QUT GP P 664.1220973 1

# JOURNAL

American Society of Sugar Cane Technologists

Volume 21 Florida and Louisiana Divisions June, 2001

ASSCT

## 2000 JOINT EXECUTIVE COMMITTEE AMERICAN SOCIETY OF SUGAR CANE TECHNOLOGISTS

#### General Secretary-Treasurer Denver T. Loupe

Office

## Florida Division

Carmen Baez-Smith David G. Hall John A. Fanjul James M. Shine Michael Damms John Dunckelman Barry Glaz Thomas Schueneman President First Vice-President Second Vice-President Chairman, Agricultural Section Chairman , Manufacturing Section Chairman at Large Past President Secretary-Treasurer

## Louisiana Division

Bill White Will Legendre Chris Mattingly Keith Bischoff Tony Parris Benjamin Legendre Manolo Garcia Denver T. Loupe

EDITORS Journal American Society of Sugar Cane Technologists Volume 21 June, 2001

## Managing Editor Ron DeStefano

Agricultural Editor Nael El-Hout

Manufacturing Editor

Manolo Garcia

## PROGRAM CHAIRMAN

**30th Annual Joint Meeting** 

American Society of Sugar Cane Technologists Michael S. Irey Honorary membership shall be conferred on any individual who has distinguished himself or herself in the sugar industry, and has been elected by a majority vote of the Joint Executive Committee. Honorary membership shall be exempt from dues and entitled to all the privileges of active membership. Each Division may have up to 15 living Honorary Members. In addition, there may be up to 5 living Honorary members assigned to the two Divisions jointly. (Article HI, Section 4 of the Constitution of the American Society of Sugar Cane Technologists).

As of May 2000, the following is the list of the living Honorary members of the American Society of Sugar Cane Technologists for Florida and Louisiana Divisions:

## Florida Division

Joint Division

Guillermo Aleman Henry J. Andreis Pedro Arellano Enrique Arias Antonio Arvesu John B. Boy David G. Holder Arthur Kirstein III Jimmy D. Miller Joseph Orsenigo Ed Rice Blas Rodrigues George H. Wedgworth Jack L. Dean Preston H. Dunckelman Lloyd L. Lauden Denver T. Loupe Harold A. Willett Louisiana Division

Felix "Gus" Blanchard Richard Breaux S.J.P. Chilton P.J. 'Pete" deGravelles Gilbert Durbin Minus Granger Sess D. Hensley James E. Irvine Dalton P. Landry Lowell L. McCormick Joe Polack Charles Savoie

## 2000 OUTSTANDING PRESENTATION AWARDS

E. P. Richard, Jr. Weed and Sugarcane Response to CGA 362622: A Potential Herbicide for Louisiana.

M. Damms and B. Anderson. Potential Sucrose Losses in Clarifier Mud and Mud Filtrate.

K. P. Bischoff, K. A. Gravois, H. P.Schexnayder and G. L. Hawkins. The Efect of Harvest Method and Plot Size on Sugarcane Yield

C. LaBorde, K. Gravois and K. Bischoff. Fertilizer Effects of Older Sugarcane Ratoon Crops in Louisiana.

## Errata

The following corrections should be made to the manuscript: Abou-Salama, Adel M. 2000. Improving Water Use Efficiency to Sugarcane Under Upper Egypt Conditions. JASSCT. 20: 41-52. The editor of Vol. 20 apologizes to the author and readership for any inconvenience.

Abstract, Paragraph 1, line 13 Value 55.81 should read 131

Abstract, Paragraph 2, line 3 Value 3,700 should read 23,095

Introduction, Paragraph 1, line 7 Value 12,000 should read 28,571

Materials and Methods, Paragraph 3, lines 3 and 4 Values 9,700; 11,000; 7,100; and 12,400 should read 23,095; 26,190; 16,904; and 29,523; respectively

Materials and Methods, Paragraph 3, lines 5 and 6 Values 9,900; 11,250; 7,100; and 12,700 should read 23,571; 26,785; 16,904; and 30,238; respectively

Table 4, Column Sugar yield (mt/ha)

Value	Correction
5.05	12.02
6.29	14.97
4.53	10.78
5.58	13.28
6.88	16.38
7.35	17.49
6.26	14.90
6.32	15.04

# TABLE OF CONTENTS

President's Message - Louisiana Division Bill White
President's Message - Florida Division
Carmen Baez-Smith
PEER REFEREED JOURNAL ARTICLES Agricultural Section
RNA Isolation and Photosynthetic Gene Expression in Sugarcane
Grown Under Elevated C0 <sub>2</sub> and High Temperature 9
Mu-Quing Zhang, Maria Gallo-Meagher, Joseph C. V. Vu, and L. Hartwell Allen
Notes on the Yellow Sugarcane Aphid Sipha Flava (Homoptera: Aphididae)
and the Lady Beetle <i>Diomis Terminatus</i> (Coleoptera:Coccinellidae) in Florida
Fertilizer Effects of Older Sugarcane Ratoon Crops in Louisiana
Effect of Silicon and Expression of Resistance to Sugarcane Borer
The Effect of Harvest Method and Plot Size on the Estimation of Sugarcane Yield51
K. P. Bischoff, K. A. Gravois, H. P. Schexnayder, Jr., and G. L. Hawkins
PEER REFEREED JOURNAL ARTICLES Manufacturing Section
Cane Juice Analysis by Near Infrared (NIR) to Determine
Grower Payment
The Presence of Total Polysaccharides in Sugar Production and
Methods for Reducing Their Negative Effects
Strategies for the Expansion of Cane Sugar Mills. 92 Peter W. Rein
A Flexible Coupling for Sugarcane Mills-Its Design Conception
Luiz Carlos Felicio

Cane Factory Process Modeling Using Sugars for Windows. L. Warner Weiss	
Natural Frequency of Torsional Vibrations in Sugar Mill Drives	
One Design, Two Pans, A Great Flexibility of Use	

G. Journet and C. Pelletan

# AGRICULTURAL ABSTRACTS

The Lady Beetle Diomis terminatus (Coleoptera: Coccinellidae) and the	
Yellow Sugarcane Aphid Sipha flava (Homoptera: Aphididae) in Florida	1
Efficacy of Soil Insecticides for Wireworm Control in Florida Sugarcane	1
Potential Impact of Sugarcane Varietal Changes On Insect Pest	
Management in Louisiana. 152	2
T. E. Reagan and F. R. Posey	
Novel Technology for Sugarcane Insect Pest Management	3
Sugarcane Yellow Leaf Virus: Possible Tactics to Develop Resistant Cultivars	4
Molecular Markers for Monitoring Transgenic Sugarcane	4
Effect of Growth Regulators on Efficient Plant Regeneration from Sugarcane Callus	5
Current Status on Biotechnology Research at Sugarcane Research Unit, Hourna, Louisiana	5
RNA Isolation and Photosynthetic Gene Expression in Sugarcane   Grown Under Elevated CO2 and High Temperature 15   Mu-Qing Zhang, M. Gallo-Meagher, Joseph C. V. Vu, and L. Hartwell Allen	6

Effect of Purple Nutsedge ( <i>Cyperus rotundus L.</i> ) Competition in Sugarcane ( <i>Saccharum</i> spp.)
CGA-362622: A New Herbicide for Weed Control in Sugarcane157 E. K. Rawls, J. W. Wells, M. Hudetz, R. Jain, and M. F. Ulloa
Weed and Sugarcane Response to CGA 362622: A Potential Herbicide for Louisiana 157 E. P. Richard, Jr.
Effect of Harvest Method and Storage Time on Sugarcane Deterioration159 G. Eggleston, B. L. Legendre, and C. Richard
The Effect of Harvest Method and Plot Size on Sugarcane Yield159 K. P. Bischoff, K. A. Gravois, H. P. Schexnayder, and G. L. Hawkins
Efficiency of First Clonal Trial Selection Using Different Plot Sizes160 S. B. Milligan, M. Balzarini, K. A. Gravois, and K. P. Bischoff
Analysis of Resource Allocation in Final Stage Sugarcane Clonal Selection
Responses of Sugarcane Plants to Water Tables
Soaking of Sugarcane Stalk Sections in Water Before Planting163 B.Glaz
Sugarcane Yield Following Long-Term Exposure to High and Flooded Water Tables
Sugarcane Cultivar Response to Two Summer Water Management Strategies in the Everglades
Photosynthesis Characteristics in Eleven Cultivars of Sugarcane and Their Responses to Water Stress During the Elongating Stage
Characterization of Possible Sulfur Sources within the Everglades Agricultural Area165 T. J. Schueneman

Si as Macronutrient for Sugarcane						
Plant-Available Si Status of Louisiana and Florida Soils						
The Effect of Pro-Sil on P Leaching in Sandy Soils						
Effect of Urea Nitrogen Rates, A Nitrogen Stabilization Package, Winter vs Spring Nitrogen Fertilization, and Varieties on Sugarcane Yields						
Fertilizer Effects of Older Sugarcane Ratoon Crops in Louisiana168 C. LaBorde, K. Gravois, and K.Bischof¥						
A Summary of Precision Farming Results for South Florida						
Application of Precision Agriculture to Target Areas of Poor Sugarcane Yields on Mineral Soils						
MANUFACTURING ABSTRACTS						
The Presence of Total Polysaccharides in Sugar Production and Methods for Reducing Their Negative Effects						
Cane Juice Analysis by Near Infrared (NIR) to Determine Grower Payment						
Strategies for the Expansion of Cane Sugar Mills						
Potential Sucrose Losses in Clarifier Mud and Mud Filtrate						
Partition Coefficients for Aconitic Acid between Aqueous Solvents and Tertiary Amine-Containing Solvents						

Factory Comparisons of Cold and Intermediate Lime Clarification in Raw Sugar Manufacture					
A Flexible Coupling for Conception and Perfor L. C. Felicio	or Sugarcan mance	e Mills-Its Desig	gn		
Chromatographic Desu C. Baez-Smith,	garization J.Alvarez	of Syrups in Car	ne Sugar Mills		
Simulation J. Gamez and M	of 1. Saska	Vacuum	Pan	Crystallizer	
Flourescence Technolo Sugar Contamination H. Herrara	gy and the	Measurement of	· · · · · · · · · · · · · · · · · · ·		
A Preliminary Evaluati Liming vs Slurry Limin A. Monge, J. G	on of Two ng at Cora- . Spinoso, a	Published Methor Fexas Factory and H. S. Birkett	ods of Sacchara	te	
The Internet-Toy or To	ol?				
M. B. Inkson					
POSTER SES	SSION				
Progress on USDA-AR	S Water M	anagement Rese	earch Project:		
Sugarcane-Water Quali T. S. Kornecki	ity- Traffica and J. L. Fo	bility. Juss			

Rules for Preparing Papers to be Printed in the Journal of the

American	Se	ociety	of	Suga	r	Cane	Te	chnologists	
Guidelines	for	Pr	eparing	Papers	for	Journal	of	ASSCT	183
Constitution	of	the	American	Society	of	Sugar	Cane	Technologists_	184
Author Index.									. 191

To order an extra copy of this volume, or a previous journal of American Society of Sugar Cane Technologists, write to:

General Secretary-Treasurer American Society of Sugar Cane Technologists P.O. Box 25100 Baton Rouge, LA 70894-5100

Copies shipped within the USA are \$10.00, others are \$15.00.

## PRESIDENT'S MESSAGE LOUISIANA **DIVISION**

William H. White USDA, ARS, SRRC Sugarcane Research Unit Houma, LA

On behalf of the membership of the Louisiana Division of the |American Society of Sugar Cane Technologists, I thank the Florida Division for hosting our Thirtieth Annual Joint Meeting here in St. Petersburg Beach, Florida. I look forward to another successful meeting of scientific and technology exchange.

Considering that with this meeting we end an old millennium and begin a new century, it is probably compulsory that the presidential message have a new millennium theme. For example, the theme of our recent ISSCT Entomology Workshop held in Thailand was Sugarcane Pest Management Strategies in the New Millennium. Unfortunately, I am not certain that I possess either the wisdom or the insight to offer you a similarly titled presidential message, maybe, Sugarcane Industry Survival Strategies for the New Millennium. But what I will do, and what I think would be equally appropriate, is to address some important trends that have occurred in the Louisiana sugarcane industry over the last century. Trends that should cause us to reflect more profoundly about what the next century may have in store for us.

As introduction to this topic, let me first relate a few production numbers from last year's harvest in Louisiana. The Louisiana industry ended the last century on a very positive note with several records being set. Louisiana growers and processors produced a record 1.67 million tons of sugar; the sixth consecutive year of production over 1 million tons. The past year's tonnage of over 37 gross tons of cane per acre was also a state record. Although the industry high for sugar content was set in 1987 at 225 pounds of sugar per ton of cane, 1999 was a very good year with yields over 211 pounds.

A striking feature of the 1999 crop was the area planted to sugarcane. The 1999 crop was the largest acreage on record for the state. In 1999, 464,000 acres were planted to cane. With an estimated 32,000 acres used for seed, a total of 432,000 acres were harvested. This is quite a contrast to the 220,000 acres harvested in 1982, my first harvest.

A notable downside to these impressive production records was the low price of sugar. In 1999 we saw a 20% reduction in the price of sugar. Fortunately much of the 1999 crop had already been priced at a higher level, but obviously pricing of the 2000 crop remains an important concern as we begin this next century. Prices for the first quarter remained far below levels needed for a healthy industry. Growers and processors will be challenged to find new ways to cut production and processing costs while maintaining profitability. Researchers will be challenged to provide the information to the industry to accomplish this task.

The last century was replete with changes for the Louisiana sugar industry. Dr. Charley Richard of the American Sugar Cane League wrote an informative article in the Sugar Journal titled The Louisiana Sugar Industry: A Century of Change. In his article Charley discussed several trends occurring in the Louisiana industry over the last century. I was particularly struck by a trend that reflected a reduction in the number of growers and processors during the last century. The number of growers in the state dropped from 12,300 in 1919 to only 700 today; an average loss of 145 growers per year. Mills were lost at a rate of almost 3 per year-300 in 1900 to 18 in 1999. What a strong statement to the efficiency of the Louisiana industry; production records continued to be set while the size of the crop increased, but not the numbers of growers or mills. Economics of size and logistics of growth have certainly played a major role in the success of the industry.

As a researcher associated with the sugarcane industry, I am also keenly aware of other losses occurring. Losses not discussed in Charley's article, but losses that he and other industry leaders are also aware of. This is the loss in the numbers of researchers associated with sugarcane and a reduction in available funds to conduct research. The reduction in funds available for research has forced the present scientific personnel to become more efficient just as their industry counterparts have as they cope with stagnant or declining sugar prices over the last 15 years. Concurrent to the generally declining sugar prices, growers and processors are being forced to shoulder an increasing share of the research dollar. This means that researchers must become more effective communicators with the industry. Those who cannot demonstrate relevance to their industry will find little support when budget cuts are proposed and cutting a program appears the only alternative.

When I first reported to work at the Houma facility, there were 11 research scientists; now there are only seven. I am the youngest of this group and I am 47. As far as I know, I am the only full-time sugarcane entomologist employed by the USDA. Within the last two years all five mainland breeding programs have hired new plant breeders. Reshuffling existing talent basically filled these positions. Only the Canal Point Station hired an individual without can experience, however; this individual was trained as a classical plant breeder. Similar scenarios can be identified for other disciplines as well.

It is sobering to think where the next generation of classically trained plant breeders will come from; or the next field entomologist; or who will be doing the basic research on cane physiology. Will that expertise be housed at research facilities supported by our competitor? If so, does that mean that we will have to purchase this technology from these competitors? Or will we acknowledge the problem and do what we can do to reverse the trend?

I think it will be the latter because this society, a society dedicated to the advancement of the mainland sugarcane industry in the U.S., has a responsibility to look beyond the next harvest or planting season. Both divisions of the society are doing much to help cultivate a new generation of technologists by supporting local science fairs and 4-H competitons by offering monetary awards to outstanding students and their mentors. At the university level, both undergraduate and graduate fellowships are being supported. All these programs have become popular and competition for awards among students has become increasingly keener. Similar concerns also exist in the sugar mill. Where will the next engineer or mill manager come from? The Louisiano Division has begun steps to expand its fellowship program to also include support for promising young talents for the factory. Much of the success of these programs can be attributed to corporate participation at our division and joint meetings. We should take the time to thank our corporate sponsors for their support and consider as individuals to also participate in these programs.

Obviously many more changes and challenges face the domestic cane sugar industry. But I have no doubt that the skills and determination to meet these challenges exist in our membership. This meeting is part of a package that helps us remain competitive. It is a package that cultivates our future leaders and technologists. It also brings us together so that we can exchange our research findings and have our ideas challenged by our peers. This meeting also strengthens the bonds between our industries. This too is important because only by standing together can we meet the challenges from our opponents within this country and competitors from without. I thank you all for your kind attention and wish you all a very successful year.

## PRESIDENT'S MESSAGE FLORIDA DIVISION

#### Carmen Baez-Smitb

Sugar Cane Growers Cooperative of Florida Belle Glade, Florida

On behalf of the Florida Division of the American Society of Sugar Cane Technologists, we welcome the Louisiana Division to the 30<sup>th</sup> Annual Joint Meeting. We hope the preparations and the program will make your attendance as pleasant as we wish it to be. Thank you very much for your presence. Once again, my warmest welcome to all of you.

It is an honor for me to be presiding today as the first woman president of an ASSCT division. I want to thank all our members, especially our Florida Members and our executive committee officers, for the support that I have received during this year. It is also an honor for me to be part of this committee. I am very fortunate to be working with such a wonderful group of people who are true professionals in their fields of expertise. I also want to thank my family for all the support they have given me, especially my husband Bill and my daughter Kimberly. Mom, I am sure that you are also watching and cheering for me from heaven, up there, thank you. I also want to thank the Lord for sparing us last year from the damage of what was supposed to be a devastating hurricane called Irene, that miraculously turned away from us. Bless the Lord.

First, I just want to pass on some traditional information, which is also very exciting at the same time. In Florida, we just completed a very successful crop with records being broken in several categories in several mills. Congratulations to all for a job well done. We grew approximately 460,000 acres of cane, of which 444, 000 were harvested for sugar production. Tons of cane sugar ground were 16,769,871, producing 1,965,747 tons of raw sugar with polarization of 98.92 percent, and 101,196,356 gallons of final molasses, with degree brix of 79.5 percent. All of this was the result of an average sugar yield of 11.32 percent and molasses purity of 35.31 percent. The overall sugar recovery was approximately 86 percent. The land productivity was approximately 37.8 tons of cane per acre and 4.43 tons raw sugar per acre harvested.

We are all aware of the recent drop in sugar prices and the volatility of the market. While certainly hoping for stable price recovery, there is no guarantee that will happen as we wish it to be. As technologists, there is not much that we can do about sugar price recovery, but we can help in another front, and that is sugar recovery in our process and production efficiency.

## Theme: Technology Improvement & Efficiency

Fellow technologists of this society, my message to you is that we must continue to improve our process, starting with land productivity and finishing up with the packaging and dispatching facilities. The new millennium has plenty of challenges, but at the same time it has come with great opportunities. I hope our industry gears up to renew its technology by approaching new technical procedures that, ultimately, will lead to increased efficiency and productivity. Good and efficient analytical methods are not enough to have better processes. We also need better performances and improvement in production technology.

Separation technologies will play a very important role in our process improvement. An efficient separation process will lead to a unified and stronger sugar technology. As technologists we agree in one common objective: sugar will be efficiently separated from the non-sugar components in the juice to obtain higher recovery. At least we already have available one useful piece of information: we know the inconveniences caused by the presence of the non-sugar components in the separation process. I am not here to tell you how far behind we have fallen, but we need to realize that we have a lot of work ahead of us. I am here to tell you that we need to move forward quickly and with energetic steps.

First, we need better process and technology documentation. We need deeper knowledge of our raw materials and processes. To give you just one example, there are hundreds of volumes worth of research in sugar technology, but the most elemental information in clarified juice composition is vague and lacking precision. Suspended solids are not even mentioned in our classical chemical description for our soluble solids in juice, does not include suspended solids, as might be expected. As it turns out later, those solids, as well as any other non-sugar solids, need to be removed from a successful operation. There are technologies available at present to remove these suspended solids. However, we need to have a clear understanding of the suspended solids quantitative value to design the proper equipment to remove them. An ultrafiltration membrane that is capable of removing suspended solids 100 times the concentration in the feed when the suspended solids is 0.01% is not capable of doing the same job when the suspended solids are increased to 0.1%. Chromatography, ultrafiltration, and softening are among the new forms of separation from non-sugar that are worth deep experimentation and study. Although chromatography and softening have been used for more than twenty years in the sugarbeet industry, they have been reserved for use in the back end of the process. The sugarbeet industry uses chromatography for molasses desugarization.

Now, I am going to present the status of the Actual Sugar Factory. As can be seen in the production records given above, our industry process is currently working on an overall recovery of 86-87% for the raw sugar factory alone. It means the 13-14% of the incoming sugar to the mill is lost and/or gone with the molasses, the majority of it gone with the molasses. For the refinery, on the other hand, they report recoveries of 93%, when the starting point is raw sugar that already has had a loss of 13-14%. That is all we need to know to push for an improvement of our current process technology. It has been like that for more than 500 years. It is time for **a** much better position. Although ambitious, but not impossible, we must strive for lower sugar losses in our process, as well as higher sucrose content in the incoming raw cane. Every change comes with its risks. We must be able to overcome these obstacles in our quest to find more successful ways of producing sugar. We need to make it happen.

Now, I am going to present the proposed "new" factory improvement: The process of the factory of the future. I do not want to sound prophetical, but let me give you the vision I have for the factory of the future. The time is coming when refined sugar will be produced directly from juice in the raw sugar mill, and overall sugar recovery will surpass 90%. All of this will happen with better **sugar** quality and a minimum production of molasses that has purity well below 20%. The **new sugar industry** will have the option of production both raw **and** refined sugar at the same time. The new sugar industry will find a place in the sophisticated market of the pharmaceutical industries. All of this and more can happen, and for those of you thinking that I am dreaming, I just want to tell you that one day you will wake up to the reality of a new sugar industry. I believe that will happen soon if we as technologists experience a vision and prepare the plan needed to fulfill that vision.

We also need to branch out with more byproducts that are commercially feasible. We need to extract more profits from the various streams of our process. That does not mean making less sugar. On the contrary, it means more sugar resulting from better ways of production separation. We must find the technology required to produce more value-added products from the whole process. Just to name a few important co-product processes, here is an interesting list of opportunities waiting for us to put the magic touch in the revival of our industry: molasses for the production of yeast, citric acid, lactic acid, aconitic acid, ethanol, as well as production of live-stock feeds. Co-generation of electricity based on bagasse, manufacture of charcoal, activated carbon from boiler ash pit, manufacture of xylitol from bagasse are other value-added products.

We must also bring more automation to our process, and thus bring production costs down as a way of improving our process profit. The use of programmable logical controllers, PLC's and computers axe sure to make the process much easier and efficient. We must switch from batch to continuous production settings. The tools are out there, but it is up to us to make the change.

Now, my vision on How to achieve all this and more. Joint research and cooperation among the major key players - that is the answer. In today's economy we are witnessing more and more mergers of huge companies. That is happening, not because it is the fun thing to do, but because it is part of the strategy and vision for a better company. Well, the sugar industry does not necessarily need mergers, but we definitely need to bring down the unguaranteed costs of research and development. Joint research is a key answer. Joint research does not necessarily mean total and complete cooperation. It could well be for just the projects related to the strategy in vision. If a merger works for the majority of the chemical process industries, joint research will be the order of the day for the sugar industry. This will avoid repetitive and costly experimental work. Otherwise, we are all going to be trying to reinvent the wheel at the same time in different mills and places.

Research and development, as you know is expensive. The price tag, contrary to the price of a determined consumer item, sometimes is higher than the simple price of achieving a set goal. The reason is because, in the process of finding a specific goal, one has to pay the price for the knowledge of certain processes in between. One example of joint cooperation re research is Brazil. As we know, Brazil has developed the ethanol technology to such a point that it is the leading producer of ethanol from sugarcane derivatives in the world. They also have one of the largest, if not the largest, sugar research and development centers in the world, which is funded by joint company participation. Fellow technologists, remember, if we want to be more efficient, we should also live up to the complete meaning of the word. It does not take a scientist to figure out that it is more productive to run one process, than two or more identical processes trying to achieve the same goal. As a scientist, I have faith in the advancement of science and my vision of the sugar industry.

To accomplish, this vision, resources must be allocated for research and development. The task should be collective. It assumes that we have efficient and skilled workers. It assumes management is committed to the advancement of our sugar technology. It is then up to us to put a plan in motion to achieve our vision. Generally, the management commitment is not a problem. We as technologists need to be capable of presenting responsible alternatives that eventually lead to more efficient ways of production. While the pressure is really upon us to be productive, what a wonderful sense of accomplishment the development of these new technologies bring. A responsible alternative means an idea with a plan that must be accompanied by an experimental design, which must also have expected results attached. Correlation of experimental results with the practical world is of paramount importance. In some cases, feasibility studies will help in pointing out the commercial potential of our ideas.

Fortunately, we have in our hands great tools to help us in our task. We are living in the information era, which will make the work cheaper, more pleasant, and easier. If our ancestors were capable of making sugar at a competitive price two hundred years ago we should, with today's achievements, be able to be much more productive and competitive.

I am looking forward to working with you in making this a reality. Thank you very much for your attention and God bless you all. PEER

REFEREED

JOURNAL

ARTICLES

AGRICULTURAL

SECTION

# RNA ISOLATION AND PHOTOSYNTHETIC GENE EXPRESSION IN SUGARCANE GROWN UNDER ELEVATED C0 $_2$ AND HIGH TEMPERATURE

## Mu-Qing Zhang, Maria Gallo-Meagher, Joseph C.V. Vu, and L. Hartwell Allen, Jr. Agronomy Department University of Florida Gainesville, FL 32611

#### ABSTRACT

Two simple protocols based on guanidinium thiocyanate (GTC) and cetyltrimethylammonium bromide (CTAB) were used for the isolation of RNA from leaves of sugarcane (Saccharum officinarum L. cv. CP 73-1547 and CP 80-1827) exposed to ambient CO20r double-ambient CO<sub>2</sub>, along with either ambient or high temperature treatments, RNA yields of 43 to 93 ug per gram leaf fresh weight were obtained. RNA quality was determined by UV spectrophotometry, denaturing agrose gel electrophoresis, and northern blot analysis. The GTC protocol resulted in high vields of quality RNA from sugarcane grown at ambient temperature, but not from tissues exposed to high temperature. The CTAB protocol was superior to the GTC protocol for isolating RNA from sugarcane exposed to high temperature, due to the inclusion of polyvinylpyrrolidone (PVP) and ethylenediamine tetraacetic acid (EDTA) in the RNA extraction buffer. RNA isolated using the appropriate protocol was of sufficient quality to produce strong hybridization signals in northern blot analysis. In studies examining the effect of high CO<sub>2</sub> and temperature on photosynthetic gene expression, results from northern blot analysis showed that when compared to ambient temperature, high temperature significantly reduced the transcript levels of sugarcane phosphoenolpyruvate carboxylase (pepc) and ribulose bisphosphate carboxylase/oxygenase small subunit (*rbcS*) genes regardless of the growth  $CO_2$  used. However, the percent reduction in transcript abundance seen in high temperature-treated plants was greater at ambient C02 than at elevated C02. At ambient temperature, pepc transcript abundance declined at the elevated CO<sub>2</sub> level. Conversely, rbcS transcript levels were increased by the enriched CO<sub>2</sub> treatment.

Key words: RNA isolation, sugarcane, pepc, rbcS, enriched CO2, high temperature

#### INTRODUCTION

Since the Industrial Revolution in Western Europe (1750-1800), the atmospheric carbon dioxide concentration ( $(CO_2)$ ) has increased from 280 ppm to more than 365 ppm (for review, see Vu et al., 2000). As atmospheric levels of  $CO_2$  continue to rise, one of the results will be higher air temperature. Therefore, there is considerable research interest in understanding the mechanisms by which plants respond to elevated  $CO_2$  and temperature, and, in particular, how changes in plant response may impact photosynthesis (Sage, 1994; Webber et al., 1994; Bowes, 1991). Research during the past twenty years on growth, as well as on mechanisms and acclimation in photosynthetic processes as a result of long-term exposure to elevated  $CO_2$ , has focused mainly on C<sub>3</sub> species (Vu et al., 2000). For C<sub>4</sub> plants, interactive effects of elevated  $CO_3$  and other adverse environmental

conditions on growth, yield, fundamental physiology, biochemistry and molecular biology of leaf photosynthesis are not well understood.

The isolation of high quality RNA from selected tissues for use in techniques such as RT-PCR, northern blot analysis and RNase protection assays is one of the first steps towards elucidating molecular mechanisms underlying physiological processes. However, the presence of polyphenolics, polysaccharides and other unidentified compounds from different plant tissues appears to cause the formation of RNA complexes that interfere with RNA isolation and hamper further analyses (Lay-Yee et al., 1990; Wang and Vodkin, 1994; Graham, 1993). The large number of published RNA isolation procedures reflects these difficulties (e.g. Bahloul and Burkard, 1993; Schultz et al., 1994; Dong and Dunstan, 1996; Geuna et al., 1998; Kiefer et al, 2000) and particular plant tissues generally have specific requirements for successful RNA isolation. The success of an RNA isolation protocol is judged by the quality, quantity, and integrity of RNA recovered.

In this study, we investigated photosynthetic gene expression of the C<sub>4</sub> plant, sugarcane, grown under elevated CO<sub>2</sub> and high temperature. Two specific RNA isolation protocols were evaluated in terms of their ability to produce high yields of quality RNA for northern blot analysis. Here we describe successful protocols for RNA isolation from the leaves of sugarcane exposed to ambient (360 ppm) and double-ambient (700 ppm) CO<sub>2</sub> and two temperature regimes, 1.5C° above ambient (referred to in the text simply as ambient temperature) and 6.0C°C above ambient (referred to in the text simply as ambient temperature) and 6.0C°C above ambient (referred to in the text simply as ambient temperature) and 6.0C°C above ambient (referred to in the text simply as ambient temperature) and for the sequence and *rbcS* genes under the above environmental conditions. The *pepc* and *rbcS* genes code for phosphoenolpyruvate carboxylase (PEPC) and ribulose bisphosphate carboxylase (Rubisco), respectively, two key enzymes responsible for CO<sub>2</sub> fixation in C<sub>4</sub> photosynthesis.

## MATERIALS AND METHODS

#### Plant materials and growth conditions:

Sugarcane (Saccharum officinarum L., cv. CP 73-1547 and CP 80-1827) was transplanted into paired companion temperature-gradient greenhouses (TGGs) under natural sunlight at Gainesville, Florida in March 1997 and grown continuously through ratooning in the TGGs. The structural characteristics, specific methods and quality of environmental controls in the TGGs are described in detail by Sinclair et al. (1995). These greenhouses, 4.3-m wide and 27.4-m long, provided four 5.5-m zones along the length with differences maintained at  $1.5^{\rm C}$  steps above ambient by a combination of heaters, solar radiation, and computer-controlled ventilation fans. The interactions of temperature and CO<sub>2</sub> treatments were provided by paired CO<sub>2</sub>-enriched (700 ppm) and ambient-CO<sub>2</sub> (360 ppm) greenhouses. The first fully expanded leaves were sampled from five second- ratoon plants of each cultivar near midday on June 18, 1999 in the TGGs at  $1.5^{\rm C}$  above ambient and  $6.0^{\rm C}$  above ambient. For June 1 - June 18,1999, the average maximum ambient temperature was  $32.6^{\circ}$ . Leaf samples were immediately frozen in liquid N<sub>2</sub> until total RNA isolation.

## **RNA** isolation protocol:

A. Modified RNA isolation procedure using guanidinium thiocyanate (GTC)

## (Guetal., 1997)

Procedure
Homogenize frozen leaves in sterile tubes with extraction buffer* containing
GTC (3 ml per gram leaf tissue).
Add 1/10 vol. of 2 M sodium acetate, pH 4.0, vortex well.
Add 1 vol. of phenol, pH 4.3, vortex well.
Add 1/5 vol. of chloroform : isoamyl alcohol (24:1), vortex well.
Place on ice for 30 min.
Centrifuge at 15000xxg for 30 min at 4°C.
Transfer upper, aqueous phase into sterile tube. Add 1 vol. of isopropanol to
precipitate RNA, vortex well.
Place at -20°C for at least 15 hr.
Centrifuge at 13000xxg for 25 min at 4°C. Remove supernatant.
Dissolve pellet in 1ml GTC solution. Add an equal volume of isopropanol,
vortex well.
Place at -20°C for at least 15 hr.
Centrifuge at 13000xg for 20 min at 4°C. Remove supernatant.
Rinse pellet two times with 70% alcohol.
Air dry the RNA pellet.
Resuspend pellet in 100 µl 50 µM aurin tricarboxylic acid (ATA).
Quantify and store at -80°C.

•Extraction buffer: 4 M GTC; 25 mM sodium citrate, pH 7.0; 0.5% (v/v) sarkosyl; \_0.1 M β-mercaptoethanol added just before use.

B. Modified RNA isolation procedure using cetyltrimethylammomum bromide (CTAB)

#### (Dong and Dunstan, 1996)

Step	Procedure
1.	Homogenize frozen leaves in sterile tubes with extraction buffer* containing
	CTAB solution preheated at 65°C (20 ml per gram leaf tissue), shake
	vigorously.
2.	Heat samples to 65°C in a water bath for better homogenization (30 min at
	least).
3.	Add an equal volume of chloroform, vortex well.
4.	Centrifuge at 12000xg for 15 min at 4°C.
5.	Remove the top layer and place into another sterile tube, add an equal volume
	chloroform, vortex well.
-	G . 16 . 10000 . C 10 . L 10 G

Centrifuge at 12000xg for 10 min at 4°C.

- 7. Remove the supernatant into a sterile tube and add 1/10 vol. of 10 *M* LiCl to precipitate RNA. Shake, but not vigorously.
- 8. Precipitate the RNA overnight at 4°C in an ice bath.
- Harvest RNA by centrifugation at 12000xg for 20 min at 4°C. Remove and discard supernatant carefully.
- 10. Air dry the RNA pellet, and dissolve the pellet in warm SSTE buffer.
- Add an equal volume of chloroform, vortex well. Centrifuge at ICOCOxg for 10 min at 4°C.
- 12. Remove the supernatant into a sterile tube and add 2 vol. of 95% ethanol.
- 13. Precipitate for a minimum of 1 hr at -80°C.
- 14. Centrifuge at 1 0000xg for 20 min to pellet the RNA.
- 15. Rinse the pellet two times with 70% ethanol.
- 16. Air dry the pellet, then resuspendin 50 μl 50 μM ATA.
- 17. Quantify and store at -80°C.

\*Extraction buffer: 2% (w/v) CTAB (hexadecyltrimethylammomum bromide); 2% (w/v) PVP (polyvinylpyrrolidone); 100 mM Tris-HCl, pH 8.0; 25 mM EDTA; 2 M NaCl; 0.5g/L spermidine; **2%** (**v**/v) β-mercaptoethanol added just before use.

Northern blot analysis:

Total RNA was isolated from approximately one gram of liquid N2-frozen leaf tissue using one of the above RNA isolation protocols. The absorbance of individual RNA samples was scanned between 320 and 220 nm and quantified spectrophotometrically using a U-2000 double beam UV/VIS spectrophotometer (Hitachi Instruments, Inc., San Jose, CA), Fifteen micrograms of total RNA was denatured in 50% (v/v) formamide, 18% (v/v) formaldehvde, and separated by electrophoresis in 1.0 % agarose gel containing 18% (v/v) formaldehvde, RNA gels were blotted onto Hybord-N (Amersham, Piscataway, NJ) by capillary transfer with 20x SSC buffer (pH 7.0) and then UV-crosslinked at 1.2 x 10<sup>5</sup> mJ/cm<sup>2</sup> using a XL-1000 UV Spectrolinker (Spectronics Corp., Westbury, NY). Duplicate samples were run on the same gels, cut and stained with 0.5 ug/ml ethidium bromide. Membranes were prehybridized at 65°C for 30 min in 0.25 M phosphate buffer (pH 7.2), 7% SDS and 100 ug/ml denatured salmon sperm DNA. Hybridization was carried out overnight in the same solution with <sup>32</sup>P-labelled probe at 65°C. The *rbcS* probe was made from a 0.9kb BamHI fragment of scrbcs-1 (Tang and Sun, 1993) inserted in Bluescript II SK+ (Stratagene, La Jolla, CA). The pepc probe was made from a Sphl/EcoRI 0.8kb fragment of a maize genomic clone inserted in pUC19 (Matsuoka and Minami, 1989). Probes were labeled by random priming (Prime-a-Gene Labeling System, Promega, Madison, WI) and had a specific activity of approximately 3×10° cpm/µµg DNA. Membranes were washed once at 65°C for 30 min in 20 mM phosphate buffer (pH 7.2) containing 5% (w/v) SDS, and then once at 65°C for 30 min in 20 mM phosphate buffer (pH 7.2) containing 1% (w/v) SDS. Autoradiographic images were obtained by exposure to Fuji medical X-ray film (Fuji, Stanford, CT) at -80°C for 5 days, Bands were quantified using the Gel-Doc 2000<sup>TM</sup> System (Bio-Rad, Hercules, CA). Following hybridization with pepc or *rbcS*, membranes were stripped twice in a large volume of 0. lx SSC and 0.5% (w/v) SDS at  $95^{00}$ C

and re-probed as described above with a randomly-labeled 381 bp fragment of a rice 18s rDNA sequence which was PCR-amplified using 5'ATGAAAGACGAACCACTGC3' and 3'CAAGAATCAACC ACCTGC5' as upper and lower primers, respectively. The radiolabeled 18s rDNA probe was used to ensure that each lane contained equal amounts of sugarcane total RNA, and all *pepc* and *rbcS* signals were normalized according to the 18s rRNA signals obtained for each blot.

#### Statistical analysis:

A repeated measures analysis of variance was used to model RNA yield and ratios using the ANOVA procedure (SAS Institute, Gary, NC).

## RESULTS AND DISCUSSION

Yields and purities of RNA isolated from sugarcane exposed to ambient as well as elevated  $C0_2$  and temperature are summarized in Table 1. Total RNA yields ranged from 43 to 93  $\mu$ g per gram leaf fresh weight. RNA isolation from sugarcane leaves was first tested using a modification of a protocol based on the classical guanidinium method (Chomczynski and Sacchi, 1987; Gu et al., 1997). Guanidinium thiocyanate (GTC) is a powerful protein denaturant that inhibits ribonucleases. With the GTC method, significantly more RNA was isolated from leaves subjected to ambient temperature (T<sub>1</sub>) than from leaves exposed to high temperature (T<sub>2</sub>), regardless of the  $C0_2$  concentration.

The high temperature treatment resulted not only in lower yields, but also in RNA of reduced quality. RNA quality was first assessed by evaluating the values obtained from the Azeroza and Anorra ratios. The Anorra ratio is an estimate of protein contamination and highly pure RNA should have a ratio in the range of 1.7-2.0. The Amora ratio is an indication of polysaccharide or polyphenol contamination and highly pure RNA should have an Azeroza ratio >2.0 (Logemann et al, 1987; Manning, 1991). Results showed that A 2007 and A 2007 atios from leaves exposed to high temperature were significantly smaller than those for RNA isolated from leaves exposed to ambient temperature (Table 1). Areas ratios of RNA extracted from high temperature treatments were less than 1.0, indicating contamination from polysaccharides or polyphenolics. Polysaccharides often result in the formation of gelatinous material that may co-precipitate with RNA and severely interfere with centrifugal isolation (Lewinsohn et al., 1994). In some other protocols, 4M LiCl is added to solubilize polysaccharides and specifically precipitate RNA, thereby reducing contamination (Puissant and Houdebine, 1990). Additionally, polyphenol can bind to protein and RNA resulting in inactivation and precipitation (Loomis, 1974; Schneiderbauer et al., 1991), and in some cases, it auto-oxidizes into quinone to increase the RNA pellet color. In RNA isolated from the high temperature treatments using the GTC protocol, brown pellets were visible, and following resuspension the maximal spectrophotometric absorbency shifted from a peak absorbance for pure RNA at 260 to 255 nm (Table 1). Quality was also checked by the appearance of the RNA on a formaldehyde-denatured agarose gel (Figure 1). GTC-extracted RNA from high temperature-treated leaves contained faint or no distinct ribosomal bands (Figure 1, Lanes 2 and 5) in contrast to the RNA from ambient temperature-treated leaves (Figure 1, Lanes 1 and 4).

Because of the inadequacy of the GTC method to produce suitable RNA from high temperature-treated sugarcane leaves, the CTAB method was investigated. The CTAB protocol did not improve the yield of RNA from the high temperature treatments, but did substantially improve quality (Table **A**<sub>360780</sub> ratios were greater than 1.7 and **A**<sub>360780</sub> ratios were greater than 1,2, which indicated that the RNA was of higher purity than that extracted using the GTC protocol. Additionally, distinct ribosomal bands could be seen on a denaturing gel (Figure 1, Lanes 3 and 6).

The superiority of the CTAB method over the GTC method was likely due to several reasons. First, PVP in the CTAB extraction buffer forms a complex with polyphenols through hydrogen bonding, allowing them to be separated from RNA, and reducing levels of polyphenol in the extracted RNA (Maliyakal, 1992). Second, the addition of PVP has been noted as being essential for isolating RNA from plant tissues containing high levels of polysaccharides and phenolic compounds (Lewinsohn et al., 1991). Third, EDTA in the extraction buffer can inhibit polyphenoloxidases (Van Driessche et al., 1984). And lastly, the LiCl extraction step used in this CTAB protocol most likely resulted in a reduction of polysaccharide contamination as previously discussed. The only disadvantage of the CTAB method is that it is more labor intensive than the GTC protocol

Once suitable RNA was obtained using the GTC protocol for ambient temperature-treated sugarcane and the CTAB protocol for high temperature-treated sugarcane, the effects of elevated C0<sub>2</sub> and high temperature *onpepc* and *rbcS* transcript abundance were examined by northern blot analyses for the two cultivars CP 73-1547 and CP 80-1827 (Figure 2). Transcript levels in plants subjected to ambient C0<sub>2</sub> and temperature dramatically decreased the transcript abundance of *bothpepc* and *rbcS* genes. Transcript levels of *pepc and rbcS* were reduced by 78% and 81%, respectively in CP 73-1547 (Figure 2, column 2), and 69% and 89%, respectively in CP 80-1827 (Figure 2, column 6). This reduction in transcript abundance was even more pronounced in plants grown at ambient C0<sub>2</sub> and high temperature, where mRNA levels of *pepc* and *rbcS* were reduced by 94% and 87%, respectively in CP 73-1547 (Figure 2, column 4), and 99% and 98%, respectively in CP 80-1827 (Figure 2, column 8).

Results from this study showed dramatic decreases in sugarcane *pepc* and *rbcS* gene expression under high growth temperature for both  $CO_2$  treatments. However, elevated  $CO_2$  may partially alleviate the adverse effects of high temperature. Under elevated  $CO_2$  and high temperature, the reduction in *pepc* and *rbcS* transcript levels was less severe, compared to the ambient  $CO_2$  and high temperature treatment.

Under ambient temperature and elevated C0<sub>2</sub>, changes in *pepc* and *rbcS* transcript accumulation varied with the cultivar examined. For CP 73-1541, *pepc* transcript levels decreased by 44% (Figure 2A, column 1), while *rbcS* transcript levels remained relatively unchanged (Figure 2B, column 1). For CP 80-1827, accumulation *of pepc* transcripts was not reduced (Figure 2A, column 5); however, accumulation of *rbcS* mRNA increased by 64% (Figure 2B, column 5).

The effect of  $C0_2$  enrichment on *pepc* transcript accumulation has not been reported until now for a  $C_4$  crop; however, it has been shown for sugarcane that PEPC activity is down-regulated in response to a doubling [ $C0_2$ ] (Vu et al., 1998). Our results suggest that such a reduction in PEPC activity may be due to a reduction in the steady-state level of *pepc* mRNA. There is more information in the literature on *rbcS* expression for  $C_3$  plants under  $CO_2$  enrichment; however, the expression of *rbcS* genes in response to elevated  $CO_2$  might be different between  $C_3$  and  $C_4$  plants. Decreases in *rbcS* transcript levels have been shown for many  $C_3$  plants grown under elevated  $CO_2$  (Cheng et al, 1998; Nie et ai, 1995; Gesch et al, 1998). However, our data showing an increase in *rbcS* transcripts under elevated  $CO_2$  in sugarcane coincide with results found for another  $C_4$  plant, maize (Moore et al, 1998).

Previous work on effects of rising  $CO_2$  and high temperature on sugarcane growth revealed that elevated  $CO_2$  and temperature increased leaf area, total above-ground biomass, and juice volume without an enhancement of leaf  $CO_2$  assimilation rates (Vu et al., 1998). For sugarcane, as well as other  $C_4$  plants, the reasons for observed growth stimulation by elevated  $CO_2$  and temperature remain uncertain. Since the photosynthetic mechanisms operating in a crop species are the major determinants of how it will respond to both rising  $CO_2$  and temperature, work on the regulation of expression of key photosynthetic and carbon metabolism genes should expand our understanding of the photosynthetic mechanisms as well as growth and yield mechanisms underlying responses of this  $C_4$  crop in a future climate-changed world.

## ACKNOWLEDGEMENTS

This work was supported in part by the University of Florida/IFAS, USDA-ARS and the Furnin Foundation. We are grateful to M. Matsuoka for supplying the *maize pepc* clone and S.S.M. Sun for providing the sugarcane *rbcS* clone. The authors also wish to thank Drs. D.S. Wofford and R.L. Smith for critical review of this manuscript. This work was approved for publication by the Florida Agricultural Experiment Station as Journal Series No. R-07697.

