NC STATE UNIVERSITY

Wolfpack's Waggle

NC State Apiculture Program Newsletter

Dedicated to the dissemination of information and understanding of honey bee biology and management

Issue 4 | Oct 2021

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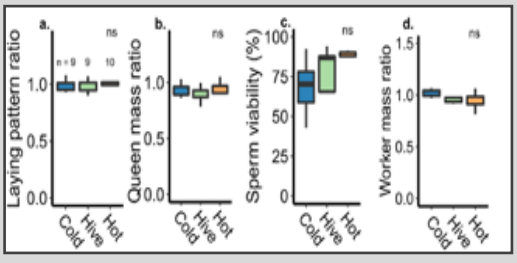
What have we been up to?

We are currently wrapping up a second challenging season, but we have been coping with the various constraints placed on all of us. Jennifer used the relatively open field season to practice and refine her skills on instrumental insemination. Brad conducted several more experiments on drone reproductive quality including a flight experiment using RFID tags. Ali was exceptionally busy in both the field and the lab, conducting both her main projects as well as several side projects in collaboration with Canadian beekeepers. Esmaeil was also very busy in the field and he is currently wrapping up his analyses in the lab before his big move to the USDA and Mississippi State. Sharon put in a tremendous effort to re-launch our latest BEES Academy this fall, in between helping in the field on various projects. Erin has been sampling her experimental hives every other week since March, so the number of samples she has obtained is quite substantial, but supply-chain issues have stalled her ability to analyze all of them because of a lack of lab supplies. We're happy to be joined by three new undergraduates in the lab—Morgan, Kaitlyn, and Glenn—now that undergraduate research is not as restricted as last season. We look forward to completing our various studies and writing them up for publication soon!



New publication clarifies and refines how temperature stress affects queens

A new paper by Ali McAfee follows up on her previous work to demonstrate that, while extreme temperatures can sterilize queens, they can also be resilient to thermal stress.

HONEY BEE QUEEN AND DISEASE CLINIC | BETTER DATA BETTER BEES

Quality Assurance

Morphometric Analyses: multiple measures of queen or drone, body and reproductive tract (rearing quality)

Semen Quality: total sperm count, and sperm viability in queens (mating success), or drones (mating potential)

Quality Report: a "grade" report of a queen or drone's reproductive quality for your quick interpretation

Troubleshooting

Mitotyping for Africanization: genetic analyses of maternal ancestry as African or European using population genetic techniques and markers

Pathogen Screening: identification of presence and relative levels of ABPV, BQCV, DWV(A&B), IAPV, LSV, Trypanosomes, and both Nosema species. Additional and custom pathogen targets available upon request.

Genotyping Analyses: full assessment of paternity for up to 48 workers and an estimate of queen mating frequency

Custom Collaboration

This highly-tailored collaboration involves custom experimental design, analyses, and interpretation. This unique partnership between science and industry has been utilized to:

- Test the impact of various agrochemicals
- Assess the effects of banking on queen quality measures
- Evaluate novel management practices' improvements in queen mating quality
- Observe the effects of shipping on queen health and sperm quality

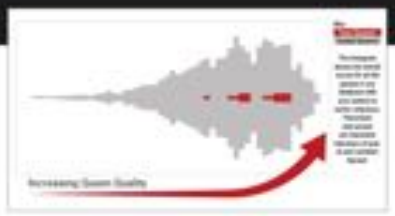
Strong Research Foundations

Established as a natural extension service leveraging basic and field honey bee research at NC State, the clinic has worked to improve colony health for over 10 years.

Queen and Disease Clinic Pricing

Five Sample Minimum • Bulk Pricing Available

ANALYSIS	PRICING	SAMPLES TESTED		
		QUEENS	DRONES	COLONIES
Reproductive Quality	$24.00	✓	✓	
Standard Pathogen Screen	$55.00	✓	✓	✓
Apiary Pathogen Screen	*$220.00			
Mitotyping (Africanization)	$25.00	✓	✓	✓
Genotyping (Mating Number)	$320.00			✓



Custom Disease Screening

Additional and custom pathogen targets available upon request.

