CALIFORNIA DEPARTMENT OF FOOD AND AGRICULTURE
SPECIALTY CROP BLOCK GRANT PROGRAM
PROGRESS REPORT

PROJECT INFORMATION				
USDA PROJECT NUMBER:	53			
RECIPIENT ORGANIZATION NAME:	California Department of Food and Agriculture			
PROJECT TITLE:	Biological Control of Bagrada Bug, Bagrada Hilaris (Pentatomidae)			
CDFA GRANT NUMBER:	SCB16053			
RECIPIENT'S PROJECT CONTACT				
NAME:	Charles H. Pickett			
PHONE:	(916) 262-2053			
EMAIL:	Charlie.Pickett@cdfa.ca.gov			
PROJECT REPORT				
ANNUAL REPORT TYPE:	2nd Annual Report			
REPORTING PERIOD:	**START DATE:**	10/1/17	**END DATE:**	9/30/18

ACCOMPLISHMENTS

OCTOBER 2017 – MARCH 2018

The estimated total percentage of work completed on this project …….. 20%

#	Objective	Activity and Accomplishment
1	Objective 1: Measure host specificity of *Ooencyrtus* sp.	The host specificity of *Ooencyrtus* sp. was tested on three other stink bug species: the invasive pests *Nezara viridula* (*N. viridula*) and *Halyomorpha halys* (*H. halys*) and the native species *Thyanta pallidovirens* (*Th. pallidovirens*). For each replication with each alternate host species, three vials were prepared: one with a card of ten *Bagrada hilaris* (*B. hilaris*) eggs, a second with a card of ten alternate host eggs, and a third with two cards of ten eggs each, one from *B. hilaris* and one from the alternate species. The first two vials constituted a "no choice" test and the third vial was a "choice" test. A wasp was added to each vial and removed after 24-hours. Then one of the "choice" egg cards in each replication was removed to a separate vial so each card was in its own vial. For *H. halys*, more wasps emerged from *B. hilaris* eggs in no-choice and choice tests. For *Th. pallidovirens*, if the wasp was given a choice, they laid more eggs on *B. hilaris*.

#	Objective	Activity and Accomplishment
		But in no-choice tests, they laid the same number of eggs in both hosts. For *N. viridula*, the number of wasps that emerged was greater on *B. hilaris* in both the choice and no-choice tests. In addition to this experiment, the project team maintained a colony of *N. viridula*-reared wasps for more than two years, and a colony of *Th. pallidovirens*-reared wasps for over nine months. *Ooencyrtus* sp. was also reared on another native species, *Chlorochroa uhleri*, for several months. The success with these colonies showed that *Ooencyrtus* sp. could reproduce successfully on these alternate host species.
2	Objective 2: Survey for resident parasitoids and predators of bagrada bug.	From 13 locations across the Sacramento region and central California, 101 sentinel cards were exposed to predation and parasitism over the season. Each card had 12 to 18 non-viable *B. hilaris* eggs glued to them to measure naturally occurring predation and parasitism. Only two foliar cards (same site, different months) had eggs that were parasitized. Three *Ooencyrtus s*p. emerged from one card, while six eggs were parasitized on the other card with no successful emergence of adult parasitoids. High levels of predation were recorded from cards placed both on the ground and on leaves of host plants. A total of 25 percent of eggs on leaves and 44 percent of eggs on the ground were preyed upon. However, this number may increase as the team begins to refine exactly what a predated egg would look like. *B. hilaris* lay their eggs just below the soil surface. By placing a thin layer of sand on top of some of the sentinel egg cards, the team measured whether buried eggs deterred predation. It did not. Based on cameras imaging cards placed on the ground at 2 of the above 13 locations, 5 predacious arthropods were identified feeding on *B.*

#	Objective	Activity and Accomplishment
		hilaris eggs, ants being the most common and active. In Riverside, the team placed 195 sentinel cards, each card containing 15 live *B. hilaris* eggs that were 1 day old. Cards were left in the field for 5 days, then collected and held for parasitoid or bug emergence. The cards deployed in October 2017 and November 2017 were placed on the soil and at a height of 30 cm. There was heavy predation on the eggs on soil cards. While the team did not identify the predators, an abundance of argentine ants, *Linepithema humile*, were observed. Due to the high predation, only upper cards placed at 30 cm off the ground were utilized on subsequent sampling. Of the total 2,925 eggs, the team recovered 63 parasitoids on 8 of 13 sampling dates. Parasitoids were identified as *Trissolcus hyalinipennis* (*T. hyalinipennis*), and *Trissolcus basalis (T. basalis). T. basalis* was a widespread and important parasitoid of southern green stinkbug, *N. viridula*, and it was clear that it will utilize *B. hilaris* eggs. The other species, *T. hyalinipennis* was known only from Pakistan, and likely was introduced with *B. hilaris*.

