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Trapping Sweetpotato Weevil, *Cylas formicarius* (Coleoptera: Brentidae), with High Doses of Sex Pheromone: Catch Enhancement and Weathering Rate in Hawaii

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Abstract. Sweetpotato, Ipomoea batatas (L.) Lamarck, one of the top ten staple crops produced worldwide, has increased in production in Hawaii in recent years. The sweetpotato weevil, Cylas formicarius (Summers) (Coleoptera: Brentidae), is a major economic and quarantine pest of sweetpotato in Hawaii as well as a pest of concern in all parts of the tropics where sweetpotatoes are grown. Sweetpotato weevil infestation can reduce marketable root yield as well as reduce root quality by inducing production of bitter tasting sesquiterpines by the sweetpotato tissue. Traps baited with a male sweetpotato weevil lure, (Z)-3-dodecenvl (E)-2-butenoate, can be used for population monitoring, or even for population suppression if mass trapping is done using high doses of this lure. Weathering rates, though, have not been documented in Hawaii for the higher septa loadings (100 to 1000 µg [=1.0 mg]) that have been proposed for use in population suppression efforts through mass trapping. Here, we present comparative catch rates and weathering rates, along the Hamakua Coast of Hawaii island, of traps baited with septa loaded with 12 µg, 120 µg, or 1.0 mg of male sweetpotato weevil lure. Traps baited with fresh 1.0 mg male lure caught over 22 times as many weevils as traps baited with 12 µg lure over an initial one-week trapping period. Based on a fitted decay curve, decline in attractiveness of the 1.0 mg treatment to 50% of fresh attractiveness occurred at 19.0 weeks, while the 120 µg treatment showed a 50% decline after 16.3 weeks, under climate conditions on the Hamakua Coast of Hawaii island. Further research is needed to test the effectiveness of mass trapping in reducing root damage by sweetpotato weevil, through the use of a high dose male lure in combination with the recently reported enhancement of trap catch by adding a green light source.

Key words: Ipomoea batatas, Cylas formicarius, detection, monitoring, mass trapping

Sweetpotato, *Ipomoea batatas* (L.) Lamarck, is one of the top ten staple crops produced worldwide, trailing only corn, rice, wheat, potatoes, cassava, and barley (FAO 2014). Among the nations of the world, the United States is the 12th greatest producer of sweetpotatoes (USDA-ERS 2012), with production greatest in the states of North Carolina, California, Mississippi, and Louisiana (USDA-NASS 2013a). Production in Hawaii has continued to increase in recent years, reaching 364 harvested ha, with a total production of 5.90 million kg in 2012 (USDA-NASS 2013b). Based on 2011 data (2012 data not available for all crops), sweetpotato is now the 10th highest-value crop in the state of Hawaii (USDA-NASS 2014). The sweetpotato weevil, Cylas formicarius (Summers) (Coleoptera: Brentidae), is a major quarantine pest of sweetpotato in Hawaii (Follett 2006) as well as a pest of concern in all parts of the tropics where sweetpotatoes are grown (Heath et al. 1986). Sweetpotato weevil infestation can reduce marketable root yield as well as reduce root quality through induction of production of bitter tasting sesquiterpines by the sweetpotato tissue (Heath et al. 1986). Monitoring of sweetpotato weevil populations is facilitated by the use of traps baited with a male sweetpotato weevil lure [(Z)-3-dodecenyl (E)-2-butenoate] (Heath et al. 1986). Traps baited with higher doses of this lure can, however, also be used for population suppression through mass trapping (Jansson et al. 1991, Pillai et al. 1993, Hwang 2000, Reddy et al. 2014). Earlier studies under field conditions on the Hamakua Coast on the island of Hawaii have shown that sweetpotato weevil catch in traps baited with rubber septa loaded with 12.0 µg of sweetpotato weevil male lure dropped to 50% of the catch of unweathered lure at 13.2 weeks, at a lower-elevation site, and at 9.0 weeks at a higher-elevation, windier site (McQuate 2011). Local weathering rates, though, have not been documented for higher septa loadings (100 to 1000 µg [=1.0 mg]) that have been proposed for use in population suppression efforts through mass trapping (Jansson et al. 1991). Here, we present comparative catch rates and weathering rates, along the Hamakua Coast of Hawaii island, of traps baited with septa loaded with 12 µg, 120 µg, or

Materials and Methods

1.0 mg of male sweetpotato weevil lure.

