

The reassembly of plant-pollinator interactions along tropical forest succession

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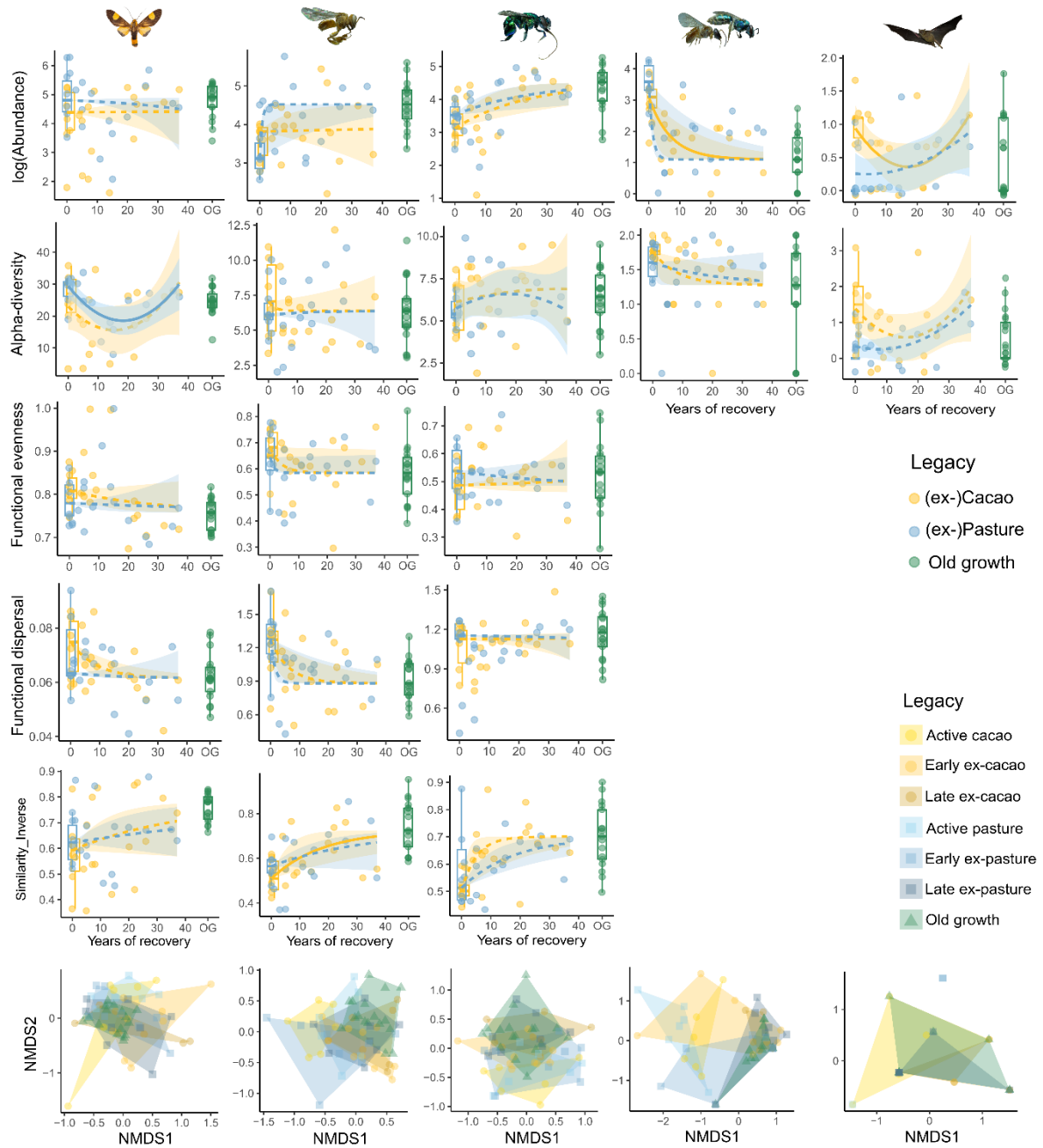
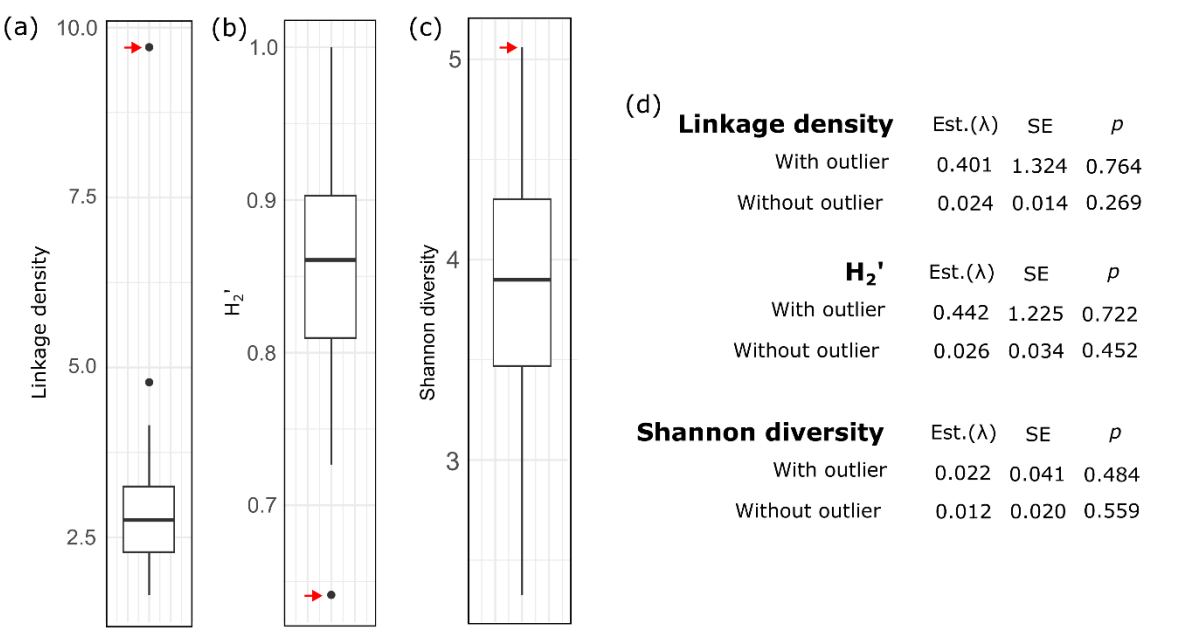


