

## AGRICULTURAL ABSTRACTS

### **Green Cane Trash Blankets: Influence on Ratoon Crops in Louisiana**

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Approximately 75% of Louisiana's 2000 sugarcane crop was harvested with a chopper harvester. A significant portion of the chopper-harvested sugarcane was harvested green, especially early in the season. Information on the impact of the post-harvest, green-cane residue blankets on subsequent ratoon crops is inconclusive, but yield reductions have been reported. To insure maximum yields, the residue is generally removed by burning during the winter months when weather conditions are more favorable in reducing the likelihood the smoke will offend the public. The effects of residue blanket management methods on ratoon crops were studied following the 2000 harvest. In one study, burning the residue in January resulted in higher (14%) sugar yields of first-ratoon LCP 85-384 compared to the no removal treatment. Delaying the burning of the residue until February or March did not significantly improve sugar yields over the no removal treatment. In a second study designed to evaluate varietal responses to dates of residue removal, first-ratoon crops of CP 70-321, LCP 85-384, HoCP 85-845, and HoCP 91-555 were found to respond similarly to the removal of the residue. The average sugar yield (6.6 Mg/ha) for the four varieties was 11% higher than the no removal treatment (5.9 Mg/ha) when the residue was removed in early January, regardless of whether the residue was mechanically removed to the row sides or completely burned off. When burning was delayed until March, the average sugar yield (5.3 Mg/ha) was 10% lower than the no removal treatment suggesting that some damage to the emerged shoots was occurring with the later burn. Soil temperature and soil moisture readings taken early in the growing season (January to April, 2002) indicate that the soil is colder and wetter under the blanket of residue. The cold and wet soil condition created by the thick blanket of residue may be affecting crop emergence in the spring and ultimately sugar yields.

### **The Effect of Combine Speed on Cane Quality at Alma Plantation in 2001**

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The parallel acceptance of a new sugarcane variety LCP 85-384 and the use of combine harvesters have significantly redefined the Louisiana sugarcane industry in recent years. The importance of high quality cane deliveries has been emphasized due to the new harvest method and the challenges faced by raw sugar

processors. This study was conducted to help determine the influence of forward speed on cane quality. Alma Plantation in Lakeland, LA agreed to participate in the experiment throughout the 2001 harvest season. Weekly sampling was conducted using the same operator and a 2000 model 7700 Case Combine Harvester. The extractor fan speed was 900 to 950 rpm in burned cane and 1100 rpm in green cane. The treatments (speeds) were 1.5, 2.5, 3.5 and 4.5 mph and were monitored with a handheld radar unit to ensure accurate ground speed. For 12 consecutive weeks, one truckload was cut at each speed and delivered to the mill to be weighed and sampled using the mill's core sampler. While the delivered tons of cane per acre was significantly less when the combine was slowed down to 2.5 and 1.5 mph, the pounds of sugar per ton of cane was only higher in the 1.5 mph treatment as compared to 3.5 and 4.5 mph ( $P = 0.05$ ). There was no significant difference in the resulting yield of pounds of sugar per acre between the treatments. The 4.5 mph treatment had the highest fiber % cane, but sediment readings were not significantly different among treatments. When the mill's incentive formula was applied to the yield results, the 1.5 mph treatment received a bonus of 3.36 pounds of sugar per ton of cane which was only significantly greater than the -1.57 pounds of sugar per ton of cane for the 4.5 mph treatment. The data demonstrates that forward speed of the combine harvester has a significant influence on delivered cane yield and quality. Practical application of this information could be used to determine other optimal combine settings to improve cane quality from combine-harvested sugarcane in Louisiana.

