

# How to Breed for Behavioral Resistance to Varroa Destructor

**Krispn Given, Purdue University**

<https://extension.entm.purdue.edu/bee hive/>

# Why **breed** bees?

- **Increase honey Production**
- **Improve overwintering ability**
- **Decrease defensive behavior**
- **Reduce swarming**
- **Reduce or increase use of propolis**
- **Change color**
- **Decrease incidence of diseases**
- **Improve defense against varroa destructor\***
- **Other personal reasons**



# Most important part of a breeding program



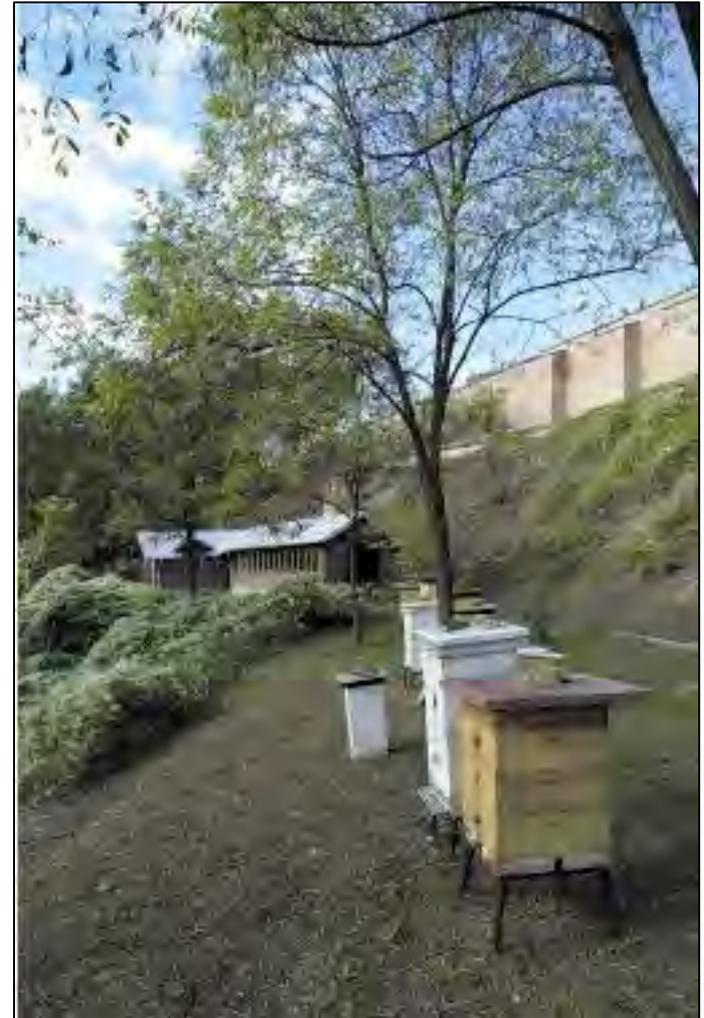
# Steps in Breeding

- **Decide on a trait (phenotype)**
- **Develop a assay**
- **Measure the foundation population**
- **Select parents**
- **Control mating using a design**
- **Evaluate, document, and verify**
- **Select parents**

# Bee Genetics + Breeding



Mendel's bee house



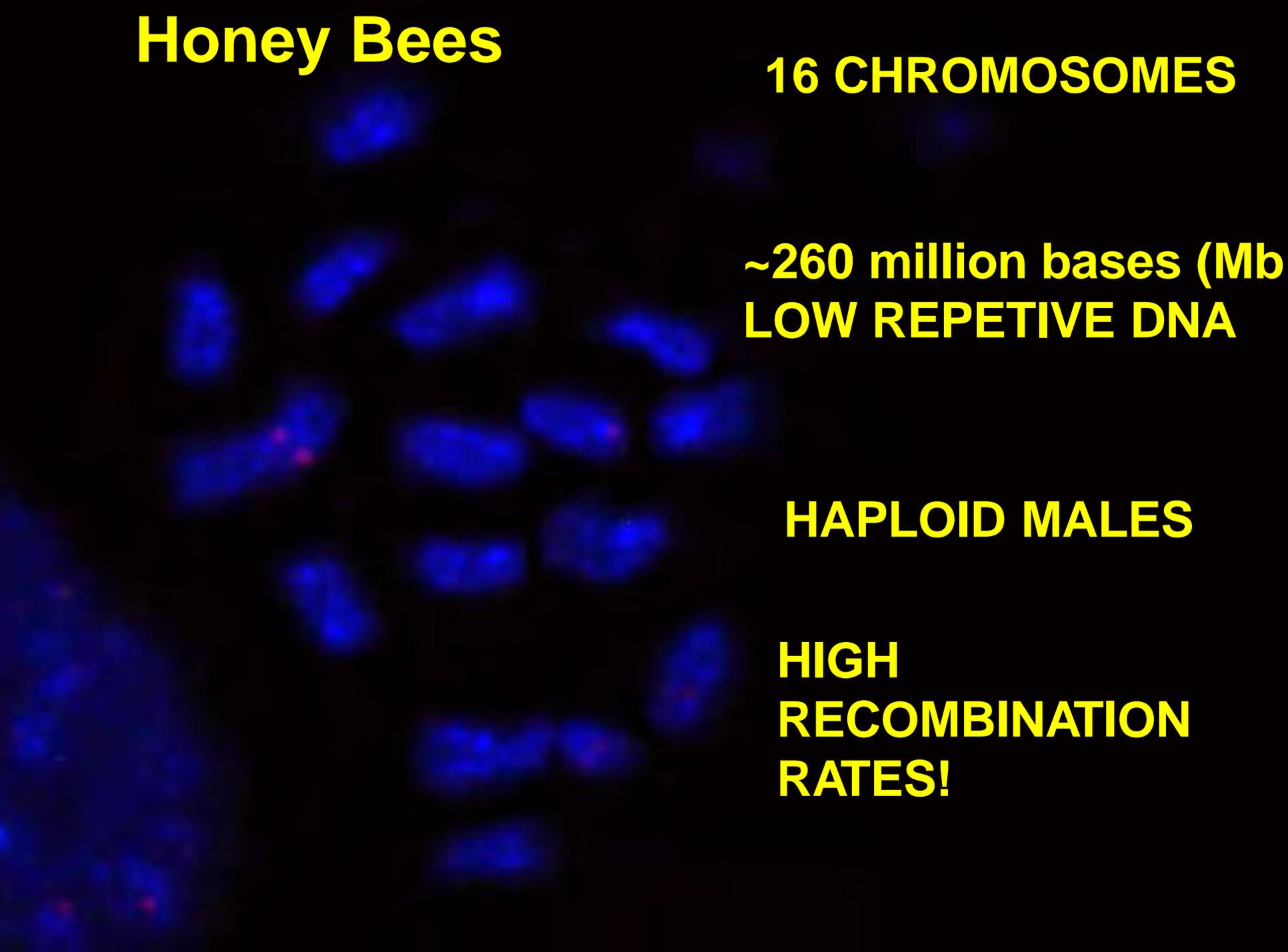
# Honey Bees

**16 CHROMOSOMES**

**~260 million bases (Mb)  
LOW REPETITIVE DNA**

**HAPLOID MALES**

**HIGH  
RECOMBINATION  
RATES!**



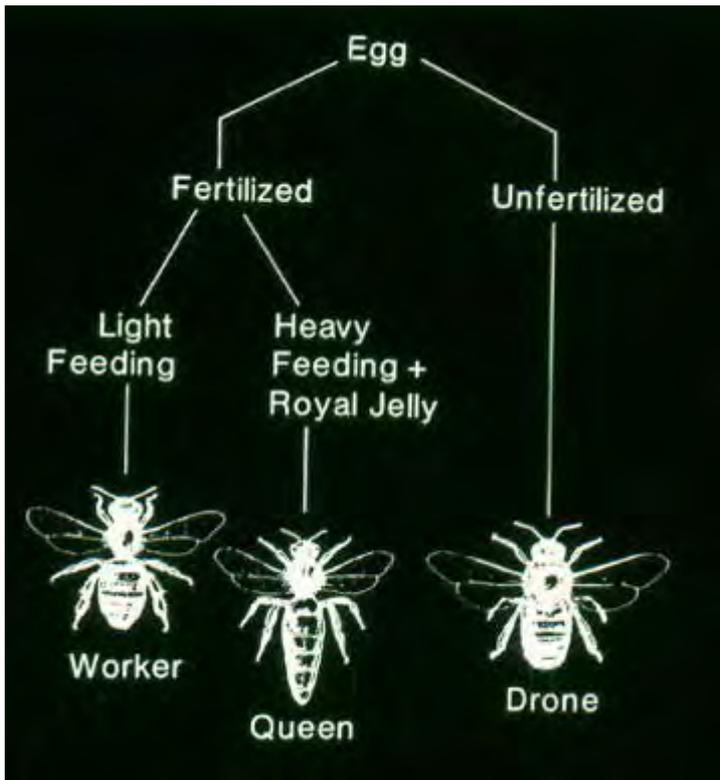
# CHROMOSOMES



- **Composed of coiled, twisted DNA and coated with proteins**
- **Each species has a specific number**
- **They are organized in pairs**

**(one comes from the mother and the other from the father)**

# Genetic gender and caste



- Queens lay two types of eggs
- Males develop from unfertilized eggs
- Females develop from fertilized eggs

# HONEY BEE GENETICS

Bees have 16 paired chromosomes.

## Haplodiploidy

- Males honey bees have one of each chromosome (haploid 16)
- Females have two of each chromosome (diploid 32)

## Polyandry

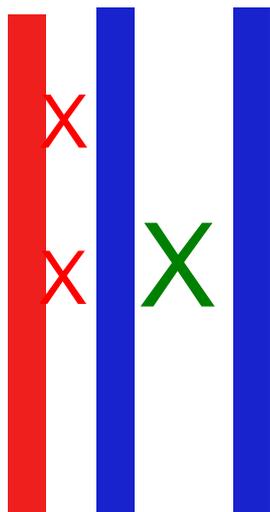
- The queen mates with 10 to 15 males, resulting in many sub-families within the hive's family.

Many important behaviors are influenced by genes!

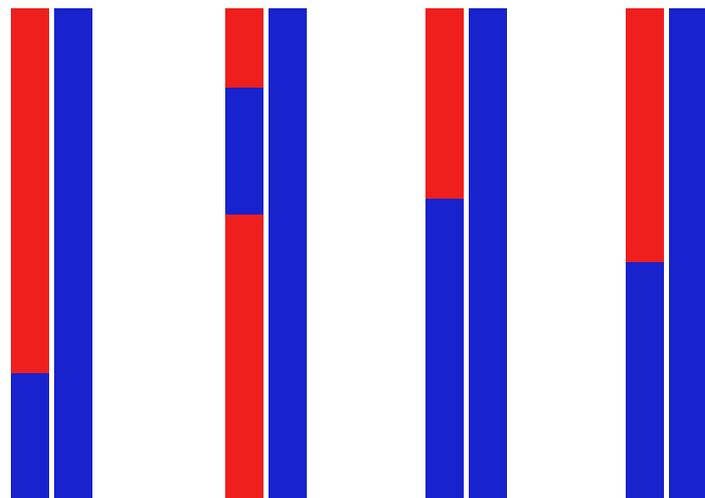
- stinging
- pollen foraging
- undertaking
- hygienic behavior
- brood rearing traits
- swarming tendency
- propolis collection
- honey production
- even learning!



