

## ARTHROPOD MANAGEMENT

### Association of Verde Plant Bug, *Creontiades signatus* (Hemiptera: Miridae), with Cotton Boll Rot

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#### ABSTRACT

Cotton yields have suffered losses from cotton boll rot during the last 10 to 15 years in areas of South Texas. Piercing-sucking insects feeding on cotton bolls, particularly stink bugs, have been implicated in introducing the bacterial disease. Along the Gulf Coast of South Texas, boll-feeding plant bugs occur, and may be associated with the disease. A replicated field survey was conducted in 2010 and 2011 to assess relative abundance of these boll-feeding species and subsequent boll injury caused by cotton boll rot. This survey was paired with a field cage experiment that isolated feeding by the verde plant bug, *Creontiades signatus* Distant (Hemiptera: Miridae). This species represented about 99% of insects collected, during peak bloom (about wk 3 to 4 of flowering) in cotton fields near the coast. It was not detected in fields located further inland. Cotton boll rot was found on up to 25% of open bolls and was concentrated in coastal fields. The proportion of green bolls with cotton boll rot estimated two weeks after insect sampling was not linearly related to verde plant bugs per plant, but the subsequent proportion of open bolls with cotton boll rot near harvest was linearly related to verde plant bugs per plant (adjusted  $r^2 = 0.53$ ,  $P = 0.007$ ). In field cages, verde plant bug-infested plants had significantly higher incidence of insect-punctured bolls (15 to 35%) and disease incidence (5 to 27%), than uninfested plants, when plants were infested for 72 h about

wk 4 of bloom. Diseased bolls tested positive for bacterial contamination. From a pest management perspective, insect monitoring for verde plant bugs provided in-season indication of subsequent boll damage from cotton boll rot, and was especially relevant for cotton fields near coastal waters.

Pest abundance of a complex of stink bugs and plant bugs (Hemiptera: Pentatomidae and Miridae, respectively) has increased in cotton, *Gossypium hirsutum* L. (Malvaceae), during the last 10 to 15 years. Insecticide sprays which indirectly controlled these piercing-sucking insects have been reduced following boll weevil eradication and the adoption of transgenic Bt (*Bacillus thuringiensis*)-cotton (Allen, 2008; Edge et al., 2001).

Stink bugs have been shown to cause damage to cotton bolls: boll abscission, lint staining and loss, and seed loss (Greene et al., 2001; Reay-Jones et al., 2010). Loss is magnified when bacteria causing cotton boll rot are introduced during feeding, as in cases involving the southern green stink bug, *Nezara viridula* (L.) (Hemiptera: Pentatomidae) (Medrano et al., 2007). Along the Gulf Coast cotton-growing region of South Texas, several species of stink bug occur in cotton and neighboring crops such as soybean, *Glycine max* (L.) Merrill (F.). Their abundance and species composition is variable throughout the region (Hopkins et al., 2009). Concurrent with this period of increased boll damage and boll rot, a mirid, the verde plant bug, *Creontiades signatus* Distant (Hemiptera: Miridae), emerged as a threat along the Gulf Coast. Like stink bugs, verde plant bug feeding injury to cotton bolls can result in lint and seed staining (Armstrong et al., 2010). The verde plant bug is a native species along the Texas Gulf Coast and is very similar in appearance to *C. dibilis* Van Duzee and *C. dilutus* Stål. The latter is the primary plant bug pest of cotton in Australia (Coleman et al., 2008). In the cotton growing region of the Texas Gulf Coast, verde plant bug has been collected from weedy plants such as London rocket, *Sisymbrium irio* L. (Brassicaceae), pigweed, *Amaranthus* spp. (Amaranthaceae), and

