



Instituto de Pesquisa Jardim Botânico do Rio de Janeiro
Escola Nacional de Botânica Tropical
Programa de Pós-Graduação em Botânica

Dissertação de Mestrado

Amofilia em escala global: características e contribuição biótica e abiótica na polinização

Danilo Alves de Carvalho

Rio de Janeiro

2021



Instituto de Pesquisa Jardim Botânico do Rio de Janeiro
Escola Nacional de Botânica Tropical
Programa de Pós-Graduação Stricto Sensu

Ambofilia em escala global: características e contribuição biótica e abiótica na polinização

Danilo Alves de Carvalho

Dissertação apresentada ao Programa de Pós-Graduação em Botânica da Escola Nacional de Botânica Tropical, do Jardim Botânico do Rio de Janeiro, como parte dos requisitos necessários para a obtenção do título de Mestre em Botânica.

Orientador: Leandro Freitas

Rio de Janeiro

2021

Ambofilia em escala global: características e contribuição biótica e abiótica na polinização

Danilo Alves de Carvalho

Dissertação submetida ao Programa de Pós-Graduação em Botânica da Escola Nacional de Botânica Tropical, Instituto de Pesquisas Jardim Botânico do Rio de Janeiro - JBRJ, como parte dos requisitos necessários para a obtenção do grau de Mestre.

Aprovada por:

Dr. Leandro Freitas (Orientador) _____

Dra. Ana Carolina Galindo da Costa _____

Prof. Dr. André Rodrigo Rech _____

Profa. Dra. Juliana Vilela Paulino _____

Em 17/09/2021

Rio de Janeiro

2021

C331m Carvalho, Danilo Alves de.

Ambofilia em escala global: características e contribuição biótica e abiótica na polinização/ Danilo Alves de Carvalho. – Rio de Janeiro, 2021.

x, 57f. : il. ; 28 cm.

Dissertação (mestrado) – Instituto de Pesquisas Jardim Botânico do Rio de Janeiro / Escola Nacional de Botânica Tropical, 2021.

Orientador: Leandro Freitas.

Bibliografia.

1. Polinização. 2. Espécies ambófilas. 3. Sucesso reprodutivo. 4. Polinização pelo vento. I. Título. II. Escola Nacional de Botânica Tropical.

CDD 582.0463

AGRADECIMENTOS

Gostaria de agradecer primeiramente ao meu orientador, Leandro Freitas, pela enorme contribuição e paciência nesses dois anos, por ter me ajudado a entender melhor sobre a história da Ciência e da Academia, pelos conselhos e críticas que me fizeram amadurecer tanto como profissional quanto cientista, pelos empréstimos de livros e pelos assopros depois dos socos.

Quero agradecer também à Dra. Marina Wolowski e ao Dr. Pedro Bergamo pela paciência, cuidado, parceria e ensinamentos fundamentais para eu desenvolver o manuscrito e minha dissertação num geral.

Quero agradecer também à Dr. Camila Souza e Dr. Andrea Sánchez-Tapia pela leitura crítica nas minhas qualificações e de antemão à banca avaliadora pela disponibilidade e pelas considerações ao meu trabalho.

Quero agradecer a Amparo Lázaro pela disponibilidade dos dados na meta-análise de *B. balearica* e mais uma vez ao Pedro Bergamo pelo intermédio.

Quero agradecer a todos os amigos de laboratório do Laboratório de Biologia Floral do JB pelas trocas e pela companhia nesses dois anos e meio e por terem feito um ambiente melhor pra desenvolver meu mestrado.

Quero agradecer à administração e pessoal responsável pelo PNI parte alta, ao pessoal do Herbário RB e ao Laboratório de Estrutura Vegetal do JBRJ pela disponibilidade e atenção comigo no projeto anterior a esse.

Quero agradecer à minha família, principalmente mãe e avós por termos conseguido passar por esse momento tão ímpar mais juntos que antes e pelo apoio contínuo no meu desenvolvimento profissional.

Quero agradecer aos meus amigos Alline, Anna, Laura, Jonata, Letícia, Luis Bernardo, Mariana, Nana, Priscila, Rafael, Rodrigo, Sávio, Virgínia, Thaísa e todos que me ajudaram e vibraram pelo meu sucesso.

Quero agradecer também ao CNPq pela bolsa concedida durante o mestrado, à CAPES e à FAPERJ pelo financiamento dos projetos do laboratório.

Por último, quero agradecer ao Danilo do passado por ter acreditado que o Danilo do futuro poderia ser o Danilo do presente.

RESUMO

A ambofilia, um sistema misto de polinização que combina a ação do vento e animais para a polinização, vem sendo negligenciada nos estudos de polinização. A distribuição taxonômica e geográfica da ambofilia e os atributos florais e da planta associados ao sistema ainda não foram acessados para um conjunto de espécies. A contribuição relativa dos componentes de polinização, biótico e abiótico, também não foi acessada. Foram conduzidas uma revisão sistemática global e uma meta-análise de espécies ambófilas para estimar a contribuição dos componentes de polinização no sucesso reprodutivo. Foram identificadas 71 espécies com potencial ambófilo na revisão, que são distribuídas em zonas tropicais e temperadas no globo. Um grupo de 46 espécies foi incluso na meta-análise, em que 44% são de espécies consistentemente ambófilas, associadas a flores actinomorfas e sistemas entomófilos generalistas de polinização. Os 56% restantes são casos em que as espécies foram consideradas funcionalmente anemófilas. A ambofilia pode ser considerada um sistema de generalização mais amplo por combinar insetos e vento como vetores de pólen, sendo o vento o agente mais comum e confiável quando os polinizadores são insuficientes.

Palavras-chave: ambofilia, polinização generalista, anemofilia, entomofilia, sistemas de polinização, interação planta-polinizador, polinização generalista, simetria floral, sistemas bimodais, sucesso reprodutivo.

ABSTRACT

The study of ambophily, a mixed pollination system in which plants depend on both biotic and abiotic pollen vectors, has been neglected. The taxonomic and geographic distributions of ambophily and the associated plant traits have not yet been assessed for a large set of species. The contributions of biotic and abiotic pollination components to reproductive success likewise require assessment. We conducted a global systematic review with a meta-analysis of ambophilous species to estimate the contribution of each pollen vector to their reproductive success. We identified 71 cases of potentially ambophilous species, which are widely distributed in tropical and temperate biomes across the globe. A subset of 46 species was included in the meta-analyses. About 44% of the cases were consistently ambophilous species, i.e., higher fruit set with combined biotic (mainly insects) and abiotic (wind) contributions than with only one vector. Actinomorphic flowers and functionally generalist pollination systems in the biotic component were associated with consistent ambophily. The remaining 56% of cases were considered functionally wind-pollinated species, because the total contribution of vectors was not significantly greater than the abiotic contribution alone. Consistently ambophilous species were associated with actinomorphic flowers, insect generalist systems, lower mean temperatures, and lower precipitation. Ambophily can be considered a broader case of generalist pollination systems because of the combination of wind and several insect species, since wind is a reliable pollen vector when pollinators are insufficient.

Key words: ambophily, generalist pollination, wind pollination, entomophily, pollination system, plant-pollinator interaction, bimodal pollination, reproductive assurance, floral symmetry, insect pollination.

SUMÁRIO

RESUMO.....	vi
ABSTRACT	vii
LISTA DE FIGURAS	ix
REFERÊNCIAS BIBLIOGRÁFICAS	13
ARTIGO CIENTÍFICO	15

LISTA DE FIGURAS

Figure 1: Evolutionary pathways in unpredictable pollination environments for a population with animal pollination as the ancestral state. At least three consequences are expected: a progressive negative demographic effect, leading to population extinction; evolution of self-fertilizing mechanisms; or changes in the pollination system. This last may lead to a shift toward a new group of pollinators (for example, from bee to hummingbird pollination); greater specialization or greater generalization of the pollination system; or a transition to abiotic pollination. In this hypothetical scenario, ambophily might be an intermediate condition in a full transition to wind pollination or a stable pollination system relying on biotic (specialist or generalist system) and abiotic pollen vectors. Ambophily may also evolve from wind-pollinated systems. Illustrations of pollinators obtained from <divulgare.net> and other elements obtained from <thenounproject.com>. Modified from Culley et al., 2002..... 32

Figure 2: Locations of study sites from 71 publications surveyed in the systematic review. The number of species reported at each location is indicated by the size of the circle or triangle. Triangles indicate studies conducted in tropical regions; circles indicate studies in extra-tropical regions. Blue indicates studies included in the meta-analysis; red indicates studies that were excluded.....33

Figure 3. A) The cumulative number of studies on ambophily from 2002–2020. B) The number of studies from 1945–2020 from Web of Science database searches for specialist pollination (speciali* AND pollinat*), wind pollination (wind AND pollinat*), generalist pollination (generali* AND pollinat*), and ambophily (amb*phil*).....34

Figure 4: Effect size and 95% confidence interval for 46 species used in the global meta-analysis of ambophilous species. Consistently ambophilous species includes cases where the biotic contribution was interpreted as significant; functionally wind-pollinated cases include species where biotic pollination did not contribute significantly to reproductive success.....35

Figure 5: Overall effect size (d) and 95% confidence interval of pollen limitation, and per category based on phylogenetically independent meta-analysis (90 cases of 49 species).....36

Figure 6: Overall relationship between size effects (Hedges' g) and a) mean annual temperature and b) mean annual precipitation. Solid line indicates the predicted relationships. Blue indicates

effect size from extratropical regions, and red from tropical regions.....	37
Figure S1: Diagnostics of traditional random-effects models. Diagnostics for model assumptions and publication bias: normal Q-Q plot, funnel plot, influence plot, symmetry test, overall effect size, heterogeneity, Rosenberg fail-safe number, and critical value for the complete dataset (104 cases from 53 species) and without outliers (84 cases from 46 species).....	38
Figure S2: Distribution of case locations across seven of the nine major global terrestrial biomes.....	39
Figure S3: Proportion of pollinator taxonomic groups represented in the animal-pollinated component of the 51 species included in the systematic review.....	40
Figure S4: Effect sizes (d) and 95% confidence interval of reproductive success and per category based on unit of sampling in the phylogenetically independent meta-analysis.....	41

INTRODUÇÃO GERAL

A polinização é um processo chave na reprodução e manutenção das espécies de angiospermas, caracterizado pela transferência de pólen entre flores (Jordano *et al.* 2003; Ollerton *et al.* 2007). O estudo da polinização subsidia o entendimento da ecologia e evolução das angiospermas, tendo influência em estudos de sistemática, conservação e dinâmica de comunidades (Lever *et al.* 2014; Arceo-Gómez *et al.* 2020). A maioria das angiospermas e uma parcela das gimnospermas utilizam espécies de invertebrados e vertebrados como mediadores do transporte de pólen, envolvendo sinais e recompensas florais (Ollerton *et al.* 2011; Ollerton, 2017). Em menor proporção, agentes abióticos também atuam como polinizadores, sendo que a polinização pelo vento ocorre em aproximadamente 18% das famílias de espermatófitas e em 12% das espécies de angiospermas (Ackerman, 2000; Ollerton *et al.* 2011).

Os estudos de biologia da polinização têm se concentrado em sistemas especializados (por exemplo, sistemas compostos por agentes pertencentes a um grupo taxonômico/funcional de polinizadores), enquanto outros sistemas, como a ambofilia, têm pouca representação na literatura (Culley *et al.* 2002; Timerman & Barrett, 2020). A ambofilia pode ser caracterizada como um sistema misto composto por dois componentes, biótico e abiótico. Na única revisão de literatura no tema, a ambofilia foi registrada para apenas 15 espécies de sete famílias de angiospermas (Culley *et al.* 2002). A ambofilia pode ser derivada de clados entomófilos e anemófilos e é incerto se é um sistema estável ou um sistema de transição entre o gradiente entomofilia-anemofilia (Culley *et al.* 2002; Friedman & Barrett, 2008; Timerman & Barrett, 2020).

Espécies polinizadas por animais e pelo vento têm morfologia que se diferenciam expressivamente, mesmo se pertencentes ao mesmo grupo taxonômico (Welsford *et al.* 2014, 2016). Na polinização por animais, são associadas espécies com corola chamativa e presença de odor e néctar, por exemplo (Raguso, 2004; Ollerton *et al.* 2011; Schiestl and Johnson, 2013). Já as anemófilas estão associadas a espécies dioicas com flores pequenas com anteras expostas e estigmas plumosos (Friedman and Barrett, 2009; Rech *et al.* 2014). Além da morfologia, os sistemas de polinização anemófilo e zoófilo são associados a diferentes escalas ambientais (Rech *et al.* 2016). A polinização por animais é esperada ser mais comum em ambientes tropicais, devido à diversidade de animais polinizadores, e fechados/florestais (Wragg & Johnson, 2011; Ollerton, 2017). Já a polinização pelo vento é associada a ambientes de altas altitudes e abertos (Regal, 1982; Rech *et al.*, 2016).

A ambofilia é esperada como resposta à limitação de polinizadores e/ou mudanças no ambiente que afetem o transporte pelo vento (Culley *et al.* 2002). Esses fatores levam à deficiência no recebimento de pólen ou na qualidade do pólen recebido (i.e., limitação polínica), o que implica na redução das medidas sucesso reprodutivo, principalmente de fecundidade feminina (Ashman *et al.* 2004; Bennett *et al.* 2018). Em sistemas de polinização em que há limitação polínica, é esperado que haja seleção de estratégias evolutivas que mitiguem a baixa frequência e/ou qualidade da transferência de pólen. Essas estratégias podem indicar processos de autofertilização, mudança no grau de especialização-generalização do sistema ou mudança no próprio sistema de polinização. Assim como a ambofilia, sistemas generalistas de polinização também são entendidos como resposta a cenários de polinização variável (Waser *et al.* 1996; Ollerton *et al.* 2011). Sistemas generalistas podem usar uma variedade de grupos funcionais e taxonômicos de polinizadores para garantir o sucesso reprodutivo. Para mediar essa interação, é esperado que espécies generalistas possuam atributos que facilitem o acesso de diferentes tipos de polinizadores até as estruturas reprodutivas (Yoder *et al.* 2020). Em sistemas generalistas entomófilos, são esperadas flores pequenas, abertas e com pouca quantidade de néctar (Moreira & Freitas, 2020). Por serem entendidos como respostas a limitação de polinizadores, é possível que o fenótipo final de espécies ambófilas seja semelhante ao encontrado em sistemas generalistas e que os dois sistemas sejam interligados.

Por possuir componentes de polinização notavelmente diferentes, é esperado que haja diferença na contribuição dos dois vetores de polinização no sucesso reprodutivo dessas espécies (q.v. Stebbins, 1970). Apesar das taxas de limitação polínica serem maiores em espécies zoófilas, a polinização por animais é considerada mais efetiva e direcionada, devido, por exemplo, à menor razão pólen-óvulo e à presença de sinais e recursos florais nas flores zoófilas (Raguso, 2004; Pellmyr *et al.* 2020). Entretanto, não foi produzida uma estimativa do conjunto de espécies ambófilas descritas desde Culley *et al.* (2002).

