Developing Specialized Tech-Transfer Team to Help Improve Honey Bee Genetics and Stocks

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Objectives:

1. Establish a specialized tech-transfer team as a pilot project to work directly with bee breeders in California to improve stock selection, enhance genetic diversity, engage in disease - and parasite-related diagnostic evaluations, and participate in collaborative and interdisciplinary research on key issues.
2. Use the pilot project experience as a basis for establishing a second tech-transfer team elsewhere in the United States.

Interpretive Summary:

Bee breeders, a small but extremely important subset of beekeepers, provide the genetic material for beekeepers nationwide through the sale of queen bees. Given the serious and chronic health problems facing bees and the increasing demand for pollination services, it is critical to provide bee breeders assistance with the production of genetically diverse stocks of bees that can defend themselves against parasitic mites and diseases.

The almond industry commonly relies on services provided by professional, independent crop consultants. These professionals help implement scientific and technological advances to enhance environmental sustainability and profitability (Jones, 2004). Based on this idea of a crop consultant, we established a Honey Bee Tech-Transfer Team for bee breeders in Northern

California. This is the first time the US beekeeping industry has utilized independent, professional consultants in an analogous manner. We established a Tech-Transfer Team to assist beekeepers with implementing new strategies by providing the following services:

1. Assisting with stock selection and breeding for resistance traits
2. Enhancing genetic diversity in bee stocks
3. Disease and pest diagnostics and information on integrated management practices to reduce chemical treatments
4. Facilitating cooperative research on relevant topics (e.g., quality of queen and drone bees; nutrition)

The Tech-Transfer Team consists of three professionals that work one-on-one with 16 queen breeders. The Team organizes and conducts on-site visits with bee breeders throughout the year to assess colonies for strength and health, to collect disease and pest samples (namely *Varroa*, *Nosema*, and viruses), and to perform a test for hygienic behavior on potential breeder colonies. Selecting for hygienic behavior is a relatively easy, quantifiable way to select for a disease resistance trait, and colonies with hygienic behavior have natural resistance to brood diseases and the *Varroa* mite, while still being productive and gentle (Spivak and Reuter, 1998). The Tech-Transfer Team encourages selection for hygienic behavior only from colonies pre-selected for other desirable traits.

The Tech-Transfer Team generates reports for the beekeepers that contain the data for each colony, including hygienic behavior, colony characteristics, and disease and mite levels. The reports are given promptly to the bee breeders as an aid in selecting the most disease and mite resistant breeder colonies from which to raise queens and for making educated treatment decisions.

To correlate colony health with management practices, the Tech-Team collects extensive data on management practices. The management data will be used to look at the best times of year to treat or to feed pollen and sugar, to determine treatment thresholds, and for other relationships between management and colony health. The management and disease sample data will be included in the BIP database. The BIP database is a vast and growing archive of disease survey records, where the individual identities of beekeepers are protected. Beekeepers and researchers will be able to access anonymous aggregate summaries of these records to give context to disease loads in specific seasons and locations.

The Tech-Transfer Team also responds to any beekeeper with an emergency situation, namely a large bee loss. The Team has the capability to respond quickly to sample bees for virus, pesticides, or disease, and properly ship the samples to research labs for analysis. In cases where an immediate response is not possible, the Tech-Transfer Team can instruct the beekeeper on how to take samples and where to ship the samples, or how to get in touch with the County Agricultural Commissioner.

In May 2011, the Bee Informed Partnership (BIP) (beeinformed.org) started supporting the Tech-Transfer Team. The Tech-Transfer Team now operates under the guidance and support of the project leader and project collaborators of BIP. In conjunction with BIP, a second Tech-Transfer Team was initiated in May 2012 for commercial beekeepers in Minnesota and North Dakota. In addition, the project leader in conjunction with an economist funded by BIP will develop a five-year business plan for the Tech-Transfer Teams.

Materials and Methods:

Potential breeder colonies were sampled for *Varroa*, *Nosema*, hygienic behavior, and viruses. The virus samples were analyzed by Dr. David Tarpy's Lab (NC State). *Varroa* and *Nosema* samples were primarily processed at the USDA Bee Research Laboratory (Beltsville, MD).

To quantify *Varroa* and *Nosema* levels, samples of 300 adult bees were collected in alcohol jars off a comb with open larvae. To estimate the *Varroa* infestation, the alcohol samples were vigorously shaken for about one minute, the bees were thoroughly rinsed, and the mites strained using a wire basket, and then counted. For each sample, 100 bees were counted out and weighed and the weight used to convert the total bee weight into a number of bees. Dividing the number of mites by the number of bees and multiplying by 100 gave mites per 100 adult bees.

