

CHARACTERIZATION OF BOLL DAMAGE POTENTIAL OF *LYGUS HESPERUS* AND ITS RELEVANCE IN PEST MANAGEMENT

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Abstract

Cotton boll feeding biology and behavior of *Lygus hesperus* has not been fully understood. Boll damage potential of *Lygus hesperus* was determined in a no-choice cup-cage study. Ten cohorts of cup-caged single bolls (10-20 days old) were each exposed to a *Lygus* adult for 48 hours and the boll damages were quantified. After bolls reached 16 days of age, *Lygus* caused very little seed damage, which as expected, also did not result in significant lint yield loss. Cotton bolls were safe from *Lygus* damage when they reached >28 mm diameter or their carpel wall hardness was 0.7 lb per square foot or greater. Cotton boll feeding preferences of *Lygus hesperus*, within-plant boll distribution profile, and *Lygus* damage to cotton bolls at various *Lygus* densities were determined in a whole-plant cage field study. Individually caged cotton plants were exposed to 4 levels of *Lygus* (0, 1, 2 and 4 adults per cage) for one week when plants were at two selected boll development stages (350 and 550 HU after first flowering). When the crop matured from 350 HU to 550 HU after first flower, the percentage of bolls vulnerable to *Lygus* feeding damage was reduced from 53% to 30%. Internal warts were mostly limited to the bolls measuring <35 mm in diameter. In this open-choice boll feeding situation, *Lygus* preferred to feed on bolls that were 10-30 mm in diameter. There were no significant yield differences between control plants and *Lygus* infested plants when plants were first infested with *Lygus* bugs at 550 HU after first flower. A detailed understanding of *Lygus* boll feeding biology and behavior will be highly valuable in improving *Lygus* management decisions during the different boll developmental stages.

Introduction

Cotton, *Gossypium hirsutum* L., is a major cash crop in the U.S. and worldwide. The U.S. is the world's third largest cotton producer and the U.S. cotton industry is valued at more than 25 billion dollars per year. In Texas, approximately six million acres of cotton have been planted annually in recent years, and Texas is the largest cotton producing state (Williams 2011). *Lygus hesperus* is an important economic pest of cotton in some regions of the United States and it is an emerging pest of Texas High Plains cotton. In 2010, a 3.9% reduction in U.S. cotton yields was attributable to arthropod pests – 0.68% due to *Lygus* species, which was ranked third among other yield-reducing pests (Williams 2011) and also cost more per infested acre because multiple applications were often required. In Texas, over 2 million acres of cotton were infested by *Lygus* in 2010 (Williams 2011). *Lygus* can cause severe cotton square loss, anther damage, and seed damage depending upon which crop growth stage the infestation occurs. Both adult and nymphal stages of *Lygus* can inflict damage to cotton fruiting structures. *Lygus* late-instar nymphs are capable of inflicting greater internal damage to maturing bolls than are adults, and this was especially true for 1-2 week old (150-250 HU) bolls (Jubb and Carruth 1971, Parajulee *et al.* 2011). In the Texas High Plains region, *Lygus* generally infest cotton fields during the latter part of the cropping season, thus causing mostly damage to the cotton bolls. Following the introduction of *Bt*-technology (Bollgard cotton), outbreaks of lepidopteran pests have been drastically reduced, and in recent years, secondary piercing-sucking pests such as *Lygus* are of increasing concern to Texas High Plains producers (Parajulee *et al.* 2008).

Cotton boll profiles change as a crop matures, and as a result, the number of *Lygus* susceptible and/or tolerant bolls to *Lygus* damage also change. As boll maturity profiles change, *Lygus* boll selection and feeding behavior may also change which can result in different levels of crop injury and yield loss. There is a strong relationship between boll maturity and *Lygus* feeding damage, thus understanding the boll maturation profile and characterizing *Lygus* damage risk dynamics is very important. Since reliable *Lygus*-resistant or tolerant cotton cultivars are unavailable, cotton producers primarily rely on pesticides for *Lygus* management. Current pesticide application decisions are based on field scouting, whereby spray applications are typically warranted when *Lygus* populations exceed locally established economic threshold (ET) levels.

Oosterhuis and Kim (2004) reported that cotton bolls that accumulated 350-450 heat units were safe from piercing-sucking insects. It is expected that *Lygus hesperus* may also be unable to damage cotton bolls once a certain boll

maturity level has been reached, after which pesticide applications would not be necessary. However, the actual boll damage potential of *Lygus hesperus* is largely unknown. One important question in this study was: At what point do maturing bolls or the entire crop become “safe” from *Lygus* feeding damage, and, consequently, when does insecticide use become unnecessary? Given the availability of tools to identify when the bolls are safe, timing of insecticide use termination may be refined to minimize unnecessary economic and ecological costs.

The objectives of our field experiments were to: 1) determine the maximum potential for *Lygus* to inflict damage to cotton bolls at various boll maturity levels (ages), 2) determine the cotton boll maturity profile during two boll development stages (at 350 and 550 HU After First Flowering [AFF]), 3) determine the boll feeding preference of *Lygus hesperus* adults as affected by the change in boll maturity profile as the crop matures from 350 HU to 550 HU AFF, 4) quantify the yield loss caused by 4 different levels of *Lygus* infestations (0, 1, 2 and 4 *Lygus* adults per plant), and 5) determine the overall yield contribution of cotton bolls from different nodal positions. The overall goal is to better understand the boll feeding biology and behavior of *Lygus hesperus* in order to further develop a dynamic economic threshold for improved *Lygus* management in Texas High Plains cotton.

