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Original Article

How a generalist bee achieves high efficiency of pollen collection on diverse floral resources

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Bees foraging for floral rewards are one of our most thoroughly studied examples of generalist foraging ecology. Generalist bees rely considerably on instrumental (associative) learning to acquire routines that allow them to collect nectar efficiently from diverse plant species. Although such bees must also collect pollen from diverse species, few studies have examined if and how high efficiency is achieved. We characterized how generalist bumble bees (*Bombus impatiens*) foraged effectively for pollen from diverse floral resources, by manipulating the presence of pollen and anther cues, in a series of experiments using pollen-bearing live flowers, flowers of a sterile pollenless horticultural hybrid, and artificial flowers. We show that generalist bumble bees exhibit flexible and effective pollen collection by switching between 2 routines: "scrabbling" when pollen is abundant and "sonicating" when pollen is scarce. Efficient switching between these behaviors is regulated by the interplay of 2 ubiquitous floral cues: chemical anther cues stimulating pollen collection behavior and mechanical pollen cues suppressing sonication (and eliciting scrabbling). Flexible pollen collection behavior is functional: When pollen on anthers was scarce, bees collected it at a greater rate by sonicating than scrabbling. This mechanism of behavioral flexibility likely allows generalist bees to handle diverse anther morphologies efficiently and may have facilitated the recurrent evolution of plant species that conceal pollen rewards via pored floral morphology. Whereas effective nectar foraging relies heavily on associative learning of unique routines for each flower type, a weighing of 2 types of cues regulates the flexible pollen collection mechanism we describe.

Key words: **behavioral flexibility, floral sonication, floral evolution, learning, pollen foraging, specialization.**

> "It is not obvious what characteristics the flowers […] might have that may assist in the acquisition of buzzing behavior by bees …" (Michener 1962)

INTRODUCTION

A fundamental question in behavioral ecology is how generalist animals forage efficiently on diverse resources (Loeuille 2010; Wright et al. 2010; Baudrot et al. 2016). This question has been extensively addressed for generalist pollinators foraging on diverse plant species that vary greatly in floral morphology. For instance, generalist pollinators use instrumental (associative) learning to acquire routines specific to each flower type to extract nectar efficiently (Lewis 1993; Laverty 1994; Gegear and Laverty 1995). Such learning allows a generalist pollinator to use novel species it may never have encountered in its evolutionary history, which is particularly relevant given how humans have increased the frequency of such encounters (Harmon-Threatt et al. 2009; Bartomeus et al. 2016). Moreover, pollinator learning may have facilitated the diversification of floral form (Schiestl and Johnson 2013). Yet nectar is not the only floral reward and instrumental learning may not be the only means by which flexible behavior is achieved. Pollen is in fact offered by hundreds of thousands of plant species to pollinators such as bees, beetles, flies, and some butterflies (Simpson and Neff 1981; Kevan and Baker 1983). Bees in particular must collect pollen, which is their primary source of protein (Nicolson 2011). Although it is commonly assumed that bees adjust their behavior to collect pollen effectively from a range of plant species, the nature of this assumed flexibility and its implications for floral evolution have scarcely been examined in comparison to nectar collection.

One obvious place to look for flexibility is in the 2 major means by which pollen is collected from biotically pollinated flowers of varying morphology. The pollen of most angiosperm flowers is exposed, to varying degrees, on the anthers (Figure 1g–i). Bees use their legs and mandibles to knock this pollen free via a behavior called scrabbling (Thorp 2000; see Russell and Papaj 2016 for a video). Six to ten percent of plant species (>22 000 species across >72 angiosperm families; Buchmann 1983; De Luca and

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Figure 1
Examples of poricidal and nonporicidal flowers. Three species conceal their pollen either within poricidal anthers: (a) *Senna covesii* and (b) *Solanum elaeagnifolium*; or a poricidal corolla: (c) *Pedicularis groenlandica*. Three species that partially conceal their pollen, to varying degrees: (d) *Dianella revolute*, (e) *Dodonaea microzyga*, (f) *Asterolasia grandiflora*. Three nonporicidal species display pollen openly on anthers: (g) *Deppea splendens*, (h) *Aloe cryptopoda*, (i) *Phacelia campanularia*. Photographs: (a, b, g–i): Avery Russell; (c): Walter Siegmund, licensed by CC BY-SA 3.0; (d–f): Kevin Thiele, licensed by CC BY 2.0.

Vallejo-Marín 2013) entirely conceal their pollen within specialized tube-like anthers or, less commonly, corollas (e.g., Houston and Ladd 2002; De Luca and Vallejo-Marín 2013; Corbet and Huang 2014; Figure 1a–c). Species with this so-called poricidal morphology are pollinated nearly exclusively by bees that extract the concealed pollen via a complex behavior termed floral sonication (the buzz pollination syndrome; Buchmann 1983; Russell, Leonard, et al. 2016). By rapidly contracting indirect flight muscles to produce powerful vibrations (i.e., buzzing) while biting the anthers or corolla, pollen is expelled onto the bee's body, where it can be collected (Michener 1962; Macior 1968; Buchmann and Cane 1989; Russell, Leonard, et al. 2016). Although bees must sonicate to collect pollen from poricidal species, scrabbling is observed only on nonporicidal species and never on poricidal species (Thorp 2000) and is inefficient for removing and transferring pollen from flowers that partially conceal pollen (e.g., King and Ferguson 1994; Javorek et al. 2002; Pomeroy and Fisher 2002; Figure 1d–f). We might therefore expect that bees are able to adjust their foraging behavior to collect pollen effectively from these types of flowers.

Although pollination via the buzz mechanism is generally considered a relatively specialized interaction (Buchmann 1983; De Luca and Vallejo-Marín 2013), reports of bees sonicating on flowers with nonporicidal morphology are found in the literature (Table 1). We have recently made many more observations of the same kind (Table 1). Rather than exclusively sonicating poricidal species, this behavior is performed on both poricidal and nonporicidal species, albeit to different degrees. On poricidal species, sonication is consistently performed whereas on nonporicidal species, sonication is variable in its expression. This variability is expressed at multiple scales. Not only might one bee scrabble on a nonporicidal species while another sonicates but individual bees have been observed sonicating some flowers and scrabbling on others of the same species (e.g., Buchmann 1983, 1985; Raine and Chittka 2007). Taken together, these observations suggest that generalist bees may be able to adjust when they use floral sonication versus scrabbling to effectively collect pollen from plant species with flowers of diverse morphologies (Buchmann 1985; Raine and Chittka 2007).

We devised laboratory experiments to examine the floral cues that regulate the flexibility of pollen foraging behavior (sonication and scrabbling) in the generalist bumble bee (*Bombus impatiens*) on nonporicidal species. We first tested how the amount of exposed pollen on the anthers affects expression of sonication. We then characterized 2 sets of cues, anther-based chemical cues, and pollen-based mechanical cues, which together account for the variable expression of, and switching between, sonication and scrabbling. Finally, we assessed whether the conditional expression of floral sonication on nonporicidal species resulted in effective pollen collection, thereby providing a mechanism by which bees might effectively collect pollen from plant species that partially conceal their pollen (e.g., Figure 1d–f). Our findings indicate that bee pollen foraging, like nectar foraging, is flexible. However, the flexibility we describe differs in key respects from that of nectar foraging and at least some flexibility in this pollen collection mechanism involves innately specified responses. We also discuss how this flexibility may facilitate the evolution of taxa with poricidal floral morphology from nonporicidal ancestors.

