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RESEARCH ARTICLE

Enhancing the Shelf Life of Sweet Sorghum [*Sorghum bicolor* (L.) Moench] Juice Through Pasteurization While Sustaining Fermentation Efficiency

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Abstract The influence of pasteurization on storage stability of sweet sorghum juice and subsequent bioconversion to ethanol was studied. Juice samples were pasteurized at three different temperatures, i.e., 70 °C for 10 min, 80 °C for 5 min and 90 °C for 2 min and were further stored at three different temperatures of 35, 40 and 45 °C. The storage shelf life of the sorghum juice was observed to be extended for 21 days without compromising the ethanol conversion efficiency. Consistent fermentation efficiencies were observed for the juice samples pasteurized at 70 °C followed by storage at 45 °C, pasteurized at 80 °C followed by storage at 40 °C and pasteurized at 90 °C followed by storage at 35 °C and these samples showed an ethanol yield in the range of 0.473–0.477, 0.461–0.47 and 0.466–0.473 g g^{-1}, respectively. Hence, the juice samples pasteurized at 90 °C and stored at 35 °C was deemed as the superior preservation condition as it was close to ambient temperature and increased the shelf life of sweet sorghum juice. The highest fermentation efficiency of 93 % was observed after 48 h of fermentation.

Keywords Sweet sorghum juice · Pasteurization · Shelf life · Ethanol · Storage

Soma Gupta and Jayalakshmi Malapaka contributed equally to this study.

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Introduction

Biomass as a primary renewable energy resource for biofuels generation has gained immense importance in the last few decades and utilization of ethanol from biomass is predicted to increase from 1.0 mboe day^{-1} in 2010 to 3.4 mboe day^{-1} in 2035 (Anonymous 1998), which includes organic and animal wastes, wastewater, energy crops, agricultural and industrial residues (Antonopoulou et al. 2008). Sweet sorghum (*Sorghum bicolor* (L.) Moench) is a C4 plant possessing high photosynthetic activity and drought tolerance that can be cultivated in all temperatures including tropical and temperate climatic areas requiring minimal quantity of water and fertilizer unlike other crops (Rao et al. 2009, 2011). It has been deemed as a potential feedstock for biofuels production since it has approximately equal quantities of soluble (glucose and sucrose) and insoluble (cellulose and hemicellulose) carbohydrates (Yu et al. 2012). These features make the production of biofuels such as ethanol from this feedstock juice advantageous (Anderson 2005).

Sweet sorghum juice has a short shelf life and prone to microbial spoilage due to the presence of sugars (Krishnakumar and Devadas 2006). Sweet sorghum juice like sugarcane juice gets affected by chemical (acid) and enzymatic inversion (Singh et al. 2006) due to the presence of both neutral invertase (NI) and acid invertase (AI). These enzymes cause sucrose inversion, the reason being their high correlation with sucrose and reducing sugar content during plant growth (Siswoyoa et al. 2007). The existing propensities for juice preservation depends on the utilization of the methods that assures qualitative products, high nutritional value and safe from a microbiological perspective at the downstream step of yeast fermentation which is critical for the viability of the whole value chain

(Gould 2000; Ranken et al. 2005; Rao et al. 2012). Thus, the preservation and storage of sweet sorghum juice is needed for its further utilization in ethanol production, as an alternate energy source that is renewable, sustainable, efficient, cost-effective, convenient and safe (Gould 2000; Chum and Overend 2001). The different preservation methods used in the food industry are the removal of water content, controlling temperature, freezing, drying, pH control, irradiation, vacuum packaging, modified atmosphere packaging, aseptic packaging, acidification, fermentation, heating (pasteurization and sterilization) and addition of chemical preservatives (Ranken et al. 2005). The potential methods employed in the food preservation can thus be divided into physical, physicochemical, microbial-derived and miscellaneous, among which, the most important ones are high temperature, low temperature, water activity, acidity, redox potential (Eh), competitive microorganism (e.g. lactic acid bacteria) and preservatives (e.g. nitrite, sorbate, sulphite) (Leistner and Gorris 1995).

Pasteurization is one of the effective and widely practised preservation method employed in the food industry since heating at higher temperature kills a major fraction of microbes in foods stored in both room and refrigerated temperatures (Karmakar et al. 2010). The method of food preservation using pasteurization has been adapted for many fruits juices such as Nagpur mandarin (*Citrus reticulata* Blanco) (Pareek et al. 2011), aonla juice (Bhattacherjee et al. 2011), kinnow juice blends (Bhardwaj and Mukherjee 2011) as well as widely used in sugarcane juice preservation (Chauhan et al. 2002; Karmakar et al. 2010; Sankhla et al. 2012). The aim of the present study is to evaluate the effect of pasteurization on the stability of sweet sorghum juice so as to enhance its shelf life and also to study their effect on ethanol fermentation by yeast.

Materials and Methods

Crop Cultivation and Management

The sweet sorghum cultivar, ICSV 93046, was grown during the post-rainy (*rabi*) season (October–February), 2010–2011 in vertisols of the experimental farm of the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), located in Patancheru, Andhra Pradesh, India (altitude 545 m above mean sea level, latitude 17.53°N and longitude 78.27°E). This cultivar was sown in a plot size of 3 m × 4 m, i.e. four rows of 4 m long spaced at 75 cm × 15–20 cm. The planting was done on ridges with a plant stand of about 100,000 ha^{-1}. Sweet sorghum was initially planted dense but 15 days after seedling emergence (DAS) thinned to one plant in each hill. Hand weeding was done followed by two inter-cultivations. Surface irrigation was applied in furrows to the crop to maintain proper growth. Standard agronomic package of practices (80-40-0 NPK ha^{-1}; 2/3rd N and total P as basal dose and 1/3rd at 25 DAS) and plant protection measures were adopted throughout the crop growth period in all the plots. At flowering, sorghum heads were covered with nylon bags for protection against bird damage on the developing grain. All the four rows were harvested at physiological maturity (when hilum turns black). The stalks were squeezed once to extract the juice on a three-roller cane press mill. The juice was sieved through a muslin cloth to remove the plant parts that may come while extracting the juice. The juice was collected into sterile sample bottles and then transported under cold ice-jacketed conditions to the laboratory for further analysis. Data on juice yield (t ha^{-1}), pH and stalk yield (t ha^{-1}) were collected following standard procedures for each plot (Rao et al. 2011). The sugar yield (t ha^{-1}) was estimated as the product of Brix % and juice yield (t ha^{-1}) (Wortmann et al. 2010).

