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## Management of Phosphine Resistance in Storage Insect Pests in Almond Storages and Processing Facilities in California

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### Introduction

- Fumigation with phosphine gas (PH<sub>3</sub>) has been the most, common practical and efficient way to control stored grain pests world-wide since 1930.
- There is a great challenge in controlling the pests due to resistance to PH<sub>3</sub> found in many countries worldwide, including Australia, Brazil, India, Bangladesh, China and the US.

### Introduction

Percent change in PH<sub>3</sub> resistance over time (USA)

Year	<i>R. dominica</i> (Lesser Grain Borer)	<i>T. castaneum</i> (Red Flour Beetle)
1990	67	13
2012	100	89

Zettler and Cuperus, 1990

Opit et al. 2012

### Introduction

- The Central Valley of California produces >1 million metric tons of almonds valued at >\$7.5 billion in 2017, which represents nearly all almond production in the United States (NASS, 2017).
- Such high production levels are associated with a high level of risk from stored product insect pest infestation.
- Fumigation is the primary choice for disinfestation of storage pests in almond storage and processing facilities.

# Overview

- Phase I: 2013–2015.
- Phase II: 2015–2017.
- Phase III: 2018 completing survey questionnaires, summarizing questionnaire data, conducting data analyses, and project wrap up.

# 2013-2015

#### PH<sub>3</sub> resistance in red flour beetle and Indian meal moth populations in California almond storage facilities





Plodia interpunctella (Hübner) Indian meal moth (IMM) Tribolium castaneum (Herbst) Red flour beetle (RFB)

#### 2013–2015 — PH<sub>3</sub> resistance in red flour beetle and Indian meal moth

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Journal of Economic Entomology, 2016, 1-9 doi: 10.1093/iee/tow221 **Research** article

Insecticide Resistance and Resistance Management

**Phosphine Resistance in Adult and Immature Life Stages** of Tribolium castaneum (Coleoptera: Tenebrionidae) and Plodia interpunctella (Lepidoptera: Pyralidae) **Populations in California** 

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#### Abstract

Phosphine resistance in stored-product insects occurs worldwide and is a major challenge to continued effective use of this fumigant. We determined resistance frequencies and levels of resistance in Tribolium castaneum and Plodia interpunctella collected from California almond storage and processing facilities. Discriminating doses of phosphine were established for eggs and larvae of P. interpunctella and eggs of T. castaneum using laboratory susceptible strains of the two species. For T. castaneum and P. interpunctella eggs, discriminating doses were 62.4 and 107.8 ppm, respectively, over a 3-d fumigation period, and for P. interpunctella larvae, discriminating dose was 98.7 ppm over a 20-h fumigation period. Discriminating dose tests on adults and eggs showed that 4 out of 11 T. castaneum populations tested had resistance frequencies that ranged from 42 to 100% for adults and 54 to 100% for eggs. LC<sub>99</sub> values for the susceptible and the most resistant adults of T. castaneum were 7.4 and 356.9 ppm over 3 d, respectively. LC99 values for T. castaneum eggs were 51.5 and 653.9 ppm, respectively. Based on adult data, the most resistant T. castaneum beetle population was 49× more resistant than the susceptible strain. Phosphine resistance frequencies in P. interpunctella eggs ranged from 4 to 20%. Results show phosphine resistance is present in both species in California. Future research will investigate phosphine resistance over a wider geographic area. In addition, the history of pest management practices in facilities where insects tested in this study originated will be determined in order to develop phosphine resistance management strategies for California almond storage and processing facilities.

Key words: phosphine, stored-product, resistance management, red flour beetle, Indian meal moth

The United States is the world's leading producer of almonds. The the reliance on PH<sub>3</sub> and SF is expected to continue into the foresee-

Central Valley of California produces nearly all almonds in the able future. However, these two important fumigants have their United States, with annual production exceeding 840,000 metric own set of challenges. Limitations with SF include species-specific **Published in** Journal of **Economic** Entomology

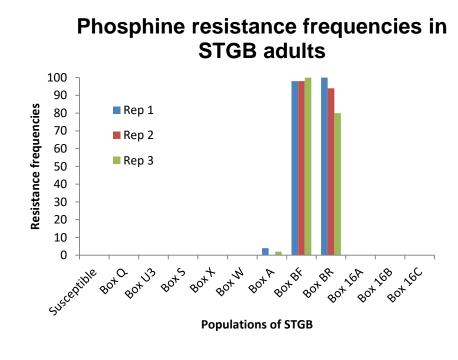
# 2015-2017

#### A survey of PH<sub>3</sub> Resistance in Indian Meal Moth, Red Flour Beetle, and Sawtoothed grain beetle in California Almond Storage Facilities



Plodia interpunctella (Hübner) Indian meal moth (IMM) *Tribolium castaneum* (Herbst) Red flour beetle (RFB) *Oryzaephilus surinamensis* (L.) Sawtoothed grain beetle (STGB)

# PH<sub>3</sub> Resistance Frequencies in STGB Adults — 11 Populations



STGB		esistanco equencie	
Populations	Rep 1	Rep 2	Rep 3
Susceptible	0	0	0
Box Q	0	0	0
Box U3	0	0	0
Box S	0	0	0
Box X	0	0	0
Box W	0	0	0
Box A	4	0	2
Box BR	98	98	100
Box BF	100	94	80
Box 16A	0	0	0
Box16B	0	0	0
Box 16C	0	0	0

8 out of 11 populations had no resistance (72.7%)

This is the first report of high PH<sub>3</sub> resistance frequencies in STGB in the United States and the world!