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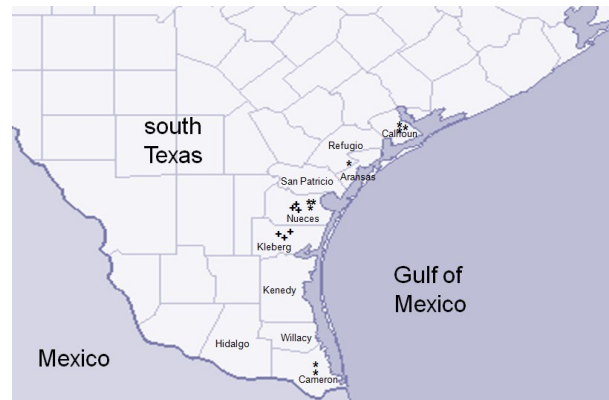
nettleleaf goosefoot, *Chenopodium murale* L. (Chenopodiaceae) (Coleman, 2007). Verde plant bug also has been found during our collecting activities on coastal seepweed, *Suaeda tampaicensis* (Standley), annual seepweed, *Suaeda linearis* (Elliot) (Chenopodiaceae), and cultivated sorghum, *Sorghum bicolor* (L.) Moench (Poaceae). Armstrong et al. (2009b) confirmed that verde plant bug can reproduce on cotton.

To complement greenhouse studies verifying that microbes were associated with verde plant bug feeding injury to cotton bolls (Armstrong et al., 2009c), we investigated whether feeding by verde plant bug was associated with cotton boll rot in the field. A replicated grower field survey was done in 2010 and 2011 along the Texas Gulf Coast to capture a range of piercing-sucking insect species and observe subsequent boll injury, including cotton boll rot. The vast majority of boll-feeding insects collected were verde plant bug, which allowed a presumptive association of this insect's feeding to any cotton boll rot that was subsequently detected. To further strengthen the association of verde plant bug and boll-rotting organisms, a controlled field cage experiment was done to compare characteristics of insect feeding and cotton boll rot on plants exposed to and protected from verde plant bug adults.

## MATERIALS AND METHODS

**Field Survey.** In 2010, stink bugs and plant bugs were collected using a beat bucket during wk 3 to 4 of flowering (about 10 nodes above white flower [Kerby et al., 2010]), from 80 to 200 plants in each of 15 commercial cotton fields. Nine coastal fields and six inland fields were sampled. Fields within 8 km of the nearest coastline, inland bay, or coastal waterway were designated coastal; fields exceeding this 8 km demarcation were designated as inland (Fig. 1). Stink bugs and plant bugs were identified and counted in the field. Two weeks later, green bolls ranging in size from 15 to 27 mm in diameter ( $n=150$ ) were randomly selected from each of 14 of these fields, eight coastal and six inland fields, and brought back to the laboratory to be inspected for internal symptoms of cotton boll rot within the locules (Medrano et al., 2007). More mature bolls, greater than 27 mm in diameter, were not selected because they are less prone to damage by verde plant bug (Armstrong et al., 2009a). The same fields were re-visited near harvest to assess damage to

open bolls. At each field, randomly selected bolls ( $n=150$ ) judged as harvestable (i.e., expected to be fully open in time for mechanical harvest [Jenkins et al., 1990]) were scored for damage using a five class locule damage scale. The scale ranged from 0 (no damage), through an incremental 1 to 3 gradation as damage progressively worsened within each locule and affected additional locules, to 4 (severe damage to all locules) (Lei et al., 2003). Presence or absence of cotton boll rot also was tallied.



**Figure 1. Locations for sampling plant bugs and stink bugs (Hemiptera: Miridae and Pentatomidae) of nine coastal (\*) and six inland (+) cotton fields of the Texas Gulf Coast, 2010 and 2011.**

In 2011, stink bug and plant bug populations were very low but verde plant bug was reported by area consultants to be at potentially damaging levels in two of the 2010 coastal fields near Rio Hondo in Cameron County, and at the Texas AgriLife Research and Extension Center in Nueces County. Drought conditions were severe during 2011: about 8.3 cm of rainfall 1 April 2011 through 30 August 2011 compared with 45.7 cm in 2010 and a 35.5 cm average over 125 years (Corpus Christi station, National Weather Service, 2011). At these fields, verde plant bug was counted during peak to late bloom, and green and open bolls were scored for damage and presence of cotton boll rot as previously described. During both years, fields were planted with multiple cultivars adapted to the region. Insecticides were used in some fields at early squaring for cotton fleahopper control, but sampling did not occur within two weeks of an application.

