

Comparing Foraging Niches of Newly Sympatric Bumble Bees in Alpine Habitats of

Colorado

By

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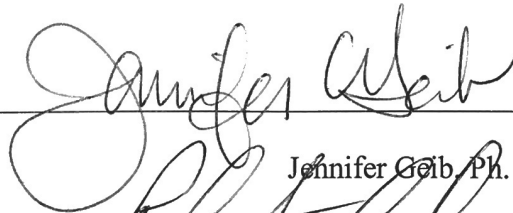
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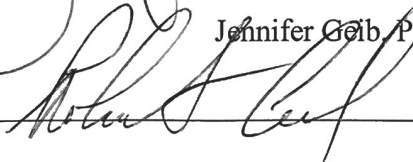
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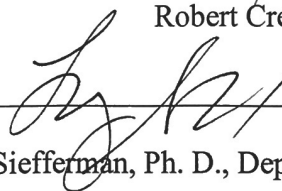
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Abstract

Global climate change has facilitated upward range shifts of bumblebees in mountainous habitats worldwide, increasing species richness and potentially competition for limited floral resources. Bumblebees are generalist pollinators but select floral resources based on cues such as nectar content, accessibility, and corolla depth. Competition for flowers is predicted to occur primarily between bees with similar tongue lengths. I assessed the potential for competition between two bumblebees in central Colorado: *Bombus sylvicola*, a short-tongued native alpine bee, and *B. bifarius*, a short-tongued subalpine species that has recently established a presence above treeline. To assess diet preferences, I allowed individuals of each species to forage on interspersed arrays of seven early season native alpine flowers. I observed bees' foraging for a single foraging bout and recorded floral visitation frequency, foraging time, and constancy of transitions among inflorescences. Results showed that, even though visitation profiles of individual bees varied, all measures of *B. sylvicola* and *B. bifarius* foraging reflected overlapping diet niches. Mean visitation frequencies to the seven floral species were nearly identical for both bee species, as were inflorescence foraging times and species fidelity during transitions. Results suggest that the arrival of *B. bifarius* above treeline has resulted in competition between the morphologically similar bees for available floral resources.

Introduction

Recent studies have projected and documented shifts in the historic ranges of numerous species mediated by climatic changes (Parmesan 2006, Lenoir et al. 2008, Hegland et al. 2009, Van der Putten et al. 2010, Chen et al. 2011, Engler et al. 2011). Species are expected to migrate towards the poles and upward in elevation (Chen et al. 2001, Parmesan 2006, Pradervand et al.

2014), the most significant of range changes being observed in mountainous species (Lenoir et al. 2008, Engler et al. 2011). While many lowland organisms are simply expected to expand their ranges, species already present in alpine and polar regions of the global are experiencing range contractions as they reach upper physical limits (Chen et al. 2011, Kerr et al. 2015). These cold-adapted species are less able to respond to climate change due to their physiology and physical constraints (Engler et al. 2011, Pradervand et al. 2014). Climate-mediated range shifts introduce novel species into habitats and bring together species that may not have previously been sympatric. This is likely to result in new interactions or increased probability of interactions between the novel species and the original inhabitants, potentially altering interaction webs and community dynamics.

Climate change impacts on pollinators have been of particular interest due the ecosystem services they provide and their mutualism with plants. Changes in plant and pollinator phenology have been documented at multiple locations (Fitter & Fitter 2002, Hegland et al. 2009, Bartomeus et al. 2011). In Europe and the Western US, pollinator species are shifting ranges upward in elevation, presumably in response to climatic warming (Ploquin et al. 2013, Kerr et al. 2015, Miller-Struttman et al. 2015). Treeline is also shifting (Xu et al. 2009), but mobile species such as bees can advance further at a more rapid pace. These upwardly mobile, mid-to-lowland pollinator species have increased species richness and altered the functional traits involved in pollination (i.e. tongue length; Ploquin et al. 2013, Miller-Struttman et al. 2015). When there is morphological similarity in such pollinator traits, it may offer functional redundancy from the plant's perspective, but it is unclear what the outcome will be for the pollinators competing for floral resources. This is especially true given global declines in flower abundance. Unless resident pollinators can disperse to even higher areas to avoid competition with their new

neighbors, competition with invaders appears inevitable. Competitive exclusion or character displacement are predicted to occur with increasing niche overlap, particularly if resources are limiting. Alternatively, coexistence between morphologically similar species has been observed in multiple organisms, largely due to differences in foraging strategy and frequency (Johnson & Hubbell 1975, Genner et al. 1999, Nakano et al. 1999). If invading pollinators differ significantly in these behaviors from resident pollinators then coexistence is possible.

This study aims to assess the potential for niche overlap between newly sympatric, morphologically similar bumblebees in the Central Rocky Mountains of Colorado. The upward movement of a short-tongued bumblebee, *Bombus bifarius*, into the alpine habitat has been documented at multiple sites (Miller-Struttman et al. 2015) creating the potential for competition with a native short-tongued congener, *B. sylvicola*. It is necessary to observe and compare species' diet preferences to evaluate the potential for competition. I hypothesized that the bee species would exhibit similar diet preferences due to their corresponding proboscis lengths. The goal of this study was to determine whether *B. bifarius* and *B. sylvicola* have overlapping diet niches via assessment of multiple indices of foraging preference among common bumblebee-pollinated alpine plants, including individual visitation frequencies, foraging time, and species fidelity during inflorescence transitions (constancy).

Methods

Study site and system

Data for this study were collected during the summer of 2016 at Pennsylvania Mountain Natural Area (Park County, Colorado, USA), a site that has experienced marked shifts in its bumblebee assemblage over the last forty years. Bumblebee inventories conducted in the 1970s

reported two permanent residents above treeline, *Bombus balteatus* and *B. sylvicola* that comprised 99% of all caught individuals. More recent semi-annual inventories conducted since 2008 show that the community now comprises at least eight species, the five most abundant of which each comprise 10-40% (Miller-Struttman et al. 2015). These five species vary in traits that would be predicted to impact their associations with host food plants, including body size and tongue length (Miller-Struttman et al. 2015).

The bees selected for this study were the alpine-adapted historical resident, *Bombus sylvicola* and *B. bifarius*, a historically subalpine species that has recently established a presence above treeline. *B. sylvicola* and *B. bifarius* are extremely similar in morphology, including their short tongue length (5.75 mm and 5.79 mm, respectively; Miller-Struttman et al. 2015). Both species, like other *Bombus*, are eusocial pollinators annually established by new inseminated queens who emerge from torpor upon snowmelt, phenologically-linked to budding flowers (Goulson 2009). Queens in the Central Rockies typically emerge around mid-June and begin searching for suitable nesting sites to establish a colony, thereafter collecting nectar and pollen to support their broods (Byron 1980, Heinrich 1979). Colony size and life expectancy are primarily dependent upon resource availability during the season (Heinrich 1979). Floral preferences of *B. sylvicola* and *B. bifarius* workers were assessed, because workers comprise the majority of flower visitors throughout the season, following their emergence.

Field experiments

Seven native plant species known to be bee-pollinated (Byron 1980, Geib 2010) and in full bloom during the experiment (July 1-31) were used to assess bumblebee diet preference (e.g., *Mertensia* spp., *Oxytropis sericea*, *Pedicularis parryi*, *Phacelia sericea*, *Polemonium viscosum*, *Trifolium dasyphyllum*, and *T. parryi*). *Mertensia* spp. are referred to only by genus, as

species present on Pennsylvania Mountain are difficult to distinguish. Virgin inflorescences of the seven floral species were cut from wild populations in the bud stage to eliminate interacting effects from other foraging pollinators. Cut inflorescences were immediately placed into filled florist water picks (4 ¾") to retain vitality. Inflorescences were then placed into mesh enclosures (~2m x 2m) to exclude pollinators until the plants bloomed. Upon full bloom inflorescences were available for inclusion in experimental foraging arrays.

Foraging arrays (Figure 1) comprised a seven-point interspersed arrangement of water picks in a 2m x 2m enclosed, bottomless mesh tent. The experimental design followed that of Geib (2010). Any flowers occurring naturally within the enclosure were cut and removed. Inflorescences were haphazardly chosen from those available to fill the array, and flower order within the array was randomized for each bout. A new array was created for each individual pollinator. Foraging worker bees were haphazardly collected with nets in the krummholz and lower alpine areas. Following capture, bees were put into vials and placed on snow or ice packs to induce torpor and allow for species identification. Each trial was conducted with an individual bee (*B. sylvicola*: $N = 13$, *B. bifarius*: $N = 10$); bees were warmed up, placed in the middle of the array to reduce distance bias effects, and observed foraging for nectar. A foraging bout was considered finished when the individual flew to the walls of the enclosure and could not be coaxed back to the middle of the array. The goal was to attain 10-20 minutes of consistent foraging, but some bees ended their bouts in less time. The plant species, position within the array, number of flowers foraged upon per inflorescence, and foraging time per plant were recorded. Bees were then marked to prevent recapture and reuse, and then released.

Statistical analysis

I used independent t-test assuming unequal variances (JMP 13.1; SAS Institute 2016) to compare *B. sylvicola* and *B. bifarius* bout lengths, including means of total time spent foraging per bout and total number of inflorescences visited per bout. Inflorescences of different species within an array were not statistically independent of each other, therefore I also used independent t-tests, assuming unequal variances and run separately for each plant species, to compare indices of preference between *B. sylvicola* and *B. bifarius*. These indices included proportion of available inflorescences visited per bout, mean number of flowers visited per bout, mean time spent foraging per inflorescence, and proportion of constant (same-species) transitions during the bouts. I then used nonparametric Kruskal-Wallis tests to assess whether the indices of preference varied among the plant species. When the Kruskal-Wallis test was significant, I used Steel-Dwass tests for nonparametric post-hoc pairwise comparisons between plant species. I used analysis of variance (ANOVA; JMP 13.1) to compare the overall prevalence of transition types (constancy and switching) during foraging sequences between *B. sylvicola* and *B. bifarius* (floral species pooled). For parametric tests, proportions were subject to arcsine square root transformations prior to analysis. Chi-squared (χ^2) analyses were conducted in SAS (Version 9.1; SAS Institute 2004) to compare observed visitation frequencies and constancy (proportion of same species transitions during a bout) to that expected based on abundance of inflorescences of each species.