Figure S1. Taxon-based analysis of the effect of recovery time (years) on the community parameters of (from left to right column) moths (Arctiinae and Sphingidae), stingless bees (Meliponini), orchid bees (Euglossini), other bees (Halictidae, Megachilidae, Collectidae, and other Apidae), and bats (Chiroptera: Phyllostomidae). From the first to the fifth row, the analyzed parameters were: abundance (log-transformed), alpha-diversity (effective Shannon diversity, except for bats, where richness was used instead due to small sample sizes), functional evenness, functional dispersal, and community similarity to old growth. See Methods for details on the parameters. Other bees and bats could not have their functional diversity or community similarity to old growth because many plots had small sample sizes. The fitted models are shown, with dotted lines depicting non-significant recovery rates. Old-growth forests are always shown on the right as the reference state. Plots are divided by legacy (cacao plantations or pastures), and models are fitted independently for each legacy. In the last row, the non-metric multidimensional scaling of the communities for each group is shown, with plots separated by successional stage.



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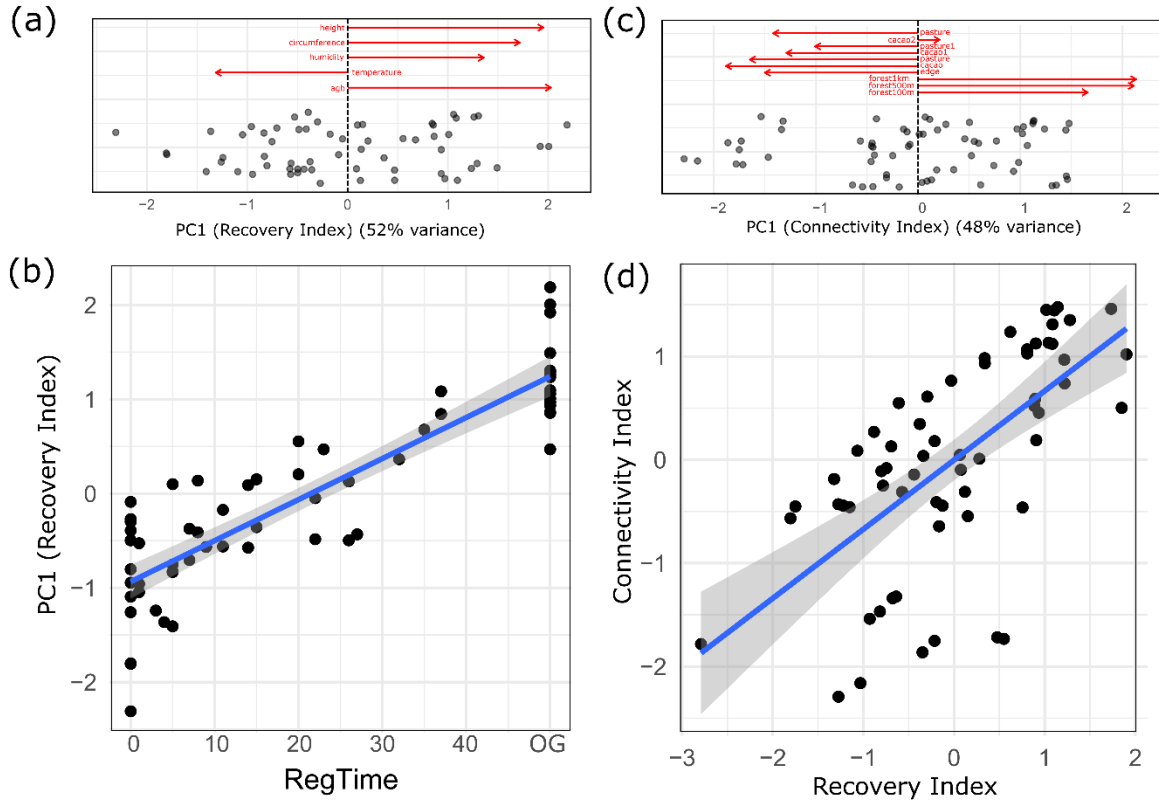
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Figure S2. Justification for the removal of a far-lying outlier plot from the network data prior to the analysis of the recovery rate. (a) linkage density, (b) specialization (H_2'), and (c) interaction Shannon diversity. The removed replicate is shown by a red arrow. In (d), the comparison of the model outputs with and without the outlier is shown in terms of the slope estimate for the constant recovery rate (λ), its standard error (SE), and the p -value. Only the results for the (ex-)cacao plantation legacy are shown, as results for the (ex-)pasture legacy were not affected.



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64 **Figure S3.** The PCA (principal component analysis) loadings of the variables used to derive the Recovery Index
65 (a) and the Connectivity Index (c). The distribution of the plots along the first component is shown. The red arrows
66 correspond to variable loadings. In (b), the relationship between the Recovery Index and plot recovery time in
67 years is shown, and in (d), the relationship between the connectivity Index and the Recovery Index is shown.
68 Variables: height = mean tree height, circumference = mean tree circumference, humidity = yearly average
69 humidity, temperature = yearly average temperature, agb = total aboveground biomass, pasture = proportion of
70 active pastures in a 1km buffer, pasture1 = proportion of early-regeneration pastures in a 1 km buffer, pasture2 =
71 proportion of late-regeneration pastures in a 1km buffer (the same descriptions apply for cacao, cacao1 and
72 cacao2), edge = distance to the nearest forest edge, forest100m = proportion of forest cover in a 100m buffer,
73 forest500m = proportion of forest cover in a 500m buffer, forest1km = proportion of forest in a 1km buffer.

