

# Theoretical predictions of plant-pollinator interactions in sympatric species of *Psychotria* (Rubiaceae) in Cerrado of Brazil

José N. Mesquita-Neto, Carlos M. Silva-Neto & Edivani V. Franceschinelli\*

Laboratório de Biologia Reprodutiva de Plantas, Departamento de Botânica, Instituto de Ciências Biológicas, Universidade Federal de Goiás, 74001-970, Goiânia GO, Brazil

\*Author for correspondence: [edivanif@gmail.com](mailto:edivanif@gmail.com)

**Background and aims** – In highly seasonal habitats, sympatric species are often constrained to flower simultaneously. Many sympatric species of *Psychotria* have similar floral traits, are pollinated by bees, moths, butterflies and flies and flower at the same time of year. This genus provides an opportunity to analyse the relationships between sympatric congeneric plants and pollinators. Interactions between potentially sympatric *Psychotria* species and their pollinators were surveyed to assess the occurrence of sharing, specialisation, or generalisation of pollinators and plants in the system.

**Methods** – A dataset compiled from studies of pollination biology of this genus was used to investigate aspects of interactions with shared pollinators and flowering overlap. These data were used to draw a bipartite graph and analyse the interaction network and degree of niche overlap.

**Key results** – In total, the dataset included eight species of potentially co-occurring *Psychotria* species, which interacted with 24 species of pollinators. The most generalised plant in the network was *Psychotria trichophoroides*. The pollinators with the highest number of links, and therefore the most important for the network, belonged to orders Hymenoptera and Lepidoptera. Pollinators were shared among *Psychotria* species, thus pollination niches did overlap ( $\theta = 0.20$ ,  $P_{\text{random} \geq \text{observed}} = 0.04$ ), and plants of this genus depended on pollinators that were generalists among them.

**Conclusion** – The peak of overlap in flowering phenology, when all of the species may potentially co-flower, coincided with the Cerrado rainy season. Since all *Psychotria* species occurred in sympatry, overlapped in flowering phenology, and had asymmetric interactions with floral visitors, it is possible that pollination facilitation is present in this system.

**Key words** – Brazilian savannah, floral phenology, network biology, niche overlap, pollination, sympatry.

## INTRODUCTION

Pollinators are essential for the reproduction of at least 90% of forest plant species (Bawa 1990). In addition, they are crucial to maintain food chains in natural environments, because they enable the production of fruits and seeds that sustain frugivores (Moreti et al. 2006). Given the evidence for a decline in pollinator numbers, particularly bees, it is especially important to study the relationships between native plant species and their potential pollinators to inform conservation and management strategies.

In highly seasonal habitats, sympatric species are often constrained to flower simultaneously (Janzen 1967, Reich & Borchert 1984, Rathcke 1988, Ollerton et al. 2003). Under these circumstances, plants may become more tolerant of competition by lengthening flower longevity (e.g. Levin 1978, Motten 1986, Rathcke 1988, Ashman & Schoen 1994,

Ashman 2000) or through a finer-grained partitioning of pollinator activity, for example throughout the day (Levin & Anderson 1970, Ollerton & Lack 1992). On the other hand, convergence of floral morphology among plants of different species that flower simultaneously may be a mutually beneficial strategy to attract pollinators (Bobisud & Neuhaus 1975, Schemske 1981), since the concentration of floral resources increases flower visitation rates (Augspurger 1980, de Jong et al. 1992, Podolsky 1992).

Many sympatric species of *Psychotria* have similar floral traits and flower at the same time of year (Hamilton 1989, Sakai & Wright 2008). Flowers in this genus are small, have a tubular corolla, whitish colouration and are pollinated by bees, moths, butterflies and flies (Almeida & Alves 2000, Coelho & Barbosa 2004). In addition, they have a number of sexual mechanisms in common, including hermaphroditism,

distyly (heteromorphic incompatibility system), and functional unisexuality (Nepokroeff et al. 1999).

*Psychotria* L. (Rubiaceae) is one of the largest angiosperm genera, including approximately 1650 species (Hamilton 1989). This genus is comprised predominantly of shrubs or treelets (Carvalho et al. 2000), which are common in the understory of tropical forests (Taylor 1996, Orians 1997, Lopes & Buzato 2005) and are important for ecosystem function (Lopes & Buzato 2005). Therefore, *Psychotria* provides a unique opportunity to analyse the relationships between plants and pollinators of sympatric congeneric species that flower synchronously (Sakai & Wright 2008, Liuth et al. 2013). In this study, interactions between potentially sympatric *Psychotria* species and their pollinators were analysed to assess the occurrence of pollinators sharing, and specialisation or generalisation in plant-pollinator interactions. In our analysis, we included interactions with shared pollinators and overlap in flowering phenology, based on a dataset compiled from studies of pollination biology of genus *Psychotria* in Cerrado.

### METHODS

Our dataset was compiled from studies with *Psychotria* species found in south-eastern Goiás State and western Minas Gerais State, Brazil, within the Cerrado domain (table 1). To minimize errors due to non-overlapping spatial distributions of the *Psychotria* species and their pollinators, only data from studies up to 200 km apart and located within the same river basin were included (<https://docs.google.com/file/d/0ByQNaWHmeZ15b21zYW5kUHA2cGs/edit>).

The survey included secondary data from scientific papers, theses and dissertations that had a list of pollinators for different *Psychotria* species (table 1). The studies analysed mostly focused on plant biology and reproductive ecology. These studies were selected because they deliberately distinguished nectar robbers from effective pollinators. Furthermore, they included one or a few plant species, resulting in a higher sampling effort and number of observations by species in comparison to studies of large communities (Olesen & Jordano 2002, Waser 1986). Altogether, five studies were included in the dataset, for a total of eight species studied. Only studies that had species-level identification of pollinators were included, and nectar robbers were excluded.

