Effect of Planting Date and Density on Insect Pests of Sweet Sorghum Grown for Biofuel in Southern Florida¹

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Abstract There is much research and resource currently being invested in sweet sorghum, *Sorghum bicolor* L. Moench, for biofuel in southern Florida. The objective of this study was to determine the effect of planting date and density on insect pests of the crop in southern Florida. Emergent damage was primarily caused by fall armyworms, *Spodoptera frugiperda* (J.E. Smith), and to a lesser extent by the lesser cornstalk borer, *Elasmopalpus lignosellus* Zeller. Damage at heading was caused by different species of stink bugs (Heteroptera: Pentatomidae). Both emergent damage and numbers of stink bugs at heading varied significantly between planting dates. Correlation analysis showed that planting density had no to little effect on percentage damage by insects to emerging or heading sweet sorghum. Estimated ethanol yield was highest in the first crop of the early planting and decreased thereafter. No consistent effect of planting density or row configuration on yield was shown.

Key Words sweet sorghum, biofuel, fall armyworm, Pentatomidae

Sweet sorghum, *Sorghum bicolor* L. Moench, is a summer annual crop which has potential for use as a biofuel feedstock. Similar to sugarcane, the juice from harvested sweet sorghum stalks can be converted into ethanol using currently available fermentation technology. Ratoon crops are even possible in the mild climate of southern Florida. Currently, there is much research and investment in raising sweet sorghum for biofuel in southern Florida.

The economics and ecological ramifications of biofuels are hotly debated. However, an area that has been greatly overlooked is the monetary and energy cost of controlling insects in biofuel crops. For example, it has been suggested by Cartwright (2008) that increased biofuel production will lead to an increased need for pest control intervention. Also, Martines-Fiho et al. (2006) noted that investing in research and development, including pest control, is one of the most important factors underlying the success and growth of Brazil's sugar/ethanol complex.

An excellent recent example of insect pests in a biofuel crop is *Miscanthus* spp. as discussed by Prasifka et al. (2009). Grasses such as *Miscanthus* X *giganteus* Greef and Deuter ex Hodkinson and Renvoize (a sterile hybrid) and *Miscanthus sinensis* Anderson (a putative *M*. X *giganteus* parent), and switchgrass, *Panicum virgatum* L., are often suggested to be low-input crops because they should require little to no management for insect pests (Parrish and Fike 2005, Semere and Slater 2007, Wang 2007, Clifton-Brown et al. 2008). However, observations of feeding by several orders

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of herbivorous insects on *Miscanthus* spp. (Gottwald and Adam 1998, Prasifka et al. 2009, Bradshaw et al. 2010) suggest rather than being pest free, the identity of insect pests and their effects on harvestable biomass are simply not yet known (Mitchell et al. 2008).

With the expansion of sweet sorghum production in southern Florida, insect pests will become increasingly important. For example, in an earlier study (Anderson and Cherry 1983) 15 sweet sorghum varieties were screened in southern Florida to determine their potential for ethanol production. Infestations of fall armyworm, *Spodoptera frugiperda* (J.E. Smith), occurred in all 15 varieties and ranged from 68 - 100% showing the insect may be an important limiting factor to ethanol production. The objective of this study was to determine the effect of planting date and density on insect pests and yield of sweet sorghum grown for biofuel in southern Florida.

Materials and Methods

Experimental design. One sweet sorghum variety, M81-E, was used in this test. The seeds were obtained from Mississippi State University, Agricultural and Forestry Experiment Station (Starkville, MS) in 2011. The experiment was conducted at Everglades Research and Education Center at Belle Glade, FL, in 2011. The test used 2 different planting dates, early planting and late planting to determine the possible effect of planting date on insect damage. The early planting date was 22 March, and the late planting date was 8 June.

