The Role of Pesticides in Honeybee Decline

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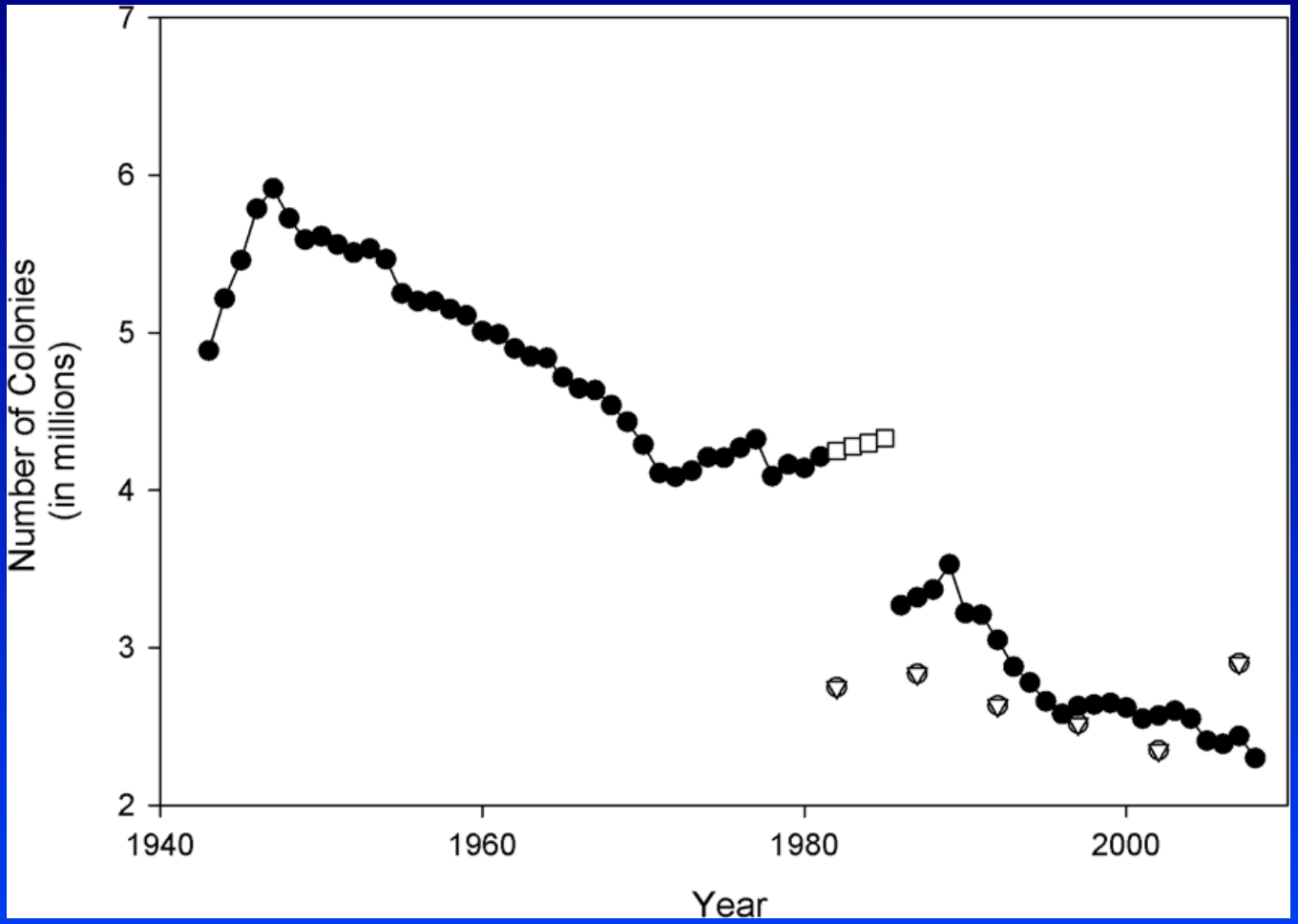
Economic Value of Pollinators

Crop Category	Worldwide Value ($ Billions)	Pollinator Value Percentage
Nuts	20	31
Fruits	307	23
Edible Oil Crops	336	16
Vegetables	585	12
Stimulant Crops	27	39

Source: Gallai et al. in Ecological Economics (2009)

The Number of Honey Producing Colonies is Decreasing



Source: vanEngelsdorp and Meixner in Journal of Invertabrtate Pathology (2010)

What are Possible Causes of Decline in Honey Bees?

- Migratory Stress
- Poor Nutrition
- New Diseases
- Varroa Mites
- Colony Collapse Disorder
- Pesticides

Pesticide Questions

- What is the toxicology of pesticide exposure?
  - Acute vs. Chronic (sub-lethal) Effects
  - Synergistic Effects
- What pesticides are honey bees exposed to?
- How does the exposure occur?
- How much of the various pesticides are they exposed to?
- How does the exposure change with time and location?
- Can pesticide exposure be correlated with hive health?

