Suitable Biomass for a Sustainable Sugarcane Industry

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Sweet Sorghum in a Sugarcane Production System

Introduction

Sweet sorghum and energycanes, have the potential to be used as feedstocks for the Australian sugar industry. In comparison to Brazil and the USA little research has been conducted in Australia on this topic.

In South America, especially Brazil, sweet sorghum has been grown as a complementary crop with sugarcane for several years [9]. Ceres and Syngenta have been major players in developing sweet sorghum genotypes that can be utilised as a supplementary crop in a sugarcane-based bioindustry. Unlike sugarcane sweet sorghum is an annual crop, growers can easily change their management strategy, depending on market conditions. It is estimated that sweet sorghum has the potential to extend the ethanol production season by more than 60 days.

Sorghum needs to be considered as a feedstock for first and second generation biofuels, extraction of water soluble components or for biogas applications.

Here we consider the possible integration of sorghum as a supplementary crop and possibilities around the utilisation of its water-soluble components.

In this Update

- *Incorporation of sorghum into the sugarcane production system.*
- *Sorghum juice as a fermentation substrate.*
- *Other potential products in the juice.*

Growing a sorghum crop

As discussed previously a major production constraint for sorghum in the tropical conditions is early flowering. This is in part the result of minimal variation in daylength in the tropics. The data in our trials showed that even genotypes like SK106 selected for very late flowering flower profusely in Northern Queensland. The genotype SK106 did not flower within the first 130 days after planting (DAP) at a lattitude of 27.5606^oS (Toowoomba) but flowers 100% 70DAP at a lattitude of 17.268 ^oS (Atherton Tablelands).

The impact of flowering on biomass is twofold [3]. Firstly, there is competition between grain filling and accumulation of soluble sugars in stalks. Removal of the inflorecences as they are formed resulted in an increase of soluble sugar accumulation in the stalk. However, this effect is only moderate demonstrating that the carbon to support flowering and grain fill is not derived from remobilisation of stalk sugars. The second, and major impact of flowering, is the creation of a new major sink for the photosyntate from the leaves and a strong suppression of vegetative biomass accumulation. In sorghum delayed flowering increases the size of stems and the potential for sucrose accumulation [4, 2, 8, 6]. Delayed flowering and long duration of vegetative growth is a key trait associated with high biomass yield and nitrogen use efficiency.

Integration of sweet sorghum into a sugarcane based farming operation, especially in the wet tropics will not be without significant challenges. Timing of planting and harvesting of both the Tablelands and Coastal (Mossman) sugarcane crops are similar. The ideal planting window is as soon as practicable after the finish of the wet season. An April/May planting will maximise the plant crop yield (Fig. 1). However, adverse weather conditions could push out planting to beginning of September, especially

Frikkie Botha, Crop Science, Queensland Alliance for Agriculture & Food Innovation The University of Queensland, St Lucia QLD 4072
☎ (+61) 048 840 0074 ✉ f.botha@uq.edu.au

at Mossman.

At both locations the crop is harvested between June and November (driest months of the year). Blocks that are scheduled for ploughout is cut at the end of the harvest season. Ideally the fallow period is from December to March. However, if the wet season lingers then it might be December to May or June (Fig. 1).

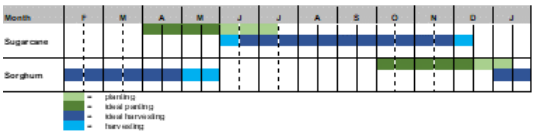

Figure 1: *Potential cropping cycles of sugarcane and sorghum tropical Northern Queensland.*

Evidently, the successful integration of sorghum into a sugarcane-based production system require selection of genotypes that have a much longer vegetative growth period in the tropics than the varieties tested in the project. There is a significantly higher biomass production when sorghum is planted in spring-early summer. However, the short growth cycle then forces a harvesting end of December early January (Fig. 1).

This implies that with the current tested germplasm planting to get a crop in March to May will have to be in December and January. The ideal situation will be a range of maturation groups that allow growth of 90 to 120 days. Research in Brazil also found that optimal biomass production is achieved with a spring early summer planting and that early-maturing varieties produce less biomass, especially when planted outside the optimal planting date period [9].

