



D. Sammataro, PhDUSDA-ARS Carl Hayden Honey Bee LabTucson, AZ

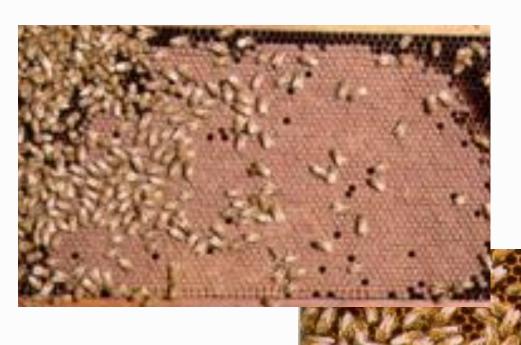
Our Website: http://gears.tucson.ars.ag.gov







Healthy Colony Brood Frames





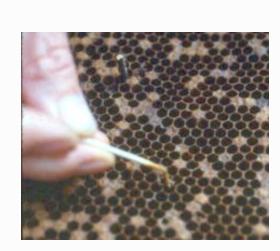


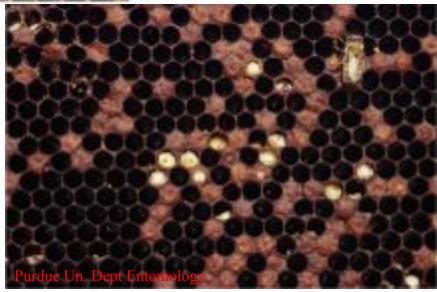




Parasitic Mite Syndrome Visible Signs on Brood









Visible Symptoms on Bees



Virus symptoms (DFW)



Bees dragging out pupae

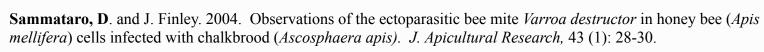




Varroa can be found on Chalkbrood mummies













Diagnosing Varroa Mite (also *Tropilaelaps*)



1. Ether or Sugar Roll (wash)

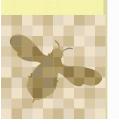


2. Uncapping drone brood



3. Sticky boards or bottom debris









- A. Rolls or shakes
- B. Brood examination
- C. Hive debris/sticky boards
- 2. Estimating Mite Densities
- 3. Thresholds for Treatment
- 4. Future work













- Brush or shake 300 bees into a wide-mouth jar from one frame with capped brood and nurse bees. NOT THE QUEEN.
 - 300 bees = 2 inch (5.08 cm) layer of bees
 - or 100ml (0.42 cup) or make your own marked container [1/3 cup (78.07 milliliters) is about 238 bees or less]









1A. Ether Roll

- Apply a two second burst of ether (automotive starter fluid) into the jar. (FLAMMABLE - keep away from fire and working smokers)
- Replace the top and shake vigorously for 30 seconds.
- Gradually rotate the jar horizontally.
- Look for any mites sticking to the sides; ether kills the bees, and causes them to regurgitate, making the sides sticky.













- Scrape 300 bees into jar, again make sure that the queen is NOT included!
- Add 50 ml (~ 2 oz) of windshield wiper fluid, rubbing alcohol or *soapy* water; shake vigorously for several minutes.
- Remove lid and pour contents through metal wire-mesh screen.
- Repeat several times on same 300 bees.
- Count mites; can also use coffee filter.



www.extension.umn.edu/honeybees







1C. Sugar Shake



Collect 300 bees (100mL beaker=300 bees)



Collect 300 bees by brushing into a measured container





Add the 300 bees into jar with screened lid









Add 2 Tablespoons (25 g or 1 oz) of:

Powdered Sugar (also called Icing sugar)

or

Ground white sugar (or Flour)







Sugar Shake (Cont'd)

Rest for 1 min.

- Shake jar vigorously; shake like a salt shaker.
- Shake sugar containing the mites on a white flat surface.
- Repeat the above two steps until virtually no more sugar shakes out.













