

## RESEARCH ARTICLE

# Wildflower plantings and honeybee competition impact nutritional quality of wild bee diets

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**Handling Editor:** Fabrice Requier**Abstract**

1. Wildflower habitats planted along field borders are a widely promoted strategy for supporting bees in agricultural landscapes. However, honeybees (*Apis mellifera*), which are often stocked at high densities in crop lands can compete with wild bees for pollen and nectar, potentially limiting the successfulness of wildflower plantings in supporting diverse bee communities.
2. Using weekly samples of five study sites in Northern California we assessed how plants in pollinator-friendly seed mixes varied in their ability to provide bees with abundant and nutritious pollen under intense honeybee competition.
3. We quantified pollen production, protein and lipid content, and end-of-day pollen availability for different plant species. We also sampled bee visits to flowers and assessed the composition of pollen on bee bodies. Using these data, we investigate how the nutritional quality of pollen in wildflower plantings and honeybee abundance impacted native bee pollen nutrition.
4. Bees collected more nutritious pollen (i.e. pollen with more protein) from plantings with more nutritious plant species (i.e. sites with more high-protein plants). However, as honeybee abundance increased, the nutritional quality of native bee diets declined. We also detected important interactions between honeybee abundance and the nutritional quality of flowers in plantings, such that, for some bee taxa, there was no impact of competition on pollen diet quality in high-nutrition plantings.
5. *Synthesis and applications:* Our study reveals that honeybee competition can reduce the nutritional quality of native bee diets. From an applied conservation perspective, we therefore recommend that honeybee introductions in natural areas be approached with extreme caution. However, our results also suggest that high-protein flower plantings could mitigate negative effects of honeybee competition in managed landscapes. Where simultaneous support of managed and wild bees is a key management objective, we recommend including high-protein plant species in plant mixes to support diverse bee populations.

**KEYWORDS**

competition, honeybee, native bee, nutrition, plant–pollinator interactions, pollen, protein, wildflower habitat

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## 1 | INTRODUCTION

Global food security depends on reliable pollination services for a wide variety of fruit, nut, and animal forage crops (Eilers et al., 2011). Managed honeybees and a diverse community of wild bees provide the bulk of these services, whilst simultaneously harvesting essential pollen and nectar resources from flowers. Unfortunately, as natural habitat is converted to agriculture, bee populations face two related threats: decreasing floral resource availability and decreasing resource quality (Goulson et al., 2015). Indeed, commercial beekeepers frequently cite nutritional stress associated with reduced diversity and availability of floral resources as a major cause of colony losses (Durant, 2019).

A widely promoted strategy for supporting bees is planting wildflowers along crop borders, which provide floral resources for pollinators whilst increasing pollination services to crops (Williams et al., 2015). In addition to increasing floral resource availability, informed plant species selection could also increase bee diet diversity and nutrition (Vaudo et al., 2024). For example, pollen ranges from 2.5% to 60% protein content (Roulston & Cane, 2000) and this protein is essential for ovary development and egg production in bees (Cane, 2016). Although proteins in pollen primarily function as enzymes involved in pollen tube germination and growth (Roulston et al., 2000), proteins can also attract and reward pollinators. For example, generalist bumblebees can assess the nutritional composition of pollen using chemical cues (Muth et al., 2016) and use this information to select plants with high-protein pollen (Vaudo et al., 2018). Beyond bumblebees, other bee taxa can also benefit from high-protein diets: increasing pollen protein content increases *LasioGLOSSUM* offspring growth and survival (Roulston & Cane, 2002) and improves immunocompetence in honeybees (Alaux et al., 2010). Recent studies suggest that the ratio of proteins-to-lipids in pollen (P:L ratio) is also an important component of bee nutrition. For example, bumblebees have improved survival when fed pollen with high P:L ratios and preferentially select pollen with high P:L ratios under both field and laboratory conditions (Vaudo et al., 2018). As such, variations in the protein and lipid content of pollen in wildflower plantings could impact the nutrition of both managed and wild bee diets.

Although wildflower plantings can benefit both honeybees (Decourtye et al., 2010) and wild native bees (Williams et al., 2015), managing plantings to support diverse bee populations is complicated by the fact that abundant honeybees can compete with native bees and reduce pollen and nectar in flowers (Cane & Tepedino, 2016; Page & Williams, 2023). Indeed, a recent literature review suggests that honeybee competition often has negative impacts on native bee reproductive success (Iwasaki & Hogendoorn, 2022), and wildflower plantings may fail to benefit native bees under intense honeybee competition (Angelella et al., 2021). Mechanistically, we generally assume that exploitative competition affects bees via quantitative changes in resource collection and energy expenditure. For example, bees may respond to honeybee competition by spatially re-locating (Herbertsson et al., 2016) or by collecting fewer resources per flower (Henry & Rodet, 2018), both of which could

increase energy costs associated with foraging (Thomson, 2004), and ultimately decrease bee fitness in cases where floral resource availability limits population growth (Thomson & Page, 2020).