crossing-over



FEMALES



# WHAT HAPPENS WHEN YOU CROSS TWO DIFFERENT STOCKS OF BEES



or



HIGH LINE

LOW LINE

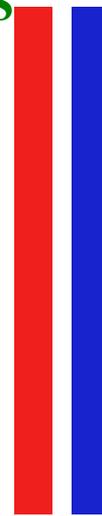
HYBRIDS



X



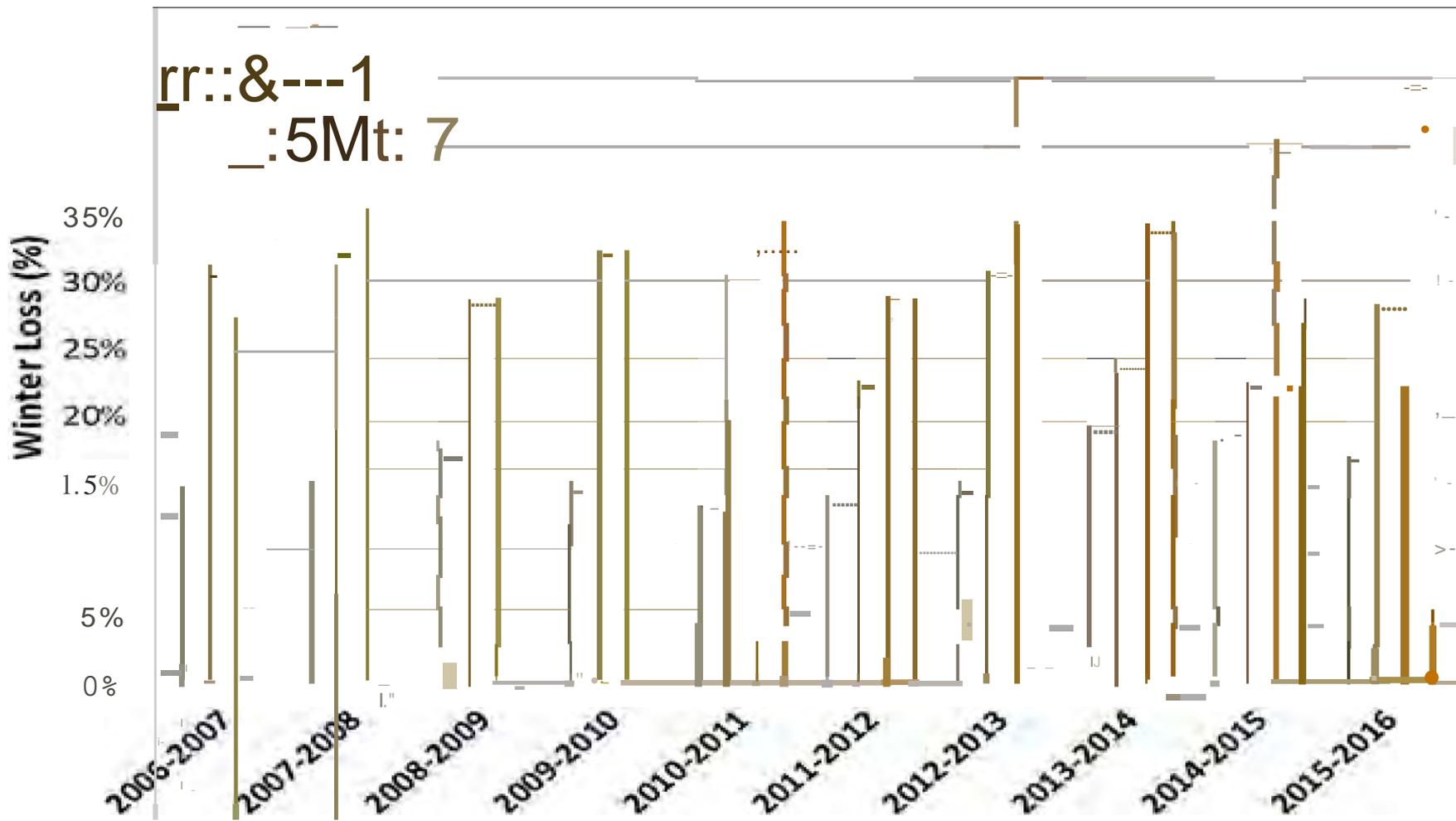
gives you



HYBRID CROSS

# Total US managed honey bee colonies Loss Estimates

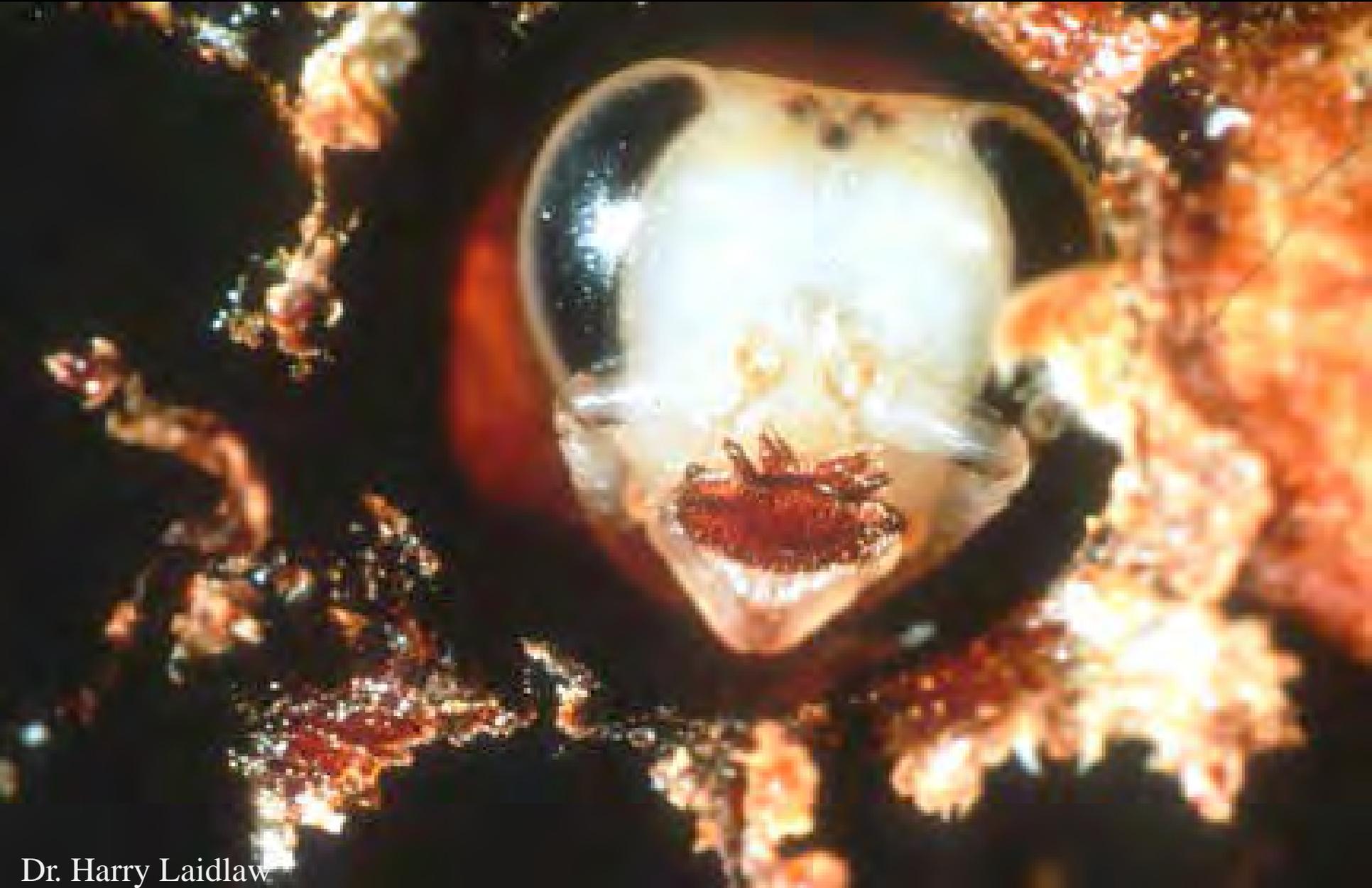
Acceptable Winter Loss Total Winter Loss Total Annual loss



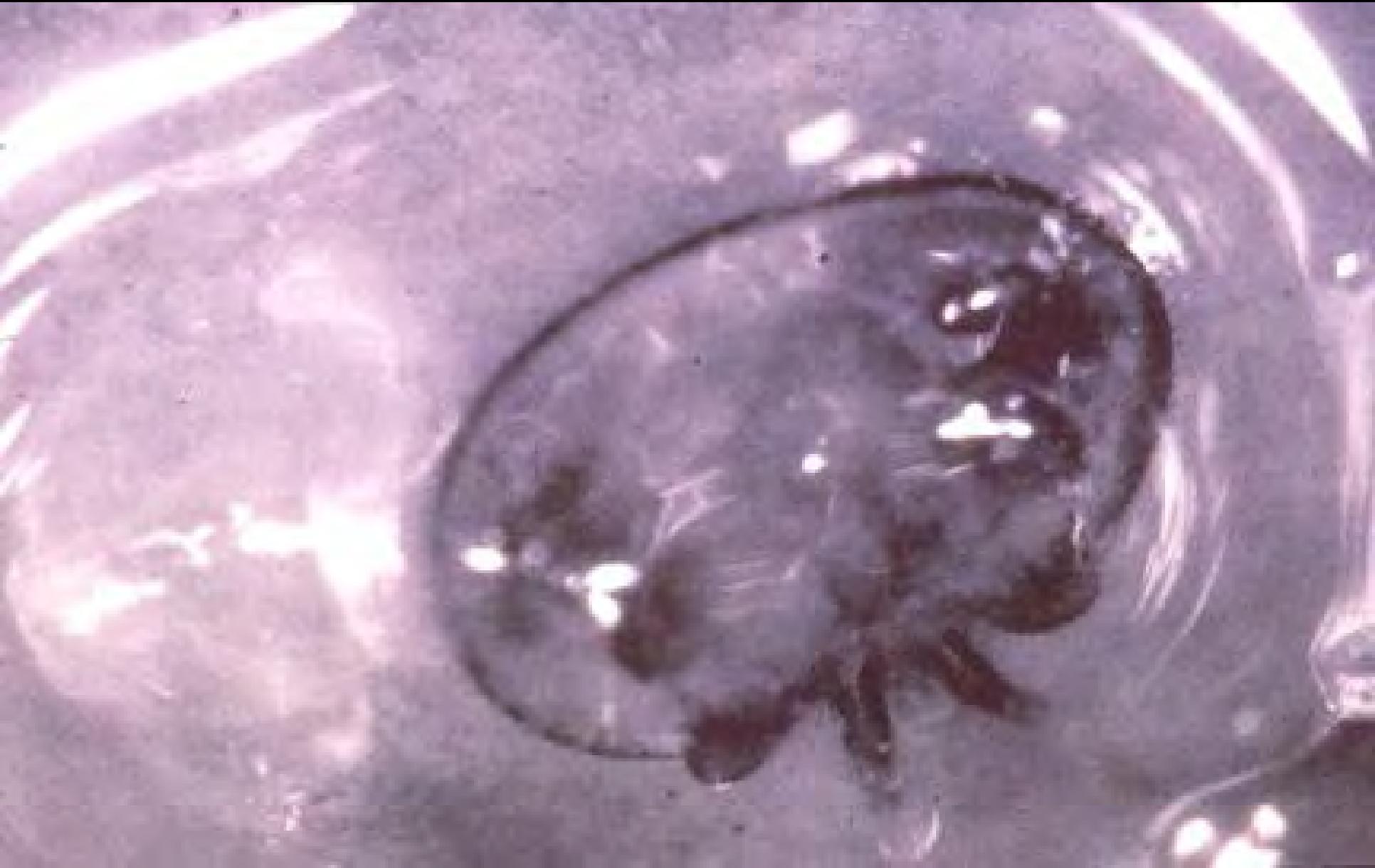


Registration #	Product Name	Active Ingredient
2724-406	ZOECON RF-318 APISTAN STRP	Fluvalinate (100%)
2724-406-62042	AP STAN ANTIVARROA MITE STR PS	
61671-3	FOR-MITE	Formic acid (65.9%)
70950-2	AVACHEM SUCROSE OCTANOATE (100%)	Sucrose octanoate (40%)
70950-2-2205	SUCROCIDE	
70950-2-8470	SUCRASHIELD	
73291- 1	API LIFE VAR	Thymol (74.09%), Oil of eucalyptus (16%), Menthol (3.73%)
75710- 2	MITE-AWAY QUICK STRIPS	Formic acid (46.7%)
79671- 1	APIGUARD	Thymol (25%)
83623-2	HOPGUARD II	Hop beta acids resin (6%)
87243-1	Apivar	Amtraz (3.33%)
91266- 1	OXALIC ACID DIHYDRATE	Oxalic acid (100%)
91266-1-73291	OXALICACID D HYDRATE	
91266-1-91832	OXALICACID D HYDRATE	
11556- 138	CHECKMITE+ BEE HIVE PEST CONTROL STRIP	Coumaphos (10%)
11556-138-61671	CHECKMITE+ BEE HIVE PEST CONTROL STRIP	

# Adult female invades cell before its sealed



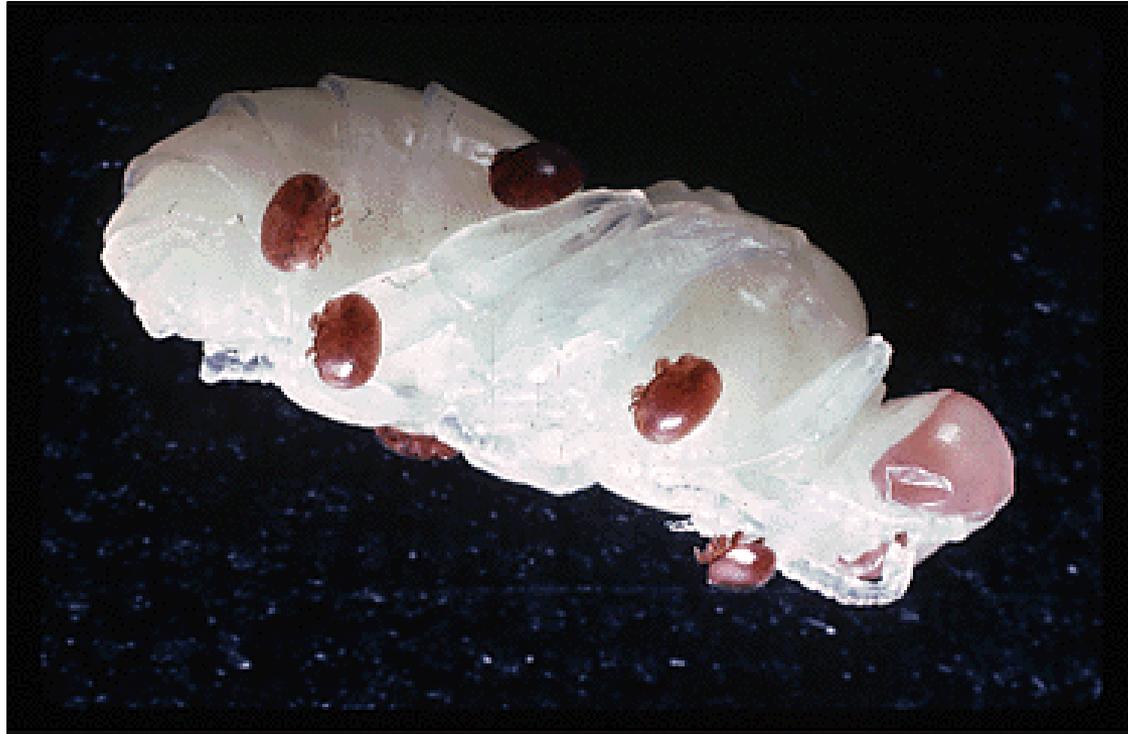
**female immerses herself in brood food,  
lays first egg after about 72 hours.**



**First egg laid is a haploid male, then mites mate in cell and must mature before the bee emerges (usually about 4 females emerge).**



**Varroa mites prefer drone brood but also invade worker cells. Most mites (80%) will be in the sealed drone and worker brood**

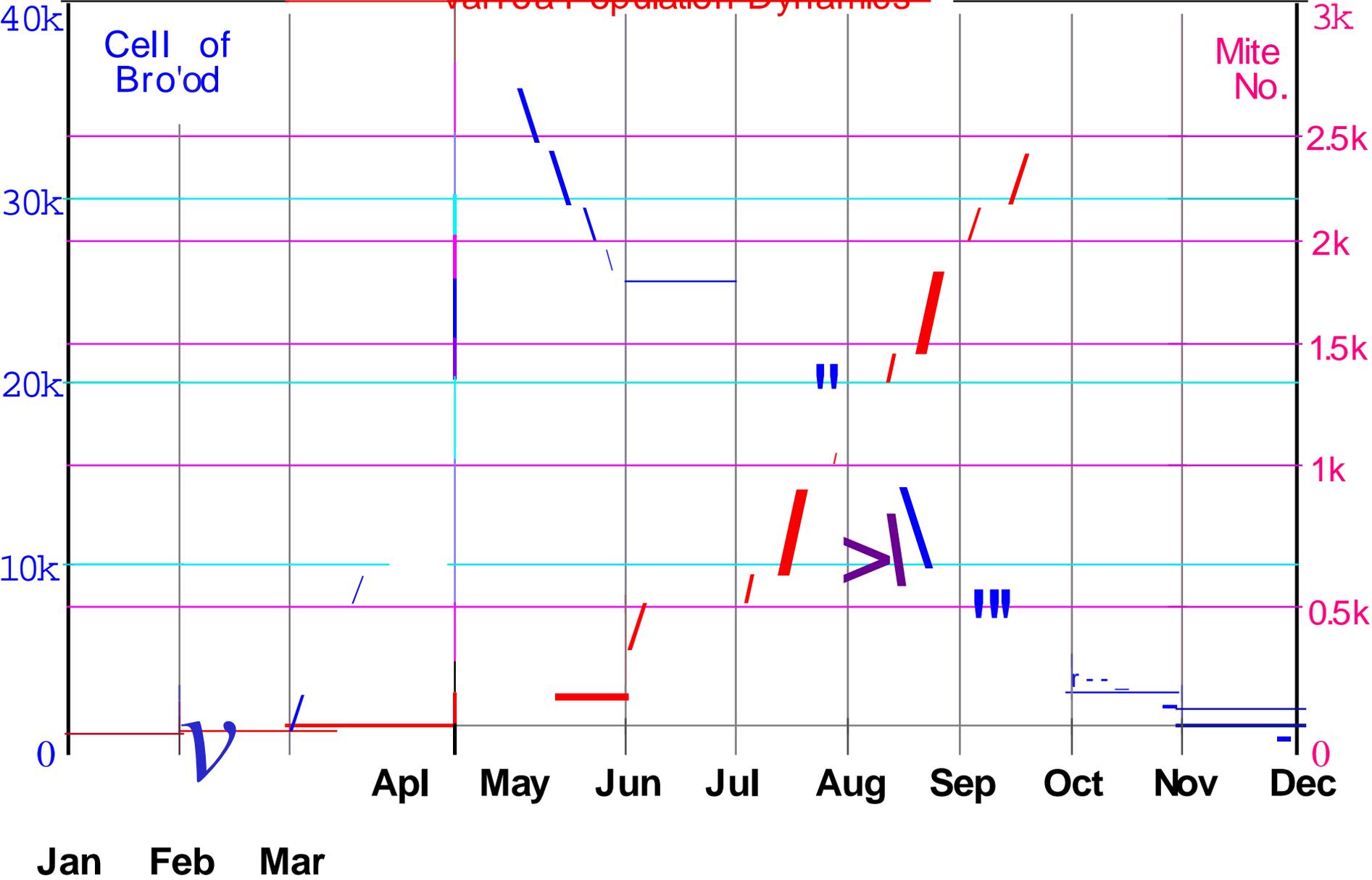


THEY CAME FROM ASIAN HONEY BEES

In *Apis cerana* Varroa only reproduces in drone brood



# Varroa Population Dynamics



# PARASITIC MITE SYNDROME



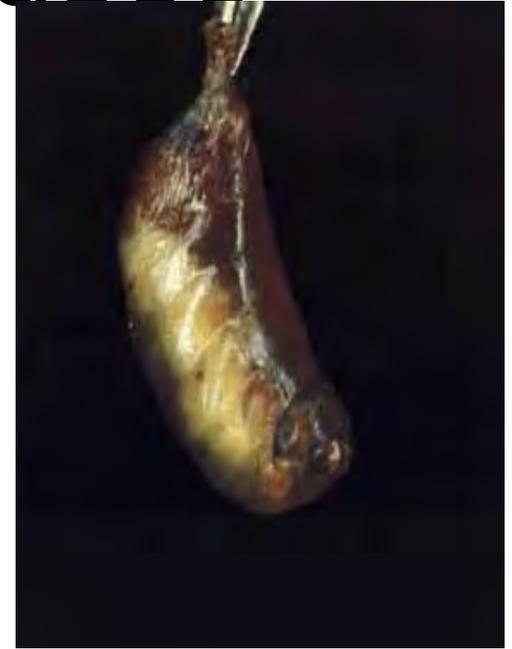
DWV

A close-up photograph of a honeycomb structure. The cells are hexagonal and arranged in a grid. Some cells are dark and empty, while others are filled with a light-colored, irregular substance, likely diseased brood. A single bee is visible on the right side of the frame, positioned on a cell. The overall color of the honeycomb is a mix of dark brown and reddish-brown.