A meta-análise é uma ferramenta amplamente usada para entender a magnitude de processos biológicos, inclusive na biologia vegetal (Wolowski *et al.*, 2014; Zhang *et al.*, 2021). Além disso, a literatura acumulada por 18 anos desde a última revisão sobre o tema, junto aos avanços conceituais e instrumentais na biologia da polinização, favorece o avanço no entendimento das lacunas do conhecimento sobre a ambofilia. Para tal, foi proposta uma revisão sistemática e meta-análise do sucesso reprodutivo das espécies ambófilas em escala global, com objetivo de atualizar o estado da arte acerca de sistemas ambófilos, identificar padrões geográficos e frequências nos atributos florais e verificar as estimativas de contribuição dos componentes de polinização das espécies ambófilas registradas.

REFERÊNCIAS BIBLIOGRÁFICAS

- Ackerman J.D. 2000. Abiotic pollen and pollination: Ecological, functional, and evolutionary perspectives. *Plant Syst Evol.* 2000;222:167-85.
- Arceo-Gómez G., Barker D., Stanley A., Watson T., Daniels J. 2020. Plant–pollinator network structural properties differentially affect pollen transfer dynamics and pollination success. *Oecologia*, 192:1037-45.
- Ashman T-L., Knight T.M., Steets J.A., Amarasekare P., Burd M., Campbell D.R., Dudash M.R., Johnston M.O., Mazer S.J., Mitchell R.J., Morgan MT, Wilson WG. 2004. Pollen Limitation of Plant Reproduction: Ecological and Evolutionary Causes and Consequences. *Ecology*, 85:2408-21.
- Bennett J.M., Steets J.A., Burns J.H., Durka W., Vamosi J.C., Arceo-Gómez G., Burd M., Burkle L.A., Ellis A.G., Freitas L., Li J, Rodger J.G., Wolowski M., Xia J., Ashman T-L., Knight T.M.. 2018. GloPL, a global data base on pollen limitation of plant reproduction. *Scientific Data*, 5:180249.
- Costa A.C.G. da, Albuquerque I., Thomas W., Machado I.C. 2018. Influence of environmental variation on the pollination of the ambophilous sedge *Rhynchospora ciliata* (Cyperaceae). *Plant Ecology*.
- Culley T.M., Weller S.G., Sakai A.K. 2002. The evolution of wind pollination in angiosperms. *Trends in Ecology & Evolution*, 17:361-9.
- Friedman J., Barrett S.C.H. 2009. Wind of change: new insights on the ecology and evolution of pollination and mating in wind-pollinated plants. *Ann Bot*, 103:1515-27.
- Friedman J., Barrett S.C.H. 2008. A Phylogenetic Analysis of the Evolution of Wind Pollination in the Angiosperms. *International Journal of Plant Sciences*, 169:49-58
- Jordano P., Bascompte J., Olesen J.M. 2003. Invariant properties in coevolutionary networks of plant–animal interactions. *Ecology Letters*. 2003;6:69-81.
- Lever J.J., Nes E.H. van, Scheffer M., Bascompte J. 2014. The sudden collapse of pollinator communities. *Ecology Letters*, 17:350-9.
- Moreira M.M., Freitas L. 2020. Review of the Pollination System by Small Diverse Insects. *Neotrop Entomol.*, 49:472-81.
- Ollerton J. 2017. Pollinator Diversity: Distribution, Ecological Function, and Conservation. *Annual Review of Ecology, Evolution, and Systematics*, 48:353-76.
- Ollerton J., Killick A., Lamborn E., Watts S., Whiston M. 2007. Multiple meanings and modes: on the many ways to be a generalist flower. *TAXON*, 56:717-28.
- Ollerton J., Winfree R., Tarrant S. 2011. How many flowering plants are pollinated by animals? *Oikos*, 120:321-6. <https://doi.org/10.1111/j.1600-0706.2010.18644.x>
- Pellmyr O., Kjellberg F., Herre E.A., Kawakita A., Hembry D.H., Holland J.N., Terrazas T., Clement W., Segraves K.A., Althoff D.M.. 2020. Active pollination drives selection for reduced pollen-ovule ratios. *American Journal of Botany*, 107:164-70.
- Raguso R.A. 2004. Flowers as sensory billboards: progress towards an integrated understanding of floral advertisement. *Current Opinion in Plant Biology*, 7:434-40.
- Rech A.R., Agostini K., Oliveira P., Machado I. *Biologia da Polinização*. 2014

- Rech A.R., Dalsgaard B., Sandel B., Sonne J., Svenning J-C., Holmes N., Ollerton J. 2016. The macroecology of animal versus wind pollination: ecological factors are more important than historical climate stability. *Plant Ecology & Diversity*, 9:253-62.
- Regal P.J. 1982. Pollination by Wind and Animals: Ecology of Geographic Patterns. *Annual Review of Ecology and Systematics*, 13:497-524.
- Schiestl F.P., Johnson S.D. 2013. Pollinator-mediated evolution of floral signals. *Trends Ecol Evol*, 28:307-15.
- Stebbins G.L. 1970. Adaptive Radiation of Reproductive Characteristics in Angiosperms, I: Pollination Mechanisms. *Annual Review in Ecology & Systematics*, 1:307-26.
- Timerman D., Barrett S.C.H. 2020. Influence of local density and sex ratio on pollination in an ambophilous flowering plant. *American Journal of Botany*, 107:587-98.
- Waser N.M., Chittka L., Price M.V., Williams N.M., Ollerton J. 1996. Generalization in Pollination Systems, and Why it Matters. *Ecology*, 77:1043-60.
- Welsford M.R., Hobbhahn N., Midgley J.J., Johnson S.D. 2016. Floral trait evolution associated with shifts between insect and wind pollination in the dioecious genus *Leucadendron* (Proteaceae). *Evolution*, 70:126-39.
- Welsford M.R., Midgley J.J., Johnson S.D. 2014. Experimental Evaluation of Insect Pollination versus Wind Pollination in *Leucadendron* (Proteaceae). *International Journal of Plant Sciences*, 175:296-306.
- Wolowski M., Ashman T-L., Freitas L. 2014. Meta-Analysis of Pollen Limitation Reveals the Relevance of Pollination Generalization in the Atlantic Forest of Brazil. *PLoS One*, 9.
- Wragg P.D., Johnson S.D. 2011. Transition from wind pollination to insect pollination in sedges: experimental evidence and functional traits. *New Phytologist*, 191:1128-40.
- Yoder J.B., Gomez G., Carlson C.J. 2020. Zygomorphic flowers have fewer potential pollinator species. *Biology Letters*, 16:20200307.
- Zhang Z., Liu Y., Yuan L., Weber E., Kleunen M. van. 2021. Effect of allelopathy on plant performance: a meta-analysis. *Ecology Letters*, 24:348-62.

ARTIGO CIENTÍFICO

Meta-analysis of biotic vs. abiotic pollination in ambophilous species

Autores: Danilo Alves de Carvalho, Pedro Joaquim Bergamo,
Amanda Pacheco dos Santos, Marina Wolowski e Leandro Freitas

Introduction

Pollination is an essential ecological process that influences plant reproduction and the structure and dynamics of plant-pollinator communities (Ollerton et al., 2007; Lever et al., 2014; Arceo-Gómez et al., 2020). Most angiosperms and a few gymnosperms are pollinated by animals (Ollerton et al., 2011; Schiestl and Johnson, 2013), while wind pollination occurs in about 12.5% of angiosperms and is dominant in gymnosperms (Lu et al., 2011; Ollerton et al., 2011). Pollination by animals or by wind is associated with distinct floral phenotypes (Friedman and Barrett, 2009; Ollerton et al., 2011). However, some plant species are pollinated by both animals and wind, and scarcely represented in the literature (Norman et al., 1997; Culley et al., 2002; Friedman and Barrett, 2009; Timerman and Barrett, 2020). These cases are examples of ambophily, defined as a mixed pollination system composed of biotic and abiotic components (Norman et al., 1997; Culley et al., 2002). Ambophily is hypothesized to be a response to the limitation of pollinators and/or changes in the environment that affect pollen transport by wind (Culley et al., 2002). Deficiencies in the amount or quality of pollen received by flowers (i.e., pollen limitation) are widespread among angiosperms and reduce reproductive success, especially through the female function (Ashman et al., 2004; Bennett et al., 2018). In scenarios involving pollen limitation, evolutionary strategies that mitigate the negative effects of low frequency and/or quality of pollen receipt, such as selfing or shifts in the pollination system, are expected to be favored (Harder and Aizen, 2010). These strategies include possible shifts from zoophilous systems toward ambophilous or anemophilous systems (Fig. 1). Generalist pollination systems (pollination by several functional groups of animals) are also hypothesized

to be responses to unpredictable pollination environments (Waser et al., 1996; Ollerton, 2017). Therefore, selection of floral traits associated with generalist pollination systems in ambophilous species is expected. However, floral attributes have not yet been assessed for a large set of species described as ambophilous.

It is uncertain if ambophily is a stable system selected because of its flexibility in a scenario of pollen limitation or is a transitional stage between the zoophily-anemophily gradient (Culley et al., 2002; Friedman and Barrett, 2009; Timmerman and Barrett, 2020). Ambophily may also represent a particular bimodal system, because ambophilous species can interact with pollen vectors of markedly different natures (biotic and abiotic). Therefore, ambophilous species are expected to exhibit distinctive traits that attract pollinators and allow interaction with wind at the same time (Friedman and Barrett, 2009; Ollerton et al., 2011; Rech et al., 2016). Animal-pollinated species commonly have flowers with showy floral parts, odors, and resources such as nectar (Raguso, 2004; Schiestl and Johnson, 2013). In contrast, wind-pollinated species are associated with dioecy and small flowers with a reduced or absent perianth (Friedman and Barrett, 2009; Rech et al., 2014). These two systems have distinct ecogeographical patterns: pollination by wind is favored in high-altitude, open, temperate environments, whereas pollination by animals is favored in tropical, dense, forested environments (Wragg and Johnson, 2011; Rech et al., 2016).

Because ambophilous species interact with two strikingly different pollination vectors, one of the components (biotic or abiotic) is expected to be more effective for plant reproduction (the most effective pollinator principle, Stebbins, 1970). Although animals are considered to be more effective pollinators than wind (e.g., lower pollen/ovule ratio, Pellmyr et al., 2020), pollen limitation may be more frequent in animal-pollinated than in wind-pollinated species (e.g., Ashman et al., 2004; Friedman and Barrett, 2009). However, the relative contributions of biotic and abiotic components to the reproductive success of a large group of ambophilous

species have yet to be assessed. In only previous review of ambophily recorded 15 species belonging to seven families of angiosperms (Culley et al., 2002). Thus, it is unclear which component (biotic vs. abiotic) is more effective and the traits or conditions that may favor one component over the other in ambophilous species. The increase in studies on ambophilous species in the last two decades, together with conceptual and instrumental advances in pollination biology, have provided sufficient information to identify general patterns and better understand ambophily. In this study, we summarize the present state of knowledge of ambophily and estimate the relative contributions of biotic and abiotic components to plant reproductive success. We assessed the occurrence of ambophilous species through a systematic review, and the magnitudes of biotic and abiotic contributions through a meta-analysis. We also tested for a phylogenetic signal in the contributions of biotic and abiotic components. Besides that, we tested if the reproductive cycle (perennial or annual), mating system (self-compatible or self-incompatible), sexual system (hermaphroditic, monoecious, or dioecious), floral symmetry (actinomorphic or zygomorphic) and presence of a perianth (present or absent), climatic zone (tropical or extratropical), and pollination system (generalist or specialist) might explain the variations in the contributions of biotic and abiotic components.

Methods

Systematic Review and Data Coding

We conducted a systematic review of published studies, using the databases Web of Science (ISI-WoS), SciVerse Scopus, Scientific Electronic Library Online (SciELO), and Google Scholar. We used the key words “ambophil*” and “ambiphil*” (search 1 and 2; Côté et al., 2013), including papers published between 1985 and April 2020, resulting in 398 published studies (88 studies were found in more than one database). We also included published studies and academic theses from the authors’ personal libraries (9). From the unique entries (301), we

screened the titles, abstracts, and main text for studies that tested species with simultaneous animal and wind pollination. Studies that classified species based on floral morphology, phylogenetic relationships, or simply tested for ambophily but reported a non-ambophilous system were excluded. In total, 47 published studies encompassing 71 species met our criteria (46 published papers and one PhD thesis).

For each study, we extracted the following data: (a) year of publication, (b) geographic and climatic zone, and (c) pollination experimental method. Specifically, the pollination experimental methodology was divided into two categories: bagging experiments and sampling of airborne pollen. For each study site we extracted the geographic coordinates and altitude. If the geographic coordinates were not available, we extracted the data using Google Earth® based on the location description. We used the geographic data to assess bioclimatic variables (mean annual temperature and mean annual precipitation) using WorldClim 1.4 (Hijmans et al., 2005), when these data were not provided. We then classified the study sites, using the terrestrial biomes described by Whittaker (1961) and dividing them into tropical and extra-tropical regions. All taxonomic nomenclature was checked with the Taxonstand R package.

In addition to the data extracted from the papers, morphological and ecological attributes were obtained, primarily from the original publication, or, if not available, using taxonomic databases (e.g., World Checklist of Selected Plant Families), regional or national floras (e.g., Flora do Brasil, 2020) and papers on taxonomy. The following attributes were recorded: i. sexual system (dioecious, monoecious with unisexual flowers, or monoecious with bisexual flowers), ii. reproductive cycle (annual or perennial), iii. life form (tree, shrub, herb, palm, or vine), iv. mating system (self-compatible or self-incompatible), v. perianth (present or absent/vestigial), vi. floral symmetry (actinomorphic or zygomorphic), vii. corolla type (tubular or open), viii. color (white, beige/cream, yellow, green, red, pink, or violet/lilac, *sensu* Machado & Lopes, 2004), and ix. floral resources (pollen, nectar, or both pollen and nectar). For the

sexual system, we then analyzed differences in the proportion of each group (unisexual flowers vs. unisexual flowers) in the review and for angiosperms (*sensu* Renner, 2014), using chi-squared tests. Two andromonoecious species were categorized as monoecious, and three bi-annual species were included as annuals. Data for flower size, corolla size, inflorescence size, and nectar volume were extracted only if this information was available in the original publication. Information about animal pollination was recorded when a taxonomic identification was provided. Floral visitors were classified as pollinators only when a legitimate visit was recorded (*sensu* Inouye, 1980). Therefore, we excluded visitors recorded as floral larcenists. Flower visitors were divided into eight functional-taxonomic groups: ants, bees, beetles, butterflies, flies, moths, thrips, and wasps. Visitors of other groups were rare, and we grouped them in the category of “others” (including lacewings, lizards, and hummingbirds). The zoophilous component of ambophilous species was also classified based on its degree of specialization-generalization, as a specialized (one functional-taxonomic group), bimodal (two), or generalized (three or more) system.

Meta-analysis

To estimate wind and animal contributions to reproductive success, we conducted a meta-analysis from a subset of 31 studies, which measured fruit and seed set of angiosperm species in natural conditions (i.e., both wind and animal pollination) and after animal exclusion (i.e., only wind pollination). When data were available in graphs, we used GetData Graph Digitizer 2.26 to extract it (<getdata-graph-digitizer.com>). We calculated effect size based on the log odds ratio for proportion data and Cohen’s *d* for data based on averages between the two treatments (wind and animal pollination vs. only wind pollination). Both values were transformed to Hedge’s *g* to facilitate interpretation and comparison among studies (Cooper et al., 2009). We also assessed which response variables were measured (fruit set or seed set) and

the level at which treatments were applied (flower, inflorescence, or whole plant). If the same study provided separate estimates for more than one species, year, location, or morphotypes, we recorded each estimate as a different case. In total, 104 cases from 53 angiosperm species composed the final dataset for the meta-analysis.

