Sugar Tech (Jan-Mar 2012) 14(1):21–25
DOI 10.1007/s12355-011-0133-x

MINI REVIEW

Feasibility of Sustaining Sugars in Sweet Sorghum Stalks During Post-Harvest Stage by Exploring Cultivars and Chemicals: A Desk Study

P. Srinivasa Rao · C. Ganesh Kumar · Jayalakshmi Malapaka · Ahmed Kamal · Belum V. S. Reddy

Received: 29 October 2011 / Accepted: 24 December 2011 / Published online: 13 January 2012

Abstract In the recent years sweet sorghum is emerging as an important feedstock for bioethanol production. It was observed that total soluble sugar yield (TSSY) increases with time in the post-anthesis phase depending on the length of crop cycle. The qualitative and quantitative sugar loss of up to 50% or more occurs due to delay in harvest during post-physiological maturity stage depending on the genotype, weather and soil conditions, and the time lag between harvest and crushing of the stalks. Hence, a desk study was conducted to identify suitable cultivars and/or explore the use of chemicals that sustain sugars in the post-harvest phase. In case of delayed harvest beyond physiological maturity stage, growing of cultivars such as SPSSV 30, ICSV 25275, ICSV 25280 and SPV 422 that sustain sugar yield at post-physiological maturity, is recommended. As there are no published reports on sweet sorghum, the literature from sugarcane and wine industries were analyzed and inferences drawn from these industries suggest the evaluation of chemicals like sodium benzoate, potassium metabisulphate, sodium metabisulphite, ammonia, SO_2, vanillin and acetic acid (vinegar) which may arrest the post-harvest deterioration of sweet sorghum stalks before juice extraction.

P. Srinivasa Rao and C. Ganesh Kumar these authors contributed equally.

P. Srinivasa Rao · B. V. S. Reddy
International Crops Research Institute for the Semi-Arid Tropics, Patancheru, Hyderabad 502324, Andhra Pradesh, India

C. Ganesh Kumar (✉) · J. Malapaka · A. Kamal
Chemical Biology Laboratory, Indian Institute of Chemical Technology, Hyderabad 500607, Andhra Pradesh, India
e-mail: cgkumar1@rediffmail.com; cgkumar@iict.res.in

Keywords Sweet sorghum · Post-harvest losses · Bioethanol · Juice quality · Chemicals

Introduction

Sweet sorghum [*Sorghum bicolor* (L.) Moench] is a high biomass and sugar-yielding crop since it has a unique characteristic of high carbon assimilation (50 g m^{-2} day^{-1}) and has a special ability to accumulate high concentrations of easily fermentable sugars (glucose, fructose and sucrose) in the stalks; and the crop is more widely adapted in both tropical and temperate climatic conditions than sugarcane or sugar beets, and is seen as a viable feedstock for ethanol production. A comparative analysis of the juice composition of sweet sorghum and sugarcane is shown in Table 1. Sorghum feedstock has lower sucrose and higher amounts of glucose and fructose as compared to that of sugarcane (Srinivasarao et al. 2009), and is also rich in starch (0.4%–5.3% vs. 0.001%–0.05%); protein (0.9%–1.3% vs. 0.5%–0.6%) and aconitic acid (3.6%–4.8% vs. 1.0%–2.1%). The first step in juice processing is clarification to remove impurities. The juice can be clarified with 3% lead acetate, but it requires 2–3 cycles of filtration after lead acetate treatment, and this affects the Brix%. Higher starch content in sweet sorghum juice also limits its clarification efficiency. Addition of α-amylase helps in clarification by hydrolysis of the starch present in the juice. Further, higher aconitic acid concentration in sweet sorghum juice also causes problems in fermentation. The TSSY increases with time at post-anthesis and with crop cycle length. The stalks comprise as major sinks of soluble sugar, with 79.4–94.6% of TSSY, and major sinks of insoluble sugar with 55.9–75.9% of the total cellulose and hemicellulose yield at physiological maturity (Zhao et al. 2009). The wide variation is due to cultivar