#### REFERENCES

- Bahloul, M., G. Burkard, 1993. An improved method for the isolation of total RNA from spruce tissues. Plant Mol. Biol. Rep. 11:212-215.
- Bowes, G., 1991. Growth at elevated C0<sub>2</sub> :photosynthetic responses mediated through Rubisco. Plant Cell Environ. 14:795-806.
- Cheng, S.H., B.D. Moore, J. R. Seemann, 1998. Effects of short- and long-term elevated CO<sub>2</sub> on the expression of ribulose-I,5-bisphosphate carboxylase/oxygenase genes and carbohydrate accumulation in leaves of *Arabidopsis thaliana* (L.) Heynh. Plant Physiol. 116:715-723.
- Chomczynski, P., N. Sacchi, 1987. Single-step method of RNA isolation by acid guanidinium tmocyanate-phenol-chloroform extraction. Anal. Biochem. 162:156-159.
- Dong, J-Z., D.I. Dunstan, 1996. A reliable method for extraction of RNA from various conifer tissues. Plant Cell Rep. 15:516-521.

- Gesch, R.W., K.J. Boote, J.C.V. Vu, L.H. Allen Jr., G. Bowes, 1998. Changes in growth CO<sub>2</sub> results in rapid adjustments of ribulOse-1,5-bisphosphate carboxylase/ oxygenase small subunit gene expression in expanding and mature leaves of rice. Plant Physiol. 118:521 -529.
- Geuna, F., H. Hartings, A. Scienza, 1998. A new method for rapid extraction of high quality RNA from recalcitrant tissues of grapevine. Plant Mol. Biol. Rep. 16:61-67.
- Graham, C. C, 1993. A method for extraction of total RNA from *Pinus radiata* and other confiers, Plant Mol. Biol. Rep. 11:32-37.
- Gu, H. Y., LJ. Qu, X.T. Lin, 1997. Plant Gene and Molecular Manipulation. Beijing University Press, Beijing, pp 25-37.
- Keifer, E., W. Heller, D. Ernst, 2000. A simple and efficient protocol for isolation of functional RNA from plant tissues rich in secondary metabolites. Plant Mol. Biol. Rep. 18:33-39.
- Lay-Yee, M., D. Della-Peima, G.S. Ross, 1990. Changes in mRNA and protem during ripening of apple fruit. Plant Physiol. 94:850-853.
- Lewinsohn, E., M. Gijzen, R. Croteau, 1991. Defense mechanisms of conifers: Relationship of monoterpene cyclase activity to anatomical specialization and oleoresin monoterpene content. Plant Physiol. 96:38-43.
- Lewinsohn, E., C. L. Steele, R. Croteau, 1994. Simple isolation of functional RNA from woody stems of gymnosperms. Plant Mol. Biol. Rep. 1:20-25.
- Logemann, J., J. Schell, L, Willmitzer, 1987. Improved method for the isolation of RNA from plant tissues. Anal. Biochem. 163:16-20.
- Loomis, W.D., 1974. Overcoming problems of phenolics and quinones in the isolation of plant enzymes. Method Enz. 13:555-563.
- Maliyakal, E.J., 1992. An efficient method for isolation of RNA and DNA from plants containing phenolics. Nuc. Acid Res. 20:2381-2386.
- Manning, K., 1991. Isolation of nucleic acids from plants by differential solvent precipitation. Anal. Biochem. 195:45-50.
- Moore, B.D., S.H. Cheng, J. Rice, J. Seemann, 1998. Sucrose cycling, Rubisco expression and prediction of photosynthetic acclimation to elevated atmospheric C0<sub>2</sub>. Plant Cell Environ. 21:905-915.
- Matsuoka, M., E. Minami, 1989. Complete structure of the gene for phosphoenolpyruvate carboxylase from maize. Eur, J. Biochem. 181:593-598.

- Nie, G.Y., D.L. Hendrix, A. N. Weber, B. A, Kimball, S. P. Long, 1995. Increased accumulation of carbohydrates and decreased photosynthetic gene transcript levels in wheat grown at an elevated C0<sub>2</sub> concentration in the field. Plant Physiol. 108: 975-983.
- Puissant, C, L-M. Houdebine, 1990. An improvement of the single-step method of RNA isolation by acid guanidinium thiocyanate -phenol-chloroform extraction. Biotechniques 8:148-149.
- Sage, R.F., 1994. Acclimation of photosynthesis to increasing atmospheric C0<sub>2</sub>: the gas exchange perspective. Photosynth. Res. 39:351-368.
- Schneiderbauer, A., H. Sandermann Jr., E. Ernst, 1991. Isolation of functional RNA from plant tissues rich in phenolic compounds. Anal. Biochem. 197:91-95.
- Schultz, D.J., R. Craig, D.L. Cox-Foster, R.O. Mumma, J.I. Medford, 1994. RNA isolation from recalcitrant plant tissue. Plant Mol. Biol. Rep. 12:310-316.
- Sinclair, T.R., L.H. Jr. Allen, G. M. Drake, 1995. Temperature gradient chambers for research on global environmental change. H. Design for plot studies. Biotronics 24:99-108.
- Tang, W., S.S.M. Sun, 1993. Sequence of a sugarcane ribulose-1,5-bisphosphate carboxylase/oxygenase small subunit gene. Plant Mol. Biol. 21:949-951.
- Van Driessche, E., S. Beeckmans, R. Dejaegere, L. Kanarek, 1984. Thiourea: The antioxidant of choice for the purification of protein from phenol-rich plant tissues. Anal. Biochem. 141:184-188.
- Vu, J.C.V., L. H. Allen, Jr., G. Bowes, 1998. Growth and photosynthesis response of sugarcane to high CO<sub>2</sub> and temperature. Plant Biology 98: Abstract No. 448 (Final Program of the Annual Meeting of the American Society of Plant Physiologists, Madison, WI, June 27~July 1,1998).
- Vu, J.C.V., L.H. Allen, Jr., M. Gallo-Meagher. 2000. Crop Plants Responses to Rising CO<sub>2</sub> and Climate Change. In: Handbook of Plant and Crop Physiology. Ed. M. Pessarakli, Marcell Dekker, Inc. New York. (In press).
- Wang C. S., L.O. Vodkin, 1994. Extraction of RNA from tissues containing high levels of procyanidins that bind RNA. Plant Mol. Biol. Rep. 2:132-145.
- Webber, A.N., G.Y. Nie, S.P. Long, 1994. Acclimation of photosynthetic proteins to rising atmospheric C0<sub>2</sub>. Photosynth Res. 39:413-425.

CO <sub>2</sub> (ppm)	Temp. (° C)	Protocol	RNA yield (µg/gFW) <sup>‡</sup>	A260/200	A <sub>260/230</sub> 1	Maximal Wavelength _Abs. (nm)
700	T,¥	GTC	93.4 ± 5.9 a	1.86 ± 0.06 a	1.24 ± 0.05 ab	260
	T2 <sup>€</sup>	GTC	55.1 ± 30.2 b	1.51 ± 0.07 b	$0.73 \pm 0.18$ c	255
	T2	CTAB	55.2 ± 4.0 b	1.73 ± 0.06 a	$1.38 \pm 0.03$ a	260
360	Τι	GTĆ	64.1 ± 14.0 a	1.75 ± 0.11 a	1.14±0.04b	260
	T <sub>2</sub>	GTC	$\textbf{43.1} \pm \textbf{10.1} \text{ b}$	$1.49 \pm 0.07$ b	0.69 ± 0.06 c	255
	T <sub>2</sub>	CTAB	44.6 ± 6.8 b	1.71 ± 0.03 a	1.26 ± 0.12 ab	260

Table 1. Total RNA isolated from sugarcane exposed to ambient and elevated C0<sub>2</sub>and temperature treatments using the GTC and CTAB protocol

<sup> $\overline{t}$ </sup> Data represent the mean values and standard deviations of two replicates for each of two cultivars for a total of four samples per treatment and values followed by the same letter within a column are not significantly different at P = 0.05.

<sup>1</sup>One microgram per gram of fresh weight

<sup>4</sup>Ratio of spectrophotometric absorbance values at 260 nm and 280 nm

Ratio of spectrophotometric absorbance values at 260 nm and 230 nm

\* 1.5C° above outside ambient

 $\mathbf{E}6.0C^{\circ}$  above outside ambient



Figure 1. Total RNA from sugarcane leaves exposed to ambient C0<sub>2</sub> or double-ambient C0<sub>2</sub>, along with either ambient or high temperature using the GTC or CTAB protocol. Five micrograms of total RNA was fractionated on a 1% agarose gel containing 18% formaldehyde, stained with ethidium bromide. Lane M: RNA molecular weight marker; Lane 1: double-ambient C0<sub>2</sub>, ambient temperature, GTC; Lane 2: double-ambient C0<sub>2</sub>, high temperature, GTC; Lane 3: double-ambient C0<sub>2</sub>, high temperature, GTC; Lane 5: ambient C0<sub>2</sub> high temperature, GTC; and Lane 6: ambient C0<sub>2</sub> high temperature, CTAB.



Figure 2. Transcript levels of *pepc* (A) and *rbcS* (B) genes in sugarcane leaves grown in paired companion temperature-gradient greenhouses. Data were expressed as a percentage of the transcript level obtained for each gene from plants exposed to ambient CO<sub>2</sub> and ambient temperature. Columns 1 and 5: double-ambient CO<sub>2</sub>, ambient temperature; Columns 2 and 6; double-ambient CO<sub>2</sub>, high temperature; Columns 3 and 7: ambient CG<sub>2</sub>, ambient temperature; Columns 4 and 8; ambient CO<sub>2</sub>, high temperature.

## NOTES ON THE YELLOW SUGARCANE APHID SIPHA FLAVA(HOMOPTERA: APHIDIDAE) AND THE LADY BEETLE DIOMUS TERMINATUS (COLEOPTERA; COCCINELLIDAE) IN FLORIDA

David G. **Hall** Research Department United States Sugar Corporation P.O. Drawer 1207 Clewiston,FL 33440

#### ABSTRACT

In a greenhouse study comparing the growth of young sugarcane plants infested versus not infested by the yellow sugarcane aphid [Siphaflava (Forbes)] (plants averaged 13.8 cm tall to the top visible dewlap leaf at the beginning of the infestations) over a 3-wk period, aphids reduced the height of infested primary shoots by 36.2%, At the end of the test after harvesting and drying plant matter, infested plants weighed 71.7% less than non-infested plants. The lady beetle Diomus terminatus Say is a common predator of the vellow sugarcane aphid in Florida. Mass-rearing the beetle on yellow sugarcane aphids was investigated. Yellow sugarcane aphids were relatively easy to raise in our greenhouse during winter and spring but difficult during summer and fall. A sorghum-Sudangrass hybrid proved a convenient plant host for mass-rearing the aphid. D. terminatus was reared in a laboratory in large glass test-tubes on yellow sugarcane aphids. Adult beetles were placed with aphids into the tubes (10 aphids per adult beetle/day) along with a small piece of wax paper, on which the beetles oviposited. Eggs on wax paper were transferred to new tubes (an average of 68.0 eggs per tube) and supplied with aphids for developing larvae to feed upon (7 aphids per larva/day). Larvae pupated in the tubes, and adults were harvested. When mated females were held individually, they laid an average of 3.0 eggs per female per day and an average total of 41.9 eggs per female. Increasing the number of beetles per oviposition tube appeared to reduce fecundity. Across all densities of eggs placed into rearing tubes, an average of 39.4% eggs developed to the adult stage. The percentage success in rearing larvae to the adult stage increased as the number of larvae per rearing tube was decreased.

#### INTRODUCTION

The yellow sugarcane aphid [Siphaflava (Forbes)] is a pest of occasional importance in Florida sugarcane. This aphid feeds on leaves, and extensive feeding by the aphid can result in a reddening and/or yellowing of leaves and sometimes leaf death. Outbreaks of the yellow sugarcane aphid may occur at anytime of the year in Florida but are perhaps most common either during late spring and early summer before the onset of the rainy season or during late summer after the summer rains. A good review of *S. flava* as a pest of sugarcane and its pest status in Puerto Rico was compiled by Gaude et al. (1965). Box (1953) lists *S. flava* as occurring in sugarcane in Bernuda, the United States of America, Mexico, El Salvador, Panama, Cuba, Jamaica, Haiti, Dominican Republic, Puerto Rico, St. Croix, Leeward Islands, St. Lucia, Barbados, Trinidad, Venezuela, British Guiana, Brazil, Colombia, Peru and Argentina. The aphid was discovered in Hawaii during 1988.

Although empirical observations indicate the yellow sugarcane aphid can be an important economic pest (Gaud et al., 1965), specific information on the impact of the aphid on the growth and yield of sugarcane is lacking. Such information is needed to develop management guidelines for the aphid. Infestations of the yellow sugarcane aphid can currently be controlled in Florida sugarcane using insecticides. As an alternative to chemical control, plant resistance to the aphid (e.g., see White, 1990) and biological control strategies might be exploited in Florida sugarcane to reduce losses to the yellow sugarcane aphid.

With respect to exploiting biological control, economic outbreaks of the yellow sugarcane aphid might be prevented by making inundative or augmentative releases of certain natural enemies. Natural enemies that may attack the yellow sugarcane aphid in Florida include; the brown lace wing Micromus subanticus (Walker); the green lacewing Chrysoperla externa (Hagan); syrphid flies including Allograpta exotica (Wiedemann); lady beetles including Coleomegilla maculata fuscilabris (Mulsant), Cycloneda scmguinea (Lin.), Diomus terminatus Say, Hippodamia convergen Guerin, and Ola v-nigrum Mulsant; and entomopathogenic fungi includingAcrostalagmus spp (Hall, 1988; Hall and Bennett, 1994). The relative importance of these natural enemies in controlling the aphid is not known. No insect parasitoids are known to attack the yellow sugarcane in Florida or anywhere else. Populations of beneficial insects attacking the aphid often increase dramatically in a density-dependent fashion toward the end of an outbreak of the yellow sugarcane aphid, often too late to prevent economic damage by the aphid. This has particularly appeared to be true with respect to the coccinellid *Diomus terminatus* (Hall, unpublished). While a number of coccinellids and other insects have been reported as predators of S.flava (Box, 1953; Gaud et al, 1965), D. terminatus has only been reported as a predator of sugarcane aphids in Florida (Hall, 1987). However, the author collected D, terminatus in Louisiana sugarcane during September 1999 (specimens identified by M. Thomas, Florida Center for Arthropod Systematics, Gainesville). This beetle is common in eastern North America and is apparently restricted to mainland North America. Among the natural enemies known to attack the yellow sugarcane aphid in Florida, D. terminatus would be a good candidate for inundative or augmentative releases from the standpoint that the beetle is often one of the most common insect predators attacking the aphid (Hall, unpublished observations).

Reported here are results of a study on damage by the yellow sugarcane aphid to young sugarcane plants, an investigation into mass-rearing *Diomus terminatus*, and an overview of the morphology and biology of *D. terminatus*.

#### METHODS AND MATERIALS

#### Aphid Damage to Young Sugarcane Plants

Plants for the study were obtained from U. S. Sugar Corporation's Research Department, Pathology Division, Clewiston, Florida. Originally intended for another study, plants derived from tissue culture of CP73-1547 had been transplanted on 1/5/00 (at which time the plants were 3 to 5 cm tall) into organic soil in seedling trays (35x24 cm plastic tray, 104 cells per tray, individual rectangular cells 9 cm deep measuring 2.5x2.5 cm at the upper opening and tapering to 1.3x1.3 cm at the base) and maintained in a greenhouse. On 4/3/00, 40 of these plants were transplanted into

7.61 plastic pots containing organic soil. The plants were paired one week later according to visual appearance, primary shoot height (to the top visible dewlap, TVD), and number of emerged tiller shoots. Six of the plants could not be reasonably paired and were discarded, leaving 34 paired plants. One plant of each pair was arbitrarily assigned to be infested by aphids and the other plant was assigned to be a non-infested check. These seventeen pairs were divided into 3 groups, 2 groups of 6 pairs and 1 group of 5 pairs. The 3 groups were placed onto three different benches in a greenhouse. Each of the 3 groups was divided into 2 subgroups, one consisting of plants to be infested by aphids and one consisting of check plants. These subgroups were positioned 1.0 to 2.4m apart in order to reduce the spread of aphids from infested to non-infested plants. On 4/10/00, a single sorghum-Sudan leaf infested by 30 to 75 aphids was placed onto each of the plants assigned to be infested, and these leaves were left to allow aphids to migrate to the sugarcane plants. Data were collected just prior to infesting plants and weekly thereafter for 3 weeks on: primary shoot height (cm) to the TVD, number of leaves with visible dewlaps per primary shoot, number of leaves infested by aphids on the primary shoot, number of aphids on the TVD leaf, number of tiller shoots, and number of tiller shoots infested. The test was terminated on 5/4/00, at which time all plant material in each pot was cut at soil level, placed in paper bags and weighed for wet weights. The bags were then placed into a drying oven for 5 days, after which the dried plant material was weighed for dry weights. For statistical analyses, a paired t-test (a=0.05) was used to compare all variables except the number of tiller shoots infested, for which a simple t-test (a=0.05) was used.

#### Rearing Diomus terminatus on the Yellow Sugarcane Aphid

Yellow sugarcane aphids were reared in a greenhouse (natural sunlight, no supplemental lighting) on sorghum-Sudan [sorghum Sudangrass hybrid, Var. Kow Chow], and these aphids were used to rear *D. terminatus* in a laboratory during 1999 - 2000. To rear the aphids, sorghum-Sudan seeds were planted in organic soil using seedling trays (described previously, 2 or 3 seeds per tray cell). The seedling trays with sorghum-Sudan were maintained in trays containing 1 or 2 cm of water replenished every other day, circumventing the need for over-plant watering. Between 150 to 300 aphids were usually introduced onto each tray of plants several days after the plants emerged. Once aphid densities increased on the plants, infested leaves were excised and taken to a laboratory. The aphids from these leaves were used to rear *D. terminatus*. A brushing machine was frequently used for collecting aphids from leaves of infested sorghum-Sudan, although the brushing procedure killed some aphids.

To obtain eggs of the beetle, adult beetles (usually 10) were placed into a glass test tube (15 cm tall, 2.2 cm inside diameter) along with aphids (a ratio of about 10 aphids per beetle were usually maintained in each tube). Prior to introducing the beetles, a 1.5x6.0 cm piece of wax paper was pushed down into the test tube so that the wax paper covered the bottom end of the tube. After introducing the beetles and aphids, the tube was plugged with a cotton ball wrapped in tissue paper. The beetles fed on the aphids and usually oviposited on the wax paper. Wax paper with eggs was harvested daily and placed into separate test tubes (plugged with a cotton ball wrapped in tissue paper) for incubation, usually no more than 100 eggs per tube. Aphids were introduced (usually 7 aphids per larva) as eggs began hatching, and new aphids were added every day or two until the beetle larvae pupated. Pupae were held in the tubes until adults emerged. To harvest newly emerged

adults, the tubes were emptied into a glove box and a vacuum pump aspirator was used to collect beetles into new test tubes.

Data were collected on numbers of eggs harvested daily from the rearing tubes and on the numbers of adult beetles recovered from eggs, from which an estimate of rearing success was made. Numbers of eggs harvested per rearing tube was used as an estimate of fecundity. Fecundity was further studied by holding 53 newly-emerged adults in a container for 24 hours, after which they were placed individually into glass microtubes (7.4 cm tall, 0.9 cm inside diameter) along with a 0.5x3.0 cm piece of wax paper pushed down into the test tube so that the wax paper covered the bottom end of the tube. Ten live aphids were maintained in each microtube to feed each beetle, and beetle eggs were harvested daily until the beetles died. All rearing was conducted at 27.7° C (std 3.9) with a 16L:8D photoperiod, and all tubes containing insects were held in an air-tight translucent box along with a moist paper towel at about 70% RH.

#### Diomus terminatus Morphology, Biology and Rearing Parameters

The general morphology and biology of *D*, *terminatus* as observed under the rearing procedure was described.

## RESULTS AND DISCUSSION

#### Aphid Damage to Young Sugarcane Plants

At the beginning of the test, there were no significant differences between paired plants with respect to the height of primary shoots (mean 13.4 cm), number of leaves per primary shoot (mean 2.3 leaves with visible dewlaps), number of leaves infested (mean 0.0), number of dead leaves per primary shoot (mean 0.0), or number of tiller shoots (mean 0.97). During 4/10/00 - 5/4/00 among infested plants, an average of 3.2 leaves per primary shoot was colonized by aphids with a mean of 99.2 aphids on the TVD leaf (Table 1). In contrast among check plants during this period, an average of only 0.2 leaves per primary shoot was colonized with a mean of only 0.4 aphids on the TVD leaf.

At the end of the 3-week infestation, the primary shoots of infested plants were 36.2% shorter and had produced 16% fewer leaves than check plants. Based on visual examinations of leaves, the extent of reddening and death of leaf tissue of infested plants was severe. The aphid infestations had a negative impact on tiller production, as infested plants averaged 0.9 live tillers per primary shoot after the 3-wk infestation while check plants averaged 5.9 live tillers per primary shoot. Plant material from the infested versus non-infested plants weighed 7.47 versus 39.61g per plant, respectively, before drying and 1.87 versus 6.6 g per plant, respectively, after drying. At the end of the test, whether or not the infested plants would have recovered from the aphid infestations had the aphids been controlled remained questionable. One week after the plants had been harvested, 94.1 % of the plants which had not been infested were regenerating shoots; in contrast, only 35.3% of the plants which had been infested were regenerating shoots, and these shoots appeared feeble. The data showed that the 3-wk infestation of yellow sugarcane aphids had a significant negative effect on the development of young sugarcane plants. Based on the study, when infestation densities of the aphid reach above 100 aphids per TVD leaf and most lower leaves are infested, growth reductions in young cane including the inhibition of tiller production may occur in some varieties of sugarcane. Infestation levels as high as this have only infrequently been observed in young sugarcane plants in Florida over the past 20 years. It is probable that infestation levels lower than 100 aphids per TVD leaf may also negatively affect plant development but to a lesser degree. The quantitative impact of the aphid on the development of older plants remains to be assessed. While this study provided insight into the potential importance of the yellow sugarcane aphid as a pest of cane, more research is needed. In particular, quantitative studies relating infestation densities of the aphid to yield of sucrose are needed, data which would be useful for identifying aphid densities which warrant control. Because low infestation densities of the yellow sugarcane aphid may be present year-round in Florida sugarcane, an ecological investigation to determine environmental conditions which favor rapid increases in aphid infestation densities might be of value in helping growers predict when aphid outbreaks might occur.

#### Rearing Diomus terminatus on the Yellow Sugarcane Aphid

Sorghum-Sudan appeared to be a good host for rearing the yellow sugarcane aphid. The sorghum-Sudan plants generally emerged within 2 or 3 days after planting, and aphids could be introduced onto the young plants as early as 5 to 7 days after planting. During late winter and spring (in 1999 and again in 2000) when average air temperatures in the greenhouse were in the range of 23 to 27°C and maximum daily air temperatures did not exceed around 34°C, the aphid rapidly colonized and reproduced on sorghum-Sudan grown in seedling trays in the greenhouse. Following introductions of 150 to 300 aphids per tray, aphid population levels in excess of an estimated 5,000 aphids per tray were often reached within several weeks. For example, among 18 trays of sorghum-Sudan into which an average of 174 aphids were introduced at a mean of 13.2 days after planting, aphid populations per tray were estimated to have reached 600, 2,000 and 5,700 aphids per tray after 1, 2 and 3 weeks, respectively. The plants sometimes began to die off quickly due to damage by the aphids when densities per tray reached 5,000 or more. Considerable difficulty was experienced during the summer and fall 1999 getting aphids to multiply on sorghum-Sudan in the greenhouse, which initially was attributed to adversely high air temperatures in the greenhouse. During late fall and early winter 1999 when average air temperatures dropped to the range of 23 to 27°C, difficulties in rearing aphids in the greenhouse continued. During late January 2000, these difficulties disappeared. Air temperature, photoperiod and/or other factors were apparently more favorable for rearing the aphid in our greenhouse during late winter and spring than during summer and fall.

## Diomus terminatus Morphology, Biology and Rearing Parameters

Individual eggs ranged from 0.6 to 0.7 mm in length and were oval with a dome-shaped appearance. The eggs were translucent-yellow when first laid and turned a dark green as they matured. Larvae were very small and greenish-yellow in color when they first emerged from the eggs. As the larvae matured, they turned dark greenish in color. In rearing tubes, pupation

occurred directly on wax paper, the inside of the glass tube, and on the cotton-tissue plug. Newly-emerged adults were light brown or tan in color but soon turned shiny black. *D. terminatus* adults measured approximately 2.0 mm in length.

Female beetles often oviposited individual eggs in a scattered pattern across the oviposition surface, with perhaps several mm up to a cm or more between the eggs; sometimes, however, a short, consecutive row of several eggs was oviposited. After the oviposition process, individual eggs were firmly attached to the oviposition surface. The developmental times at 27.7°C generally appeared to be about 3 d for the egg stage, 10 d for the larval stages, and 4 d for pupal stage, but a good quantitative assessment of developmental times was not made. Adults sometimes lived for several weeks in the laboratory. No data were collected on adult feeding rates, but general observations indicated adults consumed 5 to 10 aphids daily depending on the age of aphids. Characters for separating live female and male beetles were not determined.

Of the 53 beetles held individually in microtubes, 22 laid eggs. The beetles that did not lay eggs were presumed to be males (indicating 58% were males) but perhaps could have been unmated females or females that did not lay eggs in the tubes. An average of 3.0 eggs per female per day were laid by the 22 females, with an average total of 41.9 eggs (std 54.8) per female (minimum and maximums of 1 and 178 eggs per female, respectively). The females lived an average of 17.0 days (std 7.9). In larger rearing tubes with 10 beetles per tube, an average of 1.4 eggs (std 1.5) per female per day were laid (50% beetles per tube assumed to be female). Although fecundity appeared to be reduced by holding 10 beetles together in the larger tubes, less time was required to maintain the larger tubes and problems associated with not being able to identify sexes were reduced. Overall, egg production by *D. terminatus* appeared to be relatively low, at least in the laboratory. Supplying honey-water or sugar-water in tubes as an additional food source for beetles did not increase fecundity (data not presented).

Among rearing tubes containing an average of 68.0 eggs (range 2-210, std 60.9, stderr 8.8, n=48), an average of 39.4% eggs (std 25.4, stderr 4.04, n=48) developed to the adult stage. Percentage success in rearing eggs to the adult stage increased as the number of eggs per tube was decreased (Figure 1). Increased competition at higher densities of *D. terminatus* larvae per tube may have been responsible for relatively poor success in getting adults from eggs. A less favorable environment for larval development may have been present in tubes with large numbers of aphids. Whether or not *D. terminatus* larvae were cannibalistic was not determined. No good quantitative data were collected on larval consumption rates, but general observations indicated that larvae may have consumed 2 or 3 aphids daily during early larval instars up to 6 or 7 aphids daily during later instars.

Overall, *Diomus terminatus* was relatively easy to rear in the laboratory on yellow sugarcane aphids grown on sorghum-Sudan in a greenhouse. One individual rearing aphids in a greenhouse and *D. terminatus* in a laboratory could use the rearing procedures described in this paper to easily produce perhaps 500 adult *D. terminatus* weekly, provided a continual, abundant supply of aphids is available. As discussed in this report, we had difficulty rearing aphids during the summer and fall and, consequently during this time period, we were unable to mass-rear *D. terminatus*. Although the beetle's natural affinity for yellow sugarcane aphids and it's common occurrence in Florida
sugarcane support *D. terminatus* as a candidate for inundati ve or augmentative releases for managing the aphid, the fecundity and aphid consumption rates of other candidates may exceed those of *D. terminatus*. Also, other candidates might be easier and less expensive to rear than *D. terminatus*. It remains unknown whether releases of *D. terminatus* before or early during an outbreak of yellow sugarcane aphids would be effective in reducing economic damage by the aphid.

## ACKNOWLEDGMENTS

I thank Sherry Little, Research Laboratory Technician, for her help in rearing the aphids and beetles.

### REFERENCES

- Box, H. E. 1953, List of sugar-cane insects. Commonwealth Institute of Entomology, London. CAB. 101pp.
- Gaud, Silverio Medina, L. F. Martorell and R. Bonilla Robles. 1965. Notes on the biology and control of the yellow aphid of sugarcane, *Sipha flava* (Forbes) in Puerto Rico. Proc. 12<sup>th</sup> Congress International Society of Sugar Cane Technologists. 1307-1320.
- Hall, D. G. 1987. The sugarcane aphid, *Melanaphis sacchari*, in Florida. Journ. Am. Soc. Sugar Cane Techn. 7:26-29.
- Hall, D. G. 1988. Insects and mites associated with sugarcane in Florida. Fla. Entomol. 71 (2): 138-150.
- Hall, D. G. and F. Bennett. 1994. Biological control and IPM of sugarcane pests in Florida. In Pest Management in the Subtropics, Biological Control - a Florida Perspective. Eds. D. Rosen, F. D. Bennett and J. L. Capinera. Intercept Ltd., Andover, UK. 737pp.
- White, W. H. 1990. Yellow sugarcane aphid (Homoptera: Aphididae) resistance mechanisms in selected sugarcane cultivars. Journ. Econ. Entomol. 83(5): 2111-2114.

**Table 1.** Data collected from young sugarcane shoots (CP73-1547) infested versus not infested by yellow sugarcane aphids for 3 weeks, infestations beginning 4/10/00 (17 plants infested, 17 plants not infested)<sup>a</sup>.

	М	ean number of	aphids per TVI	۶Ľ	
	4/10/00	4/20/00	4/27/00	<u>5/4/00</u>	
Intested	0.0 a	128.1 a	141.9 a	126.7 a	
Not Infested	0.0 a	0.4 Ъ	0.1 b	1.2 b	
		Mean number	leaves infested		
	4/10/00	4/20/00	4/27/00	5/4/00	
Infested	0.0 a	3.6 a	4.7 a	4.6 a	
Not Infested	0.0 a	0. <b>4 b</b>	0.2 в	0.6 b	
		Mean hei	ght (cm) <sup>4</sup>		
	4/10/00	4/20/00	4/27/00	5/4/00	
Infested	13.4 a	13.4 в	14.1 a	14.7 a	
Not Infested	13.5 a	14.5 a	18.4 b	23.0 b	
		Mean num	ber leaves'		
	4/10/00	4/20/00	4/27/00	5/4/00	
Infested	2.4 a	4.6 a	4.7 a	4.9 a	
Not Infested	2.2 a	5.3 a	5.5 a	5.9 Ъ	
	Mean n	umber live tille	r shoots/prima	y shoot	
	4/10/00	4/20/00	4/27/00	5/4/00	
Infested	1.0 a	1.1 a	0.9 a	0.9 a	
Not Infested	0.9 a	1.8 a	3.8 Ъ	5.2 b	
	Mean number t	iller shoots infe	sted/primary sl	100t with tillers	
	4/10/00	4/20/00	4/27/00	5/4/00	
Infested	0.0 B	1.0 a	1.3 a	1.3 a	
Not Infested	0.0 a	0.1 Ъ	0.1 Б	0.2 h	

<sup>a</sup>Each pair of means in the same column followed by the same letter are not significantly different [paired t-test (a=0.05) for all variables except tiller shoots infested, for which a simple t-test was used(a=0.05)].

<sup>b</sup>TVDL is the uppermost leaf with a visible dewlap

<sup>c</sup>on primary shoots

<sup>d</sup>to TVDL of the primary shoot

eleaves with visible dewlaps on the primary shoot



Figure 1. Percent success rearing adults of *D. terminatus* from eggs on wax paper placed into glass test tubes (7-10 yellow sugarcane aphids per egg maintained daily in tubes).

## FERTILIZER EFFECTS OF OLDER SUGARCANE RATOON CROPS IN LOUISIANA

# Chris LaBorde, Kenneth Gravois, and Keith Bischoff

Sugar Research Station Louisiana Agricultural Experiment Station LSU Agricultural Center, St. Gabriel, LA 70776

#### ABSTRACT

Louisiana sugarcane (Saccharum spp.) farmers are undergoing a trend change in the ration longevity of their sugarcane crops. For the last several decades in Louisiana, the intensity of ratooning in sugarcane has usually been a plantcane crop plus two ratoon crops. Since its release in 1993, the sugarcane variety LCP 85-384 has surpassed expectations in Louisiana due to its increased ratooning ability. The main objective of this study was to determine the response of LCP 85-384 to higher than recommended rates of nitrogen (N) in third and fourth ration crops. A secondary objective was to determine potassium (K) and phosphorus (P) responses. Fertilizer treatments were evaluated at two locations during 1998-1999: Blackberry Farms near Vacherie, Louisiana, and Triple V Farms near Youngsville, Louisiana. The experiments were planted as a Latin square design with six fertilizer treatments and six replications. The year by treatment interaction was nonsignificant (P > 0.05) for each location; therefore, treatment means were averaged across the two years. For the Vacherie location, only cane yield was significantly affected by the treatments. Orthogonal contrasts indicated a significant increase in cane yield between the medium (168 kg/ha) and high (224 kg/ha) N rates. Potassium significantly increased cane yield between the low (0 kg/ha) and high (112 kg/ha) rates. At the Youngsville location, cane yield was not significantly increased by the N rates. Orthogonal contrasts indicated a numeric increase in both sugar and cane yield due to K application between the low (0 kg/ha) and high (112 kg/ha) rates. For fourth ration cane at Blackberry Farms, the higher N rate increased cane yield over the medium N rate. The third ratoon LCP 85-384 cane at Youngsville exhibited no response to higher N rates, Potassium increased cane and sugar yield in Youngsville, while it only increased cane yield in Vacherie. Phosphorus did not produce a significant cane or sugar vield response at either location.

## INTRODUCTION

Louisiana sugarcane farmers have increased the ratoon longevity of their sugarcane crops by the recent adoption of the new sugarcane variety LCP 85-384. For the last several decades, the ratooning intensity of sugarcane in Louisiana has usually been a plant crop plus two ratoon crops. Current problems inhibiting ratoon longevity consist of an increase in weeds and diseases in ratoon cane, in addition to climatic conditions that may necessitate a cold-tolerant variety (Viator et al. 1987). Finding a variety that would nullify the current problems inhibiting ratoon longevity should increase farm productivity and profitability.

Since its release in 1993, the variety LCP 85-384 has surpassed yield expectations in the Louisiana sugar industry. It has become very popular because of its high cane yields, high sucrose concentration, and excellent ratooning ability and disease resistance. The movement toward increased combine harvesting is stimulating interest in growing LCP 85-384 because of the variety's propensity for lodging. The total statewide acreage for LCP 85-384 has increased from 1% in 1994 to 58 % in 1999 (Gravois 1999). Never before in the history of Louisiana sugarcane production have yields been achieved that rival that of LCP 85-384. The year 1999 marked the fifth consecutive year that the industry will have produced more than 900,000 tonnes of sugar. Numerous individual records were set by both growers and factories in 1999. LCP 85-384 has performed so well that it is now being used as the standard check variety replacing CP 70-321 (Garrison et al. 1999). LCP 85-384 has allowed the industry to survive despite a lower price for sugar (Richard 1999). With prices as low as 8 cents per kg for sugar in 1999, many facets of farm management need to be looked at to run a profitable and efficient operation.

Current Louisiana fertilizer recommendations do not take into account third and fourth ration sugarcane crops because of the lack of a variety to make adequate crop yields beyond second ration. Currently, farmers are using recommended fertilizer rates for second ration crops on their third and fourth ration crops. Louisiana's fertilizer recommendations are based on crop, with plant cane usually receiving less N than ration crops, and on crop stands, where higher tonnage crops should receive more N than lower tonnage crops. Also, cane grown on heavier clay soils should receive higher N rates than sugarcane grown on other Louisiana soil types (Faw and Funderburg 1995). Applications of P and K are based on soil test results in Louisiana. Schroeder et al. (1998) summarized fertilizer advisory practices for South Africa and Australia. In the Australian sugar industry, fertilizer recommendations are based on general production functions and/or critical values. In contrast, the South African sugar industry is more soil- and region-specific and is based on managing nutrients and soils according to their chemical and physical properties.

Only limited information exists regarding the fertility needs of LCP 85-384 in older ration crops in Louisiana. Therefore, the main objective of this study was to determine the response of LCP 85-384 to higher than the recommended rates of N in older ration crops. A secondary objective was to determine K and P responses.

#### MATERIALS AND METHODS

A two-year fertilizer experiment beginning in 1998 was conducted on two third and two fourth ratoon crops at two locations in Louisiana. One of the locations was Blackberry Farms in Vacherie, and the other location was Triple V Farms in Youngsville. In 1998, the experiment at Blackberry Farms consisted of fourth ratoon LCP 85-384, and the experiment at Triple V Farms consisted of fourth ratoon LCP 85-384. In 1999, the experiment at Blackberry Farms consisted of fourth ratoon LCP 85-384. The soil type for the experiment at Blackberry Farms in 1998 was a Commerce silt loam (*Fine-silty, mixed, nonacid, thermic Aeric Fluvaquents*). The soil type at Blackberry Farms in 1999 was a Commerce silt yelay loam (*Fine-silty, mixed, nonacid, thermic Theric Fluvaquents*). The soil type at Triple V Farms for both years was a Memphis silt loam (*Fine-silty, mixed, thermic Typic Hapludalfs*). The experimental design at each location was a Latin square that wer 7.6 meters long with a 1.5-meter alley between each plot. An assumption for the Latin square design uses

rows and columns as blocking factors. Each treatment occurs once in each of the blocking factor level combinations as described by Freund and Wilson (1993).

The fertilizer treatments for the field experiments are shown in Table 1. The fertilizer formulations were blended by using a cement mixer to ensure proper blending. The sources of N. P, and K were urea, triple superphosphate, and muriate of potash, respectively. In addition to weighing the fertilizer formulation for each treatment, the fertilizer formulation was divided into three separate bags for each row in every experimental unit to minimize application error of the granular fertilizer. The treatments were applied by hand to simulate fertilizer banding. The fertilizer was placed in the off-bar V on each side of the row. Fertilizers were applied in late March or early April.

Data collected from each experiment included soil samples taken prior to fertilization of the experiment to identify soil-test levels and the physical properties of each soil type. In 1998, multiple soil samples were taken and mixed for a representative soil sample from each of the two experiments. In 1999, each experimental unit was soil sampled in each of the two experiments. Fertilizer recommendations were obtained from the soil-test results, Stalk counts were done by counting unliable stalks in mid-August. Fifteen-stalk samples were randomly hand-harvested from the center row of each plot. The samples from each plot were cut even with the ground and topped just below the top most visible dewlap as demonstrated by Golden and Abdol (1977). After the removal of the leaves from each of the samples, bundle weights were measured. Harvest dates for Vacherie and Youngsville in 1998 were November 30 and November 16, respectively. Harvest dates for Vacherie and Youngsville in 1999 were November 16 and September 30, respectively, Brix and pol values from the harvested samples were determined at the St. Gabriel sucrose laboratory and were used to estimate the theoretical recoverable sugar per unit weight of cane (Gravois and Milligan 1992). Stalk weight, stalk height, and stalk diameter were recorded for each sample. Estimated cane vield was calculated as the product of the stalk population and stalk weight. Sugar vield was estimated as the product of cane vield and sucrose concentration.

Leaves were sampled from the plots as described by Golden (1972). The leaf blade samples were selected from the first leaf below the top most visible dewlap in mid-July. A leaf blade sample, composed of 20 leaf blades, was sampled from each plot. The leaf blade samples were oven-dried. ground, and analyzed for their total nutrient concentration. Leaves were analyzed at the LSU Department of Agricultural Chemistry. Critical and optimal nutrient levels of sugarcane plant tissue analyses are included in Table 2 (Anderson and Bowen 1990).

Data from each location were analyzed across years with the following model:

 $Y_{ijkl} = \mu + Y_{i} + R_{j(i)} + C_{k(i)} + T_{i} + YT_{i} + 8_{ijklt}$ where  $Y_{ijkl}$  was the observed response of the lift treatment (T) in the jth row (R) and kth column (C) within the ith year (Y); YT, was the interaction of year with treatment; µ was the overall mean, and  $\varepsilon_{aat}$  was the residual error. In the model, T = R = C = 6. Year and replication were considered random effects. When analyzed across years, the error term to test for significant treatment effects is the year by treatment interaction (Gomez and Gomez 1984).

The data were subjected to analysis of variance using the General Linear Model procedure of SAS (Freund and Wilson 1993). The significance level ( $P \le 0.05$ ) was used as the maximum

acceptable probability for rejecting a true null hypothesis. If the year by treatment interaction was not significant, means were averaged across the two years. Means were compared using orthogonal contrasts on the significant yield parameters for each location across the two years. Orthogonal contrasts were used to determine the effects of N on significant yield components by contrasting the three different levels of N in treatments 1, 2, and 3. Orthogonal contrasts were also used to determine P effects on any significant yield component by contrasting the different levels of P in treatments 2 and 5 with 4 and 6. Orthogonal contrasts were used to determine K effects on any significant yield component by contrasting the different levels of K in treatments 2 and 4 with 5 and 6.

A simple projected cost analysis per hectare of sugarcane was performed for the Vacherie and Youngsville experiments. The economic assumptions included in this analysis were a payment of \$20.94 per metric ton of cane based on \$0.42 per kg, a 40% mill share, a 16% landowner share, and a 10% recovery for sugar on a metric ton of cane. The per hectare return was calculated as the product of cane yield and the price or payment per metric ton of cane. Fertilizer cost was subtracted from the per hectare return.

## RESULTS AND DISCUSSION

#### Vacherie Results

At Vacherie, cane yield was the only response variable significantly affected by the fertilizer treatments (Table 3). Orthogonal contrasts indicated that both N and K significantly affected cane yield. Nitrogen significantly increased cane yield from the medium rate to the high rate. Therefore, we showed a positive cane yield response at a higher than recommended N rate. However, the sucrose content of the cane was not significantly increased, even at the higher N fertilizer rates (Table 3). Nitrogen treatments had no significant effect on the N concentration of sugarcane leaf blades, although there were numeric increases in succession from the 112 kg/ha treatment to the 224 kg/ha treatment (Table 4).

Phosphorus treatment effects on cane yield were determined by orthogonal contrasts with and without levels of K (Table 3). Phosphorus did not significantly increase cane yield. Because of a year by treatment interaction for P concentration of the sugarcane leaf blade, means were reported and tested for significance for each year (Table 4). In 1998, the P treatment significantly increased the P concentration of the sugarcane leaf blades. This is evidence that the sugarcane plants responded to the additional P fertilizer. Although the critical nutrient level for P in the sugarcane leaf blade is 0.14 (Table 2), the P concentration of the sugarcane leaf blades never fell below the critical level. The P concentration of the sugarcane leaf blades increased from 0.15 to 0.16 with P fertilizer (Table 4), which is above the critical nutrient concentration but still below the optimum nutrient concentration. The soil recommendation for phosphorus in 1998 was 17 kg P/ha. Even with a higher-than-recommended P fertilizer rate in 1998, P uptake into the plant was not maximized. In 1999, the P treatment had no significant effect on the P concentration of the sugarcane leaf blades, although there was a numeric increase from the 0 kg/ha treatment to the 29 kg/ha treatment. Soil-test recommendations indicated no need for P fertilizer in 1999, which would correspond to the adequate P concentration of the sugarcane leaf blades where no P was applied (Table 4).

Potassium treatment effects on cane yield were also determined by using orthogonal contrasts with and without P (Table 3). Potassium significantly increased cane yield at the Vacherie location from the 0 kg/ha treatment to the 112 kg/ha treatment. Potassium also significantly increased the K concentration of the sugarcane leaf blades from the 0 kg/ha treatment to the 112 kg/ha treatment (Table 4). The K concentration of the sugarcane leaf blades increased from 1.44 to 1.53 with K fertilizer, which is in the optimum nutrient concentration range (Table 2). Soil K recommendations were 66 kg K/ha in 1998 and 100 kg K/ha in 1999.

#### Youngsville Results

At Youngsville, fertilizer treatments did not significantly affect either sugar or cane yield in 1998 and 1999 (P=0.079 and P=0.078, respectively) (Table 5). Orthogonal contrasts indicated that N numerically (P=0.081) decreased cane yield from the low to the medium N rate. The inability to detect significant differences may be attributed to the high variability associated with older sugarcane ratoon crops. The CV's for sugar yield and for cane yield in 1998 and 1999 were 14.8% and 12.0%, respectively. Nitrogen concentration in the sugarcane leaves also was not significantly affected by N fertilizer treatments (Table 6). The N concentrations of the sugarcane leaves were near or slightly above the critical concentration for each of the N treatments (Table 2).

Phosphorus did not significantly increase sugar yield, cane yield, or sucrose content between the 0 kg/ha treatment and the 29 kg/ha treatment. Because of a year by treatment interaction regarding the P concentration of the sugarcane leaf blade, means were reported and tested for significance for each year. In 1998, the P treatments had no significant effect on the P concentration of the sugarcane leaf blades, whereas there was a numeric increase from the 0 kg/ha treatment to the 29 kg/ha treatment (Table 6). Although the critical nutrient concentration of P in the sugarcane leaf blade is 0.14, the P concentration of the sugarcane leaf blades never fell below the critical nutrient concentration. The P concentration of the sugarcane leaf blades increased from 0.14 to 0.16 with P fertilizer, which is above the critical nutrient concentration but still below the optimum nutrient concentration. The soil recommendation for P in 1998 was 22 kg P/ha. Despite a higher than recommended P fertilizer rate in 1998, P uptake into the plant was not maximized. In 1999, the P treatments had no significant effect on the P concentration of the sugarcane leaf blades. The P concentration of the sugarcane leaf blades was in the optimum nutrient range.

Potassium treatments resulted in a significant increase (P = 0.039) for sugar yield and a numeric increase (P = 0.073) for cane yield (Table 5). The K treatments significantly increased the K concentration of the sugarcane leaf blades from the 0 kg/ha treatment to the 112 kg/ha treatment (Table 6). The K concentration of the sugarcane leaf blades increased from 1.38 to 1.41 with the K fertilizer, which is in the optimum nutrient range (Table 2). Soil K recommendations for 1998 and 1999 were 0 kg K/ha.

## Economic Justification

Specific contrasts from the Vacherie location indicated that cane yield significantly increased as the N rate increased from 168 kg/ha treatment to 224 kg/ha. There was also a significant response in going from the 112 kg/ha treatment to the 224 kg/ha treatment. Potassium also increased cane yield from the 0 kg/ha treatment to the 112 kg/ha treatment. Since the

application costs of fertilizer for an increased rate will not cost any more than the price of the fertilizer itself (Champagne and Salassi 1999), the question is whether this increased rate will offset the costs of additional fertilizer.