For the effect size of a given case or for the overall effect size, the biotic contribution was interpreted as significant when the effect size was positive and its 95% confidence interval did not overlap zero (hereafter “consistently ambophilous species”). Negative values of effect size or 95% confidence intervals that overlapped zero meant that biotic pollination did not contribute to increase reproductive success (hereafter “functionally wind-pollinated species”). If a species had cases belonging to the two different groups, each case was interpreted individually.

To test the assumptions of the meta-analysis model and publication bias, we analyzed the models using visual and statistical methods (normal Q-Q plot, influence plot, funnel plot, symmetry test, normality test, and Rosenberg fail-safe number) using the *metafor* package (Fig. S1). After 15 outliers were detected and removed, the normality and symmetry assumptions of the model were acceptable. For this subset (without outliers) of 84 cases from 46 species, the Rosenberg fail-safe number (12,124) was larger than the critical value (460).

Overall effect size was estimated by traditional and phylogenetically independent random-effect models, using the *metafor* package. Random-effect models were chosen, as they take into account the variation between the true effect size across study populations and the variation due to sampling error (Borenstein et al., 2009). For the phylogenetically independent meta-analysis, the phylogenetic hypothesis was incorporated using the angiosperm APG IV mega phylogenetic tree by Zanne et al. (2014) as modified by Jian and Qin (2016). The phylogeny was also used to test the phylogenetic signal of effect sizes, using Pagel’s (λ) model in *phytools* (Revell, 2020). Phylogenetically independent meta-analyses had values of Akaike’s

information criterion (AICc) lower than the traditional model (23 units), and the interpretation of results focused on the phylogenetically independent random-effect models (Table S1).

Overall heterogeneity was statistically significant (see Results), indicating that there was among-case variation among effect sizes. To investigate the possible explanation of among-case variation, we fitted meta-analytical models, using each categorical plant trait as moderators (reproductive cycle, mating system, breeding system, floral symmetry, perianth presence, climatic zone, and pollination system) and meta-regressions using continuous variables (temperature, precipitation, and altitude). Within each plant trait, a category was taken into consideration cautiously if the number of cases was lower than ten (self-compatible system, zygomorphic symmetry, and monoecious system). All statistical tests were performed in the R environment (R CORE TEAM, 2020).

RESULTS

Trends in ambophilous species

Experiments on wind and animal pollination were conducted for 71 species of vascular plants in 47 published studies, mainly in Asia (31%), South America (27%), and Europe (25%) (Fig. 2). These experiments were conducted at tropical and temperate sites and in seven of the nine terrestrial biomes described by Whittaker (1961), most of them in seasonal tropical forests (40%), woodland/shrubland (26%), and seasonal temperate forests (11%) (Fig. S2).

Although the number of published studies on ambophily has gradually increased since the previous review by Culley et al. (2002), the cumulative number of papers is equivalent to only 1% of the number of published papers about specialized animal-pollinated systems, for instance (Fig. 3). Most studies have focused on a single species (91%) or a few phylogenetically related species (9%). One-quarter of the studies evaluated temporal and/or spatial variation. Ambophily was mainly studied by using bagging experiments (91%) but also by sampling

airborne pollen (9%) (Table S2).

Taxonomic distribution and floral traits

Seventy-one species of ambophilous species belong to 33 families in 25 orders were included (APG, 2016). The best represented orders were the angiosperms Malpighiales (21%, 15 species), Poales (15%), and Arecales (9%); and the gymnosperms Cycadales (50%, 2 species), Ephredales (25%), and Gnetales (25%). Most species were perennial (71%) and shrubby (43%). Almost half of the species were hermaphroditic (46%) but unisexual flowers were more frequent in ambophilous species than in angiosperms (54% vs. 15% (sensu Renner, 2014), $\chi^2 = 31.95$, $df = 1$, $P < 0.05$). Most ambophilous flowers were apetalous (60%); petalous flowers ($n = 28$ or 40% of total) were frequently actinomorphic (89%) and dish-shaped (54%). Flowers were mostly pale (85% of 67 species), but petals and other floral structures, such as bracts and stamens, in some species were colored (see Table S3). Concerning resources, most species used both pollen and nectar as flower resources (60%), and nectar was predominantly present in tubular flowers (80%) (mean nectar volume = $1.0 \pm 2.0 \mu\text{L}$, $n = 11$) (Table S3).

Regarding the zoophilous component, the pollination system of most species was classified as generalist (52% of 50 species), followed by specialist (32%) and bimodal (16%). Flies and bees were the most frequent pollinator groups (72% and 62%, respectively). Only two species were not pollinated exclusively by insects: *Ephedra fragilis* (Ephedraceae) was pollinated by lizards, and *Triplaris gardneriana* (Polygonaceae) by hummingbirds (Table S3, Fig. S3).

Estimates of the contribution by biotic and abiotic vectors

The overall effect size calculated for 84 cases, representing 46 species, was 0.63 with a positive confidence interval (95% CI = -0.06 to 1.3, Table S2). The effect sizes of 37 cases, representing

26 species, were interpreted as consistently ambophilous systems, and 47 cases, representing 24 species, as functionally wind-pollinated species. Four species (*Aconitum gymnanthum*, *Mallotus japonicus*, *Excoecaria agallocha*, and *Rhynchospora ciliaris*) had cases falling into both groups (Fig. 4). The phylogenetic signal was not significant ($\lambda = 0.42$, $P = 0.5$).

The heterogeneity of the random phylogenetic effect model was large ($Q = 478.93$, $P < 0.001$), indicating variability in the effect size among cases. Although the heterogeneity of moderators was not significant for all plant traits, some categories within traits had positive and significant effects, namely pollination generalization and actinomorphic floral symmetry (Fig. 5). Sampling units (flowers, inflorescences, and whole plant) showed effect sizes with confidence intervals overlapping zero (i.e., did not explain among-case variation in effect sizes) (Fig. S4).

We found a negative relationship between effect size and mean annual temperature, and between effect size and mean precipitation (meta-regressions -0.003 95% CI $[-0.005$ to $-0.0006]$ for temperature and -0.003 95% CI $[-0.005$ to $0]$ for precipitation, both $P < 0.05$, Fig. 6). That is, consistent ambophily was associated with lower temperatures and drier environments. Latitude and altitude meta-regressions were not significant.

DISCUSSION

After almost two decades, cases of ambophily in the literature increased fivefold since the last review (Culley et al., 2002). However, the ambophilous pollination system continued to be neglected in reproductive biology studies, compared to the interest in more specialized pollination systems, for example. The reasons are not clear-cut, but may reflect the problem that ambophily is not predicted and may not be inferred based on classic pollination syndromes (see Dellinger, 2020). Ambophily usually requires careful observation to detect and may remain hidden as a case of wind or animal pollination. This being so, at present it is not possible to

ascertain if the occurrence of ambophily is very rare or underestimated. Feasible methods that can detect ambophily in the field are encouraged, such as observation of floral visitors in presumed wind-pollinated species (e.g., Costa et al., 2018; Dórea et al., 2018; Schulze-Albuquerque et al., 2020) and sampling of airborne pollen in presumed animal-pollinated species (e.g., Cresswell et al., 2004; Mangla et al., 2011; Yamasaki et al., 2013). Moreover, it is still difficult to separate the relative contribution of animal and wind pollination to the reproductive success of ambophilous species, because studies do not include a treatment evaluating each component separately. This may lead to overestimation of the abiotic contribution in bagging experiments, which are most often used to test for ambophily. Methodological refinements are welcome in order to advance our understanding of ambophily.

In this first meta-analysis for ambophilous species, we established that ambophily is a pollination system that can vary along a gradient of biotic-abiotic contributions to reproductive success, highlighting two groups: consistently ambophilous species and functionally wind-pollinated species. Nevertheless, we did not find a phylogenetic signal in the effect sizes, meaning that these groups were composed of distantly related species. These new findings indicate that ambophily is a context-dependent ecological process (Chamberlain et al., 2014; Rech et al., 2016). For example, in some cases ambophily may be evolutionarily stable (*sensu* Culley et al., 2002), and in other cases may be an intermediate stage from insect pollination to wind pollination from animal-pollinated ancestors, or a full reversal toward wind pollination from wind-pollinated ancestors. To evaluate the evolutionary trajectory of ambophily in a particular group, phylogenetic relationships and pollination systems from related species for each group should be taken into consideration (Friedman and Barrett, 2009; Wragg and Johnson, 2011; Costa, 2017). Moreover, the two groups (consistent ambophily vs. functional wind pollination) may occur due to particular spatiotemporal variations that lead to different assemblages of floral visitors in each reproductive event, as reflected by four species that

showed marked differences in effect sizes depending on the particular case. For each situation, ambophily may be a consistent, safe pathway to mitigate pollen loss in both wind- and animal-pollinated groups. This suggests that ambophily may be favored as a functionally flexible pollination strategy (Friedman and Barrett, 2009; Timerman and Barrett, 2020).

Nearly half of the species in this meta-analysis had wind as their only effective pollen vector. Wind-pollination systems may be favored in unpredictable pollination environments where pollinators are scarce (Goodwillie, 1999; Lázaro and Traveset, 2005; Medan and Devoto, 2005; Rios et al., 2014) or absent (e.g., pioneer habitats, Custodio et al., 2017). Moreover, some floral traits in animal-pollinated species, either morphological (e.g., "operculum type" corolla in Ericaceae; Ladd, 2006) or aerobiological, may mediate interaction with wind, favoring shifts to wind pollination. For instance, the movement of the fusiform dry pollen of *Aconitum gymnantrum* (Ranunculaceae) was more efficient in open or marginal sites (Wang et al., 2017). In any case, the wind contribution to reproductive success can be overestimated in ambophilous systems. Animal pollination is less predictable than wind pollination, so contribution by pollinators in truly ambophilous species can go undetected after some exclusion experiments, which may at least partly explain the high number of functionally wind-pollinated species in our review.

Setting aside these methodological caveats, our results indicated that animal pollinators (mainly insects) act as effective pollen vectors in half of the species. Animals may still ensure reproductive success in ambophilous species during periods of calmer winds, as well as contribute to other pollination proxies not evaluated here, such as the male function (Ashman et al., 2004; Rosas-Guerrero et al., 2014; Leal et al., 2020). The contribution of wind and animals to reproductive success may be maintained by the high frequency of floral traits across species found here, such as small, simple, pale, open, and unisexual flowers, which are related to both wind- and generalist insect-pollinated floral phenotypes (Friedman & Barrett, 2009;

Moreira & Freitas, 2020; Yoder et al., 2020). Therefore, phenotypic generalization may ensure insect pollination when wind is ineffective, without significantly compromising the efficacy of wind pollination in wind-favorable scenarios.

The relative contributions of wind and insects were not related to plant traits in our review. We did not expect these results in view of the well-established associations between morphological traits and pollination systems (Friedman and Barrett, 2009; Ollerton et al., 2011; Rech et al., 2016). However, different plant traits may be favored to function simultaneously for wind and insect pollination, resulting in even frequencies of floral traits related to each pollen vector in ambophilous species. An analogy can be drawn with the influences of abiotic and biotic non-pollinator agents on floral evolution (Strauss & Whittall, 2006).

Although the contributions of pollination components did not differ significantly between climatic zones, consistently ambophilous species were associated with lower mean temperatures and lower precipitation. Pollen release and dispersal into the atmosphere by ambophilous species have been associated with higher temperatures and humid environments (Custodio et al., 2017; Costa et al., 2018). The association between consistent ambophily and colder and drier environments may reflect the presence of more airborne pollen grains in warmer environments. Thus, a higher wind contribution to reproductive success, in addition to insects being more sensitive to environmental variation, would favor functionally wind-pollinated species in warmer and more-humid environments.

Phenotypic (actinomorphic floral symmetry) and functional-taxonomic (number of taxonomic groups of pollinators) generalization was associated with consistent ambophily, although floral symmetry overall did not explain the variation in effect sizes. Half of the species surveyed in the review are generalists in their biotic component (e.g., Duan et al., 2009; Cursach and Rita, 2012; Gulías and Traveset, 2012; Custodio et al., 2017). These findings suggest that generalist pollination systems may increase the success biotic component in ambophilous

species. Thus, ambophily can be considered a broader generalist pollination system, because ambophilous species usually combine wind pollination with several functional groups of animal pollinators. The reasons behind the association of ambophily with pollination by several insects are not clear. However, generalist pollination systems are expected to evolve in response to unpredictably fluctuating pollination environments (Waser et al., 1996; Ollerton et al., 2007), and the evolution of wind pollination in animal-pollinated plants may be more frequent in these scenarios of a high degree of generalization due to low densities of pollinators.

CONCLUSION

Based on a global-scale systematic review and meta-analysis of ambophilous species, ambophily can vary along a gradient of biotic-abiotic contributions to reproduction. We demonstrated that half of the species in this meta-analysis are effectively pollinated by wind, and the contribution of wind increased with increasing temperature and rainfall, whereas actinomorphic flowers and generalist pollination systems were associated with the contribution of biotic pollination to reproduction success. These findings indicate that simultaneously animal- and wind-pollinated species are, in fact, efficiently pollinated only by wind and may or may not exhibit floral attributes associated with animal pollination. Overall, these findings demonstrate that ambophily may be under-represented in pollination studies due to its transitory nature or lack of practical methods to properly detect this system. Pollination experiments should be encouraged in order to detect ambophily in plant communities and fill gaps in knowledge of this neglected pollination system.

Acknowledgments

We thank Amparo Lázaro for providing seed-set data for *Buxus balearica*; Sávio Cavalcante for assistance in the framework design; Janet Reid for language editing; FAPERJ for a CNE

fellowship to LF (No. E-26-202.775/2018) and a postdoctoral fellowship to PJB (No. E-26/201.867/2020; CNPq for a graduate scholarship to DAC (No. 132464/2019-5); and CAPES for a graduate scholarship to APS (No. 88887.477571/2020-00). This paper is part of the M.Sc. dissertation of DAC, presented to the Graduate Program in Botany of the Rio de Janeiro Botanical Garden.