Microorganism and Inoculum Preparation

Saccharomyces cerevisiae strain ICTY 417 previously isolated and maintained in the in-house culture collection of CSIR—Indian Institute of Chemical Technology, Hyderabad, India was cultured in yeast extract-malt extract (YM) medium at 30 °C and agitated on a gyratory shaker at 150 rev min^{-1} for 18 h. The actively growing cells in the broth with an absorbance of about 0.5 at 600 nm which corresponded to 10^6 cells ml^{-1} was used as inoculum for ethanol production.

Heat Treatment for Pasteurization

Sweet sorghum juice (1 l each) was taken in 18 Erlenmeyer flasks that were divided into three sets consisting of six flasks in each set. Each set of juice were pasteurized at three different temperatures, i.e., 70, 80 and 90 °C for 10, 5 and 2 min, respectively. One flask from each set was stored under 35, 40 and 45 °C. Additional unpasteurized sweet sorghum juice samples were taken as blank, which were also maintained at all above mentioned three temperatures. Experiments were carried out in triplicates and the analysis for different sugars (glucose, fructose and sucrose) and ethanol yield was carried out at 24 h periodic intervals for 72 h.

Fermentation Studies

Hundred milliliter aliquots of each of the pasteurized and blank sweet sorghum juice samples was used for the zero

hour study to which mineral salts [0.05 % $MgSO_4$ and 0.2 % $(NH_4)_2SO_4$] were added and autoclaved at 121 °C for 20 min. The flasks were then cooled and inoculated with 1 ml of the fresh grown yeast inoculum (OD_{600} 0.5), incubated at 30 °C with agitation at 150 rev min^{-1}. Further, at periodic intervals of 48 h till 13th day after which the interval was doubled to 96 h for the last two sets of samples (17th and 21st day), 100 ml of the juice samples from each of the three sets of pasteurized temperatures along with the blank and stored at three different temperatures was taken and processed as described above. This sampling process was continued till 72 h at every 24 h interval. The fermented samples were taken from the inoculated flasks after every 24 h, centrifuged at 8,000 rpm for 10 min for cell separation and the cell-free supernatants were subjected to gas chromatography (GC) analysis to determine the amount of ethanol produced. The reducing sugar content present in the juice samples before and after fermentation was also analyzed by dinitrosalicylic acid (DNS) method (Miller 1959).

Analytical Methods

Sugar concentration in terms of Brix (%) was measured using a hand-held pocket refractometer (Model PAL, Atago Co. Ltd., Tokyo, Japan) (Zoecklien et al. 1995). The pH was recorded using a microprocessor-based pH meter (Model DPH506, Global Electronics, Hyderabad, India). Between two different sample readings, the refractometer and the pH meter were cleaned with distilled water and dried with a paper towel. The sweet sorghum juice was centrifuged at 10,000 rev min^{-1} for 10 min and total soluble sugars (TSS) content in the supernatant was determined using the phenol sulphuric acid method (Dubois et al. 1956), while the reducing sugar content was determined using the 3,5-dinitrosalicylic acid (DNS) method (Miller 1959). Sugar profiling to determine the contents of individual hexose sugars, like glucose, fructose and sucrose, present in the extracted sweet sorghum juice were analyzed on a HPLC system (Kumar et al. 2010).

In addition, ethanol concentrations (P, g l^{-1}) in the samples were analyzed using the gas chromatograph (Model GC2014, Shimadzu, Japan) equipped with a flame ionization detector and interfaced with a Zebron ZB-624 column (Phenomenex Inc., USA) having dimensions of 30 m × 0.53 mm × 3.00 µm, and set at 60 °C. Ethanol (GR) and isopropanol (GR) were used for the standard curve and as an internal standard, respectively. Operation conditions: Oven temperature was 60 °C; injecting temperature was 250 °C using nitrogen as carrier gas and hydrogen as a flaming gas both at a flow rate of 41 ml min^{-1} with a column flow rate of 1.9 ml min^{-1}; flame ionization detector temperature was 280 °C; helium

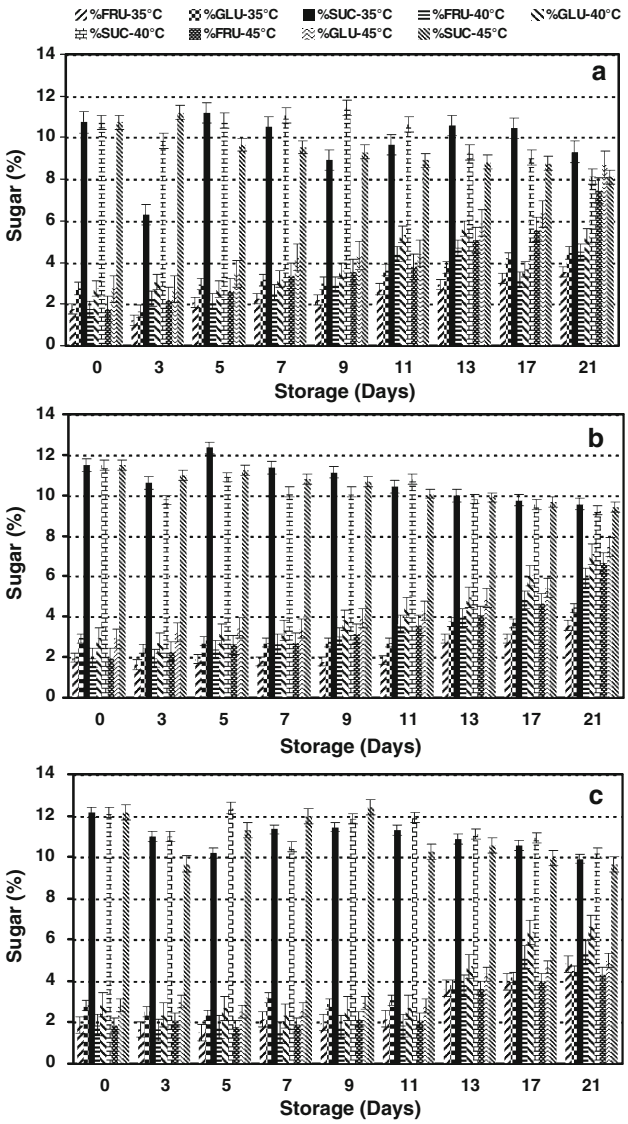

Fig. 1 Sugar analysis of sweet sorghum juice samples pasteurized at **a** 70 °C, **b** 80 °C and **c** 90 °C

gas was used for cooling the column. Head pressure was 11.5 kPa with a 25:1 split ratio; sample volume was 1 µl. All experiments were carried out in triplicates and the data values are represented as mean ± standard error (S.E.) and the S.E. values are shown as *Y*-error bars in all figures.

