FINAL REPORT **YEAR**: **2012**
WTFRC Project Number:

Project Title: Protein-based foam for applying lacewings eggs to fruit trees by ATV

PI: Thomas Unruh
Organization: USDA-ARS
Telephone: (509) 454-6563
Email: thomas.unruh@ars.usda.gov
Address: 5230 Konnowac Pass Rd.
Address 2:
City/State/Zip: Wapato WA 98951

Co-PI(2): Christopher Dunlap
Organization: USDA-ARS
Telephone: (309) 681-6339
Email: christopher.dunlap@ars.usda.gov
Address: Room 3323
Address 2: 1815 N University St
City/State/Zip: Peoria IL 61604

Cooperators: David Horton, USDA-ARS Wapato, WA
Gene Miliczky, USDA-ARS Wapato, WA
Sinthya Penn, Beneficial Insectary, Redding CA

Other funding sources
WTFRC/ Apple Crop Protection
Amt. requested/awarded Total Project Request:
Requested: $239,663 / awarded: Year 1 (2010): $79,117; Year 2(2011): $79,866;
Year 3 (2012): $80,680. **Notes:** The lacewing portion of this grant overlaps with the foam project

Pending:
Western SARE: Total request: $ 178,954
WTFRC Crop Protection: Total Request $ 237, 702

Budget History

Organization Name: USDA-ARS

Item	**2012-Unruh**	**2012-Dunlap**	**TOTAL**
Salaries			
Benefits			
Wages GS-3 (90/90 days)	$7431	$7431	
Benefits	$569	$569	
Equipment	$ 400		
Supplies	$600	$1200	
Travel		$800	
Miscellaneous			
Total	$9000	$10000	$19, 000

Footnotes:

OBJECTIVES

1. **Test formulations of various foaming agents using a foam generator and adapt foam generation to a modified 12-volt pump sprayer suitable for use on an ATV**
   We have tested *keratin and whey protein hydrolysates, saponin-containing Yucca extract and Qullaja sopninaria extract as foaming agents were tested using off-the-shelf foam sprayers and a sprayer under development. The latter device drops dry lacewing (LW) eggs into the foam stream after it leaves the pressurized portion of the sprayer and appears close to a final product. The remaining problem is in the geometry of the egg delivery system which allows the eggs to be blown out ahead of the foam and falling before the target is reached.*

2. **Test adhesion of foam to waxy, water repellent, surfaces and leaves of seedling apples and on bark**
   Initial efforts have been restricted to tests on artificial surfaces including Tyvec sheets plastic cafeteria trays (Wapato) and Plexiglass (Peoria. We have found that the foam produces by keratin, Yucca and Quillaja stick well to tree trunks

3. **Test survivability of lacewing eggs in laboratory conditions when eggs are immersed in and sprayed with these foams**
   With each new formulation of foam producing liquid, measurement of survival after 30 minute submersion in the product is compared to submersion in water. With new spray technique where eggs are dropped into a trough and swept up in a stream of foam, survival has been tested with egg sprayed onto Tyvec surface or sprinkled into foam

4. **Test adherence of LW eggs in foam on apple, pear and cherry trees in the greenhouse and the field and estimate hatch rates of eggs in those settings.**
   In field experiments using tarps below trees, collect and estimate bounce-off and drop of sprays are desirable but have not yet been addressed.

5. **Estimate colonization rates (proportion of eggs recollected as larvae) on test trees.**
   Studies remain to be conducted in pears and in apples infested with aphids and pear psylla at the Moxee Farm. Preliminary studies could not be made in 2012 because of lack of aphids in our experimental farm orchards and our incapacity to apply eggs in foam during June-July

SIGNIFICANT FINDINGS

- ✓ Keratin and whey protein hydrolysates, *Quillaja* and *Yucca* saponins can produce rich foam suitable for initial contact adherence to water repellent surfaces and tree trunks
- ✓ Passage of eggs through rotary diaphragm pumps damages >25% eggs requiring eggs be introduced into the stream of the foaming agent distal to the pump
- ✓ Eggs e introduced in a suspension medium separate from the foaming medium using Venturi aspirator has proven problematic accurate
- ✓ Eggs can be dropped into the spray stream of foam after foam leaves spray nozzle.
- ✓ Mixing of eggs with dry bulking material is necessary to meter eggs for above gravity feed.
- ✓ Long term adherence depends on volume deposited, concentration of foaming agent and presence of other additives
- ✓ Psyllium husk (Metamucil), a potential bulking agent, expands on wetting, absorbs water as foam collapses and causes eggs to stick securely to Tyvec substrate

