

# Floral traits affecting the transmission of beneficial and pathogenic pollinator-associated microbes

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Flowers provide resources for pollinators, and can also be transmission venues for beneficial or pathogenic pollinator-associated microbes. Floral traits could mediate transmission similarly for beneficial and pathogenic microbes, although some beneficial microbes can grow in flowers while pathogenic microbes may only survive until acquired by a new host. In spite of conceptual similarities, research on beneficial and pathogenic pollinator-associated microbes has progressed mostly independently. Recent advances demonstrate that floral traits are associated with transmission of beneficial and pathogenic microbes, with consequences for pollinator populations and communities. However, there is a near-absence of experimental manipulations of floral traits to determine causal effects on transmission, and a need to understand how floral, microbe and host traits interact to mediate transmission.

## Addresses

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## Introduction

By providing pollen and nectar in attractive displays, flowers serve as ‘nature’s rest stop’ in all its connotations: a source of resources but also a receptacle for waste and a way station for passing travelers that mix and then disperse. Flowers can have insect densities 10 000 times greater than surrounding foliage [1] and so may be sites where microbes are transmitted between visitors and

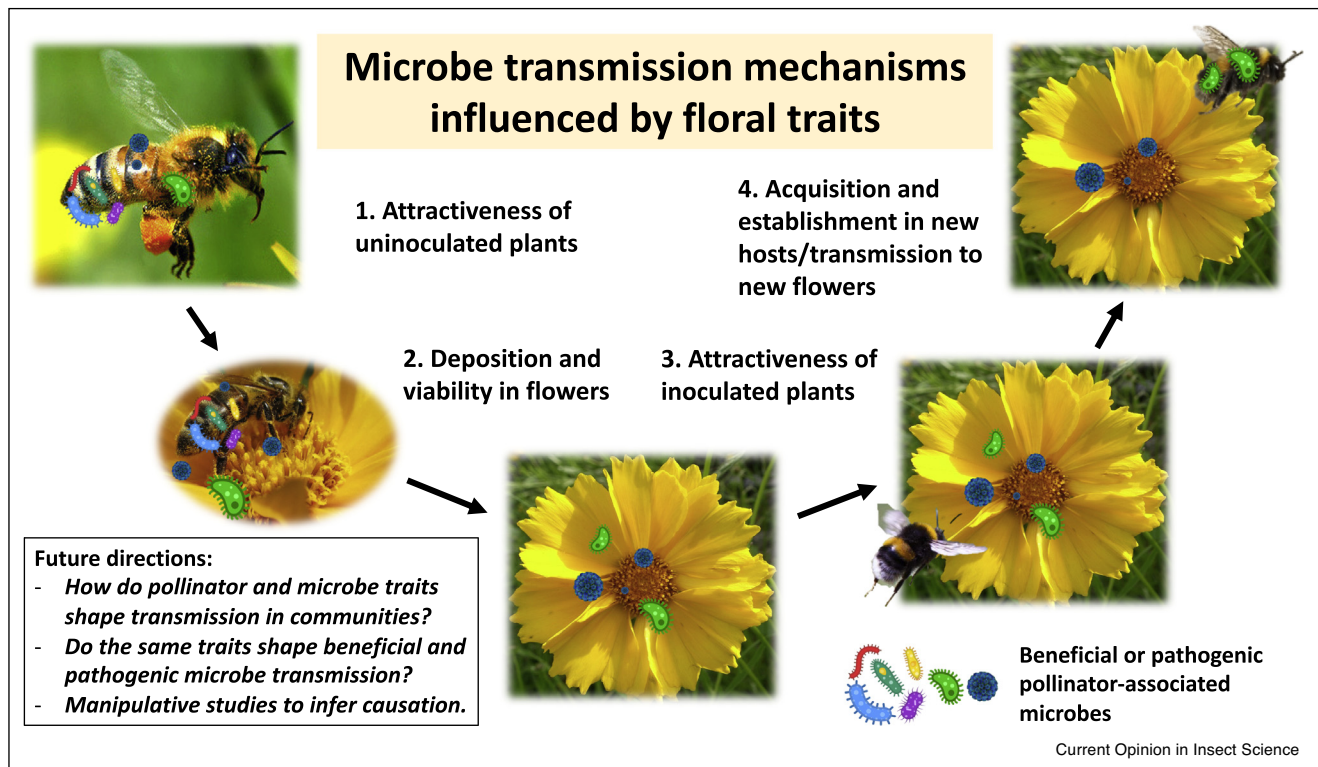
other flowers. A 2014 review assessed the role of floral traits in transmission of pathogens to plants and pollinators via flowers [2]; while there was a substantial literature for plant pathogens, at that time only a single study had experimentally demonstrated transmission of pollinator pathogens at flowers. Since then, there has been growing research on pollinator pathogens, alongside a parallel but largely separate literature understanding how floral traits affect transmission of microbes beneficial to pollinators.