Attractant. Three different loadings (12 μ g, 120 μ g, and 1.0 mg) of sweetpotato weevil male lure, ((Z)-3-dodecenyl (*E*)-2-butenoate), in rubber septa were obtained from Scentry Biologicals, Inc. (Billings, MT). Septa were deployed in sweetpotato weevil traps (universal moth traps) obtained from Great Lakes IPM, Inc. (Vestaburg, MI). The universal moth traps used were plastic traps with a 1.8 liter base white bucket (15.2 cm diameter at the top) underneath a yellow plastic funnel covered, 2.5 cm above, with a green plastic disk (16.0 cm diameter) to keep rain out. A green plastic cage inserted down through the covering disk held the treated rubber septum.

Sites. Traps were weathered in fields in which sweetpotatoes were grown on the Hamakua Coast (East side) of the island of Hawaii. Because of the long duration of the weathering trial (40 weeks, which is longer than a normal sweetpotato production cycle), it was necessary for the weathering to be conducted in more than one field. Overall, weathering was conducted in a progression of five separate fields in the vicinity of Pepeekeo, HI, with transition to new fields used as an opportunity to conduct one-week assessments of the relative strength of male lure impregnated rubber septa of different male lure loadings and different weathering duration (see Figure 1). Initial weathering (Site 1) was conducted at a site just off of Sugar Mill Road, on the makai side of the Hawaii belt road, near Pepeekeo, HI (Universal Transverse Mercator [UTM] grid [USGS 2001]: Easting 0280818, Northing 2196273 m, Zone 05 Q; 88 m elevation). Traps were moved to Site 2 (Easting 0281024, Northing 2194462 m; 105 m elevation) after eight weeks; to Site 3 (off of Kaupakuea Homestead Road on the mauka side of the Hawaii belt road: Easting 0277295, Northing 2194518 m; 304 m elevation) after 16 weeks; to Site 4 (off of Kaupakuea Homestead Road on the mauka side of the Hawaii belt road: Easting 0276728, Northing 2194394 m;



Figure 1. Map of weathering trial showing locations of fields where traps with lures were placed (developed using ArcGIS [ESRI 2012]). Traps were initially deployed at Site 1 on 14 February, 2012, and moved on to Sites 2, 3, 4, and 5 over the course of the weathering trial. The weathering time of the traps at each site was as follows: (Site 1) first 8 weeks; (Site 2) weeks 9–16; (Site 3) weeks 17–24; (Site 4) weeks 25–40; and (Site 5) week 41 (assessment). A weather station was maintained over the course of the weathering trial and was located at Site 2 for the first 16 weeks and then located at the location of the filled circle on the map for the remaining weeks of the trial.

339 m elevation) after 24 weeks; and to Site 5 (off of Kaupakuea Homestead Road on the mauka side of the Hawaii belt road: Easting 0276422, Northing 2194464 m; 368 m elevation) after 40 weeks. A Wireless Vantage Pro2 weather station (Davis Instruments, Hayward, CA) was set up to collect data on rainfall, temperature, relative humidity, and wind speed over the course of the 40-week weathering period and one-week assessment period. The weather station was maintained at Site 2 for the first 16 weeks and then moved near Site 3 (at the location of the filled circle on the map in Figure 1) for the remaining weeks of the trial.

