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Quantitative assessment of plant-arthropod interactions in forest canopies: A plot-based approach

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Data Availability Statement: We provide the data used in the analyses in S1 Table. The data on Yawan dataset that were used for the comparison of plot-based and non-plot-based analyses of herbivore-plant interaction networks were taken from Redmond et al. 2019 [20]. The publicly available sequences on insect DNA barcodes can be accessed from The Barcode of Life Data System (http://www.boldsystems.org) as DS-LANZMIK (Mikulcice and Lanzhot sites; [21]), TLR (Tomakomai), DS-CATS1, DS-SEGAR16, and

Abstract

Research on canopy arthropods has progressed from species inventories to the study of their interactions and networks, enhancing our understanding of how hyper-diverse communities are maintained. Previous studies often focused on sampling individual tree species, individual trees or their parts. We argue that such selective sampling is not ideal when analyzing interaction network structure, and may lead to erroneous conclusions. We developed practical and reproducible sampling guidelines for the plot-based analysis of arthropod interaction networks in forest canopies. Our sampling protocol focused on insect herbivores (leaf-chewing insect larvae, miners and gallers) and non-flying invertebrate predators (spiders and ants). We quantitatively sampled the focal arthropods from felled trees, or from trees accessed by canopy cranes or cherry pickers in 53 0.1 ha forest plots in five biogeographic regions, comprising 6,280 trees in total. All three methods required a similar sampling effort and provided good foliage accessibility. Furthermore, we compared interaction networks derived from plot-based data to interaction networks derived from simulated non-plot-based data focusing either on common tree species or a representative selection of tree families. All types of non-plot-based data showed highly biased network structure

ASPNA (Wanang; [48, 56]), and DS-YAWAN2 (Yawan; [23, 57]) datasets.

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towards higher connectance, higher web asymmetry, and higher nestedness temperature when compared with plot-based data. Furthermore, some types of non-plot-based data showed biased diversity of the associated herbivore species and specificity of their interactions. Plot-based sampling thus appears to be the most rigorous approach for reconstructing realistic, quantitative plant-arthropod interaction networks that are comparable across sites and regions. Studies of plant interactions have greatly benefited from a plot-based approach and we argue that studies of arthropod interactions would benefit in the same way. We conclude that plot-based studies on canopy arthropods would yield important insights into the processes of interaction network assembly and dynamics, which could be maximised via a coordinated network of plot-based study sites.

Introduction

Forest canopies represent one of the most diverse environments on the planet [1], harbouring a large proportion of terrestrial arthropod diversity estimated at 6.8 million species [2]. At the same time, canopies are among the least explored habitats due to the logistical challenges of accessibility [1]. This combination of high diversity and inaccessibility has fascinated biologists for more than 150 years [1].

The development of single-rope climbing and fogging has provided ecologists with efficient tools for researching canopy arthropod communities, generating several influential studies e.g. [3, 4, 5]. Such studies spurred the development of new methods of access that nowadays include canopy walkways, canopy rafts, balloons, cherry pickers, or canopy cranes [1]. Canopy studies have contributed to our understanding of species global diversity and biotic interactions [2, 6], but, as pointed out by Lowman et al. [1], "*. . .the real challenge is ahead. Canopy organisms, both mobile and sessile, must be surveyed and their roles measured.*"

Research into canopy arthropods has progressed from species inventories to the study of their interactions, allowing us to understand how hyper-diverse communities of canopy arthropods are maintained [7]. Particular sampling methods are suitable for different systems and questions concerning the various roles arthropods play in forest canopies [1] (Table 1). Methods that allow access to individual branches or certain parts of the canopy are suitable for exploratory studies on arthropod diversity, detailed surveys focused on specific taxa, or manipulative experiments e.g. [8, 9]. They also allow for comparative studies across various canopy microhabitats and their arthropod communities [10]. However, to fully census interactions between arthropods and plants on the level of the canopy as a whole, sampling methods must provide access to the entire canopy, from the terminal branches, through the inner canopy, to the lower branches. This is because arthropod species composition may differ considerably among various parts of the canopy [11], reflecting variation in resource availability and leaf traits [12]. Neglecting some parts of the canopy, therefore, has the potential to influence the results of the census. In addition, methods suitable for censusing canopy arthropod interactions must facilitate the sampling of arthropods in such a way that enables the reliable reconstruction of the interaction network. In the tropics, transient herbivorous arthropod species (i.e. species with no lasting association to the sampled plant) can comprise up to 20% of species found on a particular tree [13]. Thus, dead arthropods sampled from a plant do not constitute reliable interactions. To reliably reconstruct interaction networks, one needs to either sample live arthropods for feeding trials [14] or a use molecular detection of trophic interactions [15].

Table 1. Summary characteristics of forest canopy sampling methods that allow active sampling of arthropods by manual search, beating, sweeping, or fogging. The trapping methods are not listed. Characteristics include Canopy accessibility (accessibility of tree strata: T (terminal branches), U (upper canopy), L (lower canopy), I (inner canopy)); suitable Scale of sampling (whole canopy vs. individual branches), Arthropod taxa sampled (E (endophytic), T (trunk-nesting), N (non-flying exophytic herbivores and predators), F (flying).

Method	Canopy accessibility	Scale	Arthropod taxa	Team size	Costs	Replicability	Site availability	References
Canopy crane	T,U,L	Whole canopy, branches	E,N,F	Medium	High	Low	Low	Basset et al. [17]; Ødegaard [18]; Wardhaugh [19]
Cherry picker	T,U,L	Whole canopy, branches	E,N,F	Medium	High	High	Medium	Corff and Marquis [20]; Volf et al. [21]
Felling	T,U,L,I	Whole canopy	E,T,N	Large	Medium	High	Medium	Whitfeld et al. [22]; Redmond et al. [23]
Canopy rafts	T,U	Branches	E,N,F	Medium	High	Low	High	Lowman et al.[8]
Canopy walks	U,L,I	Branches	E,N,F	Medium	Medium	Low	Low	Reynolds and Crossley [24]
Fogging	T,U,L,I	Whole canopy	N*,F*	Small	Low	High	High	Erwin [3]; Kitching et al. [25]
Tree climbing	U,L,I	Branches	E,T,N,F	Small	Low	High	High	Lowman [26]; Schowalter and Zhang [27]

* indicates that dead insects are sampled); minimal required Team size; relative operational Costs; Replicability (ease and practicality of replication); Site availability (low—limited sites with crane or walkway access; medium–available access road for cherry picker, felling not permissible in protected forests and other situations; high—almost all forests can be sampled); and key References.

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Similarly, it is necessary to map ant nests rather than simply sample individual ants, as up to half of the ants foraging in a tree are tourists from surrounding vegetation [16].

Most importantly, for a quantitative analysis of arthropod interaction networks, the methods should allow structured sampling across large parts of the canopy, thus including all species in proportion to their abundance [7, 21, 28]. Previous studies often focused on sampling individual tree species, individual trees or selected constituent parts. Selective sampling is particularly beneficial for exploring insect-plant interactions in a phylogenetical or evolutionary framework as it allows the researcher to focus on particular lineages of interest [29, 30]. Methods that employ selective sampling are also valuable when assessing herbivore specialization or the effects of host-plant traits on insect community structure. This is because all focal species can be sampled with equal effort, thus allowing for direct comparisons between herbivore or host species [14, 31]. However, a drawback of selective sampling is that it does not facilitate quantitative network structure analyses, because it tends to skew interaction frequencies, over- or underestimate specialization and diversity, and biases network structure [7]. In particular, it typically omits a high proportion of the arthropod and plant taxa co-existing at the sites, hence not reflecting species diversity and network structure at the whole forest level. We argue that for interaction network analyses, a plot-based approach, where entire plots are censused for plants and arthropods, is preferable, as it more accurately reflects the diversity and abundance of the available resources [21, 23].

Plot-based approaches applied to forest vegetation have greatly benefitted plant ecology research [32]. We anticipate the study of arthropod interaction networks would benefit in equal measure [33]. We accessed canopies using tree felling, canopy crane, and cherry picker techniques (Fig 1) across biogeographic regions (Palearctic, Nearctic, Neotropical, and Australian) and forest types (tropical vs. temperate, lowland vs. montane, primary vs. secondary). We compare our plot-based methods with non-plot-based sampling and highlight the strengths and limitations of the methods for sampling mobile flightless exophytic herbivores (leaf-chewing insect larvae), endophytic herbivores (miners and gallers), and flightless invertebrate

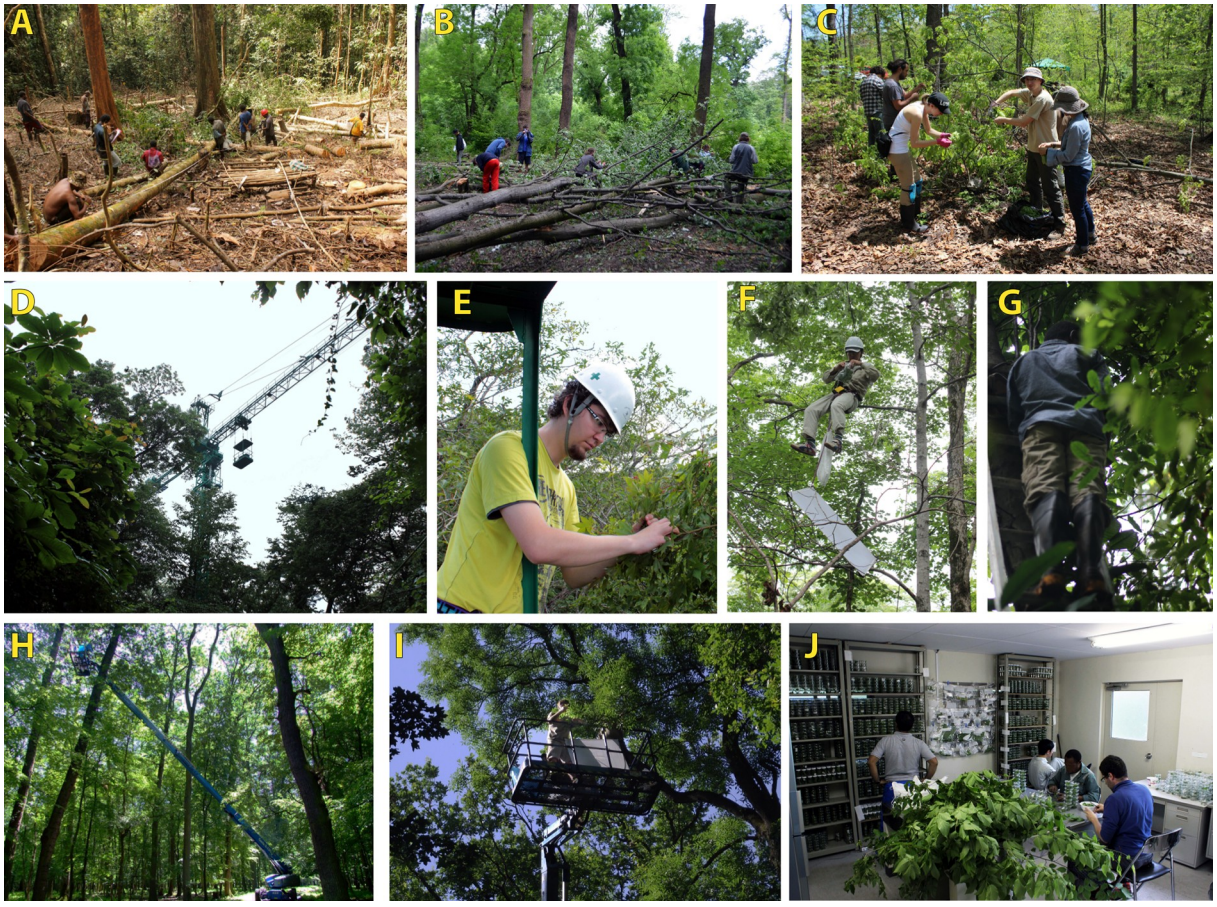

Fig 1. Photos from the field. Measuring a felled tree in Numba (A), herbivore sampling from felled trees in Mikulcice and Toms Brook (B, C), sampling from canopy crane in Tomakomai (D, E), a tree climber accessing a tree inaccessible from the crane in Tomakomai (F), sampling of an understory tree by ladder in San Lorenzo (G), sampling from cherry picker in Lanzhot (H, I), sample sorting and caterpillar rearing in Tomakomai (J). The individuals whose faces are fully or partially visible in this figure have given written informed consent (as outlined in PLOS consent form) to publish these photos.

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predators (spiders and ants). Our aim is to stimulate plot-based research by providing practical and reproducible sampling guidelines for the analysis of arthropod interaction networks in forest canopies. We expect i) plot-based data and non-plot-based data to provide largely different estimates of interaction network structure as non-plot-based sampling skews frequencies between rare and abundant species, ii) felling to be the most efficient method in terms of sampling effort as it allows employing large teams of field workers who can simultaneously access large parts of the canopy, iii) all three methods to provide similar acessibility to the canopy with access to over 75% of foliage.

Materials and methods

During our operations, we took advantage of ongoing logging operations (Mikulcice, Toms Brook) and shifting agriculture (PNG sites); no plot was cleared solely for sampling. All projects were conducted in close collaboration with the local community and land owners. We obtained all research and export permits where required. Arthropods and plants from Papua New Guinea were sampled and exported under the permits nr. 070382, 070384, 080275, 010075, 011209, 011324, 012134, 014282, 0133004, 133005, and 018060 issued by Department

of Environment and Conservation, Papua New Guinea, and 0139/2008, 0162/2010, and 0203/2013 issued by Forest Research Institute and Department of Forests, Papua New Guinea. Arthropods and plants from Panama were obtained and exported under the permits nr. SE/A-49-16, SE/AP-28-16, SC/AP-2-16, SEX/P-30-17, SEX/A-67-17, SEX/A-76-17 issued by Ministerio de Ambiente, Panama. The individuals whose faces are fully or partially visible in Fig 1 have given written informed consent (as outlined in PLOS consent form) to publish these photos.

Following a standardized protocol (Appendix 1) and workflow (Fig 2), we sampled i) lowland temperate forests in the Czech Republic (Mikulcice, Lanzhot), Japan (Tomakomai), and USA (Toms Brook); ii) lowland tropical forests in Panama (San Lorenzo) and Papua New Guinea (hereafter PNG; Wanang); and iii) highland tropical forests in PNG (Numba, Yawan) (Table 2, S1 Table).

Setting up the plot

At each location, we selected 0.1 ha plots with a vegetation structure and species composition typical for local broadleaf forests (Table 2, S1 and S2 Tables). In Wanang, Numba, and Yawan, plots were larger and subdivided into 0.1 ha sections (Table 2). Forest edges, plantations, stands with non-native vegetation, and large gaps were all avoided, as were steep slopes and swampy areas (for technical and safety reasons). We took GPS coordinates of all plot corners and used measuring tape or laser range finders to set up the plot and map all plants with DBH $\geq$5 cm. Each stem was tagged and identified to species level. It took 2–12 hours for three people to set up a 0.1 ha plot and map 24–251 trees within. In Tomakomai (4 trees), Toms Brook (8), and San Lorenzo (17) some trees proved to be hazardous to sample or were damaged by factors beyond our control, such as a hurricane, during the sampling. These trees were replaced by conspecifics or other broadleaf trees with a similar DBH adjacent to the plot. One non-native and one coniferous tree in Toms Brook were treated in the same way.