Your Bees • Your Data

Any results or interpretations from our work is held in the strictest confidentiality and anonymity

Lab Spotlight: Esmaeil Amiri

Since early 2016, Iran native Esmaeil Amiri has been an NRC Postdoctoral Fellow at the University of North Carolina at Greensboro. Housed in Olav Rueppell's lab, he has been associated with our program as well through collaborative research on honey bee stressors, virus pathogens, and factors that affect the size of honey bee eggs. Esmaeil's hard work has certainly paid off, as he will be leaving us to start his own program with the USDA-ARS and Mississippi State University in Stoneville, MS. We have every confidence that he will continue his good work and look forward to him remaining in the honey bee research community. All the best and thanks, Esmaeil!

Queens exhibit variation in resilience to temperature stress

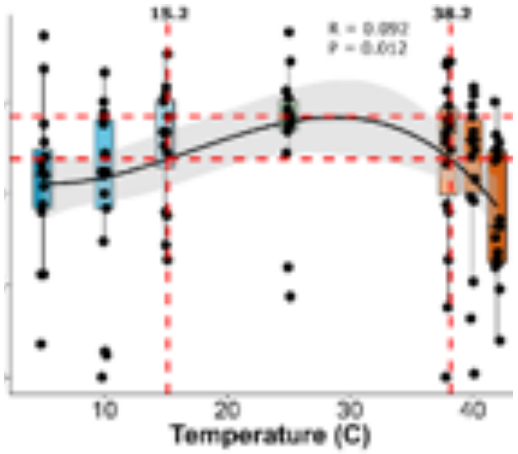
In another paper in a series of studies on the effects of temperature on queens, Ali McAfee has shown that not all queens are affected in the same way. So while deviant temperatures, both hot and cold, can kill the stored sperm in a queen's spermatheca, her new paper published in *PLoS ONE* suggests that some queens can actually be surprisingly resilient to such temperature swings.

As the sole egg layer in a colony, the queen has a clear function and important role in keeping the colony healthy and productive. Her ability to do so is predicated on numerous factors, but perhaps most importantly is her ability to lay eggs and fertilize them (that is, her reproductive capacity). Anything that jeopardizes her ability to fulfill her main role in the colony should clearly be avoided.

Previous research out of our lab has been investigating how the handling of newly mated queens can affect their down-stream reproductive quality. Specifically, we have looked at how temperature exposure to queens during shipping and handling might inadvertently affect the stored sperm in queens' spermathecae. In a previous study led by Postdoctoral Fellow Dr. **Alison McAfee**, queens exposed to high temperatures (above brood-nest temperatures) can reduce their sperm viability, but perhaps more importantly so can low temperatures (Figure 1; right). Beekeepers often avoid overheating queens because it can kill them outright, but they often over-compensate and keep them chilled. While cooling queens might not risk them dying, it can result in their partial sterilization and thus not being able to lay fertilized eggs very long.

One of the hallmarks of good science, however, is replication and repeatability. In other words, it's important to show how robust any given cause-and-effect relationship might be. As such, Ali then followed up on her study to conduct a completely independent field study to see how temperature stress might impact queens and their entire colonies. Little is known about how thermal stress may directly impact queen performance or other maternal quality metrics, such as queen mass, egg laying rate, and development of embryos within eggs. In a blind field trial, we recorded laying pattern, queen mass, sperm viability, and average callow worker mass before and after exposing queens to a cold temperature (4 °C, 2 h), hot temperature (42 °C, 2 h), and hive temperature (33 °C) to serve as a handling control. We then used proteomics to investigate potential vertical effects of maternal temperature stress on embryos, as well as to measure the abundance of previously determined protein markers for temperature stress in the queens' spermathecal fluid.

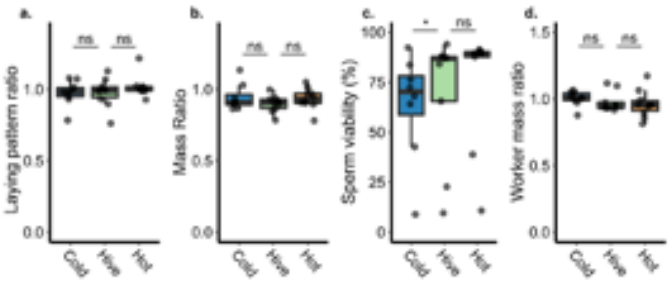
Somewhat surprisingly, we found no significant effect of abiotic stress on any of the metrics we recorded. We did not



detect lasting maternal effects of temperature stress, and the queens themselves appear to be highly stress tolerant (Figure 2; below).