Sammataro photo

To visualize mites:

•Shake onto white paper, and spray mites with water

or

•Spray paper towel with water or

•Shake mites into pan of water, mites will float; then strain out and count.







Return bees to colony











- Sample 100 250 cells.
- Locate a patch of drone cells in the purple -eye pupal stage; worker brood should also be examined.
- Slide the prongs of an uncapping fork along the comb face and into the cappings.
- Pry upward and remove the pupae.
- Carefully examine the bodies and the interior of the cells for mites.



Another method:

- Cut cappings with a knife.
- Wash out brood with water from hose through a

strainer.

• Collect mites in fine sieve.





1B. Pulling up Brood





Good method but: we need standard procedures for estimating mite populations









1C. Sticky Boards (or Hive Debris)









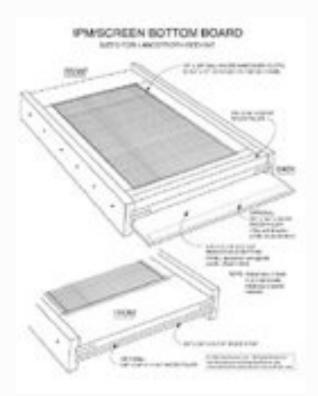




Bottom (Floor) Board Designs















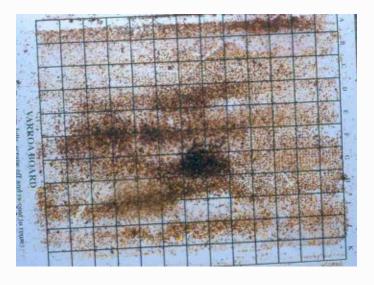






Counting Mites











Record Keeping

Varroa Mite Counts				DATE	
				Yard	
Sticky Boards					
Col. No.	Date IN	Date OUT	Mite numbers	Treatmnt Date	NOTES: (Chalk?, dead bees or pupae?, board chewed?)





- On a 24-h board
 - 12 mites (spring in NW states)
 - 23 mites (fall in NW state)
- In southeastern states
 - 1-12 mites (spring in SE states)
 - 71-224 (fall in SE states)

Obviously, this needs more research









- Time of year
- Colony size
- Hygienic behavior of bees
- Amount of brood
- Amount of drone brood
- ❖ If amount of drone brood is small, mites will invade worker brood
- Only a small percentage of mites could be in drones







- Early detection offers the best opportunity for effective Varroa control.
- Frequency of colony testing:
 - 1. If mite populations low, twice a year.
 - 2. If mite populations high, every two months (excluding winter).











- * Estimate the density of mites (per 100 bees) in individual colonies:
 - ❖ collect ~300 adult bees from a single frame in upper broodbox to estimate colony mite density (sugar shake or ether)
- * Beekeepers can extrapolate colony mite density by
 - Dividing # mites by 3 (mites per 100 bees) and multiplying by a correction factor of 2 (mites in brood).
 - \bullet e.g. 12 mites on 300 bees (12/3=4) x 2= 8% infestation
- * For greater precision examine three- 300-bee sample units

Lee et al. 2010. Practical sampling plans for *Varroa destructor* in *Apis mellifera* colonies and apiaries. J. Econ. Ento. 103(4).





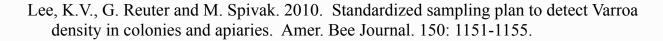
Alternative way to Determine Mite Density (Colony Infestation) in Colonies



If 12 mites were counted on 300 bees, then check that number on the chart:

From chart = 8% total colony infestation level

#Mites per 300 adult bees	Colony infestation	#Mites per 8 300 adult bee samples	Apiary infestation
1	1%	8	1%
2	1%	16	1%
3	2%	24	2%
4	3%	32	3%
5	3%	40	3%
6	4%	48	4%
7	5%	56	5%
8	5%	64	5%
9	6%	72	6%
10	7%	80	7%
11	7%	88	7%
12	8%	96	8%
13	9%	104	9%
14	9%	112	9%
15	10%	120	10%
16	11%	128	11%
17	11%	136	11%
18	12%	144	12%





Sampling Apiaries

- For sampling whole apiaries, examine 300 adult bees from one brood frame in each of 8 randomly-selected colonies/apiary, count mites.
- Check total mite numbers against the chart;









e.g. if you counted 130 mites...