Timing the sampling

Arthropod abundances and species composition can vary dramatically throughout the year in seasonal forests. For example, temperate leaf-chewing insects exhibit one major peak during spring leaf-flush, and a smaller peak in late summer [12]. Furthermore, peaks in abundance may differ among arthropod guilds, for instance leaf miners, where the major peak appears to occur later than for leaf-chewers (S1 Fig, S3 Table). A single, short sampling campaign can fail to capture all arthropod groups. Therefore, we generally sampled temperate plots at a slower pace throughout the season to mitigate this undesirable effect, returning periodically to the sites in order to sample trees (i.e. typically one to several tree individuals were sampled per day depending on their canopy sizes). Sampling effort was increased during abundance peaks if they materialised. During this period, sampling was conducted whenever the weather permitted. In this way, the variation in sampling effort mirrors the variability in insect abundance, and the probability that an insect will be sampled remains constant throughout the season. We spread the sampling seasonally within each sampled tree species to avoid a bias due to an unbalanced seasonal sampling (Appendix 1). In wet tropical forests, sampling was carried out with constant effort throughout the seasons as the effects of seasonality are much less pronounced and individual species appear throughout the year [34]. However, a variable sampling strategy would be advisable in dry tropical and subtropical forests, where seasonality asserts greater influence [35]. Such intense and relatively long-term sampling required careful logistical planning. This involved negotiating the research plan well in advance with land owners, crane drivers, chainsaw operators, and local managers so as to avoid clashing with other

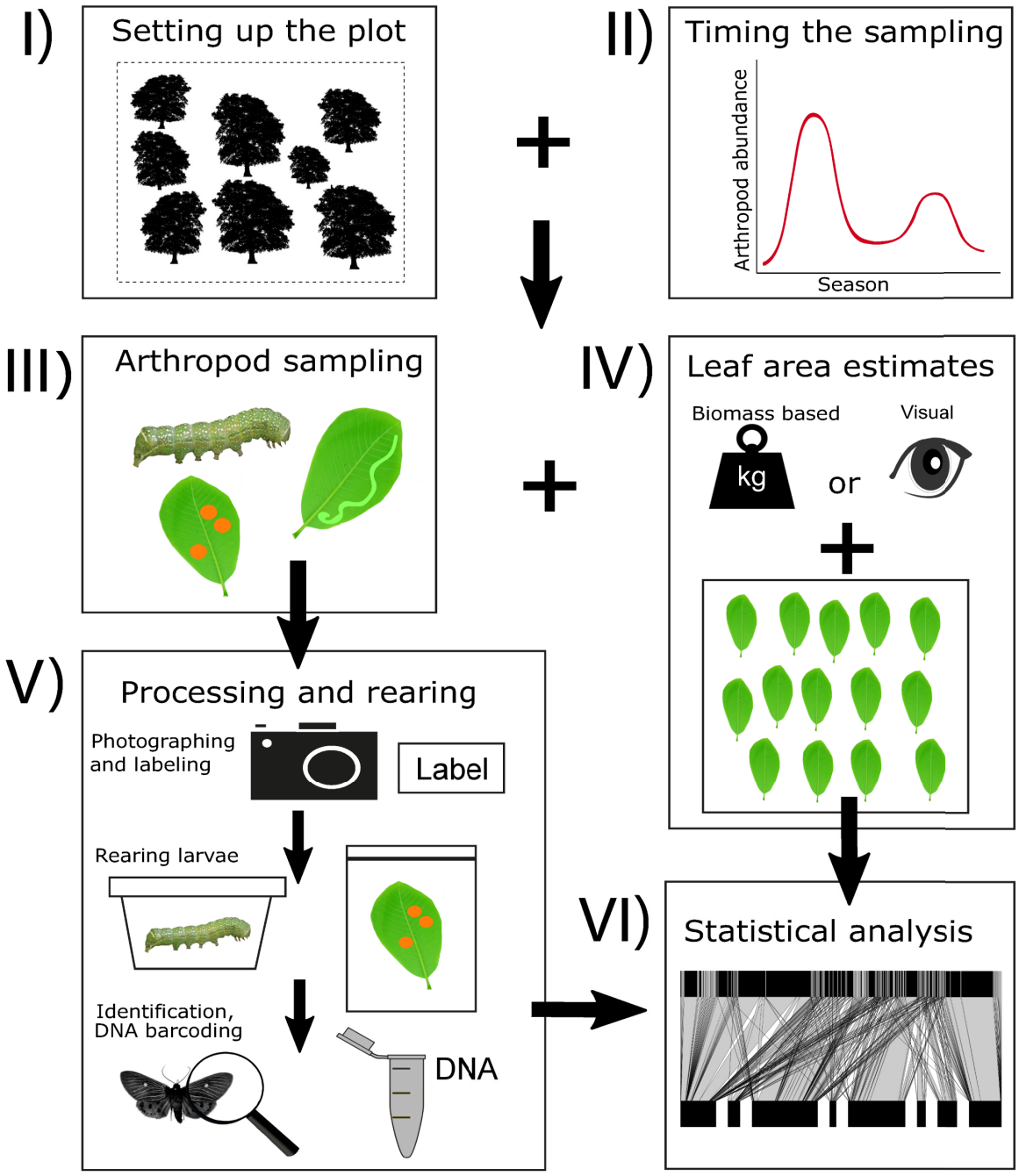

Fig 2. A workflow diagram for the proposed methods. The process starts with setting up the plot (I) and planning the sampling according to seasonality at a given site (II). The field work includes arthropod sampling (III) and estimation of leaf area (IV, including visual or biomass based estimates and processing of leaf frames). Sampled arthropods are then processed (V), which includes the labelling and photographing of morphospecies, rearing, and the sending of material for taxonomic identification or DNA barcoding. Finally, the data are analysed (VI).

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projects at the given sites. For example, for the sampling from felled trees, we specifically sought plots of forest that were scheduled for logging and paid the loggers to cut the trees on our schedule.

Table 2. Sampling site characteristics. Forest type (Trop—tropical., Temp—temperate), lowland (90–230 m a.s.l.), highland (700–1800 m a.s.l.), primary (P), and secondary (S) forests); Maximum tree height (m); Plots (number and size of sampled plots); Method of sampling; mean Number of stems with DBH≥5 cm per 0.1 ha (±SD); mean Sampled leaf area (m^2) per 0.1 ha (±SD); mean number of Leaf-chewing larvae per 0.1 ha (±SD); mean number of Active mines per 0.1 ha (±SD); mean Area-based sampling effort per 0.1 ha (ASE, person-hours; ±SD); mean Resource-based sampling effort (RSE, person-hours per 1 m^2 of foliage; ±SD); mean Accessibility (% of foliage accessed; ±SD); average Team size in the field and lab combined; and Sampling period (month and year). See S2 Table for data by individual plots and all arthropod groups.

Site	Forest type	Maximum tree height (m)	Plots	Method	Number of stems	Sampled leaf area (m^2)	Leaf-chewing larvae	Active leaf mines	ASE (person-hours)	RSE (person-hours)	Accessibility (%)	Team size	Sampling period
Tomakomai (JPN)	Temp. lowland (P)	22.8	2 x 0.1ha P	Crane	92 ±16	1,219 ±116	8,300 ±825	385 ±196	1,330 ±178	1.10 ±0.25	82.0 ±0.1	7	May-Aug 14; May-Aug 15
Lanzhot (CZE)	Temp. lowland (P)	45.0	2 x 0.1ha P	Cherry picker	29 ±6	1,208 ±194	4,891 ±576	148 ±60	1,128 ±305	0.92 ±0.10	89.3 ±6.3	8	May-Aug 13; May-Aug 14; May 15
Mikulcice (CZE)	Temp. lowland (P)	33.6	1 x 0.1ha P	Felling	53	1,137	2,352	2717	1,512	1.33	83.4	10	May—June 13
Toms Brook (USA)	Temp. lowland (P)	30.7	2 x 0.1ha* P	Felling	81 ±18	1,793 ±132	2,608 ±428	564 ±470	1,604 ±326	0.89 ±0.12	76.5 ±1.0	7	Apr-Aug 16; Apr-Aug 17
San Lorenzo (PAN)	Trop. lowland (P)	35.0	3 x 0.1ha P	Crane	91 ±6	2,023 ±303	808 ±754	1,007 ±965	2,404 ±416	1.19 ±0.03	83.3 ±5.5	5	May 16-Apr 17
Wanang (PNG)	Trop. lowland (P+S)	74.2	1 x 1.0 ha (P) 1 x 1.0 ha (S)**	Felling	120 ±30	3,377 ±1050	1,354 ±705	185 ±85	1,880 ±474	0.58 ±0.14	82.9 ±4.0	21	Jan 06-Nov 07
Numba (PNG)	Trop. highland (P+S)	49.6	2 x 0.2 ha (P) 1 x 0.2 ha (S)**	Felling	143 ±17	3,658 ±1403	1,118 ±321	60 ±32	1,800 ±642	0.52 ±0.19	81.6 ±3.5	16	May 13-Jun 14
Yawan (PNG)	Trop. highland (P+S)	65.7	4 x 0.2 ha (P) 5 x 0.2 ha (S)**	Felling	133 ±62	3,591 ±620	1,103 ±862	199 ±152	1,183 ±488	0.33 ±0.14	82.9 ±4.0	16	Jul 10-Dec 12

* one of the 0.1 ha plots consisted of a 0.06 ha plot and a 0.04 ha plot separated by a 50 m gap

** these plots were divided into 0.1 ha plots for the purpose of the analysis

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Arthropod sampling

The requirements for accessing the forest canopy and obtaining live arthropods dramatically limit the range of suitable methods for the study of quantitative arthropod interaction networks (Table 1). We sampled arthropods from felled trees, and from standing trees using canopy cranes or cherry pickers (Fig 1). Arthropods were, as far as possible, completely sampled from all trees with DBH ≥5 cm. The percentage of the canopy accessed was visually estimated for each tree (Appendix 1). We sampled on days without strong rain or wind to mitigate safety risks and lowered arthropod activity due to harsh weather. The focal arthropod groups included all live leaf-chewing insect larvae (free feeding and semi-concealed), leaf mines, galls (insects and mites), spiders, and ants (foraging and nesting; S1 Table). Some species of galls

were extremely abundant, making their complete sampling impractical. In such cases, we selected 3–5 branches each with 100–500 leaves, calculated the mean number of galls per leaf per branch, and used the resulting values to estimate the total abundance on the respective tree (Appendix 1).

Felling. Felling trees as a standardized destructive method is only suitable when it does not contribute to net deforestation. During our operations, we took advantage of ongoing logging operations (Mikulcice, Toms Brook) and shifting agriculture (PNG sites); no plot was cleared solely for sampling. All projects were conducted in close collaboration with the local community and land owners.

Sampling began with the clearing of the understory, followed by the felling of trees with DBH $\geq$5cm. One tree was felled at a time, starting with the shortest. Lianas on trees were cut prior to felling in order to free up the focal tree from its neighbours. Felled individuals were directed into gaps created by previous felling. Once felled, the entire tree (trunk included) was searched and all focal arthropods hand collected, a process taking anywhere from minutes to several hours, depending on the crown size. Prompt work minimized the loss of arthropods through dispersal or predation. It also prevented the contamination by foraging ants and spiders from the ground. Using division of labour, each team member focused primarily on one arthropod group, but would also contribute to the collection of secondary groups. Trees were always fully sampled on the day of felling, and necessitated teams of 7–21 members, dependent on study site and season (Table 2).

Unlike sampling from cranes and cherry pickers, felling allows the sampling of arthropods dwelling in large branches and trunks, such as nesting ants (Table 1). At felling sites, we intensively searched every tree for ant nests and foraging ants with a team of two to three collectors, as described in Klimes et al. [16]. Foraging ants were collected first, before searching for ant nests by cutting branches, inspecting live and dead twigs, by dissecting parts of the trunk and bark, and by inspection of epiphytic aerial soil (Appendix 1).

Conversely, felling is not suitable for mobile, flying herbivores [36]. Even non-flying herbivores may become dislodged when the crown forcefully impacts the ground. If this were a serious concern, the ratio between endophytic herbivores and leaf-chewing larvae would depend on the method. However, the ratio of leaf-chewing larvae to active miners sampled in individual 0.1 ha plots did not differ among the methods (χ^2 (2) = 2.57, p = 0.2764) when compared by linear mixed-effect models using the 'lmer4' R package [46], with site as a random effect.

Crane. We sampled arthropods from canopy cranes in Tomakomai and San Lorenzo. In Tomakomai, the crane is 25 m high, covers ca 0.5 ha of forest, and is operated by researchers from the gondola. In San Lorenzo, the crane is operated by a driver. The maximum accessible height from the gondola is 40.5 m. The crane covers almost 1.0 ha of tropical forest [37].

There were 4–7 team members working in the field, typically including 2 members sampling from the crane (canopy team), 1–2 members sorting samples on the ground (ground team), and possibly 1–2 members accessing larger mid-story trees by climbing (climbing team). The canopy team sampled branches starting at the tip and working towards the base, in order to minimize arthropod loss during sampling. Arthropods were sampled by beating onto a beating tray, followed by a visual search and hand collection of any remaining arthropods. The canopy team was assisted by an additional member during periods of peak arthropod abundance. The samples were regularly delivered to the ground team for sorting.

Sampling from the crane was augmented with other methods. The canopy team accessed understory trees from ladders. Step ladders were ideal for sampling 3–5 m tall trees. For sampling at heights up to 8 m, or on sloped terrain, modular ladder poles were more efficient and stable. In addition, more complex forest architecture, as in San Lorenzo, required the climbing

team. Using a single rope technique, they accessed those mid-story trees inaccessible from the gondola or ladders (Fig 1).

Cherry picker. A cherry picker (an elevated truck-mounted work platform) was employed in Lanzhot. The 20 ton vehicle was transported by truck to the site, thus necessitating a forest access road. We used a Platform GENIE Z-135/70 JRT (Genie Industries, Redmond, WA, USA), which was equipped with a retractable arm enabling canopy access up to 43 m. The arm was operated by researchers directly from the basket. This four-wheel drive model can operate on gravel or clay forest roads, but not on off-road terrain. Plots were set up along a forest road with a firm dirt surface (~4 m wide, and completely covered by forest canopy) in order to provide good access to the plot from a single straight trajectory and to avoid having to manoeuvre the cherry picker between trees. Two team members sampled trees starting from the base and working towards the treetop. Arthropods were sampled using a beating tray combined with hand collection of any remaining individuals, before a final manual search by both workers. Samples were delivered to the ground team for processing before transportation to the laboratory. There were 2–6 people processing samples in the ground team, depending on insect abundance.

Leaf area estimates

We calculated the leaf area of sampled trees in order to standardize arthropod abundance and allow cross-site comparisons (Appendix 1).

At the felled sites, we quantified leaf biomass directly by defoliating each tree and weighing the fresh foliage. Mature and young leaves were sampled and weighed separately immediately following herbivore sampling. Care was taken that only leaves, with no other plant parts such as twigs and flowers, were sampled. At Mikulcice and Toms Brook sites, where team size was limited, only 50% or 25% of the canopy was defoliated on the largest trees and the results extrapolated to 100%. This measure was taken to ensure the complete sampling of large trees on the day of felling.

At the crane and cherry picker sites, defoliating trees and weighting the biomass was not possible. Instead, we visually estimated the number of young and mature leaves on standing trees. These estimates were conducted separately for every branch sampled for arthropods. The estimates were carried out for branches with ca 500 leaves each by two persons from the canopy team. The mean value of the two estimates was taken. The branch level estimates for the given tree were then summed to give an estimate for the entire tree. This method yielded more reliable results than if estimating leaves on larger branches or whole trees.

At all sites, a random sample of leaves from each tree was then arranged on a 50 x 50 cm board with white background (the "leaf frame") and photographed. One frame each of young and mature leaves was processed for small trees (DBH $<$15 cm), while at least two frames were processed for larger trees. The leaf area of each sample was then calculated using ImageJ 1.48 [38]. For felled trees, we included the weight of the sample to obtain the area to weight ratio. For the trees sampled from cranes and cherry pickers, we divided the leaf area of the sample by the number of leaves in the frame to obtain the mean area per leaf.

Finally, we calculated the total sampled leaf area for each tree using (i) the total leaf biomass and the area to weight ratio from the photographed sample for the felled trees, or (ii) the estimated total number of leaves on the tree multiplied by the mean leaf size of the photographed sample for the crane and cherry picker trees.

Sample processing

In Tomakomai, Mikulcice, and Lanzhot, pre-sorting, photographing, and labelling of samples was done in the field by a team consisting of 1–6 members, depending on arthropod

abundance (Appendix 1). This made subsequent sorting in the lab much faster. Smaller trees in Toms Brook were treated the same way. Otherwise, samples were processed entirely in the laboratory.

We assigned all leaf-chewing insect larvae, galls, and mines to morphospecies according to their morphology [21]. Each morphospecies was given a unique code name and was photographed. We preferred to assign initial morphotypes *de novo* per each individual tree sampled instead of using a complex system of morphospecies across all trees within the plot or even across multiple plots (Appendix 1). This approach is rapid and resistant to errors as even incorrect morphotyping does not generate false host plant records. It requires a second step where individual morphospecies are cross-referenced across all trees on completion of sampling. It is suitable for taxonomically poorly known and species diverse samples, where per-guild richness for an entire plot could reach hundreds of morphospecies.

We reared larval insect herbivores to adults or parasitoids (Appendix 1). Only in Toms Brook, where insect taxonomy and host associations are well known, were leaf-chewing larvae immediately stored in ethanol due to the overwhelming logistics of rearing all. We preserved larvae that died during rearing, the larvae from Toms Brook, spiders, and representative samples of all ant castes from each nest or foraging event in vials with 95% ethanol for subsequent DNA barcoding. The results of DNA barcoding along with reared adults are being used to refine morphospecies concepts and assign final identifications [21, 23, 39–41]. See Data Accessibility section for details on the publicly available sequences.

Statistical analysis: Comparing methodological approaches

Non-plot-based studies typically focus on i) abundant tree species or ii) a taxonomically/phylogenetically representative selection of species e.g. [42, 43, 44]. In order to compare plot-based and non-plot-based methods, we derived both types of data from plot-based data on plant-caterpillar interactions in 0.8 ha of PNG highland primary rainforest Yawan [23]. This dataset was chosen because it is species-rich, the caterpillar species exhibit various levels of host specificity, and were already identified to an acceptable level. Only living trees identified to species and caterpillar species/morphospecies with confirmed host associations were included. The pruned dataset, representing 0.8 ha of primary forest (eight x 0.1 ha plots), included 113 tree species and 186 caterpillar morphospecies.

We computed network statistics and structure from the Yawan primary plots, and compared them with networks comprised of i) the most abundant (in terms of amount of foliage based on leaf area calculations) tree species and ii) a taxonomically representative selection of primary tree species including all tree families that had at least 200 m^2 of foliage sampled for arthropods.

For type (i) networks, we combined all 0.1 ha plots to represent a larger patch (0.8 ha) of rainforest and ranked tree species in order of decreasing amount of total foliage. We then selected the species whose cumulative total foliage represented 20%, 40%, 60%, and 80% of the total foliage of the 0.8 ha patch (3, 7, 15, and 31 species, respectively). In each threshold category, we rarefied the foliage amount of each species (F_{sp}) to equal the average total foliage of a 0.1 ha plot divided by the respective number of tree species (F_{thresh}). This was achieved by randomly selecting individual trees until $F_{sp} >= F_{thresh}$. The final trees (T_f) are only partially sampled of their caterpillars (C_f) so that $F_{sp} = F_{thresh}$ (if we use $0.25T_f$ then we take $0.25C_f$ randomly selected caterpillars, rounded to the nearest integer). The individual trees and the caterpillars found on them made up the networks from which statistics were computed. The process was repeated 100 times for each category to account for the random tree (and caterpillar) selection.

For type (ii) networks (taxonomical selection), we limited the dataset to tree species and families that had at least 200 m^2 of foliage sampled (200 m^2 equals ca 0.7% of the total foliage in the 0.8 ha primary forest patch). We selected the most abundant species per family in terms of leaf area (19 species and families selected). With this tree selection, rarefaction then proceeded as per type (i) networks.

We focused our comparison between plot-based and non-plot-based networks on: i) connectance (realised proportion of possible links), ii) web asymmetry (balance between numbers of species in the two levels; positive values indicate higher proportion of higher trophic level species), iii) nestedness (temperature of the matrix; 0 means high nestedness, 100 means chaos), iv) species richness of caterpillars, v) weighted generality (mean effective number of host species per caterpillar species), and vi) weighted vulnerability (mean effective number of caterpillar species per host species) as defined and computed in 'bipartite' package [45]. The network parameters were compared using 95% confidence intervals.