Virus and brood diseases appear

# some vectored viruses from varroa

- Acute bee paralysis (ABPV)
  - Israel acute bee virus (IAPV)
  - Kashmir bee virus (KBV)
  - Black queen cell virus (BQCV)
  - Deformed wing virus (DWV)
- 
- 22 known viruses!



# Monitoring Mites in Your Hive

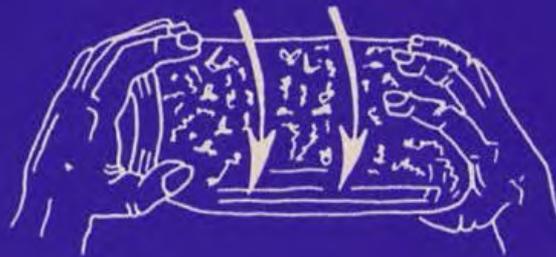
- Uncapping drone brood
- Ether Roll
- Ethanol wash
- Sugar Roll
- Sticky Boards

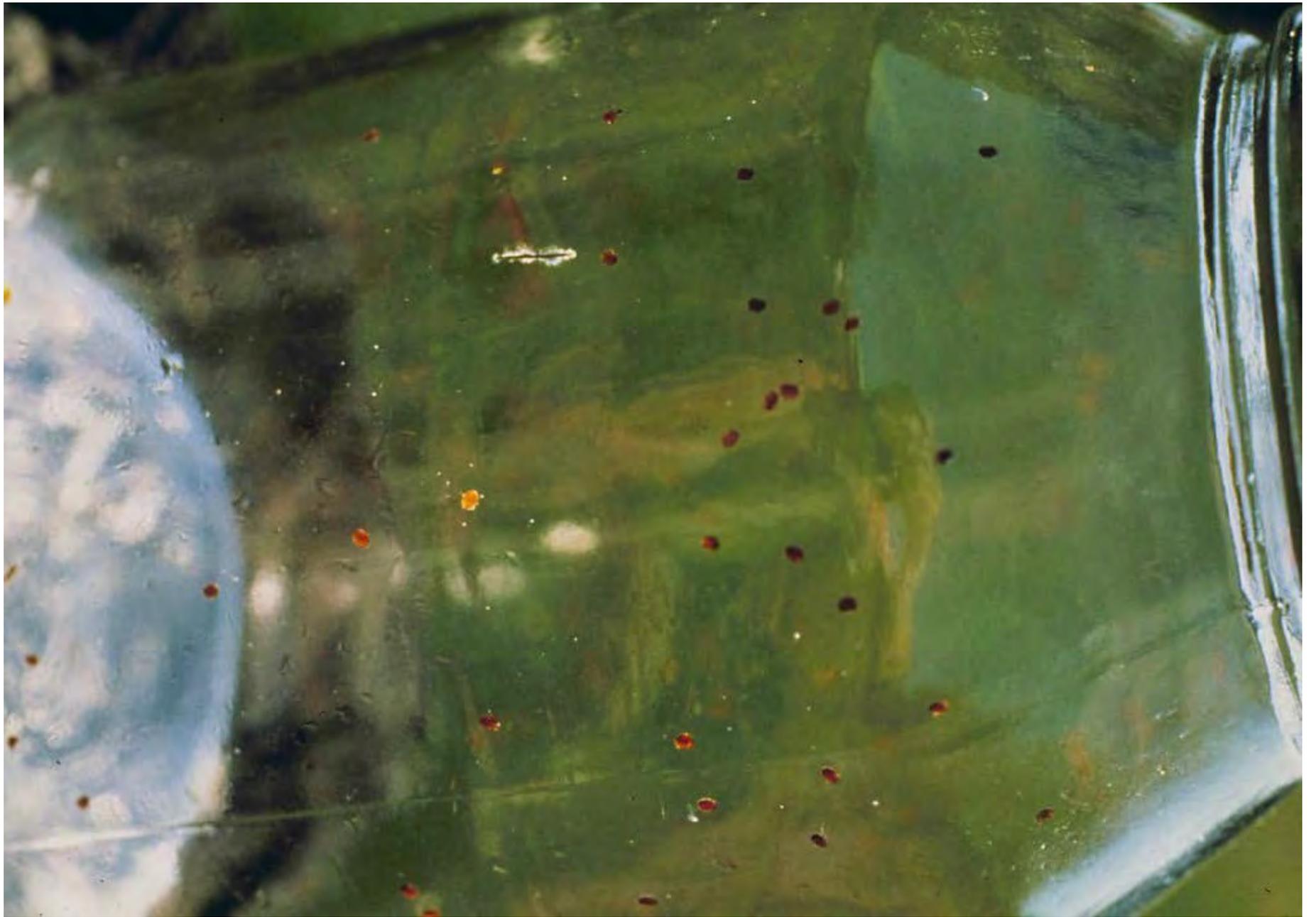
Looking for mites in drone brood





Looking for mites on adults





WHAT TRAIT WILL YOU  
MEASURE AND HOW TO  
MEASURE IT?

1)How do you choose the parents? (need to evaluate and keep good records)

2)how do you control mating's?  
(need to do I.I. or have drone source flooding in your mating yard)

# PRACTICAL WAY TO SELECT FOR VARROA RESISTANCE:



# GRAFT FROM STRONG HIVES THAT DON'T GROW MITES





**To select for traits: You need enough colonies (at least 100 would be sufficient)**

You need try to control mating's



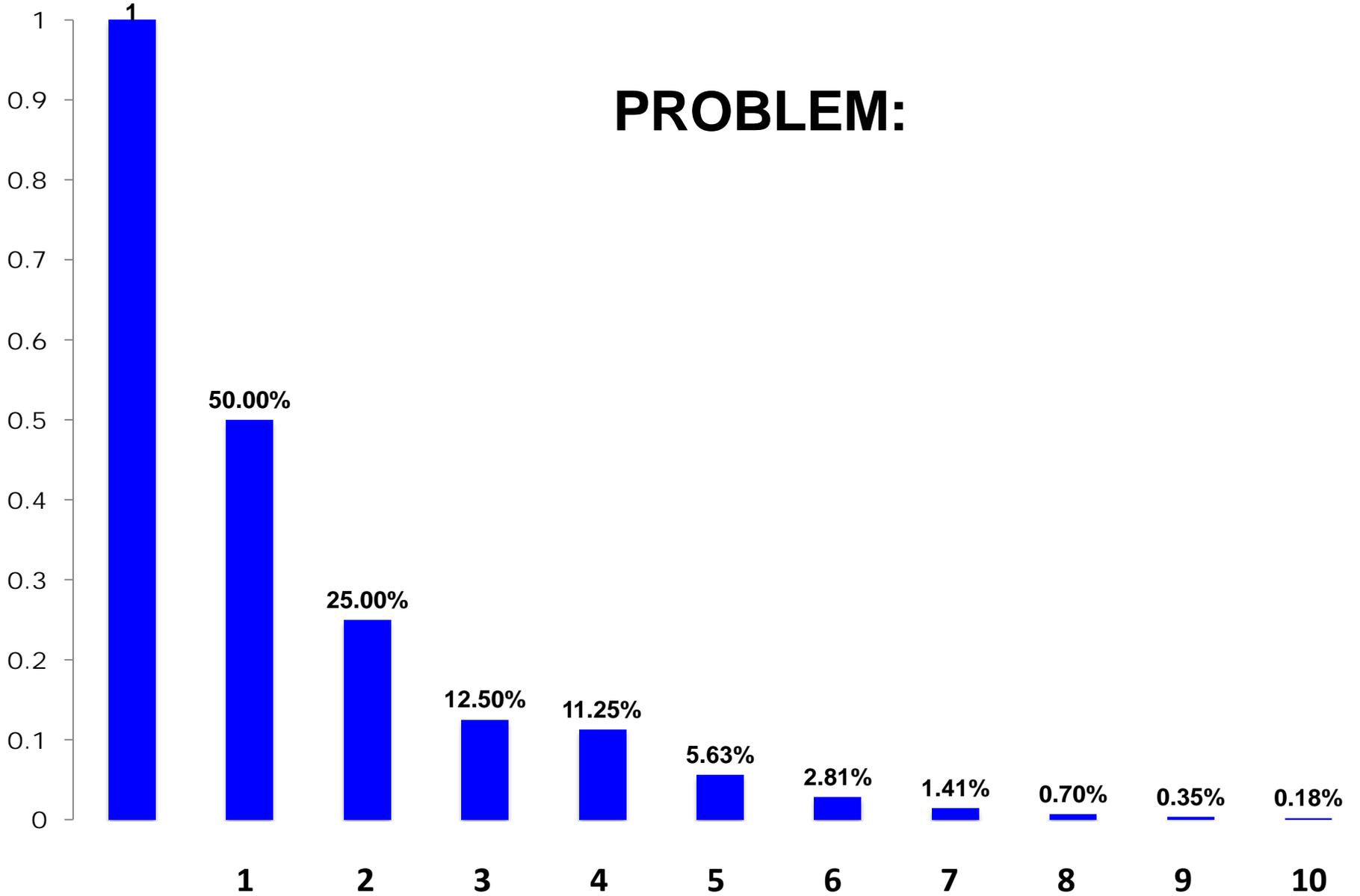
Try to have good colonies with drones in the mating yard.



**You need a trait you can evaluate,  
something that is beneficial,  
and that is heritable.**

**Start with bees that have good traits for  
your area!**

# PROBLEM:

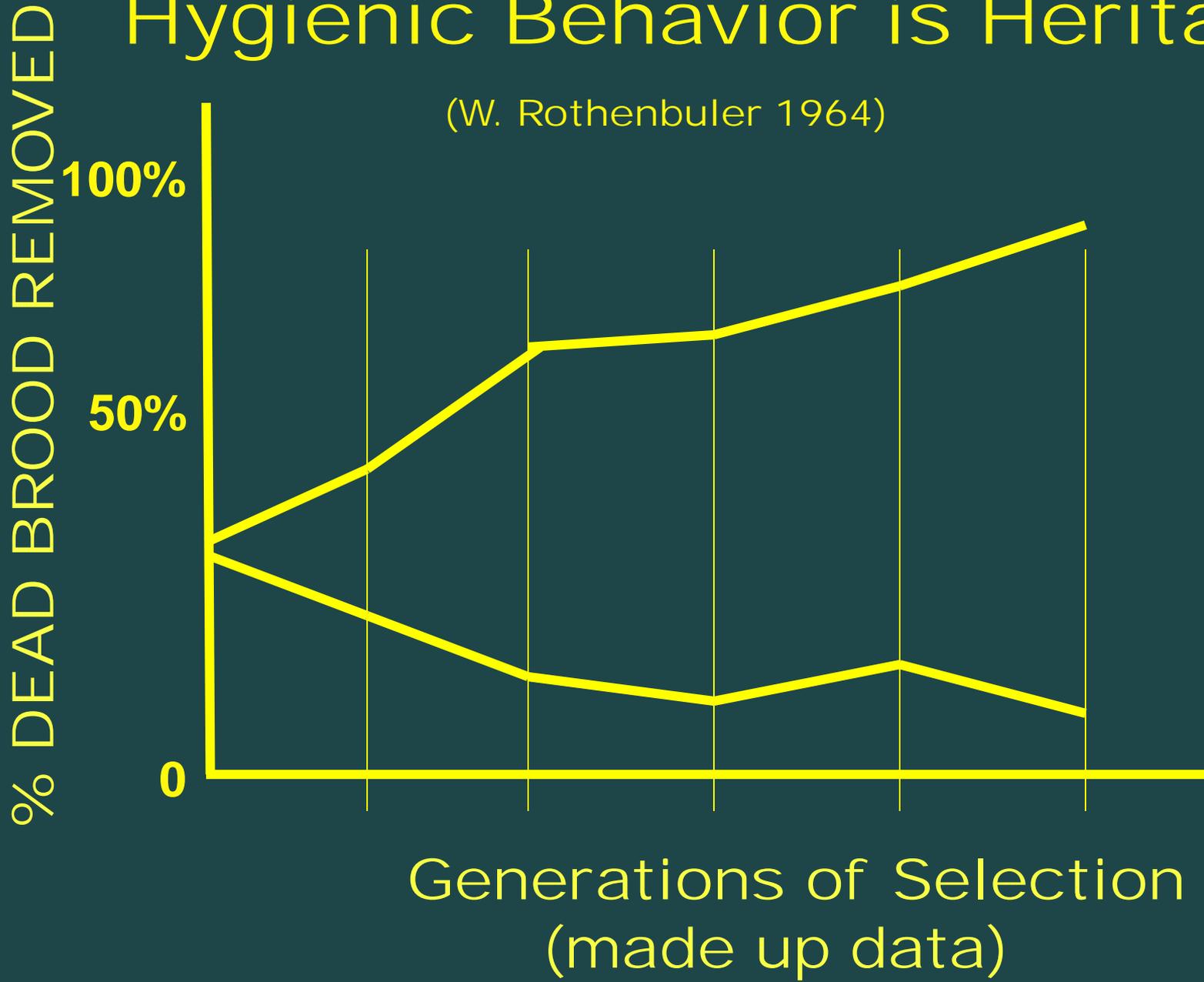




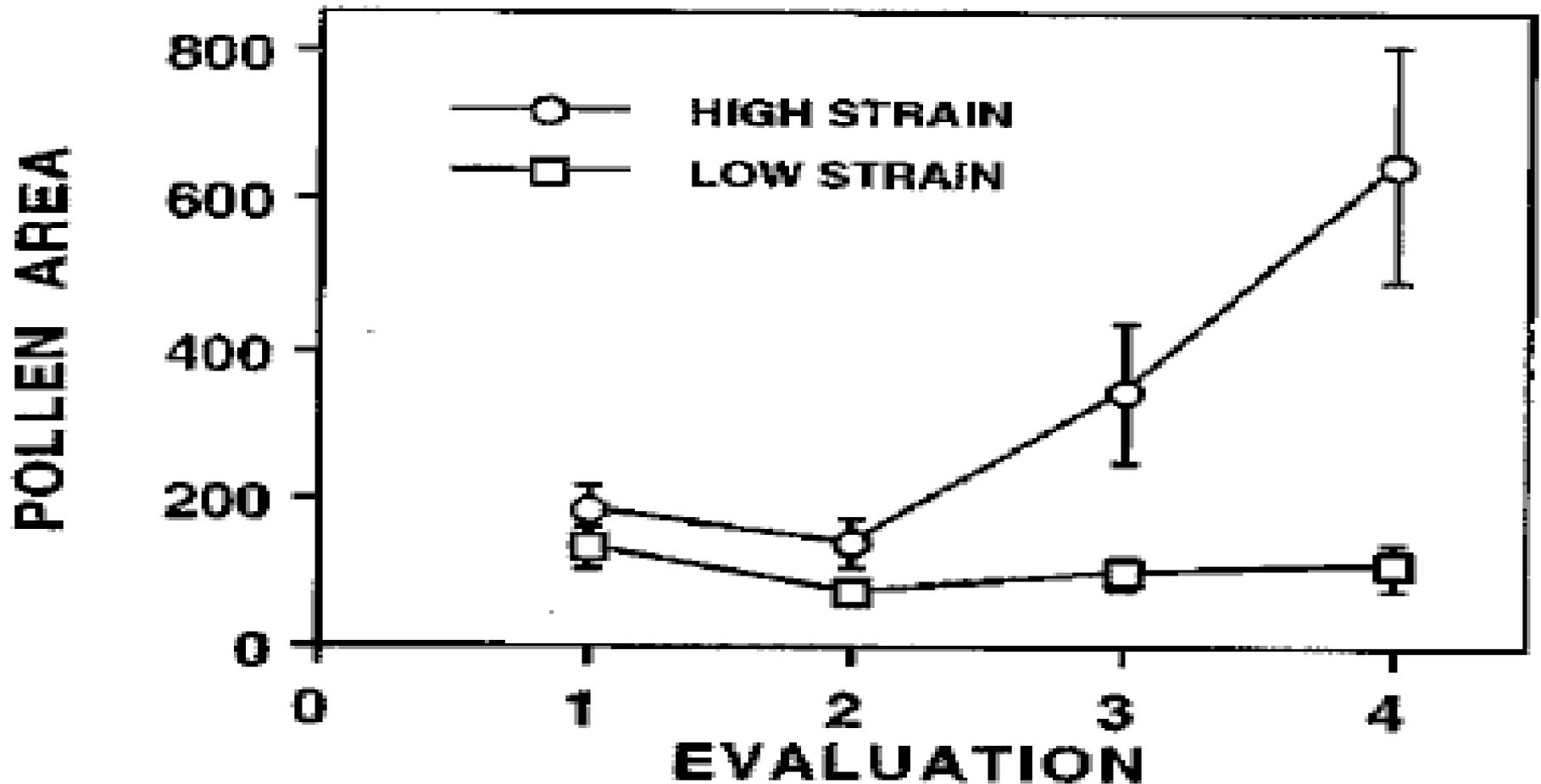


# Hygienic Behavior is Heritable

(W. Rothenbuler 1964)