METHODS

Bees

We used 154 workers from 8 commercially obtained (Koppert Biological Systems, Howell, MI) colonies of the bumble bee *Bombus impatiens* Cresson in laboratory experiments conducted between June 2014 and April 2016. Bumble bees are globally distributed generalist pollinators and species such as *B. impatiens* forage from hundreds of angiosperm species across its range (Plowright and Laverty 1984). We used approximately equal numbers of bees from each colony for each treatment. We allowed bees to forage daily for sucrose and pollen in arenas constructed of plywood (L×W×H, 82 × 60 × 60 cm). The arenas were lit from above by 40W 4400 lumen LED lights (2 × 2 LED Ultra Thin Panel; 5000K Cool

Table 1

Nonporicidal species sonicated by bees

Family	Genus	Anthers directly accessible?	Sonicating bee(s)	Bees sonicated stamens?	Reference
Asteraceae	*Senecio vulgaris*	Yes	*Bombus edwardsii*	Yes	Buchmann (1978, 1983)
Begoniaceae	*Begonia cucullata*	Yes	*Bombus impatiens*	Yes	A. Russell, personal observation
Begoniaceae	*Begonia descoleana*	Yes	*Bombus impatiens*	Yes	This manuscript
Begoniaceae	*Begonia "Angel Wing"*	Yes	*Bombus impatiens*	Yes	A. Russell, personal observation
Begoniaceae	*Begonia "Dragon Wing"*	Yes	*Bombus impatiens*	Yes	This manuscript
Begoniaceae	*Begonia odorata*	Yes	*Bombus impatiens*	Yes	A. Russell, personal observation
Bignoniaceae	*Tecoma alata*	Yes	*Bombus impatiens*	Yes	A. Russell and D. Papaj, personal observation
Bignoniaceae	*Tecoma stans*	Yes	*Bombus sonorus*	Yes	D. Papaj and K. Mauerman, personal communication
Boraginaceae	*Alkanna orientalis*	Yes	*Anthophora pauperata*	Yes	Stone et al. (1999)
Boraginaceae	*Mertensia ciliata*	Yes	*Bombus bifarius, B. flavifrons, B. mixtus*	Yes	D. Papaj and K. Mauerman, personal communication
Boraginaceae	*Mertensia paniculata*	Yes	*Bombus mixtus, B. frigidus, Bombus spp.*	Yes	Morris (1996), A. Russell, personal observation
Boraginaceae	*Symphytum officinale*	Yes	*Bombus pascuorum*	Yes	Corbet et al. (1988)
Boraginaceae	*Phacelia tanacetifolia*	Yes	*Anthophora urbana*	Unknown[a]	Buchmann (1983, 1985)
Calophyllaceae	*Kielmeyera coriacea*	Yes	*Augochloropsis spp., Exomalopsis fulvofasciata, Xylocopa frontalis, X. hirsutissima*	Yes	Oliveira and Sazima (1990)
Calophyllaceae	*Kielmeyera speciosa*	Yes	*Augochloropsis spp., Exomalopsis fulvofasciata, Xylocopa frontalis, X. hirsutissima*	Yes	Oliveira and Sazima (1990)
Clusiaceae	*Clusia spp.*	Yes	*Augocloropsis spp.*	Yes	Kaminski and Absy (2006)
Commelinaceae	*Tradescantia pallida*	Yes	*Anthophora spp.*	Yes	A. Russell, personal observation
Cucurbitaceae	*Cucurbita foetidissima*	Yes	*Xenoglossa angustior*	Yes	Buchmann (1985)
Ericaceae	*Diospyros virginiana*	No	*Bombus impatiens, B. vagans*	Unknown[a]	C. Switzer, personal communication
Fabaceae	*Astragalus spp.*	Yes	*Eucera spp.*	Yes	Z. Portman, personal communication
Fabaceae	*Desmanthus cooleyi*	Yes	*Protoxaea gloriosa*	Unknown	Buchmann (1985)
Fabaceae	*Lupinus spp.*	No	*Bombus spp.*	Yes[c]	K. Mauerman, personal communication
Fabaceae	*Lupinus lepidus var. sellus*	No	*Bombus vosnesenskii*	Yes[c]	J. Francis, personal communication
Fabaceae	*Trifolium spp.*	Yes	*Bombus impatiens*	Unknown[a]	C. Switzer, personal communication
Fabaceae	*Vicia spp.*	Yes	*Bombus impatiens*	Unknown[a]	C. Switzer, personal communication
Fabaceae	*Coronilla varia*	No	*Bombus impatiens*	Unknown[a]	C. Switzer, personal communication
Fabaceae	*Swartzia apetala*	Yes	*Bombus spp.*	Yes	Chiara Moço and Pinheiro (1999)
Fabaceae	*Swartzia pickelii*	Yes	*Bombus spp.*	Yes	Machado and Lopes (2004)
Fabaceae	*Lespedeza bicolor "Natob Strain"*	No	*Bombus bimaculatus*	Unknown[a]	C. Switzer, personal communication
Hypericaceae	*Hypericum "Hidcote"*	Yes	*Bombus affinis, B. bimaculatus, B. griseocollis, B. impatiens, B. perplexus*	Yes	C. Switzer, personal communication
Lamiaceae	*Stachys recta*	Yes	*Anthophora furcata, A. quadrimaculata, Bombus pascuorum, B. terrestris, Rophites algirus*	Yes	Müller (1996)
Liliaceae	*Polygonatum x hybridum*	Yes	*Bombus pascuorum*	Yes	Corbet et al. (1988)
Loasaceae	*Mentzelia pumila*	Yes	*Bombus sonorus*	Yes	Linsley and Cazier (1963)
Onagraceae	*Oenothera speciosa*	Yes	*Bombus impatiens*	Yes	A. Russell, personal observation
Orchidaceae	*Thelymitra antennifera*[b]	Yes	*Lasioglossum spp.*	Yes	Bernhardt and Burns-Balogh (1986)
Orchidaceae	*Thelymitra aristata*[b]	Yes	*Lasioglossum spp.*	Yes	Bernhardt and Burns-Balogh (1986)
Orchidaceae	*Thelymitra nuda*[b]	Yes	*Lasioglossum spp.*	Yes	Bernhardt and Burns-Balogh (1986)
Orobanchaceae	*Castelleja spp.*	Yes	*Bombus sps*	Yes	D. Papaj and K. Mauerman, personal communication
Orobanchaceae	*Melampyrum pratense*	Yes	*Bombus lucorum, Megachile willughbiella*	Yes	Meidell (1944)
Paeoniaceae	*Paeonia spp.*	Yes	*Bombus spp.*	Yes	A. Russell, personal observation
Papaveraceae	*Papaver rhoeas*	Yes	*Bombus terrestris*	Yes	Raine and Chittka (2007)
Papaveraceae	*Argemone arizonica*	Yes	*Xylocopa californica*	Yes	Buchmann (1985)
Papaveraceae	*Argemone spp.*	Yes	*Bombus sonorus, Xylocopa spp.*	Yes	D. Papaj, personal communication; A. Russell, personal observation
Plantaginaceae	*Chelone Glabra*	Yes	*Bombus vagans, Hylaeus annulatus*	Yes	Richardson and Irwin (2015)
Plantaginaceae	*Penstemon cyananthus*	Yes	*Osmia brevis*	Yes	Cane (2014)

