

USDA Honey Bee Pests and Diseases Survey Project Plan for 2012

Comprehensive Objective

A national survey of honey bee pests and diseases has been funded annually since 2009 by the USDA Animal Plant Health Inspection Service (APHIS). This survey is being conducted in an attempt to document which bee diseases, parasites, or pests of honey bees are present and/or likely absent in the U.S. Specifically, this survey will attempt to verify the absence of the parasitic mite *Tropilaelaps* and other exotic threats to honey bee populations (e.g., *Apis cerana* and Slow Paralysis Virus). To maximize the information gained from this survey effort, collected samples will be analyzed for other honey bee diseases and parasites known to be present in the U.S. This cross-country survey will be the most comprehensive honey bee pest and health survey to date, and provides essential disease and pest load base line information. This information will help place current and future epidemiological studies in context and thus may indirectly help investigations of emerging conditions such as Colony Collapse Disorder (CCD). Coordination of this survey is in collaboration with USDA Agricultural Research Service (ARS) Bee Research Lab (BRL) and the University of Maryland (UMD).

Primary Objective – Exotics

Tropilaelaps spp., an Asian parasitic mite (several species in the genus *Tropilaelaps* are recognized) of honey bees, feeds on honey bee brood. Their parasitic feeding actions vector viruses, weaken or kill parasitized brood, and can cause infected colonies to abscond which spreads the mites to new areas. *Tropilaelaps* mites can complete their lifecycle in one week, and thus this mite can outcompete the Varroa mite when both mites are present in a hive. Currently, there are no known *Tropilaelaps* mites in the U.S.

The exotic *Apis* species *Apis cerana*, or Asian honey bee, and Slow Paralysis Virus are also NOT known to be in U.S. apiaries; this survey will help confirm their absence. *A. cerana* are now confirmed in northern Australia. It is smaller but very similar in appearance to *Apis mellifera* is well adapted to warmer climates, builds smaller colonies, and is known to swarm many times during the year. In tropical areas (e.g., Solomon Islands) *A. cerana* has been shown to outcompete *A. mellifera* in nectar and pollen gathering and exhibits a propensity for robbing European honey bee stores. Its honey yield is far less than *Apis mellifera* making it a less valuable bee for commercial honey production.

Secondary Objective – Honey Bee Health Evaluation

A decline in honey bee health has been documented over the past 60 years. The known negative honey bee health challenges are primarily attributable to parasites, diseases, poor nutrition, stress and environmental toxins. There have been no national honey bee health surveys conducted to ascertain the scope of additional unidentified parasites, diseases, and pests that may have a negative impact on honey bee populations in the U.S. The benefit of informing and guiding the direction of honey bee parasite, disease, and pest research and mitigation recommendations to the U.S. apiculture industry would be significant. All of the data collected from the National Survey, including the pilot and limited national surveys in 2009 and 2010, will be included in the

nationwide Bee Informed Partnership database. The Bee Informed Partnership is an extension project funded by the USDA National Institute of Food and Agriculture (NIFA) for 5 years. As part of its core mission, the database endeavors to capture honey bee health and management practices from around the country to better inform all beekeepers with the goal of reducing colony losses. The data gathered in these extensive surveys are critical for capturing base line information on the status of honey bee health; this in turn will help place beekeeper disease load data in regional and temporal context.

Although the primary goal of the survey is to determine if exotic species are present in the U.S., the survey presents an important secondary opportunity. This survey is also being conducted to investigate potential causes of Colony Collapse Disorder (CCD), a significant disappearance of honey bee colonies that is one of many factors responsible for the sustained high rate of winter mortality (~30%) suffered by beekeepers across the U.S. over the last 5 winters. This unsustainable rate of loss threatens the viability of beekeeping operations and – importantly - the production of crops dependent on bees for pollination as well as honey production. Pollination is responsible for \$15 billion in added crop value, particularly for specialty crops such as nuts, berries, fruits, and vegetables. Of the 2.7 million colonies of bees in the United States, the almond crop in California alone requires approximately 1.3 to 1.5 million colonies, and this need is projected to increase significantly over the next few years. The bee industry is facing difficulty meeting the demand for pollination in almonds because of bee production shortages in California. Consequently, growers depend increasingly on beekeepers from other states to transport honey bee colonies across the country to meet the pollination demand (a phenomenon known as migratory beekeeping). If researchers cannot find a solution to CCD and other causes for high losses, beekeepers will be unable to meet demand for this and other crops.