Furthermore, we compared the efficiency of each sampling method we used across the plots. We expressed the method efficiency as i) **Foliage accessibility** per plot (the average percentage of accessible foliage), ii) **Area-based sampling effort** (**ASE**) required to sample each 0.1 ha plot (in person-hours), and iii) **Resource-based sampling effort** (**RSE**) required to sample 1 m^2 of foliage (in person-hours). Only time spent on sample collection and sorting in the field was counted towards the sampling effort. Workers helping with logistics (chainsaw operators or the crane driver in Panama) were excluded. We modelled the relationship between these components of sampling efficiency and the sampling **Method** (felling, crane, cherry picker), **Forest type** (temperate, tropical lowland primary, tropical lowland secondary, tropical highland primary, tropical highland secondary), the **Number of stems** (DBH $\geq$ 5 cm) per plot, and **Sampled leaf area** using linear mixed-effect models as implemented in the R package 'lmer4' [46]. **Foliage accessibility** was arcsine-transformed and **sampling effort** log-transformed. We used **Site** as a random factor in all mixed-effect models (S2 Table). Model simplification by forward selection was employed to produce the most parsimonious model based on Akaike's Information Criterion (AIC). All analyses were performed in R software version 3.4.0 [47].

Results

In total, we sampled focal arthropod groups from 5.3 ha of forest, representing 6,280 trees and 167,744 m^2 of foliage (Table 2). We sampled 89,243 leaf-chewing larvae, 14,547 active mines, 135,446 abandoned mines, 28,698 spiders, 35,343 ant individuals, 3,487 ant nests, and sampled or estimated abundance of 2,963,942 insect and mite galls (S2 Table).

All non-plot-based data types showed highly biased network structure towards higher connectance, higher web asymmetry, and higher nestedness temperature when compared to the plot-based data (Fig 3). Non-plot-based data using the most abundant tree species representing 20% of the foliage in the local forest had the highest caterpillar richness while those using a taxonomically representative selection of tree families had the lowest caterpillar richness. Non-plot-based data using the most abundant tree species representing 20% and 40% of the foliage in the local forest showed lower generality than the plot-based data. Differences in vulnerability were less pronounced mainly because of the high variability in plot-based data. However, vulnerability was highest in the data using the most abundant tree species representing 20% and 40% of the foliage in the local forest.

On average, **Foliage accessibility** was 82.5% ±3.9% (mean ±SD) foliage in felled plots, 82.7% ±3.3% foliage in plots sampled by canopy crane, and 89.3% ±6.3 foliage in plots sampled by cherry picker (S2 Fig). **Foliage accessibility** correlated with **Method** (χ^2 (2) = 6.91,

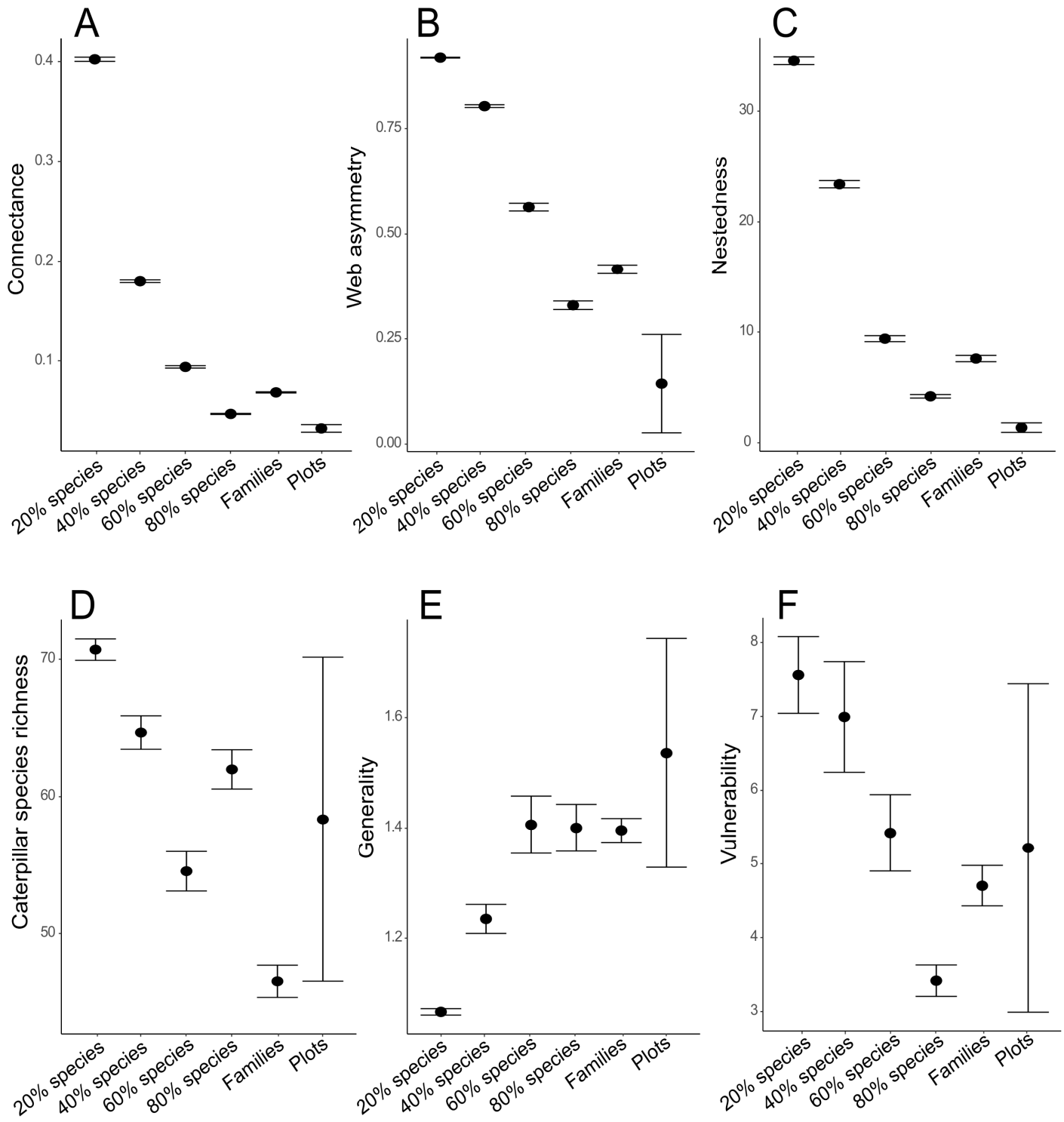

Fig 3. Parameters of plant-caterpillar interaction networks based on the plot-based data (Plots) and simulated non-plot-based data where individual tree species were sampled with equal effort. The simulated data represent a non-plot-based approach focusing on locally abundant tree species representing a certain amount of the foliage in the forest (20, 40, 60, or 80% species) or a representative selection of tree families (Families). The results are based on Yawan primary forest dataset from Redmond et al. [23]. The compared network parameters include connectance (A), web asymmetry (B), nestedness (C), species richness of caterpillars (D), weighted generality (E), and weighted vulnerability (F). All simulated datasets were rarefied to the average leaf area of a 0.1 ha plot. All rarefactions were repeated 100-times. Points show mean. Bars show 95% confidence intervals.

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p = 0.0254). The optimum model, after simplification, included the fixed effects **Forest type** (highest in lowland and highland secondary tropical forests), **Method** (highest from the cherry picker), **Number of stems** (positive correlation), and **Sampled leaf area** (negative correlation) (χ^2 (8) = 64.02, p < 0.0001) (S4 Table).

The average **ASE** required to sample a 0.1ha plot was 1583 ±579 person-hours (mean± SD) for felled trees, 1867 ±673 for sampling by canopy crane, and 1128 ±305 for sampling by cherry picker. **Method** did not have a significant effect on **ASE** (χ^2 (2) = 1.49, p = 0.4740). The optimum model that explained differences in **ASE** included the fixed effects **Number of stems** (positive correlation) and **Forest type** (highest in lowland primary tropical forests) (χ^2 (5) = 95.24, p < 0.0001; S4 Table).

The average **RSE** to sample 1 m^2 of foliage was 0.51 ± 0.24 (mean± SD) person-hours for sampling felled trees, 1.14 ±0.15 for sampling by canopy crane, and 0.92 ±0.10 for sampling by cherry picker. **Method** did not have a significant effect on **RSE** (χ^2 (2) = 3.52, p = 0.1722). The optimum model explaining differences in **RSE** included the fixed effects **Number of stems** (positive correlation), **Sampled leaf area** (negative correlation), and **Forest type** (highest in temperate forests) (χ^2 (6) = 80.75, p < 0.0001; S4 Table).

Discussion

We propose a plot-based approach to studying arthropod interaction networks, using three methods for sampling a continuous area of forest canopy. Plot-based standardisation means that frequent associations can be distinguished from those that are casual or rare [48, 49]. Focusing on a selection of abundant tree species or representative families sampled with a standardized sampling effort skews the proportions between rare and common interactions. As expected, this resulted in higher connectance, high web asymmetry and higher nestedness in the simulated non-plot-based data. This is not surprising as all these parameters are linked to the network size, which has been reduced under the selective sampling scenario. In addition to reducing network size, non-plot-based sampling focused on abundant or phylogenetically distinct hosts can affect the patterns recovered in host specificity and diversity. This is because such hosts typically harbor distinct arthropod communities. Locally abundant tree species tend to harbor higher diversity of herbivores than rarer hosts [50]. Focusing on such hosts can lead to over-estimations of diversity. On the other hand, hosts from isolated or chemically distinct families can have relatively species poor herbivore communities [51]. Emphasizing such hosts in the data can lead to under-estimations of diversity. Also, many herbivores are shared between congeneric or confamilial hosts while the amount of shared herbivores decreases with the host phylogenetic distance [44]. Specificity of interactions can thus be biased in datasets that include skewed proportions of such hosts although this trend was not particularly pronounced in our simulated data.

Plot-based sampling provides a robust description of the community structure as one can assume that the interactions are completely censused for the proportion of the canopy successfully sampled (Fig 4). One can then test and improve the performance of models that predict trophic interactions in real communities by decomposing the effects of abundance, plant characteristics and arthropod community composition [28, 41]. Derived food-web metrics are comparable on a common area basis, and may identify processes shaping communities of canopy arthropods across various habitats, ecosystems, or geographic regions [21].

Plot-based analyses thus represent an ideal counterpart to those based on a stratified selection of focal species sampled with an equal sampling effort. Methods that employ equal sampling effort are advantageous for studying the host specialization of herbivores, the effects of host traits on herbivore communities, or insect-plant interactions in a phylogenetic framework

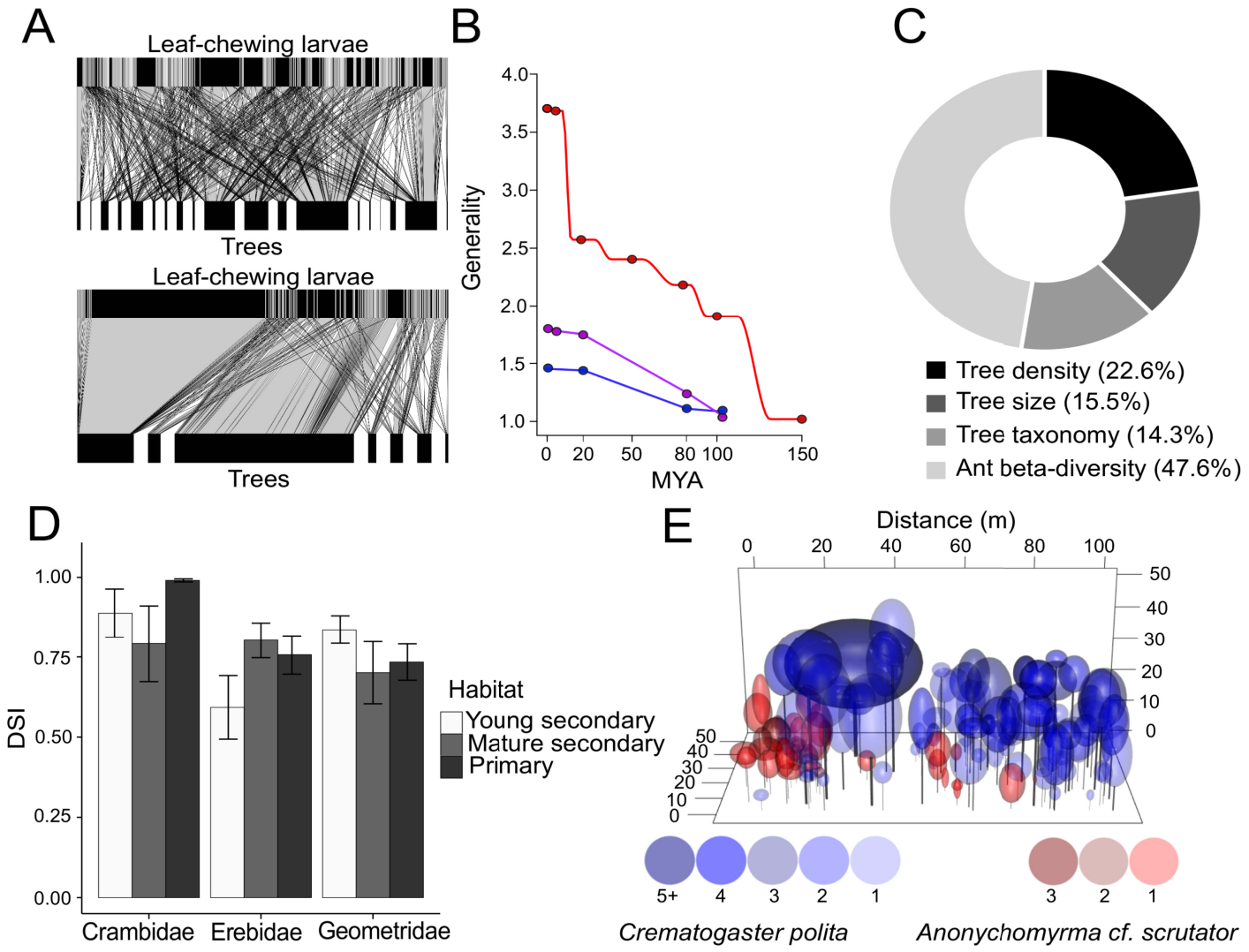

Fig 4. Example results from plot-based sampling. Construction of comparable quantitative interaction networks (A: plant-caterpillar food-webs from two 0.1 ha plots with contrasting herbivore and tree diversity; based on data from Volf et al. [21]). Such networks can be used to quantify effects of plant traits or phylogeny on arthropod communities (B: effects of host phylogeny on caterpillar food-webs quantified by change in generality from herbivore data collated according to the time of divergence of their hosts (in Tomakomai (red), Lanzhot (purple), Mikulcice (blue)); based on data from Volf et al. [21]). The relative contribution of such effects can be decomposed, allowing the prediction of arthropod community composition (C: the proportional difference in total ant species richness between primary and secondary forest in Wanang due to the effects of vegetation composition and species turnover; based on data from Klimes et al. [28]). Furthermore, standardized measures of herbivore specialization can be made, enabling meaningful comparisons across habitats and taxa with variable phylogenetic diversity and plant abundance (D: mean Distance Based Specialisation Index (DSI*) +/- SE for Crambidae, Erebidae, and Geometridae along a successional gradient in Yawan; based on data from Redmond et al. [23]. Finally, we can analyse spatial patterns in canopy arthropod communities (E: distribution of tree canopy nest density in the two most abundant ant species in 0.4 ha of Wanang forest (only trees with nests are shown); based on Klimeš and Mottl (*unpublished data*)).

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e.g. [14, 30]. However, modern methods enable the measurement of host specialization with respect to host phylogeny or chemical similarity in plot-based data also [52]. Furthermore, a plot-based approach can be used to investigate spatial distribution of arthropods across the forest canopy and their impact on competitors and other trophic levels. This is important, for instance, when considering competition among ants where canopy connectivity and structure play important roles in forming ant communities [16]. Furthermore, herbivores may have density-dependent effects on plant survival that need to be studied in a spatially explicit framework [53].

One limitation of plot-based sampling methods is that the logistical challenges necessitate relatively large teams and overall effort. Despite our expectations, however, all methods demanded comparably high sampling effort, with none being significantly more efficient. Such prerequisites stem from the need to census all parts of the canopy, including those difficult to access, in order to reconstruct truly quantitative interaction networks [7, 21]. Foliage accessibility positively correlated with the number of stems in the plot, probably because many of the trees in densely vegetated plots were small and easier to access. On the other hand, the number of stems within a plot increased both types of sampling effort that we quantified. ASE (total effort per 0.1ha plot) was highest in lowland primary tropical forests characterized by relatively high stem density and large trees difficult to sample. RSE (effort per 1 m^2 of foliage) was highest in temperate forests. This may be because arthropod density is generally higher in temperate forests [5], especially during the spring abundance peak.

High effort per site prevented a rigorous methodological comparison where the same forest is sampled by all three methods. All methods enabled access to over 80% of the foliage. But the unbalanced distribution of methods may be one reason why the cherry picker appeared to provide better access to the canopy than felling or cranes. Similarly to Corff and Marquis [20], we operated the cherry picker in almost optimal conditions in temperate forest where plots were close to an access road and the trees could be accessed from a straight trajectory. Operating in less favourable conditions would dramatically decrease foliage accessibility or require employing additional methods. Sampling from cranes also had to be supplemented by other techniques at both our crane sites. While sampling by other techniques represented a small proportion of sampling effort in the temperate Tomakomai forest, it considerably increased the sampling effort in San Lorenzo tropical rain forest. In San Lorenzo, only 49% of the trees (representing 58% of the foliage sampled) were accessed solely by crane.