Kinetic studies were also carried out for the fermented samples. The ethanol concentration estimations were performed at periodic intervals of 4 h up to 60 h. The ethanol yield ($Y_{p/s}$) was calculated as the actual ethanol produced and expressed as g ethanol per g total sugar utilized (g g^{-1}). The ethanol productivity (Q_p, g l^{-1} h^{-1}) and the percentage of conversion efficiency or yield efficiency (E_y) were calculated using the following equations (Laopaiboon et al. 2007):

$$Q_P = P/t \quad \text{and} \quad E_Y = \left(Y_{p/s} \times 100\right)/0.51$$

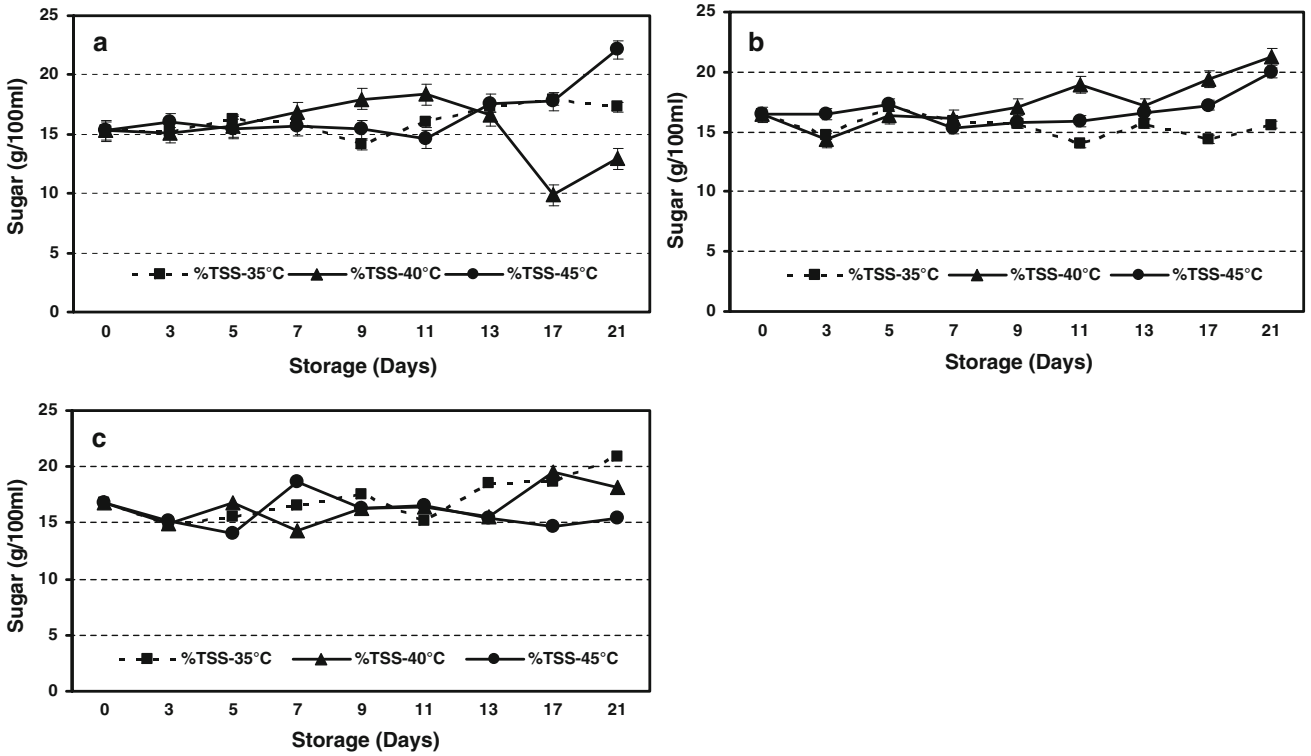

Fig. 2 TSS and reducing sugar analysis of sweet sorghum juice samples pasteurized at **a** 70 °C, **b** 80 °C and **c** 90 °C

