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Research Article

Insect functional guilds in the flowering canopy of *Myristica fatua* in a lowland swamp, central Western Ghats, India.

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Abstract

Little is known of canopy insects and pollinators of the nutmeg family (Myristicaceae), despite its being a common pantropical tree family and a commercially-grown spice. To address this lack of knowledge, an assessment was made of the insect fauna associated with the canopy of *Myristica fatua* (Myristicaceae), an endemic tree species inhabiting the rare and endangered lowland 'Myristica' swamps of the south and central Western Ghats of India. Yellow Sticky Traps were hung in tree canopies to record insect diversity. Insect visitors encountered during the flowering period of *Myristica fatua* were classified based on their functional role. The captured arthropod fauna was characterised by an abundance of parasitic, predatory, herbivorous and gall-making insects of Chalcidoidea, Platygastroidea, Bethyloidea, Ceraphronoidea, Dolichopodidae, Empididae and Cicadellidae, and a scarcity of beetles. The exclusive gall-making hymenopterans of Cynipidae formed one of the largest insect families in the canopy of *M. fatua.* The faunal assemblage appeared to be a subset of the entire insect fauna associated with the tree species, as sticky traps leave out the apterygote, sedentary and large insects, and a higher turnover in the insects at the family level could be expected during the fruiting season of the tree. The study highlights the importance of canopy arthropods and their functional roles in community structure.

Key words: canopy insects, functional role, lowland Myristica swamp, parasitic Hymenoptera, thrips, Yellow Sticky Traps.

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Introduction

Forest canopies not only guide several ecosystem processes but also are the heart of an exceptionally high diversity of organisms that are woven in a complex web of functions and interactions [1]. Erwin's canopy fogging studies in Panama [2] sent the global arthropod species richness on a tumultuous rise from 3-5 million to 30–100 million, and more recent studies suggest an estimate of 6.1 million arthropod species in the tropics alone (3,4). A number of studies throughout the tropical world have given us detailed information on canopy invertebrates [5-7], yet much of the diverse canopy fauna [8, 9] remains largely undiscovered or unquantified [10], especially outside of the intensively studied field sites. In the Indian subcontinent, in spite of the conservation value of such research, canopy biodiversity studies are rare [9]. For example, a study by Srinivasa *et al.* [11] quantifying the beetle diversity of two evergreen emergent tree species (*Vateria indica* and *Dipterocarpus indicus*) is the only published data on canopy insects from the Western Ghats, SW India. In this study we document the arthropod assemblages in flowering canopies of the dioecious tree species *Myristica fatua* var. *magnifica* (Bedd.) Sinclair (Myristicaceae).

Early descriptions of *Myristica fatua* mention "swampy grounds in lower elevation evergreen forests" of the Western Ghats with adaptive stilt roots growing high from the trunk [12-14]. These swamps are the exclusive habitat for two of the five Indian and Sri Lankan species of Myristicaceae - *Myristica fatua* var. *magnifica* and *Gymnacranthera canarica,* both of which are endemic to the Western Ghats and endangered [15-17], along with a newly described species *Semecarpus kathalekanensis* [18, 19]. The swamps are highly vulnerable to conversion to agriculture, overuse of swamp water for irrigation [15], soil erosion, invasion by Pandanus and destructive harvest of *Myristica* fruits for trade of nutmeg and mace [20]. They are critical habitats for three species of hornbill that feed on the aril (seed coat) of both species. Information on species interactions such as pollination, herbivory and predation in the canopy of *M. fatua* could be of immense value to understanding and conserving the species in the wild.

This study is an assessment of the diversity and functional role of canopy arthropods in *Myristica fatua* using a single capture technique. Insect visitors in the canopy of *M. fatua* were classified into five feeding guilds based on their functional role – fungivores, herbivores, parasitoids, predators or saprophytes. Since *M. fatua* was in flower, we also used the opportunity to make preliminary observations of pollinators and compare the guild composition and arthropod species richness between male and female trees. In flowering trees, in addition to the insect fauna routinely present in the canopy of non-flowering trees, diverse functional categories or trophic roles of insects may be found: those foraging for food resources (nectar, pollen, or the flowers themselves), which are potential pollinators [21]; those using the flowers as a space to enhance the likelihood of encounter with prey or mates; or those that are merely casual visitors [22-24].