Although most studies of honeybee competition focus on quantitative impacts (e.g. altered resource availability), qualitative changes in diet composition and nutrition may also be occurring when competitive pressures are high. For example, we know that bees forage adaptively (Valdovinos et al., 2016) and often respond to competition by visiting different resources (niche partitioning: Schoener, 1974) or by narrowing their diet breadth (Magrath et al., 2017; Page & Williams, 2023). Such niche partitioning could allow bees to avoid directly competing with one another for floral resources (Valdovinos et al., 2016), implying that diverse flower plantings could lessen or even fully negate the negative effects of competition. Indeed, beekeepers frequently cite the diverse pollen use of native bees as a rationale for being less concerned about honeybee competition negatively affecting native bee populations (M. Page, personal communications). However, if native bees respond to competition by shifting their visits to less-preferred species, they may compromise the nutritional quality of their pollen diets. Such decreases in diet quality could negatively affect bee health and offspring survival (Roulston & Cane, 2002) even when total pollen collection and energy expenditure remain unaffected by competition. However, the theory that honeybee competition and associated diet shifts would reduce the nutritional quality of native bee diets remains hereto untested. Although such interactions operate at the minute level of pollen grains, understanding how competition impacts pollen diet quality may nonetheless reveal an underappreciated facet of how competition unfolds in natural systems.

Our objectives for this study were to (i) evaluate the pollen nutrition of several plant species commonly used in 'pollinator-friendly' wildflower plantings; and (ii) assess how the nutritional quality of pollen in wildflower plantings and honeybee competition individually and interactively influence the nutritional quality of native bee pollen diets. Our null expectation was that bees would collect more nutritious pollen when high-quality pollen resources were more abundant in plantings. However, we also hypothesized that the average nutritional quality of bee-collected pollen would be higher than the average quality of available pollen (revealing selective foraging) and that the nutritional quality of pollen collected by native bees would decrease as honeybee competition increased. We use data on pollen availability and pollen collection by bees to test these hypotheses and explore potential mechanisms underpinning observed changes in native bee pollen nutrition. We also discuss results in the context of improving plant selection for wildflower mixes to support wild and managed bees.

## 2 | MATERIALS AND METHODS

### 2.1 | Study sites and pollinator surveys

We conducted this work in the California Central Valley at five replicated wildflower plantings neighbouring conventionally managed Almond orchards. Wildflower plantings were established in 2015

and 2016 as 632–3612 m<sup>2</sup> plots using a consistent seeding mix of diverse plant species native to California. Sites averaged 11.3 km to the nearest neighbouring site and were located within 0–30 m of semi-natural riparian habitat. Sites varied in honeybee abundance due to spatial variation in the placement of commercial apiaries (see [Figure S1](#) in Supporting Information), and the abundance of honeybees visiting flowers was not correlated with planting size (Pearson's correlation coefficient = 0.107,  $p = 0.865$ ). In 2017 and 2018, we surveyed bees and their visits to flowering plants over four sample rounds from April to May. During this period, Almond flowers are no longer blooming, but many beekeepers maintain apiaries adjacent to Almond groves whilst they wait to move hives to pollinate different crops or to higher elevations for honey production. We netted insects actively visiting flowers during 10-min walks of two 100 m<sup>2</sup> transects sampled once in the morning and once in the afternoon (40 min total). Netted pollinators were collected individually in separate vials to minimize pollen contamination and euthanized using dry ice, except for bumblebee queens, which we identified, swabbed for pollen, and released. We collected up to 20 honeybees during netting transects and counted any additional honeybees. On the same day and in the same transects where bees were collected, we assessed flowering species composition by counting and identifying all flowers in 10 evenly spaced 1 m<sup>2</sup> quadrats. At the end of each sampling day, we also assessed pollen availability for the most abundant and well-represented plant species: *Clarkia unguiculata*, *Clarkia williamsonii*, *Collinsia heterophylla*, *Eschscholzia californica*, *Lupinus densiflorus*, and *Phacelia californica*. We measured pollen availability as the proportion of dehiscent anthers with pollen visible to the naked eye, using one to three flowers on 10–20 'open-pollinated' (i.e. unmanipulated) plants. Bees were identified to species or morphospecies by expert taxonomists (Skyler Burrows, USDA Bee Lab, Logan, Utah, and Joel Gardner, University of Manitoba, Canada). In statistical analyses described below, we grouped bees into five categories: *Apis mellifera*, *Bombus* spp., Megachilidae, Halictidae, and 'Other bees' (comprising Andrenidae, Colletidae, and non-corbiculate Apidae). We attempted to group at the lowest taxonomic level possible without creating sparse categories, hence why some taxa are grouped at the genus level and others at the family level. The study did not require approval from an animal ethics committee or any permits. Further details regarding site selection, wildflower establishment, and bee identification are described in Page and Williams (2023).