Table 1

Continued

Family	Genus	Anthers directly accessible?	Sonicating bee(s)	Bees sonicated stamens?	Reference
Plantaginaceae	*Penstemon radicosus*	Yes	*Osmia brevis*		Cane (2014)
Plantaginaceae	*Penstemon strictus*	Yes	*Bombus nevadensis, Osmia brevis*	Yes	Cane (2014)
Ranunculaceae	*Delphinium spp.*	Yes	*Bombus spp.*	Unknown[a]	D. Papaj and K. Mauerman, personal communication
Ranunculaceae	*Aconitum spp.*	Yes	*Bombus spp.*	Yes	K. Mauerman, personal communication
Ranunculaceae	*Aquilegia caerulea*	Yes	*Bombus spp.*		D. Papaj, personal communication
Ranunculaceae	*Aquilegia chrysantha*	Yes	*Bombus occidentalis*	Unknown	Pellmyr (1985)
Ranunculaceae	*Aquilegia formosa*	Yes	*Bombus spp.*	Yes	A. Russell, personal observation
Ranunculaceae	*Cimicifuga arizonica*	Yes	*Bombus huntii, B. occidentalis*	Yes	Pellmyr (1985)
Rosaceae	*Potentilla recta*	Yes	*Bombus ternarius, B. terricola*	Yes	Heinrich (1976)
Rosaceae	*Potentilla gracilis*	Yes	*Bombus vosnesenskii*	Yes	J. Francis, personal communication
Rosaceae	*Prunus dulcis*	Yes	*Bombus spp.*	Yes	Thomson and Goodell (2001)
Rosaceae	*Fallugia paradoxa*	Yes	*Bombus pennsylvanicus*	Yes	Buchmann (1985)
Rosaceae	*Rosa* "*Bucbi*"	Yes	*Bombus bimaculatus, B. impatiens*	Yes	C. Switzer, personal communication
Rosaceae	*Rosa californica*	Yes	*Bombus edwardsii, B. vosnenskii*	Yes	Buchmann (1978, 1983)
Rosaceae	*Rosa multiflora*	Yes	*Bombus bimaculatus, B. impatiens*	Yes	C. Switzer pers. comm.
Rosaceae	*Rosa nitida*	Yes	*Bombus ternarius*	Yes	Heinrich (1976)
Rosaceae	*Rosa rugosa*	Yes	*Bombus terrestris*	Yes	Dobson et al. (1999)
Rosaceae	*Rosa virginiana*	Yes	*Bombus pennsylvanicus*	Yes	Buchmann (1983)
Rosaceae	*Rubus odoratus*	Yes	*Bombus bimaculatus, B. impatiens*	Yes	C. Switzer, personal communication
Rosaceae	*Rubus parviflorus*	Yes	*Bombus occidentalis*	Yes	Buchmann (1983)
Solanaceae	*Physalis philadelphica*	Yes	*Bombus impatiens*	Yes	C. Switzer, personal communication
Solanaceae	*Physalis longifolia*	Yes	*Colletes latitarsis*	Yes	Paine and Roulston (2012)
Theaceae	*Stewartia sinensis*	Yes	*Bombus perplexus*	Yes	C. Switzer, personal communication
Verbenaceae	*Callicarpa cathayana*	Yes	*Bombus impatiens*	Yes	C. Switzer, personal communication
Verbenaceae	*Callicarpa dichotoma*	Yes	*Bombus bimaculatus, B. impatiens*	Yes	C. Switzer, personal communication
Verbenaceae	*Callicarpa japonica*	Yes	*Bombus impatiens*	Yes	C. Switzer, personal communication
Zygophyllaceae	*Kallstroemia grandiflora*	Yes	*Bombus impatiens*	Yes	A. Russell, personal observation
Zygophyllaceae	*Guaiacum coulteri*	Yes	*Anthophora spp.*	Yes	A. Russell, personal observation

List comprises 26 families, 47 genera, and 73 species of angiosperms.

[a] Possible that sonicates to push deeper into the flower to access nectar?. [b] Rewardless. [c] Pushes open banner petal and then sonicates anthers.

White, James Industry) set to a 14 h:10 h light: dark cycle. Colonies had access to ad libitum 2M sucrose solution and pulverized honey bee-collected pollen (Koppert Biological Systems) within the foraging arena. Sucrose solution was dispensed via braided cotton wicks that extended into vials. Pollen was dispensed via custom-made feeders constructed of chenille fibers glued within 40-dram vials (Russell and Papaj 2016). Bees always scrabbled for this pollen; further, of bees naive to pollen foraging that were observed on their first few visits to feeders, none sonicated. Bees were also naive to the pure (i.e., hand-collected) pollen used in experiments: These pure pollen types were not present in the honey bee-collected pollen (Kim Skyrm, Koppert Biological Systems, personal communication), which moreover does not resemble pure pollen, as honey bee-collected pollen is adulterated with up to 60% by mass regurgitated crop sugars (Russell and Papaj 2016).

Plants and flowers

We used freshly clipped flowers with mature anthers from 8 poricidal *Solanum houstonii* Martyn (synonym *S. tridynamum*) plants, 2 nonporicidal *Begonia x hybrida* ("Dragonwing"), and 10 nonporicidal *Begonia descoleana* L. B. Sm. and B. G. Schub plants raised in a university greenhouse and fertilized weekly (Miracle Gro, NPK = 15-30-15). The anthers of *B. hybrida* are sterile (pollenless) and made it possible to precisely control pollen cues presented to bees. Conversely, *B. descoleana*, one of the species crossed to make the *B. hybrida* hybrid, is fertile and presents pollen on its anthers (its only floral reward). Likewise, *S. houstonii* only offers pollen to its pollinators. A given trial used an approximately equal number of flowers from each plant. We used approximately 2000 flowers in experiments.

General experimental protocol

All trials took place in a foraging arena (L×W×H, 82 × 60 × 60 cm) painted gray on floor and sides. In trials, freshly clipped flowers were displayed horizontally (their natural orientation) on custom-built water tubes (see Russell, Golden, et al. 2016), to prevent desiccation. The water tubes were Velcro-mounted on the arena wall, facing the flight chamber's nest entrance. Flowers were arranged on the wall in a 3 × 3 Cartesian grid with each water tube spaced 7 cm apart in the horizontal and vertical axes of the grid. Fresh flowers were used at the start of every trial and for each bee. Flowers were never reused across trials. We systematically alternated treatments that belonged to a given experiment (for experiment 1, the subexperiments) in time to control for effects of day and time of day on behavior.

To initiate a trial, a single flower-naive individual was introduced into the arena. Bees readily visited all types of flowers, live and artificial. We recorded landings made by the bee on the flowers. A landing was defined as taking place when a bee touched the flower with at least 3 legs simultaneously. Three types of landings were noted: landings with sonication buzzes, landings with only scrabbling, and landings without scrabbling or sonication buzzes. On rare occasions, in experiment 1 and 3 only, bees switched from scrabbling to sonicating on the same landing: These rare landings were also classified as landings with sonication buzzes. Sonication buzzes were identified by their distinctive sound and the stereotyped posture of the bees on the flowers (see Russell et al. 2016 for extended description) and occurred only after a bee had landed. Scrabbling involved the bee manipulating the anthers with the mandibles and legs (see Russell and Papaj 2016 for videos and extended description). Virtually all flower visits where flowers presented pollen, small 20 μm diameter cellulose powder, or 20 μm diameter plastic microspheres (Supplementary Figure S2) involved collection attempts (either by sonicating or scrabbling). Bees never scrabbled when the anthers (surrogate or live) were bare or presented large 180 μm diameter cellulose powder. We tracked whether bees landed on previously unvisited flowers or on previously visited flowers to allow comparison of behavior between these 2 categories. Bees nearly always visited all flowers in an array at least once. Bees were allowed to make up to 20, 30, or 40 landings in an array (depending on experiment), after which the trial was terminated. A trial was sometimes terminated before the maximum number of landings if the bee did not forage for a period of 5 min. Most bees (71%) made the maximum number of allowed landings (bees visiting unrewarding arrays tended not to complete the maximum number of allowed landings) and all bees were included in analyses. We euthanized each bee and all its flowers after the bee completed its trial: A given bee was tested only once in a single foraging bout (i.e., a single trial).

To facilitate recording of behavior, video for all trials was captured at 30fps with a high-definition digital camcorder (Canon VIXIA HF R400) positioned in front of the array. Audio was input to the camcorder using an external microphone (33–3013 Lavaliere Microphone, RadioShack) attached to the center of floral arrays. A Zoom H2 Handy Recorder (ZOOM Corporation) was used to amplify and verify sonication buzzes in ongoing trials.

Experiments

Experiment 1

Here, we sought to determine whether bees buzzed the flowers of nonporicidal species and the role of pollen availability in mediating this response. This experiment (composed of 3 subexperiments) used 54 bees from 7 colonies and each bee was presented with an array of 9 flowers. In experiment 1a, we used the natural species, *B. descoleana* to characterize the normal pollen collection behavior of bees; in experiments 1b and 1c, we used the pollenless hybrid, *B. hybrida*, supplemented with controlled amounts of pollen to precisely determine how pollen availability affected pollen collection behavior.

