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Observations on Ecology and Behavior of Two Species of Theraphosid Spiders from the Western Ghats of Goa, India; with Notes on Their Conservation Concerns

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Abstract This research addresses gaps in information on the occurrence, distribution, ecology and *in situ* behaviour of Indian Violet (*Chilobrachys fimbriatus* Pocock, 1899) and Lesser Goa Mustard or Karwar Large Burrowing spider (*Thrigmopoeus truculentus* Pocock, 1899) in the precincts of Western Ghats in the Indian state of Goa. Populations of these two theraphosid spiders were studied during the monsoon and winter seasons through 2018-19. Besides a comparative data on gross morphology and morphometry; observations were recorded on the habitat and microhabitat characteristics, distribution, density and construct of burrows across two seasons; and *in situ* behaviour of both the species. An interesting aspect of this study is the comparative fine morphology of spider silk; and assessment of the regional conservation threats of the two species, of which *T. truculentus* falls under 'Near Threatened' IUCN category, while *Chilobrachys fimbriatus* is considered to be a 'Least Concern' species.

Keywords: *theraphosidae, ecology, behavior, fine structure, pet trade, conservation, thrigmopoeus truculentus, chilobrachys fimbriatus, SEM*

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1. Introduction

Among the spiders of the world, the Mygalomorph spiders grab attention due to their size, colours, and pet trade value; yet remain poorly studied in India as elsewhere in the world [1]. The global count of recorded Mygalomorph species is 2731 under 328 genera and 16 families; the most diverse and dominant family being Theraphosidae represented by 124 genera and 946 species [2]. From India so far, a total of 111 species of the infra order Mygalomorphae distributed over 32 genera under 8 families have been documented [3]. A gross underestimation of their actual diversity is perhaps attributable to insufficient efforts at arachno-exploratory surveys and their patchy restricted distribution. Most of the studies have been taxonomically-centered, and very little information is available on the ecology and natural history of Mygalomorphs of India.

Among Theraphosidae spiders, endemism is high at the species level, and the endemic species are generally threatened with habitat loss, fragmentation and pet trade. Popularly called 'Tarantulas', they are one of the most traded invertebrate groups and many species are legally and illegally sold in the pet markets [4,5,6,7]. Red List assessments for 14 of the 53 known species of Indian Theraphosids have been carried out using the IUCN criteria and categories [8]. Of the 14 species assessed, 8 are categorised as 'Threatened' with extinction and the rest are typed either as 'Data Deficient' or 'Least Concern' due to their relatively wide range of distribution.

As for the state of Goa, a single arachno-faunal survey has been carried out by ZSI [9], but there are no dedicated studies on Mygalomorph spiders. The presence of Theraphosid Mygalomorphs has been sporadically reported from the state; literature survey suggesting that till date 4 Theraphosid species have been brought on record from the state of Goa viz., *Chilobrachys fimbriatus* [8,9,10] *Thrigmopoeus truculentus [11]*, *Thrigmopoeus insignis [9]* and *Poecilotheria regalis* [12]. However, 4 species is an acute depreciation of Theraphosid diversity in the state and is attributable to absence of dedicated active search.

Some workers have focused on the ecology and behaviour of Theraphosid species from different parts of the world, such as Uruguay [13], Seychelles [14], North America [15] and Australia [16]. Since very little is known about the Ethology of Indian Tarantulas, present work additionally ventures to address a few gaps in this direction. As these are typical ground-dwellers that live in silk-lined burrows, silk produced by both species has been examined for its fine structure.

2. Methodology

2.1. Study Site

Goa is a maritime state of the Indian west coast, and has an area of 3,702 km^2. The study site is physiographically a mountainous region close to Goa-Karnataka border, but well within the jurisdiction of the Goa State. The Theraphosid habitat investigated here is in the precincts of the Bhagwan Mahavir National Park, Mollem and is contiguous with the Wildlife Sanctuary, together a span of 240 km^2 of protected area in proximity of the Western Ghats (Goan Sahyadri) in Dharbandora taluka of Goa with co-ordinates 15°15"30' to 15°29"30'N and 74°10"15' to 74°20"15'E. (Figure 1).