Each method also has its own specifics unrelated to its overall efficiency. Felling generally requires larger teams [22, 23] as felled trees need to be sampled immediately. Cranes and cherry pickers allow proceeding at a slower pace with a smaller team e.g. [12, 20]. The three methods are also not completely comparable in terms of the sampled arthropod groups. All were suitable for sampling endophytic and exophytic non-flying arthropods. Less mobile flying herbivores, such as aphids or psyllids, were also well represented in our samples, although they were not the focus of our study. Felling was the only method which enabled sampling of nesting ants, which can represent an important proportion of the canopy arthropods [16]. Quantitative sampling of highly mobile macroscopic arthropods (adult beetles, flies or true bugs) was not possible by these methods, although they were better represented in crane and cherry picker samples.

Other methods, such as fogging, may be more suitable for surveying highly mobile arthropods [25]. Such methods can also strongly reduce the required team size and effort. To assess trophic interactions, however, they would need to be combined with a massive barcoding effort so the sampled arthropods could be reliably assigned to their host-plants. A molecular approach to assessing trophic interactions is becoming increasingly popular [15] and can be especially useful in well studied or less diverse communities. However, the approach may face identification limitations in diverse communities that include a high proportion of closely related and/or hybridizing hosts. Indeed, standard barcode markers may fail to provide a sufficient resolution for such hosts unless combined with specifically selected ones [54]. The implementation of such methods for large scale plot-based sampling should, therefore, be carefully considered. Furthermore, the sampling of endophytic or semi-concealed herbivores and ant nests would require the employment of additional methods.

We suggest that a global network using the methods described for area-based sampling would provide important insights into the processes of food web assembly and dynamics [33].

To that end, we propose a network of permanent plots where canopy arthropods and their interactions would be censused by non-destructive sampling. The network of permanent plots could benefit from collaboration with the global network of ForestGEO plots [32] which generates major insights into forest community ecology. We suggest that plots of 0.1 ha are an appropriate size to be sampled from cranes or cherry pickers, and which allow for repeat surveys, while keeping the required effort manageable. A single 0.1 ha plot census can yield information on more than 100,000 canopy arthropods and their interactions, thus the potential to make significant contributions to arthropod ecology research is huge.

The network of permanent plots should ideally be augmented by a larger network of temporal plots to be sampled by felling. Despite a slight revival in canopy crane construction [55], such platforms are still missing from vast regions, including Africa and North America. Similarly, opportunities for the use of cherry pickers remain limited in many forests. The sampling of 0.1 ha plots by felling thus seems to be the only widely applicable option in many regions. These plots could be highly replicated and ideally adjacent to the ForestGeo plots.

Sampling canopy arthropods by felling can become a salvage sampling strategy to obtain data on arthropod communities being lost due to ongoing deforestation. There has been considerable activity in the past decade focused on constructing large-scale experiments, such as planting forest stands of a given richness [56], or manipulation of landscape fragmentation [57], which deepen our understanding of how ongoing changes in forest structure affect ecological interactions. However, ecologists have been slow to take advantage of ongoing logging operations, urban development, or shifting agriculture for destructive arthropod and plant sampling to salvage the data. Yet, such data in combination with data from permanent plots would enable the exploration of trends in arthropod networks along major environmental gradients [23]. Furthermore, the detailed data obtained by our methods could be used for modelling forest composition and arthropod interactions. Combining such models with high-throughput methods, such as remote sensing, that allow forest composition to be assessed may enable us to predict the basic characteristics of plant-arthropod interactions over large spatial scales [58]. Ultimately, the application of the outlined methods could lead to high-impact results with far-reaching consequences, such as the prediction of the effects of forest degradation on forest arthropod communities, and the identification and preservation of arthropod diversity hotspots in the world's forests.

Supporting information

S1 Fig. Seasonal trends in abundance of leaf chewing larvae and active miners.
(DOCX)

S2 Fig. Foliage accessibility.
(DOCX)

S1 Table. Site characteristics.
(DOCX)

S2 Table. Characteristics of individual 0.1 ha plots and number of arthropods sampled.
(DOCX)

S3 Table. Monthly trends in abundance of caterpillars and active miners.
(DOCX)

S4 Table. Variables with a significant effects on Foliage accessibility, Area-based sampling effort, and Resource-based sampling effort as selected by forward selection in linear mixed

effect models.
(DOCX)

S5 Table. List of staff, interns, students, volunteers, and local assistants who helped with the sampling.
(DOCX)

S1 Appendix. Sampling protocols.
(PDF)

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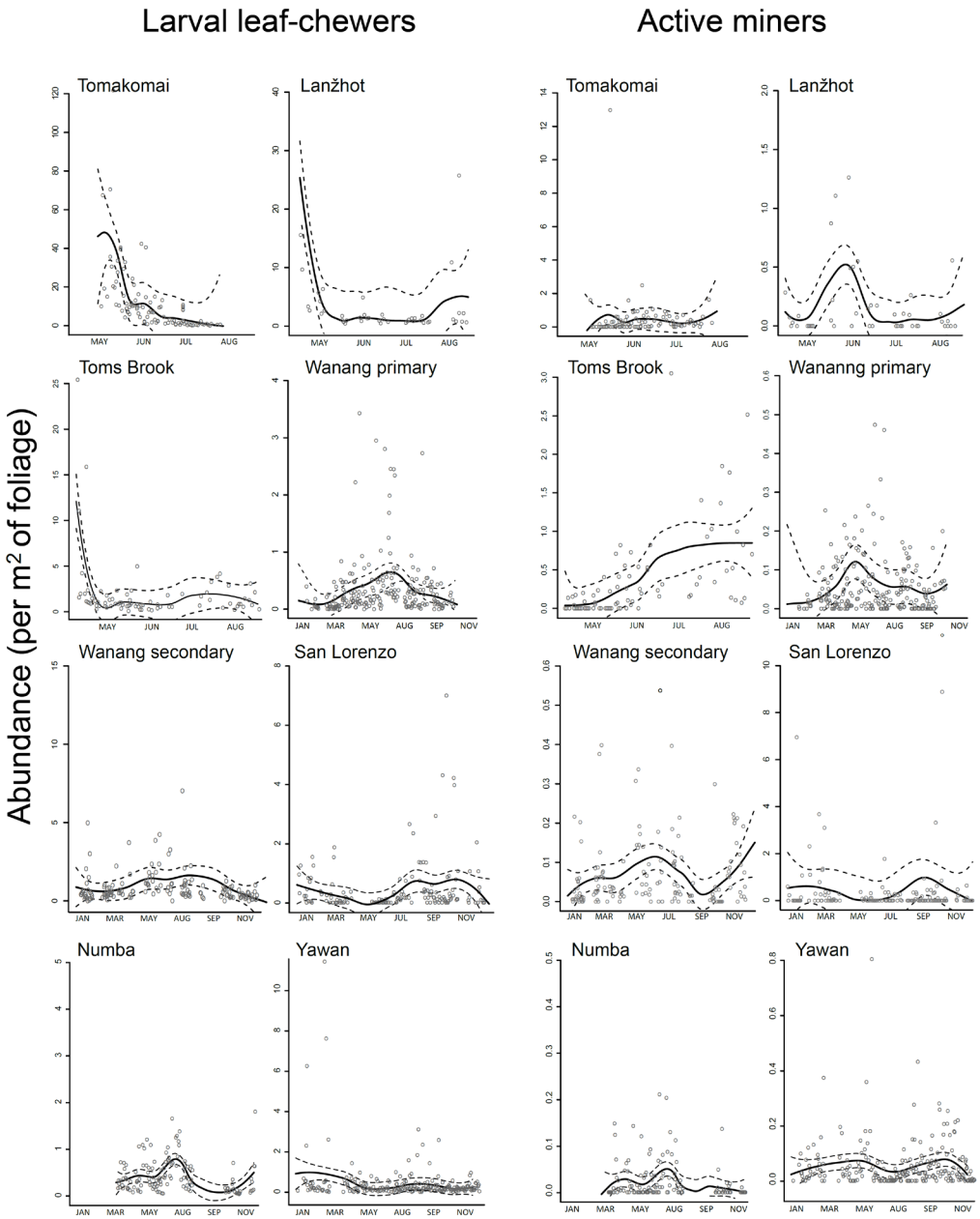

S1 Fig. Seasonal trends in abundance of leaf chewing larvae and active miners across the plots sampled for multiple months (Tomakomai, Lanzhot, Toms Brook, San Lorenzo, Wanang, Numba, Yawan). The data points represent number of caterpillars and active miners per 1 m^2 of foliage on individual days of sampling. The seasonal trend was modelled with a loess smoother (solid line). Dashed lines show confidence intervals. The abundance was standardized by leaf area. Data from individual 0.1 ha plots sampled at the listed sites were combined. The data from Wanang primary and secondary forest plots were kept separate to illustrate possible differences between primary and secondary forest. Two outlier data points with leaf-chewer abundance of 226 and 18 are not shown in the case of Tomakomai and Wanang secondary, respectively. One outlier data point with active miner abundance of 25 is not shown in the case of San Lorenzo.

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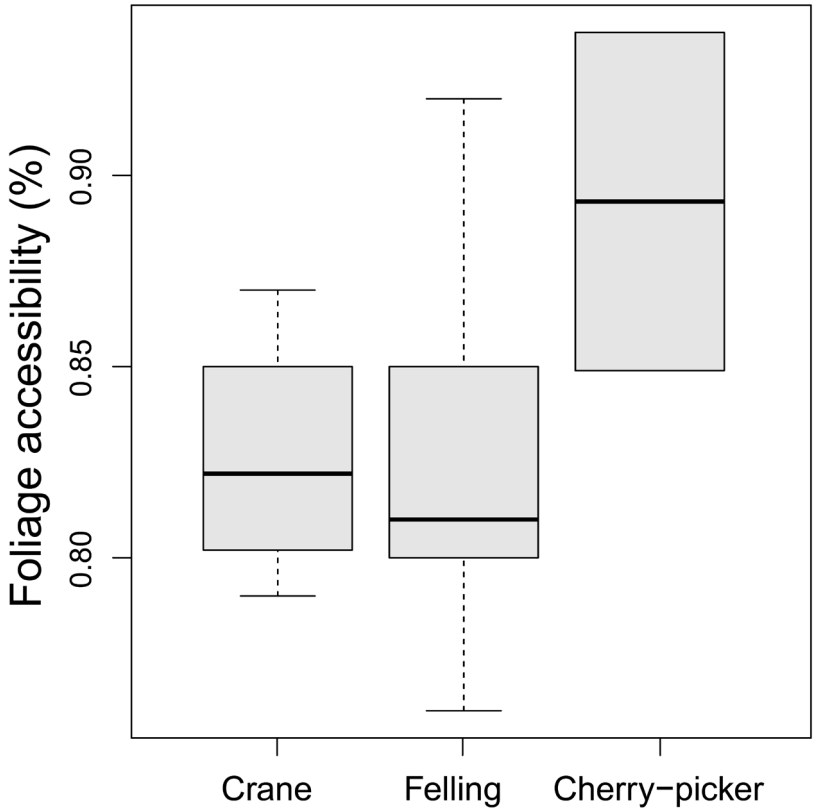

S2 Fig. Foliage accessibility (% of foliage possible to access in individual 0.1 ha plots) facilitated by individual methods. Canopy accessibility was correlated to the used method (χ^2 (2) = 6.91, p = 0.0254). The highest accessibility was achieved by the cherry-picker, which operated in optimal conditions of a temperate forest.

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S1Table. Site characteristics including latitude, longitude, altitude, average temperature, and annual rainfall.

Site	Latitude	Longitude	Altitude (m asl)	T (°C)	Rainfall (mm)	Reference
Tomakomai (JPN)	42° 43' N	141° 34'E	90	5.6	1,450	[1]
Lanzhot (CZE)	48° 48' N	17° 5'E	152	9.0	525	[2]
Mikulcice (CZE)	48°41' N	16°56'E	164	9.0	525	[2]
Toms Brook (USA)	38°55' N	78°25' W	230	12.7	970	[3]
San Lorenzo (PAN)	9°16' N	79°58' W	130	26.0	3,140	[3]
Wanang (PNG)	5° 14' S	145° 4' E	150	25.8	4,000	[4]
Numba (PNG)	5°44' S	145°16' E	700	22.3	3,000	[5]
Yawan (PNG)	6° 9' S	146° 50' E	1,800	16.2	3,000	[5]

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S2 Table. Characteristics of individual 0.1 ha plots and number of arthropods sampled. All living trees and climbers of DBH $\geq$ 5 cm with foliage and known taxonomic identity and their arthropods are included. Larval leaf-chewers refer to all free living or semi-concealed leaf-chewing insect larvae. All sampled leaf-chewing larvae, including those without confirmed host record or identification, are listed. All sampled leaf mines, including those without identification, are listed. All fungal galls and mines and galls with uncertain status (e.g. possible fungal galls and pathogen damage) were removed from the list. Gall numbers marked with an asterisk refer to insect galls only (other records include mite galls as well). Spider numbers marked with two asterisks indicate that spiders were sampled from half of the plot (i.e. 0.05 ha) only. Arthropod groups, which were not sampled or their census counts are currently not available in the respective plot are marked as NA.

Site	Plot	Method	Forest type	Area-based sampling effort (man hours)	Resource-Based sampling effort (man hours)	Foliage accessibility (%)	Stems (DBH≥5cm)	Leaf area (m^2)	Larval leaf-chewers	Active mines	Abandoned mines	Galls	Spiders	Foraging ants	Ant nests
Tomakomai	Tomakomai P1	Crane	Temperate	1,204	0.93	83	81	1,301.00	8,883	231	2.086	527,015	515	201	NA
Tomakomai	Tomakomai P2	Crane	Temperate	1,456	1.28	81	103	1,136.70	7,716	520	1.429	531,586	534	130	NA
Lanzhot	Lanzhot P1	Cherry-picker	Temperate	1,344	1.00	85	32	1,346.15	4,484	106	5.779	311,063	5,810	984	NA
Lanzhot	Lanzhot P2	Cherry-picker	Temperate	912	0.85	94	24	1,071.34	5,298	190	5.590	321,015	4,164	541	NA
Mikulcice	Mikulcice P1	Felling	Temperate	1,512	1.33	83	53	1,137.32	2,370	2.717	2.041	398,265	1,230	943	79
Toms Brook	Toms Brook A	Felling	Temperate	1,835	0.74	77	93	1,886.50	2,910	232	641	402,426	NA	582	65
Toms Brook	Toms Brook B	Felling	Temperate	1,374	0.76	76	68	1,699.70	2,305	904	2337	224,286	NA	538	77
Numba	Numba_PA12	Felling	Tropical Highland Primary	1,950	0.49	80	137	2,617.69	1,391	102	1591	942*	949	NA	432
Numba	Numba_PA34	Felling	Tropical Highland Primary	1,650	0.46	83	133	2,172.11	1,145	86	2220	615*	1,042	NA	419
Numba	Numba_PB12	Felling	Tropical Highland Primary	2,175	0.33	79	169	4,434.37	1,189	61	3231	454*	1,666	NA	NA
Numba	Numba_PB34	Felling	Tropical Highland Primary	2,775	0.34	78	145	6,038.78	1,418	56	4853	452*	1,667	NA	NA
Numba	Numba_S1+S2	Felling	Tropical Highland Secondary	1,050	0.34	84	121	3,148.80	1,028	45	1351	46*	1,089**	NA	136
Numba	Numba_S3+s4	Felling	Tropical Highland Secondary	1,200	0.41	87	154	3,533.30	539	12	2526	164*	NA	NA	115
Yawan	YPA12	Felling	Tropical Highland Primary	1,500	0.25	78	114	4,386.80	1,495	147	1186	394*	517	927	111
Yawan	YPA34	Felling	Tropical Highland Primary	1,350	0.21	80	106	3,279.60	481	68	1078	211*	77**	1,079	177
Yawan	YPB12	Felling	Tropical Highland Primary	1,050	0.31	77	104	4,223.29	675	482	2632	182*	495	NA	NA
Yawan	YPB34	Felling	Tropical Highland Primary	900	0.44	82	124	4,217.79	626	359	3812	260*	814	NA	NA
Yawan	YPC12	Felling	Tropical Highland Primary	1,200	0.45	81	139	3,931.24	3,601	266	876	171*	483	400	118

