

UCDAVIS



JULY 17-20, 2019

*Multidimensional Solutions to Current  
and Future Threats to Pollinator Health*

ABSTRACTS



# Thurs, July 18 - Session 1

## *Novel Quantitative Methods in Pollinator Ecology & Management*

### **Molecular methods for pollination ecology**

**Juan Lobaton<sup>1</sup>, Linsey Kirkland<sup>2</sup>, Sarina Macfadyen<sup>3</sup>, Rose Andrews<sup>4</sup>, Romina Rader<sup>5</sup>**

Pollination studies traditionally use ecological approaches to estimate pollinator efficiency and foraging behavior. Yet, with the advent of new genetic technologies in the past decade biologists and ecologists can now ask novel questions using molecular tools within the fields of genomics. We examined the differential gene expression during different pollination events in an apple *Mallus domestica* cv. Pink Lady orchard located in Stanthorpe, Queensland Australia. We conducted a series of cross-pollination, self-pollination and honey bee single and multiple visits events to determine the genes involved in apple pollen-stigma recognition. Through a series of hand pollination treatment we identify differentially expressed genes involved in the pollen tube development, pollen recognition by the stigma and activation of the auto incompatibility system. Furthermore we conducted the first transcriptome analysis of an apple natural pollination events at orchard conditions using single and multiple visited honey bee stigmas. In addition, we use pollen DNA sequencing data to developed PCR markers to identify pollen varieties on honey bee pollen loads to track variety pollen flow at orchard conditions.

### **Implementing a honeybee foraging model and REDAPOL fruit set predictions in Washington State's Decision Aid System**

**Vincent P. Jones<sup>1</sup>, Gloria DeGrandi-Hoffman<sup>2</sup>, Stefano Borghi<sup>1</sup>, Ute Chambers<sup>1</sup>, Peter W. Shearer<sup>1,2</sup>, Henry Graham<sup>1</sup>, Matt S. Jones<sup>1</sup>**

We have implemented a version of the honeybee foraging model sub-program of the REDAPOL model in the WSU Decision Aid System (WSU-DAS) (<https://decisionaid.systems>). This model provides users with the foraging activity and conditions over the past 3 days using near-real time environmental measurements (temperature, solar radiation, windspeed and rainfall) and over the next three days using gridded weather forecasts. We are also in the process of final validation of the REDAPOL model that predicts fruit set based on bloom density and overlap, and colony size and number. Our data so far suggests that 95% of fruit set occurs in the first ~75% of the blooms that open, regardless of the cultivar. The incorporation of the fruit set predictions should significantly improve the growers' understanding of the system and help guide them during the bloom period. These models will also be useful in evaluating the effects of climate change on pollination.

### **Using DNA metabarcoding techniques to improve plant-pollinator interaction networks**

**Victoria A. Reynolds<sup>1,2</sup>, Anya J. Cutler<sup>1</sup>, Karen L. Bell<sup>1,2</sup>, Margaret M. Mayfield<sup>1</sup>, Berry J. Brosi<sup>1</sup>**

Plant-pollinator networks have typically been composed using insect-visitation data that may not accurately depict true pollination events. Recently, there has been an increase in the number of networks created via insect pollen-load samples. These networks are often determined through light microscopy identification, requiring specialised knowledge and reference collections; yet even with these, lower levels of identification can be hard to achieve. DNA metabarcoding has been used as an alternative to identifying species-specific pollen samples from honey, hives and insects. However, this novel approach has not yet been used to test ecologically driven questions, like how varying landscape factors may be altering resource collection patterns and floral fidelity in pollinating insects. We use DNA metabarcoding and the newly developed DADA2 bioinformatics pipeline to assess the impacts of a mass-flowering crop, *Brassica napus*, on the floral fidelity of pollinating insects in wildflower communities adjacent to these fields. We collected 120 pollinating insects from remnant patches of York Gum-Jam woodland wildflower communities in SW Western Australia that were varying distances from the edge of a blooming canola field. Using DNA metabarcoding we identified the individual species composition of insect pollen-loads and created pollination networks and pollen-load composition analyses to determine how increased proximity to canola crops alters pollen-load consistency. With this research, we highlight the practical applications of using this technique and the potential for its use in ensuring improved accuracy for plant-pollinator network analyses and pollen-load analyses that will give valuable insights towards answering central questions in ecology.

### **CITIZEN SCIENCE DATA FOR MAPPING BUMBLEBEE POPULATIONS**

**Claudio Gratton<sup>1</sup>, Benjamin Zuckerberg<sup>2</sup>**

Understanding large-scale distributions of bumble bee species has increased in recent years with the ability to harness the power of observations from legions of citizen scientists. Citizen science has proven useful for detecting regional population declines, identifying climatic constraints on species distributions, and identifying traits associated with vulnerability to environmental change. Nationwide projects such as Bumble Bee Watch (Xerces Society) or regional programs (Bumble Bee Brigade, --

-Wisconsin; Beespotter, Illinois) allow citizen scientists to catalogue bee observations at unprecedented scales. By participating in these projects, observers can record and identify bee species using both casual as well as targeted surveys. The advantage of these programs is that observations come from a diversity of locations that have proven difficult to sample by trained experts (e.g., private lands, urban areas, etc.). As part of these programs, experts verify observations collected by the general public. Our goal was to explore the usefulness of these data to model bee distributions across the Upper Midwest. We modeled bee distributions using information on land cover variability and presence-background modeling. Further, quantified rates of agreement between citizen scientists and experts to explore which species were prone to misclassification. Citizen science and species distribution modeling are powerful ecological research tools that has been widely applied in the conservation of bird and mammal populations, and has untapped potential for the conservation and modeling of bee populations.

### **From Theory to Practice: The Bumble-BEEHAVE Model and its Application to Enhance Pollinator Friendly Land Management**

**Matthias A. Becher<sup>1,2</sup>, Grace Twiston-Davies<sup>3</sup>, Juliet L. Osborne<sup>3</sup>, Tonya A. Lander<sup>4</sup>**

The decline in abundance and species-richness of bumblebees raises serious concerns as they are important pollinators for crops and wild flowers. To better understand how spatial and temporal availability of nectar and pollen affects colony and population dynamics of bumblebee species, we developed the agent-based model *Bumble-BEEHAVE*. The model simulates multiple UK bumblebee species in a spatially-explicit landscape. Bees, represented as individuals or as cohorts, decide about their activity, using a stimulus-threshold approach. Activities include egg laying, foraging for nectar and pollen, and brood care. Successful colonies will produce new queens and males. Stressors such as parasites, predators and pesticides, can, to some degree, be included. *Bumble-BEEHAVE* can be used to predict and identify the variables associated with bumblebee colony success. Beesteward, a modified version of the model, was then designed to be specifically used by farmers, land managers and land advisors as a management tool for the conservation of pollinators in agricultural landscapes. We will give an overview of the model features and provide examples how they can be used. The models are freely available to download from <http://beehave-model.net/>.

### **A Laboratory System to Study the Effects of Stressors on Honey Bee Health and Fecundity**

**Julia D. Fine<sup>1</sup>, Hagai Y. Shpigler<sup>1,2</sup>, Gene E. Robinson<sup>1,3</sup>**

As the sole producer of fertilized eggs in a colony, honey bee queen health and reproduction are essential to the longevity of a colony. Research has demonstrated negative effects of agrochemicals on colony reproduction, including decreased colony expansion, queen failure and replacement, and decreased queen egg laying. This suggests that agrochemicals can have negative effects on queens and developing brood as well as workers. However, it is much more difficult to track the performance of queens and brood in the field, and previously, there were no laboratory assays to quantify queen performance. This presentation will describe a new system developed to study the effects of stressors on honey bee queen egg laying and brood development under tightly controlled, laboratory conditions using custom designed Queen Monitoring Cages (QMC) and current efforts to develop a robotic system to rear the eggs produced in these cages. The results of the first experiment in this system examining the effects of imidacloprid administered in worker diet on queen egg laying will be reported. Future applications including the tracking of brood survival and health following maternal stress exposure in QMCs and the system's use in parallel with field level studies of colony health will be discussed.

### **Using automated tracking to link individual behavior to colony performance in bumblebees**

**James Crall<sup>1</sup>, Callin Switzer<sup>2</sup>, Andrew Mountcastle<sup>3</sup>, Nick Gravish<sup>4</sup>, Robert Oppenheimer<sup>5</sup>, August Easton-Calabria<sup>1</sup>, Ashlee N Ford Versypt<sup>6</sup>, Biswadip Dey<sup>7</sup>, Andrea Brown<sup>1</sup>, Mackay Eyster<sup>8</sup>, Claire Guerin<sup>9</sup>, Sarah Kocher<sup>10</sup>, Naomi Pierce<sup>1</sup>, Stacey Combes<sup>11</sup>, Benjamin L de Bivort<sup>1</sup>**

Many critical pollinators live in complex, tightly integrated societies. A key challenge in the ecology of these social insect pollinators is studying the behavior and interactions of many animals simultaneously, and understanding how these individual-level phenomena drive colony performance. While this has traditionally posed a significant methodological hurdle, recent advances in computer vision and automation are making it increasingly feasible to (1) study the behavior of many individual workers in parallel, and (2) link these individual behaviors with the dynamics, growth, and health of entire colonies. Here, we describe recently developed tools for tracking the behavior of individual workers in bumblebee (*Bombus impatiens*) colonies across extended time scales and in multiple colonies in parallel. Our approach reveals substantial, stable individual variation in worker behavior within bumblebee colonies that plays a critical role in colony resilience. Next, we show that exposure to a common neonicotinoid pesticide alters the behavior of workers within the nest, including disrupting nursing, social interactions, and thermoregulation performance. Finally, we preview state-of-the-art, emerging techniques - including deep learning approaches - for studying social insects, and highlight the potential of these technologies to open new lines of inquiry in pollinator behavior and ecology.



# Thurs, July 18: Session 2

## Drivers of Host-Pathogen Interactions

### DWV as the driver of bee decline

Robert Paxton<sup>1</sup>

Elevated losses of honey bee (*Apis mellifera*) colonies have been widely reported in temperate zones for over a decade. Though multiple causes have been suggested, the finger of blame is often pointed at the exotic ectoparasitic mite *Varroa destructor* and viruses that it transmits, especially *Deformed wing virus* (DWV). Here I demonstrate that DWV is causal in colony loss, and that the elevated virulence of DWV genotype B (DWV-B), a relatively new variant of DWV, may explain why beekeepers have more recently had greater difficulty in maintaining healthy colonies. DWV-B and several other viruses associated with honey bees can also be found in wild bee species, with the weight of evidence suggesting that virus sharing occurs at flowers. The higher prevalence of DWV in honey bees argues for this virus spilling over into wild bee species, yet the impact on wild bee populations of viral spill-over is currently an open question. Using results from our recent laboratory infection and selection experiments, I attempt to answer this question, and, in so doing, reveal aspects of host susceptibility that can inform on the honey.

### NOVEL TRANSMISSION ROUTES AND INTENSIFICATION AS DRIVERS OF DISEASE EMERGENCE AND VIRULENCE IN HONEYBEE VIRUSES

Mike Boots<sup>2</sup>, Lena Wilfert<sup>2</sup>, Lewis Bartlett<sup>2</sup>, Robyn Manley<sup>2</sup>, Carly Rozins<sup>2</sup>

The drivers of the emergence of infectious disease and the determinants of the evolution of virulence remain central questions in disease ecology and evolution. There is some discussion and limited theory on how novel transmission modes and in particular the acquisition of vector transmission may determine the risk of emergence and may select for higher virulence. Alongside this there is an increasing interest in understanding how management practice may also influence not only disease spread but also their evolution. Honey bee viruses in the context of the acquisition of varroa mites as novel route of transmission and intensive management highlight both of these issues. I will present empirical and theoretical work on the potential impact of the acquisition of the novel vector on disease spillover and virulence and present theory on the impact of intensification on disease prevalence. Our results suggest that the acquisition of the novel vector rather than intensification has the most impact on disease.

### Viral transmission in honey bees and native bees, supported by a global BQCV phylogeny

Elizabeth A. Murray<sup>1,2</sup>, John Burand<sup>3</sup>, Natalia Trikoz<sup>3</sup>, Julia Schnabel<sup>3</sup>, Heather Grab<sup>1</sup>, Bryan N. Danforth<sup>1</sup>

Virus transmission across pollinators is an emerging area of research, and little is known of virus patterns in solitary bees. Our research addresses virus crossover in bees foraging in the same localities. We tested for the presence of three different viruses in field-collected managed honey bees and native bees (*Andrena* spp.), and we incorporated bee community data to relate those metrics to the incidence of the viruses sampled. We postulated that extensive sampling would allow us to provide evolutionary context to the viral incidence patterns in a region, and we wanted to determine if we could ascertain the geographic source of a common pollinator virus and detect species crossover. Towards this objective, we sequenced BQCV – the most common virus in our study – from across five sites and analyzed the strains in a phylogenetic framework, along with hundreds of others obtained from GenBank. We believe that a global phylogeny is essential for deciphering the origins of viral strains, due to worldwide pathogen movement. Our results substantiate viral crossover between honey bees and native bees, revealing multispecies viral transmission.

### Drivers of pathogen distributions in feral and managed honey bees Panuwan Chantawannakul

Asia hosts at least nine honey bee species and the honey bees are important in maintaining regional biodiversity and food security. Since the 1980s, the European honey bee (*Apis mellifera*) has been introduced and has successfully replaced the Asian honey bee (*Apis cerana*) in traditional apiculture in Thailand and in much of South East Asia. However, *A. mellifera* is prone to many diseases and parasites that have been co-evolved with native honey bees. One of the previous classic examples is the *Varroa* mite (*Varroa destructor*) that originated from *A. cerana*, but which has spilled over to the European honey bee and is now spreading worldwide. The *Tropilaelaps* mite (*Tropilaelaps mercedesae*) originally parasitizes giant honeybees and recently has jumped to the *A. mellifera* in southeast and east Asia. Recent disease and parasite surveys clearly showed that the feral honey bees are also affected by viral and nosema diseases as well as the parasitic mites, however appear to be more resistant than *A. mellifera*. Biotic and abiotic drivers of geographical and species distributions of bee pathogens and parasites will be described. In addition, the population decline of *A. mellifera* has been previously reported in many parts of the USA and in European – countries, resistance mechanisms are being investigated in the Asian honey bees in different levels. The information may assist beekeepers to combat current diseases and pests in commercial beekeeping operation and to assess the risks of the future introduction and spread of invasive bee pests and parasites.

## ***Serratia marcescens*: a pathobiont of honey bees**

**Kasie Raymann<sup>1</sup>, Kerri L Coon<sup>1</sup>, Zack Shaffer<sup>1</sup>, Stephen Salisbury<sup>1</sup>, Nancy A Moran<sup>1</sup>**

Only a few honey bee diseases are known to be caused by bacteria. However, pathogens of adult worker bees may go unrecognized due to social immunity mechanisms. For example, infected adult bees usually abandon the hive or are removed by guards. *Serratia marcescens* is an opportunistic pathogen of many plants and animals and is identified at low abundance in the guts of honey bee workers. It has also been isolated from Varroa mites and the hemolymph of dead and dying honey bees. However, the severity and prevalence of *S. marcescens* pathogenicity in honey bees have not been fully investigated. Here we characterized three *S. marcescens* strains isolated from the guts of honey bees and one previously isolated from hemolymph. *In vivo* tests confirmed that *S. marcescens* can be pathogenic in workers. However, expression of antimicrobial peptide and phenoloxidase genes was not elevated following infection, suggesting that these *S. marcescens* strains can evade the immune response of honey bees. By surveying four different locations in the United States we identified the presence of *S. marcescens* in the guts of over 60% of the worker bees evaluated, but at very low relative abundance. Our results indicate that *S. marcescens* is a widespread in adult honey bees and can be highly virulent under certain conditions such as perturbation of the normal gut microbiota. Therefore, we hypothesize that *S. marcescens* represents a pathobiont of honey bees (i.e. a potentially pathogenic organism which, under normal circumstances, lives as a symbiont).

## **Foreign fungi in native bees across the Commonwealth of Virginia**

**Kathryn LeCroy<sup>1</sup>, Erin Krichilsky<sup>1</sup>, T'ai Roulston<sup>1</sup>, Heather Grab<sup>1</sup>, Bryan Danforth<sup>1</sup>**

A risk of introducing exotic species into new ecosystems is introducing their pathogens along with them. A number of mason bee species (genus *Osmia*) have been introduced into the United States in the last 50 years. The Japanese horn-faced bee, *Osmia cornifrons*, was introduced by USDA researchers in the 1960's-70's for crop pollination. In 2002, another Japanese species, *Osmia taurus*, was first documented in the United States, but it is unknown how it arrived. Previous research found *Ascosphaera naganensis*, a pathogenic fungus native to Japan, present in *O. cornifrons* in New York. *Ascosphaera* is the causative agent of chalkbrood, a frequently serious disease for cavity-nesting bees, which can lead to production shortages in agricultural systems managing cavity-nesting bees. This study examined prevalence of *Ascosphaera* across the Commonwealth of Virginia in the nests of both exotic and native *Osmia*. Our trapping efforts, carried out by citizen scientists at 100 locations, yielded over 1,000 specimens for molecular analysis with Sanger sequencing. Results indicate Japanese species, *Ascosphaera naganensis* and *Ascosphaera fusiformis*, are present in native Virginia mason bees, *Osmia lignaria* and *Osmia georgica*, with high prevalence of *A. naganensis* found in *O. georgica*. Our study is the first record Japanese *Ascosphaera* in native North American *Osmia*. We also found *A. naganensis* in multiple parasitic wasps (family Chrysididae), which highlights potential for *Ascosphaera* to move across trophic levels. Future work will involve assessing pathogenicity of Japanese *Ascosphaera* for *Osmia* to better inform our understanding of these novel associations between native hosts and exotic fungi.

Session 2 - Drivers of Host-Pathogen Interactions

## **Traits as Drivers of Plant-Pollinator-Pathogen Networks**

**Scott H. McArt<sup>1\*</sup>, Quinn S. McFrederick<sup>2\*</sup>, Lynn S. Adler<sup>3</sup>, Becky E. Irwin<sup>4</sup>, Christopher R. Myers<sup>5</sup>, Stephen P. Ellner<sup>6</sup>, Paige A. Muñiz<sup>1</sup>, Ashley A. Fersch<sup>1</sup>, Peter Graystock<sup>7</sup>, Wee Hao Ng<sup>1</sup>**

Pathogens are a main contributor to declines in pollinator health, yet we know surprisingly little about how pathogens are transmitted among different species of bees. Such epidemiological information is crucial if interventions will be considered to limit disease transmission and spread, thereby improving pollinator health. We have recently found that flowers are an efficient venue for pathogen transmission among bees. Indeed, we find that about one sixth of individual flowers in eastern old-fields test positive for bee pathogens, and bees acquire pathogens when visiting contaminated flowers. However, how can disease spread possibly be predicted in highly complex plant-pollinator networks, which are often comprised of dozens if not hundreds of plant and pollinator species? In this talk, we will outline our efforts to merge a trait-based approach with network modeling to understand disease spread in diverse communities of bees that transmit pathogens at flowers. We will show new theory that predicts disease hotspots and super-spreaders in trait space. Then, we will test trait-based theoretical predictions with empirical data and computational approaches using a diverse community of bees (>100 species) that transmit pathogens at nearly 100 species of flowers. Practically, our approach can inform the design of improved pollinator-friendly wildflower plantings that maximize forage while minimizing disease transmission among bees.



# Friday, July 19: Session 3

## Variable Climates and Changing Pollinators

### Bee nutritional ecology: from genes to landscapes

Christina M. Grozinger •

Multiple interacting factors are driving bee declines, including parasites and pathogens, as well as biotic and abiotic features of the landscape, such as the availability of forage, pesticide use, and climate. Our studies evaluating the genomic responses of bees to multiple stressors have demonstrated that nutrition and metabolic pathways play a critical role in supporting bee health. Moreover, macronutrient ratios in pollen underpin bee resilience to diverse stressors, and bee foraging preferences are shaped by these nutritional factors. While these studies can inform pollinator forage and habitat restoration, bees can forage over large distances and thus be exposed to stressors at the landscape scale. To address these issues, we have developed online decision support tool, Beescape, to help beekeepers, land managers, growers, and policymakers evaluate the quality of their landscapes for supporting bee populations and obtain recommendations for improving their landscapes and management practices.

### BEE RESPONSES TO CLIMATE CHANGE: FROM MICRO- TO MACROECOLOGY

Jessica Forrest<sup>1</sup>

Anthropogenic climate change can affect bees and other pollinators through a variety of processes, from physiological effects of warming temperature through altered interactions with other species to geographic range shifts. Of course, these various effects do not operate on the same scale, and they are not independent of one another; rather, they are hierarchical. For example, if locally rising temperatures, by affecting activity rates, alter local species interactions in such a way that rates of population growth fall below zero, local extinction is likely; spatially clustered local extinctions ultimately result in range shifts. So, in principle, it should be possible to infer regional-scale (macroecological) changes from local (“microecological”) processes. However, this integration across scales is rarely achieved, at least in the pollinator context. In this talk, I will describe recent work from my lab on populations of a solitary bee species (*Osmia iridis*) distributed across an elevational (and climatic) gradient. We have focused on local-scale, “microecological” effects of temperature variation on bee activity levels, phenology, generation times, and interactions with flowers and parasites. All of these things are affected by temperature, but what will be the net effect for this bee species in terms of its global abundance, geographic distribution, and long-term fate? Here, I propose a way for researchers to clarify our thinking about climate-change impacts on pollinators (specifically, bees), and I suggest some contexts in which we can use an understanding of local-scale processes to anticipate larger-scale changes in species distributions—and perhaps even to inform management.

### A Climate Vise of Temperature Extremes May Explain Past and Predict Future Bumble Bee Range Shifts.

Michael E. Dillon<sup>1</sup>, Kennan J. Oyen<sup>1</sup>, James D. Herndon<sup>2</sup>, Meaghan Pimsler<sup>3</sup>, James P. Strange<sup>2</sup>, Jeffery D. Lozier<sup>3</sup>

Recent shifts in the geographic ranges of bumble bees appear to be tightly linked to changes in climate. Warming temperatures could explain range contraction at the southern edge, but the failure of many bees to move northward remains enigmatic. As with many other species, we know little about the mechanistic links between changing temperatures and bumblebee physiology. To address this gap, we first measured critical thermal limits of *B. vosnesenkii* workers reared from queens collected across the geographic range of the species (from southern CA to northern OR). We found strong divergence in cold (but not heat) tolerance across latitude and altitude, with CTmin closely tracking winter minimum temperatures of the queen collection sites. We predict past distribution by filtering historic climate data with measured thermal limits. Finally, we used projected climates to predict future range shifts given geographic variation in both temperature extremes and in bumble bee thermal tolerance.

### Effects of Climate Change on Nesting Habits of Megachilidae Bee Species in Northern Arizona

Lead Author (Janice Baldwin-Rowe), Co-author 1 (Lindsie McCabe)

This project examines the impacts of climate change on Megachilidae nesting habits in Northern Arizona. Changing temperature and precipitation levels in Northern Arizona impact the populations of native tree species. Many indigenous Megachilidae species nest in native trees, so when the tree populations are reduced due to climate change, Megachilidae nesting patterns are disrupted as well. We hypothesized that if bee blocks of non-native wood and native wood types were distributed at different life zones in Northern Arizona, bees would prefer to nest in native wood bee blocks. Results indicated that there is not a significant difference in bee nesting preference between non-native wood, Ponderosa pine, and aspen bee blocks. There was a significantly lower difference in the number of nests found in fir bee blocks. Nesting habitats will continue to be limited at higher elevations, but artificial nesting blocks may help stabilize Megachilidae populations in Northern Arizona.

### TESTING THE PHENOLOGICAL MISMATCH HYPOTHESIS FOR A PLANT-POLLINATOR INTERACTION

Charlotte W. de Keyser, James D. Thomson<sup>1,2</sup>, J. Scott MacIvor<sup>1,3</sup>

Phenological mismatches occur when organisms respond differently to environmental cues. Climate change can alter these cues and as a result, mismatches are predicted to become more common in the future. For most plant-pollinator interactions, detecting

mismatch is difficult because long-term data are lacking. When mismatches are observed, the mechanisms behind the phenological shifts are rarely explicitly tested. Fortunately, manipulative experiments can help to overcome these difficulties. Here, we present the first experimental test of the phenological mismatch hypothesis where both plant (*Cercis canadensis*) and pollinator (*Osmia lignaria*) are exposed to the same cue manipulations. Although experiments designed to investigate plant-pollinator mismatch exist; to date, these experiments have only manipulated one side of the interaction. Our experimental species are both generalists; however, *O. lignaria* has a strong preference for *C. canadensis* pollen and early nesting success has previously been linked to *C. canadensis* availability. To assess the mismatch hypothesis, we manipulated three abiotic cues (chilling temperature x chilling length x forcing temperature) in environmental chambers. We found that all three cues were used to time first flowering and adult bee emergence phenology, but the strength of the forcing cue was organism dependent. Because the plant was more responsive to warmer forcing temperatures than the bee, female bees were more likely to emerge after the average date of first flowering. Under warm spring conditions, bees that are unable to mate and find a nest immediately upon emergence may be at risk of missing peak flowering of this preferred pollen source.

## Phenological mismatch between bees and flowers early in the spring and late in the summer

**Gaku Kudo** 1

Changes in snowmelt regime affect phenology of many organisms in high latitude and altitude ecosystems. When the synchrony of flowering and pollinator emergence time is disturbed by climate change, symbiosis between plants and pollinators may decay. Phenological mismatch between interacting species tends to occur at the marginal growing season. I introduce how phenological mismatch occurs between bumble bees and bee-pollinated flowers in the early spring of cool-temperate forest and the late summer of alpine ecosystem in northern Japan. The mechanism of phenological mismatch between spring ephemeral and overwintered bumble bees were analyzed based on a long-term monitoring and a snow removal experiment in a deciduous forest. Flowering onset was explained with surface thermal degree-days after snowmelt, while bee emergence was determined by soil temperature. Early snowmelt intensively accelerated the flowering time more than the emergence time of bees. Phenological mismatch in early spring occurred when snow melted early but subsequent soil warming progressed slowly. Seed-set success of plants was strongly related to the extent of mismatch. In the alpine ecosystem, colony development of bumble bees depends on the floral resource of snowbed communities. Flowering period of snowbed communities highly varied from year to year reflecting the fluctuation of snowmelt time. However, seasonal dynamics of bees were more conservative probably because of eusocial lifecycle. Mismatch occurred in early-snowmelt summer in which flowering progressed rapidly and bee populations increased after the major flowering period. In both cases, therefore, acceleration of flowering phenology more than bee phenology is a major cause of mismatch.