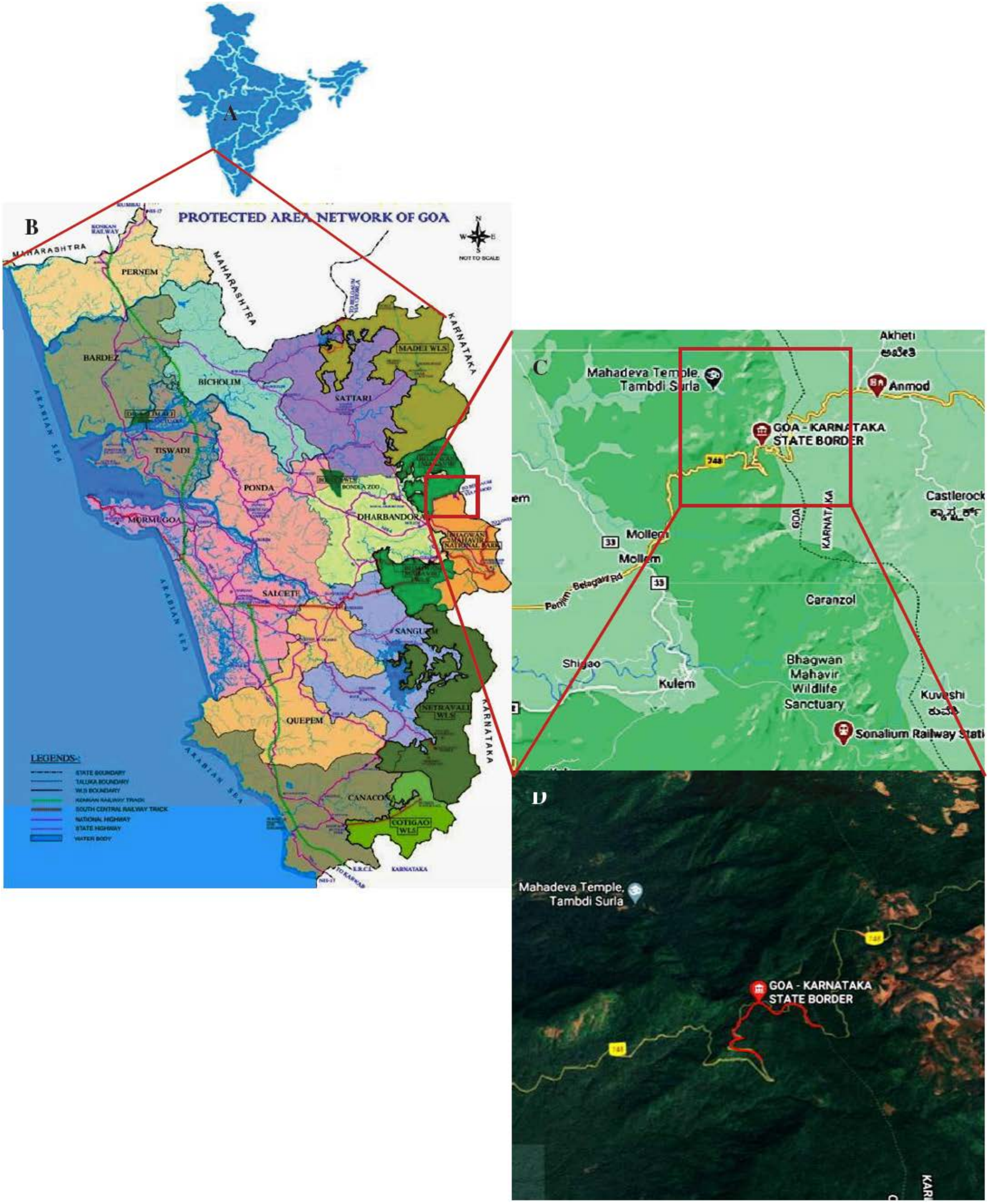

Figure 1. A- The geographic contexts of the Theraphosid spider habitat in the Western Ghats of Goa, India. **B**- The maritime state of Goa. **C**- View of the Mollem PA with a 'location pin' on the study site. **D**-Google Image with the red track marked along the NH-4A that offers the micro-habitat for the Theraphosid spiders

The area has a temperature range of 21°C-30°C and average rainfall of 2500 mm to 3000 mm. Humidity varies in the range of 75%-95%. The general vegetation here is a mosaic of Moist Deciduous forest in the lower elevations and Semi-Evergreen and Evergreen forests in the upper elevations of the Ghats [17]. The dominant trees include *Careya arborea, Dilenia pentagyna, Leea indica, Grewia tiliifolia, Lannea coromendelica, Xylia xylocarpa, Schleichera oleosa, Terminalia bellirica;* with intermittent brakes of *Dendrocalamus* and *Bambusa sps.* Owing to its tropical settings, the Goan Sahyadris shelter an impressive biodiversity. This ecological study was conducted over a stretch of 1 km linear transect along the Panaji-Anmod road (NH-4A), on the road side embankments.

2.2. Field & Laboratory Studies

Pilot bio-surveillance was carried out in consultation with experienced field staff of the state forest department to generally identify the theraphosid habitats. Perusal of secondary literature also helped in zeroing in on specific locations of Theraphosid populations in the state of Goa. The present investigation comprised of monthly visits between the months of July and December 2018 and the month of February 2019. An active search for Theraphosid burrows was carried out along the road opposite the valley, carefully scanning the embankments. For the purpose of enumeration numerous quadrats were marked over a linear stretch of 1 km. Each quadrat was examined to record the burrow densities, distribution pattern of burrows, height above the road, burrow dimensions, construction and decoration. Hygrothermal profiles of the ambience, burrow entrance and interiors were measured using a Hygrothermometer (Model HTC-1) with the accuracy of ±1°C and ±5%RH. The permanent and ephemeral flora of the embankments as well as associated fauna was recorded during every visit. Careful observations were recorded on the *in-situ* behaviour of Theraphosid spiders. After taking consent from the Goa State Biodiversity Board (Letter No.23-1-2020/GSBB/Appl/107 dated 1/7/2020), a few specimens were baited out of burrows using technique suggested by Hamilton [18], and collected in plastic containers with requisite bedding material and aeration. Later these were shifted to the laboratory for investigating morphology and morphometry using SMZ Nikon 1500-Stereomicroscope. Silk samples from webbing laid in terrariums were carefully collected and examined for fine structure using JEOL JSM 6360 LV Low Vacuum Scanning Electron Microscope.

2.3. Statistical Analysis and Pet Trade Value Evaluation

Parameters like burrow morphology and density were studied by Exploratory Statistical Analysis. Spatial distribution pattern of burrows of was investigated by Nearest Neighbour Analysis, also correlation between height from ground and width of burrows was analyzed using Pearson's correlation. To understand the collection pressure and cash value of the two concerned species in exotic animal trade, online websites were searched and reviewed in the month of September 2020 to extract data such as price, countries selling the two species and the extent of shipment worldwide. Statistical analysis was carried out using excel and IBM SPSS software.

3. Diagnosis

Both the specimens were diagnosed as female Theraphosids, and species were confirmed as *Chilobrachys fimbriatus* (Pocock, 1899) and *Thrigmopoeus truculentus* (Pocock, 1899).

3.1. Morphology & Morphometry of the Two Collected Specimen

Specimen 1:

Carapace is brown while the fovea is black. The opisthosoma is light brown but turns dark after molting in captivity. The opisthosoma shows a prominent single median and several transverse parallel black striations/chevron patterns. The legs are covered in dark violet coloured setae that are more densely packed on the femur. The uterus externus of the specimen (Figure 2A) opens out to the gonoslit. Measurements (length and width in mm): Prosoma-21 and 17, Ocular tubercle-2 and 2, Opisthosoma-22 and15, Sternum-8 and 8, Labium - 2 and 1, Maxilla - 12 and 2, Macro spinnerets -15 and 2, Micro spinnerets - 2 and 1. Pedipalp (length in mm) (coxa, trochanter, femur, patella, tibia, tarsus, total) - 9, 4, 14, 5, 9, 9, 50. Appendages (length in mm) (coxa, trochanter, femur, patella, tibia, metatarsus, tarsus, total) **Leg.1**-8,4,18,9,15,8,7,69. **Leg.2**-7,5,16,8,15,9,9,69. **Leg.3**- 6,3,15,5,14,7, 7, 57. **Leg.4**-8, 3, 19, 5, 15, 6, 7, 63. Species: ***Chilobrachys fimbriatus*** (♀).

