

Leople and bees have a long shared history. Honeybees, natives of Europe, were carried to the United States by early settlers to provide honey and wax for candles. As agriculture spread, bees became increasingly important to farmers as pollinators, inadvertently fertilizing plants by moving pollen from male to female plant parts as they collected nectar and pollen for food. Today, more than two-thirds of the world's crop plants—including many nuts, fruits and vegetables—depend on animal pollination, with bees carrying the bulk of that load.

It's no surprise that beekeeping has become a big business in the farm-rich Midwest. Wisconsin is one of the top honey-producing states in the country, with more than 60,000 commercial hives. The 2012 state honey crop was valued at \$8.87 million, a 31 percent increase over the previous year, likely due in part to the mild winter of 2011–2012.

But other numbers are more troubling. Nationwide, honeybee populations have dropped precipitously in the past decade even as demand for pollination-dependent crops has risen. The unexplained deaths have been attributed to colony collapse disorder (CCD), a mysterious condition in which bees abandon their hives and simply disappear, leaving behind queens, broods and untouched stores of honey and pollen. Annual overwintering losses now average around 30 percent of managed colonies, hitting 31.1 percent this past winter; a decade ago losses were around 15 percent. Native bee species are more challenging to document, but there is some evidence that they are declining as well.

Despite extensive research, CCD has not been linked to any specific trigger. Parasitic mites, fungal infections and other diseases, poor nutrition, pesticide exposure and even climate change all have been implicated, but attempts to elucidate the roles of individual factors have failed to yield conclusive or satisfying answers. Even less is known about native bees and the factors that influence their health.

Poised at the interface of ecology and economy, bees highlight the complexity of human interactions with natural systems. As reports of disappearing pollinators fill the news, researchers at CALS are investigating the many factors at play—biological, environmental, social—to figure out what is happening to our bees, the impacts of our choices as farmers and consumers, and where we can go from here.

At first glance,

a honeybee colony literally buzzes with activity, true to its industrious reputation. But there's much more than meets the eye.

"People forget these hives have more than just bees in them," says Kirk Grubbs, a graduate student in bacteriology. In addition to the workers, drones and queen, there are developing pupae; stores of pollen, nectar and honey used to feed the colony; and a resinous substance called propolis

that seals and protects the hive. But beehives also house complex microbial communities that bind together the entire hive-centered ecosystem. "They all come together as a larger organism," Grubbs says.

A healthy hive likely depends on a healthy community of microbes, says bacteriology professor Cameron Currie, Grubbs' advisor. Much like those in our guts, bacteria in a beehive normally exist in a balance of good and bad, where the beneficial keep the pathogenic in check.

Hive microecology is relatively new territory for scientists. Grubbs and Currie are using an approach called next-generation DNA sequencing to take a genetic census of the microbial species present and begin to define what's "normal" for a hive community. Unlike previous attempts to survey hive microbes, this high tech approach uses DNA fingerprints of all the microbes present to reliably represent the population without biasing toward familiar strains or those more amenable to growing in a laboratory environment. With the benefit of the new technique, Grubbs has been able to look at more than 100,000 DNA sequences—previously an unthinkable feat.

So far his work with this and other methods has identified dozens of microbial groups, including distinct communities associated with different parts of the hive—for example, pupae, adults, stored pollen or honeycomb. "These different components represent very different microbial communities," Grubbs says, similar to work that has identified discrete bacterial populations in different parts of the human body.

With this microbial portrait of a healthy hive, Grubbs is working to better understand the roles of these microcommunities in colony health and productivity. He's also asking how these symbiotic relationships may be affected by environmental influences such as agricultural chemicals.

Honeybees routinely encounter a stew of compounds, from pharmaceuticals used to control disease to pesticides and herbicides carried into hives by foraging adults. Grubbs is currently studying hives treated with the common crop pesticide chlorothalonil, which is frequently found in hives in large amounts. His approach allows him to look at community-level effects of exposure over a period of several weeks or months, which simulates the type of exposure faced by a real hive.

Preliminary results suggest that chlorothalonil exposure significantly changes the microbial communities associated

Kirk Grubbs, a graduate student in bacteriology, examines a hive he keeps on the sixth floor of the Microbial Sciences Building.

with adult bees, with lesser effects on pupal microbiota. He doesn't yet know what these changes will mean for the bees or how they might affect a hive's function. But he hopes to find distinct patterns that could serve as health indicators or even identify bacterial strains or new antibiotics that could be used to treat ailing colonies.