# Probit analyses of dose-response data for adults of susceptible and two PH<sub>3</sub>-resistant populations of saw-toothed grain beetle

STGB	LC <sub>50</sub>	LC <sub>95</sub>	LC <sub>99</sub>	X² (df)
Adults	(95% CI)	(95% CI)	(95% CI)	[H]
Susceptible	4.8	9.4	<mark>13.2</mark>	49.6 (20)
	(4.6 – 5.2)	(8.9 – 11.2)	(11.5 – 15.9)	[2.5]
Box BF	118.7	223.7	<mark>290.7</mark>	55.8 (19)
	(107.6 – 129.7)	(198.4 – 263.5)	(249.1 – 362.8)	[2.9]
Box BR	52.8	188.9	<mark>320.5</mark>	49.2 (19)
	(44.3 – 60.9)	(157.6 – 241.6)	(249.9 – 456.9)	[2.6]

Concentration of  $PH_3$  required to kill 99% **adults** of the most resistant population, Box BR, was **320.5** ppm based on 3-day fumigation.

Comparison of lethal concentrations (ppm) required to kill 50, 95, and 99% adults of two field populations of saw toothed grain beetle and those required to kill similar percentages of adults from the susceptible population.

Populations compared	Lethal concentration ratios (95% CI)				
	LC <sub>50</sub> LC <sub>95</sub> LC <sub>99</sub>				
Box BF vs susceptible	24.4	<mark>22.7</mark>	22.1		
	(22.9–25.9)	(20.5–25.2)	(19.2–25.2)		
Box BR vs susceptible	10.9	<mark>19.2</mark>	24.3		
	(9.8–12.0)	(16.7–22.1)	(19.9–29.7)		

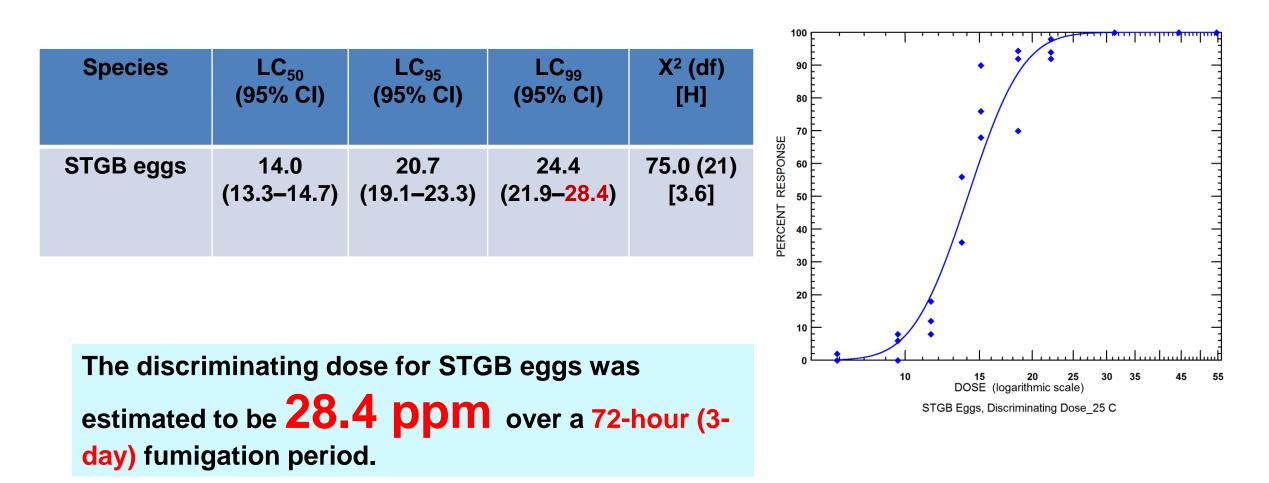
Highest level of resistance in STGB adults was 24. Adults of the most resistant population required 320 ppm of phosphine over 3-day exposure.

## **Discriminating Dose for STGB Eggs**

- FAO Protocol #16 does not have discriminating dose for eggs of STGB.
- Discriminating dose established using a laboratory susceptible strain.
- Protocol similar to that for IMM and RFB eggs were followed.
  - PH<sub>3</sub> dose response for STGB eggs during 72-h exposure to PH<sub>3</sub>.
  - Upper limit of the LC<sub>99</sub> confidence interval value is the discriminating dose.

