# Development of novel Varroa mite control methods from attractants and arrestants isolated from brood host volatiles

\*Mark J. Carroll

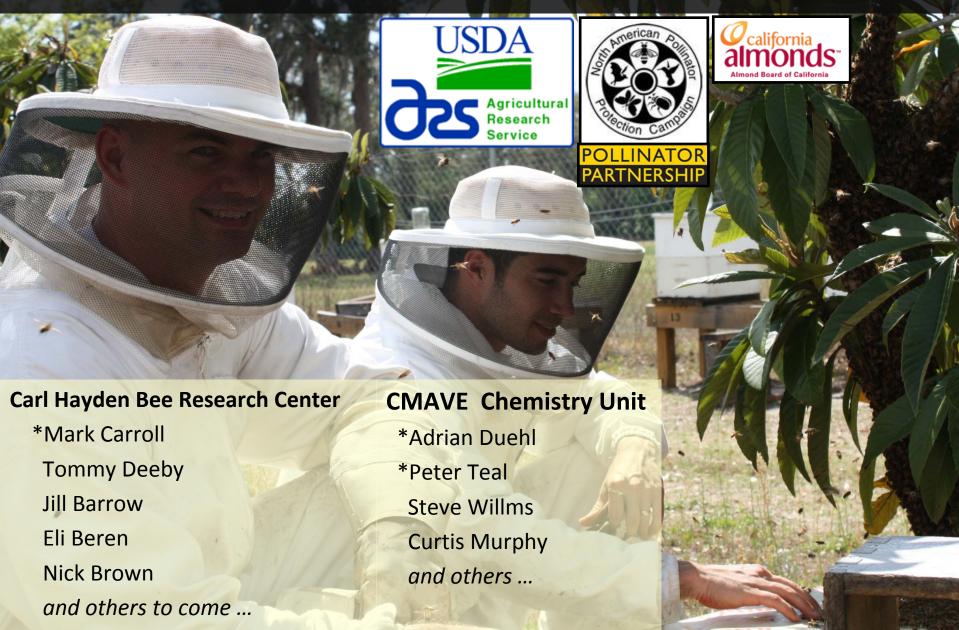
USDA-ARS Carl Hayden Bee Research Center, Tucson, AZ

Adrian Duehl and Peter Teal
USDA-ARS CMAVE, Gainesville, FL

\* formerly of USDA-ARS CMAVE and University of Florida

Department of Entomology

### Varroa semiochemicals (signaling chemical cues) – a collaborative project



#### Cell invasion behaviors – brood host acquisition



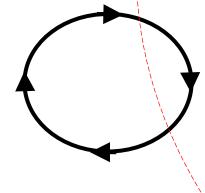
maternal mite encounters brood of various ages on adult bee (phoretic) host

mite detects and invades capping brood cell (attraction behaviors)

mites disperse to other adult bees



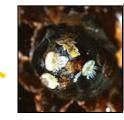
mites emerge with the new adult host





mite moves to back of cell and stops (arrestant behavior)

host emerges as adult bee



host cell capped by worker bees

maternal mite feeds on brood, produces 1-3 daughters

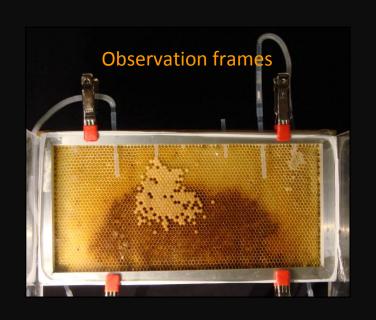
photos courtesy of USDA

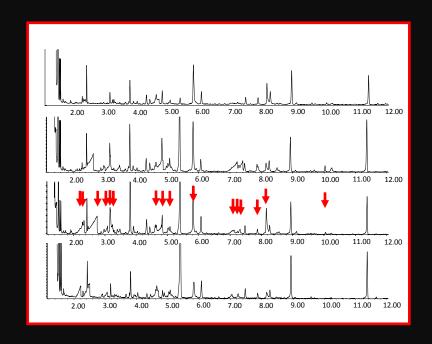
# Cell invasion by phoretic mites A rapid, very infrequent, near-contact event



- Female mite may take a week to locate an appropriate brood host
- Phoretic mite encounters hundreds of brood while on adult hosts
- Mite must be within ~7 mm of brood host for detection
- Cell invasion occurs in several seconds
- Finds appropriate host despite presence of similar non-host brood

#### Compare volatiles from host and non-host brood

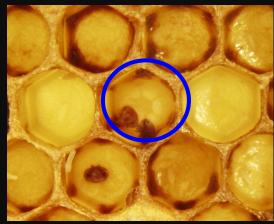




- ~ 120 volatiles from brood comb
- ~ 30 vary consistently with onset of cell capping
- phoretic mites respond to 2 compounds (CA and CB) by moving off adult worker hosts
- CA and CB emissions are higher (up to 3x) in preferred hosts