The USDA has developed a CCD Action Plan, (http://www.ars.usda.gov/is/br/ccd/ccd_actionplan.pdf). The second goal of this plan is to determine current status of honey bee colony production and health. Objective two of this goal is to develop a long-term annual APHIS survey on the overall health status of U.S. honey bees. This action was designated as a high priority in the CCD Action Plan. Current theories about the cause(s) of CCD and increased colony mortality generally include Varroa mite parasitism; new or emerging diseases, especially mortality by a new *Nosema* species (e.g., *Nosema ceranae*), a newly evolved and more virulent strains of Varroa mite vectored bee viruses; sublethal pesticide exposures (through exposure to pesticides applied for crop pest control or for in-hive insect or mite control); and poor nutrition (due to reduced forage lands caused by increased corn monocropping and changes in agricultural practices). These factors, alone or in combination, are thought to suppress bees immune systems, making them susceptible to a host of pathogens, which in turn causes increased mortality.

Despite the existence of several surveys for both honey production and bee health, these surveys are either limited in scope, fundamentally flawed, or otherwise unable to provide an accurate picture of bee numbers or products (e.g., honey and pollination services). New surveys are needed to determine the extent of CCD in the United States and the current status of honey bee colony production and health. Apicultural industry groups, researchers (Federal, State and private), and apicultural Extension specialists all agree that there is an immediate need to establish uniform and consistent data collection methodologies to provide a baseline for both bee

production and health (epidemiology) measures. While several surveys have been or are currently being conducted, none meets the criteria needed to enable researchers to evaluate increases or decreases in these measures across the U. S. or North America.

Scope of work and methodology

The 2012 National Survey has two goals, 1) identify potentially invasive pests such as the exotic mite *Tropilaelaps*, problematic *Apis* species such as *A. cerana* and viruses such as the Slow Paralysis Virus and, 2) conduct an epidemiological survey that would meet the goal of developing a long-term overall baseline picture of colony health.

To accomplish these objectives, cooperators will distribute sampling kits, identify stationary and migratory beekeepers who will participate in sampling of their honey bee colonies, collect and preserve samples, quantify parasite loads from bees collected in alcohol and forward live samples for molecular analysis. Additional samples of pollen will be collected from 10 apiaries per state to evaluate pesticide occurrence in hives. The results of these analyses will be forwarded to the participating beekeepers and the respective state apiary contacts. Beekeepers participating in this survey should expect a summary report on the average apiary level Nosema, tracheal mites, Varroa loads, presence or absence of *Tropilaelaps* mites, and viral results from the molecular analysis in the sampled apiary within 4 to 6 months of sample collection. All data collected will be maintained at APHIS, ARS and UMD. This data will be entered into the APHIS NAPIS database as well as the BIP database described above.

As part of the survey includes a visual inspection of the hives before sampling, the presence of the following is recorded at the apiaries:

1. American Foul Brood
2. Black Shiny Bees
3. Chalkbrood
4. Deformed Wing Virus
5. European Foul Brood
6. Parasitic Mite Syndrome (PMS/Snotty Brood)
7. Sac Brood
8. Small Hive Beetle Adults
9. Small Hive Beetle Larvae
10. Wax Moth Adults
11. Wax Moth Larvae

The samples taken at the apiary and preserved in alcohol are later inspected using microscopic analysis at UMD and USDA-ARS BRL for the following:

1. *Nosema* spp. spore count
2. *Tropilaelaps* Mites
3. Varroa Mite loads

The live bees shipped directly from the apiary during collection are sent to USDA-ARS BRL. There, the honey bees are frozen until molecular and visual analyses are conducted. The molecular and visual analyses includes the following:

1. *Apis cerana*
2. Acute Bee Paralysis Virus (ABPV)
3. Chronic Bee Paralysis Virus (CBPV)
4. Deformed Wing Virus (DWW)
5. Israeli Acute Paralysis Virus (IAPV)
6. Queen Cell Virus (QCV)
7. Slow Paralysis Virus (SPV)
8. *Nosema ceranae*
9. *Nosema apis*

Also, as part of the national survey, training and outreach materials have been developed in the form of videos and written information (http://www.aphis.usda.gov/plant_health/plant_pest_info/honey_bees/survey.shtml). Literature will also be developed for pest identification and finally, information on honey bee health and maintenance will be collected and distributed at the conclusion of the survey.