Specific contrasts from Youngsville indicated that nitrogen decreased cane yield from the 112 kg/ha treatment to the 224 kg/ha treatment. Potassium increased cane and sugar yield from the 0 kg/ha treatment to the 112 kg/ha treatment. The yield parameter, cane yield, was compared for both Vacherie and Youngsville.

On fourth ratoon sugarcane at Vacherie, the medium (168 kg/ha) to high (224 kg/ha) N rates and the low (0 kg/ha) to high (112 kg/ha) K rates significantly increased cane yield. The N and K responses for these treatments show a return on a per hectare basis of \$111 and \$134, respectively (Table 7). Although these results warrant more fertilizer research, our data indicate that increases in N rates of 10-20% would be economically beneficial on the Mississippi River soil types. The lack of N fertilizer response at higher than recommended N rates at Youngsville indicates that older ratoon crop response may be affected by the yield potential of that crop. The cane yield was lower at Youngsville (Table 8) than at Vacherie. These data indicate that older stubble crops on soils similar to those at Youngsville would not respond to higher than recommended N rates.

#### REFERENCES

- Anderson, D.L., and Bowen, J.E. Sugarcane Nutrition. 1990. No. R-00693. Potash and Phosphate Institute. Atlanta, GA.
- Champagne, L.P., and M.E. Salassi. 1999. Projected Costs and Returns-Sugarcane in Louisiana, 1999, Department of Agricultural Economics and Agribusiness. Louisiana Agric. Exp. Stn., LSU Agricultural Center, A.E.A. Information Ser. No. 175.
- Faw, W.F., and E.R. Funderburg. 1995. Sugarcane Fertilization. Louisiana Coop. Ext. Service Pub. No. 2473.
- 4. Freund, R.J., and W. J. Wilson. 1993. Statistical Methods. Academic Press, San Diego, CA.
- Garrison, D.D., D.P. Guillot, W.R. Jackson, and H.L. Waguespack, Jr. 1999. A Report on the 1998 Outfield Variety Tests. American Sugar Cane League of the U.S.A. Sugar Bull.77, No. 9:19-23.
- Golden, L.E., and LB. Abdol. 1977. Effects of N and K fertilizers and soil type on yield components and nutrient uptake of four sugarcane varieties. Louisiana Agric. Exp. Stat. Bull. 700.
- Golden, L.E. 1972. Relationship between fertilizer and leaf blade P and S and sugarcane yield in Louisiana. Proc. of the Int. Soc. of Sugarcane Technol., 14\* Congr., 695-701.
- Gomez, K.A., and A.A. Gomez. 1984. Statistical Procedures for Agricultural Research. 2<sup>nd</sup> ed. John Wiley and Sons, New York.
- Gravois, K.A. 1999. The 1999 Louisiana sugarcane variety survey. P. 91-96. Sugarcane research annual progress report. Louisiana Agric. Exp. Stn.
- Gravois, K.A., and S.B. Milligan. 1992. Genetic relationships between fiber and sugarcane yield components. Crop Sci. 32:62-66.
- Richard, C.A. 1999. 1998 Yields-Variety Releases. American Sugar Cane League of the U.S.A. Sugar Bull. 77:11-18.
- Schroeder, B.L., J.H. Meyer, G. Kingston, and A.W. Wood. 1998. A comparison of fertiliser advisory practices in the Australian and South African Sugar Industries. Proc. South African Sugar Technol. Assoc. 72:69-76.
- Viator, D., E. Dumgan, J. Beaud, Jr., M. Cancienne, P.J. deGravelles, Jr., P. Ellender, J. Gay, R. Gonsoulin, B. Harang, R. Judice, Jr., R. Lemann, L. Levert III, and G. Wood. 1987. Expectations of Production Research. p. 6-7. *In* the Challenge of Sugarcane Research. Louisiana Agric. Exp. Stn., L.S.U. Agric. Ctr. 6-7.

Treatment	N	Р	ĸ	
		kg/ha		
1	112	29	112	
2	168	29	112	
3	224	29	112	
4	168	0	112	
5	168	29	0	
6	168	0	0	

Table 2. Critical and optimum nutrient levels of sugarcane leaf tissue for Louisiana<sup>†</sup>.

% Dry We	eight	(months)	
1.25	1.50-1.75	3-4	
0.14	0.18-0.22	3	
1.00	1.25-1.75	3	
	1.25 0.14 1.00	1.25     1.50-1.75       0.14     0.18-0.22       1.00     1.25-1.75	1.25         1.50-1.75         3-4           0.14         0.18-0.22         3           1.00         1.25-1.75         3

<sup>†</sup>Anderson and Bowen, 1990.

		Treatment	Sugar	Cane	Sucrose	Stalk	
Treatment		(N-P-K)	Yield	Yield	Content	Weight	Population
Number	dſ	(kg/ha)	(Mg/ha)	(Mg/ba)	(g/kg)	(kg)	(stalks/ha)
1		112-29-112	17.53	115.1	137.09	0.88	131742
2		168-29-112	17.14	113.1	135.95	0.82	137958
3		224-29-112	18.23	119.7	137.31	0.87	137839
4		168-0-112	18.30	117.7	140.03	0.87	135448
5		1 <b>68-29-0</b>	17.13	111.9	137.8	0.84	132519
6		168-0-0	16.88	109.3	138.52	0.83	131323
ANOVA				P-val	1165		
Year	1		0.001	0.001	0.741	0.177	0.001
Row (Year)	10		0.005	0.004	0.001	0.148	0.001
Col (Year)	10		0.397	0.316	0.077	0.031	0.431
Trt <sup>†</sup>	5		0.143	0.019	0.631	0.427	0.448
Year* Tri	5		0.824	0.974	0.256	0.285	0.247
C. V. (%)			10.9	10.0	3.6	7.7	6.2
Contrast Prob	<u>abilit</u> y	ι					
P Response	1		0.261	0.479	0.188	0.370	0.543
Trt 2,5 vs. Trt	4,6						
K Response	ĩ		0.106	0.015	0.855	0.752	0.153
Trt 2,4 vs. Trt	5,6						
N Response	1		0.479	0.342	0.650	0.105	0.183
Trt I vs Trt 2							
N Response	1		0.085	0.018	0.591	0,193	0.977
Trt 2 vs. Trt 3							
N Response	1		0.227	0.060	0.931	0.657	0.190
Trt 1 vs. Trt 3							

Table 3. Treatment means, analysis of variance, and orthogonal contrasts for the tests conducted at Vacherie, LA during 1998 and 1999.

<sup>†</sup>Tested for significance using the Year \* Trt interaction.

Table	4.	Contrast	means	of the	macronutrient	concentrations	of the	sugarcane	leaf blades	for
Vacher	ie a	across tw	o years,	except	where denoted	l.				

Macronutrient	Contrasts		P Value		
		(112 kg/ha)	(168 kg/ha)	(224 kg/ha)	· atus
			% dry weight		
Nitrogen	kg/ha				
	112 vs 168	1.03	1.04		0.837
	168 vs 224		1.04	1.07	0.560
	112 vs 224	1.03		1.07	0.439
		Phosphor (29 kg/ha)	us Level (0 kg/ha)		
		% dry	weight		
Phospborus †					
1998	29 vs 0	0.16	0.15		0.011
1999	29 vs 0	0.18	0.18		0.249
		Potassiu	m Level		
		(112 kg/ha)	(0 kg/ha)		
		% dry	weight		
Potassium					
	112 vs 0	1.53	1.44		0.048

<sup>†</sup>Two different means were reported for each year because of a significant year by treatment interaction. The upper mean denotes 1998, and the lower mean denotes 1999.

Treatment <u>Number</u> 1 2	df	Treatment (N-P-K) (kg/ha) 112-29-112 168-29-112	Sugar Yield (Mg/ha) 14.35 13.76	Cane Yield (Mg/ba) 107.6 101.1	Sucrose Content (g/kg) 119.84 121.74	Stalk Weight (Kg) 0.70 0.69	Population (stalks/ha) 156727 149076
3		224-29-112	14.03	105.1	119.58	0.70	151826
4		168-0-112	14.53	109.5	118.77	0.71	157504
5		168-29-0	13.78	103.0	119.63	0.70	150571
6		168-0-0	13.07	98.07	119,15	0.65	151168
ANOVA			P	-values			
Year	1		0.001	0.001	0.001	0.001	0.001
Row (Year)	10		0.625	0.122	0.681	0 181	0.861
Col (Year)	10		0.720	0.661	0.632	0.023	0.001
Trt'	5		0.079	0.078	0.821	0.709	0 374
Year* Tri	5		0.964	0.883	0.721	0.920	0.807
C. V. (%)			14.8	12.0	6.1	10.5	10.0
Contrast Prob	<u>ability</u>	:					
P Response Trt 2,5 vs. Trt	1 <b>4,6</b>		0.899	0.448	0.330	0.470	0.190
K Response Trt 2,4 vs. Trt	1 5,6		0.039	0.073	0.631	0.233	0.453
N Response Trt 1 vs Trt 2	1		0.169	0.081	0.440	0.517	0.129
N Response Trt 2 vs. Trt 3	1		0.499	0.241	0.385	0.404	0.543
N Response Trt I vs. Trt 3	1		0.421	0.434	0.914	0.838	0.297

Table 5. Treatment means, analysis of variance, and orthogonal contrasts for the tests conducted at Youngsville, LA during 1998 and 1999.

Tested for significance using the Year \* Trt interaction.

Table 6. Contrast means of the macronutrient concentrations of the sugarcane leaf blades for Youngsville across two years, except where noted.

Macronutrient	Contrasts		P Values		
		(112kg/ha)	(168 kg/ha)	(224 kg/ha)	
			% dry weight		,
Nitrogen					
	112 vs 168	1.28	1.24		0.635
	168 vs 224		1.24	1.36	0.211
	112 vs 224	1.28		1.36	0.395
		Phosphor (29 kg/ha)	us Level (0 kg/ha)		
		% dry	weight		
Phosphorus†					
1998	29 vs 0	0.16	0.14		0.334
1999	29 vs 0	0.19	0.19		0.427
		Potassiu	m Level		
		(112 kg/ha)	(Okg/ha)		
		% dry	weight		
Potassium					
	112 vs 0	1.41	1.38		0.011

<sup>†</sup>Two different means were reported for each year because of a significant year by treatment interaction. The upper mean denotes 1998, and the lower mean denotes 1999.

Macronutrient	Fertilizer Bate	Fertilizer Cost	Cane Yield	Per Hectare Return for Grower with Fertilizer Cost§	
	(kg/ha)	P**	(Mg/ ha)		
Nitrogent	112	\$49.28	114.9	\$2357	
	168	\$73.92	113.1	\$2294	
	224	\$98.56	119.6	\$2405	
Potassium	0	\$0	109.3	\$2289	
	112	\$40.19	117.6	\$2423	

Table 7. Projected	l cost analysis per	hectare of sugarcane	for Vacherie.
--------------------	---------------------	----------------------	---------------

† Urea (45-0-0) was the nitrogen fertilizer source that was utilized in this experiment (\$.198 is the price per kilogram of Urea calculated from \$198/tonne)

‡ Fertilizer Cost calculated by following formula: [kgs of fertilizer source \* price per kilogram for fertilizer source]

§ Per hectare return for grower calculated by the following formula: [Cane Yield \* \$20.94/metric tonne] (\$20.94/tonne is price per tonne based on \$.42 per kg, 40% mill share, 16 % landowner rent, and assumine a 10% recovery for sugar on a tonne of cane)

Muriate of Potash (0-0-60) was the potassium fertilizer source that was utilized in this experiment. (3.18 per kilogram if potash was \$176/tonne)

Macronutrient	Fertilizer Rate	Fertilizer Cost per Hectare‡	Cane Yield	Per Hectare Return for Grower with
	(kg/ha)		(Mg/ha)	Fertilizer Cost§
Nitrogent	(12	\$49.28	107.52	\$2201
	168	\$73.92	101.02	\$2041
Potassium	0	<b>S</b> 0	97.90	\$2050
	112	\$40.19	109.31	\$2249

Table 8. Projected cost analysis per hectare of sugarcane for Youngsville.

† Urea (45-0-0) was the nitrogen fortilizer source that was utilized in this experiment (\$.198 is the price per kilogram of Urea calculated from \$198/tonne)

<sup>‡</sup>Fertilizer Cost calculated by following formula: [kgs of fertilizer source \* price per kilogram for fertilizer source]

§Per acre return for grower calculated by the following formula: [Cane Yield \* \$20.94/metric tonnes] (\$20.94/tonne is price per tonne based on \$.42 per kg, 40% mill share, 16 % landowner rent, and assuming a 10% recovery for sugar on a tonne of cane)

[Murial: of Potash (0-0-60) was the potassium fertilizer source that was utilized in this experiment. (\$.18 per kilogram if potash was \$176/tonne)

## EFFECT OF SILICON ON EXPRESSION OF RESISTANCE TO SUGARCANE BORER (DIATRAEA SACCHARALIS)

D.L. Anderson<sup>1</sup>

University of Florida Everglades Research and Education Center P.O. Box 8003 Belle Glade, Florida 33430

and

**Omelio Sosa, Jr.**<sup>2</sup> Research Entomologist (retired), USDA-ARS, Canal Point, FL Present Address: 2365 Palm Rd. West Palm Beach, Florida 33406

### ABSTRACT

The sugarcane borer (*Diatraea saccharalis*) causes damage to sugarcane (*Saccarum* spp.) in Florida and the Western Hemisphere. Association of host plant pest resistance with silicon content in plants has been shown with some insects. Under many soil conditions, calcium silicate slag, a by-product of electric furnace production of elemental phosphorus from apatite ores, is applied in Florida for increasing sugarcane yields. The objective of this study was to determine the effect of calcium silicate application on the resistance of sugarcane to the borer in field studies. Five popular cultivars (CP70-1133, CP72-1210, CP72-2086, CP74-2005, and CP80-1827) were evaluated for yield response and borer resistance to the broadcast application (1/164 ha plots) of calcium silicate slag (0 and 6.7 Mg ha<sup>-1</sup>) using a randomized complete block design and 4 replicates. Across all cultivars, Si application increased cane and sugar plant crop yields by 16.7% and 19.5%, respectively. In cultivars CP72-1210 and CP80-1827, cane yields of the other cultivars. Although nonsignificant at p=0.10, data trends for all five cultivars indicated decreased borer intensity with application of calcium silicate slag.

#### INTRODUCTION

The sugarcane borer (*Diatraea saccharalis*) is a major pest of sugarcane (*Saccharum* spp.) in the Americas (Pemberton and Williams, 1969; Sosa, 1981). Sugarcane yields in southern Florida have been demonstrated to increase from application of calcium silicate in certain soil conditions (Anderson et al., 1987; Anderson et al., 1991). Recent studies have recognized the importance of silicon content and silicon structures within the plant resulting in host plant resistance. The physical presence of pubescent leaf hairs (silicon-based structures) on leaf surfaces or stems were related to

<sup>1</sup>Deceased. <sup>2</sup>Corresponding author. Contribution from the University of Florida, Institute of Food and Agricultural Sciences. Florida Agric. Exp. Stn., Journal Series No. R-06539. Received September 22, 2000. resistance to the white fly in cotton (Gossypium species and genotypes) (Meagher et al, 1997; Butter and Vir, 1989), the weevil (Otiorhynchus sulcatus F.) in strawberry (Fragaria chiloensis, L) (Doss and Shanks, 1988), the planthopper (Sogatella furcifera, Horvath) in rice (Salim and Saxena, 1992), and

Listronontus bonariensis(Coleoptera:Curculionidae) in grasses (Barker, 1989). Sosa (1988 and 1990) determined that the oviposition preferences of the sugarcane borer occurred on clones with the least pubescence. These pubescent structures (leaf hairs, trichomes, ridges, etc.) largely consist of silicon (Hodgen and Bell, 1986; Lanning and Eleuterius, 1992; Perry et al., 1987). The objective of this study was to determine the effect of calcium silicate application on the resistance of sugarcane to the borer in field studies.

#### METHODS AND MATERIALS

The soil of the field test location was classified as a Pahokee muck (Euic, hyperthermic Typic Medisaprist). Calcium silicate slag was broadcast at 0 and 6.7 Mg slag ha<sup>1</sup> before the planting of sugarcane. Calcium silicate slag is a by-product of electric furnace production of elemental phosphorus from apatite ores and contains up to 206g kg<sup>-1</sup> Si (Anderson et al, 1992). Five cultivars were evaluated during 1996-97: CP70-1133, CP72-1210, CP72-2086, CP74-2005, and CP80-1827. A randomized complete block design with four replications was used. Treatment plots measured 6.1 m x 10 m. The total number of millable stalks ha<sup>-1</sup> was estimated from stalk and whole-plot weights. Stalk weights were estimated from 20 stalk subsamples at harvest. Top-visible dewlap (TVD) sugarcane leaf blades with midribs were collected at growth stage five (Chiarappa, 1971) for Si analysis. Growth stage five occurs during June through mid-September in Florida and is the stage characterized by rapid growth, increased leaf production, and rapid stem elongation. Leaves were dried at 70°C and ground in a stainless steel Wiley mill to pass a 60-mesh sieve. Silicon concentration in TVD leaf tissue was determined using the procedures described by Elliott et al (1988).

Subsamples of 20 cane stalks per plot were randomly collected from each plot, weighed, and passed through a three-roller sample mill for juice extraction. The crusher juice was analyzed for Brix (soluble solids) using a refractometer (Bausch & Lomb, Inc., Rochester, NY). After clarifying the juice using lead subacetate (Meade and Chen, 1977), the Pol (sucrose concentration) was determined using a saccharimeter (Rudolph Research, Flanders, NJ). The percent sucrose in juice was estimated using formulas developed from sucrose tables and Brix temperature correction tables (Mead and Chen, 1977):

% Sucrose =  $(Pol x 26)/{105.811 + [(Brix - 15) x 0.44]}$ , where the 20°C temperature correction for Brix is given:

Corrected Brix (CBrix) = Brix + (temperature - 20) x 0.075.

Recoverable 96° sugar was calculated using the Winter-Carp-Geerligs formula modified by Arceneaux (1935), and the varietal correction factor (VCF) described by Rice and Hebert (1972):

96° Sugar = [(Sucrose x 21.058) - (CBrix x 6.15)] x VCF

The sugar yield (Mg sugar  $ha^{-1}$ ) was calculated from the measured cane yield (Mg  $ha^{-1}$ ) and the theoretical recoverable  $96^{\circ}$  sugar (kg sugar  $Mg^{-1}$  cane).

Sugarcane borer infestation (intensity, %) of the plant cane crop was evaluated by randomly taking whole-stalk samples from each plot after the cane was cut by hand, topped, and piled on the ground. A total of 20 stalks from each plot was taken at random from the pile. All stalks were split with a knife down the middle, and a count was made of the total number of internodes bored and non-bored by the sugarcane borer. Sugarcane borer infestations in the second year of the study (first ratoon crop) were too low to provide sufficient selection pressure to evaluate differences between cultivars. Comparison of main treatment effects (cultivar, slag) on the collected data was made at the p=0.1 level according to the Waller-Duncan k-ratio t test (SAS, 1996). To evaluate the influence of the % borer intensity and slag application on sugarcane cultivar yields (i.e., cane and sugar), stepwise regression procedures (SAS, 1996) were used. Statistically significant (p<0.1) linear and simple 1: linteraction term variables were evaluated for use in final regression models:

# $\Upsilon = \beta_0 + \beta_1 \chi_1 + \beta_2 \chi_2 + \beta_3 \chi_1 \chi_2$

where  $\Upsilon$  is the cane or sugar yield (Mg ha<sup>"1</sup>),  $\chi_1$  is the slag rate treatment (Mg ha<sup>"1</sup>),  $\chi_2$  is the borer intensity (%), and the  $\beta$  values are the intercept ( $\beta_1$ ) and corresponding regression coefficients ( $\beta_1$ ,  $\beta_2$ ,  $\beta_3$ ,  $\beta_3$ ).

### RESULTS AND DISCUSSION

The application of calcium silicate slag significantly (p<0,10) increased sugarcane leaf Si content of all cultivars (Table 1). In past studies, it was shown that higher applications of silicate (at rates approaching or exceeding 10 to 20 Mg ha<sup>-1</sup>) would increase leaf Si contents above 10 g kg<sup>-1</sup> (Anderson, 1991; Anderson et al., 1987; 1991). Commercial applications of slag are normally within 4.5 to 6.7 Mg ha<sup>-1</sup> While commercial rates have not likely maximized Si uptake or yields, they are within acceptable application rates for growers. Significant cane and sugar yield increases were observed as a result of calcium silicate slag application (Table 2). Across all cultivars, cane and sugar yields were increased by 16.7% and 19.5%, respectively. Differences in yields between cultivars were also observed with cane and sugar yields increasing with Si applications as much as 24.0% and 27.8%, respectively.

Overall, borer intensity was low in this study, however, cultivars CP72-2086 and CP80-1827 were more heavily infested with borers than others. Borer intensity declined with slag application, but the trends (for each cultivar and slag rate) were nonsignificant at the p=0.10 level (Table 2). Differences in borer resistance among cultivars were significant (Table 2). In cultivars CP72-1210 at CP80-1827, cane yields (Mg ha<sup>-1</sup>) declined due to increased borer intensity (Table 3). In cultivar CP72-1210 a significant (p < 0.10) interaction between slag applied and borer intensity was observed. The influence of borers on sugar yields are more difficult to assess. While slag application appears to be the sole factor increasing sugar yields of three cultivars, in CP70-1133 and CP80-1827 the data suggest a positive slag x borer intensity interaction on sugar yields that merits further study (Table 3). Plant stress, particularly at lower levels, could increase sugar accumulation in sugarcane. Borer intensity in this study was at low levels. It is assumed that at higher borer infestation levels both cane and sugar yields would decline.

Data trends show that resistance to borers on some cultivars may be enhanced from the use of calcium silicate slag. Field studies such as this are difficult to assess from an insect-host plant perspective. If the density of borers is not high, data cannot be collected and assessed with assurance; this occurred in year two of this study. Also the spatial distribution of insects may be such that the statistical field design is unable to determine significance despite observed treatment trends. From the literature presented and data trends (however weak), a positive relationship may exist between insect resistance and Si fertilization. Because high insect density and uniformity is not guaranteed in field studies, artificial infestation using laboratory-reared borers may be necessary to obtain host plant resistance data relative to Si fertilization. Increasing host plant resistance to the sugarcane borer through application of silicon or breeding for development of pubescence would be more environmentally friendly than the use of insecticides, which are often ineffective and/or damaging to non-target organisms. Data indicate that Si application has beneficial results to the sugarcane plant when cultivar resistance is also considered. We suggest that additional field tests be conducted and natural field populations of sugarcane borers be augmented with laboratory-reared insects.

#### ACKNOWLEDGEMENT

Appreciation is expressed to Mr. Modesto F. Ulloa and New Hope Farms for assistance in field studies.

#### REFERENCES

- Anderson, D. L. 1991. Soil and leaf nutrient interactions with sugarcane grown in response to calcium silicate slag. Pert. Res. 30:9-18.
- Anderson, D.L., D.B. Jones, and G.H. Snyder. 1987. Response of a rice-sugarcane rotation to calcium silicate slag on Everglades Histosols. Agron. J. 79:531-535.
- Anderson, D.L., G.H. Snyder, and F.G. Martin. 1991. Multi-year response of sugarcane to calcium silicate slag on Everglades Histosols. Agron. J. 83:870-874.
- Anderson, D.L., G.H. Snyder, and J.D. Warren. 1992. Availability of phosphorus in calcium silicate slag. Commun. Soil Sci. Plant Anal, 23:907-918.
- Arceneaux, G. 1935. A simplified method of making theoretical sugar yield calculations in accordance with Winter-Carp-Geerligs formula. Int. Sugar J. 37:264-265.
- Barker, G.M. 1989. Grass host preferences of *Listronontus bonariensis (Coleoptera: Curculionidae)*. J. Econ. Entom. 82:1807-1816.
- Butter, N.S., and B.K. Vir. 1989. Morphological basis of resistance in cotton to the whitefly (*Bemisia tabaci*). Phytoparasitica 17:251-261.

- Chiarappa, L. 1971. Crop loss assessment methods, p. 4.4.6/1 IN: FAO Manual on the Evaluation and Prevention of Losses by Pests, Diseases, and Weeds. Comm. Agr, Cur., Farmham, England. Supplement 1.
- Doss, R.P., and C.H. Shanks, Jr. 1988. The influence of leaf pubescence on the resistance of selected clones of beach strawberry (*Fragaria chiloensis*, L) to adult black vine weevils (*Otiorhynchus sulcatus* F.). Scientia Hort. 34(l/2):47-54.
- Elliott, C. L., G. H. Snyder, and D. B. Jones, 1988. Rapid gravimetric determination of Si in rice straw. Commun. Soil Sci. Plant Anal. 19(13): 1543-1550.
- Hodgen, M.J., and A. Bell. 1986. The mineral relations of the lemma of *Phalaris canadensis* L., with particular reference to its silicified macrohairs. Israel J. Bot. 35:241-253.
- Lanning, F.C., and L.N. Eleuterius, 1992. Silica and ash in seeds of cultivated grains and native plants. Ann. Bot. 69(2): 151-160.
- Meade, G.P., and J.C.P. Chen. (1977). *Cane Sugar Handbook*. John Wiley & Sons, New York.
- Meagher, R.L., Jr., Smith, C.W., Smith W.J. 1997. Preference of Gossypium genotypes to Bemisia agentifolii (Homopter:Aleyrodidae). J. Econ. Entom. 90:1046-1052.
- Pemberton, C.E., and J.R. Williams. 1969. Distribution, origins, and spread of sugar cane insect pests, *In J.R. Williams, J.R. Metcalfe, R. W. Mungomery, and J.R. Mathes* [eds.], *Pests* of Sugarcane, pp. 1-9. Elsevier, Amsterdam.
- Perry, C.C., R.J.P. Williams, and S.C. Fry. 1987. Cell wall biosynthesis during silicification of grass hairs, J. Plant Phys. 126:437-448.
- Rice, E. R., and L. P. Hebert. 1972. Sugarcane variety tests in Florida during the 1971 -72 season. USDA Agr. Res. Ser. S-2. U. S. Government Printing Office, Washington, D. C.
- Salim, M., and R.C. Saxena. 1992. Iron, silica, and aluminum stresses and varietal resistance in rice: effects on whitebacked planthopper. Crop Sci. 32:212-219.
- SAS. 1996. SAS/STAT Software: Changes and Enhancements through Release 6.11, Chapter 18", SAS Institute Inc., Gary, NC.
- Sosa, O., Jr. 1990. Oviposition preference by the sugarcane borer (*Lepidoptera.Pyralidae*). J. Econ. Entom. 83:866-868.
- Sosa, O., Jr. 1988. Pubescence in sugarcane as a plant resistance character affecting oviposition and mobility by the sugarcane borer (*Lepidoptera:Pyralidae*), J. Econ. Entom. 81:663-667.

 Sosa, O., Jr. 1981. Sugarcane borer, *Diatraea saccharalis* in Florida: a review. Proceedings, Second Inter-American Sugar Cane Seminar, pp. 145-151. Inter-American Transport Equipment Co., Miami, FL.

Table 1. Sugarcane plant crop leaf (TVD) silica contents for five cultivars with and without slag treatment.

Cultivar	Slag rate (Mg ha-1)	Leaf Si (g kg-1)	
CP 70-1133	0	2.92b	
CP 70-1133	6.7	4.97a	
CP 72-1210	0	2.86b	
CP 72-1210	6.7	5.69a	
CP 72-2086	0	3.63b	
CP 72-2086	6,7	5.61a	
CP 74-2005	0	2.53b	
CP 74-2005	6.7	6.13a	
CP 80-1827	0	3.35b	
CP 80-1827	67	<u>5.75a</u>	

Cultivar	Slag rate	Cane yield (Mg-ha <sup>-1</sup> )	Sugar yield	Borer intensity
CD 70 1100	0	(	10.11	(,-)
CP 70-1133	0	199.9b	19,16	1.82a
	6.7	224.9a	21.9a	1.40a
CP 72-1210	0	125.9b	12.4b	5.62a
	6.7	156.1a	15.8a	1.56a
CP 72-2086	0	154.0b	15.8b	6.03a
	6.7	186.5a	20.2a	5.41a
CP 74-2005	0	149.4b	13.5b	3.77a
	6.7	174.7a	16.4a	2.20a
CP 80-1827	0	195.4b	18.7b	5.92a
	6.7	219.7a	20.9a	5.45a
CP 70-1133	-	212.4a	20.5a	1.61b
CP 72-1210	-	141.0d	14.1c	3.59ab
CP 72-2086	-	170.3b	18.0b	5.72a
CP 74-2005	-	162.0c	14.9c	2.98b
CP 80-1827	-	207.5a	19.8a	5.69a
	0	164.9b	15.9b	4.63a
	6.7	192.4a	19.0a	3.21a

Table 2. Treatment means and main effects of cultivar and slag application rate\*.

\*Means (column) followed by the same letter are not significantly different at p=0.10 level according to the Waller-Duncan k-ratio t test.

Journal American Society of Sugarcane Technologists, Vol. 21, 2001

	r	= β <sub>α</sub>	+ <u>β</u> ,χ,	ł	$\beta_2 \chi_2 +$	β <sub>3</sub> χ <sub>1</sub> χ <sub>2</sub>		
						Slag x		
					Borer	borer		
Cultivar	Yield	Intercept	Slag		intensity	intensity	F test	$\mathbb{R}^2$
CP 70-1133	Cane	199.9	16.68		-	-	18.47	0.75**
CP 72-1210	Cane	132.2	22.99		-1.11	-3.77	42.74	0.97**
CP 72-2086	Cane	154.0	21.69		-	-	21.19	$0.78^{**}$
CP 74-2005	Cane	149.4	16.87		-	-	15.24	0.72**
CP 80-1827	Cane	205.5	15.66		-1.69	-	14.73	0.85**
CP 70-1133	Sugar	19.4	-		-	1.06	19.68	0.77**
CP 72-1210	Sugar	12.5	2.24		-	-	12.88	0.68 +
CP 72-2086	Sugar	15.8	2.97		-	-	13.62	0.69**
CP 74-2005	Sugar	13.5	1.90		-	-	16.97	0.74**
CP80-1827	Sugar	18.9				0.23	4.39	0.42 +

Table 3. The influence of slag rate (Mg ha<sup>-1</sup>) and borer intensity (%) on cane and sugar yield (Mg ha<sup>-1</sup>) as indicated by "best-fit" multiple regression models.

\*\* and + represent statistical significance at p<0.01 and p<0.10, respectively. All regression coefficients ( $\beta_{0},\beta_{r},\beta_{p},\beta_{3})$  are significant at p<0.10.

# THE EFFECT OF HARVEST METHOD AND PLOT SIZE ON THE ESTIMATION OF SUGARCANE YIELD

## K. P. Bischoff, K. A. Gravois, H. P. Schexnayder, Jr., and G. L. Hawkins Louisiana State University Agricultural Center, Sugar Research Station,

P.O. Box 604, St. Gabriel, LA 70776.

# ABSTRACT

Harvest method and plot size are important research factors determining the accuracy of estimating sugarcane (Saccharum spp.) yield and yield components. Two studies were conducted in 1999 at the St. Gabriel Research Station, St. Gabriel, LA to compare cane yield estimates obtained via whole stalk, combine, and estimated yield harvest methods. The effect of plot size on cane yield estimates was also studied. One study was conducted using the cultivar HoCP85-845, and the other study used the cultivar LCP86-454. Whole stalk harvesting produced lower cane yield estimates than either combine harvesting or estimated cane yield methods. The sucrose concentration of hand cut samples for combine harvesting and estimated cane yield were numercially lower for HoCP85-845 and significantly lower for LCP86-454. Cane vield was underestimated and sucrose concentration overestimated for whole stalk harvesting, mainly due to the shorter length of improperly topped stalks from the whole stalk harvester. Cane yield for the combine harvesting system and the estimated method for determining cane yield were similar. Cane yield estimates from the standard infield plot size (three-row, 4.9 m) and a new plot size of two-row, 7.3 m were not significantly different, indicating that the change in plot dimensions to facilitate combine harvesting was not detrimental. Based on improved standard errors, the plot lengths of the off-station nurseries of the Louisiana breeding program were increased from a single row of 4.9 m to a single row of 6.1 m. Cane yield estimates on small single-row plots were most accurate when estimated with millable stalk counts and stalk weights. Combine harvesting of larger multi-row plots was more accurate for estimating cane vield than estimating cane vield based on stalk counts and stalk weights.

#### INTRODUCTION

For years, the accepted means of collecting plot weight data in Louisiana was utilizing the whole stalk or "soldier" type harvester and a tractor mounted weighing device (Fanguy, 1970). The accuracy of this process can be compromised due mainly to the calibration of the scale and the limited topping ability of whole stalk harvesters in sugarcane plot areas. This became even more evident with the advent of the two-row whole stalk harvesting system. Since proper topping height differs among varieties, the use of the two-row whole stalk harvester for two different varieties could impose a degree of error due to topping differences. This resulting error may affect both cane yield and sucrose concentration. Sucrose concentration will be over estimated if the stalks are topped at lower than optimum standards and under estimated if the stalks are topped higher than optimum standards. Optimum topping height of sugarcane stalks should occur at the first hard internode below the growing point.

Several studies have been conducted examining the accuracy of differing methods to determine cane yield in sugarcane research plots. Early research was conducted by hand-cutting sugarcane plots. In the Louisiana outfield testing stage of the variety development program, all plots were hand-harvested from 1926 to 1958 (Fanguy, 1967). Since 1958, some outfield experiments were harvested with whole-stalk harvesters. Fanguy (1967) reported no harvest method (hand-cut vs. whole-stalk machine cut) x variety interaction for two experiments, and a third location only reported a significant (P=0.05) interaction for cane yield.

Hebert (1963) estimated cane yield by hand-cutting plots and weighing and by estimating cane yield from population and stalk weight. In eight tests that were conducted, the two cane yield estimating methods were moderately to highly correlated (r = 0.66 to r = 0.90) with weighed plots. The best varieties were identified in each test whether the cane yield was determined by weighing or by estimation.

In the Louisiana sugarcane breeding program, cane yield is estimated based on population and stalk weight for the early stages of the variety development program. Fanguy (1971) reported that estimated cane yields and cane yields obtained from whole-stalk machine cut plots were moderately to highly correlated (r = 0.69 to r = 0.92) in four tests. The ranking of varieties for each method was similar indicating no significant interaction between harvest method and variety. In each test, the cane yield coefficient of variation (CV) was lower for the machine cut plots compared to estimated yields. In whole stalk machine harvested tests, we surmise that the CV's associated with these tests could be artificially lowered if care is not taken to individually top each variety correctly. A uniform topping height among varieties would decrease treatment differences and correspondingly the CV statistic.

As the Louisiana sugar industry moved toward a combine harvesting system, work was begun to adapt research methods to a combine harvesting system. Along with a combine harvester, a small hydraulic high-dump wagon equipped with load cells was acquired. Load cells were placed in the axle near each wheel and in the wagon hitch. Once this equipment was in place in 1999, sugarcane researchers began obtaining plot weight data using this system.

A study was initiated in 1999 to compare three sugarcane cane yield estimating methods: whole stalk and combine harvesting systems, and cane yield estimated via millable stalk counts and stalk weights. This study also investigated the effect of plot size on cane yield estimates.

#### MATERIALS AND METHODS

Two experiments were conducted during 1999 at the St. Gabriel Research Station, St. Gabriel, LA, to determine the effect of harvest method on sugarcane yields. The two varieties used for these experiments, LCP86-454 and HoCP85-845, were selected because of their erectness and better than average harvesting characteristics. This would provide a best case scenario for each harvesting system. The experimental design was a randomized complete block for each experiment and was replicated three times. Treatments were established based on the various sizes of plots used throughout the stages of the Louisiana Variety Improvement Program. Single-row, 4.9 m plots are used in the second line trial through nursery stages of the sugarcane breeding program. Three-row, 4.9 m plots are used in the outfield test stage. A single-row, 6.1 m plot was included in the study to determine if a larger plot size would improve estimated cane yield determinations in the early yield testing stages of the

sugarcane breeding program. A two-row, 7.3 m plot size was included to determine if changing plot dimensions in the infield stage would affect cane yield measurements.

To obtain estimated cane yield (Mg/ha), millable stalk counts were made in all plots during the first week of December, 1999. Hand-cut, hand-stripped ten stalk samples were taken from each plot. These samples were then weighed to obtain mean stalk weight (kg) and measured to obtain mean stalk length (cm). Estimated cane yield was obtained as the product of stalk population and stalk weight. The samples were then milled in a three-roller sample mill, and a juice sample was obtained for quality analysis. The crusher juice was analyzed for Brix (percent soluble solids w/w) by refractometer. Pol of the clarified juice was obtained with an automated saccharimeter. Sucrose concentration (g sucrose/kg of cane yield) was estimated as described by Gravois and Milligan (1992). Sugar yield (Mg/ha) was obtained as the product of cane yield and sucrose concentration and dividing by 1000.

The 4.9 m and 9.8 m plots were then harvested with a single-row whole stalk harvester and weighed using a tractor mounted weigh rig with a load cell (Fanguy, 1970). Ten-stalk samples were taken from the heap row of each whole stalk harvested plot. These samples were weighed and measured to obtain mean stalk weight and length, respectively. The samples were also milled for juice analyses as noted above.

All remaining plots were harvested and weighed using the combine harvesting system. The cane from the combine harvester was loaded into a weigh wagon having load cells in each axle and in the wagon hitch. An electronic data device recorded the weights from each plot. Cane yield estimates were not corrected for trash. No billet samples were taken for sucrose analyses on the combine harvested plots, The hand-cut whole stalk samples were used for sucrose analyses.

Data for each test were analyzed with the SAS mixed model procedure (SAS Institute, 1988). Specific contrasts were used to test various treatment mean effects, and probability differences were reported. Standard errors of the treatment means were also reported using the SAS means procedure.

### RESULTS AND DISCUSSION

For each variety, HoCP85-845 and LCP86-454, no significant differences were obtained for sugar yield among the harvesting systems (Table 1 and Table 2, respectively), although sugar yield was numerically lower for the whole stalk harvesting system. For HoCP85-845, P values indicated that cane yield was not significantly different for the three harvesting systems, although cane yield was numerically lower for the whole stalk harvesting system. For LCP86-454, whole stalk harvesting produced significantly lower cane yield than both combine harvesting and estimated cane yield. The sucrose concentration of hand cut samples that were taken for combine harvesting and estimated cane yield was numerically lower for HoCP85-845 and significantly lower for LCP86-454. For comparing harvesting methods, cane yield was underestimated and sucrose concentration overestimated for whole stalk harvesting. Although sugar yield is calculated as the product of cane yield and sucrose concentration, sugar yield was numerically lower for whole stalk harvesting. Gravois (1988) and Milligan et al. (1990) showed cane yield to be approximately three times more important in determining sugar yield than sucrose concentration. A major contributing factor to the differences between the harvest systems is illustrated in Table 3. The average height of hand cut samples was significantly greater than that of whole stalk harvested samples of both varieties. The maximum topping height of whole stalk harvesters is approximately 244 to 260 cm. The lower stalk height of whole stalk harvested samples was the likely cause of the differences observed in cane yield.

Sugar yield and cane yield for the combine harvesting system and the estimated methods of establishing yield were virtually equal. Sucrose concentration of combine harvesting vs estimated yields would be the same because both are derived from hand-cut samples.

Other harvest method and plot size treatments were examined (Table 4). One comparison contrasted cane yields derived from whole stalk harvesting of the standard infield plot size (three-row, 4.9 m) to the standard outfield plot size (three-row, 9.8 m). Cane yield was not significantly different between the two stages, indicating plot size was not a factor in determining cane yield for these stages of the sugarcane breeding program when harvesting with whole stalk machines. The results were similar for both varieties.

To facilitate combine harvesting, the standard infield plot size (three-row, 4.9 m) was changed to a two-row, 7.3 m plot size (Table 4). The longer row length would require less starting and stopping while harvesting with the combine. Cane yield estimates for each plot size were not significantly different, indicating that the change in plot dimensions to facilitate combine harvesting was not detrimental.

The new infield plot size (two-row, 7.3 m) was also compared to the standard outfield plot size (three-row, 9.8 m) to indicate repeatability between breeding program stages for combine harvesting systems. Again, cane yield from combine harvesting of the new infield plot size was not significantly different than cane yield derived from the standard outfield plot size. The results were similar for both varieties. Based on this information, the breeding program now uses an infield plot design of two-rows, 7.3 m long. This change was begun during the 1999 planting season.

Standard errors of cane yield means derived from estimated yield and combine harvesting were used to determine the accuracy of cane yield estimates based on plot sizes (Table 5). For estimated cane yield, standard errors decreased as plot size increased until the three-row, 9.8 m plot size. After attaining the lowest standard errors for estimated cane yield in the two-row, 7.3 m plots, the standard error increased for the three-row, 9.8 m plots. We surnise that the single row 4.9 and 6.1 m plots were too small to overcome the inconsistencies of stands in the field. On the other hand, the three-row, 9.8 m plots may have been too large leading to variability in millable stalk counts. The nursery stage of the Louisiana sugarcane breeding program uses single row 4.9 m plot sizes, There is an obvious advantage to extending that plot length to 6.1 m, and the effort to increase plot size would be minimal and seed requirements only slightly more than currently needed. Based on the results of this study, the plot sizes of the off-station nurseries of the Louisiana sugarcane breeding program were increased from 4.9 m to 6.1 m. These changes were put into effect during planting in 1999.

Optimum plot size was also compared for a combine harvesting system based on cane yield standard errors (Table 5). Standard errors were largest for the single row plot sizes when compared to the multiple-row, larger plot sizes. The new infield and standard outfield plot sizes had similar standard errors. Combine harvesting small single-row plots was not an accurate means of obtaining cane yield. Cane yield estimates on small single-row plots were more accurately obtained when estimated with millable stalk counts and stalk weights. Combine harvesting larger multi-row plots was more accurate for determining cane yield than was estimating cane yield.

#### REFERENCES

- Fanguy, H.P. 1967. A comparison between hand and machine cutting of sugarcane variety outfield experiments in Louisiana. Proc. ISSCT. 13:615-617.
- Fanguy, H.P. 1970. A tractor mounted hydraulic scale for weighing sugarcane plots in Louisiana. Sugar y Azucar. 65(3):38-40.
- Fanguy, H.P. 1971. Comparison of estimated and weighed yields in sugarcane variety trials; preliminary results. Proc. ASSCT 112-114.
- Gravois, K.A. 1988. Path-coefficient analysis: A study of phenotypic, broad-sense, and narrow-sense variation in sugarcane. Ph.D. diss. Louisiana State Univ., Baton Rouge, LA (Diss. Abstr. 89-17818).
- Gravois, K.A. and S.B. Milligan. 1992. Genetic relationships between fiber and sugarcane yield components. Crop Sci. 32:62-67.
- Hebert, L.P. 1963. Estimating cane yields in evaluating sugarcane seedling selections. Crop Sci. 3:277-278.
- Milligan, S.B., K.A Gravois, K.P. Bischoff, and F.A. Martin. 1990. Crop effects on genetic relationships among sugarcane traits. Crop Sci. 30:927-931.
- 8. SAS Institute. 1988. SAS User's Guide. Statistics (6<sup>th</sup> ed.). SAS Institute, Gary, NC.

Contrast	Estimated	Combine	Whole Stalk	P Value
		Sugar yie	ld (Mg/ha)	
Whole Stalk vs Combine		13.6	12.8	0.14
Estimated vs Combine	13.6	13.6		0.89
Estimated vs Whole Stalk	13.6		12.8	0.25
		Cane yiel	d (Mg/ha)	
Whole Stalk vs Combine		115.1	107.1	0.08
Estimated vs Combine	115.6	115.1		0.88
Estimated vs Whole Stalk	115.6		107.1	0.10
		Sucrose conce	ntration (g/kg)	
Estimated vs Whole Stalk	118.5		120.085	0.29

Table 1. The effect of harvest method on sugarcane yield components averaged across all plot sizes for HoCP85-845 for the test conducted in 1999 at the St. Gabriel Research Station.

Contrast	Estimated	Combine	Whole Stalk	P Value
		Sugar yie	eld (Mg/ha)	
Whole Stalk vs Combine		15.3	14.0	0.08
Estimated vs Combine	15.7	15.3		0.28
Estimated vs Whole Stalk	15.7		14.0	0.09
		Cane yie	ld (Mg/ha)	
Whole Stalk vs Combine		125.4	110.2	0.01
Estimated vs Combine	128.4	125.4		0.24
Estimated vs Whole Stalk	128.4		110.2	0.01
	Sucrose conc		entration (g/kg)	
Estimated vs Whole Stalk	122		127	0.001

Table 2. The effect of harvest method on sugarcane yield components for LCP86-454 averaged across all plot sizes for the test conducted in 1999 at the St. Gabriel Research Station.

Table 3, Height of whole stalk harvested vs hand cut samples of HoCP85-845 and LCP 86-454 for the test conducted in 1999 at the St. Gabriel Research Station.

Harvest Method	HoCP85-845	LCP86-454
	He	right (cm)
Whole stalk harvester	242	247
Hand cut	287	293
P Value	0	001 0.001

Contrast	Combine Two-row	Combine Three-row	Whole Stalk Three-row	Whole Stalk Three-row	P Value
	7.3 m	9.8 n	1 4.9 m	9.8 m	· aae
		Ca	ane yield (Mg/h	a)	
HoCP85-845					
Whole Stalk: Three-row 4.9 m vs					
Whole Stalk: Three-row 9.8 m			110.2	104.2	0.39
Combine: Two-row 7.3m vs Whole Stalk: Three-row 4.9 m	114.2		110.2		0.55
Combine: Two-row 7.3 m vs Combine: Three-row 9.8 m	114.2	114.0			0.98
LCP86-454					
Whole Stalk: Three-row 4.9 m					
Whole Stalk. Three-row 9.8 m			108.4	111.8	0.63
Combine: Two-row 7.3 m vs Whole Stalk: Three-row 4.9 m	121.6		108.4		0.30
Combine: Two-row 7.3 m vs Combine: Three-row 9.8 m	121.6	128,6			0.10

Table 4. The effect of harvest method and plot size on sugarcane yield components for HoCP85-845 and LCP 86-454 averaged across all plot sizes.

