

# Ambient Orchard Volatiles for Navel Orangeworm (NOW) Analyses

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The navel orangeworm (NOW) continues to be a major insect pest of California tree nuts. Current efforts to monitor NOW utilize virgin female NOW traps or almond meal, both of which are beneficial but have proven either unavailable commercially or inadequate, respectively. The pheromone blend, while a promising attractant, is hampered by stability problems in the field. Ambient emissions collected from 2009 almond orchards has provided a prototypical simple synthetic blend, based on relative ratios of volatiles, that has elicited promising electroantennogram responses and notable volatile trends. Data collected in 2010 include: ambient volatiles and associated SPME snapshots; pictures of kernel phenological development; and fatty acid content at collected intervals.

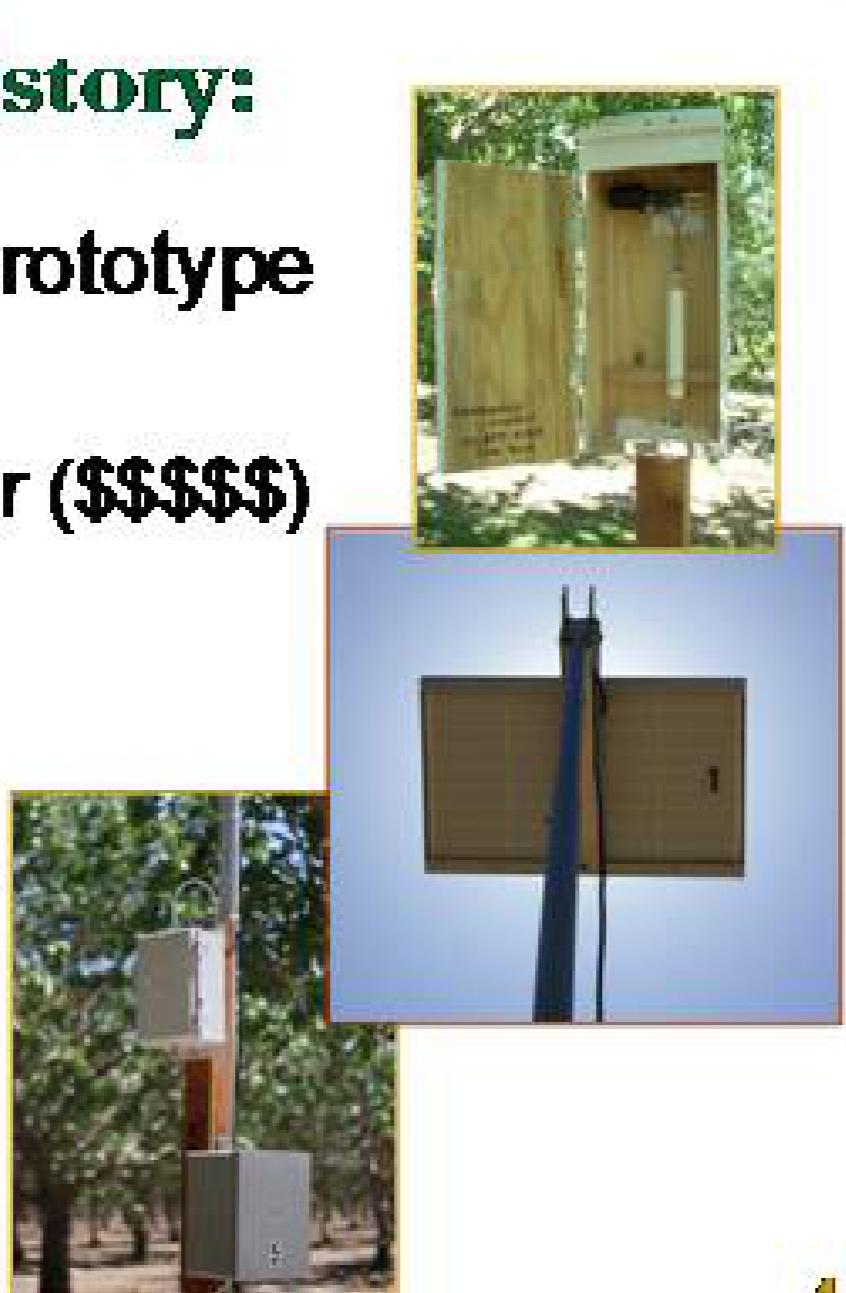
## Problem:

- "Control of NOW continues to be a top priority of the tree nut industry..."
- California Almonds
  - 80% world's needs
  - 100% U.S. needs
  - Approximately \$2 billion
- Navel Orangeworm
  - Direct feeding damage
  - Vector for toxicogenic aspergilli
  - Millions of \$ annually lost
  - Eluded effective control



## Ambient Volatiles History:

- 2008 – Developed 1<sup>st</sup> prototype
  - Proof of concept
  - Venturi & compressed air (\$\$\$\$\$)
- 2009 – 2<sup>nd</sup> Generation
  - Independent
  - Higher resolution
- 2010 – 3<sup>rd</sup> Generation
  - Sturdier cartridges
  - Diurnal capabilities



## 2009 Almond Orchard Volatiles:

#	Ambient Almond Volatile Amounts (ng/m <sup>3</sup> )			
	Collection 1	Collection 2	Collection 3	Collection 4
1	26.8	49.2	31.1	23.3
2	0.0	0.0	0.0	7.0
3	3.0	5.5	0.0	0.5
4	12.2	13.1	11.4	13.1
5	3.7	0.6	0.0	0.0
6	1.8	4.8	4.5	6.9
7	78.1	108.2	49.6	50.1
8	237.4	337.4	161.2	169.4
9	11.1	3.9	13.9	11.2
10	3.8	0.0	18.1	21.7
11	306.8	165.3	306.6	1971.5
12	3.9	1.7	3.5	10.0
13	3.4	31.1	10.8	0.2
14	7.7	9.3	14.8	7.0
15	12.6	0.0	5.3	3.2
16	11.9	10.4	19.0	25.9
17	151.5	224.8	265.8	355.4
18	51.9	79.7	23.3	31.8
19	5.4	7.2	5.3	9.8
20	4.3	9.6	13.9	13.5
21	0.0	0.0	1.4	3.0
22	122.7	191.7	76.7	77.5
23	2.0	1.0	4.8	0.0
24	74.7	83.7	74.4	87.8
25	3.5	0.0	0.0	11.3
Degree Days (days)	482-587	1586-1732	1752-1930	2592-2817
Collection Dates (days)	4/23-5/12	6/30-7/7 (7)	7/7-7/15 (8)	8/11-8/21 (10)

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## Flight Tunnel Analysis of Blend V:

Treatment Pairs*					
Experiment #1		Experiment #2		Experiment #3	
Almond meal	Control	Blend V	Control	Almond meal	Blend V
Male Captured	1.5 ± 0.8	0.2 ± 0.2	0.1 ± 0.1	0.3 ± 0.3	n/a
Female Captured	3.3 ± 1.4*	0.8 ± 0.5*	5.0 ± 0.6***	0.9 ± 0.3**	6.1 ± 0.5
N=9					5.9 ± 0.4
Eggs Deposited	30.5 ± 10.1**	12.8 ± 6.4**	52.9 ± 7.1***	1.2 ± 0.9**	51.5 ± 6.4***
N=20					74.5 ± 6.2***

\*Experimental conditions: 60 mg almond meal, gray septa impregnated with 100 µl of acetone and allowed to evaporate for control; or gray septa impregnated with 5 mg Blend V in 100 µl of acetone, which was allowed to evaporate prior to analysis. Mean values are paired. Treatments were significantly different by paired t Test: \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001.

