

Dr. Timothy Lawrence  
Dr. Allan Felsot  
Dr. Walter Sheppard



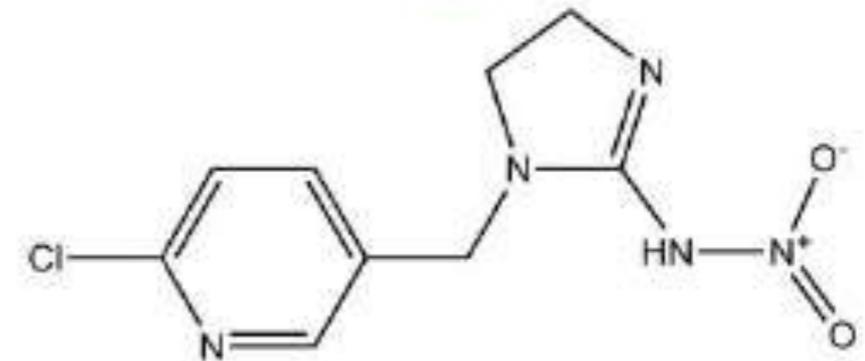
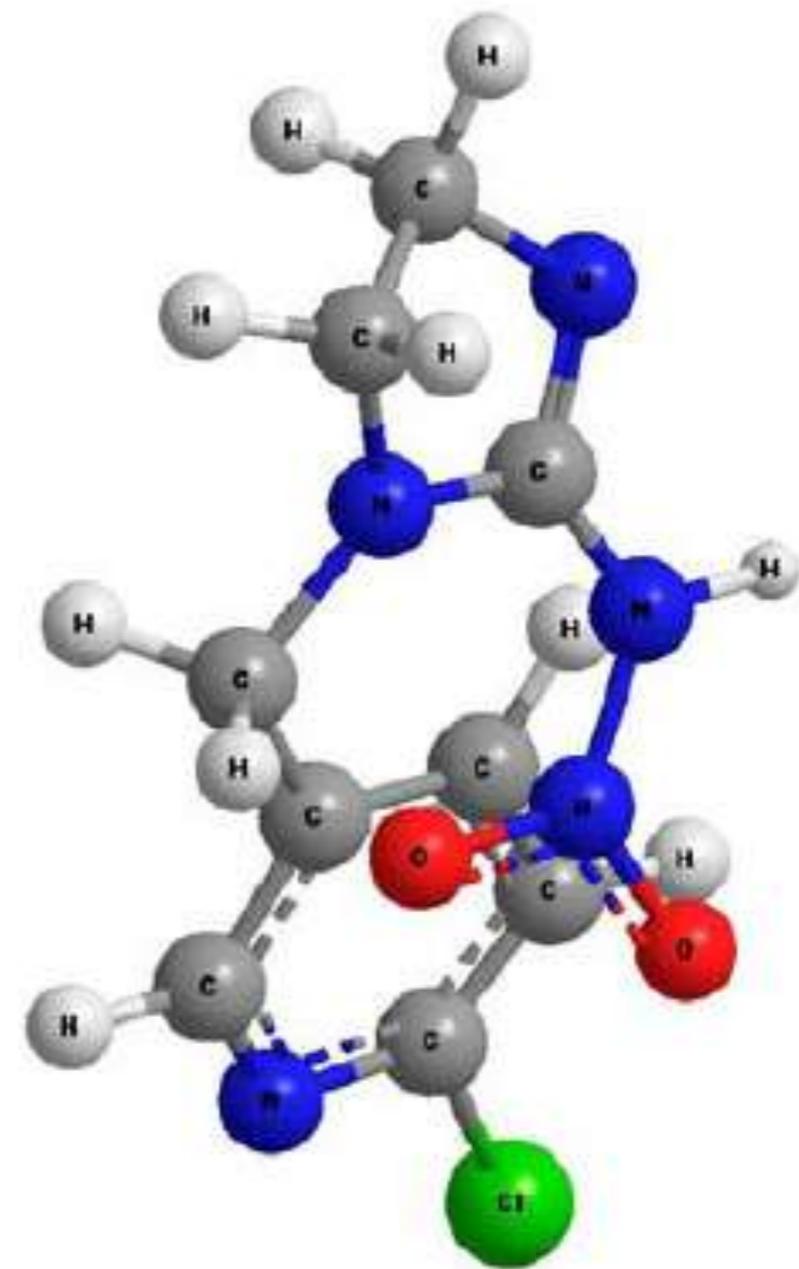
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## Field Analysis of Actual Exposure of Honey Bees to Neonicotinoids in Various Landscape Scenarios