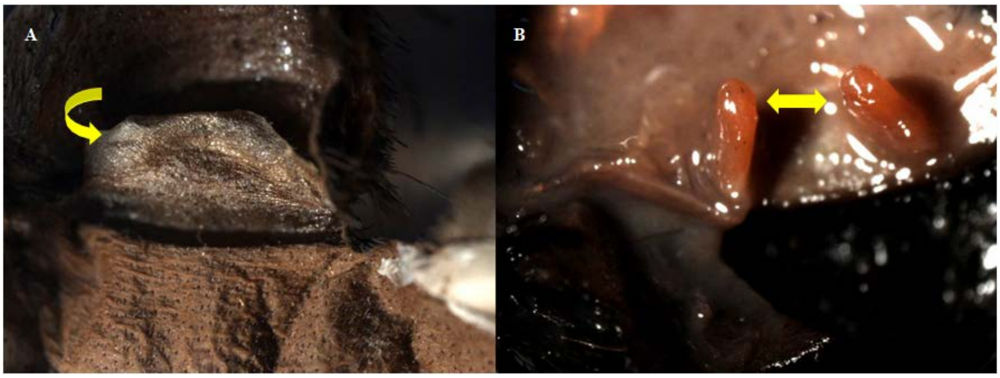

Figure 2. Stereomicroscopic Image **A.** Exuvium of *Chilobrachys fimbriatus* showing the uterus externus (Curved arrow). **B**. Bi-lobed spermathecae of *Thrigmopoeus truculentus* beneath the epigyne (Double sided arrow)

Specimen 2:

The carapace is light brown centrally and darker on the periphery. The fovea is black and two black bands on the carapace form a 'V' shaped mark. The opisthosoma is dark brown and has two spinules on the posterior end. Legs of the specimen are also dark brown colour and the femur looks black. The spermathecae were observed to have two lobes, one on each side of the structure (Figure 2B). Measurements (length and width in mm): Prosoma-19 and 18, Ocular tubercle-2 and 3, Opisthosoma - 21 and 8, Sternum - 9 and 9, Labium - 2 and 1, Maxilla - 15 and 4, Macro spinnerets - 15 and 3, Micro spinnerets - 7 and 2.

Pedipalp (length in mm) (coxa, trochanter, femur, patella, tibia, tarsus, total)-8,4,15,6,9,6,48. Appendages (length in mm) (coxa, trochanter, femur, patella, tibia, metatarsus, tarsus, total) **Leg.1**- 10,5,20,6,15,8,9,73. **Leg.2**-7,4,20,6,13,8,7,65. **Leg.3**-6,4,14,7,11,9,6,57.**Leg.4**-7,5,18,6,11,12,7,66. Species: ***Thrigmopoeus truculentus*** (♀).

4. Observations and Results.

4.1. Hygrothermal Profile, Light Intensity

The hygrothermal profiles of the habitat and numerous burrow tunnels were recorded over several visits during the monsoon and winter. The average temperature at the entrance of the burrows was 25.58°C, while he temperature inside burrows averaged at 30°C. The minimum temperature recorded inside a burrow was 22.5°C and maximum was 31.4°C. It was consistently observed that temperature inside the burrow tunnel is higher or the exactly the same as that at the entrance, difference ranging between 0 and 6.2°C. As for the Relative Humidity (RH), higher values were recorded inside the burrow than at the entrance. The relative humidity (%) at entrance ranged between 74.81% - 97.2%, while for the tunnel the range was 75.62% -98%. Burrows were preferentially excavated in the portions of embankment underlying dense tall flora and consequently a good canopy cover that shields against intense insolation. During the day the light intensity measured at the burrow entrance averaged to be approximately 9437 lux during the winter seasons and 1867 lux during the monsoons.

4.2. Floral Diversity and Associated Fauna.

Many other species share the same habitat as that occupied by the two investigated species of Theraphosid spiders and influence each other although occupying different niches. The embankments on which the burrows were dug by the spiders were predominantly covered with herbaceous flora, and in the monsoon season the micro-habitat was covered in lush green vegetation which conspicuously decreased post monsoon. The flora recorded included *Boehmeria platyphylla, Zingiveraceae sp., Strobilanthes ciliates, Adiantum sp., Caryota urens, Ipomea sp, Carallia brachiata, Macaranga peltata, Cheilocostus speciosus, Impatiens acaulis, Leea indica, Glycosmis pentaphylla.*

The faunal species documented on the embankments varied seasonally. Monsoon season shows the presence here of leeches, earthworms, snails, stick insects, variety of caterpillars, grasshoppers, millipedes etc.; while the post monsoon season brought to the embankments snakes, geckos, ants, moths, wasps, roaches etc. Other invertebrate associates were Glassy Tiger butterfly *(Parantica aglea), Owl* Moth (*Erebus macrops),* Funnel Web spider (*Hippasa agelenoides),* Giant Wood Spider *(Nephila pilipes),* Fulvous Forest Skimmer Dragonfly (*Neurothemis fulvia*), *Oriental Hornet* (*Vespa orientalis*), Stick insect (*Carausius morosus*), Giant Pill Millipede (*Arthorpshaera sp.*).), Land Snail (*Macrochlamys indica).* During nocturnal visit; herpeto-fauna was active and included Common Indian Toad (*Duttaphrynus melanostictus*), Indian Bull Frog (*Hoplobatrachus tigerinus*), Common Indian Tree Frog (*Polypedates maculatus*), Prashad's Gecko (*Hemidactylus prashadi*), and Asian House Gecko (*Hemidactylus frenatus*) were seen. The snakes documented here include Checkered Keel back (*Xenochrophis piscator*), Green Vine Snake (*Ahaetulla nasuta*), Hump-nosed Pit Viper (*Hypnale hypnale*) and Indian Cobra (*Naja naja*). Also foraging troops of Bonnet Macaques *(Macaca radiata)* were commonly spotted in the area.

4.3. Burrow Density, Diameter and Distribution Patterns

Most of the Theraphosid spider burrows were sighted on the mud bund running parallel to the road (NH-4A) in the described habitat. *C. fimbriatus* and *T.truculentus* occur in a small linear stretch of this habitat; but despite sharing the general habitat, *C. fimbriatus* lives in relative isolation and is solitary unlike the communal *T. truculentus*. The *T. truculentus* burrow density (Number/ m^2) in the stretch of 1 km linear distance using multiple quadrats was found to be 0.91±0.51/m^2.