Bioassays. Two types of trials were conducted: lure weathering trials and dose response trials. Because higher loadings of the male lure would be expected to persist longer, and it was desired to document the drop in attractiveness over time, we conducted the weathering trial over 40 weeks, deploying new traps (with fresh lure) every 8 weeks. Trap height was regularly adjusted to be just above foliage height throughout the weathering trial as done for an earlier 12 µg lure weathering trial (McQuate 2011). Every eight weeks (0, 8, 16, 24, 32, and 40 weeks), beginning on 14 Feb., 2012, a set of 15 sweetpotato weevil traps (five traps of each of three treatments: 12 µg, 120 µg, or 1.0 mg loadings in rubber septa of male sweetpotato weevil lure) was set out in a randomized complete block (RCB) assessment grid (Weeks 0, 16, and 40) or in an RCB weathering grid. Spacing among traps at any given time varied with whether they were placed in a weathering or assessment grid and with the size of the sweetpotato field. Further details on spacings used over the course of the 40-week weathering trial are presented below. Each trap held 300 ml water, with 0.5 ml Dawn Ultra dishwashing liquid (Procter & Gamble, Cincinnati, OH) added as the killing agent for captured weevils. Traps were placed in assessment grids only one week before trap servicing to assess trap catch.

Trial 1. First one-week assessment. The first set of traps, placed in a 10 x 10 m RCB design on 14 February, 2012, was serviced after one week, and all recovered weevils counted. This provided data on relative catch of fresh lure across the three septa loading rates. Traps, with lures, were all left in the field for further weathering and serviced every week to remove (but not count) any trapped weevils (counts were only done at the end of specific oneweek assessment periods). At the start of Week 8 (10 April, 2012), these traps were transferred to "field 2" and set out in an RCB design (10 m x 10 m spacing), along with another set of traps with fresh septa (= the start of Trial 2 [see below]).

Trial 2. Second one-week assessment. At the start of Week 9 (17 April, 2012), weevils were recovered from the traps with the fresh septa and counted, while the weevils from the traps that were 8 weeks older were cleared, but the weevils were not recovered and counted. The counts from the traps with the fresh septa gave a second estimate of relative one-week catch rates in traps baited with septa with the three different loadings. Weevils were cleared from traps every two weeks thereafter up until Week 16.

Trial 3. Test of weathering for weeks 0, 8, and 16. At the start of Week 16 (5 June, 2012) Week 0 and Week 8 traps, along with another set of traps with fresh septa (Week 16), were transferred to "field 3" and set out in an RCB design (10 x 10 m spacing). All traps were serviced one week later (12 June, 2012) and all captured weevils counted. This provided data on relative trap response for all three loadings for fresh (0 weeks) lure versus lure weathered for 8 or 16 weeks. The counts from the traps with the fresh septa also gave a third estimate of relative one-week catch rates in traps baited with septa with the three different loadings.

Trial 4. Test of weathering for weeks 0, 8, 16, 24, 32, and 40. Traps continued to be serviced weekly after the 16-week service, but trapped weevils were not counted until the first week after traps were deployed on Week 40 (20 Nov., 2012). At the start of Week 24 (31 July, 2012) Weeks 0, 8, and 16 traps, along with another set of traps with fresh septa (Week 24), were transferred to "field 4" and set out in an RCB design (10 x 10 m spacing) for continued weathering. This transfer was needed both because "field 3" was soon to be harvested and so that the remaining weathering time would be in a field close to where the final assessment trapping would be conducted. At the start of Week 32, another set of traps with fresh septa (Week 32) was added to the traps in "field 4." Trap locations for all traps were re-randomized and set out in an RCB design (10 x 10 m spacing) for continued weathering. At the start of Week 40 (20 Nov., 2012), Weeks 0, 8,16, 24, and 32 traps, together with another set of traps with fresh septa (Week 40), were transferred to "field 5" and set out in an RCB design (5 x 5 m spacing) to begin the final assessment trial. All traps were serviced one week later (27 Nov., 2012) and all captured weevils counted. This provided data on relative trap response for all three loadings for fresh versus aged lure at 0, 8, 16, 24, 32, and 40 weeks of weathering.