## **Discriminating Dose for STGB Eggs**

Discriminating dose — a concentration of a fumigant that kills 99% of susceptible laboratory-reared insects in a fumigation that lasts a specified period of time at 25°C



#### PH<sub>3</sub> Resistance Frequencies in STGB Eggs (vs adults) Eggs Adults

	STGB		esistar equenc	
All populations were resistant	populations	Re p 1	Rep 2	Rep 3
	Susceptible	0	0	0
	Box Q	4	6	2
	Box U3	2	10	2
	Box S	2	8	0
	Box X	0	8	2
	Box W	4	0	0
	Box A	2	0	0
	Box BR	100	100	98
	Box BF	92	94	98

STGB		esistanc equenci	
populations	Rep 1	Rep 2	Rep 3
Susceptible	0	0	0
Box Q	0	0	0
Box U3	0	0	0
Box S	0	0	0
Box X	0	0	0
Box W	0	0	0
Box A	4	0	2
Box BR	98	98	100
Box BF	100	94	80
Box 16A	0	0	0
Box16B	0	0	0
Box 16C	0	0	0

8 out of 11 populations had no resistance (72.7%)

This is the first report of high PH<sub>3</sub> resistance frequencies in STGB in the United States and worldwide!

# **Probit analyses of dose-response data for eggs of susceptible and two PH<sub>3</sub> -resistant populations of saw-toothed grain beetle**

STGB Eggs	LC <sub>50</sub>	LC <sub>95</sub>	LC <sub>99</sub>	X² (df)
	(95% CI)	(95% CI)	(95% CI)	[H*]
Susceptible	14.0	20.6	<mark>24.2</mark>	59.5 (17)
	(13.3 – 14.6)	(19.0 – 23.2)	(21.8 – 28.4)	[3.5]
Box BF	122.2	561.3	<mark>1055.9</mark>	44.08 (19)
	(105.4 – 139.5)	(439.7 – 790.6)	(755.6 – 1706.0)	[2.3]
Box BR	101.7	523.0	<mark>1030.7</mark>	50.0 (19)
	(85.2 – 118.5)	(401.5 – 763.2)	(714.9 – 1762.5)	[2.6]

Concentration of  $PH_3$  required to kill 99% adults of the most resistant population of **eggs**, Box BF, was **1,055.9** ppm based on 3-day fumigation.

Comparison of lethal concentrations (ppm) required to kill 50, 95, and 99% eggs of two field populations of STGB and those required to kill similar percentages of eggs from the susceptible population.

Samples compared	Lethal concentration ratios			
	LC <sub>50</sub> (95% CI)	LC <sub>95</sub> (95% CI)	LC <sub>99</sub> (95% CI)	
Box BF vs	8.7	27.2	<mark>43.6</mark>	
Susceptible	(7.9 – 9.6)	(23.1 – 34.1)	(34.7 – 60.1)	
Box BR vs	7.3	25.4	<mark>42.6</mark>	
Susceptible	(6.4 – 8.1)	(21.1 – 32.9)	(32.8 – 62.1)	

Highest level of resistance in STGB eggs was 43.6. Eggs of the most resistant population required 1,056 ppm of phosphine over 3-day exposure

## PH<sub>3</sub> Resistance Frequencies in RFB Eggs

Population	Percentage Survival (%)				
	Replicate 1	Replicate 2	Replicate 3		
Box L Tc	86	78	80		
Box Q Tc	0	0	0		
Box R Tc	0	0	0		
Box U3 Tc	0	0	0		
Box W Tc	0	0	0		
Box X Tc	0	0	0		
Box 16A Tc	0	0	0		
Susceptible Tc	0	0	0		

6 out of 7 RFB egg populations had no **PH<sub>3</sub>** resistance detected (85.7%)

## **PH<sub>3</sub> Resistance Frequencies in RFB Adults**

Population	Percentage Survival (%)				
	Replicate 1	Replicate 2	Replicate 3		
Box L Tc	98	96	92		
Box Q Tc	0	0	0		
Box R Tc	0	0	0		
Box U3 Tc	0	0	0		
Box W Tc	0	0	0		
Box X Tc	0	0	0		
Box 16A Tc	0	0	0		
Box 16B Tc	0	0	0		
Box 16C Tc	0	0	0		
Susceptible Tc	0	0	0		

8 out of 9 RFB adult populations had no PH<sub>3</sub> resistance detected (88.9%)