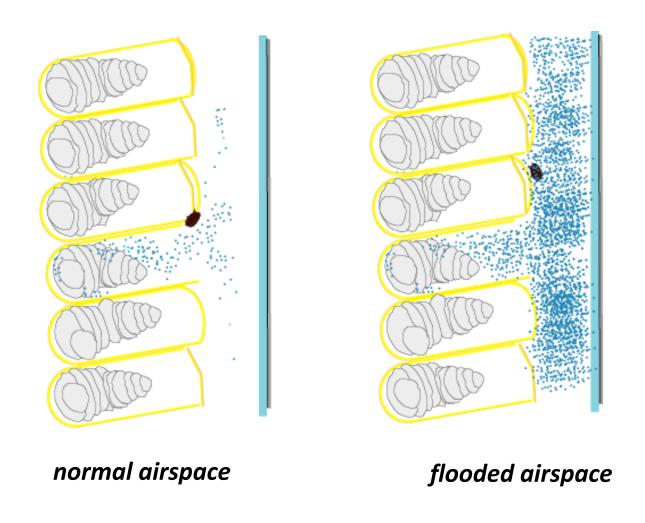
#### Evaluate mite responses to host-specific compounds





- mites detect CA and CB volatiles at high concentrations (foreleg electrophysiology)
- mites are strongly attracted to both compounds at near contact distances (~ 5 to 10 mm)
- mites show arrestant behaviors at high volatile levels
- free roaming mites are attracted to CA-treated cells
- <u>phoretic mite</u> cell invasion is partially blocked by flooding the hive airspace with CA volatiles

Flooding – disrupting behaviors by releasing enough synthetic semiochemicals to saturate chemoreceptors



The mite is overstimulated!

#### Developing in-hive trap/flooding technologies

#### 1. Identify minor volatile components that act as synergists

- EthoVision behavioral choice tests (traps)
- observation frame flooding tests (flooding)

### 2. Create slow-release formulations for sustained flooding/attracticide activity over time

- 21/42 day release? (formulation chemists)

#### 3. Develop in-hive deployment devices

- cheap and effective, but not disruptive to the bees
- devices bees tolerate (won't destroy, avoid, or wall over)

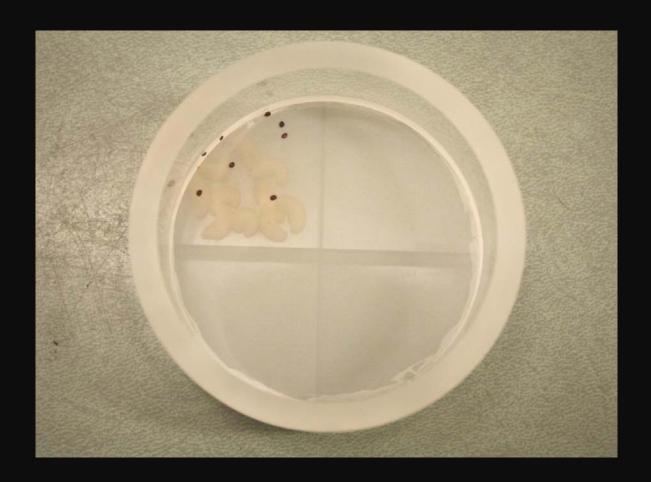
#### 4. Deployment - field tests on phoretic mites inside full-size colonies

# Looking for improvements in semiochemical activity



Provisional application for Varroa semiochemical patent granted August 2010 – becomes final August 2011

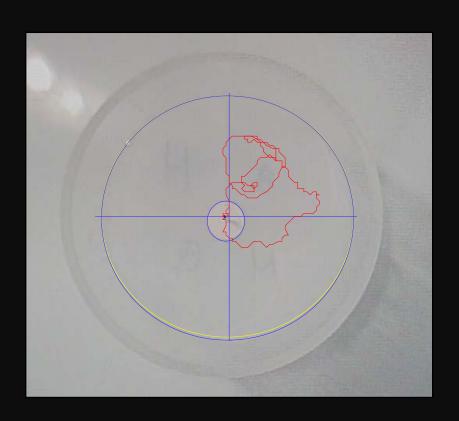
## Selecting mites that <u>consistently</u> respond to native brood odors in behavioral assays (Adrian Duehl)



85% of selected mites now respond to brood odors (~30% normal observed response rate)

### EthoVision bioassays - Video analysis of mite responses (movements) to volatiles





mite tracks in user-defined arena & odor zones

#### Identifying the odor source(s) (GC and HPLC)



What are the odor sources for the major <u>and</u> minor volatile cues? Larvae, adult worker bees, food, secretions, wastes, or hive components?