Results

Bout length

Individual bumble bees exhibited variation in their visitation frequencies and sequences (e.g see Figs. 9, 10); nevertheless, the mean bout length did not differ for *B. sylvicola* and *B.*

bifarius. They exhibited similarity in the mean number of inflorescences visited per bout and mean time spent foraging per bout ($t(1) = 1.1731, p = 0.2546$ and $t(1) = 0.7083, p = 0.4894$ for inflorescences and time per bout, respectively; Table 1A, 1B).

Visitation frequencies and foraging time

B. bifarius and *B. sylvicola* exhibited overlapping diet niches. There was no significant difference between the bee species in 1) mean proportion of available inflorescences visited (for all plants $p > 0.05$, Table 2, Fig. 2), 2) mean number of flowers visited per species during a foraging bout, (for all plants $p > 0.05$, Table 3A, Fig. 3), 3) average percentage of flowers visited per bout (for all plants $p > 0.05$, Table 3B, Fig. 4), mean time spent foraging available per inflorescences (for all plants $p > 0.05$, Table 4A, Fig. 5), and mean time spent per floral species (for all plants $p > 0.05$, Table 4B, Fig. 6).

Instead, plant species was the only significant contributor to variation in visitation frequencies when bee species were pooled for all dependent variables (mean proportion of available inflorescences visited, $\chi^2 = 35.2652, p < 0.0001$; mean number of flowers visited per species during a foraging bout, $\chi^2 = 35.3967, p < 0.0001$; average proportion of flowers visited per bout, $\chi^2 = 34.4497, p < 0.0001$; mean time spent foraging per inflorescence, $\chi^2 = 28.2972, p < 0.0001$; and mean time spent per floral species, $\chi^2 = 32.7863, p < 0.0001$; Table 8, Fig. 2-6).

Mertensia spp. and *P. sericea* received significantly more inflorescence visits, flower visits, and foraging time than almost all other species (nonparametric post-hoc pairwise Steel-Dwass tests, $p < 0.05$ for all species except *P. viscosum* when bee species were pooled).

Constancy of transitions between inflorescences

B. sylvicola and *B. bifarius* exhibited similar overall foraging constancy (floral species pooled). A two-way ANOVA revealed no differences in transition frequency among bee species

or transition types (constant vs switching) (Whole model, $F_{3,40} = 0.61$, $p = 0.61$, Table 6, Fig. 7). However, overall constancy was higher and overall switches were lower than expected under random foraging, based on proportional abundance of inflorescences of each floral species within the array (*B. sylvicola*: $\chi^2 = 25.0000$, $p < 0.0001$, *B. bifarius*: $\chi^2 = 20.8334$, $p = 0.0002$, Table 7, Fig. 7).

B. sylvicola and *B. bifarius* exhibited similar fidelity patterns among the floral species, but the plants differed among each other in proportion of constant transitions out of the total bout transitions (Constancy Index A, $\chi^2 = 40.5449$, $p < 0.0001$, Table 8, Fig. 8) and in proportion of constant transitions out of total transitions from that species (Constancy Index B, $\chi^2 = 39.8560$, $p < 0.0001$, Table 8, Fig. 8). Fidelity was significantly greater for *P. sericea* and *Mertensia* spp. when compared to all except *P. viscosum*. (Post hoc Steel-Dwass tests $p > 0.05$). Interestingly, switching back and forth, reducing overall constancy, between two floral species was commonly observed among individual bees (Figs. 9, 10).

Discussion

Our overarching objective for this study was to assess and compare the diet preferences of two newly sympatric bumble bees in the Colorado Rocky Mountains: *B. sylvicola*, a short-tongued native alpine bumble bee, and *B. bifarius*, a short-tongued lower elevation congener that has now become well-established high above treeline, likely due to climatic warming (Geib et al. 2015, Miller-Struttman et al. 2015). All indices of floral preference measured from workers of both species foraging on arrays of bee-pollinated alpine plants were nearly identical, providing support for our prediction that the species would exhibit similar foraging niches due to

similarities in proboscis length. Both bees significantly preferred *P. sericea* and *Mertensia* spp., based on visitation frequencies, foraging times, and constancy of inflorescence transitions.

How bees choose flowers

Foraging behaviors in bumbles and other insect pollinators are governed by a number of factors. Bees' floral species preferences during nectar foraging typically reflect proboscis length and corolla depth matching (Brian 1957, Ranta & Lundberg 1980, Graham & Jones 1996, Rodríguez-Gironés & Santamaría 2006), which is hypothesized to improve the energetic rewards of foraging (Waser 1986). This pattern probably represents the bees' innate preferences, which are likely to prevail in contexts similar to this study where choices offered provide their full reward potential and are not affected by prior visitors. Bee movement between inflorescences is also expected to maximize the net energy gained (Pyke 1978). Constancy of transitions (movement between same-species flowers or inflorescences) is often observed during foraging, despite other rewarding plants being accessible (Waser 1986). Specialization and constancy are advantageous to foraging as they reduce the amount of time and energy spent manipulating flowers, in addition to minimizing time taken for visual searching and learned flower handling (Heinrich 1979, Goulson 2009, Heinrich 1976). Constancy has been observed to increase as floral choices become more distinctive in morphology and color (Waser 1986, Wilson & Stine 1996).

In this study, observed constancy was higher than that expected under random foraging based on inflorescence abundance in the array, but lower than observed field study frequencies (Grant 1950, Geib 2010). Limiting resources likely contributed to the reduction in constancy of transitions in this study array, prompting bees to forage on the remaining available choices (Goulson 2009). Individual bees also displayed majoring and minoring, where bees within the

same species have preferences specific to each individual, an observation consistent with the flexible framework of foraging behavior discussed in Heinrich (1976). Several bees switched back and forth between two or three floral species while other individuals were constant on one species. Minorng is thought to help bees keep a record of resource variation throughout the season (Heinrich et al. 1977).

Potential competition among B. sylvicola and B. bifarius

Overlap in foraging niches strongly supports the idea that *B. sylvicola* and *B. bifarius*, will compete intensely and suggests that competitive exclusion of one or the other species is likely. In Pyke (1982), competition and preferential floral order for *B. sylvicola* and *B. bifarius* were based upon the relative abundance of plant species in certain areas of Colorado. Local factors influenced the density and distribution of bees, with competition between species more likely to occur in isolated areas due to limiting resources. Interestingly, in multiple North and Central European studies, coexistence between similarly tongued bees has been observed, primarily competing for pollen rewards with interspecific competition regulating colony densities (Ranta & Vepsäläinen 1981, Goulson & Darvill 2004). While not measured in this study, pollen foraging niches for *B. sylvicola* and *B. bifarius* may or may not reflect those of nectar foraging and may or may not overlap. In any case, bees regulate their consumption of certain pollen types and frequency of foraging based on individual energetic needs and available resources (Vaudo et al. 2016).

Interspecific competition often occurs between bumblebees as most are generalist foragers, pollinating several plant species with the most abundant bee species possessing the broadest diets and vice versa (Goulson & Darvill 2004). In Inouye (1978) interspecific competition for nectar was observed between *B. appositus* and *B. flavifrons*, which possess long

and medium-length tongues, and the presence the other bee altered foraging choices from observed preferences when competitors were absent. The findings suggest that coexistence is likely not solely based on tongue length and corolla depth relationships but is also influenced by the presence of competitors. Bees respond to both direct (e.g. footprint scents; Stout et al. 1998, Saleh et al. 2007) and indirect (lack of nectar; Marden 1984, Stout & Goulson 2002) cues left by prior visitors. Morphologically similar bees may partition resources through habitat selection or varying seasonal preferences (Inoue & Yokoyama 2006). Flexibility in foraging niches is a broadly common strategy for coexistence, observed in other systems and habitats; e.g. sympatric, morphologically similar fish have been observed to coexist and maintain population densities due to flexibility in niche shifts (Nakano et al. 1999). Conversely, competitive exclusion is theorized to occur between noninterbreeding, sympatric populations, functionally equivalent competitors (Hardin 1960). In Connell (1961), intertidal barnacles were artificially removed to observe interspecific interactions, with the competitively weaker barnacle experiencing higher survivorship along the lower extent of its fundamental niche following removal of its competitor. The study provides an illustration of how competitive exclusion influences the zonation of competing populations. Exclusion has also been observed in montane chipmunk populations where species were excluded primarily through interspecific aggression (Brown 1971). Localized factors of landscape composition and configuration also influenced habitat suitability for both populations.

My results provide support for competition between *B. sylvicola* and *B. bifarius* due to foraging niche overlap. Observation of species associations in true field settings is needed to further evaluate this conclusion. Bees were given limited choices in the array, with declines in nectar rewards of the innately favored species potentially influencing foraging choices.

Competitive interactions were also not included in assessing preference, allowing individual bees to visit equally rewarding plants without effects from prior visits. Nevertheless, species preferences observed here do conform with historical preference records in Colorado (Pyke 1982). It is unclear currently how other aspects of these species' life histories compare. For example, Bryon (1980) found that bumble bee colonies above treeline were likely limited in abundance and distribution by available nesting sites, and that lowland species such as *B. bifarius* were physiologically able to maintain nests when transferred into alpine environments. Further studies must also be done to assess other aspects of interactions between *B. sylvicola* and *B. bifarius* and determine if the species can coexist or competitively exclude one another.

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Literature Cited

- Bartomeus, I., J. S. Ascher, D. Wagner, B. N. Danforth, S. Colla, S. Kornbluth, and R. Winfree. 2011. Climate-associated phenological advances in bee pollinators and bee-pollinated plants. *Proceedings of the National Academy of Sciences* **108**:20645-20649.
- Brian, A. D. 1957. Differences in the flowers visited by four species of bumble-bees and their causes. *Journal of Animal Ecology* **26**:71-98.