Statistical analysis. Weevil counts were square root transformed (SQRT [catch + 0.5]) and subjected to analysis of variance (ANOVA) to test for significance of trap catch differences among the three treatments and among weathering weeks. Untransformed data are presented in the summary charts. Comparison of the effect of lure dose on trap catch response was first tested using the fresh catch data

(i.e., catch over the first week of deployment) from Trials 1, 2, and 3. Response of weevils to fresh lure over the one-week assessment period was analyzed using a 2-way ANOVA with site and dosage as main effects. Tukey HSD was used to test for significance of differences of treatment means (SAS Institute Inc. 2012). Further comparison of the dose effect, along with an initial test of the decline in attractiveness over time (weathering), was tested using the data recovered from Trial 3 (0-, 8-, and 16-week weathering data). Low weevil population in the field used for the final assessment, combined with lack of catch in many of the 12 µg-loaded traps, prevented the use of ANOVA for assessment of the trap catch data from the final assessment period. However, it was possible to estimate rate of weathering over the 40 week weathering period for both the 120 µg and 1 mg treatments by developing and statistically assessing best fit exponential decay curves based on square root transformed trap catch data and then graphing decay curves based on untransformed catch data.

Results

Weekly rainfall, average temperature, percentage relative humidity, and wind speed are presented in Table 1 for the 0-16-week weathering period, the oneweek assessment period after Week 16, the total 0-40-week weathering period, and the one-week assessment period after Week 40. Results of analyses of trap catch data are presented below and in Figures 2-4.

One-week assessments – from Trials 1, 2, and 3. There was a significant difference in weevil catch among doses (F = 3.933; df = 2,36; p = 0.0285), but differences among sites or in the dose-site interaction were not significant. Catch in traps baited with septa with a 1.0 mg lure load had significantly higher catch than in

week weathering	period and the assessment per	riod after the first 40 v	weeks.	man and an and an and an and		
Site	Date	Temp. (°C)	Relative humidity (%)	Wind speed (m/s)	Weekly rainfall (mm)	
Weeks 0, 8, and 2	16					
Weathering	14 Feb5 June, 2012	21.0 ± 0.19	87.8 ± 0.44	0.56 ± 0.04	8.1 ± 0.87	
Assessment	5 June–12 June, 2012	21.2 ± 0.15	88.1 ± 0.46	1.48 ± 0.05	5.7 ± 0.14	
Weeks 0, 8, 16, 2	4, 32, and 40					
Weathering	14 Feb21 Nov., 2012	23.8 ± 0.07	87.5 ± 0.04	1.01 ± 0.08	6.8 ± 0.52	
Assessment	21 Nov.–27 Nov., 2012	20.4 ± 0.06	85.7 ± 0.13	1.78 ± 0.12	4.5 ± 0.19	

traps baited with a 12 µg lure load. The differences in catch between the 1.0 mg and 120 µg treatments, and between the 120 µg and 12 µg treatments, however, were not significant (Figure 2), because of high variability in catch among traps within each treatment. Overall, though, the trend was for increased catch with increased lure load, with catch at traps baited with 120 µg lure loading over four times as great as catch at traps baited with 12 µg lure loading, and traps baited with 1.0 mg lure loading over five times as great as catch at traps baited with 120 µg lure loading and over 22 times as great as catch at traps baited with 12 µg lure loading.

Trial 3. Test of weathering for weeks 0, 8, and 16. There was a significant difference in trap catch among treatments (F= 5.246; df = 8,36; p = 0.0002). Differences related to lure dose were significant (F =9.422; df = 2,36; p = 0.0005), but differences related to lure aging (F = 0.037; df = 2,36; p = 0.9635) or age x dose interaction (F = 1.211; df = 4,36; p = 0.323) were not significant. Average catches are presented in Figure 3. Also presented in Figure 3 is the number of times greater the average catch is at higher loadings relative to lower loadings. Average trap catch dropped to 95.8% and 50.0% of fresh catch after 8 and 16 weeks, respectively, for 12 µg loadings. Average trap catch dropped to 58.6% and 40.4% of fresh catch after 8 and 16 weeks. respectively, for 1.0 mg loadings. Average catch with the 1.0 mg lure was always at least 1.6 times higher than the comparably aged catch with a 120 µg lure and 17 times higher than the comparably aged catch with a 12 µg lure, while average catch with the 120 µg lure was always at least 1.3 times higher than the comparably aged catch with a 12 µg lure.