In experiment 1a, bees were each allowed 40 landings. We split bees in experiment 1b into 2 treatments and allowed each bee 20 landings. In one treatment, *B. hybrida* flowers were each supplemented with 1.5 mg pistachio pollen (*Pistacia vera*; Pollen Collection and Sales; Lemon Cove, CA) added to their anthers (mean ± SE: 1.52 ± 0.02). Pollen was stored at −18 °C and weighed using a Sartorius Analytic Balance (Data Weighing Systems, Inc.) to the nearest 0.1 mg. We used pistachio pollen in experiment 1b, because it was available in large quantities, unlike *Begonia* pollen. In the other treatment, *B. hybrida* flowers were pollenless (their natural state). In experiment 1c, bees were split into 3 treatments and each bee allowed 40 landings. Each treatment varied by the quantity of added pollen: flowers presented 1, 2, or 4 mg of pollen on their anthers (mean ± SE: 1.0 ± 0.01; 2.0 ± 0.01; 4.0 ± 0.01). We discarded one bee that collected pollen only 5 times. The amount of pollen added to flowers was within the range that live flowers contain (see Russell and Papaj 2016).

Experiment 2

Here, we wanted to determine whether chemical anther cues of nonporicidal *B. hybrida* and poricidal *S. houstonii* elicited sonication by bees. To this end, we used surrogate flowers made with real corollas and surrogate foam anthers. Use of the live flower's corolla allowed us to assess whether an anther extract applied to the surrogate anthers alone elicited sonication. This experiment used 22 bees from 5 colonies.

We made surrogate flowers by cutting off and discarding the stamens from the corollas of flowers (Figure 4a, b). Pure pentane or a pentane anther extract was applied to Yellow Fibrecraft Foam (Jo-Ann Stores, LLC.), cut into cuboids (L×W×H, 1.4 × 0.2 × 0.2 cm). These surrogate anthers were hot-glued to the corollas. Surrogate flowers were arranged in a 3 × 3 grid without a central flower (8 total targets), with pentane control and extract-treated targets alternated by position. See Supplementary Material for details. We split bees into 2 treatments and allowed each bee 30 landings. Arrays and extracts were made from *B. hybrida* in one treatment and from *S. houstonii* in the other treatment.

Experiment 3

We investigated whether mechanical stimulation by odorless particles, similar in size to pollen, mediated pollen collection behavior similarly to pollen. This experiment (composed of 2 subexperiments) used *B. hybrida* and 38 bees from 2 colonies; each bee was presented with an array of 9 flowers.

In experiment 3a, bees were allocated to each of 3 treatment groups and each bee allowed 40 landings. Each treatment varied by the quantity of supplemental small cellulose powder (20 μm Cellulose Microcrystalline Powder; Sigma Aldrich, St. Louis, MO): flowers presented 1, 2, or 4 mg of cellulose powder on their anthers (mean ± SE: 0.98 ± 0.03; 2.0 ± 0.02; 3.99 ± 0.02). We discarded one bee that landed on flowers but did not collect any cellulose.

We examined pollen and cellulose powder under a compound microscope to determine whether cellulose powder physically resembled pollen and thus was a good proxy. We found that unlike pistachio, cherry (*Prunus avium*; Pollen Collection and Sales; Lemon Cove, CA), or *Begonia* pollen, 20 μm diameter cellulose powder (180 μm diameter microcrystalline cellulose powder less so) were not of uniform size or shape (Supplementary Figure S2). However, in experiment 3b, assays comparing foraging behavior on arrays of flowers offering either 2 mg 20 μm cellulose powder (results from experiment 3a, which were of bees that had been concurrently tested) or 2 mg uniformly shaped and sized 20 μm diameter polystyrene microspheres (Supplementary Figure S2c; 20 μm Polystyrene DVB Microspheres; Thermo Scientific, Fremont, CA) confirmed using cellulose did not affect patterns of behavior (Figure 5, Supplementary Figure S3d; Welch 2 sample *t*-test: proportion of visits that involved sonication, 20 μm cellulose × 20 μm microspheres, $t_{11.991} = -0.0428$, $P = 0.967$). We discarded 2 bees which landed on flowers but did not collect microspheres or cellulose, respectively.

Experiment 4

Here, we wanted to find out whether pollen collection behavior was mediated by pollen-sized particles specifically. This experiment used *B. hybrida* and 17 bees from 2 colonies; each bee was presented with an array of 9 flowers.

Bees were allocated into each of 2 treatments and each bee allowed 40 landings. In one treatment, flowers each had 2 mg of 180 µm diameter microcrystalline cellulose (Avicel PH-200 NF; FMC Corporation, Philadelphia, PA) added to their anthers (mean ± SE: 2.0 ± 0.02). These cellulose particles are larger than typical pollen offered by biotically pollinated plants and collected by bees (mean diameter: 34 µm; Roberts and Vallespir 1978). In the other treatment, flowers were bare. We discarded one bee that made only 2 landings.

Experiment 5

Here, we investigated whether bees that collected pollen from nonporicidal flowers by sonicating were able to collect it at a faster rate than those that collected pollen by scrabbling. This experiment used artificial flowers and 23 bees from 2 colonies. We used artificial flowers to control the amount of pollen presented and whether bees would sonicate or scrabble.

Bees were allocated into each of 2 treatments: in one treatment bees only scrabbled for pollen and in the other treatment bees only sonicated for pollen. Each bee was allowed 2 landings to each artificial flower (8 pollen collecting landings per bee), whereupon, the flower was removed from the foraging arena. While removing the flower, the bee continued to visit flowers and did not exhibit signs of being threatened by our activity, such as aggressive behavior or attempts to escape from the arena.

The artificial flower's corolla (diameter: 2.8 cm) was made from purple Fibrecraft Foam, cut with a puncher (Medium Plum Blossom; Punch Bunch Inc., CO). The surrogate anthers were of the same design used in experiment 2 and were hot-glued onto the center of the corolla, such that 2 of the rectangular surfaces faced up at an angle simultaneously (Figure 6a). Cherry pollen (mean mg per anther ± SE: 0.63 ± 0.05, $N = 15$ measurements) was spread evenly onto those 2 surfaces.

Flower-naive bees scrabbled for pure cherry pollen on artificial flowers and never sonicated. To ensure that bees in the comparison treatment only ever sonicated on artificial flowers, we trained bees to sonicate in response to a floral odor, prior to visiting artificial flowers. In this training, bees were allowed to make 8 rewarding visits to an array of 4 poricidal *S. houstonii* flowers (pollen is their only floral reward); sonication is the only behavior these bees use to extract pollen from the poricidal anthers. After this training, bees were labeled with unique color combinations of acrylic paint (painting does not appear to affect pollen collection behavior; Switzer and Combes 2016) and returned to their colony. After depositing its pollen loads the bee was tested by allowing it to collect cherry pollen from artificial flowers treated with *S. houstonii* anther extract. Bees tested in this way always sonicated to collect pollen. We discarded one bee that completed the training phase but did not visit the surrogate flowers in the testing phase. Training compelled bees to sonicate but should not have made bees more effective sonicators (and thus more efficient pollen collectors than if they had never sonicated previously): Sonication is fully effective at first expression and modified little with experience (see Russell et al. 2016 for details). Bees had extensive experience scrabbling, but both set of bees were naive to foraging on surrogate anthers and pure pollen.

To calculate the rate of pollen collection per bee, we divided the amount of cherry pollen collected by the total time on the flower (defined by the start of the flower visit until the end of a visit, summed across all 8 bee landings). Total time on flower was estimated from video footage viewed frame-by-frame with Avidemux software (fixounet@free.fr). The end of a visit was defined as the first video frame in which the bee no longer contacted the flower with its legs. To determine the amount of pollen collected, we euthanized each bee immediately after it completed its trial and removed and weighed its pollen load. Because the rate of pollen collection depends on its individual currency components (i.e., the amount of cherry pollen collected and time on the flower), we also analyze these components for differences among treatments.

Data analyses

All data were analyzed using R v.3.2.0 (R Development Core Team 2010).