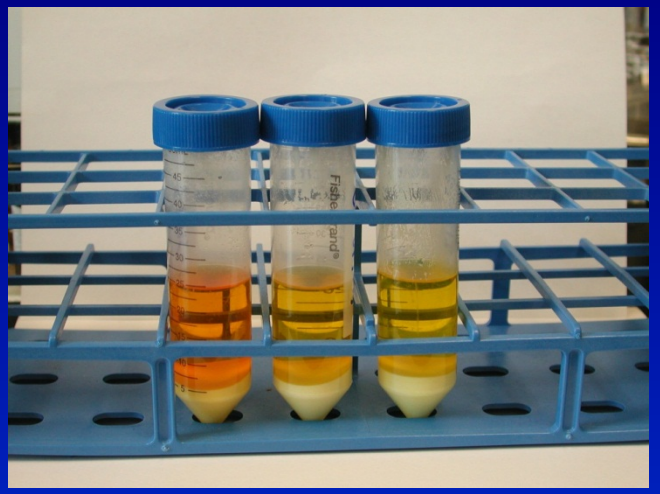
Analytical Procedures for Pesticide Analysis

Extract with a Modifed QuEChERS Procedure

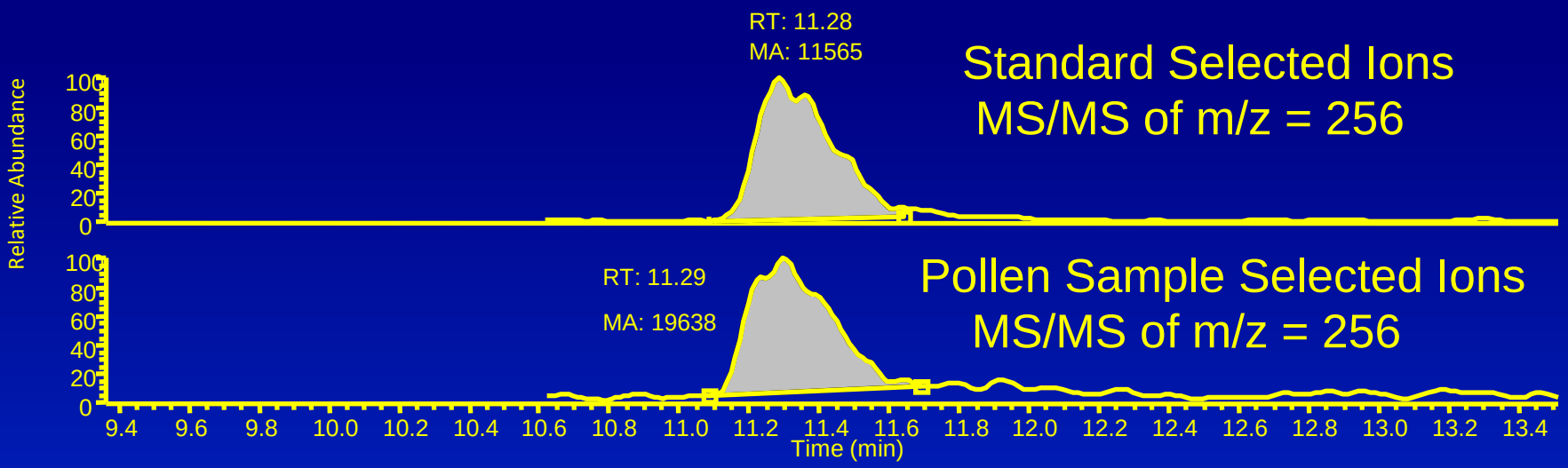
- 5 g of pollen/bees plus:
  - 6 g magnesium sulfate
  - 1.5 g sodium acetate
  - 15 mL water
  - 15 ml acetonitrile
  - C-13 Alachor and D-4 Imidacloprod I.S.
- Shake / Vortex
- Centrifuge

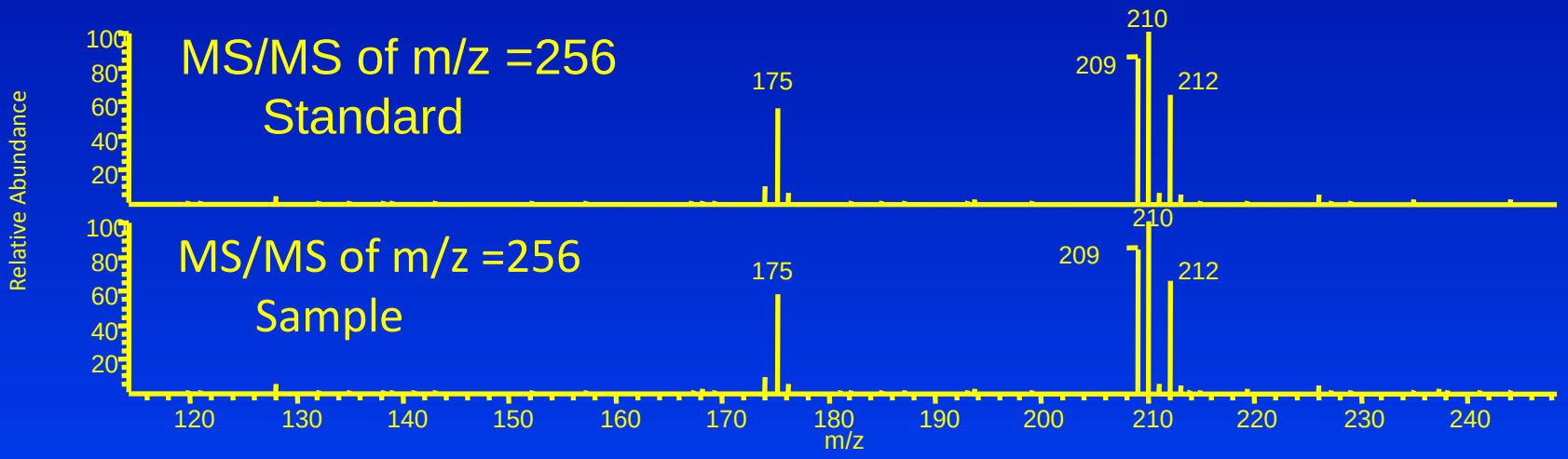
Extraction (cont.)