There are several potential explanations for this null result, but we suspect there are two things that may be going on here: (1) not all queens are equally sensitive to temperature stress, and the queens in the current study were more resilient than those in previous studies; and (2) not all sperm are equally sensitive to temperature stress, and the sperm stored in these queens were particularly resilient.

Many researchers have shown that not all queens are the same in many respects (e.g., genetics, rearing environment, adult environment), so it may not be surprising that some queens are susceptible and others are tolerant of temperature swings. Perhaps more interestingly, it is also possible that variation of thermal tolerance of the sperm themselves (i.e., "fragile sperm") could be causing these discrepant results. Similar to variation in adult queens, it is plausible that not all sperm are the same; indeed, not all drones are the same—



Apiculture Online (Continued)

they vary significantly in their reproductive quality, colony environment, and their thermal tolerance. It stands to reason that if drones of variable quality may still inseminate a queen, how well their respective sperm tolerate stressful conditions may similarly vary. As such, the queens in the present study may have happened to mate with drones that have robust sperm, whereas those in other studies may have mated with drones whose sperm that were more "fragile." Such tolerance could further explain why we did not see the expected effect of temperature, and warrants further investigation.

Queen failure is a common problem in beekeeping operations, but surprisingly little is known about the underlying factors. We confirmed that cold stress reduces stored sperm viability, but surprisingly, this could not be confirmed for heat stress. Some of these observations are contrary to previous findings and suggest that there is variation in queens' abilities to tolerate extreme temperatures. Cumulatively, the data acquired consistently demonstrate resilience of the queen herself to temperature stress, even if it may decrease the viability of the sperm she stores in some cases. Future research should focus on longer-term experiments to test queen quality after stress throughout a beekeeping season, as well as investigate the drivers of variation in sperm viability responsiveness that has been reported in the literature. Extreme temperature fluctuations should also be investigated (e.g., from extreme cold to extreme heat) or longer durations of milder temperatures, which may be realistic scenarios to occur as the climate continues to change.

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Esmaeil Amiri - NRC Postdoctoral Fellow *(UNCG)*
Brad Metz – NC State Research Associate
Alison McAfee – NCERC Postdoctoral Fellow *(UBC)*

Undergraduate Researchers
Morgan Risko (media intern), Kaitlyn Sage, Glenn Cameron (Meredith College)

Support the NC State Apiculture Program!

The Apiculture Science fund-raising efforts operate under the auspices of the North Carolina Agricultural Foundation, Inc. a 501(c)3 organization. You will receive an official receipt for your donation.

A Gift Toward Emerging Needs

Consider supporting the program with a gift that would go toward the current area of greatest importance. Flexible funding enables the Apiculture Program to address critical needs as they emerge, often enhancing the program beyond what would be possible through restricted grant funding. Funding of any amount, from $10 to $10,000, will be extremely helpful.

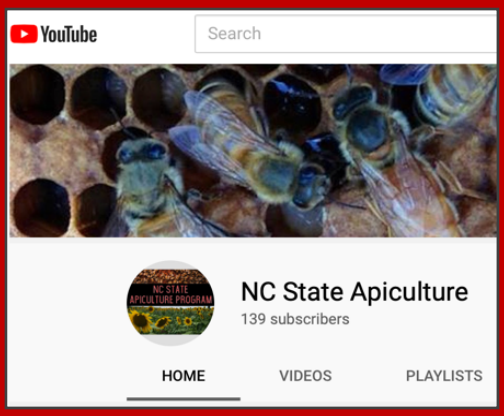
Gift-In-Kind

The Apiculture program is always seeking creative solutions to its material needs. If you have surplus equipment or other non-monetary assets to give (e.g., gently used honey extractors, microscopes, even vehicles), please consider donating them to the program. You will receive credit for the monetary value of the gift and the gratitude of our faculty and students.

