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Breeding program for *Varroa* mite resistance in Ontario honey bee populations

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The mite *Varroa destructor* is considered the main threat to honey bee health worldwide. In Canada, particularly in Ontario, *V. destructor* is responsible for the majority of overwinter colonies losses. *V. destructor* parasitizes larvae, pupae, and adult bees, feeding upon the haemolymph (blood) and fat body of its hosts, *V. destructor* also vectors and transmits viruses such as the deformed wing virus (DWV) reducing the lifespan of infested bees. Most beekeepers control mite infestations using synthetic miticides, but the mites develop resistance to their active compounds, compromising their efficacy. Accordingly, it is necessary to have different control strategies. One way of reducing the impact of *V. destructor* parasitism is to breed *Varroa*-resistant strains of honey bees. If mite infestations are reduced through selective breeding, the damage caused by DWV may also be indirectly reduced. In this project a breeding program is being implemented in Ontario to select for lower and higher rates of *V. destructor* population growth (LVG and HVG, respectively), monitoring infection rates of DWV. Collaborative institutions are the Ontario Queen Breeders Association, the Ontario Beekeepers Association, and the U of G Honey Bee Research Centre. Preliminary results of this work show a six-fold difference in mite population growth between the LVG and HVG colonies. Additionally, DWV levels and winter colony mortality are significantly lower in LVG colonies than in HVG colonies. We will select additional generations of LVG and HVG colonies to analyze behavioural immunity and other parameters that could explain the divergence between the selected genotypes.

Keywords: *Apis mellifera*, *Varroa destructor*, deformed wing virus (DWV), selective breeding, mite population growth.

To pee or not to pee: how do female *Aedes aegypti* mosquitoes regulate anti-diuresis?

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Haematophagous insects, such as the female *Aedes aegypti* mosquito, face the challenge of excess ion and water intake after engorgement on a blood meal. To cope with this, adult female *A. aegypti* have a specialized excretory system that includes the Malpighian 'renal' tubules (MTs), which are under rigorous control by several neuroendocrine factors to regulate transepithelial movement of ions/water. Produced in the CNS, the mosquito anti-diuretic hormone is a member of the CAPA peptide family, which share homology to the vertebrate neuromedin U peptide. CAPA peptides inhibit fluid secretion of MTs stimulated by select diuretic factors, 5HT and DH₃₁ through the NOS/cGMP/PKG pathway. However, the anti-diuretic signalling mechanism and downstream cellular targets remain unclear. To investigate whether

the V-type H⁺-ATPase (VA) plays a role in CAPA inhibition, we performed fluid secretion assays in MTs treated with diuretics and bafilomycin, a known VA inhibitor. Bafilomycin significantly inhibited DH₃₁-stimulated fluid secretion while delayed responses were seen in 5HT-stimulated MTs. Alkalization of the secreted fluid in response to CAPA suggests inhibition of the proton pump, which may lead to constrained cation entry across the apical membrane of the MTs. Furthermore, adult female MTs treated with DH₃₁ resulted in an increase of VA activity, while tubules incubated with both DH₃₁ and CAPA had a lower VA activity resulting in activity levels comparable to saline control levels. Investigating the pathway of CAPA inhibition and its role in countering diuresis will help provide a deeper understanding of the critical process of diuresis and its signaling mechanism.

A revision of the genus *Scipopus*: Solving the *Scipopus* problem

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Scipopus Enderlein is a large group of Neotropical flies often recognized by their orange heads and black bodies. Despite their seemingly conspicuously appearance, the genus lacks a proper generic definition and diagnosis. The goal of my research is to identify synapomorphies to define the genus and to differentiate *Scipopus* from the other three closely related genera in the *Scipopus* group: *Pseudeurybata* Hennig, *Phaeopterina* Frey and one undescribed genus. Preliminary DNA barcode data shows these four genera as distinct clades. The *Scipopus* group, along with several other Micropezid genera are relatively easily separated on the species level but on the generic level show high occurrences of homoplasy and character overlap, making them an evolutionarily interesting group to study.

Evaluating methods used in Canadian bumble bee status assessments

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Jocelyn Armistead, Miriam Richards, Cory Sheffield

Bumble bees (genus *Bombus*) play important roles in the pollination of plants, both wild and crop, and maintaining healthy ecosystems. Many insect populations have shown declining trends in abundance in recent years, including some of Canada's bumble bee species. To be considered for federal protection in Canada, status assessments outlining a species' level of risk must be completed by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC). Six such bumble bee species have already been assessed. Set criteria must be met for a bumble bee species to be considered threatened, and determining this often requires assessing abundance and distribution. There are a variety of different ways to collect and analyze bumble bee abundance and distribution; my project aims to evaluate these different methods for their strengths and weaknesses. Additionally, some newer collection methods, such as community science photographs, blue vane traps and yearly roadside surveys, may produce data that could be a valuable asset to these status assessments, and I intend to evaluate them as well. The ultimate goal will be to provide recommendations about which collection and analysis methods produce the most reliable data for Canadian bumble bee status assessments. These assessments play critical roles in ensuring species receive the protection they need, so it is essential that they are based on the most accurate data and analysis. By

identifying the most accurate methods, my research will aid in helping to protect bumble bee species across Canada.