## Climate change impact on Brazilian pollinators

**Tereza Cristina Giannini**<sup>1,2</sup>; **Wilian França Costa**<sup>1</sup>; **Rafael Cabral Borges**<sup>1,2</sup>; **Leonardo Miranda**<sup>1</sup>

Global climate change directly affects biodiversity, also affecting the interactions occurring between species. An interaction that deserves special attention is that between plants and their pollinators. In Brazil, we have conducted studies aiming to evaluate the impacts of climate change on Brazilian crop pollinating bees, and on the pollinators (bees, birds and bats) that occur in an Amazonian protected area, the Carajás National Forest. To analyze vulnerability of crop pollinators, we firstly determined that 60% of crops in Brazil are dependent on pollinators, with a greater or lesser degree of dependence. Secondly, we evaluated 95 pollinators from 13 pollinator-dependent crops and found that pollinators' occurrence probability could fall 13% by 2050. Almost 90% of the 4,975 Brazilian counties analyzed will experience loss of pollinator species in the next 30 years. As for Carajás, we determined a potential reduction of pollinating species that ranges from 70 to 100% based on different scenarios. Some Amazonian protected areas in the vicinities of Carajás presented greater climatic suitability, and we are evaluating the application of ecological corridors to connect these areas to improve the climate-forced dispersal of species. Conservation projects including climate-refuge areas and wildlife corridors are urgent to guarantee the preservation and provision of pollination in this area.

## Climate Change Effects on the Status, Distribution, and Phenology of California Bumble Bees

**Leif L. Richardson**<sup>1,2</sup>, **John Mola**<sup>3</sup>, **Kristal Watrous**<sup>4</sup>, **Neal Williams**<sup>3</sup>, and **S. Hollis Woodard**<sup>4</sup>

Climate change is altering the geography and timing of plant-pollinator interactions. Such changes are associated with increases in temperature, drought, fire frequency, extreme weather events, and decreases in winter snowpack. While some plants and bee pollinators are tracking climate change in parallel, there is evidence for other pairs of mutualists of both spatial and phenological mismatch driven by climate. California is critical to US agriculture and also hosts more than half of all bumble bee (*Bombus*) species found in North America. We constructed a database of bumble bee occurrence records spanning more than a century and used it to assess changes in bumble bee species abundance, distribution, and phenology between historic and recent time periods. We found evidence for both persistence and declines among the state's native bumble bees. We report that some species, such as *B. centralis*, appear to be shifting their distributions northward and upslope in response to warming, while others with historic ranges confined to Pacific Coast habitats, such as *B. crotchii*, may be shifting southward. Many of the state's more common bumble bees are tracking warming by advancing mean emergence date of foundress queens by up to 3 weeks, yet we find evidence that some declining species are failing to track advancement of spring in this way. Taken together, our results suggest that climate change is interacting with other threats such as agricultural intensification, development, and habitat loss to drive strong shifts in the distribution and ecology of California's native bumble bees.



# Friday, July 19: Session 4

## *Causes and Consequences of Pesticide Use: from use patterns to pollination services*

### **A new framework for environmental risk assessment of pesticides**

**Francisco Sánchez-Bayo**

Pesticides applied to agricultural land contaminate a significant proportion of the planet's land, water and air. As a result, innumerable species of animals associated with agricultural landscapes are declining at rates that may put them on the brink of extinction in a few decades. It is obvious that current risk assessments of agrochemicals have failed to protect the environment on a large scale. Despite recent improvements, current risk assessments are still deficient on three accounts: i) the effect of the time factor on toxicity at the individual level; ii) the combined effect of mixtures, and iii) the inclusion of population endpoints that are crucial for the survival and recovery of species. Taking bees as a case study, a new framework for risk assessment is proposed here that combines the mandatory introduction of new toxicity endpoints with a more logical assessment of risks within the existing tiered approach. In the first tier of the risk assessment, chronic toxicity tests designed to detect time-cumulative effects should be a requirement for assessing delayed mortality among individuals in a population. In a second tier, crucial population endpoints, such as fecundity and other reproductive outcomes, should demonstrate unambiguously that no negative effects on the viability of a species occur before a chemical can be registered. Emphasis is given to the modern tenet of toxicology: "the dose and the time of exposure makes the poison." Thus, dose and time help us understand the severity of the impacts that pesticides have on bees and their colonies.

### **Putting insecticides on the map for pollinator research and conservation**

**Margaret Douglas<sup>1</sup>, Eric Lonsdorf<sup>2</sup>, and Christina Grozinger<sup>3</sup>**

Wild and managed pollinators are essential to both food production and the function of natural ecosystems; however, multiple stressors including insecticide use threaten their populations. Because most pollinator species travel hundreds to thousands of meters to forage, recent research has stressed the importance of studying drivers of pollinator decline at the landscape scale. However, scientists' ability to do this has been limited by a lack of data on insecticide use at relevant spatial scales and in toxicological units meaningful to pollinators. We synthesized several large, publicly available datasets on insecticide use, land use, and toxicity to generate novel datasets that describe patterns of aggregate-

insecticide use in units of honey bee lethal doses from 1997-2014 for states and major crop groups in the continental US. Furthermore, we illustrate how these estimates can be mapped to existing land use data to facilitate landscape-scale research and conservation planning for pollinator populations and their ecosystem services. Lastly, we summarize variation in US agricultural insecticide use over space and time and the hypothesized drivers of this variation.

### **Estimating pollinator pesticide exposure**

**Maj Rundlöf, Nika Galic, Ove Jonsson, Björn K. Klatt, Jessica L. Knapp, Eric V. Lonsdorf, Charlie C. Nicholson\*, Ullrika Sahlin, Clara Stuligross, Neal M. Williams\* & Joachim de Miranda\***

Pesticide use has the potential to expose pollinators when they forage on crop and wild plants. Such pesticide exposure can affect the pollinator's survival and ability to provide pollination services. Our knowledge on pesticide exposure to pollinators are currently limited, particularly for non-*Apis* bee species, for mixtures and in real landscapes. In our projects, we aim to quantify exposure of pesticide mixtures to bees across crops, landscapes, regions, seasons and years. We explore two approaches to estimate pesticide exposure to pollinators: 1) quantification of pesticide residues in pollen and nectar collected by several species of bees in combination with identification of pollen to plant species origin and 2) extending bee foraging models to include temporally and spatially explicit pesticide use information combined with pesticide environmental fate information. We will present a few cases highlighting the variation in estimated pesticide exposure depending on the approach used, where the study is performed and the pollinator species in focus. The outcomes of the projects are relevant for monitoring pesticide residues in terrestrial environments and for ecological risk assessment of chemical mixtures.

### **A Risk Assessment of Neonicotinoid Insecticides in New York**

**Travis Grout<sup>1</sup>, Scott McArt<sup>1</sup>**

Like any pest management technology, neonicotinoid insecticides present both benefits and risks, which vary with location, crop, timing, and many other conditions. This study examines the trade-offs of common neonicotinoid uses in New York State. Rather than using a "no treatment" alternative, we compare the benefits and risks of neonicotinoids relative to likely substitute pest management products and/or practices. This approach permits a realistic assessment of the impacts, both positive and negative, of a range of potential restrictions on neonicotinoids. While this risk assessment is intended to support science-based regulation, it makes no recommendations or policy prescriptions. Finding the "best policy" for New Yorkers will require hard choices between competing priorities. This study aims to clarify those choices and their likely consequences.



## **Risk of exposure in soil and sublethal effects of systemic insecticides applied to crops on adult female ground-nesting bees using the hoary squash bee as a model species.**

**D. Susan Willis Chan<sup>a</sup>, Ryan S. Prosser<sup>a</sup>, Jose L. Rodríguez-Gil<sup>a</sup>, Nigel E. Raine<sup>a</sup>**

Ground-nesting solitary bees comprise 70% of bee species in temperate climates. In these species, female bees contact relatively large amounts of soil as they excavate their nests. Using the hoary squash bee (*Peponapis pruinosa*) as a model species, we evaluated the risk to adult female ground-nesting bees of exposure to lethal doses of systemic insecticide residues (clothianidin, thiamethoxam, imidacloprid, chlorantraniliprole) in agricultural soil in Ontario, Canada. To do this, we gathered agricultural soil samples at biologically relevant depths both during the bee-active period (July/August) and before insecticide application was made. Samples were analyzed for insecticide residues, and the residue concentrations were fitted to a distribution curve relating concentration to probability of exposure. Three LD50 benchmarks were then applied to the distribution curve to determine the probability of exceeding these benchmarks. Our assessment demonstrated high risk to ground-nesting bees, of exposure to lethal doses of clothianidin, thiamethoxam, and imidacloprid residues in agricultural soil based on the hoary squash bee model. No exposure risk was found for chlorantraniliprole. In parallel to our risk assessment, we introduced mated adult female hoary squash bees into net-covered hoop-houses in which a squash crop had been treated with imidacloprid, thiamethoxam, or chlorantraniliprole or not treated to evaluate the effects of exposure to these insecticides on nest establishment, reproduction, and pollen harvest. Statistically significant sublethal effects on pollen harvest, nest establishment, and reproduction were found for bees exposed to imidacloprid-treated squash plants with no effects found for bees exposed to squash plants treated with thiamethoxam or chlorantraniliprole.

## **Delayed lethality: The effects of a widely-used fungicide on honey bees (*Apis mellifera*)**

**Adrian Fisher II, Teddy Cogley, Meredith Johnson, Aurora Beans, Dena Kalamchi, Kenyan Kerman, Jon Harrison, Jennifer Fewell, Osman Kaftanoglu, Brian Smith, Gloria DeGrandi-Hoffman**

Populations of the honey bee (*Apis mellifera*) and other pollinators are declining worldwide for unexplained reasons, threatening over \$12 billion in agriculture that depends on pollination services. Fungicides, often considered harmless to pollinators and other animals, are often applied on flowers before pollination to prevent rot and disease, leading to consumption of fungicides by pollinators. Despite frequent exposure, few studies have documented isolated effects of fungicides on honey bee health and behavior. To mimic likely fungicide consumption under field conditions, we forced colonies to feed on pollen containing the fungicide formulation Pristine<sup>®</sup>, which is composed of a carboxamide fungicide, boscalid, and a strobilurin fungicide, pyraclostrobin, at four doses ranging from 0.1 to 100x concentrations previously reported for agricultural pollen. Pristine<sup>®</sup> consumption reduced adult population by 20-40% in a dose-dependent manner, with effects increasing over time, and reduced over-winter survival. Pristine<sup>®</sup> exposure increased

pollen consumption and foraging, suggesting it may interfere with protein digestion or absorption. Honey bees exposed to the fungicide also began foraging at an earlier age and had higher in-hive mortality rates. These findings suggest that fungicides may play a significant role in pollinator decline. Our results may influence current fungicide application regimes and the importance of understanding fungicides in future assessments of honey bee health.

## **Towards Next Generation Chemistries For Reducing Arthropod-Borne Disease In Honey Bee Colonies**

**Troy D. Anderson**

The honey bee plays an economically vital role in global agriculture as a pollinator of a wide variety of food and fiber crops to satisfy the needs of human and animal health. The loss of honey bees is a major environment health challenge that demands attention from the scientific community. There are numerous environment stressors that negatively impact the health and survival of honey bees, although a growing consensus identifies the high levels of parasites and pathogens, especially arthropod-borne viruses, are among the most significant threats to the health of these pollinators. A common approach to arthropod-borne virus management is the use of synthetic neurotoxicants alone or in combination with organic acids and botanical oils to reduce ectoparasite infestations. These conventional acaricides not only have adverse health effects on honey bees, but widespread acaricide resistance limits their use to reduce mite infestations and their transmission of viruses to honey bees. The development of acaricide resistance is an evolutionary phenomenon that requires appropriate and comprehensive monitoring and management strategies within an integrated vector management framework. Here, I will discuss (i) novel acaricide resistance surveillance and reporting tools, (ii) alternative interventions to mitigate acaricide resistance evolution and preserve the efficacy of existing acaricides, (iii) exploration of next generation anti-parasite and -pathogen modes of action and their applications, and (iv) building of local, national, and global response partnerships that are actively engaged in reducing arthropod-borne diseases to honey bees and improving the health and protection of these pollinators.



# Saturday, July 20: Session 5

## *Integrative Approaches to Improving Bee Health Across Landscapes*

### **BEE PROTECTION AND CONSERVATION**

**Cedric Alaux, Jean-Luc Brunet, Mickael Henry**

Due to growing anthropogenic pressures, heavy losses of honeybee colonies (*Apis mellifera*) have been reported around the world, and the species richness and abundance of many groups of wild bees have recently declined across Europe and North America. Assessing the possible impacts of ongoing and future environmental changes and developing mitigative policies are therefore at the top of the agenda. For that purpose, there is an urgent need to identify new metrics that can be used to capture the state of health of bees and therefore improve their population monitoring. Indeed, monitoring is essential for species conservation/protection as it can serve as an early warning system for detecting environmental problems, but can also help to determine how well remedial actions are working. As a proof-of-concept, we showed that the combination of landscape ecology and physiological metrics of honeybee health status is a highly valuable approach to measure the efficiency of habitat-restoration and enhancement schemes. More specifically, we found that honeybee physiology was significantly improved by the presence of flowering catch crops, which were associated with a significant increase in pollen diet diversity. The influence of semi-natural habitats on bee health was even stronger. We therefore suggest transposing this approach, combining physiological and ecological data, to wild bees to better assess habitat suitability and effectiveness of current and future conservation strategies.

### **Keeping bees in a warming world: Protein biomarkers for heat stress and queen failure diagnostics**

**Alison McAfee**

Honey bee colony health and productivity is intrinsically linked to the quality of the queen. Unfortunately, queen quality is compromised by stressors such as extreme temperatures and pesticide exposure, both of which drastically reduce their stored sperm viability. This causes colonies to dwindle, produce less honey, and ultimately fail. However, diagnostic tools for identifying root causes of queen failure are lacking, so beekeepers are often left wondering why their queens have failed and are unable to take evidence-based action to prevent recurrent failures. Here, we exposed queens to a range of temperatures (5 to 42 °C) and measured the impact on their stored sperm viability while looking for protein biomarkers that indicate temperature stress. We found that exposing queens to 42 °C for 4 h reduced sperm viability by 56% and induced expression of strictly ATP-independent heat-shock proteins (HSPs) in queens' spermathecae, but not in their ovaries. Viability of ejaculated sperm and the drones (males) themselves plummets with heat, with only 42% of drones surviving 6 h at 42 °C. Conversely, 100% of queens survive. This sex-biased heat tolerance may be a quality control mechanism to ensure that queens are inseminated only with high quality

sperm. We are currently testing if expression of specific HSPs we observed with laboratory heat-shock experiments are useful biomarkers in the field. These signatures could serve as biomarkers for heat stress and enable post-failure diagnostics for the beekeeping industry. Honey bees may yet become sentinels for heat-induced loss of sperm viability in diverse landscapes.

### **Factors influencing colony survival in migratory beekeeping based on honey bee resistance traits**

**Michael Simone-Finstrom<sup>1</sup>, Thomas O'Shea-Wheller<sup>2</sup>, Hannah Penn<sup>1</sup>, Frank Rinkevich<sup>1</sup>, Bob Danka<sup>1</sup>, Kristen Healy<sup>2</sup>**

Commercial beekeeping in the United States and Canada accounts for the majority of colonies in circulation, with migratory pollination comprising <75% of the industry. Notably, this system experiences high overwinter losses on a yearly basis. Crucially, without substantial improvements in bee health, large-scale migratory pollination is likely to become both biologically unsustainable, and commercially infeasible in its current form. Consequently, our research aims to identify the key predictors of colony loss, and their relative weightings and importance through a targeted multi-year longitudinal study of honeybee health in migratory beekeeping. As such, we analyse pathogens, agrochemicals, nutrition, and parasites, at a large spatiotemporal scale, in a real commercial pollination operation. Moreover, the study incorporates a test of Pol-Line bees, a stock bred for resistance to *Varroa destructor*, which our current data indicate to account for up to 70% of observed mortality across regions. Pol-Line bees are unique in their combination of mite resistant traits paired with favourable beekeeping characteristics, and thus represent a viable integrated solution to ongoing and intense parasite pressure. Further work showing differential responses to viral infection and how this may impact field results will be discussed. In sum, our research aims to parsimoniously quantify the relative impacts of key stressors, allowing for more effective management and predictive modelling. Furthermore, we evaluate a mite resistant commercial bee stock, providing a potentially integrated solution to the ongoing *V. destructor* pandemic.

### **TEMPORAL AND SPATIAL DYNAMICS OF POLLINATOR COMMUNITIES ACROSS NC AGROECOSYSTEMS**

**Hannah (Levenson)<sup>1,2</sup>, David (Tarpy)<sup>1,2</sup>**

Honey bees and native bees together provide important pollination services to our agricultural and natural landscapes. In agriculture, this equates to pollinating over \$18 billion worth of crops annually in the United States. In natural landscapes, bees pollinate more than 80% of our flowering natural areas. Despite their economic and ecological importance, bees are facing extreme population pressures from factors such as pathogens, pesticides, and habitat loss. Additionally, there are few native bee species whose populations have been thoroughly documented, especially when

compared to the wealth of knowledge we as a scientific community have on honey bees. This study fills the research gap on native bee populations by focusing on an initiative implemented by the North Carolina Department of Agriculture (NCDA) called “Protecting NC Pollinators.” This initiative mandates the planting of wildflower seed mixes – creating ‘pollinator plots’ – at all NCDA Experimental Agricultural Research Stations across the state in order to preserve biodiversity. For the past three years, we have surveyed native bee populations at these plots in order to measure the impacts of the plots on population growth and health over time. As such, this study is the most detailed survey of native bee populations in NC to date and is the first to empirically measure the consequences of planted habitat on native bee populations at the physiological, disease, *and* community levels. Additionally, findings from this study could be used to facilitate nationwide studies and advise national policymakers on ways to protect pollinator communities.

## **THE EFFECTS OF LAND COVER ON HABITAT QUALITY FOR NESTING BUMBLEBEES**

**Lead Author (Genevieve Pugsek), Co-author 1 (Elizabeth Crone)**

Evaluating habitat quality is a fundamental goal of conservation ecology. Ideally, evaluations of habitat quality should incorporate demographic measures of species success, because abundance, survival, and reproduction are not always positively correlated. Here, we used two metrics to evaluate the impacts of land cover on *Bombus impatiens* nesting habitat quality: colony density and colony reproductive output. Using mark recapture methods, we compared bumblebee nest densities across three different land cover types (forests, meadows, and hayfields) at three farms in Massachusetts. Nest density surveys were conducted at 30, 1500 m<sup>2</sup> plots, which were each searched on four different occasions. After locating colonies (n = 17), we evaluated reproductive output of each colony by counting newly produced queens entering or exiting nest entrances for a 30-minute period 4 times a week. Our nest density surveys suggest that natural habitats, like forests and meadows, provide more suitable nest sites for bumblebees than do hayfields. We found no nests in hayfields (0 nests per hectare), and similar nest densities in forests (6.20 nests per hectare) and meadows (4.73 nests per hectare). These results contrast our surveys of reproductive output: we encountered a greater number of queen bumblebees at colonies located in forests (16.8 queens encountered per colony) than at colonies located in meadows (5.3 queens encountered per colony). Our research is one of few to address the impacts of land cover on bumblebee nest density and showcases the value and feasibility of demographic metrics for evaluating habitat quality for ground-nesting pollinators.

## **Improving Bee Health in Canola Pollination**

**Shelley Hoover**

The link between canola and beekeeping is strong in Canada. Approximately 75% of Canadian honey production is from canola, and an estimated 78-84% of the economic value of pollination to agricultural production is due to the commodity and hybrid seed canola production, grown on over 21 million acres of land. As a result, there is a strong imperative to ensure both sustainable bee health and pollination services in this crop. This is especially critical in Alberta, which has over 40% of the managed honey bee colonies in Canada, 6.5 million acres of commodity canola, and a thriving seed production industry pollinated by both honey and leafcutter bees. Our research takes a multi-pronged approach to the issue of sustainable bee health and pollination in canola

production systems. I will present results from specific experiments examining honey bee management (including colony size, protein supplementation, hive-product production, and queen production), discuss the relationships between managed honey and leafcutter bees (pathogen transmission, and behavioural interactions), and our studies of the effects of pollination and other inputs on canola yield. We find there are many approaches through which beekeepers can increase bee health, profit, and the delivery of pollination services, and there are many areas of common ground between beekeepers and crop producers. The results of this research are used to inform government surveillance and policy decisions, to advocate on behalf of the industry, and to provide beekeepers and crop producers with best management options to maintain bee health and pollination services.

## **MITIGATING POLLINATOR FORAGE LOSSES CAUSED BY LAND USE DECISIONS**

**George Hansen**

Every year a significant amount of pollinator forage is lost due to a host of causes. Land use decisions are intended to do things that will in their own way be good, like help infrastructure, create opportunity, make money, or some other motivation. However, rarely if ever in the decision-making process is what has been lost even identified, let alone replaced or mitigated. For instance, forage, habitat, wildlife shelter, pollinator nesting sites, to name a few, are commonly lost or diminished in our rush to use land for a new, more efficient purpose. Sometimes policies, rules or laws are enacted to compensate for our activities. For example, in Oregon, when a parcel of public or private land is logged by clear cut, it must be replanted within a few years to avoid a tax penalty. But many land owners, when given the chance, will try to do the right thing even without regulations. I want in this talk to point out some voluntary actions that are a positive response to pollinator forage losses caused by land-use decisions. Many are currently being demonstrated as options, and can make a huge difference in mitigating landscape impacts from a variety of land use decisions, but without regulatory action.

## **Impacts of landscape-scale floral resource availability on pollinator communities**

**Aaron Iverson, Allyson Evans, Heather Grab, Alison Power, Scott McArt**

The surrounding landscape is often an important predictor of pollinator communities, yet a mechanistic understanding of why the landscape matters is limited, partially due to the lack of detailed characterizations of the landscape. We addressed how floral resource availability at landscape scales relates to the abundance and diversity of pollinators in New York State, USA. We modeled floral resource availability by first sampling the plant community composition in all major habitat types in the region. We then translated the plant community data into the amount of floral area present through time per habitat by measuring flower density, flower size, and phenology of each plant species. Using pollinator data collected from different studies in the same geographic region, we tested whether a detailed floral resource depiction of the landscape improved the ability to predict pollinator communities compared to coarser metrics, such as percent agricultural land. We found that floral resource availability can be an important predictor of pollinator communities, yet the strength of the relationship varies according to pollinator groups and traits.



# Saturday, July 20: Session 6

## *Pollinators in Urban Environments*

### **Floral trophic ecology of a North American metropolis revealed by honey bee foraging assay**

**Doug Sponsler**

Florivory, a trophic interaction of which pollination visits are a special case, was likely instrumental in the co-diversification of angiosperms and insects in the mid-Cretaceous and remains a trophic keystone of modern plant-arthropod communities. The ecological centrality of flowers-as-food extends to systems in which the historic floral trophic interface has been dramatically altered, such as urban landscapes characterized by novel assemblies of native and exotic flora. Urban landscapes can host surprisingly diverse communities of flowers and flower-visiting insects, sometimes even functioning as refugia for rare species. Studying floral resources at the landscape scale is technically daunting under any circumstances, but the challenge becomes especially acute in urban landscapes where the physical obstacles of the built environment and the limits of land access often render traditional approaches to floral surveying impracticable. Flower-visiting insects, having the advantage of flight and a disregard for property lines, might be harnessed as elegant environmental samplers of landscape-scale floral resources, provided the spatial and taxonomic scope of their interaction with the landscape is well-understood and the materials they collect can be identified and quantified. The western honey bee (*Apis mellifera* L.) is arguably the organism best suited to such a sampling approach due to the large spatial scale of its foraging, its ability to integrate landscape-scale information to allocate foraging effort dynamically to the most rewarding floral patches, and its amenability to standardized sampling techniques. We employed a network of 12 sentinel apiaries, sampled over two years, to characterize the floral resources of Philadelphia, PA. Using a combination of DNA metabarcoding of monthly pollen and honey samples and hourly colony weight monitoring, we inferred both the taxonomic composition and the cumulative abundance of floral resources throughout the foraging season. Our results reveal a floral resource landscape dominated by woody plants, both native and exotic, including many varieties likely cultivated as ornamentals. We also document clear temporal patterns in the cumulative availability of floral resources, with marked peaks in spring and fall and a dramatic dearth in late summer. These findings lay the foundation for understanding the floral trophic ecology of major cities with a long history of species introductions and a complex mosaic of urban land use forms.

### **Pollinators and urban warming: A landscape physiology approach**

**Elsa Youngsteadt, April L. Hamblin, Margarita M. López-Urbe, Steven D. Frank**

Cities are thermal mosaics. The warmest parts of a city often average 1 – 3 °C warmer than the surrounding landscape, placing them several decades ahead of the global-warming curve. Cities may thereby provide opportunities to test predictions about effects of global change on pollinator health and abundance. As ectotherms, bees are sensitive to environmental temperatures, and each species or population has characteristic heat tolerance limits. We asked whether bee species with low physiological heat tolerance are the ones whose populations decline the most in urban hotspots. We sampled the wild-bee community at 18 sites in Raleigh, NC, USA, using a combination of pan traps, vane traps, and netting. We monitored air temperatures at each site using iButton temperature dataloggers. To assess bee thermal tolerance in the lab, we measured critical thermal maxima (CT<sub>max</sub>) for 15 common species using a heat-ramping assay. The entire community sample included 3,593 individual bees of 113 species. Across sites, total bee abundance declined by 41% per °C urban warming. Among the 15 common species for which we measured CT<sub>max</sub>, those with the lowest thermal tolerance were those whose populations declined the most at hotter sites. Ongoing and future work aim to detect how temperature-related changes in bee behavior, abundance, and community composition alter the provision of pollination services across urban landscapes.

## **Green infrastructure to support urban wild bees: Communicating science to practitioners**

**Scott MacIvor**

Practitioners are encouraged by city staff and citizens to support bees and other pollinators in urban planning and design. Bees are especially portrayed as beneficiaries of urban green infrastructure, such as bioswales, green walls and even green roofs, all of which are commonly planted with pollinator-friendly flowers and food crops. However, individual green infrastructure projects are often isolated and small, embedded into high-density urban conditions and as a result, may be limited in the type and diversity of bees they support. This talk will describe three studies in which wild bees were evaluated on green roofs in Toronto that demonstrate 1) green roofs are limited in nesting opportunities for ground- and cavity-nesting bees, 2) green roofs are dominated by non-native plant species with restricted bloom times supporting functionally narrow bee communities, and finally 3) building height is significantly negatively correlated with bee abundance and diversity. With over 550 green roofs in Toronto, we have a tremendous opportunity to consider them as unique and novel habitat, pose questions and design experiments, and cultivate information that supports best practices and policy to enhance wild bee conservation. These findings have contributed to the development of the new City of Toronto Pollinator Protection Strategy, and this case study will be discussed within the context of green infrastructure, an increasingly common feature of all urban environments.