Literature Cited

- Abrahamczyk, S., L. S. Dannenberg, and M. Weigend. 2020. Pollination modes and divergent flower traits in three species of *Plantago* subgenus *Plantago* (Plantaginaceae). *Flora* 267: 151601.
- Arceo-Gómez, G., D. Barker, A. Stanley, T. Watson, and J. Daniels. 2020. Plant–pollinator network structural properties differentially affect pollen transfer dynamics and pollination success. *Oecologia* 192: 1037–1045.
- Ashman, T.-L., T. M. Knight, J. A. Steets, P. Amarasekare, M. Burd, D. R. Campbell, M. R. Dudash, et al. 2004. Pollen Limitation of Plant Reproduction: Ecological and Evolutionary Causes and Consequences. *Ecology* 85: 2408–2421.
- Bennett, J. M., J. A. Steets, J. H. Burns, W. Durka, J. C. Vamosi, G. Arceo-Gómez, M. Burd, et al. 2018. GloPL, a global data base on pollen limitation of plant reproduction. *Scientific Data* 5: 180249.
- Borenstein, M., L. Hedges, J. Higgins, and H. Rothstein. 2009. An Introduction to Meta-Analysis. *Introduction to Meta-Analysis* 19.
- Chamberlain, S. A., J. L. Bronstein, and J. A. Rudgers. 2014. How context dependent are species interactions? *Ecology Letters* 17: 881–890.
- Costa, A. C. G. da, I. Albuquerque, W. Thomas, and I. C. Machado. 2018. Influence of environmental variation on the pollination of the ambophilous sedge *Rhynchospora ciliata* (Cyperaceae). *Plant Ecology*.
- Cresswell, J. E., T. W. Davies, M. A. Patrick, F. Russell, C. Pennel, M. Vicot, and M. Lahoubi. 2004. Aerodynamics of wind pollination in a zoophilous flower, *Brassica napus*. *Functional Ecology* 18: 861–866.
- Culley, T. M., S. G. Weller, and A. K. Sakai. 2002. The evolution of wind pollination in angiosperms. *Trends in Ecology & Evolution* 17: 361–369.

- Cursach, J., and J. Rita. 2012. Reproductive biology of *Ranunculus weyeri* (Ranunculaceae), a narrowly endemic plant from the Balearic Islands with disjunct populations. *Flora - Morphology, Distribution, Functional Ecology of Plants* 207: 726–735.
- Custodio, T., P. Comtois, and A. C. Araujo. 2017. Reproductive biology and pollination ecology of *Triplaris gardneriana* (Polygonaceae): a case of ambophily in the Brazilian Chaco. *Plant Biology* 19: 504–514.
- Duan, Y. W., T. F. Zhang, Y. P. He, and J. Q. Liu. 2009. Insect and wind pollination of an alpine biennial *Aconitum gymnantrum* (Ranunculaceae). *Plant Biology (Stuttgart, Germany)* 11: 796–802.
- Friedman, J., and S. C. H. Barrett. 2009. Wind of change: new insights on the ecology and evolution of pollination and mating in wind-pollinated plants. *Annals of Botany* 103: 1515–1527.
- Gong, Y.-B., M. Yang, J. C. Vamosi, H.-M. Yang, W.-X. Mu, J.-K. Li, and T. Wan. 2016. Wind or insect pollination? Ambophily in a subtropical gymnosperm *Gnetum parvifolium* (Gnetales). *Plant Species Biology* 31: 272–279.
- Goodwillie, C. 1999. Wind Pollination and Reproductive Assurance in *Linanthus parviflorus* (Polemoniaceae), a Self-Incompatible Annual. *American Journal of Botany* 86: 948–954.
- Gulías, J., and A. Traveset. 2012. Altitudinal variation in the reproductive performance of the Mediterranean shrub *Rhamnus lycioides* L. *Journal of Plant Ecology* 5: 330.
- Harder, L. D., and M. A. Aizen. 2010. Floral adaptation and diversification under pollen limitation. *Philosophical Transactions of the Royal Society B: Biological Sciences* 365: 529–543.
- Inouye, D. W. 1980. The Terminology of Floral Larceny. *Ecology* 61: 1251–1253.
- Lázaro, A., and A. Traveset. 2005. Spatio-temporal variation in the pollination mode of *Buxus balearica* (Buxaceae), an ambophilous and selfing species: mainland-island comparison. *Ecography* 28: 640–652.
- Leal, R. L. B., M. M. Moreira, A. R. Pinto, J. de O. Ferreira, M. Rodriguez-Girones, and L. Freitas. 2020. Temporal changes in the most effective pollinator of a bromeliad pollinated by bees and hummingbirds. *PeerJ* 8: e8836.
- Lever, J. J., E. H. van Nes, M. Scheffer, and J. Bascompte. 2014. The sudden collapse of pollinator communities. *Ecology Letters* 17: 350–359.
- Lu, Y., B. Jin, L. Wang, Y. Wang, D. Wang, X.-X. Jiang, and P. Chen. 2011. Adaptation of male reproductive structures to wind pollination in gymnosperms: Cones and pollen grains. *Canadian Journal of Plant Science*.

- Machado, I., and A. Lopes. 2004. Floral Traits and Pollination Systems in the Caatinga, a Brazilian Tropical Dry Forest. *Annals of botany* 94: 365–76.
- Mahy, G., J. De Sloover, and A.-L. Jacquemart. 1998. The generalist pollination system and reproductive success of *Calluna vulgaris* in the Upper Ardenne. *Canadian Journal of Botany* 76: 1843.
- Mangla, Y., R. Tandon, Y. Mangla, and R. Tandon. 2011. Insects facilitate wind pollination in pollen-limited *Crateva adansonii* (Capparaceae). *Australian Journal of Botany* 59: 61–69.
- Manning, J., P. Goldblatt, and B. Krukoff. 2005. Radiation of Pollination Systems in the Cape Genus *Tritoniopsis* (Iridaceae: Crocoideae) and the Development of Bimodal Pollination Strategies. *International Journal of Plant Sciences - INT J PLANT SCI* 166.
- Medan, D., and M. Devoto. 2017. Ambophily, not entomophily: the reproduction of the perennial *Discaria chacaye* (Rhamnaceae: Colletieae) along a rainfall gradient in Patagonia, Argentina. *Plant Systematics and Evolution* 303: 841–851.
- Medan, D., and M. Devoto. 2005. Reproductive ecology of a perennial outcrosser with a naturally dissected distribution. *Plant Systematics and Evolution* 254: 173–184.
- Norman, J., S. Weller, and A. Sakai. 1997. Pollination biology and outcrossing rates in hermaphroditic *Schiedea lydgatei* (Caryophyllaceae). *American Journal of Botany* 84: 641.
- Ollerton, J., A. Killick, E. Lamborn, S. Watts, and M. Whiston. 2007. Multiple meanings and modes: on the many ways to be a generalist flower. *TAXON* 56: 717–728.
- Ollerton, J., R. Winfree, and S. Tarrant. 2011. How many flowering plants are pollinated by animals? *Oikos* 120: 321–326.
- Rech, A., P. J. Bergamo, and R. A. de Figueiredo. 2014. Polinização abiótica. In A. R. Rech, K. Agostini, P. E. Oliveira, and I. C. Machado [eds.], *Biologia da Polinização*, 183–204. Ceres Belchior, Rio de Janeiro.
- Rech, A. R., B. Dalsgaard, B. Sandel, J. Sonne, J.-C. Svenning, N. Holmes, and J. Ollerton. 2016. The macroecology of animal versus wind pollination: ecological factors are more important than historical climate stability. *Plant Ecology & Diversity* 9: 253–262.
- Rios, L. D., E. J. Fuchs, D. R. Hodel, and A. Cascante-Marín. 2014. Neither insects nor wind: ambophily in dioecious *Chamaedorea* palms (Arecaceae). *Plant Biology (Stuttgart, Germany)* 16: 702–710.

- Rosas-Guerrero, V., R. Aguilar, S. Martén-Rodríguez, L. Ashworth, M. Lopezaraiza-Mikel, J. M. Bastida, and M. Quesada. 2014. A quantitative review of pollination syndromes: do floral traits predict effective pollinators? *Ecology Letters* 17: 388–400.
- Schiestl, F. P., and S. D. Johnson. 2013. Pollinator-mediated evolution of floral signals. *Trends in Ecology & Evolution* 28: 307–315.
- Strauss, S. Y., and J. B. Whittall. Non-pollinator agents of selection on floral traits. *Ecology and Evolution of Flowers*, 120–138. Oxford University Press, Oxford.
- Timerman, D., and S. C. H. Barrett. 2020. Influence of local density and sex ratio on pollination in an ambophilous flowering plant. *American Journal of Botany* 107: 587–598.
- Waser, N. M., L. Chittka, M. V. Price, N. M. Williams, and J. Ollerton. 1996. Generalization in Pollination Systems, and Why it Matters. *Ecology* 77: 1043–1060.
- Wragg, P. D., and S. D. Johnson. 2011. Transition from wind pollination to insect pollination in sedges: experimental evidence and functional traits. *New Phytologist* 191: 1128–1140.
- Yamasaki, E., S. Sakai, E. Yamasaki, and S. Sakai. 2013. Wind and insect pollination (ambophily) of *Mallotus* spp. (Euphorbiaceae) in tropical and temperate forests. *Australian Journal of Botany* 61: 60–66.
- Yoder, J. B., G. Gomez, and C. J. Carlson. 2020. Zygomorphic flowers have fewer potential pollinator species. *Biology Letters* 16: 20200307.

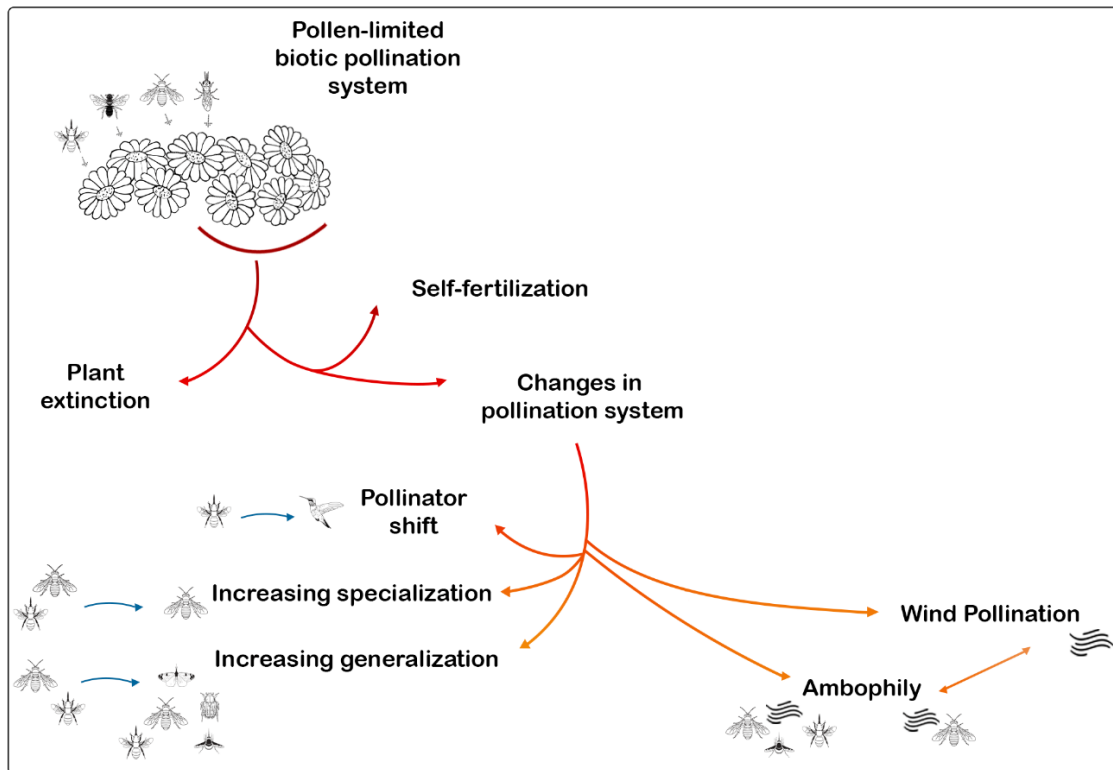


Figure 1: Evolutionary pathways in unpredictable pollination environments for a population with animal pollination as the ancestral state. At least three consequences are expected: a progressive negative demographic effect, leading to population extinction; evolution of self-fertilizing mechanisms; or changes in the pollination system. This last may lead to a shift toward a new group of pollinators (for example, from bee to hummingbird pollination); greater specialization or greater generalization of the pollination system; or a transition to abiotic pollination. In this hypothetical scenario, ambophily might be an intermediate condition in a full transition to wind pollination or a stable pollination system relying on biotic (specialist or generalist system) and abiotic pollen vectors. Ambophily may also evolve from wind-pollinated systems. Illustrations of pollinators obtained from <divulgare.net> and other elements obtained from <thenounproject.com>. Modified from Culley et al., 2002.

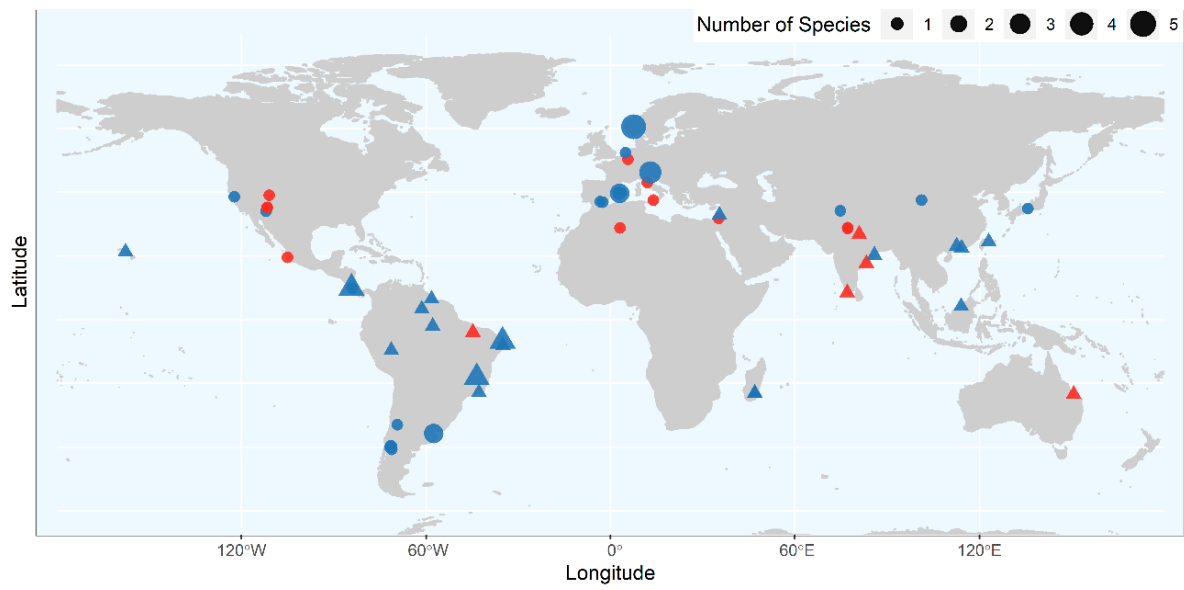


Figure 2: Locations of study sites from 71 publications surveyed in the systematic review. The number of species reported at each location is indicated by the size of the circle or triangle. Triangles indicate studies conducted in tropical regions; circles indicate studies in extra-tropical regions. Blue indicates studies included in the meta-analysis; red indicates studies that were excluded.