<sup>a</sup>Males not released

- Female NOW showed significant ovipositional preference for Blend V relative to Almond Meal

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## 2010 Data Undergoing Analyses:

- Volatile emissions similar to 2009
  - Increased # of collections providing more accurate emission trends (11 vs. 4)
  - Trends providing "dynamic blends"
- Application of GC-FID for better quantitation of volatiles
  - Allowed for enhanced analysis of emission trends
  - Allowed for more blend formulations
  - GC-MS still used for identification



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## Semiochemicals For Monitoring & Control:

- Almond meal for females (egg traps)
- Baited virgin female traps (for males)
- Female sex pheromone
  - Mating disruption



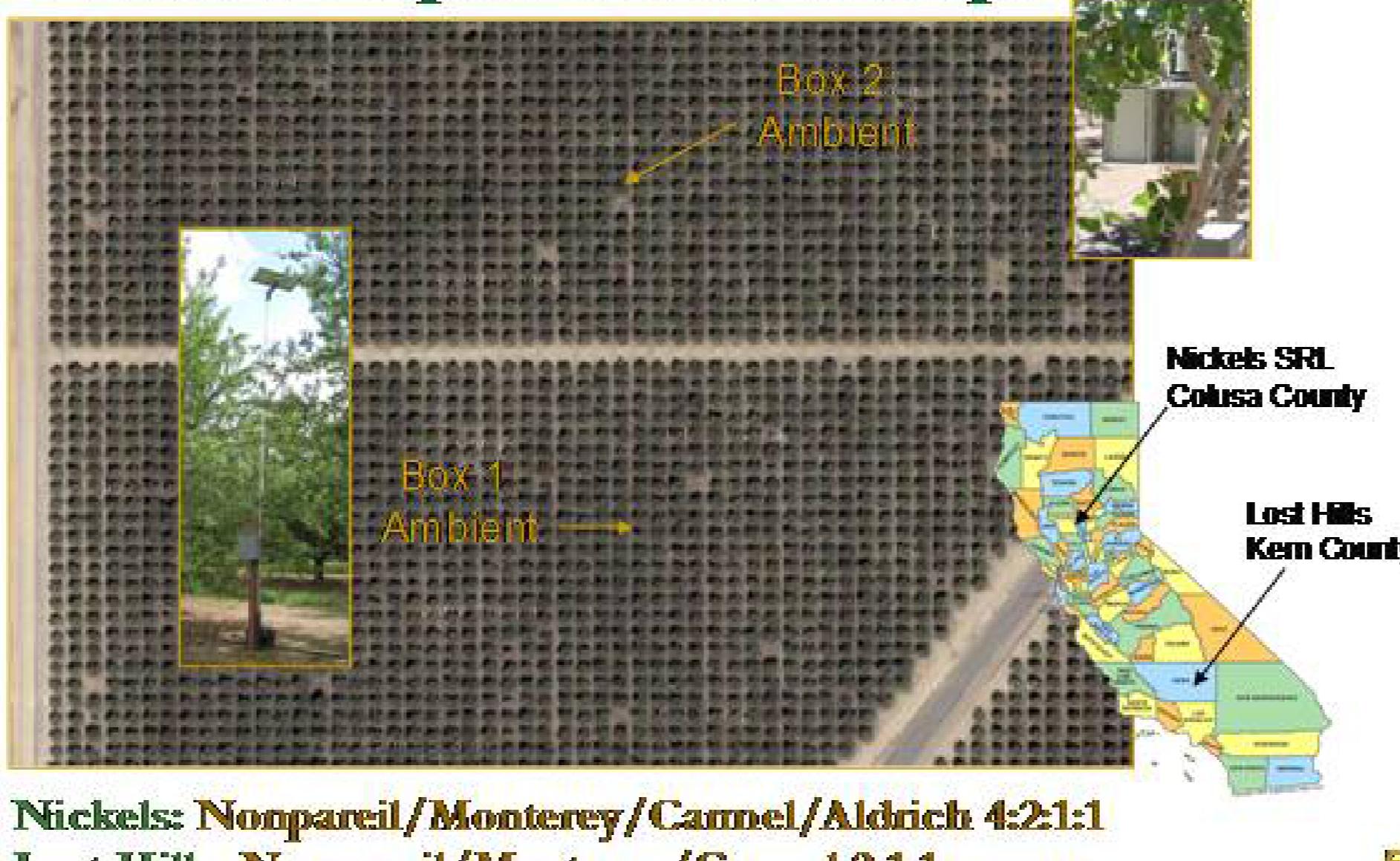
## Objective:

- To develop a non-pheromonal female NOW attractant blend/lure that is environmentally friendly, safe, and stable to harsh orchard conditions



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## LSAVC Experimental Setup:



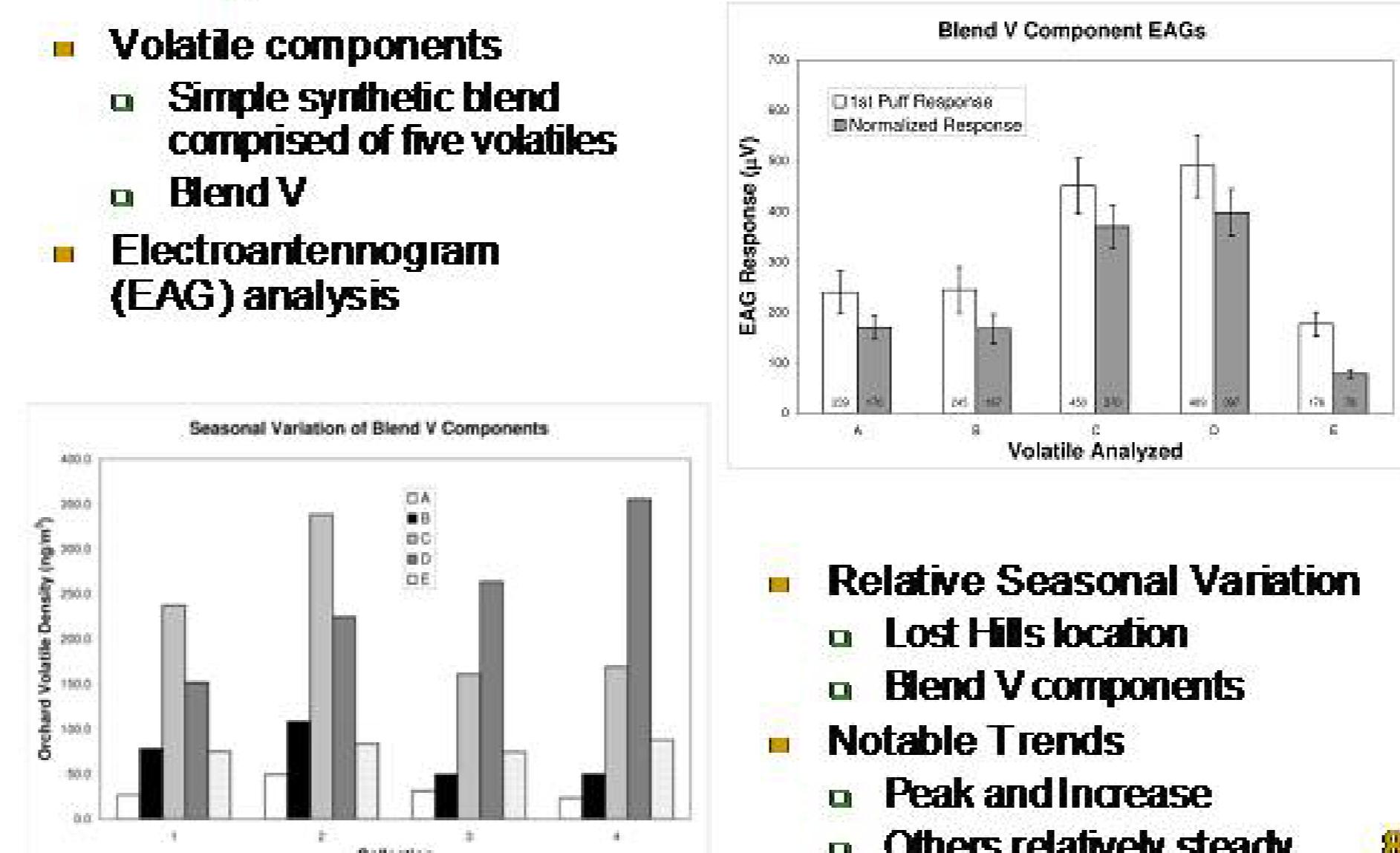
## 2009 Almond Orchard Volatile Results:

### Four Collections

- Duplicated in orchards with two boxes
- Lost Hills Collections (Kern County)
  - April 23<sup>rd</sup> – May 5<sup>th</sup> (12) 482-587 dd
  - June 30<sup>th</sup> – July 7<sup>th</sup> (7) 1586-1762 dd
  - July 7<sup>th</sup> – July 15<sup>th</sup> (8) 1752-1930 dd
  - August 11<sup>th</sup> – August 21<sup>st</sup> (10) 2592-2817 dd
- Nickels Collections (Colusa County)
  - May 12<sup>th</sup> – May 18<sup>th</sup> (6) 594-719 dd
  - June 22<sup>nd</sup> – July 2<sup>nd</sup> (10) 1242-1470 dd
  - July 17<sup>th</sup> – July 23<sup>rd</sup> (6) 1768-1908 dd
  - July 27<sup>th</sup> – July 30<sup>th</sup> (3) 1993-2057 dd

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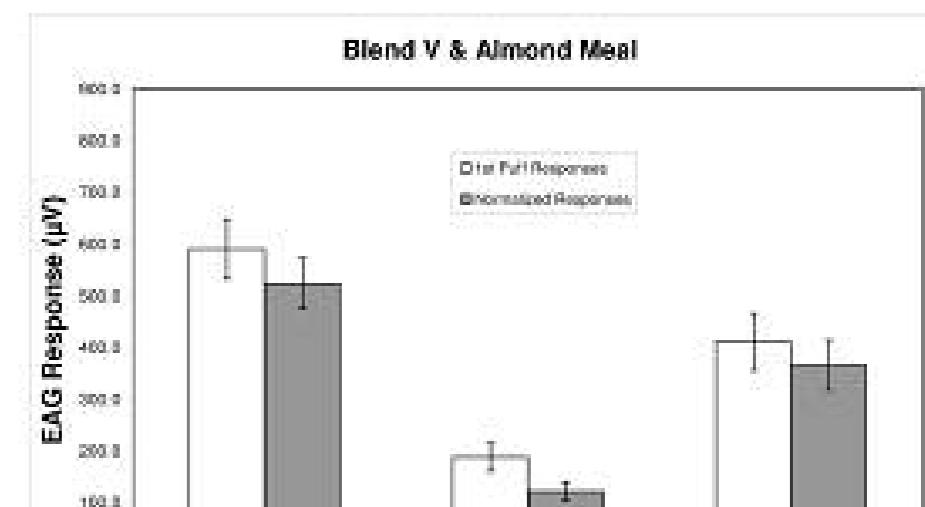
## Analysis of a 2009 Volatile Blend:



- Relative Seasonal Variation
  - Lost Hills location
  - Blend V components
- Notable Trends
  - Peak and increase
  - Others relatively steady

## EAGs of 2009 Blend V:

- Blend V & Almond Meal
  - Blend V elicits greater EAG response than almond meal
  - Blend V + Almond meal approximately equal



- Orchard Bouquet
  - Two collections evaluated
  - Nearly equal
  - Greater than synthetic blend

