ARDP PI Meeting Notes
10/15/14

Participants: Peter Ellsworth, John Palumbo (via conference, first 30 minutes), Nilima Prabhaker, Steve Castle, Yves Carrière, Wayne Dixon, Al Fournier, Bob Nichols, Ryan Kurtz (via conference)

Project Overview (Peter):

Can we predict resistance in close-to-real time? Yves published work on pyriproxyfen that related use pattern to whitefly (WF) resistance within a 3km radius (roughly 9 section grid). The APMC tracks pesticide use data on a section-level basis in cooperation with Arizona Department of Agriculture (ADA). We invest a lot of time reviewing and correcting records as they come into the database. In this project: We will develop chemical use maps for key WF chemistries, across all crops, on a regional basis. Maps will be used in outreach to help PCAs decide on chemical use based on last year's use intensity at the section level. Table 1 in the grant lists target chemistries for this project. (The table lists only 1 diamide, the one that is active against WFs. There are 2 other diamide AIs that are currently registered in alfalfa, cotton, and vegetables). One selling point for this project is that regardless of hypothesis testing, a major outcome of this project is simply teaching (and measuring knowledge and implementation of) basic principals of resistance management.

Project Objectives:
1. Develop chemical use maps and conduct outreach (Wayne, Al, Lydia, Peter and John)
2. Test resistance hypotheses (Steve, Nilima, Yves and Ben)
3. Measure changes in awareness, knowledge, skills behaviors and conditions (Al with Peter, John and others)

Timeline:
- 2013 data will serve as baseline data and needs to be cleaned by Wayne and Al (May 1)
- 2014 data cleaned and draft maps produced (May 1)
- Maps reviewed by John/Peter to verify patterns (June)
- Maps reviewed by Yves, Steve, Nilima, Peter to determine field choices for resistance monitoring during summer (June)
- Post maps online (by June 30)
- Outreach (ongoing, starts immediately) includes various annual workshops and events
- PI planning meeting (Today, ahead of schedule)
- Sampling and resistance monitoring (May – October 2015, 2016; but probably best last two weeks in July for Yuma, Imperial, 1 Aug for La Paz, Riverside Co. and 1st two weeks of August for central Arizona)
- Data analysis (Oct – Dec 2015, 2016)
- Collect data on awareness, knowledge and intention to adopt chemical use maps at Extension meetings (throughout project, with baseline data in fall 2014, spring 2015)
- Develop and deploy online survey, distribute through Extension email lists, newsletters, etc. (deploy by Jan 2015, throughout project)

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- Develop survey questions and deploy with mapping website (June 30)
- Some Y2 activities will likely be performed under a short term no cost extension
- Analyze Crop Pest Losses Data and APMC database to develop data on changes in use patterns and group adoption (Jan 2016 for Y1, Jan 2017 for Y2)

Introductions / Roles:
- **Yves Carrière**, UA Entomology – will assess the scale that is relevant for resistance to evolve (expects it to be larger than 3km and that it will vary between regions / counties). This is a very ambitious goal.
- **Bob Nichols,** Cotton Inc. – Here to advise and provide support for these efforts
- **Nilima Prabhaker,** UC Riverside – Resistance monitoring research, lab assays
- **Steve Castle,** USDA-ARS – will work with Nilima on field work and coordinate with Yves to guide locations for WF sampling
- **Wayne Dixon**, UA Assistant in Extension for IPM Assessment – will work on pesticide use data correction, development of maps, software and websites
- **Al Fournier**, UA IPM Evaluation Specialist – will develop and deploy evaluation instruments and analyze data, in collaboration with Peter, John, Wayne and Lydia
- **John Palumbo**, UA Vegetable IPM Specialist – Will conduct outreach for vegetable IPM, IRM and work with the team to accomplish goals
- **Ryan Kurtz** – entomologist with Cotton Inc. Happy to review anything and work with the team on this project
- **Lyida Brown** (not present, introduced by Peter)**,** UA Assistant in Extension for Agronomic Crops IPM – involved in supporting outreach for this project and also involved in the Crop Pest Losses Signature Program of the WIPMC. She will help with writing outreach publications.
- **Peter Ellsworth**, UA Cotton IPM Specialist and Project Director – will oversee the project, conduct outreach and work with Al on evaluation components

Budget: Due to last minute administrative challenges, Peter and John's budgets were cut significantly with almost no operation funds. Nearly all UA budget is in salary. Subcontracts to UCR and USDA were included to handle resistance work.

