Pesticide Transfer in Alfalfa Leafcutting Bee Nests

Investigating provision uptake of pesticides from alfalfa leaves

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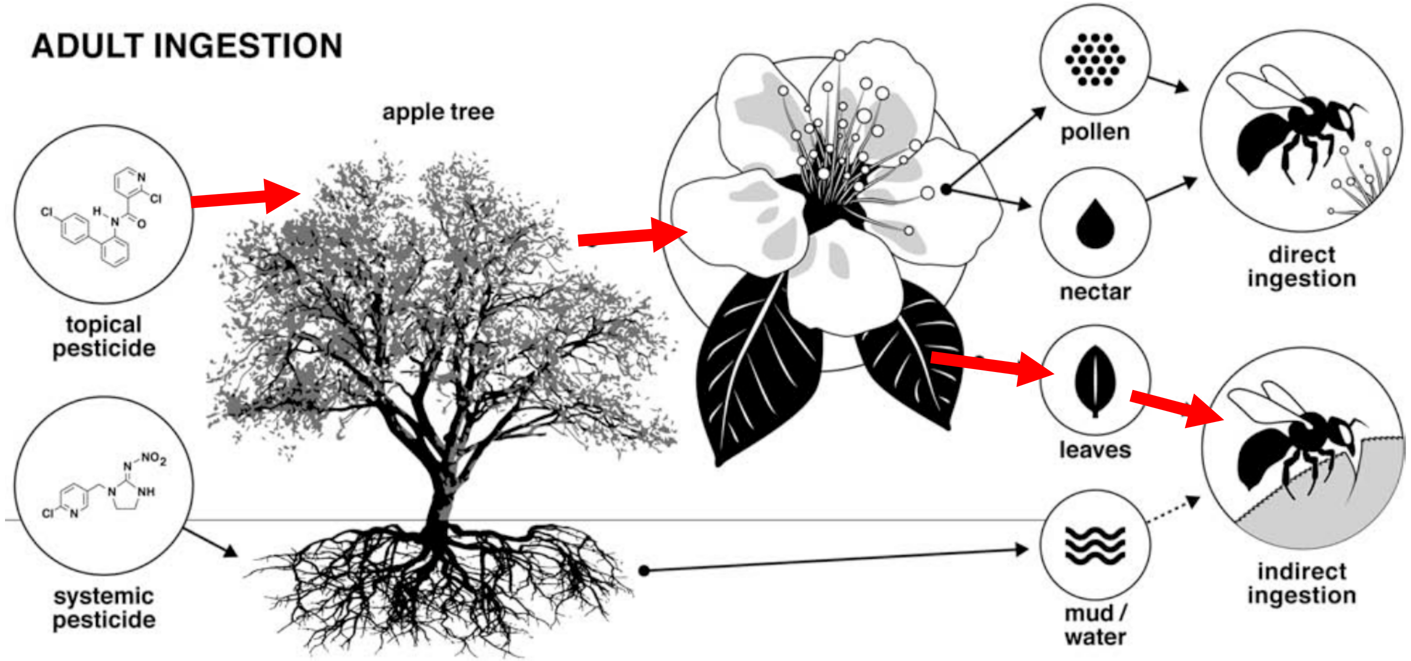
Pesticides and Pollinators

- Pesticides are a significant stressor for pollinator health.[1]
- Pollinator pesticide risk assessments for honeybees are extrapolated to all other species of bees.[2,3]
- Solitary bees have different life histories compared to honeybees.[3,4]
- Quantifying pesticide exposure routes for solitary bees is needed.



Kopit A.M. and T.L. Pitts-Singer. "Routes of Pesticide Exposure in Solitary, Cavity-Nesting Bees. Environmental Entomology. 2018.

[1]Tosi, et al. "Lethal, sublethal, and combined effects of pesticides on bees: A meta-analysis and new risk assessment tools." Science of the Total Environment. 2022

[2]Sgolastra, et al. "Pesticide Exposure Assessment Paradigm for Solitary Bees." Environmental Entomology. 2019.

[3]USEPA. "Guidance for Assessing Pesticide Risks to Bees." 2014.

[4]Kopit and Pitts-Singer. "Routes of Pesticide Exposure in Solitary, Cavity-Nesting Bees." Environmental Entomology. 2018

Pesticide Exposure Routes for Solitary Bees[4]

Ingestion of pesticides by adult bees

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Ingestion of pesticides by adult bees

Contact with pesticide contaminated surfaces by adult bees

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- Ingestion of pesticides by adult bees
- Contact with pesticide contaminated surfaces by adult bees
- Ingestion of pesticides by bee larvae

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- Ingestion of pesticides by adult bees
- Contact with pesticide contaminated surfaces by adult bees
- Ingestion of pesticides by bee larvae
- Transovarial transmission of pesticides from mother bee to eggs

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Ingestion of Pesticides by Larvae

- Larval ingestion of pesticides occurs for solitary, cavity nesting bees.[5]
- Peterson, et al. identified pesticides present in cavity-nesting bee and wasp larvae (and all other parts of the nests).

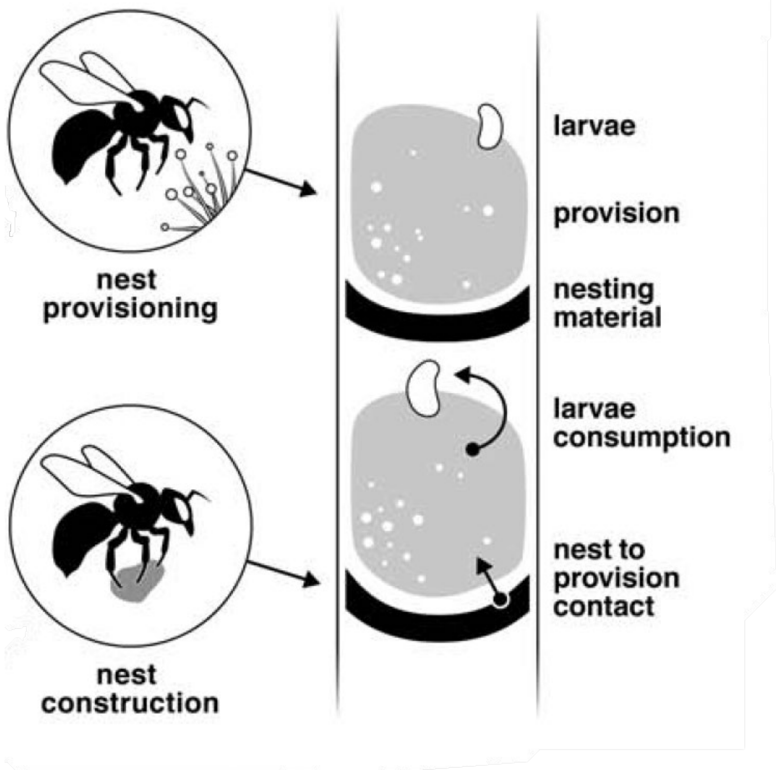
Matrix	Feed yard		Cropland	
	N	Detection Frequency (%)	N	Detection Frequency (%)
Capped leaf	14	86	16	69
Dead larvae	16	81	8	50
Feces	50	68	19	58
Larvae	45	60	13	23
Leaf capsule	20	70	19	53
Mud	74	80	26	58
Parasites	13	54	9	22
Pollen	8	87	8	38

N = number of samples.

[5]Peterson, et al. "Use of nest bundles to monitor agrochemical exposure and effects among cavity nesting pollinators." Environmental Pollution. 2021.