Nosema levels were estimated by using the reserved 100 of the 300 sampled bees. These bees were put into a plastic bag, thoroughly mashed, and 100 ml of water was added. The water and bees were mixed into slurry, and a pipette was used to collect a few drops. The drops were put on a hemocytomter slide with 25 grid squares (normally used for counting blood cells) for counting under a 400-power microscope. We counted the number of spores were counted on five of the grid squares and took an average, then a conversion factor was used to estimate the number of *Nosema* spores per bee (Cantwell, 1970). We assumed the spores found were *N. ceranae* and not *N. apis*, since *N. ceranae* is much more prevalent in the US (Klee, 2007), and is more virulent (Higes, 2007).

To test for Hygienic behavior, a comb with capped pupae was removed from each colony, a 3" PVC tube was inserted into the comb over the pupae, then 10 oz of liquid nitrogen was poured into the tube to freeze-kill the pupae. Before the addition of nitrogen, the number of cells that were not sealed pupae was counted (there are 160 cells in a 3" circle). After the tube thawed, it was removed and the comb placed back into the colony. The comb was checked in 24 hours and the number of cells not completely cleaned by the bees and the number of cells partially cleaned were counted. Since the number of initial and final cells is known, each colony can be ranked for hygienic behavior. The more cells uncapped and pupae removed, the more hygienic the colony. A hygienic breeder would completely clean out 90% or more of the sealed pupal cells.

During sample collection and hygienic testing, a number of characteristics of each colony were recorded: number of frames of bees, queen status, hive configuration (number of hive bodies and the size), brood pattern, temperament, weight, color, and the presence/absence of apparent adult or brood diseases. The Team also records information about the following management practices: migratory practices, treatment types (for *Varroa*, *Nosema*, and brood diseases), treatment dosages, seasonal applications, protein usage, sugar usage, fumigants, and other additions to the colony.

Results and Discussion:

1. Assistance with stock selection and breeding for resistance traits

The Tech-Transfer Team samples potential breeder colonies for *Varroa* and *Nosema* with the goal of the beekeeper being able to choose breeder colonies with the lowest pest and disease levels. Disease sampling of potential breeder colonies is done to differentiate colonies that may have natural resistance to *Varroa*, *Nosema*, or viruses. Colonies with low levels compared to the other colonies around them may have heritable resistance, and the beekeeper can choose colonies with apparent resistance as queen mothers.

From December 2010 – June 2012, the Tech-Team has taken 5,377 *Varroa* samples, 5,503 *Nosema* samples, 634 virus samples, and tested 1,635 colonies for hygienic behavior. After sampling each beekeeper, a report with all recorded data and sample results was given back to the beekeeper within 7-15 days.

The Tech-Transfer Team encourages bee breeders to breed from colonies that score over 90% hygienic. From 2011 to 2012, the proportion of colonies testing 90% hygienic or better increased (**Figure 1**), where the average in 2011 was 18% (±17% Stdev) and 30% (±21% Stdev) in 2012. This indicates that the hygienic trait is now more abundant within the bee breeders own stocks and that the selection process is working.

The Tech-Transfer Team has received positive feedback from the queen producers about increasing the frequency of hygienic behavior in their own stocks. The queen breeders are using the Tech-Transfer Team's results and choosing the more hygienic colonies as queen mothers. The breeders understand the need to saturate mating areas with drones exhibiting this trait. Continued selection and propagation of productive, healthy hygienic colonies will ensure that naturally mated queens could be certified and marketed as being hygienic within another 2-4 years.

2. Disease diagnostics

Beekeepers report that the disease information provided from the Tech-Transfer Team has helped in making treatment decisions. Beekeepers have reported using fewer treatments, especially with Fumigillian, when disease levels are low. One beekeeper stated that the Team has saved his operation $80,000 (McNeil 2012).

Participating beekeepers are provided with a summary report showing their average levels for *Varroa*, *Nosema*, and hygienic behavior anonymously compared to the levels of the other participating beekeepers. This summary report was provided to give the beekeepers context for their disease and hygienic behavior levels. In the future, we would like to connect the levels of disease with management practices, and, if given permission, share among bee breeders the management practices that are most effective and use the fewest chemical treatments.

3. Enhancement of genetic diversity in bee stocks

Maintenance of a genetically diverse population of foundation stock provides the potential to maximize success in selecting desirable traits, while reducing inbreeding. Commercial queen producers can benefit from selection conducted by university and government breeding and stock importation programs, while still maintaining their own identity in the queens they provide through a collaborative and coordinated selection program. The WSU/UCD stock importation program has imported germplasm from selected lines of Italian and Carniolan stocks from Europe and has incorporated the genetic material into established commercial lines specifically established to increase their genetic diversity. The imported stocks are being maintained and tested at both universities in a tightly controlled breeding program that includes hygienic behavior and the characteristics many beekeepers favor (gentle temperament, rapid growth, reduced susceptibility to *Nosema* and other diseases etc.).