Monsanto Insect Management Knowledge Research Program. Started 3 years ago. The first 2 years were dedicated to corn rootworm management. Peter first saw info on this year's program in Western Farm Press. Up to $250k per year for up to 3 years. No IDC stipulation, so it would be subjected to full 51.5% IDC. The extended deadline for the pre-proposal was this past Monday. Peter drafted and submitted a pre-proposal on behalf of the group. If invited to submit a full proposal (Nov 10), it will be due Jan 5 and funds available March 2015. This will mean having a proposal and all budgets in place and submitted to UA by mid-December.

One possibility is to extend this project to CA data (Blythe and Imperial Valley areas). Eric Natwick and Vonny Barlow may be added to the proposal. Incorporate GIS approaches? Game-training simulation similar to Lygus RAMP project? How is this related to CI-supported resistance monitoring in AZ? These are all ideas that Peter brainstormed. Yves: Monsanto's

interest is in understanding the scale of resistance, and only we have the data to do this. This is a compelling selling point. Another selling point is linking this project to Bt and refugia (as a model for future work). Bob: Monsanto will also need this type of approach for Lygus.

Reviewer Comments on ARDP Proposal

One criticism was the one-year time lag between data used to predict the potential development of resistance in the following year. This introduces a bias because Y1 data will affect Y2 behavior. Our objective 3, evaluation, is important because it will measure how people were affected by the outreach.

Bob made the point that outreach (obj 1) will be occurring before we know the scale of resistance, which seems backwards. We have interlocking objectives: it will be important for everyone to meet their timelines.

Table 1. *Major chemistries used to control whiteflies in the eco-region of AZ, CA, and northern Mexico in multiple crops and target life stages for resistance monitoring.*

Common Name	[1]IRAC Number	Chemical Group	Whitefly Crop Hosts Targeted	Resistance Status	Assayed Life Stage(s)
synergized pyrethroids	1B + 3A	Pyrethroids + Organophosphates	All	Resistance detected	Adults
acetamiprid	4A	Neonicotinoid	Cotton, Melons, Vegetables	Field complaints	Adults
clothianidin	4A	Neonicotinoid	Cotton, Vegetables	–	
dinotefuran	4A	Neonicotinoid	Cotton, Melons, Vegetables	–	
imidacloprid	4A	Neonicotinoid	Cotton, Melons, Vegetables	Resistance detected	Adults
thiamethoxam	4A	Neonicotinoid	Cotton, Melons, Vegetables	–	
pyriproxyfen	7C	Juvenoid	Brassicas, Cotton, Melons	Resistance detected	Eggs
buprofezin	16	Chitin inhibitor	Cotton, Melons, Vegetables	None	Nymphs
spiromesifen	23	Lipid synthesis inhibitor	Cotton, Melons, Vegetables	None	Nymphs
spirotetramat	23	Lipid synthesis inhibitor	Vegetables	None	
cyantraniliprole	28	Diamides	Melons, Vegetables	None	Nymphs

Review of Table 1: WF Chemistry to Examine
- We will produce maps for all key modes of action.
- How much of this should we do resistance monitoring for?
    - Pyriproxyfen – must do. It is more time consuming and challenging to achieve in the lab
    - Acetamiprid – must do. Imidacloprid possibly.
    - Synergized pyrethroids – should do (important for brown stink bug and Bagrada control)
- These 3 choices give a good variation in resistance levels for Yves to work with, providing 3 independent estimates of the scale. They all get used quite a bit, although there is variation in use throughout the state.

- Neonicotinoids are challenging in that different AIs show different results.
- Diamides are also important. There seems to be some resistance developing in the southern U.S.
- Buprofezin – the assays are challenging, but Nilima thinks it could be included.
- It may be better to have fewer chemistries but have more samples collected in more sites

Chemical use maps from 2014 will be provided to Yves, Nilima and Steve in May/June to help them select WF sampling sites. At the same time, John and Peter will review maps for accuracy before they are finalized for PCAs and posted online. We will want to sample areas with a lot of chemical use and little or no use as well as something in between for comparisons.