There are two issues and for both data is lacking. Firstly, the planting of a sorghum crop in the period that should be earmarked for a fallow or break-crop will have profound impacts on pest and disease cycles. Sugarcane and sweet sorghum share several diseases and pests. Secondly, the planting of a sorghum crop is very close to the beginning of the wet season, and ideal harvesting would significantly overlap with the wet season.

Sugars and Biofuel

Sweet sorghum juice can be used for various applications. The earliest and widest use of this fraction is the production of sugar from the liquid. The by-product, of sugar extraction is molasses, which still contains a high amount of sugars. Hence, molasses is used as a good carbon source in the fermentation industry. Sweet sorghum juice itself can be used for ethanol production as well [5] and it was reported to be an ideal substrate for the production of gaseous biofuels such as hydrogen [1].

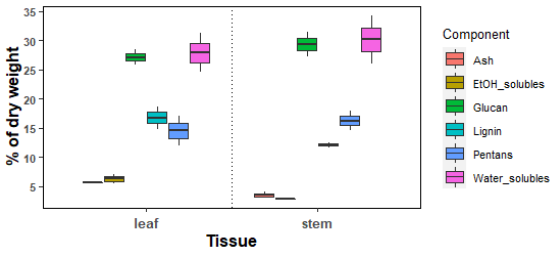

Figure 2: *Biomass composition of five sweet sorghum genotypes 70 DAP.*

A major problem with the uitlisation of sorghum juice is the fast deterioration after harvest. Several techniques can be used to limit this problem [10, 7].

The non-lignocellulosic fraction or extractives are defined as extraneous components that may be separated from the insoluble cell wall material by their solubility in water or neutral organic solvents. To extract all these constituents solvents of different polarities are required to remove different types of extractives. In the work reported here we obtained the total soluble component of the sorghum tissue by first extracting the tissue with water and the remaining solid residue then extracted using 95% ethanol.

The total soluble component varies between 21-40% in the leaves and 30-42% in the stems (Fig. 2). The wide range in values can be attributed to genotype differences (Fig. 3).

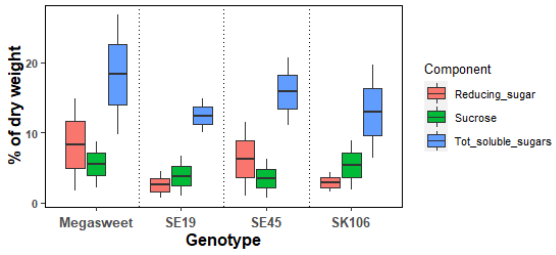

Figure 3: *Sugar levels in the stems of sorghum genotypes 70DAP.*

It is important to note that this implies that even exhaustive extraction can only recover a maximum of 40% of the total biomass.

A main differences between the leaves and stems are in the water soluble sugars. Total water-soluble sugars in the leaves vary between 3-13% and between 14-27% in the stems. The reducing sugar (glucose and fructose) levels are similar or even higher than that of sucrose in the stems. The reducing sugars represent between 40 and 60% of the total water-soluble sugar. Sucrose, glucose and fructose make up the bulk of water soluble sugar in the leaves and stems.

Frikkie Botha, Crop Science, Queensland Alliance for Agriculture & Food Innovation The University of Queensland, St Lucia QLD 4072
☎ (+61) 048 840 0074 ✉ f.botha@uq.edu.au

Traditionally sugar is extracted from sweet sorghum by squeezing the stalks through a roller mill. This process of crushing is ineffective and a substantial amount of fermentable sugar remains after a single crushing. Multistaged, extraction technologies used to process sugarcane achieve much better recovery but more energy is expended to achieve this efficiency. Diffusion extraction similar to that use for sugar beets and sugarcane is more efficient to extract most of the fermentable sugar. This latter method is not utilised by Australian sugarcane mills.

This project did not consider extraction methodologies but rather in this report we assume that all the soluble sugars and other metabolites can be recovered from the material.