Trial 4. Test of weathering for weeks 0, 8, 16, 24, 32, and 40. The field sweetpotato weevil population was low during the assessment week of the 40-week

Table 1. Average (± SEM) temperature, percentage relative humidity, wind speed, and weekly rain at sites used for sweetpotato weevil male



Treatment

Figure 2. Effect of lure loading on trap catch over one week of weathering. Average (\pm SEM) male sweetpotato weevil catch per trap per week in sweetpotato fields in the vicinity of Pepeekeo, Hawaii, in traps baited with one of three different loadings of male sweetpotato weevil attractant. Catch results are from the first week following initial trap deployment with five traps for each loading, deployed in a randomized complete block design (average of three separate trials). Bars labeled with the same letter are not significantly different at the $\alpha = 0.05$ level.

weathering trial, resulting in low catch overall. However, catch was adequate to permit some preliminary comparative assessments of septa having the different loadings tested. No fresh, or 8-week or 16week traps caught any weevils in the 12 µg treatment. At the final assessment, average sweetpotato weevil catch in the 1.0 mg treatment was higher than in the 120 µg treatment at every weathering age, ranging from 1.3 to 7.0 times higher catch for weeks for which there was catch in the 120 µg treatment. The calculated exponential decay curve based on square root transformed 1.0 mg sweetpotato weevil catch data was statistically significant (ANOVA results: F = 43.01; df = 1,4; p = 0.0028; r-square of square root transformed data = 0.91). The calculated exponential decay curve, based on untransformed trap catch data was: trap catch = $2.2962 \exp(-0.0365)$ x[no. weeks]) (Figure 4A); The calculated exponential decay curve based on square root transformed 120 µg sweetpotato weevil catch data was not statistically significant at the $\alpha = 0.05$ level, but was statistically significant at the $\alpha = 0.10$ level (ANOVA results: F = 5.28; df = 1,4; p =0.083; r-square of square root transformed data = 0.57). The calculated exponential decay curve, based on untransformed trap catch data was: trap catch = 0.7933exp(-0.0424x[no. weeks]) (Figure 4B). Based on the fitted decay curves, decline



Figure 3. Trial 3 results: Effect of weathering for 0, 8, and 16 weeks on trap catch. Average catch/trap/day of sweetpotato weevils in traps baited with sweetpotato weevil male lure of three different loadings (12 µg, 120 µg, and 1.0 mg) weathered for 0, 8, or 16 weeks. Sweetpotato weevil catch in traps baited with 1.0 mg sweetpotato weevil male lure was significantly greater than catch in traps baited with either 120 µg or 12 µg lure, and there was no significant difference in catch between traps baited with 120 µg versus 12 µg lure. Numbers presented at the tops of the columns are the number of times greater the catch is than in traps baited with lower lure dosages. Error bars are not presented in the figure because high variations in catch within each treatment led to large error bars which would adversely affect the viewing of the data. Mean and error (SEM) data, for the 1.0 mg, 120 µg, and 12 µg treatments, respectively, are as follows: Week 0 (40.31 ± 28.70, 1.86 ± 0.76, 1.43 ± 1.11); Week 8 (23.66 ± 8.32, 14.43 ± 2.34, 1.37 ± 0.85); Week 16 (16.31 ± 4.74, 6.43 ± 2.71, 0.71 ± 0.51).

in attractiveness of the 1.0 mg treatment to 50% of fresh attractiveness occurred at 19.0 weeks while decline in attractiveness of the 120 μ g treatment to 50% of fresh attractiveness occurred at 16.3 weeks, (Figure 4).