### Probit Analyses of Dose-Response Data for Box L RFB Adults and Eggs\*

	LC <sub>50</sub> (95% CI)	LC <sub>95</sub> (95% CI)	LC <sub>99</sub> (95% CI)	Slope ± SE	X² (df) [H]
Adults					
Susceptible Tc	3.4	5.9	7.4	$6.9 \pm 0.37$	12.9 (16)
	(3.3–3.6)	(5.5–6.4)	(6.8–8.2)		[0.8]
Box L Tc	31.0	105.4	175.2	$8.0 \pm 0.39$	6.6 (10)
	(23.5–37.0)	(91.9–128.6)	(140.9–247.2)		[0.7]
Eggs					
Susceptible Tc	19.2	38.2	51.5	5.4 ± 0.25	64.8 (25)
	(16.7–21.6)	(32.2–50.1)	(44.6–62.4)		[2.6]
Box L Tc	125.3	223.2	283.5	$16.6 \pm 0.4$	40.8 (15)
	(115.1–136.1)	(197.3–266.2)	(242.0–358.2)		[2.7]

\* Probit analyses of mortality for the laboratory susceptible strain and phosphine-resistant Box L populations of *T. castaneum* adults and eggs, after 3 d exposure to PH<sub>3</sub> at 25°C. Lethal concentration values (LC) are in parts per million (ppm).

Comparison of lethal concentrations (ppm) required to kill 50, 95, and 99% of adults and eggs of Box L Tc and to those required to kill similar percentages of adults and eggs from the susceptible population.

Samples Compared	Lethal Concentration ratios				
	LC <sub>50</sub> (95% CI)	LC <sub>95</sub> (95% CI)	LC <sub>99</sub> (95% CI)		
Adults					
Box L Tc vs	9.1	17.9	23.7		
susceptible	(7.1–10.3)	(16.7–20.1)	(20.7–30.2)		
Eggs					
Box L Tc vs	6.5	5.8	5.5		
susceptible	(6.3–6.9)	(5.3–6.1)	(5.4–5.7)		

# Indian Meal Moth (IMM)



- Use of CIDETRAK IMM (mating disruption) by facilities Mr. Ed Hosoda is trying to collect IMM for tests has been so successful that insects are hard to find.
- This seems to imply CIDETRAK IMM is a great tool to incorporate in IMM phosphine resistance management strategies for almond storage and processing facilities.



Indian Meal Moth (IMM)

## Findings for 2013–2015 and 2015–2017

- What is the highest concentration of PH<sub>3</sub> required for control of any of the insect populations investigated? 1,055.9 ppm for STGB eggs.
- What is the proportion of RFB populations with no phosphine resistance? 15 out of 20 populations 75% of the populations.
- What is the proportion of STGB populations with no PH<sub>3</sub> resistance? 8 out of 11 populations — 72.7% of the populations.
- What is the proportion of IMM populations with no phosphine resistance? 3 out of 3 populations — 100% of the larval populations. <u>However, eggs of all IMM populations were resistant.</u>

## Findings for 2013–2015 and 2015–2017

CIDETRAK IMM is a great tool to incorporate in IMM PH<sub>3</sub> resistance management strategies for almond storage and processing facilities.





## 2013–2017 — What Was Accomplished

- PH<sub>3</sub> resistance was evaluated in STGB for 7 egg populations and 11 adult populations.
- PH<sub>3</sub> resistance was evaluated in RFB for 18 egg populations and 20 adult populations.

## 2013–2017 Presentations and Manuscripts

- Zhaorigetu Hubhachen, George Opit, Sandipa Gautam, Charles Konemann, and Ed Hosoda. 2018. Phosphine Resistance in Saw-toothed Grain Beetle, Oryzaephilus surinamensis (Coleoptera: Silvanidae) in the United States. <u>To be presented at the 12<sup>th</sup> International Working Conference on Stored Product</u> <u>Protection. Berlin, Germany. Oct. 7–11</u>.
- George P. Opit. 2017. A survey of phosphine resistance in Indian meal moth, red flour beetle, and sawtoothed grain beetle in California almond storage facilities. Talk: <u>Workshop on Emerging Pests and</u> <u>Emerging Fumigation Technologies in Grain Stores. Crops Research Institute, Prague, Czech Republic.</u> <u>June 26–30, 2017.</u>
- Zhaorigetu Hubhachen, Sandipa Gautam, Charles Konemann, George Opit and Ed Hosoda. 2017. Phosphine resistance in Oryzaephilus surinamensis (Coleoptera: Silvanidae). Talk: <u>The 65<sup>th</sup> Southwestern</u> Branch of the ESA Annual Meeting. Austin, TX. April 9–13, 2017.
- Sandipa G. Gautam, George Opit and Kandara Shakya. 2016. Phosphine resistance in Oryzaephilus surinamensis (L.) from almond storage and processing facilities in California. Talk: <u>XXV International</u> <u>Congress of Entomology, Orlando, Florida, USA. Sep. 25–30.</u>
- Gautam, S. G., G. P. Opit, and E. Hosoda. 2016. Phosphine Resistance in Adult and Immature Life Stages of *Tribolium castaneum* and *Plodia interpunctella* Populations in California. <u>Journal of Economic</u> <u>Entomology 2016; doi: 10.1093/jee/tow221.</u>
- Zhaorigetu Hubhachen, Sandipa Gautam, Charles Konemann, George Opit and Ed Hosoda. 2017. Phosphine resistance in *Oryzaephilus surinamensis* (Coleoptera: Silvanidae) in the United States. (Manuscript in Preparation to be submitted to Journal of Economic Entomology).