Site	Plot	Method	Forest type	Area-based sampling effort (man hours)	Resource-Based sampling effort (man hours)	Canopy sampled (%)	Stems (DBH≥5cm)	Leaf area (m^2)	Caterpillars	Active mines	Abandoned mines	Galls	Spiders	Foraging ants	Ant nests
Yawan	YPC34	Felling	Tropical Highland Primary	1,500	0.36	81	117	3,433.94	1,014	133	2650	202*	602	485	72
Yawan	YPD12	Felling	Tropical Highland Primary	1,275	0.56	81	104	2,845.71	419	235	4742	202*	452	NA	NA
Yawan	YPD34	Felling	Tropical Highland Primary	1,350	0.65	80	133	3,754.99	1,886	411	4885	243*	969	NA	NA
Yawan	YSF12	Felling	Tropical Highland Secondary	1,875	0.39	91	251	3,333.99	1,524	44	554	1766*	870	NA	NA
Yawan	YSF34	Felling	Tropical Highland Secondary	2,175	0.37	92	251	3,333.11	1,071	36	630	652*	692	605	70
Yawan	YSG12	Felling	Tropical Highland Secondary	1,500	0.20	86	209	3,820.00	1,060	30	353	352*	418	NA	NA
Yawan	YSG34	Felling	Tropical Highland Secondary	1,725	0.18	85	234	4,701.39	2,619	125	1044	684	675	557	76
Yawan	YSH12	Felling	Tropical Highland Secondary	450	0.22	84	60	2,205.64	399	60	711	127*	167**	NA	NA
Yawan	YSH34	Felling	Tropical Highland Secondary	675	0.16	81	71	3,805.55	403	317	2246	119*	103**	NA	NA
Yawan	YSJ12	Felling	Tropical Highland Secondary	750	0.22	86	81	3,403.85	783	134	1737	207*	456**	NA	NA
Yawan	YSJ34	Felling	Tropical Highland Secondary	525	0.23	81	83	3,331.01	693	183	1872	273*	901	NA	NA
Yawan	YSK12	Felling	Tropical Highland Secondary	600	0.41	80	73	2,741.99	452	76	572	82*	542	NA	NA
Yawan	YSK34	Felling	Tropical Highland Secondary	900	0.54	76	136	3,894.35	663	470	2270	297*	799	NA	NA
Wanang	WP1	Felling	Tropical Lowland Primary	1,837	0.70	79	123	3,486.88	397	214	3.264	NA	NA	NA	NA
Wanang	WP2	Felling	Tropical Lowland Primary	2,285	0.63	81	124	3,792.40	1,431	167	2.333	NA	NA	NA	NA
Wanang	WP3	Felling	Tropical Lowland Primary	2,444	0.48	80	135	4,715.49	1,260	177	1.923	NA	NA	NA	NA
Wanang	WP4	Felling	Tropical Lowland Primary	2,397	0.60	80	139	4,179.61	1,128	244	1.578	NA	NA	NA	NA
Wanang	WP5	Felling	Tropical Lowland Primary	2,284	0.49	82	118	4,649.52	1,593	109	3.497	NA	NA	NA	NA
Wanang	WP6	Felling	Tropical Lowland Primary	2,519	0.51	81	135	4,008.64	918	146	2.703	NA	NA	NA	NA
Wanang	WP7	Felling	Tropical Lowland Primary	2,282	0.46	82	154	4,397.18	717	209	7.892	NA	NA	4,164	279
Wanang	WP8	Felling	Tropical Lowland Primary	2,027	0.49	77	109	4,448.03	550	219	2.654	NA	NA	4,218	142
Wanang	WP9	Felling	Tropical Lowland Primary	2,014	0.75	80	115	2,386.43	698	174	6.960	NA	NA	3,491	251
Wanang	WP10	Felling	Tropical Lowland Primary	2,159	0.90	85	129	2,219.53	1,223	362	6.462	NA	NA	9,911	197
Wanang	WS1	Felling	Tropical Lowland Secondary	1,779	0.72	87	144	2,540.76	1,203	151	2.225	NA	NA	NA	NA
Wanang	WS2	Felling	Tropical Lowland Secondary	1,988	0.45	88	172	3,328.60	1,791	167	982	NA	NA	NA	NA
Wanang	WS3	Felling	Tropical Lowland Secondary	1,833	0.46	89	123	1,688.33	475	17	294	NA	NA	NA	NA
Wanang	WS4	Felling	Tropical Lowland Secondary	1,484	0.58	87	132	2,355.16	2,605	58	1,293	NA	NA	1249	131
Wanang	WS5	Felling	Tropical Lowland Secondary	785	0.51	89	45	1,767.06	2,432	163	1,565	NA	NA	1,615	186
Wanang	WS6	Felling	Tropical Lowland Secondary	1,369	0.45	83	118	4,034.45	2,486	273	1,930	NA	NA	NA	NA
Wanang	WS7	Felling	Tropical Lowland Secondary	907	0.57	81	45	2,999.24	1,191	166	1,655	NA	NA	NA	NA
Wanang	WS8	Felling	Tropical Lowland Secondary	1,801	0.90	82	108	1,891.99	791	325	1,067	NA	NA	NA	NA
Wanang	WS9	Felling	Tropical Lowland Secondary	1,699	0.97	88	101	1,886.50	2,458	270	2,282	NA	NA	1,188	132

Site	Plot	Method	Forest type	Area-based sampling effort (man hours)	Resource-Based sampling effort (man hours)	Canopy sampled (%)	Stems (DBH≥5cm)	Leaf area (m^2)	Caterpillars	Active mines	Abandoned mines	Galls	Spiders	Foraging ants	Ant nests
Wanang	WS10	Felling	Tropical Lowland Secondary	1,704	0.81	81	127	1,699.70	1,740	82	2,270	NA	NA	1,535	222
Fort Sherman	Panama 1	Crane	Tropical Lowland Primary	2,698	1.21	87	95	2,237.41	1,341	1689	6577	193,215	NA	NA	NA
Fort Sherman	Panama 2	Crane	Tropical Lowland Primary	2,110	1.17	79	86	1,808.31	275	327	4699	45,974	NA	NA	NA

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S3 Table. Monthly trends in abundance of caterpillars and active miners across the plots sampled for multiple months (Tomakomai, Lanzhot, Toms Brook, San Lorenzo, Wanang, Numba, Yawan). The table shows averages (± standard deviation) of caterpillar and active miner abundance per m^2 of foliage encountered at individual days of sampling within given months.

Caterpillars	Tomakomai	Lanžhot	Toms Brook	Wanang primary	Wanang secondary	San Lorenzo	Numba primary+secondary	Yawan primary+secondary
January				0.3±0.2	0.3±0.2	0.5±0.5		0.9±1.7
February				0.1±0.1	0.5±0.3	0.3±0.1		1.3±3.2
March				0.1±0.2	0.7±0.5	0.3±0.5	0.4±0.2	0.7±1.1
April			8.8±10.1	0.3±0.4	1.1±1.3	0.2±0.2	0.3±0.2	0.4±0.4
May	41.9±51.4	11.4±15.2	1.5±2.6	0.4±0.7	1.5±0.8	0.0±0.0	0.5±0.3	0.3±0.2
June	10.4±9.1	1.1±1.1	1.0±1.1	0.6±0.8	1.6±1.3	0.1±0.1	0.5±0.4	0.2±0.2
July	1.0±0.8	0.7±0.4	2.1±1.1	0.8±0.8	1.2±1.7	0.4±0.6	0.6±0.4	0.3±0.4
August	0.5±0.1	5.3±8.8	1.4±1.2	0.2±0.2	2.1±4.9	0.8±0.7	0.4±0.2	0.4±0.7
September				0.4±0.6	0.5±0.3	0.5±1.1		0.4±0.5
October				0.1±0.1	0.9±1.2	0.9±1.7	0.1±0.2	0.2±0.1
November				0.0±0.0	0.3±0.2	0.3±0.5	0.5±0.5	0.2±0.2
December					0.4±0.4	0.5±0.7		0.3±0.2
Miners	**Tomakomai**	**Lanžhot**	**Toms Brook**	**Wanang primary**	**Wanang secondary**	**San Lorenzo**	**Numba primary+secondary**	**Yawan primary+secondary**
January				0.00±0.00	0.01±0.01	0.53±1.68		0.03±0.04
February				0.01±0.01	0.04±0.03	0.74±0.98		0.05±0.04
March				0.05±0.05	0.09±0.12	0.58±1.05	0.03±0.05	0.10±0.16
April			0.03±0.06	0.05±0.06	0.03±0.03	0.07±0.14	0.02±0.02	0.04±0.06
May	0.77±2.9	0.11±0.25	0.09±0.15	0.14±0.34	0.13±0.10	0.13±0.14	0.02±0.04	0.14±0.19
June	0.33±0.46	0.33±0.41	0.32±0.27	0.09±0.12	0.11±0.15	0.06±0.15	0.03±0.05	0.02±0.02
July	0.32±0.26	0.08±0.09	0.92±1.05	0.05±0.10	0.09±0.10	0.21±0.44	0.04±0.05	0.02±0.03
August	0.93±0.98	0.09±0.19	0.86±0.74	0.06±0.06	0.05±0.07	0.05±0.11	0.00±0.00	0.07±0.09
September				0.02±0.03	0.03±0.07	1.39±5.58		0.04±0.04
October				0.04±0.06	0.04±0.07	0.63±1.93	0.01±0.03	0.09±0.09
November				0.05±0.00	0.11±0.08	0.15±0.20	0.00±0.00	0.05±0.06
December					0.06±0.08	0.08±0.21		0.01±0.01

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S4 Table. Variables with a significant effects on **Foliage accessibility**, **Area-based sampling effort,** and **Resource-based sampling effort** as selected by forward selection in linear mixed effect models. The best model explaining differences in **Foliage accessibility** included fixed effects of the forest type, used method, number of stems with DBH≥5cm, and sampled leaf area (m^2) (χ^2 (8) =64.02, p<0.0001). Percentage data on **Foliage accessibility** were arcsine transformed. The best model explaining differences in **Area-based sampling effort** included fixed effects of number of stems with DBH≥5cm, and forest type (χ^2 (5) =95.24, p<0.0001). The best model explain differences in **Resource-based sampling effort** included fixed effects of number of stems with DBH≥5cm, sampled leaf area, and forest type (χ^2 (6) =80.75, p<0.0001). Effort data were log-transformed. Site was used as random effect.

Foliage accessibility				
Fixed effects	**Estimate**	**Std. Error**	**t value**	**AIC**
Null model				-114.50
Selected model				-162.52
(Intercept)	0.9259	0.0290	31.94	
Forest type				
Tropical highland primary	0.0468	0.0318	1.47	
Tropical highland secondary	0.1167	0.0307	3.80	
Tropical lowland primary	0.0569	0.0323	1.76	
Tropical lowland secondary	0.1257	0.0284	4.44	
Method				
Felling	-0.0201	0.0307	-0.66	
Cherry-picker	0.2112	0.0438	4.82	
Number of stems with DBH≥5cm	0.0008	0.0002	4.59	
Sampled leaf area	0.00003	0.00001	-3.30	
Area-based sampling effort				
Fixed effects	**Estimate**	**Std. Error**	**t value**	**AIC**
Null model				56.13
Selected model				-29.11
(Intercept)	6.788	0.106	63.95	
Number of stems with DBH≥5cm	0.007	0.001	13.11	
Forest type				
Tropical highland primary	-0.294	0.165	-1.78	
Tropical highland secondary	-0.770	0.168	-4.58	
Tropical lowland primary	0.199	0.166	1.20	

Tropical lowland secondary	-0.106	0.171	-0.62	
Resource-based sampling effort				
Fixed effects	**Estimate**	**Std. Error**	**t value**	**AIC**
Null model				-70.96
Selected model				-139.71
(Intercept)	0.709	0.058	12.32	
Number of stems with DBH≥5cm	0.002	0.000	10.42	
Sampled leaf area	-0.000	0.000	-7.66	
Forest type				
Tropical highland primary	-0.268	0.097	-2.77	
Tropical highland secondary	-0.393	0.096	-4.07	
Tropical lowland primary	-0.058	0.096	-0.60	
Tropical lowland secondary	-0.108	0.096	-1.13	

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S5 Table. List of staff, interns, students, volunteers, and local assistants who helped with the sampling.

Site	First Name	Second Name	Role
Tomakomai			
Tomakomai	Haruka	Abe	Plant and Arthropod sampling
Tomakomai	Hiroaki	Fukushima	Plant and Arthropod sampling
Tomakomai	Tsutom	Hiura	Field management
Tomakomai	Utsugi	Jinbo	Lepidoptera identification
Tomakomai	Ryosuke	Kogo	Plant and Arthropod sampling; sample processing
Tomakomai	Rajesh	Kumar	Plant and Arthropod sampling, Lepidoptera identification
Tomakomai	Roll	Lilip	Plant and Arthropod sampling (in charge of miner sampling and processing)
Tomakomai	Jan	Macek	Insect identification
Tomakomai	Junichi	Yukawa	Gall identification
Lanžhot			
Lanžhot	Denisa	Bazsoová	Plant and Arthropod sampling; Insect rearing
Lanžhot	Ondřej	Dorňák	Plant and Arthropod sampling; Insect rearing
Lanžhot	Jiří	Hodeček	Plant and Arthropod sampling
Lanžhot	David	Kaspřák	Plant and Arthropod sampling
Lanžhot	Markéta	Kirstová	Plant and Arthropod sampling; Insect rearing
Lanžhot	Nela	Kotásková	Sampling, sample sorting
Lanžhot	Jan	Macek	Insect identification
Lanžhot	David	Musiolek	Plant and Arthropod sampling
Lanžhot	Hana	Platková	Plant and Arthropod sampling
Lanžhot	Veronika	Plocková	Plant and Arthropod sampling; Insect rearing
Lanžhot	Aneta	Sajdok	Plant and Arthropod sampling
Lanžhot	Mark	Shaw	Insect identification
Lanžhot	Stefan	Schmidt	Curator of parasitoid specimens
Lanžhot	Alžběta	Suchánková	Plant and Arthropod sampling
Lanžhot	Michal	Zapletal	Insect identification
Mikulčice			
Mikulcice	Jaroslav	Baloun	Plant and Arthropod sampling
Mikulcice	Lukáš	Čížek	Help with preparation of the sampling and management
Mikulcice	Jaroslav	Dlouhý	Plant and Arthropod sampling
Mikulcice	Nela	Kotásková	Plant and Arthropod sampling
Mikulcice	Kateřina	Kuřavová	Plant and Arthropod sampling
Mikulcice	Jan	Macek	Insect identification
Mikulcice	Ivan	Mikuláš	Arthropod sampling
Mikulcice	Ondřej	Šulák	Plant and Arthropod sampling
Mikulcice	Štěpán	Vodka	Plant and Arthropod sampling

Site	First Name	Second Name	Role
Mikulcice	Jan	Vrána	Arthropod sampling, in charge of ant sampling
Mikulcice	Michal	Zapletal	Insect identification
Mikulcice	Tomáš	Zítek	Plant and Arthropod sampling
Front Royal			
Front Royal	Thomas	Blair	Plant and Arthropod sampling
Front Royal	Grace	Carscallen	Plant and Arthropod sampling, in charge of ant sampling in 2016
Front Royal	Maria Eugenia	Losada	Plant and Arthropod sampling, in charge of leaf miner-gall in 2016
Front Royal	Inga	Freiberga	Gall sample processing and dissection
Front Royal	Aaron	Goodman	Sampling, in charge of spider sampling and identification in 2017
Front Royal	Geoffrey	Nichols	Plant and Arthropod sampling, in charge of leaf miner-gall in 2017
Front Royal	Margaret	Rosati	Logistic support
Front Royal	Matthias	Weiss	Gall sample processing and dissection
Front Royal	Kate	Aldrich	Plant and Arthropod sampling
Front Royal	Clayton	Hatcher	Plant and Arthropod sampling
Front Royal	Shelby	Abbott	Plant and Arthropod sampling
Front Royal	Meghan	Melberg	Plant and Arthropod sampling
Front Royal	Amanda	Gambale	Plant and Arthropod sampling
San Lorenzo			
San Lorenzo	John	Auga	Plant and Arthropod sampling
San Lorenzo	Stefan	Curtis	Plant and Arthropod sampling, leader of the climbing team
San Lorenzo	Ondřej	Dorňák	Plant and Arthropod sampling
San Lorenzo	Inga	Freiberga	Gall sample processing and dissection
San Lorenzo	Domminik	Rabl	Plant and Arthropod sampling
San Lorenzo	Mariam	Trejos	Plant and Arthropod sampling
San Lorenzo	Matthias	Weiss	Gall sample processing and dissection
San Lorenzo	Joachim	Yalang	Plant and Arthropod sampling
San Lorenzo	Inga	Freiberga	Gall sample processing and dissection
San Lorenzo	Matthias	Weiss	Gall sample processing and dissection
Wanang			
Wanang	Darren	Bito	Research Supervisor with a focus on parasitoids
Wanang	Erik	Brus	Arthropod sampling
Wanang	Kipiro	Damas	Data cleaning/Identifications
Wanang	Jan	Hrček	Research Supervisor with a focus on parasitoids
Wanang	Sentiko	Ibalim	Arthropod sampling
Wanang	Cliffson	Idigel	Arthropod sampling
Wanang	Bruce	Isua	Botany leader
Wanang	Robin	Kalwa	Plant sampling/vouchering
Wanang	Martin	Keltim	Arthropod sampling
Wanang	Andrew	Kinibel	Arthropod sampling
Wanang	Joseph	Kua	Arthropod sampling
Wanang	Roll	Lilip	Arthropod sampling
Wanang	Martin	Mogia	Arthropod sampling
Wanang	Kenneth	Molem	Botany leader
Wanang	Rebecca	Montgomery	Herbivory measures protocol
Wanang	Aloysius	Posman	Arthropod sampling
Wanang	Maling	Rimandai	Arthropod sampling

Site	First Name	Second Name	Role
Wanang	Steven	Sau	Arthropod sampling
Wanang	Gibson	Sosanika	Plant sampling/vouchering
Wanang	Elvis	Tamtiai	Arthropod sampling
Wanang	Tim	Whitfeld	Data cleaning/Identifications
Numba			
Numba	Kenneth	Benedict	Field lab work, Sample processing (botany)
Numba	Bradley	Gewa	Field lab work, Plant and Arthropod sampling, sample processing
Numba	Amelia	Hood	Field lab work, Sample processing
Numba	Frank	Jurgen	Field lab work
Numba	Graham	Kaina	Field lab work, Plant and Arthropod sampling, sample processing
Numba	Martin	Keltim	Plant and Arthropod sampling, sample processing
Numba	Andrew	Kinibel	Plant and Arthropod sampling, sample processing
Numba	Nancy	Labun	Sample processing (plants)
Numba	Roll	Lilip	Field lab work
Numba	Grace	Luke	Sample processing (arthropods)
Numba	Gibson	Maiah	Arthropod sampling
Numba	Gibson	Mayiah	Plant and Arthropod sampling, sample processing
Numba	Frank	Philip	Plant and Arthropod sampling, sample processing
Numba	Steven	sau	Field lab work
Yawan			
Yawan	Beneth	Ara	Plant and Arthropod sampling, sample processing
Yawan	Semcars	Ara	Plant and Arthropod sampling, sample processing
Yawan	Steven	Ganya	Plant and Arthropod sampling, sample processing
Yawan	Bradley	Gewa	Field lab work
Yawan	Hendry	Ginsongne	Plant and Arthropod sampling, sample processing
Yawan	Menos	Ginsongne	Plant and Arthropod sampling, sample processing
Yawan	Henson	Gomes	Plant and Arthropod sampling, sample processing
Yawan	Wotimo	Guboingnuc	Plant and Arthropod sampling, sample processing
Yawan	Bridget	Henning	Research supervisor; Sample collection
Yawan	Amelia	Hood	Sample processing
Yawan	Cliffson	Idigel	Field lab work
Yawan	Brus	Isua	Field lab work
Yawan	Tonsep	Joseph	Management of field assistants, field management
Yawan	Samuel	Joseph	Plant and Arthropod sampling, sample processing
Yawan	Martin	Keltim	Field lab work
Yawan	Barnabas	Kombe	Plant and Arthropod sampling, sample processing
Yawan	Joseph	Kua	Field lab work
Yawan	Oberth	Kui	Plant and Arthropod sampling, sample processing
Yawan	Roll	Lilip	Field lab work
Yawan	Bill	Lodi	Plant and Arthropod sampling, sample processing
Yawan	Max	Manaono	Plant and Arthropod sampling, sample processing
Yawan	Kenny	Mangirai	Plant and Arthropod sampling, sample processing
Yawan	Markus	Manumbor	Field lab work, Team leader
Yawan	Gibson	Mayiah	Field lab work
Yawan	Martin	Mogia	Field lab work
Yawan	Robert	Mongo	Plant and Arthropod sampling, sample processing