 Table 5. Standard errors for estimated and combine harvested cane yield for HoCP85-845 and LCP 86-454 as affected by plot size for the tests conducted at the St, Gabriel Research Station in 1999.

Variety	Plot Size	Estimated Cane Yield	Standard Error	Combine Cane Yield	Standard Error	
	meters	Mg/ha	Mg/ha	Mg/ha	Mg/ha	
HoCP85-845	1.8 x 4.9	116.3	4.86	114.5	6.38	
HoCP85-845	1.8 x 6.1	118.3	3.92	117.2	8.96	
HoCP85-845	3.7 x two-7.3 rows	113.9	2.76	114.2	2.40	
HoCP85-845	5.5 x three- 9.8 rows	113.9	5.69	114.5	2.53	
LCP86-454	1.8 x 4.9	121.3	6.54	135.5	3.65	
LCP86-454	1.8 x 6.1	133.6	2.91	115.8	3.16	
LCP86-454	3.7 x two-7.3 rows	127.8	2,64	121.6	1.30	
LCP86-454	5.5 x three- 9.8 rows	130.5	11.67	128.6	1.81	

PEER REFEREED JOURNAL

ARTICLES

MANUFACTURING

SECTION

## CANE JUICE ANALYSIS BY NEAR INFRARED (NIR) TO DETERMINE GROWER PAYMENT

Tere Pi Johnson Sugar Cane Growers Cooperative of Florida Belle Glade, Florida

### ABSTRACT

As applications of NIR analysis are increasing in the grain and food industries, more options are becoming available for analysis of incoming sugar cane in the factories. For the 1999-2000 crop season, Sugar Cane Growers Cooperative of Florida (SCGC), together with Florida Crystals Corporation, has adopted Near Infrared (NIR) spectroscopy as the standard method for cane juice analysis. This paper will include a brief history of cane juice analysis at SCGC as it has evolved from polarization using lead clarification to polarization using NIR spectroscopy. Experiences with data acquisition, equation development, and equation validation necessary for implementing NIR analysis will also be discussed.

## THE GOALS OF CANE JUICE SAMPLING

The process of analyzing incoming sugar cane for grower payment can be a challenge to many factories. A uniform method is difficult to standardize throughout the industry since many variables exist between one factory and another. Variables such as the size and the number of growers should be considered when developing the process for analyzing either the sample of cane or the sample of juice.

Sugar Cane Growers Cooperative of Florida grinds close to 22,000 tons of cane per day on average and brings over 1,000 loads of cane daily from over fifty grower members. With the goal being to collect a sample from every 65 tons of cane, this presents a sampling challenge. Assuming that each trailer carries approximately 20 tons of cane, a sample will be collected from every third trailer for each grower for each field harvested on any given day. This sampling has been fine-tuned over the last two years to optimize efficiency. Where in earlier years, 600 samples were collected in 16 hours for 19,000 tons of cane, now 350 samples can be collected in 10 hours for 22,000 tons of cane. This may not seem to be a significant improvement, but before, although there were obtained for a particular grower. Now, on the other hand, the 350 samples are sufficient to complete the grower sampling. Trucks are dumped at a rate of up to 28 trucks per hour on each milling tandem. The sampling method must be able to analyze at a rate of one sample every two minutes. Where older methods could not analyze the cane juice samples at this rate, the NIR instrument can do it easily.

#### EVOLUTION OF METHODS OF CANE JUICE ANALYSIS

The procedures for sampling and analysis of incoming cane have changed over the years. The earlier method of juice analysis used the brix hydrometer for determining the juice density. The percent sucrose was determined by polariscope reading converted to sucrose using the Schmitz Table
with lead subacetate (and later Home's dry subacetate of lead) for juice clarification. The use of lead salts for clarification lasted until the late 1980's, when government regulations for disposal of heavy metals became more stringent. Formidable research (Chou, 1987; Clarke and LeGendre, 1989; Clarke, 1982; Rens, 1978) had been performed during the 1970's and 1980's for a replacement clarification method using aluminum salts. In 1988, SCGC replaced the dry lead salt with the aluminum salt, aluminum chloride, in combination with calcium hydroxide, and used this mixture to clarify cane juice samples in the laboratory. The computer programs converted the pol results from the aluminum chloride/calcium hydroxide clarified juice to the standard lead method equivalent pol, using a regression analysis equation. About the same time, paired comparison tests were performed at the U.S.D.A. - A.R.S. in Houma and at SPRI in New Orleans (Clarke, 1982) between lead subacetate and the aluminum chloride/calcium hydroxide combination. The results of the aluminum chloride/calcium hydroxide pols proved to have superior correlation with the lead method equivalent pols.

For two years, incoming cane juice was analyzed using the aluminum chloride/calcium hydroxide clarification for pol determination, and hydrometer spindle for brix determination and, although this method was reliable, it was time consuming. During this time, the total tons of sugarcane harvested at SCGC grew by almost 20%, resulting in an increase in juice samples/hour for analysis in the laboratory. At these higher sampling rates, it was nearly impossible for the chemists to keep up with the sampling requirements. A method of analysis had to found that could analyze samples at a higher rate of speed.

In 1990, the methodology changed again as the solution for faster analysis was mechanical clarification rather than chemical clarification. This mechanical clarification was found in the form of the Membrex ultrafiltration membrane (DeStefano, 1990). To help increase the speed of analysis, brix determination by spindle was changed to refractometric analysis. A flow through system was devised whereby the cane juice sample that was pumped into the laboratory flowed directly from the membrane filter to a refractometer and to a polarimeter. The entire analysis took less than three minutes. The cane juice emerged from the 0.01 micron pore size membrane a brilliant golden color, and could easily be read by the refractometer and the polarimeter. These advances in analytical techniques were advances only in the laboratory. The data that emerged from the laboratory for grower payment was correlated back to the nearly obsolete lead pol and spindle brix that the growers were accustomed to seeing. Once again, regression analysis equations were updated in the computer programs to correlate the Membrex clarified juice pol and the refractometric brix. Although the Membrex system brought with it a myriad of problems, it became the method of choice for the Florida mills for almost a decade.

## INITIAL PROPOSAL

Throughout the 1990's, SCGC investigated and tested alternative methods of cane juice analysis. During the 1998-1999 season, a collaborative effort was made between the three mills of Florida Crystals Corporation, and Sugar Cane Growers Cooperative, to develop a uniform equation where the same sample of cane juice would produce the same analytical results in the four laboratories. The NIR instrument that was selected for measuring cane juice in all four laboratories is the Routine Sugar Analyzer with beverage module and 5000 monochromator with wavelength range of 1100-2500 nanometers. In the cane juice spectrum, (see Figure 1), the wavelength range used for analysis is 1250-1820 nm, and 2120-2330 nm.

# MODEL DEVELOPMENT AND POPULATION STRUCTURING

Beginning in 1998, almost 900 samples were collected, scanned and analyzed by the four laboratories throughout the season. A flowchart for calibration and implementation of NIR analysis is shown in Figure 2. Once the cane juice samples have been collected, near infrared spectral data must be obtained, along with accurate reference method analysis for each sample. The reference method used for cane juice NIR correlation is pol clarified with ABC Sugar Clarifier ®, and spindle brix. The spectral data and the reference data are used as the basis for population structuring. The key to population structuring is the collection of representative samples. Samples that exemplify the varying characteristics anticipated during the span of the season such as maturity, soil type, quality of cane, and quantity of mud, and if available, pre-harvest (immature) cane juice samples and freeze damaged cane juice will help make the equation more robust. The purpose of population structuring is to identify the "best" samples by removing redundant samples and spectral outliers. Using Chemometrics, the software combines the spectral data and the reference data, performs a regression analysis and generates a model to predict the constituents of interest, in this case, pol and brix. The software uses second derivative math for the enhancement of the near infrared spectra, and modified partial least squares (MPLS) regression for the development of the calibration equation. The results of the preliminary equations for brix and pol are shown in Figure 3. After the equation has been developed, its performance is measured by using it in parallel with the wet chemical analysis to determine if the equation is satisfactory. If it is acceptable, it is implemented into the routine operations.

#### MODEL VALIDATION AND UPDATING

The NIR analysis performed on each sample is a prediction of the juice sample based on calibration data stored in the software, and as with most natural products, sugarcane juice will change throughout the season, due to soil composition, rainfall, and other growing conditions, Because of this, it is necessary to periodically validate the equation against the reference method. Validation will measure the prediction accuracy of the NIR calibration. It is done by acquiring spectra and performing wet chemical analysis on a separate set of samples, similar to those used for the mathematical model, but that have not been used to develop the model. Validations at SCGC are performed weekly. Figure 4 shows examples of early and recent validation results. If the validation results are acceptable, they can be included in the model. Updating the model with accurate validation data will make the equation more robust, as shown in Figure 5 where the improvement in the current equation over the preliminary equation is evident. As routine analyses are performed, the instrument may detect a sample that it does not recognize as being similar to other samples. The software will flag this sample as an outlier. Outliers can be caused by conditions such as ambient vibrations, a change in ambient temperature, a contaminated sample, a sample with an unusual temperature, or a sample from an unusually muddy field. Samples that are identified by the equation as neighborhood outliers should be analyzed by the reference method, and if they are genuine, uncorrupted samples, should be added to the calibration data to update the equation, Updating the equation with special event data will make the equation more robust. During the 1999-2000 season, the initial plan for updating the model included collecting 50 samples each week to add to the calibration equation in order to capture the subtle changes that occur in the cane juice

throughout the crop. However, there were occasions where a more aggressive sampling schedule was necessary. During these situations, 250 samples were added to the equation for the week. This occurred at the beginning and near the end of the season, when the cane juice composition was different than it had been when the equation was first developed. Immature cane, early in the season, and exceptionally muddy cane, late in the season were two of the reasons for the change in juice composition. Signs that may indicate that updating is needed include unusual NIR results, poor correlation between instruments, and an excessive number of neighborhood outliers. With the NIR, in contrast with the polarimeter and the refractometer, the chemist's ability to tune in to the instrument's sensitivity is essential.

#### INSTRUMENT DIAGNOSTICS

The NIR software is able to perform self-diagnostic tests as per the user's request. Diagnostics is composed of three separate tests; instrument response, wavelength accuracy, and repeatability. Instrument response tests the performance of the detectors. Wavelength accuracy checks the wavelength alignment of the instrument. Repeatability checks the optical data at each wavelength. It is important to not only perform the diagnostics, but to look for any trends that may occur in the diagnostic data. This can serve as a quality control tool. For analysis of grower samples, the chemists at SCGC perform the instrument diagnostics daily on each instrument. The results can be printed daily, or as often needed.

#### SUMMARY AND CONCLUSIONS

The overall performance of the NIR during the 1999/2000 crop season has been satisfactory. The NIR requires little to no maintenance, other than an annual lamp replacement. There are no harsh chemicals used for clarification, or cleaning. There is no need for sample preparation. The instrument is simple to operate for routine analysis, though the data manipulation can be challenging. There is ample support from the manufacturer while designing and implementing the calibration process. In addition, a four-day training session is included with the purchase of each instrument. Analysis on growers samples can be performed in less than one minute. Precise correlation can be obtained, and maintained with appropriate quality control measures such as diagnostic testing, equation validation and equation updating. This requires a dedicated chemist available to perform the wet chemical analysis and data manipulation on an as-needed basis. Other than the initial purchase of each instrument and the sample cups, no other expenditures have been made for the instrument to analyze incoming canejuice during this first season of operation. In contrast, the method used for ten years prior to the NIR, the Membrex membrane, polarimeter and refractometer, resulted in repairs, replacement parts, and replacement membranes totaling approximately 15,000 USD per year.

For the 2000-2001 season, development of NIR applications for the process laboratory will continue at SCGC along with plans to determine potential on-line applications in the factory.

## ACKNOWLEDGMENTS AND DEDICATION

The success of this project is the result of the efforts of many people. The author would like to acknowledge the contributions made by the chemists who were vital to the development and implementation of NIR spectroscopy at SCGC: Marcus Giddens, Aubrey Trotman, Candy Alfonso, Donald Boudreaux and Michael Carkey.

This paper is dedicated to the late Margaret A. Clarke, an extraordinary woman, friend, and scientist, and the impetus behind NIR spectroscopy in the sugar industry.

## REFERENCES

- 1. Chou, C.C. 1987. Alternate methods of polarizing sugar. Proc. Sugar Ind. Tech. 46:1 -26.
- Clarke, M.A. and B. Legendre. 1989. Replacement of lead salts in polarimetric analysis. Proc. USNC, 112-131.
- Clarke, S.J. 1982. A reagent based on aluminum for analytical clarification. Proc. 18th Session ICUMSA, 164467.
- DeStefano, R.P., E. Aguirre and H. Llorens. 1990. Ultrafiltration as an Alternative to Chemical Clarification in Cane Juice Analyses. JASSCT, 11:89-95,
- Rens, G. 1978. The use of aluminum salts as clarification reagents. Proc. 17<sup>th</sup> Session ICUMSA, 129-131.
- Winstrom-Olsen, B. 1988. Sugar analysis without lead Determination of the polarization of samples from a beet sugar factory without using lead salts. Zuckerind. 113:506-510.
- Meyer, J.H. and R. A. Wood. 1988. Rapid analysis of cane juice by near infrared reflectance. Proc.S. African Sugar Technol. Assoc. 62:203-207.
- Spencer, G.L. and G. P. Meade. A Handbook for Cane-Sugar Manufacturers and their Chemists. (1929). John Wiley and Sons, Inc., New York.









Figure 3. Scatter plots and statistics for preliminary equations relating conventional and NIR analyses for Brix and pol in cane juices.









RSQ: 0.974









RSQ: 0,987



Figure 5. Scatter plots for current equation relating conventional and NIR analyses of Brix and pol in cane juices and a comparison of statistical results for preliminary and current equations.

	Prelin	iinary E	quation	Current Equation			
Constituent	RSQ	SEC	Samples	RSQ	SEC	Samples	
Brix	.966	.246	886	.990	.196	1776	
Pol	.958	.182	886	.992	.181	1776	





# THE PRESENCE OF TOTAL POLYSACCHARIDES IN SUGAR PRODUCTION AND METHODS FOR REDUCING THEIR NEGATIVE EFFECTS

James A. Cuddihy, Jr., Miguel E. Porro, and James S. Raiih Midland Research Laboratories, Inc. 10850 Mid America Avenue Lenexa, Kansas 66219

#### ABSTRACT

A tremendous amount of research has been done on the various polysaccharides found in sugar cane and their effects on the processing and recovery of sugar. Polysaccharides found in the production process include those from the plant itself, which maybe dependent upon plant variety and weather patterns, and those resulting from deterioration processes due to cane handling methods. Efforts to deter the effects of polysaccharides have been primarily focused on starch and dextran, although there are several other polysaccharides contributing to these production inefficiencies and losses. Furthermore, while various researchers have examined the effects of polysaccharides, and considered means of decreasing their concentration, the industry has not generally taken vigorous steps to reduce their impact. The practice of adding a specific enzyme, for instance, to address a single polysaccharide often ignores the larger problem. Also, a detailed discussion is presented on how total polysaccharides contribute to production losses, the economic importance of more closely tracking their presence, and taking appropriate actions to reduce their negative influence. Because this is primarily a review paper, there is no Methodology section as no specific research was done.

#### INTRODUCTION

In an extensive review of polysaccharides, Imrie and Tilbury (1972) summarized the relevant data up to that time beginning with a general overview that, "Polysaccharides are long-chain molecules, either branched or straight, composed of simple sugar units linked in a definite fashion. A wide range of polysaccharides is found in nature and many of these are present in sugar cane and in the products derived from it. The polysaccharides found in sugar cane and its products are derived from two sources. There are those polymers, such as starch, which have arisen from the metabolic activities of the growing plant. There are also polymers, such as dextran, formed by the activities of microorganisms growing in the plant sap either during its life or at some stage in subsequent processing. All polysaccharides, by virtue of their physical properties, tend to have adverse effects on the processing of sugar cane. At best they represent a loss in terms of processable sucrose and at worst they may cause delays in processing due to excess viscosities and losses of sucrose due to modification of crystal growth. Soluble polysaccharides other than starch which occur in sugar cane and its products may be grouped into three main categories. The first group comprises structural polysaccharides originating in the standing cane plant, and includes hemicelluloses, pentosans" (presently known as 'pentans' following the standardization of polysaccharide terminology to use the simple suffix 'an' rather than 'osan') "and pectins. The second group comprises polysaccharides formed by bacteria before or during the processes of sugar manufacture and refining, e.g. dextran and levan. Thirdly, there is evidence that polysaccharides similar to dextran may be formed in harvested

cane in the absence of bacterial infection, presumably by the action of natural enzymes present in the cane juice in the parenchyma of the stalk, e.g. sarkaran."

In a later study Madhu et. al. (1984) stated that "starch and dextran present in cane juices interfere in their clarification. The extent of the presence of these polysaccharides in juices depends on the variety of cane, maturity period, harvest and transport conditions, etc. With high contents, juices show low filtration rates and poor crystallization."

And, in a more recent evaluation Godshall et. al. (1994) indicated "starch and other cane polysaccharides are significant minor components of cane juice that influence the yield of sugar in processing. Soluble polysaccharides expressed into the juice include phytoglucan, arabinogalactans, and other soluble cell-wall polysaccharides, as well as microbial polysaccharides that can be formed on cane - most commonly dextran. Polysaccharides lower the quality of the juice and raw sugar in several ways. During processing, they increase viscosity, slow or inhibit crystallization, and increase the loss of sucrose to molasses (i.e. they have a high melassigenic effect). Because of their carbohydrate nature and high solubility, they are difficult to remove in processing, and tend to be included in the raw sugar crystal, going into the refining process and causing similar problems in refining. Polysaccharides may also contribute to polarization distortion."

At the time of their review, Imrie and Tilbury (1972) offered the conclusion that "comparatively little has been published on the nature of the polysaccharides found in juice extracted from freshly harvested cane, or normal mill juices. Most workers have utilized non-specific methods for 'gum' or dextran determinations in studies of post-harvest deterioration problems. Gums constitute 0.3-0.6% of the soluble solids in sugar cane. During extraction of the juice in milling, some of the soluble structural polysaccharides of the cane are released into the juice. These include pectins and associated pentosans" (pentans) "such as arabinans. to addition, some soluble polysaccharides are suspended in the cane juice where they may later pass into solution during the alkaline liming treatment in clarification. This group includes the cellulosans" (cellulans), "such as glucosans" (glucans) "and xylans, and true hemicelluloses containing polyuronide residues."

The following discussion will focus on the origin, structure, properties and effects of polysaccharides in sugar cane from the field through processing as documented in the literature over the past 30 years, and will also address current and proposed methodology for dealing with resultant problems caused in the factory.

#### DISCUSSION

#### **Polysaccharides - Structure and Properties**

Dextrans, as summarized by Imrie and Tilbury (1972), "are homologous polymers of Dglucopyranose (glucans) containing predominantly a-( $1 - \infty 6$ ) glucosidic linkages, They are usually formed by the action of the enzyme dextransucrase on sucrose. The structure and properties of dextrans vary widely between strains of organism, and are also governed by conditions of cultivation such as sucrose concentration, pH, temperature and aeration. Dextrans consist of a basic straightchain polymer of a-( $1 - \infty 6$ ) linked glucose units, with some branches linked by a-( $1 - \infty 3$ ) or a-( $1 - \infty 4$ ) glucosidic bonds. The extent of branching and relative proportions of non a-( $1 - \infty 6$ ) linkages is variable. Most native dextrans have a high molecular weight, of the order of  $10^5$  -  $10^7$  or more. Aqueous solutions of dextrans may be quite viscous."

In another study Bose and Singh (1981) described dextran as a "homopolysaccharide made up of glucose units. It carries a basic straight-chain of a-(l-»b) linked glucose moieties with a-(l-»4), a-(l-»3) and a-(l-»b) linkages at the branching points. At least 50 percent to 60 percent of the linkages must be a-(l-»b) to define a polyglucose as a dextran. Other polysaccharides similar to dextran are presumably formed by the action of soil-bome organisms present in the cane juice in the parenchyma of the stalk, such as sarkaran, scleroglucan, pullulan and curdlan." And more recently Edye et. al. (1997) added that "dextran is a polymer of D-glucopyranose with a-(l-»b) linkages and a-(l-»3) branch linkages, and dextran is polydisperse and may contain other branch linkages such as a-(l-»2) or a-(l-»4)."

*Levans*, as described by hnrie and Tilbury (1972) "axe b-(2-w6)-linked polyfructosans" (polyfructans). "They are high molecular weight polysaccharides, mostly water-soluble, and exhibit negative optical rotation. The types found in sugar cane products are mainly formed by the action of levansucrase on sucrose, produced by bacteria of the genus *Bacillus*," And, Bose and Singh (1981) pointed out that "levan, the fructose polymer, is produced by bacterial action. Levan is of factory rather than field origin, whereas dextran, the glucose polymer is of both field and factory origin."

In their review, Imrie and Tilbury (1972), gave the following descriptions of pectins, cellulans and hemicelluloses, starch and gums:

"Pectins are gel-forming, water-soluble polygalacturonides with a-(l-»4) linkages found in the middle lamella between plant cells. D-galacturonic acid and its derivatives are the principal constituents. Various amounts of other sugars are commonly present. Neutral polysaccharides such as arabinans, galactans and arabinogalactans may accompany pectins, or perhaps form an integral part of pectins. Pectins are dextrorotatory."

"Cellulosans" (cellulans) "and hemicelluloses are associated with cellulose in the cell walls of plants. They are insoluble in water but are extracted by 4% caustic soda solution. Cellulosans" (cellulans) "comprise the homopolysaccharides, glucosans" (glucans) "and xylans. Xylan is highly laevorotatory. Hemicelluloses are hetero-polysaccharides containing uronic acids. They may be divided into two groups according to their constituent sugar units. Hemicelluloses are laevorotatory. Cellulose is bonded by b-(1-w4) linkages."

"Starch consists of two fractions, amylose and amylopectin, both of which are polymers of glucose. The fundamental linkage between adjacent glucose units is  $a-(1-\gg4)$ , the glucose being in the pyranose form. Amylose and amylopectin differ considerably in their structure and consequently have different physical properties. Amylose is essentially a linear polymer arranged as a helix, one complete turn of the helix accommodating six glucose units. The chain length, i.e. the average number of glucose units per non-reducing end group, is in excess of 2000. By contrast, amylopectin is a highly branched polymer in which branches occur by means of  $a-(1-\gg6)$  linkages. The chain length is variable within the range 19-28. Starch occurs in plants in an organized form and starches from different sources may contain differing amounts of amylose and amylopectin."

"Gums are defined as carbohydrates of high molecular weight precipitated from aqueous solutions by acidified ethanol. In sugar process materials they may include hemicelluloses, pentosans" (pentans), "pectins, dextrans, and levans. Heated materials may also contain starch in solution, but in freshly extracted cane juice starch is in granular form and is usually removed by filtration or centrifugation prior to test. Gums constitute 0.3 - 0.6% of the soluble solids in sugar cane. Gums present in the mother liquor are occluded in the crystal. Since little of the gums is removed by affination, it maybe deduced that gums originating in the raw factory process materials are carried over into the refinery and can exert their harmful effects at all stages in refining beyond the melter."

# Polysaccharides from the Metabolic Activities of the Growing Plant and Differences in Cane Varieties

Imrie and Tilbury (1972) state that "it is apparent from the literature reviewed that sugar cane and its products can contain a complex mixture of soluble and insoluble polysaccharides which varies according to external conditions. Further research is needed to clarify the identity, structure and origin of many of these polysaccharides. However, the evidence suggests that there are at least eight principal types. Fresh, healthy cane appears to contain four types of polysaccharide. One, which has been termed the natural polysaccharide of sugar cane, is a high molecular weight compound with negative optical rotation. The second type is pectin, derived from the middle lamellae of the cane plant cell walls. The third group comprises the structural polysaccharides of the plant cells, cellulosans" (cellulans) "and hemicelluloses. The fourth type is starch, the classic storage carbohydrate in cane as in other plants. Deteriorated cane and its products may contain in addition four other types of gum. Microbiological deterioration can result in the formation of bacterial dextrans and levans. Stale cane appears to form a polyglucan with a high proportion of a-(l-»4) linkages, named sarkaran."

And according to Bose and Singh (1981), "sarkaran, a polyglucan found in stale and wholestalk harvested cane has about 25 percent a:(1-»6) linkages." Imrie and Tilbury (1972) conclude that "finally, an intermediate complex of dextrorotatory, heterogeneous polysaccharides of unknown origin may be formed. They may be formed by natural cane enzymes or microbial enzymes or interactions of both."

An extensive study by Godshall et. al (1994) on polysaccharides compared cane variety, environment, location and microorganism effects reporting that "soluble polysaccharides expressed into juice include phytoglucan, arabinogalactans, and other soluble cell wall polysaccharides, as well as microbial polysaccharides that can be formed on cane - most commonly dextran. Starch, a storage polysaccharide of sugarcane, is an important part of the polysaccharide complex of cane. Starch concentration is elevated in immature and maturing cane and raw sugar produced early in the crop season can be high in starch. The soluble polysaccharide in fresh cane did not change significantly during the last three months of maturity, but location did have an effect. A study of four varieties (Australia) concluded that starch was a definite varietal character. Three varieties (Louisiana) showed a significant increase in starch between the first and second harvest dates, and none showed a significant decrease. When all varieties are averaged, there was a significant increase between the first and second, and the second and third harvest dates, followed by a significant decrease in December. Five varieties decreased significantly in total polysaccharide content from the first to the second harvest date, and three remained unchanged, leading to an average overall significant decrease between October 1 and October 15 dates. For most varieties, the total polysaccharides did not change significantly from the beginning to the end of the harvest season. All varieties had a significant increase in total polysaccharide concentration coinciding with unseasonably warm and wet weather. Cane varieties differ significantly in the juice concentration of starch (range 5 times higher) and total polysaccharides (range 2 times higher). A previous study had shown that around 30 - 40% of the juice starch will end up in the raw sugar crystal. At about 200 - 250 ppm starch level in raw sugar, the refinery may begin to experience process difficulties. While juice composition is sensitive to weather changes during the harvest (i.e. unseasonably warm and wet periods), varietal differences are more significant than seasonal changes and varieties are consistent from year to year in relative concentration of starch, total polysaccharides and proanthocyanidin."

Hidi et. al. (1976) studied the polysaccharide concentrations in diseased cane and immediately after harvesting stating that "the average dextran content of expressed juice (Australia and Fiji) was reported to be 172 ppm on Brix for cane apparently free of disease, and 279 ppm for cane that was diseased. From the results of work in Jamaica in 1969-70 Imrie and Tillbury (1972) also concluded that freshly-harvested cane often exhibited a haze by dextran test; the average initial dextran content being 1800 ppm on Brix, with values as high as 5000 ppm on Brix. It appears that from our results and others test results that there may be appreciable differences between sugar canes in regard to the nature of the alcohol precipitable impurities in the cane immediately after harvest. Linkage determinations suggest that at dextran concentrations of 1000 ppm on Brix or higher, the polysaccharide contained predominantly a(1 - 6) glucosidic linkages. This suggests that the material in the haze at high dextran concentrations was almost exclusively a true bacterial dextran. At low dextran concentrations (less than about 300 ppm) the material forming the haze contained an appreciable proportion of 1 - 4 and 1 - 3 linkages, and thus resembled natural cane polysaccharides reported previously by other researchers."

# Effects of Harvesting and Weather, Storage and Delays before Processing

Hidi et. al. (1976) also addressed the effects of cane deterioration and delayed delivery to the factory as follows, "deterioration of sugarcane between harvesting and milling has long been associated with losses of sucrose and the formation of deterioration products which are harmful to the processes of mills and refineries. A polysaccharide, usually referred to as dextran as typifying a class of compounds which occurs widely in deteriorated cane and which has a major influence on processing performance. Trials in Australia showed that the viscosity of B syrup containing high concentrations of dextran could be reduced by 20% as a result of enzymatic dextran hydrolysis. In addition to these experiments demonstrating a causal relationship between dextran and retrogressing performance and the dextran content of juices or process materials. Cane that was burnt and left standing in the field, usually in wet ambient conditions (Australia and Fiji) contained up to 280 ppm dextran on Brix three days after burning, rising to as high as 2900 ppm after 7 days."

Coll et. al. (1978) researched the effects of harvesting conditions (i.e. wet and freezing weather) as well as mechanical harvesting methods and reported that "increasing research has been directed toward understanding and correcting problems of cane deterioration, particularly those associated with chopper harvester operations. Rates of deterioration are primarily functions of the degree of mechanical damage, cut-to-crush delay, environmental conditions, degree of burn and delay of harvest after burning, degree of frost or freeze damage, and combinations of these factors. During periods of good harvesting weather, variations in cane quality are small; however, wet conditions produce extreme variations in quality. A study of dextran and total polysaccharides on Louisiana process streams (i.e. mixed juice, syrup, B-molasses, final molasses and raw sugar) included total polysaccharide analyses to study the dextran/total polysaccharide relationship. Dextran contents increase progressively from dilute juice to final molasses, as is expected with the removal of sugar solids. Harvesting conditions were unusually wet throughout the season. There were large increases between the mixed juice and syrup stages. Polysaccharide content of syrups was closely related to field conditions, and, as expected, increased in concentration as sugar solids were removed in processing. Total polysaccharide data indicate a high degree of variability that is inconsistent with dextran values. Large amounts of soil present in mixed juice samples probably trapped appreciable quantities of the polysaccharides. Although lime clarification removes a high percentage of polysaccharides during periods of low soil, the minimum content at the syrup stage is probably about 3000 ppm on Brix."

In their review, Imrie and Tilbury (1972) discussed the presence of a polysaccharide that may not be due to the presence of microorganisms but is formed by the presence of natural enzymes in the juice as "another polysaccharide, found to be a glucose polymer, but differing from the L. mesenteroides dextran in that it contained a low content of a-(1-»6) glucosidic linkages (29% approx.) and a high content of a-(1-»4) linkages (60% approx.). Further evidence that it differed from starch was shown by the absence of color formation by the iodine test, and differences in the chromatographic pattern of enzyme hydrolysates. Bacteriological studies revealed that this polysaccharide was not associated with the growth of L, mesenteroides in the stalks and was formed in the absence of infection. It was postulated that the polysaccharide may be formed either by the priming action of dextran from dextransucrase upon another polysaccharide synthesizing system or by plant enzymes alone. Later work showed that a polysaccharide was formed in stored cane in much greater amounts under dry conditions than in moist conditions. Lactic acid formation was not detected, so it was assumed that the dry cane was not infected by L. mesenteroides. It was found to be a straight chain glucose polymer containing 25% (1 ->6) and 75% (1 ->4) a-glucosidic binds. The polymer differed significantly from both starch and bacterial dextran. Since the cane polysaccharide appeared to be a new a-polyglucan it was named sarkaran. There is evidence that sarkaran may be formed in harvested cane in the absence of bacterial infection, presumably by the action of natural enzymes present in the cane juice in the parenchyma of the stalk."

## Polysaccharides Polymers, such as Dextran, Formed by the Activities of Microorganisms

hnrie and Tilbury (1972) also presented extensive observations on microbial formation of polysaccharides beginning with the fact that "it has been known for many years that post-harvest biodeterioration of cane frequently results in excessive gum formation, with subsequent harmful effects in the factory. This deterioration is generally thought to be caused by infection of the cane stalk with the lactic acid bacterium *Leuconostoc mesenteroides*, which converts sucrose to fructose and dextran. Similar changes occur in the extracted juice in the mill. The problem is often termed 'sour cane' because of the associated increase in organic acids such as lactic and acetic acid in the juice. In recent years interest in this subject has been renewed. This stems partly from a realization that sucrose losses due to sour cane may be significantly high, and that dextrans can reduce factory capacity and efficiency. The main stimulus, however, originated in Australia, when it was discovered that a changeover from manually harvested, whole-stalk cane to mechanically harvested, chopped-up cane greatly increased the degree of infection of cane with *L. mesenteroides* and hence increased the extent of sour cane. A further consideration was that raw sugar quality was adversely affected by the presence of dextrans in manufacture, thus leading to the possibility of penalties imposed by refiners. In Louisiana, sour cane is often experienced when a period of warm weather follows a freeze during the harvesting season. It is thought that freeze damage increases the susceptibility of cane to infection of some plant gums in the cell walls. When freeze damaged cane is milled, processing difficulties due to gums are usually encountered."

On the effects of billet-cut cane, Imrie and Tilbury (1972) reported that "bacterial deterioration of chopped-up cane on Queensland was first studied by Egan who named the problem 'sour storage rot'. In a series of papers Egan established that this disease was caused by infection of the cane with L. mesenteroides at the moment of harvest. Chopped cane deteriorated much faster than whole-stalk cane due to the excessive damage and infection caused by the mechanical chopperharvester. The gums were not characterized, but in view of the rapid increases in gum content accompanied by similar increases in L. mesenteroides counts of the cane, it is reasonable to assume that the new polysaccharides formed were mainly bacterial dextrans. A comparison was made between gum content and pH as measures of losses due to sour storage rot and it was concluded that gum content is a better indicator than pH. Chopped cane exhibited much greater rates of dextran formation than whole-stalk cane, probably due to the greater area of exposed stalk and hence greater degree of infection. It was estimated that with chopped cane the dextran content would cause significant processing difficulties within 18 hours of harvest in cool, dry weather or 14 hours in hot. wet weather. Dextran was not detected in fresh cane or in whole-stalk cane after 30 hours storage. Dextran was frequently detected by the haze analysis technique in freshly harvested, chopped cane. A comparison between cane diseased by red rot and healthy cane showed that the former contained slightly higher dextran contents on average than the latter. Some healthy cane contained very high dextran levels. In view of the non-specificity of the haze analysis technique for dextran, it seems unlikely that the observed 'dextran' was derived from bacterial infection. Most probably it was abnormal polysaccharide produced by natural processes within the healthy cane."

"Another significant observation" noted by Imrie and Tilbury (1972) "was that the rate of dextran formation was much higher in burnt than in green cane. There appear to be two possible explanations for this phenomenon. Burning may increase the susceptibility of cane to infection by *L. mesenteroides*, either by causing splits in the rind which facilitate entry of bacteria, or by inactivating the plants' natural antibacterial enzymes, phenol oxidases. Alternatively, the observed 'dextrans' may be non-bacterial polysaccharides synthesized by enzymes within the cane cells in response to temperature rises, In general, both dextran content and juice viscosity were directly proportional to time of storage." And quite importantly, Hidi et. al. (1976) pointed out that "the level of dextran in the juice and process materials has been found to be a sensitive indicator of the processing quality of the cane."

A study reviewed by Bose and Singh (1981) to evaluate "dextran level in raw sugars with the respective sucrose losses incurred to produce these dextran levels and the amounts of fructose and acids (about 30% yield) formed " is shown in Table 1.

Dextran	Sucrose lost	Fructose formed		
(%)	Kg/Tonne sugar	(Kg/Tonne sugar)	% Acid produced	
0.05	0.20	0.99	0.07	
0.10	0.40	1.98	0.17	
1.50	2.00	9.90	0.70	

Table 1. Dextran levels and sucrose loss in raw sugars (Bose and Singh, 1981).

Bose et. al. (1981) concluded that "these amounts of sucrose loss represent only those lost directly due to dextran formation. Three to five times these levels may be lost later in processing because of the other organic materials formed conjointly with dextran."

A recent study (Cerutti de Guglielmone et. a1, 2000) showed that *Leuconostoc mesenteroides* (strain 3A) consumes "sucrose very quickly (8.05 g/hr at 25 °C and 8.46 g/hr at 30 °C) during the first 6 hours of culture. This fermentative process implies a sucrose consumption of 59% at 25 °C and 62% at 30 °C. At higher temperatures (37 °C and 40 °C) the percentage of consumed sucrose decreases to 47% and 27% respectively." The strain of organism studied was isolated from sugarcane juice in Argentina. "Its high femientation rate consumed sucrose rapidly, stopping its growth and sugar utilization after 6 hours incubation. From the sugar industry point of view it is important to know the consumption of sucrose in short periods of time", as shown in Table 2.

 Table 2, Sucrose consumption and dextran production at different time intervals. Leuconostoc

 mesenteroides 3A; culture medium 10% sucrose basal medium; incubation temperature 30 °C

 (Cerutti de Guglielmone et. al., 2000).

Time	Consumed sucrose	Dextran		Sucrose/	Dextran yield
(hours)	(g/l)	production	(g/l)	dextran ratio	(%)
1	8.50	0.35		24.29	4.12
2	17.00	0.60		28.33	3.53
3	25.40	0.80		31.75	3.15
4	33.80	0.90		37.56	2.66
5	42.30	1.08		39.17	2.55
6	50.80	1.30		39.08	2.54
9	51.90	2.25		23.07	4.33
15	53.40	5.10		10.47	9.55
18	54.10	7.00		7.73	12.94
24	55.50	13.00		4.27	23.42
48	57.50	13.20		4.35 23.0	0

"Taking into account only the first hours of the fermentative process at 30 °C," Ceratti de Guglielmone et. al. (2000) observed that "the consumed sucrose (g/1) to produced dextran (g/1) ratio was high for *Leuconostoc mesenteroides* 3A. Analyzing the relation between consumed sucrose and produced dextran, it can be seen that the amount of sucrose required to produce 1 gram of dextran varied remarkably with the incubation time."

# Processing Difficulties in Raw Factories

"It has been known for many years that gums from sour or stale cane adversely affect the process of sugar manufacture, (Imrie and Tilbury, 1972). With the exception of starch little is known about the effects of "normal" polysaccharides present in fresh cane on processing. Since the early days of sugar manufacture there have been many reports of the formation of insoluble deposits of gum in sugar factories and refineries which cause clogging and blockage of pipelines, tanks, strainers, filters, etc. These were invariably due to the growth of bacteria in the process, with constant formation of dextrans and levans."

In their review, Imrie and Tilbury (1972), gave the following summary of effects on the process including pol analysis, clarification, evaporation and crystallization, factory capacity, scale formation, crystal shape, exhaustibility of massecuites, molasses purity, and gelling of molasses:

"Pol analysis: One effect of dextran in process materials is interference with analytical tests for sucrose and purity in process control. Dextran is highly dextrorotatory and therefore inflates the direct polarization reading of samples, unless removed prior to test. Furthermore, high dextran levels reduce the efficiency of clarification techniques used in pol determinations."

"Clarification: At the clarification stage in raw sugar manufacture, abnormally high dextran levels cause severe effects. Juice derived from deteriorated cane contains excess acids and requires extra line addition to neutralize the acidity. Scale formation in the juice heaters decreases heating efficiency. The presence of excess gums increases juice viscosity, which retards the settling time in clarifiers and prevents satisfactory removal of suspended matter. Consequently the clarified juice is cloudy, resulting in higher mud volumes at the filter station. Filtration of muds is also impeded by the excess viscosity of the juice."

"Evaporation and crystallization rate of sucrose; During the stages of evaporation and crystallization, the major harmful effects of dextran are an increase in the viscosity of syrups and massecuites and a decrease in the rate of crystallization of sucrose."

"Factory capacity: The most important economic effect of increased dextran levels is that factory capacity is reduced. This is due to the combined effects of increased viscosities, which increase boiling and evaporation times, and decrease crystallization rates. Thus an increase occurs in the time required to manufacture unit weight of sugar and the milling rate may have to be reduced." "Scale formation: Other secondary effects due to dextran include excessive scale-up of heaters and evaporators which leads to poor heat transfer. This, combined with increased viscosities increases the boiling time and hence leads to greater loss of sucrose by inversion."

"Crystal shape: It has been shown that dextran not only retards the overall crystallization rate of sucrose, but it selectively retards growth along the *a* and *b* axes. This results in needle-shaped crystals elongated along the *c* axis. Needle-shaped raw sugar crystals are highly undesirable for several reasons. Firstly, they reduce the efficiency of purging of massecuites in the centrifugals, resulting in poor separation of crystal and molasses. Secondly, the shape is less acceptable to the customer from an aesthetic viewpoint. Thirdly, and more important, the refining quality of the sugar is reduced. The degree of elongation was independent of starch, silica and phosphate content of the raw sugars, but a close relationship existed for gum content and a fairly close relationship for oligosaccharide content. It was clearly shown that gum present in the mother liquor is occluded in the crystal. Since little of the gums is removed by affination, it maybe deduced that gums originating in raw factory process materials are carried over into the refinery and can exert their harmful effects at all stages in refining,"

"Exhaustibility of massecuites and final molasses purity: The increased viscosity of massecuites coupled with reduction in crystallization rate due to dextran leads to a reduction in the exhaustibility of low-grade massecuites in crystallizers. Since crystallization takes longer in the presence of dextran, massecuites become cooler than normal, which increases the already abnormally high viscosity. Purging of such massecuites becomes difficult and inefficient, especially in batch centrifugals, due to the combination of high viscosity mother liquor and needle-shaped crystals. Raw sugar derived from these massecuites is sticky, and difficult to handle, dry and pack. The net result of these effects is an increase in the purity and volume of final molasses/ton cane, and hence a loss in tons sugar/on cane."

"Stickiness and gelling of molasses: Accumulation of gums in molasses is thought to be responsible for the phenomena of 'stickiness' and occasional gelling of molasses in transit."

Imrie and Tilbury (1972) conclude that "it is evident that polysaccharides can seriously reduce the efficiency of both raw sugar manufacture and refining. Soluble polysaccharides such as dextran and dissolved starch produce processing difficulties mainly in the factory during the production of raw sugar. This results in the production of poor quality raw sugar which has a consequent depressing effect on refining efficiency. The main harmful effects of soluble polysaccharides on the process are:

- (a) the production of excessive juice viscosities leading to poor clarification and filtration,
- (b) a reduction in the rate of crystallization of sucrose,
- (c) elongation of the c axis of the sucrose crystal with consequent inefficient separation and purging on the centrifugals,
- (d) an overall reduction in the economic efficiency of the mill and the subsequent refining.

Insoluble polysaccharides such as starch are known to pass through the factory and (together with some of the soluble polysaccharides) become incorporated into the raw sugar crystal. Their main effects are seen during refining and particularly influence the clarification and filtration adversely. Usually, the harmful effects of polysaccharides on processing only receive attention when abnormally high polysaccharide contents enter the process, such as when sour or stale cane is milled. Here the economic effects can be severe."

Hidi et. al. (1976) observed that "the level of dextran in the cane supply (Australia and Fiji) at the mills ranged, on a daily basis, from zero to over 5000 ppm on Brix, but was generally below 200 ppm. Levels in excess of 200 ppm were associated with unusually long harvesting-crushing or burning-crushing delays, excessive wet weather, or unfavorable ambient conditions in the latter stages of the season; all of which led to increased rates of deterioration between cutting and crushing. Within a particular season, there was a general tendency for dextran levels to increase as the season progressed. This was associated mainly with the increase in ambient temperature, humidity, and rainfall intensity throughout the season. Mills receiving an increasing proportion of delayed burnt cane often exhibited very high levels of dextran."

And more recently, Clarke et. al. (1997) concluded that "the major problem from dextran is that its presence indicates a loss of sucrose in cane, harvesting and transport, the mill yard and in the factory. Dextran in juice, syrups and sugars can cause false pol, because dextran polarizes about three times as much as sucrose and gives a falsely high pol. Dextran in solution increases viscosity, lowers evaporation rates and reduces heat transfer. It slows boiling times, and slows purging in centrifugals through the viscosity increase. It's estimated that for every 300 ppm dextran in syrup there is a 1 % increase in molasses purity."

"From an industrial point of view," according to Cerutti de Guglielmone et. al. (2000), "studies have shown the presence of Leuconostoc mesenteroides strains able to utilize high percentages of the sugar present in juices in short time periods. This may imply important losses if the infection level is not controlled. High loss in product yield (sucrose) and contamination of the industrial process cause various problems to sugarnills, such as: an increase of juice viscosity which produces blockage in the process line, pumps and filters; lower heat exchange; evaporation diminution; decrease in the efficiency and output of crystallization; crystal shape distortion; blockages in centrifuges and sucrose losses in the molasses and dextran is occluded in sugar crystal resulting in possible payment penalties assessed by the refiner. It should also be kept in mind that dextran increases the polarization readings and can interfere in the sucrose analysis. Therefore, it would be advisable to increase microbial controls in sugarmills to keep losses under control."

## Processing Difficulties in Refineries

Imrie and Tilbury (1972) also included refining operations in their review and observed that "in a sugar refinery, polysaccharides can only originate from two sources. Some may enter the refinery in raw sugar, either occluded in the crystal or present in the molasses film surrounding it. Other polysaccharides can be formed during the refining process itself by microbial action. The effect of starch on the processing of sugar from cane to refined sucrose is better defined than that of gums. It is clear that starch in cane juice and raw sugar influences the filterability of these materials. There is a wide divergence of opinion on the level of starch at which these effects become significant and whether they are due just to starch or to a combination of the effects of this polysaccharide and other non-sugar impurities. Much more is known about the effects of polysaccharides on the affination and filtration of raw sugars than on any other aspect of the sugar production process. In refining, the harmful effects of polysaccharides are first observed at affination. Here the presence of needle grains of uneven size, coated with a film of molasses of excessive viscosity, reduces the efficiency of molasses separation in the centrifugals. This leads to subsequent difficulties in color removal and other refining processes. Since filtration-impeding substances such as gums and insoluble matter are occluded within the sugar crystal, affination is often ineffective in removing them from the process stream. There is ample evidence that raw sugars containing exclusive amounts of polysaccharides impede filtration in the refinery."

In summarizing work performed on starch and dextran at a refinery, Johnson (1989) observed that "whatever their origin, the presence of starch and dextran in raw sugar either individually or combined, can have a devastating effect on the throughput of a refinery. High starch concentrations in conjunction with mid-range dextran could result in a synergistic increase in viscosity problems. That is, that the two combined would have much worse effects than the sum of the two would indicate. Effects that include blocking filters, increasing viscosity, reducing throughput, and decreasing crystal yield. These debilitating effects will occur in both the refinery and the mill. Starch is generally insoluble at lower temperatures, but when juice or liquor reaches 60 °C (140 °F), the starch "gelatnizes". This occurs as the starch granules swell and occupy greater volumes of space. Solubility of starch at high temperatures becomes apparent as viscosities increase."