Here, we review recent studies of transmission of both beneficial and pathogenic pollinator-associated microbes. We define ‘beneficial’ and ‘pathogenic’ microbes in terms of their effect on pollinators; ‘pathogenic’ microbes have a detrimental impact on at least some pollinators, while ‘beneficial’ microbes have a positive impact on at least some pollinators, although the ecology of many microbes is poorly understood and effects on pollinators could be context-dependent. Further, we structure this new review by the four mechanisms proposed in the 2014 review [2]: (1) Floral attractiveness of uninoculated plants, (2) Microbe deposition and viability in flowers, (3) Floral attractiveness of inoculated plants, and (4) Pathogen acquisition and establishment in hosts upon visiting inoculated flowers (Figure 1). With this organizational structure, our goal is to highlight similarities and differences in the role of floral traits on transmission of microbes that may be beneficial, commensal or detrimental to pollinators, with the goal of a more mechanistic understanding of the role of floral traits in these interactions. Although many microbes that affect plants can be transmitted via flowers [2], this is outside the scope of our review. We end by highlighting gaps in knowledge and identifying future key areas of interest.

## Traits influencing transmission of beneficial microbes

Many microbes on flowers cause no detectable harm to plants or pollinators, and may in some cases benefit them. Yeasts and bacteria are common inhabitants of flowers and are often more abundant and frequently isolated after pollinators have visited a flower compared to unvisited flowers [e.g. Refs. 3,4], suggesting that pollinators are major transmitters. Flowers and pollinator bodies are distinct environments that differ in nutrient composition, environmental conditions, and longevity, and so it is no surprise that microbial species appear to specialize on these distinct habitats. For example, the Ascomycete

Figure 1



Floral traits can influence beneficial or pathogenic pollinator-associated microbe transmission in four ways. Trait can influence 1) the attractiveness of uninoculated plants to pollinators, 2) deposition and viability in flowers, 3) attractiveness of inoculated plants, and 4) acquisition and establishment in new hosts/transmission to new flowers. Each of these four mechanisms is highlighted in the text, along with a brief discussion of promising future directions. Beneficial and pathogenic pollinator-associated microbes are represented by the caricatures of different colors.

yeasts *Metschnikowia reukaufii* and *Metschnikowia gruessii* attain high density in flowers but are detected in low numbers on or in bumble bee bodies. In contrast, Ascomycete yeasts in the *Starmerella* clade and the genus *Debaryomyces* can be detected in low numbers on flowers, but attain high densities in bumble bee gastrointestinal tracts and honeypots [5].

Some species of fungi and bacteria found on flowers can benefit pollinators. Consumption of nectar containing yeasts can increase colony growth of *Bombus terrestris* and may protect against pathogens [6]. Bumble bees can use microbial volatiles as foraging cues [7], which may increase foraging efficiency or resource acquisition. Microbes in nectar and pollen can colonize solitary bee provisions [8], where they may aid in preserving sugar and protein, or serve as food for developing larvae [9,10]. Many other insects deposit microbes on floral surfaces and/or consume microbes at flowers, and microbes could benefit flower-feeding insects in diverse ways [11].

Despite the central importance of flower–pollinator interactions in the transmission of beneficial microbes, surprisingly little is known about how floral traits affect

transmission. Our review focuses on nectar fungi and bacteria, with recognition that research on other microbial symbionts awaits detailed investigation [12].

### (1) Floral attractiveness of uninoculated plants

Because many nectar microbes require pollinator visitation for transmission, presumably any floral traits that increase floral visitation (overall or by taxa that carry specific microbes) will increase transmission. For example, nectar yeast frequency and abundance are often positively correlated with pollinator visitation. In addition, yeast abundance in nectar was positively associated with the proportion of floral visits by bumble bees, but negatively correlated with visits by solitary bees [13], suggesting that floral traits associated with particular pollinator groups may affect transmission [14]. However, research is needed that links intraspecific and interspecific trait variation to transmission [akin to Ref. 15\*\*]. One exemplar study found higher nectar microbial abundance in male compared to female flowers of *Eurya emarginata*, but the role of pollinator transmission versus filtering by the nectar environment or resource availability between flower types was not resolved [16\*].

## (2) Microbe deposition and viability in flowers

Microbes in nectar are often a subset of those found on and in pollinator bodies, suggesting strong filtering by nectar, likely favoring species that can withstand the high sugar environment [17] or grow quickly [18]. Research is accumulating on how nectar traits affect microbe viability and growth, although it is sometimes challenging to separate plant species identity versus trait-based effects, and strains of the same microbial species can vary widely in relative growth rates in the same nectar sources [19]. However, some common patterns have emerged. For example, high sugar concentration can inhibit microbial growth, and nectar secondary compounds, once thought to reduce microbial growth in nectar, drive concentration-dependent and compound-specific effects that often don't inhibit growth at natural concentrations [18,20]. Survival and growth of microbes in nectar also depends on their interactions. For example, nectar bacteria and yeast experience strong priority effects, with whoever arrives first or has higher initial abundance suppressing the other [21,22]. Mechanisms associated with priority effects are likely related to microbial growth rate [18] and subsequent effects of microbes on nectar traits, such as pH and amino acids [22,23]. Floral traits that affect microbial viability and growth in turn affect microbe acquisition. In artificial nectar arrays, transmission can depend strongly on microbial density in flowers [24]. These studies lead to the prediction that floral traits that promote microbial growth will also promote microbial transmission among pollinators (Figure 1).