Another example:  
Response to Selection for Pollen Hoarding



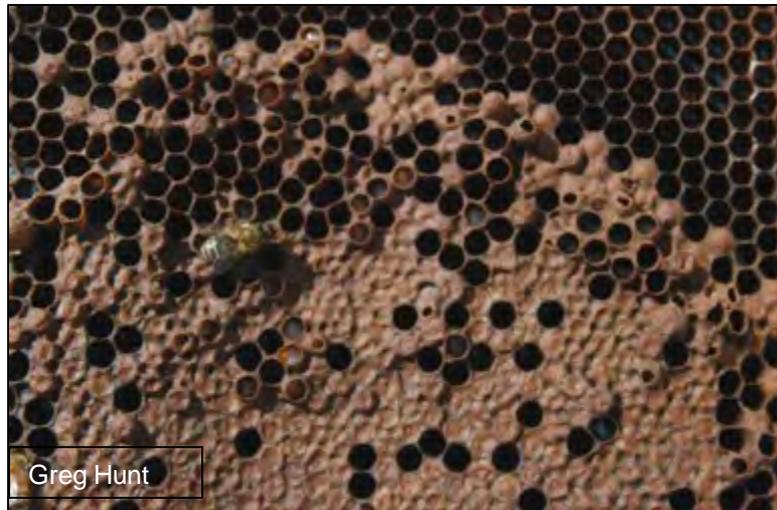




USDA Baton Rouge bee lab



# VSH BEHAVIOR



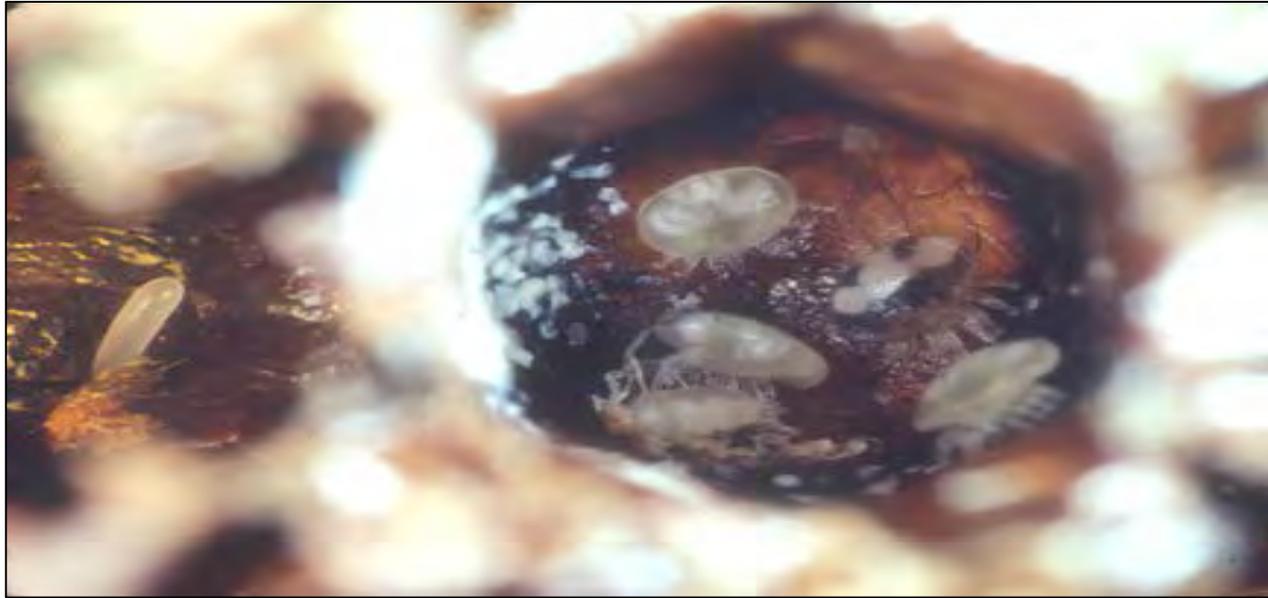
- **Varroa-sensitive hygienic behavior (VSH)**

- **Detection and removal of varroa infested brood**

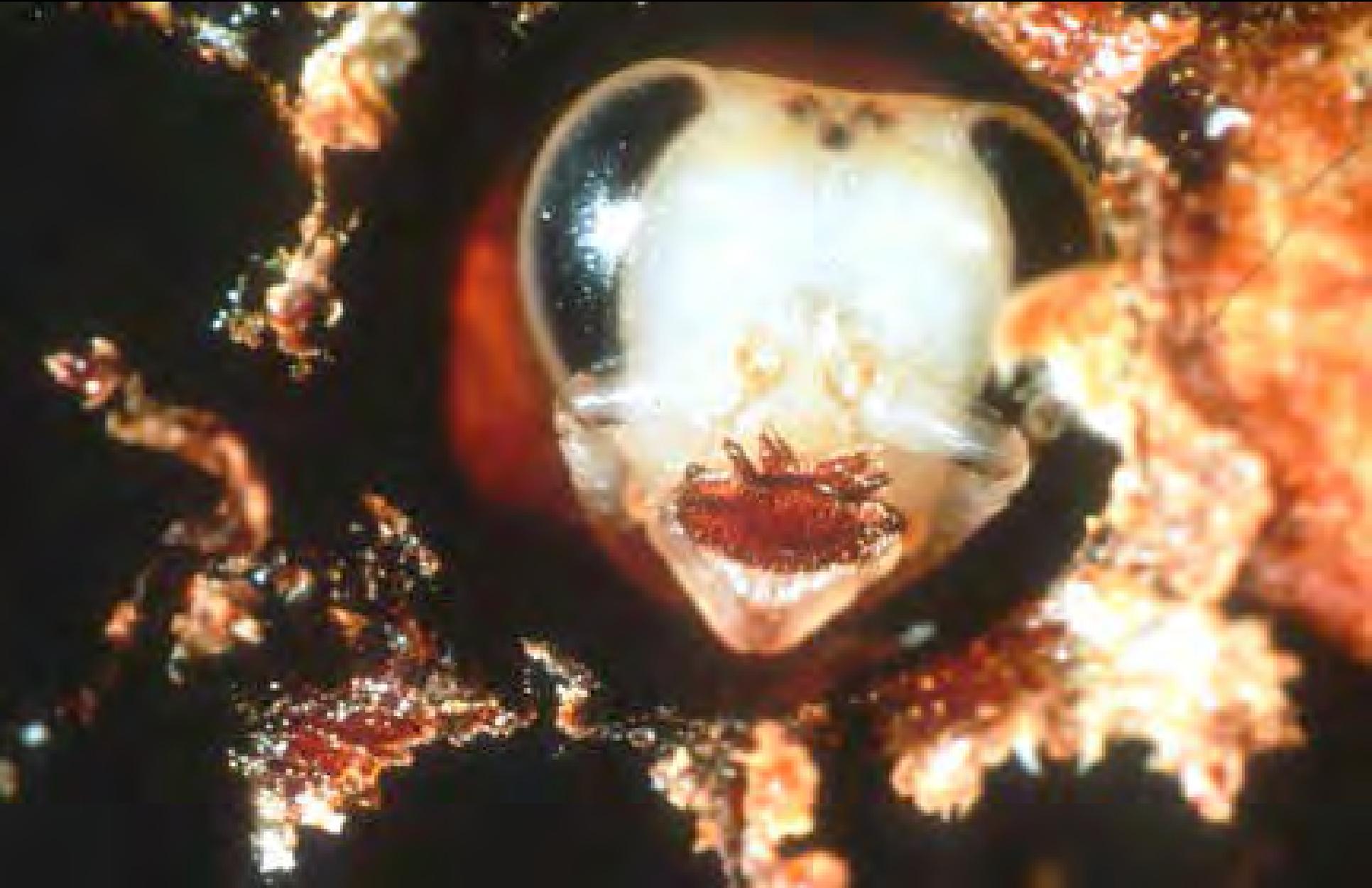
# Making uniform colonies



# VARROA MITES INSIDE THE CELL

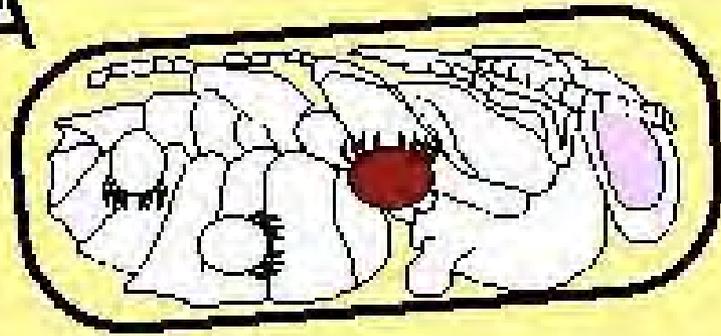


# Good age to check for reproductive mites

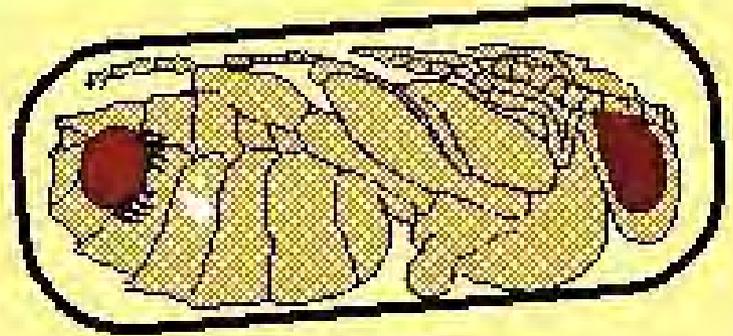
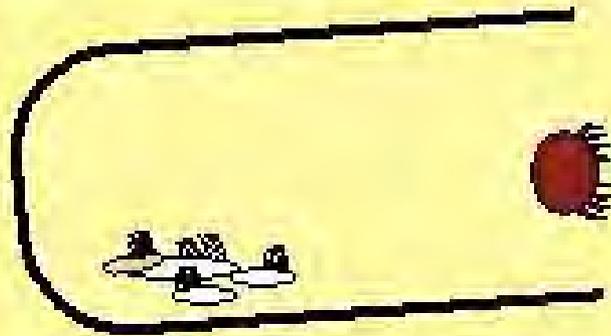
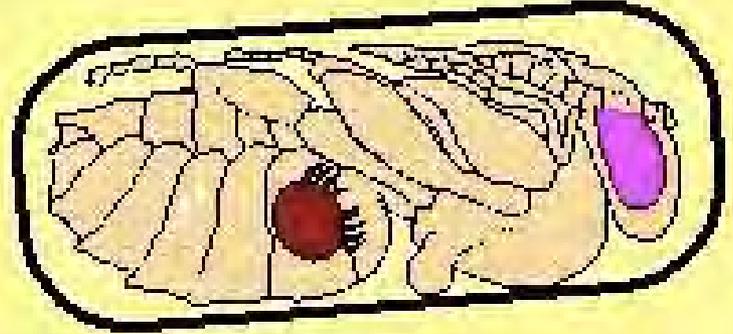
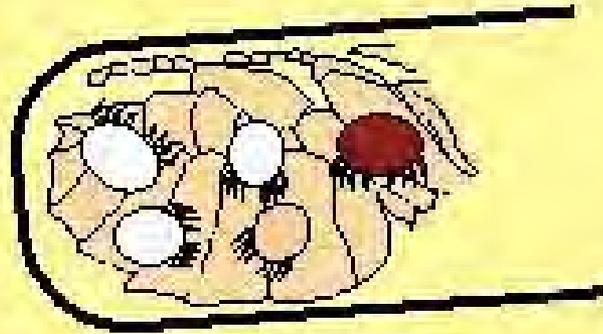
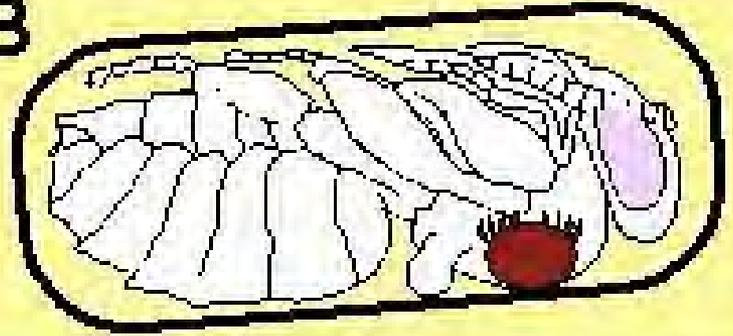


VSH is an indirect attack on Varroa

**A**



**B**





## Selecting for Varroa Sensitive Hygiene

**Bee Health**

November 12, 2010

Print

For a description of the VSH trait in honey bees, please see [Varroa Sensitive Hygiene and Mite Reproduction](#).

The [USDA-ARS Baton Rouge Bee Lab](#) has bred bees that express Varroa Sensitive Hygiene, and we would like to offer a simple technique that queen breeders could use to select for VSH. However, there is no method at this time that is both easy and accurate. We have developed several types of measurement, but the most accurate require significant time at a microscope to evaluate colony performance (Fig. 1).

### Connect with us

### Welcome

This is where you can find research-based information from America's land-grant universities enabled by [eXtension.org](#)

**Select a different institution**

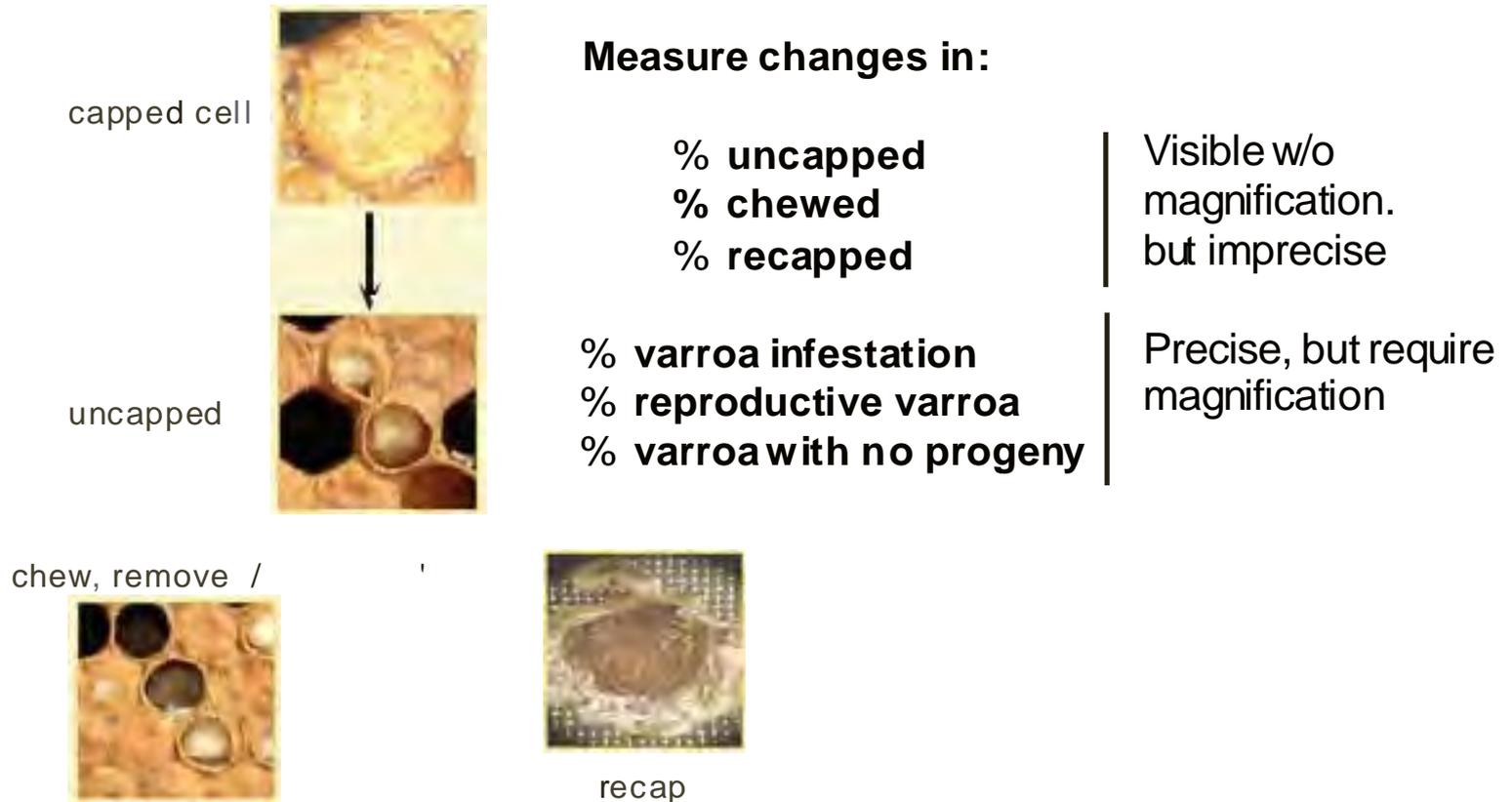
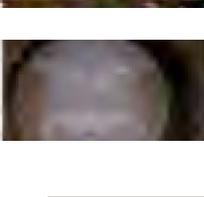
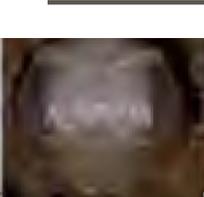
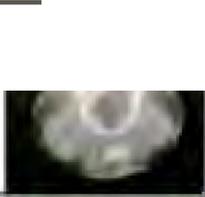


Figure 1. - Types of changes in brood combs that can be measured without and with a stereo microscope. For example, the percentage of pupae uncapped during a certain period of time could potentially be used to breed for varroa resistance. Uncapped pupae are visible without a microscope, but measuring the % uncapped pupae is highly variable and does not always predict VSH behavior. On the other hand, measuring the percentage of infertile mites (those without progeny) predicts strong varroa resistance, but it requires detailed microscopic examinations of brood to measure.