Methods

Insects were sampled during the flowering season of *Myristica fatua* in January 2008 and 2009 in a swamp in Kathlekan village in the Uttara Kannada district of Karnataka (14°16'80.3" N, 74°46'75.7" E; 586 m asl.). This is one of the three populations of *M. fatua* reported from an estimated total of fifty swamps in the central Western Ghats [25].

To assess canopy insect diversity, we used Yellow Sticky Traps (Growing Success Ltd., Wiltshire, UK, size 5 X 10 cm), and assumed that the probability of capture was proportional to the species frequency in the canopy [26]. Between eight and 10 sticky traps were hung in each of one male and one female tree near flowering branches at a height of 15 – 20 m. Traps were retrieved after 48 h in the canopy.

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The retrieved traps were scanned under a stereo zoom binocular microscope, and insects were identified initially to order and family and then to recognisable taxonomic units (RTUs) or morpho-species [27], hereafter species. Individuals that were difficult to identify were removed from the glued surface of the trap using ethyl acetate. Arthropod species were classified into one of five major guilds: fungivores, herbivores (including pollinators), parasitoids, predators or saprophytes. Insects and spiders were categorised into feeding guilds based on personal observations and family-level information from published sources [7, 28]. Because of the great diversity in gall-inducing parasitic hymenopterans and the difficulty of separating gall-inducing wasps from inquilinies and the parasitoids of gall-inducing wasps [29], all the Cynipid wasps (true gall-inducing wasps) were scored as gall-inducers and categorised as herbivores. All other parasitic wasps of the super-family Chalcidoidea were categorised as parasitoids.

Insects found with pollen loads from nutmeg flowers and those engaged in diurnal foraging within flowers were subsequently classified as pollinators of *M. fatua*.

The arthropod assemblages of male and female tree canopies were compared using both guild composition and species richness. A lack of female flowering trees with canopy access was a major constraint in the first year of the study and led to the difference in sampling effort between the sexes. Since a different number of traps was used to sample the arthropods in male (N = 21 traps) versus female (N = 9) trees, rarefied species richness, a recommended tool to negate the difference in the sampling effort [30], was used to compare arthropod richness between the sexes. Data from 2008 and 2009 were pooled for all analyses. To determine if the current sampling effort was sufficient, a species accumulation curve was constructed with the observed species richness to estimate overall species richness (Chao 2; Biodiv Pro, Beta version [31]). In a saturated sampling effort, the curve would plateau. Chao 2 calculates an estimate of species richness, calculated as: Chao 2 = S_{obs} + (L^2 / 2M); S_{obs} is the number of species observed in all samples pooled, 'L' the number of species observed in only one sample (unique captures) and 'M' the number of species seen in only two samples (duplicate captures) (EstimateS 7.5 [32]). Numbers of singletons (species with one individual in the sample) and doubletons (species with at least two individuals in the sample) are used as measures of species rarity in the canopy. The mean number of individuals of each feeding guild was compared using Kruskal Wallis rank ANOVA test in S Plus version 5.1.

Results

Species richness

A total of 725 individual insects and spiders belonging to 110 species of 61 families and 10 orders were collected in sticky traps: The observed species richness (S_{obs} = 110) of this study falls short of that expected (S_{ex} =169.8, Fig. 1). The difference in observed species richness and estimated species richness and the non-asymptotic species accumulation curve illustrates that the sampling effort was insufficient to truly represent the arthropod assemblage in the tree canopy. The rarefied species richness of arthropods in male and female tree canopies was 32 and 27 species, respectively. Almost half (46%) of the species were represented by only one or two individuals and were considered rare species in the *Myristica fatua* canopy. Most individuals were small (Fig. 2).