**Controlled Field Cage Experiment.** In 2010 and 2011 at the Texas AgriLife Research and Extension Center (Corpus Christi, TX), boll injury and cotton boll rot of green bolls from caged plants exposed to verde plant bug were compared to

green boll injury and rot from non-infested caged plants. Adult verde plant bugs used for infesting were obtained from a laboratory colony that was established and periodically replenished with field-collected bugs from several wild and cultivated host plants in the Lower Rio Grande Valley. Green beans and shucked ears of sweet corn provided the food source, harborage, and oviposition substrate for the colony (Armstrong, 2010). Field-collected adults were regularly added to the colony to assure that verde plant bugs with putative boll rot-causing microbes were available for the field cage experiment.

In 2010, infestation rates for the field experiment were 0 (control), 0.25, and 2 verde plant bugs per plant. The treatment cages were replicated 3 to 4 times in a randomized complete block design. Full plant cages made of organza cloth were placed over randomly selected groups of plants (12 plants per cage) when plants were at wk 4 of flowering (about 10 nodes above white flower). This time period corresponded with verde plant bug occurrence in commercial fields. The experiment was repeated in 2011, but infestation rates were changed to 0, 2, and 4 verde plant bugs per plant to further increase the potential for cotton boll rot. Also, plants per cage were reduced to four to avoid plant crowding that was experienced in 2010.

Prior to caging, no stink bug and verde plant bug activity was detected for the previous 3 weeks, and plant inspection also indicated no feeding activity prior to caging. Two d before infestation, all cages were sprayed with short-residual pyrethrins (0.02% by volume, United Industries, St. Louis, MO). Randomly selected adults from the colony were placed in portion cups (1 oz) in the morning and released at the base of the cages by 10 am at the designated infestation rates. Seventy-two hr after the infestation, bugs were killed with a combination of pyrethrins and a longer residual pyrethroid (zeta-cypermethrin, FMC, Philadelphia, PA). Cages were left on the plants to further avoid incidental insect feeding.

When bolls were well developed at about wk 7 of bloom, large green bolls greater than 27 mm in diameter ( $n \geq 40$  per treatment) were randomly selected across replications of each treatment for detailed insect injury and microbiological analyses, according to methods described in Medrano et al. (2007). Bolls were individually surface sterilized for 10 min in a 0.5% sodium hypochlorite solution (Clorox Bleach, Oakland, CA) then rinsed for 2

min in sterile water three times. Macroscopic evidence of insect feeding on the outer and inner boll, and symptoms of infection of lint and seed tissue were recorded after excising carpel walls with a sterile scalpel. Diseased bolls were scored for severity of symptoms using a scale of 0–5 for each locule: 0 (no disease symptoms), 1 (1 to 24%), 2 (25 to 49%), 3 (50 to 74%), 4 (75 to 99%), and 5 (100% rotted tissue). Lint and seed (~0.5 g) from locules with disease symptoms were diced and transferred into a 1.1 ml microtube that contained 0.5 ml  $\text{PO}_4$  (Sigma Aldrich, St. Louis, MO) buffer and a sterile 4 mm stainless steel ball. A second 4 mm stainless steel ball was added, the tubes capped, and the tissue was pulverized using a 2000 Geno/Grinder (SPEX SamplePrep, Metuchen, NJ) for 10 min at 1500 stroke min) then dilution ( $\text{PO}_4$  buffer, pH 7.1) plated on Luria Bertani agar (Becton, Dickson and Company, Sparks, MO) to isolate bacteria. To detect fungal growth, tissue samples were also plated on potato dextrose agar (Becton, Dickson and Company, Sparks, MO) amended with chloramphenicol (100  $\mu\text{g}/\text{ml}$ ) (Sigma Aldrich, St. Louis, MO) and tetracycline (50  $\mu\text{g}/\text{ml}$ ) (Sigma Aldrich, St. Louis, MO) to deter bacterial growth. Seed and lint tissue from bolls without an insect were processed as negative controls and plated on both media described above. After 2 wk of incubation at 28°C, colonies were enumerated and recorded as CFU (colony forming units) per g plant tissue. Subsequent pathogenicity testing and identification of the disease-causing organisms are in progress.