### **Use of Cover Crops in Rotation with Sugarcane in a South Florida Mineral Soil**

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The establishment of cover crops (grasses or legumes) prior to planting sugarcane (interspecific hybrids of *Saccharum* spp.) offers many potential agricultural and ecological benefits to the grower. These benefits include organic matter production to enrich the soil, ground cover to reduce windblown soil erosion, weed control (including less herbicide use), reduced runoff, improved infiltration, soil moisture retention, and soil tilth, nutrient enhancement, and food for wildlife. By improving soil organic matter, cover crops directly influence the soil water holding capacity by increasing water retention and lateral water movement within the soil. Rotation of susceptible agronomic crops with crops that are not nematode pest hosts or are resistant to certain nematodes has been a successful nematode management strategy. The objective of this study was to evaluate the impact of eight cover crops on sugarcane grown on sandy soils. Cowpeas, *Aeschynomene*, Hairy indigo, Sorghum sudangrass, Sterile sorghum, Sorghum sudan/cowpeas mixture, Japanese millet, and Tifleaf millet were planted in April 1992-1994 in 0.25 to 1.2 acre (0.10 to 0.50 ha) plots. Cover crop biomass was measured in August of each year, followed by sugarcane planting in September, which was subsequently harvested in November of the following year (1993-1995). Cover crop yield was significantly higher for the grasses than for the legumes in 1993 and 1994. Cool temperatures and flooded fields during the establishment period resulted in thin stands and low yields of the

cover crops. *Aeschynomene* had the best ground cover (46%) of all cover crops. Cowpeas did not tolerate periods of standing water, indicating that this crop should be planted on drier sites. Japanese millet, which tolerates wet field conditions, should not be planted until late April or early May to prevent early (within 21 days of planting) seedhead emergence. The optimum time to plant warm-season cover crops may be early May, so that at least 4 months of growth are obtained before sugarcane is planted. In the 1993-1995 crop, sugarcane yield (tonnage and sucrose content) obtained for *Aeschynomene* was numerically higher than for all other cover crops treatments and the control treatment (fallow field with no cover crop planted with sugarcane). However, significant differences (Fisher's protected L.S.D. test,  $P=0.05$ ) for sugarcane yields were only obtained between the *Aeschynomene* treatment and the Sorghum sudangrass and the Sorghum sudangrass/cowpeas mixture.

### **Evaluation of Sorghum-Sudangrass Hybrids for Biomass Potential in Southern Louisiana**

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As close relatives of sugarcane, sorghum-sudangrass hybrids are easy to establish (seed propagated), could be used as an interim crop (April - July) during the fallow season, and may have potential as an complimentary bioenergy crop. Ten sorghum-sudangrass (*Sorghum bicolor* x *S. bicolor* var. sudanese) hybrids were evaluated for biomass potential at the site of the USDA-ARS Sugarcane Research Unit in Houma, Louisiana. The experiment was designed to be largely observational with single-row unreplicated plantings. Beginning 14 May and continuing weekly through 10 July (nine weeks), 10-stalk samples of each hybrid were collected and analyzed to obtain fresh weight, dry weight, and Brix estimates. One of the hybrids known to be photoperiod sensitive, was non-flowering, and therefore expressed an indeterminate growth habit, continuing to increase in weekly cumulative dry matter content through the end of this experiment. At 97 days following planting (4 Apr 2001 – 10 Jul 2001) the nine hybrids with determinate growth habit, averaged 3 tons green matter/acre, 0.80 tons dry matter/acre, 8.5 Brix, and just over 7 ft height. By contrast the non-flowering hybrid achieved 8 tons GM/acre, 1.75 tons DM/acre, 6.7 Brix, and reached 12 ft height. During 2002, the bioenergy potential of this non-flowering hybrid will be entered into a sorghum test at Houma and directly compared with sorghum varieties considered for commercial bioenergy production in sugarcane-growing areas of Southwestern Louisiana.

## **ENVOKE: A New Herbicide for Weed Control in U.S. Sugarcane**

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Envoke® [N-(4,6-Dimethoxy-2-pyrimidinyl)carbamoyl]-3-(2,2,2-trifluoroethoxy)-pyridin-2-sulfonamide sodium salt] is a new broad-spectrum, post-emergence herbicide that Syngenta Crop Protection is developing for use in sugarcane, cotton, citrus and almonds. It has been field tested as a 75% water dispersible granule for several years in North America, South America, Africa, and Asia under the code name CGA-362622. The proposed common name is trifloxysulfuron-sodium. Envoke® will offer control of certain broadleaf, sedge, and grass weeds in cotton, sugarcane, citrus, and almonds including yellow nutsedge, purple nutsedge, flatsedge, redroot pigweed, spiny pigweed, pitted morningglory, ivyleaf morningglory, scarlet morningglory, hemp sesbania, cocklebur, sicklepod, broadleaf panicum, spurge, spanish needles, and horseweed.