Ingestion of Pesticides by Larvae

- Routes of pesticide contamination in provisions:[6]
  - Nectar and pollen are contaminated prior to provisioning.
  - Nesting material transfers pesticides into provisions.
- **Lack of evidence on the transfer of pesticides from nesting material into provisions.**

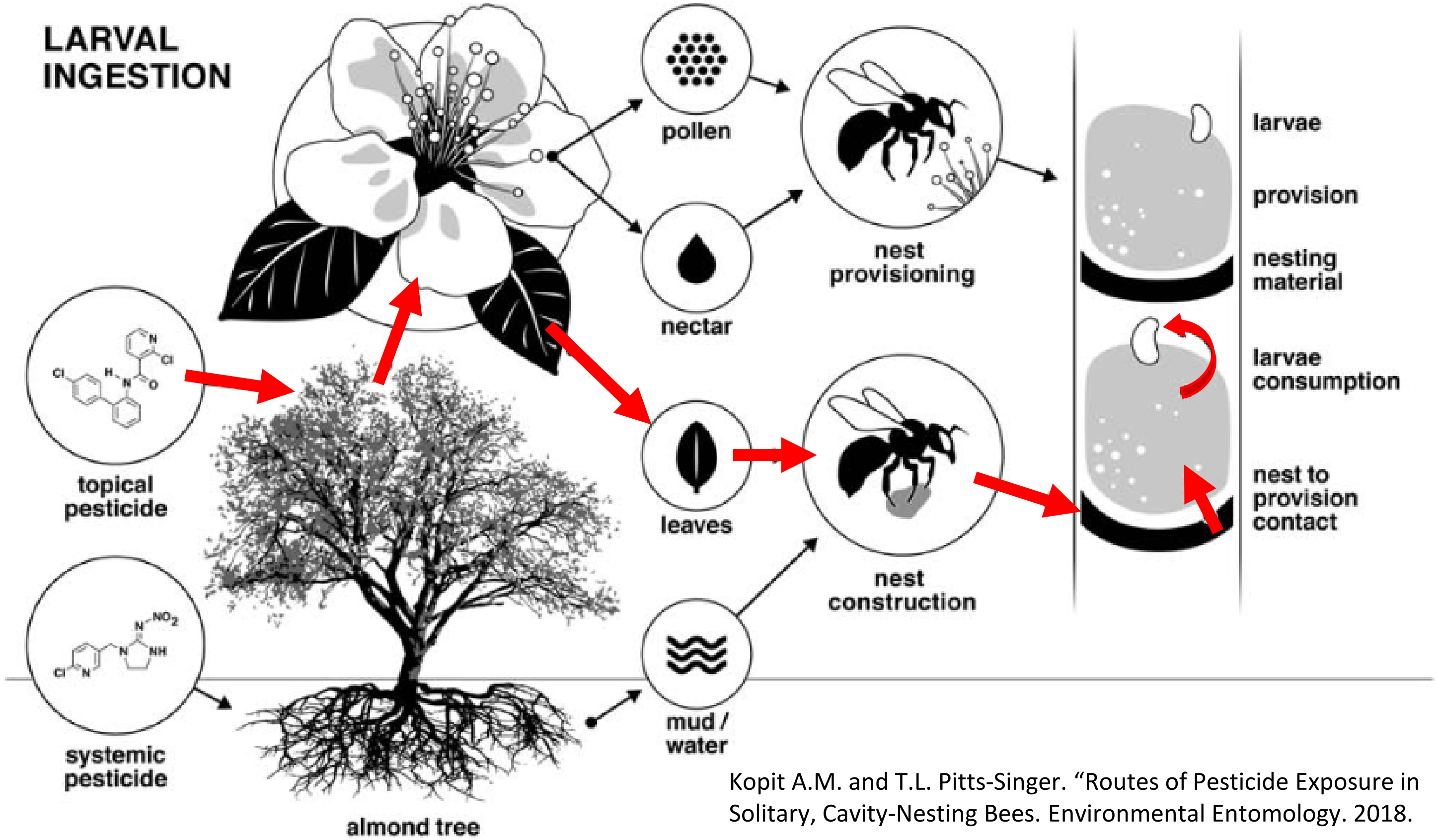


Kopit and Pitts-Singer. "Routes of Pesticide Exposure in Solitary, Cavity-Nesting Bees." Environmental Entomology. 2018

[6]Pitts-Singer and Barbour. "Effects of residual novaluron on reproduction in alfalfa leafcutting bees, *Megachile rotundata* F. (Megachilidae). Pest Manag Sci. 2017

Objective:

Determine if nesting materials are an important source of pesticides to provisions



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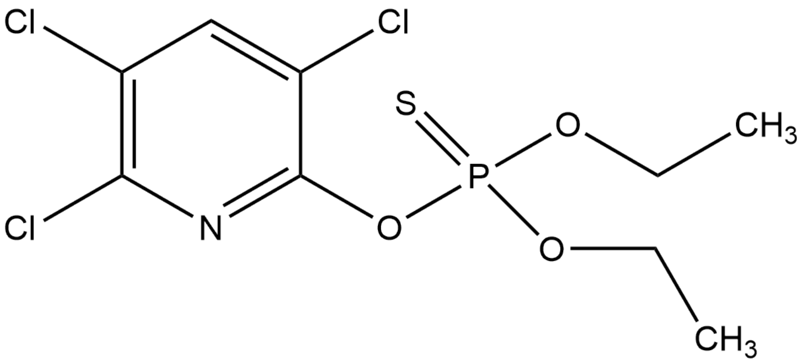
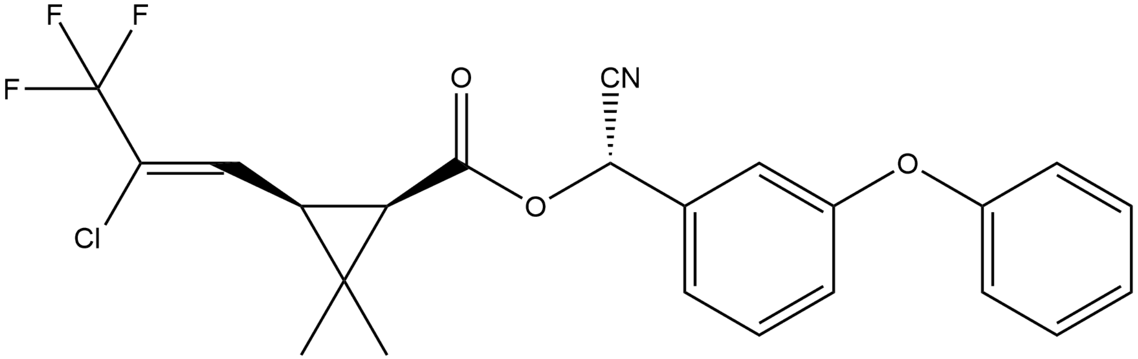
Insecticides of Interest

Active Ingredient: Chlorpyrifos
Chemical Class: Organophosphate

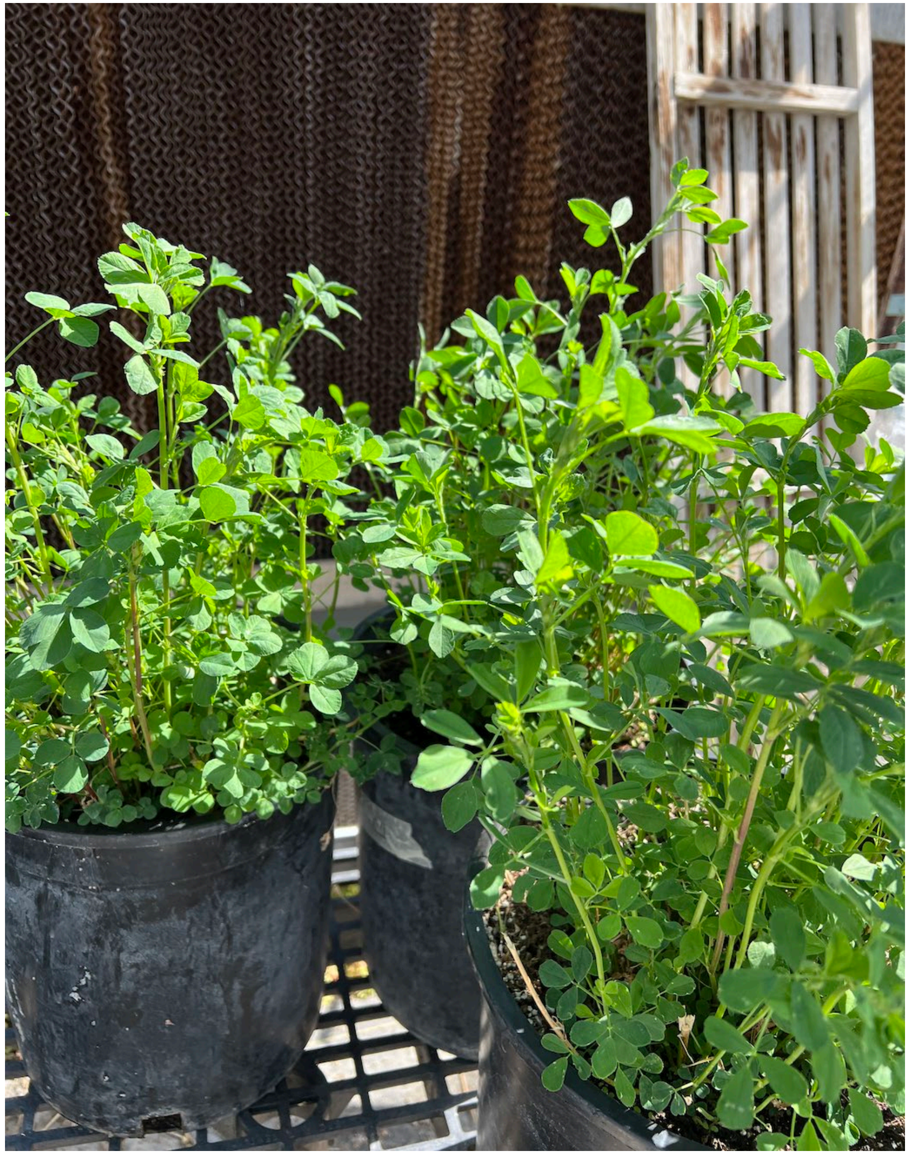
Active Ingredient: λ-cyhalothrin
Chemical Class: Pyrethroid