variability. This forms the basis for recommendation of harvesting of the crop during dough to physiological maturity stage (Srinivasarao et al. 2011; Kumar et al. 2010). The qualitative and quantitative stalks sugars in sweet sorghum during post harvest were discussed below and options of mitigation through cultivar variability and chemical usage has been detailed in the following sections. The losses are primarily due to in-stalk fermentation and desiccation. Prior to fermentation, one glucose molecule is converted to two pyruvate molecules by glycolysis. During fermentation, pyruvate is metabolized to various compounds. Homolactic fermentation is the production of lactic acid from pyruvate; alcoholic fermentation is the conversion of pyruvate into ethanol and carbon dioxide; and heterolactic fermentation is the production of lactic acid as well as other acids and alcohols. During ethanol fermentation (performed by the yeast, *Saccharomyces cerevisiae*, and some types of bacteria like *Clostridium* sp. and *Zymomonas mobilis*), the pyruvate is converted to ethanol and carbon dioxide. In alcohol production, the carbon dioxide is released to the atmosphere. The following chemical equation summarizes the course of fermentation of glucose to ethyl alcohol and carbon dioxide.

$$C_6H_{12}O_6(\text{glucose}) \rightarrow 2C_2H_5OH\ (\text{ethyl alcohol}) + 2CO_2(\text{carbon dioxide}).$$

Losses Due to Delayed Harvest

Sweet sorghum being a perishable commodity which needs timely processing of the cane juice after it is harvested. Post-harvest deterioration of the cane is troublesome and has gained increased attention in the recent years among various researchers. The nature of the problems observed in sweet sorghum cane processing are similar to that observed in sugarcane processing industry within the Indian subcontinent and many other parts of the world. Quality losses of the juice is mainly due to the delay in harvesting of the crop in time which may be linked to field problems and lack of suitable mechanical harvesters for the cane of sweet sorghum, transportation of the harvested cane, in factory storage pile or during subsequent milling operations which have been the major impeding factors for the viability of sweet sorghum value chain. The time lag between harvesting to milling of the sorghum cane ranges between 2 and 4 days, which leads to huge losses in the recoverable sugars due to deterioration of the harvested cane. Weather conditions including high temperatures and humidity also have major impact on the cane deterioration. In sugarcane, it has been observed that quality losses in cane is primarily due to chemical (acid) and enzymatic inversion wherein the sucrose could be hydrolyzed to the respective reducing sugars (glucose and fructose) by the acid invertase enzyme (acid inversion of sucrose) which is secreted by few yeast species like *Saccharomyces* (Hanko and Rohrer 2000). Juice quality losses may also be due to microbial contamination, mainly by the *Leuconostoc* spp. which originates from the cane fields and enters the interior of the cane through cut ends and/or damaged sites of the stalk and survives at the expense of stored sucrose. The *Leuconostoc* bacteria use the glucose from the sucrose to form dextran. The amount of dextran synthesis varies from the agro-climatic conditions, cane variety, method of harvesting, cut-to-crush delay and sanitary conditions prevailing in the processing unit. The presence of dextran increases the viscosity of the juice which contributes to the overall quality losses of the milled juice in terms of recoverable sugars, which also have influence on the process of crystallization during sugar manufacturing (Purchase 2001; Solomon et al. 2001; Eggleston 2002). As the cane deteriorates, the cane deterioration products reported are high invert sugars, polysaccharides (e.g., dextran, levan, etc.) and microbial contamination (e.g., ethanol and lactic acid formation). These cane deterioration products often lead to factory processing problems (Lionnet 1996; Eggleston et al. 2001; Solomon et al. 2006). Further, quality parameters such as colour and odour of the cane juice could also serve as visual indicators for deterioration of juice quality (Eggleston 2002). Similar observations in terms of juice quality losses can be drawn for sorghum cane. The qualitative and quantitative sugar losses are up to 20–50% or more due to the delay in harvest at post-physiological maturity depending on the genotype, weather and soil condition (TCL distillery, Nanded, Maharashtra, India, personal communication). In other instance, stalk weight decreased by 20% within 2 days on shelf due to rapid initial moisture loss (AICSIP 2010). However, based on the two seasons experiments conducted at ICRISAT (2009–2010) showed that under moist field conditions, the juice loss is not significant when the stalks were harvested beyond

Table 1 Juice characteristics of sweet sorghum and sugarcane (Lingle 2010)