Presence and absence of *Psychotria*-pollinator interactions were tabulated in a matrix with plants in rows and visitors in columns (Jordano 1987). Figures and network analyses were based on this data matrix. In addition, the flowering periods of the species included in the matrix were analysed to determine the degree of phenological overlap among the *Psychotria* species. Reproductive phenology data were obtained from the same studies used to analyse plant-pollinator interactions (table 1). The presence or absence of the flowering phenophase of each species was recorded to obtain the proportion of species flowering in a given time of year. This method is qualitative, but it allows the assessment of synchrony among species, given that synchrony increases with the number of species flowering simultaneously.

**Table 1 – List of studies on reproductive biology of *Psychotria* species in Cerrado (Brazilian Savanna).**

Authors	Year	Method†	Municipality (state)	Species	N° of studied plants‡	Size area	Sampling effort per specie	Day time	Phenology duration	Phenology frequency
Coelho & Barbosa	2004	fi; pr	Uberlândia (MG)	<i>P. poeppigiana</i> Müll.Arg.	450(f)	7.78 ha.	60 h	06–18 h	Jun. 2000–May 2001	monthly
Consolaro et al.	2011	fi	Uberlândia (MG)	<i>P. carthagensis</i> Jacq. <i>P. capitata</i> Ruiz & Pav.; <i>P. deflexa</i> DC.; <i>P. hoffmannseggiana</i> (Willd. ex Schult.) Müll.Arg.; <i>P. prunifolia</i> (Kunth) Steyerf.; <i>P. trichophoroides</i> Müll.Arg.	101(f) 101(p)	404 ha.	17 h	06–18h	Aug. 2002–Sep. 2003	monthly
Sá	2013	fi	Catalão (GO)	<i>P. prunifolia</i> ; <i>P. hoffmannseggiana</i> ; <i>P. gracilentata</i> Müll.Arg.; <i>P. trichophoroides</i>	40(f)	28.48; 54 ha.	24 h	06–18h	Jan. 2012–Dec. 2012	fortnightly
Mesquita-Neto	2013	fi	Catalão (GO)	<i>P. capitata</i> ; <i>P. deflexa</i> ; <i>P. hoffmannseggiana</i> ; <i>P. prunifolia</i>	20(f) 20(p)	28.48 ha.	12 h	06–18h	Nov. 2012–Jan. 2013	Weekly
Oliveira	2008	fi	Araguari (MG)	<i>P. gracilentata</i> Müll.Arg.; <i>P. trichophoroides</i>	20(f)	11 ha.	10 h	-	Mar. 2006–Feb. 2008	monthly

Graphic representations and matrix indexing were generated using the bipartite package in R 2.11.0 (Dormann et al. 2008, Dormann et al. 2010). Networks were described based on the following properties: (1) number of nodes: total number of plant and animal species involved in the interaction network; (2) number of links (k): observed number of interactions between plants and pollinators; (3) connectance (C): proportion of observed interactions, i.e., a ratio between the total number of actual links (E) and the maximum possible number of links, which is given by the product of the number of plants (P) and animals (A) in the network:  $C = E/(A * P)$ .

The qualitative data matrix used for the network analysis was also used to evaluate overlap in the pollination niches of the eight *Psychotria* species. Pianka's index ( $\theta$ ) was used to estimate similarity among the pollinator compositions of each plant species (Pianka 1973). This index ranges from 0 (total segregation) to 1 (total overlap) (Gotelli & Graves 1996). The observed overlap index was then compared to a distribution of values obtained by randomizing the original matrix 1000 times using the RA2 algorithm of the software Ecosim 7 (Gotelli & Entsminger 2006).

## RESULTS

Eight species of *Psychotria* from the pre-defined geographic region were included. These species interacted with 24 species of pollinators, establishing a total of 43 interactions with an average of 5.4 links per plant and 1.8 per pollinator. Fourteen of the visitors interacted with only one *Psychotria* species. In contrast, all plant species were visited by at least two different pollinators, revealing a lack of extreme specialisation in *Psychotria* interactions (fig. 1).

The connectance of the network ( $C = 0.22 = 22\%$ ) was higher than observed in other pollination studies (average of 11% according to Olesen & Jordano 2002). The cumulative distribution of the number of links per species (total number of species with k interactions) revealed a dominance of plants with three or more interactions (fig. 2A). The most generalist plant in the network was *P. trichophoroides* Müll. Arg. ( $k = 13$ ), and the pollinators that interacted with the largest number of *Psychotria* species were *Apis mellifera* L. and the butterfly species *Ithomia agnosia* Hewitson and *Pythonides jovianus* Staudinger (fig. 1). The pollinators with the largest number of links, thus the most important for the maintenance of the plants, belonged to orders Hymenoptera and Lepidoptera (figs 1 & 2B). However, bird pollination did occur in *P. poeppigiana* Müll. Arg., which was visited by hummingbirds (fig. 1).

The pollination niches of the eight *Psychotria* species overlapped more than expected based on the null model ( $\theta = 0.20$ ,  $P_{\text{random} \geq \text{observed}} = 0.04$ ). As a result, the plants had more pollinators in common than expected by chance.

Species of *Psychotria* flower throughout the year, but the peak of their flowering period occurred between the months of October and January, coinciding with the period of largest accumulation of rainfall in the Cerrado (fig. 3). The peak of co-flowering occurred in the months of November and December, when all the eight species analysed had flowers (fig. 2). These data demonstrate that the flowering phenol-

ogy of the *Psychotria* species included here did overlap in the rainy season. All species had their flowering period monitored year around and there was no lack of data collection during monitoring period, so the absence of this phenophase indicates that the species were not in flower (table 1).

## DISCUSSION

Rainfall seems to be a predominant factor in determining the flowering period of *Psychotria* species (Liuth et al. 2013). For instance, *P. hoffmannseggiana* (Willd. ex Schult.) Müll. Arg. flowers at different times of year in different regions of Brazil, but always in periods of elevated rainfall (Oliveira 2008, Teixeira & Machado 2004). *Psychotria* species that flower during the rainy season are generally pollinated by insects (Castro & Oliveira 2002, Coelho & Barbosa 2004, Consolaro et al. 2011, Sá 2013, Lopes & Buzato 2005, Mesquita-Neto 2013, Oliveira 2008, Ramos & Santos 2006, Teixeira & Machado 2004).