In sugarcane, 0.3 - 0.6 ounces product/A (15.8 - 31.6 g ai/ha) of Envoke® can be applied post-emergence, depending on cultivar, with excellent crop tolerance. For optimum post-emergence activity, the addition of NIS is recommended at 0.25% v/v. The very low use rate of 0.3 to 0.6 ozs/A together with its favorable toxicological, ecotoxicological and environmental properties make Envoke® an excellent tool for sugarcane farmers. Envoke® is readily absorbed by shoots and roots and is readily translocated in weeds. Susceptible weeds are inhibited following an application of Envoke® with complete death occurring within 1 to 2 weeks after application.

Envoke® is compatible with other herbicides including AAtrex® and Evik® which can be used to increase the weed spectrum and duration of control. Envoke® can be applied in combination with Evik®, post-directed only, to increase speed of activity and weed spectrum, especially the grasses.

## **Experimental Products for Weed Control in Florida Sugarcane**

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Several new herbicides are being evaluated for weed control in Florida sugarcane. Both pre-emergence (PRE) and post-emergence (POST) herbicides are being evaluated. Control of a wide range of common weeds, including fall panicum, broadleaf panicum, alligator weed, purple nutsedge, yellow nutsedge, and several other species is being evaluated. The PRE products in testing include flumioxazin and azafenidin, applied alone or in conjunction with labeled PRE herbicides. These treatments are being evaluated in comparison to standard PRE treatments. POST products under evaluation include carfentrazone, trifloxysulfuron, and flumioxazin. These products are being evaluated both alone and in conjunction with standard POST treatments, such as asulam, atrazine, halosulfuron, and ametryn.

Early results indicate potential for good control of a range of weeds utilizing these new products alone or in tank-mixture with currently labeled products. Detailed results will be presented during the conference.

## **Effect of Calcitic Lime and Calcium Silicate Slag Rates and Placement on LCP 85-384 Plant Cane on a Light-Textured Soil**

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Substantial sugarcane yield responses to silica application have been documented in Florida and Hawaii, but not in Louisiana. Our research determined the effect of calcitic lime and calcium silicate slag rates and placement on plant cane yields grown on a light-textured soil in Louisiana. Results showed that mixing 2.24 Mg ha<sup>-1</sup> and 4.48 Mg ha<sup>-1</sup> of calcium silicate slag into soil before planting, or placing 2.24 Mg ha<sup>-1</sup> of slag under cane at planting resulted in higher (P#0.10) sugar yields compared to the check. Mixing 2.24 Mg ha<sup>-1</sup> and 4.48 Mg ha<sup>-1</sup> of calcitic lime, however, into the soil before planting did not increase (P#0.10) sugar yields. Higher sugar yields obtained with calcium silicate slag vs. calcitic lime indicates that the yield response obtained with calcium silicate slag was due to its silica content.

## **Sugarcane Leaf P Diagnosis in Organic Soils**

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Most of the sugarcane production in south Florida is on organic soils. Phosphorus is an essential plant nutrient that contributes to optimum sugarcane yields, but producers are required to reduce P levels in waterways. One way to monitor P nutrition is through leaf diagnosis. The objective of this study was to determine the best time to leaf sample during the summer months and to relate optimum leaf P tissue content and yield. A 3-year field study was conducted on four organic soil locations in south Florida. An 8 by 3 factorial experimental design with four replications was used at each location with eight sugarcane (interspecific hybrids of *Saccharum* sp.) genotypes in combination with three fertilizer P rates (0, 24, and 48 kg P ha<sup>-1</sup>). Fertilizer rates were based on soil test analysis with 24 kg ha<sup>-1</sup> being the recommended rate. Upper-most fully expanded leaves were sampled in early, mid, and late summer prior to three harvests (plant cane, first ratoon, and second ratoon). Two locations had optimum cane and sugar yields at 24 kg P ha<sup>-1</sup> for all harvests. There was no response to P fertilizer at one location for any harvest year, while the other location had the highest cane yields at 48 kg P ha<sup>-1</sup> for all harvests. Analysis of variance for leaf P content showed significant interactions for location by P rate by harvest and for location by P rate by leaf-sample time. Leaf P content did not always correspond to yield data. Within each location, sometimes the leaf P content increased with increasing P rate as did yield, and sometimes yields did not show a response to P fertilizer even though leaf P increased. Consistent patterns in time of leaf sampling within locations could also not be obtained. Correlation analysis of yield vs. leaf P content across all treatment in early and mid summer were statistically significant ( $P < 0.05$ ), but coefficients were very low ( $r = 0.14$  and  $0.26$ , respectively). Correlations of harvests within location at each leaf sample time were occasionally significant ( $P < 0.01$ ) with the highest correlation of  $r = 0.79$ . But, there was no consistent pattern relating leaf P tissue content with yields. Optimum leaf P tissue content should be calibrated for each field, harvest, and sampling date for precision agriculture applications.