## (3) Floral attractiveness of inoculated plants

Many nectar microbes affect floral attractiveness to pollinators. For example, both artificial and natural flowers inoculated with the nectar yeast *M. reukaufii* receive increased bumble bee pollinator visitation relative to uninoculated flowers [25,26], whereas nectar and floral surfaces colonized by bacteria (such as *Neokomagataea* sp. formerly *Gluconobacter* sp.) can reduce visitation by hummingbirds, bumble bees, and honey bees [27–29]. Although mechanisms may vary, strong evidence suggests that pollinator attraction of yeast-inoculated flowers is driven by associative learning of yeast-derived volatiles with floral rewards [28], as well as gustatory responses once pollinators start feeding [30\*\*,31\*\*].

## (4) Microbial acquisition by pollinators and transmission to new nectar sources

Little is known about how floral traits affect the likelihood that pollinators will acquire microbes from nectar sources, but some exemplar behavioral work suggests a role. Bees acquire approximately 1% of the microbes on flowers, with less acquired from nectaring than collecting pollen [32]. Thus, floral traits that increase time spent accessing rewards or proportion of the pollinator's body contacting contaminated nectar or surfaces should increase acquisition. Three-way interactions among

flower, microbe and pollinator traits are likely important in the transmission process, and assessing how microbe traits interact with floral and pollinator traits to affect dispersal may yield unique insights [11].

## Traits influencing transmission of pathogenic microbes

Floral traits could mediate the transmission of pathogenic microbes similarly to beneficial microbes, but there may also be key differences. Some beneficial microbes can establish and grow in and on flowers, and floral traits may shape their growth. By contrast, pollinator pathogens typically cannot grow on or in flowers (although this is rarely examined) and so floral traits may affect pathogen survival and transmission, but not growth. We describe how floral traits could affect pathogen transmission using the same mechanistic structure as for beneficial microbes to facilitate comparisons.

### (1) Floral attractiveness of uninoculated plants

Pathogen deposition on flowers can differ by plant species, which could be due to attraction or how pollinators interact with flowers. The bumble bee pathogens *Nosema bombi*, *Crithidia bombi* and *Apicystis bombi* had more deposition onto bell-shaped (*Campanula cochleariifolia*) than flat (*Viola tricolor*) flowers, but deposition of *Nosema apis* and *Nosema ceranae* honey bee pathogens did not differ between plant species [33]. Two honey bee viruses had uneven deposition onto flowers of three legume species, and deposition differed when plant species were presented alone versus in mixtures [34\*\*]. Flowers with the longest but also fewest honey bee visits had the highest virus loads, suggesting that floral traits affect deposition by altering bee visitation and behavior. *Bombus impatiens* infected with *C. bombi* had variable likelihoods of depositing feces (containing infective cells) on different floral parts in three plant species [35\*], suggesting that floral morphology affects deposition, and *B. impatiens* were also more likely to defecate on a large composite flower than flowers of seven other species [36]. Bumble bees infected with *C. bombi* spent more time in flowers with high-iridoid glycoside nectar than uninfected bees and were more likely to return to other high-iridoid flowers [37], both of which could affect pathogen deposition.

### (2) Pathogen deposition and viability in flowers

In a comprehensive study, Figueroa *et al.* [38\*] found at least one bee pathogen (including neogregarines, trypanosomatids, *N. ceranae* and *N. bombi*) in flowers from 75% of 13 plant species from multiple field sites, with pathogen prevalence differing widely between plant species. These differences could be due to differential visitation or acquisition and viability between plant species, but the traits responsible are unknown. Once deposited on flowers, pathogens may contact nectar [but see Ref. 39] and as noted for beneficial microbes, the nectar environment may be challenging for pathogen survival. Exposure to increasing sugar concentrations before consumption reduced *C. bombi* infection likelihood and intensity in

bumble bees [40]. *C. bombi* exposure to the nectar iridoid glycoside aucubin before consumption also reduced subsequent bumble bee infection [41], but exposure to several other nectar secondary compounds did not [41,42]. Location on flower parts can also affect viability; *C. bombi* survival was lower on exposed bracts than inside flowers and in sun compared to shade [35\*].

### (3) Floral attractiveness of inoculated plants

Although *B. terrestris* avoided foraging on flowers inoculated with *C. bombi* [43], we are aware of no studies assessing whether floral traits mediate bee responses to inoculated plants. Given the strong evidence that beneficial microbes affect floral attractiveness, this mechanism warrants further investigation for pathogens.

