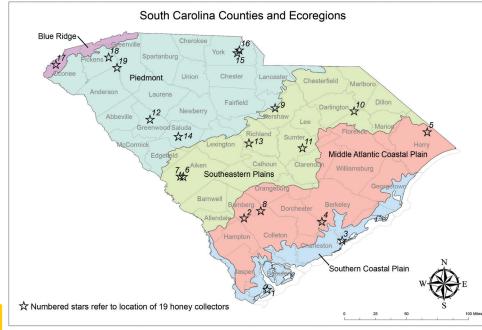
South Carolina Statewide Honey Pollen Project

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(varietal honey). Prior to the current study, most of the existing information on preferred nectar sources was anecdotal and based on direct observation by beekeepers of which plants bees were visiting. This did not permit beekeepers to differentiate between plant use by bees for nectar production vs. pollen collection. In addition to information for the beekeeper, the current study also acts as a baseline for the regional nectar ecosystems in the state, which will permit the scientific community and other interested parties to determine the potential impact of factors like urban sprawl and climate change on future honey production.

Funded by the SC Depart-

Figure 1. Locations of the 19 participating apiaries in this study. SC's Type III ecoregions are denoted by color.

In early 2021, the South Carolina Beekeepers Association (SCBA) began thinking about conducting a South Carolina (SC) statewide honey pollen analysis. Interest in such a project was sparked by the Congaree River Basin Honey Pollen Analysis (melissopalynology) project that was completed in 2021. For the Congaree project, we collected fresh honey weekly during nectar flow periods in 2020 from a single apiary within the Congaree Biosphere Reserve and analyzed the pollen in the honey to determine what nectar sources were contributing to the honey. This project was the first to reveal that plants are blooming (and being used by bees) at times that didn't match existing bloom-time reports for SC (MacFawn and O'Keefe, *in review*).

Given the success of the Congaree study, SCBA decided to attempt to replicate the analysis across the entire state of SC. This would make SC the first state to carry out honey pollen analysis on a statewide basis to determine what plants bees are using as nectar sources and, thus, contributing to our honey. Importantly, this analysis can provide beekeepers with advice on what plants bees are using across the state, when the bees are using those plants, and whether the plants used vary across the state. With this information, beekeepers can make more informed decisions about when to install honey supers (boxes) for floral and multifloral honey, when to conduct hive and super management, and when to move hives to a specific region to capture specific nectar flow if they intend to produce a specific type of honey ment of Agriculture in the Fall of 2021, the statewide project had three objectives:

- Determine *which* plants are nectar sources for honeybees by region across the state of SC via analysis of pollen from collected honey.
- Determine *when* these nectar plants bloom across the state by regular, routine collection of honey samples for one calendar year.
- Determine *whether* the maple (*Acer*) bloom (end of January/early February) is both a pollen source and a honey/nectar source.

Meeting the aims of such a large project was a team effort. Two members of the team are experienced beekeepers who orchestrated the recruitment and training of beekeeper volunteers. These team members also organized and arranged honey sample collection and shipping of those samples for processing. The team also consisted of three melissopalynologists - researchers who specialize in identifying and quantifying pollen in honey. In addition to the research team, 19 experienced beekeepers from apiaries distributed across each of the five Level III ecoregions in SC volunteered to collect weekly honey samples during the nectar flow and to send their samples for processing (Figure 1). Fifteen of the participating beekeepers were in rural portions of the state and four were in urban areas. Also, the apiaries were spread through 17 of SC's 46 counties. Honey samples were collected throughout 2022 and analyzed during 2023. The results of this analysis have been compiled into a book which is available in open-access digital form and paper form through Clemson University Press. We focused the analysis on the ecoregions because it's the local and regional ecology that impacts the

bees. Geology, soils, and water all interact to impact the plants that can grow in a habitat and, therefore, impact the resources available to the bees.