When should samples be taken? An important question. If you start measuring from May through December you introduce a lot of within-year variation. It would be better to pick a narrow window when you can collect all your samples. In August, could collect in all crops, but the problem is that some areas will have been sprayed in that year. If you sample later (e.g., Aug), after applications have occurred, then you could link within-year data to the analysis. The problem is we won't have the 1080 data ready until the following year. This could only be done retrospectively. If you try to collect earlier (e.g., May), you will not get enough populations of WF. If you did May melons, you would get info about neonics, and it would be subjected to sprays during the sampling year. If you sample in August, you have already had some cotton treated and some not treated. 12 months of selection pressure is going on. Pesticide use is almost continuous, but use data is not available in real time. Therefore, sampling is happening with only partial knowledge of what insecticides have been used. For each location where you sample, you could (in the next year) include the sample year data as a covariate in the analysis. Populations will be mostly available in August. Depending on where you sample, and which crop, sprays may have already occurred, or not. Wayne: Could you write an algorithm for each section where sampling occurs that tells you what chemicals were used for the past 12 months, 18 month, etc.? It seems possible.

We are interested in knowing if last year's use can predict this year's resistance. This is the premise. This assumes that what we see this year in a location is similar to what we see next year. (Assumes no grower change in behavior.) The smaller the scale, the larger the correlation will be. The analytical problem is that we are asking growers to change their behavior based on last year, if they do, they diminish the effect we are trying to measure: the predictability of resistance.

Steve & Nilima: How many populations by dose per chemical can you estimate per year? It depends on whether there is a discriminating dose versus LC50 approach. If you know a discriminating dose, you can process many more samples. Fewer for pyriproxyfen, because processing time is slower (8-14 days) than for other chemistries (about 2 days). Steve: normally we would concentrate on a region, e.g., La Paz, collect 10 different field sites (populations). It would be good to have at least 2 regions, better 3, to generalize. In each of 3 regions, sample high use, moderate use and low use sites (based on last year's maps). One problem is that you might not have comparable use of particular pesticides across regions. Could you do 18 populations over a 30-day period (mid-July to mid-Aug)? It depends upon how long the time

frame is for sampling. Steve is not sure about pyriproxyfen in this time frame, but for the other 2, no problem.

- 3 regions: La Paz, Yuma, Central AZ, 18 populations each (low, medium, high), 3 chemistries
- If we get additional funding, we could expand to Blythe and Imperial
- Late July: Yuma, Imperial
- 1 Aug: Parker / Blythe
- 15 Aug: Central AZ

See photo #1 & below showing no. of populations per chemistry (PI = Pesticide Intensity)

Timing	Location	PI = Low	PI = Medium	PI = High
15 July	Yuma	3	3	3
15 July (Monsanto)	Imperial	3	3	3
1 August	Parker	3	3	3
1 August (Monsanto)	Blythe	3	3	3
15 August	Central AZ	6	6	6

Bob advocates using a 3-dose approach for resistance testing. It provides more information than a single dose approach. For pyriproxyfen, 0.1 dose looks great and clearly tells a story (Peter's PPT chart; a version shown below).


Is there a relationship to intensity of use?