Variables being analyzed were a composite of each bee's responses; specifically they were proportion variables. We analyzed differences in the proportion of landings that included sonications, in the proportion of landings with sonication per flower type, and in the rate of cherry collection. When analyzing differences between 2 treatments (or 2 flower states) we used *t*-tests if assumptions of normality and equal variance were met (using Shapiro–Wilk and *F*-tests, respectively, in the mgcv package: Wood 2016) or, otherwise, Wilcoxon-signed rank tests. Where we were interested in patterns across 3 treatments, we used one-way analysis of variance (Anova) using the aov() function in R. In cases of significant effects, we ran Tukey's post hoc test, using the TukeyHSD() function in R, to determine which pairs were significant.

RESULTS

Experiment 1: bumble bees sonicate the anthers of nonporicidal flowers but pollen suppresses this response

Foraging bumble bees flexibly switched between sonication and scrabbling between floral visits when foraging from nonporicidal flowers (Figure 2). Bees rarely sonicated previously unvisited flowers of *B. descoleana*, a nonporicidal species that offers only pollen as a reward but readily sonicated flowers that had been depleted of pollen during previous visits (Figure 3a, b; Wilcoxon matched-pairs rank sum tests: first landings vs. repeated, $V = 36$, $P < 0.0143$).

This result is probably an effect of a difference in pollen availability and not of correlated factors, such as a mark left by the bee on flowers: bees readily sonicated flowers of *B. hybrida*, a nonporicidal, pollenless hybrid (Figure 3c) and sonicated them on a higher proportion of visits relative to visits to *B. hybrida* flowers whose anthers had been supplemented with pollen (Figure 3d, e; Welch 2 sample *t*-test: pollen versus pollenless, $t_{19.087} = -7.81$, $P < 0.0001$). Even naive bees on their initial visit only sonicated pollenless flowers and only scrabbled pollen-supplemented flowers (Supplementary Figure S3a). Bees sonicated pollen-supplemented flowers more frequently after they had been depleted of added pollen during previous visits (Figure 3f; Wilcoxon matched-pairs rank sum tests: first landings vs. repeated, $V = 45$, $P < 0.0092$). Additionally, bees sonicated flowers

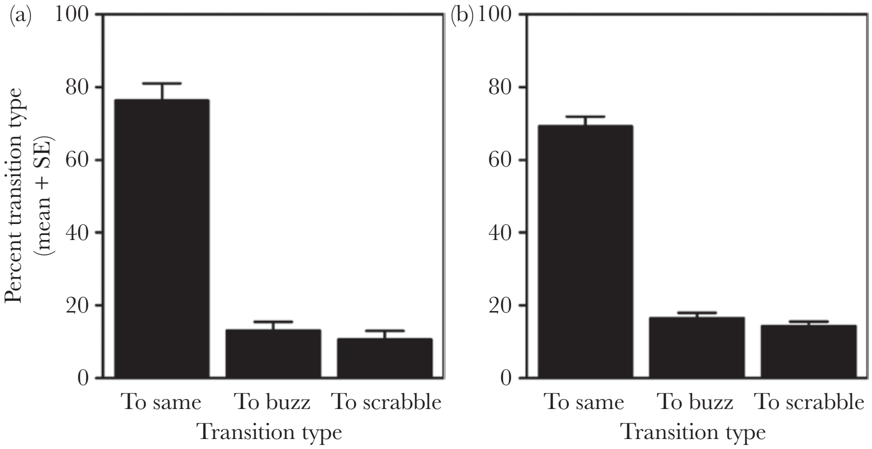

Figure 2

Mean percentage of landings (± SE) by *Bombus impatiens* that involved bees either staying with the same collection behavior or switching between collection routines from one to the next floral visit. Bees either switched from using sonication on one visit to using scrabbling on the next visit ("to scrabble"), from using scrabbling on one visit to using sonication on the next visit ("to buzz"), or did not switch collection behaviors from one to the next visit ("to same"). (a) Bees foraging on *Begonia descoleana*; (b) Bees foraging on *B. hybrida* supplemented with 1 mg of pistachio pollen. $N = 9$ and 7 bees for *B. descoleana* and *B. hybrida* treatments, respectively; data from experiment 1.

more frequently that were initially enriched with smaller amounts of pollen (Figure 3g; Anova: pollen amount effect: $F_{2,19} = 28.18$, $P < 0.0001$). In all cases, bees landing on pollen-supplemented flowers collected that pollen (Supplementary Figure S1b). Bees never scrabbled on bare flowers.

Experiment 2: chemical extracts of anthers from nonporicidal or poricidal species elicit sonication by bumble bees

Bees sonicated on a greater proportion of visits to surrogate flowers treated with a pentane extract of live anthers versus on visits to surrogate flowers treated with a pentane control (Figure 4). Surrogate flowers consisted of live corollas bearing artificial foam anthers treated with the pentane anther extract or pentane control. In both treatments (surrogates and extract either made from nonporicidal, rewardless *B. hybrida*, Figure 4a, or from poricidal *Solanum houstonii*, Figure 4b), visits to anther extract-treated surrogates resulted in sonication significantly more often than visits to surrogates treated with a pentane control (Wilcoxon matched-pairs rank sum tests, extract vs. pentane: Figure 4c; *B. hybrida*: $V = 60$, $P < 0.019$; Figure 4d; *S. houstonii*: $V = 5.44$, $P < 0.0039$). All bees but one made their first buzz on an anther extract-treated surrogate flower. Bees never sonicated corollas and never scrabbled on surrogate flowers.

Experiment 3: pollen-like mechanosensory stimuli suppress sonication

Patterns of behavior by bees collecting plastic microspheres and cellulose powder (both 20 µm diameter particles) similar in size to pollen were similar to patterns of bee behavior observed in experiment 1 (Figure 5). Bees were more prone to sonicate *B. hybrida* flowers depleted of plastic or cellulose in previous visits (Figure 5b; for both treatments, Wilcoxon matched-pairs rank sum tests: first landings vs. repeated: $V = 28$, $P < 0.016$) and were more prone to sonicate flowers supplemented with less cellulose (Figure 5c; Anova: amount of cellulose treatment effect: $F_{2,19} = 40.08$, $P < 0.0001$). Bees visiting flowers supplemented with 20 µm plastic or cellulose particles always collected the particles (Figure 5a, Supplementary Figure S1c). We used cellulose because it elicited equivalent patterns of behavior as plastic (Figure 5, Supplementary Figure S3d) and much larger cellulose particles stick to the anthers (unlike larger plastic microspheres).

Experiment 4: suppression of sonication is contingent on pollen or particle size

Cellulose particles (180 µm diameter) much larger than pollen typically collected by generalist bees (Roberts and Vallespir 1978) did not suppress sonication and bees did not scrabble for these particles, as they did for the 20 µm cellulose powder, plastic microspheres, or pollen. Specifically, the proportion of landings involving sonication by bees visiting *B. hybrida* flowers that presented large cellulose particles on their anthers was not significantly different from that by bees alighting on unmanipulated bare flowers (Figure 5f; Wilcoxon matched-pairs rank sum tests: 180 µm cellulose versus pollenless, $W = 30$, $P = 0.873$).

Experiment 5: bees removed pollen at a higher rate by sonicating than by scrabbling when nonporicidal flowers presented small amounts of pollen

Because bees switched from scrabbling to sonicating when pollen was depleted on live flowers, we investigated the benefit of using one versus the other routine when pollen was depleted. Bees that only sonicated pollen-supplemented artificial (all foam) nonporicidal flowers (Figure 6a) collected pollen at a significantly higher rate than bees that only scrabbled for pollen (Figure 6b; Welch 2 sample *t*-test: collection rate, scrabble vs. buzz: $t_{17.123} = 2.4693$, $P < 0.025$; $N = 11$ bees per treatment). The difference corresponds to a 52% higher pollen collection rate for sonicating bees compared to scrabbling bees.

