India) 1972, and Critically Endangered on the IUCN Red List. Wild collection of Costus is banned in India and permission is required for its cultivation and marketing. In the past 100 years of cultivation, various policy and management issues have impacted commercialization of the species. In 2015, we conducted surveys in the village of Kanol, Chamoli District, Uttarakhand, India to determine the status of Costus cultivation, problems associated with its marketing and the potential for expanded propagation to enhance local livelihoods. Forty-nine farmers in the study area were cultivating costus and interest in its cultivation had increased in the recent past due to the availability of governmental support. Annually, 1,250–2,950 kg roots (dried) and 20–57 kg seeds from this plant were produced by farmers within the study area. The area under Costus cultivation per farm was fairly constant (0.5 or 0.6 ha.) from 2012 to 2014, and the production of roots per farm ranged from 128 to 156 kg per year. Market prices for dried roots per kg had decreased over time. We found that regional and national marketing of the plant was not a problem for farmers, but export of cultivated products was a major challenge due to existing laws. In addition, local understanding related to post-harvest value addition, and self-reliance in Costus cultivation, was generally poor. Better national policies that increase the prospects for export, and more outreach to local villagers, are needed to improve the conservation and sustainable uses of Costus.

Keywords: Asteraceae, Costus, export, Himalaya, India, medicinal plant, wild collection.

Abbreviations: CITES—The Convention on International Trade in Endangered Species of Wild fauna and Flora | IUCN—International Union for Conservation of Nature and Natural Resources | TRAFFIC—Trade Records Analysis of Flora and Fauna in Commerce | WWF— World Wide Fund | WCCB—Wildlife Crime Control Bureau.

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Author contribution: CPK conducted field surveys, JTH contributed in manuscript preparation and editing, BSN supervised the work and JCK helped in manuscript preparation.

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INTRODUCTION

Saussurea costus (Falc.) Lipsch. [*Aucklandia costus* Falc.] syn. *S. lappa* (Decne.) Sch.Bip. (English and trade name: Costus; Hindi: Kuth, Koot, Kut; Sanskrit: Kushtha), a member of the family Compositae/Asteraceae is a threatened medicinal plant native to the Indian Himalaya (Madhuri et al. 2011; Zahara et al. 2014; USDA 2018). IUCN and the Red Data Book (RDB) of Indian Plants list the species as Critically Endangered and Endangered, respectively, due to its restricted distribution and heavy harvesting pressures (Hajra 1988; Walter & Gillet 1998; Saha et al. 2015). In the state of Jammu & Kashmir, costus grows as a wild perennial along the Indo-Pakistan border (Kuniyal et al. 2015). Rapid propagation techniques have been developed (Johnson et al. 1997), and the plant is cultivated in selected portions of the states of Uttarakhand and Himachal Pradesh for both medicinal and germplasm purposes. Its roots are used to produce aromatic oils and both leaves and roots are used in traditional and modern medicines for many purposes (e.g., Akhtar & Riffat 1991; Kala 2005; Parekh & Karathia 2006; Pandey et al. 2007). Cultivation began in 1920 in Himachal Pradesh and in 1929 in Uttarakhand (Kuniyal et al. 2005, 2015).

In 1950, the area under Costus cultivation in Himachal Pradesh was nearly 600ha, and this was the major source of the plant for both in-country trade and export to China (Kuniyal et al. 2015), which is the major market importing wild plant and animal products from neighbouring countries and from many other parts of the world (e.g., Heinen et al. 1995, 2001). During that period, the estimated annual production of Costus in Himachal Pradesh was 300 to 400 metric tons (MT; 1.0MT = 1,000.00kg; TRAFFIC 2011). In 1962, Indo-Chinese trans-border trade was stopped due to political conflicts between the two countries and the export of Costus was greatly affected. Cultivation, however, persisted and from 1988 to 2001 an estimated 304MT of Costus, at INR20.40 (USD0.30) to INR56.00 (USD0.84) per kg (INR66.64 = USD1.00, as on 08 March 2017), from Himachal Pradesh was marketed within India. By 2002, the per-farm area under Costus cultivation was quite small, ranging from 0.002 to 0.014 ha (Kuniyal et al. 2005).

In 1929, introductory cultivation of costus began at the Department of Forest's Bhuna Farm, located at about 3,150m in Chamoli District, Uttarakhand, India. Cultivated Costus from this region was presumably all exported to China at that time, but information regarding annual production is not available. From 2007 to 2010, 11.04MT of Costus (at INR53.00 to INR120.00 per kg; USD0.80 to USD1.80), was traded in national markets from Uttarakhand (Kuniyal et al. 2013). Despite market volatility, support from local communities and institutions for the cultivation of Costus remains due to consistent demand within India.

In 1975, Costus was listed to CITES Appendix II, however, India was not the party to CITES at that time. In 1978, the state government of Jammu & Kashmir, India, enacted the Kuth (Costus) Act for the conservation, preservation, protection and storage of the species. In 1980, with the consent of the Government of India, Costus was relisted on Appendix I of CITES (TRAFFIC 2011). To foster international compliance, the Government of India amended the Wildlife Protection Act (1972) (WPA) in 1991 and inserted Schedule VI for the protection of six rare medicinal plant species including Costus (WPA 1972; TRAFFIC 2011). Due to its inclusion on Schedule VI of the WPA, cultivation, possession, storage and trade of the species or its parts became illegal without permission from the chief wildlife warden.

At the time of inclusion in CITES and WPA, only the status of wild populations of Costus was considered, while the fact that it was and remains under cultivation in other Indian states was overlooked. Conflicts have, therefore, arisen about the validity of cultivated produce, and cultivators must abide by laws meant for the conservation of wild plants. This is a common issue for endangered plants otherwise under cultivation worldwide (e.g., Heinen & Chagain 2002; Shrestha-Acharya & Heinen 2006; Liu et al. 2014). Due to such provisions, herbal formulations or products containing costus can be seized at national and international destinations (TRAFFIC 2013). Therefore, validation of cultivated plant species listed in CITES and/or national conservation legislation requires much more consideration.

Here we made household surveys to explore the current status of Costus cultivation in the village of Kanol, Chamoli District, Uttarakhand, India, in an effort to assess marketing patterns of cultivated produce and prospects for expansion of cultivation to promote rural livelihoods. We also considered the possibility of local self-reliance in costus cultivation and provide suggestions for how laws can be amended to better-facilitate domestic cultivation and marketing.

MATERIALS AND METHODS

Study area

The village of Kanol, Chamoli District, Uttarakhand, India was selected for the field survey. Kanol is located about 8.0km from the department of forest's Bhuna Experimental Farm (~3,150m; 30.154°N, 79.395°E), where cultivation of costus was initiated in 1929. The village includes about 300 families in three settlements: Sarma-Badguna, Pranmati and Kanol. As is common throughout rural areas of India (e.g., Shrivastava & Heinen 2005), cultivation of staples such as potato (*Solanum tuberosum* L.), wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.), amaranth (*Amaranthus caudatus* L.), and beans (*Phaseolus vulgaris* L.) is done in self-owned or leased agricultural lands and cultivation of optional cash crops such as costus frequently takes place in small home garden plots. In addition, rearing livestock such as water buffalo (*Bubalus bubalis* L.), cattle/cows (*Bos taurus* L.), humped cattle/bullock (*Bos indicus* L.), sheep (*Ovis aries* L.) and goats (*Capra aegragus hircus* L.) is also common. The collection of *Ophiocordyceps sinensis* (Berk.) G.H. Sung, J.M.Sung, Hywel-Jones & Spatafora syn. *Cordyceps sinensis* (Berk.) Sacc. (Vern. - Kida Jadi, Eng. Winter Worm-Summer Grass, Tibetan – Yartsa Gumba) during May and June has also emerged as a significant economic activity in recent years (Kuniyal & Sundriyal 2013). In response to government programs, the cultivation of *Saussurea costus*, as well as other medicinal herbs such as *Picrorhiza kurrooa* Royle ex Benth. (Kutki) and *Aconitum heterophyllum* Wall. (Atis) has also received recent attention. Majority of the villagers of Kanol are well-accustomed to the cultivation of costus but, at present, only 49 families in the area cultivate costus from domestic germplasm.

For the past one and a half decades, programs have been initiated by the Government of Uttarakhand for promoting the cultivation of medicinal and aromatic plants (MAPs). As per rules, MAPs growers in Uttarakhand are registered with the Herbal Research and Development Institute (HRDI). On the basis of registration, transit passes (permission for transport and sale of cultivated MAPs products to anyone and anywhere in India) are granted to growers. The Chief Wildlife Warden, Government of Uttarakhand, has delegated power to the Herbal Research and Development Institute for granting permission to cultivate Costus and Indian Medicines and Pharmaceuticals Limited (IMPCL), a government-operated company based in Uttarakhand, has agreed to purchase cultivated Costus from local farmers at INR150.00/kg (Choudhary et al. 2013). The price for sale or purchase of costus seed is set at INR1,000.00/kg by the government.

Surveys

Field surveys were conducted in the Sarma-Badguna (~2,400m, 30.250°N, 79.583°E), Pranmati (~2,500m, 30.257°N, 79.566°E), and Kanol (~2,600m, 30.245°N,

Figure 1. Study area showing different settlements (Sarma-Badguna, Pranmati and Kanol) (not to scale).

79.205^{0}E) settlements of Kanol village during October 2015 (Figure 1). Costus is a perennial and harvestable produce is obtained after approximately two and a half years of growth. Therefore, the area under cultivation in respective years was considered, only, where from produce was harvested in that year. A semi-structured questionnaire, asking information regarding, i) area under Costus cultivation, ii) production of roots and seeds in the past three years, iii) prices received from the sale of roots and seeds at the village level, iv) marketing patterns at the village level, and v) key problems in marketing, was used for field surveys.

Data were analysed for total production and prices per kilogram during field surveys. General discussion with villagers as key informants (e.g., Shrestha-Acharya & Heinen 2006) was also held regarding cultivators' intentions for, or interest in, self-reliance in Costus cultivation, problems they face exporting costus, and whether fluctuations in the prices of raw material was a hindrance to production. Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) and the Wildlife Crime Control Bureau (www.wccb.org) websites were also considered for understanding national and international compliances and regulations about trade of threatened, CITES listed MAPs.

RESULTS

During the years 2012–13, 2013–14, and 2014–15, the area under Costus cultivation in the study area was 0.97ha. (18 farmers), 0.50ha. (8 farmers), and 1.18ha. (23 farmers), respectively. On an individual basis, the average area under Costus cultivation was 0.05–0.06 ha. A total of 2,425kg of Costus was produced in 2012–13, while much less (1,250kg) was produced in 2013–14 and more (2,950kg) was produced in 2014–15. On an average per annum basis, individual farmers produced about 135kg, 156kg, and 128kg of Costus in 2012–13, 2013–14, and 2014–15, respectively (Table 1). The average prices for the sale of costus at the village level were INR118.33 (USD1.77) in 2012–13, INR110.10 (USD1.65) in 2013–14, and INR74.35 (USD1.11) in 2014–15. The estimated average income per farmer per year from the sale of roots was INR15,941.41 (USD239.22), INR17,187.00 (USD257.92), and INR9,536.10 (USD143.01) in 2012–13, 2013–14, and 2014–15, respectively (Table 1).

A total 119.50kg of Costus seeds was produced in the study area during 2012–13 to 2014–15. On an individual basis, cultivators were able to produce averages of 3.17kg of seed in 2012–13, 2.50kg in 2013–14 and 1.85kg in 2014–15, respectively. Accordingly, on the basis of officially-fixed price (Rs. 1,000.00/kg; US $ 15,00, for seeds), individual farmers earned, on average, Rs. 3,170.00 (US $ 47.56), Rs. 2,500.00 (US $ 37.51) and Rs. 1,850.00 (US $ 27.76) in 2012–13, 2013–14 and 2014–15 respectively (Table 1). Only four farmers in the study area (who, in fact, were not registered traders) were engaged in collection, pooling, traditional drying and sale of Costus from the village to nearby towns. Produce, at the town-level, was then sold to any independently-registered trader with the forest department or its corporation, or to anyone authorized by the District Herbal Produce Purchasing and Selling Cooperative Association. In some instances, Costus produce may also be purchased by any unregistered trader, in which case, unreported trade is illegal under national and state law.

Cultivators informed that, in general, local, regional, and national trade of Costus is not a problem due to recent facilitation from the Uttarakhand State Government. Ever-fluctuating or generally decreasing prices were the main worry reported but, in any case, farmers were still able to sell their produce. Export-oriented marketing, however, poses many hurdles such as problems in obtaining legal procurement certificate (LPC), which is required for issuing valid export permits from the Wildlife Crime Control Bureau (WCCB). Maintaining quality standards required for export purposes was also reportedly difficult for local growers, a common problem in the region (e.g., Shrestha-Acharya & Heinen 2006). Cultivation of Costus along with Amaranth in mountain villages is innovative (Image 1a), however, misidentification of species in cultivation was another problem for the villagers (also see Heinen & Shrestha-Acharya 2011).