Discussion

As expected, higher loadings of septa produced higher trap captures. This

trend has been reported before, but trap captures at traps with higher doses may not be significantly greater in areas where weevil populations are low (Jansson et al. 1991, 1992). The dose-related trap catch increases reported here were greater than reported earlier in McQuate (2014) for the 1.0 mg vs. 120 μ g comparison (5.2 versus 1.9 times greater) and for the 1.0 mg vs. 12 μ g comparison (22.3 versus 8.3



Figure 4. Trial 4 results: Effect of weathering over 40 weeks on trap catch. Decline in sweetpotato weevil catch/trap/week over 40 weeks in traps baited with (A) septum holding 1.0 mg male lure (see text for calculated exponential decay curve), and (B) septum holding 120 μ g male lure (see text for calculated exponential decay curve). Calculated septum age where catch is 50% of fresh catch is presented for each curve.

times greater). The dose-related trap catch increases were, however, comparable in both reports for the 120 µg versus 12 µg comparison (4.3 times greater). McQuate (2014) reported that trap catch could be considerably increased at all of these tested dose levels if the trap includes a green light source in addition to the male lure. The use of light as an attraction modality is made possible because of the sweetpotato weevil's largely nocturnal activity pattern (Shimizu and Moriya 1996a, 1996b). The green light would be positioned in the trap at the point where the lure basket would be inserted with the treated rubber septum held directly under the light, held in place on the end of a firm wire (see Figure 2 in McQuate 2014). Traps which included

a green light source had 4.5x, 2.6x, and 2.2x greater catch than traps lacking a green light source when baited with 1.0 mg, 120 µg, and 12 µg doses of male lure, respectively (McQuate 2014).

The estimated longest cultivation period for a sweetpotato crop along the Hamakua Coast of the Big Island of Hawaii, from planting to harvest (winter months) is 8 months (\approx 32 weeks). Based on weathering rates reported here, traps baited with septa having either a 1.0 mg or 120 µg male sweetpotato weevil lure load would need to have two fresh charges over the course of the production cycle in order to maintain catch at a rate of at least 50% of the catch rate of freshly charged septa. Recommended trap density for sweetpo-

mass trapping may not differ much for the two doses, based on the attractive area of traps estimated by Sugimoto et al. (1994) and Yasuda and Sugie (1990) (as referenced in Sugimoto et al. 1994). Sugimoto et al. (1994), using plastic box traps (modified plastic funnel traps) (Setokuchi et al. 1991) and an area-ratio model (Hartstack et al. 1971), estimated that traps baited with 100 ug of male lure had an attractive radius of 55 m, while traps baited with 400 µg of male lure had an attractive radius of 64 m. They did not test traps baited with 1.0 mg of male lure, but noted that Yasuda and Sugie (1990) estimated that a plastic funnel trap having a 1.0 mg dose of the male sweetpotato pheromone would have an attractive radius of 50 m. This latter estimation was based on the assumption that the effective radius corresponded to the distance from the trap where there was a 30% recapture rate of released weevils. With this estimate of the distance of attractive area, densities of one trap per 0.1 ha should readily provide sufficient coverage for mass trapping purposes when using traps baited only with the male lure. The higher catch associated with the use of the septum with the 1.0 mg male lure load, especially as deployed in a trap also incorporating a green light source, would seem to offer the best potential for effective sweetpotato weevil population suppression in mass trapping programs in Hawaii. However, the economic viability of such an approach is not clear because costs associated both with the 1.0 mg male lure and with a trap incorporating a green light source are not clear. In order to test the potential effectiveness of such a mass trapping system to reduce root damage caused by sweetpotato weevils, we intend to do mass trapping trials utilizing traps which combine both a high-dose olfactory attractant (male lure) and a visual (green light) attractant.

tato weevil population suppression through

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