Neither the length of time foraging on flowers nor the amount of pollen collected alone accounted for the significant difference in the higher collection rate by sonication over scrabbling. The mean amount of time spent visiting artificial flowers supplemented with cherry pollen did not differ between treatments where bees only sonicated versus treatments where bees only scrabbled (Wilcoxon: time foraging, scrabble vs. buzz: $W = 42$, $P = 0.243$, mean seconds ± SE:

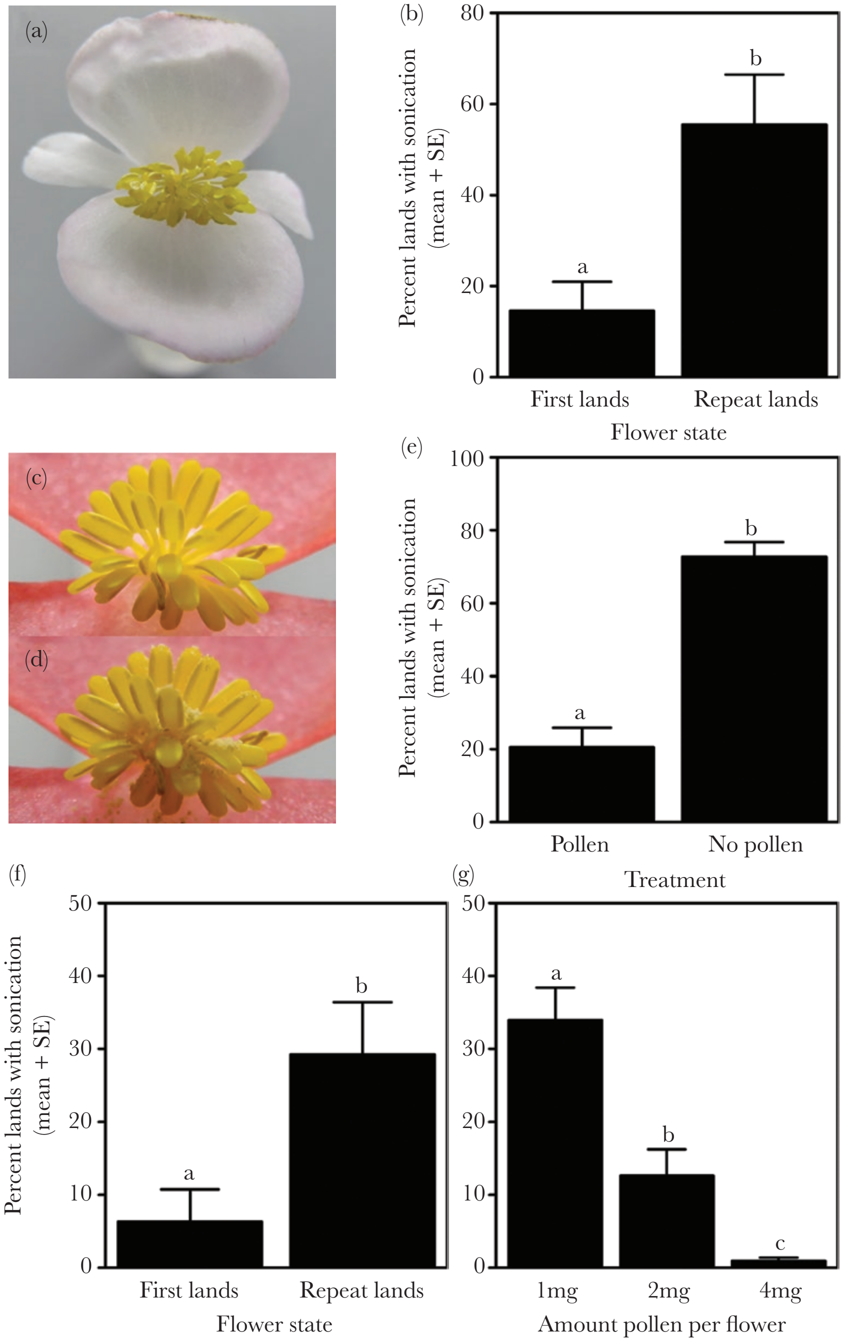

Figure 3

Mean percentage landings (± SE) by *Bombus impatiens* resulting in sonication in treatments where the availability of pollen presented by *Begonia descoleana* and *B. hybrida* was varied. (a) *B. descoleana* and (c) *B. hybrida* in its natural state and (d) *B. hybrida* supplemented with 2 mg pollen. Mean percentage landings resulting in sonication (b) of *B. descoleana* flowers on the first landings versus repeat landings; (e) of *B. hybrida* pollenless flowers or pollen-supplemented flowers; (f) of pollen-supplemented flowers on the first landings to each flower in an array versus repeat landings; and (g) of flowers initially supplemented with 1, 2, or 4 mg of pollen. (b) $N = 9$ bees for *B. descoleana* treatment. (e, f) $N = 12$, 11 bees for the *B. hybrida* pollenless and pollen-supplemented treatment, respectively. (g) $N = 7$, 7, and 8 bees for the 1, 2, and 4 mg pollen treatments, respectively. Different letters above bars indicate significant differences at $P < 0.05$ according to a Wilcoxon test, *t*-test, or Tukey's post hoc test.

sonicated, 58.79 ± 5.36; scrabbled, 78.50 ± 12.55, $N = 11$ bees per treatment). Likewise, the amount of cherry pollen collected did not differ for sonicating bees versus scrabbling bees (Wilcoxon signed rank tests: amount of pollen collected, pollen collected, scrabble vs. buzz: $W = 87$, $P = 0.0869$; mean mg ± SE: sonicated, 2.15 ± 0.30; scrabbled, 1.47 ± 0.15; $N = 11$ bees per treatment).

Results summary

Our results demonstrate that bees exhibited flexible pollen collection behavior by switching between routines (floral sonication and scrabbling). Anther chemical cues from nonporicidal *B. hybrida*, as well as from poricidal *S. houstonii*, elicited sonication behavior in flower-naive bumble bees. Mechanosensory pollen cues in turn

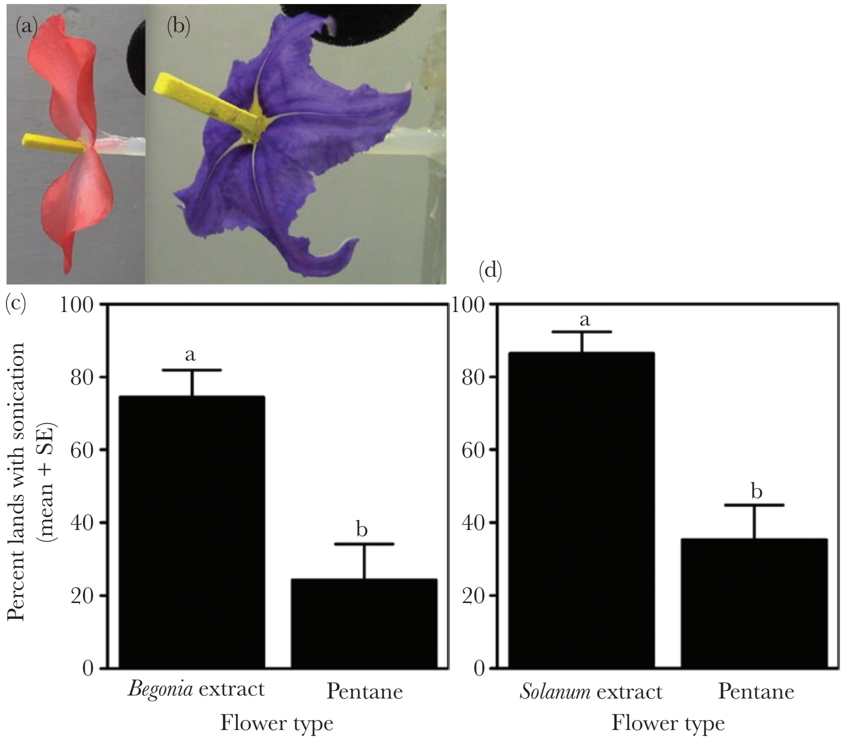

Figure 4

Mean percentage landings (±MSE) resulting in sonication by *Bombus impatiens* on anther extract surrogates and on pure pentane treated surrogates made using nonporicidal *Begonia hybrida* or poricidal *Solanum houstonii*. (a) *B. hybrida* surrogates. (b) *S. houstonii* surrogates. (c, d) Mean percentage landings (± SE) on surrogates treated with anther extract or pure pentane resulting in sonication. $N = 11$ bees per treatment. Different letters above bars indicate significant differences at $P < 0.05$ according to a Wilcoxon test.

suppressed expression of the sonication routine. Bees more often buzzed anthers that lacked pollen or had little pollen remaining; these patterns were the same when pollen was substituted for pollen-sized (but odorless) cellulose or plastic powder. Bees never scrabbled when the anthers (surrogate or live) were bare.