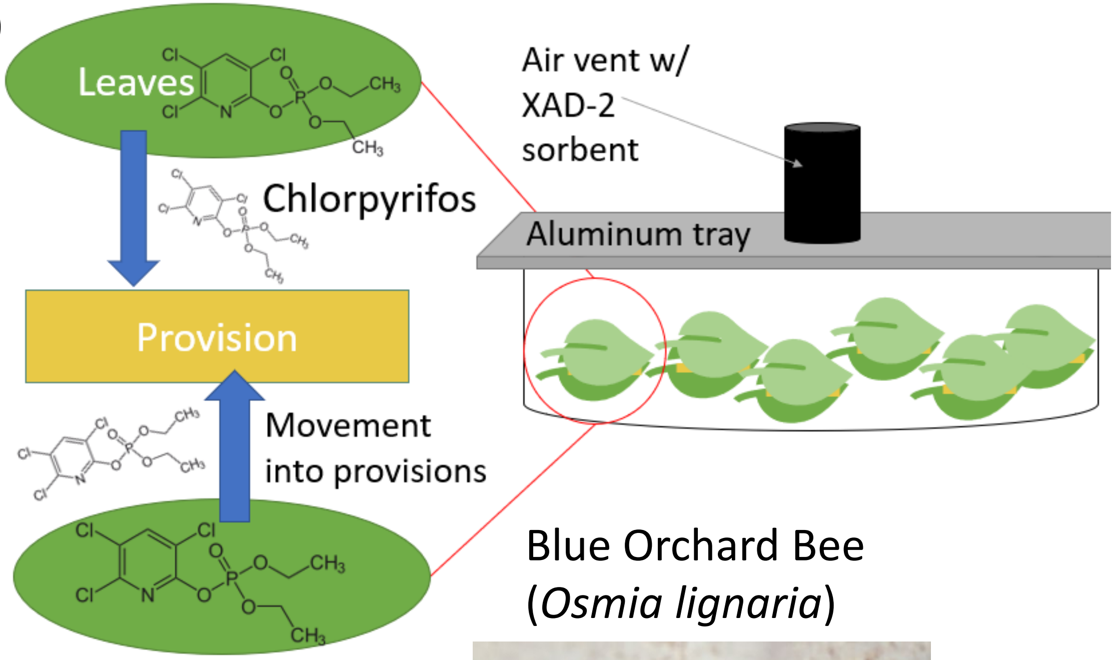
Approach

1. Determine if the transfer of pesticides from nesting materials into solitary bee provisions occurs
2. Calculate pesticide distribution ratios and transfer rates between nesting materials and provisions
3. Calculate hypothetical pesticide exposure for solitary bee larvae



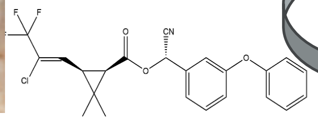
Experimental Methods

Alfalfa Leafcutting Bee
(*Megachile rotundata*)

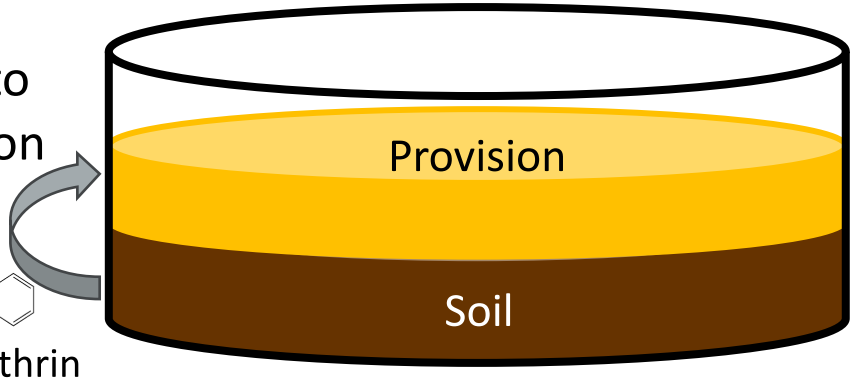


Blue Orchard Bee
(*Osmia lignaria*)