**Data analyses.** To evaluate insect species composition in the field survey, percentages for each species relative to the total number of piercing-sucking insects collected, and total number of boll-feeding piercing-sucking bugs collected was calculated separately for the coastal and inland fields in 2010. A  $\chi^2$  (Pearson's) goodness-of-fit test was used to test equality of the number of piercing-sucking insects collected in coastal and inland fields (adjusted to a single field basis of 120 plants).  $\chi^2$  statistics were calculated using standard formula (Freund and Walpole, 1980) and probabilities calculated by the  $\chi^2$  function of SAS (SAS Institute, 2003).

For fields sampled in 2010 and the two fields in 2011, boll injury data were used to calculate proportion of bolls with evidence of cotton boll rot for green bolls inspected (15 to 27 mm in diameter)

during wk 5 to 6 of bloom and for open bolls inspected near harvest. An average damage score of open bolls (0 to 4 scale) also was calculated. Verde plant bugs per plant during wk 3 to 4 of bloom was calculated using beat bucket sampling data. Three regressions with one quantitative independent variable and one qualitative independent (indicator) variable (Neter et al., 1985) were done using field averages as the data points: a) damage score (y, dependent variable) linear relationship to proportion of open bolls with cotton boll rot ( $x_1$ , quantitative independent variable) and whether the relationship differed between coastal and inland fields ( $x_2$ , qualitative independent variable), b) proportion of green bolls with internal symptoms of cotton boll rot (y) linear relationship to number of verde plant bug per plant ( $x_1$ ) across coastal and inland fields ( $x_2$ ), and c) proportion of open bolls with cotton boll rot (y) linear relationship to number of verde plant bug per plant ( $x_1$ ) across coastal and inland fields ( $x_2$ ). Analyses of residuals showed no regular pattern of deviation from linear regression assumptions; therefore no data transformations or curvilinear functions were considered. The SAS regression procedure was used (Littell et al., 1991).

For the controlled field cage experiment each year, percentages of insect-punctured bolls and locules, number of punctures per boll, percentages of diseased bolls and locules, and amount of disease per boll were calculated for each infestation level. As done above,  $\chi^2$  (Pearson's) goodness-of-fit tests were used to test equality of the frequencies of the measurements among treatments for each year.

## RESULTS AND DISCUSSION

**Field survey.** Plant bugs and stink bugs were much more numerous in coastal fields (mean = 0.61 bugs per plant, ranging from 0.045 to 1.59 per plant) than in inland fields (mean = 0.00 bugs per plant, ranging from 0 to 0.23 per plant) ( $\chi^2 = 75.3$ ,  $df = 1$ ,  $P < 0.0001$ ) in 2010 (Table 1). Verde plant bug averaged 0.42 bugs per plant in coastal fields and its abundance was quite variable among fields inspected (0 to 1.59 bugs per plant). Both verde plant bug nymphs and adults were collected. In comparison, verde plant bugs were not detected in inland fields (Table 1). In collections from the same fields earlier in the season, over 99% of the 216 insects (early-season squaring) and 140 insects (early bloom) were cotton fleahopper, *Pseudatomoscelis seriatus* (Reuter) (Hemiptera: Miridae). Cotton fleahopper feeds on squares and very small bolls, primarily causing abscission (Bell et al., 2006). The rice stink bug, *Oebalus pugnax* (F.) (Hemiptera: Pentatomidae) does not feed on cotton bolls and was likely a transient from nearby sorghum. Excluding these species, verde plant bug represented the vast majority of boll-feeding, piercing-sucking insects in coastal fields (~99% of insects collected) (Table 1). In 2011, drought conditions apparently led to much lower insect activity. Monitoring activity was limited to two coastal fields where verde plant bug was found above 0.10 bugs per plant using a beat bucket ( $n = 120$  plants). Some cotton fleahoppers were detected, but other boll-feeding, piercing-sucking insects were not detected.