Site	First Name	Second Name	Role
Yawan	Jim	Nasing	Plant and Arthropod sampling, sample processing
Yawan	Aikson	Nea	Plant and Arthropod sampling, sample processing
Yawan	Namuce	Nongi	Plant and Arthropod sampling, sample processing
Yawan	Walindong	Nonong	Plant and Arthropod sampling, sample processing
Yawan	Frank	Philip	Plant and Arthropod sampling, sample processing
Yawan	Maling	Rimandai	Field lab work
Yawan	Sawaing	Sorong	Plant and Arthropod sampling, sample processing
Yawan	Wrefords	Sorong	Plant and Arthropod sampling, sample processing
Yawan	Gibson	Sosanika	Field lab work
Yawan	Elvis	Tamtiai	Field lab work
Yawan	Alu	Tonsep	Plant and Arthropod sampling, sample processing
Yawan	Sesilin	Tonsep	Plant and Arthropod sampling, sample processing
Yawan	Maxon	Tonseph	Plant and Arthropod sampling, sample processing
Yawan	Salape	Tulai	Field lab work
Yawan	Joseph	Valeba	Field lab work
Yawan	Mangan	Witwit	Plant and Arthropod sampling, sample processing

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PLOS ONE

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S1 Appendix. Sampling protocols

Quantitative assessment of arthropod-plant interactions in forest canopies: a plot-based approach

Content:

1.0 Setting up a 0.1 ha plot

We propose a **standardized protocol for sampling 0.1ha forest plots to quantify interaction networks of canopy arthropods**. The choice of forest area depends on the characteristics of the forest structure and composition meeting all suitable requirements for your project and research questions. Allocate the necessary time to explore and find a suitable forest site. In particular, you should base your decision on the presence of invasive species, topography, and access to the plot (important for the removal of felled trees or a for cherry-picker access). Before you start your project, always inform yourself on all safety instructions applicable to working in the field. These are not included in this protocol. Anyone conducting the sampling is responsible for obtaining the safety instructions elsewhere and following them.

1. Select a plot, which represents a 0.1 ha with a structure and a species composition typical for the local forests. Avoid forest edges, gaps, heavily disturbed areas, sloped terrain, and plantations.

2. Set up the corner points of the plot and take GPS coordinates for reference. Use a measuring tape or a laser range finder to measure the distance between points. Use a compass to measure the angles between the corner points in order to set up the plot in the desired shape. You can use a standard or electronic compass for this. Artillery compasses, specifically designed for taking azimuth angles, are usually a good option.

3. Mark the trees with DBH ≥ 5 cm with labels and identify them to species level (the identifications can be improved once the canopy is accessed). Mark only the trees which are rooted in the plot. If the border of the plot goes through tree trunk, include the tree in the plot only if more than 50% of the trunk mass at breast height is within the plot perimeter.

4. Record the position of all trees within the plot. First, select a "ZERO" point within the plot from which you can see all the trees. Clear the understory vegetation to improve the visibility if necessary. You can also use brightly coloured marks (or somebody in bright clothing standing next to the trees) to further increase the visibility of individual trees. Then record the azimuth angle (using a compass) and distance (using a measuring tape or a laser range finder) of individual trees from this point. These can be later easily transformed into x and y coordinates.

5. Optional. If visibility cannot be improved by removing some of the understory vegetation, divide the plot into a grid (Fig. P1). Measure the position of their corner points and all the trees in individual sub-plots as described above. If this method is not possible, you can also take GPS coordinates of individual trees. However, this can be rather inaccurate compared to the previous method depending on the precision of your GPS.

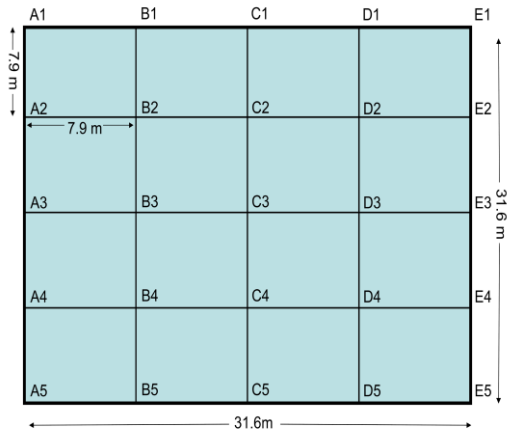

Figure P1. Example of a 0. 1ha plot divided into a grid with several reference points (A1-E5). Having such a grid improves accuracy of setting up the plot in densely vegetated sites.

2.0 Arthropod sampling

In temperate (and other seasonal) forests, sampling needs to be spread seasonally within each target tree species to capture the seasonal variability in associated arthropod communities. Create a sampling plan according to the phenology in the focal region (e.g. spread your sampling across both the spring and summer peak of arthropod abundance if such peaks are typical). Avoid sampling all conspecific trees in one part of the season if possible. Spreading sampling across the season may be problematic in the case of singleton tree species. Some methods, such as forest felling, provide limited flexibility for seasonal targeting of singleton tree species as trees cannot be resampled and the data thus represent a single time-point. On the other hand, sampling from cranes or cherry-pickers provides more flexibility. If there are any singleton tree species in your crane or cherry-picker plot, sample half of their canopy during the (spring) peak of arthropod abundance, while the second half can be sampled later in the season.

2.1 Arthropod sampling from felled trees

General notes

First, prepare a sampling plan to establish an ideal sequential order from which trees should be felled. Make sure individual tree species have a similar proportion of individuals sampled in different parts of the season. Clear the understorey. Start with felling small trees. Once enough small trees are gone and a sufficient space is opened, proceed with the larger trees. Always start with trees that are least likely to fall in a manner which may destroy other trees. This will minimize disturbance to the plot.

Trees should be felled one at a time. It is necessary to finish sampling on the same day as the tree was felled. All arthropods should be sampled as quickly as possible. This will prevent them from escaping or being predated.

Sampling should be done only during the day and when the leaves are not too wet. Avoid sampling in heavy rain, or directly after heavy rain (give the leaves some time to dry). Also avoid sampling during strong wind.

Divide sampling responsibilities within your team. If the size of your team allows, form sorting and sampling teams. Forming a sorting team, which will start pre-sorting samples in the field, will speed-up the final sorting in the lab; 2-3 team members are usually enough for pre-sorting.

There should be always skilled researchers and entomologists present in the field supervising the sampling and sample processing. Other team members should specialize primarily on a single arthropod group (leaf-chewing larvae, miners, or galls etc.) and be trained in the identification of their focal arthropod taxon prior to sampling. These specialized team members then can help other team members with assigning preliminary morphospecies and assist the skilled researcher with final morphotyping (see below).

Sampling steps

1. Select the tree to be felled according to your sampling plan. Measure its DBH (at 1.3 m).

2. Fell the tree

3. Measure its total height, trunk height, and canopy width. Trunk height is measured to the first major branch. Canopy width is measured at the widest point of the canopy. Record this into 'Plant Form'.

4. Record whether the leaves are mature or young (developing). In temperate forests, almost all leaves on a tree will be either mature or

Figure P2. Measuring a felled tree in Numba.

young at the time of sampling. In the tropics, this may not be the case so record mature and young leaves separately (see below in *Leaf area estimates*).

5. Sample the focal arthropod groups systematically by a manual search (see details on sampling of individual arthropod groups below). Hand the samples to the sorting team (if there is any) regularly during sampling. This is a much more efficient strategy than passing the samples all at once after the sampling is finished.

Figure P3. Sampling arthropods from a small felled tree in Toms Brook.

6. After the sampling, estimate what percentage of the foliage was sampled for arthropods (since part of the canopy usually gets destroyed during felling and you cannot sample herbivores from it). Record in 'Plant Form'. Estimates should be done by two trained persons independently and the mean estimated value should be used. This provides more accurate results.

These are the following arthropod groups to be sampled:

Leaf-chewing insect larvae

Search for all free-living and semi-concealed larvae. Check all rolled, tied, or folded leaves. Sample each larva in a separate rearing container. Gregarious larvae can be placed into a single large container, record their quantity. Containers should be available in various sizes suitable for larvae of different sizes. Provide a reasonable amount of leaves based on the size of the larva. The leaves should be of the same age the larva was sampled from (i.e. mature or young). Provide the larva with both young and mature leaves if you are not sure what leaves the larva was feeding on. Do not overfill the container with leaf material and keep it in the shade.

Miners

Sample all active and record all abandoned mines. When sampling mines try to assign them to preliminary morphospecies based on their shape, size, and position on the leaf. Mainly, separate blotch and serpentine mines. Keep your preliminary morphospecies in separate bags. Your preliminary morphotyping will be later corrected by an expert during final processing, but doing preliminary morphotyping and keeping your preliminary morphospecies in separate bags will speed up the final sorting.

Active mines

- Do not sample just the leaf with the mine. Mines will last longer if the leaf is attached to a twig with a couple of other leaves (but make sure that no other mine morphospecies are on the same leaves).
- Put all active mines from one morphospecies in one bag (they will be separated later). If you are not sure whether the mine is active or abandoned, sample it (it can be checked in detail later) and put it among other active mines from the respective morphospecies. Do not overfill the bag with leaf material and keep it in a shade.
- Sample up to ca 100 active mines per morphospecies only (50 will be used for rearing, 10 will be put in ethanol, and the rest will serve as a reserve in case some mines you sampled are inactive).
- The mines exceeding 100 can be simply counted (or their abundance can be estimated if there are many of them; see below). Record the number exceeding 100 into your notebook and report it to the sorting team after sampling. Always confirm with the expert assigning mines to final morphospecies that these mines are truly from a single morphospecies before you stop sampling them.

Abandoned mines

- Usually, you do not have to sample all abandoned mines. Just count their number or estimate their abundance visually in the event where there are too many of them (see below; but always confirm with the expert assigning mines to final morphospecies that these mines are truly from a single morphospecies). Record their number into your notebook and report it to the sorting team after finishing the sampling.
- Sample abandoned mines only if you do not have any active mine of that morphospecies available or assigning to clear morphospecies is problematic.

Gallers

- Sample all galls on all above-ground plant parts. When sampling galls, try to assign them to preliminary morphospecies. Mainly, focus on the plant part galled and shape of the gall. Your preliminary morphotyping will be later corrected by the expert doing the final processing, but doing preliminary morphotyping and keeping your preliminary morphospecies in separate bags will speed up the final sorting. It can be hard to distinguish arthropod and fungal galls. If unsure, sample all galls. Fungal galls can be identified in the laboratory and later removed from the analysis.
- Sample galled plant parts by detaching from the tree. If the galls are to be reared, and are in low numbers, galls will last longer if the plant part is attached to a twig with a couple of leaves. Otherwise, sample only the galled plant parts, preferably with active (inhabited) galls.
- Put different morphospecies in separate collecting bags. Do not overfill the bags and keep them in the shade.
- Sample enough galled material for each morphospecies to provide healthy quantities for rearing and dissection. What is considered a "healthy quantity" is dependent on the available resources (space, manpower, etc.) for rearing and dissecting, and the size of the galls. The more material reared and dissected, the better the chances of yielding insightful information to aid the species concept. Therefore, it would be ideal to rear at least 10 galled parts and retain at least 10 galls for dissection, per morphotype.
- Unsampled galls can be counted (or their abundance estimated if there are many of them; but always confirm with the expert assigning galls to final morphospecies that these galls are truly from a single morphospecies). Record the unsampled number into your notebook and report it to the sorting team after sampling.

Abundance estimates for very abundant mines and galls

Some abandoned leaf mines or gall morphospecies can be very abundant, which means counting them may take an excessive time investment. Instead of counting them individually, you can estimate their abundance in such cases. Mine and gall density can sometimes largely differ among various parts of the canopy. It is thus necessary to do the estimates repeatedly in various parts of the canopy.

- Select a reasonably large branch (ca 100-500 leaves) and count number of leaves and number of mines or galls on this branch. Divide their number by the number of leaves to calculate mine or gall per leaf average for this branch. Repeat this procedure at various parts of the canopy (at least three in the case of smaller trees and at least five in the case of larger trees). Use the averages to calculate a mean mine or gall density per individual leaf. Record this value. This can be used for estimating total mine or gall density once the total number of leaves is calculated.
- Some mite galls can be highly abundant (hundreds of galls per leaf). In such a case, pick only 20 leaves in random and calculate gall/leaf average. Repeat this procedure at various parts of the canopy (at least three in the case of smaller trees and at least five in the case of larger trees). Use the averages to calculate final mean gall density per individual leaf. Record this value. This can be used for estimating total gall density once the total number of leaves is calculated. Use this approach scarcely and only when really needed; e.g. in cases when more than 50% of leaves are galled.

- It is always better if the estimates are done by two specially trained persons using the mean estimate as a final value as it may provide more accurate results.

Spiders

Sample spiders into a vial with ethanol. All spiders from one tree can go into one vial but do not overfill it. Divide the spiders into more vials as needed to ensure a good proportion between ethanol and the sampled individuals. Similarly to ants (see below), sample spiders also from all lianas and epiphytes associated with the sampled tree.

Ants

Three people should be collecting ants (1 ant-trained staff member supervising 2 assistants) in tropical areas. In the temperate zone, where vegetation is less complex, two persons are enough. Sampling of foragers is done first immediately after felling. This helps to avoid contamination by ants invading the felled tree from the ground. After sampling for foragers is complete, collection continues with a search for individual nests.

Starting from the base of the tree (trunk) towards its crown, search carefully for any ants present on the fallen tree, especially those:

- foraging on the tree
- nesting on the leaves (silk or carton nest, weaved leaf nests etc.)
- living on and inside of the branches or twigs (Fig. P4)
- in the tree cavities
- under the bark
- under the lianas attached to the tree
- in the epiphytes on the tree, especially in the soil around their roots
- in any other suitable place where ants can occur

Figure P4. Ants often nest inside twigs and other host tree tissues. Do not forget to inspect even small twigs for ant nests. Use an axe or a chainsaw to cut open trunk and branches cavities for nests.

- We record several extra pieces of information for ants (such as their position on the tree, nest type etc.). This information should be recorded immediately after sampling, and recorded on both the labels and the 'Ant protocol' (see the example below). Do not wait till final processing to record this information.
- For all foragers, record their position on the tree – T (trunk below the branches) or C (crown – branches). All foraging ants (without a known nest) from one tree and similar height (T vs. C) can go together in one vial – this vial can contain a mix of different species If there is more than one vial with ants, mark each collection with a number: 1, 2, 3…
- For all nests, record their position (crown vs. trunk plus the vertical height above ground in meters), nest site type, and nest dimensions. Estimate the number of ant individuals in the nest. Record this information immediately after finding the nest. The examples of nest site types are listed below.
- Take vouchers of ant nests for photography (see *Sample processing and insect rearing*).
- Smaller colonies should be collected whole – including eggs, larvae and pupas and allates. Information as to whether the colony was collected as a whole is marked in the protocol and on labels.

- If the colony is too big (thousands of individuals), collect just part of it (20-50 individuals typically). Always try to sample all castes you can find as well as immature stages. Vials should be filled no more than halfway (1/2) with insects, the upper half should contain only ethanol to permit later molecular analysis (e.g. species barcoding). Use 2 ml vials for small samples. Use larger (e.g. 8 ml) vials for large bodied ants or larger colony samples.
- Ants from one colony (nest) should always be collected into one vial. They can be split in two, if there are too many ants for one vial – especially for big ants. In this case, each vial has to get its own label but with duplicated information. Don't mix ants from different colonies.
- Record if the host trees, or the ant-associated epiphytes, are myrmecophytes. Note if the plant contained ant domatia or nectaries (see an example of 'Ant protocol' below). Assigning plants as myrmecophytes or non-myrmecophytes can be difficult in tropical regions with poorly known flora and ant associations. Therefore, it is always crucial to record all the additional information as described above. The information on the location of the nest in dead or living tissue and trunk or branches can be especially helpful.

Nest types (write on the back side of your labels):
i) under the bark, ii) in hollow trunk, iii) in hollow live branch (= branch more than 5cm in diameter), iv) in hollow live twig (= branch less than 5cm in diameter, v) in hollow dead/dry branch, vi) in hollow dead/dry twig, vii) in hollow liana, vii) in/under epiphyte roots (or aerial soil), viii) inside of myrmecophytic epiphyte, ix) under liana, x) silk/carton nest on leaf, xi) carton nest on trunk/branch, xii) no nest (used for foraging individuals).

2.2 Arthropod sampling from cranes and cherry-pickers

General notes

First prepare a sampling plan, outlining the order in which the trees should be sampled. The primary aim here should be to account for seasonality. If the herbivore composition changes with the seasonal, ensure that you distribute sampling of conspecific tree individuals across the season. Avoid sampling all conspecific tree individuals in one part of season. If there are singleton tree species in your plot, sample 50% of their canopy in early season and the other 50% in later season.

Figure P5. Canopy sampling and ground sample sorting in Tomakomai.

Sampling should be done only during the day and when the leaves are not very wet. Avoid sampling in heavy rain, or directly after heavy rain (give the leaves some time to dry). Also avoid sampling during windy weather.

Divide sampling responsibilities within your team. If the size of your team allows, form sorting and sampling teams. Forming a sorting team, which will start pre-sorting samples in the field, will speed-up the final sorting in the lab; 2-3 team members are usually enough for the pre-sorting. Ideally, there should be a skilled researcher present in both teams.