## **Wireworm Effects on Sugarcane Emergence After Short-Duration Flood Applied at Planting**

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Sugarcane (interspecific hybrids of *Saccharum* spp.) growers in Florida normally apply a soil insecticide at planting to limit wireworm (*Melanotus communis* Gyll.) damage to planted stalk sections. Long-

duration floods prior to planting sugarcane are also used to control wireworms. A recent study found that sugarcane emergence was improved by floods of 2-12 days applied at planting. The purpose of this study was to analyze sugarcane emergence after floods of 7, 14, and 21 days applied at planting, as well as following a conventional application of an organophosphate insecticide at planting without flooding. In three outdoor experiments, wireworms were applied at the severe rate of 13 larvae per meter of row in plastic containers filled with Pahokee muck soil. In the first experiment, emergence under the flood treatments was lower than under the insecticide treatment, probably due to lower than normal air and soil temperatures. Emergence in the 14- and 21-day flood treatments and the insecticide treatment were similar in the final two experiments. However, reductions in plant weight were associated with some flood treatments. Previous work reported that wireworms damaged growing plants in containers, but damage was primarily limited to reduced emergence in field studies. The successful wireworm control of the 14- and 21-day floods and the negative effects on plant weights reported in this study need to be verified in field studies.

### **Laboratory Screening of Insecticides for Preventing Injury by the Wireworm *Melanotus communis* (Coleoptera: Elateridae) to Germinating Sugarcane**

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A laboratory bioassay was investigated for screening candidate materials for preventing stand losses by wireworms in germinating plant cane. For liquid materials, single-eye billets were dipped into different concentrations of a material and then planted in plastic containers of organic soil; wireworms were then introduced, airtight lids were placed onto the containers, and wireworm survival and damage were assessed 4 wk later. Tests with granular materials were similar except the containers were partially filled with untreated soil; 30 ml of soil treated with the granular material were then added to the container; an untreated single-eye billet was placed onto this treated soil; an additional 30 ml of treated soil was then placed on and around the billet; and finally untreated soil was added to fill the container. Conditions inside the bioassay containers appeared suitable for germination and growth of most varieties. Airtight lids were advantageous from the standpoint of maintaining soil moisture. Data indicated it may be disadvantageous to hold wireworms for a long period of time before using them to screen a material.

Bifenthrin, thiamethoxam 25WG, thiamethoxam 2G, and tefluthrin 3G appeared to have value as materials for reducing damage by wireworms to germinating eyes of seed cane planted in organic soils. However, germinated shoots of billets treated with these materials were sometimes injured by wireworms. Another material, ethiprole, was found to inhibit germination of CL77-797 when applied in solutions greater than ~ 1,000 ppm. Little wireworm mortality occurred in containers of billets treated with ethiprole at any rates tested, but surviving wireworms frequently caused injury to the billets. Another material, zeta-cypermethrin, appeared to have no value as a wireworm control material at the rates studied (75 to 125

ppm). Overall based on limited data, the most promising of these materials with respect to reducing wireworm damage to both germinating eyes and young shoots appeared to be thiamethoxam 25WG at 12,000 ppm.