- ✓ A two trigger spray gun has been developed which has a sliding plate that collects a fixed amount of eggs with bulking material, drops eggs into a trough, and toggles the spray.
- ✓ The addition of eggs to the foam stream after it leaves the spray nozzle eliminates mechanical damage from pressure and shearing in the pump, and from long term submersion of eggs in the foaming agent or other liquids. Optimization of this spray device is required.

RESULTS & DISCUSSION

Objective 1 - Initial testing evaluated the suitability of a variety of natural products and proteins to serve as a foaming agent in this application. A variety of food grade or OMRI approved proteins or natural surfactants were evaluated to serve as foaming agents. Preliminary screening evaluated keratin hyrdolysate, egg albumin, gelatin, whey protein isolate and concentrate (Glanbia inc.), β-lactoglobulin and α- lactalablumin (Davisco foods inc.) and *Yucca schidigera* extract and recently *Quillaja saponaria* extract.. The suitability was evaluated by measuring their physical properties including dynamic surface tension, expansion ratio, half-life, and density using standard procedures. This analysis was conducted us a pestifoamer PF-2 (Richway Industries) to generate foam in continuous mode.

Figure 1. Richway Pestifoamer.

In general, full size proteins evaluated lacked sufficient dynamic surface tension to produce suitable foam under these conditions. The protein hydrolysates (keratin hydrolysate and whey protein isolate) provided better dynamic surface tension due to their smaller molecular size and faster diffusion rates. Still only keratin hydrolysate produced acceptable foam characteristics under these conditions. While keratin hydrolysate had the potential to serve a suitable foam agent, it had other potential limitations. Keratin hydrolysate, while derived from agricultural products (bovine hooves and horns), is not currently OMRI approved. In addition, while it is produced on a commercial scale for fire-fighting foams, it is not readily available without antimicrobial biocides included as preservatives. These limitations and the need to use OMRI products in some field testing sites caused to take a closer look at existing OMRI certified surfactants that could be adapted with other adjuvants to serve as suitable foaming agents. This search identified *Yucca schidigera* and *Quillaja saponaria* extracts, both of which are OMRI approved agricultural surfactants(with a reputation of undesirable tank foaming in standard spray applications). Initial screening identified it as having acceptable dynamic surface tension to meet our requirements. Preliminary screening of hatch rate of eggs after being submerged in the yucca extract showed no appreciable differences from water controls up to 5% yucca. These studies suffered from low survival of eggs in the water control. The following foaming systems were evaluated Moultrie MFH-SPR15P ATV sprayer (Moultrie inc), Pump up bullet foamer, model#925008 (LaffertyEquipment) Pestifoamer (Richway Industries LTD), a variety of TeeJet venturi spray tips on a generic variable pressure sprayer. In each case these sprayers were used with 3.5% keratin hydrolysate. **Mixtures have not been tested.**

The secondary goal under this objective was to determine the best method to introduce lacewing eggs to foam. The solution to this objective was confounded by the competing engineering requirements needed for foam generation and introducing the lacewing eggs. After much trial and error, we concluded that the ideal system would produce a transient pulse of foam with some ability to cast it and introduce the eggs in a batch mode. The eggs would be metered in a dry state on a tree by tree basis. **Figure 2** shows a prototype design of an applicator sprayer that fulfills these design requirements.