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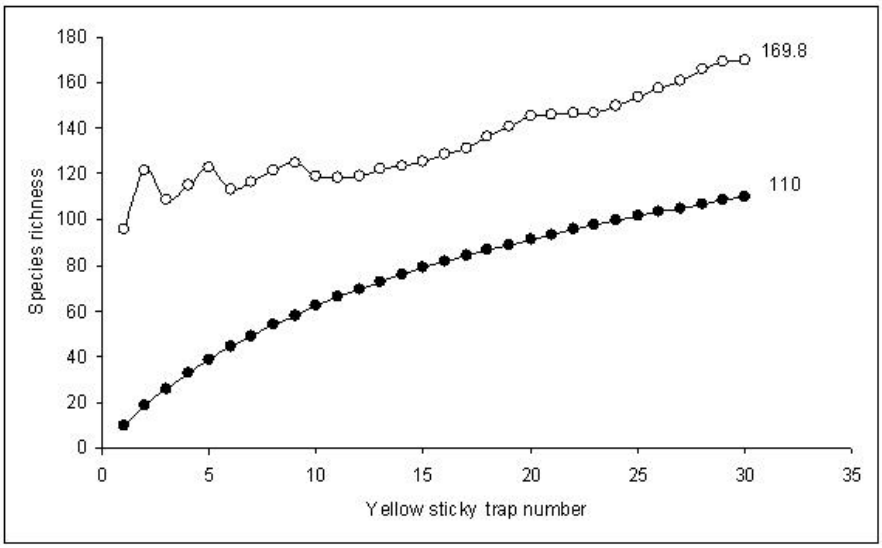


Fig. 1. Species accumulation curve illustrating the observed (closed circle) and expected (Chao2, open circle) species richness.

Order richness

The order Hymenoptera was the most diverse and abundant insect group in the canopy of *M. fatua*, represented by 222 individual parasitic wasps of 44 species and 18 families of the super-families Chalcidoidea, Platygastroidea, Proctotrupoidea, Bethyloidea, and Ceraphronoidea. The considerable number of tiny female Mymarid (N=16) and Encyrtid (N=54) wasps in the canopy suggests that the tree foliage may be providing plentiful host species of parasitoids. Only one large predatory wasp belonging to family Tiphiidae was captured.



Fig. 2. Size range of the insect fauna captured on a Yellow Sticky Trap.

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Guild diversity

Representatives of all five arthropod guilds were found (Fig. 3a). However, the two carnivorous guilds, predators (23%) and parasitoids (32%), formed the bulk of individuals and species. The parasitic insect guild was composed of 235 individuals and included Dipteran families Phoridae and Calliphoridae. While parasites, herbivores and predators were significantly more abundant per trap, herbivores and predators had more individuals per species.

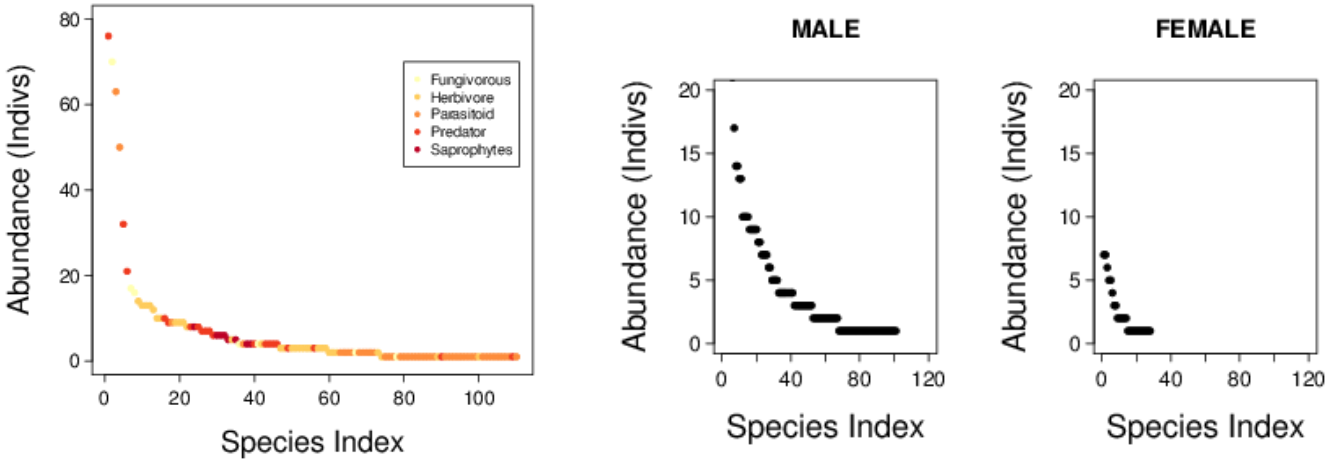


Fig. 3. Dominance-diversity curves for all trees with feeding guild identified (Left), and for males and females (Right).