Materials and Methods

Estimating *Lygus* Boll Damage Potential

A field study to quantify adult *Lygus hesperus* cotton boll damage potential was conducted at the Texas A&M AgriLife Research and Extension Center farm located near Lubbock, Texas. On May 18, 2012, cotton cultivar ST 5458B2RF was planted on 40-inch spaced rows of a furrow-irrigated field. The targeted seeding rate was 56,000 seeds per acre. On June 2, 2012, the entire test was treated with Orthene® 97S for thrips at the rate of 3.0 oz per acre and with Cornerstone Plus® herbicide (41% glyphosate) at 32 oz per acre for weed management.

The experimental design was a split-plot randomized block with three replications. Ten cotton boll age cohorts (10 to 20 days from flowering at 1-day increment) served as the main plot and two *Lygus* infestation levels (I: one adult *Lygus* feeding for 48 hours, and II: control or zero bugs) served as subplots. Thus, there were 30 main plots (3 blocks x 10 boll age cohorts), each of which consisted of 100 ft long cotton rows. In each main plot, 20 randomly selected white flowers were individually cup-caged using modified polystyrene foam and cloth-net “cup cages” (Fig. 1). Thus, a total of 600 white flowers were cup-caged (30 main plots x 20 flowers per main plot). Two treatment levels (control and single *Lygus* infestation) were applied in each main plot. Each plot contained 20 cup-caged bolls of which 5 bolls were used as controls, and the remaining 15 bolls were exposed to *Lygus* feeding. Cotton bolls in the Texas High Plains region typically accumulate 14-30 HU per day in August; thus, in ten days following cup-caging the fruit, on August 20, the August 1st cup-caged bolls received about 450 HU, whereas the August 10th cup-caged bolls had accumulated approximately 200 HU. Once the cotton bolls received 200-450 HU, individual *Lygus* adults were released in the appropriate cages and allowed to feed for 48 hours. *Lygus* adults were initially reared on artificial diet, but were “trained” on fresh green beans and cotton squares for a week prior to using them for the boll feeding experiment. Prior to release into the cup-cages, the *Lygus* adults were starved for 4-5 h. Five *Lygus* infested bolls from each plot were used for boll size, weight, carpel wall hardness and *Lygus* damage assessment (internal and external *Lygus* damage lesions), while the remaining ten *Lygus* infested bolls were kept for yield assessments. Both control bolls and the bolls kept for yield assessment were harvested during the first week of November, 2012.



Figure 1. Deployment of cup-cages. Lubbock County, TX, 2012.

Determination of Boll Maturation Profile, Feeding Preference and Economic Threshold

A field study was conducted to quantify the effect of *Lygus* density and infestation timing on cotton yield and fiber quality. On May 18, 2012, cotton cultivar ST 5458B2RF was planted in a drip-irrigated field with 40-inch row spacing at the Texas A&M AgriLife Research farm located near Lubbock, Texas. The targeted seeding rate was 56,000 seeds per acre. On June 2, 2012, this study was treated with Orthene® 97S for thrips at a rate of 3.0 oz per acre and with Cornerstone Plus® herbicide (41% glyphosate) at 32 oz per acre for weed management.

The field study was laid out in a split-plot randomized block design with three replications, two main plot factors (two cotton boll developmental stages [early boll development and late boll development]), and four subplot factors (four levels of *Lygus* infestation [control or zero bugs, one bug/plant, two bugs/plant, and four bugs/plant]). There were a total of 24 experimental units. Each experimental unit had 8 cotton plants as subsamples (4 used for damage assessment and 4 for yield and quality assessment). A total of 192 whole-plant sleeve-caged cotton plants (three blocks x two cotton boll stages x four *Lygus* densities x eight subsamples) were used for this study (Fig. 2).



Figure 2. Deployment of whole-plant cages. Lubbock County, TX, 2012.

The cotton field study site was closely monitored and kept virtually arthropod pest-free until cages were deployed on July 24, 2012. When the cotton plants reached the target maturity level (350 HU after first flower on August 7, and 550 HU after first flower on August 21), lab-reared *Lygus* were released into the whole-plant sleeve-cages at the rates of 0, 1, 2, and 4 bugs/plant. Again, the *Lygus* adults were initially reared on artificial diet, yet “trained” on fresh green beans and cotton squares for a week before using them for the boll feeding experiment. Cotton plants were exposed to the *Lygus* adults for 6-7 days, after which time, the insects were killed via a pesticide application. Four randomly selected cotton plants from each plot were cut and brought to the laboratory on August 13 and August 21 for the 350 HU and 550 HU plots, respectively. Boll positions, internal and external *Lygus* damage, boll weights, boll diameters, and boll hardness were recorded for all plants from Block 1. For plants from the other blocks, external boll damage, boll weight, and size were recorded. The cotton crop was defoliated by spraying FOLEX® 6EC (12 oz per acre) and a boll opener (Ethephon® 6; 32 oz per acre) in a tank mix on October 3, 2012. After the crop was ready to harvest, the remaining 4 caged plants from each plot, which had been maintained pest-free, were harvested manually to evaluate the lint yields and fiber quality.

Data from the whole-plant cage study were summarized by calculating average and standard errors. ANOVA, GLM model in SAS, 2010 were used to evaluate the treatment effects ($\alpha=0.1$) and treatment means were compared by LSMEAN procedure.

