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Native Pollinators (Hymenoptera: Anthophila) in Cotton Grown in the Gulf South, United States

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Abstract: Native bees (Hymenoptera: Anthophila) were sampled using bee bowls in two states to determine biodiversity in commercial cotton fields of the southern United States. In both states, native bee communities found in cotton fields were dominated by generalist pollinators in the genera *Agapostemon*, *Augochloropsis*, *Halictus*, and *Lasioglossum* (Hymenoptera: Halictidae), and *Melissodes* (Hymenoptera: Apidae). *Melissodes tepaneca* (Cresson) was the most abundant species found in cotton fields in both states. Some species collected are known specialists on other plant taxa, suggesting they may be tourist species. Here we provide a baseline species list of native bees found in cotton. Ordination indicated separation between the communities found in the two states when pooled by genus, but these differences were not significant. While cotton is grown in highly managed and disturbed landscapes, our data suggest that a community of common generalist native pollinators persists. Many of these species are also found in other cropping systems across North America.

Keywords: *Melissodes tepaneca*; Mississippi; bee diversity; Texas; pollinator community

1. Introduction

The availability of mass flowering crops across the landscape in agricultural areas can have a positive impact on the density of generalist native pollinators [1]. Cotton is an important agronomic crop, planted on 13.4 million acres in 17 states across the southern United States in 2018 [2]. Insect pest management in commercial cotton (*Gossypium hirsutum* L.) production has changed in the past few decades due to the eradication of the boll weevil (*Anthonomus grandis* Boheman) and introduction of varieties expressing toxins derived from *Bacillus thuringiensis* (Berliner) for the control of heliothine pests (Lepidoptera: Noctuidae) [3]. As heliothine pests have decreased, various plant bugs and stink bugs (Hemiptera: Miridae and Pentatomidae) have emerged as primary pests across cotton growing regions [4–8]. Although these events have led to a substantial overall reduction in total insecticide use, there remains an average of two spray applications of broad-spectrum insecticide to control insect pests on cotton each year [2,3].

Much of the available literature on pollinators in cotton grown in the United States is over 30 years old, prior to changes in cotton production mentioned above. In that time, cotton acreage in the state of Mississippi has fluctuated annually, with highs of over a million acres to a low of approximately 300,000 acres in 2013, with acreage increasing every year since that low [9]. Texas upland cotton acreages also fluctuate, with between 5 and 7 million acres planted annually [9]. Much of the historical work on pollinators in cotton was done either in the Texas panhandle or in Arizona as part of a larger effort to identify potential native pollinators for economically feasible hybrid cotton seed production [10–17]. More recent research from southern Texas showed that native bee abundance and diversity increases with cotton bloom density and the abundance of semi-natural habitat [18]. Additional studies have shown that historical land usage often has long-lasting effects on bee community composition [19].

Many methods can be used to collect and sample native bees in various habitats, and each of these potential methods have inherent biases. Bee bowls, also commonly referred to as modified pan traps, have been used across a wide variety of plant communities and crops in varying geographic regions to examine bee communities [20–23]. While these traps are efficient and easy to use, they often exhibit bias in the species they collect, often failing to collect larger bodied bees like carpenter bees (*Xylocopa* spp.) and bumblebees (*Bombus* spp.) [24]. Bee bowls also collect some groups of native bees less often than their perceived or visually confirmed abundance, especially honey bees (*Apis mellifera* L.) and cellophane bees (*Colletes* spp.) [25,26]. Given these considerations, bee bowls are the most effective and cost-efficient trap for targeting native bees in agricultural cropping systems [24,27].

While it is apparent that cotton is widely planted across the southern United States and is an economically important crop with abundant pollen through its growing season, knowledge of the biodiversity within the community of native bees utilizing this resource remains unknown. Understanding the community of native bees in this region can inform management decisions by providing baseline data with modern agricultural practices. Our research goals for this project were to document the native bee fauna present in commercially managed cotton fields and present a checklist of species as a foundation for future research studies. Here we characterize and compare the current communities of bees visiting cotton fields in two states along the Gulf Coast of the United States.