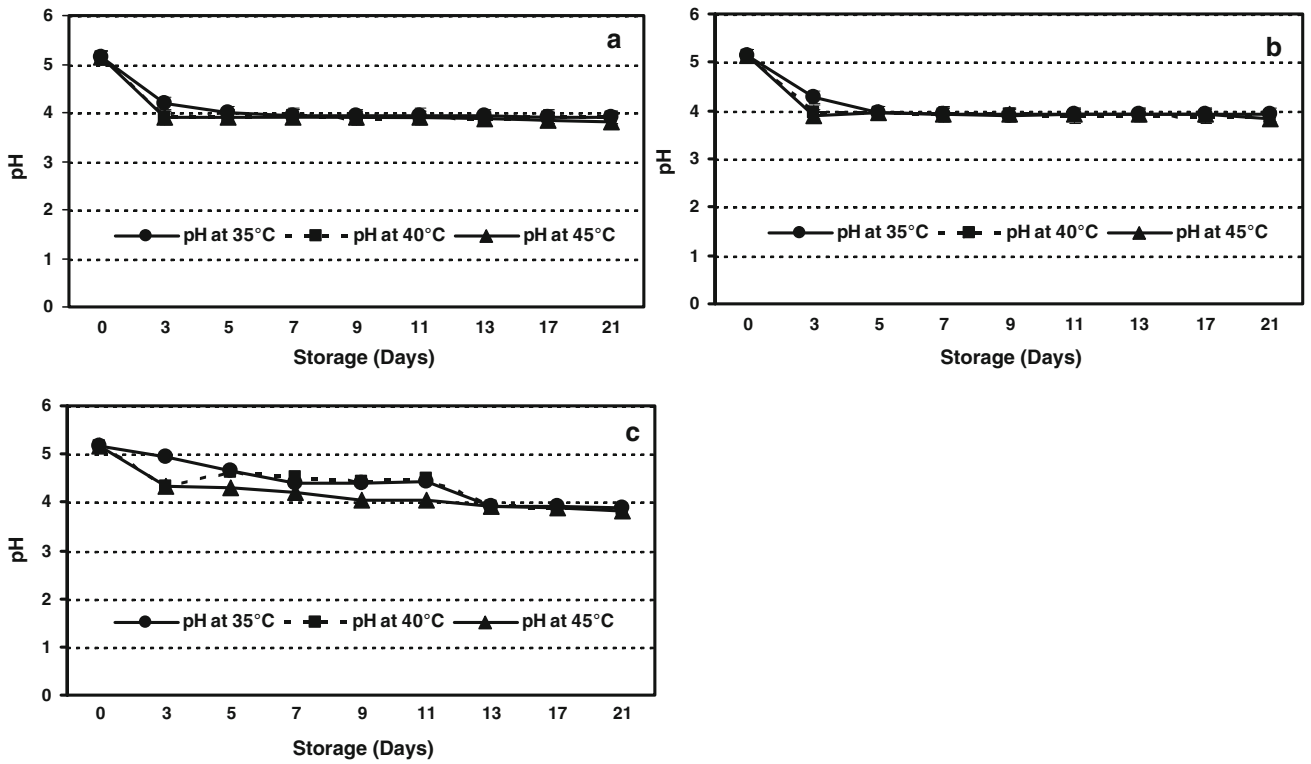

Fig. 3 pH analysis of sweet sorghum juice samples pasteurized at **a** 70 °C, **b** 80 °C and **c** 90 °C

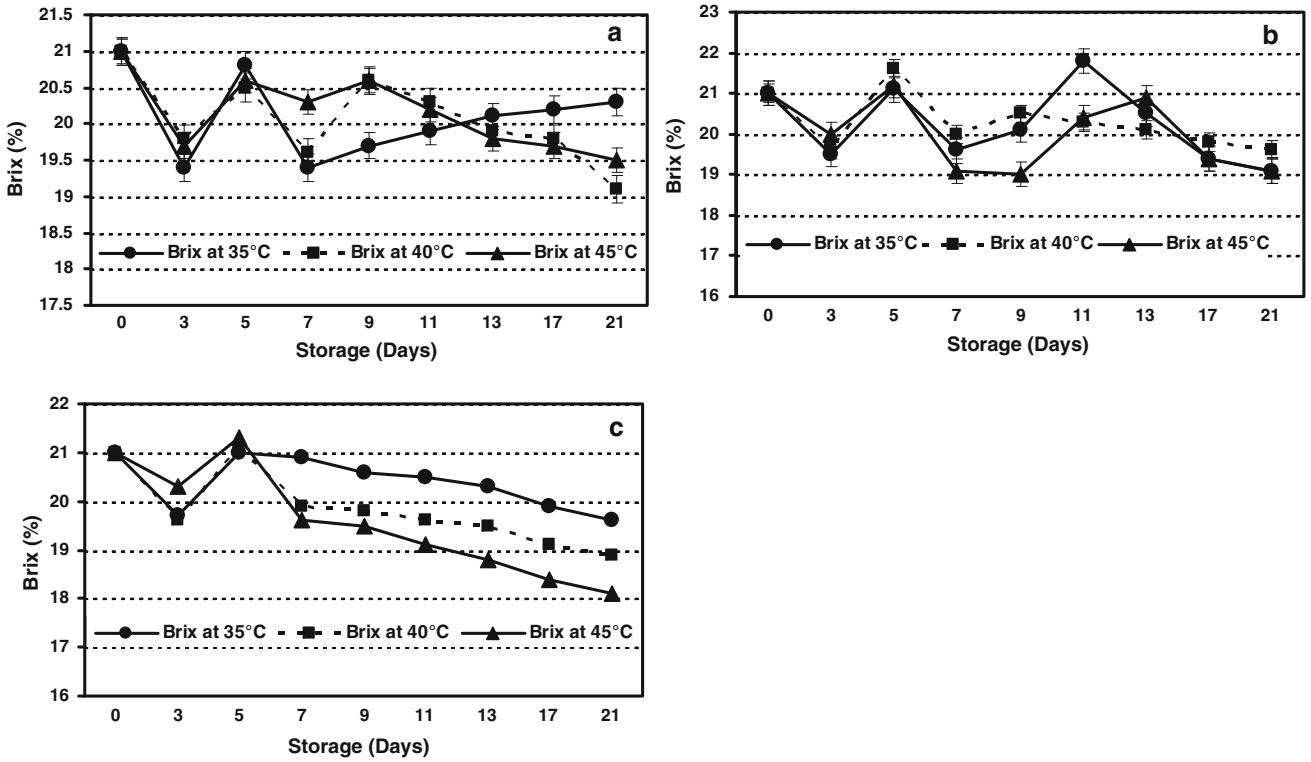

Fig. 4 Brix analysis of sweet sorghum juice samples pasteurized at **a** 70 °C, **b** 80 °C and **c** 90 °C

where P is the actual ethanol concentration produced (g l^{-1}), t is the fermentation time (h) giving the highest ethanol concentration and 0.51 is the maximum theoretical ethanol yield of glucose consumption.