## 2.2 | Assessing pollen diet composition and nutritional quality of different pollen species

In the lab, we used fuchsin-tinted jelly cubes to remove and stain pollen from bee bodies, which we then melted onto microscope slides (Kearns & Inouye, 1993). When swabbing bees, we focused our effort on scopae (i.e. pollen from specialized pollen-collection hairs), but also lightly brushed the head and thorax. For *Apis* and *Bombus*, we used tweezers to dislodge small clumps of pollen from multiple areas of corbiculae (i.e. pollen baskets). We identified

pollen to species using light microscopy (Nikon Eclipse 80i, Nikon Instruments Inc.) and a pollen reference collection. In 2018, we assessed pollen from corbiculae and scopae separately from pollen removed from other body parts but ultimately pooled pollen from both sources for each specimen in our analyses. As described in the Discussion, pooling pollen did not qualitatively affect statistical results. To assess the nutritional composition of pollen loads, we grew monospecific flower plantings for each of the 10 most-used plant species (representing 96% of floral visits and 92% of bee-collected pollen). We covered each planting with organza fabric prior to anthesis to prevent insect visitation, collected fresh pollen from flowers, and assessed pollen protein and lipid content using methods outlined by Vaudo et al. (2020). As we harvested pollen for macronutrient analyses, we also collected data on the number of florets sampled and the mg of pollen extracted from those florets, giving us an estimate of pollen production per floret. Using data on the composition of pollen on bee bodies and information on the protein and lipid content of pollen from different plant species, we calculated the mean protein content and protein-to-lipid (P:L) ratios of pollen collected by each bee specimen sampled. Throughout the paper, we refer to the protein and P:L ratios of bee-collected pollen as the quality of 'collected pollen'.

## 2.3 | Estimating nutritional quality of wildflower plantings at the time of sampling

We estimated nutrient availability in wildflower plantings for each site sample by calculating the mean protein-to-lipid (P:L) ratio and protein content of available pollen. To do this, we used data on the composition of floral resources available in netting transects for which we had data on protein and lipid content (representing 96% of visited flowers and 92% of collected pollen). To account for the fact that not all plants produce the same amount of pollen, we weighted the contribution of different plant species by their pollen abundance, where pollen abundance was calculated as the number of florets of a given species counted during a site sample multiplied by the pollen production per floret of that same species. Throughout the article, we refer to the estimated nutrition of pollen in plantings as 'planting nutrition'.

## 2.4 | Statistical analysis

We assessed whether plant species varied in their likelihood of having pollen in anthers at the end of the day by fitting a generalized linear mixed model (GLMM) with plant species sampled as a fixed effect as well as site, sample round, and plant ID (to account for cases where multiple flowers were sampled on the same plant) as separate random effects. We modelled pollen availability, our response variable, at the level of the individual flower, as the proportion of dehiscent anthers that had visible pollen using a beta-binomial error distribution. We modelled GLMMs using the `glmmTMB` package

(Brooks et al., 2017) in R (R Core Team, 2022) and tested for significance using a likelihood ratio test. Pairwise differences amongst plant species were further evaluated using post hoc Tukey tests via the 'emmeans()' function.

We assessed how the nutritional quality of collected pollen varied amongst bee taxa by fitting separate linear mixed models (LMMs) for two response variables measuring different aspects of pollen quality. These response variables were: (i) protein content, and (ii) P:L ratios of collected pollen, each measured at the level of the individual bee. These two variables are correlated (Pearson's correlation coefficient=0.408,  $p < 0.001$ ), but we present results from both because both measures are commonly used in the bee nutrition literature. Each model included year (with two levels: 2017 or 2018), bee taxon (with five levels: *Apis mellifera*, *Bombus* spp., Halictidae, Megachilidae, and 'Other bees'), and either the estimated protein content or the estimated P:L ratio of pollen available in plantings (hereafter, 'planting nutrition') as fixed effects and site and sample round as separate random effects. We modelled LMMs using the lmer() function in the lme4 package (Bates et al., 2014) and tested for significance using likelihood ratio tests. We evaluated pairwise differences amongst bee taxa using post hoc Tukey tests via the 'emmeans()' function.

We assessed how honeybee abundance and planting nutrition individually and interactively affected the quality of collected pollen by fitting separate linear mixed models (LMMs) for two response variables: (i) protein content, and (ii) P:L ratios of collected pollen, each measured as the level of the individual bee. Each model included planting nutrition, year, native bee taxon, honeybee abundance (measured as the total number of honeybees visiting flowers during morning and afternoon netting transects), the interaction of honeybee abundance and bee taxon, the interaction of honeybee abundance and planting nutrition, and a three-way interaction between honeybee abundance, planting nutrition, and bee taxon as fixed effects as well as site and sample round as separate random effects. We tested for the significance of fixed effects using a likelihood ratio test.

Analyses described above revealed statistically significant three-way and two-way interactions between honeybee abundance, planting nutrition, and bee taxon (Table S1). As such, we also fit separate LMMs for the three most abundant and well-represented native bee taxa: *Bombus* spp. Halictidae, and Megachilidae. For each taxon and for each response variable (protein content of collected pollen and P:L ratios of collected pollen), we assessed the individual and interacting effects of honeybee competition and planting nutrition by fitting LMMs with year, honeybee abundance, planting nutrition, and the interaction of honeybee abundance and planting nutrition as fixed effects as well as site and sample round as separate random effects. To correct for multiple testing, we applied Bonferroni corrections to all p-values.