# Neonicotinoids

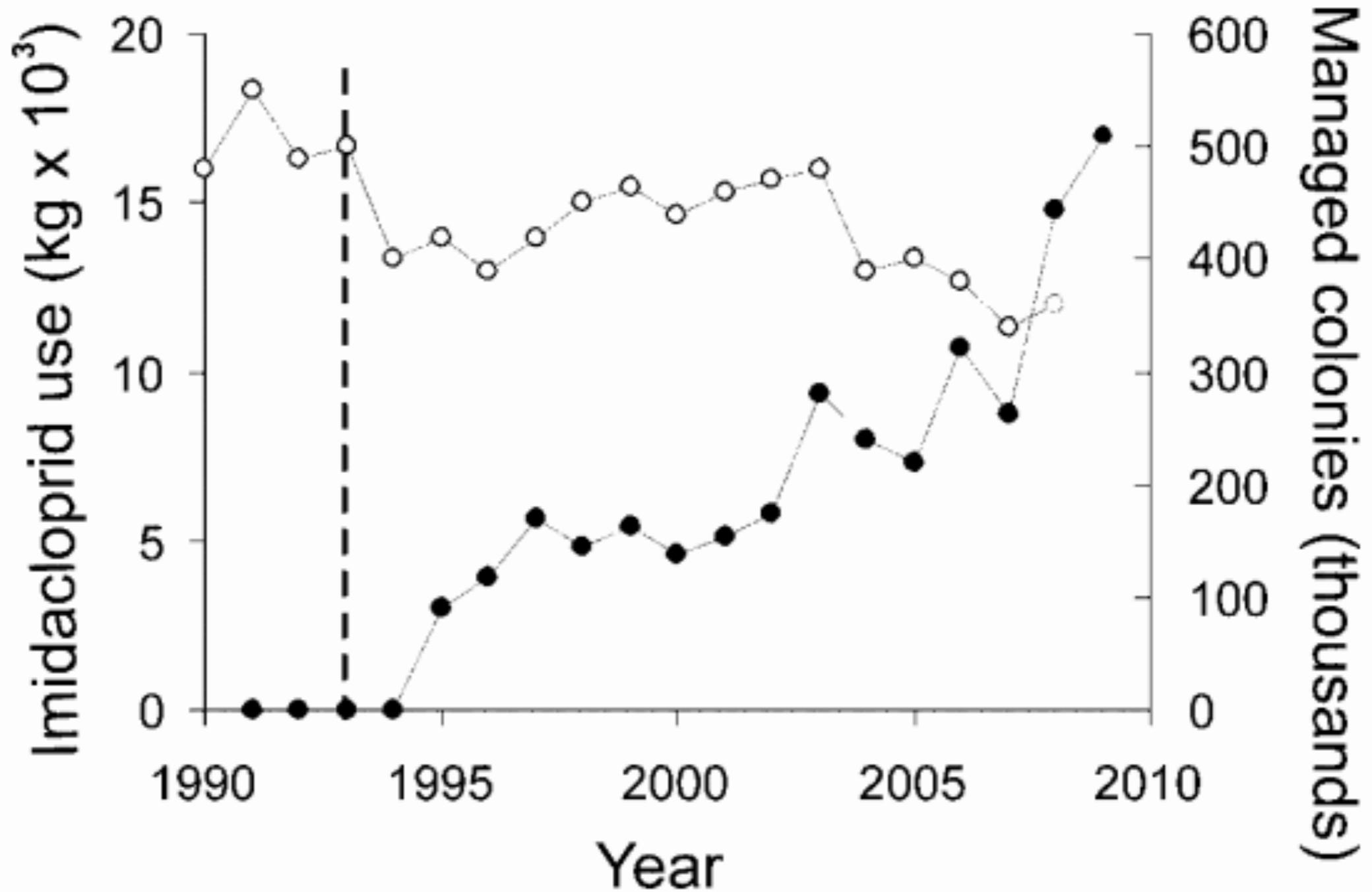
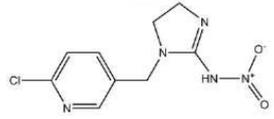
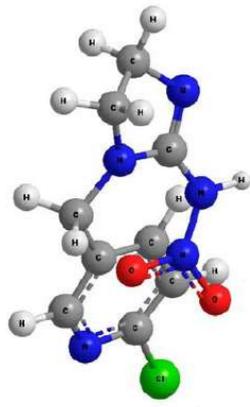
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- Imidacloprid
  - alters learning behavior
  - motor activity
  - memory
  - reduces brood production
  - foraging activity
- Clothianidin
  - impairs foraging behavior
- Thiamethoxam
  - decreases sucrose sensitivity
  - memory
- Acetamiprid
  - impairs activity
  - memory
  - sucrose sensitivity