Are the volatile cues microbial in origin?

### Examining differences in volatile emissions between Varroa-resistant and non-resistant honey bees



<u>Resistants</u> – Russians, Africanized bees, local "non-treated" lines <u>Susceptibles</u> – island honey bees (pre-invasion)

#### Increasing the margins in our favor

A 2-3x more attractive synthetic mix will probably outcompete brood odors



#### Increasing the margins in our favor

A 2-3x more attractive synthetic mix will probably outcompete brood odors



### August 2011



# Observation frames Fits inside the perimeter of any frame



### Examining differences in volatile emissions between the original host (*Apis cerana*) and *Apis mellifera*



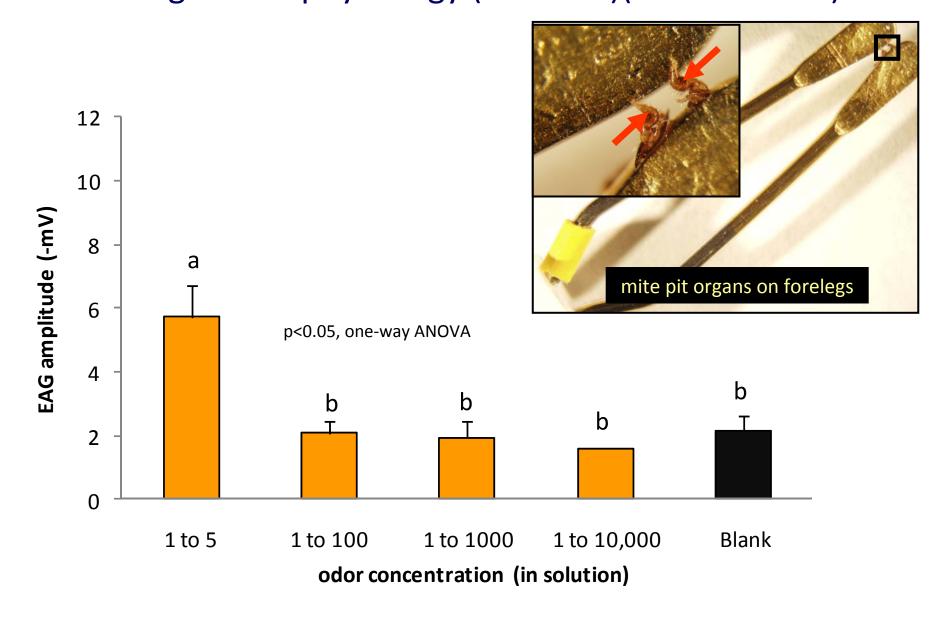
Phoretic Varroa mites invade the cells of worker and drone brood in *Apis mellifera*, but only drone brood in *Apis cerana*.

Why the difference (from a <u>mechanistic</u> point of view)?

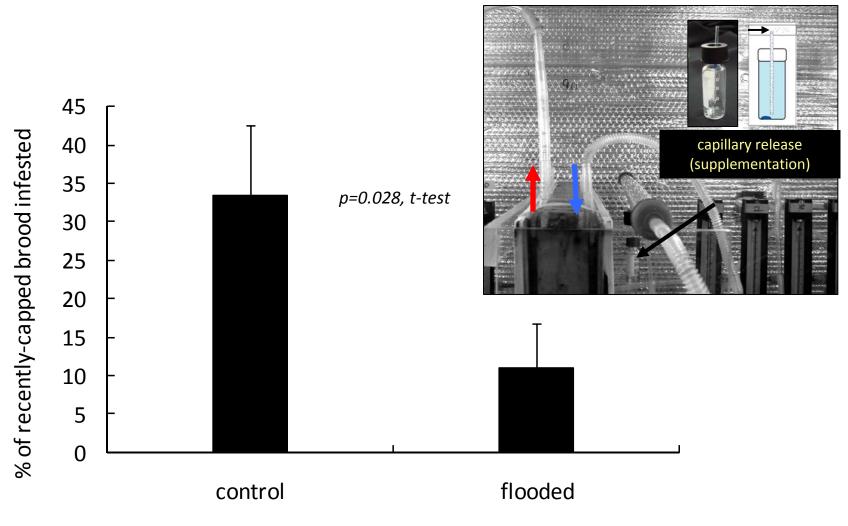
## **Observation frames** Collection and manipulation of headspace volatiles glass plate vacuum line air line (pull) (push)

a highly controlled push-pull airflow system

### Mite forelegs respond only to high concentrations of CA Foreleg electrophysiology (EAG-like)(Adrian Duehl)



Flooding the brood comb airspace with synthetic CA reduces cell invasion by mites without disrupting normal colony functions



observation frame airspace treatment