Results and Discussion

Sugar Analysis as a Function of Pasteurization Carried Out and Stored at Different Temperatures

The pasteurization studies were carried out on fresh sweet sorghum juice samples at different temperature conditions. The fresh juice samples were first pasteurized at three different temperatures, that is, 70, 80 and 90 °C which was further incubated at three different temperatures viz., 35, 40 and 45 °C for 21 days. The results suggest that the amount of total soluble sugars and the percentage of hexose sugars like glucose, fructose and sucrose as a function of time did not show significant changes over the period of time. It was also observed that the amount of reducing sugars increased, while the amount of non-reducing sugars decreased with an increase in the storage time as a result of breakdown of non-reducing sugar (sucrose) to reducing sugars (Figs. 1, 2). From the figures, it can be inferred that the fructose and glucose content increased, while the sucrose content decreased in the case of the samples pasteurized at 70 °C and stored at 35, 40, and 45 °C. Similarly, the fructose and glucose content increased in case of samples pasteurized at 80 °C and stored at 35, 40 and 45 °C. The sucrose content also decreased for the samples pasteurized at 80 °C and stored at 35, 40 and 45 °C. However, there was no much significant changes observed in case of total soluble sugar content for the samples pasteurized at 80 °C and stored at 35, 40 and 45 °C. However, in case of samples pasteurized at 80 °C and stored at 35 °C, the fructose and glucose contents increased from 1.85 to 4.84 and 2.83 to 4.46 %; at 40 °C, it increased from 1.85 to 5.41 and 2.83 to 6.61 % and at 45 °C storage temperature, it increased from 1.85 to 4.31 and 2.83 to 4.99 %, respectively. In case of samples pasteurized at 90 °C and stored at 35, 40 and 45 °C, the sucrose content decreased from 12.15 to 9.91, 12.15 to 10.18 and 12.15 to 9.62 %, respectively. The changes observed in total soluble sugar content for samples pasteurized at 90 °C and stored at 35, 40 and 45 °C were from 16.85 to 20.92, 16.85 to 18.2 and 16.85 to 15.43 %, respectively. The observed data was comparable with an earlier study where the juice samples stored at room temperature ($\approx$25 °C), resulted in a sharp decline in the sucrose content of the total soluble sugar content to 31 % after the fifth day (Wu et al. 2010). The effect of pasteurization thus, increased the storage shelf life of the fresh sweet sorghum juice from 5 h to 3 weeks. The

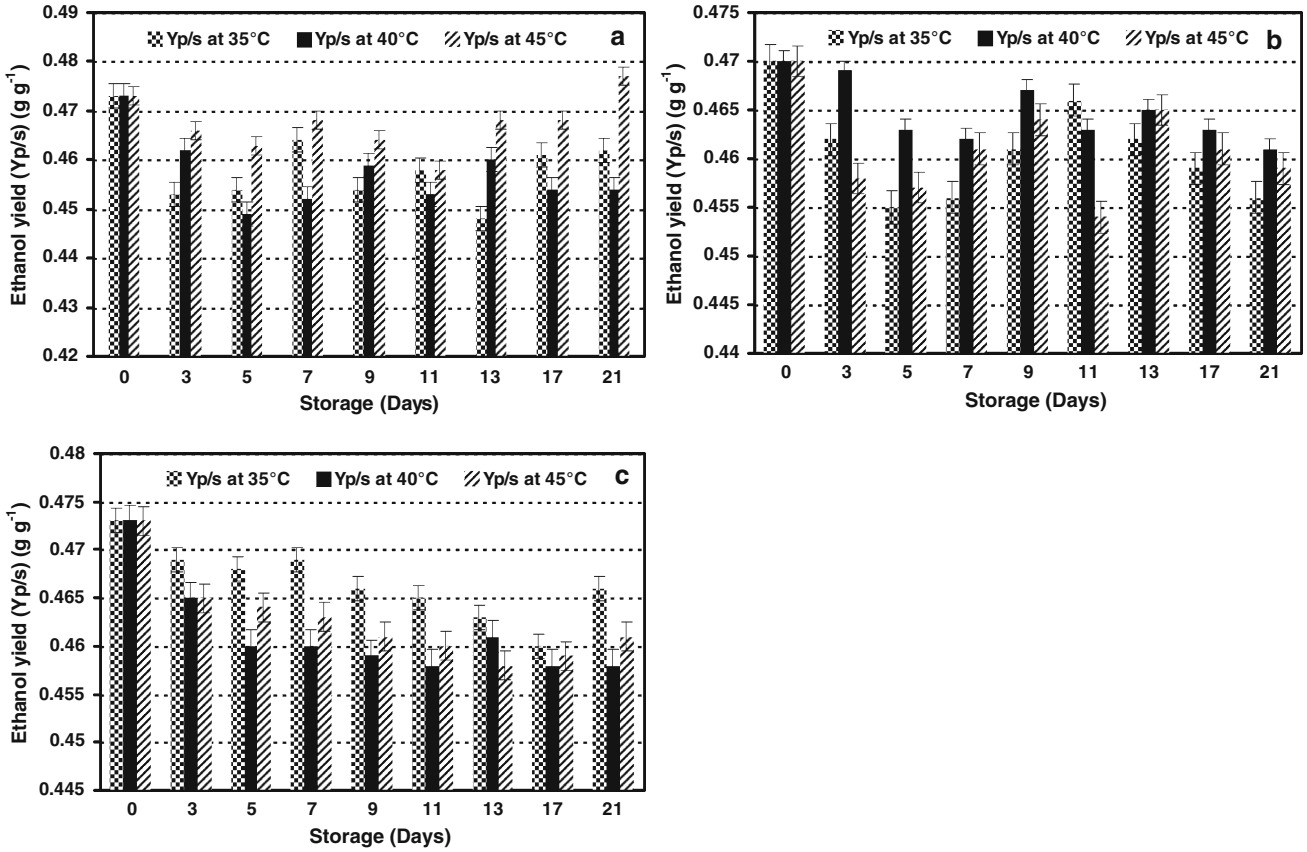

Fig. 5 Fermentation analysis of sweet sorghum juice pasteurized at **a** 70 °C, **b** 80 °C and **c** 90 °C

fresh sorghum juice (control sample) deterioration was observed with an obvious browning and rapid increase in the viscosity (visual observation) which may be due to the fermentation by spoilage microflora within 12 h. Later, all these pasteurized samples were subjected to fermentation to check their fermentation ability.