Transfer into
BOB provision



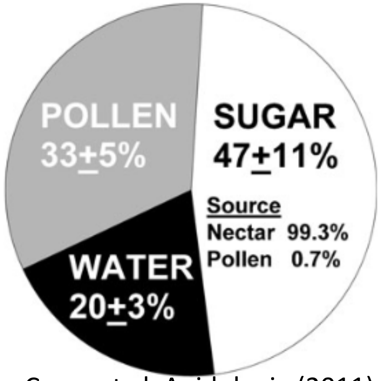
λ-cyhalothrin



Results

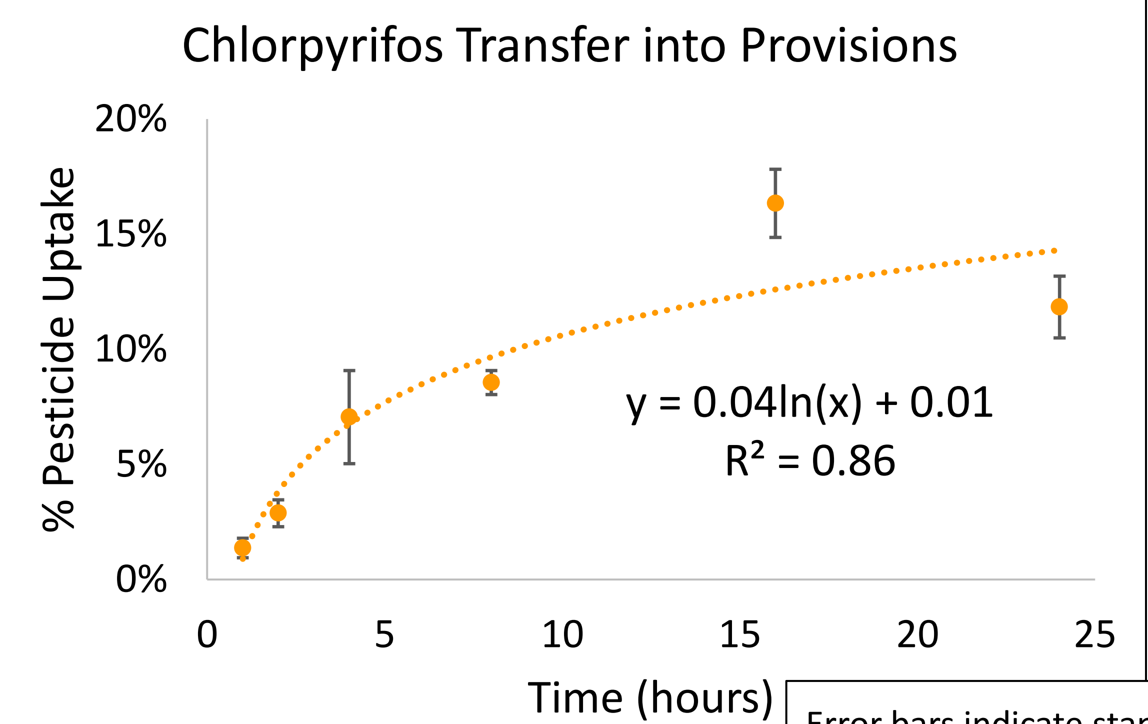
Pesticide Uptake by Provisions

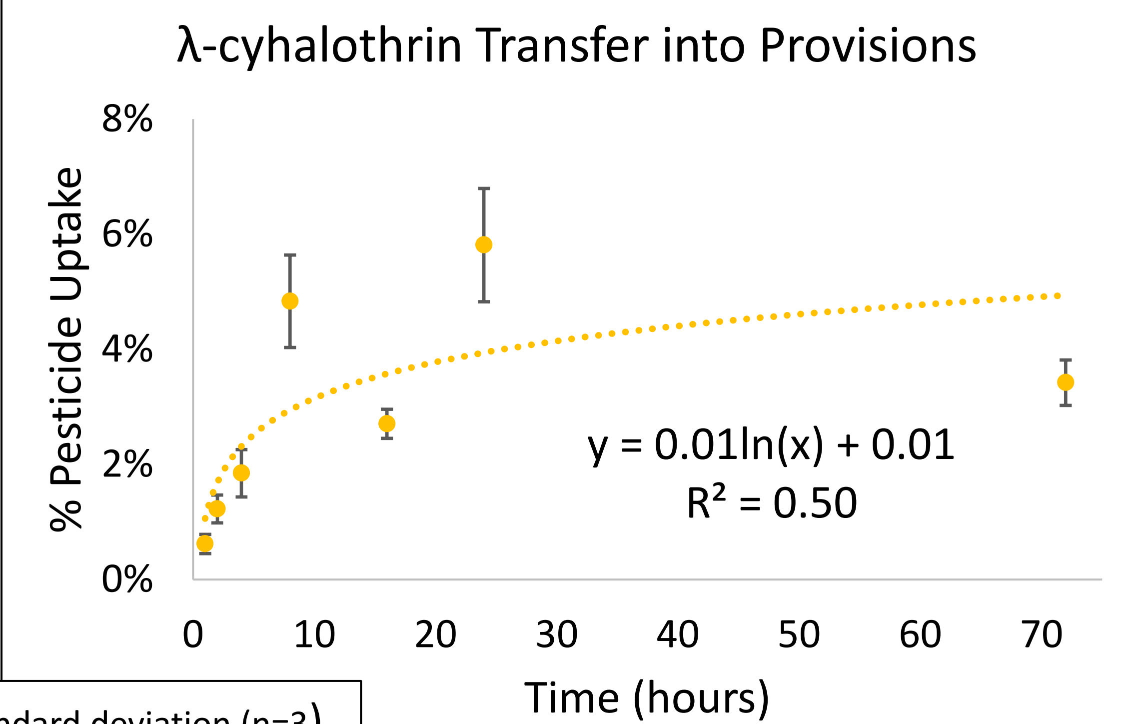
- Do pesticides transfer from alfalfa leaves to provisions?
- Yes! And chlorpyrifos had higher uptake compared to λ-cyhalothrin.
- Provisions have high water/sugar content.
  - Chlorpyrifos is more soluble in water than λ-cyhalothrin.



Cane, et al. Apidologie (2011)

Constituents of ALCB Provision





Error bars indicate standard deviation (n=3)

Approach 2: Calculate pesticide distribution ratios and transfer rates between nesting materials and provisions

Distribution Ratios and Transfer Rates

Distribution Ratio: concentration of the pesticide between two phases but does not imply that equilibrium has been achieved.

$$D_{Leaf-Provision,t} = \frac{Concentration\ in\ Leaves}{Concentration\ in\ Provision} \text{ at time } t$$

Transfer Rates: Experimentally determined.

Calculated by determining the change in pesticide concentration over time in the provisions.

Pesticide A.I	$D_{leaf\text{-}prov,72h}$	Provision uptake constant: k (h^{-1})
Chlorpyrifos	2.75	0.01
λ-cyhalothrin	25.12	0.005

Approach 3: Calculate hypothetical pesticide exposure for solitary bee larvae

Hypothetical Scenario

An alfalfa seed grower has just applied Chlorpyrifos 4E-AG at the max rate on their alfalfa field at 8 am.

To ensure the alfalfa leaf cutting bees are not harmed, they wait 24 hours before releasing their bees.

They want to know if the bee's offspring will have any pesticide exposure.

GROUP 1B INSECTICIDE

Drexel

Chlorpyrifos 4E-AG

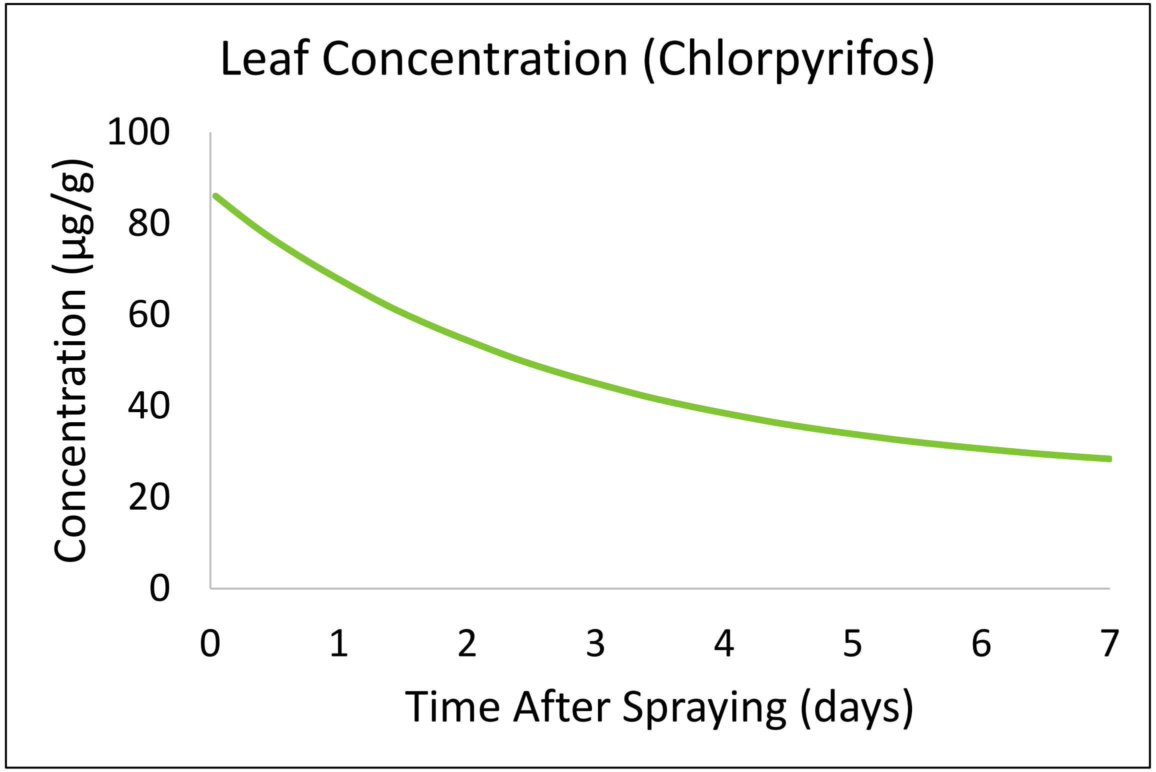
Insecticide

Pesticide Dissipation from Agricultural Lands (PeDAL) Model[7]

- Estimates pesticide concentration in plants and soil after application
  - Takes account time of application, environmental conditions, etc.

  pesticidetoolkit.usu.edu

- Pesticide concentration 24 hours after an 8 AM application
  - Chlorpyrifos: ≈ 67.8 µg/g leaf
  - λ-cyhalothrin: ≈ 1.9 µg/g leaf



Temp: between 50 °F - 77 °F **Relative Humidity:** 30%

Location: North Logan, UT.

[7]Lyons and Hageman. “Foliar Photodegradation in Pesticide Fate Modeling: Development and Evaluation of the Pesticide Dissipation from Agricultural Land (PeDAL) Model.” Environ Sci Tec. 2021.

Hypothetical Scenario

Question 2:

It take 4 hours for the alfalfa leafcutting bee to finish their first nest capsule to when they place their first provision.

How much chlorpyrifos has dissipated from the leaves before provisions are placed?

https://mdc.mo.gov/discover-nature/field-guide/leafcutter-bees

Hypothetical Scenario: Question 2

The nests are a completely different environment than in the fields.