Additionally, cellulose particles much larger than pollen typically collected by generalist bees (Roberts and Vallespir 1978) did not suppress sonication and bees never scrabbled; accordingly, simply impeding detection of the anthers by covering them with particles did not suppress sonication. Bees packed particles (pollen, small plastic microspheres, small, or large cellulose particles) from *B. hybrida* in their scopa as they would the pollen of the natural species *B. descoleana* (Supplementary Figure S1).

Finally, when anthers presented small amounts of pollen, sonication resulted in a greater pollen collection rate (i.e., weight harvested per unit time or per flower) than scrabbling behavior.

DISCUSSION

The consequences of flexibility in generalist pollinator behavior for the evolution of plant-pollinator mutualisms have been considered extensively in the context of nectar foraging. Flexible nectar foraging behavior via learning can facilitate floral trait evolution (e.g., Sargent 2004; Gómez et al. 2015; Guzmán et al. 2015; Rojas-Nossa et al. 2016) and species invasions (Harmon-Threatt et al. 2009; Bartomeus et al. 2016), for example, by allowing pollinators to use floral morphologies they have not previously encountered (e.g., Chittka and Thomson 1997; Bartomeus et al. 2016). Our findings suggest that generalist bees can also flexibly and effectively adjust pollen-collecting behavior to make use of diverse floral resources. These pollinators may thus exert similar impacts on floral diversity in the context of pollen collection, particularly as regards the form and arrangement of anthers and associated structures. For instance, the strongly curved petals of many species of *Senna* enhance pollination by sonicating bees via deflection of expelled pollen onto pollinating spots (e.g., Wolfe and Estes 1992; Westerkamp 2004). Similarly, bee-pollinated plant species often present their pollen more gradually and/or divide it among “feeding” and “pollinating” anther sets and thereby optimize pollen delivery (Castellanos et al. 2006; Vallejo-Marín et al. 2010).

Our results have particularly important implications for the widespread and repeated evolution of poricidal floral morphology from nonporicidal ancestors (Buchmann 1983; Vallejo-Marín et al. 2010; De Luca and Vallejo-Marín 2013), particularly if poricidal morphology evolves gradually, via a series of intermediate stages, as commonly presumed for traits (Futuyma 2013; Guzmán et al. 2015). Under a scenario of gradual evolution, intermediate stages will have pollen partially concealed to varying degrees (e.g., Figure 1d–f). Scrabbling alone is ineffective at removing unexposed pollen from such flowers (King and Ferguson 1994; Javorek et al. 2002; Pomeroy and Fisher 2002) and thus such intermediates are at a potential disadvantage. However, our findings suggest that a combination of scrabbling and sonication might result in effective pollen collection, and thus effective pollen transfer, for plants with anthers of these intermediate types. As pollen is progressively concealed during this evolutionary process, sonication would be expressed more and more during pollen collection, until eventually, when flowers are fully poricidal and pollen entirely concealed, it would be the only behavior used to extract pollen. To test this proposed scenario, future work should evaluate whether bees are

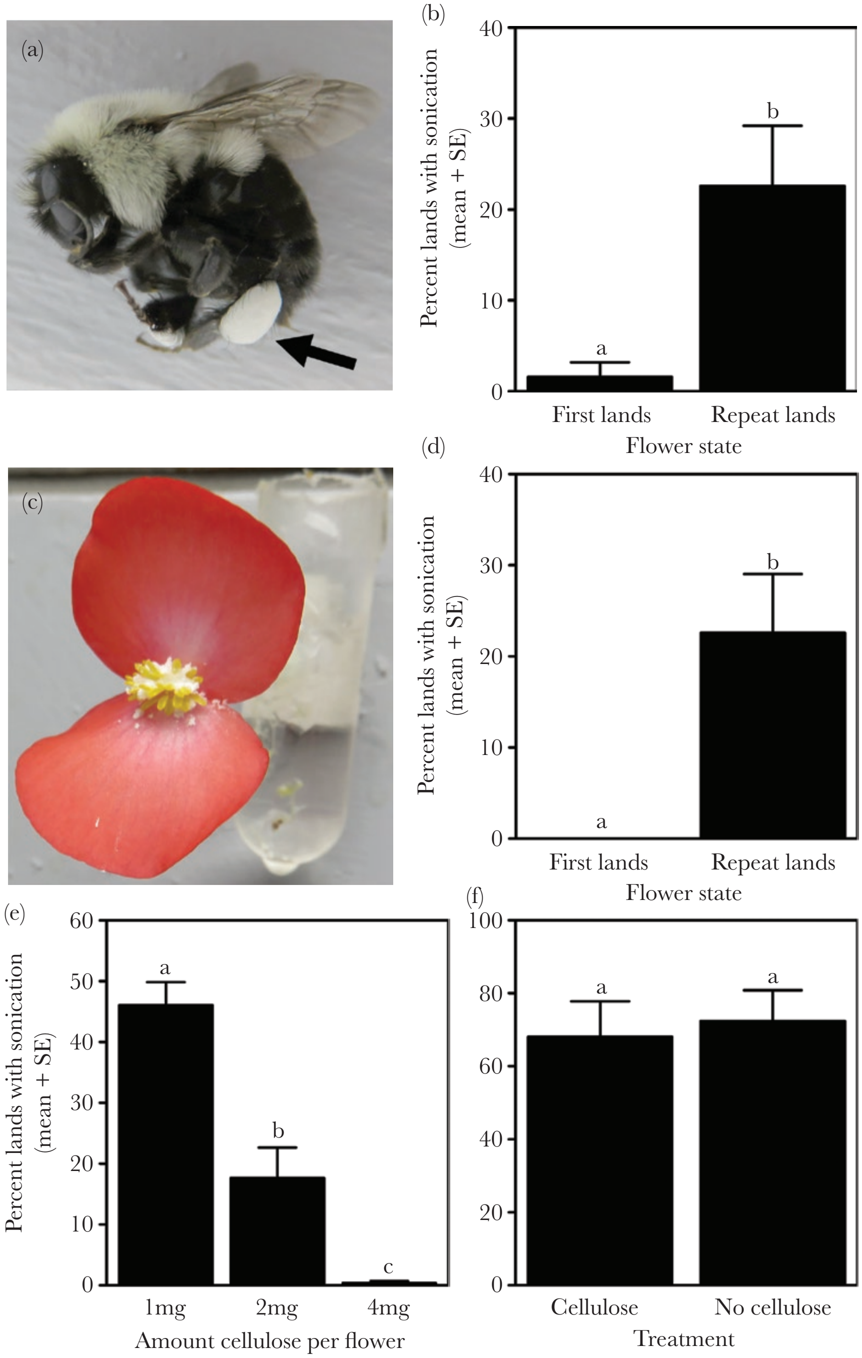

Figure 5

Mean percentage landings (± SE) by *Bombus impatiens* resulting in sonication in treatments where the availability or size of plastic microspheres or cellulose particles presented by *Begonia hybrida* was varied. (a) Representative forager that collected plastic microspheres (black arrow). (c) *B. hybrida* with 4 mg of 20 µm size cellulose particles. Mean percentage landings (± SE) resulting in sonication (b) on flowers supplemented with 2 mg 20 µm plastic microspheres on first landings to each flower in an array versus repeat landings; (d) on flowers supplemented with 2 mg 20 µm cellulose particles on first landings to each flower in an array versus repeat landings; (e) on flowers initially supplemented with 1, 2, or 4 mg of 20 µm cellulose particles; and (f) on bare flowers versus flowers enriched with 180 µm cellulose particles. (b) *N* = 7 bees per treatment. (d, e) *N* = 7 bees each for the 1, 2, and 4 mg cellulose treatments. (f) *N* = 8 per treatment. Different letters above bars indicate significant differences at $P < 0.05$ according to a Wilcoxon test or Tukey's post hoc test.