Changes in pH and Brix % Values as a Function of Pasteurization Time

The results depicted in Fig. 3, showed that the pH changes observed during the fermentation process of the sorghum juice were not that significant and comparable. The pH values in case of all the fermentation experiments of the samples pasteurized at 70, 80 and 90 °C and stored at 35, 40 and 45 °C decreased from pH 5 and remained fairly constant around pH 4. This negligible change in the pH is plausibly due to the release of carbon dioxide, which in turn was converted to carbonic acid and produced carbonate ions and protons, and thus the pH of the fermented juice maintained at a relatively constant value (Shen et al. 2004). This decrease in pH also aided in the prevention of the growth of spoilage microbes resulting in enhancement of the storage shelf life of the sorghum juice. The minimal changes in the pH values of the sweet sorghum juice for the entire period of the 21 days of experiments showed stability in the ethanol production. These results were comparable with some earlier fermentation studies carried out on sweet sorghum juice under different conditions (Khongsay et al. 2010; Ariyajarearnwong et al. 2011). Further, the changes in the brix values in the pasteurized samples were found to be comparatively in a steady state as evident from Fig. 4. The Brix values showed slight fluctuation as it reduced slightly and then again increased slightly. The almost near consistency in the brix value of the sweet sorghum juice samples evidently showed the maintenance of total soluble sugars which in turn will influence the consistency in the fermentation of the pasteurized juice samples.

Ethanol Production as a Function of Fermentation Time

The ethanol production paralleled with the growth of the yeast in the submerged culture medium. However, the fermentation of fresh juice showed maximum concentration of ethanol (0.69 g g^{-1}) after 48 h with the onset of the stationary phase of growth, after which a reduction in the ethanol concentration was observed (Fig. 5). Since the

Table 1 Kinetic parameters of batch ethanol production as a function of fermentation time by *Saccharomyces cerevisiae* strain ICTY417 using sweet sorghum juice samples (pasteurization at 70 °C for 48 h fermentation)

Juice samples (storage period / storage temperature)	Parameters (mean ± S.E.)			
	P^a (g l^{-1})	$Y^b_{p/s}$ (g g^{-1})	Q^c_p (g l^{-1} h^{-1})	E^d_y (%)
0 h Storage period for 70 °C	65.47 ± 0.074	0.473 ± 0.440	1.343 ± 0.251	93.58 ± 0.108
3rd Day (35 °C)	63.44 ± 0.374	0.453 ± 0.412	1.322 ± 0.576	89.76 ± 0.728
3rd Day (40 °C)	61.92 ± 0.209	0.462 ± 0.266	1.29 ± 0.581	90.52 ± 0.810
3rd Day (45 °C)	60.7 ± 0.308	0.466 ± 0.488	1.264 ± 0.651	91.32 ± 0.923
5th Day (35 °C)	62.94 ± 0.091	0.454 ± 0.487	1.311 ± 0.362	90.82 ± 0.705
5th Day (40 °C)	60.25 ± 0.194	0.449 ± 0.746	1.255 ± 0.680	89.45 ± 0.629
5th Day (45 °C)	61.76 ± 0.931	0.463 ± 0.407	1.287 ± 0.692	92.45 ± 0.875
7th Day (35 °C)	63.22 ± 0.340	0.464 ± 0.404	1.317 ± 0.357	91.67 ± 0.772
7th Day (40 °C)	61.25 ± 0.146	0.452 ± 0.561	1.276 ± 0.389	90.67 ± 0.795
7th Day (45 °C)	61.82 ± 0.092	0.468 ± 0.808	1.288 ± 0.491	92.06 ± 0.673
9th Day (35 °C)	63.49 ± 0.618	0.454 ± 0.297	1.323 ± 0.305	90.89 ± 0.037
9th Day (40 °C)	60.7 ± 0.610	0.459 ± 0.471	1.265 ± 0.584	92.74 ± 0.904
9th Day (45 °C)	61.38 ± 0.161	0.464 ± 0.509	1.277 ± 0.714	93.13 ± 0.273
11th Day (35 °C)	62.68 ± 0.971	0.458 ± 0.451	1.306 ± 0.728	89.76 ± 0.609
11th Day (40 °C)	60.7 ± 0.484	0.453 ± 0.847	1.265 ± 0.892	89.81 ± 0.491
11th Day (45 °C)	60.89 ± 0.114	0.458 ± 0.230	1.269 ± 0.147	89.72 ± 0.408
13th Day (35 °C)	63.22 ± 0.106	0.448 ± 0.099	1.317 ± 0.579	91.82 ± 0.627
13th Day (40 °C)	60.81 ± 0.470	0.46 ± 0.309	1.267 ± 0.672	90.21 ± 0.182
13th Day (45 °C)	60.41 ± 0.493	0.468 ± 0.467	1.259 ± 0.877	92.47 ± 0.220
17th Day (35 °C)	63.76 ± 0.368	0.461 ± 0.476	1.328 ± 0.119	90.71 ± 0.375
17th Day (40 °C)	61.79 ± 0.589	0.454 ± 0.374	1.287 ± 0.772	89.03 ± 0.191
17th Day (45 °C)	62.14 ± 0.491	0.468 ± 0.701	1.295 ± 0.684	91.33 ± 0.275
21st Day (35 °C)	64.3 ± 0.402	0.462 ± 0.905	1.339 ± 0.557	89.48 ± 0.621
21st Day (40 °C)	60.7 ± 0.835	0.454 ± 0.106	1.265 ± 0.721	90.05 ± 0.040
21st Day (45 °C)	61.71 ± 0.676	0.477 ± 0.351	1.285 ± 0.117	90.96 ± 0.237

[a] P, actual ethanol concentration produced

[b] $Y_{p/s}$, ethanol yield

[c] Q_p, ethanol productivity

[d] E_y, percentage of conversion efficiency or yield efficiency

optimal ethanol production was observed at 48 h, the later fermentation studies were carried out for 48 h. The decrease in the ethanol production after 48 h indicates the end of stationary phase of the organism. The supplementation of the sweet sorghum juice with ammonium sulphate as substrate contributed to the yield and productivity of the ethanol production (Laopaiboon et al. 2007).