### 3 | RESULTS

Honeybee abundance in wildflower plantings, measured as the total number of honeybees visiting flowering plants during morning and afternoon netting transects, ranged from 9 to 2363 bees per m<sup>2</sup>

per hour, representing 93% of all flower visits by bees (Table S2). In total, we collected 715 honeybees and 1021 native bees comprising 49 morphospecies across 2 years of sampling to assess pollen diet composition (Table S2). Both honeybees and native bees collected diverse pollen from wildflower plantings and planted forb species comprised 93% of all pollen grains counted on bee bodies (Table S3; Figure S2).

#### 3.1 | Nutritional quality of pollen from different plant species

Plant species differed in the abundance of pollen produced (mg/floret & mg/cm<sup>2</sup>) and in the protein and lipid content of their pollen (Table S4). *Lupinus* and *Phacelia* spp. had the highest P:L ratios and *Lupinus succulentus*, *Collinsia heterophylla*, and *Phacelia* spp. produced pollen with the highest protein content. *Eschscholzia californica*, *Clarkia unguiculata*, and *Lupinus densiflorus* produced the most pollen per floret. Plant species also varied in the amount of pollen that was 'leftover' in anthers at the end of a day of bee foraging activity ( $\chi^2 = 110.070$ ,  $p < 0.001$ ; Figure 1;  $N = 518$  flowers sampled). A significantly higher proportion of *Clarkia* spp., and *Lupinus densiflorus* anthers had pollen left at the end of the day when compared with other species (Table S5;  $p < 0.01$  for all comparisons). *Collinsia heterophylla* had more pollen than *Eschscholzia californica* (Table S5;  $p = 0.021$ ), and *Eschscholzia californica* had more pollen than *Phacelia californica* (Table S5;  $p < 0.001$ ).

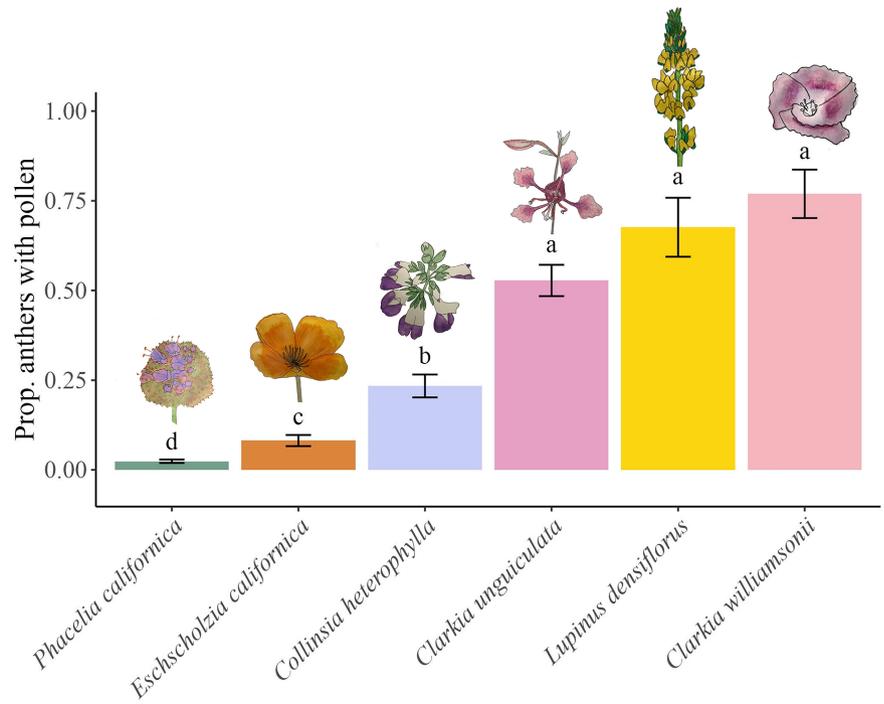
#### 3.2 | Variance amongst bee taxa in pollen diet quality

The nutritional quality of collected pollen varied by bee taxon (Table S6; P:L ratios:  $\chi^2 = 225.810$ ,  $p < 0.001$ ; Protein:  $\chi^2 = 128.565$ ,  $p < 0.001$ ;  $N = 1736$  bees sampled). *Bombus* spp. and Megachilid bees collected pollen with higher protein content when compared with *Apis mellifera* and Halictid bees (Figure 2a;  $p < 0.01$  for all comparisons). *Bombus* spp. collected higher P:L ratios when compared with all other species ( $p < 0.001$  for all comparisons). Halictid bees collected pollen with higher P:L ratios than *Apis mellifera* and Megachilid bees (Figure 2b;  $p < 0.05$  for all comparisons) and *Apis mellifera* collected pollen with higher P:L ratios when compared with Megachilid bees ( $p < 0.001$ ). For all pairwise comparisons, see Table S7. For species-level summary data, see Table S8.