Results and Discussion

Boll Development vs. *Lygus* Damage Potential

During the 2012 active boll developmental stage of August 1-20, the Lubbock area cotton crop received, on average, 24 HU per day and bolls developed rapidly. The diameter of the cotton bolls grew at an average rate of 1.2 mm per day and gained an average of 1.4 grams of weight per day. As the bolls matured and became larger, the carpel walls

became harder as evidenced by the pressure required to puncture the carpel wall, increasing at a rate of 0.018 lb per square foot per day (Fig. 3). When forced to feed on a single boll, each *Lygus* adult inflicted, averaged across all boll age cohorts, 10-28 external lesions per boll in 48 hours. Numerous external lesions were found in all bolls, irrespective of their age. It indicates that in a “no-choice” feeding situation *Lygus* can cause external feeding injury to all bolls, but the actual number of damaged seeds was significantly reduced as bolls became older, bigger and tougher to puncture. When bolls reached an age of 16 days, *Lygus* caused very little seed damage (<2 seeds per boll) that did not result in significant lint yield reductions (Fig. 4). When cotton bolls received >350 HU after first flower (or approximately 16 days of age), they were safe from *Lygus*-induced fiber yield loss. Cotton bolls were observed to be safe from *Lygus* damage when the bolls: 1) exceeded >28 mm in diameter, 2) weighed >14 g, or 3) carpel wall hardness exceeded 0.7 lb per square foot (Figs. 3 and 4).

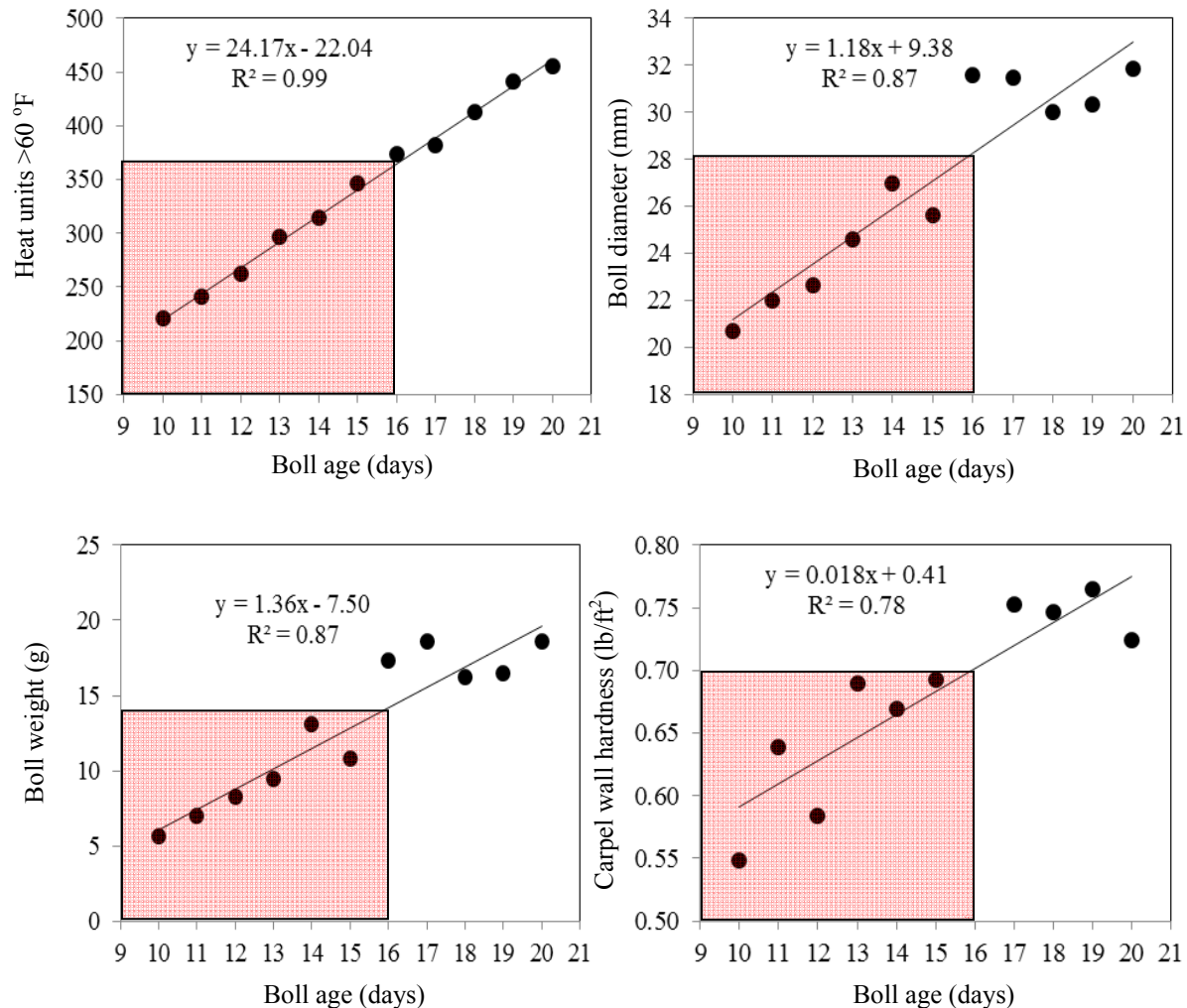


Figure 3. Cotton boll age relationships as associated to heat unit accumulations, boll size, boll weight, and carpel wall hardness. Lubbock County, Texas, 2012.