Days past capping	Brood stage		Eldest offspring normally producing mites	
3		prepupa		Varroa egg
4		pupa, white eyes		first protonymph (male)
5-6		pupa, pink eyes		second protonymph (female)
7-9		pupal purple eyes		first female deutonymph
10-11		pupal, brown head, black eyes		first adult daughter

# Uncapping of mite-infested cells due to VSH





24hrs LATER!



# GENERAL HYGIENIC BEHAVIOR

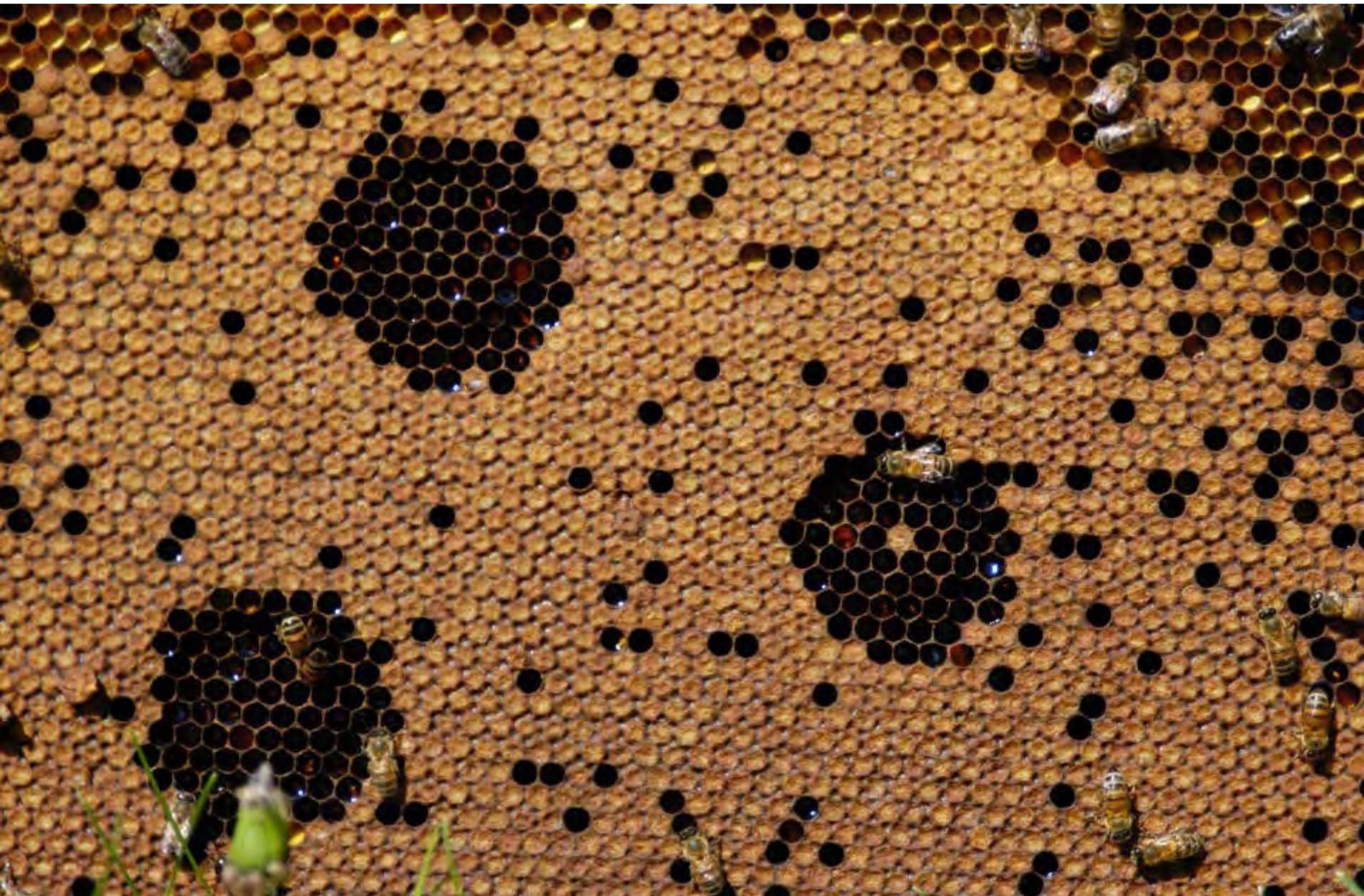
Freezing capped brood  
with liquid nitrogen  
(250 ml)

OR PIN PRICK METHOD











# GROOMING BEHAVIOR



# **MIGUEL AND ERNESTO'S STUDY**

## **In Mexico**

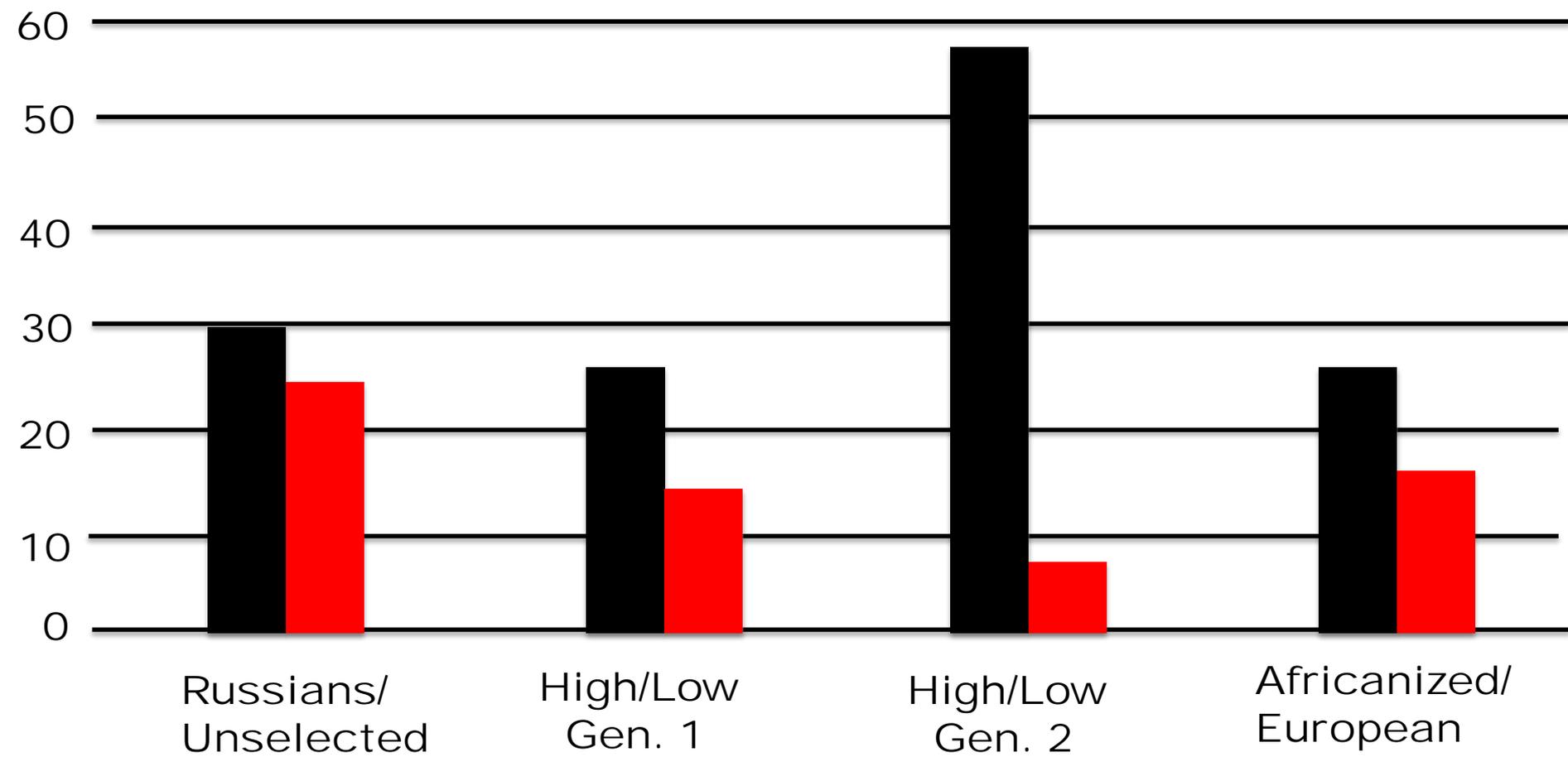
**Found that grooming behavior was important**

**Those with the lowest infestation overall had low adult infestation show highest mite drop !**

**They also had the most chewed mites on board, and also in the lab assay.**

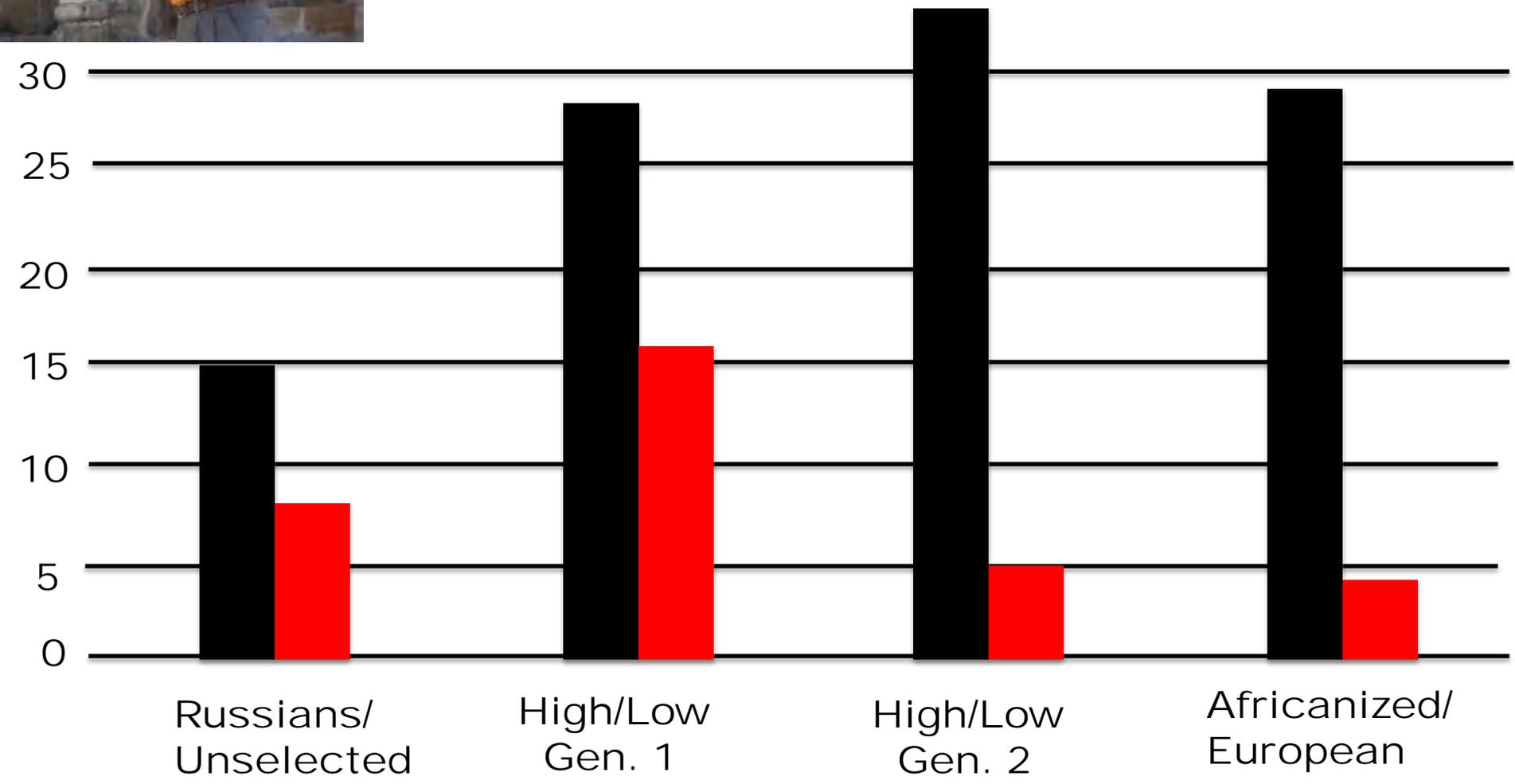


# % Mites Chewed in Hives





# % Mites Removed in Assay



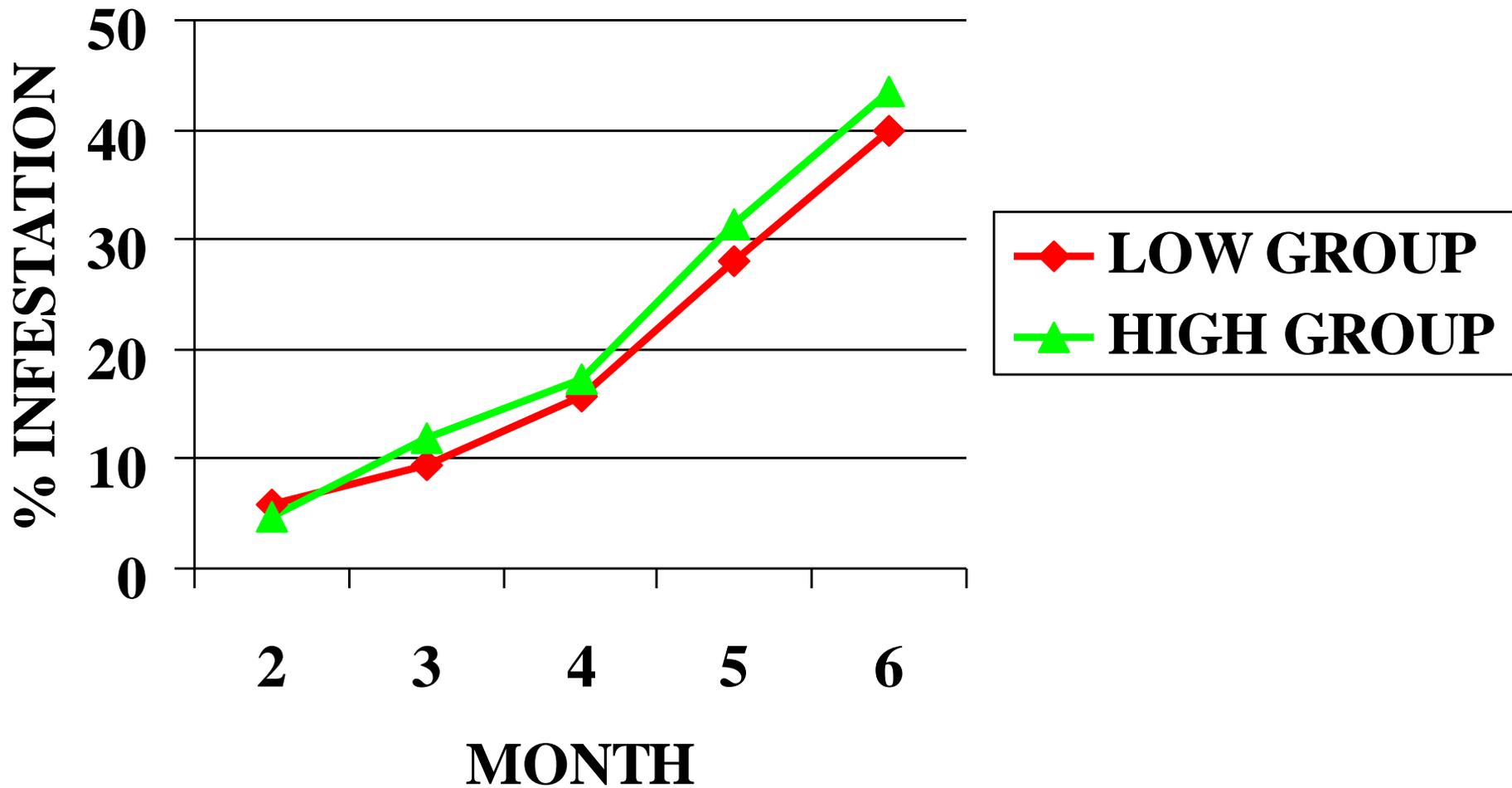
# What is responsible for behavioral traits?