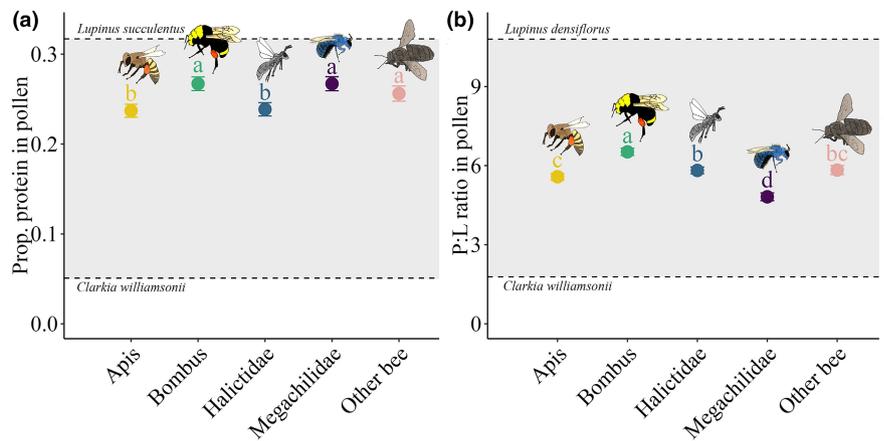
#### 3.3 | Impact of honeybee competition on native bee pollen nutrition

We detected statistically significant three-way and two-way interactions between honeybee abundance, bee taxon, and planting nutrition on the nutritional quality of native bee diets (Table S1;  $N = 1021$  native bees sampled). As such, we followed up with single

**FIGURE 1** Pollen availability, measured as the proportion of anthers with pollen available at the end a sampling day, for different plant species sampled in wildflower plantings in 2018. Letters above bars indicate significance for pairwise comparisons at  $p < 0.05$ . Error bars show standard error. Sample sizes by species: *Phacelia californica* (144), *Eschscholzia californica* (128), *Collinsia heterophylla* (125), *Clarkia unguiculata* (76), *Lupinus densiflorus* (21), *Clarkia williamsonii* (24). For all pairwise comparisons, see [Table S5](#).



**FIGURE 2** Bee taxa collected pollen with different (a) protein content and (b) P:L ratios. Letters above bars indicate significance for pairwise comparisons at  $p < 0.05$ . Grey shading depicts the range of possible values, bounded by the protein content and P:L ratios of plants with the lowest and highest values (*Clarkia* and *Lupinus* spp.). Error bars show standard error. Sample sizes by taxon: *Apis* (715), *Bombus* (273), Halictidae (439), Megachilidae (206), Other bee (103).

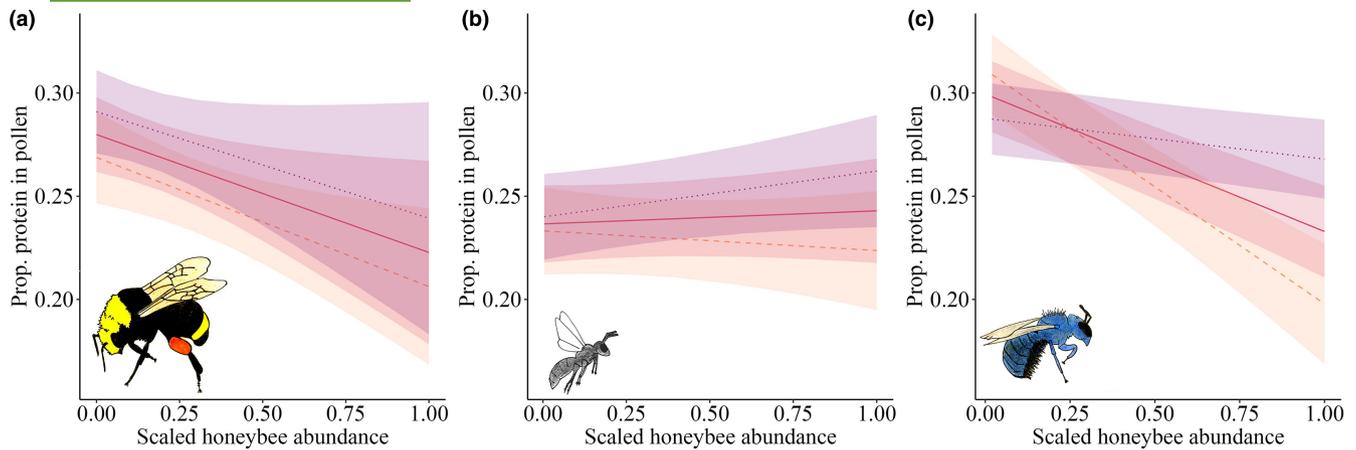


taxon analyses. For *Bombus* spp., honeybee abundance had an overall negative effect on the protein content of collected pollen and this effect was consistent in both high-nutrition and low-nutrition wildflower plantings ([Figure 3a](#); [Table S9](#);  $\chi^2 = 6.583$ ,  $N = 273$ ,  $p = 0.031$ ). For Megachilid bees, there was also an overall negative effect of honeybee abundance ([Table S9](#);  $\chi^2 = 9.976$ ,  $N = 206$ ,  $p = 0.005$ ), but the magnitude of this effect depended on the nutritional quality of wildflower plantings ([Figure 3c](#);  $\chi^2 = 37.929$ ,  $N = 206$ ,  $p < 0.001$ ), with stronger negative effects as planting nutrition decreased. For Halictidae, there was no impact of honeybee abundance on the protein content of collected pollen ([Figure 3b](#); [Table S9](#)). There was no impact of increasing honeybee abundance on the P:L ratios of collected pollen for *Bombus* spp. or Megachilidae nor was there a significant interaction of planting nutrition and honeybee abundance. For Halictidae, we observed a significant interaction between planting nutrition and honeybee abundance on the P:L ratios of collected

pollen ([Figure S3](#); [Table S9](#);  $\chi^2 = 13.307$ ,  $N = 439$ ,  $p < 0.001$ ), such that the effect of honeybee abundance was subtly positive in high-nutrition plantings and subtly negative in low-nutrition plantings.