For the project, honey samples were collected weekly



during the 2022 nectar flows, but only when fresh nectar was observed, and collection continued until the first frost of Fall. For each honey sample, a slice of the comb was removed from the uppermost frame of the top honey super and placed in a container for shipping. All samples were labeled

Figure 2. Honey sample showing the labeling used to identify all honey samples. Image courtesy of Karen Hilborn.

with the date, location, and the beekeeper's name (Figure 2) and sent to Global GeoLab in Canada for processing. During processing, the pollen was separated from the honey, acetolyzed using the methods of the late Dr. Vaughn M. Bryant and epoxied onto microscope slides. Pollen slides were then sent to the laboratories of the three melissopalynologists for counting and plant source identification. In most cases, pollen grains were identified to the level of genus, but when that was not possible, they were identified to the level of family.

Each honey sample collected provided two important pieces of information: the identity of the plants from which the bees foraged for nectar and the relative abundance of pollen from each plant present. This information (our results) was analyzed to generate two different types of charts. (1) **Apiary Pollen Occurrence Charts** provide detailed information about the plants being used by bees at a single apiary over the course of a year. (2) *Ecoregion* **Pollen Occurrence Charts** provide lists of the plants being used by bees throughout an ecoregion over the course of a year. These two ways of expressing the data also allowed us to look for trends and make comparisons both within and between the ecoregions.

Apiary Pollen Occurrence Charts were constructed from the honey samples collected for each of the 19 apiaries. These charts provide information to the beekeeper about which plant taxa's pollen were present in the honey, when each plant taxon's pollen was present in the honey, and the *relative abundance* of pollen for each plant taxon that was present in the honey. Figure 3 is a portion of the analysis for one collector as an example. For each apiary/collector, we determined all plant taxa whose pollen was present in the honey sample (left-most column) and the percentage (relative abundance) of the taxa present within each honey sample (the columns). To be very clear, these charts are not "bloom charts" as described in other publications. We do not know when the plants were blooming - we only know when the pollen was observed in the honey. From this example, you can see that honey samples were not collected every week (blank columns); remember that honey samples were only collected during nectar flows. The plant taxon list includes all taxa ever observed for this particular apiary/ collector - that's why there are many values of zero. The color indicates the proportion of the taxa in a given honey sample based on pollen categories. Samples where the taxa are highlighted in red indicate that this was the "Predominant" taxon present in the honey (present at >45% of all plant taxa) and, therefore, that honey can be classified as a "monofloral varietal" honey. Monofloral varietal honeys have special economic value because they can be sold for a premium price.

Using the Apiary Pollen Occurrence Charts, we could look for trends within the ecoregions. For example, it was an interesting observation that all ecoregions produced