Character	Sweet *Sorghum*	Sugarcane
pH	4.9–5.5	5.2–5.4
Titratable acidity[a]	3.6–4.8	2.0–3.2
Juice brix (%)	10.5–20.7	16–20
Sucrose (%)	69–74	70–88
Reducing sugars (%)	5–19	4–8
Starch (%)	0.4–5.3%	0.001–0.05
Organic acids (%)	NA	1.5–5.5
Aconitic acid (%)	3.6–4.8	1.0–2.1
Protein (%)	0.9–1.3	0.5–0.6

[a] Titratable acidity is the amount (ml) of 0.1 N NaOH required to adjust pH of 10 ml juice to pH 8.3

NA not available

physiological maturity, i.e. 14 days after attaining physiological maturity. There are genotypic differences in cultivars for juice quantity and quality sustenance *vis a vis* phenological stage. The study on sugar yield (Fig. 1) in 19 sweet sorghum cultivars (Kumar et al. 2010) showed that sucrose accounts for major fermentable sugar (about 70%) and it sharply increased by 146% from dough stage to complete maturity. The variation in the content of monosaccharides (glucose and fructose) is not statistically significant. The cultivars SPSSV 30, ICSV 25275, ICSV 25280 and SPV 422 are recommended for delayed harvesting as they were found suitable for harvesting during a wider window of time as the sugar levels are sustained at same level based on the weather conditions during physiological maturity to post-physiological maturity (Kumar et al. 2010); this helps to increase the raw material supply to distillers. Further experimentation is required to identify cultivars that sustain sugar yield during the post-physiological maturity stage for late post-rainy and summer season sorghum and the details of the factors/traits contributing for stalk sugars sustenance needs further investigation.

Minimization of Post-harvest Sugar Losses

Sodium metasilicate and sodium lauryl sulphate were found to be inhibitory to cane invertases and were able to prevent the inversion of sugars in the juice (Rosaio and Santisopasri 1977). Spraying of harvested cane with benzoic acid (100 ppm) and formaldehyde (100 ppm) significantly reduced post-harvest losses in sugarcane (Desai et al. 1985). Application of a basal preparation of zinc sulphate (25 kg ha^{-1}) reduced the post-harvest deterioration of sugarcane (Tomar and Malik 2004). Spraying of allyl isothiocyanate could also minimize the sucrose losses in the harvested cane as disclosed in a US Patent (Bretschneider et al. 1976). Frequent spraying of a solution containing potassium permanganate (0.1% or 5 ppm) and dimethyl dicarbonate (DMDC) along with sodium metasilicate (1%) on harvested cane minimized the invertase activity and retained the juice quality in mills (Janakiramaiah et al. 1967; Tilbury et al. 1977; Sharma et al. 1989). Application of a formulation of benzalkonium chloride and sodium metasilicate prevented the post-harvest staling of cane and was found effective in the retention of juice quality and improved the sugar yields (Solomon et al. 2006). In one study, spraying of a formulation comprising of glutaradehyde and benzalkonium chloride (1,000 + 250 ppm) reduced the sucrose losses by 7.1% as compared to 30.8% loss observed in case of control, thus improving the performance by 77%. It was also observed that use of these chemicals also reduced the invertase activity by 60%, which indirectly lowered the dextran formation and reduced bacterial, fungal and yeast contaminations by 68, 51 and 51%, respectively. The reduction in microbial contaminations could possibly be due to the antibacterial and antifungal activities of glutaraldehyde and benzalkonium chloride (Singh et al. 2008). Many bactericide preparations including formaldehyde, Polycide, Bacterinol-100, BD Mill sanitizer, DBAC, IFOPOL, DNBT, ABF, Actin-ID, potassium permanaganate and sodium metasilicate, Tsunami-100, KCide 800, Sucroguard, Perla soap solution (1%), etc. have also been demonstrated and recommended to control deterioration of cane and milled juice (Solomon 2009).