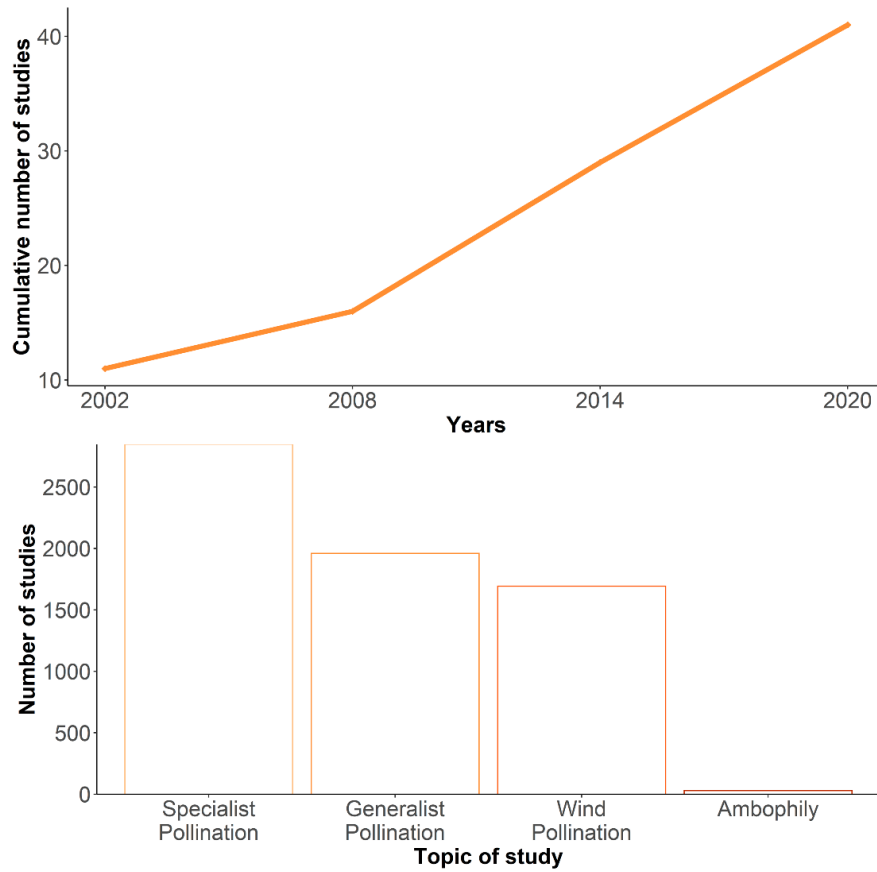


Figure 3. A) The cumulative number of studies on ambophily from 2002–2020. B) The number of studies from 1945–2020 from Web of Science database searches for specialist pollination (speciali* AND pollinat*), wind pollination (wind AND pollinat*), generalist pollination (generali* AND pollinat*), and ambophily (amb*phil*).

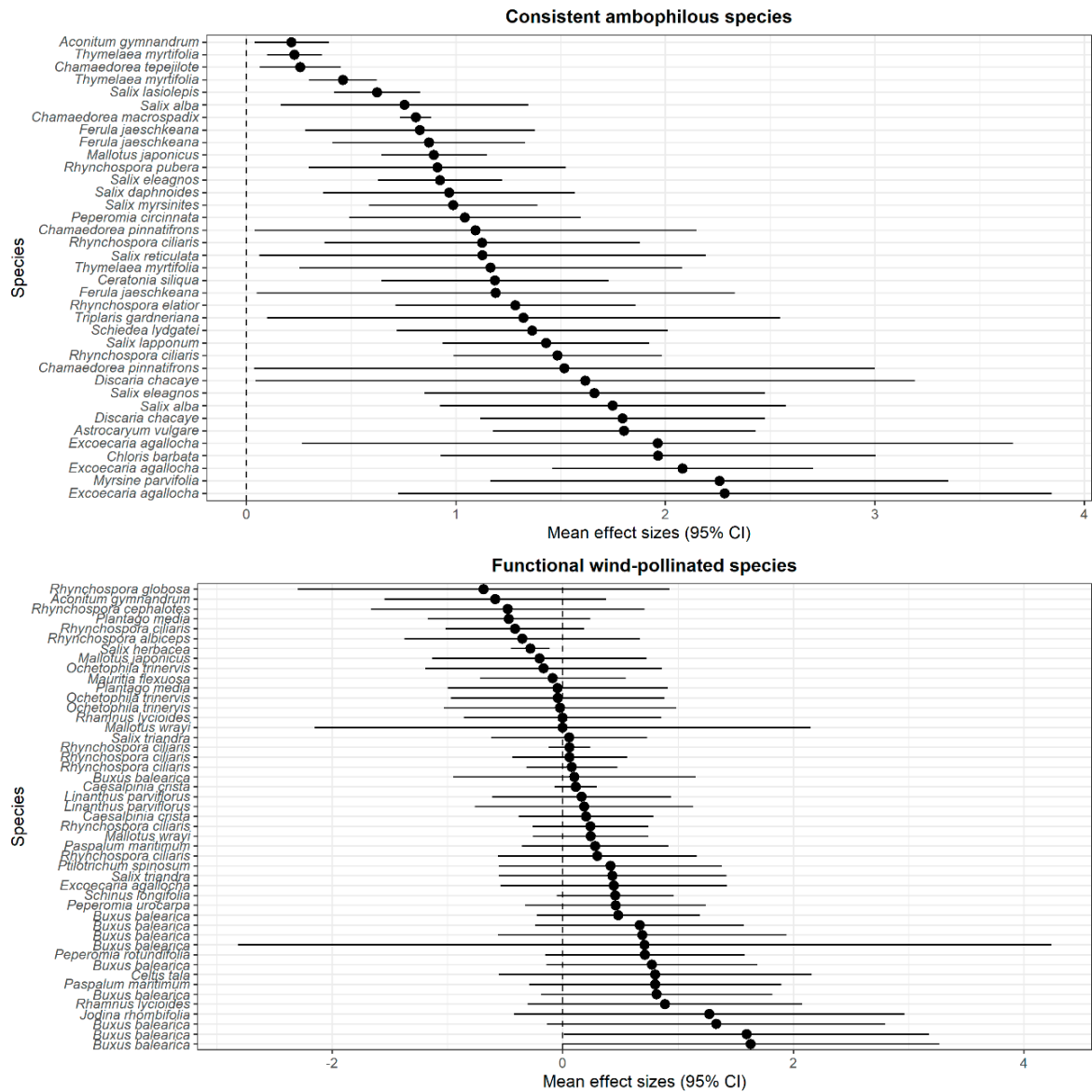


Figure 4: Effect size and 95% confidence interval for 46 species used in the global meta-analysis of ambophilous species. Consistently ambophilous species includes cases where the biotic contribution was interpreted as significant; functionally wind-pollinated cases include species where biotic pollination did not contribute significantly to reproductive success.

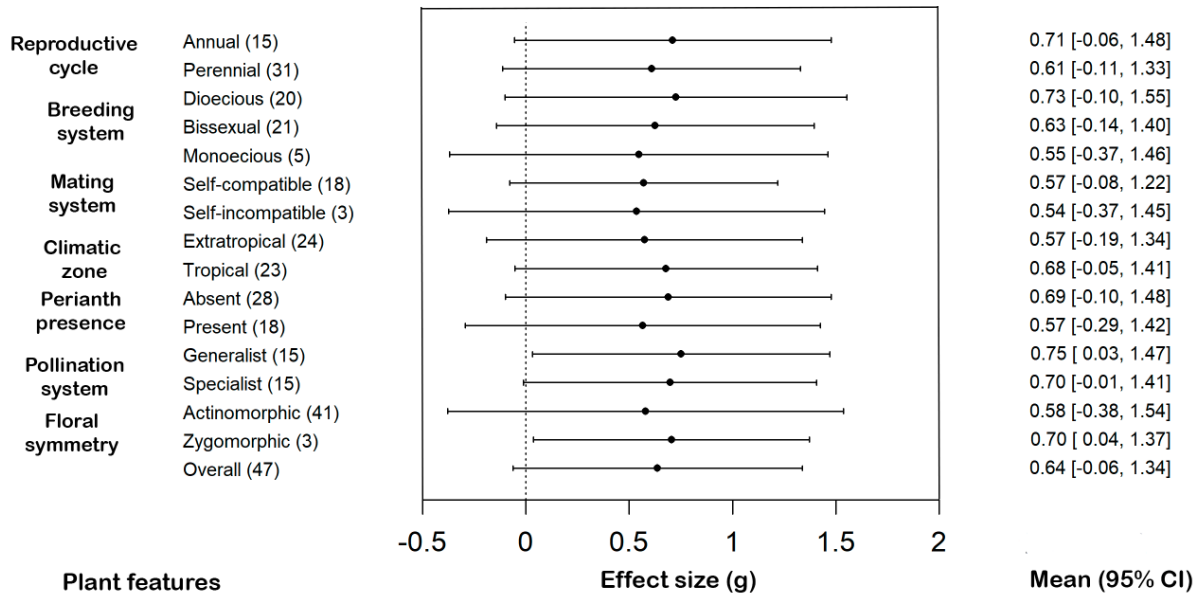


Figure 5: Overall effect size (d) and 95% confidence interval of pollen limitation, and per category based on phylogenetically independent meta-analysis (90 cases of 49 species).

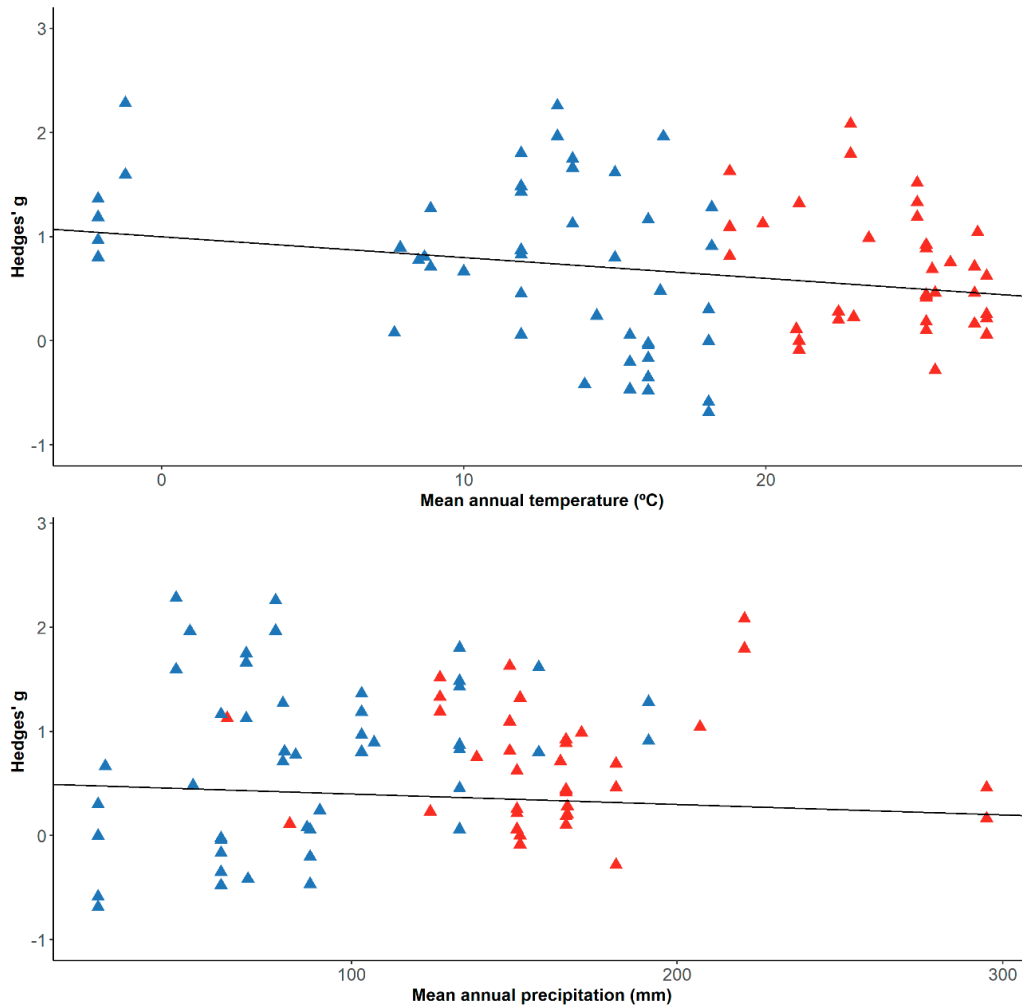


Figure 6: Overall relationship between size effects (Hedges' g) and a) mean annual temperature and b) mean annual precipitation. Solid line indicates the predicted relationships. Blue indicates effect size from extratropical regions, and red from tropical regions.

SUPPLEMENTARY MATERIAL

Appendix S1: References for the studies included in the systematic review. Data from tagged studies (*) were also included in the meta-analysis.

- Abrahamczyk, S., L. S. Dannenberg, and M. Weigend. 2020. Pollination modes and divergent flower traits in three species of *Plantago* subgenus *Plantago* (Plantaginaceae). *Flora* 267: 151601.*
- Albuquerque, A. A. E., H. A. de Lima, V. Gonçalves-Esteves, C. R. Benevides, and A. T. A. Rodarte. 2013. *Myrsine parvifolia* (Primulaceae) in sandy coastal plains marginal to Atlantic rainforest: a case of pollination by wind or by both wind and insects? *Brazilian Journal of Botany* 36: 65–73.*
- de la Bandera, M. C., and A. Traveset. 2006. Breeding system and spatial variation in the pollination biology of the heterocarpic *Thymelaea velutina* (Thymelaeaceae). *Plant Systematics and Evolution* 257: 9–23.*
- Celedón-Neghme, C., L. Santamaría, and M. González-Teuber. 2016. The role of pollination drops in animal pollination in the Mediterranean gymnosperm *Ephedra fragilis* (Gnetales). *Plant Ecology* 217: 1545–1552.*
- Cornara, L., B. Borghesi, E. Caporali, G. Casazza, E. Roccotiello, G. Troiano, and L. Minuto. 2005. Floral features and reproductive ecology in *Thymelaea hirsuta* (L.) Endl. *Plant Systematics and Evolution* 250: 157–172.*
- Costa, A. C. G. D. 2017. Atributos florais e transição entre polinização abiótica e biótica em espécies de *Rhynchospora* Vahl (Cyperaceae). Phd Thesis. Universidade Federal de Pernambuco, Recife, Brazil.*
- Costa, A. C. G. da, I. Albuquerque, W. Thomas, and I. C. Machado. 2018. Influence of environmental variation on the pollination of the ambophilous sedge *Rhynchospora ciliata* (Cyperaceae). *Plant Ecology*.*
- Costa, A. C. G. D. 2017. Atributos florais e transição entre polinização abiótica e biótica em espécies de *Rhynchospora* Vahl (Cyperaceae). PhD Thesis. Universidade Federal de Pernambuco, Recife. *Cursach, J., and J. Rita. 2012. Reproductive biology of *Ranunculus weyeri* (Ranunculaceae), a narrowly endemic plant from the Balearic Islands with disjunct populations. *Flora - Morphology, Distribution, Functional Ecology of Plants* 207: 726–735.*
- Custodio, T., P. Comtois, and A. C. Araujo. 2017. Reproductive biology and pollination ecology of *Triplaris gardneriana* (Polygonaceae): a case of ambophily in the Brazilian Chaco. *Plant Biology* 19: 504–514.*
- Dafni, A., T. Marom-Levy, A. Jürgens, S. Dötterl, Y. Shimrat, A. Dorchin, H. E. Kirkpatrick, and T. Witt. Ambophily and “super generalism” in *Ceratonia siliqua* (Fabaceae) pollination. *Evolution of Plant-Pollinator Relationships*: 344–373.*
- Duan, Y. W., T. F. Zhang, Y. P. He, and J. Q. Liu. 2009. Insect and wind pollination of an alpine biennial *Aconitum gymnandrum* (Ranunculaceae). *Plant Biology (Stuttgart, Germany)* 11: 796–802.*
- Gan, X., L. Cao, X. Zhang, and H. Li. 2013. Floral biology, breeding system and pollination ecology of an endangered tree *Tetracentron sinense* Oliv. (Trochodendraceae). *Botanical Studies* 54: 50.*