## **Linking pollinator health, microbiome composition and human provisioning in Anna's Hummingbird (*Calypte anna*).**

**Rachel Vannette\*, Lisa A. Tell, Casie Lee, Rachel Dutch**

Humans provision pollinators in many urban and suburban areas through high-density floral plantings, presentation of sugar water, and providing other pollinator resources. Despite positive effects on caloric availability for a variety of pollinator species, the potential consequences of other components of pollinator health, including disease transmission and effects on pollinator microbiome, remain poorly understood. Here, we assess if human-provisioned sugar water via feeders can serve as a source of pollinator-associated bacterial or fungal pathogens by comparing feeder and flower microbial community composition. Second, we characterize variation in hummingbird microbiome composition of multiple gastrointestinal tissues (upper intestine, lower intestine, and proventriculus) and fecal material from wild birds and deceased rehabilitated birds from environments ranging in anthropogenic influence. We compared the relative influence of bird age, geographic location (urban vs suburban) and symptoms of disease on hummingbird microbiome composition. We found that human-provisioned feeders host abundant microbial populations, which differ from floral resources in their microbiome composition. Some microbes associated with avian disease were detected in feeder sugar water, although in very low frequency and numbers, but none were detected in floral nectar. Bird gastrointestinal microbiomes were distinct among tissue types, distinct from food sources, changed over bird development, and were associated with disease. Novel taxa from bird microbiomes were described and suggest that hummingbirds, despite having incredibly rapid gastrointestinal transit times, host a

resident and potentially functionally important microbiome. Taken together, our results suggest that urbanization can affect pollinator health through diverse routes including effects on disease and microbiome composition.

## **The Effect of Land Use on a Sexually Selected Characteristic of the Cabbage White Butterfly (*Pieris Rapae*) in the United States**

**Anne Espeset<sup>1</sup>, Matthew Forister<sup>1</sup>**

Anthropogenic forces have impacted natural ecological systems over the last few decades at rapid rates. Specifically, the exponentially-increasing use of fertilizer in agricultural fields and increases in nitrogen deposition in developed areas has changed nutrient cycling and availability in these areas. How these human-induced land use and nutritional changes affect sexually selected signals has not been extensively studied. In collaboration with citizen-scientists (The *Pieris* Project), I collected the cabbage white butterfly, *Pieris rapae*, and investigated how land use and nitrogen deposition affect their sexually-selected signal (wing coloration). Butterflies were collected from varying habitats and nitrogen deposition was calculated. Wing reflectivity was compared for individuals across the US. We found that land use, but not nitrogen deposition, had a significant negative effect on pterin reflectance. Specimens from an area of high proportion of crop land are less bright and are less saturated, but do not differ significantly in hue. Our findings are consistent with the possibility that human-induced land use change is affecting the sexually selected signal of *Pieris rapae*.

## **Urban pollinator conservation opportunities: integrating research with policy and practice**

**Katherine Baldock<sup>1</sup>, Mark Goddard<sup>2</sup>, William Kunin<sup>3</sup>, Simon Potts<sup>4</sup>, Phillip Staniczenko<sup>5</sup>, Graham Stone<sup>6</sup>, Ian Vaughan<sup>7</sup>, Jane Memmott<sup>8</sup>**

Pollinators are currently the focus of international concern as numerous studies document their declines and the multiple threats they are facing. Land use change is one of the main drivers of pollinator declines, with urbanization regarded as a major threat to biodiversity. Yet urban areas could represent a fantastic opportunity for pollinator conservation: an increasing number of studies suggest that at least some urban land uses can harbor high pollinator diversity and the appetite of the general public, many of whom reside in towns and cities, for pollinator conservation seems to be ever-growing. I will outline the findings from the Urban Pollinators Project, a national scale study of insect pollinators in UK towns and cities, involving academics along with practitioner partners from local councils and Wildlife Trusts. The research addressed three questions: (1) Where is the UK's pollinator biodiversity? (2) Where are the hotspots of pollinator biodiversity in urban areas? (3) How can we help conserve pollinators in urban areas? We studied multiple urban areas across the UK and used a plant-pollinator network approach to compare urban to rural landscapes (Q1) and to identify urban pollinator hotspots and conservation opportunities (Qs 2 & 3), developing Bayesian models to assess plant-pollinator community robustness at a city scale under different management interventions. I will also describe the knowledge exchange activities that are underway to integrate our research findings with policy and practice, including partnerships with local and national governmental organizations, industry and NGOs.

## **Beekeeping Ordinances: Protecting Bees and Neighbors**

**Tracy Ellis, Jaime Garza**

Many hobbyist beekeepers are currently requesting revision to beekeeping regulations in their communities. San Diego County revised beekeeping regulations to suit local requests but also established an Apiary Inspector to ensure responsible beekeeping. These extra measures of a comprehensive beekeeping program were necessary in southern California's locally over-defensive (Africanized) honey bee zone to protect public safety. The ordinance incorporated expertise and suggestions obtained from the local beekeeping society, commercial beekeepers, community planning groups, stakeholders, and experts. It allowed reduced setback distances from roads and neighboring dwellings, and defined distances from property lines and sensitive sites compared to the previous ordinance. In addition the program recruits beekeepers to abide by state regulations and register their hives, provides educational outreach on best management practices and conducts ongoing compliance monitoring to prevent apiary infestation with over-defensive bees, pests and diseases.

## **Urban Pollinator Conservation: Bee City USA as a Model for Meaningful Community Engagement**

**Phyllis Stiles<sup>1</sup>, Mace Vaughan<sup>1</sup>, Jennifer Marshman<sup>2</sup>**

Global population is expected to grow to more than nine billion by 2050. Already, more than half of the world's population lives in urban areas and about half of the global land mass is being used for agriculture. Sustainability planning is complicated by global climate change which makes predicting future trends challenging. What is not debatable is that pollinators are needed more than ever to help sustain people and the planet. Happily, pollinator research has increased dramatically over the last decade and many nations, states, and cities are incorporating pollinator conservation into their policies and practices. There is no time to waste for engaging the general public in pollinator conservation efforts. Bee City USA was founded in 2012 to test the theory that untrained citizens would create and enhance pollinator habitat in urban and suburban areas if given ongoing encouragement. In the seven years since Bee City USA's launch, both communities and colleges have been experimenting with ways to promote, create, and enhance pollinator-friendly landscaping on public and private land. The commitment to pollinator conservation efforts includes education, celebration and habitat enhancement by reducing pesticide use, integrating more native plants and trees, and removing invasive plants, among other strategies. Annual reports from Bee City/Campus USA affiliates reveal inexpensive, creative ways of mobilizing campuses and communities to alter their landscaping paradigms to welcome pollinators into urban and suburban spaces. We will highlight some of the successful initiatives undertaken by Bee City USA's 150 affiliates to think globally, and act locally.

## **Electric Power Companies Protecting Pollinators**

**Jessica Fox and Ashley Bennett**

Electric power companies have become increasingly aware of pollinator declines and recognize they have an opportunity to manage lands such as rights of ways (ROWs), solar sites, wind energy facilities, and power stations for pollinator conservation. Power companies own and manage millions of acres throughout the United States and internationally. However, a major gap for pollinator research and conservation is translating basic academic research to on-the-ground application and decision making for power companies. In 2018, the Electric Power Research Institute (EPRI) launched a collaborative initiative, Power-in-Pollinators, to accelerate the pace, scale, and effectiveness of pollinator projects in collaboration with power companies. EPRI recognizes the potential impact power companies can have on promoting pollinator conservation by working together on research and conservation efforts. This effort is now the largest collaboration in North America designed specifically to support power companies in research, monitoring tools, and management practices needed to create and maintain pollinator habitat on utility lands. EPRI has also initiated research including: 1) measuring Integrated Vegetation Management (IVM) treatments on pollinator abundance and richness, 2) protecting and increasing monarch butterfly populations by establishing pollinator friendly habitat, 3) developing methods to remotely detect monarch habitat by mapping milkweeds, 4) establishing and maintaining pollinator friendly plantings at solar and wind energy facilities, and 5) documenting the role ROWs play in facilitating pollinator dispersal across the landscape. This presentation will discuss research with power companies and provide insights into how academic research can be developed to consider corporate application and inform land management decisions.



# Poster Sessions

## Investigating the attractiveness of native wildflowers to pollinators and natural enemies

Aaron Anderson<sup>1</sup>, Gail Langellotto<sup>1</sup>

Many organizations have published pollinator-friendly planting lists for home gardeners. However, many lists lack empirical evidence to support recommendations. In 2017-2018, we screened 23 wildflowers native to the Willamette Valley. Plants were selected based upon their potential use in ornamental gardens, as well as anecdotal reports of attractiveness to pollinators. We included four exotic comparators known to be attractive to pollinators. Species were planted in meter-squared plots spaced six meters apart. Between April and October, we monitored pollinator visits and floral bloom phenology. We also sampled insects from plots, weekly. Though this research is ongoing, we have early findings on the attractiveness of these wildflowers to bees. The most attractive plants varied between 2017 and 2018, possibly due to differences in bloom phenology and plant establishment. In 2017, four native wildflower species *Solidago canadensis*, *Symphotrichum subspicatum*, *Clarkia amoena* and *Gilia capitata* were the most attractive flowers for bees, followed by the exotic *Nepeta cataria*. In 2018, three of the top five most attractive plants to bees were exotic garden species. However, this pattern was strongly driven by European honey bee visitation. When we limited our analysis to native bee abundance, the five most attractive wildflowers were all native species. Similarly, across both seasons native wildflowers attracted the highest native bee species richness. This research will continue for a third field season, after which we will publish our own list of pollinator plant recommendations for gardeners in the Willamette Valley region of Oregon.

## Changes in the phenology of the southeastern blueberry bee (*Habropoda laboriosa*) based on historic collections data

Anderson, Sarah E., and Rachel E. Mallinger<sup>1</sup>

It is well documented that many bee species have experienced an advance in phenology over the past century. The southeastern blueberry bee (*Habropoda laboriosa*) is a large-bodied, North American bee that emerges early in spring and is thought to be oligolectic on blueberries (*Vaccinium* spp.) which have an early and relatively short bloom period. These traits make this bee particularly susceptible to changes in phenology that may decouple it from its host plant. This study uses historical data gathered from data repositories and non-databased collection specimens to examine the changing phenology of *H. laboriosa* from 1919 – 2019. Across this time period, collection date for *H. laboriosa* (Julian date) advanced in phenology by 44

days averaged across all latitudes where this bee has been collected. Much of this trend is accounted for, however, by the rapidly advancing phenology of this species at latitudes above 35°N where the advance in collection date is 83 days. Wild *Vaccinium corybosum* is one of the most widespread blueberry species in *H. laboriosa*'s geographic range. Preliminary evidence suggests that the first day of flowering (Julian date) for this blueberry species has advanced as well. Results regarding the changing phenology of this plant will be compared to those corresponding to *H. laboriosa*. Phenological mismatches between *H. laboriosa* and *Vaccinium* spp. have the potential to disrupt this pollination system, with consequences for both plant pollination success and *H. laboriosa* diet breadth.

## Plant-pollinator networks created from DNA metabarcoding data in eastern Oregon are more complex than those created from behavioral observations

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With recent declines of some pollinators, including native bees, many land managers are implementing restoration and conservation plans to enhance native bee habitat. However, information is limited about which plant species serve as major food sources for native bees. During the summer of 2018, we sampled 589 native bees from three locations in eastern Oregon: the United States Forest Service Starkey Experimental Forest and Range, The Nature Conservancy's Zumwalt Prairie Preserve, and Threemile Canyon Farms. We obtained behavioral observations of foraging by recording the flower species that each bee was visiting when sampled. We then washed all pollen from each bee, extracted DNA from the pollen mixtures, and used DNA metabarcoding to identify the plant species in each pollen mixture. These data, coupled with vegetation surveys conducted during each sampling period, allowed us to determine major food sources and preferences for native bees in these areas. The sampled bees were recorded visiting over 50 species of plants, and more than 60% of all bee visits occurred on just 10 plant species across the three sites. Most commonly visited plants included both native species (e.g., slender cinquefoil (*Potentilla gracilis*), Missouri goldenrod (*Solidago missouriensis*), and hoary tansyaster (*Machaeranthera canescens*)), and non-native species (e.g., diffuse knapweed (*Centaurea diffusa*), yellow star-thistle (*Centaurea solstitialis*), and bull thistle (*Cirsium vulgare*)). Plant-pollinator networks created from DNA metabarcoding data were more complex than those created from behavioral observations, indicating that metabarcoding provides a more complete record of bee foraging behavior relative to behavioral observations.

## Effects of Climate Change on Nesting Habits of Megachilidae Bee Species in Northern Arizona

Janice Baldwin-Rowe, Lindsie McCabe

This project examines the impacts of climate change on Megachilidae nesting habits in Northern Arizona. Changing temperature and precipitation levels in Northern Arizona impact the populations of native tree species. Many indigenous Megachilidae species nest in native trees, so when the tree populations are reduced due to climate change, Megachilidae nesting patterns are disrupted as well. We hypothesized that if bee blocks of non-native wood and native wood types were distributed at different life zones in Northern Arizona, bees would prefer to nest in native wood bee blocks. Results indicated that there is not a significant difference in bee nesting preference between non-native wood, Ponderosa pine, and aspen bee blocks. There was a significantly lower difference in the number of nests found in fir bee blocks. Nesting habitats will continue to be limited at higher elevations, but artificial nesting blocks may help stabilize Megachilidae populations in Northern Arizona.

## Radio frequency identification technology use in characterizing feeder visitations and contact network of hummingbirds in urban habitats

Ruta Bandivadekar<sup>1</sup>, Pranav Pandit<sup>2</sup> and Lisa Tell<sup>1</sup>

Despite the popularity of hummingbirds in urban environments, there are limited studies evaluating the effects of changing climate, congregation, sharing food resources and increased contact when hummingbirds visit feeders. To evaluate visitation patterns to feeders over time, we tagged two hummingbird species (Anna's and Allen's Hummingbirds), with passive integrated transponder tags and recorded their visits with commercial RFID transceivers at feeders. Data recorded included the number of feeder visits, time spent at the feeder, and simultaneous feeder visitation by different individuals. For the study period (September 2016 to March 2018), 118,017 detections were recorded at seven feeding stations located at three California sites. The rate of tagged birds returning to RFID equipped feeders at least once was 61.3% (141/230 birds). Females stayed at feeders longer than males per visit. During spring and summer, hummingbirds visited feeders most often in morning and evening hours. Although most hummingbirds visited the feeders during the daytime, our system recorded night feeder visitations (n = 7 hummingbirds) at one site. This efficient use of RFID technology to characterize feeder visitations of hummingbirds in urban habitats could be used in the future to elucidate behaviors, population dynamics and community structure of hummingbirds visiting feeders.

## Toxicity of premixed insecticide chemistries to female blue orchard bees

Joseph Belsky and Neelendra Joshi

Populations of bees have dramatically declined in recent years. At the same time, insecticide application in agro-ecosystems has largely increased worldwide. As a result, investigating the effects of insecticide exposure on bees is vital to ensure future food security and environmental stewardship. Here, we simulated a field realistic exposure scenario of female blue orchard bees, *Osmia lignaria* (Say) to four premix insecticides (containing two or more active ingredients, each with a different mode of action). A spray tower was used in a laboratory setting to simulate insecticide field application. We sprayed bees with one of four premix insecticides in petri dishes and immediately transferred them to clean cages. To simulate realistic grower insecticide application in orchards and subsequent bee exposure, we used formulated products as opposed to technical grade insecticides placed in a distilled water solvent as opposed to acetone. We quantified the resulting mortality of bees from single spray exposure to these insecticides at 24, 48, 72 and 96-hours post-treatment. Bioassay results will be discussed and presented.

## Maximizing the potential and minimizing the cost of prairie seed mix design for wild bees

Kate Borchardt<sup>1</sup>, Julia Brokaw<sup>2</sup>, Julia Schreiber<sup>1</sup>, Bethanne Bruninga-Socolar<sup>2</sup>, Michelle Vohs<sup>2</sup>, Kimiora Ward<sup>1</sup>, Daniel Cariveau<sup>2</sup>, Neal Williams<sup>1</sup>

Enhancing foraging resources for bees by planting diverse wildflower mixes is a common strategy to mitigate bee declines. Many mixes are costly, however, because seed of pollinator-friendly flower species can be expensive. Our research objectives are to determine how variation in seeding rate, wildflower species richness, and grass seeding density affect flowering performance and bee attractiveness of pollinator mixes. We chose these factors because land managers commonly decrease the species richness or seeding density of the mix to lower seed costs. To resist invasion by aggressive weeds, they sometimes add grass seed to the lower density wildflower mixes because grass seed is cheaper than most forb seed, but retains higher overall seed density of the mix. Surprisingly, none of these common strategies have been empirically tested. In Fall-Winter 2018, we initiated a field experiment in two regions, Northern California and Minnesota. The two have contrasting climates, and land use contexts, but both are areas with active pollinator conservation efforts. We sowed replicated plots of wildflower mixes that varied the forb seeding rate, forb species richness, and grass seeding rate. To evaluate the success and establishment of mixes for pollinators and their resistance to invasion by aggressive plants, we are measuring establishment of sown species and weeds, floral area and interactions between the plant and bee community. We will also compare the realized plant community with the seeded mix. Our data will help guide land managers when designing seed mixes that can be affordable and beneficial for wild bees.



## **Wildflower plantings promote blue orchard bee, *Osmia lignaria* (Hymenoptera: Megachilidae), reproduction in California almond orchards**

**Natalie K. Boyle<sup>1</sup>, Derek R. Artz<sup>1</sup>, Ola Lundin<sup>2,3</sup>, Kimiora Ward<sup>2,4</sup>, Devon Picklum<sup>5</sup>, Gordon I. Wardell<sup>6</sup>, Neal M. Williams<sup>2</sup>, Theresa L. Pitts-Singer<sup>1</sup>**

Growing concerns over the availability of honey bee (*Apis mellifera* L.) colonies to meet pollination demands has elicited interest in the use of non-*Apis* managed and wild bees to mitigate current pressures on the commercial beekeeping industry. The blue orchard bee, *Osmia lignaria* (Say), is a native bee species that shows great promise for commercial propagation and as a co-pollinator with or alternative pollinator to honey bees in managed orchards. Here, we present results of a two-year study evaluating the use of *O. lignaria* in combination with honey bees in California almond orchards, where three 0.48 ha wildflower plantings were installed and maintained along orchard edges to support native bee and honey bee forage. Plantings were seeded with native wildflower species that have flowering periods known to overlap with and extend beyond almond bloom. To examine the success and influence of the introduction of *O. lignaria* to almond orchards, we measured bee visitation to almond blossoms and wild flowers, bee reproduction and progeny outcomes, almond fruit set, and nut yield across six 16.2 ha orchard blocks during 2015 and 2016. In 2016, pollen provision composition was also evaluated to confirm and assess *O. lignaria* use of the wildflower plantings. Closer proximity to the wildflower plantings enhanced *O. lignaria* reproduction, and pollen analysis indicates regular visitation to the wildflower plots as far as 800 m from nesting sites. This study highlights the importance and benefit of providing alternative floral resources to managed solitary bees in commercial agricultural landscapes.

## **Optimization of pollinator seed mixes from low resolution data**

**Bethanne Bruninga-Socolar<sup>1</sup>, Daniel Cariveau<sup>1</sup>, Eric Lonsdorf<sup>2</sup>**

Wild bee species are experiencing declines due to habitat loss and other environmental stressors. Restoration of pollinator-friendly habitat is an important tool for mitigating these declines. Current efforts to restore wild bee populations rely on enhancing foraging resources for bees by planting diverse seed mixes of flowering plants. However, seed mixes can be prohibitively expensive, with some pollinator seed mixes costing over \$1,000 per acre. Previous research applies a genetic algorithm to plant-pollinator species interaction data to generate optimal plant species mixes. The optimization maximizes the bee diversity supported by candidate mixes while minimizing their cost. The plant-pollinator species interaction data are time-consuming and expensive to collect, requiring hundreds of hours netting individual bees and expert plant and bee species identification. One alternative to large, intensive data sets is smaller, local data sets composed of coarse plant-pollinator interaction data, e.g. records of bumblebee (*Bombus*) species visitation to plant species, or genus-level identification of other bees. Such smaller data sets are easier and cheaper to build and

may be collected by land managers or citizen scientists. To evaluate if smaller, less-intensive data sets could substitute for larger more-intensive data collection in seed optimization models, we compare the seed mixes created by an existing optimization model from smaller data sets versus a large data set. We compare how much of the total bee diversity (number of species) in the large, more intensive data set is supported by seed mixes optimized from the smaller data sets.

## **Development and validation of a bumble bee adult chronic oral test**

**A. R. Cabrera<sup>1</sup>, N. Exeler<sup>2</sup>, N. Hanewald<sup>3</sup>, A. Zicot<sup>4</sup>, E. Soler<sup>5</sup>, A. Kling<sup>6</sup>, S. Vinall<sup>7</sup>, K. Dressler<sup>8</sup>, V. Tänzler<sup>9</sup>, S. Kimmel<sup>10</sup>, D. M. Lehmann<sup>11</sup>, M. Patnaude<sup>12</sup>**

The regulation of pesticide uses is based on the local Risk Assessment frameworks, including a specific framework for pollinators. These frameworks rely on data from honey bee toxicity in a three-tiered process, from laboratory to semi-field to field settings, and exposure estimates based on application rates or refined via residue levels in nectar and pollen. In recent years, concerns about the risk to other bees such as bumble bees have been the driver for the development of new methods to address toxicity and exposure with selected surrogate species. Here, we present the results from the second international ring test for a bumble bee adult chronic oral test. Nine European laboratories conducted the 10-d test with *Bombus terrestris* workers while 3 US laboratories conducted the test with *B. impatiens*. Along with biological observations and consumption measurements, the stock solutions and feeding diets were confirmed for the concentration of dimethoate. There were 5 and 7 dimethoate test levels for the European and US ring test, respectively. The LC<sub>50</sub> endpoints derived from this test were on average 0.468 and 0.258 mg a.s./kg of diet for *B. terrestris* and *B. impatiens*, respectively. Similarly, the LDD<sub>50</sub> endpoints derived from the test were on average 0.093 and 0.032 µg a.s./bee/d for *B. terrestris* and *B. impatiens*, respectively. Our results indicate the test design is robust and replicable, and after a two-year effort, a validation report is in preparation to initiate the process to develop it into an OECD Guideline document.

Disclaimer: This presentation does not represent U.S. EPA Policy

## **Genotoxicity assessment of agrochemicals on honey bee spermatozoa using the tunel assay**

**Claire Champion, Heather North, Arun Rajamohan, Julia Bowsher**

Agrochemicals and their widespread use are among the suspected reasons for pollinator decline. Some evidence suggests that pesticides can act as contraceptives. Few studies have investigated the sublethal effects of agrochemicals on spermatozoa. Of special concern is whether agrochemicals impact drone sperm quality, in terms of genotoxicity. Spermatozoa serve as an unintentional biomarker for xenobiotics in the environment, making them an accessible way to measure reproductive impacts of agrochemicals found in the hive environment. It is unclear how honey and bee bread made from contaminated resources might affect hive members, such as drones, who

don't actively forage. Here we use terminal deoxyribonucleotidyl transferase (TDT)-mediated dUTP nick end labeling (TUNEL) assay as a measure of genotoxicity for spermatozoa. Honey bee spermatozoa were exposed to the agrochemicals Imidacloprid, Thiamethoxam, Clothianidin and Glyphosate. Spermatozoa were pooled from multiple drones. Each treatment consisted of a cell concentration of  $5 \times 10^6$  cells/ml in semen buffer. Semen buffer was made with a 10nM concentration of each agrochemical. Preliminary results suggest this is an accurate and reliable assay to measure any DNA damage agrochemicals have on spermatozoa. Thus, the purpose of this study is to assess the reproductive impacts of agrochemicals in drones, and the role this effect has on colony health.

## **Educating the public about bees: the Häagen-Dazs Honey Bee Haven**

**Christine Casey<sup>1</sup> and Elina Niño<sup>1</sup>**

The Häagen-Dazs Honey Bee Haven is a unique outdoor museum and garden dedicated to bee pollinators and the plants that support them. Located on the UC Davis campus and managed by the Department of Entomology and Nematology, the Haven is staffed by bee biologists and open daily to visitors at no charge. Numerous educational programs and social media outreach are used to inform the public about bee pollinators and the role of appropriately-designed gardens in promoting bee health. Garden scientists also conduct research on the interaction of bees and ornamental plants and that information is used to inform plant curation. The garden was installed in 2009 and an education program was initiated in 2013. Outreach is primarily to the general public, but specific training for beekeepers, Master Gardener volunteers, and teachers has also been conducted. Visitor contact in organized programs has increased steadily from 292 in 2013 to 3726 in 2018. We have also documented increased interest in growing and consuming bee-pollinated California specialty crops and improved knowledge of crop pollination in pre- and post-tests. Types of programming and the importance of garden-based learning about bee pollinators will be discussed.

## **Floral foraging traits impact pollinator susceptibility to pesticides and parasites**

**Hamutahl Cohen<sup>1</sup>, Marilia Gaiarsa<sup>1</sup>, Hollis Woodard<sup>2</sup>, Quinn McFrederick<sup>2,3</sup>, Lauren Ponisio<sup>1</sup>**

Bees have evolved mutualistic associations with plants, but flowers also expose bees to parasites and pesticides. It is unknown how floral foraging traits influence bee decline. We collected 11 species of wild bees (n=866) from sunflower farms in Yolo County. We extracted DNA from each sample and tested each bee for *Crithidia* spp., *Nosema* spp., *Apicystis* spp., *Aspergillus* spp., and *Ascospaera* spp. We analyzed pesticide residue representative samples from each species. We examined the composition of bacteria in the gut microbiome of each specimen and the plant species present in the pollen using Illumina Miseq sequencing. Molecular analysis for this project is ongoing. Preliminary data indicates that bees vary in their parasite

communities (e.g. 20.32% of all specimens tested positive for *Aspergillus* fungus (n=310) but *Svastra obliqua* significantly hosts more *Aspergillus* than other bees ( $p < 0.01$ ). To understand how flowers shape bee-microbe and bee-parasite interactions, we will use a network approach to model interaction patterns between bees, their plant partners, their parasites, and their microbes. We will examine how foraging generalization, centrality, and niche overlap influence parasite load using linear models. To examine how microbiome and parasite community composition are influenced by network characteristics, we use simple Mantel tests to compare dissimilarity correlations between the following: plant-parasite communities, microbe-parasite communities, and plant-microbe communities. Theory predicts that generalist species are less vulnerable to the extinctions of their mutualistic partners. Our work may complicate this theoretical assumption if generalists also host more parasites.