- Brown, J. H. 1971. Mechanisms of competitive exclusion between two species of chipmunks. *Ecology* **52**:305-311.
- Byron, P. A. 1980. On the ecology and systematics of Coloradan bumblebees. Dissertation. University of Colorado, Boulder, Colorado, USA.
- Chen, I., J. K. Hill, R. Ohlemüller, D. B. Roy, and C. D. Thomas. 2011. Rapid range shifts of species associated with high levels of climate warming. *Science* **333**:1024-1026.
- Connell, J. H. 1961. The influence of interspecific competition and other factors on the distribution of the barnacle *Chthamalus stellatus*. *Ecology* **42**:710-723.
- Engler, R., C. F. Randin, W. Thuiller, S. Dullingers, N. E. Zimmermann, M. B. Araújo, P. B. Pearman, G. Le Lay, C. Piedallu, C. H. Albert, P. Choler, G. Coldea, X. de Lamo, T. Dirnböck, J. Gégout, D. Gómez-García, J. Grytnes, E. Heegaard, F. Høistad, D. Nogués-Bravo, S. Normand, M. Puşcaş, M. Sebastià, A. Stanisci, J. Theurillat, M. R. Trivedi, P. Vittoz, and A. Guisan. 2011. 21st century climate change threatens mountain flora unequally across Europe. *Global Change Biology* **17**:2330-2341.
- Fitter, A. H. and R. S. R. Fitter. 2002. Rapid changes in flowering time in British plants. *Science* **296**:1689-1691.
- Geib, J. C. 2010. The impacts of partner abundance on benefits from facultative pollination mutualism. Unpublished doctoral dissertation, University of Missouri-Columbia, Columbia, Missouri, USA.
- Geib, J. C., J. Strange, and C. Galen. 2015. Bumble bee nest abundance, foraging distance, and host-plant reproduction: implications for management and conservation. *Ecological Applications* **25**:768-778.

- Graham, L. and K. N. Jones. 1996. Resource partitioning and per-flower foraging efficiency in two bumble bee species. *The American Midland Naturalist* **136**:401-406.
- Genner, M. J., G. F. Turner, and S. J. Hawkins. 1999. Foraging of rocky habitat cichlid fishes in a Lake Malawi: coexistence through niche partitioning? *Oecologia* **121**:283-292.
- Goulson, D. 2009. Bumblebees: behaviour, ecology and conservation. Oxford University Press, Oxford, UK.
- Goulson, D. and B. Darvill. 2004. Niche overlap and diet breadth in bumblebees; Are rare species more specialized in their choice of flowers?. *Apidologie* **35**:55-63.
- Grant, V. 1950. The flower constancy of bees. *Botanical Review* **16**:379-398.
- Hardin, G. 1960. The competitive exclusion principle. *Science* **131**:1292-1297.
- Hegland, S. J., A. Nielsen, A. Lazaro, A. L. Bjerknes, and O. Totland. 2009. How does climate warming affect plant-pollinator interactions? *Ecology Letters* **12**:184-195.
- Heinrich, B. 1976. The foraging specializations of individual bumblebees. *Ecological Monographs* **46**:105-128.
- Heinrich, B. 1979. Bumblebee economics. Harvard University Press, Cambridge, Massachusetts, USA.
- Heinrich, B., P. R. Mudge, and P. G. Deringis. 1977. Laboratory analysis of flower constancy in foraging bumblebees: *Bombus ternarius* and *B. terricola*. *Behavioral Ecology and Sociobiology* **2**:247-265.
- Inoue, M. N. and J. Yokoyama. 2006. Morphological variation in relation to flower use in bumblebees. *Entomological Science* **9**:147-159.
- Inouye, D. W. 1978. Resource partitioning in bumblebees: experimental studies of foraging behavior. *Ecology* **59**:672-678.

- Johnson, L. K. and S. P. Hubbell. 1975. Contrasting foraging strategies and coexistence of two bee species on a single resource. *Ecology* **56**:1398-1406.
- Kerr, J. T., A. Pindar, P. Galpern, L. Packer, S. G. Potts, S. M. Roberts, P. Rasmont, O. Schweiger, S. R. Colla, L. L. Richardson, D. L. Wagner, L. Gall, F., D. S. Sikes, and A. Pantoja. 2015. Climate change impacts on bumblebees converge across continents. *Science* **349**:177-180.
- Lenoir, J., J. C. Gégout, P. A. Marquet, P. de Ruffray, and H. Brisse. 2008. A significant upward shift in plant species optimum elevation during the 20th century. *Science* **320**:1768-1771.
- Marden, J. H. 1984. Remote perception of floral nectar by bumblebees. *Oecologia* **64**:232-240.
- Miller-Struttman, N. E., J. C. Geib, J. D. Franklin, P. G. Kevan, R. M. Holdo, D. Ebert-May, A. M. Lynn, J. A. Kettenbach, E. Hedrick, and C. Galen. 2015. Functional mismatch in a bumble bee pollination mutualism under climate change. *Science* **349**:1541-1544.
- Nakano, S., K. D. Fausch, and S. Kitano. 1999. Flexible niche partitioning via a foraging mode shift: a proposed mechanism for coexistence in stream-dwelling charrs. *Journal of Animal Ecology* **68**:1079-1092.
- Parmesan, C. 2006. Ecological and evolutionary responses to recent climate change. *Annual Review of Ecology, Evolution, and Systematics* **37**:637-669.
- Ploquin, E. F., J. M. Herrera, and J. R. Obeso. 2013. Bumblebee community homogenization after uphill shifts in montane areas of northern Spain. *Oecologia* **173**:1649-1660.
- Pradervand, J., L. Pellissier, C. F. Randin, and A. Guisan. 2014. Functional homogenization of bumblebee communities in alpine landscapes under projected climate change. *Climate Change Responses* **1**:1-10.

- Pyke, G. H. 1978. Optimal foraging: movement patterns of bumblebees between inflorescences. *Theoretical Population Biology* **13**:72-98.
- Pyke, G. H. 1982. Local geographic distributions of bumblebees near Crested Butte, Colorado: competition and community structure. *Ecology* **63**:555-573.
- Ranta, E., and H. Lundberg. 1980. Resource partitioning in bumblebees: the significance of differences in proboscis length. *Oikos* **35**:298-302.
- Ranta, E. and K. Vepsäläinen. 1981. Why are there so many species? Spatial-temporal heterogeneity and Northern bumblebee communities. *Oikos* **36**:28-34.
- Rodríguez-Gironés, M. A., and L. Santamaría. 2006. Models of optimal foraging and resource partitioning: deep corollas for long tongues. *Behavioral Ecology* **17**:905-910.
- Saleh, N., A. G. Scott, G. P. Bryning, and L. Chittka. 2007. Distinguishing signals and cues: bumblebees use general footprints to generate adaptive behaviour at flowers and nest. *Anthropod-Plant Interactions* **1**:119-127.
- SAS Institute. 2004. SAS version 9.1 SAS Institute, Cary, North Carolina, USA.
- SAS Institute. 2016. JMP version 13.1. SAS Institute, Cary, North Carolina, USA.
- Stout, J. C. and D. Goulson. 2002. The influence of nectar secretion rates on the responses of bumblebees (*Bombus* spp.) to previously visited flowers. *Behavioral Ecology and Sociobiology* **52**:239-246.
- Stout, J. C., D. Goulson, and J. A. Allen. 1998. Repellent scent-marking of flowers by a guild of foraging bumblebees (*Bombus* spp.). *Behavioral Ecology and Sociobiology* **43**:317-326.
- Van der Putten, W. H., M. Macel, and M. E. Visser. 2010. Predicting species distribution and abundance responses to climate change: why it is essential to include biotic interactions across trophic levels. *Philosophical Transactions of the Royal Society* **365**:2025-2034.

Vaudo, A. D., H. M. Patch, D. A. Mortensen, J. F. Tooker, and C. M. Grozinger. 2016.

Macronutrient ratios in pollen shape bumble bee (*Bombus impatiens*) foraging strategies and floral preferences. *Proceedings of the National Academy of Sciences* **113**:201606101.

Waser, N. M. 1986. Flower constancy: definition, cause, and measurement. *The American Naturalist* **127**:593-603.

Wilson, P. and M. Stine. 1996. Floral constancy in bumble bees: handling efficiency or perceptual conditioning? *Oecologia* **106**:493-499.

Xu, J., R. E. Grumbine, A. Shrestha, M. Eriksson, X. Yang, Y. Wang, and A. Wilkes. 2009. The melting Himalayas: cascading effects of climate change on water, biodiversity, and livelihoods. *Conservation Biology* **23**:520-530.

Tables

Table 1. T-tests comparing mean bout length for *B. sylvicola* and *B. bifarius*, including A) mean number of inflorescences visited per bout ($N = 13$ and 10 for *B. sylvicola* and *B. bifarius*, respectively), and B) mean time per bout ($N = 8$ and 10 for *B. sylvicola* and *B. bifarius*, respectively).

	DFNum	DFDen	F Ratio	Prob > F	T Test
A)	1	19.935	1.3761	0.2546	1.1731
B)	1	15.357	0.5017	0.4894	0.7083

Table 2. T-test comparing mean proportion of available inflorescences visited for *B. sylvicola* and *B. bifarius* by plant species. PS = *P. sericea*, ME = *Mertensia* spp., PV = *P. viscosum*, TD = *T. dasyphyllum*, TP = *T. parryi*, OS = *O. sericea*, PP = *P. parryi*.

Plant Species	t	DF	Sig (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
						Lower	Upper
PS	0.172544	19.75662	0.8648	0.03169	0.18368	-0.35175	0.41514
ME	-0.190290	19.41161	0.8511	-0.03323	0.17463	-0.39822	0.33176
PV	0.317371	20.85788	0.7541	0.03585	0.11295	-0.19914	0.27083
TD	-0.253510	11.37234	0.8044	-0.02308	0.09103	-0.22264	0.17648
TP	1.227366	19.21789	0.2345	0.05323	0.04337	-0.03747	0.14394
OS	0.202454	15.78224	0.8421	0.00762	0.03762	-0.07222	0.08745
PP	0.037950	19.45701	0.9701	0.00108	0.02838	-0.05822	0.06038

Table 3. T-tests comparing flower visitation frequency for *B. sylvicola* and *B. bifarius* by plant species. A) Mean number of flowers visited per species during a foraging bout, and B) mean proportion of total flowers visited. PS = *P. sericea*, ME = *Mertensia* spp., PV = *P. viscosum*, TD = *T. dasyphyllum*, TP = *T. parryi*, OS = *O. sericea*, PP = *P. parryi*.