## Survey of the History of Stored-Product Insect Pest Management Practices in Almond Storage and Processing Facilities

Objective is development of PH<sub>3</sub> resistance management strategies for almond storage and processing facilities based on laboratory data on PH<sub>3</sub> resistance and the history of pest management practices obtained through completed questionnaires

# Hypothesis

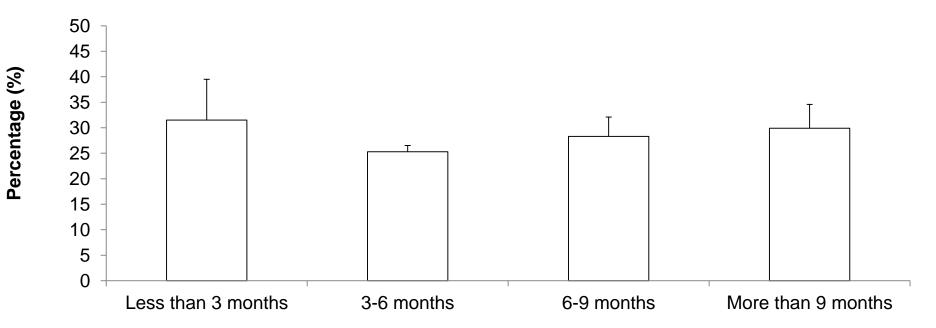
Almond storages and processing facilities with no  $PH_3$ resistant insect pest populations engage in pest management practices that keep resistance in check

# Methodology

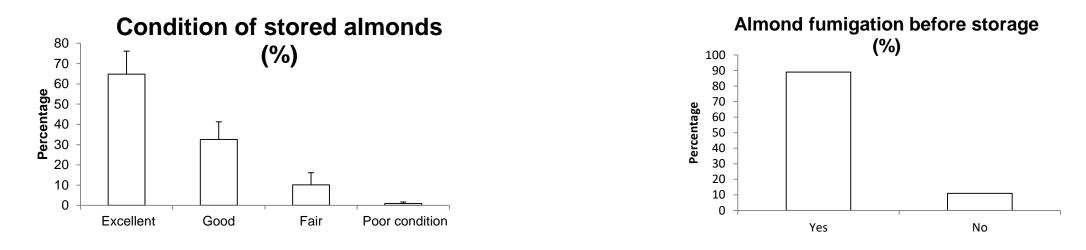
- 1. Developed questionnaire;
- Distributed questionnaires to different almond storages and processing facilities;
- 3. Collected completed questionnaires; and
- 4. Summarized, analyzed, and drew conclusions from data in the completed questionnaires.

#### Storage time

% of almonds stored for different time periods in 2014–2015



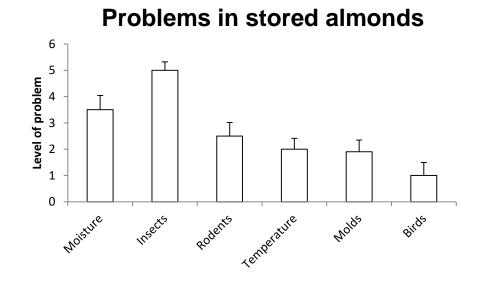
#### **Receiving and Handling**



#### Table 1. Action taken by facilities on receipt of infested almonds

	Refuse	Receive without discount	Receive with discount	Other (Fumigation)
Percentage (n)	37% (4)	27% (3)	18% (2)	18% (2)

#### Factors causing risk to stored almonds



Level 1: least important and Level 6: most important

# Insect pests are the most important factor of concern in almond storage

## **Results** — Fumigation

#### Fumigants used for control of storage pests

	PH <sub>3</sub>	Sulfuryl fluoride (SF)	$PH_3$ and SF	Ecofume
# of facilities:	10	2	3	1

#### **Concentration of fumigants (ppm) and fumigation time**

Time (h) # of facility (Dom) Facility	24	48	48-72	72	96	120	240	Note
45				1				
300	1			1				
400				2+				
300-500			1					
500			1			1	1#	
700		1						
500-1000				1				
1000				1	1*			

\*"After Dr. Opit's research on  $PH_3$  resistance to STGB, we increased the rate and exposure period and are now controlling the insects."