## Sub-Lethal Effects\*

\* As low as 0.20 ppb



## Most Common Neonicotinoids

- Imidacloprid
- Clothianidin
- Thiamethoxam
- Thiacloprid

# Public Reaction

- Portland, Spokane, Seattle, Eugene & Other Jurisdictions Banned the Use of Neonicotinoids
- Olympia Beekeepers petition the Thurston County Commissioners to Request the WSDA Limit the Use of Neonicotinoid Pesticides to Only Certified Pesticide Applicators
- WSDA Rejected the Request Due to Lack of Evidence



# Honey Bee Survey and Risk Assessment

## Funded by:

- Washington State Apiary Advisory Board
- Washington State Commission On Pesticide Registration
- Washington Department of Agriculture

- Context:  
Regulatory actions by local government entities
- Hypothesis:  
Bees from apiaries in non-agricultural landscapes are exposed to the most toxic neonicotinoid insecticides
- Predictions:  
Analysis of bee bread as a surrogate for exposure in different landscapes would yield detentions of most toxic neonics

# Site Selection

- Washington Apiaries Managed by Hobbyist, Sideliner, and Commercial beekeepers were selected to give broad geographic distribution representing urban, rural, and agricultural operations.
- Washington State Beekeepers were selected from a combined list of members of local beekeeping organizations.
- A total of 92 cooperating beekeepers (44 in 2013 and 48 in 2014) allowed collection of hive samples from 149 apiaries that are representative of bee forage areas throughout the state .