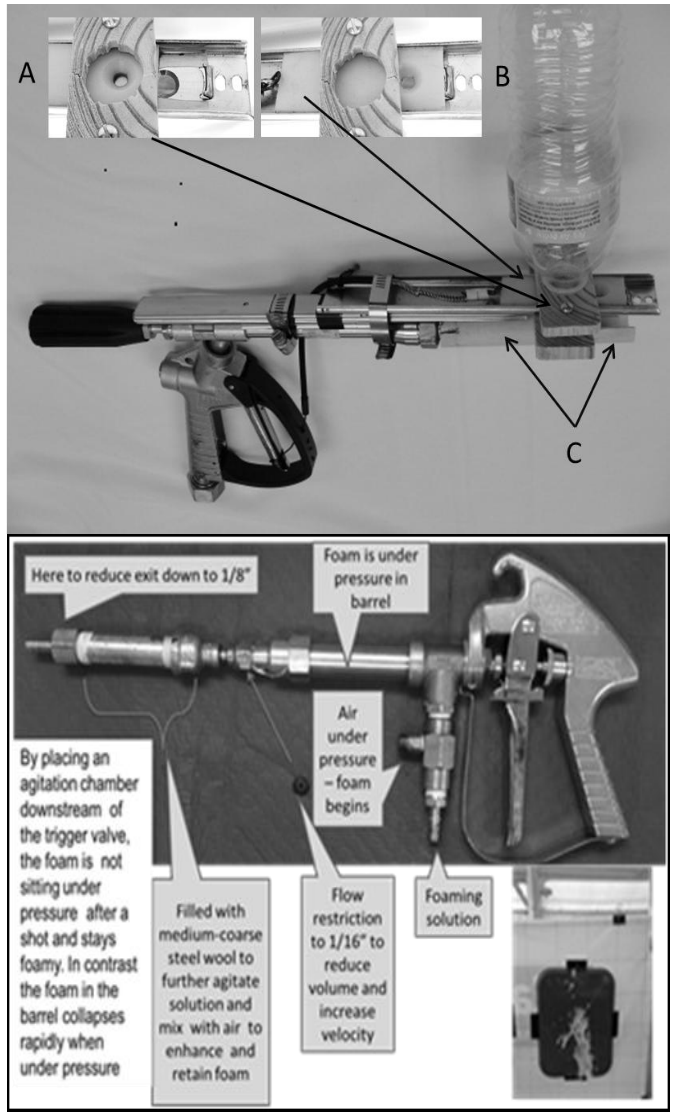

Figure 2. Two prototype designs of a two stage foam generating LW egg sprayer are shown. Sprayer in upper panel shows: A) sliding plate in the refill position - when the beveled hole would be below and this refilled by the hopper (=the plastic bottle which would contain eggs mixed with dry bulking agent); simultaneously the trigger fully squeezed (foam spray is on) or trigger is half depressed; B) sliding plate is above spray trough and has released the eggs – at this time the spray is off and the trigger is at rest; C) spray trough, inside are found air induction nozzles that produce the foam and a trough that the spray slides across picking up the LW eggs on the way out. Sprayer in lower panel does not show the egg delivery system but instead shows and describes a new design where foam quality is improved by introducing compressed air and adding a sparging system(mixing chamber)

Objective 2 was to test of adhesion of foam to waxy, water repellent surfaces in the laboratory in Peoria using a foam generator/sprayer. Fulfilling this objective was limited by the ability to settle on a preferred method of foam generation and egg introduction, which greatly impact the physical properties (such as velocity and droplet size) of the emitted foam solution. However, efforts were made to identify suitable materials that mimic the properties of apple tree surfaces. A literature survey and analysis of local tree stock determine apple leaves are generally considered easy to wet with water contact angles in the 60-80o range. The bark of local apple trees, at the estimated site of application, was variable with an average water contact angle of 74 ± 9^{o}. It was decided to use Plexiglas with a water contact angle of 76^{o} as the leaf mimic, due to its low cost and wide availability. The branches of the canopy will imitated with small diameter polyvinyl chloride pipe, which has a water contact angle of 85^{o}. Once a suitable foam generation system has been identified these mimics will be used to evaluate the influence of additional adjuvants on adhesions. These adjuvants will include viscosity modifiers, polymers to promote egg suspension and adhesion. Recent tests using psyllium as a dry bulking agent for metering out eggs shows exceptional promise in assisting sticking of the foam because the water leaving the foam as it collapses is taken up by the psyllium. In preliminary tests it appears the larges problem from psyllium is using too much, then eggs become trapped in a mat of cross-linked psyllium fibers. Finer grinding of the psyllium husks may also alleviate this problem.

Objective 3. Tests of LW egg survival following submersion in protein hydrolysates show promise, but survival less than 50% has been seen in the firefighting foam and in both the two saponin extracts from Yucca and Quillaja solutions. We have found that lower survival is cause by excessive storage of eggs prior to use for testing. Additional testing will be done once a foam generation and spraying system is finalized. The most recent test (Dec 17, 2012) is shown in Figure 3.