The Tech-Transfer Team works with S. Cobey and S. Sheppard to incorporate genetic lines of New World Carniolans and Caucasians into already established bee breeder stock by providing assistance with coordinating a drone saturation program, and conducting evaluations of the inseminated queens to ensure stock quality. To date, 112 inseminated breeder colonies were evaluated for general fitness to see if the colonies would be adequate to raise daughter queens.

4. Facilitate cooperative research on relevant topics

The Tech-Transfer Team's presence in CA helps facilitate collaborative, interdisciplinary research on important issues to beekeepers, such as queen quality and longevity and the nutritional state of colonies. Researchers or beekeepers from other states wanting to set up experiments in collaboration with bee breeders can rely on aid from the Tech-Transfer Team.

The Tech-Transfer Team collaborates with Dennis vanEnglesdorp (Maryland State University) for the implementation of the Agriculture and Food Research Initiative (AFRI) grant funded Bee Informed Partnership. The Tech-Transfer Team was incorporated into the grant due to the recognition that the Tech-Team offers insight into the bee breeding industry through on-the-ground work. BIP benefits from the fieldwork with beekeepers, and the Tech-Transfer Team benefits from having access to the BIP team of researchers, the BIP database, Bee Research Laboratory diagnostic lab, and funding two personnel for the Team.

In April 2012 one of Dr. Reed Johnson's (Ohio State) students studied the effect of a fungicide on queen development and was hosted by the Tech-Transfer Team.

Founding a Second Tech-Transfer Team

In May 2012, a second Tech-Transfer Team was established in the Midwest and supported by BIP and other funding. K. Lee (M. Spivak's former student) established the CA Tech-Transfer Team and in May 2012, began the Midwest Tech-Transfer Team to serve migratory beekeepers and queen breeders primarily in North Dakota and Minnesota. There have been multiple requests to further expand to the North East, South, and South East. Eventually, we look to establish multiple teams across the States working to assist migratory beekeepers, commercial queen breeders, or beekeepers raising local, regionally-adapted queens from survivor (untreated) stock.

Economic Plan

The BIP economist is in the process of developing a long-term business plan for the Tech-Transfer Team to become self-sustaining. In the initial phase of the project, The Team asked for a fee-for-service to offset a portion of the costs of travel and supplies, where the beekeepers provided $500 the first year and $1,000 the second year. In May 2012, the participating beekeepers made a verbal commitment to funding the salary and benefits to hire another Tech-Team member.

Research Effort Recent Publications:

McNeil, M.E.A. 2011. The Bee Informed Partnership: a vast collaborative effort to find out what's up with the bees and your bees in particular. American Bee Journal 151(7): 677-681.
McNeil, M.E.A. 2012. Boots on the Ground: A radical shift in the interface between research and the real world. Bee Culture 4: 37-40.
Spivak, M. 2011. Laying groundwork for a sustainable market of genetically-improved queens: The bee team. Managed pollinator CAP Update.American Bee Journal 151(5): 483-485.
Lee, K. Bee Informed Partnership: beeinformed.org/blog/. July 28, 2011. Titles: Selection for the Future is Underway, Katie Reports from the Field, Catching Queens, Drone Comets, Nosema, Cigar Smoke, Honey Bee Instrumental Insemination with Sue Cobey, Weather in Northern California, Origins, Present, Future, Rice Country, Hygienic Behavior, How to Make a Bee Beard, Bee Squad, Drone Fishing, Honey Extraction, Grafting, Propolis and Human Health, Test for Varroa, Pesticide Kill, The Promised Land, Propolis and Bee Health, California Bee Breeders Conference, Samoan Honey, Hive Tool to the Rescue, Honey without Pollen, The Promised Land 2: the airing, Tying Knots, Coconuts, Hive Beetles in Paradise, American Beekeeping Federation Conference, Pesticide Cost-Share, Bees in Bare Almonds, Spring Sampling Season, Queen Season, Eric Mussen, Midwest Bee Team, and Smoker Plug.
vanEngelsdorp, D., D. Caron, J. Hayes, R. Underwood, K. R. M. Henson, A. Spleen, M. Andree, R. Snyder, K. Lee, K. Roccasecca, M. Wilson, J. Wilkes, E. Lengerich, and J. Pettis. 2012. A national survey of managed honey bee 2010-11 winter colony losses in the USA: results from the Bee Informed Partnership. Journal of Apicultural Research 51: 115-124.