Farming fecund crickets: fruitful female fertility after feeding crickets royal jelly

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Canada is experiencing a boom in entomophagy. With industry-leading Entomo Farms offering delicious and safe edible cricket products over the past decade, and the recent multi-million dollar grant awarded towards the development of Aspire Foods cricket production facility, Ontario has developed into a central-hub for farming crickets. A primary goal of agricultural research is to increase yield, and while increased body size in cricket farms is desired, we have little knowledge on how to do it at such a large scale. Diet supplementation using honey bee royal jelly is a potential solution, as there is evidence that royal jelly enhances body size of other insect species. The mechanisms behind how royal jelly does this remain unclear. To determine the effect of royal jelly on a farmed cricket species, 96 *Grylloides sigillatus* were individually housed, and split into two treatment groups: half were reared on a commercially available cricket diet, while the other half were reared on the same diet mixed with 15% w/w fresh royal jelly. Body size and mass measurements were taken weekly for six weeks (approximate time to adulthood). We discovered a female-only effect of royal jelly on *G. sigillatus*: females fed the royal jelly diet grew to be 21% heavier, and this effect was driven by significantly longer abdomens containing 66% more eggs each compared to those fed the basal diet.

Mushroom-associated insects: diversity and host preferences

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Insects and fungi are two of the most diverse forms of life on Earth, and the fossil record suggests that insects have been using mushrooms for food and shelter for at least 400 million years. In temperate forests, many flies and beetles deposit their eggs in/on mushrooms, where their larvae will emerge into a nutritious food source and be relatively shielded from predators. However, the unpredictability of mushroom growth and the diverse physiochemical properties of fungi (e.g. toxicity and longevity) make the relationship between mushrooms and insects more unique and complicated than simple symbiosis or parasitism. Unfortunately, for many decades the study of these interactions has been hindered by the near-impossibility of identifying mushroom-associated insects in their egg or larval stages. As a result, mushroom-associated insects likely represent a large pool of undescribed insect species in Ontario. This study is the first in North America to use DNA-based methods to identify insects collected from wild mushrooms. We find that flies tend to lay eggs in soft, ephemeral mushrooms, whereas beetles more often lay eggs in tough, decayed fungi. However, there is wide variation in mushroom preferences among species, and more extensive sampling is needed to better characterize these interactions. We also report numerous insect species not previously known to interact with fungi, and many species that have never been recorded in Ontario. Importantly, we note that while most beetles were able to be identified to the species level from DNA

barcode sequences, reference databases for species-level identification of mushroom-associated flies are largely incomplete.

Why do caterpillars go "buzz"? Exploring the roles of vibratory communication in social *Drepana arcuata* larvae

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Larval stages of many insects species living in social groups must communicate for many contexts, including recruitment, shelter-building, and defense. However, the communication mechanisms used by social larvae have not been well studied. At present, there is little understanding about how and why social caterpillars communicate using vibrations. My research explores the role of vibratory communication amongst social larvae of the moth species, *Drepana arcuata*. Early instars of this species live in small groups and are reported to generate four vibrational communication signals: anal scrape, buzz-scrape, mandible scrape, and mandible drum. My research tested the hypothesis that these caterpillars use the buzz-scrape signal to announce their movement to conspecifics, to announce "friendly" vibrations. If this hypothesis is supported, then it is predicted that buzz-scrapes would be followed by movement of the signaller, and that the movement of the signaller causes vibrations. Videos with simultaneous laser vibrometer recordings of social groups were monitored. Ethograms were conducted to tabulate all behaviours that could be scored. These ethograms were then analyzed for the frequency of movement preceding and following a buzz-scrape, in solitary caterpillars and groups of two and four. Later, these frequencies were compared and analyzed using a paired t-test. The results confirm that buzz-scrape has a high rate of being followed by a movement, specifically crawling. Also, crawling movements followed by buzz-scrape produced vibration cues that could be detected by the laser vibrometer within the caterpillar shelter. These results support the hypothesis that caterpillars could alert conspecifics that the vibration is coming from a safe source, instead of a dangerous one. These caterpillars are small and have limited vision, so using these different vibratory communication signals could be necessary for their survival.

The effect of soil sand content on earthworm seed digestion and seed coat damage

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Earthworms are found throughout North America and can have profound impacts on seeds as seed dispersers and granivores. Earthworms use sand from their environment to help physically digest food, but it is unknown how different sand contents affect earthworm granivory. We hypothesized that higher soil sand content will increase the earthworm digestion rate of seeds and overall seed coat damage of undigested (egested) seeds. In this study, earthworms (*Lumbricus terrestris*) were kept on substrates varying in sand content by weight (0%, 25%, 50%), then fed garlic mustard (*Alliaria petiolata*) seeds. Egested seeds were visually examined and sorted into four damage classes. The results showed that sand content significantly influenced the seed digestion rate. As well, increasing sand content was associated with significantly higher seed coat damage levels. These results provide new information about the basic ecology of earthworm granivory and suggest how earthworm impacts on seed banks may vary between environments with different soil sand contents.