## 2010 Ambient Collections:

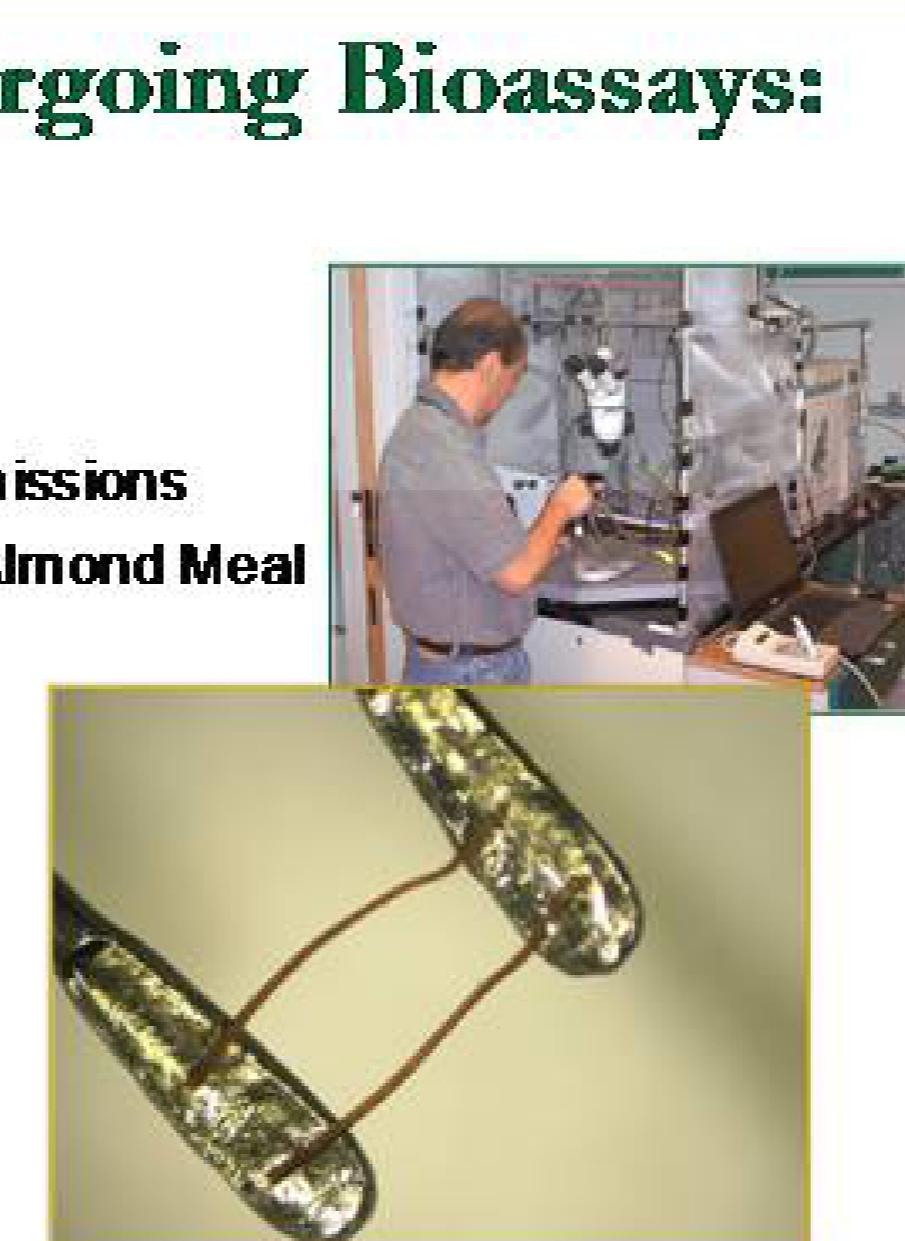
- Increased number of collections for higher resolution of volatiles and possible relation to phenology
- SPME analyses performed for corroboration and comparison (snapshot of tree emissions)
- Physical nut size at time of collections recorded
- Kernel fatty acid content will be determined
- Performed in pistachio orchards for comparison of crop emissions



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## 2010 Volatiles Undergoing Bioassays:

- EAG Analysis
  - Several blends
  - Dynamic vs. static orchard emissions
  - Comparison to BLND.V and Almond Meal
- Flight Tunnel
  - To corroborate EAG results
- Field Studies
  - Top blends for field trapping studies in 2011
- Mix with other attractants



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