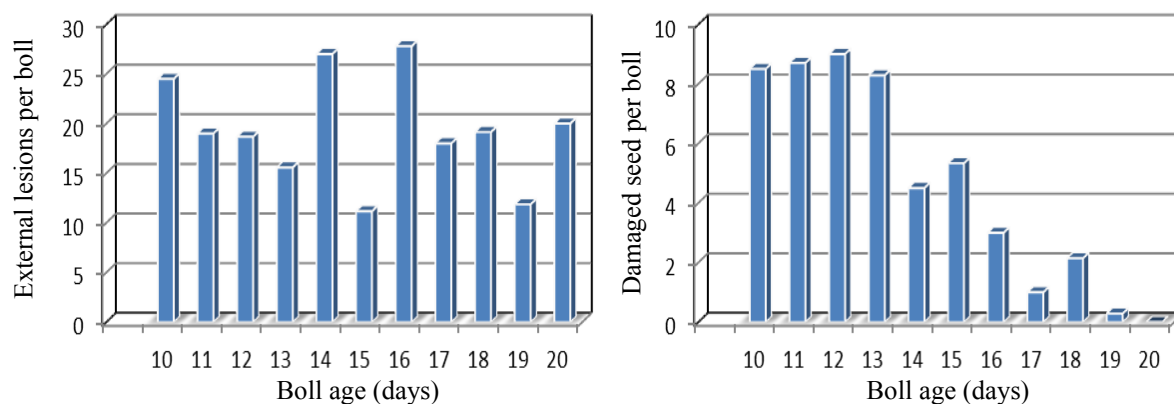


Figure 4. Following 48 hours of feeding by a single *Lygus* adult, boll injury (external lesions and damaged seeds) at various boll ages. Lubbock County, TX, 2012.

Fruiting Profile

At 350 HU after first flower, average of 56.6% fruit retention was observed, but fruit retention was decreased to 36.7% when cotton reached 550 HU after first flower. Cotton plants at 350 HU were observed to have 84% bolls, 14% squares and 2% flowers, while at 550 HU, the cotton plants had 99% bolls, 1% squares, and no flowers. Although there were a higher percentage of cotton bolls on 550 HU plants, the actual number of bolls per plant decreased from an average of 8.8 bolls per plant at 350 HU to 6.3 bolls at 550 HU. Approximately 28.4% of the bolls were naturally aborted from the plants as they matured from the 350 HU to 550 HU stage (Fig. 5).

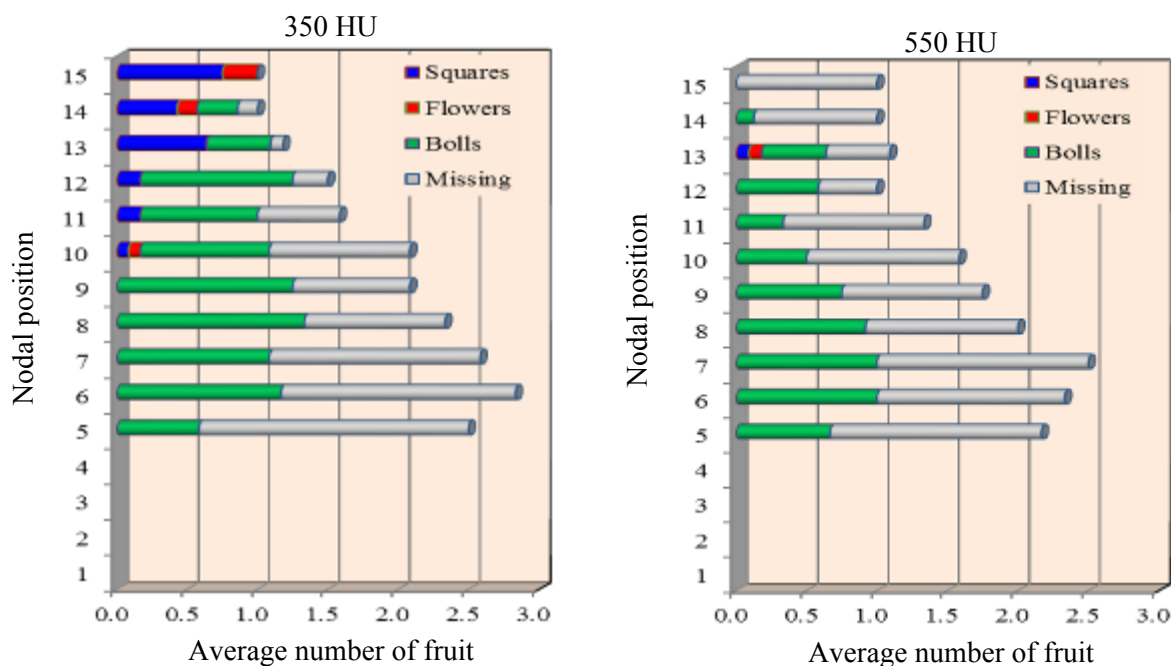


Figure 5. Cotton fruiting profile at 350 and 550 HU after first flower. Lubbock County, TX, 2012.

Most of the bolls were from first fruiting positions of the sympodial branches. At 350 HU, 66%, 24%, 8%, and 2% bolls were from the first, second, third and fourth sympodial branch fruiting positions, respectively; while at 550 HU, 81%, 16%, 3%, and 0% bolls were from the first, second, third and fourth sympodial branch fruiting positions, respectively (Fig. 6). When the cotton plants matured from 350 HU to 550 HU, they dropped all of the 4th fruiting position and most of the 3rd fruiting position bolls. Since 97% of the bolls were on first and second fruiting positions on the cotton plants at the 550 HU stage, our sampling and crop protection efforts should be focused on protecting

primarily the first and second position bolls at this stage. However, fruiting profiles may vary with cotton cultivar, cotton growing region, and crop management practices and input use patterns.