### **Management Thresholds for the Sugarcane Borer on Louisiana Varieties**

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The sugarcane borer (SCB) is responsible for greater than 90% of the total insect damage to sugarcane in Louisiana, and the process to decide when to spray is determined by many variables (i.e. infestation levels, weather conditions, economics of the grower, environmental concerns, etc.). Therefore the overall goal of this study is to provide key facts that would allow the industry to have a greater flexibility in controlling the SCB on different varieties while maintaining a high level of confidence that a reduction in sugar per acre and buildup of SCB pest populations can be avoided. SCB larval infestations were monitored weekly with leaf sheath sampling. The SCB resistant varieties CP70-321 and HoCP85-845, and the susceptible varieties LCP85-384 and HoCP91-555 with four regimes of SCB control were treated with insecticide when the designated threshold levels were reached.

Results indicated that the variety HoCP91-555 (highly susceptible) required three applications of insecticide during the growing season for both the 5% SCB infestation threshold (5%) and 5% early and 10% late season threshold (5%/10%). In comparison, LCP85-384 (susceptible) required three insecticide applications for the 5% management threshold, but only two insecticide applications for the 5%/10% management threshold. The resistant variety HoCP85-845 required two applications for the 5% threshold and only one application for the 5%/10% threshold. CP70-321 required only one application under the 5% and the 5%/10% management regimes. This study further demonstrates some positive results for the industry's leading variety LCP85-384 (it currently represents about 80% of the sugarcane grown in Louisiana) in terms of growers being able to manage this variety against the SCB with the use of timely application of insecticides. The 5%/10% threshold shows promise and supports the industry's desire to reduce unneeded insecticide applications during the season due to increasing economic and environmental concerns.

## **Yellow Sugarcane Aphid (*Sipha flava*) Colonization Strategy and its Effect on Development and Reproductive Rates on Sugarcane**

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Yellowsugarcane aphid (YSA) is an occasional serious pest of sugarcane throughout the subtropics and tropics. Leaf feeding on susceptible cultivars results in red spots of various sizes and density usually followed by chlorosis and then necrosis. Prolonged feeding results in fewer new shoots, reduced stalk diameter and yield. Field samples indicate that winged aphids (alates) normally stay in one place on favored cultivars once they start reproduction and that alates are frequently found together in groups on leaves. This aphid also prefers leaves that are about half way between the top visible dewlap (TVD) and the youngest senescing leaves. Research was begun to examine whether group feeding affected development rates, nymph production and development rates of the subsequent F2 generation. Leaf position relative to the TVD was also evaluated for its possible effect on these population parameters. Tests were conducted in a greenhouse using the susceptible cultivar CP80-1827 inoculated with YSA from a laboratory colony maintained on a Sorghum-Sudan hybrid. Individual aphids and those in small groups took longer to develop to adults and produced fewer nymphs per day than those that developed within larger groups. The F2 generation reached adulthood and started reproducing in 25% less time than did the F1. Leaf position had a minor effect on these population parameters.

## **Field Trials of a Multiple-Pathogen Bioherbicide System with Potential to Manage Guineagrass in Florida Sugarcane**

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Guineagrass (*Panicum maximum*) is a problematic weed in sugarcane in Florida due to its capacity for prolific spread and tolerance to chemical herbicides. Development of host-specific fungal plant pathogens as bioherbicides may provide a nonchemical option to manage these weedy grasses. Three fungi indigenous to Florida, *Drechslera gigantea*, *Exserohilum longirostratum*, and *E. rostratum* were evaluated in July and September 2001 in Pahokee, FL for the control of guineagrass (*Panicum maximum*). Mini-plots, each 10' x 5', with a 5' buffer zone between plots, were set up. A mixture of the three pathogens (1:1:1 v/v; total 10<sup>6</sup> spores per ml; 250 ml spore suspension per plot @54GPA) was applied to guineagrass in each plot (3 to 4 inches tall (July) and 1 to 2 inches tall (Sep.)) as follows: (1)