Until specialized harvesting equipment are produced for sweet sorghum, row spacing must be adjusted to match currently available harvesting equipment. Additionally, sweet sorghum stalks must be harvested as whole stalks or cut into smaller billets to prolong moisture and sugar purity prior to milling. Whereas commercial-scale silage corn harvesters are available in our area, the height (3.6 - 4.3 m tall) and density of mature sweet sorghum stalks can result in frequent clogging, and the normal shredding of the stalks by the combine make this equipment unsuitable for this crop. Therefore, single-row commercial sugarcane harvesters that cut the stalks into smaller billets are being considered for harvesting sweet sorghum in our area. Sugarcane is planted on 1.5-m centers to accommodate the harvester with its wide tires. Due to the 0.5-m-wide cutting throat of these harvesters, several rows could be planted adjacent to each other and still be harvested with a single pass of the harvester. Single-, double- and triple-row configurations planted on 1.5-m centers were evaluated in our trial. One of these 3 configurations was used in each plot (Fig. 1). The double-rows were planted 0.4 m apart and the triple-rows were planted 0.2 m apart. In addition to 3 row spacings, seed was planted at 3 different densities: low (60,000 seeds/acre or 148,263 seeds/ha), medium (90,000 seeds/acre or 222,395 seeds/ha) and high (120,000 seeds/acre or 296,526 seeds/ha). One seed density was used in each plot. The final dimensions of each plot were 9.1 m wide with 6 single-, double- or triple-rows 9.1 m long. Plots were separated on all sides by 4 m unplanted soil. This field study was designed in a randomized complete block design with 9 treatments (3 row configurations x 3 seed densities) with 3 replications so that 27 plots were planted at each of the 2 planting dates. Each plot consisted of 1 of 3 row configurations planted at 1 of 3 seed densities. Pesticides were not used during the test.

Agronomic sampling. Stalk counts were made to determine if higher planting densities resulted in higher stalk densities. When heads were at milk stage, we randomly selected 3 locations in each plot to count stalk number. Stalks in a 4-m-long



Fig. 1. Row configurations used in tests. Each of the three boxes represents one $9.1 \times 9.1 \text{ m}$ plot.

section of a row configuration (single, double, or triple rows) were counted at each location. Stalk density means between treatments were analyzed statistically by Least Significant Difference (LSD) test using SAS (2011). Ethanol yield for each of the 9 treatments also was estimated. Stalks were harvested at soft dough stage. Stalks within 4 m of the 2 innermost row configurations of the 6 per plot (Fig. 1) were harvested by cutting at 10 - 15 cm above the soil surface. Stalk number and weight were recorded with intact seed heads in the field to determine fresh biomass yield. Fifteen stalks were subsampled at random from each 4-m-row sample and weighed. Seed heads were excised and the stalks weighed again. These 15-stalk samples were chopped using a modular sugarcane disintegrator (Codistil S/A Denini, Mod: 132S, Piracicaba-SP, Brazil) within 2 h of harvest in preparation for measurement of yield parameters. The chopped plant material was hand mixed in a 200 L barrel, and 2 subsamples were collected from each chopped sample. The first subsample, composed of 900 - 1000 g fresh weight, was used to measure juice percentage and brix. Dry matter yield was calculated from the second subsample composed of 1500 - 2000 g fresh weight. For juice extraction, the first subsamples fresh weight was recorded and then samples were placed in a hydraulic press (Codistil S/A Dedini, Mod: D-2500-II, Piracicaba-SP, Brazil) for 30 sec at 211 kg/cm². Fresh and dry weights (dried at 50°C to constant weight) of the resulting press cakes were recorded. Juice volume and weight were measured from the extracted juice to determine the juice concentration from biomass. Brix concentration (total soluble solids) of juice was determined using a refractometer (Bellingham and Stanley Inc., RFM 91, England). Fresh and dry weights of the second subsample were similarly recorded to determine dry matter concentration. Potential ethanol yields were calculated based on several assumptions. We assumed that 75% of brix were fermentable sugars, 5.6 kg of sugar is equivalent to 3.8 | of ethanol and 95% sugar-to-ethanol conversion efficiency (Smith et al. 1987). Juice extraction percentage (%) equals juice weight / unpressed chopped sample weight x 100. Stalk weights with leaves but without heads were used in the calculation. Yield differences were analyzed by LSD tests (SAS 2011).

Insect sampling. Plots were visually surveyed for significant insect damage twice a week after plant emergence. This damage was first seen 36 d after planting (DAP) in the early planting caused by fall armyworm, *S. frugiperda,* and lesser cornstalk borer, *Elasmopalpus lignosellus* Zeller, as determined by larval identifications. Fall armyworm damage was due to larval feeding causing pinholes in leaves and ragged

leaf edges. Lesser cornstalk borer damage was due to larval feeding resulting in emerging plant deadhearts. Hence, for comparative purposes, this damage was measured at 36 DAP in the early planting and late planting and also 36 d after first harvest in ratoon crops. Plant heights at these times ranged from 30 - 40 cm in the first crops and 50 - 60 cm in ratoon crops. This emergent damage was measured by randomly sampling 20 plants/plot and recording the presence (+ or -) of damage by each of the 2 species for each plant.