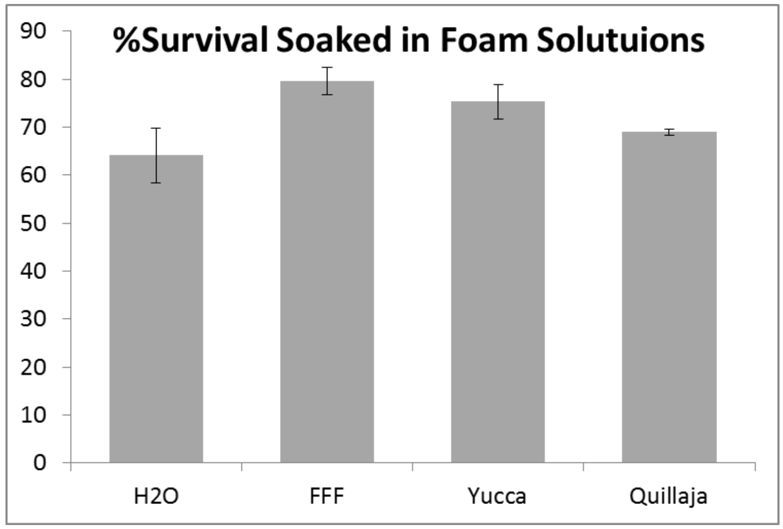

Figure 3. Survivorship of lacewing eggs following 30 minute immersion in three foaming solutions and a water control. Eggs were dried following immersion by placement on a towel and allowed to dry and then dropped on a dry adhesive (back side of an adhesive label) which allows hatch but prevents lacewing larvae from moving to other eggs and feeding on unhatched eggs.

Objective 4 and 5. A preliminary study was conducted at the Moxee farm which consisted of sprinkling dry eggs onto fresh foam solution, wet white glue and water alone painted onto leaves. Best retention of eggs occurred on leaves with glue, followed by foam and no retention at 3 days was observed with water. Improvements in adhesiveness of foam is provided using bulking agents (Metamucil) as observed by greenhouse studies subsequent to field tests. Objectives 4 and 5 remain incomplete until we have an optimal foam and egg sprayer.

EXECUTIVE SUMMARY

Project Title: Protein-based foam for applying lacewings eggs to fruit trees by ATV

Participants: Tom Unruh and Christopher Dunlap; USDA-ARS

Budget: $19,000 for 1 year.
Second year is not being requested and a no-cost extension of funding is requested for continued on project with funds remaining from year 1.

OVERVIEW
This project was designed to discover a organically useful foaming agent that could be used to apply lacewing eggs onto trees using an ATV that is only mildly modified from standard spray programs they are currently used for in apple and cherry orchards. There have been two sides to our efforts: 1) chemical, specifically to find an OMRI-approved foaming agent that preserves the health of the lacewing eggs and provides adhesion to foliage or tree bark; 2) mechanical, develop a sprayer that both produces the foam and delivers the lacewing eggs to the trees while an applicator is on the ATV. We have made progress on both fronts, but have not completed the project. We do not ask for funds for a second year because we received funds rather late in the granting cycle and had delays in hiring assistants. Given that, we intend to reach the goals stated with the funds provided in year 1.

Accomplishments:

Keratin hydrolysates, *Yucca* and *Quillaja* saponin extracts all produce suitable foam which adheres well to foliage and tree bark in test application. Only the saponin extracts are OMRI approved.

Survival of lacewing eggs in foaming agents exceeds 80% in many trials in the laboratory.

A modified hand gun that sprays foam through a cylinder where dry lacewing eggs are placed can accurately deliver the eggs in foam to a target 6-8 feet distant. Additiono of compressed air to this system has provided a very rich foam.

Eggs are dropped in the cylinder after a single spray cycle (=trigger pull and release) and addition of bulking agents (ground rice hulls or sphagnum) together with a sticking agent (dry chopped psyllim hulls – the ingredient of Metamucil) result in significant adhesion of the foam.

Work Needed:

The mechanical sprayer needs to be optimized to have reliable retrieval and carrying of the eggs from the spray gun and in metering the egg numbers accurately.

Demonstration of the utility and efficacy of application of LW eggs in foam must be demonstrated in the field

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