Plant Species	t	DF	Sig (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
						Lower	Upper
A) PS	0.179932	19.71896	0.8590	0.15960	0.8872	-1.69280	2.01210
ME	-0.149940	20.98953	0.8822	-0.10370	0.6913	-1.54140	1.33410
PV	-0.132960	19.87199	0.8956	-0.07310	0.5498	-1.22030	1.07420
TD	0.068781	16.21324	0.9460	0.03440	0.5002	-1.02490	1.09370
TP	1.009363	20.85737	0.3244	0.37410	0.3706	-0.39690	1.14510
OS	0.629706	20.95861	0.5357	0.19984	0.31735	-0.46020	0.85988
PP	0.631308	20.80834	0.5347	0.15468	0.24501	-0.35514	0.66450
B) PS	-0.201760	19.56228	0.8422	-0.04729	0.23438	-0.53689	0.44232
ME	0.076872	20.58139	0.9395	0.01343	0.17473	-0.35038	0.37724
PV	-0.337110	20.10363	0.7395	-0.04876	0.14463	-0.35036	0.25284
TD	0.355391	20.99926	0.7258	0.05661	0.15928	-0.27463	0.38784
TP	0.173221	17.84093	0.8644	0.01084	0.06257	-0.12070	0.14237
OS	0.933757	17.84206	0.3629	0.05758	0.06167	-0.07206	0.18722
PP	0.076759	18.18799	0.9397	0.00295	0.03848	-0.07783	0.08374

Table 4. T-tests comparing foraging time for *B. sylvicola* and *B. bifarius* by plant species. A)

Mean time spent per inflorescence and B) mean time spent per floral species. PS = *P.*

sericea, ME = *Mertensia* spp., PV = *P. viscosum*, TD = *T. dasyphyllum*, TP = *T. parryi*, OS =

O. sericea, PP = *P. parryi*.

Plant Species	t	DF	Sig (2- tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
						Lower	Upper
A) PS	0.031594	12.66629	0.9753	1.3680	43.311	-92.451	95.187
ME	0.008382	16.28577	0.9934	0.2210	26.340	-55.538	55.979
PV	0.199348	16.31450	0.8445	3.7580	18.849	-36.139	43.654
TD	-0.302170	8.315753	0.7699	-9.636	31.890	-82.692	63.420
TP	1.089904	13.34844	0.2950	13.670	12.543	-13.355	40.696
OS	1.437418	10.67072	0.1793	9.9320	6.9090	-5.3330	25.197
PP	0.642413	16.86683	0.5292	1.9659	3.0602	-4.4944	8.4262
B) PS	0.082815	14.16612	0.9352	14.450	174.54	-359.48	388.39
ME	0.203463	14.46750	0.8416	38.830	190.84	-369.25	446.91
PV	0.763228	15.97825	0.4564	52.140	68.310	-92.690	196.96
TD	-0.171560	8.479181	0.8678	-5.500	32.059	-78.707	67.707
TP	1.434547	11.61478	0.1778	25.534	17.799	-13.391	64.459
OS	1.437418	10.67072	0.1793	9.9320	6.9090	-5.5333	25.197
PP	0.642413	16.86683	0.5292	1.9659	3.0602	-4.4944	8.4262

Table 5. T-tests comparing foraging constancy for *B. sylvicola* and *B. bifarius* by plant species during foraging bouts. A) proportion of same-species transitions out of all transitions, and B) proportion of same-species transitions out of all transition from that species. PS = *P. sericea*, ME = *Mertensia* spp., PV = *P. viscosum*, TD = *T. dasyphyllum*, TP = *T. parryi*, OS = *O. sericea*, PP = *P. parryi*.

Plant Species	t	DF	Sig (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
						Lower	Upper
A) PS	0.437355	19.96559	0.6665	0.08571	0.19598	-0.32314	0.49457
ME	0.161580	19.25958	0.8733	0.02466	0.15262	-0.29449	0.34381
PV	0.319128	19.89161	0.7530	0.03159	0.09898	-0.17495	0.23813
TD	-1.000000	9.00000	0.3434	-0.02868	0.02868	-0.09354	0.03619
TP	-0.563980	11.77691	0.5834	-0.03174	0.05627	-0.15461	0.09113
OS	-1.000000	9.00000	0.3434	-0.91741	0.01741	-0.05679	0.02197
PP
B) PS	0.390541	19.95876	0.7003	0.09218	0.23602	-0.40023	0.58458
ME	0.471604	19.97986	0.6423	0.10673	0.22631	-0.36538	0.57884
PV	0.310167	19.99051	0.7596	0.04899	0.15795	-0.28050	0.37848
TD	-1.000000	9.00000	0.3434	-0.06119	0.06119	-0.19962	0.07724
TP	-0.294150	15.87890	0.7725	-0.02754	0.09364	-0.22618	0.17109
OS	-1.000000	9.00000	0.3434	-0.06119	0.06119	-0.19962	0.07724
PP

Table 6. ANOVA comparing transition types (constancy and switching, floral species pooled) between *B. sylvicola* and *B. bifarius*.

Source	DF	Type III SS	Mean Square	F Value	Prob > F
model	3	0.1933786	0.105147	0.6130	0.6105
bee species	1	1.1299e-33	1.1299e-33	0.0000	1.0000
transition type	1	0.08947127	0.08947127	0.8509	0.3618
bee species * trans. type	1	0.12124631	0.12124631	1.1531	0.2893
error	40	4.3992402			

Table 7. Chi-square analysis comparing observed constancy vs expected constancy for *B. sylvicola* and *B. bifarius*.

Variable	Chi-square	Df	Asymptotic Pr > ChiSq	Exact Pr >=ChiSq
<i>B. sylvicola</i>	25.0000	1	<.0001	<.0001
<i>B. bifarius</i>	20.8334	1	<.0001	0.0002

Table 8. Non-parametric Kruskal-Wallis tests comparing dependent variables by plant species (*B. sylvicola* and *B. bifarius* pooled). Steel-Dwass tests used as post-hoc pairwise comparison between plant species at $\alpha = 0.05$.

Variable (by plant species)	Kruskal-Wallis Test		
	Chi Square	DF	Prob > ChiSq
Mean prop. avail. inflor. visits	35.2652	6	<0.0001
Mean # of flowers visited/bout	35.3967	6	<0.0001
Mean % of all flower visits	34.4497	6	<0.0001
Mean foraging time/inflorescence	28.2972	6	<0.0001
Mean foraging time/plant species	32.7863	6	<0.0001
Mean prop. constant trans. (out of all trans.)	40.5449	6	<0.0001
Mean prop. constant trans. (out of species trans.)	39.8560	6	<0.0001

Figures

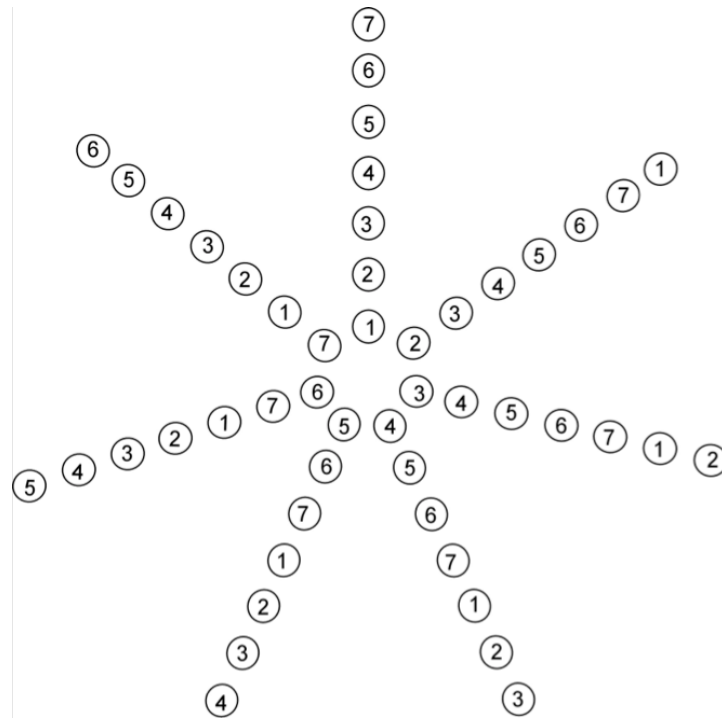
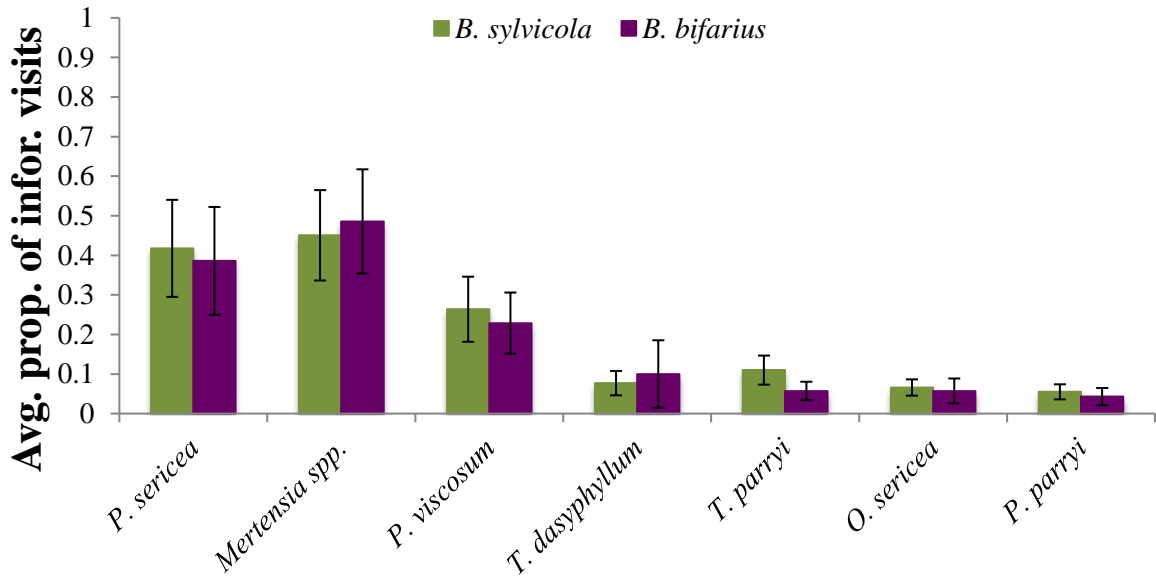


Figure 1. Seven-point star interspersed array design used to assess foraging preferences in each bout. The numbers in the array correspond to the seven alpine plant species used. Experimental design follows Geib (2010).