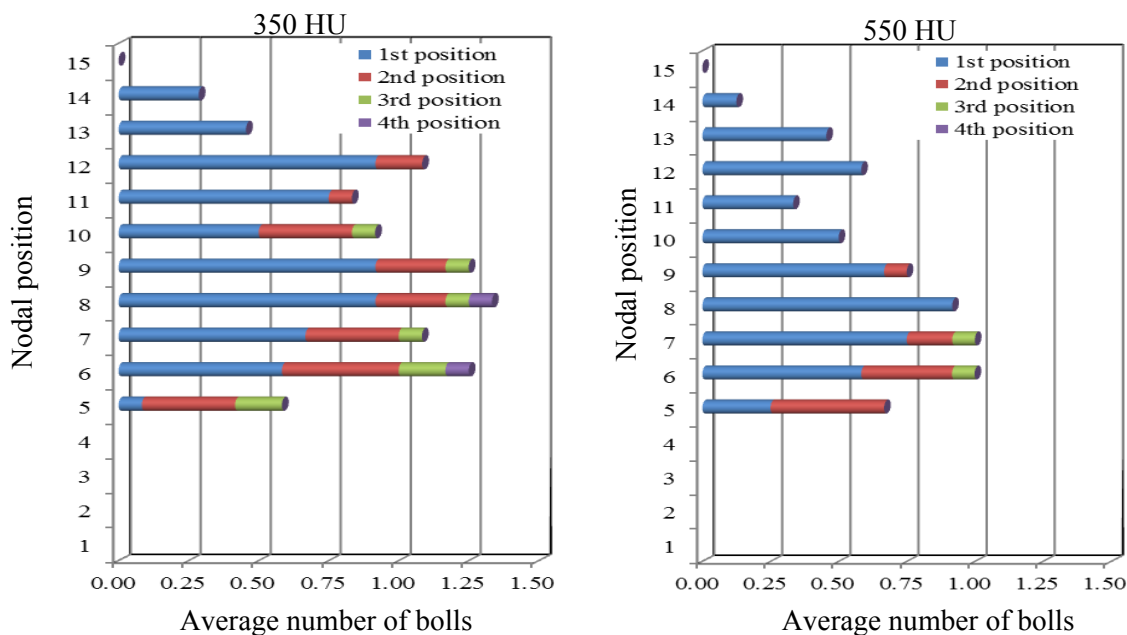


Figure 6. Cotton boll distribution on sympodial branches at 350 and 550 HU after first flower. Lubbock, TX, 2012.

Boll Maturation Profile

Thirty-two cotton plants were harvested (16 plants each from 350 HU plots and 550 HU plots) from which 643 cotton bolls were retrieved. Boll diameter was measured using a Vernier caliper and bolls were categorized into 6 boll size groups (5-10, 11-15, 16-20, 21-25, 26-30 and 31-35 mm). Our past research indicates >25 mm diameter sized cotton bolls are safe from *Lygus* damage. Plants at 350 HU had 47% of the bolls safe from *Lygus* damage (larger than 25 mm diameter), whereas after 2 additional weeks, cotton plants in the same field had 70% of the bolls safe from *Lygus* damage (Fig. 7). When the cotton crop matured from 350 to 550 HU, the proportion of bolls vulnerable to *Lygus* feeding damage was reduced from 53% to 30%. Therefore, it is likely that with a similar level of *Lygus* infestation, *Lygus* may cause a greater amount of cotton yield loss when infested to a mid-season crop (350 HU) compared to that for a late season infestation (550 HU).

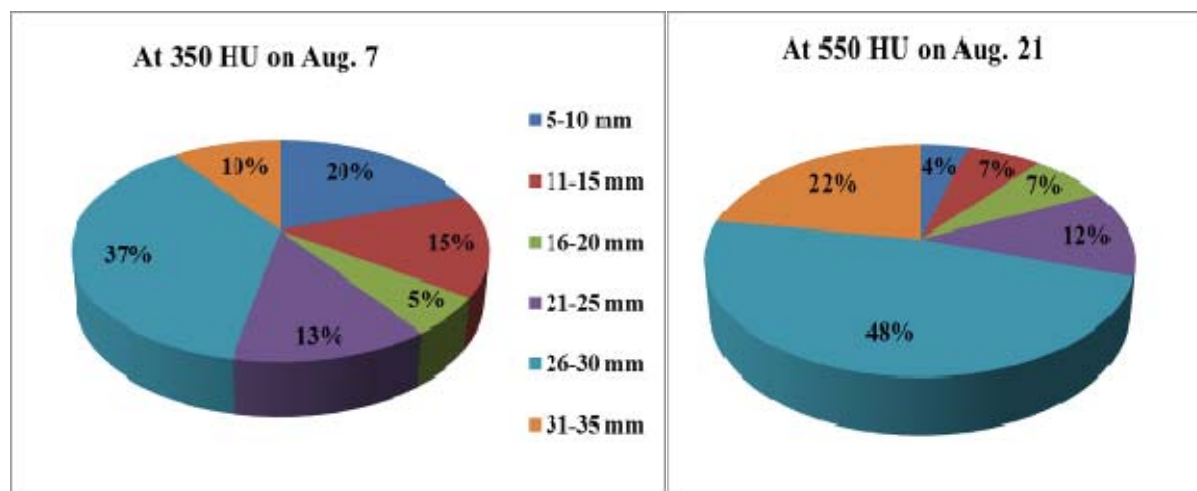


Figure 7. Within-plant boll maturity profile of cotton plants at 350 and 550 HU. Lubbock, TX, 2012.