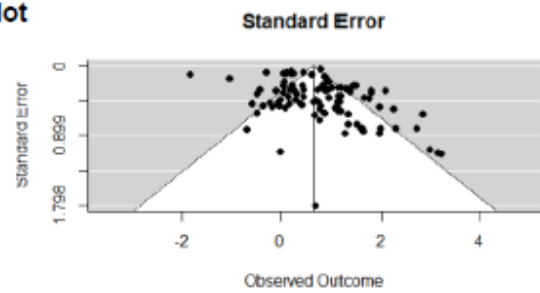
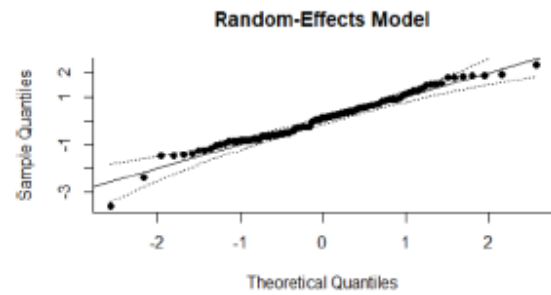
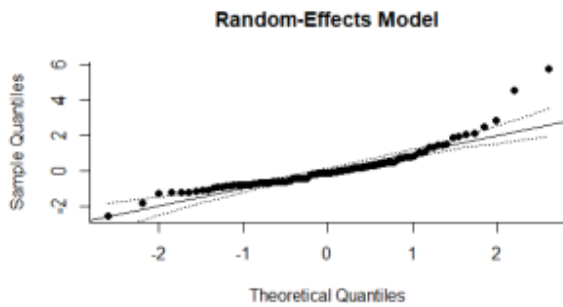
- Gong, Y.-B., M. Yang, J. Vamosi, H.-M. Yang, W.-X. Mu, J. li, and T. Wan. 2016. Wind or insect pollination? Ambophily in a subtropical gymnosperm *Gnetum parvifolium* (Gnetales). *Plant Species Biology* 31: 272–279.
- Goodwillie, C. 1999. Wind Pollination and Reproductive Assurance in *Linanthus parviflorus* (Polemoniaceae), a Self-Incompatible Annual. *American Journal of Botany* 86: 948–954.*
- Gulías, J., and A. Traveset. 2012. Altitudinal variation in the reproductive performance of the Mediterranean shrub *Rhamnus lycioides* L. *Journal of Plant Ecology* 5: 330.*
- Hall, J. A., and G. H. Walter. 2018. Pollination of the Australian cycad *Cycas ophiolitica* (Cycadaceae): the limited role of wind pollination in a cycad with beetle pollinator mutualists, and its ecological significance. *Journal of Tropical Ecology* 34: 121–134.
- Karamsetty, H. J. and Aluri, J. S. R. 2018. Ambophily in the Dioecious Weedy Mangrove Associate, (Euphorbiaceae). *Transylvanian Review of Systematical and Ecological Research* 20: 15–28.
- Karrenberg, S., J. Kollmann, and P. J. Edwards. 2002. Pollen vectors and inflorescence morphology in four species of *Salix*. *Plant Systematics and Evolution* 235: 181–188.*
- Karyamsetty, H., and J. S. R. Aluri. 2018. Ambophily in the Dioecious Weedy Mangrove Associate, *Excoecaria agallocha* (Euphorbiaceae). *Transylvanian Review of Systematical and Ecological Research* 20.*
- Khorsand Rosa, R., and S. Koptur. 2013. New findings on the pollination biology of *Mauritia flexuosa* (Arecaceae) in Roraima, Brazil: linking dioecy, wind, and habitat. *American Journal of Botany* 100: 613–621.*
- Kono, M., and H. Tobe. 2007. Is *Cycas revoluta* (Cycadaceae) wind- or insect-pollinated? *American Journal of Botany* 94: 847–855.
- Lázaro, A., and A. Traveset. 2005. Spatio-temporal variation in the pollination mode of *Buxus balearica* (Buxaceae), an ambophilous and selfing species: mainland-island comparison. *Ecography* 28: 640–652.*
- Mahy, G., J. De Sloover, and A.-L. Jacquemart. 1998. The generalist pollination system and reproductive success of *Calluna vulgaris* in the Upper Ardenne. *Canadian Journal of Botany* 76: 1843.
- Mangla, Y., R. Tandon, Y. Mangla, and R. Tandon. 2011. Insects facilitate wind pollination in pollen-limited *Crateva adansonii* (Capparaceae). *Australian Journal of Botany* 59: 61–69.
- Medan, D., and M. Devoto. 2017. Ambophily, not entomophily: the reproduction of the perennial *Discaria chacaye* (Rhamnaceae: Colletieae) along a rainfall gradient in Patagonia, Argentina. *Plant Systematics and Evolution* 303: 841–851.*
- Medan, D., and M. Devoto. 2005. Reproductive ecology of a perennial outcrosser with a naturally dissected distribution. *Plant Systematics and Evolution* 254: 173–184.*
- Meeuse, A. D. J. 1984. rate of dependence of *Plantago media* L. on entomophilous reproduction — Preliminary Report. *Acta Botanica Neerlandica* 33: 129–130.*
- Norman, J., S. Weller, and A. Sakai. 1997. Pollination biology and outcrossing rates in hermaphroditic *Schiedea lydgatei* (Caryophyllaceae). *American Journal of Botany* 84: 641.*
- Peeters, L., and O. Totland. 2011. Wind to insect pollination ratios and floral traits in five alpine *Salix* species. *Canadian Journal of Botany* 77: 556–563.*
- Pendleton, R. L., E. D. McArthur, and S. C. Sanderson. 2012. Breeding system and interaccessional hybridization of *Purshia tridentata* plants grown in a common garden. *Western North American Naturalist*. 72(2): 241-249.: 241–249.*

- Raju, A. J. S., and D. S. Rani. 2016. Reproductive ecology of *Cleome gynandra* and *Cleome viscosa* (Capparaceae). 14.
- Rios, L. D., E. J. Fuchs, D. R. Hodel, and A. Cascante-Marín. 2014. Neither insects nor wind: ambophily in dioecious *Chamaedorea* palms (Arecaceae). *Plant Biology* (Stuttgart, Germany) 16: 702–710.*
- Sacchi, C. F., and P. W. Price. 1988. Pollination of the Arroyo Willow, *Salix lasiolepis*: Role of Insects and Wind. *American Journal of Botany* 75: 1387–1393.*
- Shi-Jin, L., D. Zhang, Xiang, L. Li, and C. Zhong-Yi. 2004. Pollination Ecology of *Caesalpinia crista* (Leguminosae: Caesalpinioideae). *Acta Botanica Sinica* 46.*
- Totland, Ø., and L. Peeters. 1999. Wind to insect pollination ratios and floral traits in five alpine *Salix* species.*
- Vikas, and R. Tandon. 2011. Reproductive biology of *Azadirachta indica* (Meliaceae), a medicinal tree species from arid zones. *Plant Species Biology* 26: 116–123.
- Yamasaki, E., S. Sakai, E. Yamasaki, and S. Sakai. 2013. Wind and insect pollination (ambophily) of *Mallotus* spp. (Euphorbiaceae) in tropical and temperate forests. *Australian Journal of Botany* 61: 60–66.*
- Yaqoob, U., and I. A. Nawchoo. 2016. Reproductive ecology of an endangered monocarpic herbaceous perennial, *Ferula jaeschkeana* Vatke. *Tropical Ecology*: 849–864.

108 cases

Q-Q plot

99 cases



funnel plot asymmetry: $z = 6.37, p < .0001$
normality: $w = 0.67, p\text{-value} = p < .0001$
overall effect size 0.95 [0.72, 1.17]
heterogeneity: $Q = 1441.44$
 $df = 107, p < .0001$
Rosenberg fail-safe number: 15783
Critical value = 555

funnel plot asymmetry: $z = 3.94, p < .0001$
normality: $w = 0.97, p < .0001$
overall effect size 0.69 [0.53, 0.86]
heterogeneity: $Q = 1113.17,$
 $df = 98, p < .0001$
Rosenberg fail-safe number: 12381
Critical value = 505

Influence plot

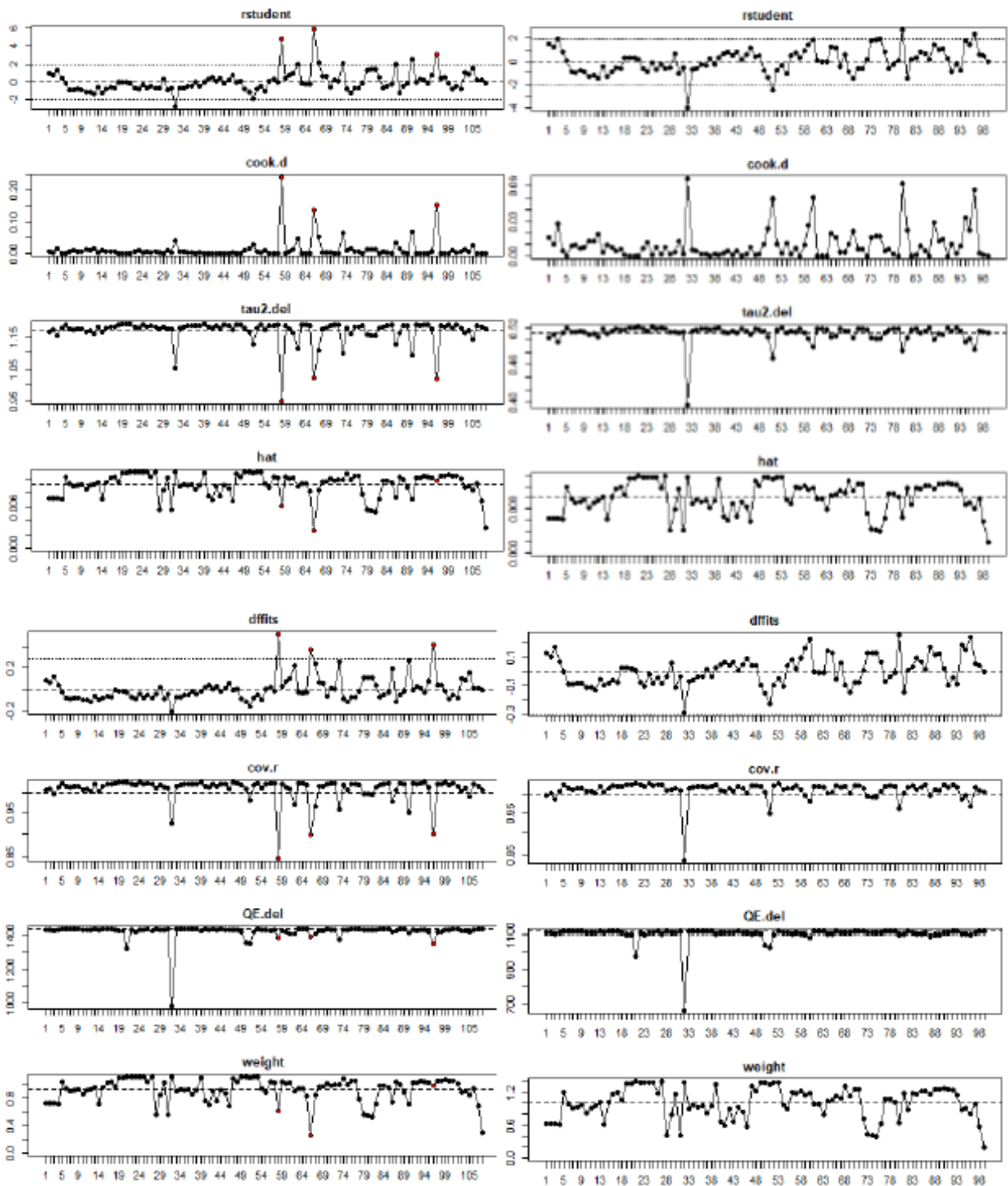


Figure S1: Diagnostics of traditional random-effects models. Diagnostics for model assumptions and publication bias: normal Q-Q plot, funnel plot, influence plot, symmetry test, overall effect size, heterogeneity, Rosenberg fail-safe number, and critical value for the complete dataset (104 cases from 53 species) and without outliers (84 cases from 46 species).

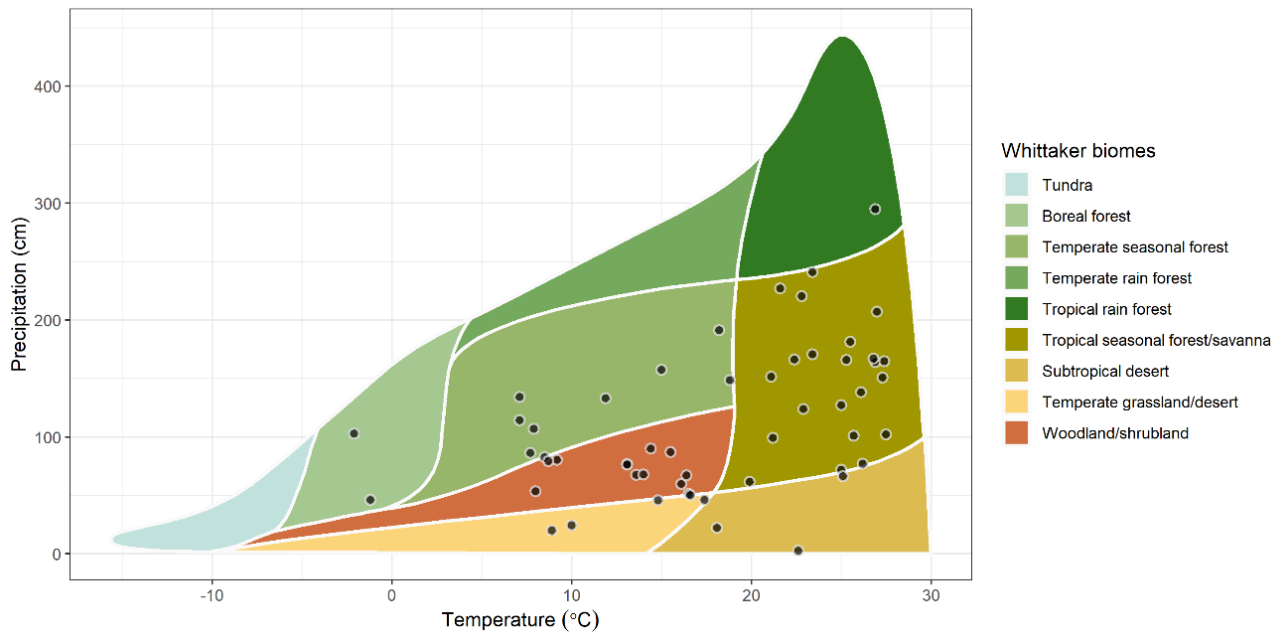


Figure S2: Distribution of case locations across seven of the nine major global terrestrial biomes.