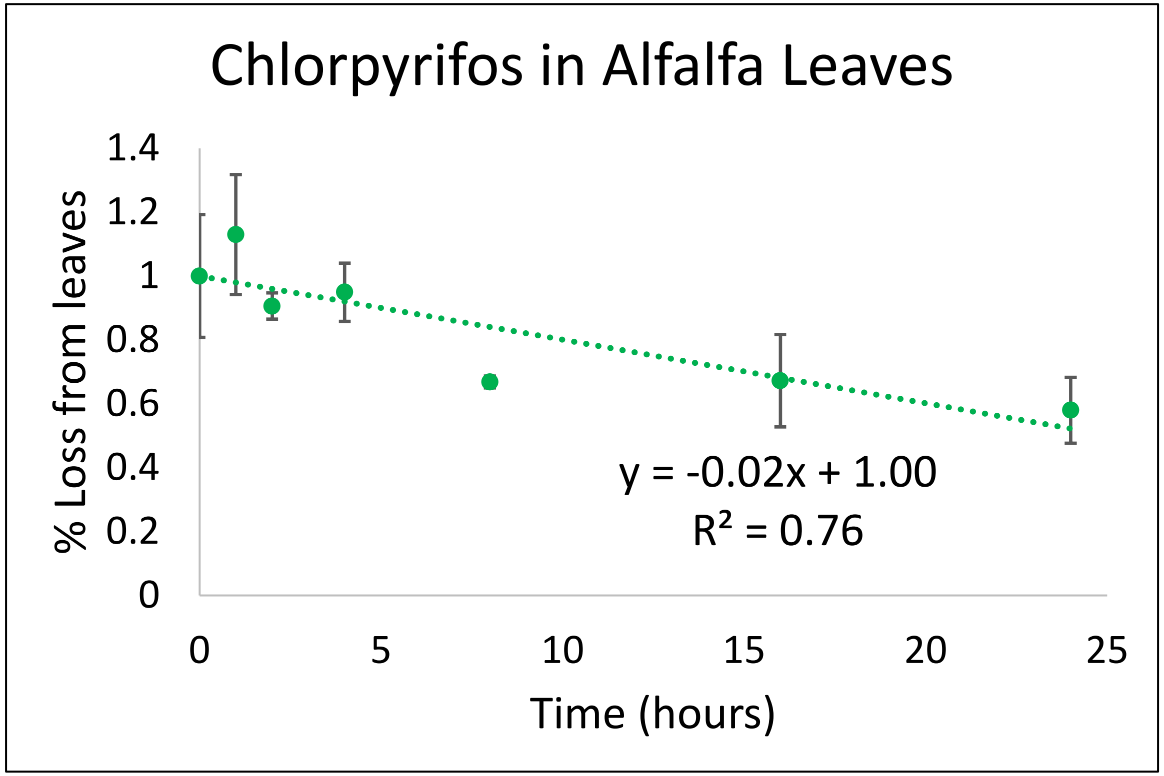
Use dissipation rate from leaves to estimate pesticide concentrations on leaves before provisions come into contact

Rate Equation: $\frac{C_0}{C} = kt + 1$

C_0 = 67.8 µg/g, k = 0.03 h^{-1} and t = 4 hours

Concentration of chlorpyrifos in leaves (the nesting material) after 4 hours

<u>**60.5 µg chlorpyrifos / g leaf**</u>



Hypothetical Scenario: Question 3

At the fastest rate, the egg takes three days for the larvae to hatch and begin eating the provision.

How much chlorpyrifos will the larvae consume?

https://www.ars.usda.gov/pacific-west-area/logan-ut/pollinating-insect-biology-management-systematics-research/docs/alfalfa-leafcutting-bee-alcb/

Hypothetical Scenario: Question 3

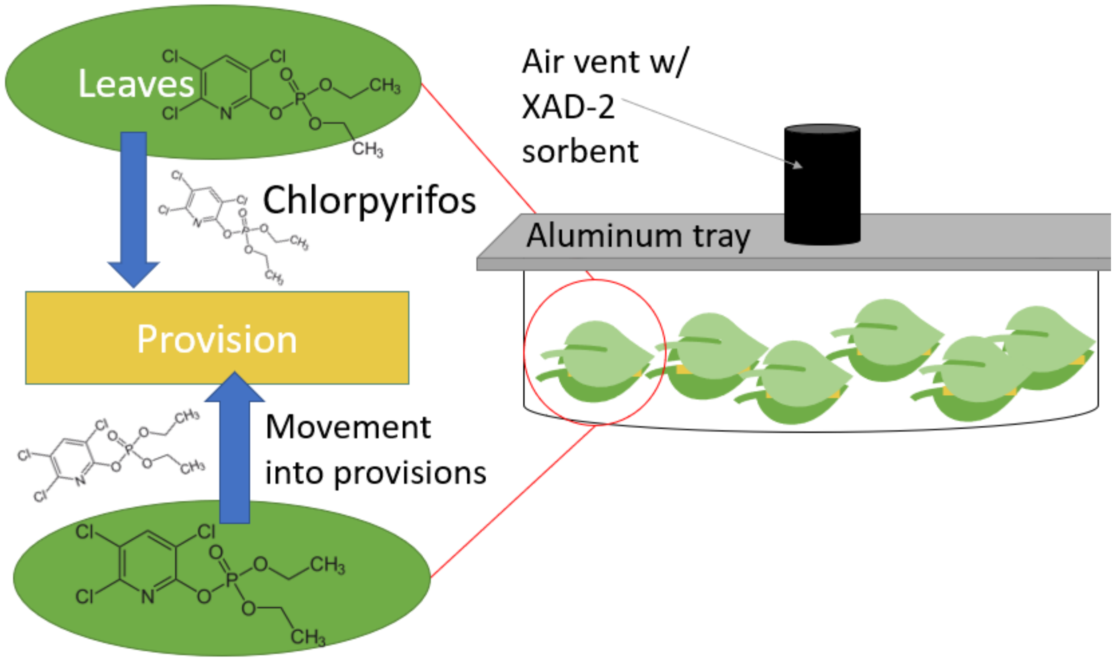
Chlorpyrifos concentration in leaves after 72 hours:

<u>19.2 µg chlorpyrifos / g leaf</u>

Use distribution ratio to estimate concentration in provision at 72 hours:

$$D_{Leaf-Provision,t} = \frac{\boldsymbol{Concentration\ in\ Leaves}}{\boldsymbol{Concentration\ in\ Provision}}\ \textbf{at time}\ \boldsymbol{t}$$

$$\boxed{D_{\text{leaf-provision,72h}} = 2.75}$$



The concentration of chlorpyrifos in provisions:

<u>7.1 µg chlorpyrifos / g provision</u>

Are chlorpyrifos levels toxic to larva?

Average mass of alfalfa leafcutting bee provision: 0.07 ± 0.02 g per provision (n=20)

Insecticide Name	Pesticide mass ingested by larva	Honeybee Pesticide Oral LD_{50}
Chlorpyrifos	**0.51 µg/larva**	Larvae: 0.46 µg[8]
λ-Cyhalothrin	**0.005 µg/larva**	*Adult: 0.909 µg[9]