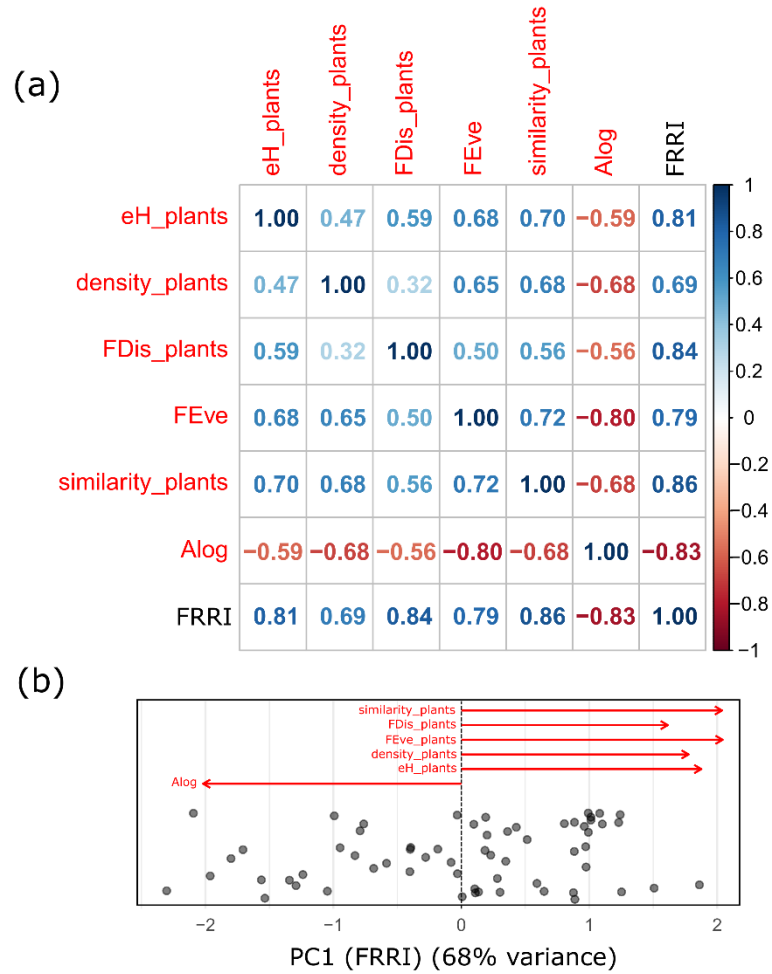


Figure S4. (a) Pearson's correlation matrix between the floral resource recovery index (FRRI) and the floral resource availability variables used to derive it. (b) Distribution of the FRRI (PC1 axis) values across the data points, and the loadings of each variable used to derive it. Variables: eH_plants = floral resource diversity (effective Shannon diversity), density_plants = resource density, FDis_plants = floral resource functional dispersal, FEve = floral resource functional evenness, similarity_plants = similarity of the community of flowering plants offering resources to that of old-growth forests, Alog = abundance (log-transformed).

Table S1. Full regression outputs of the Structural Equation Models used in the analysis. The standardized slope estimate (std. β) and its respective standard error (SE) are shown for each regression. Variables: FRRI = floral resource recovery index, RI = recovery index, Conn = connectance, Elev = elevation, A = pollinator abundance, eH = pollinator diversity (effective Shannon diversity), FDis = functional dispersal, simOG = community similarity to OG, H' = interaction Shannon diversity, H₂' = network specialization, simOG_net = network similarity to OG. Significant slopes ($p < 0.05$) are shown in bold.