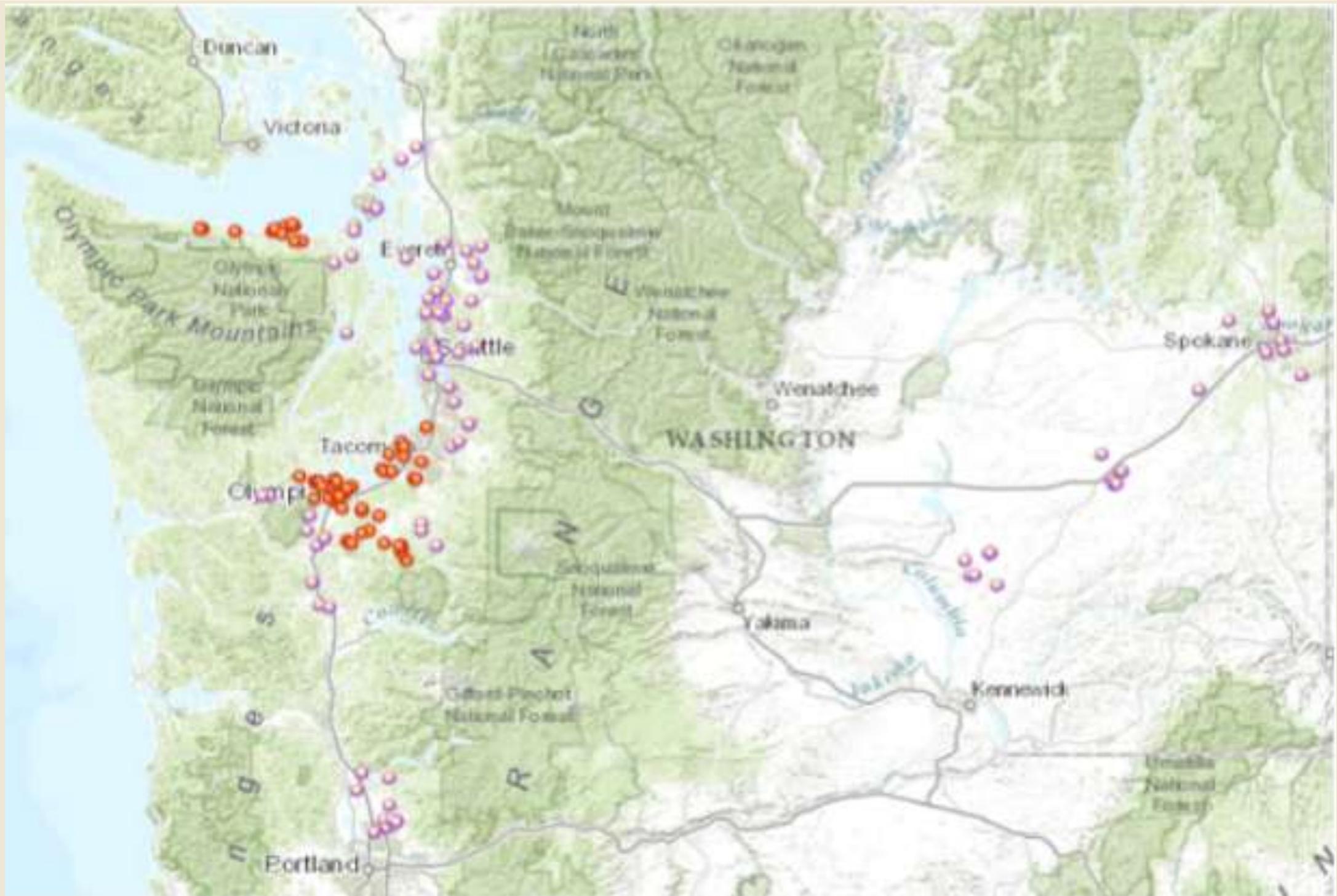


Figure 1. 2013 (orange) and 2014 (lavender) field sampling locations

## Protocol for Sampling Hives Per Apiary

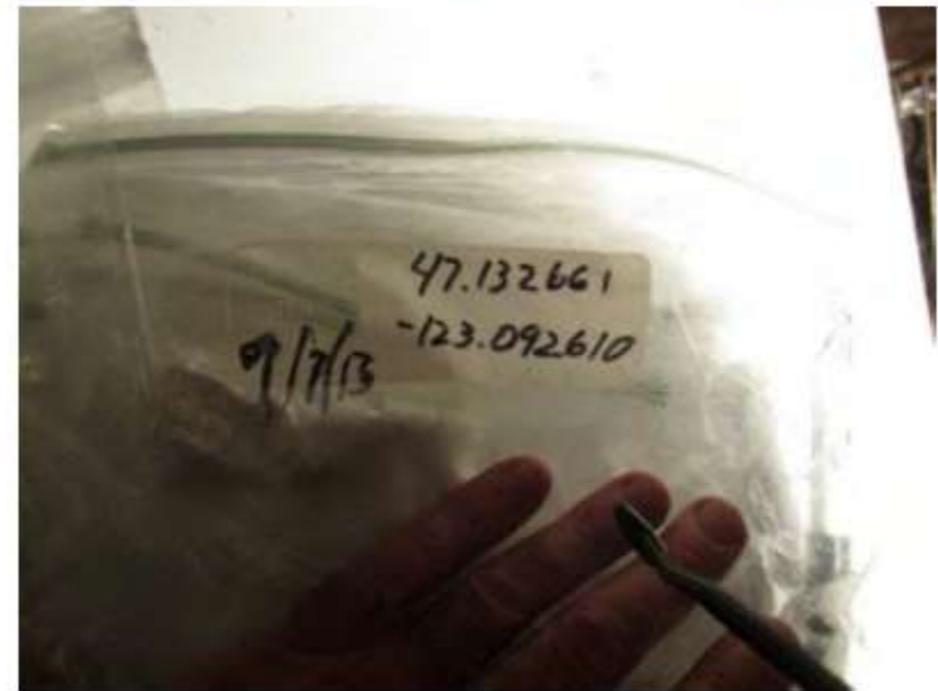
Number of Colonies	Number of Hives for Sample Collection
<3 Colonies	All Hives
3-10	2
11-20	3
>20	4

- Samples were comprised of fresh pollen
  - ✓ Avoid entombed pollen (covered by propolis)

## Comb Wax & Bee Bread Collection

- Locate the upper most super (bee box) that contains brood
- Starting from the second frame in (from either side) remove the frame and work toward the center of the brood nest until find an area with at least 2 square inches of stored pollen
- Place the frame on plywood sheet for support
- Use a two inch hole saw with a  $\frac{1}{4}$  inch pilot shaft attached to a portable drill cut a hole in the frame through the midrib
  - ✓ Use metal rod to push sample out of hole saw and into sealable plastic baggie and seal
  - ✓ Mark outside of bag with sample number
  - ✓ Place all samples from each apiary into a large sealable baggie and place in a cooler containing a sufficient number of blue ice packs to keep the samples cool
- Use fresh hole saw for each colony sampled
  - ✓ Rinse all hole saws used in the apiary with distilled water to remove any residual pollen, wax, and debris. Rinse cleaned hole saws with methanol and place in clean bag.