For example, the native weed *Arcitum lappa* L. (Asteraceae) also grows throughout the region and it is difficult to differentiate it from Costus in its vegetative stages (Image 1b,c). They are easy to differentiate only during flowering or fruiting, but that generally takes more than two years. Subsistence agro-pastoralism or other competing economic interests may be making Costus cultivation less important to villagers because the plant grows rather slowly and thus has a delayed effect on livelihoods. Even seed collection does not get the attention it deserves despite guaranteed prices and markets, so seeds sometimes go unharvested and germinate at their inflorescences due to their viviparous nature (Chauhan et al. 2018; Image 1d).

Table 1. Cultivation and production of Saussurea costus in Kanol, a remote village of Uttarakhand (the western Himalaya), India.

Year	No. of farmers (Total 49; involved in village level marketing 04)	Area under cultivation, hectare*		Production in kg (MT)		Average Price (INR/ kg)	Income at village level (in INR)		Seed production in the third year (in kg)		Income from seeds production (in INR) @ INR1000.00/kg (in USD)	
		Total area	Average area per farmer	Total production	Average production per farmer		Total income (in USD)	Average income per farmer (in USD)	Total production	Average production per farmer	Total income @ Rs. 1000.00/kg (in USD)	Average income per farmer (in USD)
2012–13	18	0.97	0.05	2,425.00 (2.43)	134.72 (0.13)	118.33 ± 26.40 (1.78 ± 0.40)	2,86,950.00 (4305.97)	15,941.41 (239.22)	57.00	3.17	57,000.00 (855.34)	3,170.00 (47.56)
2013–14	08	0.50	0.06	1,250.00 (1.25)	156.25 (0.16)	110.00 ± 26.73 (1.65 ± 0.40)	1,37,500.00 (2063.33)	17,187.50 (257.92)	20.00	2.50	20,000.00 (300.12)	2,500.00 (37.51)
2014–15	23	1.18	0.05	2,950.00 (2.95)	128.26 (0.13)	74.35 ± 6.62 (1.12 ± 0.10)	2,19,332.00 (3291.30)	9,536.10 (143.10)	42.50	1.85	42,500.00 (637.75)	1,850.00 (27.76)

*Cost of cultivation/ha., (soil and land development, seeds cost, weeding and hoeing, maintenance up to 3 years, uprooting or harvesting, and drying and packaging is approximately Rs. 150,000.00 (US $ 2250.90, @ Rs. 66.64 = US$ 1.00), and total profit after 3 years may be Rs. 3,34,566.00 (US $ 5770.80). USD1 = INR66.64 as on 08 March 2017).

Figure 2. a—cultivation of *Saussurea costus* along with Amaranth (red inflorescences) | b—flower head of *S. costus* | c—inflorescence of *Arcitum lappa* | d—germinating seeds of *S. costus* on the infructescence. © C.P. Kuniyal.

DISCUSSION

The fact that there is some continued production via traditional cultivation of *Saussurea costus* in this remote Himalayan village is encouraging for the conservation of this endangered plant. On an average annual basis, we found that individual farmers had 0.05ha under cultivation, from which about 100kg of roots and 2.50kg of seeds could be produced once the plants reached maturity (i.e., after two to three years). Therefore, the average economic benefit from Costus cultivation estimated from this study is INR16,700.00 (USD250.00). Calculated on per hectare basis, the total (gross) profit would be approximately INR334,566.00 (USD5,020.50). On the other hand, on a per hectare basis, the income from traditional cultivation of potato is around INR175,700.00 (USD2,636.55; Kuniyal & Sundriyal 2013). In order to promote Costus as an economically-attractive crop, and to make it competitive with food crops, the prices for its sale at the village level would have to increase one-and-a-half to two times more than at current prices.

A total of 49 farmers in three settlements (Sarma-Badguna, Pranmati, and Kanol) of the village of Kanol were cultivating Costus during the years under study. Almost all of them have revived this practice since 2009 from locally available seeds. A few growers also procured some seeds from the adjacent state of Himachal Pradesh; however, the productivity of both materials was virtually identical indicating that domestically-propagated plants showed no loss in fitness. This can be a concern with regard to some other medicinal plants proposed for cultivation (e.g., Liu et al. 2014). Despite the fact that this plant is established as a fully-domesticated agricultural crop, the economic potential of Costus is high and export oriented cultivation could possibly be achieved via support from government sources to maintain the gene pool in this area.

The domestication and mass cultivation of Costus in the Indian Himalaya was previously aimed at producing large quantities for export to China. Today, small-scale cultivation persists to fill local and within-country demand only. Therefore, programmes supporting entrepreneurship development should be local to regional, and involve some value addition and legal support (Kuniyal & Negi 2016, 2018). As currently practiced, Costus cultivation is more an opportunistic activity in some areas for small-income generation. The trend in marketing has also indicated that, as the availability of produce increased, prices decreased and this is of great concern for cultivators.

During the field surveys, it was noted that cultivation of Costus is as traditional as it was decades ago and farmers are not aware of, or trained about, proper cleaning, drying and storage of harvested Costus for value addition. Therefore, developing a better understanding of cultivators and training them in post-harvest management techniques are equally or more important than knowing traditional practices. Value addition and possibilities for development of Costus-based small scale industries is a possibility, but there is a long way to go. Costus cultivators and collectors at the village level, as well as local traders, tend to work independently. There is a need to bring them together in the form of a grower's and/or trader's cooperative organization to promote price sharing and training for more effective marketing.

A majority of growers in the study area still look to government for help in the cultivation and marketing of MAPs. More self-reliance would promote more innovation at all stages of production. From the part of facilitating agencies, adopting strategies for reconciling traditional farming, conservation and identification of social, legal and political actions may help in bringing about positive changes in conservation-oriented farming (Harvey et al. 2008). As compared to conventional approaches, developing socio-entrepreneurial approaches can be effective in enhancing conservation efficacy and benefit sharing (Buschke 2015). Determining management goals for threatened species, and specific policies for integrated development of the MAPs sector, are also essential (Heinen & Shrestha-Acharya 2011). Collecting baseline information allows for the evaluation of conservation practices and can be used to set interventions for future (Bull et al. 2014). Domestication and mass cultivation of MAPs takes high energy inputs, so developing supply mechanisms with lower energy input may be advantageous to growers (Smith-Hall et al. 2012).

Policy and legal efforts of parties to CITES and the Convention on Biological Diversity appear to be currently insufficient (Lambooy & Levashova 2011). Generally, developing countries are more focussed on rapid economic development despite costs, rather than on more sustainable economic ventures (Okereke & Ehresman 2015). As a result, MAPs are still a neglected commodity in local, regional and national development plans of many poor countries (Larsen & Olsen 2007). It is well accepted that policy and legal support from governments may improve multiple perspectives of any indigenous practice (Ens et al. 2015). Legal amendments would be required in conservation acts for promoting costus or any other CITES-listed species, as an export-oriented medicinal crop (Kuniyal et al. 2015).

Cultivation of Costus is a century-old practice, therefore, accepting it as an indigenous practice is inevitable. Cultivation of any CITES listed MAPs in remote, inaccessible and developing regions presents options for both conservation and socio-economic development. While preparing conservation plans for Costus, the fact that it has been under cultivation for almost a century was largely overlooked. Conservation criteria, and rules and regulations for protection, were devised only based on information about its status in the wild. As a result, farmers cultivating costus have to abide by these rules. In order to meet international compliance, while an application is filed with the office of WCCB for obtaining a legal procurement certificate for any CITES-listed species, the applicant has to provide information regarding (i) source of procurement (collected from wild/bred in captivity/artificially propagated), (ii) license number, and (iii) country in which the specimen was taken from wild/bred in captivity/artificially propagated.

Thus the procedure is complicated and the onus is on the cultivator, which creates confusion and discourages value-added, export-oriented cultivation. The cultivation and export of MAPs should be attractive to growers in remote villages given the poverty inherent to these regions, and it would be an excellent example of special conservation sites (Baral et al. 2014), involving conservation through participatory approaches and income generation. Therefore, attempts to maintain costus cultivation in this area and modifying rules and regulations as per real-world conditions are highly recommended. Capacity building of farmers for improved agricultural practices, value addition and self-reliance are good options for the conservation and sustainable uses of threatened MAPs.

CONCLUSIONS

Rejuvenation of the cultivation of Costus in remote mountain villages, and obtaining additional income from the sale of its roots and seeds, is encouraging in that it promotes the conservation of a threatened species. Villagers have conserved this species, outside of its native range, as a cash crop for about a century with rather little reward in return. Cultivation of Costus has secured its place in traditional agriculture in the study area and capacity building of farmers for better agricultural practices, as well as developing and teaching techniques for value addition, would produce better economic returns. Self-reliance of costus production, in the economic sense, could possibly be achieved through the creation of growers' cooperatives and the expansion of production into semi-processed products. Some intervention by governmental or non-governmental entities could facilitate this and thus make cultivation more attractive to local farmers. Considering 'cultivation' and 'collection from the wild' as two very separate cases, and simplifying rules and regulations for cultivated produce, will encourage farmers and promote wider cultivation (Kuniyal et al. 2015). This should be a goal for both conservation of any marketable species, and local economic development to improve rural livelihoods.

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A first photographic record of a Yellow-bellied Weasel *Mustela kathiah* Hodgson, 1835 (Mammalia: Carnivora: Mustelidae) from western Nepal

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Abstract: One live and another dead Yellow-bellied Weasel were spotted at an altitude of 2,190m and 3,078m, respectively, in Lamsung, Dhaulagiri Rural Municipality, Myagdi and Barekot Rural Municipality, Jajarkot on 1 May 2016 and 16 June 2016 in the afternoon. This is probably the first record of the species with photographs in Myagdi District of Gandaki Province, and in Jajarkot District of Karnali Province, western Nepal.

Keywords: Carnivore, Gandaki Province, habitat, Jajarkot, Karnali Province, Myagdi, traditional transhumance.

Five species of the genus *Mustela* belonging to the family Mustelidae, namely Stoat (Ermine in North America) *M. erminea,* Siberian Weasel *M. sibirica,* Yellow-bellied Weasel *M. kathiah,* Mountain Weasel *M. altaica*, and Stripe-backed Weasel *M. strigidorsa* have been recorded from Nepal (Baral & Shah 2008). The Yellow-bellied Weasel *Mustela kathiah* is found along parts of the Indian Himalaya through Nepal, Bhutan, northeastern India, southern China east to Hong Kong, and southeastern Asia in northern & central Myanmar, northern & central Thailand, Lao PDR & Viet Nam, with one series of records in the Cardamom Mountains of Cambodia (Pocock 1941; Corbet & Hill 1992; Duckworth & Robichaud 2005; Than et al. 2008; Pei et al. 2010; Ghimirey & Acharya 2012; Supparatvikorn et al. 2012; Abramov et al. 2013; Appel et al. 2013; Choudhury 2013; Phan et al. 2014).

The Yellow-bellied Weasel (Y-bW) is primarily

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associated with hill evergreen forest at elevations above 1,000m (Willcox et al. 2016) but in winter it may come down lower than 1,000m. It is evidently diurnal, probably mostly ground-dwelling but an occasional climber, and is assumed to be largely carnivorous (Wan 2014; Willcox et al. 2016) as it mostly feeds on birds, mice, rats, voles, and other small mammals.

Even if the Y-bW is known throughout Asia; only little information is known due to its inaccessible habitat. Intensive research activities focusing on small carnivores are often neglected in Nepal. Among carnivores, scientific studies on weasels are very limited. Only sketchy information is available on the abundance and distribution of these species from the country. There is dearth of distribution data and conservation efforts for Y-bW in Nepal.

There are few evidences of the Y-bW's occurrence in Nepal and it lacks scientific studies primarily focusing on this species. This paper attempts to discuss on the first record of the Y-bW from Myagdi District of Gandaki Province and Jajarkot District of Karnali Province in western Nepal.

Survey area and methods

Dhaulagiri Rural Municipality (RM) lies in Myagdi District of Gandaki Province, Nepal. The total population of Dhaulagiri RM is 14,104 (Central Bureau of Statistics 2011) that resides in an area of 1,037km². Barekot RM is located in Jajarkot District of Karnali Province, Nepal covering an area of 577.7km² with a total population of 18,083 (Central Bureau of Statistics 2011). Both rural municipalities act as a refuge for different mammal species such as the Himalayan Black Bear *Ursus thibetanus*, Red Panda *Ailurus fulgens*, Musk Deer *Moschus* spp., Northern Red Deer *Muntiacus vaginalis*, Himalayan Tahr *Hemitragus jemlahicus*, Common Goral *Naemorhedus goral*, Himalayan Serow *Capricornis thar*, and Blue Sheep *Pseudois nayaur* (District Development Committee 2011; Baral et al. 2014).