	std. β (SE)					
	Full model	Moths	Stingless bees	Orchid bees	Nocturnal bees	Other bees
Floral resources						
FRRI~RI	0.65 (0.12)	0.63 (0.12)	0.62 (0.12)	0.63 (0.12)	0.64 (0.12)	0.66 (0.12)
~Conn	0.14 (0.10)	0.17 (0.10)	0.14 (0.10)	0.15 (0.10)	0.15 (0.10)	0.12 (0.10)
~Elev	-0.04 (0.08)	-0.05 (0.08)	-0.03 (0.08)	-0.04 (0.08)	-0.04 (0.08)	-0.003 (0.08)
Pollinators						
A~RI	-0.02 (0.17)	-0.08 (0.20)	0.01 (0.17)	0.27 (0.16)	0.41 (0.17)	0.09 (0.20)
~Conn	0.36 (0.12)	0.35 (0.14)	0.11 (0.11)	0.003 (0.11)	-0.03 (0.11)	-0.09 (0.13)
~Elev	0.01 (0.09)	0.04 (0.11)	0.25 (0.09)	-0.26 (0.09)	-0.06 (0.09)	0.00 (0.11)
~FRRI	0.03 (0.14)	-0.02 (0.17)	0.30 (0.13)	0.17 (0.14)	0.25 (0.14)	-0.70 (0.17)
eH~RI	0.16 (0.25)	0.04 (0.21)	0.11 (0.18)	0.28 (0.17)	0.53 (0.23)	0.10 (0.19)
~Conn	0.14 (0.17)	0.17 (0.15)	0.13 (0.12)	-0.11 (0.11)	-0.17 (0.15)	-0.31 (0.12)
~Elev	-0.03 (0.14)	0.26 (0.11)	-0.32 (0.09)	0.11 (0.09)	-0.24 (0.12)	0.07 (0.10)
~FRRI	-0.39 (0.22)	-0.25 (0.18)	-0.29 (0.14)	0.17 (0.14)	0.15 (0.19)	-0.48 (0.16)
FDis~RI	0.19 (0.21)	-0.11 (0.17)	-0.11 (0.17)	0.16 (0.17)	-	-0.32 (0.26)
~Conn	-0.15 (0.14)	0.09 (0.12)	-0.06 (0.11)	-0.01 (0.12)	-	0.26 (0.15)
~Elev	-0.11 (0.11)	0.13 (0.10)	-0.10 (0.09)	0.03 (0.09)	-	-0.25 (0.17)
~FRRI	0.19 (0.18)	-0.33 (0.15)	-0.24 (0.13)	0.20 (0.14)	-	-0.29 (0.20)
simOG~RI	0.39 (0.14)	0.30 (0.19)	0.22 (0.15)	0.22 (0.19)	-	-
~Conn	0.21 (0.10)	0.38 (0.14)	0.14 (0.10)	0.06 (0.13)	-	-
~Elev	-0.002 (0.08)	-0.04 (0.11)	-0.17 (0.08)	-0.03 (0.10)	-	-
~FRRI	0.33 (0.12)	-0.08 (0.16)	0.39 (0.12)	0.17 (0.16)	-	-
Interactions						
H'~A	0.30 (0.16)	0.27 (0.18)	0.24 (0.18)	0.66 (0.16)	0.52 (0.18)	0.53 (0.22)
~eH	0.38 (0.11)	0.51 (0.15)	0.34 (0.17)	-0.01 (0.16)	-0.02 (0.13)	-0.11 (0.22)
~FDis	-0.14 (0.12)	-0.07 (0.11)	-0.21 (0.18)	0.17 (0.15)	-	0.16 (0.13)
~FRRI	0.21 (0.12)	0.01 (0.12)	0.05 (0.15)	-0.03 (0.13)	-0.14 (0.14)	-0.11 (0.19)
H ₂ '~A	-0.17 (0.16)	0.27 (0.22)	0.20 (0.11)	-	-	-
~eH	-0.21 (0.11)	0.18 (0.17)	0.06 (0.11)	-	-	-
~FDis	-0.14 (0.12)	0.09 (0.13)	0.14 (0.14)	-	-	-
~FRRI	-0.13 (0.12)	0.01 (0.11)	-0.08 (0.10)	-	-	-
simOG_net~simOG	0.04 (0.15)	-0.22 (0.16)	0.07 (0.14)	0.23 (0.17)	-	-
~FRRI	0.21 (0.17)	0.30 (0.14)	0.25 (0.12)	0.06 (0.12)	-	-
~A	-0.04 (0.17)	0.06 (0.29)	0.19 (0.14)	0.36 (0.16)	-	-
~eH	0.22 (0.11)	0.27 (0.22)	0.19 (0.13)	0.09 (0.18)	-	-
~FDis	-0.15 (0.12)	-0.21 (0.16)	-0.05 (0.11)	-0.02 (0.15)	-	-

Table S2. Model quality statistics of the Structural Equation Models used in the analysis. DF: degrees of freedom, RCFI: Robust Comparative Fit Index, RGFI: Robust Goodness of Fit Index (followed by 90% confidence intervals).