- Genotype of queens and workers
- Environmental factors

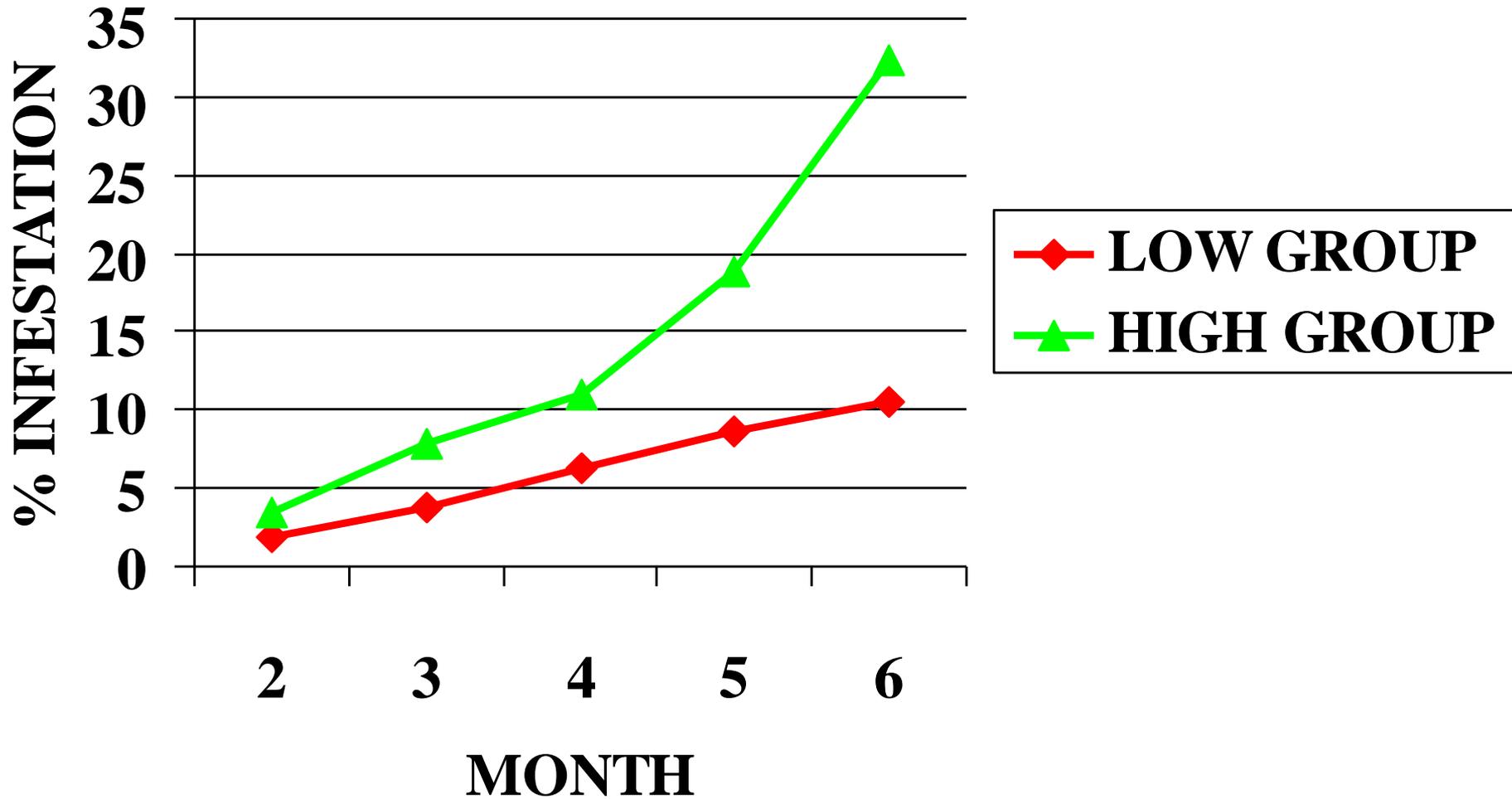


# Grooming Behavior

# MITES IN BROOD



# MITES ON ADULTS



# Selecting for chewed mites is sticky business!





68+ 65 104 117 72 78 108 5 70 33 80 18 76 17 23 14 125

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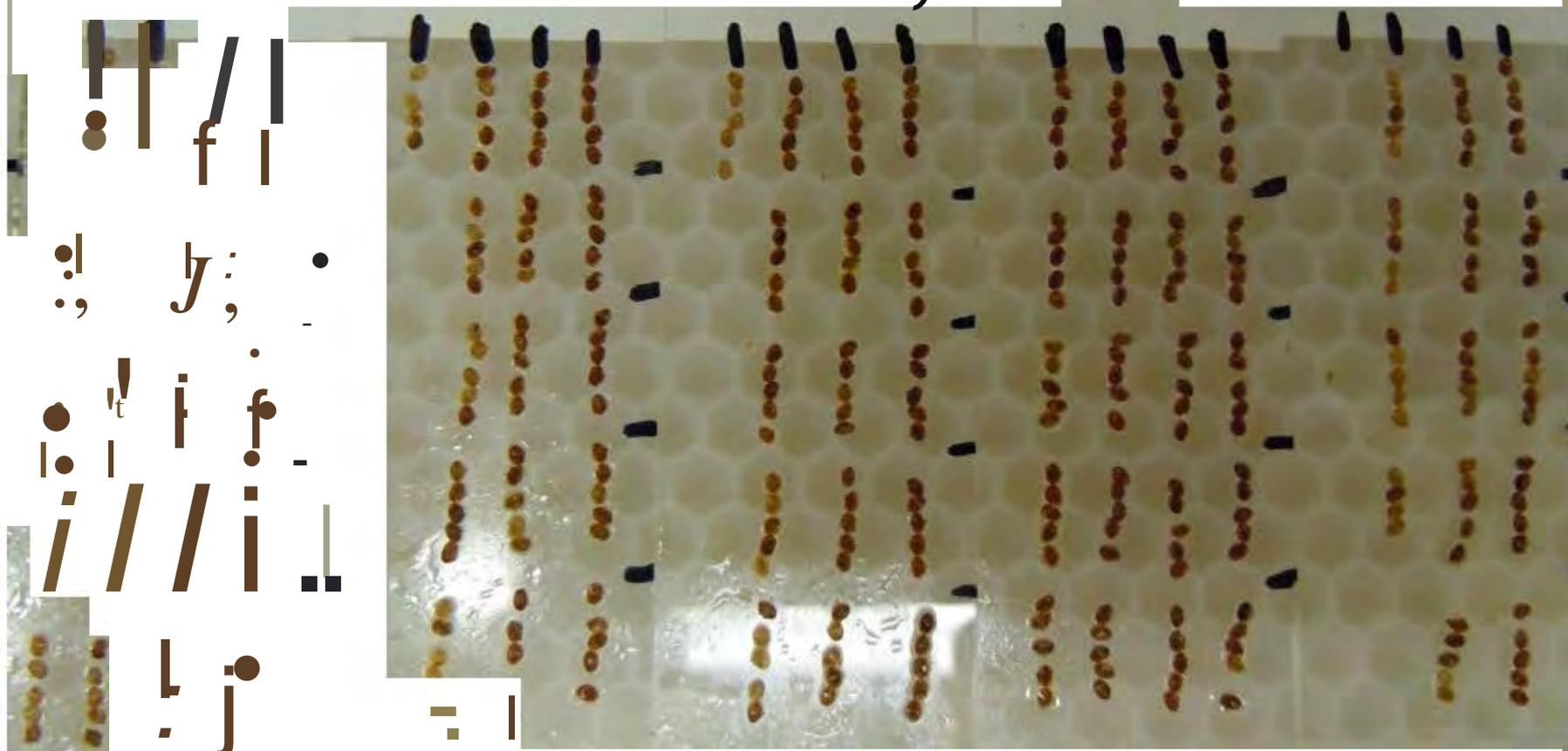
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# Current Selection Program: Grooming and Biting Behavior



# Hierarchical Selection:

Measure proportion chewed mites.



Eliminate those that show disease or did not control mite population growth.



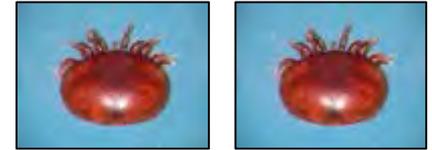
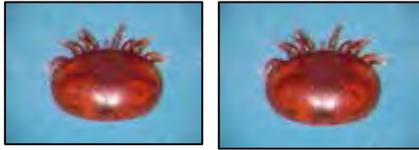
Eliminate those that do not pass freeze-killed hygienic test.



Select breeder colonies.

- **Started with SURVIVORS, VSH + Russians**
- **Method for the first 7 years:**
- **Measure mite drop (and strength) multiple times**
- **Measure honey yield**
- **Test best sources for (VSH) hygienic behavior**
- **Use instrumental insemination.**

# Response to selection at the colony level is also at the individual



**Initiating grooming dance\***

**grooming**

**Biting**



**VSH**

**General hygiene**

**Undertaker**



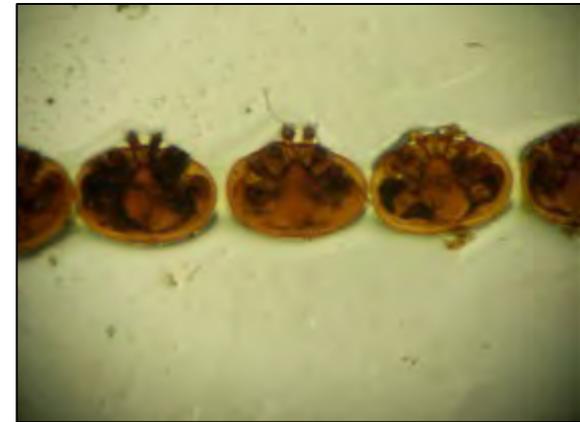
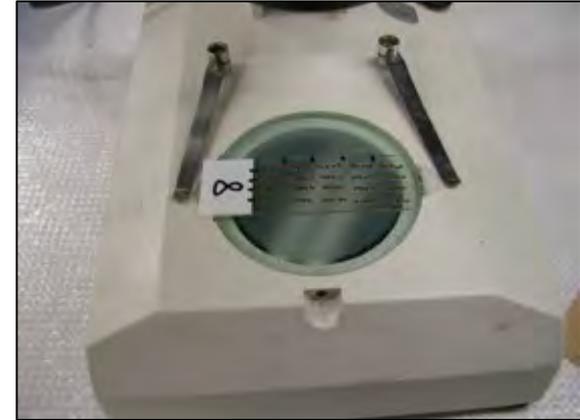
# Instrumental Insemination

High x high chewers are crossed and monitored  
For more response to selection

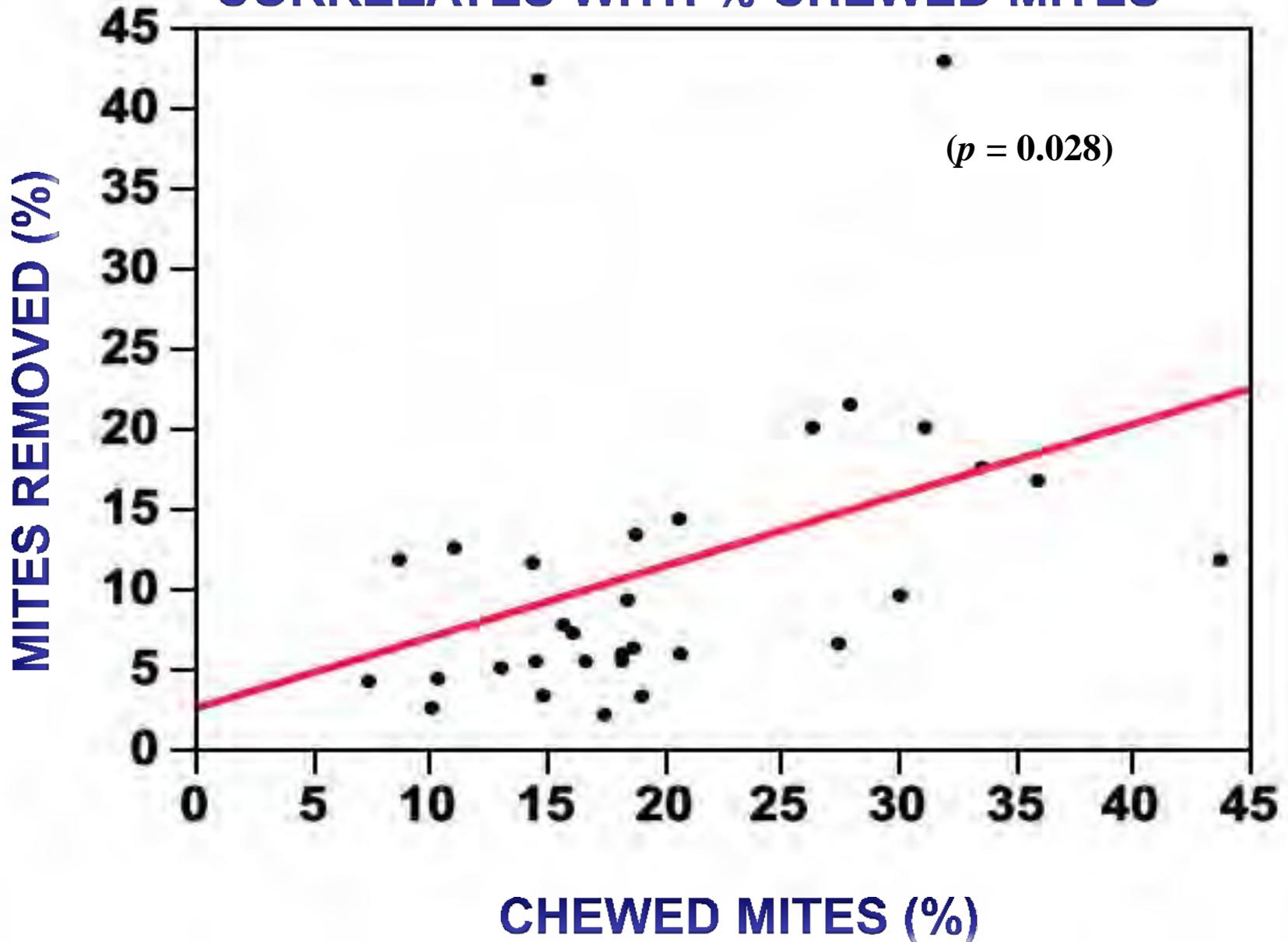
Daughter queens are then grafted  
from these\*