## **Dissecting the physiology of the nurse worker stress response**

**Vanessa Corby-Harris<sup>1</sup>, Lucy Snyder<sup>1</sup>, Charlotte Meador<sup>1</sup>, Megan Elizabeth Deeter<sup>1,2</sup>**

Nurse worker honey bees are essential to colony health because they nourish developing larvae, other worker bees, and queens through hypopharyngeal gland (HG) secretions. Stressors such as infection, pesticides, and poor nutrition cause nurse bees to have small HGs. Our research explores the physiological pathways that control this nurse stress response. Here, we discuss the mechanistic links that we find between fat body lipolysis, hormone synthesis, and autophagic HG degradation. We also discuss recent experiments that test the role of the neurohormone octopamine in the nurse stress response. As we develop a working physiological model to explain the links between stress and nurse worker health and function, we discuss how this information can be used to evaluate and improve colony health in a changing landscape. In particular, we consider synergism between stressors and the role that nutrition can play in mitigating the negative effects of environmental stress.

## **Calling all bee scientists: data needed to conserve native pollinators**

**Tara Cornelisse<sup>1</sup>**

Globally, we are experiencing a documented insect decline, including loss of pollinators. The primary cause of pollinator decline is a combination of habitat loss and agricultural intensification, leading to habitat degradation. Honey bees (*Apis mellifera*) have been the focus of pollinator loss, stemming from colony collapse disorder driven in part by increased use of neonicotinoid pesticides. These same pesticides also impact native bees, the threat of which partially caused the listing of the Rusty Patched Bumble Bee (*Bombus affinis*) as an endangered species. While honey and bumble bees are important pollinators, native, solitary bees greatly contribute to pollination services, both in agricultural fields and in natural ecosystems, including many oligolectic species propagating rare plants. Despite known threats and

importance of native, solitary bees, lacking are data sufficient enough to document clear population declines, range contractions, and/or ongoing and imminent threats; the criteria needed to list a species under the Endangered Species Act (ESA). I will present specific criteria required for an ESA listing, with examples of data needed for a successful listing of a rare bee species, including the Mojave poppy bee (*Perdita meconis*) and the Gulf Coast solitary bee (*Hesperapis oraria*), in effort to stimulate conversations and collaborations with bee (or other pollinator) scientists and natural history curators to determine species with sufficient data for conservation action and/or inspire new projects that will lead to collection of required data. With basic data, we can utilize our most effective US conservation law to conserve native bees.

## **Nectar inhabiting microbes induce pollen germination**

Megan Christensen[1], Ivan Munkres[1], Tory Hendry[2], Rachel Vannette[1]

Despite the importance of pollen in the diets of many organisms, the mechanisms underlying nutrient acquisition from pollen remains poorly understood. For most organisms that use pollen, the outer pollen wall (exine) is not mechanically broken nor enzymatically digested while passing through the digestive tract, but nutrients are still efficiently acquired, suggesting a more indirect method of exposing the more readily digestible intine layer as a means of obtaining the nutrients within. Among organisms that likely extract nutrients from pollen are the microbes found on flowers and also in bee provisions. Bacteria from the genus *Acinetobacter* are commonly isolated from both flowers and pollinators. Experiments performed with *Acinetobacter* strains revealed that incubating pollen with certain strains induces rapid pollen germination and subsequent bursting of the pollen tube. Both microbes alone and their cell-free supernatants alone produced these results, implicating an extracellular metabolite. Since nectar and pollen are mixed as bees forage as well as during offspring provisioning, we hypothesize that these microbes and their influence on pollen physiology may influence bee nutrient acquisition from pollen or shed light on the mechanisms by which pollen nutrients can be extracted.

## **Insights in the understanding of factors underlying the structure of plant-pollinator networks in tropical forests in Colombia**

**Sandra E. Cuartas-Hernández\***, Laura Gómez-Murillo, Jorge A. Rincón-Flórez

The underlying mechanisms determining the organization of mutualistic interactions are among the active questions in ecology. Here, we evaluated the variation in the topology of flower-visitor-plant networks in mountain and lowland tropical forest along time. In the mountain forest we also evaluated the effect of elevation on network structure. We recorded all flowering plants in the understory and their flower-visitors

in eight transects from 2200-2900 masl during eight months in a mountain forest, and in four transects during 12 months in the lowland tropical forest. The number of plant and flower-visitor species, their level of specialization, connectance and nestedness was estimated. In general, networks were small and showed high connectance. However, nestedness was intermediate in tropical networks, while it was very low in mountain networks. Precipitation had effect on matrix size and connectance in both ecosystems. The majority of plant and insect species were specialists and the identity of links showed a high turnover over months and transects. The partition of the whole system in small networks allowed us to detect patterns of interaction contrasting with those described for temperate cumulative networks, suggesting that the network structure is contingent to the spatial and temporal scale where the study is performed.

## **Does host lifestyle, genetics, and/or bacterial warfare impact the composition of the honey bee gut microbiome community?**

**Megan Damico<sup>1</sup>, Kasie Raymann<sup>1</sup>**

Honey bee health and populations have steadily declined due to environmental stressors such as increased pesticide use, habitat fragmentation, and novel pests and diseases. Recent work has shown the honey bee gut microbiome to be essential in aiding to immunity, digestion, behavior, and pathogen protection. However, little work has defined which factors shape the structure of a microbiome. The honey bee is a great model system to address this question because the species-level composition of the bee gut is well characterized and conserved amongst all honey bees globally. Individual bees possess multiple strains of each gut species which have been shown to display different functional capabilities. The highly conserved 16S RNA gene is commonly used for amplicon-based metagenomic surveys of microbiome community composition; however, it is not suited for capturing strain-level diversity. To satisfy this, we use a technique, metagenomic multilocus strain typing (MMST), that allows for the detection of strain variants within a species. Our preliminary studies using MMST revealed that honey bees from single colonies rarely shared the same strain-profiles (compositions of strains), yet when compared to honey bees from different geographic locations they presented nearly identical strain profiles. Here we aim to elucidate the factors that shape the community composition and structure of the honey bee gut microbiome by testing the impact of host genetic background and lifestyle as well as bacterial competition/incompatibility. Since different strains employ various metabolisms and pathogen susceptibilities, strain-profile differences may contribute to honey bee health.

## The Empire State Native Pollinator Survey – determining the conservation status of NY pollinators

Bryan Danforth (1), Maria Van Dyke (1), Erin White (2), Jeffrey Corser (2), Matthew Schlesinger (2)

A number of U.S. states have recently developed native pollinator survey programs. New York, in particular, has developing an ambitious, three-year pollinator survey program to assess the status of native bees and other pollinating insect groups including flies, beetles, and moths. The project, funded by the New York State Department of Environmental Conservation, is coordinated by the New York Natural Heritage Program with key partners including Cornell University, SUNY ESF, SUNY Cobleskill, and the New York State Museum. The project involves a number of survey approaches including (1) targeted surveys across natural areas in New York, (2) surveys focused on particular habitats (coastal sand dunes, barrens, peatlands, high elevation alpine sites, and late successional forests), and (3) surveys focused on target (focal) taxa that are considered to be in decline or at risk (*Andrena*, *Calliopsis*, *Bombus*, *Melitta*, *Macropis*, *Melissodes*, *Osmia*, and *Megachile*). Comparisons to historical collection records will help document trends in pollinator abundance, geographic distribution, and phenology. We will present an overview of the project and summarize results obtained to date on bees. A full description of the project and survey protocols can found at: <http://www.nynhp.org/pollinators>.

## The impact of viruses on honey bees at the individual and cellular levels

Katie F. Daughenbaugh<sup>1,4</sup>, Alex McMenamin<sup>1,2,4</sup>, Laura Brutscher<sup>4</sup>, Fenali Parekh<sup>2,4</sup>, and Michelle Flenniken<sup>\*1,4</sup>

Honey bee colony losses are influenced by multiple abiotic and biotic factors, including viruses. To investigate the effects of RNA viruses on honey bee health, we infected bees with a model virus (Sindbis-GFP) in the presence or absence of dsRNA. In honey bees, dsRNA is the substrate for sequence-specific RNAi-mediated antiviral defense and is a trigger of sequence-independent antiviral responses. Transcriptome sequencing identified more than 200 differentially expressed genes, including genes in the RNAi and heat shock response pathways, and many uncharacterized genes. To confirm the virus limiting role of two genes (*dicer* and *MF116383*), we utilized RNAi to reduce their expression in vivo and determined that virus abundance increased, supporting their involvement in antiviral defense. To evaluate the role of the heat shock stress response in antiviral defense, bees were heat stressed post-virus infection and virus abundance and gene expression were assessed. Heat stressed honey bees had reduced levels of virus infection compared to controls and the expression of one small heat shock protein (*Hsp*) (*protein-lethal(2)essential for life-like*) was increased. To determine if these genes are universally associated with antiviral defense, honey bees were infected with additional viruses and gene expression was assessed. In addition, the function of key genes was assessed in primary honey bee larval hemocytes that were transfected with dsRNA or infected with Lake Sinai virus 2. Together these studies indicate that *MF116383* and *Hsps* mediate dsRNA detection, that *MF116383* is involved in limiting LSV2 infection, and further our mechanistic understanding of honey bee antiviral defense.

## Pollinator disease transmission dynamics: effects of a common bee parasite on a hoverfly (Diptera: Syrphidae) host

Abby Davis, Kaitlin Deutsch, and Scott McArt

Infectious diseases heavily influence global pollinator decline, threatening the ecosystem services these insects provide. Hoverflies (Diptera: Syrphidae) are frequent floral visitors, and floral resources have been shown to be important platforms of disease. Despite evidence that hoverflies carry bee parasites, little is known about the role hoverflies play in pollinator disease transmission dynamics. *Crithidia* is an important fecal-orally transmitted parasite of bumble bees, impairing worker cognition and reducing foraging efficiency. It is important to understand how hoverflies may be impacted by this parasite, and how hoverflies may vector this parasite in pollinator communities. Using the black-shouldered drone fly (*Eristalis dimidiata*) and a bumble bee trypanosomatid gut parasite (*Crithidia bombi*), we investigated *Crithidia* replication and viability in *Eristalis*. Wild-caught hoverflies were inoculated with *Crithidia*, and the fly fecal events were screened for 10 days post-inoculation to assess *Crithidia* abundance and parasite motility. This study adds to our overall knowledge of pollinator health, while advancing our understanding of non-bee disease transmission dynamics.

## Effects of native ungulate herbivory on native bees in a Pacific Northwest forested riparian area

Sandra J. DeBano<sup>1</sup>, Mary M. Rowland<sup>2</sup>, Samantha Roof<sup>1</sup>, and Skyler Burrows<sup>3</sup>

Native bees are a diverse and functionally important group of pollinators in riparian areas of the Pacific Northwest. As in much of the US, these riparian areas have been impacted by a variety of disturbances, including logging, stream channelization, exotic weed invasions, and livestock and native ungulate grazing. While some studies have examined livestock grazing effects on native bees, little attention has been directed at understanding how herbivory by native ungulates, such as deer and elk, influences native bees. To address this question, we conducted a three-year manipulative study at 12 riparian sites on Meadow Creek in the Starkey Experimental Forest and Range in eastern Oregon. Half of the sites were excluded from deer and elk herbivory and half were open to grazing by deer and elk. Native bees were sampled using pan traps and vane traps three to four times each year. Blooming forb and shrub availability was also estimated at each site. Some common flowering forb species that are preferred forage for elk were less common in sites grazed by native ungulates. Although bee abundance did not vary between grazed and ungrazed sites, bee species richness was higher in ungrazed sites. However, we found no statistically significant differences in community composition between grazed and ungrazed sites. These results suggest that pollinator-friendly riparian management plans in the Pacific Northwest should consider not only effects of domestic livestock, but also levels of native ungulate herbivory.

## **Mite migration and increasing deformed wing virus levels in honey bee colonies in the fall**

**Gloria Degrandi-Hoffman**

Mite populations increase sharply in the fall from the migration of mites into colonies on foragers. If mite migration causes Varroa populations to grow rapidly, do levels of viruses transmitted by Varroa also increase resulting in colony loss over the winter? Can this fate be avoided by optimizing nutrition? To address these questions, we conducted an experiment establishing colonies with low mite numbers during the summer. Half of the colonies were fed pollen throughout the study and the half were nutritionally stressed. Colony size, vitellogenin levels, mite numbers, virus titers, and the frequency of capturing foragers with mites at colony entrances were recorded from July to December. Our path analysis indicated that Deformed Wing Virus (DWV) titers were significantly correlated to phoretic mite populations and infestation levels in cells both of which were correlated to the frequency of capturing foragers with mites (FWM). Levels of DWV were unaffected by supplemental feeding, possibly because FWM entered fed and unfed colonies at similar rates and transmitted both mites and DWV. Our study demonstrates difficulties in controlling both Varroa and virus levels in colonies especially in the fall.

## **Effects of a common fungicide on olfactory associative learning in honey bees**

**Nicole DesJardins, Adrian Fisher, Jon Harrison, Brian Smith**

Sublethal insecticide levels can significantly impair learning in honey bees. While insecticides directly affect the insect nervous system, fungicides target the basic biochemical processes of fungal cells. For example, a commonly-used fungicide, Pristine®, inhibits fungal cellular respiration. Honey bees are often exposed while foraging on crops treated with fungicides. Previous work on bees exposed to Pristine® has shown that it may inhibit mitochondrial function in the gut. Proboscis extension reflex (PER) is a learning assay that trains bees to associate a sucrose reward with an odor. Through its effects on feeding, Pristine® could affect PER learning directly by altering the ability of bees to taste their food, or indirectly through effects on metabolic physiology. We sampled bees from colonies exposed to varying levels of Pristine® and used PER to assess their learning. We found that bees exposed to progressively higher doses performed worse in PER assays. Although Pristine® does not directly target the nervous system and has been assumed to be safe for pollinators, our study provides evidence of negative sublethal effects. Olfactory learning is important for foraging performance; if Pristine® impairs this, it could affect foraging behavior and put colony health at risk.

## **The effect of landscape context on hoverfly communities**

**Kaitlin R. Deutsch<sup>1\*</sup>, Mahilet Kebede<sup>1</sup>, Aaron Iverson<sup>1</sup>, Paige A. Muñoz<sup>1</sup>, and Scott H. McArt<sup>1</sup>**

Global declines of insect pollinators have been extensively documented in the past few decades. An important driver of decline is the loss of natural habitat via landscape simplification. In this study, we investigated how landscape context affected the abundance, richness, and community composition of hoverfly communities. We conducted surveys at 31 agricultural, suburban, and natural sites, each with variable landscape composition. Hoverfly abundance and richness increased along a gradient of increasing agricultural area in the surrounding landscape, with higher richness attributable to an abundance-driven accumulation of species. However, richness was low overall. We also found a positive effect of increasing developed area on hoverfly abundance but not richness. Interestingly, hoverfly species composition differed between agricultural, natural, and suburban landscape types, suggesting some species may be better adapted to certain human-modified habitats but not others. Our results indicate hoverflies are robust to human-modified landscapes, potentially providing pollination insurance in the face of ongoing bee declines.

## **Eating microbes make for better bees**

**Prarthana Dharampal<sup>1</sup>, Shawn Steffan<sup>1,2</sup>, Cameron Currie<sup>3</sup>, Caitlin Carlson<sup>3</sup>**

Long considered as strict pollenivores, recent findings reveal that bees rely on pollen-associated microbes as a major dietary resource. However, little is known about the impact of microbial prey on bee fitness. Here, we examine the effects of microbe-deficient diets on bee health using in vitro reared larval mason bees. In a series of diet manipulations, microbe-rich maternally-collected pollen provisions were replaced with increasing fractions of sterilized microbe-deficient pollen before being fed to developing larvae. Convergent findings from amino acid and fatty acid trophic biomarker assays revealed that larvae derived a substantial amount of nutrition from microbial prey and occupied a significantly higher trophic position than that of strict herbivores. Larvae feeding on increasingly sterile diets experienced significant adverse effects on growth rates, biomass, and survivorship. When completely deprived of pollen-borne microbes, bee larvae consistently exhibited marked decline in fitness. Further analysis using maternal and non-maternal pollen revealed that larval health was significantly higher when reared on microbe-rich pollen, regardless of pollen source and nutritional quality. We conclude that pollen-associated microbes are central to bee health, not only as nutritional mutualists, but also as a major dietary component. In an era of global bee decline, the conservation of such bee-microbe mutualisms may represent an important facet of pollinator protection strategies.

## Identifying trends and gaps in pollinator health in Washington, D.C. through data mining

Abigail Dias<sup>1</sup>, Jesse Meiller<sup>1</sup>

Pollinator health is increasing in concern nationwide. There is a great need for quantitative measures in order to understand trends and contributing factors to pollinator health. While some regions have sufficient data records, there is a significant lack of publicly available quantitative data on pollinator health in Washington, D.C. This study analyzes quantitative data from D.C., and the nearby states of Maryland and Virginia, in order to identify possible trends and issues affecting pollinators in D.C. Analysis was conducted on publicly available data from the USDA to determine trends in colony loss, as well as trends in factors such as parasite and disease spread, pesticide use and temperature. It was found that there was a correlation between trends in Maryland and Virginia, suggesting that there are regional factors affecting both Maryland and Virginia that could also be of concern for Washington, D.C. However, in analyzing the small amount of data available for D.C. itself, there were some differences in trends that suggest D.C. faces its own unique threats to pollinator health. More studies are needed in DC in these areas of pollinator health.

## Tritrophic interactions and monarch larval success in the Great Basin, USA

Aramee C. Diethelm<sup>1</sup>, Cassidy Gosse<sup>1</sup>, Elizabeth G. Pringle<sup>1</sup>

Western monarch butterfly (*Danaus plexippus*) populations are declining, but the causes of this population decrease remain unclear. Possible causes include climate change, the loss of summer breeding habitat, and increased pesticide use, however the influence of tritrophic interactions has not yet been thoroughly examined. Food-plant species identity can be important to specialist herbivores because plants influence trophic interactions by affecting both larval development and predator recruitment. To investigate how milkweed (*Asclepias* spp.) species identity affects the development and survival of monarch larvae, two species of western milkweed (*A. fascicularis* and *A. speciosa*) were grown in a common garden in Reno, NV, during the 2018 breeding season. In September, second-instar monarch larvae (n=340) were randomly assigned to factorial combinations of the two milkweed species and either predator exclusion cages or mock controls. Larvae were monitored every two days until they reached the fourth instar, at which point all larvae were enclosed to complete development. Larvae took longer to develop and were less likely to survive on *A. speciosa* than on *A. fascicularis*, and adult weight and wing size were also smaller among individuals that ate *A. speciosa*. Interestingly, the reduced survival of monarch larvae on *A. speciosa* appeared to result in part from a higher probability of predation on this species than on *A. fascicularis*. These results suggest that monarch fitness during the breeding season is influenced by tritrophic interactions, and thus that the species of milkweed being used for habitat restoration should be considered in conservation planning.

## Exploring regional variation in blue orchard bee phenology, behavior, and reproductive success

Morgan Dunn<sup>1</sup>, Theresa Pitts-Singer<sup>2</sup>, Diane Alston<sup>3</sup>, Steve Peterson<sup>4</sup>

Orchard crops can be pollinator-dependent, and consequently, yields may suffer due to self-incompatible flowers, inclement spring weather conditions for bee flight, and a low availability of strong honey bee colonies. The blue orchard bee (BOB), *Osmia lignaria*, a solitary bee native to the U.S., is well-suited to management for pollination of rosaceous fruit and tree nut crops. BOBs are commercially available from “bee-ranchers”, who wild-trap, propagate, and sell to growers on a per female basis. However, BOBs show regional differences in development and phenology such that the population source can affect reproductive success and synchrony of emergence with crop bloom. For this fledgling industry to succeed and compete with commercial honey beekeepers, more knowledge about regional variation in BOBs is needed to develop best management practices and maximize population returns. For my graduate research, I am performing three experiments to further investigate regional differences in BOB behavior, phenology, and reproductive success. One experiment will determine consequences of pairing BOBs from regionally-distinct locations on reproduction, progeny development, and progeny performance by monitoring nesting behavior and reproductive output of first-generation bees from California and Utah in a controlled experiment. Another experiment will determine variation in developmental phenology of BOBs from three cherry-growing regions (California, Washington, and Utah) by maintaining these regionally-specific bees under controlled conditions. Finally, I am also determining whether retention of BOB females differ if they are sourced from California and Utah and used as pollinators in cherry orchards outside of their geographic origin by examining dispersal.

## The role of commercial ornamental plant varieties in supporting pollinator populations

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One of the factors underpinning pollinator declines is the reduction in the diversity and abundance of flowering plant species. In urban and suburban areas, ornamental plant varieties are often planted to mitigate these declines and provide foraging resources. However, their role in supporting pollinator biodiversity is not well established, and most studies have been conducted in landscapes with simplified pollinator communities. We observed pollinator visitation patterns to 5 ornamental annual and perennial plant genera and their cultivars over multiple years at two semi-natural sites in Pennsylvania to understand their potential for supporting diverse pollinator communities. From our preliminary studies, we found significant variation among cultivars in visitor abundance, with many cultivars varying in attractiveness based on time and year. We observed only polylectic species visiting ornamentals, despite the presence of oligolectic species in the background. We conclude that the utility of ornamental plants depends on environmental context: while their role in supporting a complex pollinator community is limited, they may provide long-lasting supplemental

foraging resources in urban and suburban environments. In future studies we will determine the relative influence of floral visual and chemical advertisement and nutritional reward on mediating patterns of pollinator attraction to these cultivated varieties. This research will (1) evaluate the potential of ornamental plants in supporting complex pollinator communities (2) determine which plant features are most critical for shaping plant-pollinator interactions (3) allow growers to develop and adapt production practices to incorporate pollinator health into breeding and production practices. This project is a collaboration between Penn State, the Pollinator Partnership, AmericanHort, the American Honey Producers Association, and the American Seed Trade Association, and is supported by funding from the Horticultural Research Institute and USDA-APHIS.

### **Does habitat quality ‘dilute’ pollinator disease risk instead of biodiversity? ‘Habitat health’, an alternate explanation for reduced pathogen prevalence in species-rich pollinator communities**

**Michelle L. Fearon<sup>1</sup>, Maryellen C. Zbrozek<sup>1</sup>, and Elizabeth A. Tibbetts<sup>1</sup>**

Growing evidence suggests that host biodiversity is linked with reduced disease risk in many diverse host-pathogen systems. These findings are commonly explained as the “dilution effect”, where species-rich communities have reduced disease risk due to reduced encounters with infected individuals or rates of transmission during species interactions. Here, we propose and test an alternative hypothesis to explain observed dilution effect patterns called “habitat health”. The habitat health mechanism proposes that areas with higher habitat quality promote greater host biodiversity and provide higher quality resources to hosts, which may allow for improved resistance to infection and stronger immune function. Therefore, we hypothesize that bees from species-rich communities may be healthier and less susceptible than individuals from species-poor communities due to better resources from the surrounding habitat. We tested the habitat health mechanism in pollinator communities infected with three viruses along a habitat gradient, and compared virus prevalence in four pollinator host species. We found that pollinators had significantly reduced virus prevalence in communities with greater proportions of natural habitat nearby and pollinator biodiversity was positively correlated with surrounding natural habitat. These results support habitat health as an important, alternative mechanism to the dilution effect and suggests that further work will be critical to tease apart how habitat quality and host biodiversity interact to influence disease risk. A better understanding of these links between habitat, biodiversity, and infectious disease could lead to additional promising management strategies that will simultaneously preserve habitats, conserve species, and reduce disease risk among humans and wildlife.

### **Mechanisms mediating bee pathogen transmission: deposition, persistence and acquisition on flowers**

**Laura L. Figueroa<sup>1\*</sup>, Malcolm Blinder<sup>2</sup>, Cali Grincavitch<sup>2</sup>, Angus Jelinek<sup>2</sup>, Emilia Mann<sup>2</sup>, Liam Merva<sup>2</sup>, Lucy Metz<sup>2</sup>, Amy Zhao<sup>2</sup>, Rebecca E. Irwin<sup>3</sup>, Scott H. McArt<sup>1</sup> and Lynn S. Adler<sup>2</sup>**

Reports of global bee decline have been linked to pathogen pressure, including potential spillover from managed colonies. Bee pathogen transmission through shared use of floral resources has been demonstrated in experimental settings, and widespread occurrence of bee pathogens on wildflowers in nature is beginning to be revealed. However, mechanisms mediating bee pathogen transmission remain largely unknown, thus impeding the development of effective conservation strategies. Here, we set out to expand the understanding of horizontal pathogen transmission through floral resources using *Bombus impatiens* and *Crithidia bombi* as the model system. Specifically, we evaluated multiple mechanisms that were hypothesized to contribute to bee disease transmission through shared use of flowers. The trials were conducted using *Monarda didyma*, *Lobelia siphilitica*, and *Lythrum salicaria*. We found that patterns of pathogen deposition, persistence and acquisition varied across plant species and floral locations. We discuss implications for disease spread in bee communities and applications for pollinator conservation.

### **Nectar changes the ecological costs of defended pollen**

**Jacob S. Francis<sup>1</sup>, Cheyenne R. Acevedo<sup>2</sup>, Felicity Muth<sup>3</sup>, Anne S. Leonard<sup>1</sup>**

Pollinators forage in a multidimensional floral marketplace: rather than collecting a single resource, floral visitors often select among flowers that differ in the composition, quality, and chemistry of multiple rewards. Though the effects of nectar chemistry are relatively well studied, the functional role of pollen chemistry in plant-pollinator interactions is surprisingly unclear. This bias is especially notable considering that pollen has a high concentration and diversity of secondary metabolites. We asked two questions about the consequences of chemically defended pollen: 1) Do bees discriminate against flowers with highly defended pollen, and 2) Can flowers overcome this discrimination by providing high quality nectar? In lab-based foraging assays we measured the preferences of individual bumblebees (*Bombus impatiens*) for two “species” of artificial flowers. Across four treatments we manipulated the amount of a defensive alkaloid in pollen alongside the quality and/or presence of nectar. We found that bees developed a strong aversion to flowers with chemically defended pollen, but this response depended on nectar context. When co-flowering species offered identical low-quality nectar rewards, bees preferred flowers with less defended pollen. Alternatively, when flowers with highly defended pollen offered higher quality nectar than co-flowering competitors, bees preferred them. Finally, we found that bees spent less time on arrays where one species offered defended pollen, raising the possibility that the costs of defended pollen might be shared by neighboring plants. These findings are a first step at understanding how recently-revealed patterns in floral reward chemistry impact bee foraging and possibly plant-plant competition for pollinators.