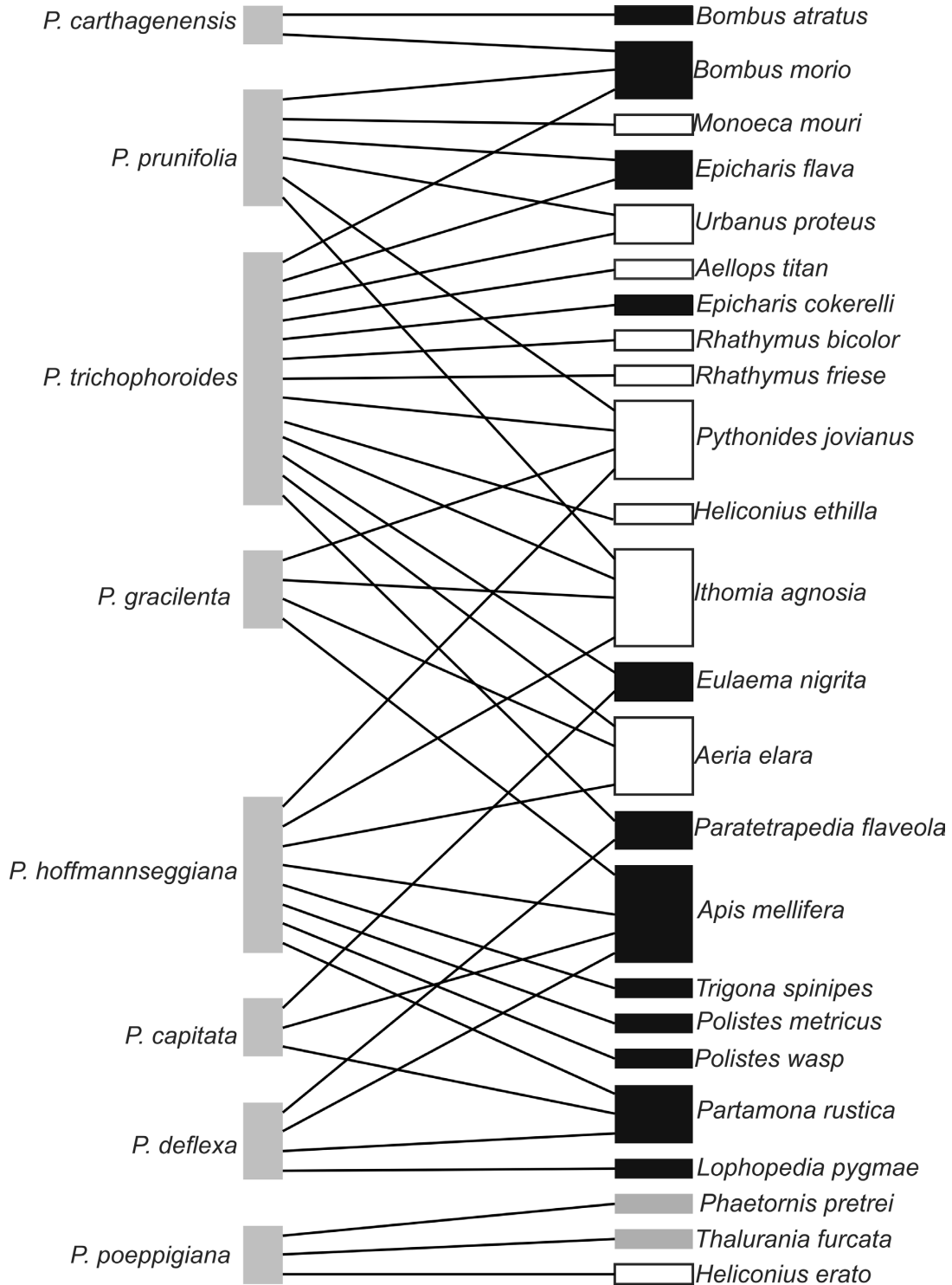
The rainy season is also the time of highest abundance and activity of Lepidoptera and Hymenoptera (Fonseca et al. 2006, Silva et al. 2011, Wolda 1988), the most common visitors of *Psychotria* flowers. Thus, the overlap in flowering of *Psychotria* species during the rainy season may be an adaptation to improved conditions for pollination, that is, species tend to flower when vector availability is higher, as suggested for other plant groups (Almeida & Alves 2000, Koptur et al. 1988, Martin-Gajardo & Morellato 2003). However, water availability may also influence the phenological pattern of *Psychotria*, because this genus includes predominantly herbaceous species with shallow roots. This morphological trait limits access to water to the most superficial layer of the soil, resulting in greater dependency of the reproductive cycle on the rainfall pattern (Batalha & Mantovani 2000).

The phenological analysis of flowering revealed the existence of synchrony among species, suggesting that the system proposed here is temporally viable through co-flowering of *Psychotria* species. Sakai & Wright (2008) highlighted that it is uncommon to find as many congeneric species with overlapping distributions as in the genus *Psychotria*, which may have more than twenty co-occurring species in a given tropical forest. The *Psychotria* species included here overlapped spatially, as well as with their pollinators, and flowered synchronously. As such, the interactions between *Psychotria* and pollinators presented here are likely to be consistent with the actual system.