- Take 10 mL aliquot
- Combine with
  - 1.5 g magnesium sulfate
  - 0.5 g PSA
  - 0.5 g C-18 silica
  - 2 mL toluene
- Shake
- Centrifuge
- Concentrate 6 mL to1 mL with nitrogen for LC/MS analysis

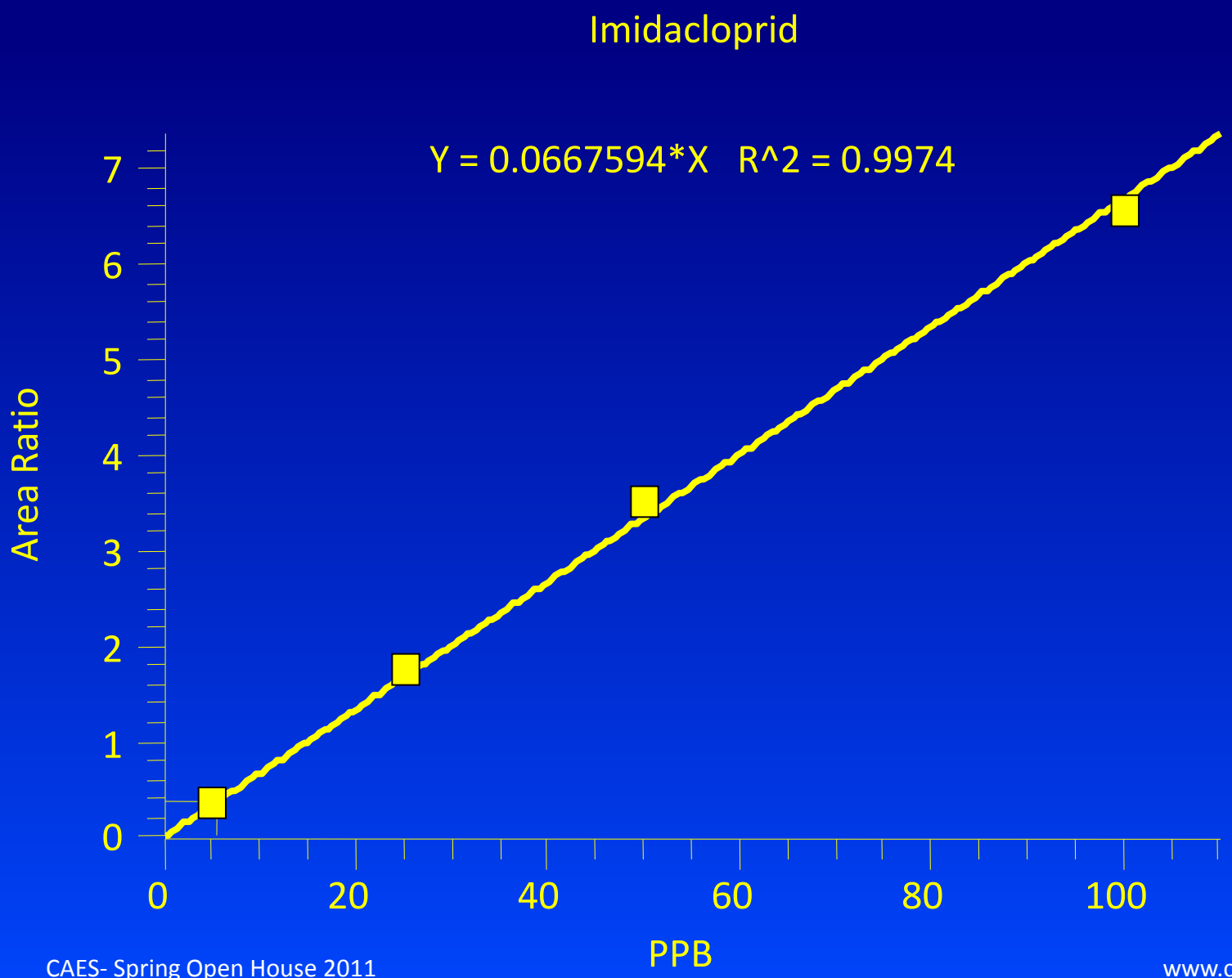


Detection of Imidacloprid at 10 PPB in Sample





Internal Standard Response Curve (no matrix present)



CAES Studies

- Suspected Honey Bee Poisoning
- Measurement of Pesticide Residues in Squash Nectar and Pollen after Agricultural Application
- Monitoring of Pollen Collected by Foraging Honey Bees
  - Coordinated Agricultural Program: Stationary Apiary Project
  - Connecticut Study

Study of an Acute Poisoning

- A honey bee researcher at Purdue University noted sick and dying bees in apiaries near recently planted cornfields during dry and windy spring.
- Young bees dying not foragers.
- As most of the corn seed in the area is treated with a pesticide toxic to bees a poisoning incident was suspected.
- Samples of honey bees and honey bee collected pollen analyzed.

Pesticides Found in Parts Per Billion

Pesticide	Pollen 84	Bee 1-P	Bee 104-P	Bee 84-P	Bee 91-P	Bee 95-P	Bee 71-C
Clothianidin	21	4.4	7.6	5.0	3.4	3.5	n.d.
Thiamethoxam	20	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Imidacloprid	2.8	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Atrazine	310	11	11	13	24	12	2.8

LD 50's for Honey Bees

- Clothianadin .04 ug/bee
- Thiamethoxam .03 ug/bee
- Imidacloprid .018 ug/bee
- Atraazine > 1000ug/bee

Follow Up

- Collect pollen from hives surrounding fields being planted with either treated or untreated seed.
- Honey bee pollen from hives around field with treated seed had 0 – 88 PPB clothianadin while those around the untreated field had only 0 -13 PPB.
- Study will continue this spring with funding from the North American Pollinator Protection Campaign.