Significant insect damage was not observed again in the first crop until heading. Stalk and leaf damage by insects were insignificant at this time. However, large numbers of stink bugs of several species were observed on the sorghum heads. Numerous species of stink bugs (Pentatomidae) are known to feed on the milky stage of grain formation in numerous crops and weeds. Hence, sampling for stink bugs was conducted at the milky stage of grain formation in all 4 crops. Ten plants were randomly selected per plot and sampled by gently bending the sorghum head into a clear plastic bag, shaking the bag, and brushing the head by hand to remove the insects. Thereafter, stink bugs which were easily identified and made up greater than 95% of stink bug adults observed. Stink bug nymphs were not counted because they were < 5% of stink bugs present and are much more difficult to identify to species. Other insects (i.e., chrysomelids, elaterids, etc.) were observed but also not counted because they also were few in number compared with stink bugs. Plant heights at heading in the 4 crops ranged from 2 - 4 m.

A Least Significant Difference (LSD) test (SAS 2011) was used to compare plant damage (emergent) and stink bug populations (heading) between each of the 4 crops. Analysis was made on number of damaged plants of 20 sampled/plot and stink bugs on 10 heads sampled/plot. Means were determined from all pooled samples taken at that time (n = 27 = 3 densities × 3 row types × 3 replications).

Linear correlation analysis was performed on seed planting density versus insect parameters (SAS 2011). Seed planting density was the 3 seed planting densities for each different row configuration of 1, 2, or 3 rows (Table 4). Insect parameters were number of plants out of 20 sampled per plot that were damaged by fall armyworm and lesser cornstalk borer in emergent sorghum and the number of stink bugs on 10 heads sampled per plot in heading sorghum. These were determined in both first and ratoon crops in both early and late plantings.

Results and Discussion

Agronomic sampling. Regardless of the row configuration (single-, double or triple-row), higher planting densities resulted in higher stalk densities subject to insect attack in all 4 crops (Table 1). Yields were highest in the first crop of the early planting and decreased thereafter being comparable in the ratoon crop of the early planting was not harvested due to damage caused by an early frost. In the first crop of the early planting, the highest yield was in the double-row x low seed density treatment. In the ratoon crop of the early planting, the highest yield was in the first crop of the late planting treatment. And, in the first crop of the late planting, the highest yield was in the double-row x medium seed density treatment. These data and other data in Table 2 show no consistent effect of planting densities or row configuration on yield.

Treatments	Early	olanting	Late	planting
(row config. x seed density)*	First crop	Ratoon crop	First crop	Ratoon crop
single-rows × low density	78.7 ± 7.9 a	74.8 ± 5.4 a	77.1 ± 7.0 a	52.6 ± 10.0 a
single-rows × med. density	99.7 ± 7.4 b	82.9 ± 2.5 b	103.8 ± 9.0 b	71.4 ± 7.0 b
single-rows × high density	126.1 ± 7.0 c	88.2 ± 6.0 bc	135.3 ± 6.1 c	90.1 ± 9.6 c
double-rows × low density	84.8 ± 9.1 a	74.4 ± 6.3 a	75.7 ± 8.3 a	65.6 ± 17.2 bd
double-rows × med. density	114.3 ± 6.8 d	90.6 ± 5.3 c	108.3 ± 5.0 b	88.8 ± 17.0 c
double-rows × high density	137.3 ± 7.2 e	103.7 ± 12.3 d	128.0 ± 8.6 d	112.8 ± 16.3 e
triple-rows × low density	86.8 ± 6.8 a	85.0 ± 10.6 bc	88.6 ± 7.7 e	57.0 ± 12.3 ad
triple-rows × med. density	123.7 ± 11.5 c	90.2 ± 5.1 c	122.3 ± 7.1 d	84.8 ± 13.2 c
triple-rows × high density	168.4 ± 13.2 f	104.4 ± 4.6 d	159.2 ± 5.4 f	103.7 ± 11.6 e

Table 1. Stalk densities per 4 m of different row spacing and seed densities.