A)



B)

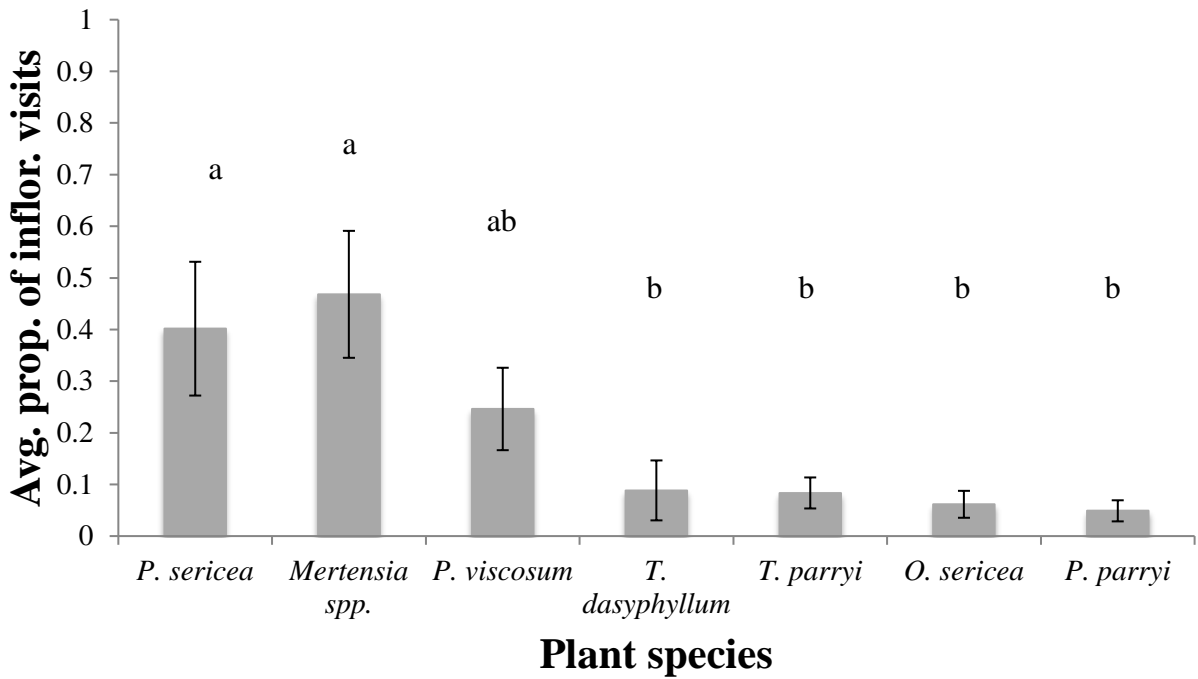
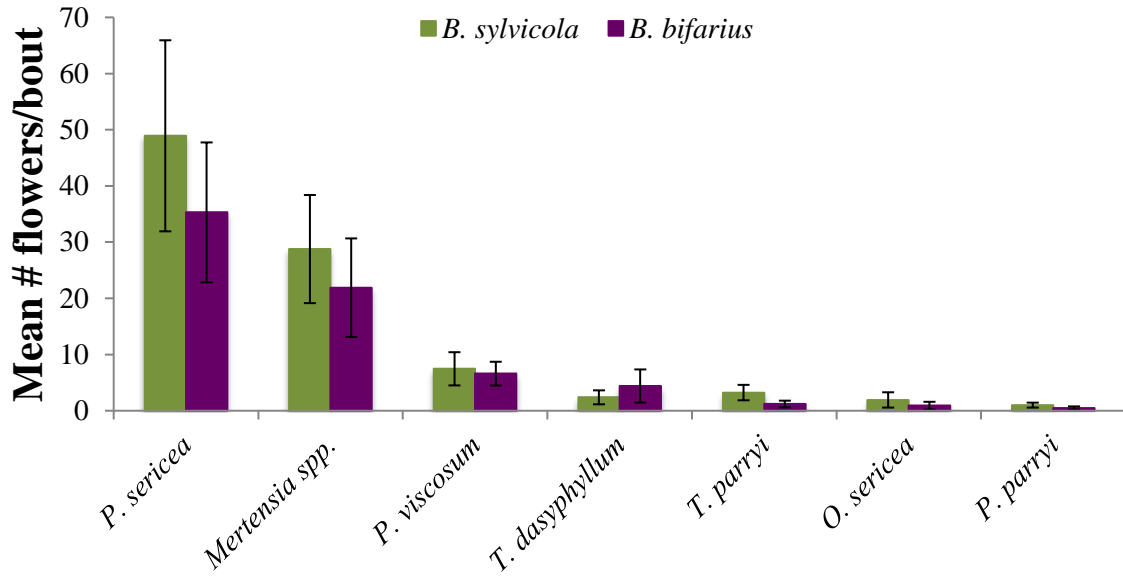


Figure 2. Mean proportion of the available inflorescences visited for each plant species during foraging bouts compared A) between bee species (*B. sylvicola* and *B. bifarius*) and B) among the plant species. Letters show significant differences among the plant species (bee species pooled) at $\alpha = 0.05$. Error bars are one standard error.

A)



B)

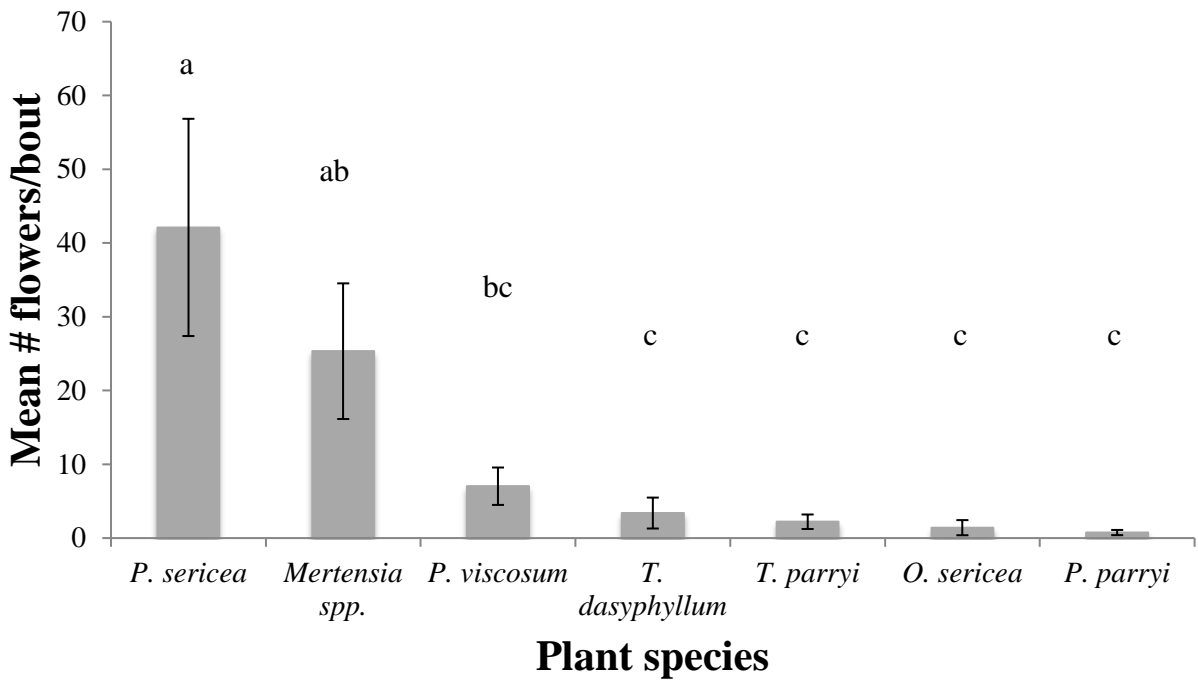
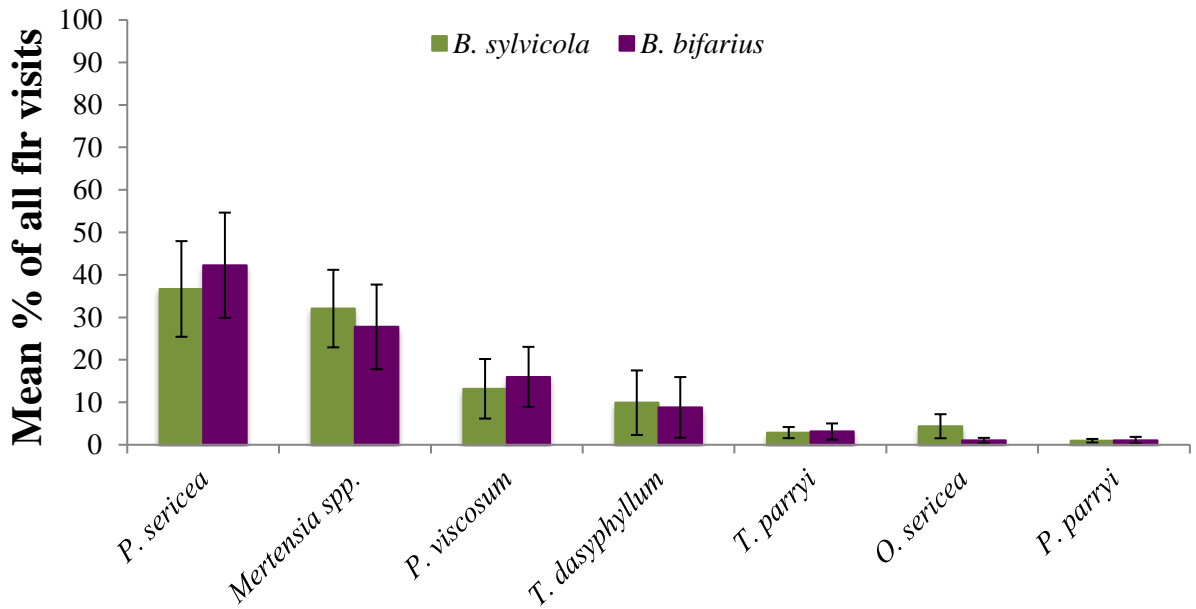


Figure 3. Mean number of flowers visited for each flower species during foraging bouts compared A) between the bee species (*B. sylvicola* and *B. bifarius*) and B) among the plant species. Letters show significant differences among the plant species (bee species pooled) at $\alpha = 0.05$. Error bars are one standard error.

