

**Agricultural Research Service & Cooperative State  
Research, Education, and Extension Service  
Colony Collapse Disorder Workshop  
April 23 & 24, 2007  
Beltsville, Maryland**

On Monday, April 23, 2007, the Agricultural Research Service (ARS) and the Cooperative State Research, Education, and Extension Service (CSREES) convened a meeting in Beltsville, Maryland, to discuss the urgent national issue of Colony Collapse Disorder (CCD). Meeting participants came from across the United States and also included participants from both Canada and Switzerland. The purpose of the meeting, which was organized on very short notice, was to identify specific research options for exploring the cause(s) of CCD, to determine high priority research needs and collaborations, and to recommend immediate and long term follow-up. Funding options were also briefly discussed.

Participants included scientists, officials, and stakeholders from ARS, CSREES, the Animal and Plant Health Inspection Service (APHIS), the Natural Resources Conservation Service (NRCS), and the Agricultural Marketing Service (AMS), the Environmental Protection Agency (EPA), the Department of the Army, numerous land grant university researchers and officials, industry groups, commodity groups, and beekeepers. In addition, numerous national media representatives covered the meeting. Many meeting participants who were unable to attend in person were included in the discussions by conference call both during the plenary discussions and during the break-out sessions.

Both the planning and the conduct of the meeting took on a sense of urgency due to growing Congressional interest and inquiries from the Executive Office of the President. Adding to the complexity of the meeting was the large number of participants with widely divergent opinions regarding the cause(s) of CCD and the potential impact of CCD on national and international agriculture.

The meeting was opened by Dr. Judith St. John, Deputy Administrator, National Program Staff (NPS), ARS. She noted the importance of the meeting and the need for the "best science" in order to respond to the emerging potential crisis posed by CCD. She stressed the need for the identification of options and ideas to solve the growing problem of CCD.

Dr. Diana Cox-Foster, Professor of Entomology from Pennsylvania State University, followed Dr. St. John. Dr. Cox-Foster summarized the potential impact of CCD on American agriculture and outlined the symptoms. She showed a map provided by Bee Alert Technology, Inc., showing State incidence of CCD. She discussed the Working Group on CCD, its membership, and its goals. The Working Group was formed in the Fall of 2006 in response to the emerging crisis. She summarized some of the work which has been completed regarding CCD and shared photos of the effects of CCD. Dr. Cox-Foster offered some working hypotheses regarding potential causes and suggested the need to create increased genetic resistance in honey bees. In closing her remarks, she spoke to the need for wide ranging collaborations among researchers while noting the important role of Cooperative Extension in bridging research and beekeepers. She noted the challenges of identifying the causes of CCD and importance of maintaining the health of honey bees.

Dr. Cox-Foster's remarks were followed by Dr. Jeff Pettis, Research Leader of the ARS Bee Research Laboratory in Beltsville, MD. Dr. Pettis' remarks focused on factors impacting bee health. He cited the recent National Academy of Sciences report (a copy of a brief summary was provided to participants) which concluded that all pollinators are in decline nationwide while pollination needs continue to increase. In addition, he noted that honey bee health has declined since the 1980s. He discussed honey bee losses around the world and noted that the first European meeting to address the prevention of CCD was held in March 2007. Dr. Pettis described the symptoms of CCD and the importance of reading the signs and symptoms. He discussed *nosema apis* vs. *nosema ceranae* and noted the presence of *nosema ceranae* in colonies in numerous States in the U.S. Further, he discussed Varroa mites and their presence in both strong and weak colonies. Additional research was summarized yet Dr. Pettis indicated that there are no common findings to date.

Dr. Pettis concluded by encouraging the participants to focus on the task and noted the limited time available for the important deliberations. He then introduced the facilitator for the meeting, Marva Nesbit, who would assist in managing the discussions. The facilitator discussed the process for the meeting and described how information would be gathered and processed. Next, participants had a facilitated round table discussion where a wide range of ideas regarding CCD were shared and debated. The breadth of ideas reflected the diverse thinking of the participants regarding CCD. Some general notes from the plenary brainstorming were recorded to help identify and focus on broad based areas for research to be distilled for further discussion.