### (4) Pathogen acquisition and establishment in hosts upon visiting inoculated flowers

Over 25 years ago, a seminal paper demonstrated that *B. terrestris* and *Bombus lucorum* became infected with *C. bombi* after foraging on flowers visited by infected bees [44]. The odds of acquiring infection differed between two plant species, and also in inflorescences manipulated to change floral architecture. This study incorporates the role of floral traits on both deposition by infected hosts and acquisition/establishment in new hosts. To our knowledge this is the only paper that has manipulated any floral trait to assess consequences for pathogen transmission in pollinators.

That said, several nectar secondary compounds consumed after pathogen acquisition can reduce *C. bombi* and *N. ceranae* infections *in vivo* [reviewed in Refs. 45–47], with one study discovering a mechanism; callumene from *Calluna vulgaris* nectar removed the *C. bombi* flagellum, preventing attachment to gut walls [48\*]. However, adding the nectar compound thymol to flowers along with *C. bombi* did not affect pathogen establishment in foraging bumble bees [49].

Flowers, by acting as deposition and acquisition venues, may also be important in pathogen transmission between host species. An RNA virus was transmitted between honey and bumble bees co-foraging on the same flowers, but this study did not eliminate the possibility of transmission via other surfaces such as cage walls [50]; a more recent study found that honey bees deposited viruses on flowers, but viruses were not subsequently acquired by bumble bees [34\*\*]. However, stingless bees (*Tetragonula hockingsi*) became infected by *N. ceranae* that was previously deposited on flowers by *Apis mellifera* [51], conclusively demonstrating the role of flowers in transmission between bee species. Vectoring of bumble bee pathogens by honey bees differed on two plant species, but vectoring of honey bee pathogens by bumble bees did not differ with plant species [33]. Furthermore, parasites can play a role in vectoring pathogens; *Varroa* mites, which transmit

deformed wing virus to honey bees, were slowest to infest honey bees foraging at *Echinacea* flowers compared to flowers of two other plant species, suggesting a role of floral shape [52].

Finally, one observational study attempted to isolate floral traits shaping pathogen acquisition. *C. bombi* was added to flowers of 14 plant species, after which individual *B. impatiens* foraged. There was a fourfold difference across plant species in pathogen acquisition and infection intensity [15\*\*]. However, floral size and shape, number of open flowers, nectar production, and inflorescence height did not explain interspecific variation in transmission; the only trait that correlated with pathogen acquisition was the total number of reproductive structures per inflorescence.

### Traits influencing microbe spread in plant–pollinator communities

The studies above indicate that floral traits can influence transmission at individual flowers, but how these individual interactions shape microbe spread in communities is just beginning to be examined. Susceptible-infectious-susceptible (SIS) models for plant–pollinator networks were recently developed with continuous trait distributions, finding that disease spread was impacted the most by selective pollinators, universally attractive flowers, and cospecialized plant–pollinator pairs [53\*\*]. Although this theory was developed for pathogenic microbes, it can also be applied to beneficial microbes.

Two recent studies show that prevalence of pathogenic microbes on flowers varies among plant species, and changes in the bee:flower ratio in communities can influence the likelihood of transmission at flowers. Graystock *et al.* [54\*\*] screened >5000 bees and flowers in old-field communities and detected bee pathogens (*N. bombi*, *N. ceranae*, *C. bombi*, *Crithidia expoeki*, or *Apicystis* spp.) in 42% of bee species (12.2% individual bees) and 70% of flower species (8.7% individual flowers). Prevalence varied by more than 80% among well-sampled flower species. In addition, prevalence on flowers was lowest late in the season when the bee:flower ratio was lowest, suggesting reduced risk of transmission via dilution. Supporting the potential importance of dilution, an experiment manipulating *B. terrestris* density in replicated plant communities found that when the bee:flower ratio was low, slow bee paralysis virus (SBPV) was transmitted less efficiently [55\*]. This pattern was not observed for *C. bombi*, indicating parasite-specific responses to bee/flower density.

New work also suggests that visitation by particular pollinator species to the flowers of particular plant species can potentially play a disproportionate role in transmission. *A. mellifera* visitation rates to nectar-rich knapweed (*Centaurea* spp.) were greater than visitation rates of the rest of the pollinator community combined to this plant



species [56], and honey bees and knapweed were both among the highest-prevalence bee and flower species, respectively, for multiple pollinator pathogens [54\*\*].

Finally, particular plant species can be associated with reduced pathogens in pollinators, and community-level changes in floral traits can shape disease intensity. Sunflower pollen dramatically reduced *C. bombi* infection in bumble bees, and farms with more sunflower had bees with lower infection intensity [57]. Bumble bees at Belgian sites invaded by *Impatiens glandulifera* had lower prevalence of infection with *Apicystis* but not other pathogens; the authors hypothesized that pollen polyphenols could decrease infection [58]. Wildflower fields increased prevalence of several bee pathogens as well as bee abundance in some landscape contexts [59\*]. Finally, Adler *et al.* [60\*] created replicated communities using plant species that had resulted in high or low *C. bombi* infection in previous flower foraging assays [15\*\*]. Colonies foraging in tents with high-infection plant species had on average twice the infection intensity compared to colonies with low-infection plant species, indicating the importance of plant communities for infection dynamics [60\*]. Identifying the floral traits mediating such interactions would provide an invaluable tool for choosing species to include in habitat restoration.