An opportunistic survey was done for the confirmation of the presence of weasel species. The field study focusing on Red Panda was conducted for a total of 40 days (20 days each in Myagdi and Jajarkot from 15 April to 4 May 2016 and 28 May to 16 June 2016, respectively). The Y-bW was observed in two different locations (Figure 1). When the species was

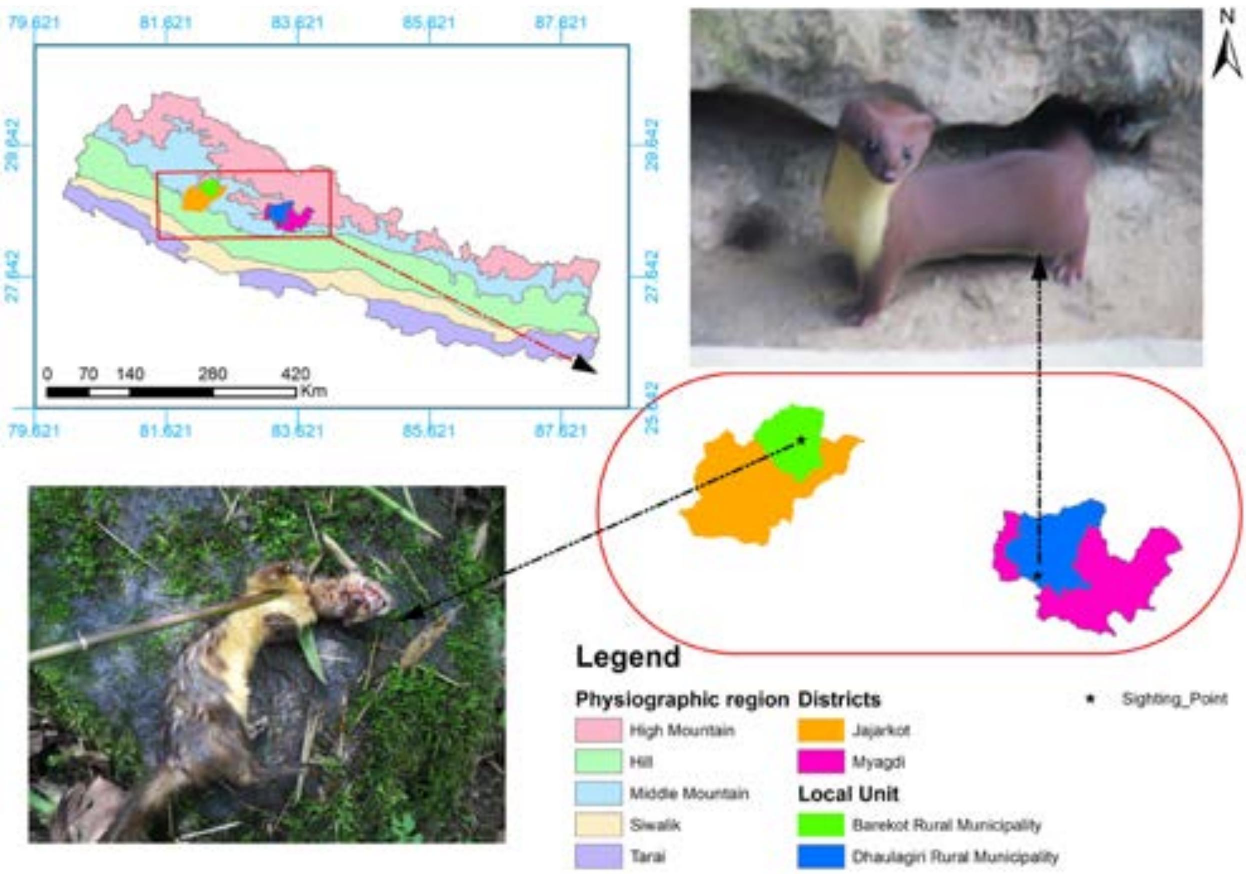

Image 1. Locations of Yellow-bellied Weasel spotting.

Image 1. *Mustela kathiah* at Lamsung, Dhaulagiri Rural Municipality, Myagdi.

Image 2. Dead *Mustela kathiah* at Dhottachaur Community Forest, Barekot RM, Jajarkot.

observed, a Canon PowerShot SX170IS camera was used to capture the photograph of the species and Garmin etrex 10(model) GPS was used to mark the location where the species was observed. Also measurements were taken where possible.

Observation

On 1 May 2016 in the afternoon, a live Y-bW was spotted at 28.518^{0}N & 83.285^{0}E at an altitude of 2,190m (Image 1). The Y-bW came out of a hole beneath a rock boulder in Lamsung Village of Dhaulagiri RM. The foot pads were well developed and exposed. The soles of the hind feet were bald. The habitat was beneath the rock boulder in the midst of Lamsung Village of Dhaulagiri RM. The nearby forest was dominated by *Quercus* sp., *Juglans regia*, and *Rhododendron arboretum*.

On 16 June 2016 in the afternoon, a dead Y-bW was spotted at 28.995^{0}N and 82.316^{0}E at an altitude of 3,078m (Image 2). A venomous Himalayan Pit Viper *Gloydius himalayanus* (Günther 1864) was found on the other side of the boulder where the dead weasel was observed. The Y-bW was found beneath the rock boulder which was kept on the boulder for the photographs. The weasel was drenched in rain. There was a severe wound on its neck, perhaps from a fight. The foot pads were well developed and exposed. The soles of the hind feet were bald. The head and body length was 250mm, and its tail length was about 130mm. The weasel weighed 1.5kg. The habitat was dominated by *Tsuga dumosa, Abies spectabilis*, and *Rhododendron arboreum* with the understory of *Thamnocalamus spthiflorus, Drepanostachyum falcatum,* and *Yushania* sp.. People from the nearest village, Nayakwada frequently visit the habitat to fetch *Thamnocalamus spthiflorus, Drepanostachyum falcatum,* and *Yushania* sp. for their household requirements. Hunting of wildlife has been an inseparable part of the local inhabitants. The area is notorious for illegal and communal hunting and it occurs throughout the year with a peak during the Dashain festival and post monsoon season which has threatened the weasels' habitat. Traditional transhumance practice of livestock management is common in the region which further affects the weasel habitat in the region.

Discussion

This paper provides a documentation of an incidental record of *Mustela kathiah*. Based on ground truthing, the Y-bW occupancy has now been confirmed from Lamsung of Dhaulagiri RM, Myagdi and Dhottachaur Community forest of Barekot RM of Jajarkot and is within the previously recorded elevational range (see Baral & Shah 2008). The Y-bW was recorded first in the Makalu–Barun National Park in eastern Nepal during a field survey in 2009–2010 (Ghimire & Acharya 2012).

There is also a report of the Y-bW within and between the protected areas of Annapurna Conservation Area, Sagarmatha National Park, Makalu Barun National Park (Jnawali et al. 2011), from Illam & Dallu, Pharping, Kathmandu (Katuwal et al. 2018), and from Hugu-Kori forests in Annapurna Conservation Area (Yadav Ghimire pers comm. 2018; Baral et al. 2019).

The Y-bW is categorized as Least Concern globally by The IUCN Red List of Threatened Species (Willcox et al. 2016) and as Data Deficient nationally under Red List criteria (Jnawali et al. 2011). The Y-bW was camera-trapped once each, in 1,184 trap-nights (Ghimire & Acharya 2012). The Y-bW has a small build and skulking behavior that makes it difficult to record it by a typical survey method and camera trap of low density (Than et al. 2008; Ghimire & Acharya 2012; Supparatvikorn et al. 2012; Willcox et al. 2016). This might have created a hindrance for an assessment of its population status.

In Nepal, the Y-bWs are commonly used to eradicate rodents and are trained to attack larger animals such as geese, goats, and sheep for sport (Sterndale 1982; Hussain 1999; Jha 1999). Local residents in the nearest villages of Jajarkot and villagers of Myagdi, however, were unaware of the existence of this species and thereby we did not find any anecdotal report on the use of weasels for any purpose. Also, villagers of both localities were unaware of the ecological significance of the species. School students in Lamsung of Myagdi were, however, reported to kill this weasel to show their bravery. School outreach and community awareness activities are recommended to conserve this small carnivore.

This study would enhance the understanding of the Y-bW's distribution and conservation status in Nepal as very less information is available on the abundance and distribution of these species from the country. This paper attempts the documentation of the first record of the Y-bW from Myagdi and Jajarkot in western Nepal emphasizing that more intensive research is needed to improve understanding of the species' characteristics, habitat and ecology.

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Mammal diversity in a montane forest in central Bhutan

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Abstract In Bhutan, knowledge of wildlife species richness in protected areas is increasing, particularly for mammals; however, the knowledge outside of protected areas typically remains poor. We conducted a camera trap survey from May 2016 to July 2017 in a montane forest outside of the protected areas network in central Bhutan and recorded 15 species of mammals (belonging to nine families and three orders), of which nearly half were listed as Endangered, Vulnerable, or Near Threatened. Our findings demonstrate that forested landscapes outside protected areas in Bhutan support a rich assemblage of wildlife species and are, therefore, deserving of comprehensive wildlife conservation plans and dedicated funding for ecological research and threat mitigation.

Keywords: Biodiversity, camera trap, Himalaya, protected areas, threatened species.

Bhutan has close to 200 species of mammals (Wangchuk et al. 2004; NEC 2011) and is a part of the Himalaya Biodiversity Hotspot (Mittermeier et al. 2004) and the Global 200 ecoregions (Olsen & Dinerstein 2002). Given the small geographical size of the country, the rich diversity of species can be attributed to its location at the junction of the Indo-malayan and the Palaearctic biogeographic realms. Apart from the protected areas which comprise more than 50% of the country, other regions lack dedicated species inventories. The lack of information deters comprehensive species conservation initiatives in the light of rapid changes to Bhutan's rich and diverse ecosystem (Dhendup & Dorji 2018a; Penjor et al. 2018). Camera traps have emerged as a successful and most frequently used tool for terrestrial species monitoring in Bhutan and have provided critical information on a few keystone and endangered species such as the Bengal Tiger (Wang & Macdonald 2009; Tempa et al. 2013; DoFPS 2015; Thinley et al. 2015) and Snow Leopard (DoFPS 2016; WCNP & WWF 2016). Here we use camera traps to document the diversity and relative abundance of mammals in a montane forest in central Bhutan and also to provide baseline information to facilitate the preservation of such sites for the conservation of globally threatened species.

Materials and methods

We conducted the study in the Lamai Goempa Research Preserve in the Bumthang District of Bhutan (Figure 1). The preserve spans an area of 1,098ha and is also used by local communities as grazing ground for cattle, extraction of timber, collection of non-wood forest products, and hiking. The area receives an average annual rainfall of 1,404mm. The summer temperatures can go up to 23°C, and the winter temperature can drop to -6°C (Pearl et al. 2015). The vegetation comprises

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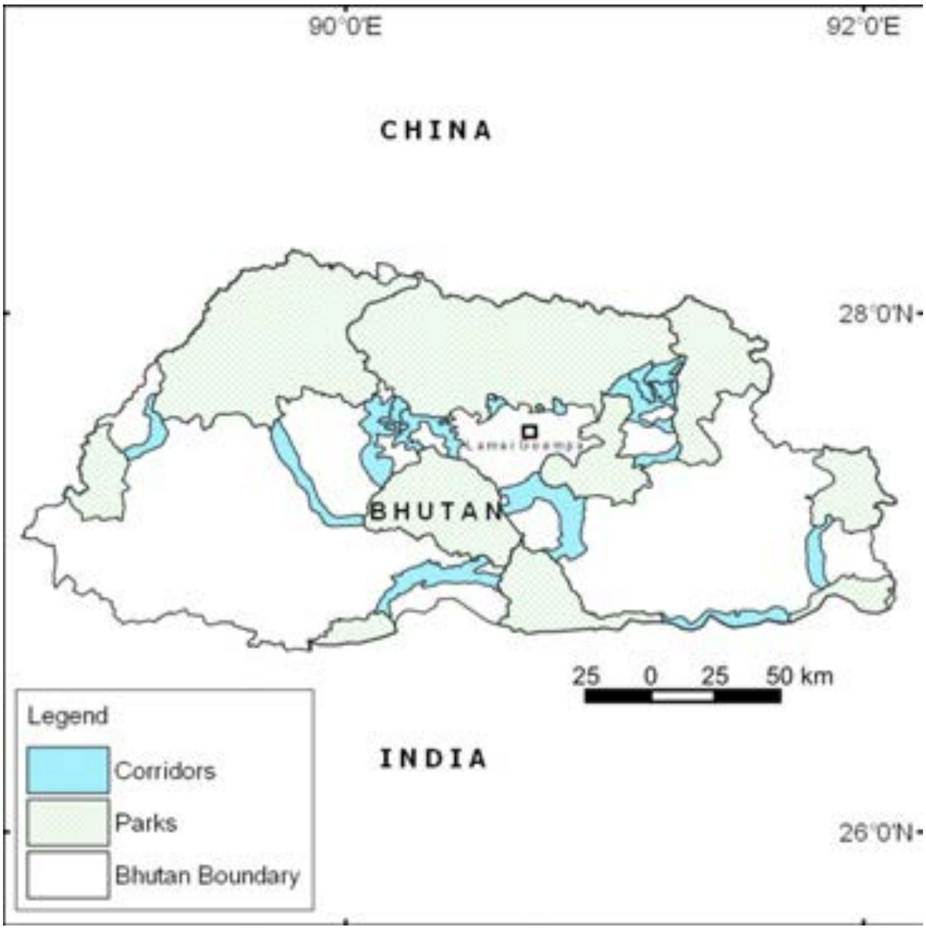

Figure 1. Study area in Bumthang against Bhutan's protected area network.

mixed conifer forest and alpine rhododendrons (Image 1 & 2).