Herbivores formed the second major group in the canopy (36% of individuals), and included sap suckers (leafhoppers in the Cicadellidae and Membracidae), flower feeders, pollen and nectar feeders, leaf miners, and gall makers. The most abundant gall makers were wasps (Cynipidae), bugs (Psyllidae), flies (Tephritidae, Cecidomyiidae and Agromyzidae), and beetles (Buprestidae). Non-parasitic gall-making wasps (Cynipidae) were particularly common: 84 individuals in two species were captured. Dipteran families (predators) were abundant in the canopy - Dolichopodidae, Empididae and Asilidae (N=82, 21 and 9, respectively), forming the next abundant feeding guild per trap – that of predators. The other two feeding guilds, fungivores and saprophytes, were much less abundant (Table 1).

The beetles trapped were either small beetles (Cucujidae, Silvanidae and Curculionidae) or those with thin, loose elytra (Cantharidae and Cleridae). Eighty-seven individual thrips belonging to Phlaeothripidae and Thripidae were captured. These, along with a few beetles (Table 1) were categorised as potential pollinators based on observations of foraging and presence of pollen load. The body length range (Fig. 2) of 96% of the fauna was 0.3 mm - 1 cm, suggesting the under-estimate of diversity was also biased to smaller body size. Some large insects like scorpion flies (Mecoptera, ~ 6 cm) were delicate bodied.

A mean of 11.2 (± 3.0 SE) individuals trapped per sticky trap were parasitoids, while herbivores and predators were 12.6 (± 2.3) and 7.8 (± 1.9) individuals, respectively (Kruskal Wallis test, H (df =4, N =105) = 24.54; $p<0.0001$) (Fig. 4a). The mean number of individuals per species (Fig. 4a) did not vary significantly among feeding guilds (Kruskal Wallis test, H (df=4, N=110) = 0.54; $p >0.05$).

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Table 1. The insect faunal assemblage captured in sticky traps represented in families as classified under five major feeding guilds. The number of individuals in each guild and family is presented alongside, and the number of species is in parentheses. There is an overlap of guilds - some beetles in the study initially thought to be strict fungivores, herbivores and predators were also potential pollinators.

Fungivore 29	**Herbivore 264**	**Parasitoid 235**	**Predator 164**	**Saprophyte 33**
Cucujidae* 11 (2)	Cynipidae 84 (2)	Encyrtidae 54 (3)	Dolichopodidae 82 (3)	Muscidae 26(5)
Sciaridae 7 (1)	Phlaeothripidae* 84 (2)	Braconidae 47 (4)	Empididae 21 (1)	Sarcophagidae 6 (2)
Psocidae 4 (1)	Cicadellidae 32 (9)	Eulophidae 23 (4)	Tachinidae 18 (3)	Coelopidae 1 (1)
Mycetophilidae 3 (1)	Tephritidae 15 (2)	Mymaridae 16 (4)	Mecoptera 13 (1)	
Silvanidae* 2 (1)	Psyllidae 13 (1)	Megaspilidae 14 (2)	Anthocoridae 9 (1)	
Trogiidae 2 (1)	Membracidae 9 (2)	Diapridae 12 (2)	Asilidae 9 (2)	
	Curculionidae* 5 (2)	Phoridae 12 (1)	Cleridae* 3 (2)	
	Aphididae 5 (1)	Scelionidae 10 (4)	Scatopsidae 3 (1)	
	Cecidomyiidae 4 (1)	Aphelinidae 2 (1)	Chrysididae 1 (1)	
	Thripidae* 3 (1)	Bethylidae 9 (3)	Ephydridae 1 (1)	
	Chloropidae 2 (1)	Platygastridae 9 (3)	Formicidae 1 (1)	
	Syrphidae 2 (1)	Eucoilidae 8 (2)	Salticidae$ 1 (1)	
	Agromyzidae 1 (1)	Trichogrammatidae 5 (2)	Tiphiidae 1 (1)	
	Anisopodidae 1 (1)	Ichneumonidae 4 (3)	Thomisidae$ 1 (1)	
	Buprestidae 1 (1)	Pteromalidae 3 (1)		
	Cantharidae* 1 (1)	Eurytomidae 2 (1)		
	Ceratopogonidae 1 (1)	Torymidae 2 (1)		
	Chironomidae 1 (1)	Calliphoridae 1 (1)		
		Ceraphronidae 1 (1)		
		Elasmidae 1 (1)		