# Collecting and counting chewed mites



# MITE REMOVAL BY GROOMING CORRELATES WITH % CHEWED MITES



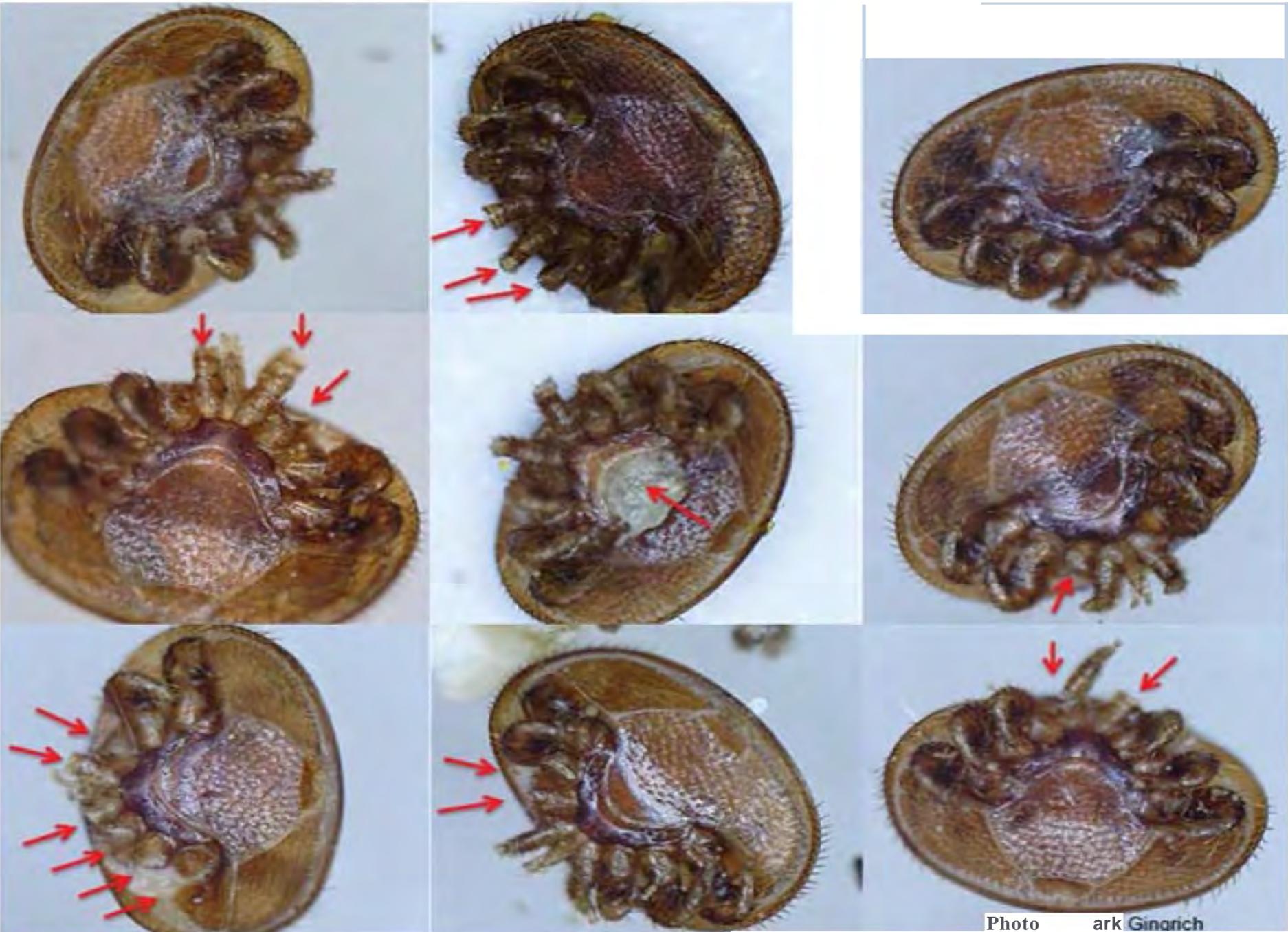


Photo ark Gingrich

*Mites that have been chewed by the mite-biters*

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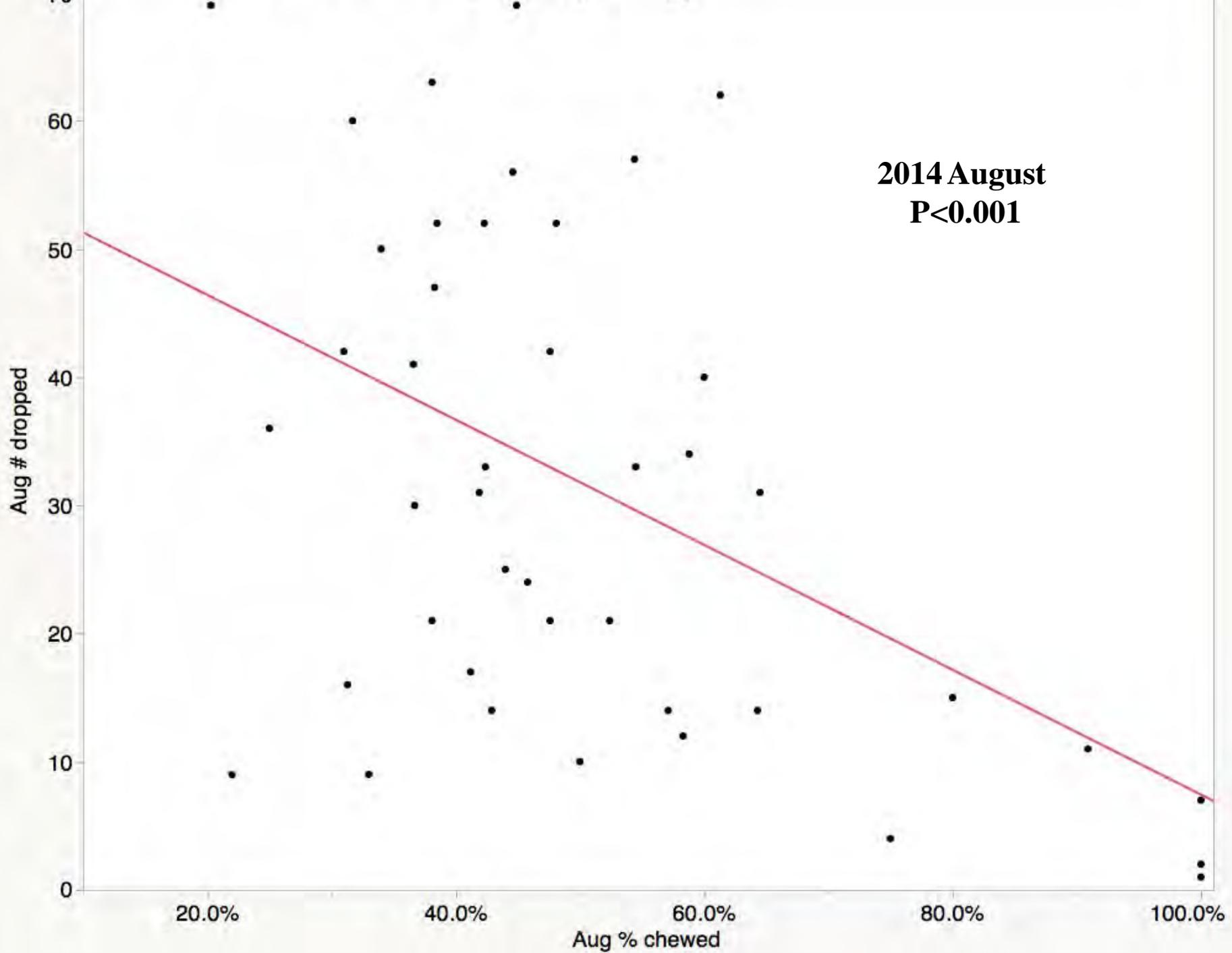
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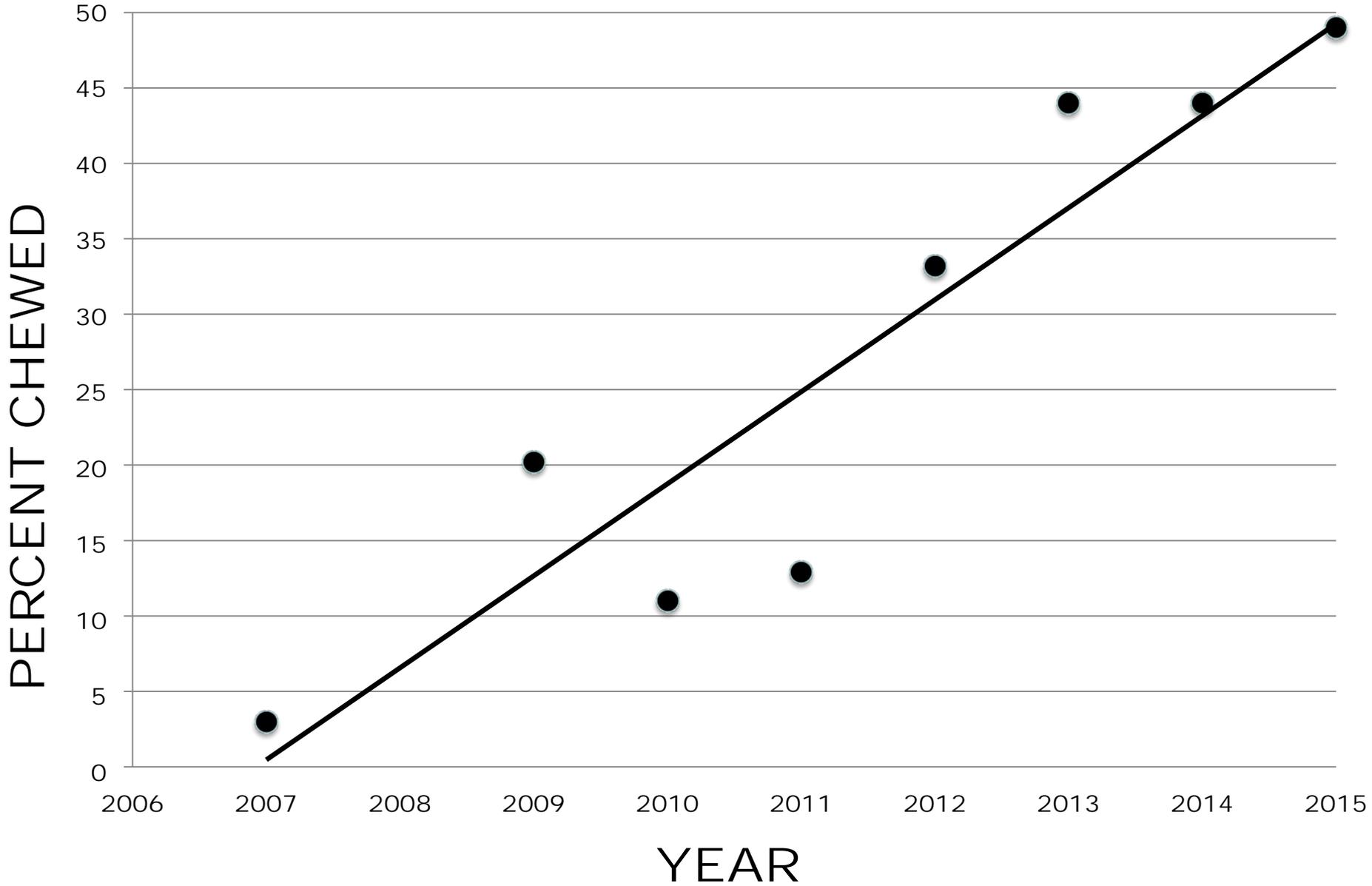
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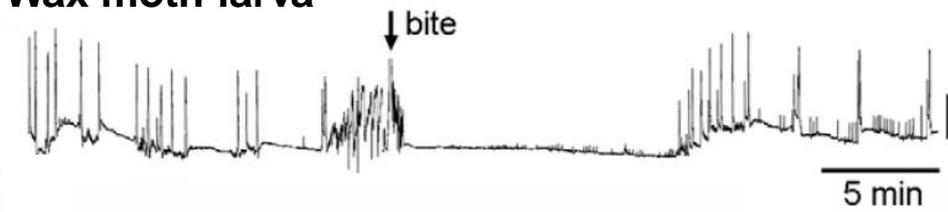




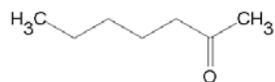
# MITE-BITING IS A HERITABLE TRAIT.



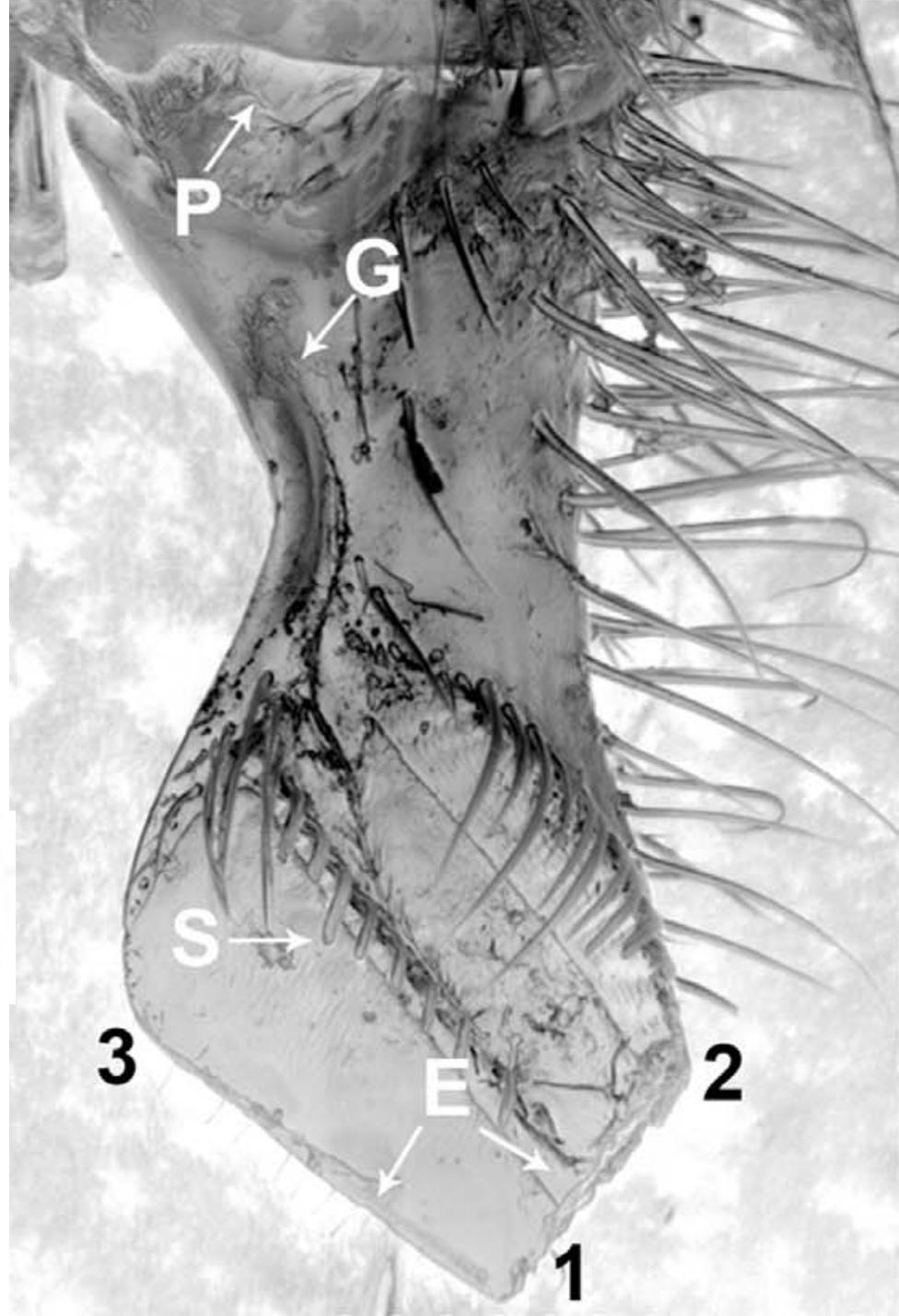
Wax moth larva



The bite of the bee  
contains 2-heptanone



Varroa



# How to select for bees that bite varroa mites and groom them from their bodies

Krispn J. Given, Greg J. Hunt, David M. Shenefield, Ginger D. Davidson Dwight C. Wells  
and Dan P. O'Hanlon

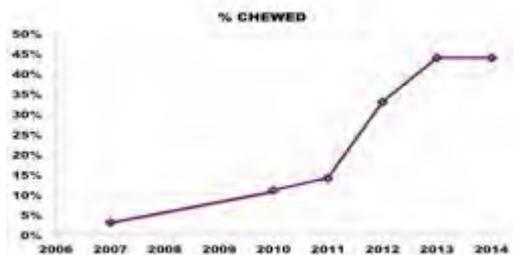
## Abstract

Grooming behavior is one of the key mechanism honey bees (*Apis Melifera*) execute to kill and remove *Varroa destructor* from their bodies. Two of the grooming behaviors identified with chewing and mite removal are responsible. Through selection with instrumental insemination we have seen *Varroa Destructor* chewing levels go from 3% to 43%. The HHHBBC has been instrumental in the selection and dissemination of northern mite-resistant stocks.