APRIL – SEPTEMBER 2018

The estimated total percentage of work completed on this project …….. 70%

#	Objective	Activity and Accomplishment
1	Objective 1: Measure host specificity of *Ooencyrtus* sp.	The project team completed 15 replicates each of choice and no-choice experiments on six pentatomid to test whether *Ooencyrtus* sp. could reproduce on hosts other than *B. hilaris* (1 rhopalid and 1 coreid species, all Heteroptera). One of the pentatomid species, *Podisus maculiventris* (*P. maculiventris*), was a native, beneficial predator. All of the other pentatomid species and the coreid species were

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		considered pests. *N. viridula, Murgantia histrionica* and *H. halys* were invasive species, while *Th. pallidovirens* and *Chlorochroa uhleri* were native pest species. In the choice test, two cards of ten eggs each were placed in a glass vial, one card with *B. hilaris* eggs and one card with eggs from the alternate species. In the no-choice test, one vial had a card of ten *B. hilaris* eggs, and a second vial had a card of ten alternate host eggs. A 3-day old *Ooencyrtus* sp. female wasp was added to each vial and removed after 24-hours. Then one of the "choice" egg cards in each replication was removed to a separate vial so that each card was in its own vial. The vials of parasitized eggs were checked every 24-hours and the number of bugs and wasps that emerged was recorded. The results were analyzed with Poisson regression analysis, with P <0.05 level of significance. *Ooencyrtus* sp. reproduced successfully on all of the species tested. For the choice test, the emergence of wasp offspring was significantly lower on the alternate species than on *B. hilaris*, except for *Th. pallidovirens* and *P. maculiventris*, which did not differ significantly. For the no-choice test, emergence was significantly lower on the alternate species except for *Th. pallidovirens*, and *N. viridula* was borderline, with P = 0.06. Thus, *Th. pallidovirens* was the only species on which reproduction was not significantly lower than *B. hilaris* in either test. These tests showed that *Ooencyrtus* sp. could reproduce on a number of species other than *B. hilaris.*
2	Objective 2. Survey for resident parasitoids and predators of bagrada bug.	Northern California: The team continued to deploy sentinel cards during this reporting period to evaluate parasitism by resident parasitoids and to measure egg predation.

#	Objective	Activity and Accomplishment
		This reporting period covered the warmer months and thus the time of year when the majority of cards were deployed. Roughly 415 sentinel cards with *B. hilaris* eggs attached were placed at 23 locations. One to two cards were placed on host plant leaves and one on the ground at each location. As last year, many cards on the ground had high levels of egg predation. The samples have yet to be processed for exact numbers of damaged eggs and degree of parasitism. Imaging of cards on the ground was done at 4 of 15 sites. Time-lapse imaging for 2 to 3 days was conducted 15 times over. To support data from imaging, the team added pitfall traps at the same sites where camera work was conducted. These will provide samples of ground dwelling predators that could be imaged feeding on eggs. These were needed for identification to species. The results from these collections will identify the species of predators attacking eggs of *B. hilaris*, fulfilling this survey objective. Southern California: The team deployed 285 sentinel egg cards, each card containing 15 live *B. hilaris* eggs that were 1 day old. After five days in the field, the cards were collected and held for parasitoid or bug emergence. Since it was discovered that cards deployed on the ground had a high percentage of parasitism, the team began using only egg cards placed at 30 cm off the ground. To date, from a total of 4,275 eggs, the team recovered 62 parasitoids on 9 of 20 sampling dates (1.5 percent). Parasitoids were identified as *T. hyalinipennis*, *T. basalis, Trissolcus hullensis (T. hullensis),* and *Trissolcus utahensis* (*T. utahensis*). *T. hyalinipennis* was known only from Pakistan, and likely was introduced with *B. hilaris*. *T. basalis* was imported from the

#	Objective	Activity and Accomplishment
		Mediterranean areas of France, Italy and Spain to the United Sates to control the invasive southern green stink bug, *N. viridula* and first was released in 1987 in Davis, California, and then in other areas throughout the state. Currently it is widespread and the team was able to demonstrate that it utilized *B. hilaris* eggs. *T. hullensis* was identified in the United States, Canada and Mexico as a parasitoid of stink bugs. *T. utahensis* was another parasitoid of stink bugs that had been only reported from the United States and Canada.

CHALLENGES AND DEVELOPMENTS

OCTOBER 2017 – MARCH 2018

Challenge	Corrective Action and/or Project Change
Defining a sentinel egg that had been preyed upon.	The team began a control study to determine what eggs looked like that had not been exposed to predation in the field. This should help clarify whether or not some eggs seen on cards exposed in the field were clear in appearance due to predation or some other factor.
Poor trap catches of *B. hilaris*.	The team was not entirely sure why the *B. hilaris* population was exceptionally low in 2017. Commercial traps for this pest have yet to be developed for it. Therefore, the team tested four traps to determine which was the best, and if color of the trap was important.

Positive Development	Project Change
No applicable.	

APRIL – SEPTEMBER 2018

Challenge	Corrective Action and/or Project Change
Not applicable.	

Positive Development	Project Change
Not applicable.	