A)



B)

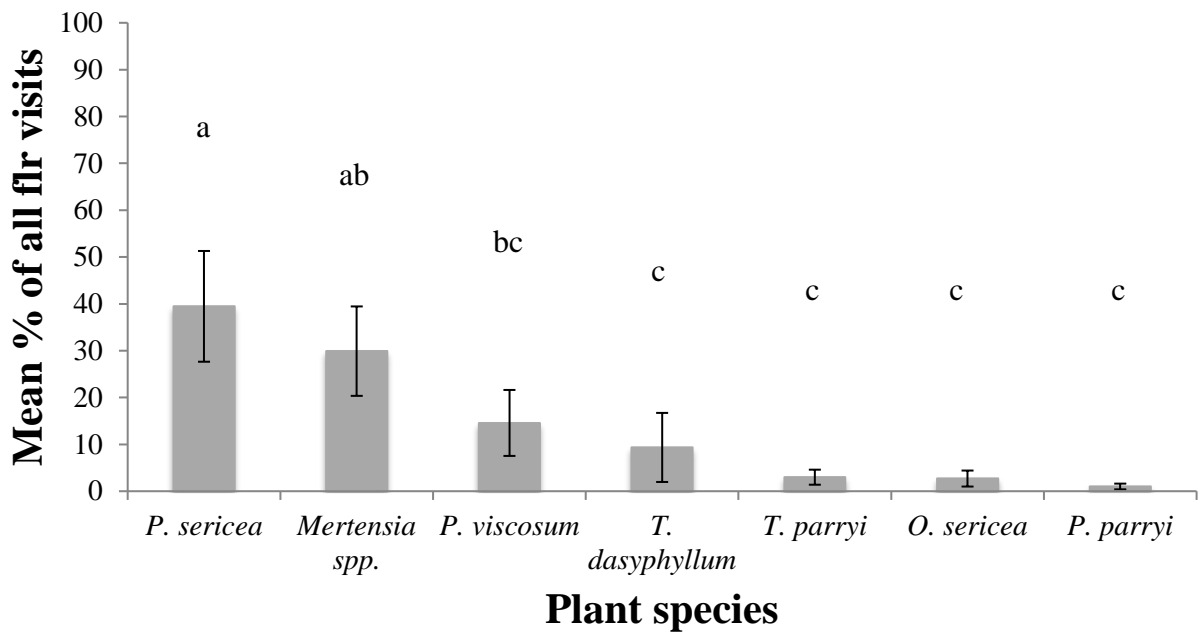
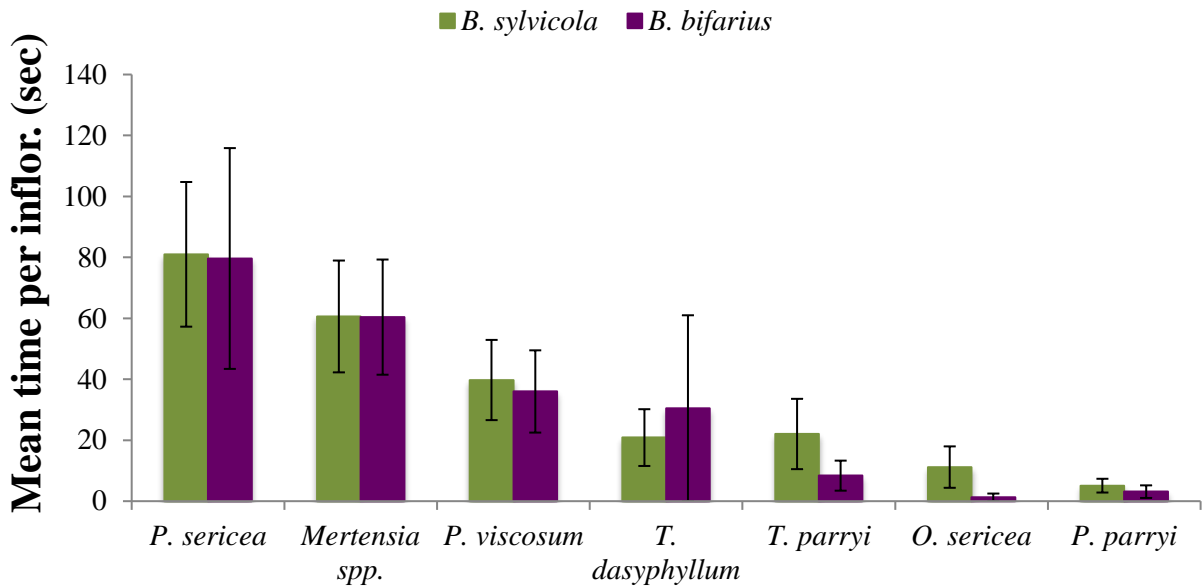


Figure 4. Mean percentage of total flower visits for each plant species during foraging bouts, compared A) between *B. sylvicola* and *B. bifarius* and B) among the plant species. Letters show significant differences among plant species (bee species pooled) at $\alpha = 0.05$.

A)



B)

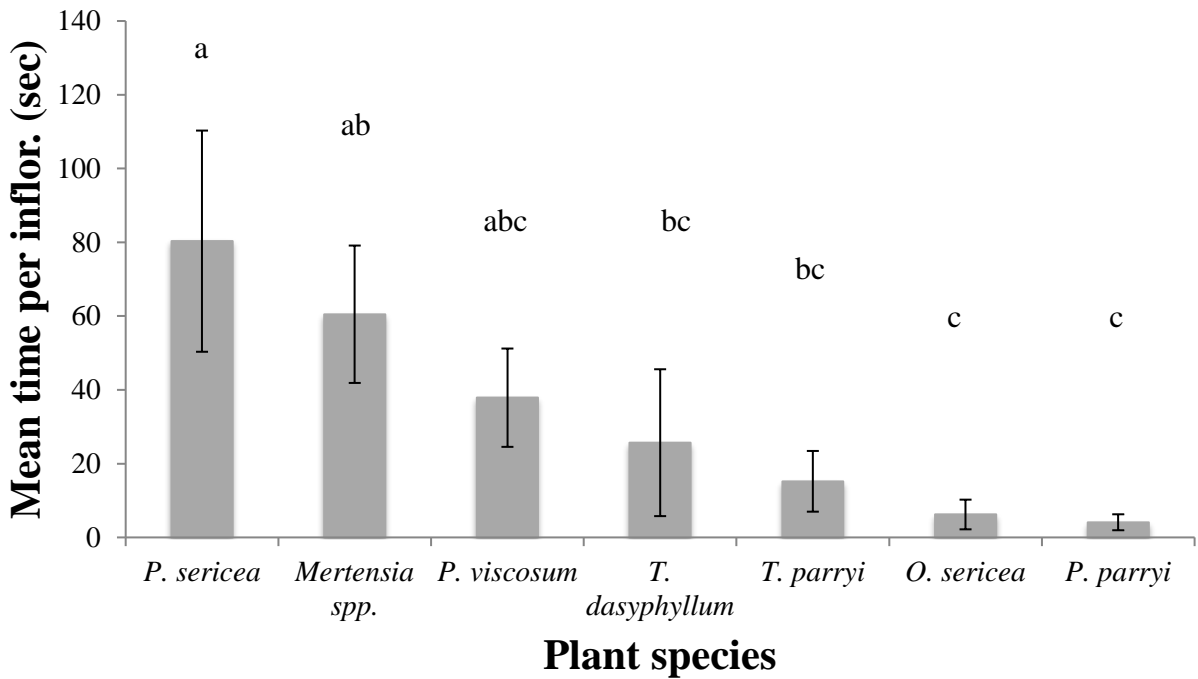
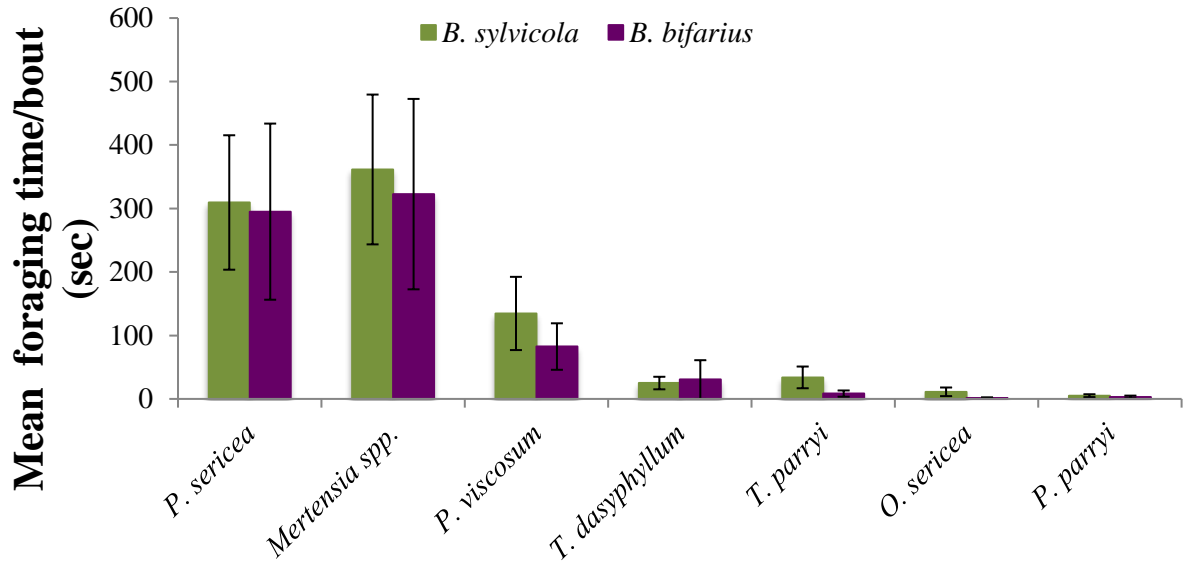


Figure 5. Mean foraging time per inflorescence during foraging bouts comparing A) the bee species (*B. sylvicola* and *B. bifarius*) and B) the plant species. Letters show significant differences among the plant species (bee species pooled) at $\alpha = 0.05$. Error bars are one standard error.