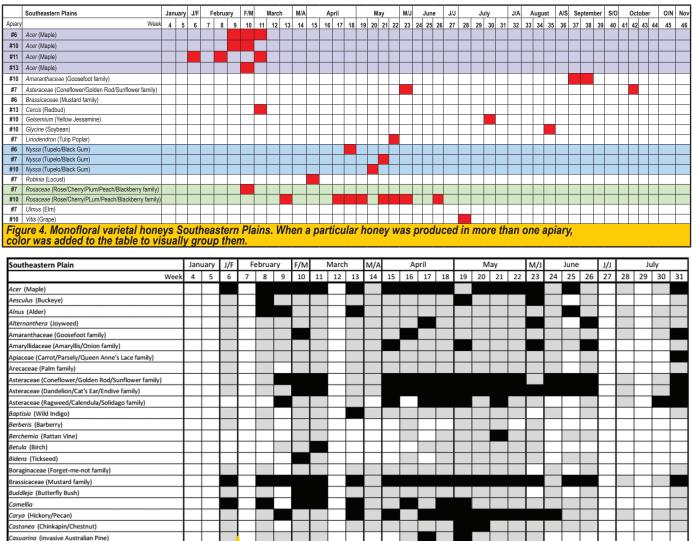
Collector 12	January			J/F		February			F/M	March						M/A	4	April							
Week	4		5	6		7		8	9	10	11		12		13		14		15		16		17	1	8
Type of Data ->				%	PC	%	PC				%	PC	%	PC	%	PC	%	PC	%	PC	%	PC		%	PC
Acer (Maple)				77.1	D	100.0	D				95.9	D	10.2	м	11.5	м	17.6	Т	1.8	L	0.0			0.0	
Alnus (Alder)				0.0		0.0					0.0		0.0		0.0		0.9	L	0.0		0.0			0.0	
Amaranthaceae (Goosefoot family)				2.6	L	0.0					0.0		27.6	1	62.0	D	1.8	L	0.0		0.0			0.0	
Amaryllidaceae (Amaryllis/Onion family)				0.0		0.0					0.0		0.0		0.0		0.0		0.0		12.9	м		0.0	
Asteraceae (Coneflower/Golden Rod/Sunflower family)				2.9	L	0.0					1.9	L	33.2	1	0.0		0.0		0.0		0.0			0.0	
Asteraceae (Dandelion/Cat's Ear/Endive family)				0.0		0.0					0.0	Π	0.0		0.0		0.0		0.0		1.0	L		0.0	
Asteraceae (Ragweed/Calendula/Solidago family)				0.0		0.0					0.0		0.0		5.8	м	2.3	L	2.7	L	0.5	L		0.0	
Brassicaceae (Mustard family)				1.3	L	0.0					0.0		1.0	L	0.0		0.0		2.7	L	0.5	L		0.0	
Buddleja (Butterfly Bush)				0.0		0.0					0.0		0.0		0.0		0.0		0.0		0.0	\square		0.0	
Camellia				1.3	L	0.0					0.0		0.0		0.0		0.0		0.0		0.0			0.0	
Carya (Hickory/Pecan)				0.0		0.0					0.0		0.0		0.0		0.0		0.0		0.0			0.0	
Caryophyllaceae (Carnation family)				0.0		0.0					0.0		0.0		0.0		0.0		0.0		0.0			0.0	
Cornus (Dogwood)				0.0		0.0					0.0		0.0		0.0		0.9	L	0.0		0.0			16.2	2 1
Corylus/Carpinus (Hazel/Hornbeam)				0.0		0.0					0.0		0.0		0.0		0.0		0.9	L	0.0	\square		0.0	
Dalea (Prairie Clover)				0.0		0.0					0.0		0.0		0.0		0.0		0.0		0.0			0.0	
Diospyros (Persimmon)				0.0		0.0					0.0		0.0		0.0		0.0		0.0		0.0			0.0	
Elaeagnus (Autumn/Russian Olive)				0.0		0.0					0.0		1.3	L	3.8	м	0.0		0.0		0.0			0.0	
llex (Holly)				0.0		0.0					0.0		0.0		0.0		1.8	L	2.7	L	0.0			0.0	
Juglans (Walnut)				0.0		0.0					0.0		0.0		0.0		0.0		0.0		0.0			0.0	
Lagerstroemia (Crepe Myrtle)				1.3	L	0.0					0.0		0.0		2.4	L	1.4	L	0.0		0.0			0.0	
Lamiaceae (Mint family)				0.0		0.0					0.0		0.0		0.0		37.1	Т	34.8	Т	3.0	м		0.0	
Ligustrum (Privet)				0.0		0.0					0.0		0.0		1.0	L	3.6	М	0.0		78.2	D		0.0	
Liquidambar (Sweetgum)				0.0		0.0					0.0		0.0		0.0		0.0		0.9	L	0.0			1.3	L
Liriodendron (Tulip Poplar)				0.0		0.0					0.0		0.0		0.0		0.0		0.0		0.5			0.0	

Figure 3. A portion of the Apiary Pollen Occurrence Chart for apiar	y/
collector #12. This apiary is located in the Piedmont ecoregion.	-

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D (predominant) >45% I (secondary) 16-45% M (important) 3-15% L (minor) <3% monofloral varietal honeys. These honeys were produced throughout the year and from different plants. Figure 4 shows the monofloral varietal honeys produced in the Southeastern Plains. In this figure, color has been added to highlight plant taxa present in more than one apiary. For example, four of the six apiaries in this ecoregion produced monofloral varietal maple (Acer) honey and, of those four apiaries, three produced this honey for more than one week. Similarly, three of the si apiaries produced monofloral Tupelo/Black Gum (Nyssa) honey. In all, 13 plant taxa produced monofloral varietal honeys in the Southeastern Plains. This is useful information to guide the timing and hive placement for a beekeeper in the Southeastern Plains who might be interested in cultivating monofloral varietal honeys. It should be noted that this project was conducted for only a single year (2022), and it is very likely that some amount of year-to-year variation occurs at any location.

