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Beebehavior and Beyond: Realizations in Research

—Sean Lombard (Editor: Brigid C. Casellini)

Most people would mistake the small carpenter bee *Ceratina calcarata* and its relatives for ants with wings, and I won't pretend that I could tell the difference before I spent a summer researching this particular bee species. We are conditioned to associate bees with three things: black and yellow coloration, honey, and stinging pain. The small carpenter bee species that I studied, however, did not exhibit a single characteristic from that list, which I found shocking, given that they are native to North America and locally abundant. Was I asleep during the lecture on the North American native insect ecosystems in biology class? Even if I reviewed every lecture since kindergarten, I doubt that information would have popped up. This was the first of many realizations stimulated by my initial experiences with research as an undergraduate.

The History and the Mystery

I became involved with research primarily because of the approach I took to college education: treat everything like a new experience, and go through every door opened for you. As an undeclared freshman, I could take whatever courses interested me, and it was through recommendations of professors like Dr. Charles Walker and Dr. Paul Tsang that I found out about and was accepted into the Research Experience and Apprenticeship Program (REAP), which offered a unique summer research experience for freshmen in the Honors Program. Curious, I stuck my head in the door... and was drawn through to the University of New Hampshire Bee Lab, headed by my REAP mentor, Dr. Sandra Rehan. Dr. Rehan's research focuses on the evolution of sociality in insects. *Ceratina calcarata* is a particularly fascinating species for an evolutionary biologist like Dr. Rehan, and ideal to study because their sub-social lifestyle provides important insight into the earliest stages of social evolution in bees and other insects.

C. calcarata are unique bees because they exhibit a combination of both solitary and social behaviors, therefore being classified as sub-social. In the sub-social system, *C. calcarata* mothers clean and care for their offspring and overwinter in the same nest with them. Part of the mother's interactions with her offspring includes an unusual behavior involving the eldest daughter.

According to research published by Rehan, when the mother makes her first pollen ball and lays an egg on it, she provisions the egg to have less sustenance than the other eggs (Rehan 2010). This results in a dwarf eldest daughter, who the mother nudges and forces to forage for pollen for the rest of her siblings. Generally the dwarf eldest daughter does not survive the winter, but her siblings do, thanks to her labor. This behavioral interaction is fascinating, because it can be viewed as a prelude to complex eusocial behavior.



Meet Darth Vader mixed with an overzealous bee researcher!
Author Sean Lombard.



The author in the field collecting bees.
(Photo credit: Jeremy Gasowsky.)

Unlike the sub-social behavior observed in *C. calcarata*, eusocial insects, exemplified by honey bees, exhibit overlapping generations, cooperative brood care and reproductive castes (i.e., individuals perform distinct tasks: queens reproduce and workers forage). The origins and maintenance of these castes are not known, but it is theorized that cuticular hydrocarbons (CHCs) on the exoskeleton of the bee may be partly responsible for maintaining the social castes. A recent study has gleaned from the presence of similar CHCs among queens of many insect species, that the CHCs have been conserved through roughly 150 million years of social evolution (Van Oystaeyen et al., 2014). This leads to the theory that the cuticular profile and behaviors of solitary bees were co-opted and repurposed during the evolution of sociality.

My REAP project had two purposes: first, to characterize the CHC profiles of *C. calcarata*, and second, to determine whether cuticular profiles are indicators of reproductive status by evaluating their behavior with CHCs present versus CHCs removed. Many bees needed to be collected, so I foraged for nests with other members of the lab early in the morning, before the bees were active. Back at the lab, I determined the sex of the bees, and if they were female, ran them in behavioral trials or obtained their CHCs by soaking them in pentane. The behavioral bees were either swabbed in pentane (to remove CHCs) or water (to act as a control) and randomly paired and placed within an "arena" of clear plastic tubing formed into a circle that forced the bees to interact. By recording previously

characterized behavioral patterns as either aggressive, avoidant, or following, we were able to analyze whether the removal of CHCs would influence the bees' behavioral patterns.

Realizations

One of the first realizations I made while working in the Rehan lab was an observation of the obvious similarities between the "caste system" of the lab and the organizational aspects of a social bee hive. In the lab there is one queen, Dr. Rehan, who manages all the projects and assigns different individuals to work on specific tasks. Besides being the "brains of the outfit" she also behaves in a maternal manner, checking up on all of the lab members to ensure that we have not come across any unsolvable problems. The entire lab formed a social hierarchy with each individual in charge of a specific task, yet prepared to help others produce more research data (rather than offspring, as the bees do).

The head worker is the lab manager, Wyatt Shell. He performs the function of general handyman and communications expert, as well as being a role model. Alongside Wyatt were the two graduate students: Nick Pizzi and Jake Withee. Nick and I worked closely together on the CHC behavioral research, and Jake focused on the gene expression in bees in aggressive social interactions. Though most of my time in the lab was spent working with Jake, Nick, and Wyatt, there were several others, including REAP student Liz Haas, who played important roles in the lab's function. The "hive" we worked in was a fantastic atmosphere, even during the most insanely overwhelming days, because every individual of the lab was focused, intelligent, and most importantly, had a sense of humor and a positive outlook.