Measuring Neonicotinoid Residues in Squash Nectar and Pollen

When we apply systemic insecticides to soil or through irrigation at labeled rates, as a farmer would, how much do we find in the nectar and pollen of the plant?

Squash bees on pumpkin flower- Liz Andrews, UMass

What we did:

- Grew squash (summer squash in 2009, summer squash and winter squash in 2010) according to standard farming methods
- Applied neonicotinoid insecticides imidacloprid (Admire Pro®) and thiamethoxam (Platinum®)
- Two methods of application:
  - To seed hole in black plastic just before seeding
  - In drip irrigation to transplants 4-5 days after transplant
- Rates:
  - Admire Pro®: 10 fl. oz. per acre (labeled range = 7 -10.5)
  - Platinum®: 8 fl. oz. per acre (labeled range = 5- 11)

Collecting pollen and nectar

What we found:

Insecticide	Average Concentration in Pollen (Overall)	Average Concentration in Nectar (Overall)
Imidacloprid	14 ppb ± 8	10 ppb ± 3
Thiamethoxam	12 ppb ± 9	11 ppb ± 6
Control	None	None

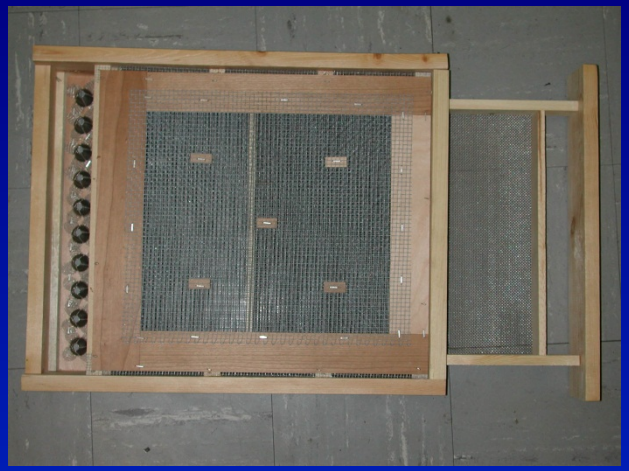
Use Pollen Collected by Honey Bees to Evaluate Exposure to Pesticides

Photo by: Kathy Keatley Garvey, UC Davis

Connecticut Study
Locations of Hives 2007 - 2010

1. Our offices in New Haven on the edge of the city, 2007 - 2010
2. Our experimental farm surrounded mostly by suburbs, 2007 - 2010
3. An orchard on the edge of a suburb with pollen collected only during the blooming season of apples and blueberries, 2007 and 2009
4. Another suburban site on the edge of a large agricultural area growing vegetable crops, 2007 – 2010
5. Mixed agricultural and industrial area, 2009 - 2010

Bee Pollen Collection



Overall Data by Year

Year	2007	2008	2009	2010
Fungicides	11	10	14	12
Herbicides	10	10	10	8
Insecticides	15	9	17	18
Avg. # per sample	4.3	5.4	5.8	4.0

Frequently Detected Pesticides

Pesticide	2007 n = 101	2008 n = 44	2009 n = 59	2010 n = 62
Coumaphos (A, I)	96	44	41	36
Carbaryl (I)	66	16	9	38
Phosmet (I)	38	15	30	20
Atrazine (H)	34	24	31	21
Imidacloprid (I)	30	23	8	3
Dithiopyr (H)	13	34	35	7
Pendamethalin (H)	11	24	30	19
Carbendazim (F)	18	20	24	22