2. Materials and Methods

2.1. Study Systems

Collections of native bees in the Mississippi Delta were made in commercial cotton fields during the summers of 2015 and 2016. Producers in the region typically plant a mixture of cotton, corn (*Zea mays* L.), and soybean (*Glycine max* (L.) Moench.). Many also plant smaller acreages of sunflowers (*Helianthus annuus* L.), sorghum (*Sorghum bicolor* L.), rice (*Oryza sativa* L.), and sweetpotato (*Ipomoea batatas* (L.) Lam.). In 2015, two commercial cotton fields in Sunflower County near the town of Indianola, Mississippi [MS] were sampled. In 2016, one cotton field near Indianola, MS (Sunflower County), and one located near the town of Charleston, MS (Tallahatchie County), were sampled for bees.

Collections made in South Texas were also in large commercial cotton fields located near the town of Kingsville, Texas in Kleberg County. Fields that were sampled spanned a total area of roughly 175 km^2. This region's commercial farms are generally composed of a rotation of dryland cotton and sorghum with natural coastal prairie habitat intermixed throughout the region. These samples were part of a larger project examining native pollinator communities at the interface of cotton fields with sorghum, semi-natural habitat, or other cotton fields. Therefore, for both regions, data were taken from cotton fields within areas with mixed croplands and semi-natural habitats.

2.2. Bee Collections

In Mississippi, commercial cotton fields were sampled starting at first bloom in July for nine weeks during the summer of 2015. In 2016, two locations were sampled beginning in July for six to eight weeks. Locations were sampled both years only while there were blooms in the field. At each location, ten bee bowl units per field were placed in two parallel transects 10 m apart, with each transect containing five bee bowl units each 5 m apart. Each unit consisted of three 3.25 oz solo cups, one of each painted a flat white, fluorescent blue, and fluorescent yellow (Figure 1). A total of 30 individual bowls (10 of each color) were collected at each location weekly. Each bowl was filled two thirds full of soapy water and placed in the field for 24 h at each sampling date.

Figure 1. Example of modified pan traps, also called bee bowls, placed in a cotton field.

In Texas, similar collections were made during the summers of 2017 and 2018. In 2017, three bee bowl units were placed in cotton at each of three sampling locations for a total of nine units per week. Traps were placed out in May at the first week of bloom for approximately six weeks, totaling 86 collection events in 2017. Some weeks, not all traps could be collected due to weather or road conditions and were collected as soon as possible at the next available date. In 2018, the sampling effort was increased to five bee bowl units in five locations for a total of 25 traps per collection week. Again, bee bowls were placed out at first bloom in July for four weeks for a total of 188 collection events.

2.3. Specimen Identification

All specimens were temporarily stored in 70% ethanol, then pinned and preserved following previously published guidelines [28]. Insect specimens were processed by sorting specimens to morphospecies, and specimens were identified to genus using general keys [29–32]. Following is a list of genera and corresponding primary literature used for identifications: *Agapostemon* [33], *Anthophora* [34], *Augochlora* and *Augochloropsis* [30,35], *Augochlorella* [35–37], *Bombus* [38], *Ceratina* [39,40], *Diadasia* [41], *Halictus* and *Hylaeus* [30], *Lasioglossum* [42,43], *Megachile* [31,44], *Melissodes* and *Svastra* [45,46], *Nomia* [47], and *Xylocopa* [48].

2.4. Data Analyses

Analyses were completed in R version 3.6.0 "Planting of a Tree" using VEGAN and ggplot2 [49]. Abundances were pooled by genus (all species within a genus combined) to account for variation in native ranges of many species across the data set for these analyses. All data was organized in a matrix containing total abundances of each genus by yearly collection location. Data ordination to determine variation between pollinators visiting cotton in the two states was conducted using non-metric multidimensional scaling (nMDS) of Bray–Curtis similarities using VEGAN and graphed in GGPLOT2 [50,51]. A one-way non-parametric analysis of similarities (ANOSIM) test of the Bray–Curtis similarity data obtained from 999 permutations was also performed using VEGAN to compare the similarity of the communities [50].