I realized that one of the best parts of working in this lab is the capacity to have a hand in everyone's research. This glimpse of how scientists working on different experiments can influence their colleagues' projects was an important realization that greatly augmented my understanding of the general research community. Researchers have great impact on one another's work, including modifications of techniques,

evaluation of data, and even interpretation of results. Without a community of peers, the progression to understanding can take much longer.

Of course things did not always run smoothly in the bee lab. Challenges, such as gathering accurate data from a winged subject (nothing is worse than when your work flies out the window), and improvising when technologies failed, provided great learning opportunities on an almost daily basis. It took me a great deal of time to become comfortable with the notion that nothing goes exactly according to plan.

Though I was unable to analyze the data before the summer ended, the CHC profiles of the bees were all determined via gas chromatography, so the first aim of the project was met. The second aim is well on its way. It is unfortunately too soon to draw any firm conclusions, except that the experiment is worth repeating. Our sample size was a little too small and over too long a season to make any judgment that is more than suggestive. The project laid the foundation for continued research on the same topic and has given valuable base-line information for the success of the next field season.

Connecting the Dots

When I first applied to REAP, I had to write several essays, one of which required me to describe a realization I had made during school and consider its significance to my education. I wrote about an epiphany I had shared with both my chemistry and biology professors when I realized that the motion of biological structures in a cell is dependent on the chemistry of the molecules that formed them and the chemistry of the molecules they interact with. I don't claim that this is any grand realization, but it was incredibly exciting for me to notice that the classes I was taking built upon each other.

During the REAP project, I had this and many other thoughts in my mind as I analyzed bees and their behavioral interactions. I realized that it's not so surprising that I'd never heard of *Ceratina calcarata* before, not because they are any more or less important than any other part of the ecosystem, but rather because the ecosystem is so incredibly complex that I could listen to lectures to the end of my days and still not discover all that there is to know about them. Further, I could spend the rest of my life researching *C. calcarata* and never fully comprehend them as a species. I saw this when I began applying the data we had collected to the subject we were studying. It was easy to get lost in the numbers and forget what they really meant. Initially I was disappointed, as it seemed that all of the work I had done during the summer had been futile, until I came to what I believe was the most interesting epiphany.

Like how the lab was like a bee hive, how the different research projects all described the same species, even like how the CHCs were thought to be the mechanism for sociality, I couldn't help but notice that everything is the same. That biology is chemistry, is math, is engineering, is art, is history, ad infinitum. All of the fields of study that we have separated into their own "subjects" are like different "pictures" of the world, using different lenses to get a "better" view. All of the views are different from each other, yet they are of the same subject matter. It is easy to get caught up in the piles of "pictures" and lose sight of the unifying theme, the essence, that gives them value. The REAP experience has helped me to use the perspective that each subject creates (in my case science and research) in order to build a more comprehensive image of the world and the forces that function within it.

With that in mind, I encourage you to go through every door, do what it is that you enjoy, and don't get stuck within one mindset, because the world is an amazing place to explore.

This research experience would not have been possible without the generous donations made by Dana Hamel to UNH undergraduates, so I'd first like to thank him for the support. The Hamel Center is run through the hard work of its members, many of whom were incredibly helpful during my experience, including Dr. Paul Tsang, Molly Doyle, and Peter Akerman (to name a few). I'd also like to give many thanks to Dr. Sandra Rehan for all of her effort and support on my behalf, and for maintaining an awesome environment in the lab, without which I would have remained ignorant of the wonderful world of beesearch.

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Author and mentor bios

Sean Lombard is an enthusiast for research. A native of Dover, New Hampshire, he is majoring in biomedical science: medical and veterinary sciences, with a minor in genetics. A member of the University Honors Program, Sean will graduate in 2017 with a bachelor of science. He conducted research in the UNH Bee Lab as part of the Research Experience and Apprenticeship Program (REAP). He learned that “anything and everything is unique and fascinating. Research is a fun experience as a whole, and much like life, it has its ups and downs.” Sean decided to submit to *Inquiry* to gain experience in writing about science in a colloquial way, and to share his research with the greater academic community. He strives to become a physician or researcher through a joint MD/Ph.D. program.

Sandra Rehan is an assistant professor of biological sciences at the University of New Hampshire. Rehan specializes in animal behavior, comparative genomics, and evolution. In her first year of teaching at the University of New Hampshire, Rehan mentored twelve undergraduates on a variety of topics, and Sean is her first *Inquiry* author. Rehan is a seasoned field biologist, having studied native bees and their social behavior for over ten years. As a result, Rehan is very aware of the highs and lows of ecological research, but overall considers the research conducted by Sean during his first field season a remarkable success. “Learning to write for *Inquiry* is a great opportunity for undergrads of all disciplines,” says Rehan. “It is particularly valuable for biologists to learn how to communicate their research to a broader audience.”