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Klee, J., et al. (2007) Widespread dispersal of the microsporidian *Nosema ceranae*, an emergent pathogen of the western honey bee, *Apis mellifera*. Journal of Invertebrate Pathology. doi: 10.1016/
Jones M. 2004. Role of the Private Crop Consultant in Implementation of IPM. Radcliffe's IPM World Textbook, Regents of the University of Minnesota http://ipmworld.umn.edu/chapters/mjones.htm
McNeil, M.E.A. 2012. Boots on the Ground: A radical shift in the interface between research and the real world. Bee Culture 4: 37-40.
Spivak, M., and G.S. Reuter. 1998. Performance of hygienic honey bee colonies in a commercial apiary. Apidologie. 29: 291-302.

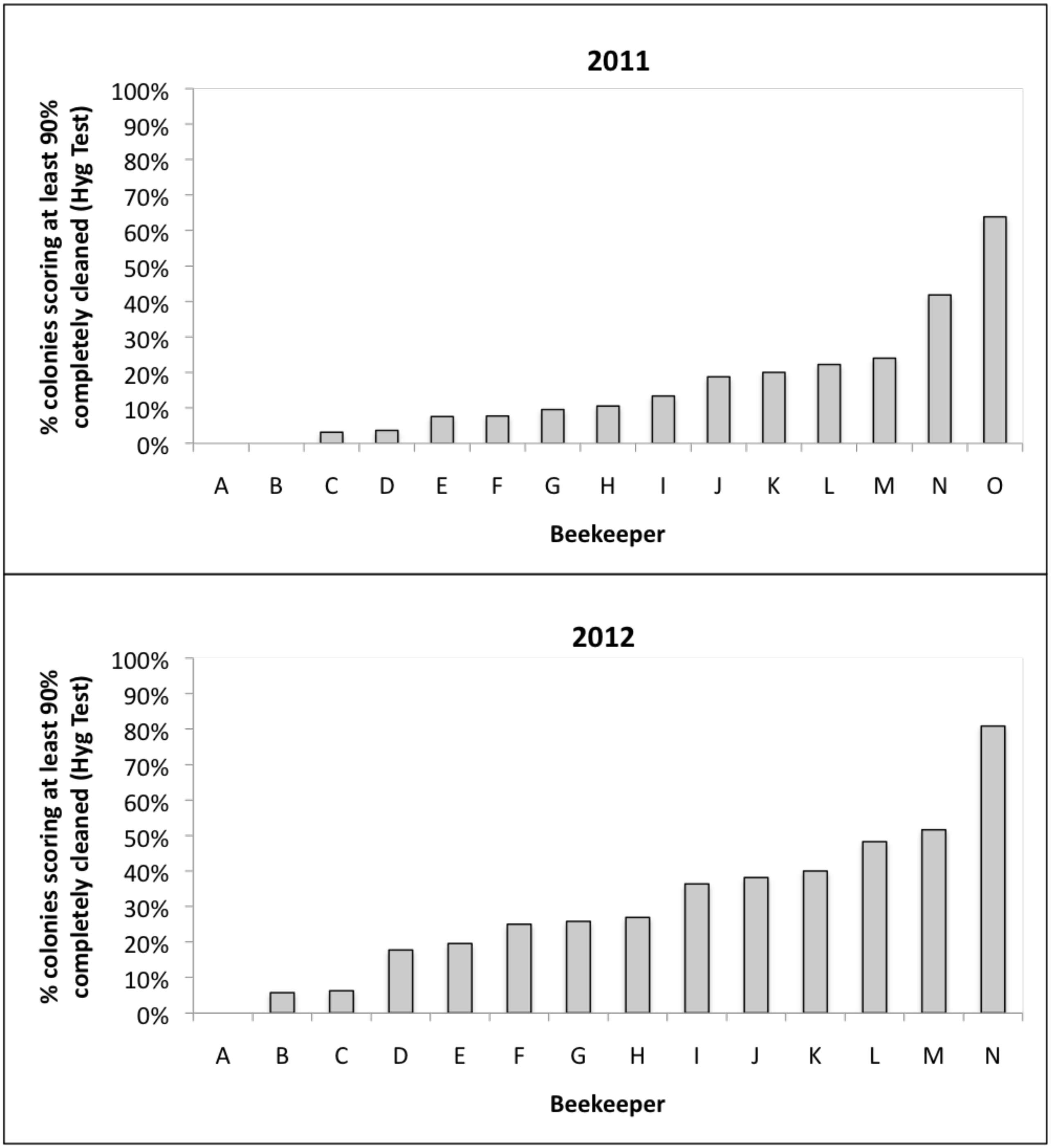

Figure 1. The percent of colonies scoring 90% hygienic or better in 2011 and 2012. Each letter indicates a beekeeper, and each bar is the percent of colonies scoring 90% hygienic or better for that beekeeper.