**Table 1. Relative abundance of plant bugs and stink bugs (Hemiptera: Miridae and Pentatomidae) detected close to peak bloom (wk 3 to 4 of flowering) using a beat bucket, in coastal and inland cotton fields of the Gulf Coast of south Texas, 2010.**

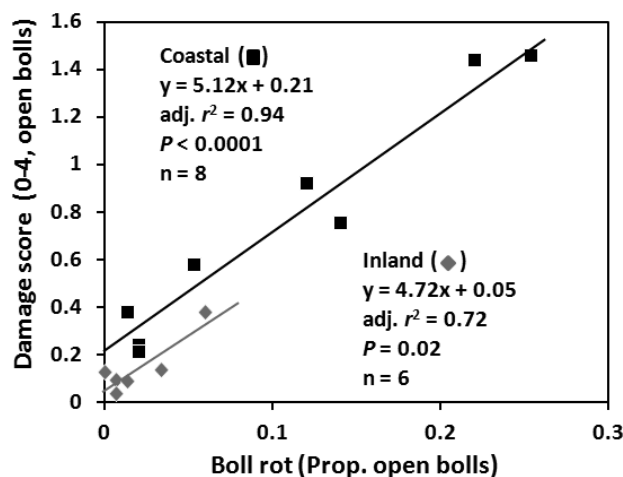
Species <sup>y</sup>	Coastal <sup>z</sup>		Inland <sup>z</sup>	
	No. insects (%)	% boll-feeding	No. insects (%)	% boll-feeding
Cotton fleahopper <sup>x</sup>	258 (30.2)	-	42 (95.5)	-
Rice stink bug <sup>x</sup>	2 (0.2)	-	2 (4.5)	-
Verde plant bug	589 (68.8)	98.9	0 (0)	0
Lygus spp.	2 (0.2)	0.3	0 (0)	0
Green stink bug spp.	5 (0.6)	0.8	0 (0)	0

<sup>z</sup> Coastal fields ( $n=9$ , total plants inspected=1,400) and inland fields ( $n=6$ , number of plants inspected=720).

<sup>y</sup> Scientific names for mirids: cotton fleahopper, *Pseudatomoscelis seriatus* (Reuter); verde plant bug, *Creontiades signatus* Distant; and *Lygus*, not identified to species, but likely *Lygus lineolaris* (Palisot de Beauvois), based on past regional records (Esquivel and Mowery 2007). Scientific names for pentatomids: rice stink bug, *Oebalus pugnax* (F.); and green stink bugs were not identified to species, but were likely a mixture of southern green stink bug, *Nezara viridula* (L.) and green stink bug, *Acrosternum hilare* (Say), based on past regional records (Hopkins et al. 2009).

<sup>x</sup> Species was excluded from the boll-feeding sucking bug calculation.

During both years, cotton boll rot and damage of open bolls was subsequently detected, and the damage was concentrated in coastal fields where verde plant bug was found. Cotton boll rot was found in up to 25% of the open bolls, and it was most common in coastal fields where verde plant bug was detected (Fig. 2). Cotton boll rot was mostly seen on bolls on the upper and outer portion of the plant. This observation was consistent with verde plant bug feeding occurring most frequently on small to mid-sized bolls during peak to late bloom (Armstrong et al., 2009a). There was a strong linear relationship of the damage score of the open bolls to the proportion of open bolls with cotton boll rot ( $F = 156$ ,  $df = 2, 11$ ,  $P < 0.0001$ ). This relationship differed between coastal and inland fields ( $t = -2.78$ ;  $df = 1$ ;  $P = 0.018$ ). Cotton boll rot detected in inland fields never exceeded 8% (Fig. 2). Cotton boll rot resulted in more damage and a stronger relationship to damage in coastal fields ( $y = 5.12x + 0.21$ ; adjusted  $r^2 = 0.94$ ;  $P < 0.0001$ ) than in inland fields ( $y = 4.72x + 0.05$ ; adjusted  $r^2 = 0.72$ ;  $P = 0.02$ ) (Fig. 2).