Means ± SD in a column followed by the same letter are not significantly different at alpha = 0.05 level (LSD test). * Row configurations on 1.5-m centers: single=1 row, double=2 rows separated by 0.4 m, triple=3 rows separated by 0.2 m. Seed densities: low=148,263 seeds/ha, med.=222,395 seeds/ha, and high=296,526 seeds/ha.

Insect sampling. Emergent damage by fall armyworm and lesser cornstalk borer was highest in the first crop of the early planting (Table 3). This is partially due to lesser cornstalk borer damage which was 39% of the damage at this time and was not found in the other later 3 crops. Exact reasons for the lesser cornstalk borer damage at this time, and not others, are not known. However, it should be noted that the first crop of the early planting was sampled at the end of April 2011 which was the second driest month based on rainfall recorded at our station in 2011. Buntin (2009) noted that lesser cornstalk borer is favored by hot, dry conditions. The driest month was November 2011, but this month was cooler than April and was preceded by months of heavier rainfall (i.e., rainy season). In contrast to lesser cornstalk borer, fall armyworm damage was found in all 4 crops ranging from April to November sampling dates. Fall armyworm damage is primarily to foliage, and Buntin (2009) noted that grain sorghum is very tolerant of defoliation and insecticide control seldom justified.

A total of 1060 stink bug adults was counted on sorghum heads during the study (Table 3). Of these, 79% were the rice stink bug, *Oebalus pugnax* (F.), 11% were *O. ypsilongriseus* (DeGeer), 6% were *O. insularis* (Stal), and 4% were the southern green stink bug, *Nezara viridula* (L.). This sequence of relative abundance closely follows that of the 4 species in Florida rice fields where *O. pugnax* is the dominant species followed by the other 3 species in lesser numbers (Cherry and Nuessly 2010). All 4

	Early p	anting	Late planti	ng
Treatments(row config. x seed density)*	First crop	Ratoon crop	First crop	Ratoon crop**
single-rows × low density	2122 ± 134 ab	1138 ± 116 ab	988 ± 23 a	
single-rows × med. density	1998 ± 239 bc	1165 ± 220 ab	1175 ± 170 abc	
single-rows × high density	1749 ± 167 c	978 ± 4 a	1195 ± 236 abc	
double-rows × low density	2659 ± 149 d	1036 ± 328 a	$1425 \pm 309 \text{ cd}$	
double-rows × med. density	2410 ± 108 ade	1264 ± 368 ab	1555 ± 27 d	
double-rows \times high density	2335 ± 176 ae	1188 ± 83 ab	1365 ± 74 bcd	
triple-rows × low density	2190 ± 78 ab	1257 ± 371 ab	1302 ± 183 bcd	
triple-rows × med. density	2300 ± 53 abe	1442 ± 80 b	1211 ± 202 abc	
triple-rows × high density	2614 ± 365 de	1171 ± 53 ab	1119 ± 143 ab	

Table 2. Estimated ethanol yield (L ha⁻¹) of different planting densities and row spacing.

Means ± SD in a column followed by the same letter are not significantly different at alpha = 0.05 level (LSD test). * Row configurations on 1.5-m centers: single=1 row, double=2 rows separated by 0.4 m, triple=3 rows separated by 0.2 m. Seed densities: low=148,263 seeds/ha, med.=222,395 seeds/ha, and high=296,526 seeds/ha. ** Yield data not obtained because of frost damage.

species are pests of rice at seed heading (Cherry and Nuessly 2010). Buntin (2009) noted that *O. pugnax* and also *N. viridula* attack sorghum at seed heading. It is likely that *O. ypsilongriseus* and *O. insularis* were causing the same damage. The relative abundance of *O. ypsilongriseus* to *O. pugnax* increased over time so that in the last 2 sampling dates the ratios of *O. ypsilongriseus* to *O. pugnax* was 8 - 1. This corresponds to Cherry et al. (1998) who reported that *O. ypsilongriseus* increased relative to *O. pugnax* in Florida rice fields from May to November.