## Assessing the vulnerability of specialty crops to pollinator decline in Wisconsin (USA)

Hannah R. Gaines-Day, Claudio Gratton

Over a third of crop plants require insect pollination to produce fruit and, in recent years, the hectares of pollinator-dependent crops have increased worldwide. At the same time, a global decline in bees, the most important pollinators, has led to concern about the future of crop production. But for many crops, the extent to which managed and wild bee declines could result in yield losses is not entirely clear. For example, even crops that require insect pollination, such as apple, don't need 100 % fruit set to produce an economically viable yield. Furthermore, farms located in a diverse landscape may receive the majority of pollination from wild bees, thereby buffering them from losses in managed bees. These same farms, however, would be vulnerable to declines in wild bees. Therefore, our primary objective was to understand the relative sensitivity of specialty crops to honey bee and wild bee declines, how this varies across crops, and where in the state of Wisconsin loss of wild or managed bees is going to have the greatest effects on crop production. We combined grower surveys, literature reviews, land cover mapping, and results from previous studies in our lab to assess pollinator supply and demand across the state. The results of this synthesis will identify crops and regions of the state most susceptible to pollinator decline and highlight areas where conservation efforts may be most important.

## Evaluating intensively managed conifer forests of the Pacific Northwest as habitat for pollinators

Sara M. Galbraith<sup>1</sup>, Andrew R. Moldenke<sup>2</sup>, Matthew G. Betts<sup>1</sup>, James W. Rivers<sup>1</sup>

A large and growing proportion of global forest area is managed for resource production, but very little is known about whether and how intensively managed conifer forest provides habitat for animal pollinators. In this study, we evaluated how pollinator communities are influenced by stand age within an intensively managed forest landscape. We hypothesized that pollinator diversity and bee reproductive output would be greatest in the years following harvest, then decline as stands move towards canopy closure, reducing flowering plant abundance. We sampled pollinators and quantified habitat in managed conifer stands of varying ages within the Oregon Coast Range from May-September 2018. Stands were sampled from four different landowners to maximize the diversity of management styles. We also monitored *Osmia lignaria* reproductive output from nesting blocks placed in each stand with a standardized number of adult cocoons. We collected >12,000 specimens representing four orders of pollinating insects and counted >105,000 flowers. As predicted, pollinator and flower abundance were greatest in younger stands, declining 3 years following harvest. Insect communities shifted along the age gradient, with relatively more bees collected in young stands and relatively more flies collected in older stands. In addition, >1/4 of nesting chambers within *O. lignaria* nesting blocks were occupied by the end of the season, and more nest chambers occupied in stands <9 years old. Our research reveals the importance of early seral forest for supporting pollinator habitat and highlights that efforts to promote pollinator habitat in managed forest should be focused on early successional forest conditions.

## Determining monarch (*Danaus plexippus*) natal site distribution in Nevada using stable isotope analysis and wing morphometrics

Cassidy Gosse<sup>1</sup>, Aramee Diethelm<sup>1</sup>, Elizabeth G. Pringle<sup>1</sup>

Monarch butterfly (*Danaus plexippus*) populations have been significantly declining west of the Rockies over the last 30 years. Site-specific information for Western monarchs outside of the overwintering areas in California is largely unavailable, yet this information is critical for understanding monarch population biology and movement. Patterns in deuterium and carbon isotopic ratios produced by local precipitation can be linked to monarchs via their *Asclepias* milkweed host plants, and wing morphometrics may indicate flight abilities. To characterize the movement of monarch butterflies during the summer breeding season and determine larval host plant preference, we analyzed 16 wild-caught monarchs and 150 milkweeds from across a precipitation gradient in Northern Nevada for deuterium (monarchs only) and carbon (monarchs and plants) isotopes. We also characterized variation in wing aspect and wing roundness, components of wing morphology that are suggested to affect the efficiency of gliding flight. Deuterium values were variable among monarch wings and were positively correlated with precipitation, whereas monarch wing measurements showed reductions in both wing aspect (length/width) and wing roundness over the season. There were no geographic or seasonal patterns in wing  $\delta^{13}\text{C}$ , but wing roundness was negatively correlated with its  $\delta^{13}\text{C}$  content. Our results suggest that Northern Nevada contains multiple natal ground regions, and that wing shape changes over the breeding season and may depend on the water relations of the food plant. Determining the geographic distribution of Western monarch natal sites and their consequences for successful migration will be essential for developing effective conservation strategies for this threatened species.

## Pesticide exposure for bees during blueberry bloom, and strategies for mitigating risk

Kelsey K. Graham, Meghan Milbrath, Philip Fanning, and Rufus Isaacs

Blueberry growers must manage insects and diseases that have direct effects on crop yields. Some pests are active during bloom when managed and wild bees are in the fields. There is therefore a critical need to balance pollinator safety with pest suppression, but first we need to better understand exposure during this potentially precarious time. In 2018, we quantified the presence and abundance of pesticides in bee collected pollen from honey bees and bumble bees placed in Michigan blueberry farms. Residue levels were compared between fields managed with conventional pesticides and farms that did not receive these sprays (organic or unmanaged). To illuminate the source of pesticide exposure, we also identified where bees were foraging through pollen identification. Bumble bees collected around 10% of their pollen, on average, from blueberry, while honey bees collected less than 2%. We detected eight pesticides in bee collected pollen, with greater exposure in bumble bee collected pollen, and at sites with conventional management. Increased exposure for bumble bees may be due to their greater fidelity to blueberry for pollen foraging.



Pesticides detected at unmanaged sites also indicates that bees are foraging outside the field where they are placed, resulting in exposure from distant fields, and highlighting the need for careful selection of pest management tools during bloom. These results will be combined with our current research on night spraying, drift reduction, and weed control to develop guidelines for reducing pesticide exposure in blueberry production, to protect yields and bee health.

## Effect of native vegetation proximity on bee diversity in alfalfa (*Medicago sativa*)

Scott VC Groom<sup>1</sup>, Jay Iwasaki<sup>1</sup>, Elisabeth Williamson<sup>1</sup> & Katja Hogendoorn<sup>1</sup>

Managed honey bees (*Apis mellifera*) meet the majority of pollination requirements in Australian agriculture. However, a considerable and typically underappreciated proportion of this requirement is often provided by unmanaged pollinators. The impending arrival of the ectoparasite *Varroa destructor* threatens much of the pollination provided by these wild populations, predominantly through the loss of feral honey bee colonies. Small colony sizes and densities of native pollinators can impede their ability to replace feral honey bees entirely but supporting strong and diverse communities likely limits the severity of pollination loss. Ensuring a healthy pollinator community requires resources for a diversity of life histories that also encourage species' presence within crops. Here we look to investigate how the proximity of native vegetation influences the abundance and diversity of unmanaged species and resulting crop yield within an alfalfa (*Medicago sativa*) seed production area of southeast South Australia. Lucerne seed is a near hundred-million dollar, pollination-dependent industry with strong ties to both livestock and dairy industries when sown for pasture. We find that the presence of native vegetation has a significant impact on the diversity of unmanaged bees within the crop, particularly when that native vegetation is integrated (i.e. scattered *Eucalyptus*). Our results demonstrate value in retaining existing vegetation for mitigating a proportion of pollination lost with feral honey bees, but also benefits to initiatives to revegetate and diversify agricultural production landscapes.

## Use of video in honey bee management

Bridget Gross<sup>1</sup>, Doug Golick<sup>1</sup>, Judy Wu-Smart<sup>1</sup>

The decline of honey bees has had significant impacts on the pollinating economy, as honey bee decline means fewer crops are pollinated, and subsequently a loss of profit for farmers. To combat colony loss, beekeepers can track colony health over the course of the season and manage them appropriately with tools such as hive scales or notes sheets. While these are traditional management and record keeping tools, we explore a novel approach of using a point - of - view video to record management techniques and perform hive inspections. Preliminary data suggests that this tool may be valuable as a hands-free way of evaluating hive health, in supplementing other management techniques, and in educating beekeepers about hive management strategies.

## Diverse plates and picky eaters: On-farm diversification in an agriculturally dominated landscape positively influences specialist pollinators

Guzman, Aidee<sup>1</sup>, Chase, Marissa<sup>1</sup>, Kremen, Claire<sup>1,2,3</sup>

Agricultural practices can either contribute to pollinator decline or provide opportunities to support pollinator communities. At the landscape-scale, agriculture can have negative impacts on pollinators, especially pollinators that specialize on limited floral or nesting resources. While increasing floral resources at the field-scale is positive for pollinator communities, little is known about how it affects specialist bees that depend on a specific pollen source (oligoleges). We studied pollinators on small-scale farms that contrasted in crop diversity (monocultures versus polycultures), embedded in the intensively managed agricultural region of the San Joaquin Valley in California, to understand how wild bee communities and specialist bees would respond to field-scale diversification practices. We used squash (*Cucurbita pepo*), which has associated oligolectic pollinators ("squash bees" in the genera *Peponapis* and *Xenoglossa*) as our focal crop. We hypothesized that a greater number of squash bees would occur on monoculture farms than polyculture farms, due to greater numbers of squash flowers on monocultures. Despite our predictions, we found that increasing the number of non-squash floral resources at the field-scale in agroecosystems supports a greater abundance of squash bees but has no effect on the diversity of bees visiting squash flowers. This pattern of increased abundance was consistent for other wild bees and the total number of bees (i.e. including honey bees), but not for honey bee abundance alone. Thus, on-farm diversification may be an important refuge for specialist bees and other pollinator species that are vulnerable in landscapes dominated by agriculture.

## Deformed wing virus induces a metabolic switch in honey bees that prevents key biochemical and genetic changes required for sustained flight

Matthew C. Heerman, Steven C. Cook, Olubukola Banmeke, Cristina Rodríguez-García, Samuel C. Ramsey, Jianghong Li, Eugene Ryabov, and Yanping Chen

Reduced flight efficacy is a preeminent phenomenon associated with asymptomatic *Deformed wing virus* (DWV) infection in the European honey bee *Apis mellifera*. While there are many factors that contribute to this occurrence, little is known about the biochemical and genetic regulation of lipids and carbohydrates with respect to pathogen infection. Our results provide a mechanism of dysfunction for biochemical, posttranslational protein modification, and genetic regulation of lipid metabolism in infected bees. In field caught honey bees displaying DWV infections greater than 10<sup>10</sup> viral genome copies, we observed depletion of their total fat stores. In response to rapid loss of fat, bees suppressed transcription of lipases *HA lipase-like*, *Uncharacterized pancreatic lipase*, and *Pancreatic TAG lipase-like*. Additionally, we

observed an increase in posttranslational phosphorylation at position threonine-358 of RAC-alpha serine/threonine protein-kinase (AKT), a key negative regulator of lipase transcription. In tandem, we observed lower transcript levels for the negative regulator of AKT, *Phosphatase and tensin*, and the lipase transcription factor *Forkhead box protein O*. Reductions in glycogen, glucose, and trehalose levels in deformed bees aside reductions in *Glycogen phosphorylase*, *Trehalose phosphate synthase*, and *Trehalose synthase* transcription were also observed, indicating a slower production of flight carbohydrates. Our results demonstrate that overt, DWV induced, consumption of lipid fuel reduces fatbody cellular outcomes. Honey bee fatbodies are the primary nutrient storage tissue. With decreased output of this cell type, honey bees are less able to convert glycogen into glucose, and finally to secreted trehalose, the energy source for honey bee flight muscles.

### **Saved by the pulse: temporal resource pulse rescues microcolony reproductive development despite differential growth patterns**

**Jeremy Hemberger<sup>1</sup>, Grant Witynski<sup>1</sup>, Agathe Frappa<sup>2</sup>, Claudio Gratton<sup>1</sup>**

Bumble bee declines have been linked to habitat loss via reduction in floral resources. Formerly flower-rich natural habitats have been supplanted by crop monocultures as agricultural lands have expanded over the last century. While some cultivated crops provide flowers for foraging bumble bees (e.g., canola, sunflower), they do so only for a short period of the growing season. These so-called "mass-flowering" events provide bumble bee colonies with an abundance of food during bloom but are typically followed by dearth periods of low to no flowers - a contrast that could have negative consequences for bumble bee colony success. To untangle the impact of total resource abundance and temporal availability on the development bumble bee colonies, we simulated agricultural landscape resource abundance in the lab, exposing microcolonies of *Bombus impatiens* to conditions that covaried total resource abundance with temporal availability. Colonies fed constant, high-amounts of diet (pollen and nectar) grew the most and produced the most drones. However, microcolonies exposed to dearth-pulse conditions were able to produce statistically equal numbers of drones, provided the total amount of food received over the experiment was equal to the constant, high-fed colonies. Our results highlight the physiological plasticity of *B. impatiens* and provides a mechanism for how *B. impatiens* is able to tolerate the resource conditions of agricultural landscapes and increase in abundance. Our results can help identify landscapes suitable to developing bumble bee colonies, while also highlighting problematic landscapes that might benefit from altered management or conservation.

### **Bees on the MAPP: establishing the Minnesota Agriculture for Pollinators Project landscape experiment**

**Christina Herron-Sweet<sup>1</sup>, Kiley Friedrich<sup>1</sup>, Katie Lee<sup>1</sup>, Elaine Evans<sup>1</sup>, Robert Koch<sup>1</sup>, Eric Lonsdorf<sup>2</sup>, Marla Spivak<sup>1</sup>, Daniel Cariveau<sup>1</sup>**

Declines in managed bee health and wild pollinator populations have prompted numerous government and non-government groups to promote and implement pollinator habitat enhancements throughout the United States. While increasing floral resources is undoubtedly important for conserving and sustaining populations of beneficial insects, key questions remain. Critical research gaps exist in how to provide effective, economically-feasible floral resources for multiple groups of pollinators and other beneficial insects. To evaluate how local and landscape factors influence the success of pollinator plantings, we recently established a landscape-scale experiment where we manipulated the plot size, seed mix, and landscape context of experimental plantings. Such experimental approaches offer a robust method to address these research gaps and are the only way to establish causal relationships, but are understandably rare in ecology because of high cost and logistical challenges. We are implementing this experiment at 38 sites in southwest Minnesota, which we planted in November-December 2018. Over the next four years, we will measure how plot size, seed mix and landscape context interact to influence, a) honey bee health and honey production, b) native bee communities and bumble bee reproductive output, and c) natural enemy communities in pollinator plantings and neighboring soybean fields. Finally, we will develop economic models that quantify the return on investment for plantings of different sizes and seed mix types across different landscape contexts. Our results will enable land managers to better prioritize placement and planning of floral plantings to generate a high return on investment, especially given increasing costs of creating pollinator habitat.

### **Disease ecology of native bees in Sacramento**

**Lauren Hisatomi<sup>1</sup>, Anjali Agarwal<sup>1</sup>, Kit T. Keane<sup>1</sup>**

Due in large part to the onset of colony collapse disorder across the United States, a panoply of parasites and diseases have been characterized from the European honey bee. However, we still have a limited understanding of the biology of most native species of bees. Recently, it has been shown that several of the pathogens present in honey bees are also present in other pollinators, and can be harbored in pollen. Thus, bee species assemblages serve as a highly compelling model for which to study both inter- and intra-species disease transmission dynamics. We are currently developing screening methods for a number of bee pathogens and applying these tools to assess the relative prevalence and distribution of pathogens across environmental gradients.

## **The Unintended Antibiotic Target: Honey Bee Reproductive Health**

**Alexis Hoopman<sup>1</sup>, Kasie Raymann<sup>1</sup>**

Research has shown that multiple factors are contributing to honey bee decline, including the implication of common hive treatments such as antibiotics and fungicides. Additionally, colony collapse has been linked to poorly mated queens and highly variable sperm quality in drones. Colonies have been replacing their queens more frequently in recent years; the replacement of a queen or any negative queen event doubles the risk of hive collapse. Antibiotics, like tetracycline, have been regularly used by commercial beekeepers for over 50 years. Tetracycline is used as a preventative treatment for bacterial brood diseases, like Foulbrood. Tetracycline has been shown to perturb the gut microbiome of honey bees, which plays an important role in metabolism, immune function, growth, and development. Tetracycline also increases pathogen susceptibility and mortality in honey bees. Some antibiotics, like tetracycline, have recently been classified as mitochondrial-endocrine disruptors. Endocrine disrupting chemicals, including tetracycline, have been found to decrease spermatogenesis and cause infertility in mammals. To our knowledge, no studies have evaluated the impacts of tetracycline on the reproductive health of bees, but these findings give urgency to the matter. In fact, our preliminary *in vitro* results indicate that even very low concentrations of tetracycline can significantly reduce honey bee sperm viability. In addition to *in vitro* studies, we are also investigating the reproductive toxicology of tetracycline in both drones and queens *in vivo*. Here we will present our most recent findings on the effects of tetracycline on honey bee reproductive health.

## **The regulation of pheromone biosynthesis by micrornas in the mandibular gland of the honey bee (*Apis mellifera*)**

**W. Cameron Jasper<sup>1</sup>, Elina L Niño<sup>1</sup>**

Using small RNA sequencing and *in silico* target prediction, I have identified multiple microRNAs which putatively regulate caste-specific pheromone biosynthesis in the honey bee (*Apis mellifera*). Statistically significant caste-specific biases in the expression patterns of microRNA were identified in the mandibular gland, a site of colony regulatory pheromone biosynthesis. Multiple microRNA target prediction programs were then used to investigate potential regulatory activity of microRNAs with a caste-specific bias in expression upon pheromone biosynthesis pathway genes. Previously identified putative pheromone biosynthesis genes also with a caste-specific expression bias showed multiple potential microRNA regulatory elements (MREs) for microRNAs with caste-specific expression bias. Currently, I am developing *in vivo* methods to verify the gene targets of microRNAs with a caste-specific bias as well as conducting luciferase assays, an *in vitro* method. RNAi constructs for microRNAs upregulated in both queens and queen-less workers were fed to queen-less workers and I am assessing differences in gene expression using 3' TAG-Seq. Given that pheromone biosynthesis pathways in insects are generally derived from modifications to conserved metabolic pathways, these results potentially offer additional insights into

the roles of these microRNAs in metabolic diseases as well as the evolution of sociality in insects. Further, many of the identified microRNAs with a bias in caste-specific expression have established roles in diseases including metabolism-related cancers and other metabolic disorders.

## **Effects of management techniques on reproductive strategy in crop-pollinating bees, *Megachile rotundata***

**Makenna Johnson<sup>1</sup>, Natalie Boyle<sup>2</sup>, Theresa Pitts-Singer<sup>1,2</sup>, Karen Kapheim<sup>1</sup>**

Following honey bees, alfalfa leaf cutting bees, *Megachile rotundata*, are the most important commercially managed pollinator in the United States. Alfalfa leaf cutting bees (ALCB) are effective pollinators of a variety of crops, but are most widely known for alfalfa crop pollination. Though they are solitary, ALCB nest aggregately in above-ground cavities, allowing for convenient maintenance and management by commercial bee producers and farmers. The most crucial step in their management is ensuring that emergence aligns with crop bloom for optimal pollination. This can be achieved by slowing development of the bees by returning them to cold storage after incubation has been initiated. As the climate continues to change, it may be necessary to slow bee development for longer periods of time than done previously. We investigated the effects of this management technique on patterns of reproductive investment among females. We found no significant differences in the amount of nest cells made by bees returned to cold storage for 1, 7 or 14 days, but found that females returned to cold storage for 7 or 14 days provided their offspring with significantly smaller pollen balls, suggesting a decrease in the efficacy of pollination by bees that were suspended in development to account for changing weather. We are further investigating these effects on offspring development to better understand how climate change will influence these crucial pollinators.

## **Value of insect-mediated pollination service to apples in us industrial sectors using an input-output framework**

**Alex Jordan<sup>1</sup>, Harland Patch<sup>2</sup>, Christina Grozinger<sup>2</sup>, Vikas Khanna<sup>1,3</sup>**

In the United States, insect-mediated pollination is a highly valuable asset to nutritional and economic welfare, accounting for over 36 billion US dollars of pollination-dependent crop production alone. Additionally, related agricultural sectors (fertilizers, pesticides) rely upon pollination-dependent crop production, and non-agricultural industrial sectors (pharmaceutical, fuel) share complex linkages with crop production and farming sectors, leading to intricate, indirect dependence upon insect-mediated pollination service throughout the economy. We present an input-output (IO) modeling based framework for quantifying dependence of industry sectors on insect-mediated pollination. Using available pollination-dependent crop field data, and publicly available data from the USDA (United States Department of Agriculture) and NASS (National Agricultural Statistics Service), we establish bounds on economic

value of agricultural crops in farming sectors attributable to insect pollinators. Using this information along with 2012 Benchmark I-O Account data, we quantify the economic dependence of non-agricultural sectors on insect-mediated pollination. Focusing on apple farming, we disaggregate the fruit and nut farming sector to represent the apple farming sector with greater detail. While the direct economic value of apples attributable to insect pollinators is 3 billion dollars, indirect economic dependence of non-agricultural sectors on insect pollinators is equally as significant, though lacking previous quantification. We further identify industrial sectors most vulnerable to loss of insect pollinators. These results are compelling and highlight need for better understanding the role of insect pollinators in agricultural and non-agricultural product life cycles. The implications of these findings for quantifying the value of ecosystem services in product life cycles will be described.

### **Toxicity of some ready-to use garden pesticides to non-*Apis* bees**

**Neelendra Joshi<sup>1</sup>, Olivia Kline<sup>1</sup>, Joseph Belsky<sup>1</sup>, John Adamczyk<sup>2</sup>**

Non-*Apis* bees are important pollinators of numerous wild and cultivated crops. In commercial crop production systems, they are often exposed to various agricultural chemicals that pose serious threats to their survival, and over a period, such regular exposure may subsequently affect biodiversity and abundance of these bees. In other settings, such as backyard gardens and urban landscapes, bees are also regularly exposed to numerous toxic pesticides, and impact of such exposure often goes unnoticed. In this study, we examined toxicity exposure effects of three commonly available ready-to-use (RTU) pesticide formulations to the blue orchard bees (*Osmia lignaria*), alfalfa leafcutter bees (*Megachile rotundata*) and bumblebees (*Bombus impatiens*) in two different exposure scenarios. The results of these bioassays will be presented, and potential implications for ecotoxicological risk assessment of RTU pesticide formulations in urban landscapes will be discussed.

### **Wild bee responses to landscape resources and topography vary seasonally**

**Melanie Kammerer, Eric V. Lonsdorf, Margaret R. Douglas, John F. Tooker, Christina M. Grozinger**

Several interacting factors are driving bee declines, including loss of habitat, pesticide use, increased pathogen and disease pressure, and climate change. Despite this complexity, most models evaluating landscape quality for wild bees only include information on floral and nesting resources. We used an extensive monitoring dataset (> 90,000 specimens) from the mid-Atlantic USA to study how wild bee communities respond to landscape factors, including floral resources, nesting resources, honey bee (*Apis mellifera* L.) abundance, and insecticide toxic load. We also considered topographic location, represented by elevation, slope, and aspect. We found that wild-bee community responses to landscape indices varied seasonally. For spring bee communities, sites with medium- to high-quality nesting resources had higher species richness and abundance of wild bees. Wild bee abundance in the spring was also greater at higher elevation sites. For wild bee communities captured in the summer and

fall, landscape composition and topography were poor predictors of wild bee abundance or richness. Overall, we found bee communities are most sensitive to landscape quality early in the season. We hypothesize that the life history of spring bee species (univoltine, short foraging range and flight season) drives this pattern. Identifying how wild bee communities differentially respond to resources and stressors is a key step in determining effective management strategies. Leveraging information from large-scale bee monitoring programs is an efficient approach for evaluating how biotic and abiotic factors influence bee communities and designing effective management.

### **Native bees exhibit species- and ecosystem-specific changes in abundance with aridity**

**Melanie R. Kazenel<sup>1</sup>, Karen W. Wright<sup>2</sup>, Terry L. Griswold<sup>3</sup>, Julieta Bettinelli<sup>1</sup>, Jennifer A. Rudgers<sup>1</sup>, Kenneth D. Whitney<sup>1</sup>**

Despite widespread concern about pollinator declines, evidence of how climate change may influence bee communities is scarce. Bees may be susceptible to changes in both climate mean and variability, including the frequency of extreme weather events such as drought. We studied relationships between native bee abundance and climate at the Sevilleta National Wildlife Refuge (New Mexico, USA), where multiple dryland ecosystem types converge and climate-induced ecosystem state transitions are predicted. We asked: 1) How does native bee abundance vary with aridity (drought) over time? 2) Do bee abundance relationships with aridity differ among three ecosystem types? and 3) Are bee abundances better predicted by present or past year's aridity? We sampled bees in March-October from 2002-2014, aggregated climate data from co-located weather stations, and focused our analyses on five abundant bee species. Bee abundance relationships with increasing aridity ranged from linear (positive and negative) to quadratic and cubic. All species had nonlinear relationships with aridity in one or more ecosystems, suggesting differential susceptibility to changes in climate mean and variance. One species showed consistent abundance optima at average aridity, and thus might respond negatively to climate shifts. The remaining species had differing abundance relationships with aridity among ecosystems; plant communities may thus mediate their climate responses. Lag effects with climate occurred, and the relative importance of present versus prior year's aridity varied among species. Understanding climate change effects on bees will require attention to bee species identity, ecosystem types and transitions, past and present climate conditions, and possible nonlinear responses.

### **Bee diversity and abundance in urban landscapes**

**Kit T. Keane<sup>1</sup>, Anjali Agarwal<sup>1</sup>, Lauren Hisatomi<sup>1</sup>**

In the face of recent news highlighting the decline of insects worldwide (including many bees), it is reassuring that studies are beginning to show that robust bee communities can seemingly persist even in highly urbanized landscapes. Surveys indicate that urban bee assemblages are able to maintain relatively high diversity by relying on hotspots of floral resources and nesting habitat (ex. gardens and wild

spaces). However, most of these data represent qualitative observations, and more long-term and standardized studies on bee diversity and abundance are needed to assess the health and persistence of bee communities over time, especially with respect to human land-use decisions. Here, we use a GIS-based approach to test for patterns in abundance and diversity of bees collected passively via blue-vane traps.

### **Assessing the effects of common garden pesticides on alfalfa leafcutter bees (*Megachile rotundata*)**

**Olivia Kline<sup>1</sup>, Joseph Belsky<sup>1</sup>, Neelendra Joshi<sup>1</sup>**

The alfalfa leafcutter bee (*Megachile rotundata*) is a solitary, tunnel-nesting bee that is known for its efficient pollination of alfalfa, as well as other crops such as canola and melons. Because of this pollination ability, it is one of the few solitary bees to be commercially managed. In recent years, however, there has been a decline in the populations, ranges, and pollination services of many bee species. Multiple risk factors have been proposed to contribute to these losses, including several commonly used agricultural insecticides. Past research has primarily focused on the toxicity of these insecticides - and to a lesser extent the synergistic toxicity of fungicides and insecticide mixes - on honey bees, but there is a lack of research on solitary bees such as leafcutter bees. In this study, we exposed leafcutter bees to four pesticide products: (a) zeta-cypermethrin, (b) carbaryl, (c) tau-fluvalinate + tebuconazole, and (d) captan + malathion + carbaryl. These pesticide formulations are widely available and marketed for use around the home and garden. We assessed the effects of direct contact exposure by recording bee mortality at 24, 48, 72, and 96-hours after the treatment. The results of the preliminary analysis of this study will be discussed.