However, even in full recovery of all the sugars the juice purity will be low as a result of the very high reducing sugar content (Fig. 4). In fact, in most cases the reducing sugar content exceeded the sucrose content making sucrose recovery impossible.

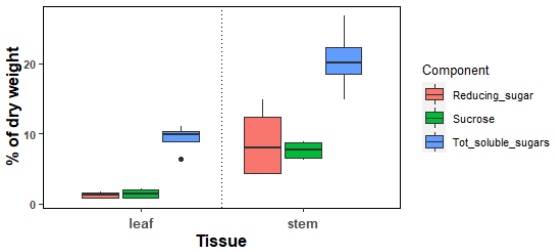

Figure 4: *Reducing sugars, sucrose and total sugar content of sorghum tissues 70 DAP.*

We therefore assume that juice extraction from sweet sorghum will be aimed at maximising the fermentation potential. It is important to note that fermentation is not only for biofuel production but could form the basis for the production of a raft of high value products.

$$litreEtOH.t^{-1} = kgSugar.t^{-1} * 0.58 \quad (1)$$

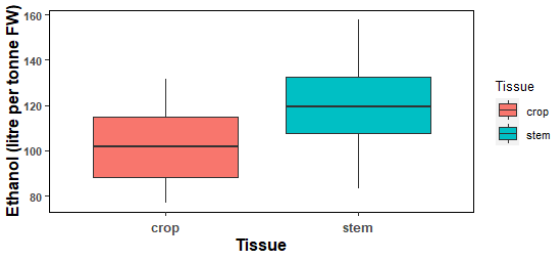

Figure 5: *Ethanol production potential from the soluble sugars present in sorghum biomass 70 DAP.*

Table 1: *Sorghum yield, sugar content and ethanol production potential. The numbers are based on total crop harvested and used for extraction of sugars*

Parameter	Min	Max	SD[1]
Yield (Tonne.hectare^{-1} FW)	20	58	-
Yield (Tonne.hectare^{-1} DW)	4	11.6	-
Kg Sugar.tonne^{-1}FW	132.6	226.7	33.19
KgSugar.tonne^{-1}DW	32.7	59.2	9.25
Litre EtOH.tonne^{-1}DW	76.9	131.5	19.25
Litre EtOH.tonne^{-1}FW	18.7	34.3	5.37

Metabolites

To determine the polar metabolite profile of sorghum tissue was sequentially extracted with methanol and then water. The resulting extract was dried under vacuum and the pellet methoximated and trimethylsilylated prior to GC-MS analyses. Two internal standards were included in all analyses ($^{13}C_5$,^{15}N Valine and $^{13}C_6$ Sorbitol. We routinely identified 132 metabolites in the sorghum tissue (Table 2).

Only eighteen of these metabolites are present at levels > than 0.05% of total dry mass and could potentially be recovered from the tissue (Table 3). An assessment of the potential commercial value of these metabolites has not been done yet.

Frikkie Botha, Crop Science, Queensland Alliance for Agriculture & Food Innovation The University of Queensland, St Lucia QLD 4072
☎ (+61) 048 840 0074 ✉ f.botha@uq.edu.au