Project Management, Cooperators and Other Participating Institutions

This National Survey is funded and lead by USDA APHIS and ARS. A Steering committee comprised of personnel from APHIS, ARS and UMD determined the sampling protocol, and determined the optimal distribution of sample analysis. Sampling is conducted under cooperative agreements with state apiary inspectors and university scientists. Beekeepers have been identified whose colonies will be used for sampling. Some of these beekeepers may also participate in conducting the survey. The states being sampled in the 2012 National Survey are:

Alabama	Maryland	Oklahoma
Arkansas	Massachusetts	Pennsylvania
California	Michigan	South Carolina
Colorado	Minnesota	South Dakota
Florida	North Carolina	Tennessee
Georgia	North Dakota	Texas
Hawaii	Nebraska	Utah
Iowa	New Jersey	Virginia
Illinois	New Mexico	Wisconsin
Indiana	New York	West Virginia
Louisiana	Ohio	

University of Maryland personnel, in close collaboration with USDA ARS personnel are responsible for the sample kit fabrication and distribution. They are also the contact for

receiving all alcohol samples and apiary data information forms from the field. The alcohol samples containing any dislodged mites from the hive frames are forwarded to USDA ARS BRL where scientists microscopically analyze the sample for the presence of *Tropilaelaps* mites.

USDA-ARS BRL scientists are the contact for receiving all live bee samples. The bees are immediately frozen and held until molecular analysis is conducted. USDA and UMD are responsible for all pest, diseases and exotic species and subspecies reporting to the beekeeper and the apiary contact for the selected states.

Guidance for Choosing Apiaries and Hives to Sample for the USDA Honey Bee Pests Survey

- 24 apiaries should be sampled in all states except California where 48 apiaries should be sampled.
- 8 hives should be sampled in each apiary.
- Apiaries sampled should have at least 10 colonies.
- If you are sampling an apiary with a large number (hundreds or thousands of hives), please try to provide a composite sample from all locations within the apiary. This may require that you divide the apiary into quadrants and then sample a few hives in each quadrant. It is critical that hives that appear 'sick' are sampled along with hives that appear 'healthy'.
- 10 queen producers should be sampled unless there are fewer than 10 willing queen producers in your state.
- Of the remaining apiaries sampled, when possible 1/2 of the apiaries sampled should be from migratory operations (move out of state and return prior to sampling) and 1/2 should be from stationary operations (Do not move out of the state but move within the state).
- Apiaries should be chosen in order to give as close to an equal representation of the entire state as possible. Ideally, a state will be sectioned into 4 quadrants with apiaries randomly chosen within a quadrant.
- Additional apiaries that may occur near ports or other areas that could be considered high risk should also be considered for sampling.

Milestones and Project Timeline

A pilot survey program funded from the APHIS Farm Bill funding was initiated in 2009 and samples were collected from three states to test the inspection and collection process, assess the infrastructures related to shipping, storing and analyzing the specimens, and to gather baseline data for a survey of honey bee pests and pathogens. Hawaii, Florida and California were part of this pilot program. These are high-risk areas that have key ports, long growing seasons and diverse agricultural crops. A summary report of the 2009 pilot survey is available on the APHIS website at

www.aphis.usda.gov/plant_health/plant_pest_info/honey_bees/downloads/2009_Pilot_Summary_Report.pdf.

The 2010 limited national survey, focusing on 13 states, was performed to expand and augment the baseline pest and pathogen data collected from the pilot study conducted in 2009. A summary report of the 2010 Survey is available on the APHIS website at

http://www.aphis.usda.gov/plant_health/plant_pest_info/honey_bees/downloads/twg_report_july_2010.pdf. In 2011, a full-scale national survey was funded for 34 states and is the most comprehensive U.S. honey bee pest and disease survey to date. The primary focus of this survey was to verify the absence of the parasitic mite *Tropilaelaps* and other exotic threats to the U.S. bee population (e.g., *Apis cerana*). Establishing the absence of threats to honey bee populations not thought to be present in the U.S. was the primary objective of this effort; however, to capitalize on the information gathered from this survey, samples were analyzed for other honey bee diseases and parasites known to be present in the U.S.

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