The total soluble sugars and total soluble solids present at the beginning of the fermentation were 170 g l^{-1} and 13.03 Brix %, respectively. The fermentation was carried out for all the samples pasteurized at 70, 80 and 90 °C further stored at three different temperatures of 35, 40 and 45 °C. The ethanol yield (g g^{-1}) as a function of fermentation time was estimated for the pasteurized samples (Fig. 5) at different time durations like 0, 24, 48, 72 and 96 h. The ethanol yield was found to be highest after 48 h of fermentation in case of all the three pasteurized samples. After 48 h, the ethanol production decreased which might be due to the ethanol feedback inhibition. The blank (unpasteurized sample) showed similar tendency. The ethanol production in the pasteurized samples in all conditions showed a relatively constant trend whereas, the ethanol production in control was found to be comparatively lower than that of the pasteurized samples. The kinetic parameter studies of the control (unpasteurized sample) showed an initial ethanol production efficiency value of 88 % with the yield of 0.453 g g^{-1} on the first day sample at 48 h of fermentation. The second day sample showed lower ethanol production efficiency and yield, that is 75 % and 0.332 g g^{-1} at 48 h of fermentation. The

Table 2 Kinetic parameters of batch ethanol production as a function of fermentation time by *Saccharomyces cerevisiae* strain ICTY417 from sweet sorghum juice samples (pasteurization at 80 °C for 48 h fermentation)

Juice samples (storage period /storage temperature)	Parameters (mean ± S.E.)			
	P^a (g l^{-1})	$Y^b_{p/s}$ (g g^{-1})	Q^c_p (g l^{-1} h^{-1})	E^d_y (%)
0 h Storage period for 80 °C	63.98 ± 0.209	0.471 ± 0.581	1.332 ± 0.692	93.03 ± 0.875
3rd Day (35 °C)	63.98 ± 0.140	0.462 ± 0.615	1.332 ± 0.581	90.11 ± 0.717
3rd Day (40 °C)	62.46 ± 0.676	0.469 ± 0.412	1.301 ± 0.181	92.63 ± 0.609
3rd Day (45 °C)	60.48 ± 0.351	0.458 ± 0.925	1.26 ± 0.752	89.78 ± 0.273
5th Day (35 °C)	62.94 ± 0.117	0.455 ± 0.557	1.311 ± 0.365	90.81 ± 0.118
5th Day (40 °C)	59.17 ± 0.237	0.463 ± 0.611	1.232 ± 0.221	92.23 ± 0.857
5th Day (45 °C)	61.76 ± 0.040	0.457 ± 0.265	1.287 ± 0.172	89.64 ± 0.671
7th Day (35 °C)	64.3 ± 0.721	0.456 ± 0.674	1.339 ± 0.626	91.56 ± 0.578
7th Day (40 °C)	61.25 ± 0.106	0.462 ± 0.711	1.276 ± 0.438	91.33 ± 0.145
7th Day (45 °C)	61.82 ± 0.835	0.461 ± 0.481	1.287 ± 0.491	90.39 ± 0.892
9th Day (35 °C)	63.49 ± 0.728	0.461 ± 0.106	1.326 ± 0.037	89.49 ± 0.374
9th Day (40 °C)	62.33 ± 0.714	0.467 ± 0.454	1.299 ± 0.673	91.32 ± 0.412
9th Day (45 °C)	61.38 ± 0.374	0.464 ± 0.971	1.278 ± 0.808	90.03 ± 0.581
11th Day (35 °C)	64.3 ± 0.476	0.466 ± 0.161	1.339 ± 0.491	90.77 ± 0.651
11th Day (40 °C)	62.87 ± 0.467	0.463 ± 0.471	1.309 ± 0.772	93.30 ± 0.362
11th Day (45 °C)	60.89 ± 0.309	0.454 ± 0.584	1.268 ± 0.357	90.05 ± 0.209
13th Day (35 °C)	63.76 ± 0.098	0.462 ± 0.904	1.328 ± 0.424	91.92 ± 0.576
13th Day (40 °C)	62.44 ± 0.231	0.465 ± 0.305	1.301 ± 0.610	92.57 ± 0.680
13th Day (45 °C)	63.12 ± 0.837	0.465 ± 0.297	1.315 ± 0.340	91.23 ± 0.629
17th Day (35 °C)	63.76 ± 0.451	0.459 ± 0.618	1.328 ± 0.875	89.70 ± 0.705
17th Day (40 °C)	61.79 ± 0.519	0.463 ± 0.092	1.287 ± 0.692	90.03 ± 0.923
17th Day (45 °C)	62.14 ± 0.569	0.461 ± 0.795	1.294 ± 0.407	90.32 ± 0.810
21st Day (35 °C)	64.3 ± 0.453	0.456 ± 0.389	1.339 ± 0.728	88.79 ± 0.266
21st Day (40 °C)	60.7 ± 0.471	0.461 ± 0.561	1.265 ± 0.308	93.39 ± 0.488
21st Day (45 °C)	61.71 ± 0.104	0.459 ± 0.146	1.285 ± 0.487	89.93 ± 0.746

[a] P, actual ethanol concentration produced

[b] $Y_{p/s}$, ethanol yield

[c] Q_p, ethanol productivity

[d] E_y, percentage of conversion efficiency or yield efficiency

production of ethanol further reduced with each consecutive day and was found to be minimal by fifth day that showed a fermentation efficiency of just as low as 40 % and yield of 0.102 g g^{-1}. The reduced ethanol production exhibited by the control is as a result of rapid degradation of the fermentable components of the juice by microorganisms.

The kinetic parameters of ethanol production for the pasteurized samples as a function of fermentation time is shown in Tables 1, 2 and 3 for samples pasteurized at 70, 80 and 90 °C, respectively. The fermentation efficiency for the samples stored at 70 °C was in the range of 89–93 %. The juice sample pasteurized at 80 °C, showed a variation in efficiency values ranging from 88 to 92 % and the samples pasteurized at 90 °C, the variation was in the range of 88–93 %. The ethanol yield for the samples pasteurized at 70 °C and stored at 35, 40 and 45 °C was in the range of 0.473–0.462 g g^{-1}, 0.473–0.454 g g^{-1} and 0.473–0.477 g g^{-1}, respectively. The samples stored at 35, 40 and 45 °C for juice samples pasteurized at 80 °C showed the range of 0.47–0.456 g g^{-1}, 0.47–0.461 g g^{-1} and 0.47–0.459 g g^{-1}, respectively, for ethanol yield. The ethanol yield for samples pasteurized at 90 °C and stored at 35, 40 and 45 °C was in the range of 0.473–0.466 g g^{-1}, 0.473–0.458 g g^{-1} and 0.473–0.461 g g^{-1}, respectively. On fermentation, the ethanol yields were almost consistent in all the three pasteurized temperatures and kept at a different storage conditions. However, the efficiency was more consistent and subsequent ethanol yield was observed in case of samples pasteurized at 70 °C and stored at 45,