Detection Frequency Depends on Hive Location

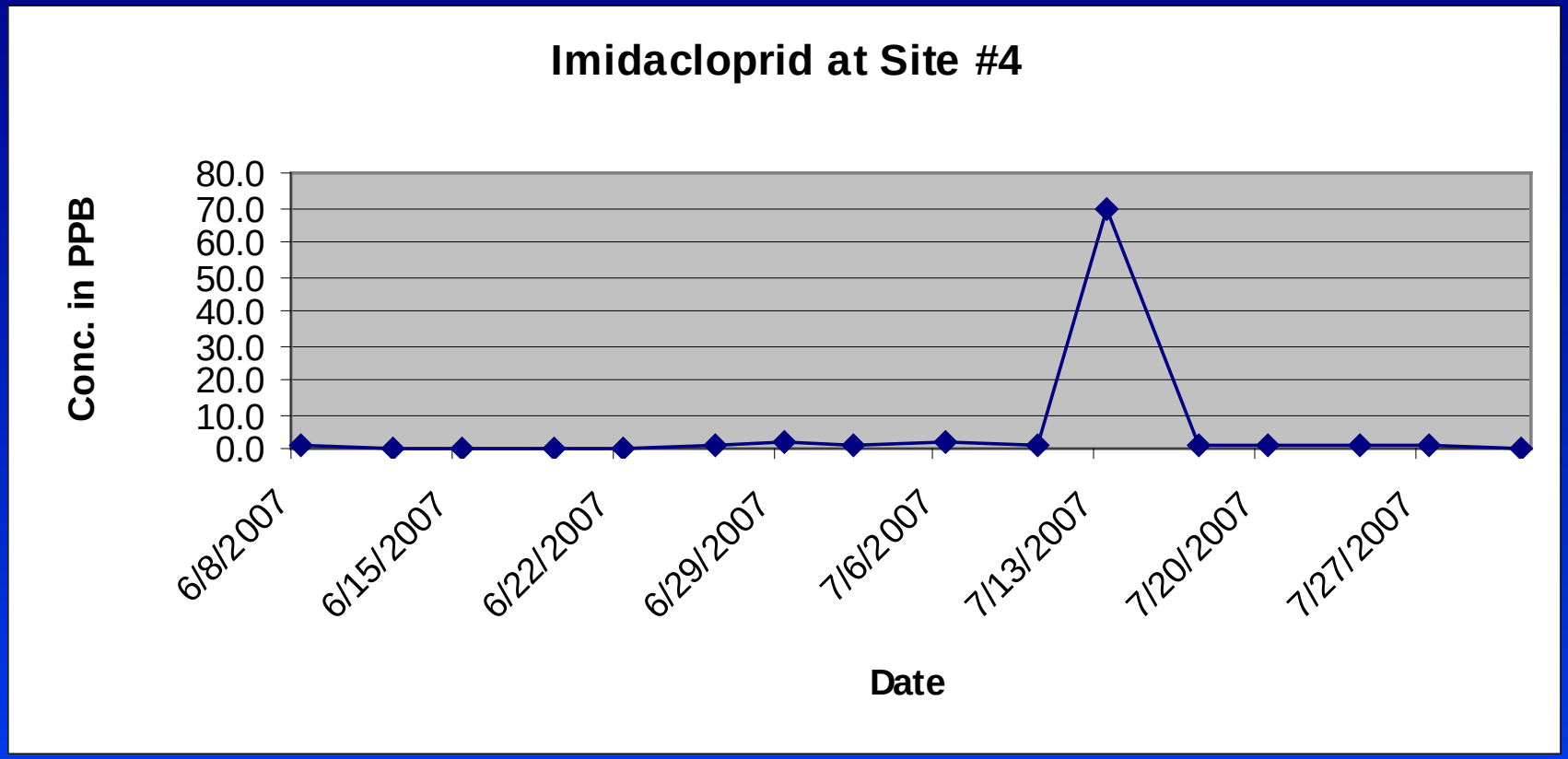
- In 2009 the orchard location averaged 12.0 residues per sample; the average of the other four sites was 5.4 residues per sample.
- In 2009 all 4 samples from the orchard location had difenconazole and 3 of 4 had myclobutanil; neither of these pesticides were seen at any of the four other hive locations that year.

High Frequency is not the same as **High Concentration** (2009 Data)

Pesticide	# Detec. (n = 59)	Max. Conc. PPB	Avg. Conc. PPB	Median Conc. PPB
Atrazine (H)	32	15	1.5	1.0
Phosmet (I)	30	540	44	7
Imidacloprid (I)	8	19	5.7	4.7
Azoxystrobin (F)	8	55	18.3	8.8
Myclobutanil (F)	3	4190	1490	270
Trifloxystrobin (F)	5	160	42	22

A = acaricide, F = fungicide, H = herbicide, I = insecticide

Pesticide Concentrations Can Change Rapidly



Hive Location

Sorted Pollen – 7/13/2007

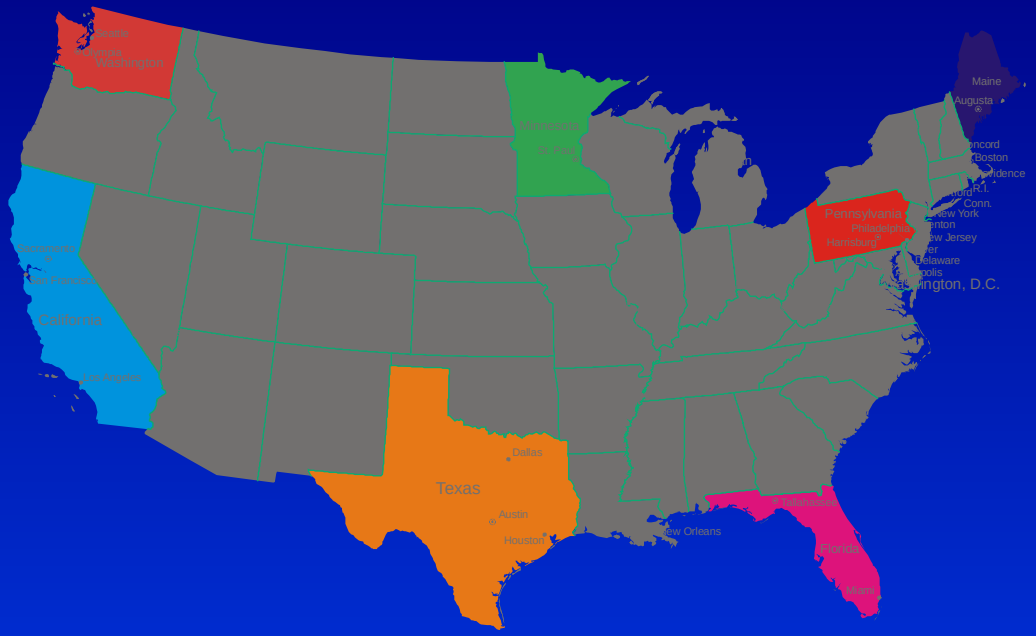
Selected Pesticides in Sorted Pollen

Pollen Color	Pollen Type(s)	Imid	Carb	Carben	Meth	Atr	Phos
Dark Brown	white clover	**611**	n.d.	n.d.	n.d.	3	**34**
Reddish-Brown	alfalfa	**199**	n.d.	5.8	n.d.	2.1	6.3
Yellowish-Brown	black locust	**114**	n.d.	6.4	n.d.	3.8	6.9
Orange	ragweed	21	15	8.7	n.d.	**38**	18
Orange-Yellow		6.6	n.d.	5.5	8.4	13	15
Medium Brown		3.9	182	n.d.	n.d.	16	**55**
Greenish-Yellow		3.1	23	n.d.	17	14	10
Round		2.4	n.d.	n.d.	17	8.6	3.1
Pure-Yellow		1.7	13	2.4	n.d.	6.5	4.1