Sunspray 6E 40% - Paraffin Oil 10% (Inoc-40E-10P); (2) Sunspray 6E 30% - Paraffin Oil 10% (Inoc-30E-10P); (3) Sunspray 6E 20% - Paraffin Oil 10% (Inoc-20E-10P); (4) Sunspray 6E 40% (Inoc-40E); and (5) Paraffin Oil 10% (Inoc-10P). Guinea grass in uninoculated control plots were treated with the respective carriers alone. The treatments were applied on July 03 and 18 and Sep. 02 and 22. A completely randomized block experimental design with four replicates for each treatment was used. At 3 weeks after initial inoculation (WAI), disease severity ranged from 15 to 27 % in July, and 52-90 % in Sep. on guinea grass applied with Inoc-40E, Inoc-20E-10P, Inoc-30E-10P, and Inoc-40E-10P fungal mixture treatments. Uninoculated guinea grass plants treated with the carriers alone, were healthy. At 4 WAI, plant growth was stunted, and reduction in panicle number per sq. m. area was 82%, 90% and 93% in July, and 99%, 99%, and 99% in Sep in Inoc-30E-10P, Inoc-40E, and Inoc-40E-10P treatments, respectively. The reduction in panicle number was higher ( $P=0.05$ ) than the control treatments. Thus, the mixture of *D. gigantea*, *E. longirostratum*, and *E. rostratum* has potential to be developed as a bioherbicide system for guinea grass in sugarcane.

### **Molecular Identification of Virus Isolates Causing Mosaic in Louisiana Sugarcane**

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Ten strains of sugarcane mosaic virus (SCMV) and three strains sorghum mosaic virus (SrMV) have been reported to cause mosaic in Louisiana; however, only strains H, I, and M of SrMV were recovered from commercial fields during surveys conducted between 1973 and 1995. Annual surveys were discontinued because of the large amount of labor required to identify strains using host differentials. At the time of these surveys, this was the only technique available to identify strains of these viruses, and results had changed little during the last 10 years. Recent advances in technology have led to the development of a laboratory procedure capable of distinguishing the mosaic virus strains. A survey was conducted in 2001 using reverse transcriptase-polymerase chain reaction-based restriction fragment length polymorphism (RT-PCR-RFLP) analysis to determine if changes have occurred among the strains of virus causing mosaic of sugarcane in Louisiana. Strain I and strain H of SrMV were associated with approximately 65% and 21% of the sugarcane plants with mosaic symptoms, respectively. In the earlier surveys, more than 80% of the plants were infected with strain H each year. The remainder of the plants (14%) surveyed in 2002 appeared to be infected by a new strain with a distinctive RFLP banding pattern. Nucleotide sequencing is being conducted to identify the virus strain. Sugarcane plants with mosaic symptoms will be collected in 2002 from a wider geographical area of the state and virus strains infecting the plants will be determined by RT-PCR-RFLP analysis.

## **Incidence of Sugarcane Yellow Leaf Virus in Clones of *Saccharum* spp. in the World Collection at Miami and in the Collection at the Sugarcane Field Station, Canal Point**

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Sugarcane yellow leaf virus (SCYLV) was detected in clones of *Saccharum* spp. in the World Collection and in the collection at Canal Point using a leaf mid-rib tissue blot immunoassay. The incidence of infection varied by the species of *Saccharum*. At Miami, approximately half the clones in the collection for each *Saccharum* spp. were sampled and the incidence of SCYLV in the clones was 7.0% for *S. spontaneum*, 74.5% for *S. officinarum*, 62.5% for *S. robustum*, 46.2% for *S. sinense*, and 14.0% for *S. barberi*. At Canal Point, there were only sufficient numbers of *S. officinarum*, *S. robustum* and *S. spontaneum* clones to sample and the incidence of SCYLV was 59.7% for the 134 clones of *S. officinarum* sampled, 60.7% for the 28 clones of *S. robustum* and 15.4% for the 52 clones of *S. spontaneum*. The results clearly indicate that SCYLV is present in clones present in the World Collection in Miami and that *S. spontaneum* and *S. barberi* are the two most resistant of the five species of *Saccharum*.