"Molecular characterization of this experiment could not have been done even three years ago," Currie says. "It's an exciting time to

do this work because next-generation sequencing is allowing us to get these insights we couldn't have gotten at before."

While Grubbs and Currie

peer into the microenvironments of a beehive, entomology graduate student Hannah Gaines is taking a wider view of bees in the context of their macroenvironments.

Though the familiar honeybee originated in Europe, there are hundreds of native bee species that play critical roles in both agricultural and natural landscapes. "When we think of pollination we think of crop plants, but 95 percent of all flowering plants require insect pollination—and most of those are being visited by native pollinators rather than honeybees," says Gaines, who is conducting her work in the lab of entomology professor Claudio Gratton.

Her research has shown that, in general, more diverse landscapes have more bees. She has documented more than 200 species of native bees in Wisconsin cranberry fields—a surprisingly high number for a single crop.

Contrast this to a vast almond orchard in central California, where the only bees in sight are imported honey-



What you see: worker bees and cells filled with pollen (lower half) or pupae (upper half), with the dark queen in the middle. What you don't: the complex microbial communities that make for a healthy hive, which bacteriologists Cameron Currie and Kirk Grubbs are studying using next-generation DNA sequencing.

bees trucked in during bloom season. Though they may appear lush to human eyes, the vast monocultures that dominate major agricultural areas are virtual wastelands to a bee for the majority of the year.

Bees need just two primary resources: food and shelter. But the intensively managed landscapes of heavily agricultural areas often have neither. Groomed to maximize efficiency, such fields bloom simultaneously and have little uncultivated land with suitable bee habitat—undisturbed soil for ground nesters, hollow stems and snags for cavity nesters. Consider a large watermelon farm, Gaines says. "When the watermelon is in flower, there's a huge resource for the bees, but when the watermelon's not in flower, it's a desert."

As a result, commercial pollination has become big business. Beekeepers truck their hives around the country, hitting each crop when it is in bloom. A profitable hive may cover thousands of miles in a year, traveling between the Midwest and California or Florida and Maine.

In contrast, native bees cover relatively little ground, generally foraging within a few miles of their nests and often specializing in one or a few types of flowers. Because of this, they need more biodiversity in a small area, including plants that bloom at different times of year.

Natural woodlands and prairies may be the ideal environments for these natives, but "if we want to talk about conservation, we have to talk about conservation in agricultural landscapes because that's what we have," Gaines says.

Her research shows that cranberry marshes in wooded areas have higher diversity and abundance of native bee species. "Within a certain radius, if you have more natural habitat you have more bees," she says. She and other researchers published a paper this spring showing similar results for dozens of other types of global agricultural landscapes.

Native bees make a big difference. A study published in the journal Science this past spring found that wild pollinators significantly increased yield in 41 different cropping systems around the world—from coffee to cotton—whether honeybees were there or not. In contrast, honeybees enhanced yields in just

one-seventh of those cropping systems. Other studies have documented that honevbees become even more effective pollinators when wild bees are present, leading to more and better fruit.

Unfortunately, native bee populations may also be shrinking. Some bumblebees are known to be in decline; researchers believe other species also are experiencing drops, but they often lack historical data for conclusive studies. CCD is not the culprit since most natives are solitary rather than social and do not have hives, but it is likely that many of the same triggers—disease, poor nutrition and pesticides—may underlie the problems. A recent study conducted in Illinois by researchers from three universities implicated changing climate and land use in regional losses of dozens of bee species over the past century.

Gaines hopes that her landscapelevel work will lead to research-based management recommendations that can benefit both farmers and pollinators. She and others have shown that specific agricultural practices can make a positive difference. Restricting use of pesticides and fertilizers, diversifying





Hannah Gaines hand-pollinating cranberry plants in a campus greenhouse as part of her research studying the effects of various factors on cranberry pollination.

fields and integrating bee habitat in or near fields can boost wild bee populations and productivity.

Amid growing recognition of this value, some farmers, especially in agriculturally dense areas, are experimenting with planting flowers along field edges and on unused land in an attempt to attract and support native bees and, in turn, honeybees. Ultimately, Gaines says, "Native bee management is really habitat management."

Lhese findings emphasize

the critical role that people play in the bee ecosystem. Both natives and honeybees are enmeshed in human activities, highlighting the need to engage beekeepers, farmers, regulators and scientists toward supporting healthy bee populations.