Corrected mortality for pyriproxyfen (0.1µg/ml)   *Ellsworth/UA*
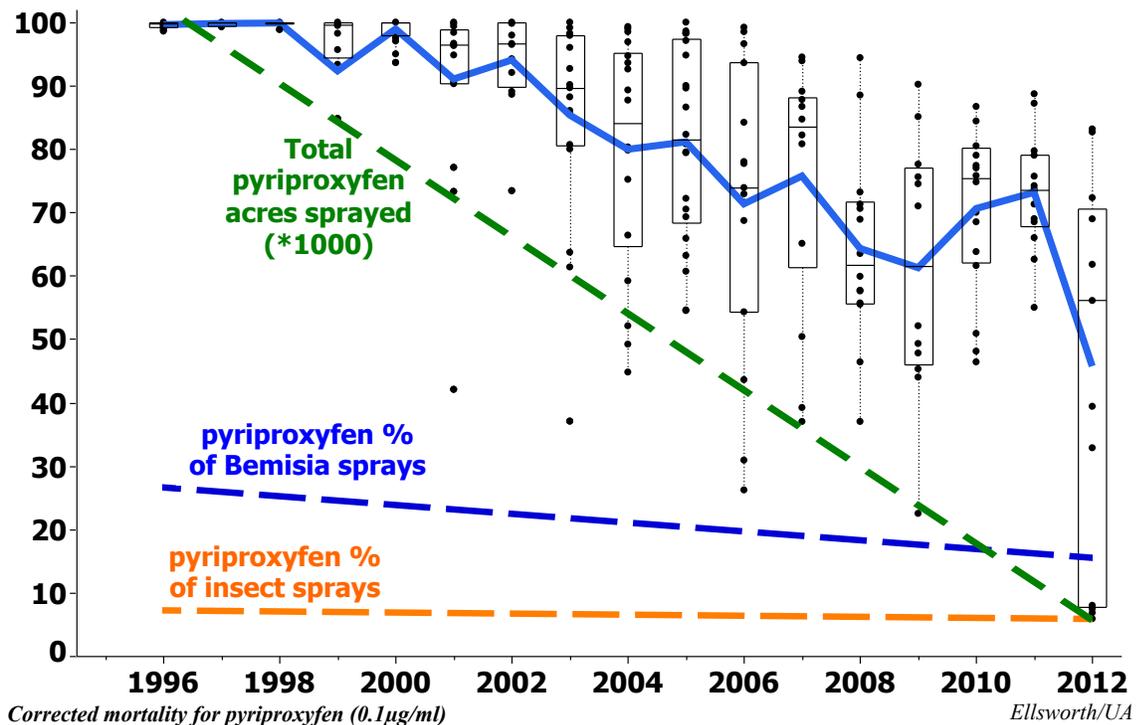
Another thorny question: What variable are we mapping? Chemical use intensity as represented by…what?

- TAC: Total acres sprayed of any or all ai's by section. TAC is not a bad measure: some chemicals are used repeatedly on the same piece of ground within a section.
- Proportion acreage of a crop treated would be ideal, but is hard to get at (but see whiteboard discussion between Yves and Peter). We could do it based on all sprays (all crops) per cotton acre.
  - The problem with this (beyond cotton) is that we don't know what a field is.
  - How do you identify a field? What we know on 1080: application data, chemical sprayed, crop, grower, PCA, acres treated. On the same 1080 form, a PCA could identify additional multiple fields (e.g., 4 * 40 acres) or one entry of 160 acres. They may combine multiple fields into a total acre amount that was treated. Is this one application or 4? Conclusion: treat all acres on a single 1080 as one application.
  - There is an open-ended field where people can identify a name of a field, but it is for their local use. We do not have a Rosetta stone to accurately identify field level sprays and match it to any kind of standard like GPS coordinates or ACRPC field codes.
- We can do percentage of all sprays: %TAC pyriproxyfen / %TAC all sprays.
- Unit of resolution is the section. Within section you can calculate the acres exposed. "Aggregate exposure" is part of the deterministic model. A spray in September hits more WFs than a spray in April. That has to be considered.
- If a field never got sprayed or if a spray went unreported, we would not know the field exists, based on the 1080 data. So we cannot easily get at refugia.
- We do have the GPS data for the corners of each section and can provide the center point for each section and the total land mass acres for each section. Each section is NOT 640 acres.
- We have total number of applications within a section (= number of 1080s for each chemistry sprayed). We have total acres treated. We can provide proportions of each chemistry within each section.

This project creates a parallel system to the resistance monitoring that Dr. Li is doing. Bob will be provided the notes from the meeting, including the discussions of research design.

Suggested steps for location of resistance monitoring effort and series of models to test for spatial associations.
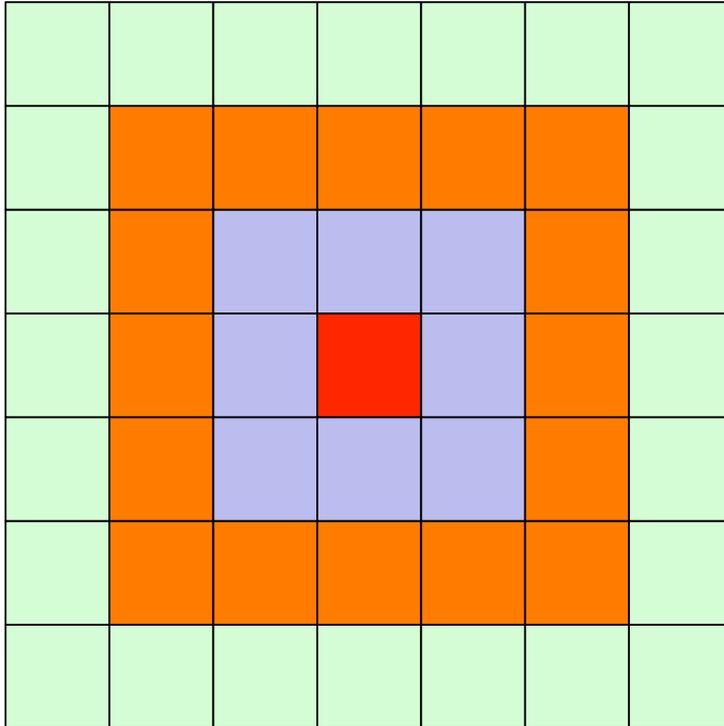
Model 1 based on 1 section
Model 2 based on 9 sections
Model 3 based on 25 sections
Model 4 based on 49 sections