#Mites per 300 adult bees	Colony infestation	#Mites per 8 300 adult bee samples	Apiary infestation
1	1%	8	1%
2	1%	16	1%
3	2%	24	2%
4	3%	32	3%
5	3%	40	3%
6	4%	48	4%
7	5%	56	5%
8	5%	64	5%
9	6%	72	6%
10	7%	80	7%
11	7%	88	7%
12	8%	96	8%
13	9%	104	9%
14	9%	112	9%
15	10%	120	10%
16	11%	128	11%
17	11%	136	11%
18	12%	144	12%

Estimated mite load is 11%







Treatment Thresholds Rolls and Shakes (MN)

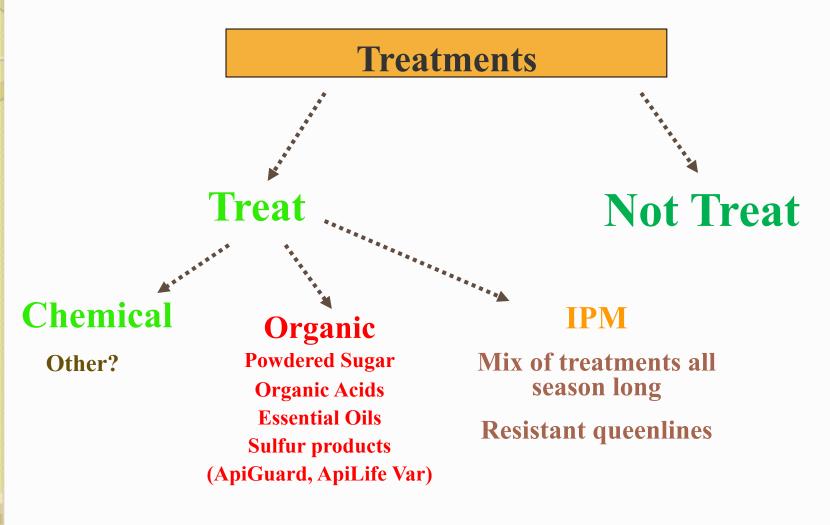
- * Controls will be needed if >10-12% adult bees infested
 - Stationary colonies, not migratory
- ❖ If less than 10%, no treatment
- ❖ If threshold is 10-12%: gray area, could treat
 - *Keep sampling, monitor mite densities until it changes



Lee et al. 2010. Practical sampling plans for *Varroa destructor* in *Apis mellifera* colonies and apiaries. J. Econ. Ento. 103(4).