Additionally, Anyangwa et. al. (1993) reported in their findings that "starch is a natural polysaccharide of sugar cane juice. Its concentration depends on many factors such as cane variety, growth condition and the age of the cane before harvest. Starch is not soluble in water except when treated as in clarification. During the clarification process the starch becomes partially soluble leading to some starch removal while some remains in the syrup. If the amount of starch that remains in the syrup is high (above 150 ppm), starch can delay crystallization in the vacuum pans preventing molasses exhaustion. Starch may become occluded with the sucrose crystals or absorbed in it: and these conditions slow down filterability in the refinery. The starch content of raw sugar A decreases as the sugar is filtered. This explains the fact that the filtration process traps roughly 75% of starch present in raw sugar and also explains the effects of starch in filtration (starch depresses the filtration rate). The amount of starch in filtered syrup is not negligible. The great reduction of starch content from the filtered syrup to the refined sugar also explains the fact that the process of crystallization is retarded by starch. If the amounts remaining in the filtered syrup are high, starch can retard crystallization in the vacuum pans degrading as well the final refined sugar. The consequence of this problem is that if the final starch content of raw sugar A is more than 150 ppm (based on dry matter), the defects will be noticed."

#### Current and Proposed Methodology for Dealing with Resultant Problems in the Factory

Madhu et. al. (1984) substantiate that "the application of enzymes for the removal of starch and dextran from the juices improves the quality of the sugar produced. The enzymes being highly specific in nature, a mixture can be used for simultaneous degradation of the polysaccharide constituents starch and dextran. This will be especially helpful in cases of mill breakdown and supply of stale cane. A long reaction time and low Brix are favorable and the enzymes should thus be added as early as possible during milling and evaporation. At 60 Brix sucrose concentration about 30 -40% less enzyme activity was observed. Higher doses of enzyme or more time were required at pH and temperature other than the optimum and also for higher Brix syrups. The presence of coagulated proteins inhibited the amylase activity but not the dextranase activity. This showed that the dextranase enzyme was able to hydrolyze all types of dextrans present or formed in cane juices during different harvesting conditions, mill breakdowns and deterioration. The decreased hydrolysis of polysaccharides in soiled parts of cane may be due to the presence of metallic inhibitors in the sample."

Furthermore, Anyangwa et. al. (1993) also concur that "the use of enzymes for eliminating starch has now been recognized to be the most effective method in sugar juice processing. It is known that the removal of starch in cane sugar processing increases the factory capacity, the product yield and product quality. The elimination of starch in the juice at the evaporators will reduce the content of starch in sugar A and as a result decrease the purity and increase the starch content of the final molasses and consequently lead to an increase in the filtration and crystallization rates."

# SUMMARY

"Usually, the harmful effects of polysaccharides on processing only receive attention when abnormally high polysaccharide contents enter the process, such as when sour or stale cane is milled. Here the economic effects can be severe. It is not known what economic losses are caused by normal levels of polysaccharides in the process. Further study of the harmful effects of polysaccharides in processing and methods of minimizing these appears to be a worthwhile topic for research. This is especially so because there is an increasing consciousness on the part of refiners concerning raw sugar quality." (Imrie and Tilbury, 1972).

The above words were written in 1972, and yet the industry still struggles with these problems today. Enzyme development has progressed, and their initial high cost has begun to decline to reasonable levels. Recent research has determined more effective means of handling and feeding to overcome the negative influence of pH, temperature and Brix which can quickly inactivate the enzymes.

Currently there are two predominant enzymes being used in sugar processing, a-amylase for starch reduction and dextranase for dextran reduction, both of which have the primary effect of reducing viscosity of process streams. The predominant linkage in starch that a-amylase attacks is 0.(1\_»4). xhe predominant linkage in dextran that dextranase attacks is a-(1-»6).

While much of the early investigation covered the broad scope of the polysaccharides, most of the recent research has generally focused on starch and more recently on dextran. The data in the following tables, again taken from the literature, show a definite relationship between starch or dextran and the presence of other polysaccharide compounds. Current technology can be modified to address the negative effects of total polysaccharides in the sugar factory with the result of reducing further losses of sucrose during production and alleviating the production problems caused by the presence of total polysaccharides in the process stream.

Table 3 shows the results of seasonal mean concentrations of TPS and starch for selected varieties. The difference from year-to-year among the different varieties is quite apparent. The concentration of starch is generally one-third the concentration of the TPS present. Using only one enzyme for starch reduction ignores the larger problem presented by the higher concentration of TPS. A combined enzyme program would address the predominant species of a-(1-»6) and a-(1-»4) linkages among all TPS present in the factory.

	TPS (	TPS (ppm on solids)		Starch	(ppm c			
Variety	1990	1991	1992	1990	1991	1992		
CP 72-370	3234	3308	4957	1460	1867	1548		
CP 79-318	2577	3082	3126	986	1092	986		
LCP 82-89	2161	3107	2841	566	701	538		
CP 65-357	2128	2849	2908	506	745	557		
CP 74-383	1931	3162	2671	611	590	636		
CP 70-321	1544	2496	2862	275	239	220		

Table 3. Comparison of total polysaccharide (TPS) and starch concentration of selected sugarcane varieties over several years (Godshall et. al, 1994).

Table 4. Percent increase in gum content of juice during storage of harvested cane (Imrie and Tilbury, 1972).

Cane storage time (days)	Whole stalk	Chopper	
1	16.1	75.0	
2	58.1	237.5	
3	103.2	384.4	
4	1413	5344	

The data in Table 4 show the significant increases in gums (total polysaccharides) not only between whole stalk and chopped cane, but also over time. In as little as 24 hours there is significant formation of gums in the stored cane, hi some parts of the world, 48 hours between harvesting and milling is not uncommon. Although sucrose that has been inverted is not recoverable, one can again make a reasonable argument for using multiple enzymes in the process.

Table 5. The effect of burning and storage on gum content of crusher juice (Imrie and Tilbury, 1972.)

Cane storage time (days)	Cane	Gum content (mg/10 ml)
0	Green	0.83
	Burnt	0.82
15	Green	1.81
	Burnt	3.53
25	Green	2.27
	Burnt	6,61

It has long been known that delays in processing of cane lead to process materials handling problems as clearly shown in Table 5. Most often these problems result from increases in viscosity leading to increased boiling times and elongated crystals. The larger impact to the factory, however, is loss of recoverable sucrose. Although the sucrose lost to inversion during storage and other delays cannot be recovered, the removal of these process contaminants will allow increased recovery of the available sucrose. The recovery of this economic loss can offset the cost of using enzymes effectively to recover more sugar, having higher quality, along with the elimination of penalties. The refinery will enjoy greater throughput when processing raw sugar containing fewer total polysaccharides, so there is economic justification for using enzymes.

Table 6. Gum content (mg/10 ml juice) of first expressed mill juices before and after storage of cane over the weekend (Imrie and Tilbury, 1972).

	Whole	Whole stalk cane		l-up cane	
Year	Fresh	Stored	Fresh	Stored	
1965	6.9	10.0	10.3	47.1	
1966	6.8	<u>9.0</u>	6.4		231

In the United States we have seen the mechanization of cane harvesting take place. And more recently, we have seen an increase in billet harvesting. Along with this trend, increases in starch and dextran have been reported. Often the increased starch levels are due to operation of the harvesting equipment or excess trash delivered to the factory. But dextran increases have been associated with the increased exposure of surface area of the cane stalk to bacterial contamination. Even 35 years ago, as shown in Table 6, this was known to occur from excessive damage and infection caused by the mechanical harvester.

It is very unlikely that mechanical harvesting practices will change significantly in the near future. We can therefore state with some certainty that total polysaccharide problems associated with harvesting will continue. This also supports the use of multiple enzymes, including some not currently used, in the factory to reduce the effects of total polysaccharides on the production process.

Table 7. Analysis and effect on crystallization of native dextrans (Imrie and Tilbury, 1972).

	-Linkage analysis-			Effect o	Effect on crystallization—		
	%1-6	%1-4	%1-3	% Added	Elongation (c/b)		
Refractory mill syrup	83	11	6	1.0	1.27		
Refractory refinery syrup	60	22	18	0.6	0.64		
Deteriorated juice	79	7	14	0.5	0.98		
Stored cane I	32	66	2	1.75	0.57		
Stored caneH	33	64	4	0.75	0-61		

The data in Table 7 show that the predominant linkage, 60 - 83%, in refractory syrups and deteriorated juice is a-(1-»6), while the a-(1->4) linkage accounts for between 7 - 22% of the molecule. In the case of stored cane, this relationship is reversed with 64 - 66% a-(1-»4) linkages and 32 - 33% a-(1-»6) linkages. These relationships clearly show why it is important to use at least two different enzymes whenever old or stale cane is being processed.

	Strike	Polarization	Starch (ppm)	Total gums (ppm)
Factory 1	А	99.7	365	1160
	В	99.6	610	1440
	С	98.2	650	1670
Factory 2	А	99.7	180	870
•	В	99.3	230	1390
	С	98.5	-	2030
Factory 3	А	99.7	195	910
	В	99.5	310	1560
	С	993	310	2440

Table 8. Variation in starch and gums levels in A, B and C sugars (Imrie and Tilbury, 1972).

Total polysaccharides (i. e., starch and gums), as shown in Table 8, increase in each successive strike. From these data it is also quite apparent that the concentration of the starch averages only 25% of the concentration of the total gums (polysaccharides) in the samples.

If enzyme for starch removal is being used, a large portion of the total polysaccharides present is most likely not being affected by the applied a-amylase enzyme dosage for starch reduction. A greater benefit in viscosity reduction may be achieved by adding dextranase enzyme to the process to attack the total combined concentrations of a-(1->6) and a-(1->4) linkages.

 Table 9. Dextran and total polysaccharides (ppm on Brix) in random sugar samples (Coll et. al., 1978).

Sample	Dextran	Polysaccharides	
Louisiana factory B raws	20-500	660-2600	
Texas raw	90	1270	
Phillipine bulk raw	60	520	
Brazil plantation white	990	1570	

The comparison of dextran to TPS in raw sugar in Table 9 shows that if only one enzyme is used to reduce the effects of dextran, again, the larger problem presented by the higher concentration of TPS is ignored. A combined enzyme program would address the predominant species of a-(1-»6) and a-(1->4) linkages among all TPS present in the factory.

Once Leuconostoc mesenteroides has entered the factory, it continues to proliferate with two consequences: the further loss of sucrose, and, the formation of additional dextran. The use of biocide in the milling process has been proven many times to have positive results. A combined program of biocide addition and physical cleaning with hot water or steam every eight hours is the most economical means of achieving control of microbial infestations in the factory. The use of biocide and mill cleaning should have high priority in every factory.

Of importance is the comment in the discussion regarding the fact that metallic ions present in the juice may result in decreased hydrolysis of the polysaccharides by the enzymes. This further substantiates the need for an adequate program of microbiological control in the milling process with a particular biocide. The most commonly used biocide for controlling microorganisms in juice is based on carbamate. Since carbamates are efficient metal ion chelants, using continuous feed at appropriate levels in milling will reduce the negative effect of metallic ions on the activity of the enzymes.

		First series (%)			Second series (%)		
Materials	Gums	Starch	Gums-Starch	Gums	Starch	Gums-Starch	
Raw sugar	0.259	0.091	0.168	0.272	0.093	0.179	
Washed sugar	0.179	0.078	0.101	0.222	0.084	0.138	
First crop recovery sugar	-	-	-	0.283	0.091	0.192	
Double cured second and							
third crop recovery sugars	-		-	0.272	0.088	0.184	
Remelt liquor	0.304	0.116	0.188	0.329	0.108	0.221	
Carbonatation supply	0.215	0.091	0.124	0.252	0.098	0.154	
Filtered liquor	0.173	0.053	0.120	0.195	0.053	0.142	
Fine liquor	0.184	0.052	0.132	0.182	0.054	0.133	
Refined sugar	0.075	0.028	0.047	0.080	0.030	0.050	
Greens	1.323	0.243	1.080	0.366	0.276	1.090	
Tail-end syrup	2.015	0.378	1.637	1.569	0.379	1.190	
Molasses	5.329	1.000	4.329	5.498	1.171	4.327	

Table 10. Gum and starch determinations in a sugar refinery (Imrie and Tilbury, 1972).

Comparing the data on gums and starch in Table 10, we find that throughout a refinery, the excess quantity of gums over starch is quite significant and the ratio increases further into the process. Since gums from the raw factory are not removed by affiliation, the refinery suffers all the negative effects imparted by the gums (other polysaccharides) in addition to the starch. This confirms the need for the raw factory to consider using two enzymes, and even higher dosages, to reduce the contaminant level on their raw sugar and avoid penalties.

Polysaccharides-			—Linkages			
in canea>(1->6)	a-(1->4)	a-(1->3)	a-(1->2	2)	3-(1->4)	3-(2->6)
Dextrans	Х	Х	Х	Х		
Starch-amylose		Х				
Starch-amylopectin	Х					
Sarkaran	Х	Х				
Cellulose					Х	
Pectins		Х				
Levans						Х
Polysaccharides						
in the factory						
Stored cane		X X	Х			
Deteriorated juice		х х	Х			
Syrup	Х	X X				

#### Table 11. Summary of total polysaccharide linkages found in sugar cane and in the factory.

Table 11 summarizes those linkages identified in the literature as being associated with the list of polysaccharides and factory process shown. Although specific reference to linkages for other "gums" were not identified, it is highly probable that these polysaccharides also contain some percentage of the linkages shown in the table.

## CONCLUSIONS

The factory, as well as the refinery, can benefit greatly from all of the following:

- Recognizing that total polysaccharides, including natural dextran, are present at all times, even though their detrimental effects may not be readily apparent.
- 2) Utilizing a coordinated chemical program of biocide and physical cleaning of the mill.
- Continuously feeding biocide at levels appropriate for the cane and processing conditions and to also enhance the activity of enzymes.
- Utilizing a coordinated chemical program of mixed enzymes applied appropriately throughout the factory.
- 5) Continuously feeding multiple enzymes at levels appropriate to the cane and processing conditions to hydrolyze the linkages among all TPS present in the factory.

The increase in recovery and improvements in processing conditions will have a return on investment that will offset and likely exceed the costs of using multiple enzymes and bilocide continuously and at higher dosages.

#### REFERENCES

- Anyangwa, E.M., C. Kapseu, Elemva and P. Musonge, 1993, The effect and removal of starch in the sugar refining industry, Int. Sugar J, 95:210-213
- Bose, S. and L, Singh, 1981, Problems associated with dextran, the major microbiological cause of sucrose loss and their remedies, Indian Sugar, December, 603-608
- Cerutti de Guglielmone, O. Diez, G Cardenas and G. Oliver, 2000, Sucrose utilization and dextran production by *Leuconostoc mesenteroides* isolated from the sugar industry, Sugar Journal, 62:36-40.
- Clarke, M.A., L.A. Edye, F. Cole and J. Kitchar, 1997, Sugarcane factory trials with dextranase enzyme, Sugar Journal, 60:20-22.
- Coll, E.E., Roberts, E.J., and M.A. Clarke, 1978, Deterioration of sugarcane: levels of dextran and total polysaccharides in process streams at two Louisiana sugar factories, Sugar Journal, 41:21-25
- Edye, L.A., F. Cole, M.A. Clarke and J. Kitchar, 1997, Trials on dextranase enzyme (from *Chaetomium gracile*) treatment of sugarcane syrups, presented at Sugar Industry Technologists Meeting, May 26-29,1997, Durban, South Africa
- GodshalLM.A.,B.L. Legendre,M.A. Clarke,X.M.Miranda, and R.S. Blanco, 1994, Starch, polysaccharide and proanthocyanidin in Louisiana sugarcane varieties, Int. Sugar Jnl, 98:144-148
- Hidi, P., J.S. Keniry, V.C. Mahoney and N.H. Paton, 1976, Observations on the occurrence and nature of polysaccharides in sugar cane, The Sugar Journal, 39:25-31
- Jmrie, F.K.E., and R.H. Tilbury, 1972, Polysaccharides in sugar cane and its products, Sugar Technology Reviews, !: 291-361
- Johnson, T.P., 1989, Experience with starch and dextran in a refinery, Presented at SIT Meeting, May 7-11,1989, New Orleans, LA.
- Madhu, G. L. Shukla and K. A. Prabhu, 1984. Application of dextranase in the removal of dexran from cane juice. IS J, 86:136-3 8.

# STRATEGIES FOR THE EXPANSION OF CANE SUGAR MILLS

Peter W Rein Audubon Sugar Institute Louisiana State University Baton Rouge, Louisiana

## ABSTRACT

Faced with the need to crush more cane, sugar mills have various options open to them. This paper explores the possible strategies for improving capacity. The first step in each case is to maximize throughput and performance of existing equipment. The best options for expanding each section of the plant, namely extraction, clarification, evaporation, pan house activities and steam and power generation are discussed. Installation of new equipment can be undertaken in a way to simplify plant and reduce operating costs, particularly if adequate forward planning is possible. Future options for making better use of capital assets are mentioned, including power islands for steam and power generation and improved cane quality.

# INTRODUCTION

Most raw sugar cane mills in the world today have been subject to expansion over the years. There has been a natural tendency for the average capacity of sugar mills to increase. This has largely been cost-driven, as mills have high fixed costs and the economics of milling operations are improved by economies of scale. Economic pressures on cane sugar enterprises have been increased by persistent low sugar prices, and there is no relief in sight.

Louisiana is typical of most sugar producing areas in that the number of sugar mills has steadily declined and the average crushing rate per mill has steadily increased. This process of consolidation is expected to continue. In addition, in the case of Louisiana specifically, new cane varieties have significantly increased the yield per unit area, putting additional pressure on milling capacities.

Larger size mills should always be lower cost producers than smaller mills. For mills of equivalent size however, the lower cost mills are those which have been expanded wisely, with due consideration being given to planning for efficient operation. Expansions provide the opportunity to reduce costs, by choosing to install the right equipment, and making use of larger items of process plant which reduce operating costs per ton of output. Good planning for future expansion will always pay off in the long term.

## FACTORS AFFECTING THE OVERALL CAPACITY OF THE MILL

The capacity of a mill can be defined in terms of its sugar output or its cane crushing rate. The former determines the revenue to the miller, and is therefore the capacity figure of most importance. However the cost of production is generally determined by the cane crushed, and the crushing rate is an important number. Profit is maximized if maximum sugar is made from the minimum amount of cane. This is achieved only if the system for payment of cane encourages the

grower to produce good quality cane.

Three elements need to be taken into account in assessing the capacity of a mill in relation to the cane that it has to crash:

The fiber throughput determines the size of the extraction plant needed to process the cane. The Brix throughput in mixed juice determines the size of the high-grade panhouse equipment required.

The non-sucrose throughput determines the size of the back-end equipment needed.

A factory short of capacity in the extraction plant or the back-end can find relief by incentivizing the growers to produce good quality cane. While payment for recoverable sugar in cane is far better than payment for just tons of cane or tons of pol in cane, and provides some of the right incentives to growers, additional incentives in relation to mill capacity could be cheaper than installing new plant, and would at the same time reduce milling costs significantly.

In most cases, there is a trade-off between capacity and performance. A drop in extraction due to lower residence time in a diffuser or reduced imbibition % cane due to evaporator constraints can be accepted instead of investing in additional equipment. Also, molasses purities can be allowed to rise, in the interests of processing higher quantities of low-grade massecuite, rather than invest in additional low-grade centrifugals. This however does not apply to all areas of the plant, specifically the high-grade pan station. If the high-grade pansecuite to be processed rise even further. This puts the miller into a vicious circle from which there is no escape other than to slow down the crushing rate.

Season length has a major impact on the high milling fixed costs. Where season length is restricted by weather conditions, there is little that the miller can do. Beet sugar factories prolong their campaigns by storing thick juice or syrup, for processing after beet processing has ended. This could be an option for cane mills, particularly where cogeneration is being practiced.

In most cases where the combined operation of growing and milling the cane is considered as a whole, the season length is often somewhat shorter than the economic optimum for the combined operation.

Good process control and instrumentation can play a part in improving capacity in each part of a factory. Good controls can always run a plant closer to its maximum throughput more consistently, and can obviate operational problems such as chokes and minimize other problems such as scaling.

## EXPANSION OF PLANT AND EQUIPMENT IN VARIOUS FACTORY AREAS

## **Cane Preparation**

There are no simple alternatives to uprating prime movers, or widening carriers or replacing preparation equipment. In some cases cascading of prime movers as part of an overall upgrading can be cost-efficient. There is generally a trend with both milling and diffusion to move to more intensive shredding and less knifing. This leads to a preparation which has longer fibers but very few "chunks" of cane. The longer fibers facilitate mill feeding and assist in the formation of an open bed in diffusers which promotes percolation. Indeed where only billeted cane is processed, as in Australia, only shredders and no knifing are used. An expansion is a good time to change to this approach; belt conveyors replace apron carriers, costs decrease and improved extraction is achieved.

# Extraction

Small increases in capacity can be obtained from a milling tandem by fitting pressure feeders or by replacing critical milling units with larger units, say the first and the last mill, but the increase obtainable by this means is limited. A cost-effective method of expansion involves installing a diffuser and utilising the best of the existing milling units as dewatering mills for a diffuser. Rivalland (1984) has confirmed the effectiveness of this approach from a capital cost point of view.

With this approach, the better milling units are kept and the old ones are scrapped. If the mills are arranged in parallel, fiber load on each can be significantly reduced. Parallel operation is most easily achieved by choke-feeding the first, and allowing the overflow to cascade over this mill into a conventional intercarrier feeding the second mill. Bagasse from the first de-watering mill is taken out sideways on a small belt conveyor.

It should be noted that a diffuser does not need any more imbibition than a milling tandem i.e. there is no need to increase evaporator capacity because a diffuser is to be installed.

Once a diffuser is installed and further expansion is required, the installation of a complete additional diffuser can be expensive. Alternatively, a reduction in extraction as a result of reduced cane residence time is an option. However if it is envisaged that an expansion will be required at some time in the future, it is probably wise to pre-invest in incorporating headshaft and chain designs which can operate with an expanded diffuser. It is relatively cheap to increase the length of a diffuser to obtain additional capacity in this way.

## Clarification

In most cases, the clarification station can be uprated by modification rather than by installing new units. Scott (1988) quotes the example of a mill installed to crush 225 metric tons cane/hr with four 24" Rapidorr clarifiers being able to crush at 385 metric tons cane/hr using only two of the clarifiers after modification.

Studies reported on the application of computational fluid dynamics studies to clarifiers (Steindl et al, 1998) are likely to show that capacity of both short retention time and conventional clarifiers can be significantly improved with only minor modifications.

# Filtration

Overloading of filters is often due to excessive soil in cane. Incentives in the form of a penalty / bonus scheme applied to cane deliveries help to avoid the installation of additional filters as well as to reduce costs in other areas - surely something to be considered by millers.

An advantage provided by diffusion is the fact that the filter requirement is only half of that required for a milling factory. The installation of a diffuser generally leads to surplus filter capacity.

Following the lead at Maidstone mill (Meadows et al., 1998), a number of mills now return clarifier muds directly to the diffuser, thus eliminating a filter station altogether. The process scheme is shown in Figure 1. No adverse effect on extraction or juice quality has been observed.

Bagacillo supply to filters can be easily augmented by sucking bagacillo from bagasse conveyor transfer points. At Triangle mill in Zimbabwe, the existing bagacillo system was shut down completely, as more than sufficient bagacillo, of excellent quality, could be obtained in this way. This approach has the added advantage of significantly reducing the fail out of bagacillo dust around the **mill**.

## Evaporation

Very few mills are in a position to measure heat transfer coefficients on a routine basis, to ensure that all vessels are working to their maximum capacity, or that cleaning has been effective. An easier alternative which can assist in this objective monitors intermediate juice and steam temperatures, and draws conclusions from changes in temperature profiles (Jones and Pozzetti 2000). Ideally, with good process control and adequate instrumentation, this process could be taken further, incorporating an on-line evaporator balance to calculate vessel heat transfer coefficients on a continuous basis, to highlight vessels which are under-performing for special attention.

Most mills have inherited a collection of different vessels, often leading to awkward operating arrangements. An expansion should be used as an opportunity to rationalize the arrangement, rather than add further complication. The obj ective must be to end up with fewer larger evaporator vessels. If very large vessels are required, long tube climbing film evaporators (referred to as Kestner evaporators) are more cost effective (Rein and Love, 1995). These large vessels are generally placed at the front end of the evaporators, particularly if vapor bleeding is significant. In this position, Kestners have the additional very important advantage of low residence time. Since it use Kestners in this position. With new Kestners in the first effect position, the remaining evaporators should if possible be configured to match the optimum heating surface arrangement

It is not widely appreciated that a tail of evaporators all the same size is a particularly inefficient way of utilizing heating surface. While it is convenient from a manufacturing point of view to make all the vessels the same size, the use of the optimum arrangement of the same heating surface can increase evaporation by almost 10%. This is illustrated by the results of some evaporator simulations shown in Table 1. Two cases are considered, the first with Vapor 1 bleeding and a large first effect, and the second where no vapor bleeding is practiced. In the two cases, evaporator area can be reduced by 7 to 9 %, to get the same evaporation rate, if the area is optimally distributed. Alternatively, the same aoptimally distributed can lead to an increase in throughput of 4.1% or 8.6% in the two cases.

	Case 1: With vapor bleeding			Case 2: No vapor bleeding		
Juice flow (mt/hr)	509	530	509	360	391	360
Vapor bleed (mt/hr)	156	156	156	0	0	00
Areas (m <sup>2</sup> )						
1 <sup>st</sup> effect	6000	6000	6000	2560	1880	1730
2 <sup>nd</sup> effect	2400	1600	1580	2560	2000	1820
3rd effect	2400	2200	2040	2560	2560	2340
4 <sup>th</sup> effect	2400	3400	3090	2560	3800	3530
Total						
2nd-4th effects	7200	7200	6710	10240	10240	9420
Area reduction (%)			7.3			8.7
Flow increase (%)		4.1 8.6				

Table 1. Results of evaporator simulations	, based on he	eating steam to	o the first	effect at	210 kPa
(15.8 psig) and final effect absolute pressure	e of 16.6 kPa	(4.9" Hg),			

It is unlikely in practice that all vessels will be installed with different heating surfaces. In Case 2, a possible practical compromise would be to install 5 vessels, each of the same size, with the last 2 in parallel. Thus 5 vessels of 2048  $m^2$  each give a 6.9% increase in flow over the case of 4 equal size vessels of the same total area.

An additional disadvantage of an arrangement with equal sized vessels is the fact that the last vessel operates with a very large pressure difference between calandria and vapor space. This often results in large vapor volumes generated in evaporator bodies that are too small, leading to excessive liquid entrainment losses.

In cases where cleaning on the run is necessary, additional flexibility and cost needs to be built in to allow cleaning with minimal disruption to crashing rate. This may lead to non-optimal arrangements of vessel heating surfaces.

Falling film tubular evaporators are widely used in the beet sugar industry, but have not found wide use in cane sugar factories. They provide the same advantages as the Kestner in terms of low first effect residence time, but have been found to have disappointing heat transfer capability (Rousseau et al 1995).

Rising film plate evaporators have been used with advantage as "add-on" capacity, but their use to replace Robert evaporators has been disappointing, and fraught with problems (de Beer and Moult 1997). Falling film plate evaporators appear to have more promise in the future (Grant et al 2000), particularly as they may be used to replace old calandrias in existing vessels, providing a cheap method of increasing capacity with minimal disruption.

# Pan Boiling

Improvements can be made to the performance and capacity of batch pans by improving circulation, particularly in the case of high head or flared pans. Either the installation of circulators or the use of circulation-assisting direct injection of steam (often called jigger steam) can increase boiling rates and enable the full pan design volume to be used. In most raw sugar mills, the use of vapor bled from the evaporators is to be preferred, as it is simpler and cheaper and is easily accommodated in the factory steam balance at no additional fuel cost. A neat alternative is to bleed incondensibles from the calandria through the massecuite as jigger steam.

A common philosophy employed when a marginal expansion is required has been to install a continuous C pan, and cascade batch C pans on to B or A duty. Subsequently, when some of the older and smaller batch pans become unserviceable, a continuous B or a continuous A pan can be installed. This results in an increased capacity pan floor with fewer pans and less complication.

A comprehensive expansion plan incorporating continuous pans for Triangle mill (Zimbabwe) involving more than doubling capacity is outlined in Table 2 (Rein and Msimanga 1999). Progressive installation of continuous pans allows older small batch pans to be taken out of

Table 2. Pan floor expansion strategy for Triangle Sugar Mill showing number and sizes (in m<sup>3</sup>) of pans required.

Crushing rate (mtoh	)	_350	430	550	640	740
Batch pans A	А	3x56 1x85	3x56 1x85 2x30	2x56 1x85	3x56 1x85	2x56
	B C	3x30 2x30	1x30 2x30	1x56 4x30	2x30 2x30	1x56 1x85
Continuous pans	A B C	- - 1x86	- 1x120 1x86	1x160 1x120 1x86	1x160 1x168* 2x86	2x160 1x168 2x86
Total number of	pans	10	11	11	12	9

\*120m3 pan expanded to 168m3 by increasing from 10 to 14 compartments

service, ultimately ending up with fewer pans in the expanded case than the initial installation. Some batch pan capacity is maintained for seed production. Triangle is presently in the third phase of this expansion.

The general strategy of replacement of small pans with larger ones is available for batch pan options as well. However, there is a limit to batch pan size. In most cases, the largest batch pans are 85m<sup>3</sup> (3000ft<sup>3</sup>), and although larger sizes are in operation elsewhere in the world, they generally do not approach the size of large continuous pans now in operation. The effective capacity of a continuous pan is of the order of 40-50% higher than an equivalent batch pan, since the pan is always full, and since no time is lost on striking and filling, a 180m<sup>3</sup> continuous pan equates in terms of capacity to a batch pan of about 290m<sup>3</sup>.

Since continuous pans do not have the peak loads experienced with batch pans, pan ancillaries such as vacuum pumps and injection water systems generally do not have to be uprated when a larger continuous pan is installed. The use of a double magma (or double Einwurf) system is particularly well suited to continuous pans. Then only C seed has to be produced in a batch pan.

#### Crystallisers

Vertical crystallisers are finding increasing use, as they are generally cheaper to install for the same capacity. In particular they are sited on the ground without need for supporting steelwork. However care needs to be taken to ensure that the configuration of crystallisers and the design of internals promote plug flow with no chanelling and sufficient shear to get good cooling performance.

## Boilers

Boshoff (1995) has outlined modifications to boilers which can be considered in order to increase output or efficiency. In most cases these are small, but are worth considering. If a boiler is relocated however a considerable increase in output can be achieved at virtually no extra cost. Examples are given of increase in output of between 10 and 30%, byre-designing the air and gas flow systems, and/or feeders, spreaders and generating or superheater banks. Increases in efficiency can also be achieved at the same time, particularly with modifications to air heaters and economizers.

It is often possible to improve the steam economy and reduce the steam required when a mill is expanded or refurbished. It may be cheaper and better to spend money in the factory to achieve this than to spend a large amount of capital on the boiler plant. This could lead to bagasse surpluses, which would be acceptable in some circumstances but not in others.

#### **Power Generation**

A simple expedient as an alternative to installing additional generator capacity is to buy in more power. The economics of purchase vs own generation of power need to be looked at in each individual case.

In the case of diffusion mills, the letdown of steam to the exhaust steam range is often quite considerable, since less prime mover power is required. This facilitates the route to cogeneration of power for sale to other users or utility companies. Cogeneration as a favorable option is promoted in cases of long crushing season and high alternative fuel costs.

Cogeneration of power from renewable resources is becoming a well-regarded activity from an environmental point of view, as it reduces greenhouse gas emissions relative to power generation from fossil fuels. This is encouraged in some areas by paying a premium for power generated from renewable resources. In Australia for instance, all electricity retailers will have to generate 2% of their power from renewable resources by the year 2010 (Dixon and Burbidge 2000). This has resulted in the first announcement of a large co-generation plant, at Rocky Point mill, where the utility company will invest in new plant to be able to generate 30 MW of "green" power.

The concept of "power islands", where an outside organization owns and operates the steam and power generation plant, is likely to become more popular. The mill owner may experience some discomfort in being dependent on an outsider for these vital services, but the reduction in capital
required on expansion should be very attractive when expansions involve expensive steam and power raising plant. This arrangement also facilitates on-going activities during off-crop such as thick juice or syrup processing, ethanol distillery operation or off-crop white-end refining. A system of greenhouse gas emissions credits which can be traded internationally, like any other commodity, would be a further driver to adopt large-scale cogeneration.

## CONCLUSIONS

Good planning of future expansion is important if low-cost milling operations are to be achieved. Options for expansion are wider than in the past, but the trend is to incorporate larger process units that provide reductions in capital costs, simpler plant to operate and improved plant efficiencies. Strategies to improve cane quality and increase the effective length of milling seasons may prove more advantageous in increasing effective capacity.

#### REFERENCES

- Boshoff, T.L. 1995. Improving older sugar mill boilers. Proc. S. Afr. Sugar Technol. Assoc. 69: 122-124.
- De Beer, T. and J. Moult. 1998. Experiences with plate evaporators at Ubombo Ranches. Proc. S. Afr. Sugar Technol. Assoc. 72:
- Dixon, T.F. and D.Burbidge. 2000. The future for cogeneration in the sugar industry. Proc. Aust. Soc. Sugar Cane Technol. 22:38-45.
- Grant G., B. Morgenroth, and K. Niepoth. 2000. The Balcke-Duerr falling film plate evaporator - an innovative concept for evaporation in the cane industry. Proc. Aust. Soc. Sugar Cane Technol. 22:355-360.
- Jones, A. and C. Pozzetti. 2000. Evaporator performance monitoring at Farleigh mill. Proc. Aust. Soc. Sugar Cane Technol. 22:334-340.
- Meadows, D. M., G. T. Schumann,and C. Soji, 1998. Farewell to filters: the recycle of clarifier mud to the diffuser. Proc. S. Afr. Sugar Technol. Assoc. 72: 198-203.
- Rivalland, J.F.R. 1984. Milling and diffusion in Mauritius. Rev. Agr. Sucr. Ile Maurice 63: 22-30.
- Rein, P.W. and D. J. Love. 1995. Experiences with long tube climbing film evaporators. Proc. Int. Soc. Sugar Cane Technol, 22: 251-259.
- Rein, P.W. and M. P. Msimanga. 1999. A review of continuous pan development in the Southern African sugar industry. Proc. Int. Soc. Sugar Cane Technol. 23: 124-136.

- Rousseau, E., L. Sifunda, and J. R. Fitzgerald. 1995. Practical experiences operating a first effect falling film evaporator unit at Pongola. Proc. S. Aft. Sugar Technol. Assoc. 69: 127-131.
- Scott, R.P. 1988. Modifications to and experiences with Rapidorr clarifiers including saccharate liming at Amatikulu. Proc. S. Afr. Sugar Technol. Assoc. 62: 32-35.
- Steindl, R.J., A. L. Fitzmaurice, and C. W. Alman. 1998. Recent developments in clarifier design. Proc. Aust Soc. Sugar Cane Technol. 20:477-483.



Figure 1. Schematic diagram of clarifier mud recycle to a diffuser.

## A FLEXIBLE COUPLING FOR SUGARCANE MILLS - ITS DESIGN CONCEPTION AND PERFORMANCE

Luiz **Carlos Felicio** Unip-Universidade Paulista - Campus Ribeirao Preto Simisa - Simioni Metalurgica LTDA P.O.Box 641, CEP 14160-000, Sertaozinho, SP, Brazil Icfelicio@hotmail.com

# ABSTRACT

Traditional mill couplings frequently cause serious problems to the mill driver components. Shaft cracks, flattened points of the shaft square surfaces and bearing deformations causing damages to the gears are the most usual troubles. Measurements of these coupling effects under actual operation condition show a significant bending moment (same magnitude of the torque) applied on the shaft square tips and an oscillatory bending stress having amplitude of 105 Mpa. In order to minimize those problems a new coupling was developed based on a mechanism similar to a crank and arm. The coupling uses four crank-arm arrangements such that a pair of cranks (like a fork) works with a pair of arms (like a "T") on each shaft. This configuration allows shafts and tail-bar to have lateral, axial and angular motions that cause a reduction in axial and bending moment loads.

This work analyzes the effects of the traditional coupling, explains the design conception of the new coupling and discusses its performance based on measured data. The most important results are a 78% reduction in the bending stress (from 105 MPa to 23 MPa) and a decrease in the bending moment to the level of 25% of the driving torques.

# INTRODUCTION

The common coupling widely used in sugar factories can be considered as a semi-rigid type. Those couplings, Figure 1, make the mechanical connection using rigid elements like rigid couplings. The semi-rigidity is achieved through the clearances that exist between the parts. Those semi-rigid couplings have the advantage of being simple and having a relatively initial low cost; however, they constantly cause serious problems to the mill components.







Figure 2. Common coupling forces on the shaft.

As shown in Figure 1, the spatial arrangement of the contact points that transmit the torque is not symmetrical. That force configuration, Figure 2, causes two undesirable effects:

1 - High compression pressures because of the applied forces on small areas; and

2 - High values of the bending moment  $\{M_B\}$ , Figure 2, due to the fact that the forces which produce the torque do not lie on the same cross section plane.

The consequences of these two effects are as follows:

a) Damages caused by the high compression pressure:

The high stress concentration yields the material by compression (Figure3) (Felicio and Cordeiro, 1982). The parts never work with a large contact area and those small regions end up with permanent deformations. The plastic deformation makes the coupling working condition even worse since that instantaneous accommodation creates some kind of constrain between the parts. Moreover, with the shaft rotation and lifts, the contact points change their places and that fact implies in new stress concentrations with new deformations that will keep continuously occurring up to the premature shaft failure.







Figure 3: Crushed Figure 4: Repairing a Figure 5: Failure by material. fissured shaft. rupture.

b) Damages caused by the bending moment:

The two important damages directly caused by the bending moment are the high bending stresses and the high vertical forces on the bearing.

b.1) The high bending stresses caused by the bending moment:

The stresses caused by the bending moment have a cyclic behavior and that means fatigue problems. After a short period of time cracks appear and a repairing process is required (Figure 4), or they will propagate up to the point of rupture (Figure 5).

b.2) High vertical forces on the bearing:

The high vertical forces on the bearing cause the following effects:

b.2.1- Premature wear of the bearings (with overheating tendency): Large forces on the bearings increase the wear.

b.2.2- Bearing vertical displacement:

The vertical displacement causes serious problems to the gears. When the bearing vertical

displacement happens, the gears stop having uniform tooth contact and only a fraction of the teeth start working. Since the loads are high, the pitting phenomenon takes place and that problem is even worse for the pinion.

The effects and damages caused by the common coupling are shown in Figure 6.



Figure 6: Diagram of the effects and damages caused by the common coupling.

Table 1. Summar	y of the ex	perimental	evaluation of	f the	common cou	pling	g effects.

Parameters	Values
Ratio: (Bending Moment)/(Drive Torque)	100%
Amplitude of the cyclic bending stress	105 MPa (15.2 kpsi)
Vertical force on the bearing	70 tf (154.3 klbf)

In the 80's decade these coupling effects were measured (Felicio and Miayesi, 1985; D'elia and Mendes, 1986; D'elia and Mendes, 1987) and a summary of the results is shown in Table 1.

## DESIGN CONCEPTION

In order to minimize the inconvenient effects of the common coupling a new device was made up and its design conception is described below.

The new coupling was developed based on a mechanism similar to a crank and an arm (Felicio, 1991), Figure 7, where:  $\{1\}$  = the first shaft;  $\{2\}$  = the second shaft;  $\{3\}$  = the arm;  $\{4\}$  = the crank;  $\{RI\}$  = the crank radius;  $\{R2\}$  = the contact radius; and  $\{P\}$  = the contact point. If the shafts  $\{1\}$  and  $\{2\}$  have a relative axial, angular or lateral motion, or any combination of them, the power transmission continues taking place. Under this misalignment condition the transmission ratio

varies because the contact radius {R2} changes and does not remains equal to the crank radius {R1}, as the shaft turns.

The axial, angular and lateral freedom makes this device interesting, however it has three problems which are the high bending stresses in the shafts {1} and {2}, the oscillatory transmission ratio, and the stress concentration at point {P},





Figure 7: Crank-arm system

Figure 8: Duplicated crank-arm system.

In order to eliminate the bending moment on the shafts, the coupling of Figure 7 was modified to have a duplicated crank-arm system as shown in Figure 8, where:  $\{1\}$  = the first shaft;  $\{2\}$  = the second shaft;  $\{3\}$  = the double arm (like a "T");  $\{4\}$  = the double crank (like a fork); and  $\{C1\}$  = the arm center point.

The new configuration has the advantage of eliminating the shaft bending stress, but it does not allow a lateral misalignment. That is, the arm center point  $\{C1\}$  has to stay on the axial centerline of the shaft  $\{1\}$  (Figure 8). The lateral misalignment can be recovered using two sets of duplicated crank-arm systems (Figure 9), where:  $\{1\}$  = the first shaft;  $\{2\}$  = the second shaft;  $\{3\}$ = the tail-bar; and  $\{C1\}$  and  $\{C2\}$  = the arm center points.



Figure 9: Two sets of duplicated crank-arm systems



Figure 10. Centered arm with axial and angular freedom.

This configuration allows the axial, angular and lateral motions of the shafts. Moreover, if the shafts are parallel, as in Figure 9, the transmission ratio is constant since the tangential speed of the point {D} is equal to the tangential speed of the point {E}. Considering that the shaft tilting is usually very small, the transmission ratio between the shafts {1} and {2} remains practically constant.

Under the point of view of constructing the device, the axial and angular freedom is obtained making a spherical surface of the arm work inside a cylindrical surface of the crank (Felicio, 1991), Figure 10. The point {C1} is always on the crank centerline, but the arm has axial and angular freedom.

Concerning solving the problem of the stress concentration on the point {P}in Figure 7 (or on points {D} or {E} in Figure 9), two semi-cylinders with their axes at 90° are placed at each contact point (Figure 11). Considering only one pair of semi-cylinders, one of them is mounted on the crank and the other on the arm. If for any reason there exists a punctual contact, the semi-cylinders will move in such way that their flat surface will get full contact.



Figure 11: The angle between the two semi-cylinder axes is 90°.



Figure 12: The new square profile increases the contact area.

In addition to all the details described above which aim to minimize the bending moment, the new coupling should also avoid crushing the shaft surfaces (see Figure 3). This property is attained by means of choosing an adequate dimension for the square holes which allows an easy assembly operation and prevents flattening points of the shaft and tail-bar. Besides, a new square profile (Figure 12) increases the contact area by a factor of four; consequently, the stress concentration is eliminated.

Figure 13 shows the first coupling built under working conditions. That coupling was mounted in Usina Sao Geraldo, Sertaozinho, S.P., Brazil, in 1997. As can be seen in Figure 13, one of the sets is mounted at the mill side and the other at the gear box side, following the sketch of Figure 9. The cranks are assembled on the shafts and the arms on the tail-bar. The mounting operation is performed in the same way as for the common coupling; i.e., after getting the two sets

on the tail-bar, they are translated to the shafts, but the arms remain on the tail-bar (see Figure 13).



Figure 13. General view of the first flexible coupling working in a sugar factory.

# THE FLEXIBLE COUPLING PERFORMANCE

The evaluation of the flexible coupling performance was done using two requisites: the observation of the damage on the shaft square surfaces and the values of the bending stress.

a) The observation of the damage on the shaft square surfaces:

Two new flexible couplings to drive two mills were installed in Usina Aralco (Aracatuba, SP, Brazil) in 1998. They were mounted on new shafts; therefore, all the surfaces began working in perfect condition. After the work done in 1998 and also in 1999 the shafts did not show any crushed point at all. Therefore, the proposed new square profile, Figure 12, attained the goal of avoiding crushing the shaft square surfaces.

b) The values of the bending stress :

Table 2 shows a comparison between the values of the main effects caused by the common coupling and those caused by the flexible coupling (Gago and Godoy, 2000). The flexible coupling performance resulted in a 78% reduction in the bending stress and a decrease in the bending moment to the level of 25% of the driving torque.

With respect to the gears, the smaller the bending moment, the smaller will be the bearing vertical displacement and the gear disturbing effects. Therefore, having smaller bending moment, the gearing problems should be minimized.

 Table 2. Summary of the experimental evaluation of the effects of the common coupling and the flexible coupling.

Parameters	Common coupling	Flexible coupling
Ratio: (Bending Moment)/(Drive Torque)	100%	25%
Amplitude of the cyclic bending stre	ess 105 MPa (15.2	kpsi) 23 Mpa (3.3 kpsi)

## CONCLUSION

The flexible coupling was built to transmit high torque and to allow axial, angular and lateral misalignment of the shafts, without damaging the parts. These features resulted in an extreme reduction in crashing, axial and bending loads, which are undesirable and harmful for all mill and gear box components,

### REFERENCES

- Felicio, L.C. and I. C. Cordeiro. 1982. Medidas de torque, rotacao e potencia de moendas de cana de acucar. Ref. CDF-RT-048/82, Institute de Pesquisas Tecnologicas do Est. de Sao Paulo, Brazil.
- Felicio, L.C. and M. Miayesi. 1985. Medidas de tensao e torque do eixo superior da 1º. terno de um conjunto de moenda 37" x 78". RT-190-85/86, Copersucar, S.P., Brazil.
- D'elia, M.A.G. and F. B. G. Mendes. 1986. Medicao de tensao nos tirantes do mancal da volandeira. Ref. CDF-RT-010/86, Instituto de Pesquisas Tecnologicas do Est. de Sao Paulo, Brazil.
- D'elia, M.A.G. and F. B. G. Mendes. 1987. Medicao de diversas grandezas fisicas no eixo superior do 1°. terno de conjunto de moenda 37"x 78". Ref. CDF-RT-024/87, Instituto de Pesquisas Tecnologicas do Est. de Sao Paulo, Brazil.
- 5. Felicio, L.C. 1991. Carta patente no. MU-7102328-3, Brazil.
- Gago, J.S.N.and D. R.A. Godoy. 2000. Medicoes de torque e fexao em acionamento de moenda 84" com acoplamento do tipo "de castanhas", fabricacao Simisa - Usina Sao Martinho 1999. RT-1058-00/01, Copersucar, Brazil.