For our 2012 cotton crop, within-plant cotton boll maturation profile shows that bolls distributed from the 5th to 13th nodes (Fig. 8). At the 350 HU stage, the top 4 bolls (from 10-13th node) were <25 mm diameter size and were vulnerable to *Lygus* damage if bugs were present. When the cotton reached 550 HU, only the top 3 bolls (nodes 11-13) were <25 mm diameter size and therefore vulnerable to *Lygus* damage, if present. Bolls from the 5th to 9th nodes were larger and less vulnerable to *Lygus* feeding damage. There was a very strong positive relationship between boll size (diameter) and the hardness of the boll carpel wall. As we move from the top to bottom nodes of a cotton plant, as expected, we found larger bolls with harder carpel walls (Fig. 8). The vertical boll profile suggests that cotton growers or crop consultants need to focus their *Lygus* damage evaluations primarily during the 350-550 HU, and mostly on the top 3-4 bolls, since they are the most vulnerable to *Lygus* feeding injury.

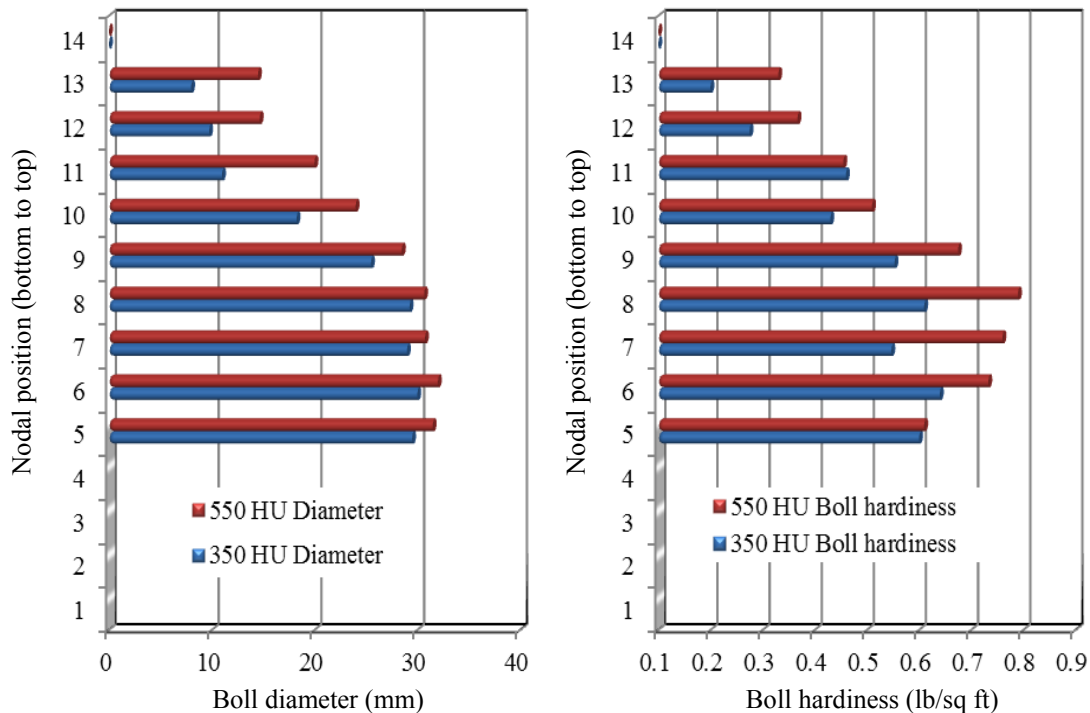


Figure 8. First position boll size profiles of 350 and 550 HU (after first flower) cotton. Lubbock County, TX, 2012.

Lygus Boll Feeding Preference and Boll Damage

In the whole-plant caging study, *Lygus* external feeding lesions were found in bolls of all sizes, indicating *Lygus* attempted to feed on cotton bolls irrespective of boll size. Nevertheless, successful punctures and the resulting internal warts were limited to the bolls <35 mm in diameter. A significantly higher proportion of bolls had internal warts (>20% of bolls) for <30 mm bolls, indicating that in an open-choice situation, *Lygus* preferred to feed on bolls that were <30 mm in diameter (Fig. 9). Cotton plants at the 350 HU had 90% of the bolls measuring <30 mm in diameter, whereas plants at the 550 HU had 78% of the bolls at <30 mm diameter (Fig. 7). The no-choice cup-cage study showed bolls that are >25 mm diameter were safe from *Lygus* damage, whereas in the open-choice whole-plant caging study, *Lygus* preferred to feed on bolls up to 30 mm in diameter. This slight discrepancy might be due to difference in cotton boll development inside cup-cages versus whole-plant cages, or due to differences in *Lygus* behavior in the presence of different boll size options and containments. Evaluation of internal lesions and internal warts suggests there is not a significant relationship between external *Lygus* feeding lesions and actual seed damage due to *Lygus* feeding (Fig. 10), but there were strong relationships between the number of internal warts and number of *Lygus* damaged seed. It clearly indicates that estimating *Lygus* damage by using external lesions can be misleading; therefore, it is best to use the number of internal warts to estimate the degree of *Lygus* crop damage.