### **Options for chalkbrood control in multiple bee systems**

**Lead Author (Ellen Klinger)<sup>1</sup>, Co-author 1 (Diana Cox-Foster)<sup>11</sup>**

Chalkbrood disease is caused by fungi in the genus *Ascosphaera* and occurs in a multitude of managed bee systems (honey bee, alfalfa leafcutting bee, orchard bees), as well as in some native bee communities. Control measures to reduce chalkbrood in bee populations are lacking and can be hazardous to pollinators and the humans who manage them. Safe but effective control measures are desperately needed. We investigated the potential use of hypochlorous acid (HOCL) on control of several species of *Ascosphaera*, each causing chalkbrood in separate managed bee populations. While higher levels of HOCL were effective in reducing spore viability in all species, the effect of HOCL on each species differed. This illustrates that chalkbrood disease dynamics differ between fungal species within the same genus, and these differences should be considered when developing control measures in pollinator systems.

### **The Georgia Tech Urban Honey Bee Project: An integrative approach To education, research, and community engagement**

**Jennifer Leavey<sup>1</sup>**

The Georgia Tech Urban Honey Bee Project (bees.gatech.edu), located in downtown Atlanta, Georgia, has provided a framework for community-engaged research and education across a wide variety of disciplines for the last seven years. The program's multidimensional approach includes education through problem-based courses such as BeeSNAP in which interdisciplinary teams of students develop technologies to facilitate crowdsourcing of data acquisition of plant-pollinator interactions, predict optimal bee habitat using geographic information systems, and design devices to monitor both native bee nests and honey bee hives. The project also maintains partnerships with community gardens and urban farms to provide pollination services, educational outreach, and beekeeper training. For the past three years the project has run a unique summer undergraduate research program, Bee-INSPIRED (INtegrating Service Projects Into REsearch and Design), in which students spend 4 days/week conducting research and 1 day/week observing urban pollinators in action while volunteering with our community partners. The program has encouraged a number of Georgia Tech faculty to pursue bee-related research projects on topics such as the biomechanics of pollen grooming, the use of nanoparticles as mite control, and predicting the range of native bee species using models of climate change. Finally, the project maintains a large volunteer program in which students, faculty, staff, and community members can learn fundamentals of urban pollinator ecology, participate in the creation and conservation of pollinator habitats, assist in maintaining honey bees, and participate in educational outreach to the community.

### **Pollination effects of honeybees (*Apis mellifera* L.) and bumblebees (*Bombus terrestris* L.) in different cultivars of Asian pear (*Pyrus pyrifolia* nakai)**

**Kyeong Yong Lee, Hyung Joo Yoon, Hyeonjin Ko**

Various cultivars of Asian pears (*Pyrus pyrifolia* Nakai) have been cultivated in Korea. Because foraging behaviors of insect pollinators for nectar and pollen determine the fruit production, we investigated comparative pollination efficiency of the honeybee (*Apis mellifera* L.) and bumblebee (*Bombus terrestris* L.) on pear cultivars, including 'Niitaka', 'Gamcheonbae', 'Wonhwang', 'Whasan', and 'Whangkeumbae', that are grown in Korea. The foraging rates and time spent on the flower of honeybee and bumblebee were significantly different among cultivars. The foraging rate of the honeybee was highest in the vars. Hwasan, followed by the Whangkeumbae, Manpungbae, and Niitaka; whereas that of the bumblebee was highest in the vars. Hwasan, followed by the Manpungbae, Niitaka, and Whangkeumbae. In particular, the foraging rate preference of the honeybee was 1/3 in the Niitaka, which was lower than that of the other cultivars. Honeybee spent the longest time in flower of Hwasan, followed by the Niitaka, Whangkeumbae, and Manpungbae; whereas bumblebee spent longest time in flower of Hwasan, followed by the Niitaka, Manpungbae, and Whangkeumbae. Bumblebee showed a 2.8-fold higher rate of foraging in the pollen-

producing cultivars than in the non-pollen cultivars. Fruit set by the honeybee and bumblebee was similar in most of the cultivars, except in Niitaka. Fruit set by artificial pollination was more effective than that by bee pollination in the Niitaka. However, fruit set by bee pollination was similar to that by artificial pollination in other cultivars. In fruit quality from each cultivar was not different from different pollination treatment. Therefore, it is considered that the pollination method using honeybees and bumblebees is a good option instead of the general artificial pollination in various Asian pear cultivar.

## **The effects of copper on the growth of a bumble bee parasite, *Crithidia bombi***

**Laura Leger<sup>1</sup>, Evan Palmer-Young<sup>1</sup>, Quinn McFrederick<sup>1</sup>**

Bees are often exposed to environmental contaminants such as pesticides and metals through flowers via contaminated soils and water. Some metals, such as copper, are known to have antimicrobial properties and have been used to prevent or treat pathogens and parasites in vertebrates. A common bumble bee parasite, *Crithidia bombi*, resides in the guts of bumble bees, and has been shown to reduce the fitness of new, colony-founding queens. It is possible that the small amounts of metals like copper that bees ingest in the wild may reduce parasite loads in the gut. Interestingly, copper has also been shown to alter the gut microbiota of bumble bees. We therefore hypothesize that the exposure to copper can reduce loads of *C. bombi* in bumble bee hosts but may also induce dysbiosis in the microbial communities in bumble bee guts. Here, we will investigate just one aspect of this hypothesis: the impact of copper exposure to cultured strains of *C. bombi*. We will culture strains of *C. bombi* in copper-spiked medium with six different concentrations: 100ppm, 10ppm, 1ppm, 0.1ppm, 0.01ppm, and 0ppm (control). To measure growth of *C. bombi*, we will take optical density readings of cultured strains at 24-hour increments for five days post-exposure to copper. We predict that the toxic effects of copper will reduce parasite loads in bumble bees and that as concentrations of copper increase, *C. bombi* growth will decrease.

## **Managing pollination services: a model of substitution between wild pollinator habitat and honey bee hive rental**

**Elinor M. Lichtenberg<sup>1</sup>, Brian J. Gross<sup>2,3</sup>, David Zilberman<sup>2</sup>, Claire Kremen<sup>4,5</sup>, Erik Lichtenberg<sup>6</sup>**

Agricultural intensification has increased dependence on renting honey bee hives to ensure sufficient pollination of many high-value food crops. That dependence has become more problematic in recent years due to colony collapse, mite infestations, diseases, and overwintering stress. An alternative is setting aside land for wild pollinator habitat. Understanding the feasibility of commercial scale implementation of wild pollinator habitat as a complete or partial substitute for hive rental requires novel quantitative methods. We investigate the economics of managing pollination services by renting honey bee hives versus setting aside land for wild pollinator habitat. To

accomplish this, we develop a theoretical model of the profit maximizing choice of habitat land set asides and hive rentals as functions of crop value, season, hive rental costs, field location, and other factors. We draw on the existing literature to parameterize the model. We use the parameterized model to estimate the relative importance of rented honeybees and wild pollinators for crops of different kinds in various locations. Our model provides insights into where and when maintaining wild pollinator habitat is a feasible strategy for enhancing the sustainability of farming operations.

## **Think big: Landscape variables predict at-risk bumblebee habitat more than local-scale variables**

**Lead Author (Amanda R. Liczner)<sup>1</sup>, Co-author (Sheila R. Colla)<sup>1</sup>**

Some bumblebee species are in decline from habitat loss, pathogen spillover, and climate change. Creating pollinator-friendly habitat has been suggested as a method to mitigate these declines, but habitat requirements for bumblebees are not well understood. We provide the first thorough description of the habitat for two at-risk bumblebee species (*Bombus terricola* and *B. pensylvanicus*) across southern Ontario. We asked: 1) Are local or landscape habitat variables more important, 2) what local and landscape variables describe *B. terricola* and *B. pensylvanicus* habitat, and 3) do important local-scale habitat variables change over a season. Habitat variables were surveyed at 25 sites with a recent occurrence of *B. terricola*, *B. pensylvanicus*, or both species. Landscape variables were extracted from a 1-km buffer around each site. Local-scale habitat surveys were conducted over in spring, mid-summer, and late-summer in 2017. Landscape-scale variables explained more of the variation between *B. terricola* and *B. pensylvanicus* habitat. There were habitat characteristics that were species-specific at both the local and landscape-scale. We did not find any evidence of important local-scale variables changing over the season. These results provide a detailed description of the habitats for *B. terricola* and *B. pensylvanicus* that can be used to inform future habitat models and be used to aid in planning conservation management for these declining species. Furthermore, our results highlight the importance of considering landscape-scale over local-scale habitat variables and considering habitat requirements at the species-level.

## **The Assessing the Effects of Fluctuating Temperature Regimes on Commercial and Wild *Bombus***

**Lead Author (T. Lindsay), Co-author 1 (J.Strange)<sup>1</sup>, Co-author 2 (K.Kapheim)<sup>2</sup>, Co-author 3 (J.Rinehart)<sup>3</sup>, Co-author 4 (J.Knoblett)<sup>4</sup>**

The successful management of bumble bee colonies is crucial to *Bombus* rearing operations. Currently one of the most critical components of effective bee management includes the obligatory artificial winter storage of mated *Bombus* queens. Under natural conditions, wild queens can hibernate up to six to nine months depending on species (Alford, 1969). In captivity, the conventional practice for captive hibernation period is three months and generally yields a high mortality rate. A potential way to mitigate this high mortality rate is interrupting cold storage

with brief pulses of warmth applied on a regular basis. In contrast constant extended low temperature treatments can hinder the benefits of antioxidant enzymes, and thus increase oxidative stress, causing damage to DNA, protein and lipid molecules (Monaghan *et al.*, 2009). FTR's periodic pulses of warmth are thought to be involved in the repair, reverse, or protection against damaged DNA from extended exposure to low temperatures (Kostal *et al.*, 2007). Therefore, my hypothesis was *Bombus* queens survived longer in the fluctuating temperature treatments compared to their constant temperature counterparts. By comparing survival analysis and fat content with queens from two different thermal storage treatments, I gained more insight on premature deaths of queens. The results have important implications for *Bombus* physiology, longevity, and management, which have the potential to improve the *Bombus* rearing industry.

### **Plant drought stress alters floral volatile emissions, and reduces floral rewards, pollinator attraction, and plant reproductive success**

**Rachel Mallinger<sup>1</sup>, Jose Franco<sup>2</sup>, Caitlin Rering<sup>3</sup>**

Potential effects of climate change on plant-pollinator interactions include both spatial and temporal mismatches as well as changes in floral traits. Changes in floral traits can have consequences for pollinator attraction and plant pollination success as well as for pollinator fitness by reducing nectar and pollen rewards. In this study, we examined the effects of drought stress on buckwheat, a globally cultivated plant that is prone to drought stress, moderately dependent on insect pollinators for reproduction, and increasingly utilized in on-farm conservation. Between drought-stressed and control plants, we compared: nectar quantity and sugar composition, pollen quantity, the relative abundance and chemical composition of floral volatile emissions, visitation rates by both managed and wild pollinators, and plant reproductive success. Flowers on drought-stressed plants produced 42% less nectar, and nectar from drought-stressed plants contained significantly lower ratios of sucrose to hexose sugars. Drought-stressed plants received 47% and 45% fewer visits by honey bees and flies, respectively, though bumble bees showed no preference. While there was no significant difference in the quantity of total floral volatile emissions, the compositions differed, with drought-stressed plants having higher emissions of cis-3-hexenol, a volatile that has been shown to repel pollinators in other studies. Finally, both drought stress and reduced pollinator visitation significantly, negatively affected seed set, and reduced pollinator visitation additionally resulted in lower average mass per seed. Our results show that drought stress can have significant, negative effects on floral traits and pollinator attraction, reducing plant reproductive success and the floral resources available to pollinators.

### **Land-use effects on the delivery of ecosystem services and dis-services by insects in agricultural landscapes**

**Yael Mandelik, Tal Shapira, Tohar Roth, Adi Bar, Moshe Coll**

Natural habitats in agricultural landscapes are critical for species persistence and for beneficial ecological functions, such as pollination and pest control. However, these natural habitats may also provide dis-services such as the harboring of pest species. These different taxa are likely to interact in a way that affects their functioning, and ultimately may determine yield quantity and quality. We studied the combined and interactive effects of insect pollinators, pests, and natural enemies of pests, on seed-set, and how the diversity and functionality of these guilds are affected by land use in agricultural landscapes in central Israel. In each of 15 landscapes, spanning a gradient of land use intensity, we established a pair of arrays of potted model plants that were either aphid-infested or aphid-free. In each array, we recorded the diversity of flower visitors, the diversity of natural enemies (predatory insects and parasitoids) and their predation and parasitism rates, and monitored seed-set. We found indirect effects of herbivory on honeybee activity and seed-set: honeybee visitation activity was lower on infested compared to un-infested plants, resulting in lower seed-set in infested plants. The activity of wild pollinators was unaffected by herbivory. The activity of wild pollinators, as well as seed set, were affected by land use at both local and landscape scales, while pest predation and parasitism rates showed only a weak land use effect. In sum, we detected interactive effects of different insect guilds on seed set. These effects varied across landscape gradients and affected the provision of ecosystem services.

### **URBAN BEE DIVERSITY AND ABUNDANCE MONITORING USING CITIZEN SCIENCE**

**Lisa Mason<sup>1</sup>, Arathi H.S.<sup>2</sup>**

This study determined the efficacy of citizen science protocols for monitoring of bee diversity in urban and natural areas in the City of Fort Collins, Colorado. Citizen science provides a variety of benefits including reduced research costs, increased efficiency of personnel and resource use, and increased opportunities to connect people to nature, facilitating habitat protection and pollinator conservation. In 2016, 22 citizen scientists were recruited and trained to identify bees in three gardens, working closely with trained researchers to ensure accuracy. They also collected plant names and tallied floral resources while monitoring. Researchers replicated the citizen scientists' activities on different days, creating two different data sets. Researchers also collected specimens to corroborate the observational data. In 2017, the same study was repeated with 25 citizen scientists. Returning volunteers improved their bee identification skills and helped new volunteers monitor. Researchers repeated the same methods and collected specimens using pan traps, blue vane traps, and netting. Including citizen science volunteers proved to be an effective method to collect data as indicated by the congruency of the citizen science and researcher data sets. We conclude that investing sufficient time training volunteers ensures accuracy of data collected by citizen scientists.

## **Pesticide risk reduction for honey bees through applicator training: opportunities and obstacles**

**Andony Melathopoulos<sup>1</sup>, Rose Kachadoorian<sup>2</sup>, Gilbert Uribe<sup>2</sup> and Matthew Bucy<sup>3</sup>**

The Oregon Department of Agriculture created incentives for licensed pesticide applicators to be trained in how to reduce pesticide exposure to bees, following a string of high-profile poisonings of bumble bees following the treatment of shade-trees with insecticides in 2013 and 2014. We initiated a new training program in 2016 to determine the extent that applicators could be trained to understand honey bee warnings on pesticide labels. We used electronic clicker technology to administer pre- and post- training evaluations and discovered that: 1) while fewer than a quarter of applicators could interpret honey bee toxicity or residual toxicity statements on pesticide labels, 2) their comprehension increased to over 95% following a 60 minute training. The effectiveness of our training, however, was predicated on pesticide labels communicating acute and residual toxicity using standardized language recommended by the US Environmental Protection Agency (USEPA). To determine the extent to which pesticide labels reflect this standardized language, we compiled a database of 232 insecticide labels reflecting 16 high-risk exposure scenarios to Oregon honey bee colonies. We found that: 1) 31.2% of these labels contained at least one deviations from USEPA's recommended language, 2) these deviations were not limited to any one particular scenario or insecticide subgroup and 3) the most common deviation was between the language used to signal the level of toxicity and laboratory-derived acute toxicity values for honey bees. While our findings underscore the importance of applicator education in reducing pesticide exposure to honey bees, it suggests trainings need to incorporate existing inconsistencies in label language to be more effective.

## **Native bee and shrub interactions in a restored riparian landscape**

**Scott R. Mitchell<sup>1,2</sup>, Sandra J. DeBano<sup>1,2</sup>, Mary M. Rowland<sup>3</sup>, Skyler Burrows<sup>4</sup>**

While some woody shrubs are well known to provide forage for bees, others such as willow (*Salix* spp.), are often overlooked but may be important resources for bees. In this study we examined interactions between native bees and native shrubs in a restored riparian system to understand how shrubs can provision native bees throughout the season. We performed targeted handnet sampling throughout the season (April to September) on blooming shrubs in the US Forest Service Starkey Experimental Forest and Range in northeastern Oregon to better understand these interactions. Over the season we caught 297 bees on 6 woody shrub species that are commonly used in restoration plantings. In the early season, shrubs such as wax currant (*Ribes cereum*) and willows may serve as particularly important forage resources in our area because few other plants are blooming. To compare shrub versus forb provisioning of early season bees, we quantified abundance of blooms at our sites and estimated the total number of observed bees on those plants. We found that shrubs

provide a dense floral resource that is readily available and used by a diverse set of bee species in this critical early season period. Our work has implications for managers trying to restore land with woody plants and for landowners that wish to create low-maintenance, long-lived patches of floral resources for bees on their property to enhance pollination services.

## **Postfire environment reveals floral limitation of bumble bee body size, colony abundance, and reproductive output**

**John M. Mola, Michael R. Miller, Sean O'Rourke, Neal M. Williams**

One of the leading causes of pollinator declines is a loss of flowering habitat leading to resource limitation within populations. Understanding floral resource limitation can be tricky as counts of foragers on flowers may simply reflect aggregation of individuals to areas of higher relative resource abundance. Studies which demonstrate resource limitation through metrics associated with elevated fitness (e.g. reproductive output, population size, etc) are needed beyond forager counts alone. Wildfires present unique opportunities to investigate floral limitation, as there is often a substantially increased blooming environment in the years immediately following burning. Within our study area, the University of California McLaughlin Reserve, genetic samples of *Bombus vosnesenskii* before and after fire within burned and unburned areas allow us to investigate how these postfire pulses affect estimated population sizes, foraging range, relatedness, and reproductive output. Using a combination of genetic mark-recapture techniques and body size measurements of netted foragers, we find that the resource-rich postfire environment releases *B. vosnesenskii* from floral limitation resulting in increased forager size, colony abundance, and reproductive output (queen abundance). Furthermore, we find that although long-distance foraging does occur in our study area, most siblings are clustered within 500 m, suggesting that the local floral abundance within burned areas explains the observed results.

## **Investigating the potential for pathogen spillover and pesticide exposure for honeybees and wild bees foraging from blueberry flowers in SW Ontario**

**Ana Montero-Castaño<sup>1</sup>, Cristina Botías<sup>2</sup>, Raquel Martín-Hernández<sup>2</sup>, Mariano Higes<sup>2</sup> and Nigel E. Raine<sup>1</sup>**

Pollinators are declining around the world, and among their main threats are pesticides and pathogens, which can interact synergistically. In agroecosystems, pollinators are simultaneously exposed to these and other threats (e.g. habitat loss). In this study we explore the role of crop flowers as reservoirs of pathogens and traces of pesticides, and the combined threat they pose to wild pollinators. Although the main direction of pathogen spread remains unclear, there is some evidence of transmission from managed to wild pollinators. Most of the known pathogens (protozoans, fungi, bacteria and viruses) are faecal-orally transmitted. Therefore, while foraging, managed bees can contaminate the flowers they visit through deposition of faeces, regurgitation of gut contents or saliva. Wild pollinators subsequently visiting these flowers could be



infected, and chronically exposed to pesticides, through their cuticles or by consuming nectar or pollen of treated plants. Pollinator interactions do not occur in isolation but embedded in complex interaction networks. Therefore, the threat that crop flowers pose to pollinators will depend on the interacting communities in which interactions occur. For instance, honeybees might increase pathogen loads by visiting them; however, if they deplete floral resources, their activity might reduce the attractiveness of contaminated flowers to wild pollinators, thereby reducing risk of infection. I will present preliminary results of my postdoctoral project exploring the risk that of crop flowers pose to pollinators and the role of the interacting community modulating such risk. I expect this study will be useful for conservation and for informing sustainable management of agroecosystems.

### **Mechanics of honey bee pollen pellet removal**

**Marguerite E. Matherne<sup>1</sup>, Suraj Puvvada<sup>1</sup>, Xi Li<sup>1</sup>, David L. Hu<sup>1,2</sup>**

Honey bees are masters at collecting pollen that varies vastly in size and shape. They do this by mixing the pollen particles with nectar and forming a pellet in the pollen basket of their hind legs. How do these pellets stay attached? In our experimental study, we measure the force required to remove the pollen pellets from the basket at different speeds. We compare our results with the rate at which real honey bees remove pellets in the hive. We use fluid mechanical models to explain how the hairs embedded in the pellet keep it attached while the bee is flying and navigating through the hive.

### **Temporal dynamics of multi-host parasite prevalence in species-rich plant and pollinator communities**

**Paige Muñiz<sup>1</sup>, Wee Hao Ng<sup>1</sup>, Peter Graystock<sup>1</sup>, Ashley Fersch<sup>1</sup>, Kyle Parks<sup>2</sup>, Amber D. Tripodi<sup>3</sup>, Christopher R. Myers<sup>1</sup>, Quinn McFrederick<sup>2</sup>, and Scott H. McArt<sup>1</sup>**

How multi-host parasites spread in species-rich communities is challenging to study and poorly understood. One approach is to track parasite prevalence through time, looking for temporal signatures of superspreaders, disease hotspots, and/or dilution effects. Here, we assessed temporal variation in parasite prevalence in a community comprised of 127 species of bees that interact at 90 species of flowering plants. We quantified abundance of each species during a growing season, then screened nearly 6000 bees and flowers for 5 pollinator parasites. We found widespread prevalence of pollinator parasites in bees and on flowers; 42% of bee species were positive for at least one parasite, and 70% of flower species were positive for bee parasites. Mean prevalence of pollinator parasites in the genera *Crithidia* and *Apicystis* generally increased over the collection period in the pollinator community. This was almost entirely driven by the late-season increase in the relative abundance of *Bombus* (bumblebees), which exhibit higher parasite prevalence compared to the other pollinator species. In contrast, the mean prevalence of pollinator parasites on flowers decreased over time, which may be caused by dilution effects from higher floral abundance later in the season, counteracting the increased prevalence among pollinators. Our results suggest that *Bombus* may play a disproportionate role in the transmission of parasites in the community, and that temporal variation in floral abundance is an important factor to consider when modelling disease dynamics.

## **Corridors through time: does resource continuity impact pollinator communities, populations and individuals?**

**Charlie C. Nicholson<sup>1,2,3</sup>, Taylor H. Ricketts<sup>1,2</sup>**

Spatial aspects of connectivity have received considerable attention from both ecologists and conservation biologists, yet temporal connectivity – the periodic linking of habitat patches – likely plays an equally important, but largely overlooked role. Different ecosystem properties underpin temporal connectivity, but here we focus on the uninterrupted availability of foraging sites. This resource continuity is expected to be particularly important in highly dynamic landscapes, such as agriculture, however its influence remains untested. We present a framework, grounded in ecological principles of connectivity and metapopulation dynamics, that explains how diversified agriculture, by promoting resource continuity, may support beneficial organisms. We then use a novel natural experiment consisting of farms that either grow blueberry and raspberry crops or raspberry without blueberry to investigate the response of pollinators to resource continuity at community, population and individual levels. We do not observe a strong signal of resource continuity on any of these measures but the effects of resource continuity are expressed most strongly when considering population level impacts. Though far from definitive, our results suggest that agricultural landscapes composed of sequentially flowering crops may bolster local populations through temporal complementarity of flowering resources, but more research is needed.

### **The impact of Thiamethoxam on drone navigation and behavior**

**Heather North<sup>1</sup>, Claire Campion<sup>1</sup>, Arun Rajamohan<sup>2</sup>, Julia Bowsher<sup>1</sup>**

Since the market introduction of imidacloprid, neonicotinoids have become the fastest growing class of insecticides. Neonicotinoid treated seeds give rise to plants that are protected from pests throughout the plants entire life. The plant can metabolize the neonicotinoid through all parts of the plant, including its pollen and nectar. When non-target insects such as honey bees forage on these plants they are bringing contaminated resources back to their nest for consumption. With respect to the male bees (drones), the effects of dietary thiamethoxam on their physiology and behavior has not been well studied. Drones participate in mating flights with virgin queens. The mated queen stores the spermatozoa in her spermatheca for the duration of her life, using it to build a strong healthy colony. Since a drone's only role is reproduction, the ability to fly and navigate to the mating congregation area (MCA) is critical for successful mating. If neonicotinoids interfere with the ability of a drone to find and navigate to the MCA, then reproduction has been disrupted before it can even take place. The overall loss of drones due to an impaired ability to navigate and or control motor function once it reaches the MCA, can lower the genetic diversity of colonies and thus impact the future success of a colony. This study aims to assess thiamethoxam's impact on drone navigation and ability to fly on the basis that it will significantly affect the overall colony health and fitness.

## Preferred plants for pollinators: a case for their conservation

Onyeka Peter Nzie<sup>1,2</sup>, Millicent A. Cobblah<sup>1,3</sup>, James K. Adomako<sup>4</sup>

Reports of the decline of pollinators have necessitated the need to maintain and boost their diversity in both natural and agricultural areas. Studies was conducted in four different habitats in the Eastern and Greater Accra regions of Ghana, to ascertain the preferred plants for pollinator species which could be used for their conservation. This was investigated through field observation of plants the pollinators foraged on and laboratory analysis of the pollen types on the pollinators. The analysed pollen types were viewed using both compound light microscope and scanning electron microscope. The pollen types collected from the pollinators were compared with a reference pollen library of flowering plants from the study areas. Twenty-five plant species were identified based on pollen observations. These plant species belong to 14 families and 23 genera. The family Fabaceae had the highest representation of foraged plant species (9) followed by Malvaceae (3) while other plant families were represented by single plant species. The agricultural site in the University of Ghana had more foraged plant species (42%), followed by a grassland study area (26%). At the individual plant level, plants such as *Cassia rotundifolia*, *Commelina diffusa* and *Jatropha gossypifolia* had a high number of diverse pollinators foraging on them. We conclude that these plants may be valuable in meeting the nutritional requirements of these pollinators as well as their conservation. It is therefore recommended that the nutritional content of pollen of these plants be analyzed in future research to determine how they influence bee health.