Table 1. Descriptive analysis of burrow dimensions

Descriptive parameter ⇨ / Sample Characteristics* ⇩	N	Min	Max	Range	Mean	Median	Standard Deviation	Variance	Skewness
Burrow Entrance Diameter	40	0.4	4.1	3.7	1.775	1.5	0.01	1.023	1.088
Burrow Tunnel Diameter	40	0.4	3	2.6	1.26	1.1	0.561	0.315	0.992
Difference between Entrance and Tunnel Diameter	40	0	2.3	2.3	0.512	0.2	0.631	0.399	-
Height from the Ground	28	24	92	68	59.079	56	-	-	0.1738
Nearest Neighbour Analysis (Rn value)	**Inference**								
1.053	Randomness								

*All values in Inches, N: Number of observations, s: Standard Deviation, v: Variance, skp: Skewness.

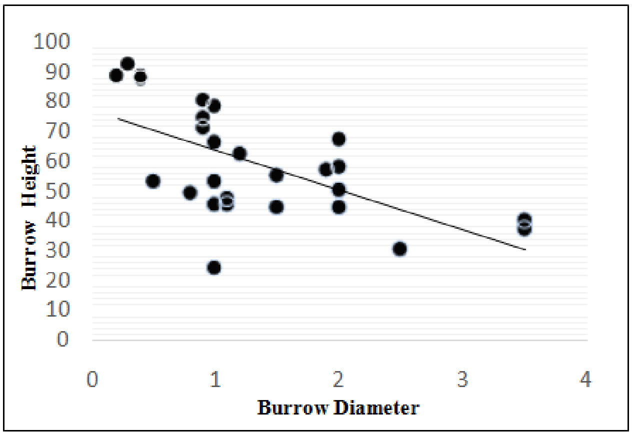

Figure 3. Scatter plot showing correlation (r= -0.614) between burrow diameters and their corresponding heights from the ground level. N=28.

Nearest Neighbour (Rn) analysis was carried out to evaluate spatial distribution and results indicated a random distribution, with Rn value of 1.053 at significance 95%. (Table 1). The entrance diameter of 40 burrows on examination revealed the median diameter to be 1.5" (s=1.088), while the smallest entrance diameter was 0.4" and biggest burrow had a diameter of 4.1". The analysis shows a moderately positive skewed data with skp = 1.088, which indicates that the number of burrows with smaller entrance diameters are more. The average value of tunnel diameter was 1.26"(s=0.561) with minimum being 0.4" and maximum being 3". The variance in entrance diameters was much higher than tunnel width values. The difference between tunnel and entrance diameter was 0-2.3" with very low variance. (s=0.631, v=0.399). The height of the embankment was not uniform and on measuring location heights of individual burrows from ground level, it was revealed that average value was 59.079" while lowest and maximum height value were 24" and 92", respectively. Skewness value recorded was 0.173 indicating that almost all burrows were approximately located close to the mean height. The association between diameter and height of the respective burrows has been found to be negative as r value is - 0.614. The correlation data represented in Figure 3 indicates that as the height from ground increases, the burrow diameter decreases; which means the smaller burrows are located higher on the embankment than the larger burrows.

4.4. Comparative Ethogram of the Two Theraphosid Species *in situ*

Chilobrachys fimbriatus though wide spread in its range, its population has a random distribution and the species is more solitary (Figure 4A) than *Thrigmopoeus truculentus;* with which it shares the habitat. Unlike *T. truculentus* whose burrows are located close to one another, *C. fimbriatus* conspecifics do not excavate burrows in the immediate proximity of each other, and were also seen occupying crevices on tree trunks (Figure 4B) The entrance of *T. truculentus* burrow is often shaped like a trumpet-bell; the rim of which is consolidated with wet mud and spider silk (Figure 5A). In some cases a porch like entrance is seen due to extended dorsal rim of the burrow. Often the burrow entrance is reinforced with adjacent twigs, dry leaves (Figure 5B) and other such artifacts that prevents it from collapsing, as also offers an extended passage during entry and exit for the inmate of the burrow.