**Figure 2. Regressions of damage of open bolls near time of harvest (average of a 5 class damage score, 0 [no damage] to 4 [severe damage]) to presence of cotton boll rot near time of harvest (proportion of open bolls with evidence of cotton boll rot) for coastal and inland cotton fields of the Texas Gulf Coast, 2010 and 2011.**

Following disease development in-season was challenging, but the final intensity of cotton boll rot at harvest related back to verde plant bug density estimates taken in-season. The proportion of green bolls with internal symptoms of cotton boll rot during wk 5 to 6 of flowering was not linearly related to the number of verde plant bug per plant estimated during wk 3 to 4 of flowering (adjusted  $r^2 = -0.16$ ,  $P = 0.91$ ). But the subsequent proportion of open bolls with signs of cotton boll rot at harvest time was linearly related to

the number of verde plant bug per plant during wk 3 to 4 of flowering ( $y = 0.193x + 0.057$ ; adjusted  $r^2 = 0.53$ ,  $F = 8.21$ ;  $df = 2, 11$ ;  $P = 0.007$ ) (Fig. 3). In this regression model, a test for differences in the linear relationship between coastal and inland fields was not relevant because verde plant bug was not found in inland fields.

**Controlled Field Cage Experiment.** In both 2010 and 2011, the plant cages were able to considerably restrict boll feeding and subsequent boll damage in cages where verde plant bugs were introduced. The percentage of punctured bolls and locules and the number of punctures per boll were much higher at all infestation levels compared with the uninfested treatment ( $\chi^2 > 14.2$ ;  $df = 2$ ;  $P < 0.0009$ ) (Table 2). The greatest  $\chi^2$  cell contributions came from the uninfested treatment in all comparisons, and differences were not as great across the verde plant bug infestation levels. The subsequent percent diseased bolls and locules, and amount of disease per boll, followed the same pattern of much greater disease detected in the two infestation levels compared with the uninfested plants ( $\chi^2 > 9.1$ ;  $df = 1$ ;  $P < 0.02$ ) (Table 2). All boll sample preparations from diseased bolls plated positive for bacteria (range  $10^2$  to  $10^8$  CFU per g tissue) and fungi were not detected. All reference plates from the uninfested treatment ( $n = 12$  for each year) were negative for microbe detection. The results were consistent with findings of the field survey, which alleviated concerns of the uncontrolled nature of field surveys and caging effects of field cage experiments. Percentage of cotton boll rot detected in the uninfested field cage control was low ( $< 2.5\%$ , Table 2) as was cotton boll rot detected in commercial fields where verde plant bug was not detected (up to 8%, Fig. 2). In contrast, percent of cotton boll rot in the infested cages and commercial fields where verde plant bug occurred was relatively high (up to 27.5% in the cage study and 25% in commercial fields).

Overall, verde plant bug was the dominant boll-feeding piercing-sucking insect species in cotton along the coastal cotton-growing region of South Texas in 2010 and 2011 (Table 1), and substantiated earlier field observations of its activity and damage to cotton (Coleman, 2007). Both nymphs and adults were collected, as expected based on previous reports of verde plant bug oviposition on cotton (Armstrong et al., 2009b). In regard to the strong concentration of verde plant bug in coastal fields, coastal seepweed and annual seepweed growing in saline and alkaline soils were in the vicinity of the cotton fields located near coastal waters. These plants along with weedy annual hosts may have provided a resource for verde

plant bug populations to increase before migration to cotton. We note that verde plant bug was detected in inland cotton fields during other collecting activities, at much lower levels than in coastal fields (MJB & JSA, personal observation).

The relationship of in-season verde plant bug density to early evidence of cotton boll rot in green bolls was poor, but as the disease progressed to near harvest the association of in-season verde plant bug density to cotton boll rot in open bolls was significant (Fig. 3). Isolating verde plant bug feeding further implicated it in introducing cotton boll rot (Table 2), as occurred with the southern green stink bug (Medrano et al., 2007, 2009). The better relationship of verde plant bug densities to evidence of disease when bolls were open, in contrast to the relationship of verde plant bug densities to in-season signs of boll rot in green bolls, was consistent with observations of Medrano et al. (2009). They found that expression of cotton boll rot was not visible in green bolls for several weeks after feeding by southern green stink bug. For those utilizing the practice of opening green bolls, we caution that inspection for early signs of cotton boll rot in green bolls may lead to false readings. The practice may still be useful to verify internal feeding because more mature bolls (> 27 mm in diameter) are less prone to injury (Armstrong et al., 2009a).