## Wax vs. Bee Bread

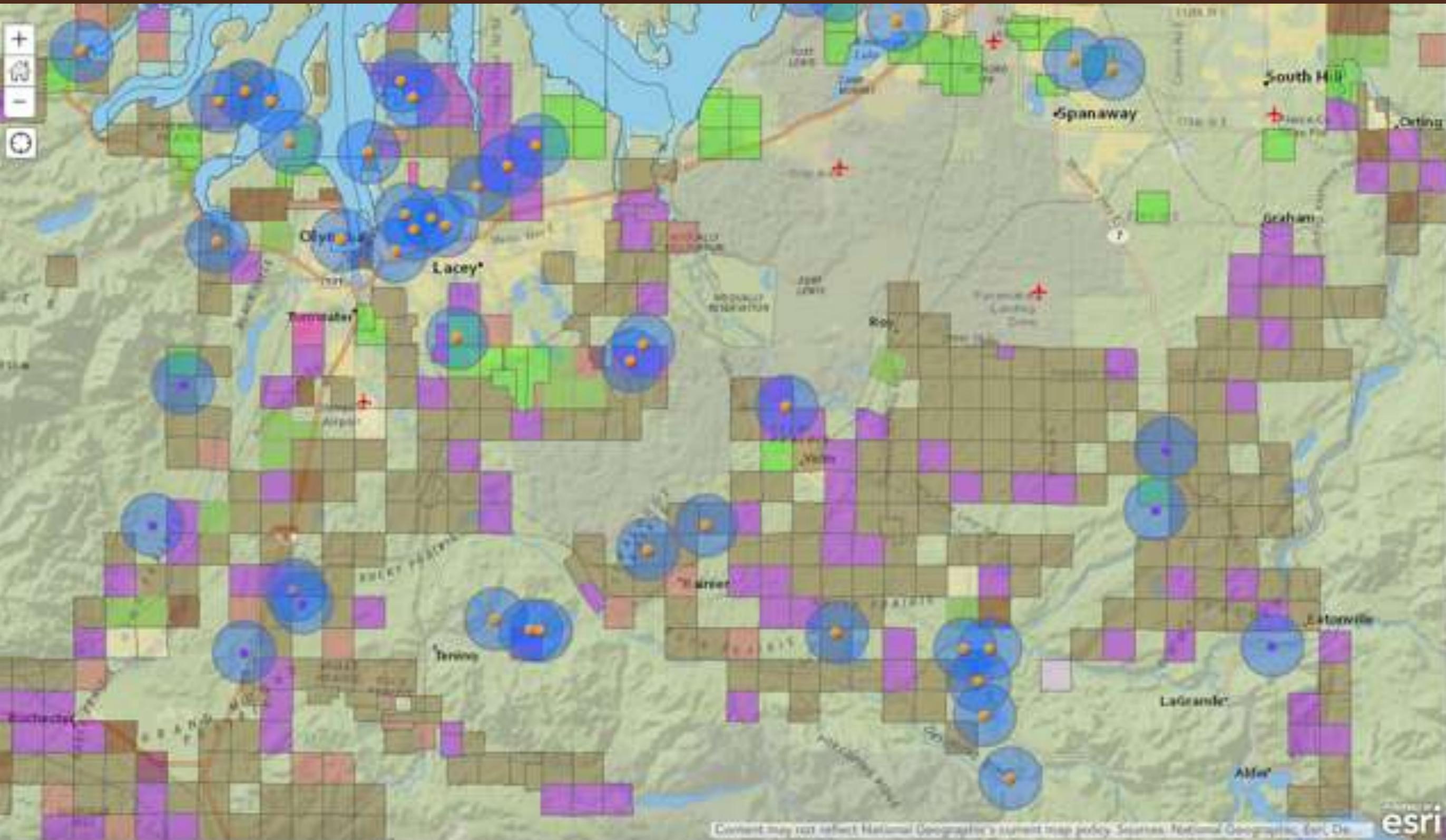


# Analytical Method

- Modification of Bayer Crop Sciences enforcement method for Imidacloprid to include dinotefuran, clothianidin and thiamethoxam Imidacloprid
- Homogenize 3-gram sample with 75% MeOH, filter, and bring to 60 mL volume
- Evaporate methanol from 30 mL of extract then clean aqueous remainder on buffered diatomaceous earth
- Second cleanup on Sep-Pak silica gel SPE with acetonitrile elution of analyses