Fermentation Inhibitors

The continued life of yeast cells depends on the availability of sufficient nutrients and sugars, and non-poisoning of the

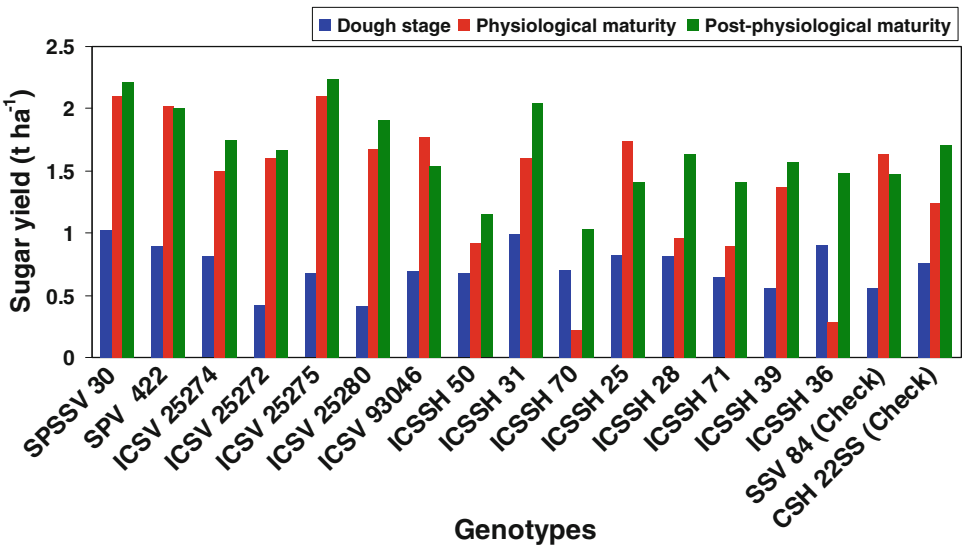

Fig. 1 Performance of sweet sorghum genotypes for sugar yield (t ha^{-1}) in three phenological stages, i.e. dough stage, physiological maturity and complete (post-physiological) maturity

enzyme systems. The osmotic pressure must be lower than that required to rupture the cell wall. As pasteurization of bulky stalks is not feasible at the farm level, chemically induced inhibition of fermenting yeast and bacteria could be a viable option. Based on a critical review of the available literature in wine and sugar industry, we short-listed the following potential chemicals that reduce and/or prevent both yeast and bacterial growth, leading to un-deteriorated stalks. The identification of potential fermentation inhibitor was decided based on the compound's toxicity, or the degree of inhibition. Inhibitory effects are broad and impact cell membrane, synthesis of macromolecules and glycolytic and fermentative enzymes. Furan and aromatic aldehydes in particular, are toxic to microbes and the inhibitory effects of furfurals act synergistically with other compounds such as lignin monomers. The inhibition mechanism of the compound to *S. cerevisiae* falls in either one of the three categories: chemical interface with cell maintenance function, direct inhibition of ethanol pathway and through osmotic pressure of cells. The degree of inhibition follows the same order from high to low concentration (Palmqvist and Hahn-Hagerdhal 2000; Luo et al. 2002; Nichols et al. 2010; Huang et al. 2011). A wide spectrum of potential inhibitors reported for other crop biomass comprises mainly of aromatic aldehydes and acids, aliphatic aldehydes and acids, and furan compounds (Table 2). The two furan aldehydes, such a 2-furaldehyde (furfural) and 5-hydroxymethylfurfural (HMF) are derived as degradation products of xylose and glucose, respectively. In addition, some of the chemicals routinely used and identified as effective inhibitors of fermentation in wine industry are ammonia, potassium metabisulphate and sulfur dioxide (SO_2).

Conclusion

In case of delayed harvest beyond physiological maturity stage, it is recommended to grow cultivars including SPSSV 30, ICSV 25275, ICSV 25280 and SPV 422 that sustain sugar yield during post-physiological maturity. There is no published information available on the in-stalk fermentation in sweet sorghum. However, from the available information in the sugar and wine industry, it seems worthwhile to explore some of the chemicals such as sodium benzoate, potassium metabisulphate, benzalkonium chloride, sodium metasilicate, ammonia, SO_2, vanillin and acetic acid (vinegar) to arrest post-harvest deterioration of sweet sorghum stalks prior to juice extraction. Hence, in order to minimize post-harvest losses of sweet sorghum one needs to explore both the cultivar and chemical options concurrently.