Table 3 Kinetic parameters of batch ethanol production as a function of fermentation time by *Saccharomyces cerevisiae* strain ICTY417 using sweet sorghum juice samples (pasteurization at 90 °C for 48 h fermentation)

Juice samples (storage period /storage temperature)	Parameters (mean ± S.E.)			
	P^a (g l^{-1})	$Y^b_{p/s}$ (g g^{-1})	Q^c_p (g l^{-1} h^{-1})	E^c_y (%)
0 h storage period for 90 °C	63.39 ± 0.140	0.475 ± 0.615	1.320 ± 0.591	93.01 ± 0.717
3rd Day (35 °C)	63.98 ± 0.629	0.469 ± 0.576	1.333 ± 0.362	92.11 ± 0.488
3rd Day (40 °C)	61.92 ± 0.618	0.465 ± 0.923	1.29 ± 0.581	90.52 ± 0.808
3rd Day (45 °C)	61.24 ± 0.810	0.465 ± 0.728	1.276 ± 0.471	90.23 ± 0.209
5th Day (35 °C)	64.02 ± 0.692	0.468 ± 0.357	1.333 ± 0.407	91.70 ± 0.308
5th Day (40 °C)	60.79 ± 0.680	0.46 ± 0.404	1.267 ± 0.389	90.24 ± 0.037
5th Day (45 °C)	61.76 ± 0.746	0.464 ± 0.904	1.287 ± 0.875	90.98 ± 0.194
7th Day (35 °C)	64.3 ± 0.091	0.469 ± 0.305	1.339 ± 0.487	92.83 ± 0.795
7th Day (40 °C)	61.25 ± 0.651	0.460 ± 0.673	1.276 ± 0.772	89.99 ± 0.705
7th Day (45 °C)	60.73 ± 0.374	0.463 ± 0.266	1.265 ± 0.412	90.83 ± 0.795
9th Day (35 °C)	60.24 ± 0.676	0.446 ± 0.106	1.255 ± 0.040	91.44 ± 0.375
9th Day (40 °C)	60.7 ± 0.402	0.459 ± 0.905	1.264 ± 0.191	89.62 ± 0.772
9th Day (45 °C)	61.38 ± 0.491	0.461 ± 0.557	1.278 ± 0.368	90.72 ± 0.476
11th Day (35 °C)	62.68 ± 0.275	0.465 ± 0.684	1.306 ± 0.119	92.76 ± 0.374
11th Day (40 °C)	60.7 ± 0.237	0.458 ± 0.351	1.264 ± 0.835	88.81 ± 0.701
11th Day (45 °C)	60.89 ± 0.161	0.460 ± 0.467	1.268 ± 0.117	89.39 ± 0.621
13th Day (35 °C)	63.22 ± 0.309	0.463 ± 0.493	1.317 ± 0.147	93.03 ± 0.721
13th Day (40 °C)	60.81 ± 0.470	0.461 ± 0.297	1.266 ± 0.484	89.89 ± 0.220
13th Day (45 °C)	61.43 ± 0.491	0.458 ± 0.610	1.275 ± 0.451	91.08 ± 0.877
17th Day (35 °C)	60.41 ± 0.609	0.460 ± 0.728	1.258 ± 0.584	91.33 ± 0.099
17th Day (40 °C)	63.76 ± 0.892	0.458 ± 0.273	1.328 ± 0.092	88.37 ± 0.106
17th Day (45 °C)	62.11 ± 0.971	0.459 ± 0.509	1.348 ± 0.714	90.56 ± 0.579
21st Day (35 °C)	61.79 ± 0.114	0.466 ± 0.847	1.287 ± 0.627	92.31 ± 0.672
21st Day (40 °C)	62.14 ± 0.230	0.458 ± 0.561	1.294 ± 0.491	90.24 ± 0.182
21st Day (45 °C)	62.01 ± 0.931	0.461 ± 0.340	1.301 ± 0.146	90.13 ± 0.408

[a] P, actual ethanol concentration produced

[b] $Y_{p/s}$, ethanol yield

[c] Q_p, ethanol productivity

[d] E_y, percentage of conversion efficiency or yield efficiency

40 °C for samples pasteurized at 80 and 35 °C for samples pasteurized at 90 °C. Therefore, we can say that the samples pasteurised at 90 °C and stored at 35 °C which is approximately close to room temperature is the best preservation condition for storage and increasing the shelf life of the sweet sorghum juice. Some of the earlier studies indicated that the ethanol fermentation efficiency of >90 % was observed in frozen, autoclaved and juice containing 25 % sugar samples, whereas less than the above was observed in normal juice fermentation (Imam and Capareda 2011). The fermentation efficiency of around 90 % was also reported in the fermentations carried out under very high gravity conditions (Nuanpeng et al. 2011). Therefore, the conditions employed in the present work is beneficial for the enhancement of the shelf life of sweet sorghum juice and pasteurization was suggested as an efficient preservation method of the sweet sorghum juice samples and the fermentation efficiency was also maintained.

Conclusion

The results observed in the present study identified a suitable pasteurization temperature that was capable of preserving the fermentable sugars in sweet sorghum stalk juice and maintained the sugar profiles reasonably well at near room temperature, i.e. pasteurization at 90 °C followed by storage at 35 °C. The storage shelf life of the juice was extended up to 21 days and also enabled efficient bioconversion of the juice to ethanol. On the other hand, the control juice sample which was not preserved well

showed a significant reduction in the total soluble sugar content and thus resulted in a sharp decrease in the ethanol yield due to reduced fermentation efficiency. The highest fermentation efficiency of 93 % was recorded after 48 h of fermentation. Hence, the pasteurization method identified in the present study can be a cost-effective strategy to preserve fermentable sugars and retain the processing properties of the sweet sorghum juice during processing, transportation, and storage under normal conditions as compared to refrigerated conditions.

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