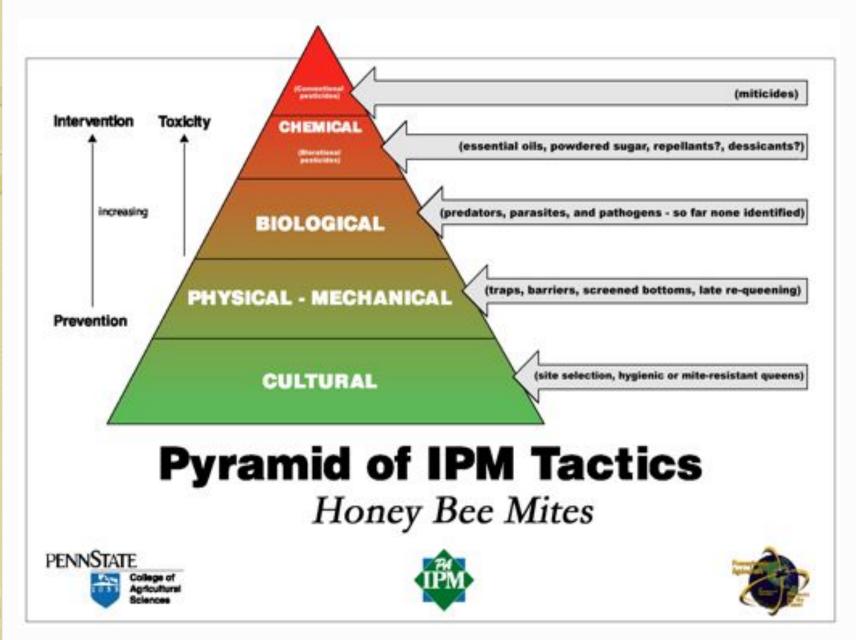
Treatment Options for Varroa Mites















- Varroa Sensitive Hygiene (VHS formerly SMR)
- Russian bees, both mites
- * Other lines?
- Grooming behavior (in conjunction with other traits)
- *AHB?







Product Trade Name	Active Ingredient	Chemical Class
Apiguard	thymol	essential oil
Exomite Apis	thymol	essential oil
Apilife VAR	thymol, eucalyptol, menthol, camphor	essential oils
Apistan **	fluvalinate	synthetic pyrethroid
Apitol	cymiazole	iminophenyl thiazolidine derivative
Apivar **	amitraz	amadine
Bayvarol **	flumethrin	synthetic pyrethroid
Check-Mite+ **	Perizin coumaphos	organophosphate
Folbex	bromopropylate	chlorinated hydrocarbon
Sucrocide	sucrose octanoate	sugar esters
Hivestan	fenpyroximate	pyrazole (alkaloid)
generic	formic acid	organic acid
generic	lactic acid	organic acid
generic	oxalic acid	organic acid

^{**} No Longer Effective in some areas



Compiled from Rosenkranz et al. 2010 and http://www.maf.govt.nz/biosecurity/pests-diseases/ animals/varroa/guidelines/control-of-varroa-guide.pdf.







Biological Treatment

Acaripathogenic fungi:

- Beauveria bassiana
- ❖ Metarhizium anisopliae var. anisopliae
- ❖ Verticillium lecanii
- Hirsutella thompsonii
- Drawbacks:
- take several days to kill their hosts,
- may not be adapted to the brood nest environment

Chandler et al., 2001, Shaw et al., 2002, James et al., 2006, Kanga et al., 2002, Kanga et al., 2006, Meikle et al., 2007 and Meikle et al., 2008.





- 1. Bacterium Serratia marcescens, isolated from A. cerana bees; secretes proteins that act as a biological degrader of chitin.
- 2. New delivery systems for oils: porous ceramics also encapsulating oils in starch
- 3. RNA interference (or RNAi), current research underway: silence particular genes; regulate gene expression
- 4. Attractiveness of brood and the release of brood volatiles
 Varroa attractant and arrestment responses (traps); currently
 being studied





Other Treatments

- Chlorfenvinphos (organophosphate), effective but residues may cause problems
- Azadirachtin (Neem), needs more work, some products can be effective
- Plant-derived monoterpenoids, some report bee toxicity; other essential oils are being tried
- Food Grade Mineral oil, not effective
- Powdered sugar dust but has potential, good for sampling; not for large scale beekeeping
- Screen wire bottom boards; not effective in keeping mite populations low (Ellis et al. 2001)
- Smoke, burning different plant materials; some success but may be harmful to bees
- Thermal treatments, such as heating frames in an 'oven' (Rosenkranz et al., 2010,).
- Cell size modifications, small cell size does not work (Berry et al. 2010,).
- Mite traps or killing with specialized frame, e.g. Mite Zapper (Huang, 2001).
- Propolis barriers
- Antioxidants, currently under investigation





Consequences of Resistance

- Experimentation of chemical cocktails
- Over-application of some chemicals
- Contamination of hive products
 - ❖If selling wax products, get wax tested for residues