Table 2 Chemicals reported for inhibiting fermentation in biomass of other crops (Adapted from Luo et al. 2002; Nichols et al. 2010; Huang et al. 2011)

S. No.	Fermentation inhibitors
Aromatic compounds	
1	3-Methoxy-4-hydroxy-benzaldehyde (vanillin)
2	4-Hydroxybenzoic acid
3	3,5-Dimethoxy-4-hydroxy-benzaldehyde (syringaldehyde)
4	2,5-Dihydroxy-benzoic acid
5	3-Methoxy-4-hydroxy-benzoic acid (vanillic acid)
6	3,4-Dihydroxy-benzoic acid
7	3,5-Dimethoxy-4-hydroxy-benzoic acid (syringic acid)
8	4-Methoxy-3-hydroxy-cinnamic acid (isoferulic acid)
9	G-CO–CH(OH)-CH_3[a]
10	G-CH_2-COOH[a]
11	3-Methoxy-4-hydroxy-cinnamic acid (ferulic acid)
12	4-Methoxy-o-hydroxy-benzenacetic acid
13	G-CH(OH)-CO-CH_3[a]
14	5-Methoxy-2-hydroxy-benzoic acid
15	2-Methoxy–hydroxy-benzenacetic acid
16	4-Methoxy–hydroxy-benzenacetic acid
Aliphatic acids	
17	Acetic acid
18	4-Oxo-pentanoic acid
19	2-Methyl-2-hydroxybutanoic acid
20	3-Hydroxy-propanoic acid
21	Methyl propanedioic acid
22	Methyl butanedioic acid
23	2-Butanedioic acid
24	Hydroxybutanedioic acid
25	Hexanedioic acid
26	2-Hydroxypentanedioic acid
27	2-Hydroxy-2-pentenedioic acid
28	Hexadecanoic acid (palmitic acid)
29	Ethanedioic acid
30	9,12-Octadecadienoic acid
31	9-Octadecanoic acid (oleic acid)
32	Octadecanoic acid (stearic acid)
33	2,3-Dihydroxypropandioic acid
34	2,4-Hexadienedioic acid
35	Dimethyl-propanedioic acid
36	2-Methyl butanoic acid
37	3,3-Dihydroxy-2-propenoic acid
38	3-Methyl-2-hydroxy-2-butenoic acid
39	Sebacic acid
Furan compounds	
40	2-Furancarboxylic acid
41	2-Furanacetic acid
42	5-Hydroxymethylfurancarboxylic acid

[a] G, guaiacyl group

Acknowledgments The authors wish to express their sincere thanks for financial assistance from the National Agricultural Innovation Project (NAIP)-Indian Council of Agricultural Research (ICAR) under component 2 and International Fund for Agriculture Development (IFAD) through Grant no. 974.

References

AICSIP (All India Coordinated Sorghum Improvement Project). 2010. Sweet sorghum physiology. All India coordinated sorghum improvement project annual progress report for 2009–2010. In: Proceedings of the 40th Annual Sorghum Group Meeting-Agm10 (Book 2 of 4), p. 156. Directorate of *Sorghum* Research, Rajendranagar, Hyderabad, Andhra Pradesh, India.

Bretschneider, R., Rehor., J., and Zdarsky, J. 1976. Method of post-harvest treatment of sugarcane. US Patent 3975204.

Desai, B.B., P.B. Sangle, and S.L. Gaur. 1985. Chemical control of post harvest losses in sugarcane. *Current Research Report* 1: 33.

Eggleston, G. 2002. Deterioration of cane juice-sources and indicators. *Food Chemistry* 78: 99–107.

Eggleston, G., B.L. Legendre, and C. Richard. 2001. Effect of harvest method and storage time on cane deterioration. I. Cane quality changes. *International Sugar Journal* 103: 331–338.

Hanko, V.P., and J.S. Rohrer. 2000. Determination of carbohydrates, sugar alcohol and glycols in cell cultures and fermentation broths using high performance ion exchange chromatographs with pulsed amperometric determination. *Analytical Biochemistry* 283: 192–199.

Huang, H., X. Guo, D. Li, M. Liu, J. Wu, and H. Ren. 2011. Identification of crucial yeast inhibitors in bio-ethanol and improvement of fermentation at high pH and high total solids. *Bioresource Technology* 102: 7486–7493.

Janakiramaiah, K., Satyanarayana, N., and Murty, T.S.N. 1967. The deterioration of cane juices by *Leuconostoc mesenteroides.* In: Proceedings of Annual Conference V STAI 35 (part I), vol. 127, pp. 85–90.