A)



B)

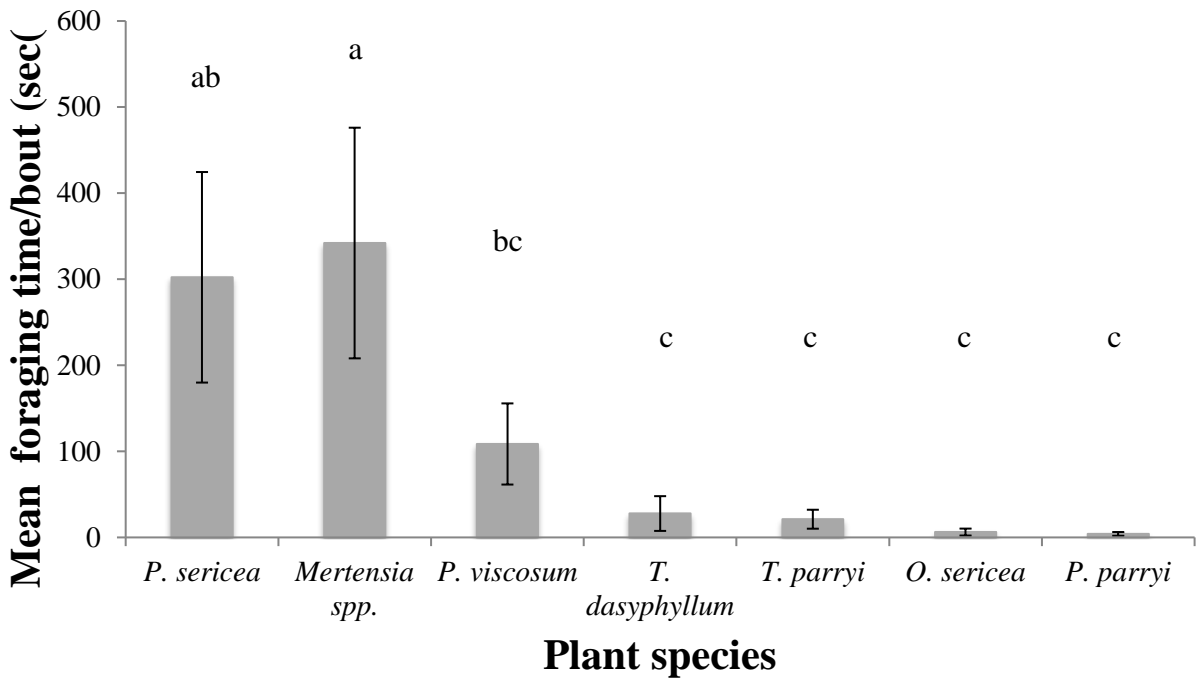


Figure 6. Mean accumulated foraging time per plant species during foraging bouts comparing A) the bee species (*B. sylvicola* and *B. bifarius*) and B) the plant species. Letters show significant differences among the plant species (bee species pooled) at $\alpha = 0.05$. Error bars are one standard error.

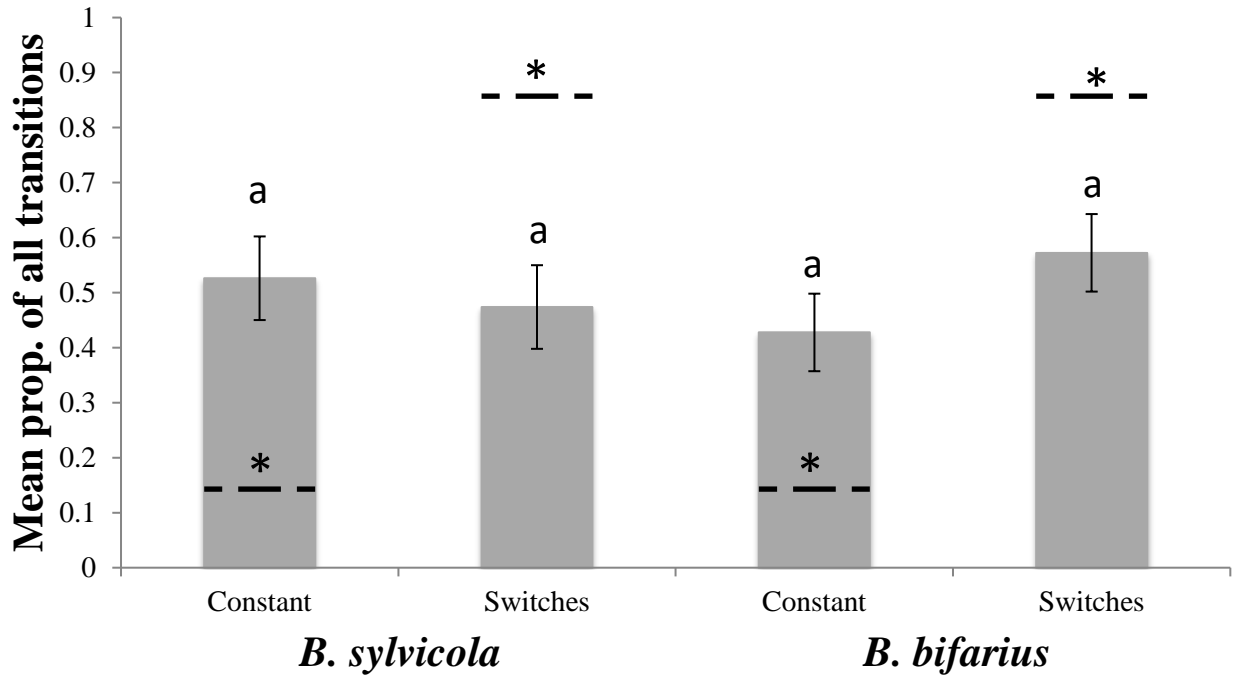
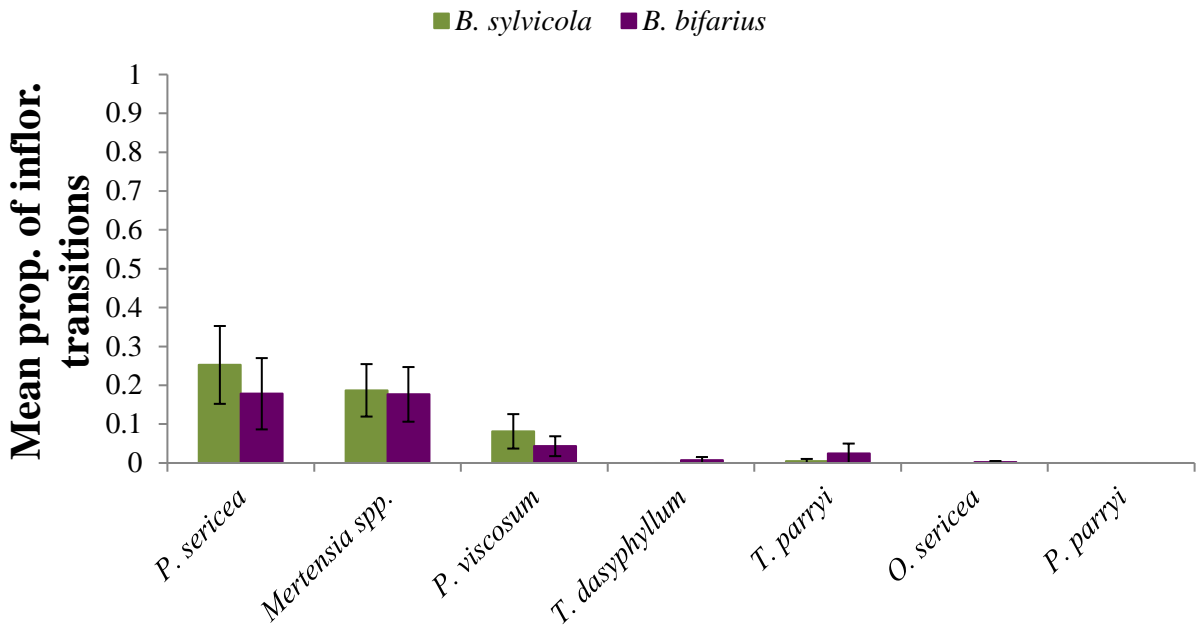


Figure 7. Overall proportion of transition types occurring during movements between inflorescences (plant species pooled). Error bars are one standard error. Dashed lines represent expected proportions based on abundance. Asterisks indicate significant differences between observed and expected proportions.

