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Investigation of gene and protein expression based on Honey Bee (Apis mellifera) aging, flight experience, and behavior

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Honeybees undergo a process of adult behavioral development, spending their first 2-3 weeks working inside the constant environment of the hive. At about 3 weeks of age workers leave the hive as foragers who gather pollen and nectar. Previous research found that bees show an enormous decline in immunity as a result of their transition from regular hive jobs to more difficult foraging activities. Foragers can be forced to go back into hive-tasks, thus becoming “reverted nurses” which may also allow a reversal of immunosenescence. Understanding how this happens could prove to be useful because if there is flexibility in honeybee immunity it could lead us to a better understanding of the human immune response since the honeybee has a very similar genome to that of humans. I plan to use protein and gene expression analysis, along with other measurements to understand how forager bees change back into nurses and how this effects their immune response and their process of senescence and aging.
Investigation of gene and protein expression based on honey bee (Apis mellifera) aging, flight experience, and behavior

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Objectives
- Determine differences in gene and protein expression between age-matched bees of differing behavioral types (precocious foragers and typical age-nurses - 7-9 days old, typical age foragers and over-aged nurses - 19-32 days old, and reverted nurses - 24-26 days old)
- Measure global gene expression of these groups using microarray and quantitative real-time PCR technology to identify genes involved in aging
- Measure expression of different proteins from the genes identified using western blotting

Introduction

Honey bees, uniquely made up of three different castes: a reproductive queen, male drones, and sterile female workers. Adult bees undergo a process known as temporal polyphenism (age-related behavioral development) which causes the bees to change their tasks in the hive based on their age. One to two week-old nurse bees are responsible for feeding the larvae. After three weeks, older nurse bees help in the brood care. This trend continues as the bee ages, performing tasks such as foraging and nurse care, or moving into the colony. When workers reach the age of one month, they transition into foragers, which involves foraging for food and returning to the hive with nectar and pollen. This process continues until the bee reaches the age of four to five months, when it becomes a forager and stops foraging for the colony.

Table 1. Honey bee sampling groups

<table>
<thead>
<tr>
<th>Honey Bee Type</th>
<th>Behavior</th>
<th>Age</th>
<th>Flight Experience</th>
</tr>
</thead>
<tbody>
<tr>
<td>Queen</td>
<td>Out of hive</td>
<td>1-5 days</td>
<td>1 week</td>
</tr>
<tr>
<td>Forager</td>
<td>Out of hive</td>
<td>1-5 days</td>
<td>1 week</td>
</tr>
<tr>
<td>Nurse</td>
<td>Out of hive</td>
<td>19-22 days</td>
<td>1 week</td>
</tr>
<tr>
<td>Queen</td>
<td>In hive</td>
<td>3-6 days</td>
<td>1 week</td>
</tr>
<tr>
<td>Forager</td>
<td>In hive</td>
<td>18-20 days</td>
<td>1 week</td>
</tr>
</tbody>
</table>

Figure 1. Honey bee behavioral development

Materials and Methods

- Colony Management: We maintain our honey bees in a warm, temperature-controlled room. We maintain the colony by providing fresh water and sugar syrup for the bees. We also monitor the colony size and provide additional brood rearing when necessary.
- Biometric data: We measured the size of the honey bee colony by collecting data on the number of bees, brood, and honey stores. This data was used to determine the colony's health and vigor.
- Microarray analysis: We used Affymetrix microarrays to analyze gene expression in the honey bee colony. These microarrays contain probes for all known honey bee genes. We analyzed the expression levels of genes related to aging and flight.
- Western Blotting: We used western blotting to validate the microarray results. We extracted protein from the honey bee colony and separated the proteins using SDS-PAGE. The proteins were transferred to a nitrocellulose membrane where they were probed with antibodies specific to the proteins of interest.

Acknowledgements

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References


Future Directions

- These experiments are ongoing – in the next few weeks we will be dissecting collected bees and running the next arrays.
- Expression of genes of interest will be verified using real-time quantitative PCR followed by reverse transcription-quantitative PCR (RT-qPCR).