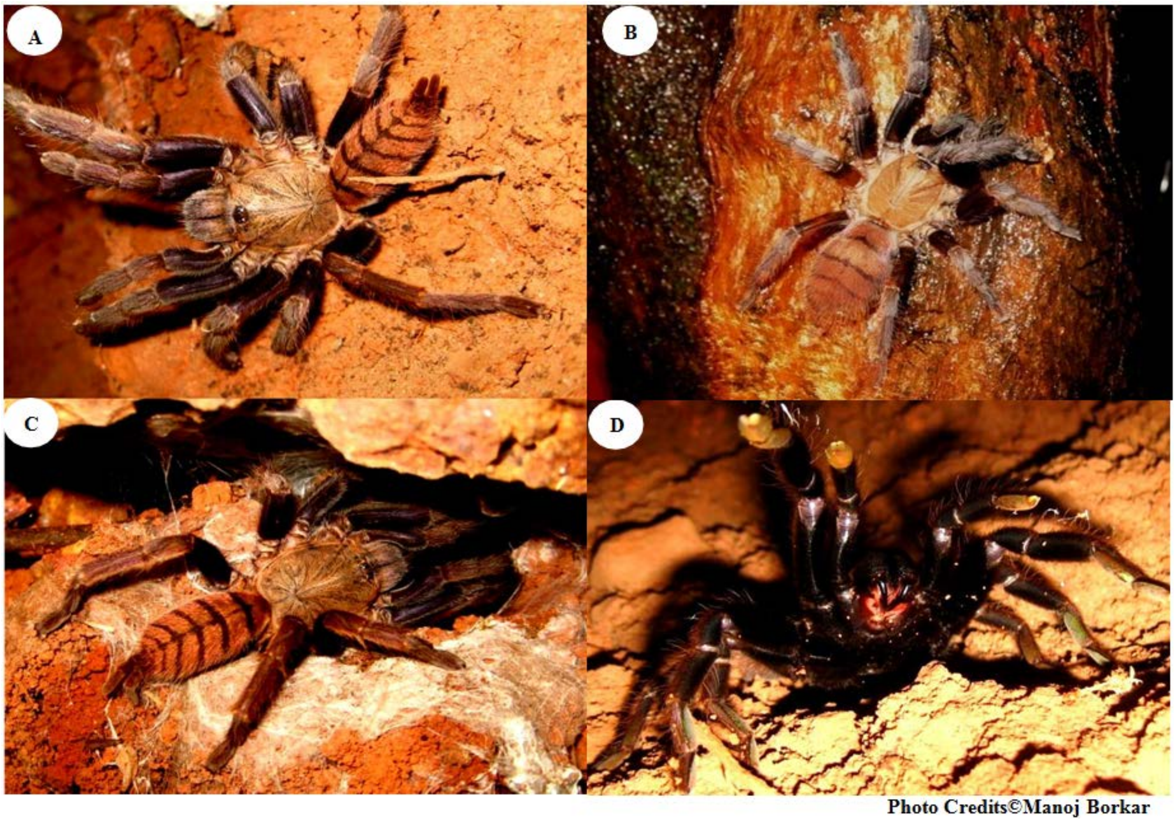

Figure 4. A. *Chilobrachys fimbriatus* on mud embankment. **B**. Arboreal individual of *C. fimbriatus*. **C.** *In situ* burrow occupancy of *C. fimbriatus*, note the silk at the entrance. **D**. An individual in with a deimatic display

Figure 5. A. *In situ* burrow occupancy of *Thrigmopoeus truculentus.* **B.** Entrance buttressed with a dry leaf. **C.** An individual on mud embankment**. D**. Threat posture of *T.truculentus* with a deimatic display

There is a striking difference in the way the two species use silk in their burrows. *C. fimbriatus* is a liberal Webber and exudes expansive and criss-cross sheets of silk, lining the burrow cavity and over the entrance (Figure 4C), as against *T. truculentus* which is conservative in its use of silk. In case of the latter species, the deeper chambers of the burrow are lined with thick layers of silk. The SEM studies of silk of both the species reveal clear structural differences that could be of strategic significance. Also, it was observed during the nocturnal field visits that *C. fimbriatus* takes relatively longer excursions away from the burrow than *T. truculentus* that stays close to the burrow for a quick retreat (Figure 5C). Theraphosid species do not weave webs as such, but use silk only for lining burrows internally; losing out on advantage of web strand light reflection if any. Interestingly, when both the species sit at the entrance of their burrows, the pedipalp and the first pair of legs protrude out with their scopulate metatarsal and tarsal segments and the pedipalpal tarsi displaying iridescence. *Chilobrachys fimbriatus* if continuously nudged resorts to brief thanatosis (posture feigning shock or death), though anachoresis (living hidden from predators in crevices, beneath bark or in holes in the ground) seen in both the species remains the most effective anti-predatory strategy. However, for food and mating the individuals do emerge from their hideouts.

When threatened, both the species engage in stridulatory hissing followed by deimatic (startling or frightening) display; raising themselves with their front legs, pedipalps spread apart and fangs bared (Figure 4D, 5D). The tarantula activity pattern is linked to changes in light intensity. The initiation of activity is always at sunset. Most individuals of *T. truculentus*, were observed plugging the entrance of the burrow with a mix of mud and silk as the winter progressed. The light intensity at the time of resumption of activity immediately after sunset was averaged at 448 lux.

4.5. Ultra-morphology of Silk in Thrigmopoeus Truculentus and Chilobrachys Fimbriatus

In captivity; *C. fimbriatus* is an aggressive weaver and covers the entire terrarium with its silk overnight, crafting a cantilever burrow entrance made entirely of silk fibres; whereas *T. truculentus* is more conservative with expenditure of silk; however the configurations of silk fibres were strikingly different and species-specific.

In both the cases the species weave simple ‘sheet webs’ without any tactical designs as such. Two distinct structural elements can be visualized; namely Attachment Fibres (AF) and Connecting Fibres (CF) distinguished by their density, orientation and thicknesses.

The foundation on the substratum comprises of attachment fibres, upon which the connecting fibres are attached to consolidate the silken sheet. In *T. truculentus* the AF are dense and occur in a criss-cross pattern with regularly interspersing CF (Figure 6A). Regions with the highest concentration of nanofibrils appear in the scanning electron micrograph as whitish spots without clear borders. Also it is seen that in case of *C. fimbriatus* silk (Figure 6B), that before joining with the silk of attachment fields, the thick connecting fibers branch out into several thinner fibers with fewer nanofibrils.

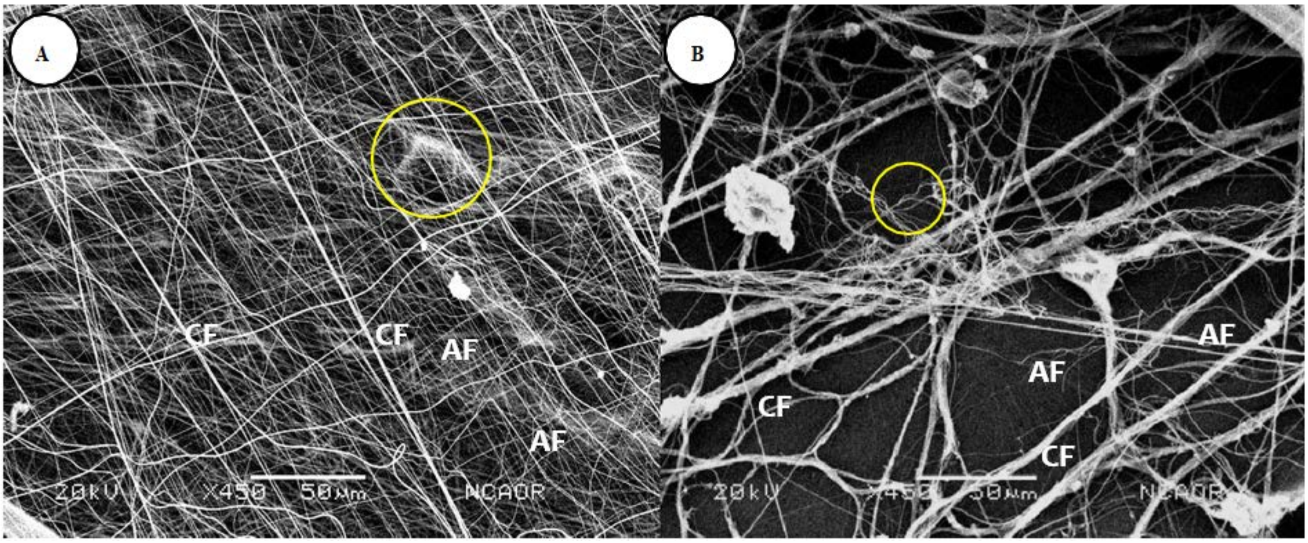

Figure 6. SEM of silk sheet(X 450) collected from the terrariums of Theraphosids in captivity. **A & B.** Scanning Electron Micrographs of silk sheet of *Thrigmopoeus truculentus* and *Chilobrachys fimbriatus* respectively. **CF**-connecting fibres. **AF**-attachment fibres. **A. Yellow Circle**-Dense concentration of Nanofibrils. **B. Yellow Circle**-Thinner fibers with few Nanofibrils