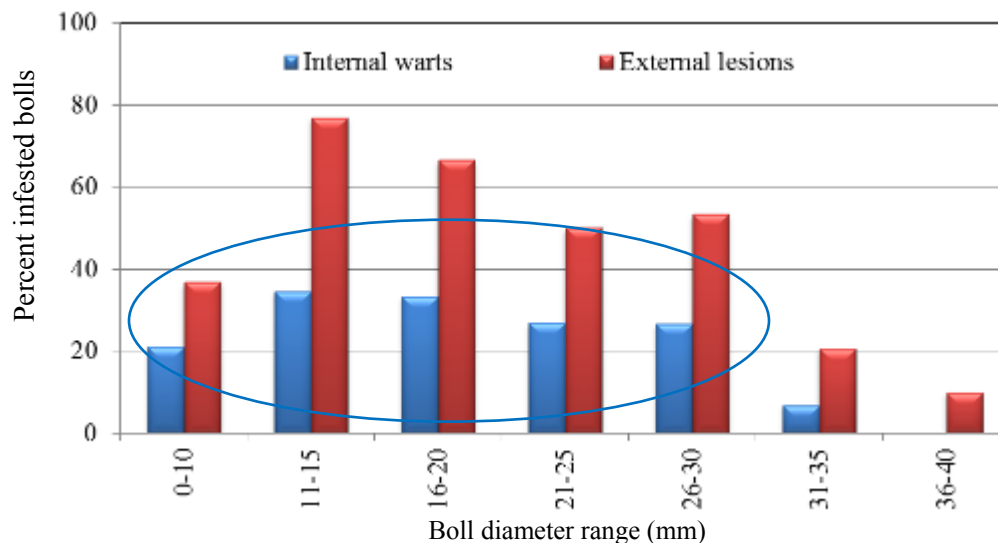


Figure 9. Boll feeding preference of *Lygus* in whole-plant cages based upon the proportion of external and internal boll damage. Lubbock County, TX, 2012.

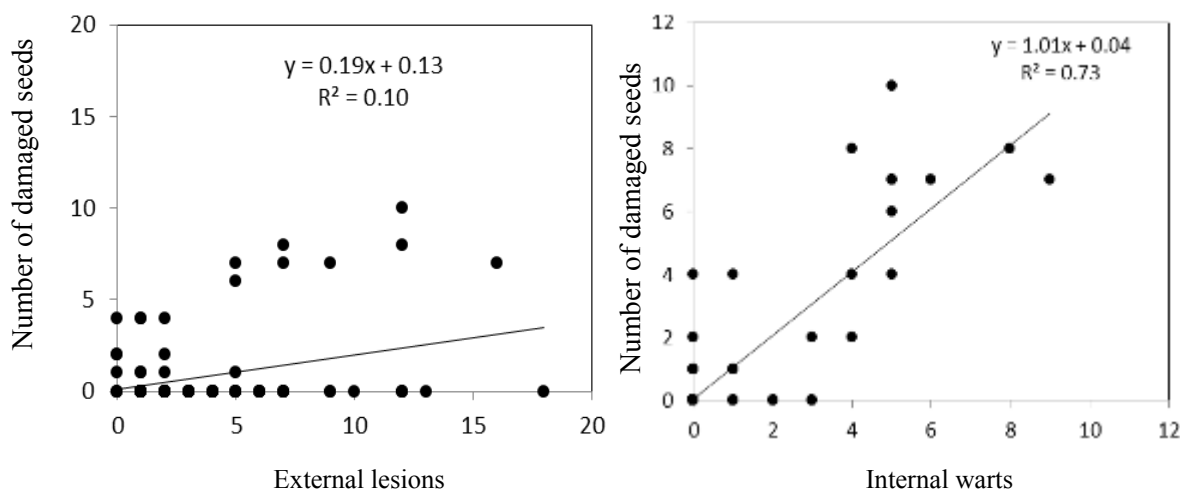


Figure 10. Relationships between the number of damaged seeds per boll and the number of external lesions or internal warts. Lubbock County, TX, 2012.

Yield Loss

Artificial infestation of 2-4 *Lygus* bugs per plant at 350 HU after first flower significantly reduced the cotton lint yield, but the same level of *Lygus* infestation at 550 HU did not result in significant lint yield reduction compared with that in uninfested control plants (Fig. 11). Although it is expected that the degree of yield loss, due to the same levels of *Lygus* infestation, varies with the crop stage the *Lygus* infestation occurred, it was rather a marked change in the crop tolerance to *Lygus* injury from 350 to 550 HU. Because potential yield loss risks due to certain *Lygus* density infestations vary with boll maturation profile, the *Lygus* management economic threshold should be optimized for a dynamic ET to accommodate for within-plant fruit maturity profiles. More detailed research is needed to characterize the interaction between crop phenology and *Lygus* feeding-induced yield loss. Our continuing project is expected to address some of these issues.