A)



B)

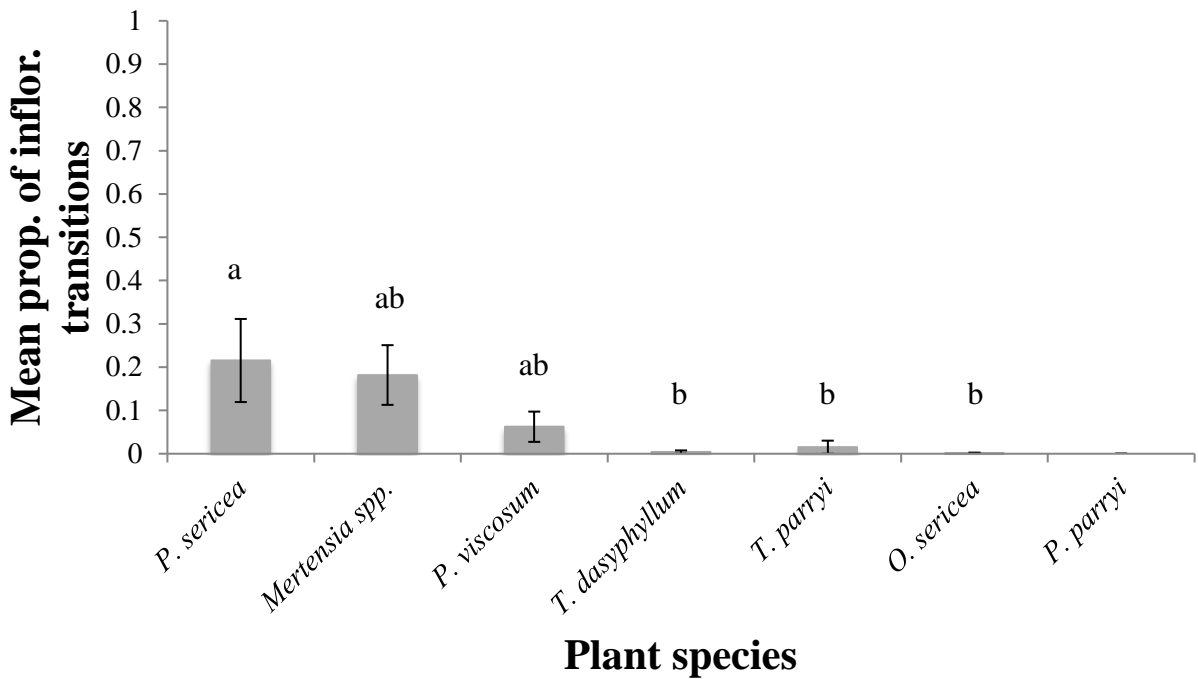


Figure 8. Proportion of same-species transitions for each flower species during foraging bouts comparing A) the bee species (*B. sylvicola* and *B. bifarius*) and B) the plant species. Letters show significant differences among the plant species (bee species pooled) at $\alpha = 0.05$. Error bars are one standard error.

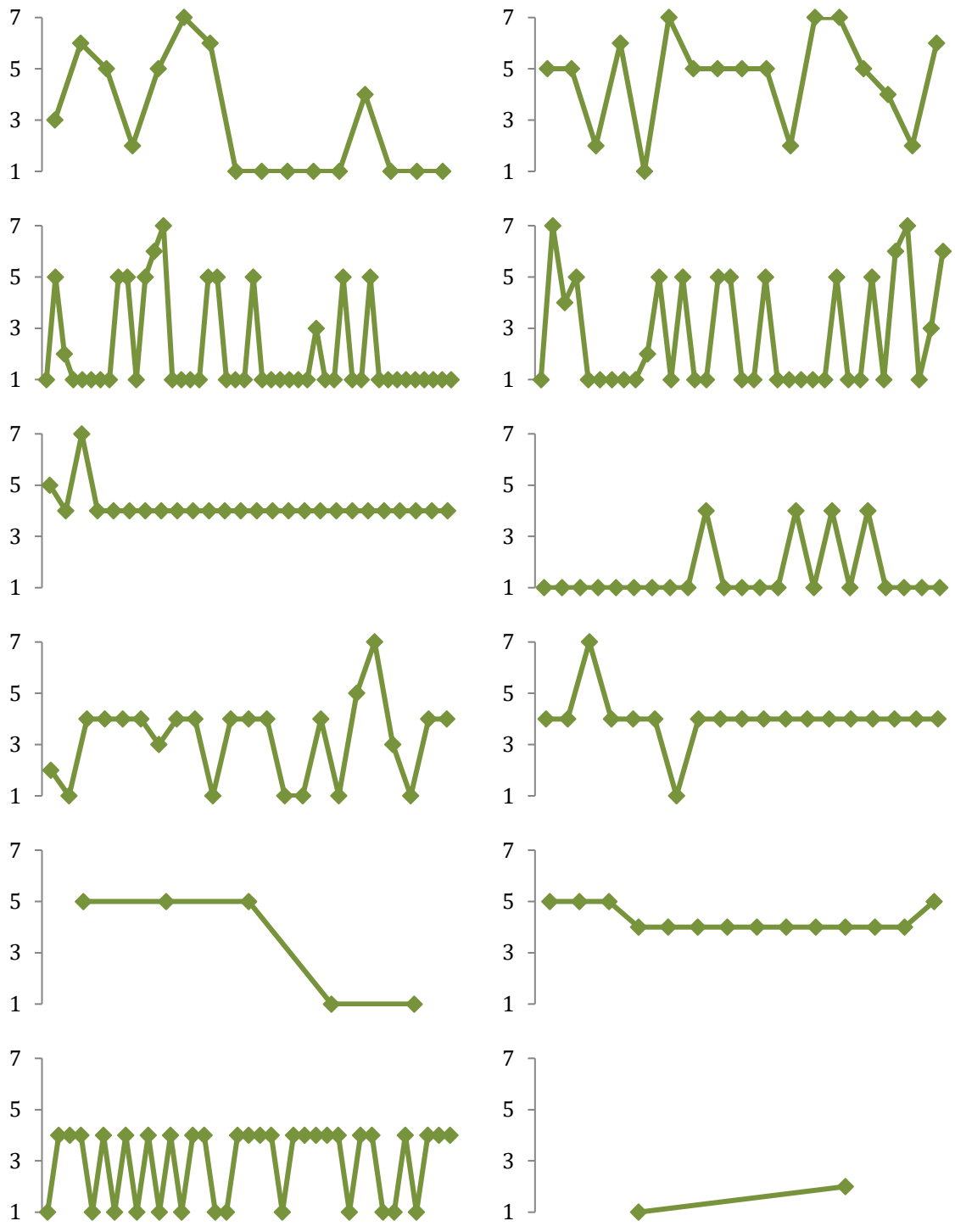


Figure 9. Variance in the sequence of foraging patterns of individual *B. sylvicola* workers ($N = 12$). 1 = *Mertensia* spp., 2 = *O. sericea*, 3 = *P. parryi*, 4 = *P. sericea*, 5 = *P. viscosum*, 6 = *T. dasyphyllum*, 7 = *T. parryi*.

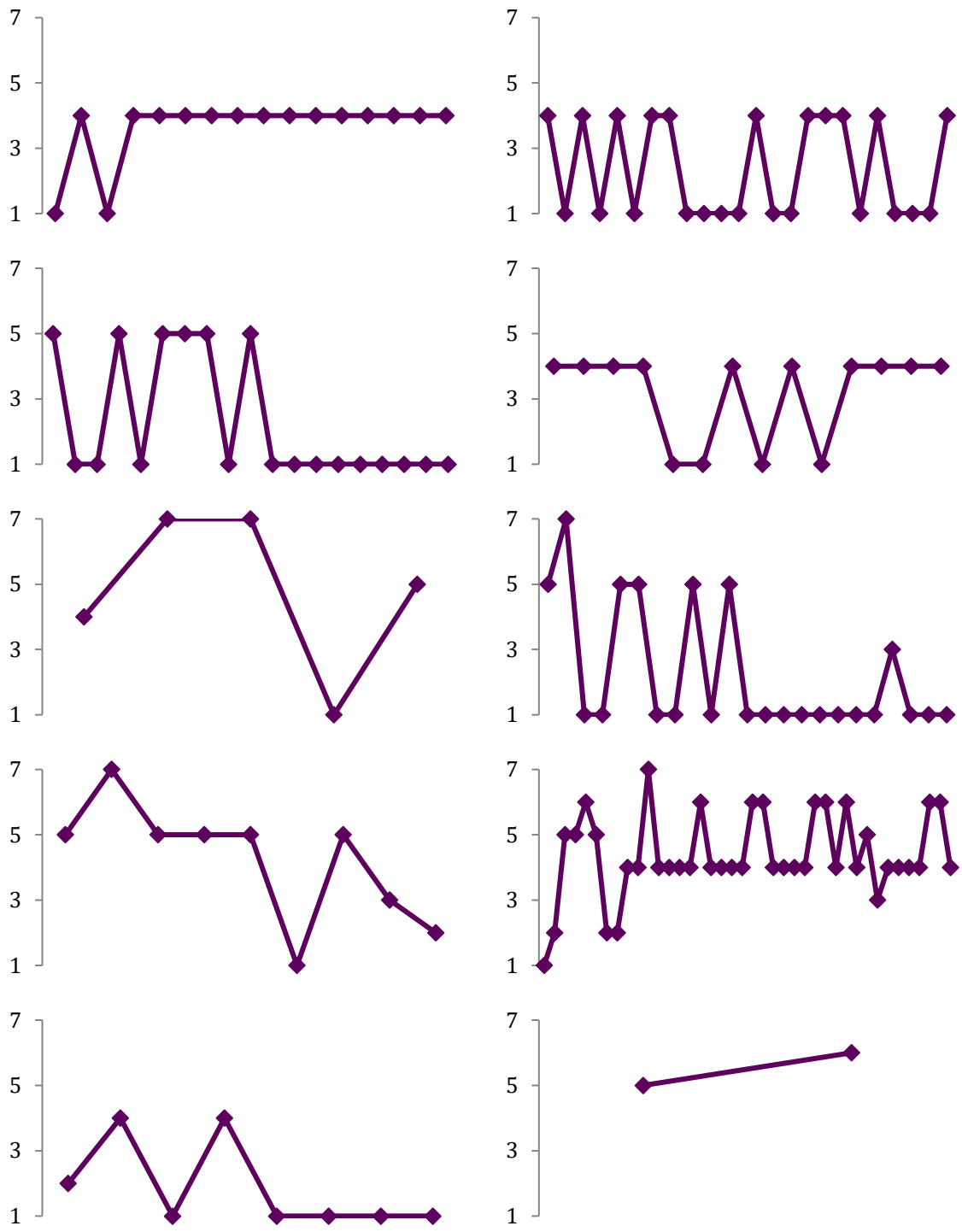


Figure 10. Variance in the sequence of foraging patterns of individual *B. bifarius* workers ($N = 10$). 1 = *Mertensia* spp., 2 = *O. sericea*, 3 = *P. parryi*, 4 = *P. sericea*, 5 = *P. viscosum*, 6 = *T. dasyphyllum*, 7 = *T. parryi*.