Grooming behavior is one of the mechanism of defense against *Varroa destructor*. Mite grooming behavior is the ability to remove mites and is associated with chewed mites.



Mites are placed on there idiosoma and inspected for damage.



This shows a response to our selection. We may be near the peak for grooming behavior but need make sure every colony has a high levels as possible by requeening the low chewers. We are working on a better measure for grooming behavior, and have a better correlation between % chewed mites and reduced mite populations in 2014 than we did in 2013, in 2013 the mite populations were higher and more variable between colonies.



Inaugural HHHBBC at the Purdue University bee laboratory, where 75 queens were in distributed. Breeders from PA, WV, OH, attended the three day



Annual queen rearing classes taught at the Purdue bee lab, in 2014 we had 34 students.



Instrumental insemination classes have also been a good resource for breeders to learn more about the importance of selection at the Purdue Bee Lab.

# 2016 Insemination Fest

PA, OH, WV, MI, IN, IL, KY, TN

130 queens inseminated



# COMMUNITY STOCK EVALUATION IN 2014-2015

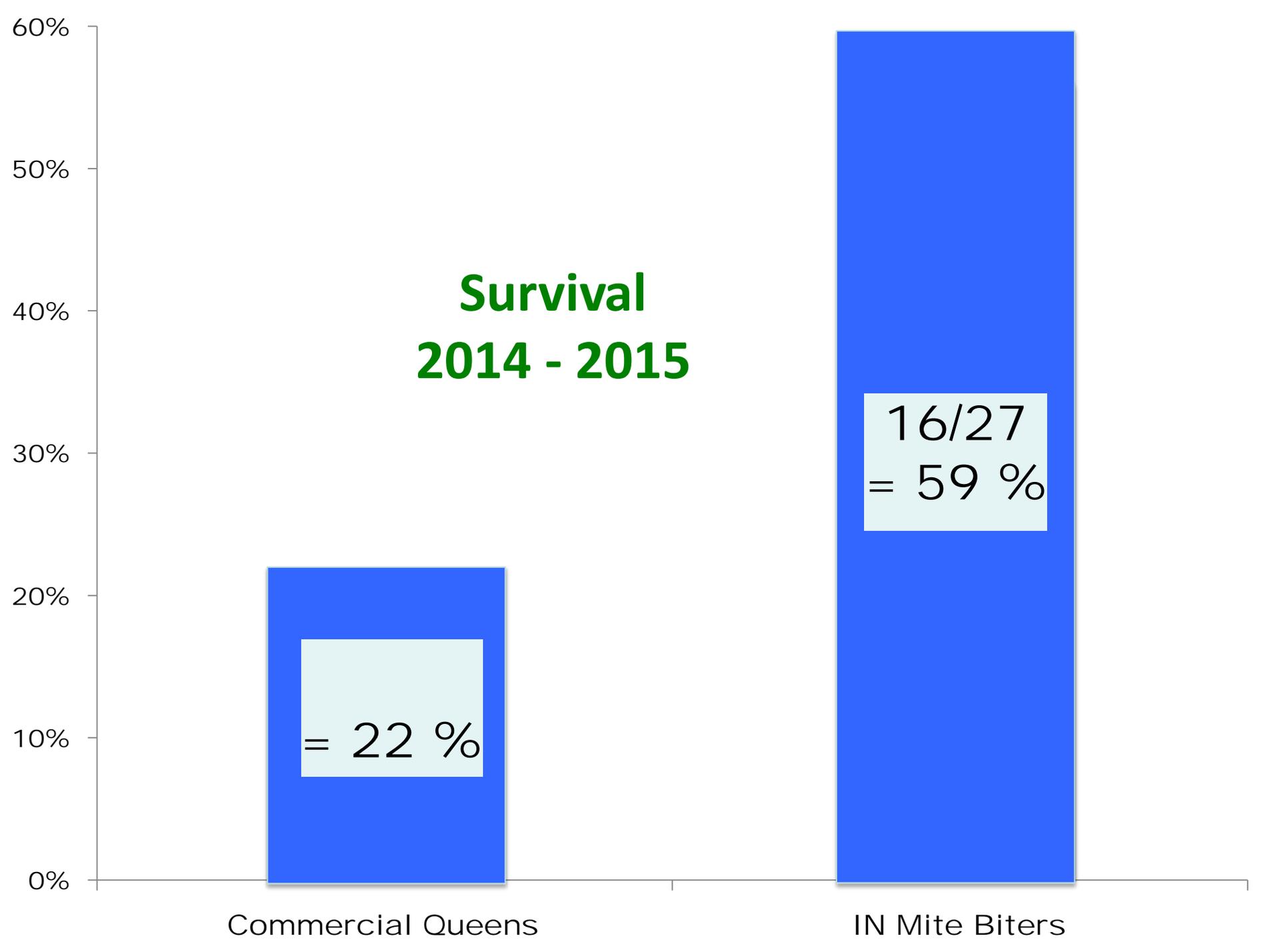
54 queens, 23 beekeepers,  
3 commercial sources,  
2 Purdue sources.

Commercial hives had 3X more mites



PHOTO: RATNA THAPA

# Survival 2014 - 2015

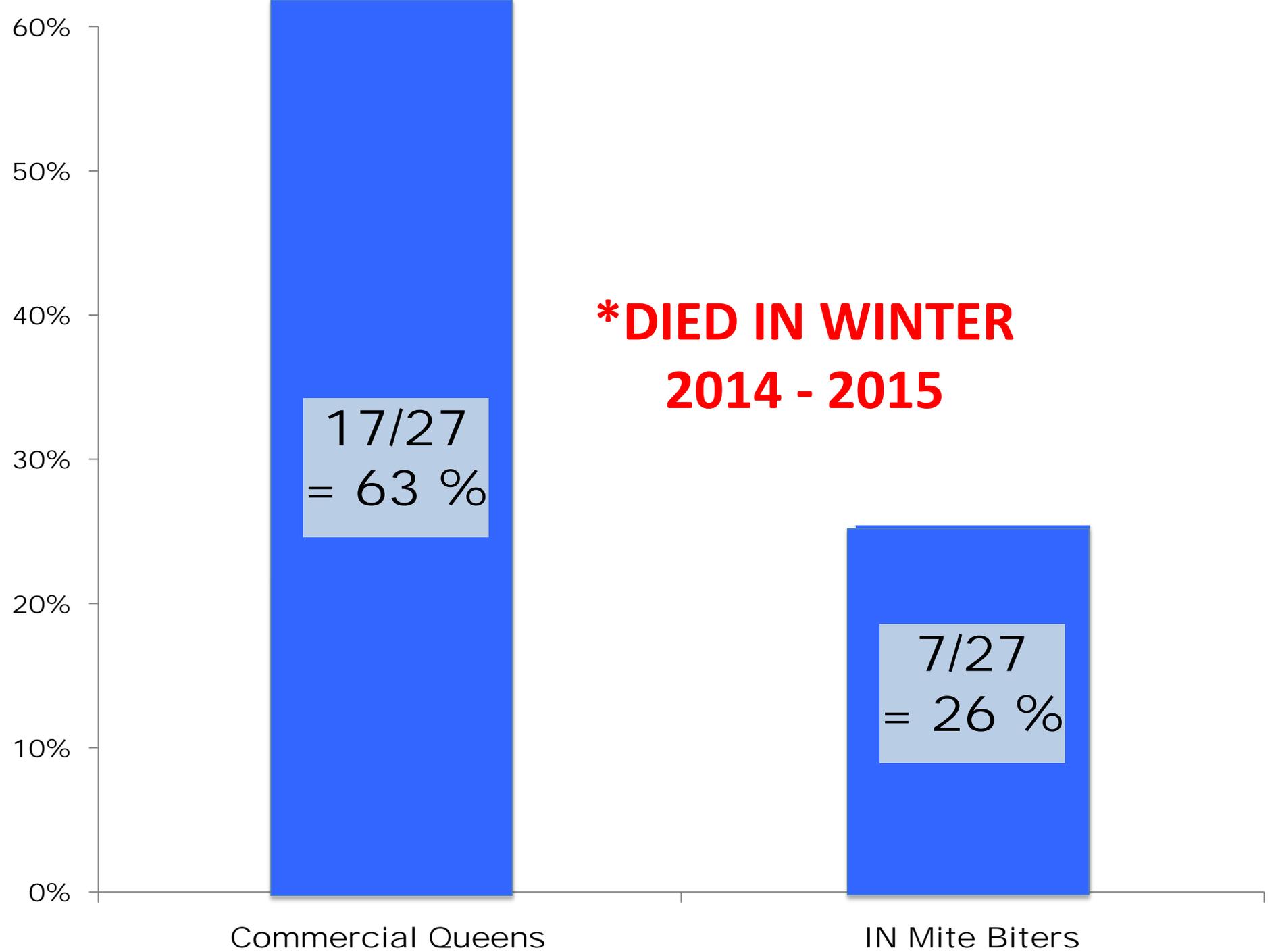


= 22 %

16/27  
= 59 %

Commercial Queens

IN Mite Biters



# IN QUEENS MADE 40 LB MORE HONEY



**Ten of eleven beekeepers preferred IN mite biters**

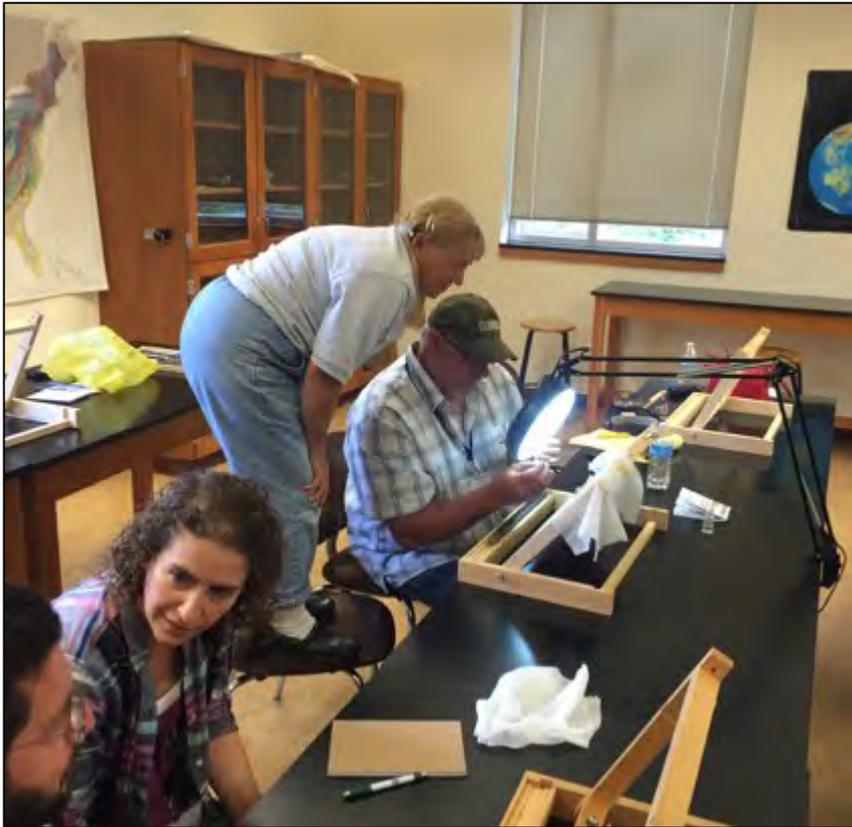




# HAS July 13-15, 2017

## Evansville, IN

July 13



# 2016 I.I. Class





# Future plans:

Cage bee assays for grooming!





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0 Madison Milwaukee

0 Cedar Rapids  
0 Iowa City

0 Chicago  
0 Naperville

0 IOWA  
0 Des Moines

0 ILLINOIS  
0 Springfield  
0 Champaign

0 Kansas City  
0 Lawrence

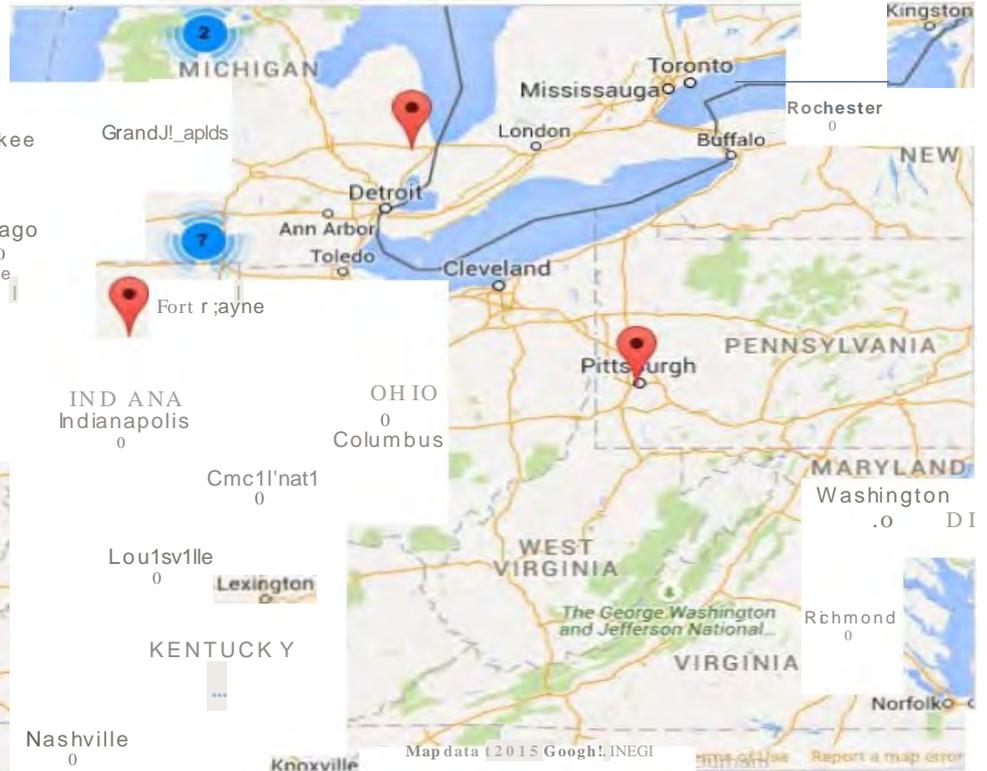
0 St. Louis

MISSOURI

0 Springfield

0 Fayetteville

0 Nashville



# The Bee Hive

Home   Beekeeping   Bee Genetics   Bee Diseases   Bee Health   Bee Links   Publications   Photo Gallery

Welcome to the Hunt lab's Bee Hive. We are involved in honey bee research and education.



W is you were here   C   l  
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heartlandbees.org

## 14TH ANNUAL H.A.S. CONFERENCE

featuring sessions with:

Dennis vanEngelsdorp, Sue Cobble, Larry Connor, Joe Traynor, Clarence Collison, Kim Flottum, Renata Borba, Roger Hooplin, and Ken Schramm, and Brian Milbrath

conference  
sponsored  
by:

Bee Culture

MANN LAKE



begin to  
adv. wort.shop...  
beginning nucs splits,  
queen raising,  
drone catching,  
new propolis discoveries  
honey tasting  
bring 3 jars to swap!

**CHILDREN'S PROGRAMMING:**  
candle-making, murals, seed bombs

**REGISTER NOW!**  
www.heartlandbees.org

Events

Do you have a swarm to pick up? Find a local swarm catcher here:  
<http://www.in.gov/Ldnr/entomoloLS755.htm>.

When bees swarm the queen and most of the live bees leave after filling up on honey. Bees in a swarm are usually very gentle but leave them alone! With all those bees flying one could get stuck in your hair. The swann sends out scout bees to find a new cavity to rest in. They usually stay for a few days and then they are off to their new home.

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# Thank you

## Heartland Honey Bee Breeders Coop

### National Honey Board Funding

### NEW USDA Funding !!!

**Greg Hunt**



**Jennifer Tsuruda**



**Gladys Andino**



**Jeff Berta - PA**  
**Matt Evans - IN**  
**Tammy Horn - KY**  
**Joe Kovalevski - OH**  
**Scott Martin - IL**  
**Megan Milbraith - MI**  
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**Dave Shenefield - IN**  
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