## **General Issues for Consideration Identified During the Plenary Discussion**

- ❖ Stock certification program
- ❖ Pesticides
- ❖ Pathogens/virus
- ❖ No clear patterns of cause based on survey
- ❖ Wild pollinators
- ❖ Nutrition
- ❖ Expanded discovery phase
- ❖ Mite management
- ❖ Climatic issues
- ❖ Info gathering (centralized)
- ❖ Is this real?
- ❖ GM crops
- ❖ Environmental stress
- ❖ Media approach
- ❖ Beekeeper management: cumulative
- ❖ Genome project involvement

Following the round table discussion, the participants concluded that the identified issues fell into four broad based potential research areas. Participants then worked in break-out groups to explore each category in greater depth. Participants were allowed to select their break-out group topic. Rooms were assigned for each break-out group and call-in numbers were shared for those participating on teleconference.

### **Broad-based categories:**

- ❖ Parasitic mites and pests ( Room 4-2223)
- ❖ Pesticides/environment (Room 4-2240)
- ❖ Pathogens/viruses (Room 4-2101)
- ❖ Stress/management (Room 3-1175)

Break-out sessions were lively with lots of discussion and idea generation. After the break-out deliberations, participants reconvened in plenary session to hear reports from each break-out group session. The reports by category from the break-out group sessions follow (exactly as reported).

## Break-out Session #1: Parasitic Mites and Pests

- ❖ Varroa
  - Association between Varroa mites and CCD
  - Interaction with viruses and other microbes (European collaboration?)
  - Interaction with nutrition
  - Interaction with management (migration)
  - Breed for resistance:
    - Use genome tools: SNP chips
    - Discover more traits that confer resistance (grooming)
  - Varroa genotypes/virulent types
- ❖ Small hive beetle/wax moths
  - Mechanical transmission or vectoring of viruses/pathogens
  - Indicator of pre- or past- CCD
- ❖ Tracheal mite
  - Improved diagnostic tools
  - Prevalence and association: CCD

## Break-out Session #1: Pesticides/Environment

Goal – Put real science behind this

- (1) Beekeepers (Inside Hive)
  - Sub-lethal efforts (beekeeper inputs)
  - Antibiotics and other medications
  - Carriers
  - Miticides (labeled & off-label)
  - Bee feed contaminants (GMO chemical)
  - Off-gassing from plastics
  - Supplements
  - Wood treatments
  - Comb preservation treatments
- (2) Environmental inputs (Outside Hive)
  - Residues in pollen and nectar
  - Bee body exposure/retention
  - What new chemicals are in use
  - Multiple/unrecorded exposures through transport to various locations
  - Neonicotinoids and pyrethroids
  - Misapplication of a variety of pesticides

- Lack of consistent labeling (e.g., Ag application, homeowner application)
- Fungicides

### Needs Assessment

- Potential accumulation in hives of:
  1. Miticides (labeled and off-label)
  2. Antibiotics (new and old)
  3. Bee feed contaminants
  4. Pesticides
- Environmental
  1. Chemical survey
  2. EPA label consistency review for bee concerns
  3. Methods available for chronic and sub-lethal exposures (SOP – standard protocols)
- Resurvey with additional concerns (off-going from plastics, supplements, wood treatments, comb preservation treatments)

### Break-out Group Session #1: Pathogens/Viruses

#### Historical Samples and time of season

- Apis mellifera
- Other sps
- Pinned vs 80°C (DNA only)

#### Is there a mutant/shift in a pathogen?

- Change in genetic stocks – susceptibility? – abrupt changes, may not be shift in genetics totally
- Seq. comp? Determine ID & area variation – time geography

#### Discovery – of new ones—need non-biased techniques

- High through put and pre existing – 454 – Columbia
- Edgewood

#### Virology/Bacteria/Fungi/Protozoa

- Need for methods to propagate/study
  - Cell line needed
- What effect do individual pathogens have
- Define samples/colony status (symptoms) to mirror with pathogen presence

- Management Needs Reaction
  - o Assay in field for beekeeper
    - Rapid
    - Conc. level
      - Define normal levels (how to sample)
      - Move to cost effectiveness
      - What are the organisms effects of other stressors

What causes changes immunity/susceptibility—effects of other stressors?

- How to detect changes
  - o Genetic system?
    - All pathogens
    - Viruses only

Discuss – Current experiments

- Comb treatment
- Migratory stress
- Controlled experiments – other stresses
  - Heat shock
  - Nutrition

Future experiments

- Koch's post. on pathogens, other stresses, combo
- Standardize or experiment design that has consensus
- How to evaluate pathogens

Data flow

- How to coordinate and evaluate data and relevance (potential importance/concern)
- Develop systems to evaluate pathogens – native bees
- Antibiotic or pharmaceutical treatments
  - o Affects on symbiotics/normal gut flora
  - o Resistance of microbes/nosema/etc.