Sampling steps

1. Follow your sampling plan to select the tree to be sampled.

2. Measure the tree. First, measure the DBH (at 1.3 m). Then measure total height, trunk height, canopy width using a laser range finder. Trunk height is measured to the first major branch. Canopy width is measured at the widest point of the canopy. Record these values in 'Plant Form'.

277 **3.** Record whether the leaves are mature or young (developing). In temperate forest, almost all leaves on a tree will
278 be either mature or young at the time of sampling. In the tropics, this may not be the case so record mature and young
279 leaves separately (see *Leaf area estimates* for more details).

280 **4.** Sample the focal arthropod groups. First, use a beating net to obtain free living arthropods. Second, do a manual
281 search to obtain remaining caterpillars, ants and spiders and also herbivores concealed in rolled or tied leaves, galls
282 and mines. Hand the insect samples to the ground team during the sampling regularly. This is much more efficient
283 strategy than passing the samples all at once after finishing sampling.

284 **5.** After the sampling, estimate what percentage of the foliage was sampled for arthropods. Record it into the 'Plant
285 Form'. This should be done by the canopy team. Estimates should be done by two trained persons independently and
286 the mean estimated value should be used. This provides more accurate results.

287 **6.** Record the number of leaves inspected for arthropods (see the instructions below in *Leaf area estimates*). Canopy
288 team should report this value to the ground team immediately after sampling.

289

Sampling low accessibility parts of the canopy

Some parts of the canopy (usually understory trees or lower branches of large trees) can be inaccessible from cranes or cherry-pickers. In such cases, you can use sampling from the ground, from ladders, or by climbing. If climbing is necessary, it usually requires forming a specialized climbing team consisting of 1-2 specially trained team members.

- Trees with height of 2-3 m can usually be sampled directly from the ground. Be careful not to break any branches or the trunk. Rather than bending such a tree by a brutal force, use a ladder.
- We used "A" shaped step ladders for sampling up to 3-5 m above ground (depending on the type, its stability, and terrain). In the case of large trees with sufficient trunk diameter, extension ladders fixed to the trunk can be also used for reaching similar heights. Always make sure the ladder is stable. During our sampling, the person on the ladder was always assisted by at least one person on the ground. We avoided using this type of ladder on sloped terrain.
- For sampling at greater heights or on sloped terrain, modular ladder poles are more efficient and stable. We used ladder poles for sampling at up to 8 m above ground. But note that this may differ depending on the type you use and its maximum load. The ladder poles should be ideally equipped with a steel fork at the basis that ensures good stability of the pole in the ground. We secured the ladder pole to the trunk of the tree with harnesses to prevent it from slipping. The person on the ladder was always assisted by at least one person on the ground.
- Trees even higher above ground, which are inaccessible from cranes or cherry-pickers, can be sampled by climbing. Descending from the gondola can ensure that even the terminal branches can be reached. But this method is usually time consuming. Also, it can only be carried out by a skilled person with proper training.
- Untrained or inexperienced team members should never sample from ladders or climb the trees.
- Always read and carefully follow safety instructions which may apply to working in the field, to working at heights, to working from ladders, or to climbing. This protocol cannot be used as a source of such information. You must obtain all the safety regulations from elsewhere and follow them.

290

These are the following arthropod groups to be sampled:

Leaf-chewing insect larvae

Collect all leaf-chewing larvae from the beating net. Then search for all free-living and semi-concealed larvae. Check all rolled, tied, or folded leaves. Sample each larva in a separate rearing container. Gregarious larvae can be sampled into a single large container, record their quantity. Containers should be available in various sizes suitable for larvae of different sizes. Provide a reasonable amount of leaves based on the size of the larva. The leaves provided should be of the same age as those the larva was sampled from (i.e. mature or young). Provide the larva with both young and mature leaves if you are not sure what leaves the larva was feeding on. Do not overfill the container with leaf material and keep it in a shade.

Miners

Sample all active and record all abandoned mines. When sampling mines try to assign them to preliminary morphospecies based on their shape, size, and position on the leaf. Specifically, separate blotch and serpentine mines. Keep your preliminary morphospecies in separate bags. Your preliminary morphotyping will be later corrected by an expert during final processing, but doing preliminary morphotyping and keeping your preliminary morphospecies in separate bags will speed up the final sorting.

Active mines:

- Do not sample just the leaf with the mine. Mines will last longer if the leaf is attached to a twig with a couple of other leaves (but make sure that no other mine morphospecies are on the same leaves).
- Put all active mines from one morphospecies in one bag (they will be separated later). If you are not sure whether the mine is active or abandoned, sample it (it can be checked in detail later) and put it among other active mines from the respective morphospecies. Do not overfill the bags with leaf material and keep them in a shade.
- Sample up to ca 100 of active mines per morphospecies only (50 will be used for rearing, 10 will be put in ethanol, and the rest will serve as a reserve in case some mines you had sampled are inactive).
- The mines exceeding 100 can be simply counted (or their abundance can be estimated if there are many of them). Record the number exceeding 100 into your notebook and report it to the sorting team after sampling. Always confirm with the expert assigning mines to final morphospecies that these mines are truly from a single morphospecies before you stop sampling them.

Abandoned mines:

- Usually, you do not have to sample all abandoned mines. Just count their number or estimate their abundance visually in the event where there are too many of them (see below; but always confirm with the expert assigning mines to final morphospecies that these mines are truly from a single morphospecies). Record their number into your notebook and report it to the sorting team after finishing the sampling.
- Sample abandoned mines only if you do not have any active mine of that morphospecies available or assigning to clear morphospecies is problematic.

Gallers

- Sample all galls on all above-ground plant parts. When sampling galls, try to assign them to preliminary morphospecies. Specifically, focus on the plant part galled and shape of the gall. Your preliminary morphotyping will be later corrected by an expert during the final processing, but doing preliminary morphotyping and keeping your preliminary morphospecies in separate bags will speed up the final sorting. It can be hard to distinguish

arthropod and fungal galls. If unsure, sample all galls. Fungal galls can be identified in the laboratory and later removed from the analysis.

- Sample galled plant parts by detaching from the tree. If the galls are to be reared, and are in low numbers, galls will last longer if the plant part is attached to a twig with a couple of leaves. Otherwise, sample only the galled plant parts, preferably with active (inhabited) galls.
- Put different morphospecies in separate collecting bags. Do not overfill the bags and keep them in the shade.
- Sample enough galled material for each morphospecies to provide healthy quantities for rearing and dissection. What is considered a "healthy quantity" is dependent on the available resources (space, manpower, etc.) for rearing and dissecting, and the size of the galls. The more material reared and dissected, the better the chances of yielding insightful information to aid the species concept. Therefore, it would be ideal to rear at least 10 galled parts and retain at least 10 galls for dissection, per morphotype.
- Unsampled galls can be counted (or their abundance estimated if there are many of them; but always confirm with the expert assigning galls to final morphospecies that these galls are truly from a single morphospecies). Record the unsampled number into your notebook and report it to the sorting team after sampling.

Abundance estimates for very abundant mines and galls

Some abandoned leaf mines or gall morphospecies can be very abundant, which means counting them may take an excessive time investment. Instead of counting them individually, you can estimate their abundance in such cases. Mine and gall density can sometimes largely differ among various parts of the canopy. It is thus necessary to do the estimates repeatedly in various parts of the canopy.

- Select a reasonably large branch (ca 100-500 leaves) and count number of leaves and number of mines or galls on this branch. Divide their number by the number of leaves to calculate mine or gall per leaf average for this branch. Repeat this procedure at various parts of the canopy (at least three in the case of smaller trees and at least five in the case of larger trees). Use the averages to calculate a mean mine or gall density per individual leaf. Record this value. This can be used for estimating total mine or gall density once the total number of leaves is calculated.
- Some mite galls can be highly abundant (hundreds of galls per leaf). In such a case, pick only 20 leaves in random and calculate gall/leaf average. Repeat this procedure at various parts of the canopy (at least three in the case of smaller trees and at least five in the case of larger trees). Use the averages to calculate final mean gall density per individual leaf. Record this value. This can be used for estimating total gall density once the total number of leaves is calculated. Use this approach scarcely and only when really needed; e.g. in cases when more than 50% of leaves are galled.
- It is always better if the estimates are done by two specially trained persons using the mean estimate as a final value as it may provide more accurate results.

Spiders

Sample spiders into a vial with ethanol. All spiders from one tree can go into one vial but do not overfill it. Divide the spiders into more vials as needed to ensure a good proportion between ethanol and the sampled individuals.

Ants

Sample ants foraging on the foliage and canopy branches into a vial with ethanol. All foraging ants from one tree can go into one vial but do not overfill it. Divide the ants into more vials in such a case to ensure a good proportion between ethanol and the sample. Note that while the sampling from a crane or a cherry picker allows to do a rapid assessment of ant foragers in the canopy, it is not comparable to the ant census using felling. In the case of felling, both whole

trunk and canopy, as well as individual nests outside and inside the host tree tissue and the associated epiphytes and lianas can be sampled, measured, and distinguished from foragers (see 2.1).

3.0 Leaf area estimates and plant vouchers

Sample leaves for leaf area estimates as specified below. We estimate leaf area of mature and young leaves separately as they can harbour different herbivores. We define mature leaves as fully developed in terms of their size and thickness. Young leaves are still developing. We define young leaves as leaves which haven't reached their full size or are much softer than mature leaves. Usually, they are also more lightly coloured than mature leaves.

In addition to the leaf area estimates, use this step to obtain herbarium vouchers, which will help with confirming host-plant identification, or to measure herbivory damage. Follow standard protocols for sampling plant vouchers (e.g. Funk *et al.* 2017). Sampling plant vouchers is especially useful in areas with high tree diversity. To avoid wilting, sample vouchers in plastic bags and mark them with tags. A voucher should include a stem bearing multiple leaves and an apical bud. Always sample flowers or fruits if present. Obtain at least three vouchers from around a canopy of each tree sampled. Press and dry the vouchers on the same day they were collected. The vouchers can be later used for DNA isolation and DNA barcoding to provide additional information on species identification.

Note: Although not discussed in this study, the sampled leaves can also be used for measuring leaf physical traits and nutrient content that can be relevant for structuring insect-plant interaction networks. Sampling leaves for measuring secondary metabolites usually requires special protocols and a separate sampling campaign. For example, the samples need to be cooled or frozen immediately after the sampling to avoid degradation and oxidation.

3.1 Leaf area estimates for felled trees

1. Sample foliage for biomass estimates (Fig. P6).

i) After you have sampled the tree for arthropods, place all foliage from the canopy into bags and weigh it. For large trees (ca DBH>30 cm), you can sample 25% or 50% of the foliage and extrapolate the results if your team is small in order to speed up the process. Record the weight into the 'Plant Form'. Sample and weigh mature and young leaves separately if both young and mature leaves are present. These values will be used for separate estimates of young and mature leaf area.

Figure P6. Leaf biomass sampling in Mikulcice.

ii) Avoid sampling leaves for biomass estimates when the foliage is wet and only sample leaves which have no other plants attached.

2. Sample leaves for calculating leaf area.

i) This includes obtaining individual leaves from across the canopy. A good method is to use the leaves sampled for the biomass estimate for this. Mix the leaves sampled for the biomass estimate in a bag and randomly pick some of them for calculating leaf area. Only use leaves which were not mechanically damaged during the sampling (but include those damaged by herbivores, pathogens, etc.).

ii) For small trees (ca. DBH < 15 cm), pick enough leaves (depending on their size) to fill a 50x50 cm white frame (Fig. P7). For larger trees or trees with large leaves, pick enough leaves to fill two frames (this is to cover the variability in

leaf sizes and shapes across the canopy of such trees). Sample young and mature leaves separately if there are both mature and young leaves present.

3. Take a photo of the leaves for the leaf area estimate.

i) Place the leaves for calculating leaf area into a 50x50 cm white frame (Fig. P7). Use as many leaves as possible but make sure they do not overlap or cross the frame border line.

ii) Leaves should be flat. Use some dark heavy objects (e.g. stones or coins) to flatten the leaves if necessary (but do not cover herbivory damage).

iii) Place a paper label with the tree number, the frame number (in case you take photos of more than one frame), and the leaf stage next to the frame so it is visible in the photo.

iv) Position the camera on a tripod right above the frame so that the frame appears on the camera display as a square.

v) Avoid strong light and shade contrasts during the photographing. Try to carry out this task with same camera settings to keep light levels consistent throughout the project.

vi) Once you take the photo, weigh the leaves. Record their total weight and their total number into the 'Plant Form'.

vii) If present, repeat this procedure for young and mature leaves separately.

viii) The resulting photos will be processed in ImageJ, Photoshop or other suitable software. In summary, the measurement is based on counting the number of pixels occupied by leaves vs. the number of pixels occupied by the background within a known area (here 2500 cm^2)). Missing leaf area or the area damaged by galls and mines can also be quantified using a similar approach in order to measure herbivory damage. Do not forget to correct for lens distortion, if needed. This can be especially important if you use a wide-angle lens. See existing protocols for details on leaf processing (e.g. Bito *et al.* 2011). The total sampled leaf area will be calculated using the total leaf biomass and the area to weight ratio from the photographed sample.

Figure P7. Preparing 50x50 cm leaf frames for photography and a final photograph of the frame. Note that the leaves are flattened by dark stones and there is a label with the tree number in the bottom corner of the frame.

3.2 Leaf area estimates for trees sampled from cranes and cherry-pickers

1. Estimate number of leaves on the tree.

i) Leaf number estimates must be done during the arthropod sampling.

ii) After you have sampled a part of the canopy for arthropods, select a reasonably large branch (with ca 500 leaves) within it and count how many leaves there are exactly (= value "A").

iii) Count how many branches of that size there are in the part of the canopy you have just sampled (= value "B"). Do this regularly. Avoid doing this across large parts of the canopy ("B" should be 5- 10, optimally).

iv) Multiply "A" with "B". Record this into your notebook as a local number of leaves ("C").

v) Repeat this procedure for each part of the canopy you sample**.**

vi) Once you finish sampling, count the sum of "C" values and report it to the ground team who will record it into 'Plant Form' as the total number of sampled leaves.

vii) Visually estimate what percentage of leaves is young and what percentage is mature if both young and mature leaves are present.

2. Sample leaves for calculating leaf area.

i) Drive the gondola all around the canopy and sample leaves in random and bring them to the ground.

ii) In the case of small trees (ca. DBH < 15 cm), pick enough leaves (depending on their size) to fill a 50x50 cm white frame (Fig. P2). In the case of larger trees or trees with large leaves, sample enough leaves to fill two frames (this is to cover variability in leaf sizes and shapes across canopy of such trees). Sample young and mature leaves separately if there are both mature and young leaves present.

3. Take a photo of the leaves for the leaf area estimate.

i) Place the leaves for calculating leaf area into a 50x50 cm white frame (Fig. P7). Use as many leaves as possible but make sure they do not overlap or cross the frame border line.

ii) Leaves should be flat. Use some dark heavy objects (e.g. stones) to flatten the leaves if necessary (but do not cover herbivory damage).

iii) Place a paper label with the tree number, the frame number (in case you take photos of more than one frame), and the leaf stage next to the frame so it would be visible on the photo.

iv) Position the camera on a tripod right above the frame so that the frame appears on the camera display as a square.

v) Avoid strong light and shade contrasts during the photographing. Try to carry out this task with same camera settings to keep light levels consistent throughout the project.

vi) Once you take the photo, weigh the leaves. Record their total weight and their total number into the 'Plant Form'.

vii) If present, repeat this procedure for young and mature leaves separately.

viii) The resulting photos will be processed in ImageJ, Photoshop or other suitable software. In summary, the measurement is based on counting the number of pixels occupied by leaves vs. the number of pixels occupied by the background within a known area (here 2500 cm^2)). Missing leaf area of the area damaged by galls and mines can be also quantified using a similar approach to measure herbivory damage. Do not forget to correct for lens distortion, if needed. This can be especially important if you use a wide-angle lens. See existing protocols for details on leaf processing (e.g. Bito *et al.* 2011). The total sampled leaf area will be calculated using the estimated total number of leaves on the tree multiplied by the mean leaf size of the photographed sample.

4.0 Sample processing and insect rearing

Figure P8. Sorting team in Tomakomai.

There can be a dedicated sorting team in the field (Fig. P8). Typically it may consist of 2-3 team members. If all team members are occupied by arthropod sampling, sample processing should be done immediately after returning from the field. The sorting team's main responsibilities are recording information into spread-sheets, sample sorting, labelling, and photographing of morphospecies and leaves.

The sorting team should include team members skilled and trained in morphotyping arthropods. The initial morphotyping is done *de novo* within each individual tree. The morphospecies will be cross-referenced across all individual trees once the sampling is finished. This reduces the amount of error compared to using a system of creating morphospecies across all trees within the plot or even multiple plots. Make sure that all arthropod individuals from a given group are always morphotyped by the same person when sorting arthropods from a single tree. Minimize the number of persons involved in the morphotyping. Give this task only to the team members with a proper training. This will increase the consistency in morphotyping and lower the amount of errors.

General notes

1. Record all information about the host-plant into the 'Plant Form'.
2. Label and sort all arthropod specimens. When taking arthropod vouchers, follow available standard protocols (e.g. Millar, Uys & Urban 2000; Schauff 2001).