	Full model (all groups)	Moths	Stingless bees	Orchid bees	Nocturnal bees	Other bees
Test User Model						
Test Statistic	12.61	15.22	17.71	12.70	0.59	6.86
DF	10	10	13	9	3	4
<i>p</i> -value (Chi-squared)	0.25	0.12	0.17	0.178	0.90	0.14
Test Baseline Model						
Test Statistic	301.94	353.22	257.92	227.62	172.28	257.80
DF	55	55	55	45	21	28
<i>p</i> -value (Chi-squared)	0.00	0.00	0.00	0.00	0.00	0.00
RCFI	0.94	0.99	0.99	0.98	1.00	0.96
RGFI	0.99 (0.96- 1.00)	0.99 (0.93- 1.00)	0.99 (0.95- 1.00)	0.99 (0.95- 1.00)	1.00 (0.99- 1.00)	0.96 (0.90- 0.99)

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Table S3. Interaction traits measured for each plant morphotype recorded in the chronosequence plots, followed by a description and observations when applicable (considering sources when necessary).

Variable	Description	Observation
Corolla width (mm)	The diameter of the perianth at its outermost position. In gamopetalous tubular flowers, it corresponded to the tube opening or the corolla diameter at petal/sepal separation. In dialipetalous flowers, it corresponded to the diameter of the perianth base.	Own manual measurements from fresh material collected in the field.
Corolla depth (mm)	The vertical length of the perianth. In tubular flowers, it corresponds to tube length. In non-tubular gamopetalous flowers (e.g., funnelform, campanulate, cyanthiform), it corresponds to the distance between perianth base and petal separation. Dialipetalous or incomplete flowers received a value of zero.	Own manual measurements from fresh material collected in the field.
Symmetry	The number of symmetry planes in a flower: actinomorphic or radial (several symmetry planes), zygomorphic or bilateral (one symmetry plane), asymmetric (no symmetry planes), and incomplete (no perianth).	Own classification based on fresh material collected from the field.
Morphology of the pollination unit	Categorical classification of the pollination unit's shape, which may comprise individual flowers or pseudanthia (densely aggregated flowers in an inflorescence forming a single flower head, e.g., capitula in Asteraceae, and spike inflorescences in Araceae). Categories were:	Classification based on and adapted from Durka (2002) and Heß (2019). Species and corresponding unit morphologies can be found in Table S2.

Brushform: diverse perianth shapes, but possessing long and numerous exert anthers (e.g., *Inga* spp., Fabaceae).

Campanulate: bell-shaped, wide gamopetalous and parallel perianth and opening wider than base (e.g., *Chelonanthus* spp., Campanulaceae);

Capitulate: flowers organized in dense capitula, with or without differentiation of ligulate flowers. Common in the family Asteraceae and Rubiaceae.

Coronate: crown-shaped flowers, usually formed by a layer of inner cup-shaped and fused projections of the tepals over a flat platform formed by the tepals.

Crateriform: shallow, bowl-shaped flowers, consisting of either dialipetalous or partially gamopetalous corollas.

Cruciform: cross-shaped flowers, usually dialipetalous and with four perpendicular petals. Common in cruciferous plants (e.g., Brassicaceae).

Cyanthiform: cup-shaped flowers with a wide perianth opening, often gamopetalous.

Funnelform: Trumpet-shaped gamopetalous flower, with a widening tube diameter towards the corolla flare (e.g., *Brugmansia*, *Allamanda*).

Labiate: zygomorphic gamopetalous corollas where petals are fused and differentiated into two or more lip-shaped structures.

Labellate: Inner perianth tepals differentiated into a labellum, common in Orchidaceae.

Rotate: dialipetalous, flat perianth. Disk-shaped.

Saccate: gamopetalous tubular flower, with the ventral surface of the corolla expanded in a sac-like structure (e.g., *Gasteranthus* spp., Gesneriaceae)

Spathate: flowers densely organized on a vertical spike, surrounded by a differentiated, chamber-shaped bract (spathe). Common in Araceae.

Spikeform: similar to spathate, but lacking the spathe (e.g., Piperaceae).

Tubulate: tubed flower, formed by a long gamopetalous or gamosepalous perianth (length approximately 3x or longer than diameter) with a relatively constant diameter.

Urceolate: urn-shaped gamopetalous flower, with tube opening narrower than the bulging midsection of the tube.

Main floral resource

The main resource produced by floral structures for attracting and rewarding pollinators.