Adult stink bugs were most abundant on sorghum heads in the first crop of the early planting which was sampled 18 July. July is the period of maximum flight activity

Early Planting	Emergent*	Heading**
First crop	13.4 ± 2.5 A	25.4 ± 22.5 A
Ratoon crop	10.7 ± 1.9 B	3.4 ± 2.5 C
Late Planting	Emergent	Heading
First crop	4.6 ± 2.2 D	11.9 ± 14.3 B
Ratoon crop	$6.6 \pm 2.0 \text{ C}$	0.4 ± 0.6 C

Table 3. Insect parameters in different sweet sorghum crops.

Means ± SD in a column followed by the same letter are not significantly different at alpha = 0.05 (LSD test).

* Number of plants damaged per 20 plants/plot.

** Number of stink bugs per 10 heads/plot.

			Early D	anting		
		First crop		Bunna	Ratoon crop	
	single-rows	double-rows	triple-rows	single-rows	double-rows	triple-rows
Emergent**	-0.75	-0.23	-0.26	-0.10	0.67	-0.30
Heading⁺	-0.12	0.39	-0.22	-0.73	0.23	-0.42
			Late PI	anting		
		First crop			Ratoon crop	
	single-rows	double-rows	triple-rows	single-rows	doub!e-rows	triple-rows
Emergent**	-0.07	-0.10	-0.40	-0.07	-0.32	0.24
Heading⁺	0.48	0.25	0.52	-0.32	-0.44	-0.27
* Three planting dens	ities times 3 replicates (n -	= 9) occurred within each of t	he 24 row configurations	. A row configuration is sin	gle row or double row or trip	ole row (See text for

Table 4. Correlations (r)* of seed planting density of different row configurations with insect parameters.

details). Correlations greater than \pm 0.66 are significant (alpha = 0.05).

** Total number of plants damaged per 20 plants/plot correlated with three planting densities.

[†] Total number of stink bugs on 10 heads/plot correlated with three planting densities.

in southern Florida for the 4 species of stink bugs in this study (Cherry and Wilson 2011). The insects were second most abundant in the first crop of the late planting sampled 19 September. Stink bugs were least abundant in both ratoon crops which were sampled at later dates. The declining stink bug numbers at sorghum heads from the July maximum also corresponds to flight activity reported by Cherry and Wilson (2011) in light trap catches.

Plant spacing may affect plant parameters such as plant density, vine size, plant size, etc. which may, in turn, affect resistance of plants to insect damage (Smith 1989). For example, Buntin (2009) noted that higher seeding rates make stand losses in grain sorghum less severe to soil insects than in corn. Correlations of planting density of sweet sorghum with insect parameters are shown in Table 4. Of 24 correlations, only 3 were statistically significant. Of these 3, 2 were negative correlations and 1 was a positive correlation. These data show that planting density had no to little effect on percentage damage by insects to emerging or heading sweet sorghum.

Lastly, although not the objective of this study, the potential damage of the insect pests observed in this study should be noted. The fall armyworm is recognized as an economically important pest of grain sorghum throughout the Americas (Andrews 1980, Ashley et al. 1989). Andrews (1988) reported that whorl infestations of fall armyworm reduced grain yields of susceptible sorghum lines by 55 - 80%, and that stand loss by their feeding on 13- to 22-d-old sorghum plants caused 50% yield loss. Grain vield reductions of 76 - 85% due to whorl feeding by fall armyworm have been observed on the susceptible sorghum line 'Huerin Inta' (Diawara et al. 1991). The Pentatomidae (Heteroptera) Chlorochroa ligala (Say) and N. viridula, and the Coreidae (Heteroptera) Leptoglossus phyllopus (L.), have also been shown to cause reductions in yield and germination of sorghum seed (Hall and Teetes 1982). The mean density of the four most common stink bugs collected in the current study (0.04 - 2.5 adult stinkbugs per panicle) were < 50% of those determined to cause grain damage during the milk stage by O. pugnax, N. viridula, or L. phyllopus by Cronholm et al. (2007). Whereas yield losses in grain sorghum are tied to effects on grain production, economic damage to sweet sorghum grown for bioenergy production would likely result from reduction in biomass and sugar. Therefore, economic injury levels in grain sorghum may not be well correlated with losses in sweet sorghum. We are not aware of any economic injury levels published for sweet sorghum. A manuscript in preparation by the authors of the current study will compare insect density and damage, biomass, sugar and potential ethanol yields between control and insecticide-protected plots for 8 sweet sorghum varieties.

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