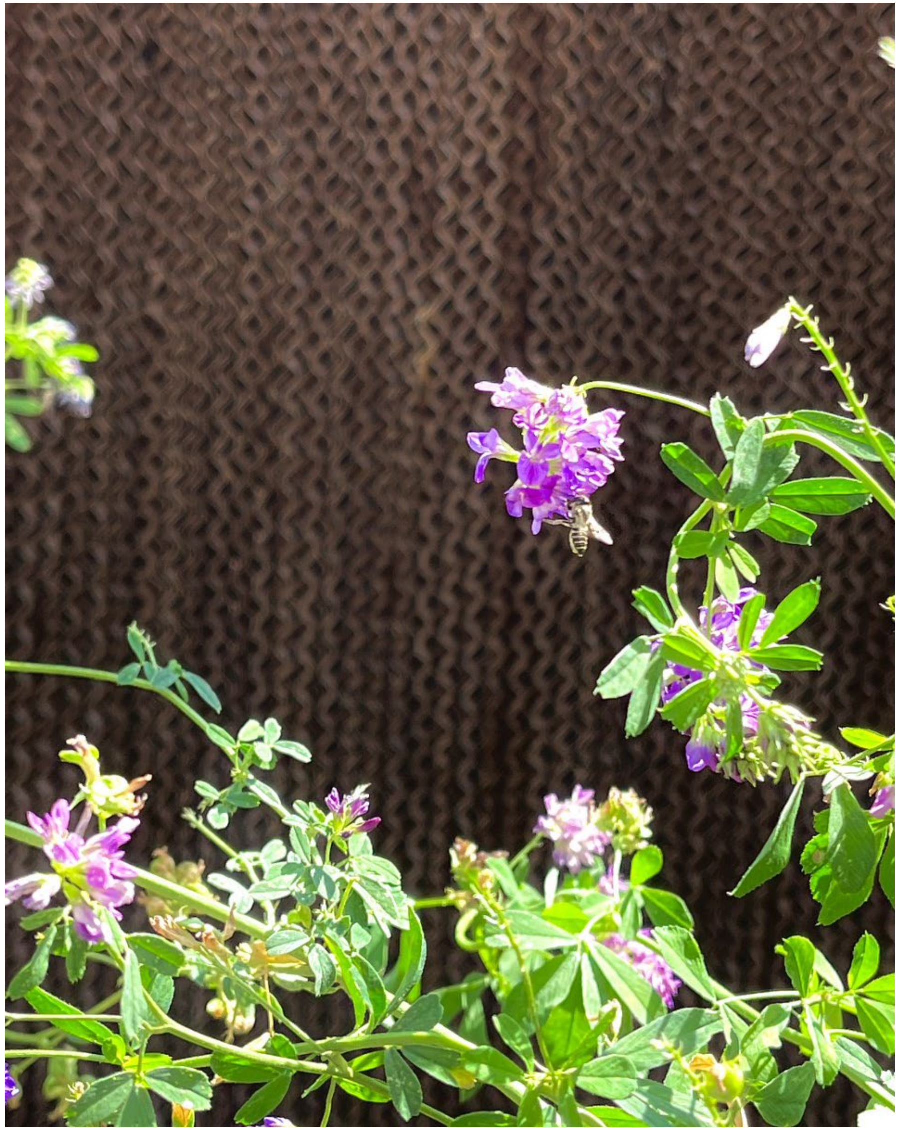
Response to grower: Yes, bee larvae can be exposed to chlorpyrifos, and the concentrations present could result in 50% of the larva population.

However, we need more toxicity data for alfalfa leafcutting bees and their larva.

[8]Dai, et al. "Acute toxicity of five pesticides to *Apis mellifera* larvae reared *in vitro.*" Pesticide Management Science. 2017
[9]https://www.mda.state.mn.us/protecting/bmps/pollinators/beetoxicity

Conclusion

- Yes, pesticides can transfer from alfalfa leaves into alfalfa leafcutting bee provisions.
- The magnitude of pesticide transfer is dependent on pesticide properties.
- Need field experiments to verify lab experiments are representative of pesticide transfer rates for alfalfa leafcutting bees.



Thank You!!!

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UtahStateUniversity

Collaborators at USDA-ARS Pollinating Insects Research Unit:

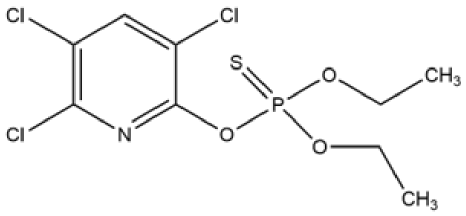
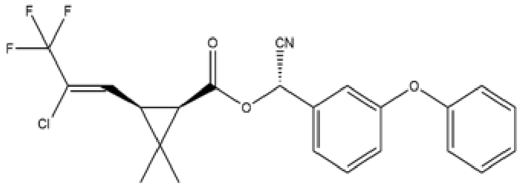
Ellen Klomps, Byron Love, and Lindsie McCabe

Lastly, pesticide formulations used in this research are NOT endorsements or recommendations

Questions?

Email: calvin.luu@usu.edu

Physicochemical Parameters

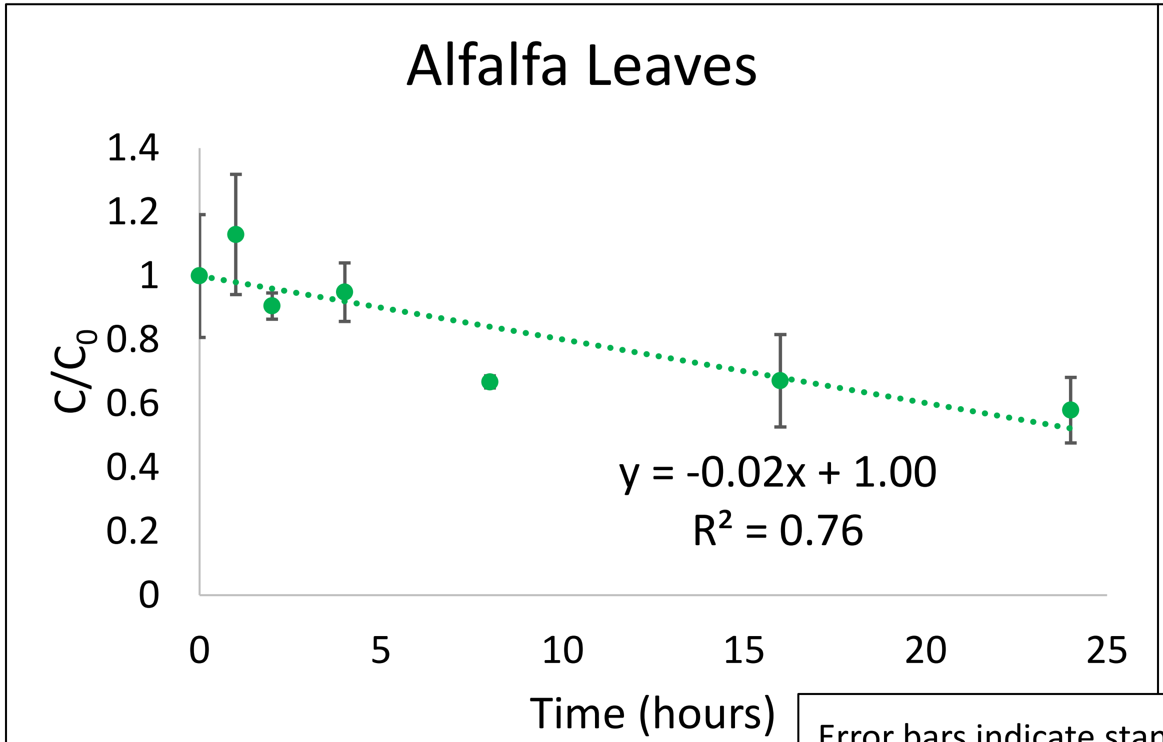
Active Ingredient	Chlorpyrifos	Lambda-cyhalothrin
Commercial Name	Chlorpyrifos 4E-AG	Warrior II
Chemical Class	Organophosphate	Pyrethroid
Structure		
Molecular Weight (g/mol)	350.6	449.86
Vapor Pressure (mmHg) @ 25˚C	2.02E-05	1.50E-09
logKow (KOWWIN) @ 25˚C	4.96	6.85
Boiling point (˚C)	379.5	436.49