Liquid chromatography - mass spectrometry

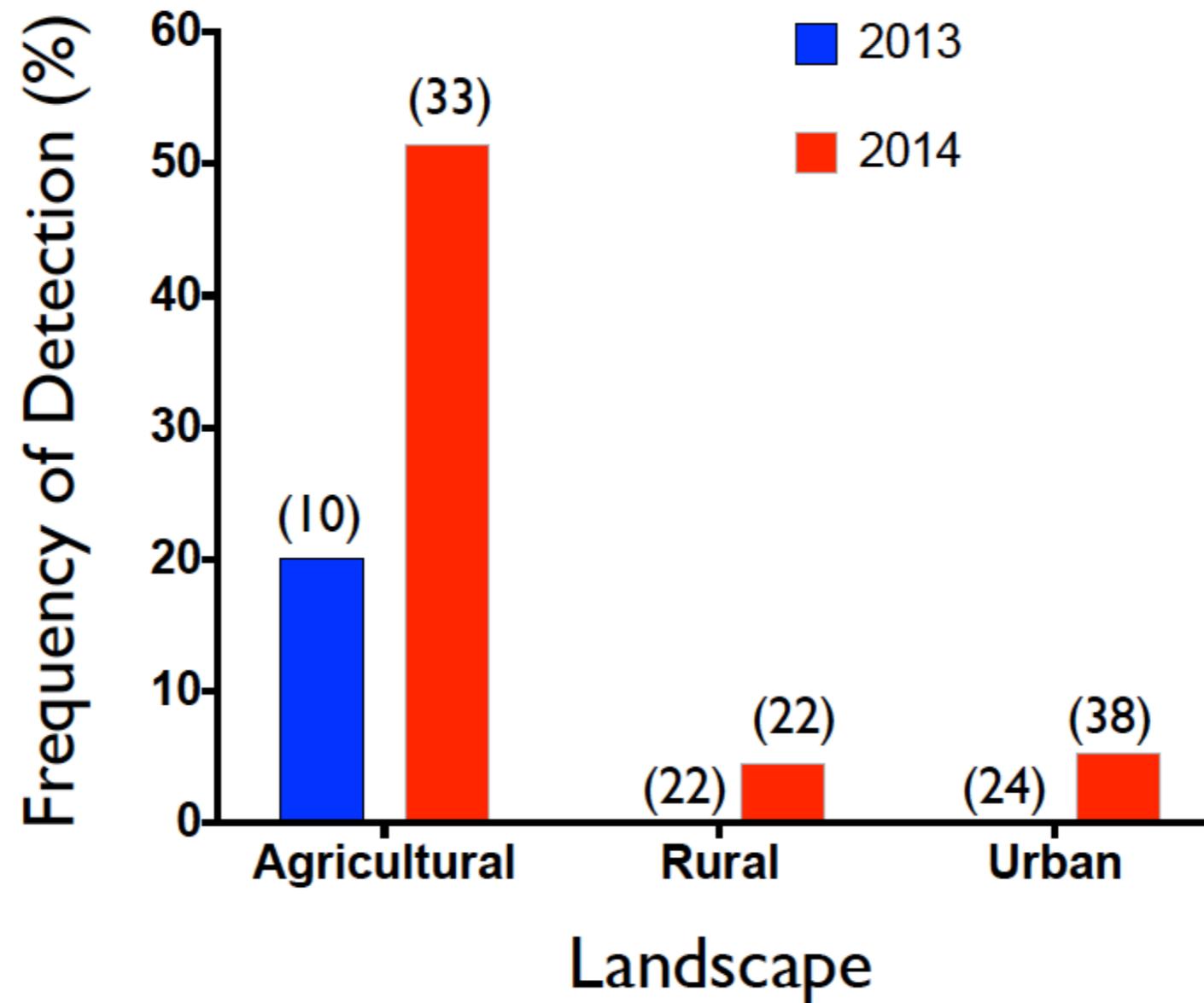


## Concentrations of Neonicotinoid Insecticides Found in Bee Bread and Wax Samples

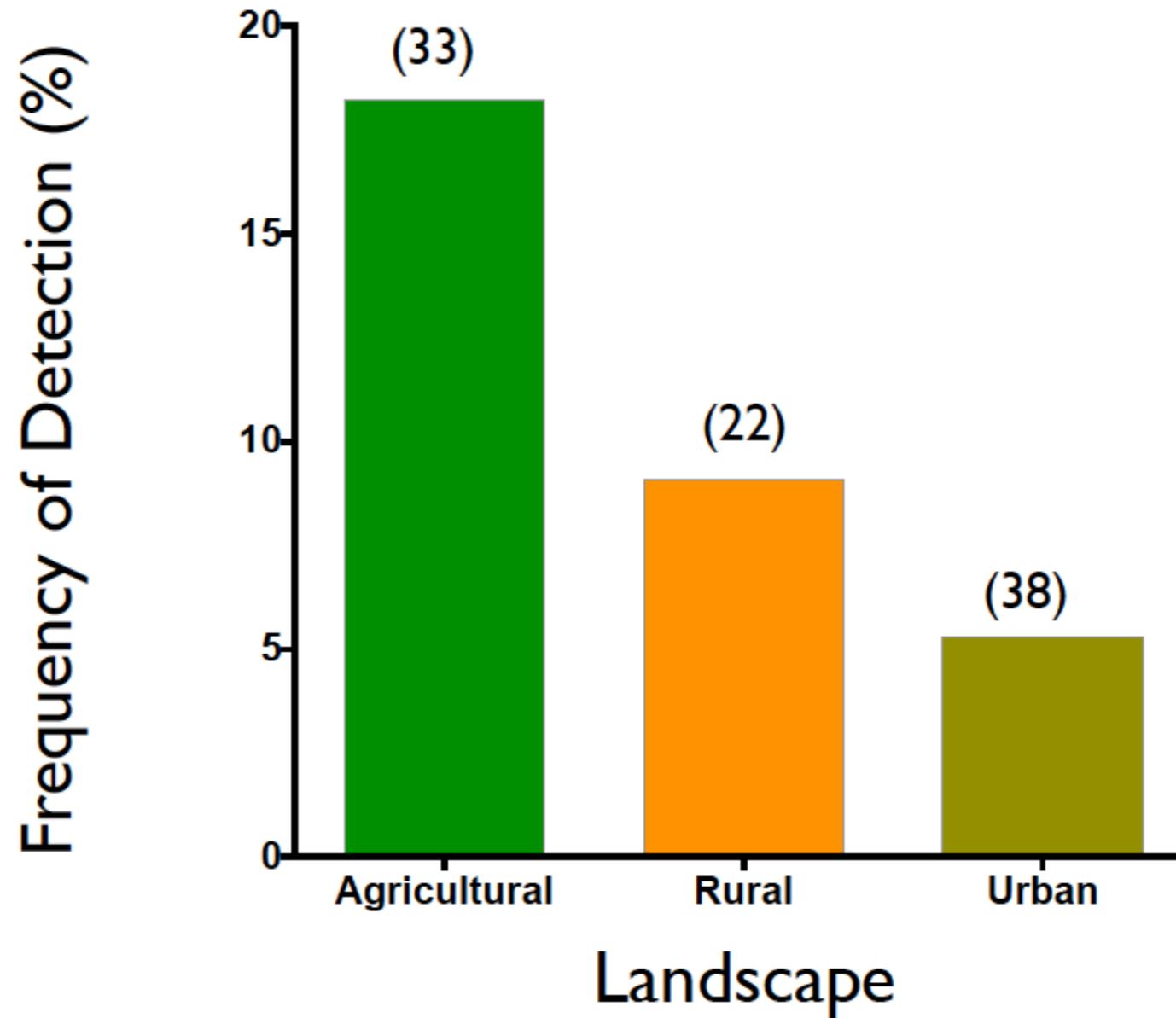
Mean  $\pm$  Standard Deviation (ppb)

Matrix	Dinotefuran	Imidacloprid	Thiamethoxam	Clothianidin
<b>Wax</b>	<b>1.94</b>	<b>2.85 <math>\pm</math> 1.04</b>	<b>1.06 <math>\pm</math> 0.38</b>	<b>1.52 <math>\pm</math> 0.94</b>
<b># samples</b>	<b>1</b>	<b>2</b>	<b>5</b>	<b>2</b>
<b>Bee Bread</b>	<b>1.56</b>	<b>2.68 <math>\pm</math> 1.09</b>	<b>1.44 <math>\pm</math> 0.97</b>	<b>0.99 <math>\pm</math> 0.43</b>
<b># samples</b>	<b>1</b>	<b>4</b>	<b>7</b>	<b>14</b>

Detection of Neonicotinoid Insecticides in Bee Bread Samples Collected from Apiaries in Different Landscapes of WA State During 2013 and 2014