### Future directions

Although there are many gaps in our understanding of how floral traits affect microbe transmission, here we briefly highlight ideas that may be particularly productive.

### Assessing the effect of floral traits on multiple microbes

Various pathogens may interact with hosts and flowers differently, and some have been more strongly associated with pollinator decline than others. Which floral traits are most important for the transmission of the most detrimental pathogens or the most beneficial microbes, which are also critical for pollinator health? Currently, observational studies suggest patterns but there are almost no experimental manipulations of floral traits to determine causal relationships. Such information is essential to provide a general framework that could guide choices of plants for pollinator habitats, as well as understand trait-mediated host–parasite dynamics.

### The role of floral traits in a community context

Do beneficial microbes and pathogens frequently co-occur in particular plant species? Are plant species with more similar floral traits more likely to share microbial communities, and are such correlations structured by floral traits and/or pollinator visitation? Are some traits generally anti-microbial, reducing both pathogen and beneficial microbe growth? Similarly, are pollinator species with more similar traits or behaviors more likely to share pathogens or transmit beneficial microbes?

Research on beneficial and pathogenic microbe transmission has grown almost independently, but combining them could yield important insights for bee health.

### Interactions among flower, host, and microbe traits

Our focus has been on how floral traits affect microbe transmission, but the transmission process will also depend on traits and behaviors of pollinators and microbes. For example, the morphology or other properties of particular microbes may make them more conducive to dispersal by particular pollinators [11], or among particular flower morphologies. In addition, transmission could occur via biotic or abiotic vectors, and the biology of the microbe may affect the efficacy of each of these routes. If this is the case, studying the traits of flowers alone may misrepresent the drivers of microbial transmission. Increased study of pollinator and microbial species using a trait-based approach may help identify the 3-way community trait space most conducive or susceptible to microbial transmission and whether such traits can be generalized across interactions.

### Credit author statement

**Rachel Vannette** and **Rebecca Irwin**: wrote the ‘Traits influencing transmission of beneficial microbes’ section. **Scott McArt**: wrote the ‘Traits influencing microbe spread in plant–pollinator communities’ section and drafted the figure. **Lynn Adler**: wrote the abstract, introduction and ‘Traits influencing transmission of pathogenic microbes’ section. **All authors**: co-wrote the Highlights, Future Directions, and provided feedback on all sections and the figure.

### Conflict of interest statement

Nothing declared.

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### References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as

- of special interest
- of outstanding interest

1. Wardhaugh CW, Stork NE, Edwards W, Grimbacher PS: **The overlooked biodiversity of flower-visiting invertebrates.** *PLoS One* 2012, **7**.
2. McArt SH, Koch H, Irwin RE, Adler LS: **Arranging the bouquet of disease: floral traits and the transmission of plant and animal pathogens.** *Ecol Lett* 2014, **17**:624–636.