Decontamination/sterilization

- o Methods – need new research
- o How to define sterility – microbe test strips, etc.
- o Need for availability across the country

What is the “normal distribution”

- APHIS – Disease survey

- 2009 (lower 48 and Hawaii) proposed \$1.4 – 2 million (2008 dollars)
- Proposed ethanol samples
- 10 reasons had been for new parasitic mites
- Potential to mites size and ultra cold frozen samples (need add ext. funding) – expand to other pathogens/parasites
- 10 apiaries/states
- Trial run with 2-3 states ASAP – need funding management
- APHIS survey
  - Disease priorities
  - Reportable vs. “normal” known diseases
  - Annual basis?
  - Not all States have the infrastructure in place – apiary inspectors
  - Couple with national bee survey (#colonies/death)
- Need for bee disease diagnostic lab network and service for processing samples
  - DHS? Jurisdiction or help with funding
  - CSREES
  - Hub – sequencing/PCR
  - State PCR
  - Potential to connect to survey data (GIS with other stressors, croplands, etc. – way to correlate other factors with diseases)

### Break-out Session #1: Stress/Management

#### Mite management – acaricides / ag chems?

Standard sampling, wherein hive - correlations – devices  
 Optimize delivery and timing of acaricide treatments – sub-lethal effects on bees  
 Mite suppression techniques

#### Nutritional management and value of crops, etc., pollen quality

Single pollen sources versus some pollens mixed  
 “Clean” pollens versus similar contaminated pollens  
 Demonstrate effects of honeybees on areas closed for “environmental” reasons  
 Collect pollens to determine what is collected, locally  
 Plants in greenhouse – effects of weather

## Stock type, maintenance and production

### Short term research:

Tracheal mite exposure of today's stocks

Hygienic behavior of today's stock

Longevity of queens and colonies – highly migratory operation – today's stocks

### Long term research:

Stock certification/maintenance

Importation issues

Africanization

## Migratory stress / ag chems

Run stocks side by side and evaluate over the year

Comparing bee feeds in various regions of the country

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Following the reports from the first break-out sessions, participants were again allowed to select break-out sessions by topic for the second session. Once again, the sessions were filled with interesting idea exchange. The reports from the second break-out session follow (as reported) using the same format and the same topics.

## Break-out Session #2: Parasitic Mites and Pests:

Archival sampling [expanded APHIS sampling]

- Current situation
- For future research sampling
- Symptomology !
- ICC (Idiopathic colony collapse)
- Systems research

## Break-out Session #2: Pesticides/Environment

1. Sub-lethal EFFECTS of systemic insecticides
  - A. Seed treatments / esp. neonicotenoids
  - B. Transport Accumulation
    1. Vapor columns?
    2. Trichomes

2. Accumulation of Pesticides in Wax
3. Define the sub-lethal Effects
  - A. Systematic Survey
  - B. Dose Response Studies
  - C. Likely Candidates
    1. Inside and Outside
4. Food Grade Plastic Foundation?
5. Mosquito Control? Malathion?
6. Timing of Supplemental Feeding - need to be willing to change management tactics
7. Climate Change?
8. Utilize honey bees and non-Apis bees for experimentation on cuts and sub-lethal effects from pesticides
9. Need to look for the common pesticide trend
10. Need to collect information on when, where, and amount of pesticide applied on crops and in the hive and correlate to CCD levels
11. Need more insect toxicologists working on bees

### Break-out Session #2: Pathogens/Viruses

#### Nosema

- Which species is normally present?
  - Etoh specimens
    - Troy F. - Venezuela 1985
    - Jerry B - freeze dried bees
    - 1980's - NIST - National Bureau of Standards
- Fumagillan - effectiveness?
  - Need research to address pathogenicity of different species?
- Research on transmission and disease development
- Longevity of spores
- Determine if immunosuppression or nutritional physiology/stress

## Bacteria

- Saprophytes vs. Pathogens?
  - What are the normal gut flora – symbionts(?)
  - Pathogens?
    - Examine putative pathogens in multiple operations/colonies
    - Role of European Foulbrood – stress
      - Genetic strain differences in suscep.

## Protozoa

- Amoeba
  - o Beltsville
- Trypanosomes

## Fungi

- What species?
- Only a stress-related disease?
- Honeybee genetic stock – susceptibility?
- Potential toxin production?