The overlap of the *Psychotria* pollination niches suggests that there is pollinator partition, which points to the occurrence of a generalised pollination system within the group of species. Given the importance of pollinators for plant reproduction, plant species with generalised pollination systems are expected to be less vulnerable to extinction than plants with specialised systems (Harris & Johnson 2004). The genus *Psychotria* is predominantly heteromorphic and self-incompatible, meaning that fruit formation can only occur through crosses between long-styled and short-styled flowers. Heterostylous flowers are mostly pollinated by bees, but may also be pollinated by many other animal groups, such as flies, moths, butterflies and hummingbirds (Rodrigues & Consolaro 2013, Castro & Oliveira 2002, Ganders 1979,

Richards & Koptur 1993). *Psychotria* species are pollinated more efficiently by lepidopterans and bees (Castro & Oliveira 2002, Coelho & Barbosa 2004, Consolaro et al. 2011, Sá 2013, Lopes & Buzato 2005, Mesquita-Neto 2013, Oliveira 2008, Ramos & Santos 2006, Teixeira & Machado 2004). Some authors suggested that insects with longer mouthparts,

such as lepidopterans and long-tongued bees, were more efficient in pollinating short-styled flowers (Beach & Bawa 1980, Lloyd & Webb 1992). In contrast, short-tongued hymenopterans, especially bees, should be more efficient pollinators of long-styled flowers (Beach & Bawa 1980). Pollen transfer from long stamens to long styles is facilitated by the



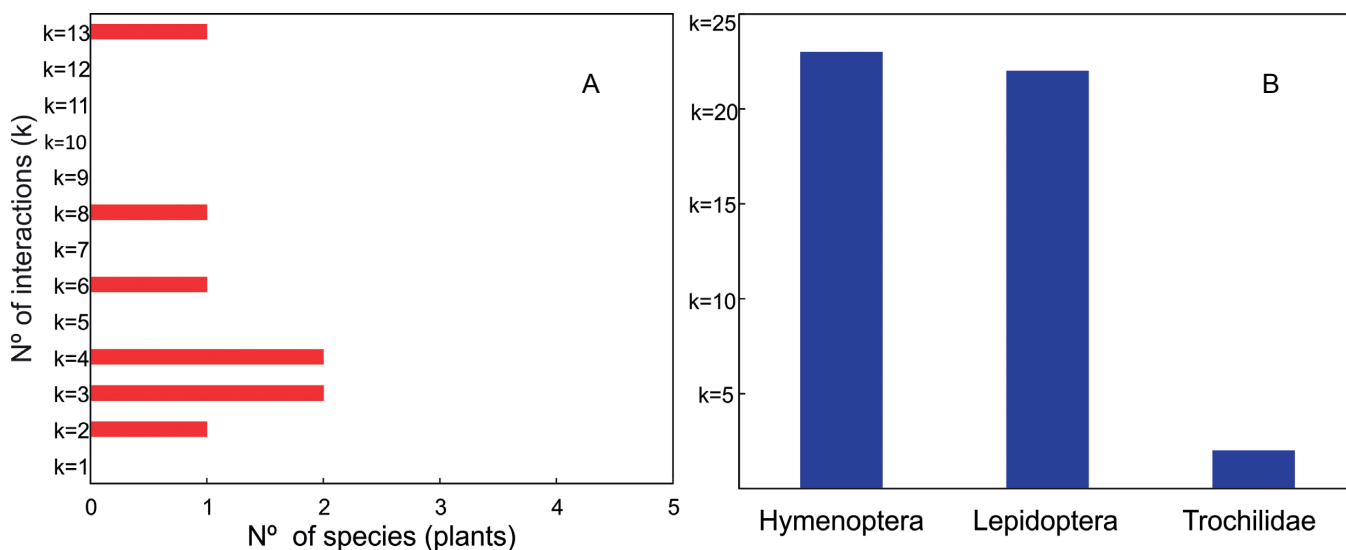
**Figure 1** – Bipartite graph including the eight co-occurring *Psychotria* species and their interactions with pollinators. Legend for pollinator nodes: grey: hummingbirds; white: butterflies; black: bees.

interaction of a much larger range of visitors with those reproductive organs (Pailler & Thompson 1997). As a result, it is necessary to have a pollination system involving more generalised pollinators, at least for plant species of the same genus, as found here.

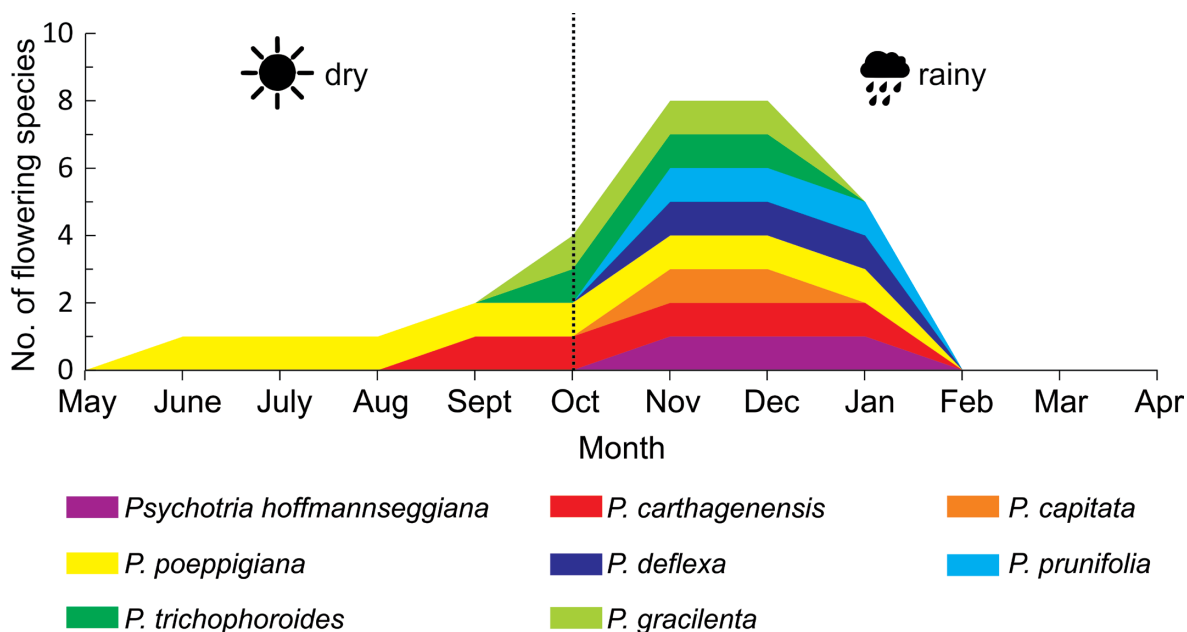
Lepidopterans and bees shared a prominent role in the pollination of *Psychotria*. *Ithomia agnosia* and *P. jovanus* were the main native pollinators of this genus. Butterflies and moths are somewhat constant and faithful to certain plant species (Fonseca et al. 2006, Goulson & Cory 1993, Goulson et al. 1997, Lewis 1989). The presence of large bracts in inflorescences may facilitate visitation by butterflies, which

use these structures as landing and support platforms during nectar removal (Coelho & Barbosa 2004).