We carried out the camera trapping survey from May 2016 to July 2017 using 25 camera stations established along footpaths, and game trails and the camera trapping array covered elevations from 2,892 to 4,120 m. We placed one passive infra-red Reconyx HC500 Hyperfire camera (RECONYX, Inc., Wisconsin, USA) at each station and was set to operate for 24h (Image 3). Cameras were at least 500m away from each other and were placed at the height of 45–50 cm above the ground. No bait/lure was used. For a series of images of the same species occurring at a camera trap location, we classified the captures as a notionally independent event only if they were taken at one-hour intervals (Sanderson & Harris 2013; Hodge & Arbogast 2016). Memory cards from two camera stations were lost. We calculated the relative abundance index (RAI), naïve occupancy, and the latency to initial detection for each species in the study area (Table 1).

Results and Discussion

The complete survey resulted in an effort of 4,501 trap nights and produced 34,237 photographs, of which 7,617 pictures were taken of 15 mammal species representing nine families and three orders (table 1) (Image 4–18). The species detected were Tiger *Panthera tigris*, Asiatic Golden Cat *Catopuma temminckii*, Marbled Cat *Pardofelis marmorata*, Leopard Cat *Prionailurus bengalensis*, Asiatic Wild Dog *Cuon alpinus*, Red Fox *Vulpes vulpes*, Himalayan Serow *Capricornis thar*, Barking Deer *Mutiacus muntjac*, Sambar *Rusa unicolor*, Wild Boar *Sus scrofa*, Yellow-throated Marten *Martes flavigula*, Asiatic Black Bear *Ursus thibetanus*, Himalayan Crestless Porcupine *Hysterix brachyura*, Orange-bellied Squirrel *Dremomys lokriah*, and Weasel *Mustela* sp. Two are listed as Endangered, three Vulnerable, two Near Threatened, and the rest as Least Concern on the IUCN Red List of Threatened Species. Although we did not record the Red Panda *Ailurus fulgens*, the River Otter *Lutra lutra* and other small mammals such as picas, rats and voles, these species are known to occur in the area and will require a species-specific survey protocol. Wild

Image 1. Typical weather in the study area during summer.

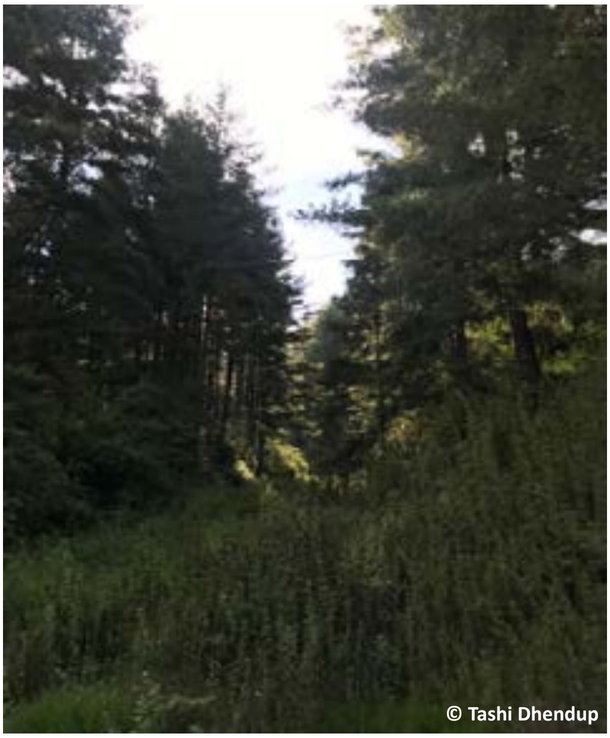

Image 2. Some parts of the study area has Blue Pine with bamboo understorey.

Image 3. Setting up a camera trap in the field.

Image 4. Himalayan Serow *Capricornis thar.*

Image 5. Tiger *Panthera tigris.*

Image 6. Asiatic Golden Cat *Catopuma temminckii.*

Table 1. Details of mammal species recorded in Lamai Goempa Research Preserve, Bumthang, Bhutan during 2016–2017 with camera trap records, total capture events, relative abundance index (RAI), naïve occupancy and latency to initial detection.

Species	Red List category[1]	Family	Camera trap records	Total capture events	RAI/Trap success[2]	Naïve occupancy[3]	Latency to the initial detection[3]
Carnivora							
Tiger *Panthera tigris*	EN	Felidae	13	3	0.07	0.09	172
Asiatic Golden Cat *Catopuma temminckii*	NT	Felidae	258	54	1.20	0.59	72
Marbled Cat *Pardofelis marmorata*	VU	Felidae	469	48	1.07	0.18	185
Leopard Cat *Prionailurus bengalensis*	LC	Felidae	71	19	0.42	0.27	36
Asiatic Wild Dog *Cuon alpinus*	EN	Canidae	620	44	0.98	0.64	105
Red Fox *Vulpes vulpes*	LC	Canidae	161	53	1.18	0.32	32
Yellow-throated Marten *Martes flavigula*	LC	Mustelidae	46	14	0.31	0.32	87
Asiatic Black Bear *Ursus thibetanus*	VU	Ursidae	225	32	0.71	0.59	32
Weasel *Mustela* sp.	LC	Mustelidae	8	3	0.07	0.09	54
Cetartiodactyla							
Himalayan Serow *Capricornis thar*	NT	Bovidae	529	70	1.56	0.55	41
Barking Deer *Muntiacus muntjac*	LC	Cervidae	766	85	1.89	0.82	33
Sambar *Rusa unicolor*	VU	Cervidae	1363	44	0.98	0.50	33
Wild Boar *Sus scrofa*	LC	Suidae	2643	152	3.38	0.91	29
Rodentia							
Himalayan Crestless Porcupine *Hysterix brachyura*	LC	Hystricidae	402	81	1.80	0.41	29
Orange-bellied Squirrel *Dremomys lokriah*	LC	Sciuridae	43	15	0.33	0.09	105

[1] LC—Least Concern | NT—Near Threatened | VU—Vulnerable | EN—Endangered
[2] Relative Abundance Index (RAI) was calculated as the number of captures divided by the total sampling effort in days multiplied by 100 (O'Brien 2011; Hedwig et al. 2018).
[3] Naïve occupancy was quantified as the number of camera trap locations at which we detected each species divided by the total number of camera trap locations (Jenks et al. 2011; Rovero et al. 2014; Hedwig et al. 2018).
[4] Latency to initial detection was determined as the number of trap nights between the start of the survey and the first record of a species (Gompper et al. 1999).

Boars had the highest relative abundance index of 3.38 and also enjoyed the highest naïve occupancy among all the species. Among the carnivores, the Asiatic Golden Cat and the Red Fox were the most common. Tiger and weasel were the least common and were found in two camera stations each. People were observed in 21 of the total 23 camera stations indicating a prominent level of human presence in the study site.

The current study was one of the first systematic camera trapping for mammal inventory outside protected areas in Bhutan and has significant conservation implications for the country as most of the available information on fauna for management decisions come from protected areas. The rich assemblage of mammals, including globally threatened species such as the Tiger and Wild Dog, suggests that the preserve is a primary habitat for many species and hence, requires protection and appropriate conservation interventions. The preserve is located near to two biodiversity-rich protected areas: Wangchuck Centennial National Park in the north and Phrumsengla National Park in the south. Given the rich diversity of mammals in the area, putting up appropriate conservation strategies could provide additional biodiversity gains. The area, however, is currently not fully gazetted as a research preserve and is under the jurisdiction of the Bumthang territorial division. As such, the area is used for selective logging, grazing, and other resource allocations for communities living nearby. Landscapes outside protected areas worldwide are facing an increase in human-induced land-use changes. This may affect species both outside and within nearby protected areas. The rich diversity of mammals in the area despite the strong human presence also demonstrates the possibility of human-

Image 7. Red Fox *Vulpus vulpus*

Image 8. Leopard Cat *Prionailurus bengalensis*

Image 9. Marbled Cat *Pardofelis marmorata*

Image 10. Himalayan Crestless Porcupine *Hysterix brachyura*

Image 11. Wild Boar *Sus scrofa*

Image 12. Sambar *Rusa unicolor*

wildlife coexistence (Moo et al. 2017). Although the tiger and a few other species were detected only a few times, the region could be an important wildlife corridor and a part of their home range (Hodge & Arbogast 2016). Therefore, we strongly feel the need to convert the area into a research preserve and be used for long term research, outreach, teaching, among others.

Image 13. Asiatic Black Bear *Ursus thibetanus*

Image 14. Yellow-throated Marten *Martes flavigula*

Image 15. Asiatic Wild Dog *Cuon alpinus*

Image 16. Barking Deer *Muntiacus muntjac*

Image 17. Orange-bellied Squirrel *Dremomys lokriah*

Image 18. Weasel *Mustela* sp.

The National Tiger Survey of 2015 recorded more tigers outside protected areas (DoFPS 2015) and also documented the presence of six species of felids and five species of small carnivores in a forest division in western Bhutan (Dhendup & Dorji 2018a,b). Unfortunately, these landscapes are highly vulnerable to habitat degradation and conversion and poaching. Therefore, as home to many threatened and endangered species, lands outside protected areas also require comprehensive conservation management plans and critical funding to ensure that these landscapes continue to sustain biodiversity in the future.

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First record of Otter Civet *Cynogale bennettii* (Mammalia: Carnivora: Viverridae) kept as a pet in Indonesia, representing a possible new threat to the species

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Otter Civet *Cynogale bennettii* is a nocturnal, semi-aquatic species of small carnivore that resembles an otter in appearance. Very little is known about the species. It was believed to be largely confined to lowland peat swamp forests in the Sunda region: Sumatra (Indonesia), Borneo (Indonesia, Malaysia, Brunei Darussalam), peninsular Malaysia, and peninsular Thailand (Cheyne et al. 2016), however, it has been recorded in lowland dipterocarp forests, secondary forests, bamboo forests, logged forests, freshwater swamp forests and limestone forests (Ross et al. 2015). It is assumed to hunt fish, crustaceans, molluscs, small mammals, and birds (Lekagul & McNeely 1977). The IUCN Red List of Threatened Species has assessed it as Endangered and in decline (Ross et al. 2015). This species is mainly threatened by habitat loss, although there are records from degraded and fragmented environments that suggest the species adapts to some degree to these altered environments (Evans et al. 2016). Other threats are silting and pollution of waterways and hunting for wild meat, as non-selective hunting and trapping methods are commonly used throughout its range. There has been no evidence of any selective hunting, including that for the pet trade and numerous research on wildlife trade in Indonesia and other range countries have not recorded them in trade before (e.g., ProFauna Indonesia 2009; Shepherd & Shepherd 2010; Nijman et al. 2014; Krishnasamy & Stoner 2016; Phassaraudomsak & Krishnasamy 2018).

During a survey carried out by TRAFFIC on the online trade of otters in southeastern Asia, a picture of a juvenile Otter Civet in a cage was discovered. The picture was posted on 6 January 2017 on the ‘Otter Lovers Indonesia X Facebook Group’, a group where people keeping otters as pets exchange information and pictures. Further research uncovered another picture of probably the same animal from 6 June 2016 on the owner’s Facebook page, and two Youtube videos from 9 June 2016 on the owner’s Youtube channel. The owner was located in Bekasi on Java, and it is unclear if the animal was being kept as a pet or whether it was for sale. The man uses an alias and posts pictures of him with different species of nationally protected animals, which raises the suspicion that he is a wildlife trader. Information on his Instagram, like his alias name, more pictures of the same animal (Image 1) and the other species of animals he displays, suggests he obtained it from the island of Borneo.

This is the first time this species has been recorded as being kept as a pet. On 15 August 2016, however, police seized two Otter Civets, among other wildlife,

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Image 1. Instagram screenshot of a juvenile Otter Civet in a cage with signboard of keepers' alias, to show actual possession and possible availability to potential buyers.

in Makassar's Soekarno Hatta Harbour (Mappesona 2016). The animals arrived on a truck from Balikpapan, East Kalimantan Province, and were hidden in boxes and baskets without destination or sender details. The police arrested two men when they were about to pick the animals up. The result (e.g., if there was a conviction) of this enforcement action is unknown.