Inferred from Gentry and Vasquez (1996), Jørgensen and León-Yáñez (1999) and Lozano et al. (2024) as well as databases such as the Plants of the World Online (<https://powo.science.kew.org/>) and the Global Biodiversity Information Facility (GBIF) (<https://www.gbif.org/>). Based on the classifications of (Proctor and Yeo 2009; Wilmer 2011; Rech *et al.* 2014).

Nectar	The most common floral resource for pollinators, consisting of a sugary solution (e.g., sucrose, hexose) and produced by floral glands (nectaries). Nectar is considered as the main floral reward unless one of the other specialized floral rewards is present and surpasses nectar production and/or when nectar production is absent, e.g., incomplete flowers without a perianth.	Extrafloral nectaries are not considered in this study. In the case of small incomplete flowers with no nectar production but where there is no other form of specialized floral resource, such as in Piperaceae (De Figueiredo and Sazima 2000), pollen was assumed as the main reward.
Pollen	Reward aimed at female bees, which require pollen for raising progeny. Pollen is often rewarded via feeding anthers, which differ from pollination anthers. Most pollen-rewarding flowers either lack nectar or produce very small quantities of it (Proctor and Yeo 2009)	Assumed for flowers with specialized pollen-rewarding systems (e.g., buzz-pollination in Melastomataceae and Solanaceae), or when nectar and other resources are lacking (e.g., Piperaceae).
Oil	Production of fatty acids by floral glands (elaiophores), characteristic of the family Malpighiaceae and targeted at female bees (Torretta <i>et al.</i> 2022).	
Resin	Rare floral resource produced by at least two plant families (Clusiaceae, genus <i>Clusia</i> ; Euphorbiaceae genus <i>Delachampia</i>). (Armbruster 1984).	In our study, this category is exclusive to the genus <i>Clusia</i> (Clusiaceae). Extrafloral resin production in other species is not considered.
Fleshy parts	Carbohydrate, lipid or protein-rich floral tissues usually scraped or bitten off by insect mouthparts, commonly by beetles. Normally produced at the base of the petal or at the tip of the stamens. Common in, e.g., Annonaceae.	
Scent	Produced at specialized cells (osmophores), these volatile oils are produced to attract insects that use them for reproductive functions (e.g., Euglossini bees or beetles).	
Deception	The absence of floral reward- The attraction of insects occurring via visual or olfactory cues that mimic similar species with resources, or females of certain insect species (leading to the copulation of males with the flower, common in wasp-pollinated Orchidaceae).	
Pollination syndrome	The main functional group of animals expected to be the plant's most effective pollinators, inferred via the set of floral traits and rewards offered by the plant.	Estimated via the set of floral traits presented by the plant, using the criteria of Faegri and Pijl (2016) and Proctor and Yeo (2009)
Anemophily	Wind pollination, marked by small inconspicuous flowers with no floral rewards and pollen adapted for wind dispersion. Common in pioneer grasses and forbs.	

Ambophily	Wind pollination, complemented by the attraction of insects through the offering of nectar or pollen (ambophily). Nectar guides normally present. Common to the families Poaceae and Cyperaceae (Abrahamczyk <i>et al.</i> 2023)
Cantharophily	Flowers showing traits associated with beetle pollination, including nocturnal or crepuscular anthesis, drab coloration, robust disk-shaped flowers or pollination-chamber inflorescences that produce strong scents at the beginning of the anthesis. Floral tissues frequently used as resources.
Chiropterophily	Flowers with traits associated with bat pollination, characterized by nocturnal anthesis, large brush-, cup-, or campanula-shaped flowers of drab colors that produce copious and diluted nectar and usually emit musky odors.
Melittophily	Flowers adapted to attracting diurnal bees by offering nectar, pollen, resin or oil as main rewards, by showing morphologies adapted to the bodies of bees (e.g., quill-shaped and labiate flowers in Papilionoideae, or feeding anthers in several families) or pollen-releasing mechanisms that require bees (e.g., poricidal anthers). Other traits include diurnal anthesis, usually blue, pink, or white flowers with nectar guides and/or high anther-corolla color contrast.
Ornithophily	Flowers with traits associated with bird (hummingbird) pollination, characterized by long floral tubes, diurnal anthesis, large quantities of diluted nectar, and usually bright red, yellow, lilac, white or blue corollas or bracts with no odor emission. Brushform or labiate flowers are also common.
Phalaenophily	Flowers with traits associated with settling moth pollination, showing nocturnal anthesis, white or drab coloration and small, long- and narrow-tubed or hipocrateriform flowers with relatively concentrated and hidden nectar and the emission of sweet scents. Often congested inflorescences that serve as a landing platform. May produce a sweet and strong scent.