3. Aizenberg-Gershtein Y, Izhaki I, Halpern M: **Do honeybees shape the bacterial community composition in floral nectar?** *PLoS One* 2013, **8**.
4. Schaeffer RN, Vannette RL, Irwin RE: **Nectar yeasts in *Delphinium nuttallianum* (Ranunculaceae) and their effects on nectar quality.** *Fungal Ecol* 2015, **18**:100-106.
5. Brysch-Herzberg M: **Ecology of yeasts in plant-bumblebee mutualism in Central Europe.** *FEMS Microbiol Ecol* 2004, **50**:87-100.
6. Pozo MI, van Kemenade G, van Oystaeyen A, Aledón-Catalá T, Benavente A, Van den Ende W, Wäckers F, Jacquemyn H: **The impact of yeast presence in nectar on bumble bee behavior and fitness.** *Ecol Monogr* 2020, **90**:e01393.
7. Russell AL, Ashman T-L: **Associative learning of flowers by generalist bumble bees can be mediated by microbes on the petals.** *Behav Ecol* 2019, **30**:746-755.
- Bombus impatiens* workers were able to learn to associate microbial presence on flowers with the presence of flower rewards. Bumble bees were innately deterred by bacterial cues but not by yeast cues, but could use either to guide foraging decisions.
8. Rothman JA, Andrikopoulos C, Cox-Foster D, McFrederick QS: **Floral and foliar source affect the bee nest microbial community.** *Microb Ecol* 2019, **78**:506-516.
9. Steffan SA, Dharampal PS, Danforth BN, Gaines-Day HR, Takizawa Y, Chikaraishi Y: **Omnivory in bees: elevated trophic positions among all major bee families.** *Am Nat* 2019, **194**:414-421.
10. Dharampal PS, Carlson C, Currie CR, Steffan SA: **Pollen-borne microbes shape bee fitness.** *Proc R Soc B* 2019, **286**:20182894.
11. Madden AA, Epps MJ, Fukami T, Irwin RE, Sheppard J, Sorger DM, Dunn RR: **The ecology of insect-yeast relationships and its relevance to human industry.** *Proc R Soc B* 2018, **285**.
12. Rebolleda-Gomez M, Forrester NJ, Russell AL, Wei N, Fetters AM, Stephens JD, Ashman TL: **Gazing into the anthosphere: considering how microbes influence floral evolution.** *New Phytol* 2019, **224**:1012-1020.
13. Herrera CM, de Vega C, Canto A, Pozo MI: **Yeasts in floral nectar: a quantitative survey.** *Ann Bot* 2009, **103**:1415-1423.
14. Zemenick AT, Vannette RL, Rosenheim JA: **Linked networks reveal dual roles of insect dispersal and species sorting for bacterial communities in flowers.** *Oikos* in preparation.
15. Adler LS, Michaud KM, Ellner SP, McArt SH, Stevenson PC, Irwin RE: **Disease where you dine: plant species and floral traits associated with pathogen transmission in bumble bees.** *Ecology* 2018, **99**:2535-2545.
- Added *C. bombi* inoculum to flowers of 14 plant species and allowed uninfected *B. impatiens* to forage, finding a fourfold difference in acquisition between plant species. Assessed several floral traits and found that only total reproductive structures per inflorescence was positively associated with acquisition.
16. Tsuji K, Fukami T: **Community-wide consequences of sexual dimorphism: evidence from nectar microbes in dioecious plants.** *Ecology* 2018, **99**:2476-2484.
- Sexual dimorphism in dioecious shrubs affected nectar microbial communities, with male flowers having more prevalent and abundant microbial communities. Different mechanisms may affect microbial communities in the flower sexes; floral visitation affected microbial communities more strongly in male flowers whereas priority effects affected microbial communities more strongly in female flowers.
17. Herrera CM, Canto A, Pozo MI, Bazaga P: **Inhospitable sweetness: nectar filtering of pollinator-borne inocula leads to impoverished, phylogenetically clustered yeast communities.** *Proc R Soc B Biol Sci* 2010, **277**:747-754.
18. Pozo MI, Lachance MA, Herrera CM: **Nectar yeasts of two southern Spanish plants: the roles of immigration and physiological traits in community assembly.** *FEMS Microbiol Ecol* 2012, **80**:281-293.
19. Herrera CM: **Population growth of the floricolous yeast *Metschnikowia reukaufii*: effects of nectar host, yeast genotype, and host 3 genotype interaction.** *FEMS Microbiol Ecol* 2014, **88**:250-257.