### CANE FACTORY PROCESS MODELING USING SUGARS™ FOR WINDOWS®

L. Warner Weiss Sugars International LLC 30 Glenmoor Drive Englewood, Colorado **80110** USA <u>sugars@sugarsintl.com</u> www.sugarsintl.com/sugars

# ABSTRACT

The Sugars<sup>™</sup> computer program is used extensively for sugar process modeling and simulation to make improvements to existing factories, or design new factories. The new Windows version of Sugars features a full graphical interface that uses drag-and-drop techniques to draw the flow diagram and build a model of the process. It is fast, flexible and very user-friendly. Stencils containing shapes of stations are provided with the program and these shapes are used to draw the flow diagram. Connections are made between shapes using a connector tool with automatic line routing and crossovers. Data for each station and flow stream in the model is entered on dialog screens that are displayed by double clicking on the station shape, or flow stream. All of the data for a model is stored in a Microsoft® Access database that can be addressed by other programs, Heat, material and color balances are quickly obtained from simulations of the model to predict the performance results for the process. A revenue screen shows the net process revenues generated by the process to assist with financial decisions, The new Sugars for Windows computer program is a major upgrade to assist cane sugar factory and refinery process engineers and management with decisions for making improvements to their operations.

## INTRODUCTION

The Sugars<sup>TM</sup> Computer Program is used to model sugar factories and refineries, and it has successfully modeled thousands of sugar processes since it was first introduced in 1986. It is a reliable program that is used by sugar companies and engineering firms to improve existing factories, or to design new ones. Sugars is the most widely used program in the world for modeling and simulating sugar processes.

Originally, the program was designed with a text-based interface for IBM compatible personal computers using the DOS operating system. A flow diagram was made of the process model for simulation by using a separate CAD, or diagramming program. The flow diagram was used as a reference to build text files with a text editor to describe the model for simulation by Sugars. This method worked well for users that were familiar with personal computers, and simulations from Sugars were found to be an accurate representation of the process. However, modern Windows® based software provides opportunities for dramatic improvements in the user interface and tools were developed that made it possible to integrate the drawing and model building steps.

In August of 1997, Visio Corporation released version 5.0 of their diagramming software. The Visio® program was known to be an excellent program for making process flow diagrams; however, it lacked some of the necessary features for integration with Sugars until the release of version 5. Once it was determined that this new release had the necessary functionality, development work was directed to a new Windows version of Sugars that would use Visio to give Sugars a robust graphical interface. Early versions of the integrated programs were shown last year at the SIT meeting in Marseille, France and at the ICUMS A meeting in Berlin, Germany. A beta test version of the new program was released in December 1998 to some sugar companies in the USA and the first showing of the complete program was made at the ASSET meeting on February 11, 1999 in Orlando, Florida. The new Sugars for Windows program is an exciting change. Now, using Sugars, it is a simple procedure to build a model and obtain material, energy and color balances that can be used to evaluate a process.

#### NEW USER INTERFACE

The new graphical interface for Sugars makes model building a simple matter of dragging pre-drawn shapes from a Shapes Stencil to the drawing surface. The shapes are designed to represent stations for modeling the process by Sugars. Connections between the shapes are drawn using a connection tool that automatically routes connections between associated stations. Data-entry dialogs for controlling the performance of each station are displayed by double clicking on the station shape. Flow-

stream properties are entered and displayed on a dialog screen by double clicking on the flow stream. Sugars for Windows is now a fully functioning Windows 95/98/NT program that gives all of the benefits to a user that are available in the Windows environment; such as, drop-down menus, online help, print preview, scrolling, object design and database storage. It is a dramatic change for users of Sugars and it makes the program much easier to learn and use while still providing all of the flexibility needed for modeling sugar processes.

The new interface is shown in Figure 1. Stencils on the left contain shapes of stations that are used to build a model. Different stencils are selected by clicking on the gray bar with the name of the stencil. Shapes on a stencil are dragged from the stencil to the drawing surface and dropped in place. Drawing tools are available to design shapes of factory equipment and add them to the stencils as needed. Connections between stations are drawn using a connection tool that features automatic routing around shapes and crossovers (line jumps) for crossing connection lines. Drawings can be composed of different layers and pages. Full zoom-in, zoom-out and panning features are provided in the interface. The menu selections at the top of the window include a Sugars menu entry that gives a drop-down menu containing selections for:

Model properties	- Overall properties of the model such as molecular weights for										
	flow stream components, colors, iteration accuracy and										
	atmospheric pressure are set on this screen.										
Default units	- Units used for all entries and when a model is saved.										
Synchronize database	- Check/correct synchronization between drawing and database.										
Reports	- Selection of reports for print preview and printout.										
Balance	- Model iteration balance calculations (single, or full).										

Figure 2 shows the Model Properties screen when it is selected from the Sugars drop-down menu. Also shown in Figure 2 is the Sugars toolbar. The Sugars toolbar provides icons for

(numbered from left-to-right); (1) Balance - full, (2) Balance - single step, (3) Display last error or warning messages, (4) Summary report, (5) Detail report and (6) Revenues report. The tool bar icons provide a quick method for selecting items that appear on the Sugars menu without having to go to the menu to make the selections. Item (1) Balance - full, gives a full iteration balance of the model. If any errors occur during the balance calculations, they will be displayed on an error, or warning, message screen. Sometimes revisions to the design of the model, or data controlling the performance of stations in the model, maybe needed to bring the model into balance. The last error, or warning, message screen can be referenced as corrections are made to the model. Item (2) Balance - single step, can be used to see how the model changes between iterations.

At the bottom of the screen, in the lower left-hand comer, a small box indicates whether the model is in, or out of, balance. When the box is green (as shown in the figure above), the model is in balance. When the box is red, the model is unbalanced. Any changes to a model will cause the balance indicator box to change to red for a model that was in balance (box was green).

Figure 3 shows the Units Selection screen for selecting the system of units to be used with Sugars for entering data and displaying the results from the balance calculations. The default system of units is used when a model is saved, even if the displayed units are different. Units are available for SI, US with temperature in °C and US with temperature in °F systems.

As shown in Figure 3, the stencils on the left contain shapes of stations that can be used to build a model of a sugar factory, or refinery. Each of the station types in Sugars is represented on the stencils and many of the stations have more than one shape that can be used for the process flow diagram of a model. Also, other unique shapes can be drawn and added to the stencils, if needed. There is no limit to the number of different shapes that can be used to represent the Sugars station types.

Complete online help systems are available from the main menu for both Visio and Sugars as shown in Figure 4. Help for Visio describes all of the drawing features available in Visio, and help for Sugars describes each of the stations, input properties, examples and features of each station. Also, the Sugars help system describes how to design shapes so that they will work with Sugars and other features of the program. The help system includes features such as a table of contents, index and full search for key words.

The input and output connections for the shapes stay glued to the shapes even when the shapes are moved, or the flow streams are rerouted. Stations can be copied from one part of the flow diagram and pasted into another part with the data for the station repeated in the copied station. Also, groups of stations maybe combined and placed on the drawing as a group instead of having to add each station individually (station numbers for each station are entered when the group is dropped on the drawing).

# MODEL BUILDING

Every model of a factory, refinery, or portion of a process is built by selecting stations from the available station modules in Sugars and connecting the stations together using flow streams. Flow streams between stations, and flow streams that leave a model, are called *internal flows*. Flow

streams that go into the model from outside sources (e.g., beets, steam, water, milk of lime, etc.) are called *external flows*. All external flows must be fully specified before a balance can be done. Internal flows are calculated by Sugars during the balance calculations.

Sugars has sixteen different stations that can be used to simulate a process. The sixteen stations are: centrifugal (batch and continuous), heater (or heat exchanger), incitier, pan, distributor, dryer, crystallizer, evaporator body, tank, receiver, flash tank, cooler, separator/filter, blender (also used to model a contact condenser), reactor, and compressor/pump. When a shape is draged from a stencil to the drawing surface, a dialog screen appears to assign a number to the station (Figure 5).

The station number for each station in a model must be unique. Sugars will automatically index the station numbers by 10 as each new one is added to the flow diagram; however, the number may be changed on the dialog screen to any unused number in the model.

Each Sugars station type has a data input screen (dialog) that is associated with the shapes for that type of station. The data input screen defines the properties of each station (object) in a model. Drawing the process flow diagram defines the model. The output flows emitted from a station are a result of the properties of the station. Changing the properties of a station will control how the station processes flows coming into the station. For example, an evaporator station, with juice and steam input flows, will process the juice flow to evaporate water while using steam, or vapor, to boil the juice. Thus, the properties of the stations in a model and the characteristics of the external flows into the model control all of the internal flows between stations, and the flows leaving the model. Each station type has a properties dialog screen for entering the appropriate data, and a dialog screen is provided for entering the data to define external flows into the model.

Shapes on the stencils are associated with their respective Sugars stations by naming the shapes in Visio with station names from Sugars and identifying the input and output ports for the stations. The Sugars help system provides full details on how to make shapes that can be identified by Sugars.

The shape color turns to blue after the station number is entered. Double click the shape (or right click and select "Sugars properties" from the drop-down menu - see Figure 6) to get the data entry screen (Evaporator Properties screen in Figure 7).

All of the necessary data to describe the performance of the station is entered on the Properties screen. The station turns yellow to indicate that data for the station was entered after the Properties screen is closed by clicking on the OK button. Shapes are added to the drawing until all of the stations for the model are on the drawing and data is entered for each station until all of the stations on the drawing are yellow.

Connections are made between input and output ports on each station using the connection tool until all of the ports on the stations have connecting flows. Some stations, such as distributors, melters, tanks and receivers, do not need to have all of their ports connected (extra ports are provided), but these stations must have at least one input and output flow.

Data for external flows into the model is entered on the External Flow Properties screen. The External Flows Properties screen (Figure 8) is accessed by double clicking on the external flow, or right clicking and selecting "Sugars properties" from the drop-down menu. Every external flow stream going into the model must be specified with pressure, temperature and flow stream components. If the flow stream contains dissolved non-sucrose components, the solubility coefficients must be defined. And, if the flow is not a required flow, the quantity of the flow stream must be specified. *Required* flows are flows that have their quantity determined by the properties of a station. For example, the vapor flow into a pan is a required flow because the quantity of the vapor needed by the pan is determined by the properties of the pan. A flow stream color entry must be made for the external flow if the color balance is being done. And, a currency value is entered if the net process revenues are being calculated for the model.

#### SIMULATION

The model is ready for balancing once the data for every station in the model and every external flow stream into the model is entered. The balance calculations give a simulation of the process and provide all of the details of the internal flow streams in the model.

Clicking on the fall balance icon will initiate the balance calculations and give a small window showing the progress of the iterations (Figure 9). Single balance iterations can be done (to observe the progress of the balance) by clicking on the single balance icon. An error message screen will appear if any errors are detected during the iterations. Fatal errors have to be corrected before the calculations can be completed. Sometimes warning, or informative, messages appear giving information about the calculations, or observations about the model.

Many of these messages do not require any action by the user, but are merely displayed as information that might be important, or helpful to the user. The error/warning message screen may be recalled at any time after the balance calculations so that it can be referenced as corrections are made to remove any errors that occurred during the balance calculations. Clicking on the error message screen icon will recall the display of the last error messages that occurred during a balance.

## RESULTS

The results of the calculations can be displayed after the balance calculations are completed. Sugars provides several different presentations of the results.

Figure 10 shows the display of the Internal Flow Properties screen after a balance. This screen shows all of the details of an internal flow, such as pressure, temperature, flow quantity, components, color and solubility coefficients. In addition, the % total dry matter (TDM), % sugar, % dry substance (DS), % purity, % crystals, % insoluble solid non-sugars (ISNS) and % gas are given in the middle of the screen with the quantity of flow in tons per hour. Clicking the DS and Purity button, under the Flow Stream Components, in the Liquid phase section, will give the % dry substance and purity for the liquid portion of the flow. For a massecuite, this would be the mother liquor %DS and % Purity.

Clicking the Parameters button on the Internal Flow Properties screen gives additional characteristics of the flow, such as: volume flow, specific weight, enthalpy, specific heat capacity, supersaturation and boiling point elevation (Figure 11). The Flow Stream Parameters feature is also available for external flows.

Every internal flow in the model can be displayed using the Properties screen. Also, flow streams that are required and/or pressure feedback (flow streams that need a pressure value from another station) are identified on the flow diagram by a small 'R' for required and a small 'P' for pressure feedback. Sugars will place these indicators on the flow streams after the balance calculations are completed.

The Internal Flow Properties screen has a small internal flow selection window (scrollable) that shows every internal flow stream in the model. Using this window, individual flow streams can be displayed quickly without having to close the Internal Flow Properties screen and double clicking on another flow stream to display its properties. The External Flow Properties screen also has a similar external flow selection window for viewing external flows. The internal and external flow properties screens provide valuable information about the process that can be used for engineering design and/or data reconciliation.

Printouts are available for the Revenues report, Summary report and Details report. These reports provide an overview of the balance calculations. The Details report shows every flow stream in the model with the fraction of each component listed along with the station the flow originates from and the station to which it goes. Also, the pressure, temperature, flow quantity, color, solubility coefficients and currency value is printed for each flow stream. The Details report is a listing of most of the information that is on the Internal Flow Properties and External Flow Properties screens with component fractions displayed instead of percentage values for purity, dry substance, etc. The summary information on the Internal Flow Properties and External Flow Properties screens is basically the same as shown on the Summary report for each flow (Figure 12).

Some of the most useful information from Sugars is given on the Process Net Revenues report (Figure 13). As changes are made to the model, the process net revenues will either increase, or decrease. New process equipment with new station properties, or changes in the process routing, can be evaluated quickly to see now the revenues will change. The Process Net Revenues are calculated from the values for all flows leaving the model (internal flows) minus the cost of all flows into the model (external flows). This information is very useful for making investment decisions. Also, it is the most convenient way to show how changes to the model will affect the operating performance of the process.

The value entries for all external flows into the model and internal flows that leave the model are entered on the External Flow Properties and Internal Flow Properties screens. These values are used to calculate the process net revenues. The entered values can also be changed on the Process Net Revenues screen as shown in Figure 13.

# SUMMARY

Sugar companies and engineering firms use the Sugars<sup>™</sup> program to make process and investment decisions. They use it to increase the yield and reduce the energy consumption of existing sugar factories and refineries and to design new ones. Also, it is used to evaluate the

feasibility of R&D projects, train process engineers, reconcile factory data and give process information that cannot be measured. The new Windows® interface for Sugars greatly enhances the use of the program and makes it much easier to build models for process simulation. A model is built by simply dragging shapes from one of the shape stencils to the drawing surface. The shapes represent actual stations in the factory or refinery. External flows into a station and flows leaving a station are connected to other stations by using a connector tool with automatic line routing and crossovers. A dialog screen to enter data for controlling the performance of a station, or to describe an external flow, is displayed when the station, or external flow, is double clicked. The model is balanced by clicking on a balance icon after the flow diagram of the model is drawn and the data is entered. The results of the balance calculations provide material, energy and color values for all flows in the model. Also, the overall net revenues generated by the process are calculated. Flow stream values given by the calculations provide information about the process and engineering data (density, enthalpy, boiling point rise, sucrose supersaturation, etc) that is useful for process design. Process net revenue results are used to make investment decisions for new equipment, or process decisions. All of the data and calculated results for a model are saved in a Microsoft® Access database. The new Sugars for Windows program is a dramatic improvement to the Sugars program that makes it much easier to build models and show the calculated results to assist with decisions to improve the efficiency of a factory or refinery.

#### ACKNOWLEDGMENTS

Sugars<sup>TM</sup> is a trademark of Sugars International LLC

Visio® is a registered trademark of Visio Corporation

Microsoft® is a registered trademark of Microsoft Corporation

Windows® is a registered trademark of Microsoft Corporation





Figure 2. Model Properties screen and Sugars toolbar.







Figure 4. Online help systems for Visio and Sugars.



Figure 5. Station numbering dialog screen.



Figure 6. Sugar properties screen.



Figure 7. Data entry on Evaporator Properties screen.



# Figure 8. External Flow Properties screen.



Figure 9. Screen showing progress of balance calculations.













							LS	แสกมัน	агу го	port				
						1					_			
	್ಷಣ ವಿಷಣ	- EF		2.	360		2						5 a a a	
ŝ.			-	1.	4	2 A. 1	di 2.4				470	-	1	· · 4-
	A	10 JUN												P71-
~				aners)		Herbert -			-		199.94	aver, et	- sec. 19. 19. 19	( - ) ~
1				×	Camer In		Awr							
				_	and trape Cale	-	Terral I	-						
1					1,1,11	1.15								
							-							_
											• ···			
- X		Manual I		Tr Fm1	FLore (2b/b)	100	frager's	-	Planet.	1.00	115	TEMPS	-	
-														
5		-		Tarres &	194, 417.1	10.01	13.4	×	1.12			- H. 14		
21	-	il.												
8	125	Labora .	tate:	Lapas, da	R.999.L	10.00	1.0					10.10		
22	States of the local division of the local di	EL app	#14	R.comp.		8.48		0.00	8. H	4.00	<b>B</b> .8	P. 18		
34	-	Pines.			248. DE-4	1.22		1.00		1.49	MI.	÷. H		
2						- 22	22	12		12				
		-							• •		** *		•	
7								_						
-						-								
4											-			
-	Han.		Pin.	7s Pert.	Jim(18/h)	-	hgar's	Suma and A	Playet.	-	110	100	Colos	
									**	·····				
÷				-	44.491									
8			<u><u> </u></u>	Inc. in	87,164.1	10.41	34.94	3.11	\$2.57	4.00			D. 4 M	
	-	. <u> </u>		Taine B	E11, 411. I	47.40	K3. M	38 YL	10.17	1.80	12.1	24.87	6,466	
3	512 JAN 183	-jela			-									
1					PL TA. I	89.33	24.44	22	R. 77	12				
	111 144 111	1.200			28.49.1	10.31	14.00	8.91	87.87	1.00				
-	10 (m 4z a		124	1.0	416.914.1	17.49	P. N	4.9	10.10	4.0	79.0	34.13	6.494	
÷	530 <b>346</b> 853													
			- 12	144.00	at 10.1	111.00				1.22	31	100.00		
1			-		<b>600</b> , 067. 1	<b>L</b> . <b>H</b>	13.4	ш.п	R. P7		74.6			
														<del></del> .
- R	- 17 Te			6 G É	خصصت ا	11.	1.1	÷		1.1	: N.	، ار پ	1. N. P.	NGA DA
10.002			1.5	. •		- 14			E	1.00	÷	÷ .		
	2.00.1.00	1.1					Stitus	.m	51:	يولون .	· · · ·		the Card	
	and the second	ii		4									1	
		43 <sup>Ka</sup>	er 4	- <b>/</b> 🕯 🔽					. 4	· •			هما بدء	. д
								· · · · · · · · ·						





#### NATURAL FREQUENCY OF TORSIONAL VIBRATIONS IN SUGAR MILL DRIVES

#### Saul Herscovici

Power Engineering and Manufacturing, Ltd. Waterloo, Iowa 50704-4055

#### ABSTRACT

Natural frequencies of vibration are present in all mechanical drive line systems. This property is derived from the shafts that represent a torsional spring with mass plus the gears, turbine rotor, and rolls, each of which possess mass. Such a mass spring system can be defined mathematically for a roll drive installation from the tip of the turbine all the way to the end of the rolls. The natural frequency of the entire drive system can thus be calculated. It is desirable to have a natural frequency of vibration higher than the operating speed then a vibration damper must be incorporated in the drive system to reduce the "amplitude of vibration" to zero or as close as possible to zero. If the drive system is operated at or near the natural frequency of vibration without damping, then the system will vibrate and possibly fail.

#### INTRODUCTION

Two identical failures occurred on a planetary drive one year apart where an internal gear tooth in a planetary ring gear broke in a wedge shape about three inches long and a half inch wide at one end and curved to a point at the other end. When the design and manufacturing analysis showed no weakness or reason for breakage, we resorted to study the system's natural frequency.

## What is a Natural Frequency?

The natural freqency of vibration is a law of nature that can best be illustrated by imagining that a ping pong ball is bouncing back and forth between two walls, but every time the ball hits the wall, the wall pushes the ball away. As a result, the ball moves away from the wall faster than it came. If the ball speed increases with every stroke between the walls, eventually it will reach a speed that will have sufficient energy to crush the ball. All systems do not increase the energy of the vibrating member until destruction occurs. Some vibrating members are excited and the vibrations fade away slowly.

The best example of a device that operates at the natural frequency of vibration is a musician's tuning fork. When excited, it vibrates at a fixed frequency for several seconds. It produces a reliable musical note because it always vibrates at the same frequency because its geometry is fixed. This is precisely why the musicians use it. They can produce the same musical note all around the world.

A good example of a damper is the shock absorber in the car. If all shock absorbers were removed from the car, it would undulate until the passengers become seasick. When good shock absorbers are present in the car, car travels in a stable manner. The energy transferred from the axle housing through the springs when traveling over rough terrain, or a bump, is absorbed by the fluid in the shock absorber and converted into heat. The amount of heat is very small, and for that reason, the shock absorbers do not overheat.

Another example of a good damper, but never used except for demonstration, is a rubber band tightly set on a musician's tuning fork.

In most cases, the vibration energy that needs to be captured by a damper to eliminate vibrations or reduce the amplitude is a minuscule fraction of the total energy traveling through the system.



Figure 1. Vibration amplitude vs. frequency ratio of actual damping to critical damping.

Figure 1 shows a series of curves of vibration amplitude vs frequency ratio of actual damping to critical damping. Two characteristics can be observed: One is that the amplitude of vibration diminishes as the amount of damping increases. Note that when the damping is at its maximum level, as shown on the lowest curve marked 10, there are no vibrations present. As the amount of damping is reduced, the vibration amplitude increases as shown on Figure 1. C denotes the amount of available damping and C<sub>c</sub> is the critical damping that is required to eliminate vibrations. All systems have some amount of damping, but most of the time it is too small to eliminate damaging vibrations. Even the tuning fork has friction damping from two sources: One is the friction with the air when the forks move rapidly, and the second is the steel internal friction represented by its hysteresis property. Damping also exists inside the gear box and that is the viscous damping provided by the oil. Some friction damping is also present in the gear box because the gears and bearings experience sliding as they rotate. More damping is provided by the sugar cane as it passes through the rolls. But, here again, this damping is insufficient to eliminate vibration when any of the rotating shafts operate at the natural frequency. For this reason, additional damping is required.

The tuning fork example maybe the simplest vibrating device, and the vibrations are initiated by pinching the forks one single time. In a sugar mill drive, not only is there a complex system with masses and many torsional springs, but torque is applied continuously to every drive member. The force imposed upon every member can and does excite vibrations.

Figure 2 shows that a phase angle change of the vibrating masses occurs as a function of damping. The smaller the amount of damping, the larger the phase angle change becomes.



Figure 2. Phase angle change of the vibrating masses as a function of damping.

Figure 3 shows a schematic representation of a sugar mill drive that has eight rotating masses and seven torsional springs. The arrows on the rotating masses show an arbitrary but possible instantaneous direction of vibration motion of each member. The members can have vibration movement in opposite direction to each other inside a gear box and cancel each other to show no vibration at the input or output where it can be observed. Such a vibrating condition can still lead to damage, and it is very difficult to detect.

Another consideration is that the natural frequency of vibration occurs at a single point. This feature often permits a simple modification of geometry to a shaft to move the natural frequency of vibration above or below the operating speed. Unfortunately, this is not the case with sugar mill drives because the shafts are long and thus make good torsional springs, and also because the mass of the rolls is enormous.



Figure 3. Schematic representation of a sugar mill drive.

Equation 1 shows how to calculate the natural frequency of vibration for a simple system that consists of one mass and one spring as shown on Figure 4. K is the spring rate, M is the mass, and *f* is the system natural frequency. It can be seen from Equation 1 that as the spring rate, K, increases, the natural frequency *f* increases, and as the mass, M, increases, the natural frequency, *f*, decreases. It is best to make the spring rate as high as possible and at the same time to make the mass as small as possible in order to maximize the natural frequency of vibration to make it substantially above the operating speed range

(Equation 1).  $f = \sqrt{K/M}$ 



Figure 4. Simple systems consisting of one mass and one spring.

To further complicate the phenomenon of natural frequency, the mass moment of inertia increases or decreases with the square of the speed of the rotating members. This means that, for two identical shafts, if one shaft rotates twice as fast as the other, Equation 2 shows that the mass moment of inertia of the faster shaft is four times that of the other.

# (Equation 2). $(I_2 / I_1)^2 = (2 / 1)^2 = 4$

The amplitude of the torsional natural frequency of a mechanical system that has many springs and masses can be solved by trial and error by the Holzer method Once the natural frequency is calculated, the amplitude of vibration can be calculated.

The amplitude of vibration can be reduced by the addition of damping, as shown on Fig. 1. It is most effective to incorporate damping in the high speed gear coupler that is located between the steam turbine and the gear box. Some gear couplers incorporate a rubber member that has vibration damping capacity. The manufacturer specifies the damping, torque and speed operating characteristics.

One hundred percent reduction in amplitude of vibration may not be achievable but a reasonable target is over 90 percent at the input gear coupler. Some additional damping is provided by the lubricating oil in the gear boxes and the sugar cane as it passes between the rolls. This amount of damping provided by the gear oil and sugar cane is insufficient to provide trouble free operation at the natural frequency of vibration. Furthermore it is necessary to allow for operation of without load (i.e. without cane) and thus without the damping provided by the cane.

#### RESULTS

During the 1996 and 1997 sugar cane seasons, two gearboxes, our WH-325005 (Figure 5), were operated at an input speed near or at the natural frequency speed without any damping. Vibrations could be felt at the base of both gear boxes and even on the concrete floor. During the 1998 season, these two gear boxes were equipped with input gear couplers that have a rubber damper. Vibrations at gear box base or at the concrete could not be observed.

The WH-SM gear box installed at the Patout Sugar Mill as shown on Figure 6, was also operated during the 1998 sugar cane season. Calculations show that the turbine speed is at or near the natural frequency speed. All efforts made to change shaft geometry to raise the natural frequency above the operating speed were in vain. To reduce or eliminate vibrations, the input gear coupler was equipped with a rubber member that has damping characteristics. No vibrations were observed at any speed, including the natural frequency speed.

It is possible that there may be vibrations inside a gearbox with such phase angles that they are self cancelling before reaching the input shaft.

The WH-500002 (Figure 7) that operates at the New Iberia Sugar Mill has a turbine speed that is substantially above the natural frequency speed.

We also manufactured one shredder drive gear box (Figure 8) and two knive drive gear boxes (Figure 9), The torsional natural frequency of these gear boxes are substantially higher than the operating speeds because there is only one gear reduction in the drive and the shafts are short.


		OUTPUT TORQUE RATING, FT-LB				
		AGMA 1	ACMA 2			•
MODEL	RATIO	INTERMITTENT	CONTINUOUS	STALL	WEIGHT (LBS)	OIL CAPACITY:
WH-325005	118.102:1	375000	175000	500000	90004	26.5 GAL

<sup>750</sup> HP AT 3600 RPM INPUT AT AGMA 3.0

Figure 5. WH-325005 sugar mill gear box with turbine speed near the natural frequency..

WH-8M WHEEL HUB



		OUTPUT TORQU			
		AGMA 1	AGMA 2		
MODEL	RATIO	INTERMITTENT	CONTINUOUS	STALL	WEICHT (LBS)
₩H-8M	1147.3:1	375000	175000	500000	112,500#

750 HP AT 3600 RPM INPUT AT AGMA 3.0

Figure 6. WH-8M sugar mill gear box with turbine speed near natural frequency.



Figure 7. Gear box with turbine speed substantially below natural frequency.

## sugar mill shredder drive gear box FMSR-16 foot mounted speed reducer



3500 HP AT 4000 RPM INPUT AT AGMA 3.9

Figure 8, Shredder drive gear box with natural frequency higher than operating speed.



Figure 9. Knife drive gear box with natural frequency higher than operating speed.

## ONE DESIGN, TWO PANS A LARGE FLEXIBILITY OF USE

## G. Journet and C. Pelletan FCB-Sugar Department BP2047, 59015 LILLE, France <u>dsucom@fcb-group.com</u>

## ABSTRACT

In 1996, the new CCTW continuous vacuum pan replaced the older CCTR pan. The new continuous vacuum pan shows clearly improved performance. Its design allows installation of mechanical agitation. A variant of the CCTW continuous vacuum pan, the CCTWD double pan, has been developed which allows a great flexibility of use, like, for example, two strikes in the same equipment, or two pans running in parallel, or the ability to make continuous vacuum pans of very small capacity.

The first true continuous vacuum pan used in sugar mills dates back to 1967. It was the FCB prototype installed in the sugar refinery in Nassandres, France. Since then FCB has built and installed 150 continuous vacuum pans (CCTR type) in every strike of beet and cane sugar mills throughout the world.

Since 1996 a new continuous vacuum pan, the CCTW, has replaced the old CCTR models. Thirty-five CCTW pans are already in operation or in the course of installation. This new model marks a significant advance relative to the older one.

## DESIGN BASIS

## Efficient calandria

In both CCTR and CCTW continuous vacuum pans, the calandria is formed of horizontal steam tubes organized vertically in rows. Simulations have proved that this type of caladria offers the best trade-off between the heat exchange coefficient and the head loss. Furthermore, as far as we know, this type of calandria is the only one to process white sugar massecuite without mechanical stirrers. The ratio of the exchange surface to the massecuite volume is normally 10  $m^2/m^3$ .

Circulating the steam in three passes enables the entire surface of the calandria to be swept systematically. The steam always circulates fast enough to ensure the complete impulsion of the condensate and the noncondensable gases. The extraction of noncondensable gases is rigorously controlled, since the extraction point is located just at the end of the steam circulation.

#### Very low hydrostatic load

The cross-sectional geometry of the calandria has been changed and specially designed to ensure the lowest possible maximum hydrostatic load (vertical distance between the level of the massecuite and the lowest point in the calandria) as shown in Figure 1. This effect is enhanced by the sloping top of the calandria, which facilitates the recirculation of the massecuite at the same time that it places the calandria as high as possible within it. The gain relative to the former CCTR vacuum pans is from 400 to 900 mm, according to the size of the pan. This corresponds to a practical gain in At in the range of 2 to  $4 C^{\circ}$ .



Figure 1. Comparison of cross-sectional geometry of CCTR and CCTW pans.

Wide passage for recirculation

The lateral recirculation passages have been improved and made as wide as possible in order to facilitate the natural recirculation of the massecuite. By way of comparison, note that the width of these passages, expressed in terms of the diameter of a central downtake in a batch vacuum pan, is equivalent to 55% of the diameter of the shell, whereas the ratio for batch pans generally runs from 30 to 40 %, and 45% for the former CCTR vacuum pans.

## Large overflow threshold

By sloping the top of the calandria toward the side passage it is possible to employ a maximum exchange surface while facilitating the recirculation of the massecuite toward the passage. This arrangement reduces the hydrostatic load on the calandria at the same time that it maintains a high overflow threshold onto the side passage (almost equivalent to the width of the recirculation passage itself), thus achieving an excellent recirculation flow.

A large overflow threshold is a necessary condition for obtaining high recirculation flow. When the top of the calandria is horizontal and the height of massecuite over the calandria is kept low in order to minimize the hydrostatic load, that inhibits the recirculation of the massecuite, whose flow rate is a direct function of the cross-section of the overflow. This is evident if we imagine a very low height of massecuite over the calandria (close to zero). In these conditions the hydrostatic load is obviously reduced to the height of the calandria. However, it is equally obvious that the recirculation flow is also close to zero.

## Installation of mechanical circulation

The bottom of the shell is conceived so as to allow the optimal installation in each compartment of a propeller for mechanical agitation; that is to say, under the calandria, in the center of and perpendicular to the calandria, without dimensional constraints.

A large flexibility of installation is possible; it is indeed possible to install mechanical stirrers just in the last compartments where the massecuite tightens, or in the last half of the pan, or even in the whole pan.

The power consumed by mechanical agitation in the last compartments are in the order of 0.5 to 0.6 kW/m<sup>3</sup> (massecuite A in Vietnam), 0.65 kW/m<sup>3</sup> (massecuite B in Vietnam), 0.5 to 0.65 kW/m<sup>3</sup> of massecuite C in Vietnam) and 0.5 to 0.65 kW/m<sup>3</sup> of massecuite (CCTW 200m<sup>3</sup> - impellers 0900mm - massecuite B in Argentina).

#### STANDARD PAN CCTW

Thus we end up with the schematic conception in Figure 2. The shell surrounds the calandria leaving a large recirculation passage on the side, which narrows slightly toward the bottom (load increase) and closes gradually toward the center underneath the calandria.



Figure 2. Standard CCTW vacuum pan design.

The CCTW continuous vacuum pan is a horizontal pan which is divided into thirteen volumetrically graduated cells and is constructed with a longitudinal partition running the entire length of the pan and with transverse partitions.

The magma is fed into the first cell, and the massecuite flows from cell to cell by means of openings located alternatively in the upper level of the clandria and underneath it. The final massecuite is drawn off from the last cell. The syrup/liquor feeds into the bottom of each cell.

The CCTW CVP is currently offered in five base diameters and nine pan lengths. The useful volume of the pans ranges from  $22 \text{ m}^3$  to  $200 \text{ m}^3$ . This vacuum pan can be used for all strikes in cane and beet sugar mills and refinery.

Given the excellent natural circulation of the massecuite, it is not necessary to add steam agitation. This fact has been verified.

## Main advantages

In summary, the CCTW vacuum pan incorporates the following advances relative to the CCTR:

- Much lower hydrostatic loads,
- Wider recirculation passages (+25% on the average),
- Better condition of natural recirculation,
- Easy installation of mechanical agitation.

These advances make the following gains possible:

- Significant reduction of Dt,
- Improved exhaustion,
- Improved granulometry,
- Elimination of steam agitation,
- Less sensitivity to fouling.

## Results in low grade product

The first two CCTW pans were operating in 1997 to the entire satisfaction of their users. The first pan started in June 1997 in cane low grade product at the Cruz Alta sugar mill in Brazil. The second started in July 1997 in Quentin low grade product at the Jaen beet sugar factory in Spain. Nine other pans started in cane in 1998; three in India on A, B and C massecuite, three in Vietnam on A, B & C massecuite (partially with mechanical agitation), two in Brazil on B and C massecuite and one in Argentina on B massecuite (with mechanical agitation on the last half of the pan). In 1999, eight other pans have already been commissioned in cane applications, mainly in Vietnam, with one pan to be installed in Mexico at the end of this year. In beet sugar applications, four pans (one in Spain, one in Sweden, and two in the Netherlands) will be put in operation next September.

In India and Vietnam pans are controlled by conductivity. In all other cases the pans are controlled predictively, i.e., by controlling the entry (namely, the flow of feeding syrup, or liquor). A permanent calculation of the pan's materials balance keeps the final brix of the massecuite at a constant level.

The Cruz Alta vacuum pan has a useful volume of 150 m<sup>3</sup> with an exchange surface of 1504 m<sup>2</sup>. It does not use mechanical agitation. The proportion of magma is approximately 30% of the flow of the produced massecuite, which is in the range of 30 to 40 tons/hour.

We measured the performance of this pan at two different times: after about two months of operation and again after three months, when the pan was fouled and needed cleaning. Table 1 below illustrates the average balance for four days in August 1997, while Table 2 illustrates the average balance for three days in September 1997. Table 3 shows the exhaustion obtained. Considering that the Cruz Alta mill was very poorly equipped for mixing low-grade product, with no means of cooling, this exhaustion can be considered excellent.

Tuble 1. Avenuge building for Cruz And CCT of pull ther two months operation. August 1997,						
	Footing		Evaporated			
	magma	Molasses	Massequite	water	Vacuum	Calandria
Flow(T/hr)	9.2	27.9	31.9	5.2		
Brix(%)	90.0	79.1	95.0			
Purity (%)	71.6	60.7	61.5			
Temperature (C°)			62.6		56.4	91.5
Pressure (bar abs.)					0.169	0.743

Table 1. Average balance for Cruz Alta CCTW pan after two months' operation. August 1997,

Table 2. Average balance for Cruz Alta CCTW pan after three months' operation. September 1997.

	Footing	Evaporated				
	magma	Molasses	Massequite	water	Vacuum	Calandria
Flow(T/hr)	10.0	26.8	31.7	5.1		
Brix(%)	90.0	79.0	95.0			
Purity (%)	71.4	61.3	64.3			
Temperature (C°)			62.6		56.8	96.0
Pressure (bar abs.)					0.172	0.878

	Averages			
	August 1997	September 1997		
Apparent massequite purity	62.23	63.70		
Apparent molasses purity	42.44	43.10		

Table 3. Exhaustion results for Cruz Alta CCTW pan during the two test periods.

## DOUBLE PAN CCTWD

19.79

20.60

#### Two pans in one

Purity drop

The basis idea is to extend the longitudinal partition up to the top of shell in order to completely separate the two symmetrical sides of the pan (Figure 3). This longitudinal partition, calculated by finite elements, is thickened and reinforced so as to support atmospheric pressure on a side and full vacuum on the other. Thus, with the same equipment, two equivalent and distinct symmetrical volumes can be separately operated. Of course, the calandria is also separated into two parts in accordance with the separate volumes. For each pan, i.e. for each side, massecuite circulation is linear from one extremity of the equipment to the other one. The number of compartments is reduced to 11.

The result is that the two volumes can operate with two different strikes : A and B or B and C. The benefits are more compactness of installation and an investment cost reduction. Figure 3 illustrates this design.



Figure 3. Double pan CCTWD design.

## Two parallel pans

At a reasonable increase in cost, this type of equipment also permits operation as two pans in parallel on the same strike. This configuration allows the following benefits:

When the two pans operate on the same strike, it is possible to continue to work at 50 to 60% of nominal, with only half a pan, during momentary stops of the other half for cleaning. Consequently, the liquor and massecuite storage capacities can be significantly reduced.

- This pan also facilitates adjustment to reductions of the factory output in case, for instance, of severe weather conditions.
- This design is also well adapted to small factories which may process two different strikes independently in the two separated parts of the CCTWD pan.

## FLEXIBILITY OF THE CALANDRIA

The calandria of the CCTW pan formed by horizontal steam tubes organized in rows presents a high flexibility of use which has not been fully exploited until now. FCB is offering new applications allowing a larger versatility of its equipment and a close adaptation to the customer needs.

#### Increase of the CCTW capacity

In particular cases where increase of production in a factory is foreseen, the CCTW vacuum pan can offer a simple and reliable solution for a large variation of massecuite throughout. The Ortofta factory (Danisco Sugar Group) in Sweden is currently processing 10,000 tons of beets per day. Due to reorganization in the Swedish sugar industry in the coming years, the capacity of the factory is foreseen to reach 16,000 tons of beets per day by the year 2002 (+60%). A single 150 m<sup>3</sup> CCTW continuous vacuum pan (diameter 4,600mm) has ben installed this year at the Ortofta factory in order to process the third strike massecuite at both 10.000 TB/D and 16.000 TB/D. In a CCTW vacuum pan, it is very easy to remove or add tubes in the calandria. Our idea was to take advantage of this specificity in order to exactly adjust the exchange surface of the calandria to the needs of the crystallization operation (Figure 4). At 10,000 TB/D, the pan is equipped with 786 heating tubes corresponding to a 1,078 m<sup>2</sup> heat exchange surface and the level of massecuite in the pan is set at 2.2 m corresponding to a total masecuite volume of 115 m<sup>3</sup>. At 16,000 TB/D, the heating surface will be increased up to 1,504 m<sup>2</sup> by adding 310 new tubes. The level of massecuite in the pan will be set at 2.7 m corresponding to a total massecuite volume of 150 m<sup>3</sup>. The pan is purposely designed from the beginning to receive all 1.096 tubes. The upper holes in the tube plates that will not be connected to heating tubes during the first step are plugged with special blind sleeves. Figure 4 illustrates this evolution.



Figure 4. CCTW pan designed for eventual increase in capacity.

## Higher heating surface/volume ratio

The standard surface to volume ratio is  $10 \text{ m}^2/\text{m}^3$ . The vacuum pan is designed to allow this ratio to increase up to  $13 \text{ m}^2/\text{m}^3$  which corresponds to + 30%, This feature may be of some interest in order to improve the thermal efficiency of the crystallization station and to achieve steam savings.

## OUTLOOK FOR THE NEW CONTINUOUS VACUUM PAN RANGE

Drawing on these preliminary results, we can project the performances that can be expected from this new vacuum pan. For any immediately foreseeable applications, these may be summarized in four areas:

- Thermal performance.
- Exhaustion.
- Granular Quality.
- Energy savings.

## Thermal performance

We customarily characterize heat exchange in terms of the evaporation rate, expressed as the kg of water evaporated per hour per  $m^2$  of exchange surface (TEV in kg/h.m<sup>2</sup>), and set in relation to

the total Dt (the difference of temperature between the calandria and the vacuum). Figure 5 shows the relationship between TEV and total Dt for CCTW and CCTR pans in cane applications.



Figure 5. Relation between evaporation rate and total Dt in cane CVP applications.

By way of comparison, we show on the same graph the performance curve of the former CCTR type vacuum pans (with an average fouling). The performance of the new vacuum pan appears as a net improvement on the former model, since its operating results are clearly above the former model's even when its calandria is fouled.

## Exhaustion

Very good exhaustion is obtained at Cruz Alta and Jaen (cf.Table 3 above). As with the improved heat exchange, this is due to better circulation and mixing of the massecuite in the equipment. The measure of massecuite temperatures demonstrates the excellent circulation. Indeed, our measures show that there is virtually no hydrostatic temperature elevation in the calandria. This improvement in the circulation and mixing of the massecuite will necessarily promote optimal exhaustion.

#### Granulometric quality

Granulometric quality in a multi-celled continuous vacuum pan is related to the main parameters:

- the granulometric quality of the footing magma,
- the number of compartments,

the quality of the mixing of the massecuite in each compartment.

The CCTW continuous vacuum pan has thirteen cells; the CCTWD pans, eleven. These values are a good compromise, being sufficiently high to minimize the dispersion of residence time, yet low enough to keep the pan and its process control simple.

A definite improvement has been obtained in the quality of the mixing within each compartment, relative the former CCTR vacuum pan. Considering that granulometric performances of the former continuous vacuum pan were already good, like illustrated by *table 4* hereafter. It is clear that those of the new one will be even better.

## Energy savings

In its present design the new CCTW vacuum pan already shows excellent heat exchange performances, as we have just demonstrated. These performances can be significantly enhanced. The standard surface to volume ratio is 10 m<sup>2</sup>/m<sup>3</sup>; the vacuum pan is designed to allow this ratio to be pushed up 30% to 13 m<sup>2</sup>/m<sup>3</sup>. The expected heat exchange performances permit very low Dt between the calandria and the vapor space, i. e., from 20 to 25 ° C with high purity massecuite. Given this, several ways of saving energy are possible:

- Steam bleeding for crystallization can be deferred to the fifth or sixth effect. For
  instance, with a vacuum at 250 mbar abs (65 ° C), the necessary pressure for the
  calandria is close to 700 mbar abs (90 ° C).
- It is relatively easy to operate vapor recompression with a compression ratio of about 3 (for example 250 to 750 mbar abs).
- Crystallization in two effects can be envisaged, as in the schematic example given in Figure 6 below:



Figure 6. Crystallization in two effects with CCTW pans.

This process can be used both in first strikes and in refined strikes of sugar mills. The vaporsteam produced by the second continuous vacuum pan is condensed in the first pan calandria, where it produces a second evaporation. This type of arrangement allows significant energy savings, since it cuts the steam required for crystallization by about half.

## CONCLUSIONS

This new continuous vacuum pan shows significant advances in every area of crystallization. We believe that this type of pan is particularly well adapted to cane sugar mills and refinerys, where its features can offer excellent exhaustion and granulometric quality while at the same time providing energy savings which translate into lower operating costs.

## REFERENCES

- Journet, G. 1996. Mechanical stirring in a FCB continuous evaporating crystallizer. Zuckerind. 121(5):357-359.
- Hill, S. 1965. A method of determining a "crystal regularity index" for white sugar. Int. Sug. J., July 1965, pp. 201-203.

## AGRICULTURAL ABSTRACTS

## The Lady Beetle *Diomis terminates* (Coleoptera: Coccinellidae) and the Yellow Sugarcane Aphid *Sipha flava* (Homoptera: Aphididae) in Florida.

D.G. Hall

United States Sugar Corporation Clewiston, FL

The yellow sugarcane aphid is occasionally an important pest of sugarcane in Florida. In a greenhouse study comparing the growth of young sugarcane plants infested versus not infested by aphids over a 3wk period, the height of infested primary shoots was reduced by 36.2% and infested plants produced fewer leaves and tillers. At the end of the test, 71.7% less dry matter was present per infested plant. The lady beetle Diomis terminatus is a common predator of the yellow sugarcane aphid in Florida. Mass-rearing the beetle on vellow sugarcane aphids was investigated. Yellow aphids were relatively easy to raise in a greenhouse during winter and spring 1998-1999 and 1999-2000, but the aphid was difficult to rear in the greenhouse during summer and fall 1999, apparently due to excessive air temperatures. Sorghum-Sudan grass proved a convenient plant host for massrearing the aphid. D. terminatus was reared in a laboratory in large glass test-tubes on vellow sugarcane aphids, To obtain beetle eggs, aphids were transferred into the tubes along with adult beetles and a small piece of wax paper, onto which adults oviposited. Eggs on wax paper were transferred to new tubes and supplied with aphids for larvae to feed upon. Larvae pupated in the tubes, and adults were harvested. A ratio of about 7 live aphids per D. terminatus larva or 10 live aphids per D. terminatus adult was maintained in the tubes. Morphological characters for distinguishing male and female beetles were not known. In rearing tubes with 10 beetles per tube, an average of 1.4 eggs per female per night were laid (50% beetles per tube assumed to be female). When mated females were held individually, they laid an average of 3.0 eggs per female per night, with an average total of 41.9 eggs per female. Across all densities of eggs per tube studied, an average of 33.9% eggs developed to the adult stage. Higher percentages of success in rearing larvae to the adult stage were achieved when fewer than around 40 eggs were placed in a tube.