Exclusive pollination by Lepidoptera does not ensure the maintenance of reproductive success of both morphotypes and the isoplethy of heterostylous *Psychotria* species. As a consequence, bee pollination is particularly relevant for *Psychotria*. In fact, they had the highest visitor richness and the largest proportion of interactions with this genus. Bees had a significant role among the exclusive pollinators of this system, since they were responsible for most of the exclusive interactions. These bees may have a prominent role in the pollination of plants with overlapping flowering phenology,



**Figure 2** – Number of observed interactions (k) between *Psychotria* species and pollinators. The panels show: A, the number of links per plant species, with a large number of species with few links and a few highly connected species; B, the number of links per pollinator group, with Hymenoptera and Lepidoptera having the largest number of interactions.



**Figure 3** – Flowering phenology of eight co-occurring *Psychotria* species, illustrating the overlap in flowering phenology during the rainy season in the Cerrado biome.

since they are faithful to their resource and are thus less likely to deposit heterospecific pollen on stigmas (Raine et al. 2007, Waser 1986). Our data show the relative importance of bees as effective pollinators of *Psychotria*.

Among the bee species with the largest number of interactions, *A. mellifera* was the most important. This species is broadly considered a super generalist and had a large role in pollinating the *Psychotria* species analysed here. This exotic bee was the most common visitor of *P. hoffmannseggiana*, *P. capitata* and *P. carthagenensis* (Sá 2013, Mesquita-Neto 2013, Faria 2010, Oliveira 2008) and was reported as a pollinator of *P. gracilentata* (Oliveira 2008). Other studies, however, have not found *A. mellifera* to be an effective pollinator of genus *Psychotria* (Almeida & Alves, 2000, Castro & Araujo 2004, Castro & Oliveira 2002, Coelho & Barbosa 2004, Teixeira & Machado 2004). Thus, *A. mellifera* is likely to have occupied a pollination niche of *Psychotria* that was empty or had little competition. In addition, habitat fragmentation and human disturbance may also have contributed for changes in this scenario. Santos et al. (2012) demonstrated that the generalist nature of *A. mellifera* could have a negative impact on an ecological network, interfering on network structure and monopolizing many interactions, often without pollinating effectively (e.g. Carmo et al. 2004). Thus, a more refined approach is necessary in future investigations of the pollination efficiency of *A. mellifera* in comparison to native pollinators. In particular, studies that use single-visit tests and quantification of interspecific and intraspecific pollen load on the body of floral visitors are needed. However, all studies included here focus on the behaviour of the floral visitors, treating all visitors that contacted anthers and stigma as pollinators.

Besides, the reliability of these analyses is limited by the resolution of taxonomic studies; the pollinators unidentified at species level were not included in our analysis. *P. carthagenensis*, for instance, is pollinated by a much larger spectrum of secondary pollinators, but without identification (Faria 2010, Consolaro et al. 2011); this could interfere with our considerations. Then, we adopted a requirement that the main pollinators of *Psychotria* included here possessed species identification. Unfortunately, the taxonomy of certain taxa is limited, especially in the tropics, where there is a lack of specialists, identification keys, bibliography and many species have still not been described (Novotny et al. 2007). Another factor that can influence our analyses is the large size of the study area and the different sampling effort of each study. This is particularly important for species included in only one study (*P. poeppigiana*, *P. gracilentata* and *P. carthagenensis*). However, most species appear in more than one study, which contributes to the reduction of these interferences. Thus, future studies should give more attention and care to the correct identification of suitable pollinators and deposit of vouchers in entomological collections.

The *Psychotria* species included here can be considered synchronopatric, since they co-occur within a limited geographic region and overlap considerably in flowering phenology during the rainy season in Cerrado. In addition, the pollination niches of these species overlap, and they are predominantly pollinated by bees and diurnal butterflies. The connectance of the network was higher than the average of

previous pollination studies (Olesen & Jordano 2002), characterising generalisation of the interactions. However, most systems analysed by other pollination networks are more extensive and involve more complex communities, which include a larger number of species.

Our data suggest that interactions between congeners and their pollinators tend to be generalist. The trend for asymmetric plant-pollinator networks (Vázquez & Aizen 2004), on the other hand, implies that there are some specialist species. Bascompte et al. (2003) and Vázquez & Aizen (2004) suggest that asymmetric specialisation is common in plant-pollinator interaction networks, and that it is more frequent than expected based on a null model. Hence, it is possible that these *Psychotria* species are involved in a process of pollination facilitation; because they frequently occur in sympatry, overlap in flowering, and interact asymmetrically with pollinators, leading to overlap in pollination niches. In facilitation, the increased availability of floral resources around an individual has a positive influence on visitation rates, pollinator diversity, pollination levels and reproductive success (Brown & Kodric-Brown 1979, Callaway 1995, Feinsinger et al. 1991, Waser & Real 1979). Therefore, facilitation should be particularly important for specialised plants that depend exclusively on one or a few generalist pollinators, as is the case for some of the *Psychotria* species included in our study. However, few studies address facilitation of pollination empirically (e.g. Ghazoul 2006). Given this scarcity, our research group is currently carrying out experiments to test the occurrence of facilitation among sympatric *Psychotria* species.

#### ACKNOWLEDGEMENTS

We thank Helder N. Consolaro and Mário Almeida-Neto for the manuscript contributions, the CNPq for financial support (Chamada Universal-MCTI/CNPQ N° 14/2012) and CAPES for the master scholarships provided to the first and second authors.