Comparing diversity by tree sex

659 insect individuals per trap (±0.36) were collected from male trees and 66 individuals per tree (±1.2) were collected from female trees (Fig. 3b). The mean number of individuals trapped per sticky trap on male and female trees was also different for fungivores (Mann-Whitney U test, U=23, p=0.02), herbivores (U=13.5, p=0.004), parasitoids (U=2, p=0.0001) and predators (U=14, p=0.004); the difference was insignificant for saprophytes (U=40.5, p=0.33) (Fig. 4b). These differences were only present when the pooled data from the two years were analysed, and not when the 2009 data were analysed in isolation. Captures from male trees had parasitoids and herbivores composing the major functional group (58% of individuals), while captures from the female tree had herbivores as the major guild (57%) with parasitoids and predators comprising only 25% of the fauna.

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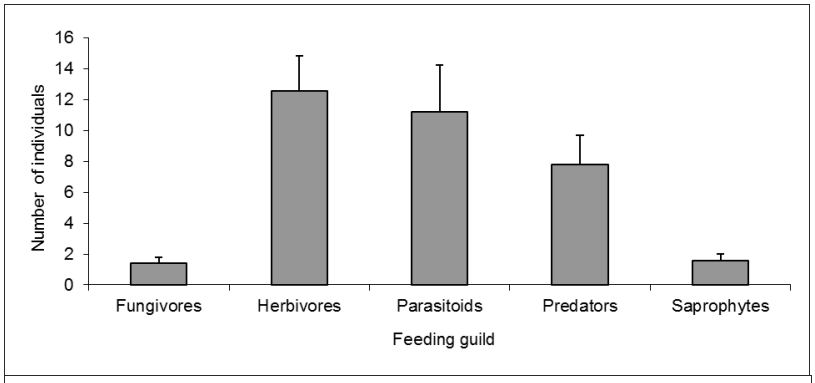


Fig. 4a. Mean (± SE) number of individuals of each feeding guild trapped per sticky trap.

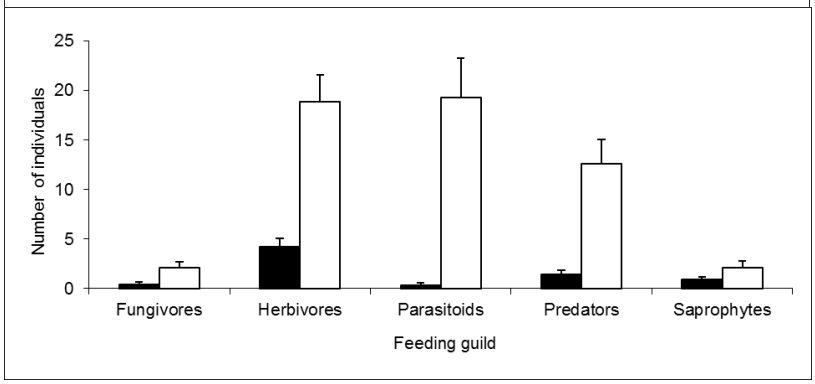


Fig. 4b. Mean (± SE) number of individuals of each feeding guild trapped per sticky trap on female (closed bar) and male (open bar) trees. There was no difference between sexes when the 2009 data was used in isolation, and the difference presented here is driven by the 2008 data.