## Efficacy of Soil Insecticides for Wireworm Control in Florida Sugarcane

R. Cherry Everglades Research and Education Center, University of Florida Belle Glade, FL

Wireworms are important insect pests of Florida sugarcane. To prevent wireworm damage, soil insecticides are routinely applied in the furrow with seedpieces at planting in Florida sugarcane. Currently the two soil insecticides most commonly used for wireworm control in Florida sugarcane are ethoprop applied as Mocap 20-G and phorate applied at Thimet 20-G. The objective of this study was to evaluate different insecticides for wireworm control in Florida sugarcane. Preliminary tests were conducted in a laboratory from January to April 1999. *Melanotus communis* were held seven days at 20°C and exposed to different doses of insecticides in moist soil. Deltagard G (AI=0.1 %) and Talstar PL-GR (AI=0.2%) caused little mortality and were not selected for further testing. Aztec 2.1%G (AI=2.1%) and Force 3G (AI=3.0%) caused some mortality to wireworms and showed potential for further testing.

Field tests were conducted from December, 1999 to March, 2000 in a newly planted sugarcane field to evaluate two rates of Aztec 2.1G versus Mocap 20G versus Thimet 20G for wireworm control. Tiller counts and wireworm counts were made during March 2000, which was three months after planting. There was no significant difference in number of tillers or wireworms between the control and any of the treatments. Quite simply, as indicated by low wireworm numbers in control plots, there were very low numbers of wireworms present in the plots so that insecticide efficacy could not be determined in the field test.

Laboratory tests were conducted from December, 1999 to April, 2000 using dose mortality regression to determine LD50 values of Aztec 2.1G, Mocap 20G, and Thimet 20G to *M. communis* wireworms. Wireworms were exposed to different doses of these insecticides for 14 days in moist soil at 25 °C and then survival determined. Based on the weight of granular formulations, Thimet 20G had the lowest LD50 value, Aztec 2.1G had an intermediate LD50 value, and Mocap 20G had the lowest LD 50 value. Based on the weight of active ingredients in formulations, Aztec 2.1G had the lowest LD 50 value. Thimet 20G had an intermediate LD50 value, and Mocap 20G had the lowest LD 50 value. These data show that the active ingredients in Aztec 2.1G are very toxic to *M. communis* wireworms and that the insecticide is a potential candidate for wireworm control in Florida sugarcane.

## Potential Impact of Sugarcane Varietal Changes On Insect Pest Management in Louisiana

#### T. E. Reagan and F. R. Posey

Department of Entomology, LSU Agricultural Center Baton Rouge, LA

Sugarcane resistance to the sugarcane borer, *Diatraea saccharalis*, (SCB) is categorized as a combination of physical characteristics that hinder boring (i.e. rind hardness, leaf-sheath appression), cultivar specific tolerance to boring, and antibiosis mechanisms that contribute to the differential survival of bored in larvae. The extent of this resistance is infestation level dependent. Heavy borer pressure results in more bored intermodes even in cultivars considered highly resistant. Several factors contribute to seasonal area-wide SCB infestation levels such as weather conditions, predator and parasite numbers, and indigenous borer populations. Expansive acreage of cultivars with elevated moth production increases endemic SCB populations and imposes additional pressure on the remaining resistant varieties. For this reason, we also report moth production for each cultivar in the various experiments.

Entomology research on Louisiana sugarcane has a long history of emphasizing development of resistant varieties, both at the LSU agricultural Center and at the USDA facility in Houma. Until the production of the two most recently released SCB susceptible varieties (LCP 85-384 and HoCP 91-555), all recently recommended varieties exhibited good levels of SCB resistance. Assessment of the L 1997 series early line trials indicates that likely high yielding candidates for advancement in the variety development program (including L 97-137) have the potential to be more severely damaged and produce significantly higher most populations than the highly susceptible HoCP 91-555. The entomology rating of the 1999 plant cane outfield variety test had six of the ten candidates for release in the most SCB susceptible group.

While the USDA entomology program focuses on varieties emphasize, assessment of various classes of SCB plant resistance (antibiosis, tolerance) in the basic breeding program, and yield relationships of candidate varieties, the LSU program avoids duplication. This complementary program, at the earliest practical stages, assesses relative SCB injury of the L cultivars, later stage USDA candidates, as well as adult emergence affecting area-wide SCB population management.

## Novel Technology for Sugarcane Insect Pest Management

## J. C. Legaspi, B. C. Legaspi, Jr. and T. E. Mirkov Texas Agricultural Experiment Station, The Texas A&M University System Weslaco, TX

The development and evaluation of transgenic lines of sugarcane resistant to insects is currently underway at the Texas Agricultural Experiment Station. An insecticidal gene (GNA) from the snowdrop lily, Galanthus nivalis, was incorporated in sugarcane, var. TCP65-357. This gene causes sugarcane to produce a protein called a lectin which interferes with insect digestion, although numerous tests indicate the lectin to be nontoxic to birds and mammals. A series of laboratory, greenhouse and field tests were conducted to determine the efficacy of transgenic sugarcane against the main insect pest in south Texas, the Mexican rice borer (MRB), Eoreuma loftini and a secondary pest, the sugarcane borer, Diatraea saccharalis (Lepidoptera: Pyralidae). In the first field evaluation of transgenic sugarcane in 1998, we found that the transgenic treatment had significantly lower damage due to the Mexican rice borer compared to the non-transgenic (check) treatment. Two fields (1<sup>st</sup> ration and plant cane) of transgenic and non-transgenic sugarcane were monitored in 1999. Transgenic plants had significantly lower damage than the non-transgenic plots in the ratoon crop. However, no significant differences were found in the percentage of bored internodes between the two treatments in the plant cane field. Analysis of the transgenic plants using the Western blot and the Southern blot analyses showed medium to high expression of GNA. The impact of transgenic sugarcane to biological control agents in the field was also assessed. We studied the percentage of parasitism of Mexican rice borer and parasite species abundance in the two treatments. In general, there seemed to be no difference in the parasitism of MRB in the field between the two treatments. The parasites collected from field samples were mainly the native species, Chelonus sonorensis and Digonogastra solitaria (Hymenoptera: Braconidae).

#### Sugarcane Yellow Leaf Virus: Possible Tactics to Develop Resistant Cultivars

J. C. Comstock<sup>1</sup>, J. D, Miller<sup>1</sup> and T. E. Mirkov<sup>2</sup> <sup>1</sup>USDA,ARS Sugarcane Field Station Canal Point, Florida <sup>2</sup>Dept Of Plant Pathology and Microbiology Texas A&M Agricultural Experiment Station Weslaco, Texas

Sugarcane yellow leaf virus (ScYLV) rapidly infects sugarcane clones in the CP-variety development program at the Sugarcane Field Station, Canal Point, Florida. In 1997, the incidence of ScYLV was 1.6, 30.4, 38.0 and 50.0% for clones in the seedling stage, Stage I, Stage II and Stage HI, respectively. Similarly, the incidence of ScYLV was 16.1 and 38.3% for clones in the seedling stage and Stage II in 1999. The incidence of ScYLV was 16.1 and 38.3% for clones in the seedling stage and Stage II in 1999. The incidence of ScYLV was 16.1 and 38.3% for clones in the seedling stage and Stage II in 1999. The incidence of ScYLV in grower fields of CP commercial cultivars is usually above 70%. Only a small percentage of clones in the domestic and foreign collection at the Sugarcane Field Station is free of the virus. Clones imported for crossing from Louisiana and Texas are initially virus-free and subsequently a portion becomes infected. Available resistance to ScYLV appears limited. The following tactics for identifying ScYLV resistance will be discussed: 1) Detection of ScYLV-free clones in domestic and foreign collection, 2) Frequency of ScYLV infection in progeny of parental clones used in the crossing program, 3) Relative incidence of infection in Stage III and Stage IV clones, 4) Relative rate of infection of clones established from virus-free plantings, and 5) Transgenetic resistant plants mediated by the ScYLV coat-protein gene.

## Molecular Markers for Monitoring Transgenic Sugarcane

A. M. Abouzid, M. Gallo-Meagher and K. Chengalrayan University of Florida, Agronomy Department Gainesville, FL

In order to move away from the use of antibiotic resistance genes as markers for transgenic sugarcane, we have tested different vectors containing the green fluorescent protein (GFP) reporter gene, as well as the phosphomannose isomerase (PMI) selectable marker gene in sugarcane transformation experiments. Using flourescense microscopy, GFP could be visualized in calli 24 h post bombardment and transformed sectors could be detected in one week. GFP appears to be a useful, non-destructive, and rapid screenable marker for transgenic sugarcane cells. In order to utilize the PMI selectable marker gene, the sensitivity of non-transformed calli to mannose as a selection agent was determined. Results showed that all concentrations of mannose (0, 1-5, 8, and 10g/L suppressed calli growth and shoot formation to various degrees. Addition of 3g/L mannose to the callus maintenance medium was sufficient to cause browning and death of the callus cells. Expression of the PMI gene in sugarcane calli in the presence of 5g/L mannose may be adequate for the selection of transgenic sugarcane plants.

## Effect of Growth Regulators on Efficient Plant Regeneration From Sugar Cane Callus

#### K. Chengalrayan and M. Gallo-Meagher

University of Florida Gainesville, FL

Efficient shoot regeneration of sugar cane (Saccharum spp. Hybrids cv. CP84-1198) from embryogenic callus cultures has been obtained using thidiazuron (TDZ). Callus was cultured on modified Murashige and Skoog's (MS) basal media containing 0.5, 1, 2.5, 5 or 10 uM concentration of one of five different growth regulators namely, 6-benzylaminopurine (BAP), kinetin (KN), 6-(yy-dimethylallylamino)-purine riboside (2iP), zeatin (Z) or TDZ with or without 22.3  $\mu$ M (x-naphthaleneacetic acid (NAA). Results indicate that modified MS basal medium supplemented with 2.5  $\mu$ M TDZ and 22.3  $\mu$ M NAA was optimal with 92% of calli turning green and inducing shoot formation with an average of 1132±41 shoots per 500 mg calli in 75 days. However only 10% of these shoots were more than 1 cm in length at the time of observation. In contrast, 73% of the shoots induced in 1  $\mu$ M KN were more than 1 cm in length. Further experiments are in progress to obtain culture conditions which maximize both shoot number and shoot elongation.

#### Current Status on Biotechnology Research at Sugarcane Research Unit, Houma, Louisiana

Y.B. Pan<sup>1</sup>, D. M. Burner<sup>2</sup>, M. P. Grishani<sup>1</sup>, B. L. Legendre<sup>1</sup>, and Q. Wei<sup>1</sup> USDA, ARS, 'SRRC, Sugarcane Research Unit, Houma, LA <sup>2</sup>SPA, Dale Bumpers Small Farms Research Center, Booneville, AR

The objectives of the biotechnology program at Sugarcane Research Unit of the Southern Regional Research Center, Houma, LA are threefold: nucleic acid-based disease diagnosis: molecular marker-assisted breeding; and sugarcane transformation. Polymerase chain reaction protocols for specific detection of the bacteria that cause sugarcane leaf scald and ratoon stunting diseases were developed. Reverse transcription-polymerase chain reaction procedures have been adopted to test for the strains of sugarcane and sorghum mosaic viruses and the newly discovered sugarcane vellow leaf viruses. Three DNA markers including Eri3/Eri4, Gigl/P2 and OPA-11 -366 have been identified that are species specific. In 1999, the marker OP A-11 -366 was used in the basic breeding program to confirm the selection of 10 interspecific F. hybrids between a Saccharum spontaneum clone and two sugarcane cultivars for further genetic improvement through backcrosses. Another useful marker system, microsatellite or Simple Sequence Repeats, is being evaluated on Louisiana varieties. Progress has also been made on sugarcane transformation. Embryogenic callus tissue is being produced routinely. A biolistic gene gun is being used to deliver into sugarcane tissue gene constructs that offer resistance to a particular herbicide or infection by sugarcane mosaic virus H strain. Potentially transformed sugarcane seedlings that were regenerated on selective media are being screened. Success of the transformation project will lead to the development of sugarcane genotypes that could have a significant impact on the breeding and selection programs in the future. Overall, biotechnology research has already begun to yield dividends, especially in the area of disease diagnosis and marker-assisted selection. However, the impact of transgenic sugarcane has not yet been realized.

## RNA Isolation and Photosynthetic Gene Expression in Sugarcane Exposed to Elevated C0<sub>2</sub> and High Temperature

#### M.Q. Zhang, M. Gallo-Meagher, J. C.V. Vu, and H. Allen, Jr

Agronomy Department, University of Florida Gainesville, FL

Two simple and effective protocols based on guanidinium isothiocyanate (GTC) and cetyltrimethylammonium bromide (CTAB) have been developed for the isolation of RNA from sugarcane exposed to enriched C0, and high temperature. The GTC protocol resulted in high yield of high-quality RNA from sugarcane grown at the ambient temperatures (Ta, °C), but not from tissues exposed to high temperatures (Ta+6,  $^{\circ\circ}$ C) because of their high levels of polysaccharides, polyphenolics or other unidentified compounds. The CTAB protocol was superior to the GTC protocol due to the inclusion of polyvinylpyrrolidone (PVP) and ethylenediamine tetraacetic acid (EDTA) in the RNA extraction buffer. However, the CTAB method is more labor intensive. RNA isolated using suitable protocols was of high quality and produced good hybridization signals in northern blot analysis. Results from northern blot analysis also showed that high temperature significantly inhibited the transcript levels of phosphoenolpyruvate carboxylase (PEPcase) and Ribulose-1,5-Bisphosphate Carboxylase/Oxygenase small unit (rbcS) genes in plants grown both at enriched C0, (700 ppm) and at ambient C0, (360 ppm); However, their transcripts in plants exposed to high temperature were reduced to a greater extent at ambient C0, when compared with the plants at elevated C0<sub>2</sub>. Growth at ambient temperature appeared to reduce the transcript levels for PEPCase, but to increase for rbcS.

# Effect of Purple Nutsedge (Cyperus rotundus L.) Competition in Sugarcane (Saccharum spp.)

#### M. J. Duchrow and J. M. Shine, Jr.

Sugarcane Growers Cooperative of Florida Belle Glade, FL

Purple nutsedge (*Cyperus rotundus* L.) causes yield losses in many agronomic crops. Yield losses associated with purple nutsedge in sugarcane (*Saccharum* spp.) have not been studied extensively. Partial additive studies were conducted in containers to determine the effect of interference of different initial population densities of purple nutsedge on sugarcane. Five commercial cultivars were evaluated; CP72-2086, CP80-1827, CP80-1743, CP88-1762, and CP89-2143. Purple nutsedge tubers were planted at densities of 50, 100, 150, 200, and 250 tubers/m<sup>2</sup>. Purple nutsedge densities as low as 50/m<sup>2</sup> significantly (*p*<0.05) reduced sugarcane fresh weight, number of tillers, primary stalk fresh weight, and primary stalk diameter in the plant cane crop. Total fresh weight reduction at 250 purple nutsedge tubers/m<sup>2</sup> ranged from 19% in CP80-1827 to

29 % in CP80-1743. Tiller numbers were reduced from 4% in CP80-1827 to 37 % in CP80-1743. Losses in primary stalk diameter ranged from 4% in CP80-1743 to 14% in CP80-1827 and CP88-1762. Primary stalk fresh weight losses ranged from 16% in CP72-2086 to 36% in CP80-1827.

## CGA-362622: A New Herbicide for Weed Control in Sugarcane

## E. K. Rawls, J. W. Wells, M. Hudetz, R. Jain, and M. F. Ulloa

Novartis Crop Protection, Vero Beach, FL., Greensboro NC., Basle, Switzerland and Sugar Farms Cooperative, Loxahatchee, FL.

CGA-362622 [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 Novariis Crop Protection is developing for use in sugarcane and cotton. It has been field tested as a 75% water dispersible granule for the past several years in North America, South America, Africa, and Asia under the code name CGA-362622. The proposed common name is trifloxysulfuron sodium.

CGA-362622 will offer control of dicotyledonous and monocotyledonous weeds and sedges. It is highly active on several important weeds in sugarcane, including yellow nutsedge (*Cyperus esculentus*), purple nutsedge (*Cyperus rotundus*), pigweed species (*Amaranthus* spp.), horse purslane (*Trianthema portulacastrum*), momingglory species (*Ipmea* spp.), cocklebur (*Xanthium* sp.), spurges (*Euphorbia* spp.), alexandergrass and signalgrass species (*Brachiaria* spp.), and panicum species (*Panicum* spp.). In sugarcane (plant and ratoon cane) up to 30 g ai/ha of CGA-362622 can be applied post-over-the-top, depending on location and cultivar without negative effects on crop tolerance. For optimum post-emergence activity, the addition of NIS is recommended at 0.25% v/v. The very low use rate of 15 to 30 g ai/ha together with its favorable toxicological, ecotoxicological and environmental properties make CGA-362622 an excellent tool for sugarcane farmers. CGA-362622 is readily absorbed by shoots and roots and is readily translocated in weeds. Susceptible weeds are inhibited following an application of CGA-362622 with complete death occurring within 1 to 2 weeks after application.

CGA-362622 is compatible with other herbicides including AAtrex® and Evik®. The combination of CGA-362622 and AAtrex® can be used to increase the weed spectrum and length of control. CGA-362622 can be applied in combination with Evik®, post-directed only, to increase speed of activity and weed spectrum, especially the grasses. Crop tolerance to Post applied CGA-362622 is generally excellent, but there is some variation in tolerance among varieties.

## Weed and Sugarcane Response to CGA 362622: A Potential Herbicide for Louisiana

## E. P. Richard, Jr. Sugarcane Research Unit, USDA-ARS

Houma, LA

In Louisiana, interference from a broad spectrum of grass and broadleaf weeds can have a major impact on cane and sugar yields. To control these weeds, the industry depends on multiple applications of preemergence (PRE) herbicides from primarily the dinitroaniline and triazine classes of chemistry. The impact of the continued use of these herbicides on both the development of weed resistance and the environment concerns the industry. In an attempt to alleviate these concerns, field studies were conducted to evaluate the potential use of preemergence (PRE) and postemergence (POST) applications of CGA 362622, a member of the sulfonylurea class of herbicides, for the control of seedling johnsongrass, itchgrass, momingglory, and nutsedge within the sugarcane crop.

Johnsongrass control 28 days after treatment (DAT) with CGA 362622 at 0.21 to 0.43 oz ai/A was similar regardless of whether CGA 362622 was applied PRE (58%) or POST (68%). Johnsongrass control with CGA 362622 PRE was equivalent to the levels of control obtained with a standard application of atrazine, but lower than the levels of control obtained with diuron, metribuzin, and pendimethalin applied at standard rates. When applied POST, control with CGA 362622 was equivalent to the control obtained with diuron and better than the control obtained with standard applications of atrazine, metribuzin, and pendimethalin. Morningglory (red and entireleaf) control 28 DAT with CGA 362622 was equivalent to the control obtained with the standards, especially when applied POST at 0.43 oz/A. Purple and yellow nutsedge control with CGA 362622 PRE was equivalent to the control obtained with metribuzin and superior to the control obtained with diuron, atrazine, and pendimethalin. As a POST treatment, nutsedge control was observed in one study and appeared to be greatest (57 to 90%) when CGA 362622 was applied PRE.

Some sugarcane injury in the form of a chlorotic band on leaves that were still part of the whorl at the time of treatment was observed by 14 DAT. The degree of chlorosis depended on rate, environmental conditions at the time of treatment, and cultivar. Injury ranged from 0 to 28% by 28 DAT and was greater for the cultivar 'CP 70-321' than for the cultivars 'LCP 85-384' and 'HoCP 85-845'. Crop injury from CGA 362622 applications of less than 0.86 oz/A did not result in significant reductions in sugarcane stalk numbers or heights for all years and cultivars evaluated and in sugar yield in one mechanically-harvested study in 1999 planted to 'LCP 85-384' sugarcane.

The labeling of CGA 362622 for sugarcane would provide the Louisiana sugarcane grower with an additional class of chemistry that provides POST and in some cases PRE control of seedling johnsongrass, morningglory, and nutsedge at rates that would pose a minimal threat to the environment. Because of the difficulty in controlling these weeds POST with currently labeled herbicides, growers apply PRE herbicides in the spring at relatively expensive rates in the anticipation of weed development. With CGA 362622's POST activity, growers could eliminate this application and would have to apply a herbicide treatment only if the weeds develop. Before CGA 362622 can be recommended for use in Louisiana sugarcane, additional information will be needed regarding the influence of weed size and application timing on CGA 362622's efficacy.

## Effect of Harvest Method and Storage Time on Sugarcane Deterioration

**G. Eggleston<sup>1</sup>, B, L, Legendre<sup>2</sup> and C. Richard<sup>3</sup>** <sup>1</sup>Southern Regional Research Center, USDA-ARS, New Orleans, LA <sup>2</sup>Sugarcane Research Unit, USDA-ARS, Houma, LA <sup>3</sup> American Sugar Cane League, Thibodaux, LA

The recent increase of billeted cane being mechanically harvested in Louisiana has often meant an increase in deteriorated cane being processed. Some of this deterioration in cane quality, i.e., the increase in associated trash (i.e. leaves, tips and field soil) is not necessarily a function of the newer harvest method, per se, but rather a function of mechanical harvesting in general. Further, there is the occurrence of sugar destruction in the cut cane between harvesting and crushing, regardless of the harvest system. There is a real need to establish new criteria for levels of deterioration in Louisiana cut cane, in order to better predict: 1) the quality of the cane to be processed and, 2) the effect of harvest methods and storage conditions. In this study, there were eight cane supply treatments, with samples taken on each day for four consecutive days (0, 24, 48 and 72h) before laboratory milling. Treatments included handcut (control) net, green and burnt standing whole-stalks taken from field plots each day. Soldier harvested burnt, green, and stored burnt whole-stalks were chosen to simulate cane from a heap or transloader stack each day. Burnt and green billeted cane were also taken to simulate cane from a billet wagon each day. Initial color for all cane treatments was associated with leaves and tops; color formed dramatically in the burnt billeted cane with storage time. Glucose and fructose were consistently greater in billeted than whole-stalk cane. Dextran and oligosaccharide formation was also greater and more rapid in billeted cane than whole-stalk cane, and concomitant with a decrease in pH. Billeted cane deterioration occurred earlier than in whole stalk cane, with burnt billeted cane deterioration more rapid and extensive than in green billeted cane. Oligosaccharide formation in cane is described, with emphasis on both kestoses and dextran oligosaccharides. Optimum postharvest handling conditions to minimize grower and factory losses are discussed.

#### The Effect of Harvest Method and Plot Size on Sugarcane Yield

K. P. Bischoff, K. A. Gravois, H. P. Schexnayder and G. L. Hawkins Louisiana State University Agricultural Center, Sugar Research Station

St. Gabriel, LA

For years, the accepted means of collecting sugarcane plot weight data was utilizing the whole stalk or "soldier" type harvester and a tractor mounted weighing device with a load cell. As the Louisiana sugar industry moved toward the combine harvesting system, Louisiana sugarcane researchers began work toward adapting to combine harvesting. This study was initiated in 1999 to compare plot yields obtained with whole stalk and combine harvesting systems and estimated yields. This study not only investigated method of obtaining plot yields, but also the effect plot size may have on the accuracy of yield data.

The two varieties used (LCP86-454 and HoCP85-845) were selected because of their erectness and better than average harvestability. This would provide a best case scenario for

harvesting methods. Plots were set up based on the various sizes of plots used throughout the stages of the Louisiana "L" Variety Improvement Program. Sixteen foot, single row (six foot wide row) plots are used in the second line trial stage through the nursery stages of the program. Two row, twenty-four foot plots are now being used in infield tests, while three row thirty-two foot plots are the outfield test plot sizes. The test was replicated three times. Stalk counts were made in all plots during the first week of December. In the combine harvested plots, ten-stalk samples were hand cut, and stripped of all leaf material. Each sample was weighed to obtain mean stalk weight and then measured to obtain stalk height. The samples were then milled to obtain Brix and pol values for estimating theoretical sugar per ton of cane. The 16 and 32 foot plots were soldier harvested and weighed with the traditional tractor mounted weigh rig. Ten stalk samples were taken from the heap row fro each soldier harvested plot. These samples were also weighed, measured, and milled for juice analysis. Four plot sizes (single row 16 foot length, single row 20 foot length, two row 24 foot length, and three row 32 foot length) were combine harvested. The combine harvested plots were weighed with a small self-dumping weigh wagon (three load cells), which was manufactured by Cameco Industries, Inc. Estimated cane yield was obtained as the product of stalk number per acre and mean stalk weight.

The data indicated that cane yields obtained from the whole stalk soldier harvesting method were significantly lower than cane yields obtained from estimated cane yields. The soldier harvesting method also had significantly lower stalk weight and stalk height and significantly higher sugar per ton of cane. This is a result of a low topping height even though the harvester topping height was set at its maximum setting. Cane yields from combine harvested plots, regardless of plot size, were not significantly different from estimated cane yields. For estimated cane yields, standard errors for cane yield decreased as plot size increased up to the two row 24 foot length, but increased again at the three row 32 foot length. As a result, the Louisiana "L" Variety Improvement Program increased its nursery plot sizes from 16 feet to 20 feet and changed its infield plot sizes to two row 24 foot length.

## Efficiency of First Clonal Trial Selection using Different Plot Sizes

S. B. Milligan<sup>1</sup>, M. Balzarini<sup>2</sup>, K. A. Gravois<sup>3</sup> and K.P. Bischoff<sup>3</sup> 'United States Sugar Corporation, Clewiston, FL <sup>2</sup>Dept. of Agricultural Science, National University of Cordoba, Cordoba, Argentina <sup>3</sup>Sugar Research Station, LSU Ag. Ctr., Baton Rouge, LA

Personnel in the LAES sugarcane variety development program select about one-third of 3000 first-clonal genotypes to advance to the second clonal trials. First clonal plots are planted using two stalks into single-row plots 1.82 m long. Second clonal trials are planted with six stalks into single-row, 4.88 m long plots. The larger plot is expected to provide a more effective basis for selection since end-row effects and sample size constraints of smaller plots tend to inflate sampling error.

The relative selection efficiencies of the different plot sizes have not been quantified. To measure such efficiencies we planted 28 randomly selected first-clonal genotypes into 1.82 m (2

stalks), 3.35 m (4 stalks) and 4.88 m (6 stalks) single-row plots. A replicated split-plot treatment arrangement with plot size as the main plot was planted in two locations and two years.

The error variance of the 1.82 m plots provided significantly larger error variances than the larger plot sizes for sugar yield, cane yield and stalk number. Stalk weight and sugar content error variances were not significantly affected. The proportion of the original population that needed to be selected to retain the top one percent of the population with 95% confidence improved with larger plot size but was still quite high for sugar yield (99% for a 1.82 m plot to 74% for a 3.35m plot).

The confidence of retaining the top one percent of the population by selecting the top third of the population for sugar yield was increased 33% from 50% confidence for a 1.82 m plot to about 66% confidence for the two larger plot sizes. The other yield component increases in selection confidence varied from an 8% increase for stalk weight to an increase of 28% for cane yield.

Maintaining the same confidence in selection associated with the 1.82 m plot but using the larger plots, breeders need advance less than half the number of first clonal plots. If the resources saved were used to replicate the second clonal stage, the selection process from cross to release could be shortened by a year. However, the additional resources to plant the larger first clonal plots need to be considered. Using the 3.35m plot would be most efficient. A further consideration is that the selection effectiveness for sugar yield at the first clonal stage is quite low. The best course may be to maintain the 33% advancement rate from the first to second clonal stages but use a 3.35 m first clonal plot size.

#### Analysis of Resource Allocation in Final State Sugarcane Clonal Selection

## J. S. Brown and B. Glaz USDA-ARS, Sugarcane Field Station Canal Point, FL

The objective of this study was to assess the allocation of resources in the final selection phase of clonal development of the Canal Point, Florida, sugarcane breeding program. Agronomically superior genotypes with adequate pest resistance must continue to be developed with the current level of resources while selection criteria change and expand. Analyses were conducted on elite genotypes from four selection series using data collected from 1992 through 1998. Each selection series had two planting sequences. To observe relative magnitudes of sources of variation, variance component analyses were performed. Estimates of the experiment-wise %CV, genetic repeatability, and  $R^2$  values were calculated as metrics of experimental precision. The source of variation next to experimental error, especially in the first planting sequence. The contributions to variance due to genotype x crop and genotype x location interactions were low, through genotype x location interaction cannot be discounted in clonal release decisions. Reducing replications from eight to four did not compromise experimental precision. Removing the second planting sequence would also further enable redirection of resources toward more beneficial ends. These changes

on genotypic performance or on genotype x environment interaction. Resources would also be available for testing P uptake efficiency and high water table tolerance.

#### **Responses of Sugarcane Plants to Water Tables**

## P. Y. P. Tai, J. D. Miller, B. Glaz, and D. Morris USDA-ARS Sugarcane Field Station

Canal Point, Florida

Waterlogging affects sugarcane growth and can cause a significant reduction in productivity. Tolerance to high water table, however, is found to be present in Saccharum germplasm and commercial sugarcane cultivars. This increased level of waterlogging tolerance is desired under some sugarcane production environments. Two experiments were conducted to assess the responses of physiological and morphological traits to varving depths of water tables on cold-tolerant clones and commercial sugarcane cultivars. Three commercial cultivars and nine cold-tolerant clones derived from crosses between commercial cultivars and S. spontaneum were used in these studies. Single-budded cuttings of each clone were transplanted to 19-gallon plastic cans of sand and organic soil mixture (1:2) in July 1997 (Experiment 1) and April 1999 (Experiment 2). Water tables were maintained at three levels, 0, 15, and 30 cm from the bottom of the can by dripping irrigation three times a day from mid-August 1997 to the time of harvest in February 1998 (Experiment 1) and from mid-May 1999 to February 2000 (Experiment 2). A split-plot design was used with three blocks in both studies. Tillering, growth rate, plant height, stalk number, stalk diameter, total stalk weight, dry root weight, Brix, sucrose, and purity were measured. In Experiment 2, the soil of can was divided into half, bottom (from 0 cm to 15 cm of the soil in the can) and top (from 15 cm to 30 cm of the soil in the can), for the measurement of roots. Combined analyses of variance of the two-year data were conducted for all traits. Data on weekly growth of plant height after being exposed to water-table treatments were fitted with an exponential growth curve. The results indicated that clones, water tables, and years affected the performance of all morphological and physiological traits except stalk number, whereas only clones and years influenced the performance of juice traits. There were no significant interactions between clones and water tables in tillering, stalk number, stalk diameter, growth rate, and plant height, but there were significant interactions between clones and years for all these traits except stalk number. The high water table (flooding) treatment (30 cm) had the lowest mean performance in all characters compared to the low and intermediate water tables, The exponential growth curves from clone to clone and from year to year. The significant interaction between clone and water table in total dry root weight indicates that differences in the responses among clones varied over water tables. Among the three water-table depths, the 0-cm water table had the most total dry root, the 30-cm water table had the least total dry roots, and the 15-cm water table was between these two water tables. The 30-cm water table produced a more marked shallow rooting than did either 0-cm or 15-cm water table. Cold-tolerant clones did not show superior performance over the commercial clones under the high water table treatment. The results indicate that, with these conditions imposed by the use of 10-gallon plastic cans, a water table at 30 cm could effectively measure the clonal response to a stress. The information obtained from this study could benefit the development of increased stress tolerance to high water tables in sugarcane through breeding approaches.

#### Soaking of Sugarcane Stalk Sections in Water before Planting

## B.Glaz USDA-ARS Sugarcane Field Station Canal Point, FL

Complementary objectives for preserving profitable sugarcane (interspecific hybrids of Saccharum spp.) production in the Everglades Agricultural Area (EAA) are to reduce rates of organic soil subsidence, reduce rates of P export to natural areas, and to increase water storage for environmental, urban, and agricultural uses. Maintaining or improving sugarcane vields with cyclic flooding would help achieve these objectives. The purpose of this study was to evaluate the effects on shoot emergence of soaking sugarcane stalk sections in water before planting. Ten genotypes, three stalk sections, and several stalk-storage durations were investigated at Canal Point on a Torry muck soil. Stalk sections were either stored aboveground on drained soil or immersed in water in plastic pools. Mean water temperatures ranged from 18° C in the mornings to 28° C in the afternoons. Soaking from 2 to 12 days caused more and faster emergence than not soaking. Four days of drained storage followed by 4 days of soaking resulted in improved emergence similar to that from other soaking treatments. Pre-plant soaking caused improved shoot emergence of four genotypes, and did not significantly affect emergence of six genotypes. Drained storage of 0 to 6 days resulted in more emergence than drained storage of 8 days. Emergence from the upper stalk section was more consistently high than from the middle or bottom stalk sections. The next research step will be to evaluate the effects on shoot emergence of flooding fields of newly-planted sugarcane.

#### Sugarcane Yield Following Long-Term Exposure to High And Flooded Water Tables

## J. D. Ray, T.R. Sinclair, and B,S. Glaz USDA-ARS Stoneville, MS. Gainesville, FL, and Canal Point, FL

Environment concerns in south Florida have raised the possibility that past management practices of pumping excess rainfall from sugarcane fields will be limited. This research was undertaken to investigate the consequences on sugarcane yield of long-term exposure to a high water table (15 cm below the soil surface) and to flooded conditions (2 cm above the soil surface). Individual plants of two cultivars (CP72-2086 and CP70-1133) were grown outdoors in large pots (25 cm diameter x 60 cm tall) and each of the water treatments were imposed on three dates (4 April, 11 June, 26 My). There were no significant differences between the two cultivars in their responses to the treatments. Weekly counts of leaf numbers showed that water treatments had no influence on the rate of leaf production relative to the control plants (water table at 50 cm). At the final harvest (22 November), however, there were significant differences in stalk weight among treatments. The flooded conditions resulted in greatly decreased stalk weight with the decrease being progressively grater with increased duration of flooding. Harvested stalk weight for the 15-cm water table was actually greater than the control for the treatment where treatment was initiated on 4 April and 11 June. The stalk weight for the 26 July treatment was equivalent to the control Brix and pol, and consequently theoretical recoverable sugar, were not different among treatments. Consequently, these results showed that sustained flooding was deleterious to sugarcane yield but that a sustained

water table held at a 15-cm depth resulted in no yield loss compared to a sustained water table at a 40-cm depth.

## Sugarcane Cultivar Response to Two Summer Water-Management Strategies in the Everglades

## B. Glaz<sup>1</sup>, S. Edme<sup>2</sup>, J.D. Miller<sup>1</sup>, D. G. Holder<sup>2</sup>, and S. Milligan<sup>2</sup> <sup>1</sup>USDA-ARS Sugarcane Field Station, Canal Point, FL <sup>2</sup>United States Sugar Corporation, Clewiston, FL

Producers of sugarcane (interspecific hybrids of Saccharum spp.) in the Everglades Agricultural Area (EAA) are seeking to maintain or improve profits and exemplify positive environmental stewardship. Maintaining or improving sugarcane yields with more of the soil profile wet for longer periods would help achieve these objectives. The purpose of this study was to evaluate the effects of two water-management strategies on nine sugarcane cultivars from the plantcane through the second-ratoon crops. Experiments were planted in two fields in February 1997 and again in the same two fields in February 1998. The management strategy of Field 1 was to maintain its water at or above 15 cm below the soil surface; the strategy for Field 2 was at or above 38 cm. Water treatments were intended to be imposed from 1 June through 1 October from 1997 through 1999. Target water levels were achieved in Field 1 for 40 days in 1997, 63 days in 1998, and 56 days in 1999. Target levels were achieved in Field 2 for 48 days in 1997, 105 days in 1998, and 50 days in 1999. Mean water levels below the soil surface from 25 July 1997 to 1 Oct. 1997 were 13.4 cm in Field 1 and 29.0 cm in Field 2; from 1 June 1998 to 1 Oct. 1998 were 17.1 cm in Field 1 and 29.0 cm in Field 2; and from 15 June to 1 Oct. 1999 were 17.9 cm in Field 1 and 35.8 cm in Field 2. Both experiments were harvested in the plant-cane and first-ration crops, and the first experiment was also harvested in the second-ration crop. Over all cultivars, crops, and years, the sugar per hectare yield of Field 1 was 91.6% that of Field 2. CP 72-2086 and CP 82-1172 generally had similar yields, regardless of water treatment. CP 80-1827 and CP 81 -13 84 were also relatively unaffected by water treatments in most years and crops, but each of these cultivars had one low sugar per hectare yield on Field 1. CP 73-1547 and CP 78-1628 had variable responses to water tables. Consistently across years and crops, CP 80-1743 in particular, and CP 70-1133 to a lesser degree, had higher yields on Field 2 than on Field 1. CP 85-1308 yielded more sugar per hectare on Field 1 than on Field 2 in two of five harvests; no other cultivar achieved equal success on Field 1. Using the mean of all five harvests in this study, CP 80-1743 yielded only 74.7% as much sugar per hectare on the "wetter" as on the "drier" field. Conversely, CP 72-2086 had equal sugar per hectare yields on both fields. These data are not offered as commercial recommendations because they are from only one location. However, this research suggests that for genotype selection programs, there maybe substantial shortterm economic benefits by routinely testing for yield responses to varying water tables.

## Photosynthesis Characteristics in Eleven Cultivars of Sugarcane and Their Responses to Water Stress During the Elongating Stage

## R Chen, G. Li, and M. Zhang

Key Lab of Sugarcane Genetics and Breeding under Ministry of Agriculture, Fujian Agricultural University, Fuzhou, P. R. of China

Photosynthesis characteristics were determined with attached leaves of nine new promising varieties and two widely cultivated cultivars in CHINA at the different age of field-grown or different water stress intensity of pot-grown plants during the elongation of sugarcane. On the basis of these data, water use efficiency and leaf conductance were calculated. The highest photosynthesis, namely net photosynthetic rate (Pn), Hill reaction and photophosphorylation rate, transpiration rate (E) and leaf conductance (gs) were found in the early stage of elongation. Light saturation point and maximum photosynthetic rate were quite different in the tested varieties. FA81-745, YT 86-368 and CT81-548 performed no evident light saturation and kept higher Pn during the elongating stage. The other varieties reached their saturation at PAR of 1000µmol Photons.m<sup>2</sup>. s<sup>-1</sup> or more. However, water use efficiency, measure of Pn/E, were highest in July 3 and Sep. 11, resulting from the interplay of Pn and E. The results here also showed leaf Pn, water use efficiency significantly declined under water stress, though E and gs kept slight increase in the mild water stress. From the above results, it was considered that higher water use efficiency was more dependent on higher photosynthesis rather than lower transpiration when sugarcane were subjected to mild water stress. YT86-368 and FA81-745 kept higher Pn and WUE in the short-term of water stress, resulting from the strong resistance to water stress, which were implied by the small increment of MDA content after water ceased

## Characterization of Possible Sulfur Sources within the Everglades Agricultural Area

#### T, J. Schueneman

University of Florida Belle Glade, FL

Public officials were quoted as attributing high levels of methyl mercury (MeHg) in Water Conservation Area (WCA) 3A to agricultural sulfur (Ag-S) use in the Everglades Agricultural Area (EAA). The sulfur use rate claimed was 4,000 lbs/A/yr, which would amount to 1.1 million tons/yr for the 550,000 acres farmed in the EAA. There is not a published budget for sulfur in the EAA. **Objective:** Assemble and publish current estimates and factual data on S sources for the EAA. **Methods:** University of Florida Institute of Food and Agricultural Sciences (IFAS) recommendations were reviewed, growers on high pH soils were interviewed, and the fertilizer distributors servicing the area were contacted to characterize Ag-S use. Literature was reviewed to determine size of the sulfur pool resulting from soil mineralization, rainfall, and Lake Okeechobee water used for irrigation, well-field recharge, and natural-lands maintenance. **Results:** Ag-S is recommended by IFAS for soil pH adjustment. Rates are 500 lbs/A for pHs over 6.6 for a sugarcane crop (3-4 years) and 500 lbs/A per 0.2 pH unit reduction desired, up to 4,000 lbs/A/year, on vegetable crops. This could result in 15,000 tons applied to sugarcane fields and half this amount on

vegetable fields. Sugarcane growers reported applying sulfur on high pH soils to every other crop (6-7 years), for an annual rate of 33 lbs/A, or 3,300 tons. Vegetables growers reported the industry practice was not to use sulfur to lower pH, but rather to apply soluble micro-nutrients to crops. Total annual Ag-S sales were estimated at 10,400 tons, which extrapolates to 37 lbs/A for the entire region including the EAA. Soil subsidence was found to contribute nearly 60,000 tons of sulfur, or 217 lbs/A, to the environmental load annually. Rainfall, Lake Okeechobee water, and chemical carriers were less important sources. Conclusions: The Ag-S application rate of 4,000 pounds per acre per year for the entire EAA, as stated publicly by both South Florida Water Management District and the United States Geological Survey, was more than 100 times too high. Natural sources of sulfur contribute at least6 times more S to the environment than grower applications.

#### Si as Macronutrient for Sugar Cane

V. V. Matichenkov, G. H. Snyder, and D. V. Calvert University of Florida, Indian River Research and Education Center Ft. Pierce, FL

A number of field and greenhouse studies have clearly demonstrated that Si is an important macronutrient for sugar cane. Effective management practices utilize Si fertilization on soils deficient in plant-available Si. This far, the mechanism of the direct effect of Si fertilizers on sugarcane is not as sufficiently advanced as it is for rice. The content of Si in cultivated plants ranges from 0.3 to 8.4%. a range of 210-224 mln. tons of Si or 70-800 kg of plant-available Si are harvested with crop from arable soils annually. Crop removal of Si by sugar cane varies from 0.1 to 3.2%. Greater yield of sugarcane is attended with higher content of Si in the leaves. In field and greenhouse experiments if Florida, Hawaii and Mauritius demonstrated that application of Si fertilizers has a positive effect on the disease-, pest-, and frost-resistance of sugar cane. It has been shown that sugar cane productivity increased from 17 to 30%, whereas the production of Su is its the stimulation of the plants defense abilities against abiotic and biotic stresses. Improved sugar cane nutrition brought about by fertilization with Si is shown to reinforce the plants protection properties against leaf freckle, sugarcane rust and sugar cane ringspot. In addition, Si fertilization has a more positive effect than liming on the chemical and physical soil properties.

#### Plant-Available Si Status of Louisiana and Florida Soils

B. Ande, E. A. Bocharnikova, D. V. Calvert, and V. V. Matichenkov\* University of Florida, Indian River Research and Education Center Ft. Pierce, FL

Si is an integral part of plants. Plants absorb Si in the form of monosiiicic acid or its anion. The content of monosiiicic acids in the soil depends on the numerous factors (mineral composition, pH level, plant cover, climatic conditions). Soluble monosiiicic acids can be transformed into polysilicic acids or amorphous silica and absorbed by minerals or microorganisms. Various forms of Si (monosiiicic acids, polysilicic acids, biogeochemically active Si) were tested in some soils of Louisiana and South Florida. Soil samples were collected in the surface (0-20 cm) and subsurface

(20-40 cm) horizons. The sandy soils in Florida (Entisols, Spodosols and Alfisols) were extremely low in monosilicic acids and biogeochemically active silicon compounds. The Louisiana soils were characterized by low or medium content of monosilicic acids and acid-extractable Si. The deficient Si nutrition may be responsible for reduced sugar cane productivity. The data obtained demonstrate that investigated soils could benefit from silicon fertilization thereby improving soil chemical and physical properties and optimization of Si plant nutrition.

## The Effect of Pro-Sil on P Leaching in Sandy Soils

## B. Ande, P. Ande, E. A. Boeharnikova, D. V. Calvert, and V. V. Matichenkov

University of Florida, Indian River Research and Education Center Ft. Pierce, FL

Phosphorus (P) contamination of natural surface and subsurface waters draining from agricultural soils is an acute environmental and economical problem in Florida. Our preliminary studies showed that soil treatment with Pro-Sil, a commercial fertilizer containing Si-rich materials, can reduce P leaching by 30 to 90% while allowing P to remain in plant-available forms. The comparison of Pro-Sil and lime on P leaching from cultivated Spodosols, Entisols and Alfisols was studied in column and greenhouse experiments with Bahia grass grown under various levels of P fertilization. The results obtained showed that Pro-Sil reduced P leaching considerably more than lime in all soils investigated. Lime transformed plant-available P into plant-unavailable forms, while Pro-Sil kept P in plant-available form. As obvious from the results of the greenhouse experiment, plant growth response was more significant from Pro-Sil soil treatment than that from P fertilization. Also, Si fertilizer had a positive effect on the evolution of Bahia grass root system. The application of Pro-Sil to sandy soils could improve plant nutrition, increase sugar cane productivity and protect natural waters from P contamination.

## Effect of Urea Nitrogen Rates, A Nitrogen Stabilization Package, Winter Vs Spring Nitrogen Fertilization, and Varieties on Sugarcane Yields

W. B. Hallmark<sup>1</sup>, G. J. Williams<sup>1</sup>, L. P. Brown<sup>1</sup> and G. L Hawkins<sup>2</sup>

<sup>1</sup>LSU Agricultural Center, Iberia Research Station, Jeanerette, LA <sup>2</sup>LSU Agricultural Center, Sugar Research Station, St. Gabriel, LA

Sugarcane in Louisiana is usually fertilized in April or May. However, urea is 10-15% cheaper when it is purchased in the fall or winter. Due to the high amounts of clay in most of Louisiana's sugarcane soils, water is frequently trapped in the furrows between sugarcane rows after harvest (especially when sugarcane is harvested under wet conditions so that the fields are rutted up and drainage ways are not reopened). If liquid urea could be stabilized (by using a urease or nitrification inhibitor) and mixed with liquid calcium chloride it may be possible to add nitrogen between sugarcane rows (in the furrows) in a narrow band in the winter after harvest. This could improve water drainage through the effect of calcium and ammonium in improving the permeability of the soil to water movement so that sugarcane yields are increased. Objectives of the research (on a Baldwin silty clay loam soil) were to determine the effect of: 1) nitrogen-stabilized liquid urea on sugarcane yields when applied in the winter vs. the spring; and 2) varieties and nitrogen fertilizer rates on sugarcane yields. Research across a two-year study (CP 70-321 plant and first stubble cane) showed that adding 135 or 202 kg N/ha (as liquid urea) with a nitrogen stabilization package (calcium chloride plus a urease inhibitor) in December of 1997 and 19098 had statistically (P>0.10) equal and numerically higher (P>0.25) sugar yields compared to where urea was side-dressed the following May. Results also showed that variety LCP 85-384 yielded higher than varieties CP 70-321 and HoCP 85-845 at four urea nitrogen fertilizer rates (67, 112, 157, and 202 kg N/ha) with plant cane. Also, adding liquid urea nitrogen (containing calcium chloride and a urease and nitrification inhibitor (to row furrow of HoCP 85-845 and LCP 85-384.