R^2 examined for each

Resistance monitored in center of middle section

a, $\dfrac{\text{TAC}_{\text{All-pyr(cotton)}}}{\text{No.1080s}_{\text{All-pyr(cotton)}}}$ = **Mean Cotton Acres / Spray$_{\text{(all)}}$**

b, $\dfrac{\text{TAC}_{\text{pyr(cotton)}}}{\text{No.1080s}_{\text{pyr(cotton)}}}$ = **Mean Cotton Acres / Spray$_{\text{(pyr)}}$**

c, $\dfrac{a}{b}$ **% acres treated in cotton with pyr**

Alt. $\dfrac{\text{TAC}_{\text{pyr(cotton)}}}{\text{TAC}_{\text{All(cotton)}}}$

Resistance = (TAC$_{\text{pyr(cotton)}}$ * No.1080s$_{\text{pyr(cotton)}}$ * c)$_{2014}$
+ (TAC$_{\text{pyr(cotton)}}$ * No.1080s$_{\text{pyr(cotton)}}$ * c)$_{2015}$
+ Region

Some ideas of what is calculated for each section of the analysis based on APMC Pesticide Use Database, where pyr = pyrethroids and All = all whitefly insecticides, and is used in the statistical model.

Yves provided these additional comments post-meeting with Peter on the subject of pesticide use intensity:

I conclude that it will not be easy to get a perfect index of "pesticide use intensity" across sections.

I can see 2 measurements that make sense given the data we have.

1) The first is actually what you suggested. "Intensity" = TAC for a pesticide (either pyrethroids, pyriproxyfen, or acetamiprid) / TAC for all whitefly pesticides. Intensity is calculated for each section, by summing TACs for all whiteflies crops (cotton, melon, cauliflower, broccoli, others?).

2) The second measurement calculated for each section is ("intensity") X (area of all crops). Area of all crops is estimated for each section based on the ACRPC template of agricultural fields.

Measurement 1) above does not take into account the fact that sections can differ importantly in number of fields treated, and thus in their potential contribution to resistance development over a larger region. For example if you have 40 fields in one section and 4000 in another one, contribution of the section with 4000 fields should be larger than contribution of the section with 40 (more whiteflies come out of the section with more fields).

Measurement 2) somewhat takes into account differences in contribution of sections due to variation in crop acreage. The problem with 2) is that we do not know crop composition in sections, so there could be errors creeping in if much of the ag fields are whitefly hosts in some sections and few of the ag fields are whitefly hosts in others. Ideally, % acreage of whitefly hosts should be relatively constant across sections to use 2). The advantage of 2) is that crop acreage is estimated independently of 1). I do not think that 1080 data could be used to get an idea of whitefly crops acreage because this would result in confounding "intensity" and whitefly crop acreage.

So in summary, I would replace a, b, and c in what you sent by 1) and 2) above.

Then the models to predict resistance to a pesticide are:

a)   Resistance = region + (Intensity)$_{2014}$ + (intensity)$_{2015}$, or
b)   Resistance = region + (intensity X crop acreage)$_{2014}$ + (intensity X crop acreage)$_{2015}$,

with these variables measured over 1, 9, 25 and 49 sections centered on the section where we will measure resistance (2 populations measured in central section). I predict b) will work better

than a) if there is not too much error in estimating whitefly crop acreage from global crop acreage.

Photo 1: Whiteboard from meeting discussions.

Photo 2: Side discussions during lunch between Yves and Peter.