Estate Gift

If you are interested in planning an estate gift to benefit Apiculture, please let us know! We can provide you with the tools you and your attorney will need to ensure that your wishes are fulfilled. Please go to our website for more information: www.ncsuapiculture.net

go.ncsu.edu/apiculture



Try us on YouTube!

For several years now, we've been adding video content onto our very own YouTube channel. From beekeeping advice to some of our latest research, this *free* resource is perfect to plug into your monthly beekeeper meetings or to watch during your downtime. Subscribe and view us today!

Congratulations Alison!

Ali McAfee, a postdoc in our program housed at the University of British Columbia in Dr. Leonard Foster's lab, has been awarded the highly coveted L'Oréal Canada For Women in Science Research Excellence Fellowship. She is only one of two awardees in all of Canada, so the honor is very prestigious and certainly deserving of all of her excellent work. Well done, Ali, we're proud of you!

Random Notes

New Publications

McAfee, A., D. R. Tarpy, and L. J. Foster. (2021). Queens exhibit variation in resilience to temperature stress. *PLoS ONE*, **16**: e0255381.

Presentations

We held our first 'Intermediate' BEES Academy since early 2020 in Union County on October 2nd-3rd. While we had a couple notable hiccups, it was well received by the attendees. It was also the first time we have held a BEES Academy on a Saturday-Sunday (rather than a Friday-Saturday). While COVID has continued to keep everyone guessing as to the future of in-person extension events, we hope to continue offering similar trainings at both the intermediate and advanced levels, so stay tuned!

David has also given presentations to the Tennessee Valley Beekeepers Association and the Huguenot Beekeepers in Virginia. Jennifer also attended the Eastern Apiculture Society conference in Kentucky in August, where she was one of the main attractions in the apiary.

Finally, we have continued to hold our monthly webinar series Apiculture Online—Hive Chat with NC State. We've had terrific topics, guests, and interactions with our online audience, and we have been posting the recordings on our YouTube channel. While we have wrapped up for this season, we will see if the demand is still high in 2022 to resume them.

Welcome aboard!

We've been joined this fall semester by three new undergraduates. First, **Morgan Risko** is our newest Media Intern and has been helping us with digital marketing, FaceBook posts, and communicating through MailChimp. Second, **Kaitlyn Sage** has joined us in both the genetics lab and in the field, which suits her well as one of the co-Presidents of the NCSU beekeeping club. Finally, **Glenn Cameron** is our first-ever undergraduate researcher from Meredith College, where she is doing an independent project on mapping drone congregation areas (DCAs), which is quite ambitious. We look forward to working with all of them going forward!

Teacher's Corner: Courses at NC State

We initially attempted to teach ENT 203 entirely online again this semester, but we were compelled to offer it in-person as we typically have in the past. While we have had several COVID cases among the students, they have been largely contained with no significant outbreaks. The course has been going extremely well so far thanks in large part to the two TAs (**Melissa Pulkoski** and **Yu-Hui Wang**), and the students have been exceptionally engaged asking lot of excellent questions. This will be the last time we offer ENT 203, although we hope to continue the course in the new Applied Ecology curriculum as AEC 203, so stay tuned!

go.ncsu.edu/honeybees

Tarpy's Back Page

As I have been discussing in previous newsletters, our field lab on the Lake Wheeler Research Farm complex was partially condemned and as a result we are needing to relocate. We are fortunate to be able to move into another structure on the other side of the farm, previously used as an event space for picnics and social gatherings. It was formerly used as a retreat for the Dorothea Dix hospital until it was donated to the college, and as such it is commonly referred to as the Dix Facility.

The personnel at Lake Wheeler have been giving the main building a make-over in time for our move, and we're currently formulating a plan to move most of our lab equipment, freezers, and incubators over to the new site. One potential difficulty is that we won't have much space to store any of our beehive equipment, so we're in the process of determining where and how to maintain the hive equipment. We also likely won't be able to have as many active beehives at the new site, but we should be able to maintain our other out-yards on the farm complex and elsewhere.

The good news with moving to the Dix Facility is that we hope to resume some extension trainings at the new building. We had stopped doing so at the original site because it was so unsightly and less conducive to hosting beekeepers, but we hope the new space will renew our ability to have field days or other events. Stay tuned as things continue to develop!