In Indonesia, Otter Civet is categorised as Protected by the Government Regulation No. 7/1999 on Preserving Flora and Fauna Species. The Government Act No. 5/1990 Concerning Conservation of Living Resources and their Ecosystems article 21(2) states that it is prohibited for any person to catch, injure, kill, keep, possess, care for, transport and trade in a protected animal. Article 40(2) states that a person who intentionally violates those provisions is liable to punishment by imprisonment of up to a maximum of five years and a fine of up to a maximum of IDR100 000 000 (USD 7,533 at 15 September 2017 rates at https://www.oanda.com/). Information on the individual with the Otter Civet observed online was passed on to Interpol in April 2017. Since keeping an Otter Civet is against Indonesian law and the species is threatened, the poaching of any individual animal should be taken seriously. It is unknown whether notifying authorities has resulted in an investigation and what action may have been taken.

The trade in and keeping of civets in Indonesia in general has been largely unregulated, even though quotas are in place for several species and full protection status in place for others (Shepherd 2008). The trade and keeping of civets (Nijman et al. 2014) and other small carnivores, like mongooses, ferret badgers (Shepherd 2012) and otters (Gomez & Bouhuys 2018) as pets is increasing in popularity in the country. As the keeping of small carnivores is increasing, the desire to keep threatened species, like Otter Civet, may increase, leading to increased poaching and trafficking. It is therefore essential that Indonesian authorities ensure that perpetrators are prosecuted according to the full extent of the law. This account attests to how social media and the internet in general are being used to show off endangered animals, often openly, and how platforms such as Facebook are enabling the illegal wildlife trade. Enforcement against the online illegal wildlife trade is becoming increasingly important in species conservation in southeastern Asia, including in Indonesia.

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NOTE

An observation of the White-bellied Sea Eagle *Haliaeetus leucogaster* preying on Saltwater Crocodile hatchlings *Crocodylus porosus* in Bhitarkanika Wildlife Sanctuary, India

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The White-bellied Sea Eagle *Haliaeetus leucogaster* (WBSE) of the family Accipitridae is a monotypic species, closely related to other eagles, kites, hawks, and harriers. It is a resident in India and its world distribution stretches from India and Sri Lanka through southeastern Asia and the Philippines to Australia and Tasmania (del Hoyo et al. 1994). WBSE is native to New Guinea and China and all of the coastal countries of mainland southeastern Asia (Thailand, Malaysia, Singapore, Indonesia, and the Philippines), Australia and India. This species is also found in other island groups, from Bangladesh, Sri Lanka, Burma, Andaman, Laos, Wallacea, Bismarck Archipelago, Nicobars, and Greater Sundas in the west to Hainan, Taiwan, New Ireland, New Britain, and Louisiades in the east, and south around Australia to Tasmania (Strange 2000; Ferguson-Lees et al. 2001). According to IUCN Red List it is categorized as Least Concern (Birdlife International 2016). It is listed in Schedule I of the Indian Wildlife (Protection) Act, 1972 and Appendix II of CITES.

WBSE is occasionally seen in island waters along tidal rivers and in fresh water lakes (Ali & Ripley 1987). Bhitarkanika mangrove ecosystem lies along the eastern coast of India and harbours mangrove forests, rivers, creeks, estuaries, sand bars and mud flats. The resident population of WBSE in Bhitarkanika was estimated 10–15 in 2005 (Gopi & Pandav 2006), and 17–20 in 2007 (Palei et al. 2014). Apart from Bhitarkanika the species has been reported from Chilika Lake and Konark-Balukhanda Wildlife Sanctuary (Rahmani & Nair 2012)

Bhitarkanika Wildlife Sanctuary is located between 86.766–86.050 ^{0}E and 20.500–20.800 ^{0}N covering an area of 145km^2. It occupies unique habitat of mangrove forests, numerous creeks and mud flats located in Kendrapara District of Odisha. The deltaic region is a habitat with mangrove vegetation on either side of the creeks and tidal mudflat. The mangrove ecosystem is one of the largest in the Indian sub-continent and the floral diversity is the second highest in world after Papua New Guinea. Bhitarkanika is home to diverse flora & fauna out of which some are endemic. It is an ideal habitat for reptiles like Estuarine Crocodile, Water Monitor Lizard, King Cobra, and Python. Important avifauna include the kingfishers, storks, ibises, waders, and a variety of migratory ducks like Bar-headed Goose, Brahminy Duck, Gadwall, and Northern Pintail. Estimation of Saltwater Crocodile *Crocodylus porosus* population is carried out every year during the month of January.

On 13th January 2019 at about 10.00h while surveying

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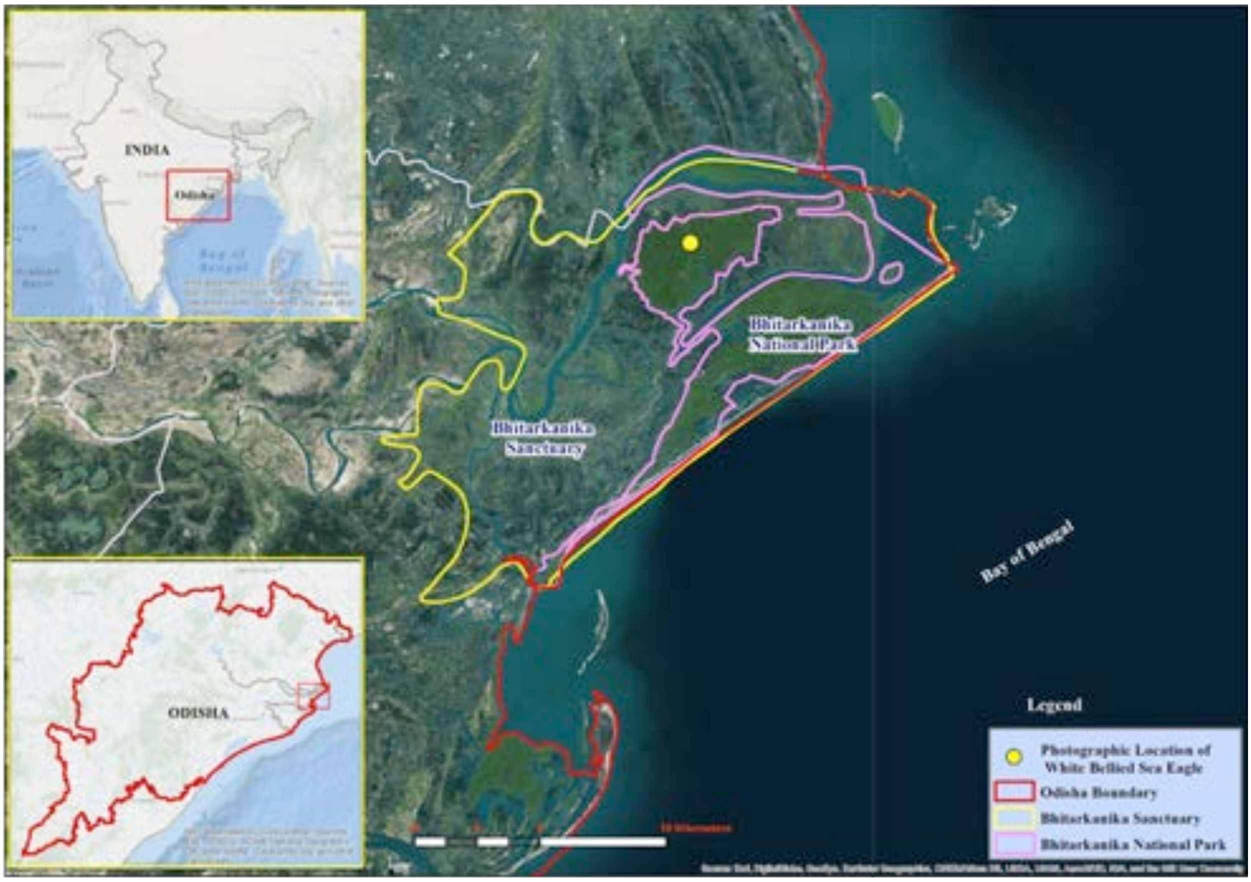

Figure 1. Sighting location of White-bellied Sea Eagle in Bhitarkanika Wildlife Sanctuary, Odisha, India.

Images 1 & 2. White-bellied Sea Eagle preying on Saltwater Crocodile hatchling in Bhitarkanika Wildlife Sanctuary, Odisha.

for the Saltwater Crocodiles, it was observed that a WBSE was gliding down to capture a crocodile hatchling in the main river of Bhitarkanika (Fig. 1; 20.733^0N, 86.869^0E). The WBSE mostly hunts and scavenges during dawn and dusk. The WBSE was able to capture the Saltwater Crocodile hatchling with precision and technique (Images 1 & 2). After capture the eagle flew to a nearby perch and started feeding on the soft dorsal portion of the body. After feeding for about 15mins the White-bellied sea Eagle flew away leaving a little portion of the body i.e. the ventral part. The hatchling was less than 2 feet long (Image 1). The WBSE was identified as an adult from its white head, breast, under-wing coverts and tail (Image 1). The upper parts were grey and the black under-wing flight feathers were in contrast with the white coverts; the tail was short and wedge-shaped.

Earlier Gopi & Pandav (2006) reported an incident of predation of a crocodile hatchling by WBSE in Bhitarkanika, but there was no photographic documentation of the incident. Iqbal et al. (2013) reported WBSE attempting to prey upon a Water Monitor *Varanus salvator* in southern Sumatra, Indonesia. Ali & Ripley (1987) and Dharmakumarsinhji & Lavkumar (1956) reported WBSE take crabs, rats, dead fish and lift domestic duck and piglets in Karwar. del Hoyo et al. (1994) also report rabbits, fruit bats, seagulls (Laridae), cormorants (Phalacrocoracidae) and gannets (Sulidae) in the diet of WBSE. Murthy & Rao (1989) observed WBSE feeding on Dog-faced Water Snake *Cerberus rhynchops* and a large-sized Wart Snake *Achrochordus granulatus* in Chilika Lake, Odisha. Rajawat (2019) captured the photographs of an adult Purple Heron *Ardea purpurea* prey upon a hatchling Mugger *Crocodylus palustries* in its beaks, along the river bank of Chambal, Palighat area.

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Elusive, rare and soft: a new site record of Leith's Softshell Turtle *Nilssonia leithii* (Reptilia: Testudines: Trionychidae) from Bhadra Tiger Reserve, Karnataka, India

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Leith's Softshell Turtle *Nilssonia leithii* (Gray, 1872) is a large freshwater turtle with a carapace length growing up to 720–1,000 mm. Being rare within its range, it occurs in most rivers and reservoirs of peninsular India, replacing the Indian Softshell Turtle *Nilssonia gangetica* of northern India (Sirsi 2010; Das et al. 2014). Except for the projected native distribution by Das et al. (2014) using GIS-defined hydrologic unit compartments, there have been no explicit records of *N. leithii*'s presence from Bhadra River flowing through Bhadra Tiger Reserve. This makes our sighting of *N. leithii*'s hatchling, the first-ever record of its presence, and indirect evidence of its nesting along the banks of Bhadra River within Bhadra Tiger Reserve (Figure 1). Based on the review conducted by Das et al. (2014) and us, our record is the third ever documented record of Leith's Softshell Turtle's hatching from the wild, with previous records from Moyar River in Tamil Nadu (Das et al. 2014) and Chalakudy River in Kerala (Nameer et al. 2007), over 25 and 12 years ago, respectively.

The individual that we observed, was a hatchling, with an approximate carapace length of 6cm (Image 1a). It was found on 7 July 2019 at 14:46h inside a puddle on a forest road that runs beside the Bhadra River, within the tiger reserve (13.435^{0}N, 75.510^{0}E). The carapace was greyish-green in colour with prominent vermiculation, running from top to bottom. The carapace also contained four eye-like concentric circular spots, with a blackish core surrounded by a reddish circle (Image 1b). The spots on the carapace were arranged as three in one row near the middle of the carapace and one near the bottom. The head of the hatchling was greenish with black streaks running from eye to the neck. The corner of the mouth was found to have yellow patches on either side (Image 1a). We observed the hatchling for over 10 minutes and, released it into the Bhadra River.