Social and historical contexts are key for this process, says entomologist Sainath Suryanarayanan, a postdoctoral fellow in community and environmental sociology. Rooted in research at land-grant universities, agricultural entomology has largely focused on

controlling crop pests and improving yields. Common experimental methods were designed to look for large, rapid effects in a controlled environment—for example, acute toxicity of high levels of a single substance.

These approaches have not found consistent toxic effects on pollinators. Chemical companies arguing that their products are safe for bees routinely point to the lack of rapid lethality. Regulatory agencies have adopted a similar stance, accepting a lack of evidence of harm as evidence for no harm. But you only get answers to the questions you ask, cautions Suryanarayanan, and these questions are not the right ones when the goal is long-term health of a population.

For instance, the traditional research approach is poorly suited for detecting impacts of the types of exposures pollinators actually receive in fields—that is, chronic exposure to low levels of many different chemicals over a long period of time.

This shortcoming is especially problematic in light of widespread use of newer systemic pesticides that persist in crop plant tissues and lead to prolonged

The decline of honeybees makes the role of native bees even more crucial. And diverse landscapes tend to have more bees, says CALS entomology student Hannah Gaines, who documented more than 200 species of native bees in Wisconsin cranberry fields.

exposures. Residues can also accumulate over time in hives and on beekeeping equipment. Concerns about these compounds have led European regulators to ban neonicotinoids, a prevalent class of systemic pesticides, on beepollinated crops.

"The kinds of studies that are being done would not allow us to know with any degree of certainty whether they are hurting bees because of this complex set of interactions," says Daniel Kleinman, a professor of community and environmental sociology. "There is essentially an area of ignorance that has been produced about this, things that we simply don't know—and yet policy and practice are proceeding on the basis of that ignorance."

In essence, the current system is biased against finding the majority of biologically relevant effects, yet our regulatory structure establishes policies based on the premise that harm we do not measure does not exist.

To be able to understand—and thus protect—pollinators, scientists and regulatory agencies alike must take a broader view of the issues at hand. acknowledge the complexity of the system and begin to explore some of the other perspectives involved, Kleinman and Suryanarayanan say.

Farmers who grow pollinatordependent crops already tend to be more aware of bee-friendly practices, such as limiting chemical use during bloom season or spraying at night, says Gaines.

Eugene Woller, owner of Gentle Breeze Honey (and a CALS Short Course grad), keeps some 600 hives mostly at dairy farms in and around southcentral Wisconsin and "pays" farmers with honey (though he charges a small number of crop farms for pollination services). Bigger operations rent out hives and truck them all over the country.

Now it makes sense, she says, to start looking at other cropping systems and management practices. For example, corn, though not a pollinator-dependent crop, has large impacts on bees because the planting process generates clouds of pesticide-laden dust that can hurt downwind insects. The direction the growing biofuel industry takes may also have a big impact on resource availability for pollinators. Cornfields for biofuel feedstock offer neither food nor shelter for bees, diverse prairie-style plantings offer an abundance of both, and switchgrass falls somewhere in between.

The amount of complexity you find is determined by how much you look for, Grubbs notes. "The more questions you ask, the more questions you have," he says. "This is no exception." As he sorts through thousands of snippets of microbial DNA, he is looking for patterns and clues as to how outside influences may change hive microbiota. By focusing on the ecological impacts to the bees, he hopes to remove the emphasis on any individual stressor. "When you hear 'disorder' you think one cause," he says. "But it's a whole suite of things that set up susceptibility for something to take over the hive."

Kleinman and Suryanarayanan would like to see a regulatory and policy-making system that can accommodate multiple types of information, including laboratory studies, multivariate analyses and even empiric evidence such as beekeeper observations. They are currently developing a project to bring together groups with different backgrounds but common interests. By linking beekeepers, regulators and scientists with a range of expertise they hope to improve methods of understanding the true impacts of different factors.

"Everybody agrees to some extent that it's a multifactorial issue," Suryanarayanan says. "What's not resolved is which factors are more prominent and which factors are less."

However, he thinks it's clear that sound policy should arise from the intersection of these types of work. One key step is to rethink both the science and the sociology on which current regulatory policies are based. "Given the ambiguity of the evidence here, a precautionary approach would be the appropriate one to take as a policy matter," says Kleinman.

Both our bees and our agriculture depend on it. 🖁