## 4 | DISCUSSION

Our study confirms that the nutritional quality of wildflower plantings and honeybee competition individually and interactively impact the nutritional quality of wild bee diets. Previous work in this system revealed that honeybee competition led to important shifts in floral visitation patterns (Page & Williams, 2023), and here we show that these diet shifts resulted in reduced protein content of native bee diets. Importantly, the tendency for honeybee competition to decrease native bee nutrition varied by bee taxon, with stronger negative effects observed for *Bombus* spp. and Megachilid bees in



**FIGURE 3** The effect of increasing honeybee abundance on the protein content of collected pollen for (a) *Bombus* spp., (b) Halictidae, and (c) Megachilidae at different levels of estimated planting nutrition: Median nutrition (solid pink lines), median – SD (dashed orange lines), and median + SD (dotted purple lines). Lines show predicted model fit and shading depicts 95% confidence intervals around point estimates. For *Bombus* spp. and Megachilid bees, honeybee abundance had a significantly negative effect on the protein content of collected pollen (*Bombus*:  $\chi^2 = 6.583$ ,  $N = 273$ ,  $p = 0.031$ ; Megachilid:  $\chi^2 = 9.976$ ,  $N = 206$ ,  $p = 0.005$ ). We also observed a significant interaction between planting nutrition and honeybee abundance for Megachilid bees ( $\chi^2 = 37.929$ ,  $p < 0.001$ ), but not for *Bombus* or Halictidae.

wildflower plantings dominated by less nutritious plant species. This result is, to our knowledge, amongst the first to experimentally demonstrate that honeybee competition may negatively impact native bees via changes in nutrient availability, leading to altered nutritional quality of native bee diets. Although less well-studied than resource scarcity, decreased diet quality may negatively impact native bee populations. For example, nutritional stress can amplify the negative effects of other stressors such as disease (Dolezal & Toth, 2018) and pesticide exposure (Stuligross & Williams, 2020), and decreased pollen protein content can reduce larval survival and adult body size (Roulston & Cane, 2002). Ultimately, we can only speculate as to whether the decreased pollen nutrition associated with honeybee competition negatively impacts native bee fitness, but the effect is likely to be negative for some bees, particularly in environments where bees face multiple stressors.

On a more positive note, our study also suggests that the negative effects of honeybee competition could be partially mitigated by planting diverse, nutritious floral resources. For example, Megachilid diet quality was seemingly unaffected by honeybee competition in high-nutrition wildflower plantings. This finding complements other recent studies which demonstrate that competition between honeybees and wild bees is more likely to occur in low-quality floral resource landscapes (Casanelles-Abella et al., 2023). We also found no evidence that honeybee competition negatively impacted the nutritional quality of Halictid pollen diets. This is curious because these bees strongly overlap with honeybees in their use of pollen resources (Figure S2). On the other hand, the Halictid bees in our data set primarily comprise small-bodied species (e.g. *Lasioglossum* spp.) and a recent review suggested that smaller bees may be less impacted by honeybee competition relative to bigger taxa like *Bombus* spp. (Iwasaki & Hogendoorn, 2022). Large-bodied bumblebees have high energy needs (Henry & Rodet, 2018), and this may explain why they are more sensitive to honeybee competition.

Another possibility is that observed effects are partially driven by species turnover in the bee community. This is unlikely to be the case for bumblebees; we find a strong negative effect of honeybee competition even when we restrict analyses to only include the most abundant species, *Bombus vosnesenskii* (Figure S4;  $\chi^2 = 10.885$ ,  $N = 185$ ,  $p < 0.001$ ). However, the most abundant Megachilid bee in our dataset is *Osmia glauca*, and negative effects for this species are subtle and not statistically significant. Importantly, *Osmia glauca* is an oligolectic bee specializing on *Collinsia heterophylla* (Rust & Clement, 1972), thus, although negative results could be attributable to species turnover, they could also be driven by generalist Megachilids, whose diets are more flexible. Ultimately, our results suggest that the nutritional quality of floral landscapes and traits like bee body size and diet breadth likely mediate the impacts of honeybee competition, but more work is needed to understand which species are most susceptible.