CAP Stationary Apiaries 7 State Project

2009 (April 09): FL, ME, MN, PA, TX, WA

2010 (April 10): CA, ME



Objective:

Identify causal factors or interactive effects of these factors (pests, pathogens and pesticides) in causing losses in stationary honey bee colonies across the United States.

Standardized Data Collection

I. weather conditions
II. landscape composition
III. pesticide contamination (pollen, wax)
IV. package source and queen genetic makeup
V. colony productivity and survival
- frames of adult bees and sealed brood
- egg laying and brood pattern quality
- queen status (presence / absence)
- supercedure

VI. infestation
1. *Varroa* mites – mites per 280 adult bees
2. SHB – adults and larvae
3. Tracheal mite dissections
4. *Nosema* (spp. ID and spore counts)
5. chalk brood symptoms

MN, August 2009

6. bacterial pathogens
7. viral symptoms and molecular markers: DWV, IAPV, SBV, BQCV

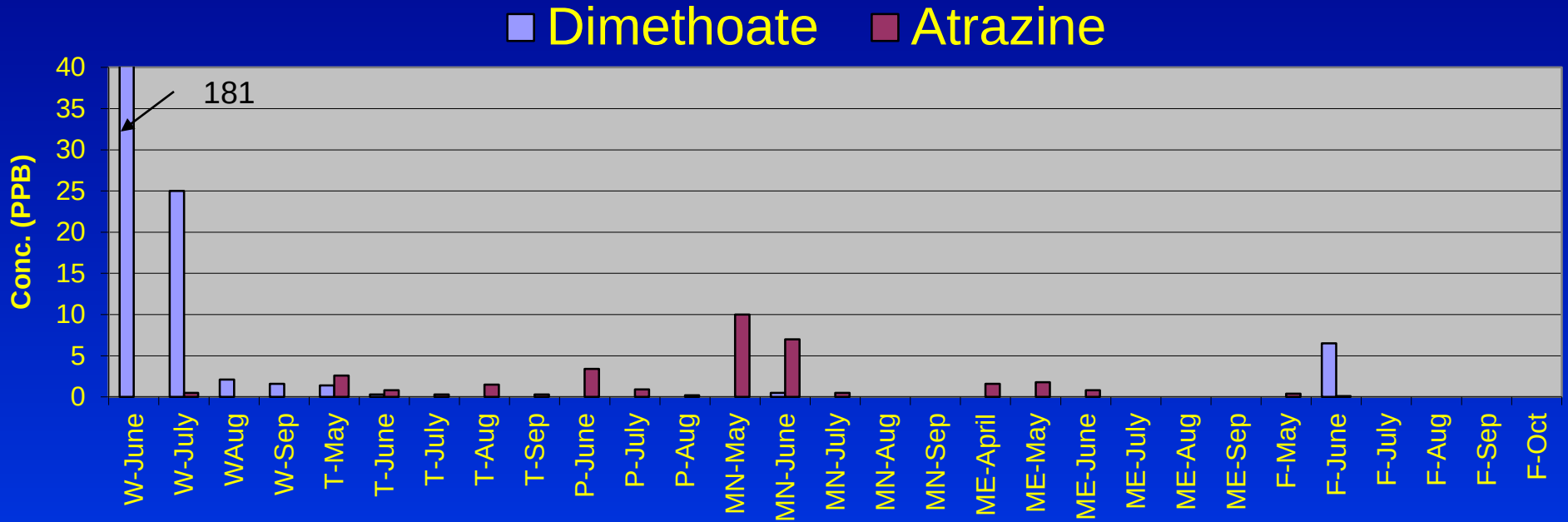
CAP Stationary Apiaries, 2009

MN, August 2009

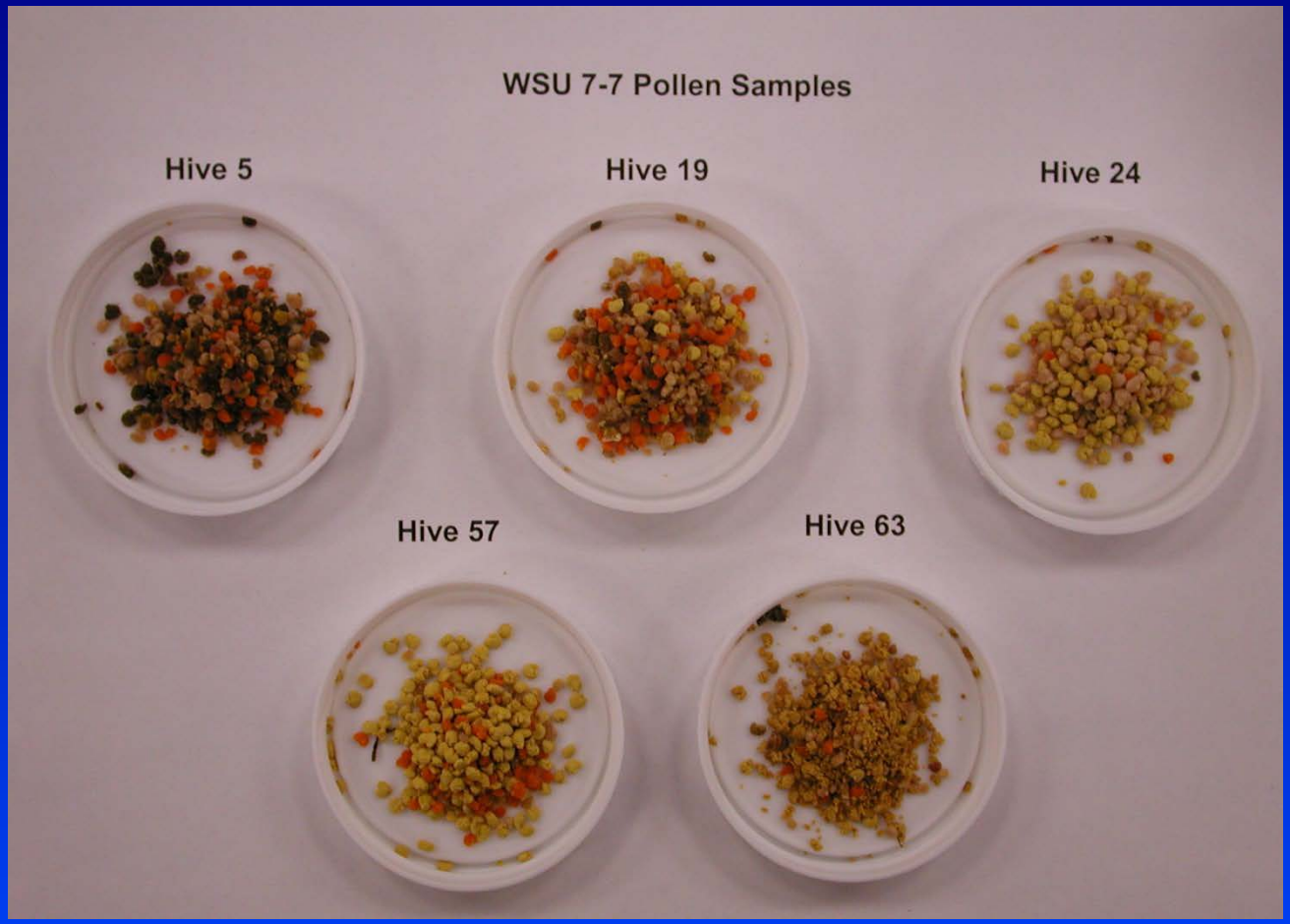
Taking Samples

TX, November 2009

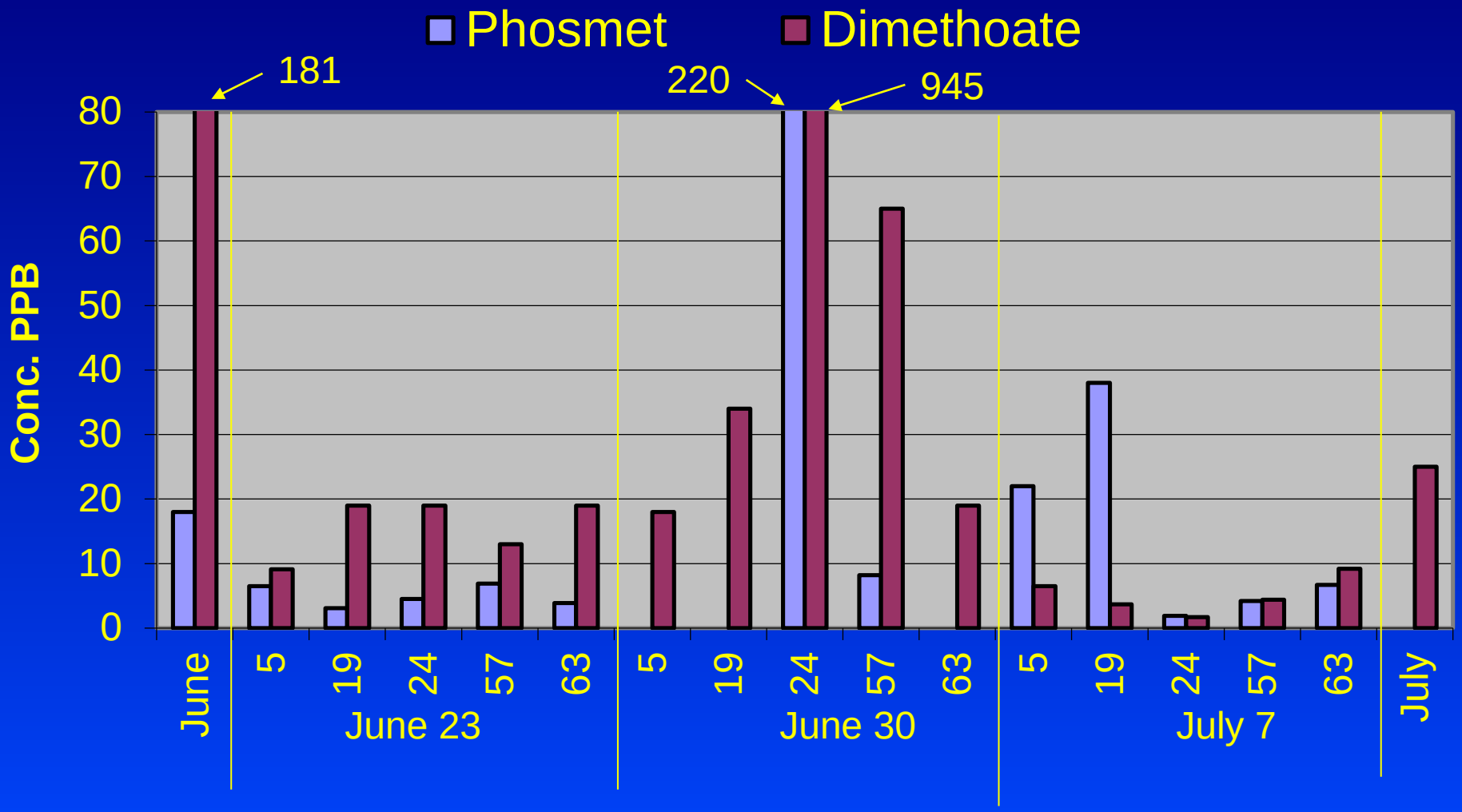
Pesticides Vary with Sample Location and Time (2009 Data)