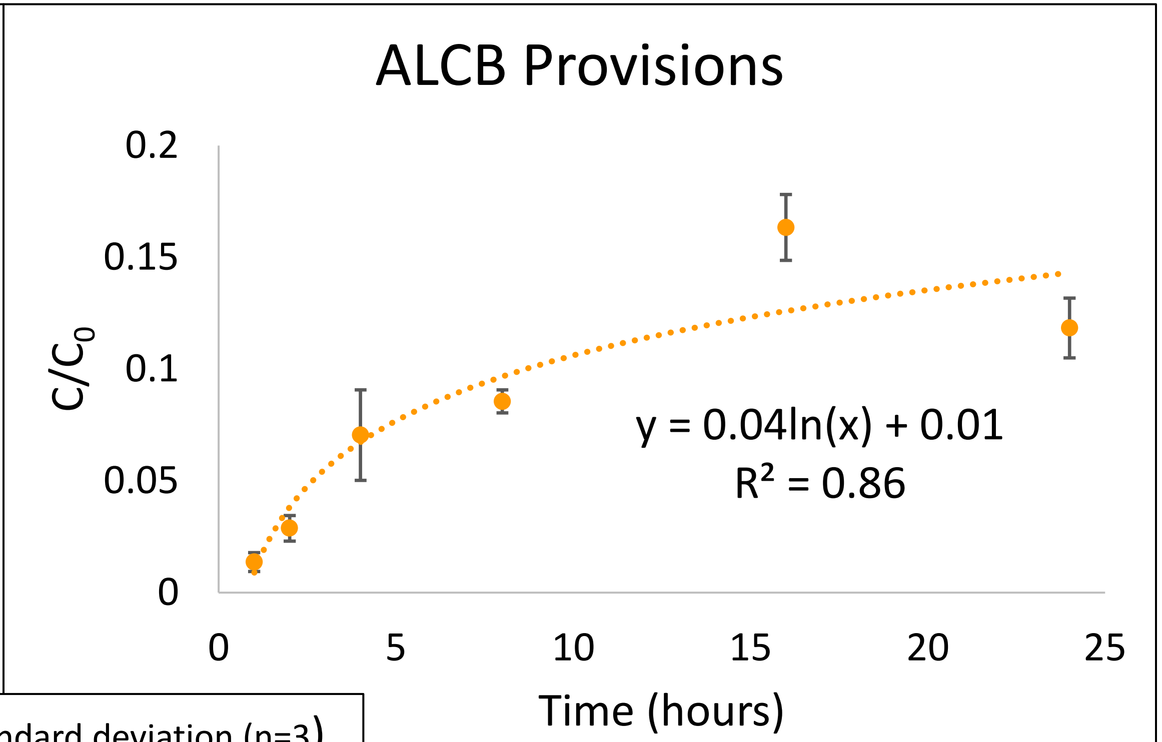
***all values are retrieved from EPIsuite, which predicts physicochemical properties from the chemical composition and structure of the compound

Chlorpyrifos 4E-AG

- Active ingredient: chlorpyrifos
- Yes, uptake by provisions occurs, reached up to 16%
- ≈40% of chlorpyrifos dissipates from leaves

GROUP 1B INSECTICIDE
Drexel
Chlorpyrifos 4E-AG
Insecticide



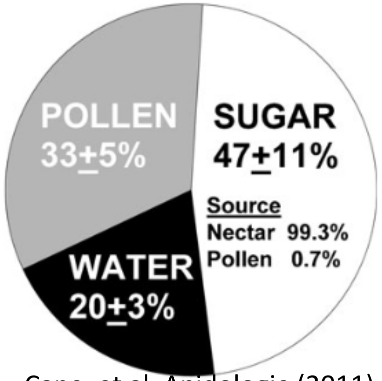


Error bars indicate standard deviation (n=3)

Warrior II®

- Active ingredient: λ-cyhalothrin
- Yes, but not as much uptake by provisions, reached up to 5.8%
  - λ-cyhalothrin is more hydrophobic than chlorpyrifos
- Negligible loss of lambda-cyhalothrin from leaves, less volatile than chlorpyrifos

GROUP 3 INSECTICIDE
Warrior II
with Zeon Technology®
syngenta



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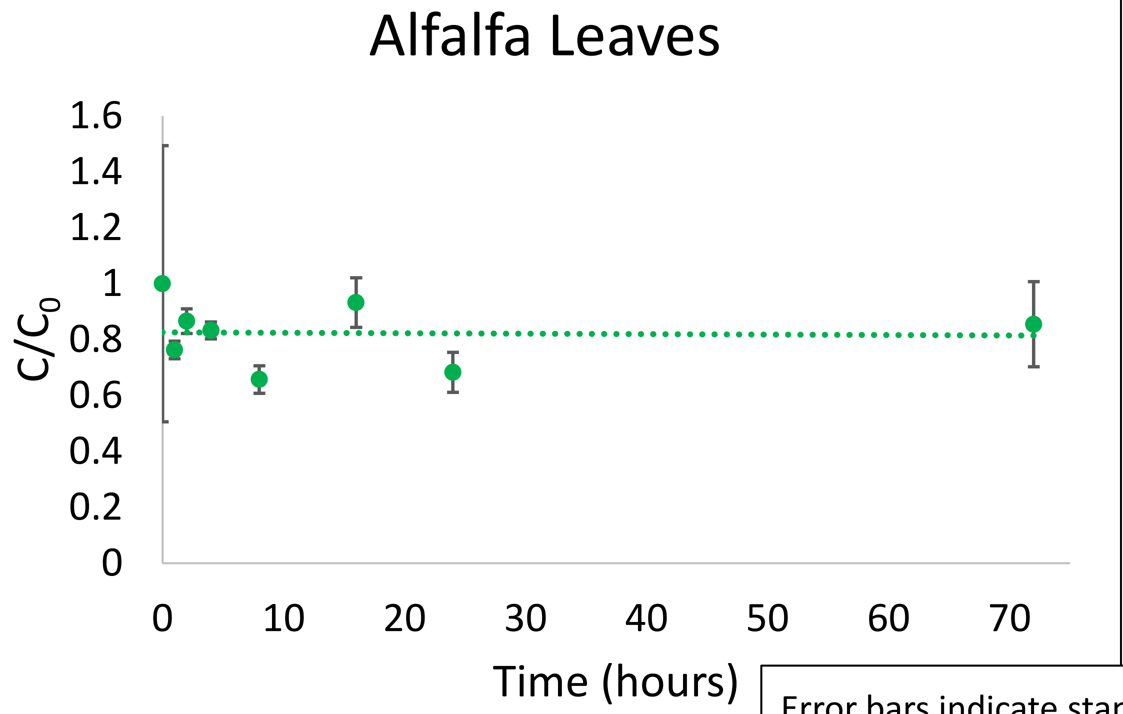