## **Selection of Interspecific Sugarcane Hybrids using Microsatellite DNA Markers**

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Three types of species-specific DNA markers, namely, PCR, RAPD, and microsatellites, have been recently developed at the USDA-ARS, SRRC, Sugarcane Research Unit, Houma, Louisiana. Among these, the microsatellite markers are the most polymorphic and can produce distinctive fingerprints (or molecular alleles) among sugarcane varieties as well as their wild relatives. In 2001, 11 wild x elite biparental crosses were made that involved 10 clones of *Saccharum spontaneum* and six commercial-type sugarcane varieties. The *S. spontaneum* clones were used as maternal parents to explore the possible impact of their cytoplasm on our varietal development program. A problem associated with sugarcane breeding is the potential for self-pollination of the maternal wild parents. We have demonstrated in earlier work that self-pollination can occur even after a hot-water treatment to emasculate the maternal tassels. Therefore, some of the seeds were selfed progeny. Since *S. spontaneum* is on the Federal noxious weed list, direct planting of *S. spontaneum* (including selfed progeny) to the field is prohibited. To circumvent the planting of selfed *S. spontaneum*, we used microsatellite markers to screen the seedlings from these crosses while they were still in the greenhouse. In this presentation, we will show the percentage self-

pollination in these crosses where the *S. spontaneum* flowers were hot-water treated. We also will demonstrate how microsatellite markers can be used to eliminate at the seedling stage unwanted selfs from the basic breeding and selection program.

## **Development of Microsatellite Markers from Sugarcane Resistance Related Genes**

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Microsatellites are arrays of short DNA sequence motifs, with 1 to 6 base pairs in length and are characterized by their hyper variability, abundance, reproducibility, Mendelian inheritance and co-dominant nature. The Microsatellite marker technique is simple, robust, reliable and suitable for a large throughput system. It is also applicable when the plant material available for analysis is limited in quantity and sufficiently quick to allow early decisions to be made prior to further screening. These advantages make the microsatellite technique a suitable tool for molecular selection in large breeding programs.

Expressed Sequence Tags (EST) in the sugarcane database were electronically searched for microsatellites and 402 were identified. Out of 267 (245 disease and 22 pest) resistance-EST investigated, 37 (34 disease and 3 pest) were positive for the presence of microsatellites. PCR primers flanking these microsatellites were designed and tested as markers on ten sugarcane genotypes – four commercial hybrids and 6 wild genotypes. Polymorphisms were evident both at the commercial clones, as well as among the *Saccharum* species. The presence of microsatellites within disease resistance genes could be the flexible mechanism that sugarcane possesses to ensure response to a new pathogen. DNA rearrangements, resulting from slippage during replication, which is characteristic of microsatellite sequences, would be allowing the cane plant to generate novel resistance to match the changing pattern of pathogen virulence. In humans, a few disease genes carry tri-nucleotide microsatellites. A novel mechanism for the amplification of these microsatellites sequences seems to be the root cause of these genetic abnormalities. Should the same mechanism work in plants, mapping microsatellites markers from disease resistance EST may increase the probability of tagging resistance genes in sugarcane commercial as well as in wild germplasm.

Microsatellites were also found in other 75 EST coding for proteins not related to disease resistance, such as sugar metabolism, and can be used as molecular markers for linkage mapping and tagging of other genes.

## **The Effect of Temperature on Flowering and Seed Set in Sugarcane at Canal Point.**

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South Florida experiences wide variation in the frequency and intensity of flowering in sugarcane in different years. The crossing program at Canal Point has maintained about 2000 pot cultures of at least 150 cultivars per year for each of the past 10 years. The individual cultivars have varied throughout the period but they are representative of the same genetic background. The number and time of emergence of tassels based on the number of tassels cut for use in crosses will be correlated to the minimum temperatures from September through January. The effect of low temperature on pollen fertility is well documented, but little information is available about the effect of low temperatures on tassels to be used as females. The plants used to produce the male tassels used in these crosses were protected from low temperatures by being moved into the crossing and photoperiod houses at night. The effect of temperature on flowering and seed set in sugarcane at Canal Point will be discussed.

## **Characterization of *S. Spontaneum* Collection for Juice Quality**

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In order to utilize a wider germplasm sample and more efficiently explore wild *Saccharum* species for breeding purposes, we initiated the characterization of 94 *S. spontaneum* and 2 *S. sinense* clones from the Copersucar germplasm collection at Piracicaba, SP, Brazil. Laboratory analysis was carried out for juice quality of these genotypes. Data were collected for Brix, Purity, Reducing Sugar, Pol and Fiber. Within the *spontaneum* genotypes, values ranged from 7.2 to 16.5 for Brix, from 0.4 to 7.8 for Pol and from 21% to 45% for Fiber.