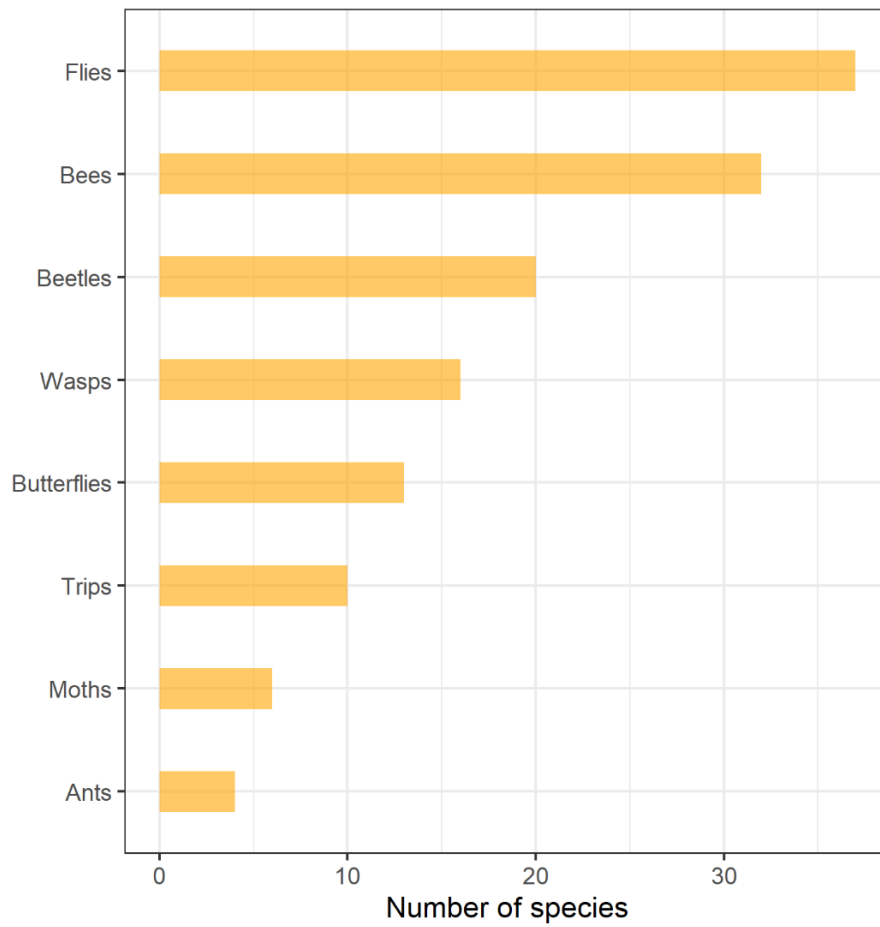


Figure S3: Proportion of pollinator taxonomic groups represented in the animal-pollinated component of the 51 species included in the systematic review.

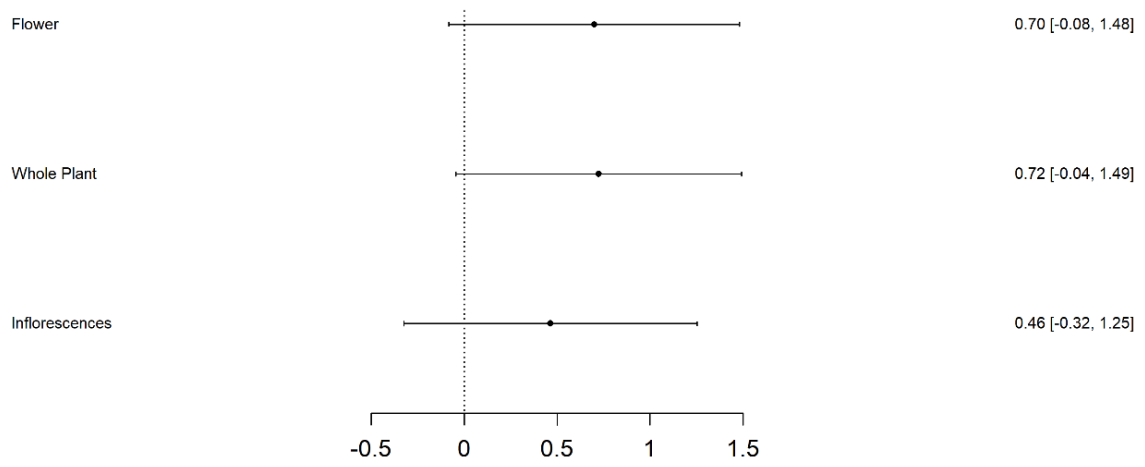


Figure S4: Effect sizes (d) and 95% confidence interval of reproductive success and per category based on unit of sampling in the phylogenetically independent meta-analysis.

Table S1. Results (effect size (g) and 95% confidence interval (95% lower and upper CI)) of traditional and phylogenetically independent meta-analyses based on random-effects models. Heterogeneity between categories (QM), Akaike's information criterion (AICc) per plant trait, number of angiosperm species (n) and p value per category were presented.

	Traditional meta-analysis								Phylogenetically-independent meta-analysis						
	n	QM	p	AICc	g	l.ci	u.ci	p	QM	p	AICc	g	l.ci	u.ci	p
Overall	47	478.46	0.01	177.08					478.46	***	160.76				
					0.64	0.5	0.79	***				0.63	-0.06	1.3	0.1
Plant traits															
Breeding system	6.55	0.03	174.67					0.24		0.885	161.65				
Dioecious	20				0.84	0.63	1.06	<0.0001				0.72	-0.1	1.55	>0.999
Hermaphroditic	21				0.56	0.34	0.78	<0.0001				0.62	-0.14	1.39	>0.999
Monoecious	5				0.35	0.01	0.70	0.05				0.54	-0.36	1.46	>0.999
Mating system	0	0.96	93.45					0.00		0.92	71.23				
Self-compatible	19				0.52	0.31	0.72	<0.0001				0.56	-0.07	1.21	0.10
Self-incompatible	3				0.51	0.06	0.95	0.05				0.53	-0.37	1.44	>0.999
Reproductive cycle	1.13	0.28	178.89					0.23		0.62	162.38				
Annual	15				0.52	0.25	0.79	<0.0001				0.70	-0.05	1.47	>0.999
Perennial	33				0.69	0.53	0.86	<0.0001				0.60	-0.11	1.32	>0.999
Climatic zone	0.54	0.46	177.69					0.21		0.64	162.07				
Extratropical	24				0.69	0.50	0.88	<0.0001				0.57	-0.19	1.33	>0.999
Tropical	23				0.59	0.38	0.81	<0.0001				0.67	-0.05	1.41	0.10

Pollination system	0.84	0.35	138.85					0.04		0.82	120.41				
Specialist	15			0.75	0.48	1.02	<0.0001					0.69	-0.01	1.40	0.10
Generalist	15			0.59	0.37	0.81	<0.0001					0.75	0.03	1.46	0.05
Perianth presence	3.1	0.07	176.88					0.08		0.76	160.99				
Absent	28			0.76	0.57	0.95	<0.0001					0.68	-0.09	1.47	0.1
Present	18			0.50	0.29	0.71	<0.0001					0.56	-0.29	1.42	
Flower symmetry	1.5	0.2	172.54							0.74	149.08				
Zygomorphic	41			0.43	0.02	0.84	0.05	0.10				0.57	-0.37	1.53	
Actinomorphic	3			0.71	0.56	0.56	<0.0001					0.70	0.03	1.37	0.05

Table S2. Effect sizes (g), variance (vg), unit of sampling and floral attributes (reproductive cycle, breeding system, mating system, climatic zone, pollination system, floral symmetry perianth presence) for 56 species included in the systematic review and in the meta-analysis.

id	statistical.level	lat_dec	lon_dec	country	whit	climate	g	vg	seg	species
aav3	flower	6.499	-58.218	Guyana	tropical seasonal forest	tropical	0.754758	0.091	0.302	<i>Astrocaryum vulgare</i>
acm5	inflorescences	9.906	-84.275	Costa Rica	tropical seasonal forest	tropical	1.795023	0.1200	0.346	<i>Chamaedorea macrospadix</i>
acp6	inflorescences	9.888	-83.964	Costa Rica	temperate seasonal forest	extratropical	1.283151	0.0855	0.2925	<i>Chamaedorea pinnatifrons</i>
acp6	inflorescences	9.888	-83.969	Costa Rica	temperate seasonal forest	extratropical	0.910438	0.097	0.312	<i>Chamaedorea pinnatifrons</i>
act7	inflorescences	9.906	-84.275	Costa Rica	tropical seasonal forest	tropical	2.082388	0.100	0.317	<i>Chamaedorea tepejilote</i>
afk2	individuals	34.132	74.8375	India	woodland/shrubland	extratropical	1.74869	0.177	0.420	<i>Ferula jaeschkeana</i>
afk2	individuals	34.132	74.8375	India	woodland/shrubland	extratropical	1.660849	0.172	0.414	<i>Ferula jaeschkeana</i>
afk2	individuals	34.132	74.8375	India	woodland/shrubland	extratropical	1.124602	0.147	0.383	<i>Ferula jaeschkeana</i>
amf8	individuals	3.3558	-61.4297	Brazil	tropical seasonal forest	tropical	0.712486	0.193	0.439	<i>Mauritia flexuosa</i>
asl1	individuals	-35.75	-57.5177	Argentina	woodland/shrubland	extratropical	0.056504	0.118	0.344	<i>Schinus longifolia</i>
bbb12	individuals	39.616	2.98333	Spain	woodland/shrubland	extratropical	-0.04227	0.223	0.472	<i>Buxus balearica</i>
bbb12	individuals	39.616	2.98333	Spain	woodland/shrubland	extratropical	-0.16723	0.273	0.523	<i>Buxus balearica</i>
bbb12	individuals	39.616	2.98333	Spain	woodland/shrubland	extratropical	-0.02491	0.263	0.512	<i>Buxus balearica</i>

bbb12	individuals	39.616	2.98333	Spain	woodland/shrubland	extratropical	-0.044	0.236	0.486	<i>Buxus balearica</i>
bbb12	individuals	39.616	2.98333	Spain	woodland/shrubland	extratropical	-0.478	0.366	0.605	<i>Buxus balearica</i>
bbb12	individuals	39.616	2.98333	Spain	woodland/shrubland	extratropical	-0.350	0.271	0.520	<i>Buxus balearica</i>
bbb12	individuals	36.833	-2.45	Spain	subtropical desert	extratropical	-0.584	0.240	0.490	<i>Buxus balearica</i>
bbb12	individuals	36.833	-2.45	Spain	subtropical desert	extratropical	0.301	0.193	0.439	<i>Buxus balearica</i>
bbb12	individuals	36.833	-2.45	Spain	subtropical desert	extratropical	-0.687	0.675	0.821	<i>Buxus balearica</i>
bbb12	individuals	36.833	-2.45	Spain	subtropical desert	extratropical	-0.0025	0.190	0.436	<i>Buxus balearica</i>
bps11	inflorescences	37	-3.3	Spain	temperate seasonal forest	extratropical	0.0786	0.040	0.201	<i>Ptilotrichum spinosum</i>
cct13	individuals	-35.75	-57.5177	Argentina	woodland/shrubland	extratropical	-0.202	0.224	0.474	<i>Celtis tala</i>
cra73	individuals	-18.183	-43.5667	Brazil	tropical seasonal forest	tropical	0.8131	0.261	0.511	<i>Rhynchospora albiceps</i>
crc19	inflorescences	-8.1166	-34.9833	Brazil	tropical seasonal forest	tropical	0.101	0.288	0.536	<i>Rhynchospora ciliaris</i>
crc19	inflorescences	-8.1166	-34.9833	Brazil	tropical seasonal forest	tropical	0.184	0.233	0.483	<i>Rhynchospora ciliaris</i>
crc19	inflorescences	-8.1166	-34.9833	Brazil	tropical seasonal forest	tropical	0.431	0.254	0.504	<i>Rhynchospora ciliaris</i>
crc19	inflorescences	-8.1166	-34.9833	Brazil	tropical seasonal forest	tropical	0.413	0.243	0.493	<i>Rhynchospora ciliaris</i>
crc19	inflorescences	-8.1167	-34.9833	Brazil	tropical seasonal forest	tropical	0.886	0.368	0.606	<i>Rhynchospora ciliaris</i>

crc19	inflorescences	-8.1167	-34.9833	Brazil	tropical seasonal forest	tropical	0.442	0.2507	0.5007	<i>Rhynchospora ciliaris</i>
crc19	flower	-8.1166	-34.9833	Brazil	tropical seasonal forest	tropical	0.923	0.023	0.151	<i>Rhynchospora ciliaris</i>
crc19	individuals	-6.7166	-35.1333	Brazil	tropical seasonal forest	tropical	1.189	0.338	0.5821	<i>Rhynchospora ciliaris</i>
crc68	individuals	-6.7166	-35.1333	Brazil	tropical seasonal forest	tropical	1.329	0.559	0.748	<i>Rhynchospora cephalotes</i>
cre70	individuals	-18.183	-43.5667	Brazil	tropical seasonal forest	tropical	1.092	0.290	0.538	<i>Rhynchospora elatior</i>
crg69	individuals	-18.183	-43.5667	Brazil	tropical seasonal forest	tropical	1.627	0.697	0.835	<i>Rhynchospora globosa</i>
crp71	individuals	-6.7166	-35.1333	Brazil	tropical seasonal forest	tropical	1.518	0.571	0.755	<i>Rhynchospora pubera</i>
cs115	individuals	21.3113	-157.796	EUA	tropical seasonal forest	tropical	0.986	0.042	0.205	<i>Schiedea lydgatei</i>
eea23	flower	20.296	85.824	India	tropical seasonal forest	tropical	0.256	0.009	0.099	<i>Excoecaria agallocha</i>
eea23	flower	20.296	85.824	India	tropical seasonal forest	tropical	0.059	0.008	0.0924	<i>Excoecaria agallocha</i>
eea23	flower	20.296	85.824	India	tropical seasonal forest	tropical	0.623	0.010	0.104	<i>Excoecaria agallocha</i>
eea23	flower	20.296	85.824	India	tropical seasonal forest	tropical	0.214	0.008	0.090	<i>Excoecaria agallocha</i>
ema24	individuals	34.833	135.83	Japan	temperate seasonal forest	extratropical	0.801	0.480	0.692	<i>Mallotus japonicus</i>
ema24	individuals	34.833	135.83	Japan	temperate seasonal forest	extratropical	1.61	0.644	0.802	<i>Mallotus japonicus</i>
emw25	individuals	4.198	114.04	Malasya	tropical rain forest	tropical	0.164	0.1559	0.394	<i>Mallotus wrayi</i>