#### REFERENCES

- Almeida E.M., Alves M.A. (2000) Fenologia de *Psychotria nuda* e *P. brasiliensis* (Rubiaceae) em uma área de Floresta Atlântica no sudeste do Brasil. *Acta Botanica Brasilica* 14: 335–346. <http://dx.doi.org/10.1590/s0102-3306200000300010>
- Ashman T.L. (2000) Pollinator selectivity and its implications for the evolution of dioecy and sexual dimorphism. *Ecology* 81: 2577–2591. <http://dx.doi.org/10.2307/177476>
- Ashman T.L., Schoen D.J. (1994) How long should flowers live? *Nature* 371: 788–791. <http://dx.doi.org/10.1038/371788a0>
- Augsburger C.K. (1980) Mass flowering of a tropical shrub (*Hybanthus prunifolius*): influence on pollinator attraction and movement. *Evolution* 34: 475–488. <http://dx.doi.org/10.2307/2408217>
- Bascompte J., Jordano P., Melián C.J., Olesen J.M. (2003) The nested assembly of plant-animal mutualistic networks. *Proceedings of the National Academy of Sciences of the United States of America* 100: 9383–9387. <http://dx.doi.org/10.1073/pnas.1633576100>
- Batalha M.A., Mantovani W. (2000) Reproductive phenological patterns of cerrado plant species at the Pe-de-Gigante re-

- serve (Santa Rita do Passa Quatro, SP, Brazil): a comparison between the herbaceous and woody floras. *Revista Brasileira de Biologia* 60: 129–145. <http://dx.doi.org/10.1590/s0034-7108200000100016>
- Bawa K.S. (1990) Plant-pollinator interactions in tropical rain forests. *Annual Review of Ecology and Systematics* 21: 399–422. <http://dx.doi.org/10.1146/annurev.ecolsys.21.1.399>
- Beach J.H., Bawa K.S. (1980) Role of pollinators in the evolution of dioecy from distyly. *Evolution* 34: 1138–1142. <http://dx.doi.org/10.2307/2408294>
- Bobisud L., Neuhaus R. (1975) Pollinator constancy and survival of rare species. *Oecologia* 21: 263–272. <http://dx.doi.org/10.1007/BF02404676>
- Brown J.H., Kodric-Brown A. (1979) Convergence, competition, and mimicry in a temperate community of hummingbird-pollinated flowers. *Ecology* 60: 1022–1035. <http://dx.doi.org/10.2307/1936870>
- Callaway R.M. (1995) Positive interactions among plants. *The Botanical Review* 61: 306–349. <http://dx.doi.org/10.1007/bf02912621>
- Carmo R.M., Franceschinelli E.V., dSilveira F.A. (2004) Introduced honeybees (*Apis mellifera*) reduce pollination success without affecting the floral resource taken by native pollinators. *Biotropica* 36: 371–376. <http://dx.doi.org/10.1111/j.1744-7429.2004.tb00329.x>
- Carvalho L.M.T., Fontes M.A.L., Oliveira Filho A.T. (2000) Tree species distribution in canopy gaps and mature forest in an area of cloud forest of the Ibitipoca range, south-eastern Brazil. *Plant Ecology* 149: 9–22. <http://dx.doi.org/10.1023/A:1009836810707>
- Castro C.C., Araujo A.C. (2004) Distyly and sequential pollinators of *Psychotria nuda* (Rubiaceae) in the Atlantic rain forest, Brazil. *Plant Systematics and Evolution* 244: 131–139. <http://dx.doi.org/10.1007/s00606-003-0036-8>
- Castro C.C., Oliveira P.E. (2002) Pollination biology of distylous Rubiaceae in the Atlantic Rain Forest, SE Brazil. *Plant Biology* 4: 640–646. <http://dx.doi.org/10.1055/s-2002-35433>
- Coelho C.P., Barbosa, A.A.A. (2004) Biologia reprodutiva de *Psychotria poeppigiana* Muell. Arg. (Rubiaceae) em mata de galeria. *Acta Botanica Brasilica* 18: 481–489. <http://dx.doi.org/10.1590/s0102-33062004000300008>
- Consolaro H., Silva S.C.S., Oliveira P.E. (2011) Breakdown of distyly and pin-monomorphism in *Psychotria carthagenensis* Jacq. (Rubiaceae). *Plant Species Biology* 26: 24–32. <http://dx.doi.org/10.1111/j.1442-1984.2010.00300.x>
- de Jong T.J., Klinkhamer P.G.L., van Staalduinen M.J. (1992) The consequences of pollination biology for selection of mass or extended blooming. *Functional Ecology* 6: 606–615. <http://dx.doi.org/10.2307/2390059>
- Dormann C.F., Fründ J., Blüthgen N., Gruber B. (2010) Indices, graphs and nullmodels: analyzing bipartite ecological networks. *The Open Ecology Journal* 2: 7–24. <http://dx.doi.org/10.2174/1874213000902010007>