3. Results

3.1. Species Richness

The 1200 individual specimens collected from Mississippi [MS] were collected over two years. These specimens represent at least 33 species (as *Lasioglossum (Dialictus)* were not identified past subgenus), which includes 21 genera in four families (Table 1). A total of 5246 individuals were collected in Texas (TX) cotton fields over two years. These specimens represent 41 (23 species are included in Table 1, with an additional 18 morphospecies of *Lasioglossum (Dialictus)*) species in three families. Apidae was the most abundant family in both locations (1028 individuals in MS and 4145 individuals in TX), including 10 genera in each state with 16 species in MS and 11 species in TX, followed by Halictidae (Figures 2 and 3). *Anthophora* and *Diadasia* were not collected in Mississippi, while *Xenoglossa* and *Ptilothrix* were not collected in TX. Members of the genus *Melissodes* were the most abundant and dominant taxa in both locations (Table 1). In particular, *Melissodes tepaneca* (Cresson) was the most abundant species found in both states.

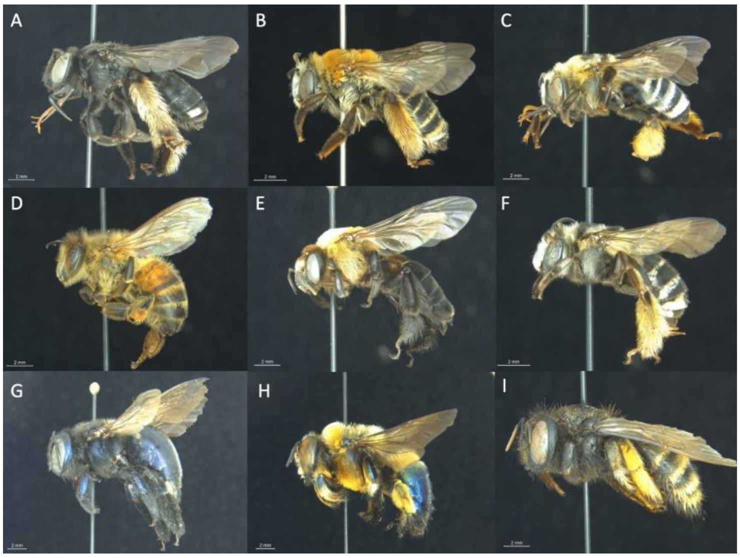

Figure 2. Some of the members of the family Apidae observed in cotton fields: (**A**) *Melissodes (Melissodes) bimaculatus;* (**B**) *Melissodes (Melissodes) tepaneca;* (**C**) *Melissodes (Melissodes) communis;* (**D**) *Apis mellifera;* (**E**) *Ptilothrix bombiformis;* (**F**) *Florilegus condignus;* (**G**) *Xylocopa (Schonnherria) micans (female);* (**H**) *Xylocopa (Schonnherria) micans (male);* (**I**) *Xylocopa (Notoxylocopa) tabaniformis parkinsonae.*

Table 1. Native bees (Hymenoptera: Anthophila) collected in cotton fields in several locations in both Mississippi and Texas. Sampling efforts and times varied among years and locations.