- First paper: Ogata, J.N., Bevenue, A. 1973. Chlorinated pesticide residues in honey. Bulletin of Environmental Contamination and Toxicology; 9 (3): 143-147.
- □ Varroa-related: Laub, E., Metzler, B., Putz, A., Roth, M. 1987. Outstanding situation of permitted insecticides against Varroatosis in honey. Lebensmittelchemie und Gerichtliche Chemie. Volume 41: 107-109
- Over 400 journal entries on residues in bee colonies
- Latest findings: 170 compounds found in hive products (Mullin CA, Frazier M, Frazier JL, Ashcraft S, Simonds R, et al. (2010) High Levels of Miticides and Agrochemicals in North American Apiaries: Implications for Honey Bee Health. PLoS ONE 5(3): e9754. doi:10.1371/journal.pone.0009754







- Learn to recognize other mites
- New mite on the horizon:
 - Troplilaelaps sp.







Tropilaelaps spp.

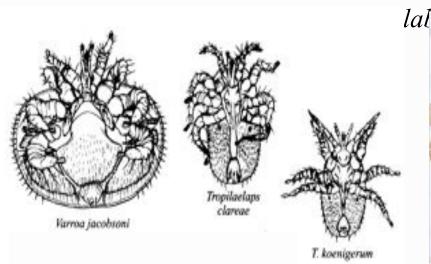
Currently (2010-2011), USDA is conducting a survey for this mite and Asian bees; importation of Australian packages bees now prohibited.

1.T. clareae (Delfinado & Baker) found on A. dorsata breviligula AND A. mellifera in the Philippines

2.T. koenigerum ex A. dorsata, A. laboriosa and A. mellifera

3.T. mercedesae ex A. d. dorsata













Life Cycle

- ❖ Similar to *Varroa* but brood is essential for survival, will not feed on adults
- Very serious in tropics, not so much in temperate climates (yet)
- ❖ Larval form is mobile, feeds actively
- ❖ Can carry virus DWV













Parasitic Bee Mites and Apis Hosts 2011

	MITE SPECIES									
BEE HOST	Varroa destructor	V. jacobsoni	V. underwoodi	V. rindereri	Euvarroa sinhai	E. wongsirii	Troplilaelaps clareae	T. koengerum	T. mercedesae n.sp.	T. thaii n.sp.
Apis florea			X Nepal, S. Korea		X					
A. andreniformis						X				
A. cerana	X	X	X							
A. koschenikovi				X Sumatra						
A. nuluensis, Borneo		X	X ?							
A. nigrocincta, Sulawasi		X ?	X							
A. dorsata dorsata, Asia, Indonesia, Palawan	## Korea							X Sri Lanka	X Palawan and Sri Lanka	
A. d. breviligula							X Philippines (not Palawan)			
A. d. binghami Sulawesi									X **	
A. laborisoa Nepal								X	X Vietnam	X Vietnam
A. mellifera	X Japan & Korean haplotypes	X, Papua N.G, Irian jaya	##				X Philippines		X	
A. m. scutellata in Africa	X		** 0 1							

X —Positive identification,

** — Currently unresolved;

— Incidental visitor.

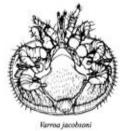
Compiled by D. Sammataro and D.L. Anderson; (from Anderson and Morgan 2007 and Navajas et al. 2010).





Parasitic Mites on Honey Bees

Family: Varroidae (Delfinado & Baker, 1974)



Genus: Varroa



Varroa jacobosoni Oudemans 1904 (ex A. cerana, A. nuluensis)