- Dormann C.F., Gruber B., Fründ J. (2008) Introducing the bipartite Package: Analysing Ecological Networks. *R news* 8: 8–11.
- Faria R.R. (2010) Fenologia de floração, sistema reprodutivo e efetividade de polinização da espécie distílica *Psychotria carthagenensis* Jacq. (Rubiaceae) em áreas de Cerrado. Ph.D. thesis, Universidade Federal do Mato Grosso do Sul, Campo Grande, Mato Grosso do Sul.
- Feinsinger P., Tiebout Iii H.M., Young B.E. (1991) Do tropical bird-pollinated plants exhibit density-dependent interaction? Field experiments. *Ecology* 72: 1953–1963. <http://dx.doi.org/10.2307/1941550>
- Fonseca N.G., Kumagai A.F., Mielke O.H.H. (2006) Lepidópteros visitantes florais de *Stachytarpheta cayennensis* (Rich.) Vahl (Verbenaceae) em remanescente de Mata Atlântica, Minas Gerais, Brasil. *Revista Brasileira de Entomologia* 50: 399–405. <http://dx.doi.org/10.1590/s0085-56262006000300010>
- Ganders F.R. (1979) The biology of heterostyly. *New Zealand Journal of Botany* 17: 607–635. <http://dx.doi.org/10.1080/0028825x.1979.10432574>
- Ghazoul J. (2006) Floral diversity and the facilitation of pollination. *Journal of Ecology* 94: 295–304. <http://dx.doi.org/10.1111/j.1365-2745.2006.01098.x>
- Goulson D., Cory J.S. (1993) Flower constancy and learning in foraging preferences of the green-veined white butterfly *Pieris napi*. *Ecological entomology* 18: 315–320. <http://dx.doi.org/10.1111/j.1365-2311.1993.tb01107.x>
- Goulson D., Ollerton J., Sluman C. (1997) Foraging strategies in the small skipper butterfly, *Thymelicus flavus*: when to switch? *Animal behaviour* 53: 1009–1016. <http://dx.doi.org/10.1006/anbe.1996.0390>
- Gotelli N.J., Entsminger G.L. (2006) EcoSim: null models software for ecology. Version 7. Available from <http://www.garyentsminger.com/ecosim/> [accessed 16 Feb. 2015].
- Gotelli N.J., Graves G.R. (1996) Null models in ecology. Washington, Smithsonian Institution Press.
- Hamilton C.W. (1989) A revision of mesoamerican *Psychotria* subgenus *Psychotria* (Rubiaceae), part I: introduction and species 1–16. *Annals of the Missouri Botanical Garden* 76: 67–111. <http://dx.doi.org/10.2307/2399343>
- Harris L.F., Johnson S.D. (2004) The consequences of habitat fragmentation for plant pollinator mutualisms. *International Journal of Tropical Insect Science* 24: 29–43. <http://dx.doi.org/10.1079/ijt20049>
- Janzen D.H. (1967) Synchronization of sexual reproduction of trees within the dry season in Central America. *Evolution* 21: 620–637. <http://dx.doi.org/10.2307/2406621>
- Jordano P. (1987) Patterns of mutualistic interactions in pollination and seed dispersal: connectance, dependence asymmetries, and coevolution. *The American Naturalist* 129: 657–677. <http://dx.doi.org/10.1086/284665>
- Koptur S., Haber W.A., Frankie G.W., Baker H.G. (1988) Phenological studies of shrub and treelet species in tropical cloud forest of Costa Rica. *Journal of Tropical Ecology* 4: 323–346. <http://dx.doi.org/10.1017/s0266467400002984>
- Levin D.A. (1978) Pollination behavior and the breeding structure of plant populations. In: Richards A.J. (ed.) *The pollination of flowers by insects*: 133–150. London, Linnean Society of London by Academic Press.
- Levin D.A., Anderson W.W. (1970) Competition for pollinators between simultaneously flowering species. *The American Naturalist* 104: 455–467. <http://dx.doi.org/10.1086/282680>
- Lewis A.C. (1989) Flower visit consistency in *Pieris rapae*, the cabbage butterfly. *Journal of Animal Ecology* 58: 1–13. <http://dx.doi.org/10.2307/4982>
- Liuth H.S., Talora D.C., Amorim A.M. (2013) Phenological synchrony and seasonality of understory Rubiaceae in the Atlantic Forest, Bahia, Brazil. *Acta Botanica Brasilica* 27: 195–204. <http://dx.doi.org/10.1590/s0102-33062013000100019>
- Lloyd D.G., Webb C.J. (1992) The selection of heterostyly. In: Barrett S.C.H. (ed.) *Evolution and function of heterostyly*: 179–208. Monographs on Theoretical and Applied Genetics. Berlin,