## Stingless bee nutrition altered by landscape simplification in the Andes

Diana Obregon<sup>1</sup>, Katja Poveda<sup>1</sup>

*Tetragonisca angustula* is the most common stingless bee species found from Southern Mexico to Northern Argentina. In Colombia this species is particularly abundant in the Andean region. The Andean Mountains are experiencing a rapid loss of natural habitats due to their transformation into cropland and grazing areas. In temperate regions, these drastic changes in land use have been linked to pollinator decline, but their effects on the Andean bees remain mostly unknown. In the eastern Colombian mountain range, we located 16 natural colonies of *T. angustula* and placed them in hives. With drone imagery, we calculated the proportion cropland and natural land covers in a 500m radius around each colony. For three different sampling dates, we determined the diversity of the pollen collected, the nutritional composition of the larval food and the workers' body size. We also weighted the brood, the food provisions and the total colony weight through time. We found that as forest area decreased and agricultural area increased the pollen richness decreases in the provisions collected by the bees. We also found that larval food contained less protein and trehalose and that the worker's body contained less glucose. This altered nutrition is also correlated with smaller bee size and lower weight gain in the brood. Our data from this important tropical wild bee supports what has been found in temperate zones, showing that as land use shifts from natural to agricultural areas, bee nutrition is negatively affected with potential devastating effects on bee health and pollination services.

## A comparison of acute toxicity endpoints for adult honey bees with technical grade active ingredients and typical end-use products as test substance

Bridget F. O'Neill<sup>1</sup>, Susan E. Spruill<sup>2</sup>, Silvia Hinarejos<sup>3</sup>, Ana R. Cabrera<sup>4</sup>

The honey bee, *Apis mellifera*, serves as a model organism for pollinators in risk assessment frameworks globally. The acute toxicity tests with adult honey bees for contact and oral exposure are part of the requirements for pesticide registration in the United States, and are conducted with active ingredient. A question often asked is, is the formulated product (TEP) more toxic than the active ingredient (TGAI) to honey bees. We explored this question by mining publicly available databases from regulatory agencies in North America and Europe, where testing with the TEP is required. The objective of this study was to determine if TEPs are equal in toxicity to the TGAI. Of the 151 active ingredients with reported endpoints for contact exposure, 28 were classified as either moderately or highly toxic. Of the 151 TEPs evaluated for contact exposure, 137 had the same toxicity classification as the TGAI. Of the 141 active ingredients with reported endpoints for oral exposure, 23 were classified as moderately or highly toxic. Of the 141 TEPs evaluated for oral exposure, 121 had the same toxicity classification as the TGAI. More than 95% of TEPs had toxicities classified consistent with, or deemed to be less toxic than, the TGAI, leaving the remaining 5% to be more toxic than the TGAI. Most of these occurred in the oral exposure testing. In general, the TEPs toxicity classifications were largely the same as their TGAI toxicity classifications, suggesting that the risk assessments of TGAIs would be sufficiently protective to pollinators.

## Are urban pollinator plantings the bee's knees in Fargo, Nd?

Mia G. Park<sup>1</sup>, Vincent A. Oliveras<sup>2</sup>, Joseph P. Rinehart<sup>3</sup>, Julia H. Bowsher<sup>1</sup>, and Kendra J. Greenlee<sup>1</sup>

Most flowering plants, including a third of our crops, rely on animal pollination, notably insects. In the United States, both commercial and native pollinators are in decline and habitat loss is a primary driver. Inadequate food and nesting resources reduces population growth and exacerbates a myriad of other stressors. Pollinator habitat enhancements have become a popular pollinator conservation tool, designed to provide a safe, diverse and abundant foraging community to sustain pollinators throughout their active season. While wild pollinator communities and managed honey bees have been shown to respond positively to habitat enhancements in agricultural landscapes, few studies have examined whether such benefits are realized in urban settings and whether these benefits extend to other managed pollinators. This study partners with the Audubon Society to investigate whether recent (1-4 yr old) pollinator plantings established in urban parks measurably benefit both wild and managed pollinators. In 2018, we surveyed a total of 8 sites (4 control and 4 enhanced with plantings) along the Red River in Fargo, ND and Moorhead, MN. Floral resources, wild pollinator visitation, and nesting success of the commercial alfalfa leafcutting bee (*Megachile rotundata*) were monitored through the growing season. Both wild pollinator community structure and *M. rotundata* nest performance increased with floral

abundance but not diversity. Contrary to predictions, pollinator enhancement efforts did not improve availability of floral resources, nor did they measurably increase pollinator abundance, diversity or reproduction. Flowering weedy species were commonly observed at control sites and pollinator plantings did not always establish well due to competitive grass species. Our preliminary results suggest that active land management is needed in our study system to ensure pollinator plantings realize intended benefits for pollinators. Finally, because flowering weeds presented a major food source for pollinators in urban parks, conservation efforts may need to consider potential trade-offs of weed control programs.

## Habitat prescriptions for safeguarding wild bees in a North American landscape

Alana Pindar<sup>1</sup>, Nigel E. Raine<sup>1</sup>

Habitat loss and fragmentation are among the most significant factors contributing to global pollinator declines. Although the importance of habitat quality and connectivity, as well as landscape composition and configuration, are comparatively well understood for wild bee communities, the actual amount of habitat needed to sustain bee community diversity in landscapes remains virtually unknown. Using comprehensive land cover and bee species data sets from agricultural and wild environments in southern Ontario (Canada), representing 63,000 individual records collected over 15 years, we assessed the amount of habitat need to maintain bee community diversity. We also tested which of the 27 different habitat types were most influential in maintaining expected bee community structure across a range of foraging distances. Our results suggest that safeguarding wild bee communities requires approximately 8% land cover from a range of habitat types in a landscape, irrespective of whether the conservation aims are species richness or abundance. Sensitive lands such as riparian and wetland habitat were very significant predictors of bee species richness and abundance, even more so than hedge-row and semi-natural habitats. This suggests that environmental and conservation policies should consider including restoration or creation of wetland habitats in order to safeguard pollination services provided by wild bee species in agricultural landscapes.

## Routes of exposure to solitary bees for pesticides used in almond and alfalfa seed production

Theresa Pitts-Singer<sup>1</sup>, Diana Cox-Foster<sup>1</sup>, Ellen Klinger<sup>1</sup>, and Andi Kopit<sup>2</sup>

Conventional U.S. crop production include the use of pesticides (insecticides, miticides, herbicides and fungicides). Our work with *Osmia lignaria* (blue orchard bees) and *Megachile rotundata* (alfalfa leafcutting bees) has revealed exposure routes and pesticide effects that are not considered or observed for *Apis mellifera* (honey bees). *Osmia lignaria* are exposed to flowers and moist soil during almond bloom that may be sources of contamination to adults and larvae through ingestion and contact. Pesticide analyses of orchard flowers and soil revealed that, likely due to chemical properties, pesticides applied months to years prior to current bloom season may be found in bee resources. *Megachile rotundata* forage on alfalfa flowers whose staminal columns are enclosed until “tripped” by a bee by applying pressure on the

flower’s keel. Therefore, recently sprayed pesticides do not tend to contaminate nectar and pollen. However, pesticides that are systemic may occur in alfalfa nectar and pollen, and pesticides that are translaminar may penetrate leaves and have long-lasting residuals. By gathering floral resources and cutting leaves for forming nest cell cups, *M. rotundata* may be exposed to pesticides via adult and larval contact and ingestion. We show examples of bee exposure in field and laboratory studies using crop relevant pesticides and show effects such as altered behavior while foraging or at nest sites, increased incidence of dead eggs, and altered larval developmental times. Understanding exposure routes and pesticide effects is critical to assuring bee safety, while acknowledging the need for pesticide use in crop protection.

## The bees of Minnesota, progress to date

Zachary M Portman<sup>1</sup>, Mike Arduser<sup>2</sup>, John S. Ascher<sup>3</sup>, Elaine C. Evans<sup>1</sup>, Joel D. Gardner<sup>4</sup>, Jessica D. Petersen<sup>5</sup>, Daniel P. Cariveau<sup>1</sup>

Here we report on the status of the Bees of Minnesota project, a collaborative effort between the Cariveau Lab at the University of Minnesota, the Minnesota Department of Natural Resources (MNDNR), and multiple bee taxonomists. The bee fauna of Minnesota is particularly interesting because it contains a mix of species from different biogeographic areas, including the Eastern US, Western US, and Great Plains. We are compiling the species list using a combination of museum specimens and recent surveys by the Cariveau Lab and the MNDNR. So far, we have recorded a total of six bee families and 45 genera: Andrenidae (5 genera), Apidae (14 genera), Colletidae (2 genera), Halictidae (10 genera), Megachilidae (13 genera), and Melittidae (1 genus). The total number of species is still being compiled, but we have a number of new state records for Minnesota. Three genera are newly recorded from the state: *Chelostoma*, *Neolarra*, and *Pseudoanthidium*. In addition, we have recorded over 20 new state records at the species level. Many difficulties remain, as we continue to resolve the identification of species in difficult taxonomic groups. This is particularly challenging because most genera have either never been revised or were last revised over 50 years ago. This work will help to better our understanding of species distributions and the changes to our bee fauna over time.

## Do herbaceous land enhancements enhance honey bee (*Apis mellifera*) forager visitation and colony performance in Michigan?

Gabriela Quinlan<sup>1</sup>, Meghan Milbrath<sup>1</sup>, Clint Otto<sup>2</sup>, Matthew Smart<sup>2,3</sup>, Rufus Isaacs<sup>1</sup>

Floral enhancements funded by USDA have been promoted as a means of supplementing available forage for honey bees (*Apis mellifera*) in the landscape, thereby improving colony productivity to the benefit of the beekeeping industry. Various studies have explored the benefits of floral availability and land cover types on forager behavior and visitation. Other studies have assessed how conservation landcovers improve colony performance. However, the effects of floral enhancements

are often addressed independently for either forager visitation or colony performance, with few studies investigating both in the same system. We assessed whether Conservation Reserve Program (CRP) land affected honey bee visitation and colony performance in Michigan. While more honey bees were observed foraging in CRP land compared to analogous unmanaged land, the amount of CRP land within a colony's foraging range did not have a significant effect on colony performance. This suggests that while attractive and rewarding to honey bees, the current investment in CRP plantings is not sufficient to have a measurable benefit for the beekeeping industry in Michigan.

### **How does consumption of nectar secondary compounds impact bumblebees exposed to a neonicotinoid pesticide?**

**Sarah K. Richman<sup>1</sup>, Denyse Marquez Sanchez<sup>2</sup>, Angela M. Smilanich<sup>1</sup>, Anne S. Leonard<sup>1</sup>**

In recent years, there has been mounting evidence of detrimental effects of pesticides on pollinator populations. Specifically, neonicotinoid pesticides found in nectar and pollen can negatively impact bee reproduction, foraging, and immune function. The majority of published studies examining the physiological or behavioral consequences of neonicotinoid consumption are conducted using a sucrose solution containing sugar and water. In nature, floral nectar has a complex chemistry, frequently containing secondary compounds which can themselves have impacts on bee health and survival. In a lab-based study of the bumblebee *Bombus impatiens*, we examine the potential of nectar chemistry to interact with the effects of neonicotinoid consumption on bee performance. We ask whether bees that consume artificial nectars offering field-realistic levels of the nectar alkaloid caffeine and terpenoid thymol exhibit differences in immune function and survival probability after an acute dose of imidacloprid, a widely used neonicotinoid pesticide, compared to bees that consume artificial nectars lacking these compounds. Preliminary results point towards indirect effects of both nectar compounds and pesticides on these response variables via changes in sucrose consumption and activity levels. Bees that consumed nectar secondary compounds experienced compound-specific differences in immune function, and the difference between dosed and non-dosed bees differed across diet treatments. Bees that received an acute dose of imidacloprid consumed less sucrose on average than non-dosed bees; however, the reduction in consumption did not translate to a difference in survival probability. Our findings suggest a complex pathway of direct and indirect effects on bee performance and, ultimately, colony fitness.

### **Dissecting the role of nectary-specific membrane transporters in modulating floral nectar elemental profiles**

**Rahul Roy<sup>1</sup>, Emilie Snell-Rood<sup>2</sup>, John Ward<sup>1</sup>, Clay Carter<sup>1</sup>**

Floral nectar is a carbohydrate rich reward produced by plants to attract pollinators. Studies have revealed the presence of a range of metabolites such as proteins, lipids, amino acids and alkaloids in nectar. Inductively coupled plasma mass spectrometric

analysis of nectar from *Brassica sp* & *Cucurbita pepo* has also revealed significant presence (up to mM levels) of ions such as P, K, Na, B, Li, Fe, Mg and Al. We also analyzed gene expression in the nectaries of these flowers using RNA-Seq. Among the genes displaying high expression during nectar production, a number of them are specialized membrane transporters involved in the transport of metal ions. This has led us to hypothesize their role in modulating the elemental profile of nectar for optimum pollinator visitation. We are currently utilizing a genetic approach to test the function of these transporters in affecting nectar elemental composition. We are also trying to ascertain elemental profile differences in nectar collected from Brassica plants exposed to various ionic stresses or metal contamination. Subsequently the composition of the nectar will be mimicked in pollinator preference studies to ascertain whether changing the elemental profiles actually affects feeding preference in pollinators. For example, preliminary data suggests that cabbage white butterflies prefer artificial nectars supplemented with Na at biologically relevant concentrations. The final goal of this study is to inform strategies by which we can control the appearance of high levels of metal stressors in the diet of pollinators.

### **The effect of temperature on *Penstemon heterophyllus* nectar and pollination success**

**Kaleigh A Russell<sup>1</sup> and Quinn S McFrederick<sup>1</sup>**

Climate change can have longstanding effects on inter-species interactions within pollination systems. For example, as temperatures increase, certain flowers may bloom earlier in the season; this has impacts on native bee health, as bees that emerge later in the season and are left with less forage. Microorganisms, such as bacteria, fungus, and viruses, which associate with plants and pollinators play an important role in pollinator networks and can also be affected by temperature change. Here, we test whether passively heating the California native perennial, *Penstemon heterophyllus*, would lead to differences in nectar volume and sugar concentrations, nectar-inhabiting microbial communities, and pollination success compared to those left to natural temperatures. We sampled nectar twice a week for the 6-week flowering season ranging from early June to late July 2018. We found that extreme temperatures in Southern California summer 2018 influenced nectar sugar concentrations and nectar volume dramatically, along with passive heating treatment. We expect, as this study continues to unfold, to find differences in nectar-inhabiting microbial communities between treatments as climate change disrupts plant-pollinator-microbe interactions.

### **Effects of fungicide on *Bombus vosnesenskii* microbiome composition and foraging choices**

**Danielle Rutkowski<sup>1</sup>, Isabelle Maalouf<sup>1</sup>, Eliza Litsey<sup>1</sup>, Rachel Vannette<sup>1</sup>**

Agrochemical application can influence pollinator health in diverse ways. Although insecticides have received much research focus, fungicides are frequently applied to control plant pathogens and may also influence pollinator health, particularly for native bees. Previous research suggests that bee-associated fungi may play important roles in the physiology and ecology of bees. Fungi, including yeasts, can impact foraging choices of bees and are found in both individual bee microbiomes as well as in the wider colony microbiome, often present in collected pollen or nectar. Bumblebees

exhibit particularly well-characterized ecological associations with yeasts. Queens often host fungal propagules during overwintering, and worker attraction to yeast volatiles has been documented in multiple floral systems. However, it remains poorly understood how fungicide application influences colony-level health. The yellow-faced bumblebee, *Bombus vosnesenskii*, is native to the Western US, and is associated with fungi in its microbiome (unpubl). Here, we hypothesized that fungicide application will shift the internal and colony microbiome of *B. vosnesenskii*, influence worker foraging choices, and influence colony performance. We collected nest-seeking *B. vosnesenskii* queens, reared them to a stage of brood production and created microcolonies. Microcolonies were exposed to the fungicide propiconazole, a commonly used fungicide, or a control solution. We will examine the effect of propiconazole on worker foraging preference, survival, feeding rate, offspring production, and offspring size using these microcolonies.

### **Utilizing whole genome sequencing to identify genomic signatures across US commercial honey bee lines**

**Perot Saelao<sup>1</sup>, Arian Avalos<sup>1</sup>, and Michael Simone-Finstrom<sup>1</sup>**

Molecular markers have been a powerful tool in breeding for traits of interest. Their use enables greater resolution and efficacy of the selective process. Implementation of marker-assisted selection in honey bees has historically met with limited success. An important barrier has been poor characterization of the genetic diversity in the greater US honey bee population, as well as within economically relevant stocks. In this study, begin to overcome this limitation by examining four common-use breeding stocks (Koehnen Italian (CA), Kona Italian, Strachan Carniolan, Koehnen Cariolan,) and three stocks from distinct selection programs focusing on improving honey bee colony health (Russian, Minnesota Hygienic, Pol-line). We use a pooled sequencing approach to identify markers associated with stock variation and examine signals of artificial selection. Analysis of stock specific variation can serve efforts to easily identify and categorize genetic groups with a high degree of discriminatory power. Our approach will also allow for a greater understanding of the landscape of selection in those stocks bred for critically important traits such as *Varroa* Sensitive Hygiene. Results of this sequencing effort will produce a robust data set that can serve as reference for analyses of genetic diversity across honey bee populations and also localize regions of interest in honey bee health traits facilitating future efforts for the development of marker assisted selection techniques.

### **The consequences of worker size variation on demography in bumble bees**

**Timothy Salazar<sup>1</sup>, Steve Ellner<sup>1</sup>, Scott McArt<sup>2</sup>**

Bumble bees (*Bombus* spp.) are primitively eusocial and exhibit large variation in worker size that contrasts starkly with size distributions observed in other castes and species such as honey bees. This variation has inspired numerous empirical studies which have highlighted general trends in bumble bee ecology. In particular, smaller workers tend to remain in the hive and perform tasks involved in broodcare while larger workers tend to perform tasks such as foraging, guarding, and fanning. Various

hypotheses have been explored in relation to the general trend of size polyethism; empirical results have yet to adequately explain the maintenance of size variation in workers. Indeed, many studies have found large workers outperform small workers at most tasks. Furthermore, workers remain labile in their duties, switching between in-hive and out-of-hive tasks during their lifetime with this switching sometimes a result of stressors on hive health. Theoretical studies meanwhile have highlighted the existence of optimal intermediate sizes that contrast with observed bimodal distributions of worker size. While this size variance in workers has been explored empirically, it has not been incorporated into demographic models. Here we explore the consequences of a bimodal distribution in worker size in a discrete time, stage structured population model with worker size included as a continuous trait influencing parameter values.

### **Analysis of nutrient profile of pollen from honey bee colonies**

**Brooke (Sayre-Chavez)<sup>1</sup>, Elisa (Bernklau)<sup>2</sup>, Louis (Bjostad)<sup>2</sup>, Arathi (Seshadri)<sup>1</sup>**

Honey bees provide an essential service, that of pollination in both natural and agricultural ecosystems, which makes them environmentally and economically incredibly important. However, there are a lot of factors affecting honey bee health, of which nutrition is a key issue. Poor nutrition can lead to decline in honey bee populations by increasing their susceptibility to pests, pathogens and other abiotic stresses. Bees depend on plants for pollen and nectar, which is the source of food for developing larvae. Pollen from the plants are collected by bees and stored in the colony. The protein from pollen ensures proper larval development, which means under poor nutrition, health of bees will be adversely affected. In addition to proteins, pollen is also a source of several micronutrients that are key components for healthy physiological functioning of bees. Currently, not a whole lot is known about the nutrient profile of pollen in colonies of beekeepers, which was a major motivation for this research. We collaborated with members of the Colorado Professional Beekeepers Association (CPBA) to gather pollen samples from 5 of their colonies over the season when bees are active. We assayed the pollen samples collected to quantify proteins, carbohydrates, lipids, and micronutrients such as phytochemicals. Preliminary results show a shift in nutrient composition in pollen across the season. We will compare seasonal variation in pollen nutrient content and explain our findings based on the geographical location of the colonies. The implications of our findings on colony performance will be discussed.

### **Microbial metabolites mediate bumble bee attraction and feeding**

**Robert N. Schaeffer<sup>1</sup>, Caitlin C. Rering<sup>2</sup>, Isabelle Maalouf<sup>3</sup>, John J. Beck<sup>2</sup>, Rachel L. Vannette<sup>3</sup>**

Pollinators such as bumble bees rely upon chemosensory cues to effectively localize and evaluate essential resources in both natural and agricultural landscapes. Increasingly, it is recognized that microbes can alter resource quality, as well as

produce chemosensory cues such as volatile and non-volatile metabolites that may alter the quality of sensory information received by pollinators in a species-specific manner. Here, we test the hypothesis that species of nectar-inhabiting microbes differentially influence pollinator attraction and feeding via microbial metabolites in nectar. We examined electrophysiological potential of bumble bee antennae to respond to volatile microbial metabolites, followed by behavioral responses using choice assays. Moreover, gustatory responses to nectar-inhabiting microbes were assessed through both no-choice and choice feeding assays. *Bombus impatiens* antennae responded to a subset of volatile metabolites produced, with 2-ethyl-1-hexanol eliciting the strongest response. Naïve workers also displayed a clear preference for *Asaia* bacteria compared to *Metschnikowia* yeast based on volatiles alone. However, *B. impatiens* consumed significantly more *Metschnikowia*-inoculated nectar, suggesting distinct roles for non-volatile and volatile microbial metabolites in mediating feeding decisions, with potential to affect associative learning and future foraging. The distinct roles of microbial olfaction and gustatory responses suggest bumble bees may use multiple microbial chemical cues to assess floral attractiveness and reward quality, with potential consequences for forager economics and host reproduction.

### **The importance of pesticide exposure in the pollinator risk assessment**

**Daniel Schmehl<sup>1</sup>, Ana Cabrera<sup>1</sup>, David Fischer<sup>1</sup>**

Honey bees and other pollinators are frequent visitors to most flowering plants to collect nectar and pollen for their nutritional needs. Pollen and nectar may contain residues from pesticides used to protect crops and other plants from damaging pests. The registration of a pesticide requires a Pollinator Risk Assessment, which is based on toxicity and exposure estimates. While toxicity of a pesticide is innate (i.e. does not change), the exposure to a pesticide is dependent upon the use pattern, application timing, the physical and chemical properties, and the biology of the particular pollinator. In this presentation, we will provide a detailed overview on how we utilize exposure data to characterize the risk of a pesticide to pollinators. We will present empirical data highlighting the design of exposure studies, how different use patterns and soil types may result in different residue concentrations within the bee relevant matrices, and how minimum pre-bloom spray intervals are determined. These exposure data are critical for risk mitigation when establishing the label instructions for how a pesticide is used in a way that is safe to visiting pollinators.

### **Antimicrobial Lipid Transfer Proteins are a Common Feature in Floral Nectar**

**Anthony Schmitt<sup>1</sup>, Andrew Sathoff<sup>2</sup>, Catherine Holl<sup>1</sup>, Deborah Samac<sup>2,3</sup>, Clay Carter<sup>1</sup>**

The primary solutes in nectars are sugars, but proteins also often accumulate to high concentrations. Here we report that non-specific lipid-transfer proteins (nsLTPs) are a common feature of floral nectars across species and that they likely prevent microbial growth *in vivo*. For example, SDS PAGE analysis of raw nectar from two species, *Brassica rapa* and *Cucurbita pepo*, revealed an array of proteins in each nectar. Major

bands at ~10 kDa for both *B. rapa* and *C. pepo* nectar were found to contain non-specific lipid transfer protein (termed BrLTP2.1 and CpLTP2.1, respectively). The genes encoding both nsLTPs were predicted to have signal peptides required for secretion from the cell and eight cysteines, which are characteristic of all nsLTPs. Heterologously expressed and purified BrLTP2.1 and CpLTP2.1 both bound strongly to saturated free fatty acids and had strong direct antimicrobial activity, particularly against necrotrophic fungi. Cumulatively, our findings suggest that nsLTPs are a widespread feature of floral nectars that may help protect reproductive tissues from infection. Furthermore, previous reports have shown that pollinators are the primary vectors for transporting microbes into nectar. Antimicrobial peptides may be important for reducing the microbial load in nectar and mitigating the exploitation of nectar by harmful fungal and bacterial species to protect both plants and their pollinators.

### **Spatio-territorial networks in male carpenter bees (genus *Xylocopa*)**

**Stefan Schoof<sup>1</sup>, Kyle Go<sup>1</sup>, Kit T. Keane<sup>1</sup>**

Male territorial behavior in *Xylocopa virginica* is both aggressive and conspicuous. It has been shown that males tend to patrol territories close to active burrow openings, where they may encounter females. While this behavior has been well-documented, we still lack clear picture of the variation in male competitive strategies and effort. Is there variation in territory fidelity between individual *X. virginica* males? How do resident and satellite males differ? Do larger males attempt to control larger territories or do they focus their energy on smaller, more desirable territories? Here we conduct a mark-recapture study to quantify the spatio-territorial behavior of males in a large and dense population of *X. virginica*. We apply network analysis in an attempt to identify individual territorial patterns.

### **What factors affect the foraging pattern of bumble bees in an alpine environment of northern Japan?**

**Akari Shibata<sup>1</sup>, Gaku Kudo<sup>1</sup>**

Bumble bees are among the most common and effective pollinators in cool-temperate, alpine, and sub-arctic ecosystems. Because bumble bees are eusocial insects with an annual lifecycle, their colony growth depends on floral resource throughout much of the growing season. The abundance and species composition of floral resource used by bumble bees dramatically change as the season progresses. In the short growing season of alpine environments, activity of worker bumble bees is maximized in the middle of season, in which competition for floral resource may be intense. We predict that the foraging pattern of bumble bees is affected by both of floral resources and interactions with other bumble bee species. Field surveys were conducted at Mt. Asahidake in the Taisetsu Mountains, Hokkaido, northern Japan. Floral abundance of each species and the foraging frequency of bumble bees to those species were recorded during the

summer. In total, 23 surveys were conducted during the flowering season, from early July to the middle of September in 2015, 2016, and 2018. Two bumble bee species, *Bombus hypocrita* and *B. beaticola* were commonly recorded throughout the summer. We analyzed the seasonal changes in floral resources and activity of two bumble bee species to clarify the factors affecting the foraging patterns of individual species.