## Insect Immunity/Susceptibility

- How to promote “stronger” bees – preventative measures
- Genetic resistance
  - Potential genetic reservoir in Africanized

## Break-out Session #2: Stress/Management

Meta factor analysis  
Weather

How to identify stress

Validation of stress indicators  
Detox genes  
Physiological indicators

Comb replacement test

Development of standardized high-quality diet, nutritionally complete

The first day of the meeting ended with the break-out group session #2 reports. The meeting adjourned after a very long day with intense discussions and deliberations.

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On Tuesday, April 24, the meeting opened with remarks from Dr. Kevin Hackett, National Program Leader for Biological Control, NPS, ARS. He reviewed the broad based research categories which had been identified the previous day. Further, he emphasized the high level of interest and urgency created by the CCD issue and further emphasized the need to identify research priorities for this critical agricultural problem.

Following Dr. Hackett's remarks, participants discussed in a plenary format what they saw as the most immediate research priorities for CCD. Initially, the discussions were brainstorming in nature. The general notes from the brainstorming include:

- ☆ Pathogens
  - Confirm or eliminate
  - Samples
- ☆ Pesticides and Mites
  - Samples
- ☆ Environmental
  - Samples
- ☆ Resurvey beekeepers
- ☆ Stress
- ☆ Collect samples (new) – systematic
- ☆ Why some States and not others?
- ☆ Statistically valid survey
- ☆ Expanding APHIS survey
- ☆ Define CCD
- ☆ Measure extent – systematic
- ☆ Causes
- ☆ Remedies
- ☆ Prevention
  - Stock
  - Practices
- ☆ Involve NASS in data collection
- ☆ Pathogen
- ☆ Parasitic mites
- ☆ Environmental stresses
- ☆ Analysis of whole farm systems

- ☆ Enhanced survey
- ☆ LT tracking but utilize existing data
- ☆ Tools
- ☆ Extensive survey: protocol for sampling
- ☆ Survey and monitoring
- ☆ Web database
  - [www.beealert.info](http://www.beealert.info) for survey results
- ☆ Survey: NASS and APHIS (urgent but LT)
- ☆ Infrastructure enhancement through competitive grants
- ☆ Honey bee toxicologist needed
- ☆ Coordinated research efforts among partners
- ☆ Reliable data, standardization of methods, web-based database

As the meeting drew to a close, participants focused more specifically on the prioritization of the next steps for CCD. Those priorities (including some hypotheses) follow.

### **Priorities**

- I. Survey and (sample) data collection
  - Dissemination
  - Re-sample
  - Refine CCD symptomology
  
- II. Analysis of the existing samples
  - Identify potential pathogens, pests, and pesticides in the immediate samples
  - Sharing data on the web (including environmental data)
  
- III. Hypothesis driven experimentation
  - Identify environmental stressors  
(Hypothesis: Crude protein levels between crashed and non crashed hives)  
(Hypothesis: Look at sublethal effects of pesticides on bee physiology and behavior)
  - Confirm pathogens (urgent!!), pests, and pesticides (number of queen matings?) Determine if CCD is caused by a common pathogen, a new pathogen, or a combination of both.
  - Share data on the web (including environmental data)

- Test alternative hypotheses  
(Hypothesis: Change in agricultural practices)
- Stock interactions  
(Hypothesis: Some strains might be more resistant or susceptible than others)

#### IV. Mitigative and preventative measures

- Best management practices
- Bee breeding and stock maintenance
- Surveillance
- A regulatory framework should be developed to better protect against introduction of new pathogens, pests, and parasites of bees (Status of Pollinators in North America)

Laurie Davies Adams from conference call:

- Establish best practices for migratory bee keepers (funding)
- Access to nutritional resources in transit
- Economic assessment (beekeeping and agricultural impact)

With only a few minutes remaining, the participants spent the last portion of the meeting discussing the difficult issue of research funding. In a brainstorming mode, the following suggestions were offered regarding possible funding for the research on CCD.

#### **Funding Strategies:**

- Emergency Funding from Congress
- Money from the continuing resolution money that went to the land grant universities
- End Users, Industry
- A small working group volunteered to spend some time at the conclusion of the meeting discussing possible funding sources: J. Pettis, N. Calderone, Gordon Wardell, M. Purcell-Miramontes, D. Weaver
- CSREES will share potential funding information

As the meeting drew to a close, participants were thanked for their attendance and their participation. CCD is an issue of critical national and international importance. The CCD Working Group will benefit from the

insightful deliberations and brainstorming on the critical issue of CCD from the workshop. The information will be used in the development of a CCD Plan of Action.

*Respectfully Submitted*

*Marva Nesbit, Facilitator  
1 May 2007*