Psychophily	Flowers with traits associated with butterfly pollination. Marked by diurnal anthesis, bright colors (yellow, red, orange, white, blue or purple), nectar production and delicate, tubular flowers, often brushform, hipocrateriform, and organized in inflorescences that serve as landing platforms (e.g., capitula).
Sphingophily	Flowers with traits associated with hawkmoth pollination. Similar to phalaenophily, but flowers are larger and longer, produce more copious and diluted nectar and often a very sweet, fermented odor.
Small diverse insects (SDI)	An agglutinative syndrome, defined by small, inconspicuous, and accessible flowers that usually produce small quantities of nectar and that do not show a specific pollination syndrome (Bawa <i>et al.</i> 1985; Moreira and Freitas 2020). Species are thus assumed to be pollinated by a variety of small bees, flies, ants, thrips, beetles, etc.

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98 Literature Cited

- 99 Abrahamczyk S, Struck J-H, Weigend M. 2023. The best of two worlds: ecology and evolution of
100 ambophilous plants. *Biological Reviews of the Cambridge Philosophical Society* 98: 391–420.
- 101 Armbruster WS. 1984. The Role of Resin in Angiosperm Pollination: Ecological and Chemical
102 Considerations. *American Journal of Botany* 71: 1149–1160.
- 103 Bawa KS, Perry DR, Beach JH. 1985. Reproductive Biology of Tropical Lowland Rain Forest Trees.
104 I. Sexual Systems and Incompatibility Mechanisms. *American Journal of Botany* 72: 331–345.
- 105 De Figueiredo RA, Sazima M. 2000. Pollination Biology of Piperaceae Species in Southeastern
106 Brazil. *Annals of Botany* 85: 455–460.
- 107 Durka W. 2002. Blüten- und Reproduktionsbiologie In: *Schriftenreihe für Vegetationskund.* Bonn:
108 Bundesamt für Naturschutz, 133–175.
- 109 Faegri K, Pijl L van der. 2016. *Principles of Pollination Ecology*. Oxford ; New York: Pergamon.
- 110 Gentry AH, Vasquez R. 1996. *A Field Guide to the Families and Genera of Woody Plants of*
111 *Northwest South America: (Colombia, Ecuador, Peru) : With Supplementary Notes on Herbaceous*
112 *Taxa*. Chicago: University of Chicago Press.
- 113 Heß D. 2019. *Die Blüte. Struktur. Funktion. Ökologie. Evolution*. Stuttgart: Eugen Ulmer KG.
- 114 Jørgensen PM, León-Yáñez S. 1999. *Catalogue of the vascular plants of Ecuador*. St. Louis: Syst.
115 Bot. Missouri Bot. Gard.
- 116 Lozano P, Roa L, Neill DA, Simpson N, Klitgaard BB. 2024. *Flora, ecología y fitogeografía de la*
117 *Reserva Canandé, Chocó ecuatorial*. Universidad Estatal Amazónica, Puyo, Ecuador, y Royal Botanic
118 Gardens, Kew, Reino Unido.

- 119 Moreira MM, Freitas L. 2020. Review of the Pollination System by Small Diverse Insects.
120 *Neotropical Entomology* 49: 472–481.
- 121 Proctor MCF, Yeo P F. 2009. *The Pollination of Flowers*. New York: HarperCollins.
- 122 Proctor M, Yeo P. 2009. *The Pollination of Flowers*. Collins.
- 123 Rech AR, Agostini K, Oliveira PE, Machado IC. 2014. *Biologia da Polinização*. Editora Projeto
124 Cultural.
- 125 Torretta JP, Aliscioni SS, Marrero HJ, Avalos AA. 2022. Oil flowers of Malpighiaceae and its oil-
126 collecting bees: loyalty and robbery in a highly specialized system. *Apidologie* 53: 30.
- 127 Wilmer P. 2011. *Pollination and Floral Ecology*. New Jersey: Princeton University Press.
- 128