emw25	individuals	4.198	114.042	Malasya	tropical rain forest	tropical	0.459073	0.159	0.399	<i>Mallotus wrayi</i>
fcc27	flower	23.1666	112.566	China	tropical seasonal forest	tropical	0.280741	0.1051	0.3243	<i>Caesalpinia crista</i>
fcc27	flower	23.1667	112.566	China	tropical seasonal forest	tropical	0.203135	0.089	0.298	<i>Caesalpinia crista</i>
fcs28	individuals	32.882	35.357	Israel	tropical seasonal forest	tropical	1.126	0.295	0.543	<i>Ceratonia siliqua</i>
pcb73	flower	-8.050	-34.948	Brazil	tropical seasonal forest	tropical	0.460	0.006	0.082	<i>Chloris barbata</i>
plp41	individuals	38.5	-122.32	EUA	woodland/shrubland	extratropical	0.242	0.06	0.25	<i>Linanthus parviflorus</i>
plp41	individuals	38.5	-122.32	EUA	woodland/shrubland	extratropical	0.238	0.06	0.255	<i>Linanthus parviflorus</i>
pmp43	flower	-22.866	-42.816	Brazil	tropical seasonal forest	tropical	0.228	0.004	0.067	<i>Myrsine parvifolia</i>
ppc35	inflorescences	-23.1	46.916	Brazil	tropical seasonal forest	tropical	1.322	0.390	0.624	<i>Peperomia circinnata</i>
ppm40	individuals	50.733	7.749	Germany	woodland/shrubland	extratropical	1.270	0.747	0.864	<i>Plantago media</i>
ppm40	individuals	50.733	7.749	Germany	woodland/shrubland	extratropical	0.710	3.233	1.798	<i>Plantago media</i>
ppm70	flower	-8.272	-35.68	Brazil	tropical seasonal forest	tropical	0.113	0.008	0.094	<i>Paspalum maritimum</i>
ppm70	flower	-8.0503	-34.948	Brazil	tropical seasonal forest	tropical	-0.28183	0.007	0.085	<i>Paspalum maritimum</i>
ppr37	inflorescences	-23.1	46.916	Brazil	tropical seasonal forest	tropical	-0.08601	0.104	0.322	<i>Peperomia rotundifolia</i>
ppu39	inflorescences	-23.1	46.916	Brazil	tropical seasonal forest	tropical	0	1.204	1.097	<i>Peperomia urocarpa</i>

ptg42	flower	-2.019	-57.8675	Brazil	tropical seasonal forest	tropical	1.042	0.0791	0.2814	<i>Triplaris gardneriana</i>
rag44	individuals	37.483	101.2	China	boreal forest	extratropical	2.283	0.633	0.796	<i>Aconitum gymnandrum</i>
rag44	individuals	37.483	101.2	China	boreal forest	extratropical	1.595	0.6522	0.8075	<i>Aconitum gymnandrum</i>
rdc46	flower	-39.570	-71.441	Argentina	temperate seasonal forest	extratropical	0.894	0.0165	0.1287	<i>Discaria chacaye</i>
rdc46	flower	-39.798	-71.212	Argentina	woodland/shrubland	extratropical	0.807	0.0014	0.038	<i>Discaria chacaye</i>
rot47	individuals	-40.815	-71.106	Argentina	woodland/shrubland	extratropical	0.773	0.2181	0.467	<i>Ochetophila trinervis</i>
rot47	individuals	-33	-69.283	Argentina	temperate grassland	extratropical	0.666	0.2139	0.4625	<i>Ochetophila trinervis</i>
rot47	individuals	-9.610	-71.354	Argentina	tropical seasonal forest	tropical	0.68	0.4077	0.6385	<i>Ochetophila trinervis</i>
rrl48	individuals	39.733	2.7	Spain	woodland/shrubland	extratropical	-0.415	0.0937	0.3061	<i>Rhamnus lycioides</i>
rrl48	individuals	39.3833	2.8333	Spain	woodland/shrubland	extratropical	0.480	0.1300	0.3606	<i>Rhamnus lycioides</i>
sjr62	individuals	-35.75	-57.517	Argentina	woodland/shrubland	extratropical	-0.466	0.1298	0.3603	<i>Jodina rhombifolia</i>
ssa51	individuals	46.2	12.966	Italy	temperate seasonal forest	extratropical	0.827	0.0782	0.2797	<i>Salix alba</i>
ssa51	individuals	46.2	12.966	Italy	temperate seasonal forest	extratropical	1.802	0.1020	0.3194	<i>Salix alba</i>
ssd53	individuals	46.2	12.966	Italy	temperate seasonal forest	extratropical	1.42	0.063	0.2517	<i>Salix daphnoides</i>
sse54	individuals	46.2	12.966	Italy	temperate seasonal forest	extratropical	1.484	0.0644	0.2537	<i>Salix eleagnos</i>

sse54	individuals	46.2	12.966	Italy	temperate seasonal forest	extratropical	0.87	0.055	0.23	<i>Salix eleagnos</i>
ssh55	inflorescences	60.533	7.533	Norway	boreal forest	extratropical	0.802	0.310	0.557	<i>Salix herbacea</i>
ssl57	inflorescences	60.533	7.533	Norway	boreal forest	extratropical	0.966	0.094	0.308	<i>Salix lapponum</i>
ssl58	flower	34	-112	USA	woodland/shrubland	extratropical	1.9626	0.749	0.869	<i>Salix lasiolepis</i>
ssm59	inflorescences	60.533	7.533	Norway	boreal forest	extratropical	1.363	0.10	0.326	<i>Salix myrsinites</i>
ssr60	inflorescences	60.533	7.533	Norway	boreal forest	extratropical	1.185	0.076	0.276	<i>Salix reticulata</i>
sst61	individuals	46.2	12.966	Italy	temperate seasonal forest	extratropical	0.060	0.064	0.254	<i>Salix triandra</i>
sst61	individuals	46.2	12.966	Italy	temperate seasonal forest	extratropical	0.455	0.066	0.258	<i>Salix triandra</i>
ttn64	individuals	39.8	2.8	Spain	woodland/shrubland	extratropical	2.257	0.3108	0.5575	<i>Thymelaea myrtifolia</i>
ttn64	individuals	39.8	2.8	Spain	woodland/shrubland	extratropical	1.9648	0.2799	0.5291	<i>Thymelaea myrtifolia</i>
ttn64	individuals	39.616	2.983	Spain	woodland/shrubland	extratropical	1.1652	0.2174	0.4662	<i>Thymelaea myrtifolia</i>

Table S2 – Continuação.

species	breeding	order	family	habits	sexual	color	symmetry	perianth	visits	cycle
<i>Astrocaryum vulgare</i>	AC	Arecales	Arecaceae	Palm	Monoecious	Pale	Radial	Absent	generalists	perennial
<i>Chamaedorea macrospadix</i>	NA	Arecales	Arecaceae	Palm	Dioecious	Pale	Radial	Absent	bimodal	perennial
<i>Chamaedorea pinnatifrons</i>	NA	Arecales	Arecaceae	Palm	Dioecious	Pale	Radial	Absent	bimodal	perennial
<i>Chamaedorea pinnatifrons</i>	NA	Arecales	Arecaceae	Palm	Dioecious	Pale	Radial	Absent	bimodal	perennial
<i>Chamaedorea tepejilote</i>	NA	Arecales	Arecaceae	Palm	Dioecious	Pale	Radial	Absent	especialists	perennial
<i>Ferula jaeschkeana</i>	AC	Apiales	Apiaceae	Herb	Monoecious	Yellow	Radial	Present	generalists	perennial
<i>Ferula jaeschkeana</i>	AC	Apiales	Apiaceae	Herb	Monoecious	Yellow	Radial	Present	generalists	perennial
<i>Ferula jaeschkeana</i>	AC	Apiales	Apiaceae	Herb	Monoecious	Yellow	Radial	Present	generalists	perennial
<i>Mauritia flexuosa</i>	NA	Arecales	Arecaceae	Palm	Dioecious	Pale	Radial	Absent	generalists	perennial
<i>Schinus longifolia</i>	AI	Sapindales	Anacardiaceae	Shrub	Dioecious	Pale	Radial	Present	NA	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Buxus balearica</i>	AC	Buxales	Buxaceae	Shrub	Monoecious	Pale	Radial	Present	generalists	perennial
<i>Ptilotrichum spinosum</i>	AC	Brassicales	Brassicaceae	Shrub	Hermaphroditic	White	Radial	Present	NA	annual
<i>Celtis tala</i>	AC	Rosales	Cannabaceae	Tree	Monoecious	Pale	Radial	Present	NA	annual

<i>Rhynchospora albiceps</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	bimodal	annual
<i>Rhynchospora ciliaris</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Rhynchospora ciliaris</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Rhynchospora ciliaris</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Rhynchospora ciliaris</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Rhynchospora ciliaris</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Rhynchospora ciliaris</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Rhynchospora ciliaris</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Rhynchospora cephalotes</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Rhynchospora elatior</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	generalists	annual
<i>Rhynchospora globosa</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	generalists	annual
<i>Rhynchospora pubera</i>	AC	Poales	Cyperaceae	Herb	Hermaphroditic	Pale	Radial	Absent	especialists	annual
<i>Schiedea lydgatei</i>	AC	Caryophyllales	Caryophyllaceae	Shrub	Hermaphroditic	White	Radial	Present	bimodal	perennial
<i>Excoecaria agallocha</i>	NA	Malpighiales	Euphorbiaceae	Tree	Dioecious	Pale	Bilateral	Absent	generalists	perennial
<i>Excoecaria agallocha</i>	NA	Malpighiales	Euphorbiaceae	Tree	Dioecious	Pale	Bilateral	Absent	generalists	perennial
<i>Excoecaria agallocha</i>	NA	Malpighiales	Euphorbiaceae	Tree	Dioecious	Pale	Bilateral	Absent	generalists	perennial
<i>Excoecaria agallocha</i>	NA	Malpighiales	Euphorbiaceae	Tree	Dioecious	Pale	Bilateral	Absent	generalists	perennial
<i>Mallotus japonicus</i>	NA	Malpighiales	Euphorbiaceae	Tree	Dioecious	Pale	Radial	Absent	generalists	perennial
<i>Mallotus japonicus</i>	NA	Malpighiales	Euphorbiaceae	Tree	Dioecious	Pale	Radial	Absent	generalists	perennial
<i>Mallotus wrayi</i>	NA	Malpighiales	Euphorbiaceae	Tree	Dioecious	Pale	Radial	Absent	generalists	perennial
<i>Mallotus wrayi</i>	NA	Malpighiales	Euphorbiaceae	Tree	Dioecious	Pale	Radial	Absent	generalists	perennial
<i>Caesalpinia crista</i>	AI	Fabales	Fabaceae	Vine	Hermaphroditic	Yellow	Bilateral	Present	generalists	annual
<i>Caesalpinia crista</i>	AI	Fabales	Fabaceae	Vine	Hermaphroditic	Yellow	Bilateral	Present	generalists	annual
<i>Ceratonia siliqua</i>	NA	Fabales	Fabaceae	Tree	Hermaphroditic	Pale	Radial	Present	generalists	annual
<i>Chloris barbata</i>	NA	Poales	Poaceae	Herb	Hermaphroditic	green	NA	Absent	NA	annual
<i>Linanthus parviflorus</i>	NA	Ericales	Polemoniaceae	Herb	Hermaphroditic	White	Radial	Present	NA	annual
<i>Linanthus parviflorus</i>	NA	Ericales	Polemoniaceae	Herb	Hermaphroditic	White	Radial	Present	NA	annual
<i>Myrsine parvifolia</i>	NA	Ericales	Primulaceae	Shrub	Dioecious	White	Radial	Present	NA	annual

<i>Peperomia circinnata</i>	AC	Piperales	Piperaceae	Shrub	Hermaphroditic	Pale	Radial	Absent	especialists	perennial
<i>Plantago media</i>	NA	Lamiales	Plantaginaceae	Herb	Hermaphroditic	Pale	Radial	Present	NA	perennial
<i>Plantago media</i>	NA	Lamiales	Plantaginaceae	Herb	Hermaphroditic	Pale	Radial	Present	NA	perennial
<i>Paspalum maritimum</i>	NA	Poales	Poaceae	Herb	Hermaphroditic	Purple	NA	Absent	especialists	perennial
<i>Paspalum maritimum</i>	NA	Poales	Poaceae	Herb	Hermaphroditic	Purple	NA	Absent	especialists	perennial
<i>Peperomia rotundifolia</i>	AC	Piperales	Piperaceae	Shrub	Hermaphroditic	Pale	Radial	Absent	especialists	perennial
<i>Peperomia urocarpa</i>	AC	Piperales	Piperaceae	Shrub	Hermaphroditic	Pale	Radial	Absent	especialists	perennial
<i>Triplaris gardneriana</i>	NA	Caryophyllales	Polygonaceae	Tree	Monoecious	pink	Radial	Present	generalists	perennial
<i>Aconitum gymnandrum</i>	AC	Ranunculales	Ranunculaceae	Shrub	Hermaphroditic	Purple	Bilateral	Present	especialists	annual
<i>Aconitum gymnandrum</i>	AC	Ranunculales	Ranunculaceae	Shrub	Hermaphroditic	Purple	Bilateral	Present	especialists	annual
<i>Discaria chacaye</i>	AI	Rosales	Rhamnaceae	Shrub	Hermaphroditic	White	Radial	Present	generalists	perennial
<i>Discaria chacaye</i>	AI	Rosales	Rhamnaceae	Shrub	Hermaphroditic	White	Radial	Present	generalists	perennial
<i>Ochetophila trinervis</i>	AC	Rosales	Rhamnaceae	Shrub	Hermaphroditic	Pale	Radial	Present	generalists	perennial
<i>Ochetophila trinervis</i>	AC	Rosales	Rhamnaceae	Shrub	Hermaphroditic	Pale	Radial	Present	generalists	perennial
<i>Ochetophila trinervis</i>	AC	Rosales	Rhamnaceae	Shrub	Hermaphroditic	Pale	Radial	Present	generalists	perennial
<i>Rhamnus lycioides</i>	NA	Rosales	Rhamnaceae	Shrub	Dioecious	Pale	Radial	Present	bimodal	perennial
<i>Rhamnus lycioides</i>	NA	Rosales	Rhamnaceae	Shrub	Dioecious	Pale	Radial	Present	bimodal	perennial
<i>Jodina rhombifolia</i>	AC	Santalales	Cervantesiaceae	Tree	Hermaphroditic	Pale	Radial	Present	NA	annual
<i>Salix alba</i>	NA	Malpighiales	Salicaceae	Tree	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix alba</i>	NA	Malpighiales	Salicaceae	Tree	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix daphnoides</i>	NA	Malpighiales	Salicaceae	Tree	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix eleagnos</i>	NA	Malpighiales	Salicaceae	Shrub	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix eleagnos</i>	NA	Malpighiales	Salicaceae	Shrub	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix herbacea</i>	NA	Malpighiales	Salicaceae	Herb	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix lapponum</i>	NA	Malpighiales	Salicaceae	Shrub	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix lasiolepis</i>	NA	Malpighiales	Salicaceae	Shrub	Dioecious	Pale	Radial	Absent	bimodal	perennial
<i>Salix myrsinites</i>	NA	Malpighiales	Salicaceae	Shrub	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix reticulata</i>	NA	Malpighiales	Salicaceae	Herb	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix triandra</i>	NA	Malpighiales	Salicaceae	Shrub	Dioecious	Pale	Radial	Absent	NA	perennial
<i>Salix triandra</i>	NA	Malpighiales	Salicaceae	Shrub	Dioecious	Pale	Radial	Absent	NA	perennial

<i>Thymelaea myrtifolia</i>	NA	Malvales	Thymelaeaceae	Shrub	Dioecious	Pale	Radial	Present	generalists	perennial
<i>Thymelaea myrtifolia</i>	NA	Malvales	Thymelaeaceae	Shrub	Dioecious	Pale	Radial	Present	generalists	perennial
<i>Thymelaea myrtifolia</i>	NA	Malvales	Thymelaeaceae	Shrub	Dioecious	Pale	Radial	Present	generalists	perennial