<sup>#</sup>Resistant population of RFB from Box L

\*Box 18A: New sample; Resistance frequency of the population will be determined

#### Fumigation

#### Fumigation practices in almond storages

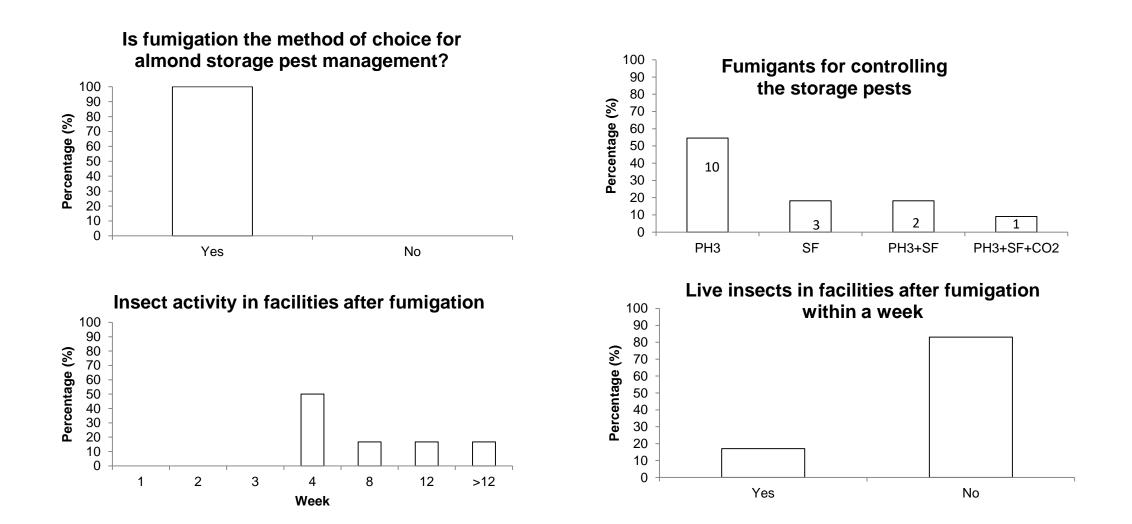
	Yes (%)	No (%)	Note
Are the storage structures sealed checked for sealing before PH <sub>3</sub> fumigation?	100	0	
Are PH <sub>3</sub> concentrations monitored during fumigation?	92	8	
Does PH <sub>3</sub> fumigation last the specified period of time?	90	10	
Are the facilities scouted for insects to determine effective control after fumigation using PH <sub>3</sub> ?	50	40	10 (not regularly)

### Fumigation

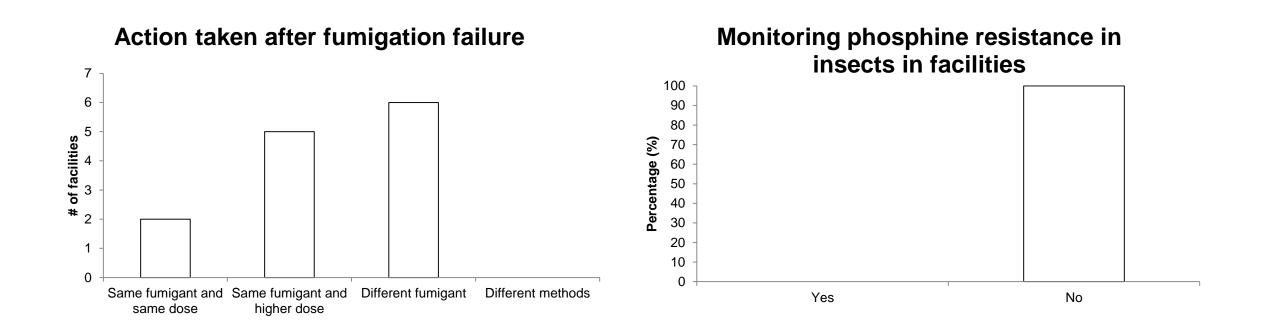
#### Effectiveness of fumigation in almond storages

When was the last time	1.8 years ago; RFB and CFB (1 facility)
you noticed fumigation	2. RFB (1 facility)
failure when using PH <sub>3</sub> ?	3. IMM and STGB (1 facility)
What insects were not	4. 2016, STGB; Then increased the dose
controlled?	and exposure time (1 facility)

## **Results** — Fumigation and its Effectiveness



# **Results** — Fumigation and its Effectiveness

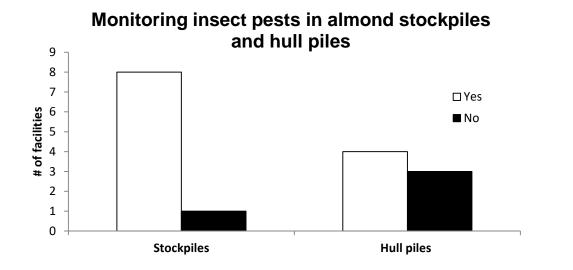


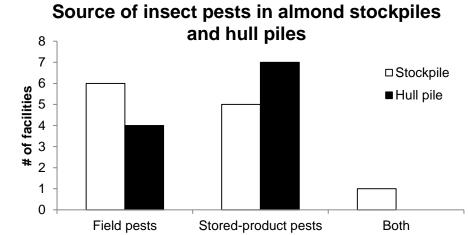
# **Results** — Fumigation and its Effectiveness