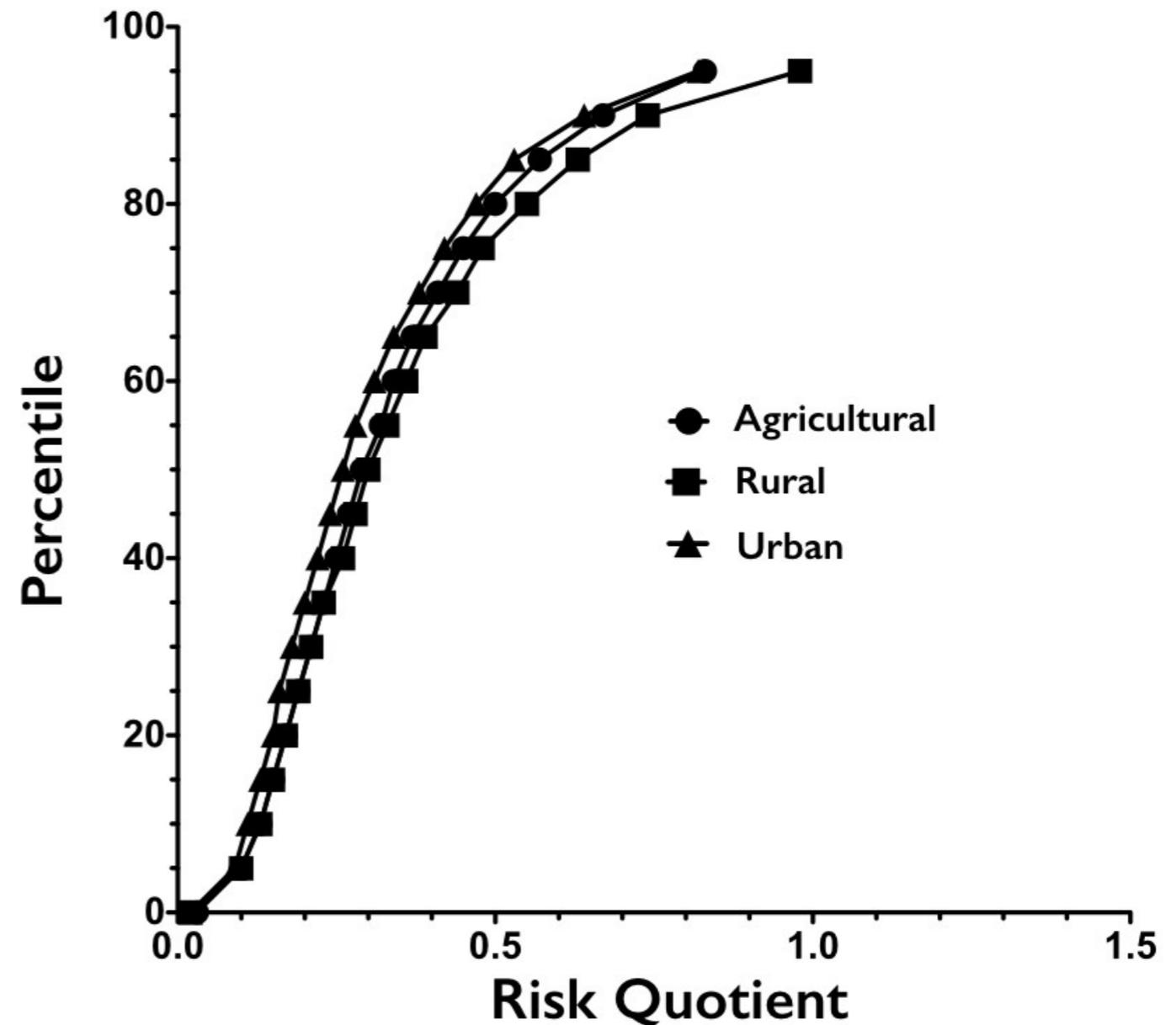


Detection of Neonicotinoid Insecticides in Bee Wax Samples Collected from Apiaries in Different Landscapes of WA State During 2014



# Neonicotinoids

- Imidacloprid
- Thiamethoxam
- Clothianidin
- Dintefuran
- Our assessment found no risk to honey bees for presence of trace dietary neonicotinoids in urban and non-agricultural rural areas; There is a very low risk to bees in agricultural areas of Washington State.
- Our NOAEC - 5 ppb
- EPA recommended NOAEC 25 ppb



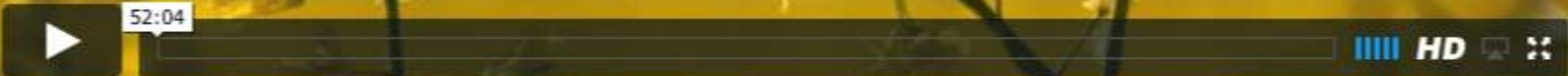
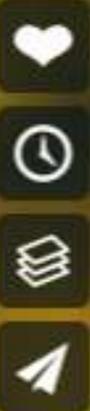
# Conclusions and Recommendation

- Insufficient Justification to Further Restrict the use of Neonicotinoids in the State of Washington
- Focus on Education of Growers and Pesticide Applicators on the Use of Neonicotinoids to Minimize the Impact on Pollinators
- Focus on Habitat Enhancement and Pollinator Friendly AgroEco Systems



<https://vimeo.com/146957716>

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## Pollination and Protecting Pollinators

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**Thanks!**  
**Questions?**