V. underwoodi (ex A. cerana, A. nigrocincta, A. florea)



V. rindereri (ex A. koschevnikovi)



V. destructor Anderson & Trueman 2000 (ex A. cerana, A. mellifera, A. m. scutellata, A. dorsata dorsata)

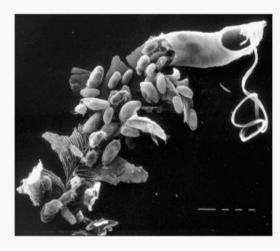




Size Comparisons of Bee Mites with a Flea

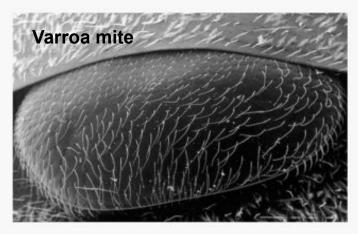
All photos are approx. the same scale





Tracheal Mites



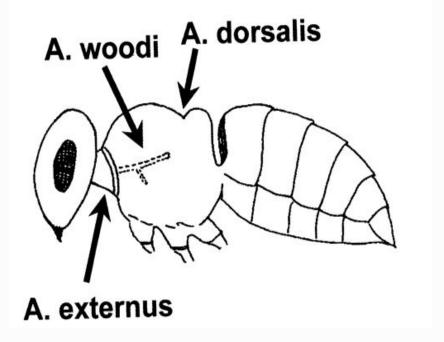


Photos by W. Styer Ohio State, Wooster





- Three mites in this Genus
 - A. dorsalis
 - A. externus
 - A. woodi





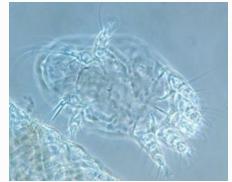


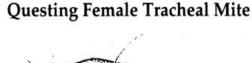




Tracheal Mite: female

- Females move off old host to quest and migrate onto new younger bee
 - Can distinguish young bees from older bees
 - Enters tube and after one to two days, lays about 1 egg/day for 12 days







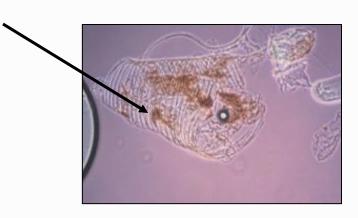
Drawing © by D. Sammataro 1997







- Damage to Tracheal Tube
 - Blotching and scabs in tube
 - Debris inside
 - Puncture wounds



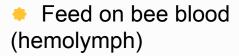


- Lowers ability to use wing muscles
 - Cannot keep cluster warm in winter
 - Bees die over winter, crawling out of colonies

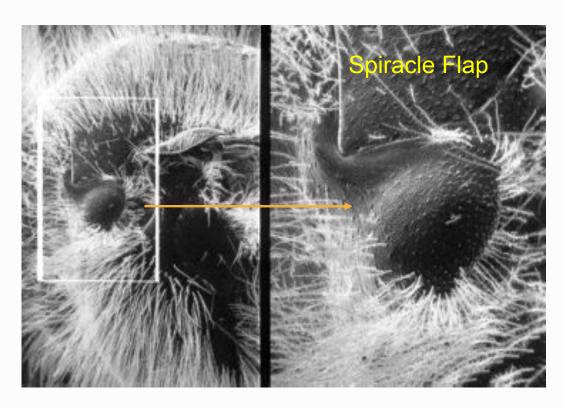




Tracheal Mites



- Attack all adults bees
- Live in the respiratory system, enter through the spiracle flap on thorax

















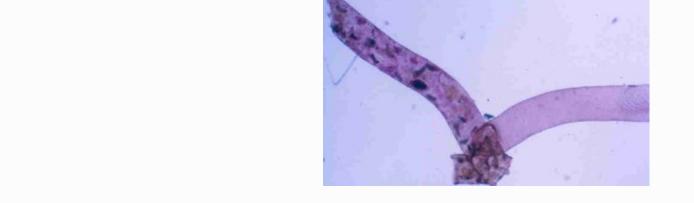
MITE DISSECTION VIDEO: http://www.ars.usda.gov/pandp/docs.htm? docid=14370)



Tracheal Mite ID



- Collar Removed
 - Main tracheal tube exposed
 - Look at spiracle opening
 - If infestation light, female is here
 - If no shadows, no mites







Future Work: What we need

- 1. Uniform estimation for mite densities
- 2. Frequency of testing guidelines
- 3. Treatment thresholds
- 4. New treatments that don't contaminate hive products