Species of Bees Collected by Family	Abundance in MS	% of Pop in MS	Abundance in TX	% of Pop in TX	Oligolectic
COLLETIDAE					
Hylaeinae					
Hylaeus (Prosopis) nelumbonis (Robertson)	1	<1	-	0	No [30]
HALICTIDAE					
Augochlorini					
Augochlora aurifera Cockerell	-	0	1	<1	No [30]
Augochlora pura pura (Say)	19	1.6	-	0	No [30]
Augochlorella aurata (Smith)	11	<1	9	<1	No [30]
Augochloropsis metallica (F.)	16	1.3	-	0	No [30]
Halictini					
Agapostemon melliventris Cresson	-	0	36	<1	No [52]
Agapostemon sericeus (Forster)	4	<1	-	0	No [30]
Agapostemon splendens (Lepeletier)	-	0	17	<1	No [30]
Agapostemon texanus Cresson	2	<1	39	<1	No [30]
Agapostemon virescens (F.)	26	2.2	-	0	No [30]
Halictus (Nealictus) parallelus (Say)	8	<1	-	0	No [30]
Halictus (Odontalictus) ligatus Say	23	1.9	7	<1	No [30]
Lasioglossum (Dialictus) nr. coactum (Cresson)	-	0	18	<1	Unknown
Lasioglossum (Dialictus) connexum (Cresson)	-	0	24	<1	Unknown
Lasioglossum (Dialictus) disparile (Cresson)	-	0	35	<1	No [43]
Lasioglossum (Dialictus) hartii (Robertson)	3	<1	-	0	No [43]
Lasioglossum (Dialictus) spp.*	49	4.1	883	16.8	-
Lasioglossum (Evyleaus) nelumbonis (Robertson)	2	<1	-	0	Nymphaeaceae [42]
Nomiini					
Nomia (Acunomia) nortoni Cresson	1	<1	6	<1	No [30]

Table 1. *Cont.*

Species of Bees Collected by Family	Abundance in MS	% of Pop in MS	Abundance in TX	% of Pop in TX	Oligolectic
MEGACHILIDAE					
Megachilini					
Coelioxys (Boreocoelioxys) sayi Robertson	1	<1	-	0	No [31]
Megachile (Leptorachis) petulans Cresson	4	<1	-	0	No [31]
Megachile (Litomegachile) brevis Say	1	<1	26	<1	No [31]
Megachile (Litomegachile) lippiae Cockerell	-	0	27	<1	No [44]
Megachile (Litomegachile) gentilis Cresson	-	0	26	<1	No [53]
Megachile (Litomegachile) mendica Cresson	1	<1	-	0	No [31]
Megachile (Sayapis) policaris Say	-	0	132	2.5	No [31]
APIDAE					
Anthophorini					
Anthophora californica Cresson	-	0	1	<1	No [54]
Apini					
Apis mellifera L.	11	<1	47	<1	No [31]
Bombini					
Bombus pensylvanicus (DeGeer)	1	<1	-	0	No [31]
Ceratini					
Ceratina (Zadontomerus) dupla Say	1	<1	-	0	No [31]
Ceratina (Zadontomerus) sp.	-	0	21	<1	-
Emphorini					
Diadasia rinconis Cockerell	-	0	14	<1	*Opuntia* spp. [55]
Melitoma taurea (Say)	2	<1	-	0	*Ipomoea* spp. *Calystigia* spp. [56]
Melitoma sp.	-	0	1	<1	-
Ptilothrix bombiformis (Cresson)	33	2.8	-	0	*Hibiscus* spp. [56,57]

Table 1. *Cont.*

Species of Bees Collected by Family	Abundance in MS	% of Pop in MS	Abundance in TX	% of Pop in TX	Oligolectic
Eucerini					
Florilegus condignus (Cresson)	8	<1	12	<1	*Pondenteria* spp. [56]
Melissodes (Eumelissodes) boltoniae Robertson	1	<1	-	0	Asteraceae [56]
Melissodes (Eumelissodes) trinodis Robertson	2	<1	-	0	Asteraceae [56]
Melissodes (Melissodes) bimaculatus (Lepeletier)	129	10.8	-	0	No [31]
Melissodes (Melissodes) communis Cresson	2	<1	19	<1	No [31]
Melissodes (Melissodes) comptoides Robertson	24	2	-	0	No [31]
Melissodes (Melissodes) tepaneca Cresson	803	66.9	3987	76	No [31]
Svastra (Epimelissodes) obliqua (Say)	4	<1	27	<1	Asteraceae [56]
Svastra (Epimelissodes) petulca (Cresson)	-	0	14	<1	Asteraceae [56]
Xenoglossa strenua (Cresson)	5	<1	-	0	*Cucurbita* spp. [56,58]
Xylocopini					
Xylocopa (Notoxylocopa) tabaniformis Smith	-	0	2	<1	No [47]
Xylocopa (Schonnherria) micans Lepeletier	1	<1	-	0	No [31]
Xylocopa (Xylocopoides) virginica (L.)	1	<1	-	0	No [31]

* *Lasioglossum (Dialictus)* spp. includes multiple species at both locations (18 spp. in Texas and an unknown number in Mississippi) that often cannot be reliably identified using available keys.