OUTCOME AND INDICATOR RESULTS TO DATE

OCTOBER 2017 – MARCH 2018

Outcome and Indicator	Quantifiable Results
Outcome 4: Enhance the competitiveness of specialty crops through greater capacity of sustainable practices of specialty crop production resulting in increased yield, reduced inputs, increased efficiency, increased economic return, and/or conservation of resources. Indicator 2: Adoption of best practices and technologies resulting in increased yields, reduced inputs, increased efficiency, increased economic return, and conservation of resources.	Identifying the right biological control agents for *B. hilaris* had direct impact on this outcome. Effective natural enemies contribute to sustainability through reducing inputs, particularly insecticides. This studies showed that *Ooencyrtus* sp., an imported parasitoid from Pakistan was a generalist. While it may prefer *B. hilaris* in choice studies, it will oviposit on other exotic and native stink bugs. This information was beneficial to determine if this parasitoid belonged in an integrated program for *B. hilaris*.
Outcome 5: Enhance the competitiveness of specialty crops through more sustainable, diverse, and resilient specialty crop systems. Indicator 8: 2,500 growers/producers that gained knowledge about science-based tools through outreach and education programs.	This research was presented to various stakeholders. During this reporting period, there were 352 "views" to the *B. hilaris* web page hosted by the Center for Invasive Species Research. In addition, presentations were given on *B. hilaris* and *Ooencyrtus* sp.: • Entomological Society of America meeting in Denver Co., November 2017. (50 attendees). • Riverside Master Gardeners, March 2018. (40 attendees). In addition, the following papers/chapters on *B. hilaris* were published during this reportin period: • Reed, D.A., Ganjisaffar, F., Palumbo, J., Perring, T.M. 2017. Effects of temperatures on immature development and survival of the invasive stink bug,

Outcome and Indicator	Quantifiable Results
	Bagrada hilaris (Hemiptera: Pentatomidae). Journal of Economic Entomology. Vol. 110: 6 p.2497-250. • Bundy, C.S., Perring, T.M., Reed, D.A., Palumbo, J.C., Grasswitz, T.R., Jones, W.A. 2018. Bagrada hilaris. Pp. 205-241 in (J. McPherson, ed.) Biology of Invasive Stink Bugs and Related Species. CRC Press/Taylor and Francis Books, Boca Raton, FL.

APRIL – SEPTEMBER 2018

Outcome and Indicator	Quantifiable Results
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Outcome 5: Enhance the competitiveness of specialty crops through more sustainable, diverse, and resilient specialty crop systems. Indicator 8: 2,500 growers/producers that gained knowledge about science-based tools through outreach and education programs.	This research had been presented to various stakeholders. During this reporting period, there were 353 "views" to the *B. hilaris* web page hosted by the Center for Invasive Species Research. This pest was also discussed with the general public and displayed as part of a poster on Integrated Pest Management at the 4th Annual City of Riverside and the University of California, Riverside Insect Fair (March, 28, 2018). An estimated 10,000 people attended this fair. In addition, the following presentations were given on *B. hilaris* and *Ooencyrtus* sp.: • Riverside Master Gardeners, May 2018. (35 attendees).

Outcome and Indicator	Quantifiable Results
	Parts of this project were summarized in an online magazine article via an interview: • Specialists seek natural enemies for bagrada bug. Ag Alert. September 19, 2018. The following paper related to *B. hilaris* management was also published during this review period: • Ganjisaffar, F., E.J. Talamas, M.C. Bon, L. Gonzalez, B.V. Brown, and T.M. Perring. 2018. Trissolcus hyalinipennis (Rajmohana & Narendran (Hymenoptera, Scelionidae), a parasitoid of Bagrada hilaris (Burmeister) (Hemiptera, Pentatomidae), emerges in North America. J. Hymen. Res. 65: 111-130.

DISCUSSION OF ACTIVITIES PERFORMED (IF NEEDED)

OCTOBER 2017 – MARCH 2018

No additional information.

APRIL – SEPTEMBER 2018

No additional information.

UPCOMING ACTIVITIES

OCTOBER 2017 – MARCH 2018

No information was provided at this time.

APRIL – SEPTEMBER 2018

Activity	Anticipated Completion
Analyze results	Jan. 2019
Prepare final reports	Mar. 2019

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FEDERAL EXPENDITURES

Cost Category	Amount Approved in Budget	Actual Federal Expenditures (Federal Funds ONLY)
Personnel	$47,134.00	$41,799.869
Fringe Benefits	$21,211.00	$19,221.44
Travel	$15,343.00	$2,600.98
Equipment	$0.00	$0.00
Supplies	$5,229.00	$5,133.41
Contractual	$285,258.00	$203,612.40
Other	$1,200.00	$0.00
Direct Costs Sub-Total	$375,375.00	$272,368.09
Indirect Costs	$0.00	$0.00
Total Federal Costs	**$375,375.00**	**$272,368.09**

Percentage of total federal funds expended to date ……..…..…73%

PROGRAM INCOME

Source/Nature (i.e., registration fees)	Amount Approved in Budget	Actual Amount Earned
N/A	$0.00	$0.00
	$0.00	$0.00
	$0.00	$0.00
Total Program Income Earned	**$0.00**	**$0.00**