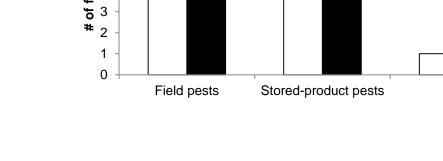
Management of PH<sub>3</sub> resistance in insect in almond storages

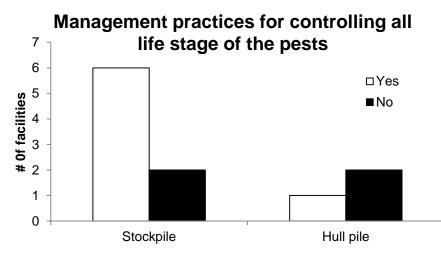
Question	Answer
When was the last time	1. 8 years ago (1 facility)
you noticed that there	2. 2 years ago (1 facility)
may be PH <sub>3</sub> resistant	3. Have known about Lesser grain
insects in your storage	borer resistance to PH <sub>3</sub> , but have
facility?	suspected resistance with other
	insects (1 facility)
Did you change the pest	1. Yes, switched to SF
management practices	2. Used higher doses of $PH_3$ and
after you noticed there	longer exposure time at the
may be PH <sub>3</sub> resistant	recommendation of Ed Hosoda
insects? If yes, what did	3. Increased monitoring and dosage
you do?	

#### **Results — Management of Almond Stockpiles and Hull Piles**









#### **Results — Management of Almond Stockpiles and Hull Piles**

Management of insect pests in stockpiles and hull piles by fumigation

Fumigant For facility type	PH <sub>3</sub>	PH <sub>3</sub> +SF	PH <sub>3</sub> + RI	$PH_3 + SF + RI + CO_2$
Stockpile	11	2	1	1
Hull pile			1	

#### Pest management in almond stockpiles and hull piles

	Yes (%)	No (%)	Note
Do you monitor concentration during fumigations?			
	92	8	
Do you maintain stockpiles/hull piles to prevent contamination from trash, sanitary facilities, dusts, and other potential source of contamination?	100	0	
Do you maintain a distance between storage areas (processing facility, warehouse, storage bins) and trash, sanitary facilities, dusts, and other potential source of contamination?	92	8	

# Results

### Personal Training for Fumigation

	Yes (%)	No (%)	Note
Did you attend a "Pest management workshop" and/or "Fumigation workshop" in the past five years?	100	0	
Do you dispose waste regularly?	100	0	

# Summary — Laboratory PH<sub>3</sub> Resistance Tests

- Twenty two healthy cultures out of 29 different cultures have been established since 09/18/2013.
- Twenty populations of RFB were tested for the evaluation of PH<sub>3</sub> resistance.
- Five RFB populations (25%) had strong resistance to PH<sub>3</sub> (resistance frequencies ~90% or higher) 75% of the populations were <u>susceptible</u> to PH<sub>3</sub>.
- Eleven populations of STGB were evaluated for  $PH_3$  resistance.
- Two STGB populations (27.3%) had strong resistance to PH<sub>3</sub> (resistance frequencies ~90% or higher) 72.7% of STGB populations were <u>susceptible</u> to PH<sub>3</sub>.
- $LC_{99}$  for eggs of the strongest population of STGB was 1,055.9 ppm.
- Larvae and eggs from 6 populations of IMM were evaluated for  $PH_3$  resistance.
- Larvae from all 6 populations of IMM were <u>susceptible</u> to PH<sub>3</sub> however, eggs from 3 populations showed weak resistance to PH<sub>3</sub> (Gautam et al., 2016).

## Summary — Laboratory PH<sub>3</sub> Resistance Tests

- Since 2015, we have not made new IMM cultures from any almond storage and processing facility.
- According to Mr. Hosoda, use of CIDETRAK for IMM control has been so successful making it difficult to find and collect IMM.
- CIDETRAK is a great tool to incorporate in IMM PH<sub>3</sub> resistance management strategies for almond storage and processing facilities.

### **Summary — Questionnaire Information**

- >90% of almonds in the field were in excellent or good condition.
- >90% of facilities fumigate almonds after receipt.
  - $\circ$  10 out of 16 facilities (62.5%) fumigate almond storages using PH<sub>3</sub>.
  - $\circ~$  2 facilities (12.5%) use SF.
  - $\circ$  3 facilities (18.8%) use both PH<sub>3</sub> and SF.
  - 1 facility (6.2%) uses EcoFume.
- All storage structures are properly sealed and then checked for good sealing before PH<sub>3</sub> fumigation.
- PH<sub>3</sub> concentrations used ranged between 45–1000 ppm and exposure periods between 24–240 h (1–10 d) low dose and short exposure time can cause development of PH<sub>3</sub> resistance.