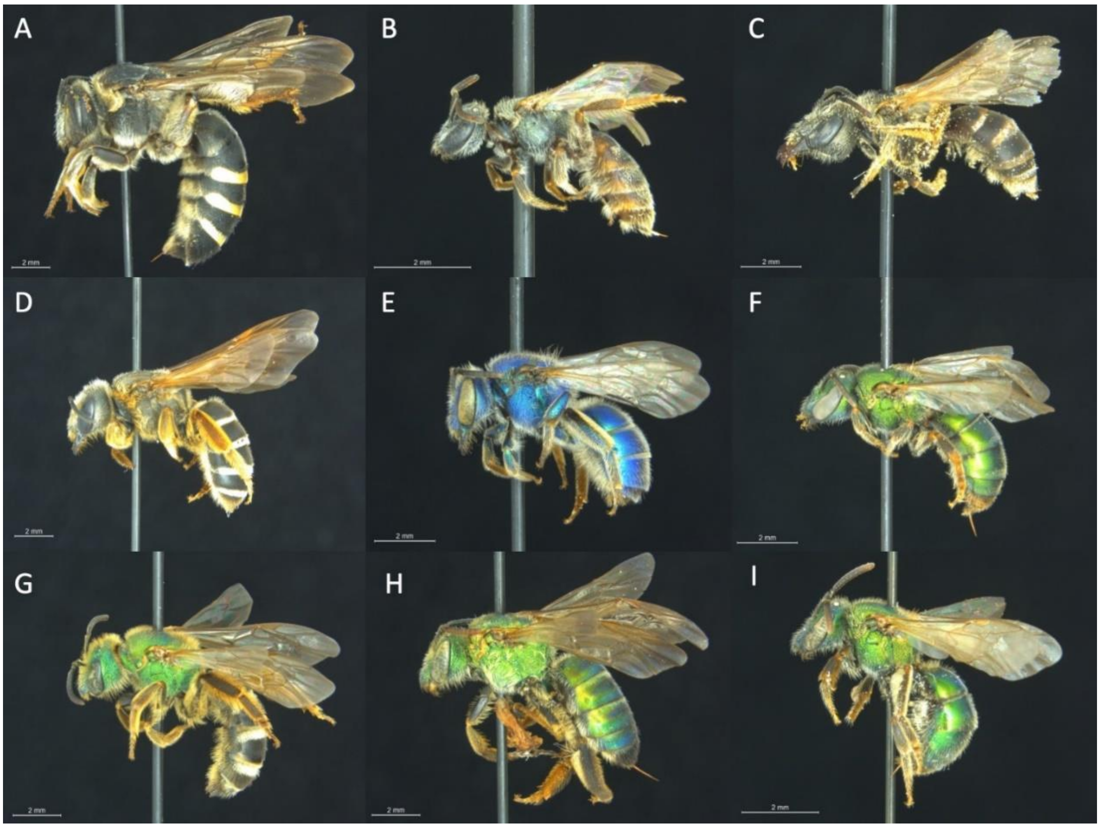

Figure 3. Some of the members of the family Halictidae observed in cotton fields: (**A**) *Nomia (Acunomia) nortoni*; (**B**) *Lasioglossum (Dialictus)* spp.; (**C**) *Halictus (Odontalictus) ligatus*; (**D**) *Halictus (Nealictus) parallelus*; (**E**) *Augochloropsis metallica*; (**F**) *Augochlorella aurata*; (**G**) *Agapostemon virescens*; (**H**) *Agapostemon splendens*; (**I**) *Augochlora pura pura*.