Bees collected more nutritious pollen than we would expect if they were exclusively foraging based on the relative abundance of pollen and flowers in plantings, but only when the estimated nutritional quality of available pollen was low (Figure S5). In low-nutrition plantings, where the mean protein content of available pollen was 19% (averaged across all plantings with a nutrition score below the median), bees collected pollen with a mean protein content of 23%. In contrast, in high-nutrition plantings, the mean protein content of available pollen was 27% and bees collected pollen with a mean protein content of 27%. Although many things influence bee foraging decisions, these data lend support to the theory that insects forage to optimize macronutrient intake (Pulliam, 1975), but also suggest that increasing the protein content of available pollen resources may be less important beyond a certain threshold (Roulston & Cane, 2002). We also observed considerable variation amongst taxa in the protein content and P:L ratios of the pollen they collected. *Bombus* spp. collected higher

P:L ratios than all other species and *Bombus* spp. and Megachilid bees collected pollen with higher protein content when compared with honeybees and Halictid bees. The tendency for *Bombus* to collect more nutritious, protein-rich pollen relative to other taxa is consistent with other studies (e.g. Vaudo et al., 2020) and it is tempting to speculate as to whether high protein requirements render *Bombus* more susceptible to nutritional competition.

Plants varied in their ability to provide native bees with nutritious and abundant pollen under intense honeybee competition. Anecdotally, species which produce more pollen seem more likely to have pollen left in anthers at the end of the day. Unfortunately for bees, plant species which have abundant pollen “leftover” are either of low nutritional quality (e.g. *Clarkia* spp.) or have hard-to-access pollen (e.g. *Lupinus* spp.). *Lupinus* spp. was rarely visited by honeybees in our system (less than 1% of their visits and 1.5% of collected pollen), and although honeybees frequently visited *Clarkia* spp. (nearly 16% of floral visits) they collected very little *Clarkia* spp. pollen (only 1.3% of their pollen diet). Given that honeybees represented nearly 93% of all flower visits from bees in our system, their distaste for these species could partially explain why these plants often had leftover pollen.

The data we provide on the pollen quality of several plant species commonly used in pollinator-friendly wildflower plantings could be useful in optimizing plant mixes to support diverse bee communities. However, bees also need high-quality nectar and three of the plants with the highest pollen quality (*Eschscholzia californica*, *Lupinus densiflorus*, and *Lupinus succulentus*) do not produce nectar. In fact, when measuring different aspects of nectar quality (including nectar refill rates, nectar volume, and sugar concentration per floret: see Table S10) we found that *Clarkia* spp. provide abundant high-quality nectar despite having relatively poor pollen. This makes it difficult to “rank” plants but also highlights the importance of providing bees with diverse flowers to account for both the diverse dietary needs of different species and the different types of resources provided by different plants. Ultimately, plant mixes should be tailored to offset whatever nutrient is most lacking in the landscape (Filipiak et al., 2022), and we hope the data we provide on pollen and nectar quality (Table S4 and S10) can inform plant species selection for pollinator-friendly habitats. Given that phylogenetically related plants have similar protein levels (Vaudo et al., 2024), these data could also be adapted to other regions by substituting locally adapted but phylogenetically similar species.

An important caveat to this study is that we included all pollen on bee bodies in our assessment of their diets. This measure would also include pollen from plants that bees were visiting for nectar (i.e. cases where a small amount of pollen was incidentally transferred to the bee during its visit). However, when considering only scopal and corbicular pollen, we still find a statistically significant negative effect of honeybee abundance on the protein content of native bee pollen diets (Figure S6;  $\chi^2 = 6.906$ ,  $N = 539$ ,  $p = 0.009$ ). As such, our findings are robust, but it is also possible that observed changes in pollen diet quality are partially reflective of bees spending more time nectar foraging and less time pollen foraging. This mechanistic explanation is further supported by the fact that plants with poor pollen quality also had

high nectar refill rates (Table S10), and the fact that honeybee abundance decreased nectar availability in wildflower plantings (based on a study at the same sites and time-period: Page & Williams, 2023). In other words, native bees may be responding to competition by shifting their visits to plants with high nectar quality, and in so doing, compromising the quality of their pollen diets. Although speculative, this explanation has good support from the literature, and many studies demonstrate that bees must balance nectar and pollen needs and redistribute foraging visits to acquire whichever resource is more lacking (Francis et al., 2016; Plowright et al., 1993).

Another important caveat is that protein is not the only measure of pollen quality. Micronutrients are important for the detoxification of pesticides and immune functioning (Dolezal & Toth, 2018), and the chemical and morphological qualities of some pollen can reduce bee pathogen loads (Figuerola et al., 2023). Additionally, some of the bees we sampled are oligolectic (e.g., *Osmia glauca*), and their diets may be more reflective of the abundance of preferred host plants. Nonetheless, protein levels have generally been considered a universally good measure of pollen quality. Protein is essential for ovary development and egg production (Cane, 2016), and diets with more protein increase offspring body size (Roulston & Cane, 2002). Additionally, bee-pollinated plants have higher protein, suggesting the evolutionary importance of protein in guiding bee foraging choices (Hanley et al., 2008). Furthermore, although bees like *Osmia glauca* are considered oligolectic, they collect less pollen from their preferred host plant (*Collinsia heterophylla*) when honeybees are abundant (based on scopal loads: Figure S7). As such, competition-mediated declines in the protein content of native bee pollen diets have the potential to negatively impact bee health across multiple taxa with different life histories and may even disrupt highly specialized plant-pollinator mutualisms.