Table 2: *Soluble metabolites present in Sweet Sorghum leaf and stem tissues.*

2-Aminoethanol	Beta-Gentibiose	Isocitric acid	Proline
2-Aminopimelic acid	Caffeic acid	Isoleucine	Protocatechuic acid
2-Hydroxybutyric acid	Cellubiose[1]	Itaconic acid	Psicose
2-Hydroxyglutaric acid	Citramalic acid	Lactic acid[1]	Putrescine
2-Hydroxyisobutyric acid	Citric acid[1]	Lauric acid	Pyruvic acid
2-Hydroxyisovaleric acid	Coniferyl alcohol	Leucine	Quinate[1]
2-Isopropylmalic acid	Cysteine	Linoleic acid	Raffinose[1]
3-Aminoglutaric acid	Decanoic acid	Lysine	Ribitol[1]
3-Aminoisobutyric acid	Dihydroxyacetone phosphate	Lyxose	Ribonic acid
3-Aminopropanoic acid	Dihydroxyacetone	Malic acid	Ribose
3-Dehydroshikimic acid	Erythrulose	Malonic acid	Sedoheptulose[1]
3-Hydroxybutyric acid	Fructose 1-phosphate	Maltose[1]	Serine
3-Hydroxyisobutyric acid	Fructose[1]	Mannitol	Shikimic acid
3-Hydroxyisovaleric acid	Fumaric acid	Mannose[1]	Sorbitol
3-Hydroxypropionic acid	Galactose[1]	Mesaconic acid	Spermidine
3-Methoxy-4-hydroxybenzoic acid	Galacturonic acid	meso-Erythritol	Stearic acid
4-Aminobutyric acid	Gentiobiose[1]	Methylsuccinic acid	Stigmasterol
4-Hydroxybenzoic acid	Glucaric acid	myo-Inositol[1]	Succinic acid
4-Hydroxyphenylacetic acid	Gluconic acid	Monostearin	Sucrose[1]
4-Hydroxyproline	Glucose 6-phosphate	Myristic acid	Tagatose
5-Hydroxymethyl-2-furoic acid	Glucose[1]	N-Acetyl-D-glucosamine	Tartaric acid
5-Oxoproline	Glucuronic acid	N-Acetylglutamine	Threonic acid
Aconitic acid[1]	Glutamic acid	Nicotinic acid	Threonine
Adenine	Glutamine	Nonanoic acid	Thymine
Adipic acid	Glutaric acid	Octanoic acid	Trehalose
Alanine	Glyceric acid[1]	Oleic acid	Tryptamine
Arabinose	Glycerol	Ornithine	Turanose[1]
Arabitol	Glycine	Palmitic acid	Uracil
Ascorbic acid	Glycolic acid	Pantothenic acid	Valine
Asparagine	Glyoxylic acid	Phenylacetic acid	Xanthosine phosphate
Aspartic acid	Hydroquinone	Phenylalanine	Xylitol
Azelaic acid	Hydroxylamine	Phosphoric acid	Xylose
Benzoic acid	myo-Inositol	Pipecolic acid	Xylulose

[1]Metabolites highlighted in green are present in concentrations higher than 0.05% of total dry weight

Table 3: *Water soluble metabolites content in leaves and stem tissue of Sweet Sorghum tissue.*

	Leaves			Stems		
Metabolite	Min	Max	SD[1]	Min	Max	SD[1]
	Kg.tonne^{-1}Dry weight					
Aconitic.acid	2.93	10.88	2.71	2.37	4.22	0.58
Cellobiose	5.74	6.42	0.23	5.97	6.83	0.27
Citric.acid	2.03	3.03	0.31	1.92	2.11	0.06
Fructose	4.85	9.58	1.42	17.23	49.41	9.84
Galactose	5.12	11.93	2.18	28.88	137.48	47.33
Gentibiose	3.44	3.75	0.11	3.88	5.00	0.38
Glucose	1.02	1.12	0.04	5.72	6.94	0.46
Glyceric.acid	0.89	1.03	0.04	0.85	0.94	0.03
Lactic.acid	1.09	1.51	0.12	1.04	1.63	0.18
Malic.acid	1.43	3.24	0.57	0.89	1.31	0.13
Maltose	7.33	8.34	0.29	7.67	8.79	0.40
Mannose	1.80	5.18	1.09	13.55	61.48	21.16
Myoinositol	1.31	1.69	0.11	1.17	1.33	0.05
Quinic.acid	1.21	1.49	0.09	1.20	1.44	0.08
Raffinose	5.80	6.35	0.18	5.85	6.50	0.23
Ribitol	1.08	3.32	0.80	1.06	1.20	0.04
Sedoheptulose	2.49	5.89	1.06	15.16	44.02	8.83
Sucrose	3.75	16.75	4.83	29.24	67.66	12.60
Turanose	4.49	7.15	0.81	6.60	7.46	0.29

[1] Standard deviation of the mean value

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