Molecular marker analysis (southern) with an EST from Sucrose synthase as DNA probe on the DNA of 11 *S. spontaneum* genotypes is presented, showing polymorphism at this locus. Electronic search on sugarcane DNA sequence database shows Simple Sequence Repeats within genes controlling sugar metabolism.

The analysis on juice quality showed a wide variation for sugar content among *spontaneum* genotypes, which suggests genetic variation for these traits within this species. The molecular data shows high polymorphism at the chromosome locus where the gene controlling the Sucrose synthase enzyme is located, suggesting that cane breeders could use molecular markers for marker-assisted selection to

introduce positive alleles into commercial genotypes. Such a strategy would speed up the Back Cross method to introduce wild alleles in commercial varieties aiming to widen the narrow sugarcane genetic basis.

### **Family Selection in Sugarcane: Notes from Australia**

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Sugarcane breeding programs typically commence by evaluating a large number of seedlings derived from true seed. Mass selection applied at this stage of the program has been shown to be inefficient due to lack of replication, and the associated confounding effects of the environment. In Australia, the introduction of mobile weighing machines made it possible to implement family selection. Several research projects demonstrated that family selection when followed by mass selection was superior in terms of genetic gain and more cost effective than either family or mass selection alone. This combination of family and mass selection is now used routinely in all the Australian programs. Families are evaluated using replicated plots for cane (mechanically harvested and weighed) and sucrose yield in the plant crop. Individual clones are selected (mass selection), based mainly on visual appraisal for cane yield, from selected families in the first ratoon crop. Family selection is usually liberal with about 30 – 40 % of families selected. More clones are selected from the best families with progressively fewer clones being selected from the moderate to average families. The availability of objective family data makes it possible to estimate the breeding value of parents using the Best Linear Unbiased Predictor (BLUP). This information is used to retain or drop parents from the crossing program and to plan better cross combinations.

### **Assessment of Trends and Early Sampling Effects on Selection Efficiency in Sugarcane**

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Quantitative data on agronomic traits are normally affected by field trends or spatial heterogeneity, which often mask the genetic potential of the tested varieties. To identify promising selections from Stage II clones with some degree of confidence, a moving means analysis was performed on 754 experimental sugarcane clones (CP 2000 Series) tested along with five check varieties distributed across three fields with unequal frequencies. The data were subjected to three different methods (linear, quadratic, and row x column) to remove any potential field trend, as revealed by the variance of the checks, and to approximate the true genotypic values of the clones under selection. The best method was chosen as the one that accounts for the greatest variance of trends and the least variance of checks. In field A (16 blocks of 43

plots each), cane (TCA) and sugar tonnage (TSA) were more efficiently assessed by the quadratic method (2 neighbors). For the clones in fields B (16 blocks of 23 plots each) and C (14 blocks of 10 plots each), a row x column method was more appropriate in analyzing TCA and TSA. The ranking of varieties changed significantly when comparing the adjusted values with the field data. Though positive and significant ( $r_{tsa}=0.44$  and  $r_{brix}=0.28$ ,  $p=0.001$ ), the correlation between early and late sampling revealed that the former is not indicative and predictive of the latter. Consequently, a late March sampling yielded 32 additional clones for advancement to Stage III, with Brix values ranging from 18.6 to 22.3. Further analyses are warranted to ascertain the benefit of these approaches as prediction methods for identifying the most promising clones.

### **Selection and Advancement of Sugarcane Clones in the Louisiana “L” Sugarcane Variety Development Program**

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The primary objective of the Louisiana “L” Sugarcane Variety Development Program is to efficiently develop improved sugarcane cultivars for the Louisiana sugarcane industry. Each year, 300 to 600 crosses are made at the sugarcane breeding facilities of Louisiana State University Ag Center’s Sugar Research Station located in St. Gabriel, La. This begins a process of selection, advancement and testing which spans a period of 12 years culminating with the release of new sugarcane varieties to growers of the Louisiana sugar industry. Although the main goal of the program has never changed, procedures and techniques have evolved and improved over the years to the extent that this program is operating more economically efficient than ever.

This paper will outline the procedures and techniques used by LSU personnel in the seedling production through infield testing phases of the Variety Development Program. For purposes of discussion, the numbers of clones moving through the program during the year 2001 will be used.