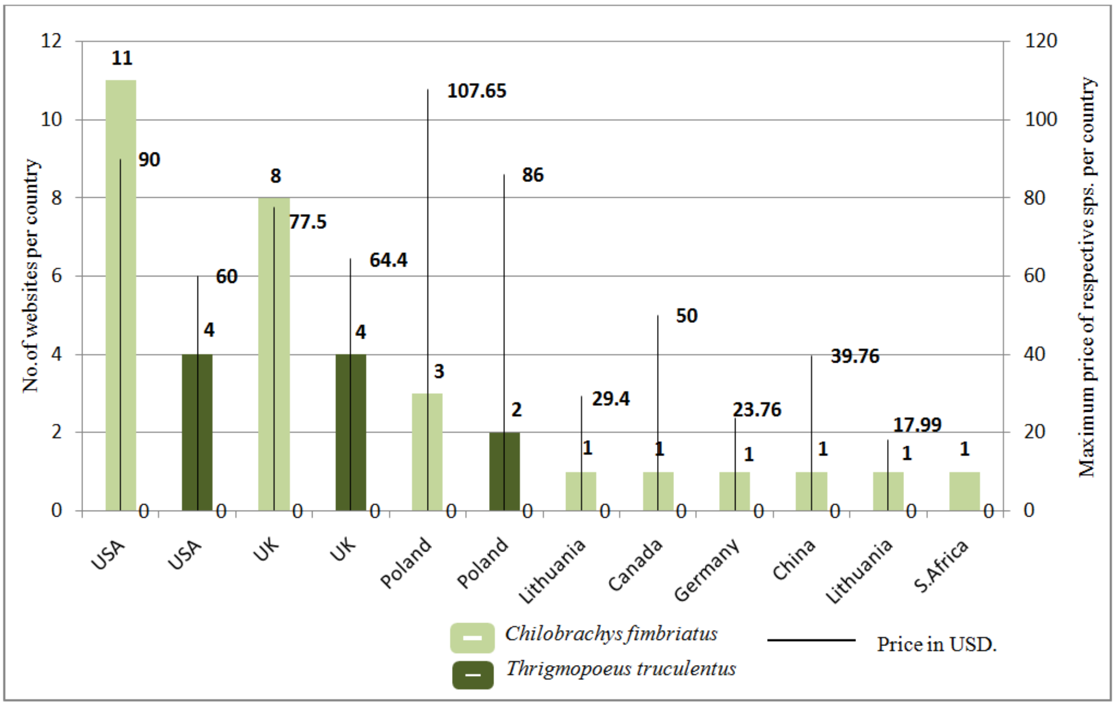

Figure 7. Data on Pet Trade (2020) involving *C. fimbriatus and T. truculentus.* Bar data represents the number of websites per country (refer the left axis) selling the respective species. The lines overlaying bars indicate the maximum price (in USD) per country (refer the right axis)

4.6. Conservation Concerns of the Theraphosidae Spiders

4.6.1. Exotic pet Trade

While *Chilobrachys fimbriatus* has been in the limelight for exotic pet trade internationally, *Thrigmopoeus truculentus* is in lesser demand and was 'in stock' for purchase only on 3 websites (Figure 7). Analysis of the online pet-trade portals confirms that *C. fimbriatus* is sold on more sites and at competitive price as less as 17.99 USD/per spider in Lithuania, Europe. The online websites selling the two species were addressed to numerous international locations; with the United States having the highest number of sites. This online surveillance revealed that tarantulas of all sizes and both the sexes were being sold, along with accessories for captive maintenance. Many websites candidly stated that the exotic spiders for sale were indeed collected from the wild. *Chilobrachys fimbriatus* is being shipped to countries that include USA, Canada, UK, UAE, Australia, China, Japan, South Africa, Indonesia, Germany, Vietnam, Ireland, Singapore, and Lithuania. However, the demand for *T. truculentus* is much lesser, with buyers mainly from UK, USA, Germany and Poland. With increasing demand for tarantulas as pets, there has been an exponential increase in illegal trade and captive breeding effort.

There are indications that locals are usually hired for collection, the spiders are than smuggled to breeders/collectors.

Figure 8. Theraphosid spider habitat degradation and destruction during road widening of section of NH4 (Panaji-Anmod road) near Goa-Karnataka border. A. Embankment with Theraphosid burrows in the year 2018. B. Same area of the embankment in February 2019 after being scrapped for road widening. C. Site of road widening project on NH- 4A. D-Excavator used for the scrapping of embankment

4.6.2. Linear Infrastructure, Habitat Destruction and EIA Context

Currently in the state of Goa, three projects have brought to the fore, the apprehensions on the impact of linear infrastructure on biodiversity of the state's Protected Areas. Of these, the four-laning of the National Highway 4A (153 km in total length and 70.07 km falling within Goa) shall have 13 km transecting the Bhagwan Mahavir National Park, Mollem. This project has already been granted 'Wildlife Clearance' by the Standing Committee of the National Board of Wildlife (NBWL) through a virtual appraisal meeting during the Pandemic lockdown period. The EIA for the four-laning project in its 'Biological Environment' section does not mention under the 'Endemic or the Threatened categories', any of the Theraphosid species studied here; despite their occurrence in the core zone of the project site.

The authors have been closely monitoring this site since 2016, where both the investigated Theraphosid species occur. While in 2016 and until late 2018, the Theraphosid habitat was secure and population dynamics normal (Figure 8A); from February 2019 large stretches of these roads opposite the valley have been worked upon for widening purpose (Figure 8C). Specifically, the continuous bench of stabilized soil strata that presents as a road side embankment has been intensely scrapped with hydraulic excavators, damaging the burrows and destroying the vegetation (Figure 8B, Figure 8D). Recognition and integration of ecosystem services in the developmental blueprint and the associated land-use planning is unceasingly sidelined in our rush for manufactured capital. Though all the stakeholders accept that some extent of biodiversity loss is inevitable, it is rarely explicitly and empirically expressed during the impact appraisal process. Apprehensions have been red flagged by the opponents on the perceived impacts of road widening of NH-4A. The project proponents have submitted the EIA report as mandated by law. However in this report the baseline data of the biological environment of the proposed site does not even have a cursory mention on the occurrence of these two Theraphosids.