Discussion

Insect faunal assemblages sampled during the flowering of *M. fatua* were dominated by parasitic wasps, which demonstrates the need for further studies on the diversity of host insect–parasitoid wasp interactions in rainforest canopies. An extensive inventory of the canopy arthropod fauna of an Australian tree species reported a greater diversity of canopy parasitoid wasps [7]. It would also be of interest to explore the diversity of parasitic wasps that can induce gall formation on the tree. Narendran *et al.* [29] consider the entire family of Cynipid wasps as gall insects of the parasitic super family Chalcidoidea (Hymenoptera). They reviewed parasitic wasps of 18 families under the super family. Their compendium suggests that certain tribes or genera in each of the 18 families can be potential gall makers. The relatively high abundance of the functional guild of gall-making Cynipid wasps, otherwise tricky to capture, emphasises the usefulness of sticky traps in canopy insect surveys. Predatory flies of Dolichopodidae, Empididae and Asilidae were abundant in the *M. fatua* canopy, unlike studies elsewhere [7], suggesting that Diptera play a more significant role in arboreal insect community interactions than their featured category as “tourists” in the canopy [33]. Unlike some canopy insect diversity studies, the fauna in the present study included few beetles (Coleoptera), which might be a drawback of using Yellow Sticky Traps. Some beetles in the study initially thought to be strict fungivores, herbivores or predators were found to be potential pollinators based on observations of foraging and pollen load on insect bodies. Such an overlap of functional guilds is only natural in tropical canopies, and it is useful to be warned that a change in faunal assemblage could have a cascading effect on pollination processes.

Sticky traps are often used in practical pest management to monitor whiteflies, thrips, fungus gnats,

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leafminers, psyllids, shore flies, winged aphids, and parasites [34]. Few studies have used them to monitor visits and record diversity of canopy insects, although they have proven to be useful tools in rapid surveys since they give a relative measure of insect density and diversity, as well as pollinator activity [26, 35]. They are especially handy in recording nocturnal or canopy insect visitors in studies where direct observations are impractical.

Minute insects stick tightly to the glue, making identification a challenge; only an expert familiar with a diverse range of insects can arrive at the level of family. Most specimens were identified without removal from the trap. Canopy fogging with knockdown insecticides, although demonstrated as a reliable method to sample large insects, apterygote insects and beetles, has a drawback: the chance of missing micro arthropods such as parasitoid wasps that drift away while falling into the collection screen beneath the tree [10, 36]. Our study demonstrates that sticky traps work well to sample and quantify delicate insects often missed in fogging experiments. Armstrong [26] and Sakai [35] experienced success with sticky traps, capturing minute beetle and thrip pollinators. Canopy foliage sweeping is an important collection method for such insects, but would require either canopy walkways or cranes, neither being cost effective and both having operational hurdles in the Indian tropics.

The primary shortcoming of the study was insufficient sampling. The faunal assemblage for the *M. fatua* canopy was thus not exhaustive; transient and large insects, particularly beetles, field crickets, and sedentary or apterygote insects such as ants were not captured. The arthropod faunal composition would likely be altered if the sampling intensity were increased and extended to the fruiting period and might then be dominated by beetles, as reported by Srinivasa *et al.* [11] on *Vateria indica* and *Dipterocarpus indicus*. A predominance of curculionid weevils on fruiting trees explained the dominance of seed predatory insects.

Implications for conservation

The last fragments of lowland Myristica swamps in the Western Ghats with dwindling populations of *Myristica fatua* and its associated tree species present a strong case for conservation [37]. While recruitment efforts are underway, it has become more crucial than ever before to understand the basic community composition and species linkages in these rare habitats. Arthropods, particularly insects, play a crucial role in the interaction of plants with their environment and form an important link in most terrestrial food webs. Documenting canopy arthropod diversity is important in understanding the functioning of a forest system [11]. Studies such as this that focus on arthropod functional roles in the canopy of an endangered tree species add a new dimension to insect ecological research in a biodiversity hotspot region.

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