3.2. Similarity of Fauna between Locations

The nMDS analyses indicated two group of samples when species were pooled to the genus level. Samples from Mississippi and Texas were separated and formed distinct groups on the plot (Figure 4). Each point on the plot represents a yearly collection location, and each color represents a state. The reasonably low stress level (0.012) indicates a good representation of multidimensional space. However, the ANOSIM test showed only weak differences (r = 0.1452) and was overall non-significant (0.144) indicating that while populations separate in the nMDS plot, there is no statistically significance between the communities found in cotton fields in Mississippi and Texas. This lack of significance indicates that the space in Figure 4 between the locations from the two states is not as great as the distance within a given state.

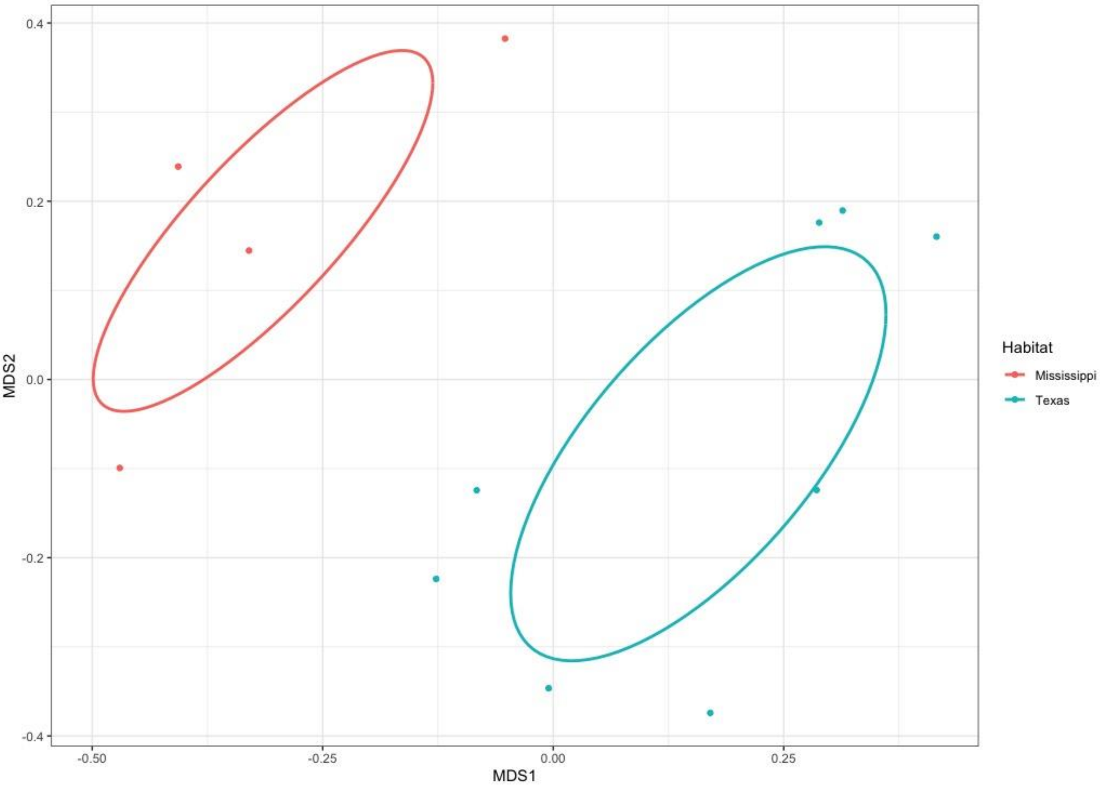

Figure 4. Non-metric multidimensional scaling (nMDS) of Bray–Curtis similarity data performed on total abundance by genera. Individual points are a representation of a collection location in a given year.