- Springer-Verlag. [http://dx.doi.org/10.1007/978-3-642-86656-2\\_7](http://dx.doi.org/10.1007/978-3-642-86656-2_7)
- Lopes L.E., Buzato S. (2005) Biologia reprodutiva de *Psychotria suterella* Muell. Arg. (Rubiaceae) e a abordagem de escalas ecológicas para a fenologia de floração e frutificação. *Revista Brasileira de Botânica* 28: 785–795. <http://dx.doi.org/10.1590/s0100-84042005000400013>
- Martin-Gajardo I.S., Morellato L.P.C. (2003) Fenologia de Rubiaceae do sub-bosque em floresta Atlântica no sudeste do Brasil. *Revista Brasileira de Botânica* 26: 299–309. <http://dx.doi.org/10.1590/s0100-84042003000300003>
- Mesquita-Neto J.N. (2013) Plant-pollinator interactions in synchropatric species of *Psychotria* (Rubiaceae). Ph.D. dissertation, Universidade Federal de Goiás, Goiânia, Goiás.
- Motten A.F. (1986) Pollination ecology of the spring wildflower community of a temperate deciduous forest. *Ecological Monographs* 56: 21–42. <http://dx.doi.org/10.2307/2937269>
- Moreti A.C.C.C., Anacleto D.A., Ávila M.D., Vieira G.H.C., Marchini L.C. (2006) Abelhas visitantes em vegetação de diferentes áreas remanescentes de Cerrado. *Magistra* 18: 229–248.
- Nepokroeff M., Bremer B., Sytsma K.J. (1999) Reorganization of the genus *Psychotria* and tribe Psychotrieae (Rubiaceae) inferred from ITS and rbcL sequence data. *Systematic Botany* 24: 5–27. <http://dx.doi.org/10.2307/2419383>
- Novotny V., Miller S.E., Hulcr J., Drew R.A.I., Basset Y., Janda M., Setliff G.P., Darrow K., Stewart A.J.A., Auga J., Isua B., Molem K., Manumbor M., Tamtiai E., Mogia M., Weiblen G.D. (2007) Low beta diversity of herbivorous insects in tropical forests. *Nature* 448: 692–695. <http://dx.doi.org/10.1038/nature06021>
- Olesen J.M., Jordano P. (2002) Geographic patterns in plant–pollinator mutualistic networks. *Ecology* 83: 2416–2424. <http://dx.doi.org/10.2307/3071803>
- Ollerton J., Lack A.J. (1992) Flowering phenology: an example of relaxation of natural selection? *Trends in Ecology & Evolution* 7: 274–276. [http://dx.doi.org/10.1016/0169-5347\(92\)90175-b](http://dx.doi.org/10.1016/0169-5347(92)90175-b)
- Ollerton J., Johnson S.D., Cranmer L., Keller S. (2003) The pollination ecology of an assemblage of grassland asclepiads in South Africa. *Annals of Botany* 92: 807–834. <http://dx.doi.org/10.1093/aob/mcg206>
- Oliveira A.S. (2008) Fenologia e Biologia Reprodutiva de cinco espécies de *Psychotria* L. (Rubiaceae), em um remanescente florestal urbano, Araguari, MG. Ph.D. dissertation, Universidade Estadual de Campinas, Campinas, São Paulo, Brazil.
- Orians G. (1997) Evolved consequences of rarity. In: Kunin W., Gaston K. (eds) *The biology of rarity*: 190–208. London, Chapman & Hall. [http://dx.doi.org/10.1007/978-94-011-5874-9\\_11](http://dx.doi.org/10.1007/978-94-011-5874-9_11)
- Pailler T., Thompson J.D. (1997) Distyly and variation in heteromorphic incompatibility in *Gaertnera vaginata* (Rubiaceae) endemic to La Reunion Island. *American Journal of Botany* 84: 315–327. <http://dx.doi.org/10.2307/2446005>
- Pianka E.R. (1973) The structure of lizard communities. *Annual Review of Ecology and Systematics* 4: 53–74. <http://dx.doi.org/10.1146/annurev.es.04.110173.000413>
- Podolsky R.D. (1992) Strange floral attractors: pollinator attraction and the evolution of plant sexual systems. *Science* 258: 791–793. <http://dx.doi.org/10.1126/science.258.5083.791>
- Ramos F.N., Santos F.A.M. (2006) Floral visitors and pollination of *Psychotria tenuinervis* (Rubiaceae): distance from the anthropogenic and natural edges of an Atlantic forest fragment. *Biotropica* 38: 383–389. <http://dx.doi.org/10.1111/j.1744-7429.2006.00152.x>
- Raine N.E., Pierson A.S., Stone G.N. (2007) Plant-pollinator interactions in a Mexican *Acacia* community. *Arthropod-Plant Interactions* 1: 101–117. <http://dx.doi.org/10.1007/s11829-007-9010-7>
- Reich P.B., Borchert R. (1984) Water stress and tree phenology in a tropical dry forest in the lowlands of Costa Rica. *Journal of Ecology* 72: 61–74. <http://dx.doi.org/10.2307/2260006>
- Richards J.H., Koptur S. (1993) Floral variation and distyly in *Guettarda scabra* (Rubiaceae). *American Journal of Botany* 80: 31–40. <http://dx.doi.org/10.2307/2445117>
- Rathcke B. (1988) Flowering phenologies in a shrub community: competition and constraints. *Journal of Ecology* 76: 975–994. <http://dx.doi.org/10.2307/2260627>
- Rodrigues E.B., Consolaro H. (2013) Atypical distyly in *Psychotria goyazensis* Mull. Arg. (Rubiaceae), an intramorph self-compatible species. *Acta Botanica Brasilica* 27: 155–161. <http://dx.doi.org/10.1590/S0102-33062013000100016>
- Santos G.M.M., Aguiar C.M.L., Genini J., Martins C.F., Zanella F.C.V., Mello M.A.R. (2012) Invasive Africanized honeybees change the structure of native pollination networks in Brazil. *Biological Invasions* 14: 2369–2378. <http://dx.doi.org/10.1007/s10530-012-0235-8>
- Schemske D.W. (1981) Floral convergence and pollinator sharing in two bee-pollinated tropical herbs. *Ecology* 62: 946–954. <http://dx.doi.org/10.2307/1936993>
- Sá T.F.F. (2013) Distylous system and reproductive biology of five species of *Psychotria* L. (Rubiaceae) in two fragments south-east Goiás. Ph.D. dissertation, Universidade Federal de Goiás, Goiânia, Goiás, Brazil.
- Sakai S., Wright S.J. (2008) Reproductive ecology of 21 coexisting *Psychotria* species (Rubiaceae): when is heterostyly lost? *Biological Journal of the Linnean Society* 93: 125–134. <http://dx.doi.org/10.1111/j.1095-8312.2007.00890.x>
- Silva N.A.P., Frizzas M.R., Oliveira C.M. (2011) Seasonality in insect abundance in the “Cerrado” of Goiás State, Brazil. *Revista Brasileira de Entomologia* 55: 79–87. <http://dx.doi.org/10.1590/s0085-56262011000100013>
- Teixeira L.A.G., Machado I.C. (2004) Biologia da polinização e sistema reprodutivo de *Psychotria barbiflora* DC. (Rubiaceae). *Acta botânica brasilica* 18: 853–862. <http://dx.doi.org/10.1590/s0102-33062004000400016>
- Taylor C.M. (1996) Overview of the Psychotrieae (Rubiaceae) in the Neotropics. *Opera Botanica Belgica* 7: 261–270.
- Vázquez D.P., Aizen M.A. (2004) Asymmetric specialization: a pervasive feature of plant–pollinator interactions. *Ecology* 85: 1251–1257. <http://dx.doi.org/10.1890/03-3112>
- Waser N.M. (1986) Flower constancy: definition, cause, and measurement. *The American Naturalist* 127: 593–603. <http://dx.doi.org/10.1086/284507>
- Waser N.M., Real L.A. (1979) Effective mutualism between sequentially flowering plant species. *Nature* 281: 670–672. <http://dx.doi.org/10.1038/281670a0>
- Wolda H. (1988) Insect seasonality: Why? *Annual Review of Ecology and Systematics* 19: 1–18. <http://dx.doi.org/10.1146/annurev.es.19.110188.000245>

Manuscript received 20 Feb. 2014; accepted in revised version 16 Feb. 2015.

Communicating Editor: Renate Wesselingh.