indeed able to collect and transfer pollen relatively efficiently from plant species that partially conceal their pollen, using a combination of scrabbling and sonication. Additionally, our results strongly suggest that rather than there being a relatively tight coevolutionary relationship between sonicating bees and poricidal floral morphology (Macior 1964; Buchmann 1983), floral sonication probably first evolved in the context of collecting pollen from diverse nonporicidal ancestors, as a strategy for collecting pollen efficiently from partially depleted or just dehiscing anthers (see Raine and Chittka 2007). Subsequently, poricidal morphology evolved repeatedly in diverse lineages, its evolution facilitated by the pre-existing capacity of bees to extract pollen via sonication (see Buchmann 1985; De

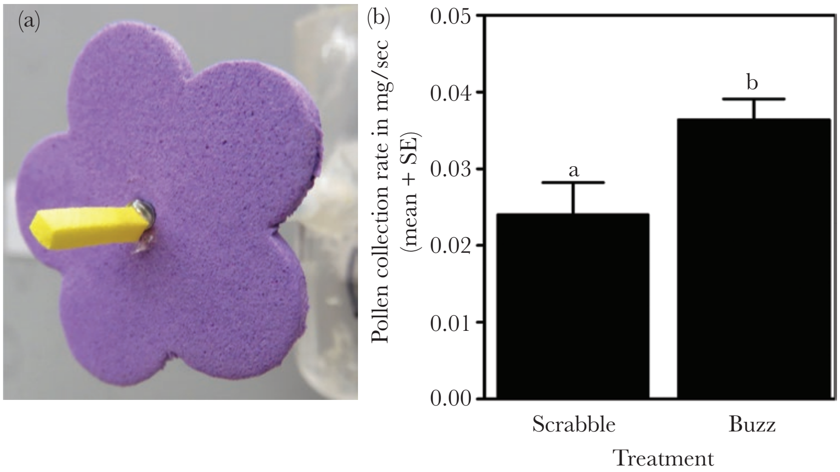

Figure 6

Pollen collection rate for *Bombus impatiens* using scrabbling versus sonication behaviors to forage from artificial flowers. (a) Nonporicidal artificial foam flower (pollen is exposed). (b) Pollen collection rate for bees that scrabbled and bees that sonicated. $N = 11$ bees in each treatment. Letters above bars within a panel indicate significant differences at $P < 0.05$ according to a *t*-test.

Luca and Vallejo-Marín 2013) and by the pre-existing occurrence of sonication elicitor chemistry in emerging poricidal forms.

Our results suggest that flexibility in pollen collection involves a different mechanism than that commonly studied in nectar collection. Whereas bees use instrumental learning to acquire and refine unique routines specific to particular flower types to extract nectar (e.g., Lewis 1993; Laverty 1994; Gegear and Laverty 1995), the flexibility in pollen collection described here involved switching between 2 seemingly stereotyped motor routines, floral sonication and scrabbling. Switching is regulated by the interacting effects of 2 floral cues: chemosensory anther cues and mechanosensory pollen cues. Importantly, chemosensory anther cues elicited sonication and mechanosensory pollen cues suppressed sonication (and elicited scrabbling) by even naive bees on their first floral visit (Supplementary Figure S3a–c), suggesting that use of either routine is innately specified.

Although switching did not appear to require instrumental learning in our study, the effectiveness of collection routines might nevertheless be altered with experience. To improve pollen collection efficiency, bees may learn when to switch between scrabbling and sonicating or adjust subtle characteristics of the 2 routines themselves (e.g., Morgan et al. 2016; Russell et al. 2016). Sonication has a strong innate component and is affected little by experience (Russell et al. 2016); however, scant work has examined whether bees learn to scrabble (see Raine and Chittka 2007). Furthermore, experience could influence how bees collect pollen in response to naturally varying floral features other than pollen presentation. Across taxa, pollen varies greatly in stickiness, clumping, size, and surface structure (Roberts and Vallespir 1978; Hesse 1981; Vaissière and Vinson 1994; Pacini and Hesse 2005; Lunau et al. 2015). Finally, pollen-foraging bees likely use instrumental learning when they must gain access to anthers concealed within a corolla (e.g., keel flowers; Reinhardt 1952; Westerkamp and Weber 1999). Bees probably also use appetitive learning in response to pollen guides, analogous to the role of learning in nectar guide use (Leonard and Papaj 2011), as well as other floral features such as color, pattern and scent. However, studies on instrumental and appetitive learning in pollen foraging are rare, compared to studies of these processes in nectar foraging (Muth et al. 2015; Muth et al. 2016; Nicholls and Hempel de Ibarra 2016; Russell, Golden, et al. 2016; Russell, Leonard, et al. 2016).

It is somewhat surprising that sonication results in a higher rate of pollen removal than scrabbling from nonporicidal flowers given that scrabbling is the preferred strategy for nonporicidal flowers. However, we are relatively ignorant of the relative costs of scrabbling and sonication. Although we have shown that sonication results in higher rates of collection of exposed pollen at low levels (when bees would switch from using scrabbling to using sonication on live flowers), we have not assessed what costs might be involved in achieving these higher rates. For example, sonication is likely energetically expensive relative to scrabbling: Flight (which, like sonication, uses indirect flight muscles) is on average 50% more energetically expensive than walking locomotion (which, like scrabbling, involves leg motion) (Balfour et al. 2015). We have also not demonstrated whether sonication results in higher rates of collection of exposed pollen at high levels. It would be useful to evaluate the extent to which energetic and other costs influence the relative use of sonication and scrabbling, especially on different anther morphologies. Likewise, each component of pollen collection rate (amount of pollen collected and time spent collecting that pollen) might conceivably vary on flowering plant species with differing anther morphology and amounts of available exposed pollen.

Studies of the common floral cues involved in the collection of nectar have been important in elucidating how shifts from nectar rewards to deception may evolve (e.g., Raguso 2004; Jersáková et al. 2006; Pohl et al. 2008). Our results likewise shed light on repeated and common shifts from pollen rewards to deception and the evolution of pollen and anther mimicry generally (e.g., Vogel 1978; Schemske and Agren 1995; Lunau 2007). Our evidence suggests that the floral cues mediating pollen collection behavior by generalist bees may be simple. These cues may also be widely shared among angiosperms (e.g., chemical anther cues from at least 2 distantly related species elicited sonication and we have since successfully tested many more (A. Russell and D. Papaj, unpublished data; see also Dobson et al. 1999). Mimicry of anthers by staminodes and pistils (Schemske and Agren 1995; Lunau 2007) may only require emulation of chemical cues. Similarly, mechanical cues alone may be sufficient to generate pollen mimicry. Consistent with this, generalist bees collect pollen-sized particles from flowers, such as pseudopollen and even fungal spores (e.g., Simpson and Neff 1981; Bernhardt and Burns-Balogh 1986; Eltz et al. 2002; Davies and Turner 2004).

In closing, examples of learning in mediating flexibility of foraging behavior by animals are common (Kamil and Roitblat 1985; Krebs and Inman 1992; Papaj and Lewis 1993). However, there is comparatively little research on mechanisms of flexibility that may not rely on learning, especially with respect to pollinator behavior. In this study, we found that pollen foraging generalist bumble bees were able to cope effectively with variability in their floral resources by switching between their 2 pollen collection behaviors. The switching mechanism involved a weighing of 2 types of general floral cues. Although this mechanism is different from learning, it is analogous in the sense that it prepares generalist bees to forage even from floral forms they may never have encountered in their evolutionary history. Bees foraging from flowers constitute a model system for examining how generalists manage collection of different food types on different resources. Accordingly, we expect that generalist taxa, including other invertebrates, as well as vertebrates that forage on a wide range of biotic resources may also exhibit flexible foraging not dependent on learning.

SUPPLEMENTARY MATERIAL

Supplementary data are available at *Behavioral Ecology* online.

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Data accessibility: Analyses reported in this article can be reproduced using the data provided by Russell et al. (2017).

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