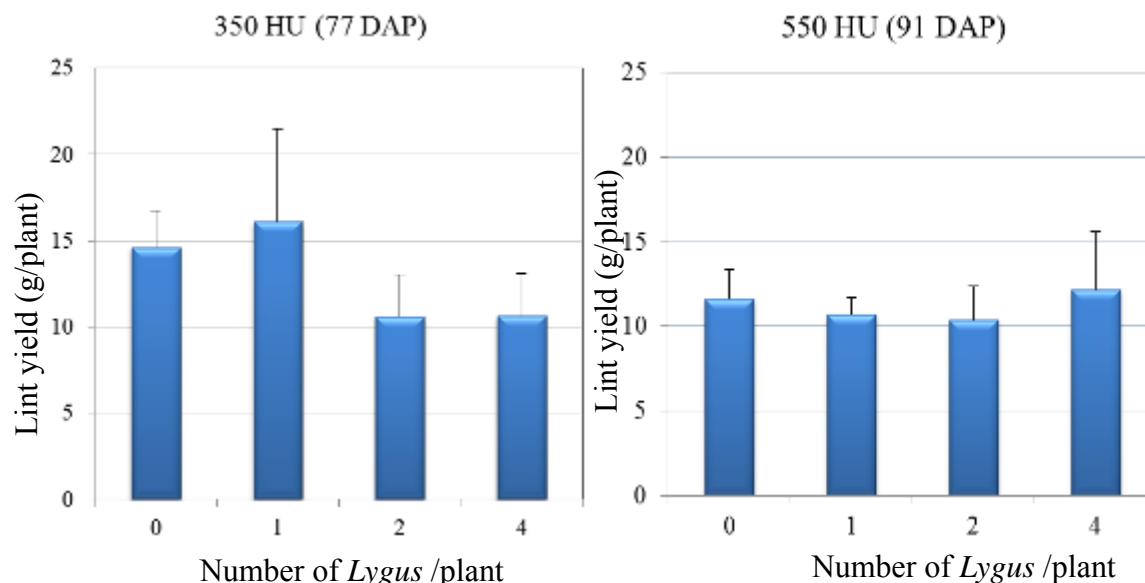


Figure 11. Influence of varying levels of *Lygus* infestations on lint yields at two crop phenological stages. Lubbock County, TX, 2012.

Our pre-harvest complete plant mapping (box mapping) data indicated that the cotton bolls from different nodal positions had different levels of contribution to the total final lint yield (Fig. 12). Nevertheless, bolls from the 5th to 14th nodes cumulatively contributed >95% of the lint yield. Of these 10 nodes, on average, 6-12 node bolls contributed significantly more lint per boll compared to the bolls from remaining nodes.

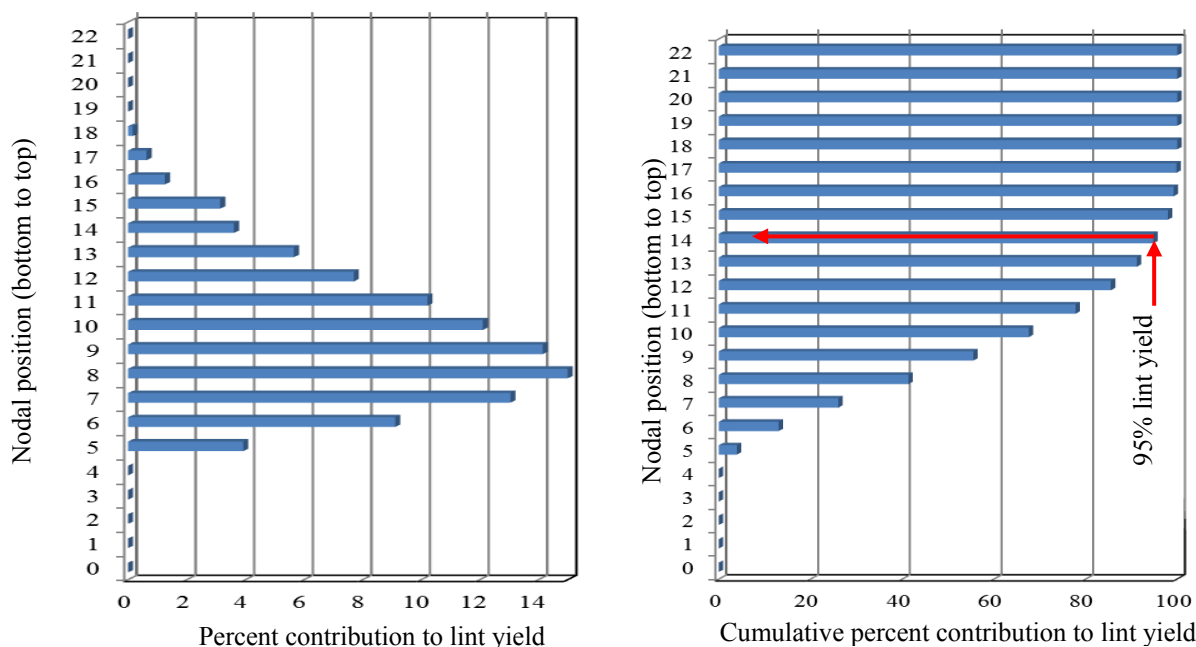


Figure 12. Percent final lint yield contributions of cotton bolls from different nodal positions. Lubbock, TX, 2012.

Summary

There was a significant change in boll composition (boll profile) between the cotton plants at 350 and 550 HU from first flower. Despite a subtle variation between no-choice (cup-caged single boll feeding) versus choice (whole-plant cage with access to all boll types for feeding) situations, it appeared that bolls were relatively safe at 28-30 mm

diameter size or 350 HU, which was approximately equivalent to two weeks old bolls. Cotton boll developmental rates may vary depending on the crop cultivar and crop management system, therefore the interactions between *Lygus* damage potential and other cotton cultivars and various crop management systems need to be investigated to determining the *Lygus* safe boll developmental stages.

Acknowledgements

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