## 5 | CONCLUSIONS

Our study demonstrates that competition may reduce diet quality, particularly when few nutritious alternatives are available. Although this is theoretically well-founded (DeBach, 1966) and well-documented in other ecological systems (Pimm et al., 1985), our study is amongst the first to document this phenomenon in bees. The idea that the negative impacts of competition would be more severe in low-nutrition and resource-limited landscapes has been proposed by reviews and conceptual papers (Thomson & Page, 2020), and our study empirically confirms that resource competition is indeed a product of both competition and variation in background resource availability. Our results also have important applications for the conservation and management of bees. First, our results show that honeybee introductions may reduce native bee nutrition and negatively affect wild pollinator populations, and these negative effects need to be carefully considered when deciding whether to place apiaries in natural ecosystems, especially in areas that support rare and endangered species. Second, and more optimistically, our results suggest that high-protein flower plantings could be used as

a tool to relax honeybee competition in managed ecosystems, potentially reducing the need for beekeepers to migrate to protected areas (Durant, 2019). Lastly, our study suggests that honeybee competition and protein availability should both be considered when selecting plants for restoration. We hope our study will motivate others to investigate impacts of competition and other stressors on bee nutrition and improve the optimization of plantings to support diverse pollinator populations.

#### AUTHOR CONTRIBUTIONS

Maureen L. Page and Neal M. Williams planned the study. Maureen L. Page collected and analysed the data and led the writing of the manuscript. Jacob S. Francis and Uta Müller helped collect pollen nutrition data. All authors read and approved the final manuscript.

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#### CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

#### DATA AVAILABILITY STATEMENT

Data available from the Dryad Digital Repository: <https://doi.org/10.5061/dryad.br15dvjt> (Page et al., 2024).

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## SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

**Table S1.** Impact of honeybee (HB) abundance, bee taxon, estimated nutritional quality of plantings, and their interactions on the nutritional quality of pollen collected by native bees.

**Table S2.** Summary data for honeybee and native bee abundance across sampled sites, where abundance is measured as the number of bees collected or counted per hour per m<sup>2</sup> summed across morning and afternoon netting transects.

**Table S3.** Summary information on pollen grains counted across all bee specimens.

**Table S4.** Summary information on pollen production per floret, floral surface area, and pollen nutritional data for species in wildflower plantings.

**Table S5.** Pairwise comparisons of the proportion of anthers with pollen available at the end of the day for different plant species sampled in 2018.

**Table S6.** Impact of estimated nutritional quality of pollen in wildflower plantings, bee taxon, and year on protein content and protein:lipid ratios (P:L ratios) of bee-collected pollen.

**Table S7.** Pairwise comparisons of the protein content and protein:lipid ratios (P:L ratios) of pollen collected by different bee taxa.

**Table S8.** Average protein content and protein:lipid ratios (P:L ratios) of bee-collected pollen, summarized at the level of genus and species.

**Table S9.** Effect of honeybee abundance ('HB abundance' or 'HB'), planting nutrition, and their interaction on protein content and protein:lipid ratios (P:L ratios) of bee-collected pollen for different native bee taxa.

**Table S10.** Summary information on nectar sugar concentration (measured in the lab using a handheld refractometer from cut flowers harvested from unvisited mono-specific flower plantings in 2020), nectar production per floret (measured in the field using 1 µL microcapillary tubes in 2021 from flowers that had been bagged with organza pollinator exclusion bags for 24 h to allow for nectar accumulation), and nectar refill rates (also measured in 2021, on the

same flowers from which nectar production was measured, assessed by re-bagging flowers after removing all nectar and then waiting 1.5–3 h before re-measuring nectar using 1  $\mu$ L microcapillary tubes).

**Figure S1.** Sites and apiary locations. Blue stars are sampled sites and orange markers are where apiaries are located.

**Figure S2.** Average composition of pollen diets for different pollinator taxa when honeybee abundance was (A) low (<300 honeybees foraging during netting transects per hour per m<sup>2</sup>), and (B) high (>300 honeybees foraging per hour per m<sup>2</sup>).

**Figure S3.** The effect of increasing honeybee abundance on protein:lipid (P:L ratios) of bee-collected pollen for (A) *Bombus* spp., (B) Halictidae, and (C) Megachilidae at different levels of estimated planting nutritional quality: median nutrition (solid pink lines), median – SD (dashed orange lines), and median + SD (dotted purple lines).

**Figure S4.** The effect of increasing honeybee abundance on protein content of *Bombus vosnesenskii* pollen.

**Figure S5.** Effect of estimated nutritional quality of pollen in wildflower plantings on (A) protein content and (B) P:L ratios for bee-collected pollen.

**Figure S6.** The effect of increasing honeybee abundance on (A) protein content and (B) protein:lipid (P:L) ratios of pollen in scopal and corbicular loads from native bees collected in 2018.

**Figure S7.** Comparison of *Osmia glauca* pollen diets when honeybee abundance was (A) low (<300 honeybees foraging during netting transects per hour per m<sup>2</sup>), and (B) high (>300 honeybees foraging per hour per m<sup>2</sup>).

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