Using the same data, **Ecoregion Pollen Occurrence Charts** were constructed for each of the five EPA Type III ecoregions. Figure 5 is a portion of the analysis for one ecoregion – the Southeastern Plains – as an example. This type of analysis gives a broader picture of what is happening in an ecoregion. Since it is less localized, it might be more relevant to the average beekeeper. For this type of analysis, the results from all beekeepers in the ecoregion were combined. Because of this, the information available to the beekeeper is different. Notice that the relative abundance values and pollen categories present in the Apiary Pollen Occurrence Charts (Figure 3 is an example) are replaced with only color-coding to indicate the presence or absence of the plant taxa. The color/information for each collection week (column of data) is the combined data for all apiaries from which honey samples were collected during that week, and might represent data from a single apiary, a few apiaries, or all apiaries in that ecoregion. Thus, information for the beekeeper is limited to presence or absence of the plant taxa in the honey samples. The exception to this is in the Blue Ridge because there was only one participating apiary in this ecoregion. Like the Apiary Pollen Occurrence Charts, Ecoregion Pollen Occurrence Charts provide information about *which* plant taxa bees are preferentially foraging nectar from and *when* they are using these plant taxa throughout the year.



Key:				
no data collected				
taxon absent on this week				
taxon present on this week				

Figure 5. A portion of the Ecoregion Pollen Occurrence Chart for the Southeastern Plains.

Using the Ecoregion Pollen Occurrence Charts, we could look for trends among the ecoregions. Across SC, we observed 141 plant taxa (Figure 6). The majority of these plant taxa (53 or 37.6%) were found in only a single ecoregion, while 26 plant taxa (18.4%) were present in all 5 ecoregions. Plant taxa unique to a single ecoregion included pea (Pisum sativum) in the Piedmont, the primrose family (Primulaceae) in the Southeastern Plains, elderberry (Sambucus) in the Middle Atlantic Coastal Plain, and the Ranunculaceae (buttercup family) in the Southern Coastal Plain. No plant taxa unique to the Blue Ridge were observed, but this could be due to collecting honey from only a single apiary in that ecoregion. Plant taxa present in all five ecoregions included maple (Acer), crepe myrtle (Lagerstroemia), tupelo/black gum (Nyssa) and red/white clover (Trifolium/Melilotus).

Plant Taxa Distribution	Number of Taxa	Taxa Percengate				
found in only 1 ecoregion	53	37.6%				
found in 2 ecoregions	24	17.0%				
found in 3 ecoregions	25	17.7%				
found in 4 ecoregions	13	9.2%				
found in all 5 ecoregions	26	18.4%				
	141	100.0%				
Figure 6. Plant taxa present in 5 ecoregions.						

Of the five ecoregions, the ecoregion with the most plant taxa was the Southeastern Plains – 72% (102 of 141 total) of all the plant taxa observed in the honey samples were found in this ecoregion (Figure 7). The Southeastern Plains include the Sand Hills region and it is a mosaic of plains between shallow stream valleys. Rainfall is generally high in this region, and soils range from well-drained, especially the Sand Hills region, to boggy with sandy rims close to the ecoregion's southern boundary. Thus, with this region's varied habitats, it is probably not surprising that so many plant taxa are used by the bees in this ecoregion.

Ecoregion	Total Taxa Present							
Blue Ridge	34							
Piedmont	93							
Southeastern Plains	102							
Middle Atlantic Coastal Plain	54							
Southern Coastal Plain	75							
Figure 7 Number of plant taxa observed in honey								

samples for each ecoregion.





Privet (Ligustrum)

Tulip Poplar (Liriodendron)

Two of the project objectives were to determine *which* plants the honeybees are using as nectar sources and *when* those plants are being used. We now know that honeybees in SC collect nectar from a wider variety of trees, shrubs, and herbaceous plants than previously reported. Pollen from a total of 141 distinct plant taxa were observed in the honey samples collected. Some plants were observed regionally, but others were important statewide such as privet (*Ligustrum*), tulip poplar (*Liriodendron*), holly (*Ilex*), and members of the rose family (*Rosaceae*) like blackberry (Figure 8). The plants just listed are used from very early Spring through the Summer.