4. Discussion

Global declines of pollinators and/or other insects have been reported in recent decades, but a lack of both historical and current documentation about pollinator abundance and community structure limits the assumptions that can be made in many geographic areas and specialized habitats [59,60]. Mass flowering crops, like cotton and soybean, can support generalist pollinator populations by providing floral and other resources [1], but these commonly grown southern row crops also pose risks to bees (including honey bees), notably insecticide exposure [61]. Baseline data on community structure is important for monitoring landscape level changes and risk to populations that frequent these areas [62]. Native bee communities that share those habitats likely are exposed to similar benefits and risks, making information about the biodiversity and community structure in these areas essential.

Similar to older studies from cotton producing regions of Arizona, native bees in the genus *Melissodes* dominate the cotton community in both states during this study [10,63–65]. Visual observations of floral visitation were made in cotton grown in the state of Georgia, and of those observations 83.9% were bees in the genus *Melissodes*, and most of those were of a distinctive species, *Melissodes bimaculatus* (Lepeltier) [66,67]. Studies from the high plains of Texas suggested that *Agapostemon* was the most abundant genus, but *Melissodes thelypodii* Cockerell was also abundant in the fields [13,14]. No species of *Agapostemon* was dominant in our study, and the most frequently encountered species in MS only made up 2.2% of the total specimens collected. While this study utilized bee bowls for sampling, additional recently published data demonstrates that *M. tepaneca* and *A. mellifera* co-dominate collections made by hand netting in cotton fields in southern TX [68], suggesting that *Melissodes* dominance is not limited by observational or collecting methodologies. These observations do also suggest that using bee bowls in cotton undersampled *A. mellifera* in these fields. This dominance within the community by *Melissodes* is not limited to the United States, as *Melissodes nigroaenea* (Smith) is an important pollinator in Brazilian cotton fields [69].

Several of the species collected in both states during this study are known to be oligolectic (floral specialists) or kleptoparasites that are likely tourist species attracted to the bee bowls. Bees in the genus *Xenoglossa* are known specialists on cucurbits [58], which grow on field edges in many parts of the south. *Ptilothrix bombiformis* (Cresson) is a malvaceous specialist on native wild *Hibiscus* sp. [57], a genus closely related to cotton, and these bees were frequently seen in cotton fields in Mississippi. *Diadasia rinconis* are oligolectic on *Opuntia* spp. cacti [55]. While these species, and others listed in Table 1, are not known pollinators of cotton, they were exposed to similar potential risks by regularly traversing fields and their regular presence should be investigated further in the future.

Even though a variety of agricultural practices in crops, including pesticide applications and tilling can negatively impact pollinator populations, a community of native bees persists across and within these landscapes [70–74]. Many of the same cosmopolitan generalist groups that are found here are also found in other agricultural crops in North America including corn and soybeans, suggesting that these species may be adapted to living with the risks in agricultural landscapes [20,75–77]. Similar to the results presented here, abundant pollinators in crop fields often consist of a few common genera of native bees, while species that are threatened or more rarely observed are infrequently or never observed in crop fields. This suggests that potential management programs focused on pollinator conservation in these regions should differ for common generalist agricultural pollinators and those more rarely encountered or oligolectic species that are not frequently observed in fields [78].

Native pollinators offer potential benefits to producers, in spite of risks they face in agricultural fields. While cotton is known to self-pollinate, there are also many benefits to increasing cotton pollination by both native and honey bees [79]. Floral visitation by pollinators in cotton can increase boll set, seed weight, and lint weight [79–83]. Intensive visitation by honey bees in particular increased yield in some cotton experiments by up to 15.8% [84]. For example, of 26 native bee species collected in cotton fields in Burkina Faso, not all species provided similar pollination services and six species did not cause fruit set and were excluded as pollinators. Visitation by both honeybees and *Tetralonia fraterna* Friese (Hymenoptera: Eucerini) significantly increased both seed weight and fiber weight, suggesting similar patterns could exist in other cotton growing regions of the world [85]. Future studies in both states involving native bees in cotton should focus on potential benefits of these native bees, including abundant species in the genera *Melissodes*, that are visiting and pollinating cotton in the United States. As the communities of native bees are not significantly different between Mississippi and Texas, this baseline data taken with modern agricultural practices can be utilized for future studies and used to refine management recommendations in cotton production systems across the southern United States.

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