20. Vannette RL, Fukami T: **Nectar microbes can reduce secondary metabolites in nectar and alter effects on nectar consumption by pollinators.** *Ecology* 2016, **97**:1410-1419.
21. Dhami MK, Hartwig T, Fukami T: **Genetic basis of priority effects: insights from nectar yeast.** *Proc R Soc B Biol Sci* 2016, **283**.
22. Tucker CM, Fukami T: **Environmental variability counteracts priority effects to facilitate species coexistence: evidence from nectar microbes.** *Proc R Soc B Biol Sci* 2014, **281**.
23. Vannette RL, Fukami T: **Contrasting effects of yeasts and bacteria on floral nectar traits.** *Ann Bot* 2018, **121**:1343-1349.
24. Hausmann SL, Tietjen B, Rillig MC: **Solving the puzzle of yeast survival in ephemeral nectar systems: exponential growth is not enough.** *FEMS Microbiol Ecol* 2017, **93**.
25. Herrera CM, Pozo MI, Medrano M: **Yeasts in nectar of an early-blooming herb: sought by bumble bees, detrimental to plant fecundity.** *Ecology* 2013, **94**:273-279.
26. Schaeffer RN, Mei YZ, Andicoechea J, Manson JS, Irwin RE: **Consequences of a nectar yeast for pollinator preference and performance.** *Funct Ecol* 2017, **31**:613-621.
27. Good AP, Gauthier MPL, Vannette RL, Fukami T: **Honey bees avoid nectar colonized by three bacterial species, but not by a yeast species, isolated from the bee gut.** *PLoS One* 2014, **9**.
28. Russell AL, Ashman TL: **Associative learning of flowers by generalist bumble bees can be mediated by microbes on the petals.** *Behav Ecol* 2019, **30**:746-755.
29. Vannette RL, Gauthier MPL, Fukami T: **Nectar bacteria, but not yeast, weaken a plant - pollinator mutualism.** *Proc R Soc B Biol Sci* 2013, **280**.
30. Rering CC, Beck JJ, Hall GW, McCartney MM, Vannette RL: **Nectar-inhabiting microorganisms influence nectar volatile composition and attractiveness to a generalist pollinator.** *New Phytol* 2018, **220**:750-759.
- Nectar-inhabiting fungal species differed from each other and from bacteria in volatile emissions, honey bees could detect many microbially produced volatile compounds, and microbial compounds could be detected in flowers that contained high densities of fungi.
31. Schaeffer RN, Rering CC, Maalouf I, Beck JJ, Vannette RL: **Microbial metabolites elicit distinct olfactory and gustatory preferences in bumblebees.** *Biol Lett* 2019, **15**.
- Characterized bumble bee detection of microbial volatile compounds and behavioral responses to microbial effects on nectar. The bacteria *Asaia* was preferred over a nectar yeast by naive bumble bee workers when only volatile cues are available, but when bees were allowed to feed on solutions, the yeast solution was consumed to a greater extent.
32. Russell AL, Rebolleda-Gomez M, Shaible TM, Ashman TL: **Movers and shakers: bumble bee foraging behavior shapes the dispersal of microbes among and within flowers.** *Ecosphere* 2019, **10**.
33. Graystock P, Goulson D, Hughes WOH: **Parasites in bloom: flowers aid dispersal and transmission of pollinator parasites within and between bee species.** *Proc R Soc B Biol Sci* 2015, **282**:20151371.
34. Alger SA, Burnham PA, Brody AK: **Flowers as viral hot spots: honey bees (*Apis mellifera*) unevenly deposit viruses across plant species.** *PLoS One* 2019, **14**.
- The first study to conclusively demonstrate that honey bees deposit viruses on flowers. Deposition differed with both the virus and the plant species, and whether the plant species was in a single-species or mixed-species plot. Bumble bees did not become infected after visiting these flowers, but this study is important for demonstrating differential virus deposition between plant species.
35. Figueroa LL, Blinder M, Grincavitch C, Jelinek A, Mann EK, Merva LA, Metz LE, Zhao AY, Irwin RE, McArt SH et al.: **Bee pathogen transmission dynamics: deposition, persistence and acquisition on flowers.** *Proc R Soc B Biol Sci* 2019, **286**:20190603.
- Demonstrated that deposition of feces containing pathogens occurred on different floral parts in different plant species, and that location on flower parts affected pathogen survival and acquisition by new hosts.