## Deep learning for image-based bee identification

Brian J. Spiesman<sup>1</sup>, Claudio Gratton<sup>2</sup>

Reliable taxon identification is critical for comparative studies of biodiversity. However, for insects, such as bees, genus- and species-level identification can be time consuming and often requires the specialized knowledge of expert taxonomists. Another approach is to use machine learning techniques to identify bees from photos based on, for example, the geometry of wing venation. New deep learning methods, however, do not require such prior domain knowledge. Instead, when a deep learning model is trained on a set of known subjects, the model learns on its own how to evaluate images and distinguish subjects belonging to different genera or species. We analyzed a preliminary data set of 1193 images of bee forewings belonging to species in six genera: *Agopostemon*, *Bombus*, *Certina*, *Lasioglossum*, *Megachile*, and *Osmia*. We used convolutional neural networks (CNN), to classify images to the genus- and species-level. At the genus level, our models achieved high training and validation accuracy. But because the data requirements for CNN models are high, species-level accuracy was low with our small data set. Our preliminary analysis shows great promise for expanding these deep learning methods to identify a broader range of bee genera or, potentially, species. We hope to eventually scale up our models and apply them to images of whole bees captured in the field.

## Bees, microbial ‘meat,’ and other sacrilege

Shawn A. Steffan<sup>1,2,\*</sup>, Prarthana S. Dharampal<sup>2</sup>, Bryan N. Danforth<sup>3</sup>, Hannah R. Gaines-Day<sup>2</sup>, Yuko Takizawa<sup>4</sup>, Yoshito Chikaraishi<sup>4</sup>

Bees are thought to derive all protein directly from pollen and nectar sources. Recent findings suggest this is largely untrue. It appears that larval bees feed extensively on pollen-borne prey. These prey are heterotrophic microbial communities and are often suffused throughout a fermenting pollen mass. Because microbes register within a trophic hierarchy just as animals do, a pollen-borne herbivorous microbe represents heterotrophic biomass (i.e., ‘meat’) enmeshed within autotrophic detritus. When a larval bee consumes this complex of trophic groups, the bee assimilates the amino acids of microbial prey as well as those of the dead plant material. The degree to which the bee has assimilated microbe-derived amino acids can be measured empirically as the trophic position of the bee. To assess the ubiquity of such omnivory in bees, we examined the trophic positions of bees representing six of the seven major bee families on Earth. We found significant evidence of omnivory across the 14 species (12 genera) of our study, suggesting omnivory is rampant among bee fauna. Given widespread

evidence of microbivory, we examined whether the community compositions of microbes in fermenting pollen were critical for bee health, focusing on the potential for fungicides to alter the community of pollen-borne microbes. In multiple studies involving bumble bees (*Bombus impatiens*), we found evidence that fungicide residue alters the microbial community and elevates larval mortality. Collectively, these findings suggest bees require microbial protein and that fungicides indirectly cause bee mortality by interfering with external microbial symbionts.

## Direct and interactive effects of nutrition and pesticide stressors on the solitary bee *Osmia lignaria*

Clara Stuligross<sup>1</sup>, Neal Williams<sup>1</sup>

Bees are threatened by many factors including floral resource limitation, pathogen infection, habitat loss, and pesticide exposure, and they can be exposed to risks across landscapes as they forage widely for pollen and nectar. These drivers rarely act in isolation, and understanding their interplay can have important consequences for pollinator conservation, especially in agroecosystems where limited floral diversity and pesticide use may be at odds with the demands for crop pollination services. We investigated the direct and interactive effects of nutritional and pesticide stressors on the solitary bee, *Osmia lignaria*. We established nesting *O. lignaria* females in 16 field cages using a crossed resource x pesticide design; cages contained spring wildflowers at high or low densities, treated with or without imidacloprid, the most widely used neonicotinoid insecticide. Nutrition and pesticide exposure both directly and interactively affected bee nesting activity. Bees provided with abundant floral resources constructed more nests than those in low-resource treatments. However, the difference between resource treatments was much smaller for bees exposed to imidacloprid. Pesticides appear to limit bees’ ability to take full advantage of the additional resources. Our research is among the first to provide experimental evidence of this nutrition-pesticide interaction, a critical step in understanding mechanisms underlying pollinator health.

## Exploring the impact of prescribed fire on gyne overwintering survival and nest initiation

Taylor Tai<sup>1,2</sup>, Claudio Gratton<sup>1</sup>

In the US alone, at least seven native bumble bee species (genus *Bombus*) have experienced population declines or range contractions in recent years. Given the vulnerability of this genus, knowledge of how habitat management practices impact bumble bees is crucial for conservation efforts. In particular, prescribed fire is a key management tool for maintaining prairie habitat, but its impact on bumble bees across the colony life cycle is largely unknown. Fire may contribute to mortality for gynes overwintering directly below the soil surface, and has the potential to alter the ground substrate so that it is more or less attractive to nest-searching gynes. To investigate this, I am surveying *Bombus* gyne abundance during the period of hibernation emergence and nest initiation on grasslands following spring burns. I predict that burning will decrease the abundance of gynes emerging from hibernation, but that this

effect will not carry over to nest-searching gynes due to their dispersal from surrounding non-burned habitat. The results of this study will allow land managers to develop burning practices that are more sensitive to the needs of bumble bees.

### **Effect of food on the honey bees foraging fitness and efficiency**

**A. S. Tanda\***

In case the honey bees will not be kept in good health world survival may be under threat due to lack of quality food production. The collective endeavors provide the best utilization of available tools and resources for improving the fitness and protection of honey bees. A coordinate program to develop practical and effective techniques for a variety of people are fruitful. Foraging efficiency of honey bees and their health are intimately related to the crop ecosystem. Latest observations show that the conservation efforts have a direct impact on the environmental suitability for supporting honey bee pollinator industry. However, relatively little is known about the effect of habitat on the health of honey bees. Knowing main factors that influence bee fitness at the colony level monitoring may explain various types of ecobioeffects. Quality of food and pressures influenced the foragers. Food distance improved the efficiency in visitation. Preliminary studies highlights the potential of crop foraging area may improve the bee fitness, efficiency and good use of honey bees.

### **Impacts of neonicotinoid pesticides on insect olfactory processing**

**Anna (Tatarko)<sup>1</sup>, Anne (Leonard)<sup>1</sup>, Dennis (Mathew)<sup>1</sup>**

Insect pollinators use olfaction to find and assess floral resources, as well as to find nesting habitat and mates. Neonicotinoid pesticides are the most widely used pesticides in the world due to their ease of application and low cost. In addition, plants incorporate these pesticides into every type of plant tissue, including the nectar and pollen that insect pollinators consume. Recent work suggests neonicotinoids can impair olfactory processing, with potential downstream effects on foraging behavior, social interactions, and possibly survival. However, it is unclear whether the impairment occurs during peripheral olfactory information processing in the sensory neurons or during information processing in deeper regions of the brain, or both. We aim to resolve this issue using the well-studied *Drosophila melanogaster* olfactory system as a model of insect olfaction to test the potential for neonicotinoids to disrupt olfactory neuron function. After establishing a pesticide exposure protocol and confirming exposure of flies to sublethal doses, we used single unit extracellular recordings in the fly antenna to look for reduced olfactory neuron activity in flies dosed with the neonicotinoid imidacloprid. We discuss our findings in relation to potential impacts on pollinators, including how work in model systems may allow us to predict sensory impairments in bees and the potential consequences for key foraging behavior such as host plant recognition and floral resource evaluation.

### **Integrating pest and pollinator management: the impacts of pesticide use on pollinators in commercial watermelon production**

**Ternest, J.J.<sup>1,2</sup>, Ingwell, L.L.<sup>1</sup>, Kaplan, I.<sup>1</sup>, and Foster, R.E.<sup>1</sup>**

Fruit set in cucurbit crops such as watermelon is entirely dependent upon pollinators, which makes them an important aspect of grower management. This reliance on pollinators means that growers must consider them when making pest management decisions, especially when using pesticides, which have a negative impact on pollinators. Thus, pest management in watermelon production faces a potential trade-off between pests and pollinators. The ways in which growers manage this trade-off, could have a large impact on the communities of both groups and the yield of the crop. During the 2017 and 2018 growing seasons, we worked with 16 commercial watermelon growers on 30 fields in Indiana and Illinois. Each of these growers implemented unique strategies for pest and pollinator management. We set out to investigate grower decision making, how to better implement integrated pest and pollinator management, and how these decisions impact pest and pollinator communities and grower outcomes. A diverse array of pollinators was identified, with communities being highly variable between sites. Pollinator communities were also far more sensitive to pesticides and grower management than were pests, which were never observed in damaging densities regardless of management. These results provide evidence that growers using threshold-based pest management experience greater pollinator diversity and potentially better fruit set without negative economic impacts due to pests.

### **A side-by-side comparison of honey bee health in colonies managed using conventional, organic, and chemical-free systems**

**Robyn Underwood<sup>1</sup>, Brenna Traver<sup>2</sup>, Margarita Lopez-Uribe<sup>1</sup>**

Honey bees are the most important managed pollinator for crop production, making the issue of colony losses a food security concern. Various practices are used to manage colonies, including conventional, organic, and chemical-free systems. An improved organic system is needed, as parasitic mite pressure and mite control chemicals are detrimental to honey bee health and sustainability. There is a critical need to develop scientific data that support organic beekeeping management practices with the goals of improving honey bee health and creating a profitable economic opportunity for beekeepers. This study is a stakeholder-driven, integrated systems-based project to rigorously test the effects of conventional, organic and chemical-free honey bee management systems on honey bee health. By measuring winter survival, honey production, varroa mite, nosema disease, and virus levels, and immunocompetence, we aim to determine the differences between the three systems. In addition, the impacts of chemical mite treatments on bee bread microorganisms is being explored. Our long-term goal is to generate evidence-based best management practices for a sustainable organic beekeeping system that will improve honey bee colony health, reduce environmental impacts, and increase economic returns to beekeepers.



## Bees in the trees: forest canopy resources for orchard pollinators

Katherine R. Urban-Mead<sup>1</sup>, Paige A. Muniz<sup>1</sup>, Scott H. McArt<sup>1</sup>, Bryan N. Danforth<sup>1</sup>

Previous studies of bee diversity in eastern apple orchards have found orchards with nearby forest had higher wild bee richness and abundance than those near agriculture. Apple-visiting bees often carry forest tree pollen, which we hypothesize bees also use before apple trees bloom. Mass-blooming forest canopies in early spring may thus be an important resource pulse for pollinators. Yet, temperate canopies are difficult to access and rarely studied. To characterize early-spring canopy bees, we sampled in orchards and in orchard adjacent forest canopies and understories using bowl traps. Samples were taken weekly from March to June at nine field sites over three years, and we also tracked forest bloom phenology. Bee abundance peaked in the forest prior to apple bloom, declined as the canopy leafed out, and increased in apple orchards until after peak apple bloom. Of over 45 species caught in the canopy, nearly all were also “apple bees” based on our data and a ten-year dataset of regional orchards. Some species of *Lasioglossum* were strongly associated with forest habitat, while several *Andrena* had canopy-associated males while later-emerging females were primarily found in orchards. To understand how adult bee forage preferences changed over time, we also dissected guts of bowl-caught bees. Using microscopy, we identified the tree genera preferentially consumed by different bee species. Our results support the importance of early-spring mass blooming forest canopies for bees, and suggest that ecological forest management should consider both wild pollinator habitat and forage resources.

## Sustainable landscape enhancement to reduce pesticide exposure in agricultural landscapes

Surabhi Gupta Vakil<sup>1</sup>, Judy Wu Smart<sup>1</sup>

Pollinators are critical for the production of various crops and serves as an important link in the food chain and ecological cycles. Recently there were reports of high losses of pollinators such as wild bees (abundance has declined by 23%), commercially managed honey bees (40% annual winter mortality), and monarch butterflies (abundance reduced by 15%). Major factors responsible for pollinator health decline include diminishing habitat and exposure to agrochemicals. To mitigate these losses recommendations are made to establish pollinator habitat in agricultural areas, however, improperly designed landscape enhancements can expose pollinators to non-target agrochemicals. Of particular concern are systemic neonicotinoid insecticides and *Bacillus thuringiensis* (*Bt*) toxin-based biocides. This project examines the use of existing tree lines as potential drift barriers to mitigate neonicotinoid and *Bt* contamination of pollinator habitats established near crop fields. Neonicotinoids residues were collected using sticky traps near tree lines and from flowers within pollinator habitats with and without drift barriers to determine the field level exposure

to bees. Similarly, leaves were collected from the milkweed plants growing at corn field margins to evaluate the exposure risk to *Bt* pollen and *cry* protein concentration. The field realized values will inform relevant dosing concentrations for supplemental laboratory toxicity bioassays. Preliminary results suggest drift barriers were effective in reducing *Bt* pollen exposure in milkweed leaves and had higher abundance and diversity of pollinators and beneficial insects. This study will improve current recommendations for landscape management with special consideration to protecting pollinator resources in high agricultural production areas.

## Big bees spread disease: a trait-based approach to measuring pathogen transmission

Jennifer I. VanWyk<sup>1</sup>, Lynn S. Adler<sup>1</sup>

Body size impacts the rate of pathogen transmission via behavioral and physiological mechanisms. Pollinator networks are highly variable in trait space for both flowers and pollinators; determining traits that result in high rates of transmission is key to understanding and predicting disease dynamics in a network. Body size could affect pathogen transmission via differences in behavior (foraging rates and preferences) and physiology (defecation amount and rate). We took advantage of the 10-fold variation in worker body mass within *Bombus impatiens*, manipulating bee size using microcolonies in field experiments to test effects on disease transmission. Inside 48 tent arenas, we introduced an infected ‘donor’ microcolony of large or small workers, and an uninfected ‘recipient’ microcolony of average-size workers. Bees foraged on pairs of plant species that varied in floral morphology for 10-14 days. We measured foraging behavior, worker pathogen loads and colony reproduction after each trial. Small donor bees were significantly more infected than large donor bees. However, large donor bees were twice as likely to transmit *Crithidia bombi* as small donor bees. Both behavioral and physiological mechanisms may underlie this apparent paradox. Large bees foraged significantly more often than small bees, although bout length did not differ, and had higher reproduction. In the lab, body size was significantly positively correlated with fecal volume, suggesting large bees had more potential to transmit pathogens. Thus, body size had significant impacts on disease transmission mediated by behavior and/or physiology, demonstrating the multifaceted impacts of this trait on transmission dynamics.

## Drivers of bee-mediated pollen dispersal in a fragmented landscape

Amy Waananen<sup>1</sup>, Daniel Cariveau<sup>2</sup>, Stuart Wagenius<sup>3</sup>

Bee-mediated pollen movement is a critical dispersal mechanism for many plants. In fragmented landscapes, such dispersal can have tremendous demographic and evolutionary consequences, especially for small populations and obligate out-crossing plants. Strategies to maintain movement amidst ongoing habitat loss would advance conservation in fragmented landscapes. However, the challenges of tracking pollinator movement at large spatial scales mean that the drivers of pollen movement in fragmented habitats remain poorly understood. By tracking the pollen movement of a bee-pollinated forb, *Echinacea angustifolia*, in a severely fragmented prairie

landscape, I measured a subset of realized pollinator movements within a 780 ha study area. Pollen movement *within* populations of *Echinacea* is influenced by the spatial isolation and timing of flowering of individual plants, so I hypothesized that pollen movement *among* populations would also be influenced by individual plants' isolation in space and time. I mapped all *Echinacea* individuals and tracked their flowering phenology in 8 populations separated by 50-3000 m, and used microsatellite paternity analysis to identify the pollen source of seeds, indicating pollen movement. These patterns reveal how bee-mediated pollen movement is influenced by the spatial and temporal distribution of flowering plants in a florally-depauperate environment and shed light on the extent to which pollinators maintain connectivity of floral populations in fragmented landscapes.

## **Pesticide exposure of wild bees visiting pollinator hedgerows**

**Laura T. Ward<sup>1</sup>, Claire Kremen<sup>1</sup>, Michelle L. Hladik<sup>2</sup>, Nicholas J. Mills<sup>1</sup>**

To better understand the exposure of wild bees to pesticides in an agricultural landscape, samples were collected from fields in northern California. Hedgerows are known to provide habitat for wild bees, but these bees may also be exposed to pesticides from nearby agricultural fields. The current study targeted eight hedgerow sites located in an intensively managed agricultural landscape near Sacramento, California that includes almonds, (wine) grapes, rice, tomatoes, and walnuts. In addition to collecting both wild bees and honey bees, soil, flowers, and silicone passive sampling devices (PSD; staked near the hedgerows to sample the air) were also included. Sampling was conducted from April to June 2016, to coincide with peak bloom and bee activity. Samples were analyzed for >150 pesticides and degradates using both gas and liquid chromatography-tandem mass spectrometry. Overall, 38 pesticides were detected in all matrices (10 insecticides and degradates, 12 fungicides, 15 herbicides and degradates, and 1 plant growth regulator). The number and type of pesticides detected varied by matrix; 25 compounds were detected in the soil, 24 in PSDs, 18 in wild bees, 17 in flowers, and 10 in honey bees. These results help determine how pesticide residues in wild bees compare with honey bees, flowers, soil, and PSDs.

## **Plasticity in the pre-diapause nutrient sequestration period of bumble bee (*Bombus impatiens*) queens**

**Kristal M. Watrous<sup>1</sup>, S. Hollis Woodard<sup>1</sup>**

Bumble bees have an annual colony cycle, wherein queens overwinter in a diapause state. Queens must sequester enough nutrients during the pre-diapause period to metabolize during diapause, making this period critical for survival. We examined how timing of food availability during the pre-diapause period impacts nutrient storage in bumble bee (*Bombus impatiens*) queens. We aimed to identify whether there is a time point during early adulthood beyond which queens are unable to recover from dietary setbacks and store sufficient nutrients for diapause. We assigned newly-eclosed queens to one of three diet treatments (pollen-starved, nectar-starved, or a control diet of 50% sucrose and honey bee-collected pollen), each lasting for one of four treatment durations (3, 6, 9, or 12d). After this period, all queens were fed control diet until day 12, then collected. We examined queen survival, weight change, and abdominal

glycogen and lipid levels in response to treatment. We found evidence of increased mortality and a net decrease in weight for bees deprived of nectar for six or more days, whereas pollen starvation had a less profound impact on survival and weight at all but the longest starvation durations. Our findings suggest that queen bumble bees are somewhat able to recover from brief dietary limitations, but there is an early age beyond which they cannot sequester sufficient nutrients for diapause. This study sheds light on plasticity in queen bumble bees in response to dietary limitations in early adulthood, and timescales upon which nutrient limitation has resounding consequences for queen survival.

## **Potential resource competition between managed honey bees (*Apis mellifera*) and native bees within natural areas in Florida**

**James R Weaver<sup>1</sup>, Rachel E Mallinger<sup>1</sup>**

Managed honey bees are an important commodity worldwide and are integral for providing crop pollination services within and outside their native ranges. During times of the year when crop pollination services are not required, commercial beekeepers need large tracts of land as temporary habitat for their apiaries, which are often situated within or near natural areas. Unfortunately, there is limited information on the competitive effects between non-native honey bees and wild bees within these natural areas. In this study, we deployed managed honey bee (*Apis mellifera*) colonies at two different natural sites with contrasting histories of commercial apiary presence and measured floral visitation rates of both honey bees and wild bees before, during, and after deployment, and at varying distances from the deployment point. Before honey bee colony deployment, there were significantly more honey bees foraging at the site with a history of commercial apiaries than at the site without, suggesting establishment of feral honey bee populations as a result of historical apiary presence. Prior to deployment, we also found higher wild bee abundance and richness at the site without a history of commercial apiaries. Wild bee visitation rates, diversity, and resource use will be analyzed as a function of honey bee colony deployment (before/during/after) and distance to honey bee colonies. Our results highlight resource use and potential competition between managed honey bees and wild bees in native Florida ecosystems, and may be used to inform both native bee conservation and honey bee management.

## **Evaluating shared pollinator taxa that provide services across multiple crops and regions**

**Bryony K. Willcox 1\*, Brad G. Howlett 2, Andrew J. Robson 3, Linley Jesson 4, Brian Cutting 5, Lisa Evans 5, Lindsey Kirkland 1, Malou Jean Meyzonier 6, Victoria Potdevin 6, Manu E. Saunders 1,7 and Romina Rader 1**

Global crop production is dependent on both managed and wild pollinating insects, yet we know little about the patterns of important taxa at local and regional scales. Some pollinator taxa are likely important across multiple crops and regions, and conversely some floral resources, like cultivated crops are grown in many locations. In this study, we surveyed avocado, mango and macadamia crops (n = 13,200 insect observations)

across different spatial and temporal scales to identify shared common taxa. Across the three crops in Bundaberg, QLD, two wild pollinators, a fly, *Stomorhina discolor* and a stingless bee, *Tetragonula carbonaria* were abundant and present in both mango and avocado crops. Macadamia orchards were dominated by *Apis mellifera* (88% of flower visits). Even though shared taxa were identified between crops, pollinator assemblages were distinct across the three crops due to varying proportions of shared and rare taxa. Flower visitation was a good indicator of pollination contribution with the of mango crops, whereby *Tetragonula carbonaria* had a greater contribution than the more frequent visitor, *Apis mellifera*. Sixteen managed and wild taxa were common and consistently present across three years in avocado, yet their importance along with the remaining wild taxa varied among years. Shared family taxa were identified between growing regions; however, their importance to network structure differed across regions. Identifying the taxa shared across multiple crops, regions and years will facilitate the development of strategies that are region-specific, inclusive of bee and non-bee taxa and co-flowering crop vegetation, to optimize crop pollination services in agricultural systems.

## **Pollinator Diversity in Northeastern Utah, a Longitudinal Study**

**Mary-Kate F. Williams, Frank D. Parker, Diana L. Cox-Foster**

For solitary native bees, little data exists on longitudinal distributions across time. A unique data set does exist from a study using nest traps in northeastern Utah performed by Frank Parker and Vincent Tepedino, beginning in the 1970's, and then again in the 1980's and 2000's. In the 1984 traps, Tepedino and Parker found 29 species of xylophilous bees, ten of which exhibited some level of parsivoltinism at differing elevations. Nine of ten parsivoltine species were from the genus *Osmia*, a group of species that is increasingly used for commercial pollination. A follow-up study is in progress to compare species diversity across time, to ask how diversity has changed and how it is associated with land-use changes. At the same sites used by Parker and Tepedino, block traps were placed in 2018 and 2019: ten sites within Logan Canyon, one site at Bear Lake, and one site at Mendon (northeastern UT; Cache and Rich counties). At each site, ten traps were placed 20 m apart. Each block contained 40 holes of mixed sizes (4, 6, 8, and 10 mm) randomly distributed in ten rows with five columns. Traps were placed for six months; and upon retrieval, traps were opened to collect overwintering organisms to rear out for identification and emergence data. A new addition to this repeated study is the identification of pathogens found in natural solitary bee populations from unmanaged areas in Utah. Molecular diagnostics will be used to identify pathogens both from historical samples and current samples.

## **Improved spring low-temperature storage of *Megachile rotundata***

**George D. Yocum, Arun Rajamohan, Julia H. Bowsher and Kendra J. Greenlee**

Managed solitary bee species used in pollination services can be subjected to low-temperature storage (6°C) multiple times throughout their life cycles. The primary period of storage occurs during overwintering and may last for 8 to 9 months. A second period of storage may occur in the spring to slow bee development, so that bees' emergence will be synchronized with the crop bloom. This spring storage is problematic, because the bees are developing and therefore, likely maladapted to survive suboptimal temperature exposure. Survival of the spring stored *Megachile rotundata* pupae can be significantly increased by interrupting the exposure to constant 6°C by a daily, one hour pulse at 20°C (Fluctuating Thermal Regime, FTR). Under commercial conditions, an FTR storage protocol would be impractical due to stress placed on the refrigeration compressors. Therefore, we investigated storage protocols designed to decrease the stress on the compressors. Two new temperature profiles were designed. In the first, the temperature changed slowly over time in a wave-form thermoprofile (peak temperatures of 12 or 18°C). The second thermoprofile was square in shape with the pulse duration increased from 1 to 6 or 12 hours at 12 or 18°C. The wave forms and 12°C square thermoprofiles had similar or lower survival rates as compared to the FTR controls. Both of the 18°C square thermoprofiles (6 and 12 hours) more than doubled the survival rate over that seen in the FTR controls. These results demonstrate that more compressor-friendly spring storage protocols are actually beneficial to the bees.

## **Quantifying pesticide contamination of bee-collected pollen in diverse landscapes**

**Jon Zawislak<sup>1</sup>, Gus Lorenz<sup>1</sup>, John Adamczyk<sup>2</sup>, Neelendra Joshi<sup>3</sup>**

Honey bees are the most important managed pollinators in commercial agriculture. Large scale mixed agricultural landscapes in the mid-south are vital to maintaining commercial honey bee operations. Severe declines in bee populations have generated concerns about the role of seed-treated crops and honey bee health issues. To investigate the role of pollen in seed-treated crops as a component of bee diet, honey bee colonies were established in both agricultural and urban areas. The agricultural site was surrounded by a predominate mixture of soybeans, corn, cotton, grain sorghum and fallow fields within the foraging range of the bee hives. Urban hives were located within a large urban community garden, containing diverse fruit, vegetable and ornamental plants, as well as pasture, woodland, wild areas, and residential areas within foraging range of bee hives. Pollen traps were used to sample pollen loads directly from foraging bees, approximately every two weeks for an entire honey production season (March-September). Pollen collection began before crops were planted, and continued until crops had ceased blooming. Pollen samples were identified taxonomically to determine the proportion of the bees' pollen diet that was derived from agricultural crops for each sample. Pollen samples were also screened for pesticide residues. Results from preliminary analysis will be presented and discussed.

## **Shared flowers, shared parasites? Honey bee floral visitation is linked with *Nosema ceranae* prevalence in bumble bees**

**Maryellen C. Zbrozek<sup>1</sup>, Michelle L. Fearon<sup>1</sup>, Elizabeth A. Tibbetts<sup>1</sup>**

Community factors such as species richness and host abundance are often important for understanding variation in infection risk among hosts in different communities. As community biodiversity and abundances are changing due to the impact of humans on ecosystems, it is becoming critical to understand how these changes will impact the dynamics of parasite transmission in ecological communities. Although many factors influence pollinator population decline, one key factor is the prevalence of the microsporidia *Nosema ceranae* in both managed and wild pollinator populations. Thus far, it is unclear whether the prevalence of *N. ceranae* is influenced by the rapid changes in pollinator communities. We tested whether community factors such as species richness, host abundance, and frequency of honey bee and bumble bee visits to flowers are linked with *N. ceranae* prevalence in honey bees and bumble bees from seven different pollinator communities. We found that *N. ceranae* prevalence in bumble bees is correlated with the frequency of floral visitation by honey bees, but other community factors such as species richness and abundance are not correlated with *N. ceranae* prevalence. These findings suggest that spillover of *N. ceranae* from honey bees to bumble bees may occur through shared floral resources. Understanding floral visitation as a potential interspecific transmission mechanism for *N. ceranae* in pollinator communities may be useful in preventing further spread of the parasite, which could help slow down rates of pollinator population decline and boost the health of pollinator communities.