Leaf-chewing insect larvae

- Morphotype leaf-chewing larvae based on their morphology (e.g. size, coloration, descriptions of hairs/ spines etc.). Record morphological characteristics of each morphospecies in your notebook. It will help you to morphotype further larvae.
- A maximum of up to 50 larvae per morphospecies should be kept for rearing. Each larva is to be kept separately in a rearing container with the exception of gregarious larvae. Keep gregarious larvae from one nest together in one large zip-lock bag or container. Record the number of gregarious larvae on the label in this event.
- If there are more than 50 larvae per given morphotype (this happens rarely):
  - i) Larvae 51-75 should be preserved in ethanol. Each larva should be kept in a separate vial and labelled with a standard label.
  - ii) Larvae 76-x can be discarded. Fill the number of discarded larvae into the 'Plant Form'.
- Label each kept larva (use only one label per nest of gregarious larvae). Record the following information on the label:
  - i) Unique Identifier (it can be pre-printed)
  - ii) Locality
  - iii) Tree ID number (unique number for each tree in the plot)
  - iv) Morphospecies
  - v) Body length (in mm)
  - vi) Feeding on the host (yes/no) – to be confirmed later in the laboratory
  - vii) Leaf age (record whether the larva was found on mature or young leaves)
  - viii) Mode of feeding (chewing, rolling, tying, skeletizing)
  - ix) Parasitized (yes/no) – to be filled in later based on the result of the rearing
  - x) Reared to adult (yes/no) – to be filled in later based on the result of the rearing
  - xi) Preserved in ethanol (yes/no)

- Photograph at least one larva per morphospecies. First, take a photo of the larva in detail. All important morphological characteristics (number of prolegs, setae, dorsal and lateral lines, head capsule etc.) should be visible. Take pictures from both the dorsal and lateral view (Fig. P9). Afterwards, take a photo of the same larva together with its label including all information.

Figure P9. Morphospecies photos of a caterpillar.

Mines

- Morphotype mines based on their morphology. Record morphological characteristics of each morphospecies in your notebook. (Specifically, record whether it is a blotch or a serpentine mine, on what side of the leaf is it visible, and colour of the frass if there is any). It will help you with morphotyping future mines.
- Separate inactive mines and count them. Add this number to the number of inactive mines of the respective morphospecies reported by the sampling team and record their number into the 'Plant Form'. If you have only abandoned mines for some morphospecies, keep a mine of that morphospecies for labelling and photographing.
- Up to 50 active mines per morphospecies should be reared in zip-lock bags.
- Up to 10 other mines of the same morphospecies should be dissected. If there are less than 60 active mines in total, dissect every second mine out of first ten mines and every fifth mine of the rest. Put the dissected larvae (or any other larger remains, e.g. head capsules) in a vial with ethanol and a standard miner label.
- If there are more than 60 active mines, discard them. Add the number of mines you discarded to the number of active mines counted (but not sampled) by the sampling team (Sampling team should report this number to you). Record this number in the 'Plant Form'.
- Mines will last longer if the leaf is attached to a branch with a couple of other leaves. Do not separate them if you plan to rear them.
- Each mine is to be reared in a separate zip-lock bag. However, if there are several miners per one leaf, do not separate them. You may keep them in one zip-lock bag but put a corresponding number of labels inside.
- Label each morphospecies or larva preserved in a vial. Record following information on the label:
  - i) Unique Identifier (it can be pre-printed)
  - ii) Locality
  - iii) Tree ID number (unique number for each tree in the plot)
  - iv) Morphospecies
  - v) Leaf age (record whether the mine was found on mature or young leaves)
  - vi) Active/abandoned
  - vii) Parasitized (yes/no) – to be filled in later based on the result of the rearing
  - viii) Reared to adult (yes/no) – to be filled in later based on the result of the rearing
  - ix) Preserved in ethanol (yes/no)

- Take a photo of one mine per morphospecies (Fig. P10). First, take a photo of the dorsal side in detail. Second, take a photo of the ventral side of the leaf in detail. Third, take a photo of the same mine together with its label with all information filled in and visible.

Figure P10. Morphospecies photos of a mine.

Galls

- Morphotype galls based on their morphology (mainly, record the type of the gall according to literature (e.g. Yukawa 1996; Redfern & Shirley 2002), on what side of the leaf is it visible, and its colour).
- Use the available literature and reference collections to identify fungal galls. Dissecting and examining under a microscope can be necessary for identification of fungal galls. Once you are absolutely sure about the identification, remove the fungal galls from further processing. However, if still unsure, process all galls with uncertain status. Make sure you take vouchers of such galls for further identifications by specialists.
- If galls of a morphospecies are low in number (e.g. < 15), prioritise putting them in ethanol for dissection rather than rearing.
- Select plant parts with the best looking galls (i.e. fresh, mature, no exit holes) for each morphospecies and rear them in one or more large zip-lock bags. All rearings of one morphospecies can be given the same label. Do not rear mite galls.
- Select, preferably, 10-30 individual galls per morphospecies, remove excess plant tissue, and place in ethanol for future dissection. Don't forget to add a vial label.
- Record the following information for each gall morphospecies in a separate sheet:
  i) Locality
  ii) Tree ID number (unique number for each tree in the plot)
  iii) Date
  iv) Gall morphospecies code
  v) Morphospecies description or a diagram
  vi) Plant part which was galled
  vii) Number of plant parts galled and the average number of galls per plant part. (This can be made exact if all individual galls are counted). This should also include the number of galled parts left on the tree (the sampling team should tell you if there were any). Alternatively, record the average number of galls per plant part. The number of plant parts galled can be estimated as % cover of plant parts galled (this approach is used for very abundant galls, and where the total number of tree parts will be known).
  viii) Number of galled plant parts (or individual galls) used for rearing.
- Label each morphospecies or larva preserved in a vial. Record the following information on the label:
  i) Locality
  ii) Tree ID number (unique number for each tree in the plot)
  iii) Date
  iv) Gall morphospecies code

- Take a photo of one gall per morphospecies. First, take a photo of the dorsal side in detail. Second, take a photo of the ventral side in detail. Third, take a photo of the same gall together with its label with all information filled in and visible.

Figure P11. Morphospecies photos of galls.

Spiders

All spiders from one tree can go into one vial. Divide the spiders into more vials in the event of high spider abundance, this will ensure there is a good proportion of ethanol. Label each vial with a spider label including:

i) Locality
ii) Tree ID number (unique number for each tree in the plot)
iii) Date

Ants

When sampling from felled trees, the information on foraging ants should be directly recorded during the sampling by the person responsible (see above). In the case of sampling from cranes and cherry-pickers, the information can be recorded once the sampling of the respective tree is finished. All vials with foraging ants should be labelled with an ant label including:

Foraging ants:

i) Locality
ii) Tree ID number (unique number for each tree in the plot)
iii) Date
iv) Trunk/Canopy (record whether the ants were foraging on the trunk or in the canopy).
v) Vial number (in case there are multiple vials with foraging ants from the respective tree)

Ant nests:

Ant nests are sampled only when sampling from felled trees. We record several extra pieces of information for ant nests (such as position on the tree, nest type etc.). This information should be recorded by the responsible person directly during the sampling in 'Ant protocol' and ant labels. Once the sampling of the respective tree is finished, check whether the following information was recorded for all ant nests:

i) Locality
ii) Tree ID number (unique number for each tree in the plot)
iii) Date
iv) Position on the tree (vertical height in m from the ground)
v) Type (description of nest site, see above)
vi) Dimensions of a nest (width times height in cm, where possible to measure)
vii) Number of individuals in the nest (assessment using categorical scale of number of workers, see example of the ant protocol)
viii) Vial number (in case there are multiple nests collected from the respective tree, each nest should have its own vial)

After the tree is searched and all samples collected, make sure that all the vials have the proper information written on their labels, and that all information is also described in the ant protocol for each tree (and that both the protocol, and labels match). Make sure all vials are full of ethanol. Check that vials are well closed/not leaking!

Take a photo of each different nest type for the common ant species, or their association with plant/symbiont species (see below). It is not necessary to take photos of all nests, but all common cases should be documented at least 3 times. The photograph should include the nest label (tree number + vial number), the voucher itself, and a scaler in cm.

Optional additions to the ant protocol:

Although not discussed in this study, the protocol for sampling ant nests can be also used for sampling other arthropods. Apart from ants, this protocol can be used for sampling termites, and the ant/termite associated trophobionts and symbionts (aphids, scale insects, beetles, bugs etc.). If the ant protocol is extended in this way, the same procedure is followed. In this case, mark if the sample contains ants, termites, or symbionts in the protocol (see example of the protocol (*"Ant, Ter, Sym"* marks) and examples of the labels). A small sample of ant individuals (1-5 workers) should be always collected with the symbionts to confirm host associations.

Insect rearing

All sampled larval insect herbivores should be reared to adults or parasitoids. Always protect rearing containers and bags from direct sunlight. Appropriate temperature and humidity are key factors affecting the rearing success. Always keep your rearing containers clean. Check them frequently and remove any frass or other waste to prevent growth of fungi. When taking vouchers of the reared arthropods, follow standard protocols (e.g. Millar *et al.* 2000; Schauff 2001), unless otherwise specified (see below).

Leaf-chewers

- Leaf-chewers should be reared in either plastic containers or zip-lock bags for large nests of gregarious larvae (Fig. P12). Write the most important information (host tree individual, morphotype number) on the container. This will serve as a back-up source of the most important information if the label gets mouldy or eaten by the larva.
- Inspect the containers every day.
- Provide larvae with fresh leaves and clean the boxes if necessary. This is usually needed every second day at least.
- Put some tissue paper into the bags or containers to absorb condensed water if needed.
- Record whether the larva feeds on mature or young leaves (mark it in the label). Record the mode of feeding if it hasn't been recorded already.
- Once the larvae pupate, clean the container. Remove any remaining old leaves, unless the pupa is directly attached to them. If this occurs, remove as much of the leaf tissue as possible without damaging the pupa. This will reduce the risk of fungal infection. Put some paper tissue or toilet paper inside the containers. This can either be used to absorb extra moisture (if you rear the pupae in a humid environment) or can be moistened if you rear the pupae in an environment with low air humidity. Separate the pupated individuals from the active larvae and check the container every day.
- Record if the larva died or was reared to an adult or a parasitoid. If it died, mark whether it was preserved in a vial with ethanol or not.
- Kill and mount every reared Lepidoptera adult. Killing by freezing will assure the best quality of DNA for barcoding. Abundant species with a known identification can be just pinned. Store adults in a dryer overnight. Place them in storage boxes once they are dry.

- Store reared parasitoids in ethanol. Label them with all of the host information as well as a unique parasitoid code.
- Note that many temperate insect species overwinter as pupae and you won't be able to rear their larvae into adults within a single season. Plan your project accordingly.

Figure P12. Insect rearing in Tomakomai. Note the caterpillars being reared in plastic containers placed in the shelves. Gallers and miners are reared in plastic bags hanging on the wall. All larvae are checked regularly

Mines and galls

- Mines and galls are reared in plastic bags (Fig. P12). Inspect the bags every day.
- Put some paper tissue or toilet paper into the bags to absorb condensed water.
- Record if the larva died or was reared to an adult or a parasitoid. If it died, mark whether it was preserved in a vial with ethanol or not.
- Kill and immediately mount every reared Lepidoptera adult. Store adults in a dryer overnight. Place them in storage boxes once they are dry. Mining and galling Microlepidoptera may die relatively quickly after emerging. It is thus essential to check for emerging adults regularly, ideally twice a day.
- Once dead, Microlepidoptera adults dry quickly due to their small size and are hard to relax for mounting. Therefore, if they die spontaneously in the rearing bag or container they are very difficult to mount. Store such individuals dried and fixed in Eppendorf tubes (but try to avoid such a situation in general!).
- Importantly, mounting mining and galling Microlepidoptera adults requires training. Study and follow standard protocols on Microlepidoptera mounting (e.g. Landry & Landry 1994).
- Adult Hymenoptera, Diptera, and Coeloptera should be preserved in vials with ethanol.
- Store reared parasitoids in ethanol. Do not forget to add a label with all information on the original herbivore larva.
- Mines and galls which do not emerge in 30 days can usually be discarded in tropical areas. If you are working in temperate regions, inform yourself if there are any ovewintering species associated with your focal host plants. Such species should be kept over winter. In addition, dissect a representative number of mines and galls per morhospecies before discarding. If there are any macroscopic remains of the larvae (e.g. head capsulas), preserve them in a vial and ethanol with a standard label.

Rearing rare mine or gall morphospecies

In the case of rare morphospecies of galls and mines, which were sampled as a single leaf (without sufficient other plant parts attached) follow the rearing protocol by Ohshima (2005):

- Remove the basal part of the leaf and expose the central vein.
- Prepare 1% sucrose solution and dip a piece of clean wiping paper in it.
- Wrap the petiole and exposed part of the central vein with the wiping paper.
- Store the leaf in a plastic container
- Check the container twice a day.
- Replace the wiping paper regularly (usually in two day intervals).

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Example 'Plant Form' (felling)

TREE ID nr.: _____ SPECIES:_____

CUT DOWN DATE: ____ _____ **RECORDED BY: _____**

PLANT SIZE: DBH _____ HEIGHT _____

NUMBER OF PLANT VOUCHERS TAKEN:

TRUNK HEIGHT Meters
[from the ground to the first big branches]

CROWN HEIGHT Meters
[from the first big branches to the top]

CROWN WIDTH Meters
[across the branches]

LEAVES: MATURE/YOUNG

PERCENTAGE OF THE FOLIAGE SAMPLED:

MATURE LEAVES:

WEIGHT of the foliage KG

% of the foliage sampled for biomass estimate:

LEAVES FRAME WEIGHT GRAMS

LEAVES FRAME - leaf area:____ cm^2

NUMBER OF LEAVES IN FRAME:_ _____

Leaf area before herbivory ___ cm^2 Leaf area after herbivory ___ cm^2

No. of discs _____ diameter: _____

YOUNG LEAVES:

WEIGHT of the foliage KG

% of the foliage sampled for biomass estimate:

LEAVES FRAME WEIGHT GRAMS

LEAVES FRAME - leaf area:_____ cm^2

NUMBER OF LEAVES IN FRAME:_ _____

Leaf area before herbivory ___ cm^2 Leaf area after herbivory ___ cm^2

No. of discs _____ diameter: _____

NUMBER OF ABANDONED MINES (can be a real number or % of the foliage attacked):

CAT001	CAT002	CAT003	CAT004	CAT005	CAT006	CAT007	CAT008	CAT009	CAT010

NUMBER OF DISCARDED ACTIVE MINES (can be a real number or % of the foliage attacked):

CAT001	CAT002	CAT003	CAT004	CAT005	CAT006	CAT007	CAT008	CAT009	CAT010

NUMBER OF DISCARDED CATERPILLARS:

CAT001	CAT002	CAT003	CAT004	CAT005	CAT006	CAT007	CAT008	CAT009	CAT010

CAT011	CAT012	CAT013	CAT014	CAT015	CAT016	CAT017	CAT018	CAT019	CAT020

NOTE:

Example 'Plant Form' (canopy cranes and cherry-pickers)

TREE ID nr.:_____ SPECIES:_____

SAMPLING DATE: ____ _____ 2016 **RECORDED BY: _____**

PLANT SIZE: DBH _____ HEIGHT _____

NUMBER OF PLANT VOUCHERS TAKEN:

TRUNK HEIGHT Meters
[from the ground to the first big branches]

CROWN HEIGHT Meters
[from the first big branches to the top]

CROWN WIDTH Meters
[across the branches]

LEAVES: MATURE/YOUNG

PERCENTAGE OF THE FOLIAGE SAMPLED:

ESTIMATED NUMBER OF MATURE LEAVES:

MATURE LEAVES FRAME - leaf area:____ cm^2

NUMBER OF MATURE LEAVES IN FRAME:_ _____

Leaf area before herbivory ___cm^2 Leaf area after herbivory ___ cm^2

No. of discs _____ diameter: _____

ESTIMATED NUMBER OF YOUNG LEAVES:

YOUNG LEAVES FRAME - leaf area:____ cm^2

NUMBER OF YOUNG LEAVES IN FRAME:_ _____

Leaf area before herbivory ___cm^2 Leaf area after herbivory ___ cm^2

No. of discs _____ diameter: _____

NUMBER OF ABANDONED MINES:

CAT001	CAT002	CAT003	CAT004	CAT005	CAT006	CAT007	CAT008	CAT009	CAT010

NUMBER OF DISCARDED ACTIVE MINES:

CAT001	CAT002	CAT003	CAT004	CAT005	CAT006	CAT007	CAT008	CAT009	CAT010

NUMBER OF DISCARDED CATERPILLARS:

CAT001	CAT002	CAT003	CAT004	CAT005	CAT006	CAT007	CAT008	CAT009	CAT010

CAT011	CAT012	CAT013	CAT014	CAT015	CAT016	CAT017	CAT018	CAT019	CAT020

NOTE:

Example 'Ant Protocol'

Tree No:		No. of Vials:	
Date:	2018	Collector:	

Myrmecophyte: YES / NO (Domatia/Nectaries) **No.:** vial number. Ant - ants, Ter - termites, Sym - symbionts

T = collection on the trunk bellow branches, C = collection in the crown; N = nest, F = only foraging ants/termites

No		Nest characteristic and notes					Nest location
1	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm
2	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm
3	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm
4	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm
5	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm
6	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm
7	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm
8	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm
9	Ant-Ter Sym T - C N - F						Distance from the ground: ……, … m
		Is whole colony collected?	Estimation of workers number (for a nest)				Size of colony w x h
		YES / NO	<100	100-500	501-1000	>1000	…… x …… cm

Example 'Ant Labels' and how to fill them. For examples of herbivore labels, see Figures P9-P11.

Frontal side of the label:	Back side - hand notes examples:
PNG, Madang prov.Wannang vill.	
Tree: W S -- 1A -- 1000	Ant foraging on crown
T/C # 1 | 22 Apr 2018	
Height .... m Whole col. Y / N	
PNG, Madang prov.Wannang vill.	
Tree: W S -- 1A -- 1000	ant nest in soil under epihyte roots
T / C # 2 | 22 Apr 2018	
Height 12 m Whole col. Y / N	
PNG, Madang prov.Wannang vill.	
Tree: W S -- 1A -- 1000	termite carton nest on trunk
T / C # 3 | 22 Apr 2018	
Height 10 m Whole col. Y / N	
PNG, Madang prov.Wannang vill.	
Tree: W S -- 1A -- 1000	ant nest in live hollow twig
T/C # 4 | 2 Apr 2018	
Height 22 m Whole col Y / N	
PNG, Madang prov.Wannang vill.	
Tree: W S -- 1A -- 1000	symbionts of nest #4 inside live twig
T/C # 5 | 2 Apr 2018	
Height 22 m Whole col. Y / N	