36. Bodden JM, Hazlehurst JA, Rankin EEW: **Floral traits predict frequency of defecation on flowers by foraging bumble bees.** *J Insect Sci* 2019, **19**.
37. Richardson LL, Bowers MD, Irwin RE: **Nectar chemistry mediates the behavior of parasitized bees: consequences for plant fitness.** *Ecology* 2016, **97**:325-337.
38. Figueroa LL, Grab H, Ng WH, Myers CR, Graystock P, McFrederick QS, McArt SH: **Landscape simplification shapes pathogen prevalence in plant-pollinator networks.** *Ecol Lett* 2020, **23**:1212-1222.
- Investigated how landscape simplification alters plant-pollinator networks and pathogen prevalence among bees. Landscape simplification reduced pathogen prevalence in bee communities via increased diet breadth of the dominant species.
39. Cisarovsky G, Schmid-Hempel P: **Combining laboratory and field approaches to investigate the importance of flower nectar in the horizontal transmission of a bumblebee parasite.** *Entomol Exp Appl* 2014, **152**:209-215.
40. Folly AJ, Barton-Navarro M, Brown MJF: **Exposure to nectar-realistic sugar concentrations negatively impacts the ability of the trypanosome parasite (*Crithidia bombi*) to infect its bumblebee host.** *Ecol Entomol* 2020 <http://dx.doi.org/10.1111/een.12901>.
41. Michaud KM, Irwin RE, Barber NA, Adler LS: **Preinfection effects of nectar secondary compounds on a bumble bee gut pathogen.** *Environ Entomol* 2019, **48**:685-690.
42. Manson JS, Otterstatter MC, Thomson JD: **Consumption of a nectar alkaloid reduces pathogen load in bumble bees.** *Oecologia* 2010, **162**:81-89.
43. Fouks B, Lattorff HMG: **Recognition and avoidance of contaminated flowers by foraging bumblebees (*Bombus terrestris*).** *PLoS One* 2011, **6**.
44. Durrer S, Schmid-Hempel P: **Shared use of flowers leads to horizontal pathogen transmission.** *Proc R Soc Lond Ser B* 1994, **258**:299-302.
45. Stevenson PC: **For antagonists and mutualists: the paradox of insect toxic secondary metabolites in nectar and pollen.** *Phytochem Rev* 2019, **19**:603-614.
46. Stevenson PC, Nicolson SW, Wright GA: **Plant secondary metabolites in nectar: impacts on pollinators and ecological functions.** *Funct Ecol* 2017, **31**:65-75.
47. Koch H, brown MJF, Stevenson PC: **The role of disease in bee foraging ecology.** *Curr Opin Insect Sci* 2017, **21**:60-67.
48. Koch H, Woodward J, Langat MK, Brown MJF, Stevenson PC: **Flagellum removal by a nectar metabolite inhibits infectivity of a bumblebee parasite.** *Curr Biol* 2019, **29**:3494-3500.
- Developed a bioactivity-directed fractionation assay for nectar metabolites, allowing testing of secondary compounds from 17 nectar plants against *C. bombi*. Discovered an effective compound from heather nectar and are the first researchers to elucidate a mechanism by which a secondary compound reduces *C. bombi* infection.
49. Rothchild KW, Adler LS, Irwin RE, Sadd BM, Stevenson PC, Palmer-Young EC: **Effects of short-term exposure to naturally occurring thymol concentrations on transmission of a bumble bee parasite.** *Ecol Entomol* 2018, **43**:567-577.
50. Singh R, Levitt AL, Rajotte EG, Holmes EC, Ostiguy N, Vanengelsdorp D, Lipkin WI, Depamphilis CW, Toth AL, Cox-Foster DL: **RNA viruses in hymenopteran pollinators: evidence of inter-taxa virus transmission via pollen and potential impact on non-*Apis* hymenopteran species.** *PLoS One* 2010, **5**.
51. Purkiss T, Lach L: **Pathogen spillover from *Apis mellifera* to a stingless bee.** *Proc R Soc B Biol Sci* 2019, **286**.
52. Peck DT, Smith ML, Seeley TD: ***Varroa destructor* mites can nimbly climb from flowers onto foraging honey bees.** *PLoS One* 2016, **11**:e0167798.
53. Truitt LL, McArt SH, Vaughn AH, Ellner SP: **Trait-based modeling of multihost pathogen transmission: plant-pollinator networks.** *Am Nat* 2019, **193**:E149-E167.
- Developed new theory and susceptible-infectious-susceptible (SIS) models to predict how disease spreads in plant-pollinator networks that are defined by continuous trait distributions instead of species identities of bees and flowers.
54. Graystock P, Ng WH, Parks K, Tripodi AD, Muñiz PA, Fersch AA, Myers CR, McFrederick QS, McArt SH: **Dominant bee species and floral abundance drive parasite temporal dynamics in plant-pollinator communities.** *Nat Ecol Evol* 2020 <http://dx.doi.org/10.1038/s41559-020-1247-x>.
- Screened >5000 bees and flowers from fields, revealing that 42% of bee species (12.2% individual bees) and a surprising 70% of flower species (8.7% individual flowers) had at least one bee pathogen in or on them. Turnover in the bee community impacted community-wide prevalence, while turnover in the plant community impacted when pathogen transmission was likely to occur at flowers.
55. Bailes EJ, Bagi J, Coltman J, Fountain MT, Wilfert L, Brown MJF: **Host density drives viral, but not trypanosome, transmission in a key pollinator.** *Proc R Soc B Biol Sci* 2020, **287**.
- Manipulated the bee:flower ratio in experimental communities, finding that slow bee paralysis virus was transmitted more quickly and resulted in higher prevalence and infection intensity when bumble bee density was high. In contrast, there was no impact of bee density on transmission of *C. bombi*.
56. Urbanowicz C, Muniz PA, McArt SH: **Honey bees and wild pollinators differ in their preference for and use of introduced floral resources.** *Ecol Evol* 2020.
57. Giacomini JJ, Leslie J, Tarpay DR, Palmer-Young EC, Irwin RE, Adler LS: **Medicinal value of sunflower pollen against bee pathogens.** *Sci Rep* 2018, **8**:14394.
58. Vanderplanck M, Roger N, Moerman R, Ghisbain G, Gerard M, Popowski D, Granica S, Fournier D, Meeus I, Piot N et al.: **Bumble bee parasite prevalence but not genetic diversity impacted by the invasive plant *Impatiens glandulifera*.** *Ecosphere* 2019, **10**.
59. Piot N, Meeus I, Kleijn D, Scheper J, Linders T, Smagghe G: **Establishment of wildflower fields in poor quality landscapes enhances micro-parasite prevalence in wild bumble bees.** *Oecologia* 2019, **189**:149-158.
- Added wildflower strips to agricultural fields and found that the prevalence of several pathogens in *Bombus pascuorum* hosts increased with wildflower strip size, but only in landscapes with few semi-natural elements. The first study to indicate that adding pollinator habitat could increase pathogen infection and demonstrates the importance of landscape context.
60. Adler LS, Barber NA, Biller OM, Irwin RE: **Flowering plant composition shapes pathogen infection intensity and reproduction in bumble bee colonies.** *Proc Natl Acad Sci U S A* 2020, **117**:11559-11565.
- Used replicated tents with 'high-infection' and 'low-infection' wildflower strips and infected *B. impatiens* colonies. Found that 'high-infection' strips nearly doubled average colony infection intensity over two weeks compared to 'low-infection' strips. Important for suggesting that plant species in pollinator habitat may structure pathogen dynamics.