#### Fertilizer Effects of Older Sugarcane Ratoon Crops in Louisiana

#### C. LaBorde, K. Gravois, and K. Bischoff

Sugar Research Station, Louisiana State University Agricultural Center St. Gabriel, LA

Louisiana sugarcane (Saccharum spp.) farmers are currently undergoing a trend change in the ratoon longevity of their sugarcane crops. For the last several decades in Louisiana, the intensity of ratooning in sugarcane has usually been a plantcane crop plus two ratoon crops. Since its release in 1993, LCP 85-384 has surpassed the expectations of the Louisiana sugar industry, with one of the reasons being its excellent and increased ratooning ability. The main objective of this study was to determine the response of LCP 85-384 to higher than recommended rates of nitrogen in third and fourth ratoon crops. A secondary objective was to determine potassium and phosphorus responses. Fertilizer treatments were evaluated at two locations during 1998-1999: Blackberry Farms near Vacherie, Louisiana and Triple V Farms near Youngsville, Louisiana, The experiment was planted as a Latin square design with six fertilizer treatments. The year by treatment interaction was not significant for either location; therefore, treatment means were averaged across the two years. For the Vacherie location, cane yield was the only yield parameter significantly affected by the treatments. Orthogonal contrasts indicated a significant increase in cane yield between the medium (168 kg ha<sup>-1</sup>) and high (224 kg ha<sup>-1</sup>) nitrogen rate. Potassium significantly increased cane yield between the low (0 kg ha<sup>1</sup>) and high (134 kg ha<sup>-1</sup>) rate. For the Youngsville location, cane yield was not significantly increased by the higher nitrogen rates. Orthogonal contrasts indicated a significant increase in both sugar yield and cane yield due to potassium between the low (0 kg ha<sup>-1</sup>) and high (134 kg ha<sup>-1</sup>) rate. For fourth ratoon cane on light to medium textured soils of the Mississippi river, the higher nitrogen rate increased cane yield over the medium nitrogen rate. The third ration LCP 85-384 cane at Youngsville exhibited no response to higher nitrogen rates. Potassium increased cane yield in both soil types, whereas phosphorus did not produce a significant cane yield response in both soil types.

## A Summary of Precision Farming Results for South Florida

#### T. Murray, R. Price, and M. Loekhart Lockhart Ag Technologies Lake Harbor, FL

The increase in sugarcane acreage in south Florida during he past ten years has been mostly to sandier soils which are west of the Everglades Agricultural Area. Aerial infrared photographs of the sugarcane in this area have demonstrated a high degree of variability in the photosynthetic activity of the crop. There are multiple reasons for reduced plant vigor in some areas of these fields. For example, statistical analysis of more than 800 sand land samples collected in central Hendry County has shown a high correlation among soil pH, Si, Ca, Mg, organic matter, K, and P. During the past four years, thousands of acres of sandy soils in south Florida have been sampled using precision farming techniques. It has been determined that some flatwood soils of central Hendry County require a sampling intensity of one sample per acre to sufficiently identify the variability of soil pH and available silicon. Soil pH values have been shown to routinely vary from 3.8 to 8.0 within the same field. Laboratory experiments have been conducted which show that mixing equal volumes of soil from high and low pH areas can result in unpredictable results for the composite sample. Multiple sites within selected fields have been soil sampled and aerially photographed over a three-year period. Using these tools, we have tried to verify the effectiveness of the variable rate application to improve the uniformity of soil pH and overall crop vigor. Liming rates for the acidic areas of the fields now far exceed the present two tons per acre lime recommendation from the University of Florida. Lime calibration studies are presently underway on both the west and east sides of Lake Okeechobee. In one experiment, the soil Ph in the treatment receiving ten tons of dolomite per acre increased from 4.0 to 6.3 a year after application.

#### Application of Precision Agriculture to Target Areas of Poor Sugarcane Yields on Mineral Soils

#### R. M. Muchovej, J.J. Mullahey, and M. Lockhart

University of Florida Immokalee, FL

Sugarcane growers have repeatedly observed that sandland fields exhibit spots of poor growth and development. The high variability presented by these mineral soils warrants intensive water, fertility, and crop management for profitable growth of the crop. These poor areas, adjacent to very productive sugarcane, can comprise up to 25% of a field. The reasons for these yield depressions are not thoroughly understood. Without knowledge on the causes of low yield and accurate maps of the location of these areas, application of corrective treatments to site specific areas of the field is not possible. The objective of this study was to investigate prevailing soil characteristics for a number of fields and determine a correlation matrix for these characteristics. This was accomplished by collection of a number of soil samples (in excess of 800) according to a grid, analyzing these samples chemically and utilizing the data for a multiple correlation analysis. Contour maps for the fields were also constructed based on the individual characteristics. Knowledge on geographical location of potentially low production areas and on factors that contribute to this variability, combined with new

technologies such as precision agriculture, could allow growers to have a more cost-effective and environmentally friendly production system.
# MANUFACTURING ABSTRACTS

# The Presence of Total Polysaccharides in Sugar Production and Methods for Reducing Their Negative Effects

## J, A. Cuddihy, Jr., M, E. Porro, and J. S. Rauh

Midland Research Laboratories, Inc. Lenexa, Kansas

A tremendous amount of research has been done on the various polysaccharides found in sugar cane and their effects on the processing and recovery of sugar. Polysaccharides found in the production process include those from the plant itself, which may be dependent upon plant variety and weather patterns, and those resulting from deterioration processes due to cane handling methods. While efforts to deter the effects of polysaccharides contributing to these production inefficiencies and losses. The practice of adding a specific enzyme to address a single polysaccharide often ignores the larger problem. A new methodology is presented for a treatment protocol that addresses the total polysaccharide problem. Also, a detailed discussion is presented on how total polysaccharides contribute to production losses, the economic importance of more closely tracking their presence, and taking appropriate actions to reduce their negative influence.

# Cane Juice Analysis by Near Infrared (NIR) to Determine Grower Payment at Sugar Cane Growers Cooperative of Florida

T. P. Johnson Sugar Cane Growers Cooperative of Florida Belle Glade, Florida

As applications of NIR analysis are increasing in the grain and food industries, more options are becoming available for analysis of incoming sugar cane in the factories. For the 1999-2000 crop season, Sugar Cane Growers Cooperative of Florida (SCGC), together with Florida Crystals Corporation, has adopted Near Infrared (NIR) spectroscopy as the standard method for cane juice analysis. This paper will include a brief history of cane juice analysis at SCGC as it has evolved from polarization using lead clarification to polarization using NIR spectroscopy. Experiences with data acquisition, equation development, and equation validation, necessary for implementing NIR analysis, will also be discussed.

## Strategies for the Expansion of Cane Sugar Mills

# P. W. Rein Audubon Sugar Institute, Louisiana State University Baton Rouge, LA

Faced with the need to crush more cane, sugar mills have various options open to them. This paper explores the possible strategies for improving capacity. The first step in each case is to maximize throughput and performance of existing equipment. The best options for expanding each section of the plant, namely extraction, clarification, evaporation, pan house activities and steam and power generation are discussed. Installation of new equipment can be undertaken in a way to simplify plant and reduce operation costs, particularly if adequate forward planning is possible. The trade off between capital costs, on-going maintenance costs and plant performance efficiencies is considered. Future options for making better use of capital assets are mentioned, including power islands for steam and power generation and off-season refining or syrup/molasses processing.

# Potential Sucrose Losses in Clarifier Mud and Mud Filtrate

## M. Damms and B. Anderson

United States Sugar Corporation Clewiston, Florida

It has been noted at several sugar cane factories in the United States that there is a considerable temperature drop across the mud filter station. There is potential for significant microbial sucrose losses associated with the low temperatures being attained. This paper describes findings at the Bryant Sugar Mill when an investigation was carried out to try and quantify these sucrose losses. Reference is made to similar findings in the South African Sugar Industry and what steps have been taken to minimize the microbial destruction of sucrose.

## Partition Coefficients for Aconitic Acid between Aqueous Solutions And Tertiary Amine Containing Solvents

A. G, Hill and M. K. Pampatwar

Department of Chemical Engineering, University of Louisiana at Lafayette Lafayette, LA

This paper will present preliminary data on the evaluation of partition coefficients of aconitic acid in two phase liquid-liquid mixtures of aqueous solutions and an organic phase containing tertiary amines. The tertiary amines will be blended approximately in a 15-20 percent by volume mixture with an organic diluent, 2-octanol. The two tertiary amines to be considered include triisooctylamine, (C8H17)3N, and Armeen 380, a commercial product of Akzo Nobel with the formula R3-N, where R = C8 - CIO alkyls. This data will assist in the evaluation of these solvents for use in recovering aconitic acid from molasses by solvent extraction.

# Factory Comparison of Cold and Intermediate Lime Clarification In Raw Sugar Manufacture

**G. Eggleston<sup>1</sup>, A. Monge<sup>2</sup>and A. Pepperman** <sup>1</sup>USDA-ARS Southern Regional Research Center, New Orleans, LA <sup>2</sup>Cora Texas Manufacturing Co., White Castle, LA

In Louisiana and Florida cold lime clarification remains the clarification process of choice. A comparative study of cold versus intermediate lime clarification was undertaken at a Louisiana factory, which currently operates intermediate liming (~3G% mixed juice of pH 5.2  $\pm$  0.3 [MJ] is preheated to 190-200°F to maintain clean lime juice heaters, mcubated at ~130°F, then limed), but still had the pipes to revert to cold liming (MJ incubated and limed at ambient temperature ~105°F) for this study. Hourly samples were collected over a six-hour sampling period across cold and intermediate clarification processes on two alternate days, respectively. The sampling periods for each process were repeated three times across the 1999 grinding season.

Although there were no marked differences in sample pH values and standard deviations (process control) between the two processes, clarified juice (CJ) and final evaporator syrup (FES) pHs were slightly higher in intermediate than cold liming. This led to less sucrose inversion occurring in intermediate liming, indicated by higher sucrose, and fewer glucose and fructose true purity values, as measured by gas chromatography. Because of the lower (by ~25°F) liming temperature in cold liming, -4.6% more lime was added than in intermediate liming; this caused no significant differences in CJ calcium contents.

For both processes ~10% color was removed in the incubator tank. Color (~10%) was also removed when 30% MJ was pre-heated in intermediate liming. This was offset by color formation on liming in both processes, because of the alkaline degradation of invert. However, overall more color was removed than formed in intermediate liming.

Major differences existed between the two processes for turbidity removal and control. Across the season there was 4.6% more turbidity removal (MJ to CJ) in intermediate that cold liming. Intermediate CJ turbidity (season av. 2028 ICU  $\pm$ 675) was approximately half of cold CJ turbidity (av. 3952 ICU  $\pm$  1450) with over twice as much better CJ turbidity control. This led FES turbidity values to be lower in intermediate (av. 4887 ICU  $\pm$  659) than cold (av. 6808 ICU  $\pm$ 1081) liming, with much better control.

For both processes, starch was reduced in the incubator tank because filtrate contains natural diastase enzyme from the cane. More starch was removed in intermediate liming when 30% of the MJ was pre-heated. More dextran was removed (~10%) in the pre-heated MJ, in intermediate liming. Some dextran may sometimes have formed in the incubator tank, in both processes. Settling performance was also reported. Summed across measured parameters, intermediate liming would appear to offer several advantages over cold liming.

# A Flexible Coupling for Sugarcane Mills - Its Design Conception and Performance

# L. C. Felicio Ribeirao Preto, SP, Brazil

Traditional mill couplings frequently cause serious problems to the mill driver components. Shaft cracks, flattened points of the shaft square surfaces and bearing deformations causing damages to the gears are the most usual troubles. Measurements of these coupling effects under actual operation condition show a significant bending moment (same magnitude of the torque) applied on the shaft square tips and an oscillatory bending tension having amplitude of 105 MPa.

In order to minimize those problems a new coupling was developed based on a mechanism similar to a crank and arm. The coupling uses four crank-arm arrangements such that a pair of cranks (like a fork) works with a pair of arms (like a "T") on each shaft. This configuration allows the shafts and the tail-bar to have lateral, axial and angular motions which cause a reduction in axial and bending moment loads.

This work analyzes the effects of the traditional coupling, explains the design conception of the new coupling and discusses its performance based on measured data. The most important results are a 78% reduction in the bending tension (from 105 MPa to 23MPa) and a decrease in bending moment to the level of 25% of the driving torque.

# Chromatographic Desugarization of Syrups in Cane Sugar Mills

# C. Baez-Smith, J, Alvarez

Sugar Cane Growers Cooperative of Florida V. Kochergin, M. Kearney, W. Jacob, L. Velasquez

Molasses desugarization has been widely applied within beet sugar industry for almost two decades. It has been proven to be a very economically feasible and powerful method of recovering additional sucrose in a sugar plant. The method, however, does not appear feasible for most applications within cane sugar industry. Sugar content is lower than in beet molasses, thus reducing potential benefits of chromatographic recovery. In addition, it is extremely costly and difficult to remove suspended solids and soften cane molasses from most sources.

A novel approach has been proposed making chromatographic recovery of sucrose from cane syrups look more attractive for cane sugar technologists. The process involves membrane filtration and softening of the clarified juice, evaporation and chromatography of the obtained syrup. The process has been piloted for several years in Sugar Cane Growers Cooperative of Florida sugar mill. Sugar recovery of 99% (based on feed material to the chromatographic separator) was achieved in the pilot trials. The extract with purity exceeding 98% was crystallized, resulting sugar satisfying the standards of white sugar. The effect of membrane filtration and chromatography on final sugar

quality is discussed. An alternative approach resulting in higher quality raw sugar coupled with higher overall recovery will be presented.

# Simulation of Vacuum Pan Sugar Crystalizer

J. Gamez and M. Saska JMG Programs Lewisville, TX

A beta version of a new software to simulate operation of batch sugar vacuum pans is presented, and the underlying principles, such as the dynamic heat and material balances, and crystallization and evaporation rates are discussed. The program consists of three subprograms, simulating respectively, the manual, semi-automatic and fully automatic pan operations. The program is well suited for training of pan operators and as a tool for consideration of various pan automation strategies and can interface with pan automation hardware.

# Fluorescence Technology and the Measurement of Sugar Contamination

# H. Herrera

Nalco

The purpose of the study was to monitor sugar contamination in the return condensate from the boiler feedwater, evaporators and pan condensate. The trials were also conducted in the cooling water application. This paper will present the results recorded from the instruments during the 1999 - 2000 season.

Fluorescence sensing is the operating basis of the unit. The water being monitored is passed through a polished, quartz tube which is known as the flow cell. An excitation light is directed through the flow cell and the emitted light is read by a photomultiplier. The quantity of light emitted is proportional to the amount of the fluorescing juice present in the sample.

The instrument is most commonly used for measurement of the fluorescent components inherent in the cane juice. The juices emit light intensely at a specific wavelength when excited with light at another specific wavelength. Therefore, within the unit excitation and emission filters are used which are specific to the juice being measured. The concentration of the juice in the boiler and cooling water therefore, is proportional to the measured fluorescence. The instrument controls the fluorescence set points, by opening the pumps and sending the water to the drain and avoiding the contaminations.

The instrument uses the technology of the fluorescent properties of non-sugars in the cane juice impurities to detect the presence of sugar. Each unit is identical for each mill, except for the excitation and emission filters are changed to suit the fluorescent properties of each mills cane juice. The instrument monitors the stream on-line and gives real-time detection of sugar leaks for the condensate and cooling water systems being sampled. The built-in data logger was used to record the fluorescence and temperature data. Each data point was logged as an average for 1 minute. The units used for the measured fluorescence of the impurities, were Relative Fluorescence Units (RFU).

Throughout the season, the sugar spikes were typical for most of the mills. However, the spikes were regularly peaking above 150 RFU when juice was detected in the sample. The frequency of spikes was related to the evaporator vessel changes that occurred in the mills or an upset in the steam demand that forced carryover into the clean water sample.

# A Preliminary Laboratory Evaluation of Two Published Methods of Saccharate Liming Vs Slurry Liming at Cora-Texas Sugar Factory

# A. Monge<sup>1</sup>, J. G. Spinoso<sup>1</sup>, and H. S. Birkett<sup>2</sup>

Cora-Texas Sugar Factory, Schaffer & Associates, Baton Rouge, LA <sup>2</sup>SchafFer & Associates, Audubon Sugar Institute

The calcium saccharate process is mainly used for the separation of sucrose from the nonsucrose constituents of beet molasses. It is extensively practiced in the beet sugar factories for increasing the yield of beet sugar beyond that obtainable in the basic diffusion-defecationcrystallization operations. In the cane sugar factories, Honig (1953) reports the usage of calcium saccharate instead of milk of lime for the treatment of cane juices as first proposed by Van Der Jagt in Java in 1929. Beet juices, containing practically no reducing sugars, can withstand high pH's and high temperatures, whereas cane juices cannot. Amatikulu in South Africa did extensive factory trials in 1983 and 1984 and decided not to adopt the saccharate liming process. Pineda, at Ingenio El Angel, El Salvador, Central America, implemented a variation of the South African method in 1998. (ATACORI, 12th Congress, 1998, pp. 122-124, Costa Rica). Laboratory tests were conducted at Cora-Texas to compare these two published methods of saccharate liming with conventional slurry liming at the end of the 1999 cane crop. The results were sufficiently encouraging for the South African method that further extensive evaluation will be conducted in the coming 2000 crop year. including the preparation of lime saccharate with 99.0 purity raw sugar in order to minimize the destruction of reducing sugars. Tests with 2nd year stubble, 1st year stubble and plant cane will be carried out, under both good and bad weather and cane quality conditions.

# The Internet: Toy or Tool?

## M. B. Inkson Sugar Knowledge International Limited

The Internet - a combination of the World Wide Web and electronic communication - has become a dominant force in certain aspects of our daily life but can it really be useful to us in the sugar industry and where is it likely to go next?

The 'web', as it is normally called, is a vast array of inter-connected databanks or 'sites'. Much of the data is frivolous and little is of direct use to the professionals of the sugar industry but what is available is certainly useful. Some of the sites are examined and it can be seen that they might perhaps be classified into three groups: professional resources, libraries of past papers and commercial, semi-sales, sites.

One of the difficulties of the web and its almost infinite nature is navigation. A fourth type of site is therefore also examined: the navigation aid, as specifically designed for sugar technologists. The usual aid is called a portal, a doorway through to other sites. Sometimes sites are just portals but more frequently they are combinations of portals and one of the previously described groups. The newest approach to navigation, still in its infancy, is the web ring which links related sites in an unbroken circle. There is now a sugar technology web ring.

There is little point reviewing email, now an essential tool for most of us. However one special aspect of the Internet, which combines electronic communication and the web, does need to be examined: specialist forums that allow us to share problems and solutions. The Sugar Technology forum, the most relevant one for the sugar industry, is examined.

Finally the future of the Internet with respect to sugar technologists is examined. The future is very difficult to predict - who would have even predicted the Internet ten years ago? - but what is clear is that, as sugar technologists, we need to participate in its development if we are to gain the maximum from it

## POSTER SESSION

## Progress on USDA-ARS Water Management Research Project: Sugarcane - Water Quality - Trafficability

T. S. Kornecki and J. L. Fouss USDA-ARS, Mid South Area Baton Rouge, LA

This new field research has been initiated to evaluate subsurface drainage of sugarcane land for improving water quality of runoff and soil trafficability when conducting field operations. Currently water quality of runoff from sugarcane land, and its potential of polluting downstream water resources, is a major concern of growers. During past harvest seasons when wet weather occurred, trafficability for the modern and heavier chopper harvester equipment was also a concern for the growers. This project is intended to evaluate the costs and benefits of subsurface drainage on sugarcane land in helping to alleviate both of these problems or concerns. Past research results on corn cropland has shown subsurface drainage reduced nitrate and pesticides carried in runoff. Additionally, the project may show the potential benefits of higher cane yields on subsurface drainage is provided to control water table depth during the wet winter months of the domant season.

The new project is located on the Louisiana Agricultural Experiment Station (LAES) Sugar Cane Research Station at St. Gabriel, LA. Three drainage treatments, replicated three times, were installed in the field site during April 1999: I - Surface Drainage Only (SDO); II - Conventional Subsurface Drainage (CSD); and III - Water Table Control (WTC). Surface drainage on the experimental site was provided by precision land grading, and the CSD and WTC treatment plots (each about 0.5 ac. in size) were also precision graded for surface drainage. All surface runoff from the plots is intercepted by special drainage ditches excavated on the site which are being equipped with flumes and instrumentation to measure and sample runoff events. All subsurface drainage lines are connected into sump structures since gravity outlets to ditches are not practical because of the frequent high water levels in the drainage ditches. Subsurface drainage water is pumped out of the sumps into surface drainage ditches or channels. Irrigation water can be pumped into the sumps to maintain water table depth by subirigation during drought periods, provided a source of irrigation water is available.

Surface runoff from the plots that is measured and sampled proportional to flow rate is analyzed for agrochemical (fertilizer and pesticide) content. The volume of subsurface drainage discharge (pumped from the drainage sumps) will be measured and also sampled proportional to volume for agrochemical analyses. Any sediment carried in runoff or subsurface discharge will also be quantified, and sediment will be analyzed for attached chemicals. Key trafficability quantifying data will include soil penetrometer readings and measurements of traction and slippage parameters on the ground surface during wet harvesting seasons. Potential delays in conducting harvesting operations with modern, heavier equipment may become more critical in future years because of increasing use of this modern harvesting equipment in the Southeastern United States.

# AMERICAN SOCIETY OF SUGAR CANE TECHNOLOGISTS EDITORIAL POLICY

### Nature of papers to be published:

Papers submitted must represent a significant technological or scientific contribution. Papers will be limited to the production and processing of sugarcane, or to subjects logically related. Authors may submit papers that represent a review, a new approach to field or factory problems, or new knowledge gained through experimentation. Papers promoting machinery or commercial products will not be acceptable.

## Frequency of publication:

The Journal will appear at least once a year. At the direction of the Joint Executive Committee, the Journal may appear more frequently. Contributed papers not presented at a meeting may be reviewed, edited, and published if the editorial criteria are met.

## Editorial Committee:

The Editorial Committee shall be composed of the Managing Editor, Technical Editor for the Agricultural Section, and Technical Editor for the Manufacturing Section. The Editorial Committee shall regulate the Journal content and assure its quality. It is charged with the authority necessary to achieve these goals. The Editorial Committee shall determine broad policy. Each editor will serve for three years; and may at the Joint Executive Committee's discretion, serve beyond the expiration of his or her term.

# Handling of manuscripts:

Four copies of each manuscript are initially submitted to the Managing Editor. Manuscripts received by the Managing Editor will be assigned a registration number determined serially by the date of receipt. The Managing Editor writes to the one who submitted the paper to inform the author of the receipt of the paper and the registration number which must be used in all correspondence regarding it.

The Technical Editors obtain at least two reviews for each paper from qualified persons. The identities of reviewers must not be revealed to each other nor to the author during the review process. Instructions sent with the papers emphasize the necessity for promptness as well as thoroughness in making the review. Page charges will be assessed for the entire manuscript for non-members. Members will be assessed for those pages in excess often (10) double spaced Times New Roman (TT) 12 pt typed pages of 8 12<sup>7</sup> x 11<sup>n</sup> dimension with one (1) inch margins.

When a paper is returned by reviewers, the Technical Editor evaluates the paper and the recommendations of the reviewers. If major revisions are recommended, the Technical Editor sends the paper to the author for this purpose, along with anonymous copies of reviewers' recommendations. When the paper is returned to the Technical Editor, he/she will Judge the adequacy of the revision and may send the paper back to any reviewer for further review. When

the paper has been revised satisfactorily, it is sent to the Managing Editor for publishing. A paper sent to its author for revision and held more than 6 months will be given a new date of receipt when returned. This date will determine the priority of publication of the paper.

A paper rejected by one reviewer may be sent to additional reviewers until two reviewers either accept or reject the paper. If a paper is judged by two or more reviewers as not acceptable for the Journal, the Technical Editor returns it to the author along with a summary of the reasons given by the reviewers for the rejection. The registration form for the paper is filled out and returned to the Managing Editor along with copies of the reviewers' statements and a copy of the Technical Editor's transmittal letter to the author. The names of all reviewers must be shown on the registration form transmitted to the Managing Editor.

If the paper as received is recommended by two reviewers for publication in the Journal, it is read by the Technical Editor to correct typographical, grammatical, and style errors and to improve the writing where this seems possible and appropriate, with special care not to change the meaning. The paper is then sent by the Technical Editor to the Managing Editor who notifies the authors of the acceptance of the paper and of the probable dates of publication. At this time, the Managing Editor will request a final version in hardcopy and on diskette in WordPerfect format from the corresponding author.

## Preparation of papers for publication;

Papers sent by the Technical Editor to the Managing Editor are prepared for printing according to their dates of original submittal and final approval and according to the space available in the next issue of the Journal.

The paper is printed in the proper form for reproduction, and proofs are sent to the authors for final review. When the proofs are returned, all necessary corrections are made prior to reproduction. The author will be notified at the appropriate time to order reprints at cost.

Any drawings and photographs for the figures in the paper are "scaled" according to their dimensions, the size of lettering, and other factors. They are then sent to the printer for camera work. Proofs of the illustrations are sent to the authors. Any changes requested at this stage would be expensive and authors will be expected to pay the cost of such changes.

Reprinting in trade journals has the approval of the Editorial Committee provided: a) no article is reprinted before being accepted by the Journal; b) credit is given all authors, the author's institutions, and the ASSCT; and c) permission of all authors has been obtained. Summaries, condensations, or portions may be printed in advance of Journal publication provided the approval of the Editorial Committee has been obtained.

# RULES FOR PREPARING PAPERS TO BE PRINTED IN THE JOURNAL OF THE AMERICAN SOCIETY OF SUGAR CANE TECHNOLOGISTS

## Format

Unless the nature of the manuscript prevents, it should include the following sections in the order listed: ABSTRACT, INTRODUCTION, MATERIALS and METHODS, RESULTS, DISCUSSION (OR RESULTS AND DISCUSSION), CONCLUSIONS, ACKNOWLEDG-MENTS, and REFERENCES. Not all the sections listed above will be included in each paper, but each section should have an appropriate heading that is centered on the page with all letters capitalized. Scientific names shall be italicized.

All material (including tables and figures) shall be submitted on 8% X 11 inch paper with one inch margins on all sides. If using WordPerfect, set the bottom margin at 0.5 inches. This will set the page number at 0.5 inches and the final line of text at 1 inch from the bottom margin. Exactness in reproduction can be insured if electronic copies of the final versions of manuscripts are submitted. Authors are encouraged to contact the managing editor for specifics regarding software and formatting software to achieve ease of electronic transfer.

## Authorship

Name of the authors, institution or organization with which they are associated, and their locations should follow the title of the paper.

## Abstract

The abstract should be placed at the beginning of the manuscript, immediately following the author's name, organization and location.

## Tables

Number the tables consecutively and refer to them in the text as Table 1, Table 2, etc. Each table must have a heading or caption. Capitalize only the initial word and proper names in table headings. Headings and text of tables should be single spaced. Use TAB function rather than SPACE BAR to separate columns of a table.

## Figures

Number the figures consecutively and refer to them in the text as Figure 1, Figure 2, etc. Each figure must have a legend. Figures must be of sufficient quality to reproduce legibly.

## Drawings & Photographs

Drawings and photographs must be provided separately from the text of the manuscript and identified on the back of each. Type figure numbers and legends on separate pieces of paper with proper identification. Drawings and photographs should be of sufficient quality that they will reproduce legibly.

## Reference Citations

The heading for the literature cited should be REFERENCES. References should be arranged such that the literature cited will be numbered consecutively and placed in alphabetical order according to the sumame of the senior author. In the text, references to literature cited should be made by name of author(s) and year of publication from list of references. Do not use capital letters in the titles of such articles except in initial words and proper names, but capitalize words in the titles of the periodicals or books.

Format Example

# ITCHGRASS (ROTTBOELLIA COCHINCHINENSIS) CONTROL IN SUGARCANE WITH POSTEMERGENCE HERBICIDES

# Reed J. Lencse and James L. Griffin

Department of Plant Pathology and Crop Physiology Louisiana Agricultural Experiment Station, LSU Agricultural Center Baton Rouge, LA 70803

and

# Edward P. Richard, Jr.

Sugarcane Research Unit, USDA-ARS, Houma, LA 70361

# ABSTRACT

# INTRODUCTION

# MATERIALS AND METHODS

# RESULTS AND DISCUSSION

Table 1. Visual itchgrass control and sugarcane injury as influenced by over-the-top herbicide application at Maringouin and Thibodaux, LA, 1989.

# CONCLUSIONS

# ACKNOWLEDGMENTS

# REFERENCES

## GUIDELINES FOR PREPARING PAPERS FOR JOURNAL OF ASSCT

The following guidelines for WordPerfect software are intended to facilitate the production of this journal. Authors are strongly encouraged to prepare their final manuscripts with WordPerfect 6.0 or a later version for Windows. Please contact the Managing Editor if you will not use one of those software packages.

**Paper & Margins:** All material (including tables and figures) shall be submitted on  $8^{1}/_{2} \times 11$ inch paper with one inch margins on all sides. To achieve this with WordPerfect, set the top, left, and right margins at one inch. However, set the bottom margin at 0.5 inches. This will place the page number at 0.5 inches and the final line of text at one inch.

Fonts: Submit your document in the Times New Roman (TT) 12pt font. If you do not have this font, contact the Managing Editor.

<u>Alignment:</u> Choose the full alignment option to prepare your manuscript. The use of SPACE BAR for alignment is not acceptable. As a general rule SPACE BAR should only be used for space between words and limited other uses. Do not use space bar to indent paragraphs, align and indent columns, or create tables.

Do not use hard returns at the end of sentences within a paragraph. Hard returns are to be used when ending paragraphs or producing a short line.

Place tables and figures within the text where you wish them to appear. Otherwise, all tables and figures will appear after your References section.

Styles: Italicize scientific names. Do not use underline.

Tables: Use Tab stops and the Graphics line draw option when constructing tables. Avoid the space bar to separate columns (see alignment). All lines should begin with the left most symbol in their left most column and should end with the right most symbol in their right most column.

Citations: When producing Literature Citations, use the indent feature to produce text as below.

 Smith, I. M., H. P. Jones, C. W. Doe, 1991. The use of multidiscipline approaches to control rodent populations in plants. Journal of American Society of Plant Management. 10:383-394.

#### CONSTITUTION OF THE AMERICAN SOCIETY OF SUGAR CANE TECHNOLOGISTS

#### As Revised and Approved on June 21,1991 As Amended on June 23,1994 As Amended on June 15,1995

## ARTICLE I

#### Name. Object and Domicile

Section 1. The name of this Society shall be the American Society of Sugar Cane Technolo-gists.

- Section 2. The object of this society shall be the general study of the sugar industry in all its various branches and the dissemination of information to the members of the organization through meetings and publications.
- <u>Section 3.</u> The domicile of the Society shall be at the office of the General Secretary-Treasurer (as described in Article IV, Section 1).

# ARTICLE 11

#### Divisions

The Society shall be composed of two divisions, the Louisiana Division and the Florida Division. Each division shall have its separate membership roster and separate officers and committees. Voting rights of active and honorary members shall be restricted to their respective divisions, except at the general annual and special meetings of the entire Society, hereinafter provided for, at which general meetings active and honorary members of both divisions shall have the right to vote. Officers and committee members shall be members of and serve the respective divisions from which elected or selected, except the General Secretary-Treasurer who shall serve the entire Society.

# ARTICLE 111

#### Membership and Dues

- Section 1. There shall be five classes of members: Active, Associate, Honorary, Off-shore or Foreign, and Supporting.
- Section 2. Active members shall be individuals residing in the continental United States actually engaged in the production of sugar cane or the manufacture of cane sugar, or research or education pertaining to the industry, including employees of any corporation, firm or other organization which is so engaged.
- Section 3. Associate members shall be individuals not actively engaged in the production of sugar cane or the manufacture of cane sugar or research pertaining to the industry, but who may be interested in the objects of the Society.

- Section 4. Honorary membership shall be conferred on any individual who has distinguished himself or herself in the sugar industry, and has been elected by a majority vote of the Joint Executive Committee. Honorary membership shall be exempt from dues and entitled to all the privileges of active membership. Each Division may have up to 15 living Honorary Members. In addition, there may be up to 5 living Honorary members assigned to the two Divisions jointly.
- Section 5. Off-shore or foreign members shall be individuals not residing in the continental United States who may be interested in the objects of the Society.
- Section 6. Supporting members shall be persons engaged in the manufacturing, production or distribution of equipment or supplies used in conjunction with production of sugar cane or cane sugar, or any corporation, firm or other organization engaged in the production of sugar cane or the manufacture of cane sugar, who may be interested in the objects of the Society.
- Section 7. Applicants for new membership shall make written application to the Secretary-Treasurer of the respective divisions, endorsed by two members of the division, and such applications shall be acted upon by the division membership committee.
- Section 8. Minimum charge for annual dues shall be as follows:

Active Membership	\$10.00
Associate Membership	-\$25.00
Honorary Membership	-NONE
Off-shore or Foreign Membership	\$20.00
Supporting Membership	-\$50.00

Each Division can assess charges for dues more than the above schedule as determined by the Division officers or by the membership at the discretion of the officers of each Division.

Dues for each calendar year shall be paid not later than 3 months prior to the annual meeting of the member's division. New members shall pay the full amount of dues, irrespective of when they join. Any changes in dues will become effective in the subsequent calendar year.

- Section 9. Dues shall be collected by each of the Division's Secretary-Treasurer from the members in their respective divisions. Unless and until changed by action of the Joint Executive Committee, 50 percent of the minimum charge for annual dues, as described in Section 8 for each membership class, shall be transmitted to the office of the General Secretary-Treasurer.
- Section 10. Members in arrears for dues for more than a year will be dropped from membership after thirty days notice to this effect from the Secretary-Treasurer. Members thus dropped may be reinstated only after payment of back dues and assessments.
- Section 11. Only active members of the Society whose dues are not in arrears and honorary members shall have the privilege of voting and holding office. Only members (all classes) shall have the privilege of speaking at meetings of the Society.

# ARTICLE IV

## General Secretary-Treasurer and Joint Executive Committee

- Section 1, The General Secretary-Treasurer shall serve as Chief Administrative Officer of the Society and shall coordinate the activities of the divisions and the sections. He or she will serve as ex-officio Chairperson of the Joint Executive Committee and as General Chairperson of the General Society Meetings, and shall have such other duties as may be delegated to him or her by the Joint Executive Committee. The office of the General Secretary-Treasurer shall be the domicile of the Society.
- <u>Section 2.</u> The Joint Executive Committee shall be composed of the elected members of the two division Executive Committees, and is vested with full authority to conduct the business and affairs of the Society.

# ARTICLE V

## Division Officers and Executive Committee

- Section 1. The officers of each division of the Society shall be: a President, a First Vice-President, a Second Vice-President, a Secretary-Treasurer or a Secretary and a Treasurer, and an Executive Committee composed of these officers and four other members, one from each section of the Division (as described in Section 3 of Article VII), one elected at large, and the President of the previous Executive Committee who shall serve as an Ex-Officio member of the Division Executive Committee. The office of the Secretary-Treasurer in this constitution indicates either the Secretary-Treasurer, or the Secretary and the Treasurer.
- Section 2. These officers, except Secretary-Treasurer, shall be nominated by a nominating committee and voted upon before the annual division meeting. Notices of such nominations shall be mailed to each member at least one month before such meeting. Ballots not received before the annually specified date will not be counted.
- Section 3. The Secretary-Treasurer shall be appointed by and serve as a non-voting member at the pleasure of the Division Executive Committee. The Secretary-Treasurer may not hold an elected office on the Executive Committee.
- Section 4. The duties of these officers shall be such as usually pertain to such officers in similar societies.
- Section 5. Each section as described in Article VII shall be represented in the offices of the President and Vice-President.
- Section 6. The President, First Vice-President, and Second Vice-President of each Division shall not hold the same office for two consecutive years. Either Section Chairperson (as described in Section 3 of Article VII) may hold the same office for up to two consecutive years. The terms of the other officers shall be unlimited.
- Section 7. The President shall be elected each year alternately from the two sections hereinafter provided for. In any given year, the Presidents of the two Divisions shall be nominated and elected from different sections. The President from the Louisiana Division for the year beginning February, 1970, shall be nominated and elected from the Agricultural Section. The president from the Florida Division for the year beginning February.

1970, shall be nominated and elected from the Manufacturing Section.

- Section 8. Vacancies occurring between meetings shall be filled by the Division Executive Committee.
- Section 9, The terms "year" and "consecutive year" as used in Articles V and VI shall be considered to be comprised of the elapsed time between one annual division meeting of the Society and the following annual division meeting of the Society.

## ARTICLE VI

#### **Division Committees**

- Section 1. The President of each division shall appoint a committee of three to serve as a Membership Committee. It will be the duty of this committee to pass upon applications for membership in the division and report to the Secretary-Treasurer.
- Section 2. The President of each division shall appoint each year a committee of three to serve as a Nominating Committee. It will be the duty of the Secretary-Treasurer of the Division to notify all active and honorary members of the Division as to the personnel of this committee. It will be the duty of this committee to receive nominations and to prepare a list of nominees and mail this to each member of the Division at least a month before the annual meeting.

## ARTICLE VII

#### Sections

Section 1. There shall be two sections of each Division, to be designated as:

- 1. Agricultural
- 2. Manufacturing
- Section 2. Each active member shall designate whether he or she desires to be enrolled in the Agricultural Section or the Manufacturing Section.
- Section 3. There shall be a Chairperson for each section of each Division who will be the member from that Section elected to the Executive Committee. It will be the duty of the Chairperson of a section to arrange the program for the annual Division meeting.
- Section 4. The Executive Committee of each Division is empowered to elect one of their own number or to appoint another person to handle the details of printing, proofreading, etc., in connection with these programs and to authorize the Secretary-Treasurer to make whatever payments may be necessary for same.

# ARTICLE Vffl

## Meetings

Section 1. The annual General Meeting of the members of the Society shall be held in June each year on a date and at a place to be determined, from time to time, by the Joint Executive Committee. At all meetings of the two Divisions of the Society, five percent of the active members shall constitute a quorum. The program for the annual meeting

of the Society shall be arranged by the General Secretary-Treasurer in collaboration with the Joint Executive Committee.

- Section 2. The annual meeting of the Louisiana Division shall be held in February of each year, at such time as the Executive Committee of the Division shall decide. The annual meeting of the Florida Division shall be held in September or October of each year, at such time as the Executive Committee of that Division shall decide. Special meetings of a Division may be called by the Executive Committee of such Division.
- Section 3. Special meetings of a Section for the discussion of matters of particular interest to that Section may be called by the President upon request from the respective Chairperson of a Section.
- Section 4. At Division meetings, 10 percent of the active division members and the President or a Vice-President shall constitute a quorum.

## ARTICLE IX

## Management

- <u>Section 1.</u> The conduct and management of the affairs of the Society and of the Divisions including the direction of work of its special committees, shall be in the hands of the Joint Executive Committee and Division Executive Committees, respectively.
- Section 2. The Joint Executive Committee shall represent this Society in conferences with the American Sugar Cane League, the Florida Sugar Cane League, or any other association, and may make any rules or conduct any business not in conflict with this Constitution.
- Section 3. Four members of the Division Executive Committee shall constitute a quorum. The President, or in his or her absence one of the Vice-Presidents, shall chair this committee.
- Section 4. Two members of each Division Executive Committee shall constitute a quorum of all members of the Joint Executive Committee. Each member of the Joint Executive Committee, except the General Secretary-Treasurer, shall be entitled to one vote on all matters voted upon by the Joint Executive Committee. In case of a tie vote, the General Secretary-Treasurer shall cast the deciding vote.

## ARTICLE X

#### Publications

- Section 1. The name of the official journal of the Society shall be the "Journal of the American Society of Sugar Cane Technologists." This Journal shall be published at least once per calendar year. All articles, whether volunteered or invited, shall be subject to review as described in Section 4 of Article X.
- Section 2. The Managing Editor of the Journal of the American Society of Sugar Cane Technologists shall be a member of either the Florida or Louisiana Divisions; however, he or she shall not be a member of both Divisions. The Division affiliation of Managing Editors shall alternate between the Division from term to term with the normal term being three years, unless the Division responsible for nominating the new Managing Editor reports that it has no suitable candidate. The Managing Editor shall

be appointed by the Joint Executive Committee no later than 6 months prior to the beginning of his or her term. A term will coincide with the date of the annual Joint Meeting of the Society. No one shall serve two consecutive terms unless there is no suitable candidate from either Division willing to replace the current Managing Editor. If the Managing Editor serves less than one year of his or her three-year term, another candidate is nominated by the same Division, approved by the other Division, and appointed by the General Secretary-Treasurer to a full three-year term. If the appointed Managing Editor serves more than one year but less than the full three-year term, the Technical Editor from the same Division Will fill the unexpired term of the departed Managing Editor. In the event that the Technical Editor declines the nomination, the General Secretary-Treasurer will appoint a Managing Editor from the same Division to serve the unexpired term.

- Section 3. The "Journal of the American Society of Sugar Cane Technologists" shall have two Technical Editors, which are an Agricultural Editor and a Manufacturing Editor. The Managing Editor shall appoint the Technical Editors for terms not to exceed his or her term of office. Any Technical Editor shall be a member of either the Louisiana or Florida Division. Each Division will be represented by one technical editor at all times unless the Executive Committee of one Division and the Managing Editor agree that there is no suitable candidate willing to serve from that Division.
- Section 4. Any member or nonmember wishing to contribute to the Journal of the American Society of Sugar Cane Technologists shall submit his or her manuscript to the Managing Editor. The Managing Editor shall then assign the manuscript to the appropriate Technical Editor. The Technical Editor shall solicit peer reviews until, in the opinion of the Technical Editor, two responsible reviews have been obtained that either accept (with or without major or minor revision) or reject the manuscript. For articles accepted with major revision, it shall be the responsibility of the Technical Editor to decide if the authors have satisfactorily completed the major revision(s). The Technical Editors shall not divulge the identity of any reviewer. The Managing Editor shall serve as Technical Editor of any manuscript which includes a Technical Editor of any an author.

## ARTICLE XI

#### Amendments

Section 1. Amendments to this Constitution may be made only at the annual meeting of the Society or at a special meeting of the Society. Written notices of such proposed amendments, accompanied by the signature of at least twenty (20) active or honorary members must be given to the General Secretary-Treasurer at least thirty (30) days before the date of the meeting, and he or she must notify each member of the proposed amendment before the date of the meeting.

#### ARTICLE XH

#### Dissolution

Section 1. All members must receive notification from the General Secretary-Treasurer of any meeting called for the purpose of terminating the Society at least thirty (30) days prior to the date of the meeting. After all members have been properly notified, this organization may be terminated at any time, at any regular or special meeting called for that purpose, by an affirmative vote of two-thirds of the total honorary and active members in good standing present at the meeting. Thereupon, the organization shall be dissolved by such legal proceedings as are provided by law. Upon dissolution of the Joint Society, its assets will be divided equally between the two Divisions of the Society. Dissolution of the Joint Society will not be cause for automatic dissolution of either Division. Upon dissolution of either Division, its assets will be divided in accordance with the wishes of its members and in conformity with existing IRS regulations and other laws applicable at the time of dissolution.

#### ARTICLE XIII

#### Assets

Section 1. No member shall have any vested right, interest or privilege of, in, or to the assets, functions, affairs or franchises of the organization; nor any right, interest or privilege which may be transferable or inheritable.

# AUTHOR INDEX

Abouzid, A. M.	.154
Allen Jr., L. Hartwell.	9, 156
Alvarez, Jose	
Ande, B	166, 167
Ande, P	
Anderson, Bazil	.172
Anderson, David L	43
Baez-Smith, Carmen	
Balzarini, Maria	.160
Birkett, Harold S	176
Bischoff, Keith P	30, 51, 159, 160,168
Bochamikova, E. A.	166, 167
Brown, J. S.	
Brown, L. P	
Burner, David M	
Calvert, D. V	166,167
Chen, R	.165
Chengalrayan, K	154,155
Cherry, Ron	.151
Comstock, J. C	.154
Cuddihy, J. A.	73,171
Damms, Michael	.172
Duchrow, M.J,	.156
Edme, Serge B	.164
Eggleston, Gillian	159,173
Felicio, Luiz Carlos	102,174
Fouss, James L	178
Gallo-Meagher, Maria	9, 154, 155, 156
Gamez, L. P	175
Glaz, Barry	161, 162, 163, 164
Gravois, Kenneth A	30, 51, 159, 160, 168
Grisham, Michael P	155
Hall, David G	21,151
Hallmark, William B	167
Hawkins, G. L	51,159,167
Herrara, H	175
Herscovici, Saul	129
Hill, A. G	172
Holder, David G	164
Hudetz, M	157
Inkson, M. B	.176
Jain, R	
Johnson, Tere P	62,171
Journet, G	

Komecki, Ted S	
LaBorde, Chris	30, 168
Legaspi, Jesusa C	153
Legaspi Jr., Benjamin C.	153
Legende, Benjamin L	155, 159
Li, G	.165
Lockhart, Mike	
Matichenkov, V. V	166, 167
Miller, J. D	154,162,164
Milligan, Scott B	160,164
Mirkov, T. E	153, 154
Monge, A	173, 176
Morris, D. M	162
Muchovei, Rosa M.	169
Mullahey, J. F	169
Murray, Travis	.169
Pampatwar, M. K	172
Pan, Yong,-Bao	155
Pelletan, Christophe	140
Pepperman, A	173
Porro. Miguel E	73.171
Posev. F. R.	152
Price R	169
Rauh James S	73 171
Ray J D	163
Rawls, E. K	157
Reagan TE	152
Rein Peter	92 172
Richard Ir Edward P	157
Richard C	159
Saska Michael	175
Schexnavder H P	51 159
Schueneman Thomas I	165
Shine Ir I M	156
Sinclair T R	163
Snyder G H	166
Sosa Ir Omelio	43
Spinoso I G	176
Tai P Y P	162
Illioa Modesto E	157
Vu Joseph C V	9 156
Wei O	155
Weiss L. Warner	109
Wells 1 W	157
TT 0110, 1 TV	



Williams, G. J	 					.167
Zhang, Mu-Qing					9,156,	165