5. Discussion

Studies of the biology, ecology, and taxonomy of spiders have been delayed compared to other groups [19,20,21]. Until now, most studies on faunal taxa of the Western Ghats; including those pertaining to Mygalomorphs spiders have been species documentation, inventorisation and brief observations of their natural history [1]. Effort at species description is comparatively smaller and a glaring lacuna exists in so far as ecology of these arachnids is concerned**.** In tarantulas, these studies are even scarcer; chiefly due to their reclusive habits and the difficulty of obtaining specimens [22]. Besides their species diversity value, ground-dwelling spiders are also implicated in eco-energetics, linking subterranean detritus food webs to surface terrestrial food webs [23]. Spiders are also seen as having a high bio-indicative value as they are emphatically associated with biotopes [24].

Present study is the first attempt to document and investigate populations of two Theraphosid spider species from the state of Goa, with analysis of their ecology, behaviour and conservation future. Interestingly, the presence of *T. truculentus* from Mollem in Goa had been reported [11], though the paper describes the specimens collected from Coorg in Karnataka and Amboli in Maharashtra. It has also been reported that in Mollem and Cotigao WLS of Goa, burrows of *C. fimbriatus* are commonly encountered on tree holes, trunks and in depression of forked branches in addition to those on the roadside bunds [11]. These researchers have also confirmed that in the Northern W. Ghats of these three states, the species is reportedly endemic.

The 2 species of Theraphosidae studied here have contrasting appearances and distinct morphological features as studied using the stereo-microscope. Apart from the conspicuous violet colour and opisthosomal striations of *Chilobrachys fimbriatus*, the structure of the spermathecae (uterus externus for *Chilobrachys* sps.) helped identify and distinguish the species from the *Thrigmopoeus sps.* , which has a characteristic two lobed spermathecae. The diagnostic morphological and morphometric characters of both the investigated specimens tally with those recorded previously for the respective species [11,25].

For a species, a habitat is not alone a physical space for occupancy, but also a resource base that offers food, mates and retreat from environmental adversities. Evidence is growing that within the larger habitat, microhabitats serve as ‘rescue sites’ that allow the species to repopulate in event of catastrophic decline in population in the larger environment [26,27]. As such the selection of habitat is critical dimension of behaviour and can decide the species vulnerability to environmental changes [28,29]. Co-habitation is normally reported among conspecifics and is usually a mating strategy [30]. There have been reports of co-habitation between a microhylid and a mygalomorph spider on Rameshwaram Island, off the south-east coast of India [31]. However, in the present study no organisms were found to be cohabiting the theraphosid burrows, though Owl moths (*Erebus macrops*) were seen around the burrow entrance, as also slimy trails of snails (*Macrochlamys indica*) were found along the rim of burrow. In absence of direct observations, trophic relationships between these species and the spiders are speculative.

Defense mechanisms that have evolved in spiders in response to predation have been largely ignored in Arachnology literature [32]. Though crypsis (camouflage) operates in reducing predatory pressure and most cryptic spiders prefer barks of trees; arboreal individuals of *Chilobrachys fimbriatus* with their strikingly dark opisthosomal bands do not seem to be at an advantage from this perspective. Even though the arboreal habitat spider sub-guild corresponds best with desiccation resistance, it is well established that morphological traits also match with the ecological preferences of these species [33].

Climatic variables of the habitat such as sunlight, hygro-thermal profile, and precipitation are known to influences spider behaviour such as microhabitat selection and also reproductive success [34,35,36,37]. In the forest ecosystem the light passing through the canopy, reaching the understory as also the ground, varies in intensity and spectral quality. Such differences directly affect visual communication and plant-animal interactions, thus influencing habitat selection [38]. Moderate correlation suggests that smaller burrows are occupied by juveniles at higher levels on the embankment. This hints at ‘site selection behaviour’ by the juveniles which could be the due to the heavy rainfalls during the monsoons here; which results in splatter of water and mud into the burrow. That the upper half of the embankment being shaded by forest canopy receives lower light intensity could also be the incentive for burrows being preferentially excavated in the upper portions of embankment underlying dense tall flora. But the overall distribution pattern of burrows evaluated using ‘Nearest Neighbour’ analysis revealed randomness of burrow distribution generally.

Changes in light intensity offer a cue for activity pattern for theraphosids [38]. In fact tarantulas were considered to be nocturnal animals [39], until that impression was challenged [40]. After a prey meal, satiated tarantulas may stay within the burrows remaining out of sight for considerable length of time. Though in the present study, both the species of theraphosid spiders were visible at the entrance of their burrows even in the daylight hours; with *T. truculentus* exhibiting a characteristic waiting posture with the front legs and pedipalps projecting out of the tunnel.

In so far as buttressing of burrow entrance with dry leaves by *T. truculentus* is concerned, it is tempting to suggest that the leaves used in constructing the burrows could also serve as an enticement for insect prey that could simply crawl over it into the burrow; however, this needs to be tested. Some workers have suggested use of spider silk near the burrow entrance as ‘Trip-lines’ that could assist in prey detection [41]. It has been reported that portions of spider webs with certain reflective pattern in UV light may resemble flowers thus attracting and trapping insect [42]. However, Theraphosid species do not weave webs as such, but use silk only for lining burrows internally, losing out on advantage of photonic reflection if any. The display of iridescent tips of appendages at the burrow entrance as described previously could be a tradeoff for non-reflective web. The iridescent ventral tips of the legs when raised certainly amplify the deimatic function, but it is also tempting to implicate this light reflectance in prey attraction. The plugging of burrows through winter as observed in *Thrigmopoeus truculentus* has also been previously reported in *Aphonopelma chalcodes [38]*.

The complexity of constituents in the silk fibers remains poorly understood [43]. Types of silk that spiders secrete depends on the purpose and various types have been documented [44,45]. Despite the diversity of fibres, the underlying mechanisms of silk production remain similar [46]. Functional diversity of spider silk strands is attributable to differences in protein configuration and morphology. There were differences observed in silk weaving behaviour, which also reflected in the SEM studies of silk of both the species. Our observations are in agreement with those recorded previously in a species of Neotropical tarantula [47].