From a pest management viewpoint, the dominance of verde plant bug in the region (Table 1), the major contribution of cotton boll rot to harvest-

relevant economic damage (Fig. 2), the relationship of verde plant bug density to subsequent damage of open bolls (Fig. 3), and results from isolated verde plant bug feeding (Table 2) justified in-season monitoring of verde plant bug. On-going economic threshold, boll injury, and pathogenicity work will assist in further quantifying cotton boll rot and harvest risk associated with verde plant bug feeding. Here, we conclude that verde plant bug is associated with cotton boll rot, the association is especially relevant for cotton fields near coastal waters, and in-season verde plant bug monitoring is an indicator of subsequent boll damage.

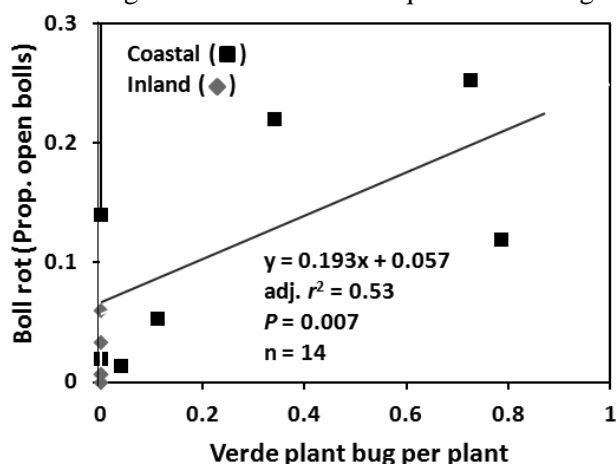


Figure 3. Regression of verde plant bug detected during wk 3 to 4 of flowering to presence of cotton boll rot near harvest (proportion of open bolls with evidence of cotton boll rot) for coastal and inland cotton fields of the Texas Gulf Coast, 2010 and 2011.

Table 2. Field cage experiment comparing insect-punctured bolls and locules and diseased bolls and locules from verde plant bug-infested and uninfested plants, Corpus Christi, Texas, 2010 and 2011.

	Infest <sup>y</sup>	Boll count <sup>x</sup>	Locule count <sup>w</sup>	% punctured bolls	% punctured locules	Puncture per boll	% diseased bolls	% diseased locules	Amount disease per boll
2010 <sup>z</sup>	0	84	341	11.90	2.93	0.12	0.00	0.00	0.00
	0.25	96	387	35.42	17.05	2.69	17.71	9.82	1.28
	2	67	271	43.28	21.96	4.49	16.42	9.23	0.85
	$\chi^2$			14.2 **	46.4 ***	319 ***	14.5 **	32.7 ***	41.3 ***
2011 <sup>z</sup>	0	81	324	2.47	0.93	0.037	2.47	0.62	0.25
	2	40	166	35.00	15.06	1.30	27.50	9.64	0.60
	4	40	160	35.00	18.12	1.08	17.50	5.00	0.42
	$\chi^2$			22.9 ***	46.2 ***	89.7 ***	14.6 **	22.9 ***	9.1 *

<sup>z</sup>Data for each year followed by  $\chi^2$  statistic for test of equality across infestation levels, df = 2, \*  $P < 0.05$ , \*\*  $P < 0.005$ , and \*\*\*  $P < 0.0005$ .

<sup>y</sup>Infest, number of verde plant bug per plant infested for 72 hr starting at about wk 4 of flowering.

<sup>x</sup>Total bolls randomly selected across replications for each infestation level. Used to calculate % bolls punctured and diseased, and punctures and amount of disease (0 [no disease] to 5 [100% rotted tissue] scale).

<sup>w</sup>Total locules inspected in the bolls. Used to calculate % locules punctured and diseased.

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