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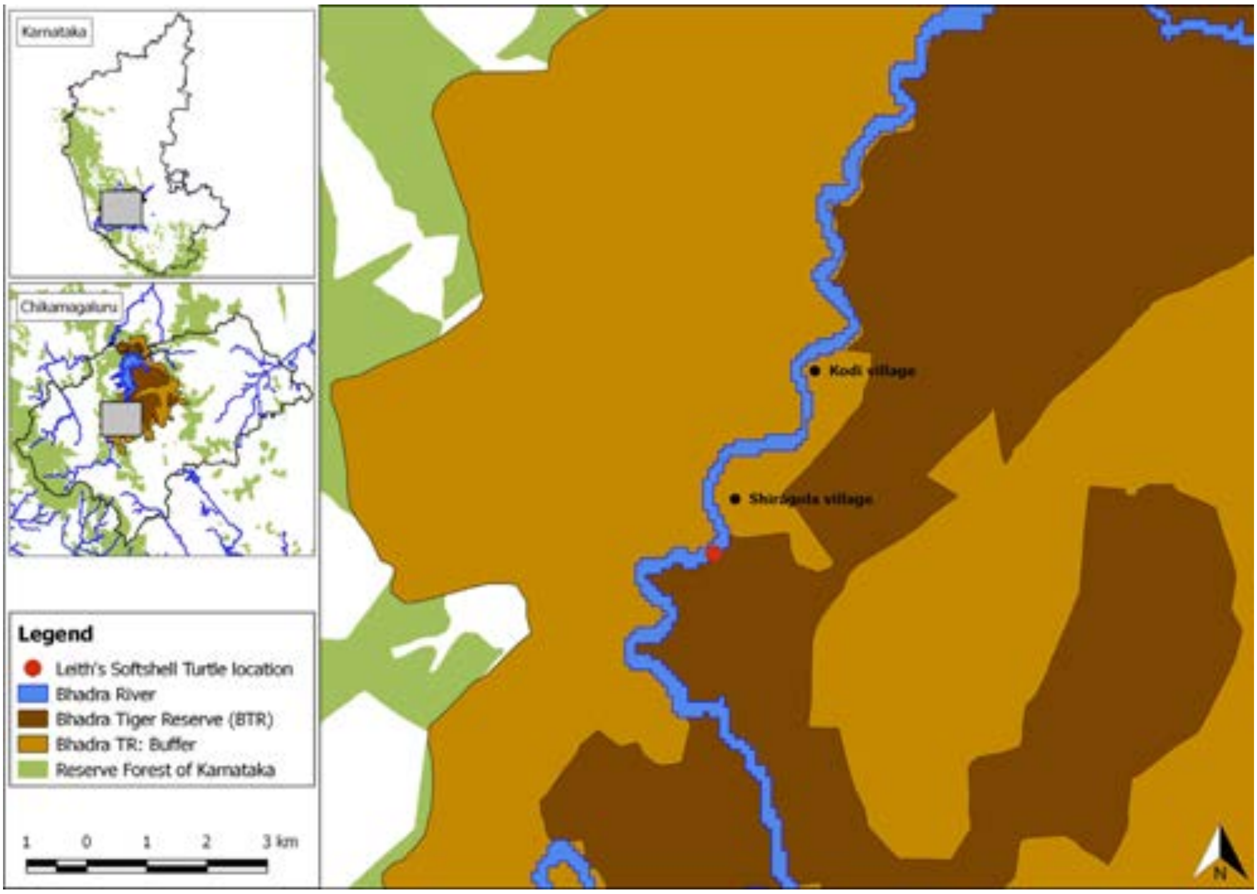

Figure 1. Location where the Leith's Softshell Turtle *Nilssonia leithii* was observed in Bhadra Tiger Reserve.

Image 1a. The observed Leith's Softshell Turtle *Nilssonia leithii* showing the yellow patch at the end of its mouth, and a reference for its size.

Image 1b. Picture of the observed Lieth's Softshell Turtle *Nilssonia leithii* showing the carapace containing four eye-like concentric circular spots, with a blackish core surrounded by a reddish circle.

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NOTE

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A new distribution record of the Pentagonal Sea Urchin Crab *Echinoecus pentagonus* (A. Milne-Edwards, 1879) (Decapoda: Brachyura: Pilumnidae) from the Andaman Islands, India

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For the first time, the Pentagonal Sea Urchin Crab *Echinoecus pentagonus* (A. Milne-Edwards, 1879) is recorded from the rocky intertidal region of the Andaman Islands. It is a symbiotic crab that lives with sea urchins of the genus *Echinothrix*. This species is recorded from the Lakshadweep (Prakash et al. 2012) and Nicobar Islands (Sastry 1981) but there is no record from mainland India. A detailed description of the species, high quality photographs and line diagrams are provided.

All eumedonines (subfamily Eumedoninae, family Pilumnidae) are obligate symbionts of echinoderms (Castro 2015). Eumedoninae consists of a total of 33 species under 13 genera (WoRMS 2019). The genus *Echinoecus* under this subfamily contains three species, *Echinoecus nipponicus* Miyake, 1939, *E. pentagonus* (A. Milne-Edwards, 1879), and *E. sculptus* (Ward, 1934) (Ng et al. 2008). They are commonly called 'sea urchin crabs' as they are obligate symbionts of sea urchins. *Echinoecus pentagonus* has a wide distribution, from eastern Africa to the Hawaiian Islands (Chia et al. 1999). Males and pre-adult females of *E. pentagonus* mostly live on the surface of sea urchins but sometimes they can be found near the rectum while adult females are restricted to the rectum in a calcified gall-like structure (Castro 1971, 2015). For the first time in India a berried female *E. pentagonus* was collected from the rectum of the sea urchin *Echinothrix diadema* (Linnaeus) from Malacca Beach, Car Nicobar in 1959 by Tiwari (see Sastry 1981). Later Prakash et al. (2012) reported a male *E. pentagonus* clinging on the ventral side of the sea urchin host *Echinothrix calamaris* from Agatti Island, Lakshadweep.

During the survey of brachyuran crabs in the intertidal regions of the South Andaman Islands, from December 2014 to September 2018, a single male specimen of *E. pentagonus* was collected in December 2015 at Corbyn's Cove, Port Blair (Figure 1). The specimen was found in rocky substratum in a free-living state. After collection, it was preserved in 10% buffered formaldehyde. Standard literature (Chia et al. 1999; Ng & Jeng 1999) were referred for identification of the species. Photographs and morphological measurements were taken with the help of a stereo zoom microscope (Leica M 205A). The specimen was deposited in the Department Museum, at Pondicherry University, Port Blair.

Material examined: One male collected from Port Blair Coast, Andaman Islands; locality: rocky intertidal region, Carbyn's Cove (11.657^{0}N, 92.753^{0}E); collected on 14 December 2015 by Balakrishna Meher and T. Ganesh; dt.02.i.2019, deposited at Museum of Department

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of Ocean Studies and Marine Biology, Pondicherry University, Port Blair (PU/MB/501).

Taxonomy

Order Decapoda Latreille, 1802
Infraorder Brachyura Linnaeus, 1758
Family Pilumnidae Samouelle, 1819
Subfamily Eumedoninae Dana, 1852
Genus *Echinoecus* Rathbun, 1894

***Echinoecus pentagonus* (A. Milne-Edwards, 1879)**
(Image 1, Figure 2)

Description: Carapace almost pentagonal in shape; length of carapace slightly more than width; surface without hairs, granules and very weakly punctuate when observed under microscope; anterior surface of carapace with white margin and two almost vertical white bands on the posterior surface (Image 1a, Figure 2a); different regions of carapace surface weakly marked; antero- and postero-lateral margins well defined. Rostrum elongated, bend downward and with a depression on centre of it (Figure 2c). Pterygostomial and sub orbital regions pitted in large amount (Figure 2b). Antennules obliquely folded (Figure 2b). Basal segment of antenna rectangular. External maxilliped quadrate; rectangular ischium and almost squarish merus; oblique suture between the ischium and merus; a vertical groove on the ischium and a large pit on the merus (Figure 2b). Surface of chelipeds smooth; upper margin of palm with a blunt spine at its distal end; carpus with one inner and one outer spine, outer spine reduced; merus with a single spine (Figure 2f, g). Walking legs smooth to poorly pitted, unarmed and subcylindrical; dactylus thorny, inner margin with a small bunch of bristles (Figure 2h). Anterior portion of thoracic sternum comparatively narrow; sutures between sternites 1 and 2 indistinct, 2 and 3 well defined, between 3 and 4 interrupted (Figure 2d). Abdomen with seven distinct segments (Figure 2e). First gonopod stout and S-shaped.

Stevcic et al. (1988) established Eumedonidae Dana, 1853 as a distinct family whereas Ng & Clark (2000) recognized Eumedonine as a subfamily of Pilumnidae family.

In the present study, the classification of Eumedonine is based on recent standard literature (Ng & Clark 2000; Ng et. al. 2008) and considered as a subfamily of Pilumnidae. The three species of genus *Echinoecus* look very similar. Sharp and longer rostrum of *E. pentagonus* readily differentiates it from *E. nipponicus* and *E. sculptus*. *E. pentagonus* is the only member of its genus represented from the coastal waters of India. Though it is mostly exclusively obligate symbiont with diadematid sea urchins (Castro 2015), in the current

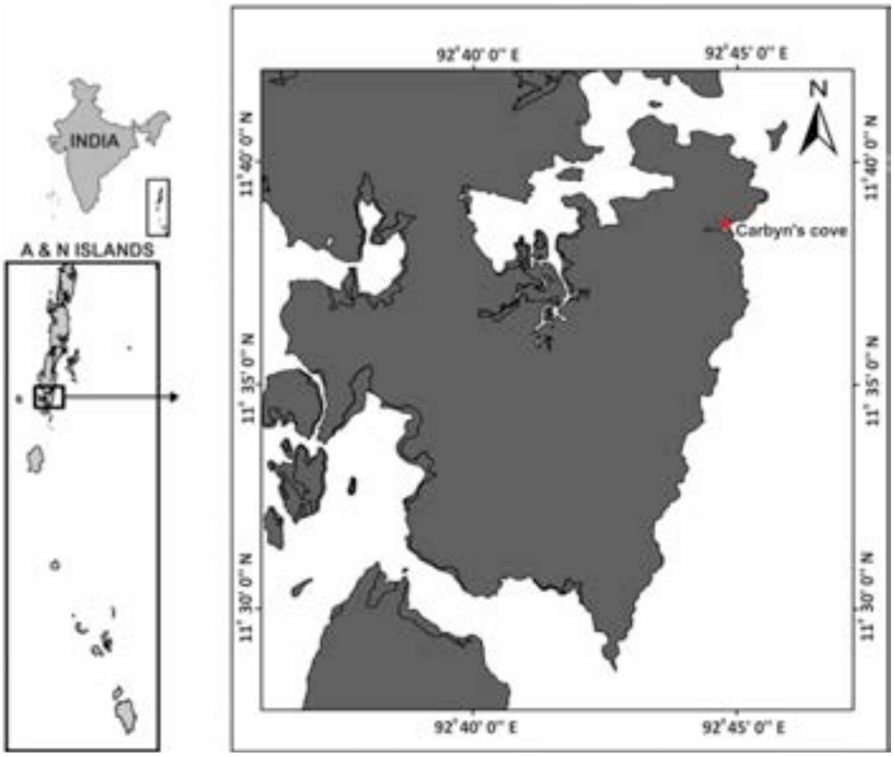

Figure 1. Sampling location of *Echinoecus pentagonus* (Herbst, 1801) at Carbyn's Cove, Port Blair, South Andaman Island.