From conservation perspective, the international pet-trade is a significant driver of biodiversity loss [48], although regrettably the invertebrate species are usually ignored when formulating conservation actions and policies [49]. That tarantula trafficking has low priority for enforcement authorities, complicates the issue further. The Gooty Sapphire Ornamental and Psychedelic tarantula are examples of heavily trafficked theraphosids for pet trade, endemic to India.

The spiders in family Theraphosidae have gained considerable reputation as pets due to their large size, striking colours, and longevity. Websites on the internet constitute only a small fraction of theraphosid trade. Several ambiguities in the legislative system and laxity in enforcement allow pet trade to subsist in India. Notwithstanding the fact that tarantulas occupy critical niches and play a significant role in ecosystems; their collection for pet trade goes on unabated, many of the websites advertising that *Chilobrachys* for sale is specifically a Goan species. In spite of being a prominent theraphosid in global pet trade, the IUCN status of *Chilobrachys fimbriatus* remains 'Least Concern'. Shockingly, Indian Theraphosid spiders are neither listed under the Indian Wildlife Protection Act (1972) nor are CITES listed [7]. As many as 13 species of Indian tarantulas are in the pet trade, though Indian legislation specifically does not preclude the collection, possession, transport, export, and commercialization of tarantulas. Fortunately many species have now been evaluated for their conservation concerns as per IUCN categories [8]. Ironically, our scientific understanding on vital issues like evolutionary history, species limits, population structure, and abundance of natural populations regarding many of the traded tarantulas is insufficient to devise pragmatic conservation strategies [50].

It has been categorically established that anthropogenic changes interfere with ecosystem functioning and services, and thus affect and threaten biodiversity [51]. The pace of linear infrastructure development in the state is fast. Key to development being connectivity, up gradation of existing Goa/Karnataka Border- Panaji Goa Section of NH-4A (Anmod to Panaji section) to four Lane configurations is currently under way. The entire stretch of NH- 4A lies in the states of Karnataka and Goa. It provides an important link between NH-4 and NH-17. The entire alignment passes through hilly and rolling terrain, except few stretches towards Goa. Also the NH-4A passes through the Bhagwan Mahavir National Park, Mollem. Reports of this specific project site in the confines of Mollem Protected Area as being potential theraphosid habitat for both species viz., *Chilobrachys fimbriatus* and *Thrigmopoeus truculentus* [8,11] undermines their conservation future. Like most species with precarious conservation status, the primary threat to tarantulas is the destruction of their habitat across their range.

This project specific EIA has come under the scanner of the stakeholders opposing this proposal, due to its insufficiency in impact assessment on biodiversity. The extent to which EIA fully addresses the identification of impacts and conservation stakes associated with biodiversity loss has been debatable [52]. Despite high scientific tractability, the suggested mitigation measures only result in compromised implementation, defeating the very purpose of EIA. Linear Infrastructure projects are notorious drivers of degradation, fragmentation and destruction of habitat; accompanied by the loss of genetic and species diversity and arrested ecosystem functioning [53,54]. The conservationists contend that without a thorough assessment of the impacts, these linear infrastructure proposals shall have disastrous consequences for the rich and endemic biodiversity of this area [55]. It is proven beyond doubt that linear infrastructure transects wilderness thereby reducing the habitat complexity and species richness of flora. This cascades into disproportionate impacts on spider density and diversity within such altered and fragmented habitats [56,57,58,59,60].

Mygalomorph spiders are considered 'relictual', because despite being genetically diverse, they have remained in a similar ecological niche on a long geological time scale [61]. Also, their low dispersal rate makes a case for high diversity of restricted-range species over evolutionary time scales [61], classifying them as conservation- significant 'Short-Range Endemics' (SREs) [62]. This poses a critical challenge by magnifying their loss even over small spatial expanse as in the present case, where their microhabitat has already been severely impacted and burrows smothered or destroyed due to scraping of embankments. Mygalomorph spiders, as SREs [63] represent a largely unrecognized contribution to biodiversity. That skewed baseline data is a limitation of EIA documents internationally has been established [64]. In case of the linear infrastructure projects proposed through the Mollem PAs; critical gaps and deficiencies in multiple functional areas including Ecology and Biodiversity have come under stakeholder scrutiny. Exclusion from the EIA of invertebrate groups like arachnids accentuates a bias against likely findings of significance. For instance *Thrigmopoeus truculentus* is both, endemic and 'Near Threatened' and yet does not find a mention in the EIA report.

Both *C. fimbriatus* and *T. truculentus* reside in their burrows on the roadside embankment in high density and prefer very specific habitat areas and need a contiguous habitat for dispersal. Theraphosid mygalomorphs are long-lived spiders that are poor at dispersal, and the species are restricted to few locations. Poor vagility, limited dispersal mechanisms and sedentary habits of these spiders has been confirmed [65]. The alteration and degradation of habitat can have a major impact on small populations as in the present case.

Scientific evidence is piling up to suggest that changes in land use and associated pressures have already taken a frightening toll on the environment and reduced the local terrestrial biodiversity [54]. Impacts from land use practices such as deforestation for development, and urbanization continue to degrade and fragment remaining pockets of habitat and accelerate loss of such isolated species. Final blow could come from development activity that disrupts the phenology and change the microclimatic parameters, by impacting the recruitment dynamics of the species.

6. Conclusion

This research has addressed an important gap in our understanding of Ecology and behavior of the two species of Theraphosidae spiders viz. (*Chilobrachys fimbriatus* Pocock, 1899) and Lesser Goa Mustard or Karwar Large

Burrowing spider (*Thrigmopoeus truculentus* Pocock, 1899) found in the state of Goa. The investigation has relied on morphology and Morphometry to confirm the taxonomy, besides having documented *in situ* behavior of the two species. Habitat of these species overlap, yet they differ in their burrow density, architecture and fine morphology of silk fibres. The study also highlights trade trends of the two species and confirms that *Chilobrachys fimbriatus* is in much demand in global pet trade. The conservation future of the two species in the state is precarious given that linear infrastructure projects have been rolled out in their extant habitat in the precincts of the Protected Areas of Mollem, without assessment of cumulative impacts and species specific mitigation. It is suggested that a long term monitoring of populations of these Theraphosid species will allow for conservation assessment and management planning.

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Statement of Competing Interests

The authors declare no competing interests.

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