While we observed many plants that had previously been reported as nectar sources in SC, such as privet (*Ligustrum*), tulip poplar (*Liriodendron*), holly (*Ilex*), and members of the rose family (*Rosaceae*) like blackberry, we also observed pollen from plants that had not been previously reported such as dogwood (*Cornus*) and sweetgum (*Liquidambar*) (Figure 9).

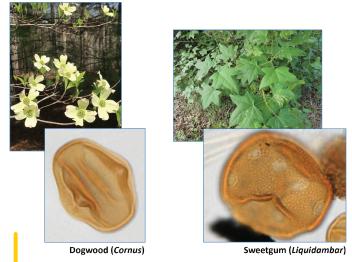


Figure 9. Examples of plants not previously reported as nectar sources for bees in SC. Plant photos obtained from iNaturalist.

Wind-blown pollen from non-nectar-producing trees such as crepe myrtle (*Lagerstroemia*), pine (*Pinus*), oak (*Quercus*), and elm (*Ulmus*) was discovered in some honey samples (Figure 10). It's possible that bees picked up these pollen types secondarily as they visited other flowers during nectar collection, or it could be the result of accidental incorporation of pollen cells into the honey sample.

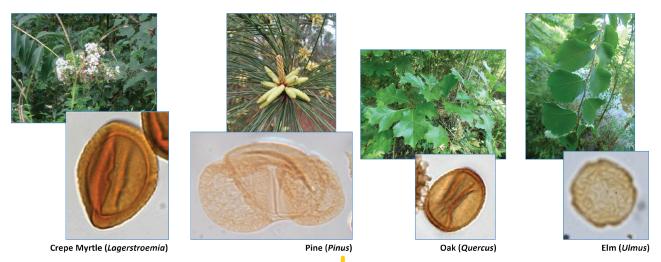
Figure 8. Examples of plants used by bees statewide. Plant photos obtained from iNaturalist.





Holly (*Ilex*)

Blackberry (Rosaceae)



Maple (*Acer*) pollen, already known to be a protein source for bees in late Winter/early Spring, also appeared in the honey. Maple (*Acer*) pollen was found in honey in all five ecoregions. In all ecoregions other than the Southern Coastal Plains, the relative abundance of maple (*Acer*) pollen was high enough to categorize the honey are monofloral. Examples of the timing for the monofloral maple (*Acer*) honey can be seen in Figures 3 and 4. This finding is evidence that maple (*Acer*) is a very important nectar source.

The results of this research provide the beekeeper with important information about *which* plants the honey bees are using as nectar sources and *when* those plants are being used by bees; this knowledge can be used to increase overall honey production. These results can also be used to facilitate monofloral varietal honey production. During 2022, monofloral varietal honeys were produced in all five ecoregions. Some monofloral varietal honeys were observed in multiple ecoregions, like maple (*Acer*); while others were unique to a single ecoregion, like *Magnolia* in the Middle Atlantic Coastal Plain.

We are excited to describe the first year-long statewide analysis of the pollen found in honey for SC. Nineteen beekeepers provided 302 honey samples during the nectar flow period of 2022 to make this project possible. Our analysis, in the form of Apiary Pollen Occurrence Charts and Ecosystem Pollen Occurrence Charts, provides the interested beekeeper with helpful information about what plants honey bees prefer to forage as nectar sources and when honey bees are making use of these plants. It is our hope that, armed with this information, beekeepers will be able to increase their honey production and increase the value of their honey by producing varietal honeys, and that anyone interested in bees will improve their local ecosystems by cultivating more bee-friendly plant varieties.BC

References

MacFawn, DE. and O'Keefe, JMK., *in review.* 2024 *Melissopalynology of honey produced in the Congaree Biosphere Reserve biodiversity hotspot*, South Carolina, USA.

Acknowledgments

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Figure 10. Examples of plants with wind-blown pollen whose pollen was observed in the honey samples in SC. Plant photos obtained from iNaturalist.

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