Kumar, C.G., A. Fatima, P. Srinivasa Rao, B.V.S. Reddy, A. Rathore, R. Nageswar Rao, S. Khalid, and A. Kamal. 2010. Characterization of improved sweet sorghum genotypes for biochemical parameters, sugar yield and its attributes at different phenological stages. *Sugar Tech* 12: 322–328.

Lingle, S.E. 2010. Opportunities and challenges of sweet *sorghum* as a feedstock for biofuel. In: Sustainability of the sugar and sugar—ethanol industries, chap. 11, ACS Symposium. Series 1058, pp. 177–188.

Lionnet, G.R.E. 1996. A review of cane quality in South Africa and its impact on factory performance. *Proceedings of ISSCT* 22: 103–113.

Luo, C., D.L. Brink, and H.W. Blanch. 2002. Identification of potential fermentation inhibitors in conversion of hybrid poplar hydrolyzate to ethanol. *Biomass and Bioenergy* 22: 125–138.

Nichols, N.N., B.S. Dien, and M.A. Cotta. 2010. Fermentation of bioenergy crops into ethanol using biological abatement for removal of inhibitors. *Bioresource Technology* 101: 7545–7550.

Palmqvist, E., and B. Hahn-Hagerdhal. 2000. Fermentation of lignocellulosic hydrolysates. II. Inhibitors and mechanism of inhibition. *Bioresource Technology* 74: 25–33.

Purchase, B.S. 2001. Losses caused by microorganisms. *Proceedings ISSCT* 24: 379.

Rosaio, E.J., and S. Santisopasri. 1977. Characterization and inhibition of invertase in sugarcane juice. *Phytochemistry* 16: 443–445.

Sharma, K.P, Rattana, B.K, Kanwar, R.S. 1989. Improvement in sugar recovery by preventing bacterial polysaccharide production in cane juice. Bharatiya Sugar, 15–18.

Singh, P., N. Arya, P. Tiwari, A. Suman, R.K. Rai, A.K. Shrivastava, and S. Solomon. 2008. Use of glutaraldehyde and benzalkonium chloride for minimizing post-harvest physio-chemical and microbial changes responsible for sucrose losses in sugarcane. *Journal of Agricultural and Food Chemistry* 56: 7176–7183.

Solomon, S. 2009. Post-harvest deterioration of sugarcane. *Sugar Tech* 11: 109–123.

Solomon, S., H.N. Shahi, A. Suman, A. Gaur, S. Deb, and I. Singh. 2001. A survey of post-harvest biological losses in Indian sugar factories: An emerging challenge. *Proceedings of ISSCT* 24: 380–381.

Solomon, S., R. Banerji, A.K. Srivastava, P. Singh, I. Singh, M. Verma, C.P. Prajapati, and A. Sawnani. 2006. Post-harvest deterioration of sugarcane and chemical methods to minimize sucrose losses. *Sugar Tech* 8: 74–78.

Srinivasarao, P., Rao, S.S., Seetharama, N., Umakanth, A.V., Sanjana Reddy, P., Reddy, B.V.S., and Gowda, C.L.L. 2009. Sweet sorghum for biofuel and strategies for its improvement. Information Bulletin no. 77, International Crops Research Institute for the Semi-Arid Tropics, Patancheru 502324, Andhra Pradesh, India. pp 80. ISBN 978-92-9066-518-2.

Srinivasarao, P., P.S. Reddy, A. Rathore, B.V.S. Reddy, and S. Panwar. 2011. Application GGE biplot and AMMI model to evaluate sweet sorghum hybrids for genotype × environment interaction and seasonal adaptation. *Indian Journal of Agricultural Sciences* 81: 438–444.

Tilbury, R.H., Hollingsworth, B.S., Graham, S.D., Pottage, P. 1977. Mill sanitation—a fresh approach to biocide evaluation. In: Proceedings of XVI Congress ISSCT, pp. 2749–2767.

Tomar, B.S., and S.S. Malik. 2004. Micronutrients impact on growth, yield and staleness of sugarcane variety CoS 8436. *Co-operative Sugar* 36: 255–256.

Zhao, Y.L., A. Dolat, Y. Steinberger, X. Wang, A. Osman, and G.H. Xie. 2009. Biomass yield and changes in chemical composition of sweet sorghum cultivars grown for biofuel. *Field Crops Research* 111: 55–64.