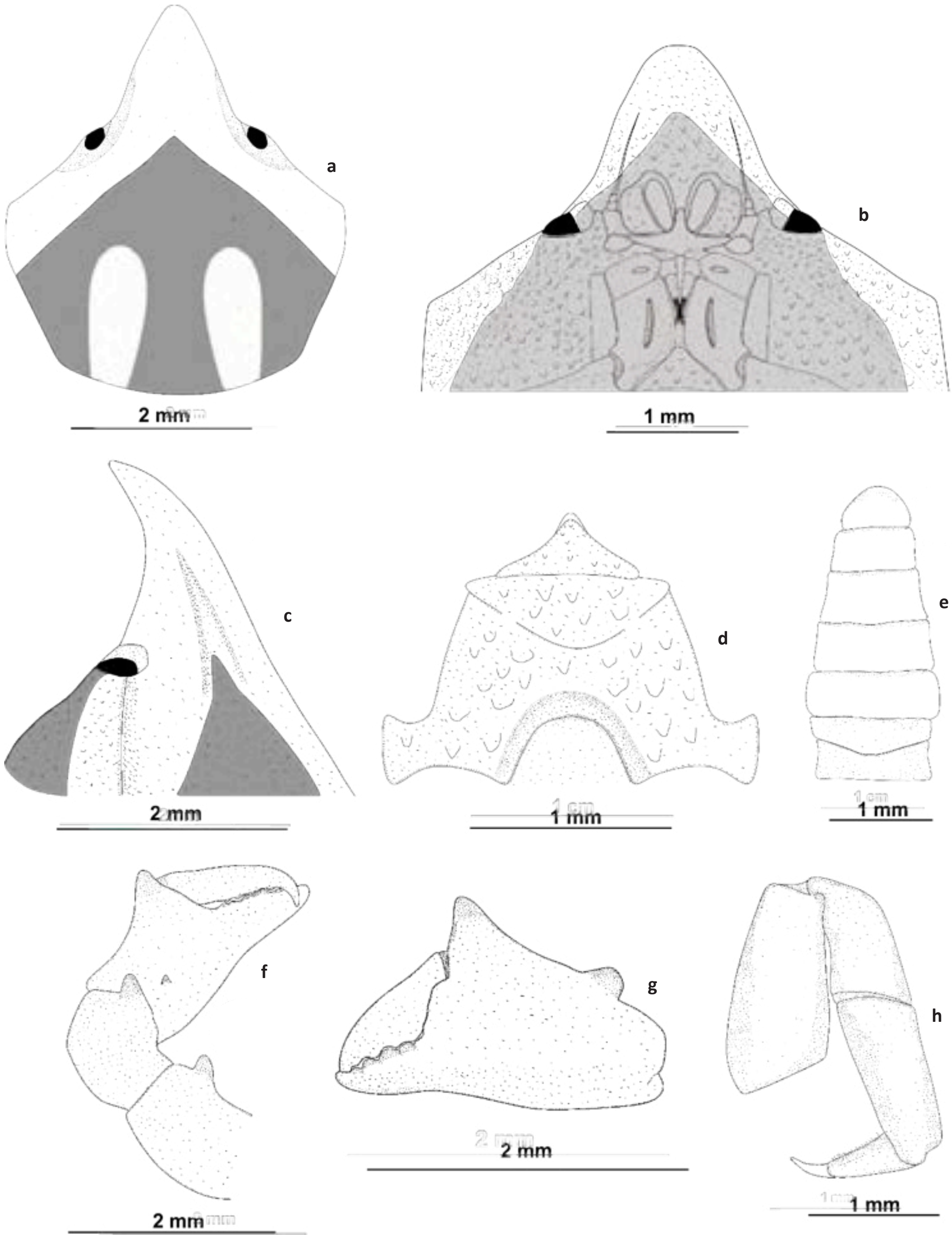

Figure 2. ***Echinoecus pentagonus*: a—dorsal view | b—ventral view | c—lateral view of rostrum | d—anterior portion of thoracic sternum | e—abdomen | f—cheliped | g—palm of cheliped | h—walking leg.**

study it was found in a free-living state, without a host, in the rocky intertidal region. It may be due to death of host or accidental separation from host. Symbiotic brachyurans can be rarely found on non-living substrates (Castro 2015).

There is almost no information on the ecology and behaviour of *E. pentagonus* from India; *E. pentagonus* sometimes shows parasitic behaviour and can be lethal to certain species of diadematid sea urchins (Castro 1971). Therefore, it is very important to understand their diversity, distribution, ecology and biology in the coast of Andaman Islands, where the sea urchin is a protected animal.

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Image 1. Echinoecus pentagonus: a—dorsal view | b—ventral view. Scale 2mm. © Balakrishna Meher.

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PLATINUM
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First records of the ghost moth genus *Palpifer* Hampson, [1893] (Lepidoptera: Hepialidae) from the Indian subcontinent south of the Himalaya

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Ghost moths are distributed over most of the world where suitable habitats are present and the family is globally represented by about 77 genera and 600 species. The family is of general phylogenetic interest because it is the largest of the families basal to many of the more diverse and derived lepidopteran lineages (Regier et al. 2015). The global geographic diversity of Hepialidae is concentrated in the general regions of central and South America, Australasia, and eastern Asia. The Indian subcontinent and Sri Lanka have a smaller diversity of only three genera – *Endoclita, Palpifer*, and *Hepialiscus* with about 26 species. These genera are also widespread across other parts of eastern Asia (Grehan 2011). Within India, the most geographically restricted genus is *Hepialiscus,* which is not recorded outside the Himalaya or its immediate vicinity (Grehan & Ismavel 2017). The most well-known genus is *Endoclita,* which comprises larger-bodied moths and species in India. There are at least seven species of *Endoclita* known from the Western Ghats, one from Sri Lanka, and a further 11 species from northern India, mostly in the northeast (Grehan & Mielke 2017; Grehan & Ismavel 2017). It is very likely that further species of *Endoclita* remain to be described as other distinct specimens from southeastern India have been reported to JRG.

In contrast to the widespread occurrence of *Endoclita*, records for Indian and Sri Lankan *Palpifer* have previously been limited to five species along the Himalayan region and a single species from Sri Lanka (Hampson 1893, 1896; Grehan & Ismavel 2017). This is a surprising distributional gap since much of western and southern India supports forested areas that would seem to provide suitable habitat. It is likely that this distributional gap represents a collecting artifact as the moths are small (wingspan less than about 33mm) and lack prominent wing markings. *Palpifer* species are also rarely reported as agricultural pests. This expectation of a collecting gap was recently demonstrated for southeastern Asia with the description of two new species from Malaysia and northern Laos, respectively. And some locality records for the distribution range of *Palpifer* are limited to personal communications or photographic records only (Grehan & Mielke 2019).

The new record of *Palpifer* in subcontinental India was made of a live individual (Image 1) that flew into a house on 21 March 2017. This occurred in a residential area interspersed with trees and shrubs and located about 10–12 km from the nearest forests. SAK photographed the moth at about 22.00h under conditions of light rain, which is typical for collecting many hepialids. Pathanamthitta is located on the lower slopes of the Pandalam Hills that

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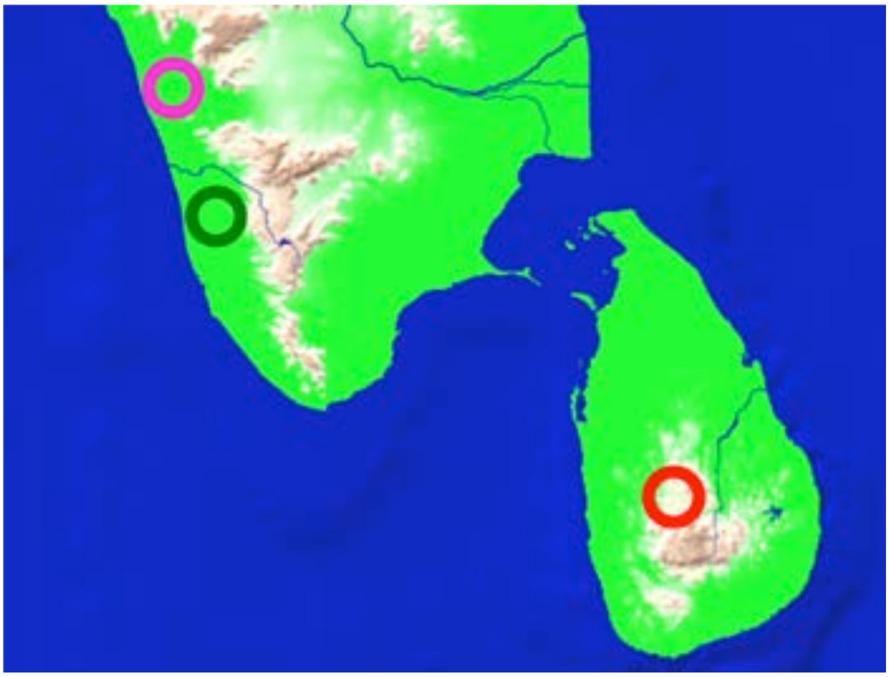

Figure 1. Distribution records for *Palpifer* in southern India and Sri Lanka: *Palpifer* sp. at Pathanamthitta (green circle) and Thumboor (crimson circle) and *P. taprobanus* at Wattegama (red circle).

Image 1. *Palpifer* sp. at Pathanamthitta, Kerala. © Siyad Karim, 21 March 2017.

Image 2. *Palpifer* sp. at Thumboor, Kerala. © Rison Thumboor, 01 December 2017.

form part of southern Western Ghats, a region that includes the Ranni Forest Division and Konni Forest Division that is the state's first reserve forest. The forests mostly comprise evergreen, semi evergreen and moist deciduous trees. Rubber and teak plantations are also very common. There are many species (mostly odonates and ants) newly described from these reserves.

Identification of the moth as a species of *Palpifer* was confirmed by the diagnostic dark chocolate brown forewings, the large white basal spot and a dark spot at the center of the posterior wing margin (Grehan & Mielke 2019). Another notable feature of the live specimen is the dorsal arching of the posterior abdomen, which might only occur in males. This behavior occurs to some extent in various other resting Hepialidae, but is particularly noticeable in *Palpifer* (Grehan & Mielke 2019). This record also came to public attention in *The Hindu* newspaper (https://www.thehindu.com/news/national/kerala/student-stumbles-upon-new-moth-species/article26156090.ece)

Two other *Palpifer* live individuals have also since been observed in Kerala State, at Thumboor on 1 December 2017 by Rison Thumboor (pers. comm. 06 December 2017) (Image 2), and at Elanthoor in January 2018 by Mebin Varghese (pers. comm. 29 January 2018). The Elanthoor record is located within an area of residential housing interspersed with forested patches while Thumboor also includes a mixture of farmland, housing,

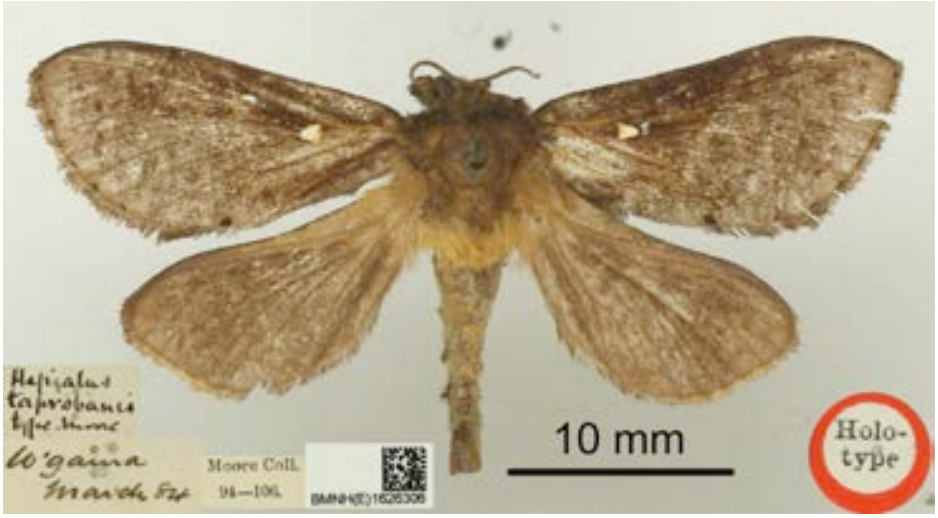

Image 3. *Holotype of Palpifer taprobanus*, Sri Lanka. (© Natural History Museum, United Kingdom). Photo by David Lees.

and forested patches. These records altogether indicate that *Palpifer* is widespread in Kerala, and likely also to be present across much of the southern Indian continent along the Western Ghats and surrounding lowlands, and perhaps also the Eastern Ghats where suitable habitat is present. The Kerala records also suggest that *Palpifer* is able to persist in residential areas in southern India, at least where adequate vegetation cover is present. Species of *Palpifer* in other parts of Asia appear to all occur in areas with forest climates ranging from tropical (India, Sri Lanka, southeastern Asia) to temperate (northern China, Korea, Japan) (Grehan & Mielke 2019).

The March and December records coincide with the beginning (March) and end (December) of the monsoon season (https://en.climate-data.org/asia/india/kerala/pathanamthitta-34524/ last accessed 09 March 2019). This periodicity is not surprising for moths where eggs drop to the ground and may not survive excessive or prolonged ground water and flooding although they require high humidity to successfully enclose. The early instars most likely live among plant debris and humus on the surface of the ground before tunneling into the soil to feed on host plant roots as this pattern of development is widespread among Hepialidae (Grehan 1989). It is very possible that *Palpifer* is an unrecognized agricultural pest in southern India. Larvae are subterranean root feeders of monocotyledonous plants and in Java and Japan; they are known to infest the tubers of some food crops (Grehan & Mielke 2019).

Future assessment of the taxonomic status of the Pathanamthitta record would ideally involve collecting specimens and making a detailed morphological comparison with the Sri Lanka species *P. taprobanus* (Moore, 1887) which is currently known from only the type specimen (Image 3) from Wattegama (Figure 1). Dr. Krushnamegh Kunte at the National Centre for Biological Sciences, Bengaluru, is interested to receive any future specimens for the purposes of description and naming if the southern Indian *Palpifer* populations prove to represent a new species.