## **Summary — Questionnaire Information**

- In 1 facility a PH<sub>3</sub>-resistant population had been detected 8 years ago but no resistant insects from these facility were found in our testing.
- Highly PH<sub>3</sub>-resistant populations of both RFB and STGB were found in 1 facility but they were controlled effectively application of 1,100 ppm for 5–7 days <u>a recommendation that resulted from tests in our lab</u>.
- 11 out of 13 facilities applied the "same fumigant at higher dose" or a "different fumigant" whereas 2 out of 13 facilities applied "same fumigant at same dose" after fumigation failure.
- All individuals fumigating have attended a "Storage pest management Workshop" and/or "Fumigation workshop."
- Regular handling and disposal of waste practiced by all facilities.

	Resistance			Management practice		
Eggs	frequency	LC <sub>99</sub> (95% CI)	Level of resistance	Dose (ppm)	Exposure time (h)	
Susceptible STGB	0	<mark>24.2</mark> (21.8 – 28.4)	1			
Box BF STGB	99.3 ± 1.2	<mark>1055.9</mark> (755.6 – 1706.0)	<mark>43.6</mark> (34.7 – 60.1)	500	48-72	
Box BR STGB	94.7 ± 3.1	<mark>1030.7</mark> (714.9 – 1762.5)	<mark>42.6</mark> (32.8 – 62.1)	500	48-72	
Susceptible RFB	0	<mark>51.5</mark> (44.6–62.4)	1			
Box L RFB	81.3 ± 4.2	<mark>283.5</mark> (242.0–358.2)	<mark>5.5</mark> (5.4–5.7)	500	240	

	Management practices							
Eggs	Are the storage structure sealed/checked before fumigation?	Are PH <sub>3</sub> concentration monitored during fumigation?	Does PH <sub>3</sub> fumigation last the specified period of time?	Are the facilities scouted for insects to determine effective control after PH <sub>3</sub> fumigation?	How long after the fumigation is completed, have you noticed insect activity in your facility?	After you notice that the fumigation is not effective, what action do you take?	Did you attend storage pest management/fumig ation workshop?	Do you dispose waste regularly?
Susceptible STGB								
Box BF STGB	Yes	Yes	Νο	Νο	4 wk	Same fumigant ; higher dose	Yes	Yes
Box BR STGB	Yes	Yes	Νο	No	4 wk	Same fumigant ; higher dose	Yes	Yes
Susceptible RFB								
Box L RFB	Yes	Yes	Yes	Yes	4 wk	Different fumigant (SF)	Yes	Yes

All these facilities that had insect populations with strong resistance seemed to have good management practices. The main reason for the strong phosphine resistance in pest populations appears to be under-dosing.

		Management practice				
All STGB	Resistance frequency	Dose (ppm)	Exposure time (h)			
Box A	0	500	72			
Box 16A	0	300	72			
Box 16B	0	500	48–72			
Box 16C	0	700	48			

	Management practices							
Adults	Are the storage structure sealed/checked before fumigation?	Are PH <sub>3</sub> concentration monitored during fumigation?	Does PH <sub>3</sub> fumigation last the specified period of time?	Are the facilities scouted for insects to determine effective control after PH <sub>3</sub> fumigation?	How long after the fumigation is completed, have you noticed insect activity in your facility?	After you notice that the fumigation is not effective, what action do you take?	Did you attend storage pest management/fumig ation workshop?	Do you dispose waste regularly?
Box A	Yes	Yes	Yes	Yes		N/A	Yes	Yes
Box 16A	Yes	Yes	Yes	Yes	4 wk	N/A	Yes	Yes
Box 16B	Yes	Yes	Yes	Yes	8 wk	N/A	Yes	Yes
Box 16C	Yes	Yes	Yes	Yes	8 wk	NA	Yes	Yes

All these facilities with susceptible insect populations appeared to have good management practices and pests were controlled effectively using  $PH_3$  at 300–700 ppm and 48–72 h exposure time.

# Conclusions

- The cause of PH<sub>3</sub> resistance in almond storage insect pests may most likely be due to under-dosing and short exposure time instead of gene flow.
- For the almond industry, the recommended dose is 500–1,000 ppm for a minimum of 3 d, but 5–7 d are highly recommended, at 20-30°C.
- Based on our research, perhaps this should be changed to a recommended dose of 500–1,100 ppm for a minimum of 3 d, but 5–7 d are highly recommended, at 20–30°C.
- Alternatives such as SF should be used for controlling PH<sub>3</sub>-resistant insect populations.
- CIDETRAK is a great tool to incorporate in IMM PH<sub>3</sub> resistance management strategies for almond storage and processing facilities.
- The majority of the facilities (>90%) applied good pest control practices.

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