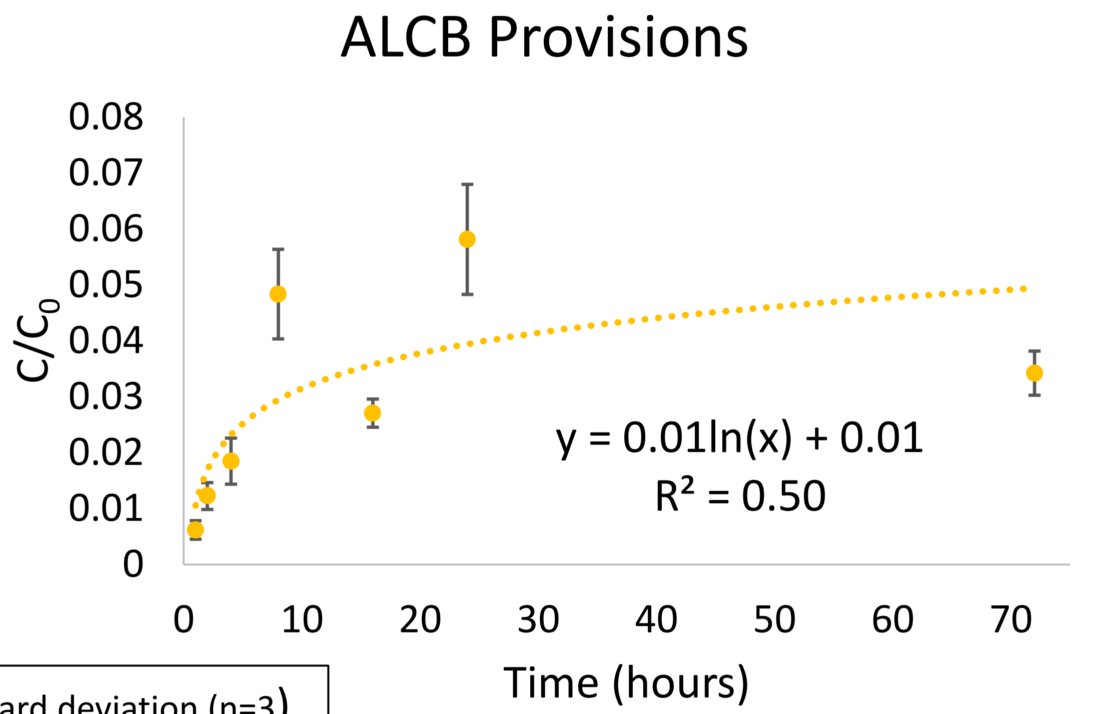

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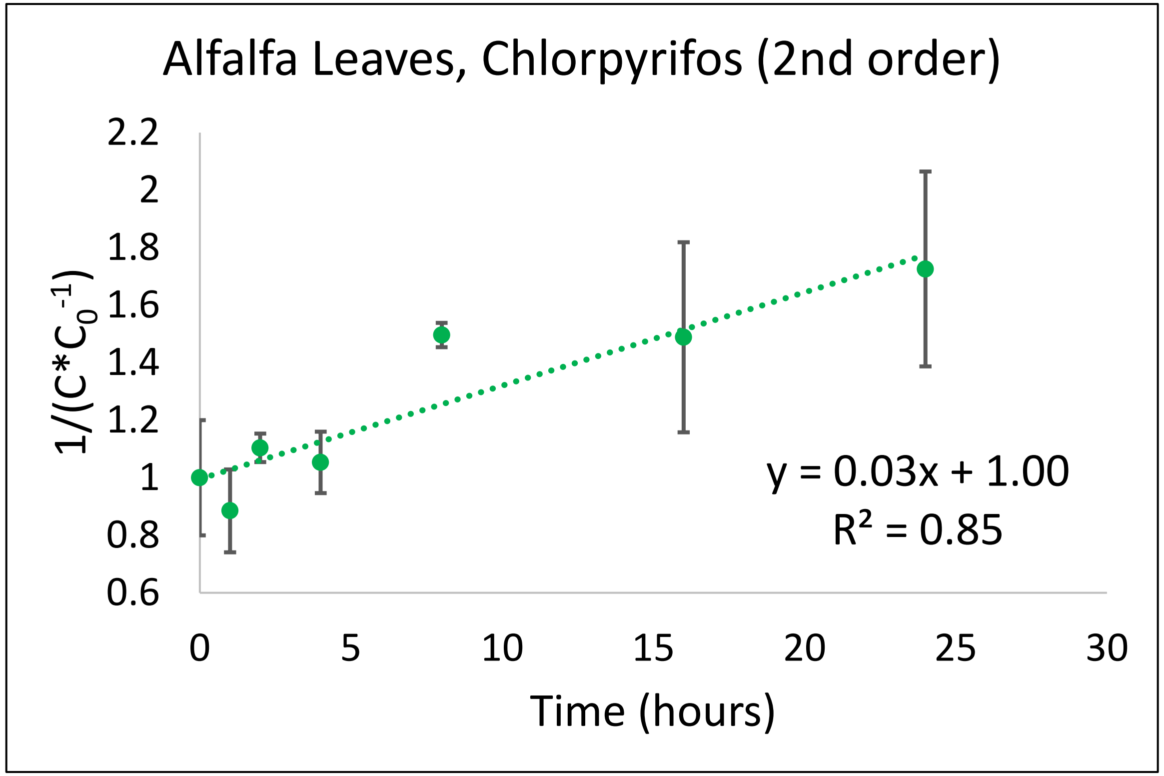
Approach 3: Calculate rate of transfer of pesticides from nesting materials into provisions

Rate Constant: Dissipation from leaves



2nd order rate of dissipation for chlorpyrifos

Lambda-cyhalothrin was indeterminate for 0th, 1st, and 2nd order

2nd order integrated rate law:

$$\frac{1}{\left(\frac{C}{C_0}\right)} = 1 + kt$$

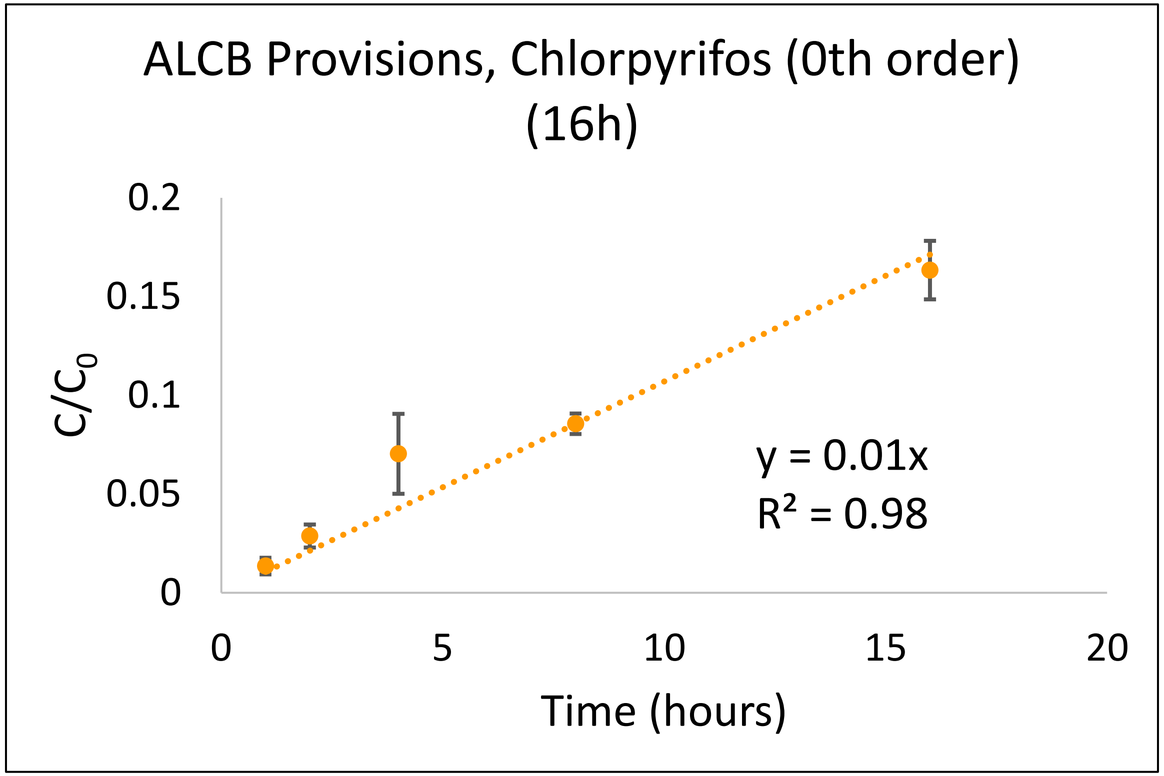
Compound	k (h^{-1})
Chlorpyrifos	0.03

Two dissipation mechanisms are occurring:

Volatilization from leaves

Uptake by provisions

Rate Constant: Uptake by Provisions



Determined to be 0th order for both chlorpyrifos and lambda-cyhalothrin

Compound	k (h^{-1})
Chlorpyrifos	0.01
λ-cyhalothrin	0.005

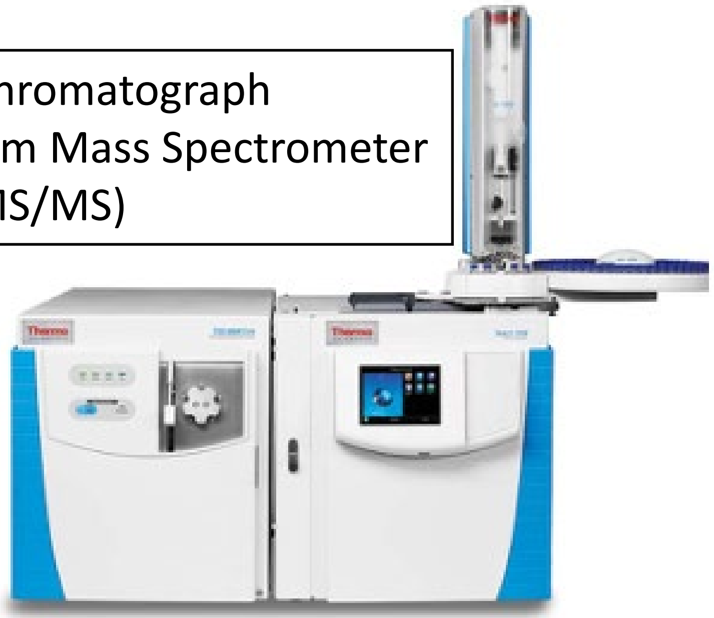
Indicates that uptake is limited by rate of absorption into provision

0th order integrated rate law:

$$\frac{C}{C_0} = kt$$

Instrumentation

Gas Chromatograph
Tandem Mass Spectrometer
(GC-MS/MS)



Energized Dispersive
Guided Extractor (EDGE)

Accelerated Solvent
Extractor (ASE) 350

Turbovap II