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First record of longhorn beetle *Calothyrza margaritifera* (Cerambycidae: Lamiinae: Phrynetini) from western India

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A specimen of a beautiful cerambycid beetle, intact but in moribund state, was collected in Konkan area (Chiplun, Maharashtra, date 25.vii.2018) during monsoon of 2018, by the first author. A similar specimen was only photographed, some 200km north, in Pen, Maharashtra a few years earlier (please see https://thebutterflydiaries.files.wordpress.com/2009/07/dsc04976.jpg), but it was not collected; a blog on this species, *Calothyrza margaritifera,* was also floated on the internet. Presence of the same species again in Western Ghats indicated presence of a viable population of this Cerambycidae member in Konkan area. The purpose of this short note is to provide the first illustrated record of this species from western part of India. Since the original description of the species is quite good, we are only adding some salient points and additional illustrations in support.

The beetle was studied under Leica SMZ 6 and photographed using Canon Powershot S50 as described by Sarode et al. (2018).

The recent specimen was identified as a member of the Lamiinae tribe Phrynetini (claws divaricate, scape without cicatrix, metasternum of normal length, metepisterna short) based on classification by von Breuning (1950). It was further identified as *Calothyrza margaritifera* (Westwood, 1848) based on keys to the genera / species of Phrynetini by von Breuning (1937).

Original description of this beetle by Westwood (1848), under the genus *Phryneta*, is short but sufficient to identify this species as the colour illustration provided is equally perfect. Westwood's original diagnosis and description is given below verbatim.

In the words of Westwood: "Diagnosis: *Phryneta* of a velvety brown colour; the sides of the pronotum with two white lines and the elytra with six large irregular-shaped patches of a pearly white colour; the legs incrassated. Length of the body one and fourth of an inch. Breadth of the base of the elytra one half inch. Inhabits Nepal".

Description: "The entire insect is clothed with a fine velvety pile, with the base of the elytra rather coarsely punctured. The parts of the mouth are very short, the eyes strongly incised at the base of the antennae, which are rather short and thick, as are also the spines at the sides of the pronotum. This part of the body has two white lateral lines on each side, and the elytra have six large pearly-white spots, of irregular form, besides two minute white dots. The sterna are simple; and the legs (especially the femora) are thickened. The tips of the elytra are unarmed. The specimens in my collection, presented to me by the Rev. F.W. Hope, are smaller than

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Image 1. *Calothyrza margaritifera*: A—Dorsal view | B—Ventral view | C—close up dorsal view of head and pronotum | D—Head in frontal view | E—Head in lateral view. © Hemant V. Ghate.

the one figured, and have the sides of the pronotum occupied by a large white oblong patch instead of two lines, as in Mr. Melly's specimen".

Description of our Chiplun specimen (female):

All measurements are in mm. Total length 34; breadth at humeral angles 12; breadth of prothorax at spine 11; foreleg: femur 8, tibia, 8, tarsus 6.5; midleg: femur 8.5, tibia 9, tarsus 7; hindleg: femur 9, tibia 9, tarsus 8; antennal segments 1 to 11: 5, 1.5, 5.1, 4.6, 3, 2.5, 2.5, 2.4, 2,1.5, 1.7

Robust, medium size beetle with thick antennal segments and strong, incrassate femora. Overall colour dark brown to blackish with a bilaterally symmetrical pattern of thick, yellowish white or white patches of pubescence dorsally; this includes lateral white lines on pronotum and large patches on elytra, which do not extend to lateral margin (Image 1 A, C; Image 2A). Entire other body, dorsally and ventrally (Image 1B), uniformly dark brown and thickly covered with pubescence.

Head vertical, typical of Lamiinae; vertex convex above with a fine sulcus that extends up to base of antennae; eyes large, deeply emarginated with lower lobe much large and more than twice long as gena (Image 1C,D,E), one of the character that separates this species from similar looking *C. sehestedti* (Fabricius). Frons squarish, lower lobe of eye partly visible from front. Mandibles strong, curved at tip; labium setose; clypeus leathery and pale brown. Antenniferous tubercles strong, elevated, slightly divergent, with shallow groove in between. Antennae shorter than body, antennomeres moderately thick, scape without cicatrix.

In thorax pronotum broader than long, with maximum breadth only slightly less than base of elytra; distinct transverse grooves present near anterior and

Image 2. *Calothyrza margaritifera*: A—Lateral view showing elytral maculae | B—Mid leg tarsus and claw in dorsal view | C—As in B ventral view | D—Claw. © Hemant V. Ghate.

posterior boders; disc elevated, slightly convex above; lateral spine strong with broad base and blunt apex; a median longitudinal, smooth and shining sulcus present (Image 1C). Prosternum narrow, with rugulose, smooth and shining area near anterior margin; prosternal process slightly elevated between procoxae, flat in middle but its lateral borders elevated. Prosternal process dilated behind procoxae with its distal part almost vertical. Mesosternum with anterior border shining and smooth, with half-moon shaped shining and sunken area without pubescence; lateral parts rugulose punctate. Mesosternal process broad, raised between mesocoxae and bifid at distal tip. Metasternum broad, long, with median shining smooth sulcus (Image 1B). Elytra elongate, slightly narrowed posteriorly, rounded at apex; each elytron sparsely but coarsely punctured near base but punctures very sparse and indistinct in posterior half, thick pubescence masking all punctures. All legs rather short and robust; all femora incrassate or 'claviform', as stated by von Breuning. Tarsal segments distinct, apical one more deeply cleft dorsally, with dense light brown setae underside; claws dark brown, divaricate (Image 2 B,C,D)

Abdomen, as seen ventrally, with first segment longest and broad, its anterior tip projecting forward between metacoxae.

The type locality of *C. margaritifera* is Nepal; von Breuning (1937) mentions 'Cochinchina' (sometimes spelled Cochin-China, a colony of former French Indochina, encompassing the Cochinchina region of present southern Vietnam); a recent checklist of longicorn beetles of India (Kariyanna et al. 2017) notes only *C. sehestedtii* (Fabricius, 1798) but not C. *margaritifera*. A website on Cerambycidae, namely 'Worldwide Cerambycoidea Photo Gallery' Vitali (2019), however records 'India, Nepal, Myanmar and Thailand' as distribution of *C. margartifera*. 'ICAR-National Bureau of Agricultural Insect Resources' website also lists this beetle and an image (probably of other species) on their website, without locality data, (see ICAR 2013). Apparently, there seems to be no published record of *C. margaritifera* from India. *C. sehestdtii* has, however, been recently recorded from Madhya Pradesh (Majumder et al. 2015) and the distribution was stated as present in 'Himalayas, Madhya Pradesh (Jabalpur) within India and Sri Lanka'.

This illustrated record should, therefore, be treated as the first authentic report of *Calothyrza margaritifera* from India. It remains to be seen if this species has recently invaded or is present for a long time but was overlooked so far.

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PLATINUM OPEN ACCESS

Extended distribution of *Ceropegia mahabalei* Hemadri & Ansari (Apocynaceae) to the state of Gujarat, India

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Ceropegia L. with more than 200 species is distributed in tropical and subtropical regions of the world. Maximum diversity of *Ceropegia* occurs in southeastern Asia, India, Madagascar, tropical Arabia, South Africa, and Kenya (Meve 2002). The genus is represented by 53 species, two subspecies, and six varieties in India, of which 41 taxa are endemic to India. A majority of the species are under threat as per Kambale & Yadav (2019).

Ceropegia mahabalei Hemadri & Ansari is one of the endemic and rare species of Maharashtra and so far known from only a few localities. It was described from Ralegaon Shinde and Bhivade Khurd, the villages nearby Junnar (Pune District) by Hemadri & Ansari (1971). After type collection it has been frequently collected from type locality by botanists and amateurs for their studies and interests. Malpure et al. (2006) mentioned its occurrence from Kasara Ghat (Thane District). Pethe & Tillu (2016) collected this species from Ramshej fort (Nashik District). A new variety of *Ceropegia mahabalei* was described by Rahangdale & Rahangdale (2012) but has been merged under *Ceropegia oculata* Hook. by Kambale & Yadav (2015).

During botanical explorations of the higher ranges of Western Ghats in Maharashtra and Gujarat, the senior author collected some interesting specimens in vegetative stage from Salher (Nashik District) and Chichali Ghat (Dang District, Gujarat). When they flowered in the botanical garden of HPT Arts & RYK Science College, Nashik they were confirmed as *Ceropegia mahabalei*.

Recently, Bhagat (2018) reported it from Mulashi Tehsil of Pune District. This report shows the extension of the distribution of this endemic species from Mulashi in the south to Chinchali Ghat (Gujarat) in the North.

Ceropegia mahabalei Hemadri & Ansari
(Images 1–3)

in Indian Forester 97(2): 105. 1971; Ansari, Fasc. Fl. India 16: 24. 1984; Nayar & Sastry (eds.), Red Data Book Indian Pl. 2: 49. 1988.

Perennial erect tuberous herbs with the beaked corolla lobes.

Flowering & Fruiting: July–October.

Distribution: India: Maharashtra, Gujarat (present report); Endemic to northern Western Ghats (Fig. 1).

The localities Panchgani, Satara and Kalsubai peak mentioned by Pethe & Tillu (2016) are erroneous and mentioned neither in Mishra & Singh (2001) nor in

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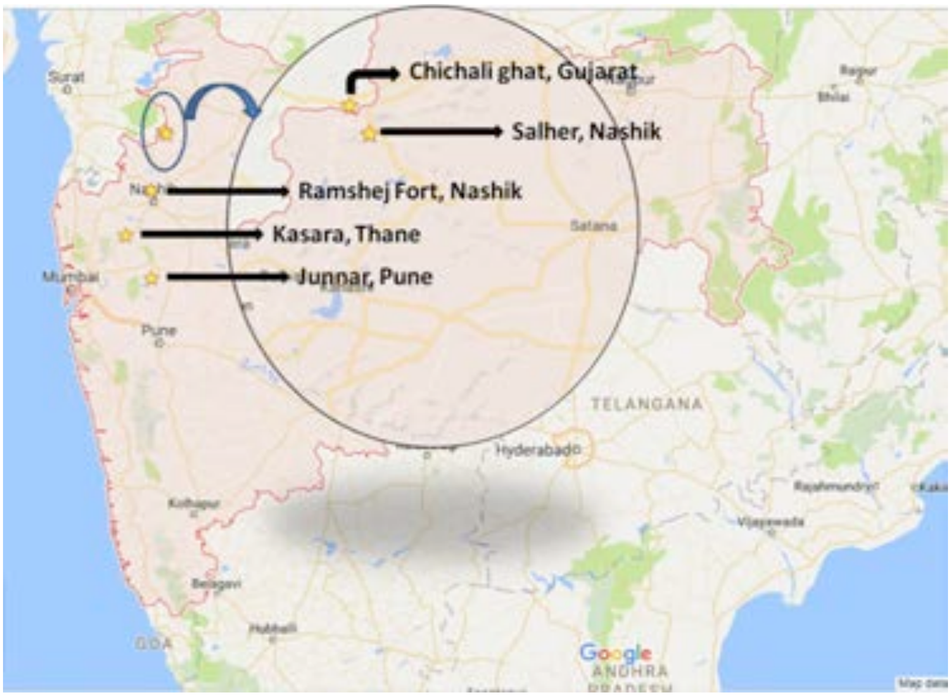

Figure 1. Distribution map of *Ceropegia mahabalei* Hemadri & Ansari.

Malpure et al. (2006).

The occurrence of this species in Kasara is not corroborated by any specimen in the Indian herbaria though mentioned by Malpure et al. (2006). Its occurrence in Kasara has been confirmed with the collection (S. More s.n.) by Sushant More. He has collected the species (Sushant More pers. comm., 2017) from Kasara and located only one individual.

Notes: *Ceropegia mahabalei* Hemadri & Ansari, commonly known as 'Gavati Kharpudi', is found growing on grassy slopes of hills. It has been reported as Critically Endangered and endemic to Maharashtra (Nayar & Sastry 1988; Mishra & Singh 2001; Yadav & Kamble 2008).

Specimens examined: India: Maharashtra, Pune District, Junnar, Ralegaon Shinde, s.d. S.R. Yadav 5812; Tuber collected from Ralegaon Shindi & grown in Garden, 19.ix.2011, S.S. Kambale & A.A. Adsul SUK-2600; 8.x.2012, S.S. Kambale & A.A. Adsul SSK-25; 19.ix.2013, S.S. Kambale & A.A. Adsul SSK-91 (SUK); Thane District; Kasara, 17.ix.2015, S. More s.n.; Nashik District; Salher, 2.viii.2016 *M*. Bhamare MB 01. Gujarat; Dang District, Chinchali Ghat, 21.viii.2016, M. Bhamare MB 02. (Herbarium, Dept. of Botany, RYK Science College, Nashik).

Image 1. *Ceropegia mahabalei* Hemadri & Ansari: A—Habitat | B—Flower | C—Flower of Junnar population | D—L.S. of flower (Gujarat population). © A—Mukta Bhamare; B–D—K.V.C. Gosavi.

Image 2. Habit of *Ceropegia mahabalei* Hemadri & Ansari.

Image 3. Herbarium image of *Ceropegia mahabalei* Hemadri & Ansari. (Reg.no: MB02).

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