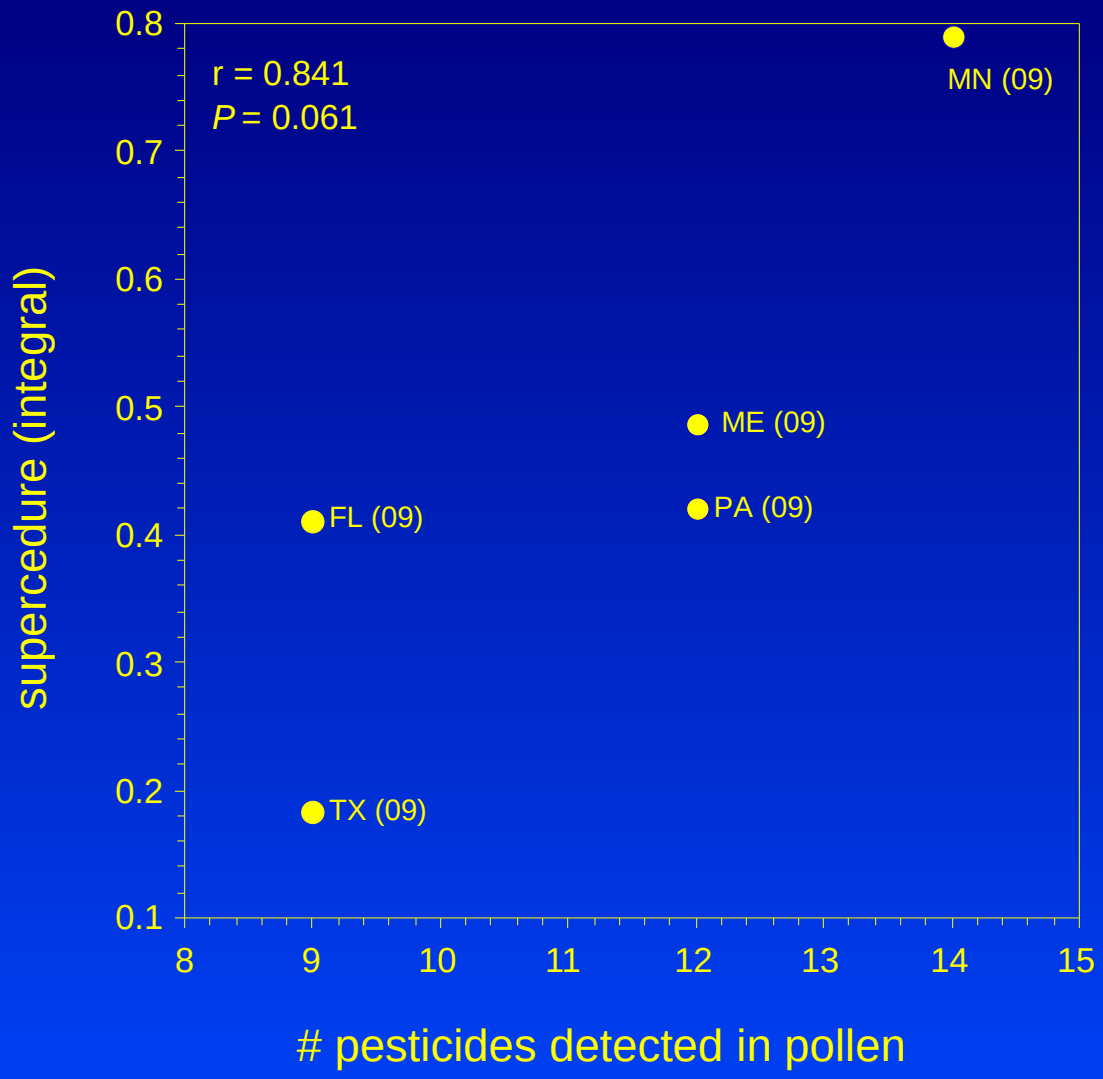
Pollen Samples Differ by Hive Even at Same Location and Date



Compositing vs. Individual Samples



Number of pesticides in pollen loads (2009)



Conclusions

- Methods used can detect low concentrations of pesticides in pollen
- Pollen samples are heterogeneous, causing an increase in variability of data
- Honey bees are exposed to pesticides while foraging for pollen
- Pesticide concentrations vary with hive, time, and location
- Pesticides are a contributing factor to the problems faced by honey bees

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Managed Pollinator CAP
Coordinated Agricultural Project

A National Research and Extension Initiative to Reverse Pollinator Decline

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